Royal Government of Bhutan

BHUTANESE JOURNAL OF AGRICULTURE

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Editorial

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FOREWORD

The Department of Agriculture is delighted to launch the fifth volume of the Bhutanese Journal of Agriculture (BJA). BJA is a print open access English language journal on agriculture and publishes research articles annually with the primary purpose of providing an additional mechanism to disseminate appropriate technologies, and knowledge and information in the agriculture sector.

Across cross-cutting themes like agronomy, postharvest, economics, agro-biodiversity, soil and water management, farming systems, pest and disease management and climate change, the journal covers research findings that are innovative and relevant to sustainable agriculture development in Bhutan.

These past couple of years have been difficult with the Covid-19 pandemic affecting everyone across the board. The department has, in its own little measures, made significant reforms in its approach to agriculture development so as to help deliver immediate recourse to our farmers as well as the economy. At the same time, our efforts in empirical and objective research to help develop data-driven technology should continue unabated. This volume which we are able to release uninterrupted is thus a testament to our collective resolve in seeking new and relevant technologies and effectively communicate them through a peer-reviewed platform.

Our editorial team, comprising members entirely from within the Department of Agriculture, is pushing hard to conform to international standards. Concerted efforts are underway to continuously improve the quality of the journal and we are glad that with every passing issue we have come out a step better. The experience has no doubt enriched our colleagues who seem to be closing the gap in designing research, carrying out analysis and putting across effective communication of their results, which in the final analysis underpin agriculture development.

I thank the authors and the reviewers alike for their contribution as well as the BJA Editorial Board for their added efforts in successfully taking out this edition. I wish everyone a resourceful reading.

Stay safe and best wishes.

(Yonten Gyamtsho)
DIRECTOR

EDITORIAL

The Bhutanese Journal of Agriculture (BJA) focuses on the documentation of newly generated technologies and understandings for improving agricultural processes, increasing crop yields and conserving agricultural resources in the Bhutanese context. The COVID-19 pandemic has underscored and reiterated the key message that we need to build a resilient and locally sustainable food production and distribution system to reduce dependency on imports. The strengths and positive attributes of domestic food production have become clearer as substantial quantities of vegetables that we consume now originate domestically. Hence, we hope that the articles presented in this edition will add to the current understanding of, and thereby help enhance food security and food systems without depleting our resource base.

The journal received 32 manuscripts that were reviewed by 17 experts including the editorial board members. The review reports were deliberated by a panel of reviewers in a three-day workshop. All the manuscripts that were accepted with major or minor revisions were returned to authors for improvement. A total of 16 manuscripts that successfully passed through a rigorous revision process have been accepted and are featured in this volume. Over the last two years, the journal received more quality articles. This positive trend is the direct outcome of the several rounds of training programmes organized by the Department of Agriculture on scientific paper writing and research methodologies for professionals with various agencies within the department.

This set of 16 interesting articles ranges from crop germplasm evaluation, testing of the innovations for our most sought-after clients -the farmers, to postharvest processing. Interesting research findings on rice, maize, finger millet, buckwheat, adzuki bean, dill, tomato, kiwi-fruit, citrus and apple are covered. We hope there is something for everyone, and interesting enough to whet our appetite for research, innovation and learning. With innovation from the AMC, farmers can now maneuver power-tillers up to 20 degrees gradient to mechanize more land for food production. For automation enthusiasts – a remote monitoring and control system for hydroponic may make for an exciting read.

Once again, we thank all authors, reviewers, facilitators and the journal editorial board members for their concerted effort and diligence in making this volume happen. On behalf of the editorial board, I would like the extend our sincere gratitude to all contributing institutions including the Agriculture Research and Development Centres at Wengkhar and Samtenling; the National Post Harvest Centre, Paro; the Agriculture Machinery Centre, Paro; and the National Centre for Organic Agriculture, Yusipang. I put on record my appreciation to the Agriculture Research and Extension Division and the DoA for providing the resources required to not only implement the research but also to make this edition a success. We hope that such an earnest effort to document evidence-based studies and their results will lend added credibility to our plans and programs aimed at helping us realize our mission to secure food for all Bhutanese people.

We wish you an intuitive reading.

(Yadunath Bajgai, PhD)
Editor-in-Chief

Table of Contents

Clean Citrus Planting Materials Production through Shoot tip Grafting and Biological Indexing

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ABSTRACT

Huanglongbing (Citrus Greening) is the most devastating citrus disease caused by phloem-inhabiting bacteria carried by the insect vector, Asian citrus psyllid (Diaphorina citri). In Bhutan, the production and movement of citrus seedlings and new orchard establishment have been restricted after the Citrus Greening disease was detected in most citrus orchards. To address this problem, Agriculture Research and Development Centre-Wengkhar has initiated research on clean citrus propagation through shoot tip grafting. This study assessed the efficiency of shoot tip grafting for the production of clean planting materials. Sweet orange was used as an indicator plant to study disease transmission and symptom expression in some of the potential and released citrus varieties. A total of 15 plants, 3 plants of each cultivar (AREP-1, AREP-2, Aoshima, Wengkhar Tshelu-2 and Yoshida Ponkan) produced through shoot tip grafting were used for bio-indexing. The cultivars were grafted on indicator plants using different grafting techniques (T-budding, side grafting, wedge, and split grafting). A total of twenty-six samples with two samples per cultivar was sent to the National Plant Protection Centre for PCR analysis. The result of PCR showed negative result for Huanglongbing. Thus, shoot tip grafting could be one potential method for clean citrus planting material production.

1. Introduction

The citrus fruit is one of the most important cash crops - both for local as well as exports - among which the mandarin (*Citrus reticulata* Blanco) is pre-dominate and the most widely distributed in Bhutan, both in quantity and value. It occupies more than 5,086 hectares of land (RSD, 2020) in Bhutan. Though it is grown in 17dzongkhags, certain dzongkhags like Samtse, Sarpang, Dagana, Tsirang, Chhukha and Pemagatshel have emerged as the leading producers. The average annual production of citrus is about 38,100 Mt, but both annual production and yield have decreased over the years due to various factors such as incidence of pests (citrus fruit fly, leaf miner) and diseases (Huanglongbing, citrus canker, tristeza, exocortis,

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phytophthora) and improper management of orchards by the farmers. Most of the citrus orchards in Bhutan are grown in hilly areas where farmers cannot manage their orchards timely and properly. In some dzongkhags like Sarpang and Tsirang, the citrus orchards were established illegally on state land near forest areas which encourage wild animals, particularly monkeys to feed on the crop, further jeopardizing citrus production in the country. The low productivity of the citrus is also attributed to the use of seedlings produced from seeds of unknown sources instead of using seedlings produced through grafting and budding where rootstocks such as Wengkhar Rato-1, Wengkhar Rato-2 and C-35 play a significant role in combating abiotic and biotic stresses. The seedlings produced through seeds not only have long unproductive juvenile phases but also the young plants are more prone to pests and diseases, thereby resulting in further decline of the citrus industry in Bhutan. The most common factor associated with citrus decline is Huanglongbing (HLB) disease, which was first reported in Bhutan in 2003 (Doe, Om, Dorji, Dorji et al., 2003). The disease is caused by *Candidatus Liberibacter asiaticus* (a fastidious phloem limited bacterium), an obligate gram-negative bacterium (Sheng Li, Wu, Duan, Singerman et al., 2020) and is transmitted through infected budwood and the citrus psyllid, *Diaphorina citri (Batool, Iftikhar, Mughal, Khan et al., 2007)*. The disease is also transmitted by the parasitic weed dodder (Sheng Li et al., 2020). Following the detection (PCR test report from NPPC, 2015) of HLB from the Regional Seed Centre at Samtenling, Sarpang, the Department of Agriculture re-emphasized that only the National Seed Centre (NSC) is authorized to produce citrus seedlings. Currently, the Regional Seed Centre in Jachephu, Trashiyangtse (located about 1,800 m above mean sea level) carries out citrus seedling production as Asian citrus psyllid remains active at an elevation of 1000 m and below (Manandhar, Malla, & Sah, 2004).

Huanglongbing results in significant fruit yield and quality losses due to severe deterioration of tree health (Vidalakis, Garnsey, Bash, Greer et al., 2004). The HLB has become one of the major challenges for both farmers and the Royal Government of Bhutan, especially for the Ministry of Agriculture and Forests, as the infected trees become less productive within 2 to 6 years of infection (Kinley Dorji, Lakey, Chophel, Dorji et al., 2016). Asian citrus psyllid, the main vector of the disease in Bhutan (Halbert & Núñez, 2004) is active only below 1000 m, so symptoms of citrus greening do not occur or are not very well expressed or simply disappear at higher elevations (Ajene, Khamis, Ballo, Pietersen et al., 2020), especially in places such as Wengkhar (Mongar) which is located at around 1,700 m above sea level.

Thus, against this backdrop, ARDC Wengkhar initiated the production of clean citrus seedlings through shoot tip grafting followed by bio-indexing and PCR to rule out any possibility of HLB. This is also believed to be the best techniques to obtain seedlings free of virus and other disease such as tristeza, psorosis, exocortis and also HLB (Navarro, Roistacher, & Murashige, 1975). The plants produced through shoot tip grafting are found to be disease free, particularly the HLB virus, when the shoot tip grafted citrus samples are bio-indexed and PCR analysed. The production of citrus through shoot tip grafting is the best techniques in producing disease-free citrus which will ultimately help revive the citrus industry in the country. However, due to the lack of laboratory facilities and limited knowledge and technical expertise in biotechnology, production of clean citrus seedlings both in terms of quality as well as the numbers is a huge challenge.

2. Materials and Method

Disease-free rootstocks (seeds imported from Australia), mainly C35, were raised under laboratory conditions with a room temperature of 25 -27ºC. After 2 to 3 weeks, once they have attained graft size, shoot tip grafting was carried out. The vigorous, healthy, and successful STG plants (that attained 2 to 3 leaves stage) were potted in bigger pots and shifted to protected greenhouse for further approach grafting. Out of 55 successful shoot tip grafted plants, to encourage faster growth, 26 plants were further approach-grafted onto the bigger and vigorous C35 rootstocks maintained in an insect-proof greenhouse.

One of the methods to confirm disease (HLB) free status of plants is biological indexing (Razi, Khana, Jaskania, & Basrab, 2012). Biological indexing is a method to detect pathogens before laboratory analysis is carried out. Though there are other indicator plants (plants that can easily express signs and symptoms of the disease) for HLB, such as sour lime, mandarin and grapefruit, sweet orange is considered a more effective and good indicator for greening disease as it expresses greening symptoms very easily as compared to other citrus species (Das, 2008). Hence, in this study, scion woods from the above successfully grafted plants were collected and again grafted (Bhandari, Basnet, & Khanal, 2021) onto the indicator plants (sweet orange) to observe if any signs and symptoms of HLB are expressed. For further reconfirmation of disease-free status, leaves and stem samples were collected from those grafted onto indicators plants and sent to the National Plant Protection Centre for PCR analysis.

2.1 Shoot Tip Grafting

Shoot tip grafting was carried out from January to May 2018 and 2019, where two to three-week-old, etiolated rootstocks were topped and shoot tips were grafted. Grafted plants were grown in a controlled growth room at a temperature of 25-27°C and humidity of 70-75%. The one- to two-month-old STG plants with a height of about 15 to 20 cm and a stem thickness of 0.5 to 1.5 mm were re-potted and kept in a controlled insect-proof citrus greenhouse (K. Dorji & Lakey, 2015). The following are the standard procedures for grafting shoot tips.

(A)   (B)

Figure 2. Rootstock seeds planted in potting media (A) and two weeks old rootstocks (B)

2.1.1 Shoot tip grafting rootstock preparation

The rootstock seeds imported from Australia were first de-coated and sterilized for 10 min with a 0.5% sodium hypochlorite solution containing 2-5 drops of 0.1% Tween-20. Then, seeds were thoroughly rinsed 3-4 times with sterile distilled water and planted in potting soil (perlite, coco peat) and allowed to germinate for 2-3 weeks under dark conditions at 25-27°C in the germination chamber. The 2-3 weeks old, etiolated rootstocks were used for grafting the shoot tips.

2.1.2 STG scion preparation and grafting

Citrus buds of 15 released cultivars (Table 2) were collected from the germplasm collection greenhouse. The collected bud sticks were 5-10 cm long and contained at least 2-3 opened buds. The leaves of the bud canes were removed, and surface sterilized with 0.5% sodium hypochlorite solution containing 2-5 drops of 0.1% Tween-20 for 5 minutes. They were then thoroughly rinsed (3-4 times) with sterile distilled water. The 3-week old rootstock was decapitated at about 1.5 cm from the hypocotyl (Starrantino & Caruso, 1988). The cotyledons and root tips were cut off. Using a sharp surgical knife, an inverted T-cut is made on the rootstock under the dissecting microscope. Again, under the microscope, the shoot tip (apical

meristem) with a length of about 0.2 mm is removed from the bud stock and aseptically placed on the inverted T-cut of the rootstock. Then the inverted T-cut is carefully wrapped with para-film to prevent immediate drying of the shoot tip (Figure 4 shows the details of shoot tip grafting). The grafted plants were transplanted into the planting medium and kept in a germination chamber with 16 hours light/day at a temperature of 25-27ºC.

(A)   (B)

(C)   (D)
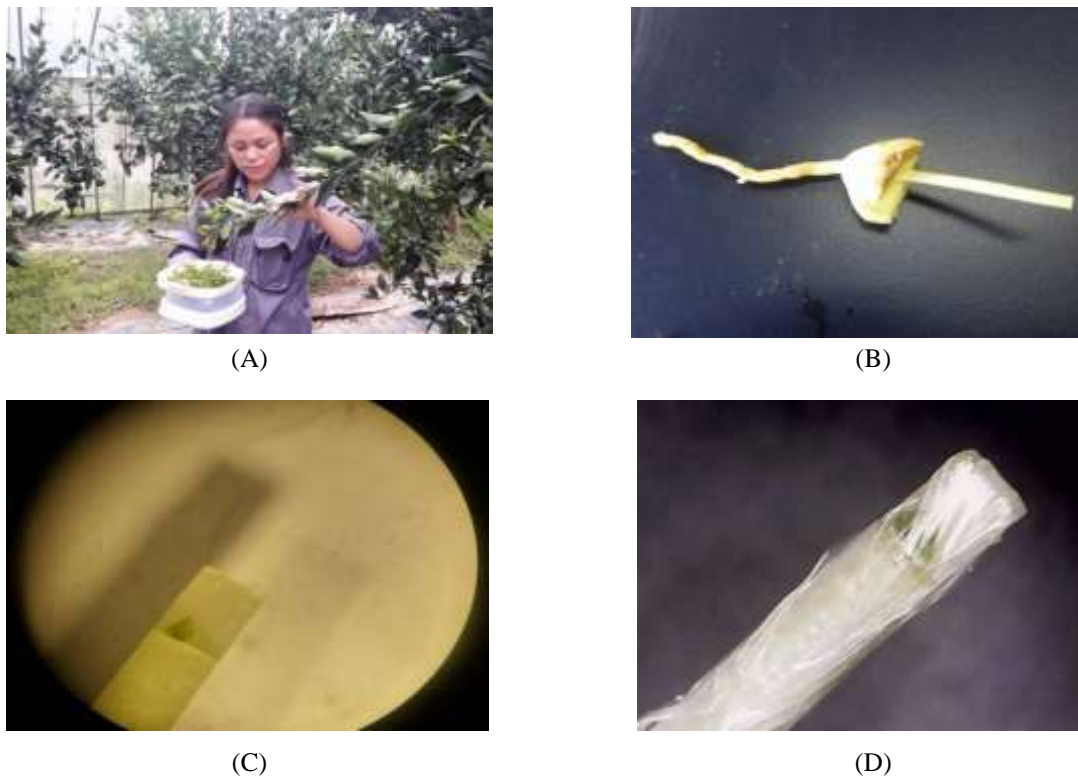
Figure 3. STG Scion collection (A), Decapitated rootstock (B), Apical meristem aseptically placed on the inverted T-cut of the rootstock (C) and T-cut is carefully wrapped with parafilm (D)

2.2 Bio-indexing of citrus varieties

T-budding, side grafting, wedge grafting, and split grafting were used to graft onto indicator plants.

T-budding: In this method, an annual rootstock with a height of 25-30 cm and a diameter of 1-2 cm is used for budding. The T-shaped cut is made on the rootstock 12-15 cm from the base with a vertical cut of 1-2.3 cm and a horizontal cut of 0.8-1 cm. Similarly, a 3.4-cm bud is inserted into the T-shaped cut with two flaps of bark and wrapped with a 300-gauge polyethene strip.

Side grafting: In this method, a similar size and height are used. A shallow inward cut of about 2-5 cm is made on the mother plant at the desired height. On the scion, a similar inward cut and a small slanting cut is made as on the rootstock so that the cambium layer of both sides match as much as possible and is wrapped with polyethene and parafilm.

Wedge and split grafting: In this method, the upper part of the rootstock is topped at a height of 15-20 cm from the base and a 3-cm split cut is made. A wedge-shaped cut of 2-3 cm is made on the scion wood, which is put on the base and fixed with a strip of polyethene.

Table 1. Grafting methods employed

S.N.	T-Budding	Side grafting	Wedge and cleft grafting
1	Aoshima	Aoshima	Aoshima
2	AREP-1	AREP-1	AREP-1
3	AREP-2	AREP-2	AREP-2
4	Wengkhar Tshelu-2	Wengkhar Tshelu-2	Wengkhar Tshelu-2
5	Yoshida ponkan	Yoshida ponkan	Yoshida ponkan

The success rate of grafting was promising for all the methods used. However, the new shoots emerging from the side grafting were more vigorous and grew faster compared to those from T-budding, wedge and cleft grafting.

3. Results and Discussion

The results are discussed under three topics: shoot tip grafting, biological indexing, and PCR analysis.

3.1 Shoot tip grafting

Fifteen different cultivars were shoot tip grafted. Out of 167 plants that were grafted, 55 plants were successful, of which 26 were successfully approach-grafted in the protected greenhouse for biological indexing and PCR analysis.

(A)   (B)

<div align="center">(C)   (D)</div>

Figure 4. Plantlet 1-2 week after STG (A), 2-3 weeks old STG plant (B), Successful STG plants (C) and then potted plants (D)

Table 2. Details of shoot tip grafting (STG) and success

S.N.	Citrus variety	Rootstock used	No. of plants grafted (STG)	No. of Successful grafts (STG)	No. of successful approach grafts
1	Tarku	Grapefruit	07	00	None
2	Wengkhar Tshelu II	Grapefruit	08	06	06
3	AREP I	Grapefruit	13	03	None
4	AREP I	C-35	11	03	None
5	Wengkhar Tshelu Ngarm	C-35	10	03	02
6	Otshu-4	C-35	19	04	None
7	Wengkhar Tshelu Drukchu	C-35	06	03	None
8	Local Selection	Swingle Citrumelo	13	04	None
9	Wengkhar Tshelu Ngarm	Swingle Citrumelo	28	10	04
10	Wengkhar Tshelu Ngarm	*Poncirus trifoliata*	08	02	01
11	Aoshima	C-35	10	04	04
12	Yoshida Ponkan	C-35	09	01	01
13	AREP II	C-35	11	07	05
14	Kiyomi	C-35	05	02	02
15	Junar	C-35	09	03	01
		TOTAL	167	55 (32.9%)	26 (47.27%)

3.2 Biological indexing

Visual symptoms were observed 30 days after grafting and persisted for up to 3 months (Table 3) at 30-day intervals. At the first observation, 30 days after grafting, some plants started showing symptoms on the leaves and the symptoms tend to decrease after 40-45 days. The symptoms were blotchy leaves and resembled zinc deficiency. However, at 90 days after grafting, all plants were very healthy. This indicated that no greening pathogens were present, and the symptoms were likely caused by nutrient deficiency or micronutrient deficiency (Silva-Stenico, Pacheco, Pereira-Filho, Rodrigues et al., 2009).

Table 3. Visual observations on indicator plants

S.N.	Time interval	Citrus varieties	Symptoms expressed	Healthy plants
1	30 days after grafting	Aoshima	Normal leaf size, symmetrical chlorosis in some leaves (1)	2 and 3
		AREP-1	Blotchy appearance of some of the leaves with thickened veins (2&3)	1
		AREP-2	Yellowing and varied chlorotic pattern with downward curling of leaves (1,2 &3)	None
		Wengkhar Tshelu-2	None	All
		Yoshida Ponkan	Symmetrical bright yellowing of outer leaf portion and thickening of the vein (1,2 &3)	None
2	60 days after grafting	Aoshima	Normal leaf size, symmetrical chlorosis in some leaves (1,2&3)	None
		AREP-1	None	All
		AREP-2	Yellowing and varied chlorotic pattern with downward curling of leaves (1&2)	3
		Wengkhar Tshelu-2	Yellowing and blotchy on some leaves (2)	1&3
		Yoshida Ponkan	Symmetrical bright yellowing of outer leaf portion and thickening of the vein (3)	1&2
3	90 days after grafting	Aoshima	None	All
		AREP-1	None	All
		AREP-2	None	All
		Wengkhar Tshelu-2	None	All
		Yoshida Ponkan	None	All

3.3 PCR analysis

In the final analysis, to fully confirm the absence or presence of HLB pathogens, leaves and stem samples from 15 bio-indexed plants (leaves and stems) were sent to the National Plant Protection Centre (NPPC) for polymerase chain reaction (PCR) analysis. Real-time polymerase chain reaction analysis was performed using Rotor-Gene Q (Qiagen) with the primer-probe combinations, for Clas and the internal control, cytochrome oxidase gene (COX), (Shalan Li, Zhang, Liu, Liu et al., 2020). A reaction volume of 25 µL with cycling conditions of pre-incubation at 50 °C for 2 min followed by 95 °C for 10 min and 40 amplification cycles of 95 °C for 30 sec and 58 °C for 40 sec were used. Positive and non-template controls were included. Samples were considered positive if both the Clas and COX probes showed threshold cycle (Ct) values in the range of zero to less than or equal to 36. All the samples were negative for HLB pathogen (Table 4), which shows that citrus plants grafted through the shoot tip are free from HLB pathogens.

Table 4. PCR analysis report from NPPC

Sample Number	Ct values	
	Clas	**COX**
437-21	0	14.58
438-21	0	16.64
439-21	0	14.81
440-21	0	13.41
441-21	0	11.59
442-21	0	13.96
443-21	0	15.3
444-21	0	13.6
445-21	0	11.02
446-21	0	9.79
447-21	0	10.14
448-21	0	16.55
449-21	0	12.26
450-21	0	12.53
451-21	0	14.32
452-21	0	13.41
453-21	0	10.4
454-21	0	10.45
455-21	0	10.63
456-21	0	9.18
457-21	0	10.23
458-21	0	9.17
459-21	0	14.93
460-21	0	12.18
461-21	0	10.36
462-21	0	12.38
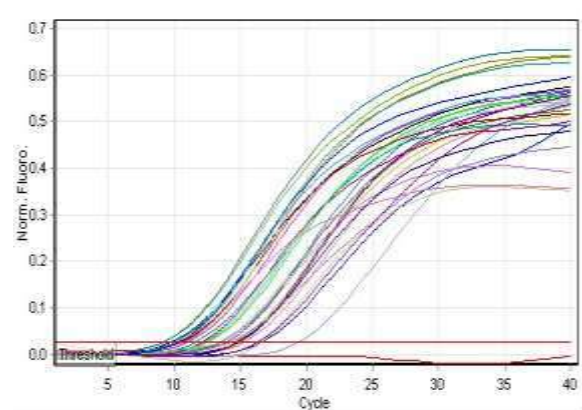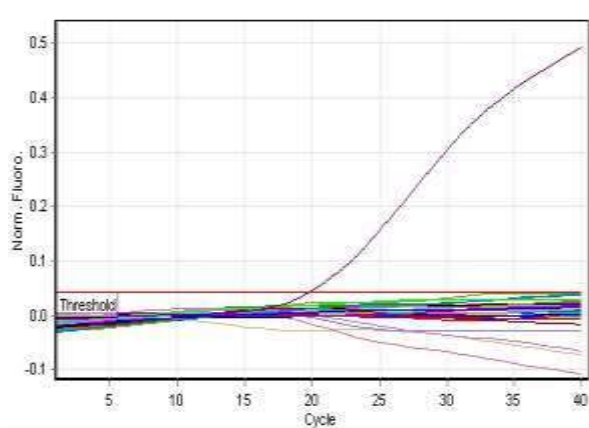
Figures 5 and 6. The standard curve derived during the PCR analysis with all samples showing Clas Ct values equal to zero and the COX probe Ct values for all samples ranged from 9.17 to 19.40

4. Conclusion

The spread of citrus greening has significantly affected the citrus industry in Bhutan since its initial detection, especially in the major citrus growing areas of Bhutan. Therefore, in this research, we studied some of the techniques to produce HLB free seedlings. Citrus plants produced using the shoot tip grafting method proved to be one of the most effective techniques for producing disease-free plants in all crops, especially citrus. The study reconfirms the precept that eliminating pathogens and keeping the same plants in a standard insect-proof greenhouse is very effective in protecting the plants from further spread of the HLB vector. The successful grafts produced through shoot tip grafting and then biological indexing showed no symptoms of greening except some nutritional deficiency symptoms. Besides production and use of cleaning planting materials, proper orchard management and protection from HLB vectors are critical. Therefore, the integrated application of all the above methods will not only help to manage the vectors and HLB disease but also help to revamp the citrus industry in the long run.

References

Ajene, I. J., Khamis, F., Ballo, S., Pietersen, G., van Asch, B., Seid, N., Azerefegne, F., Ekesi, S., & Mohamed, S. (2020). Detection of Asian Citrus Psyllid (Hemiptera: Psyllidae) in Ethiopia: a new haplotype and its Implication to the proliferation of huanglongbing. *Journal of Economic Entomology, 113*(4), 1640-1647. doi:10.1093/jee/toaa113

Batool, A., Iftikhar, Y., Mughal, S. M., Khan, M. M., Jaskani, M. J., Abbas, M., & Khan, I. A. (2007). Citrus greening disease-a major cause of citrus decline in the world-a review. *Hort. Sci. (Prague), 34*(4), 159-166.

Bhandari, N., Basnet, M., & Khanal, S. (2021). Standardization of grafting time of mandarin (*Citrus Reticulata* Blanco) in central mid hill of Nepal. *International Journal of Fruit Science, 21*(1), 599-608. doi:10.1080/15538362.2021.1875964

Das, A. K. (2008). *Citrus greening (Huanglongbing) disease in India: present status and diagnostic efforts.* Paper presented at the International Research Conference on Huanglongbing, Florida, USA.

Doe, D., Om, N., Dorji, C., Dorji, T., Garnier, M., Jagoueix-Eveillard, S., & Bové, J. M. (2003). First report of "*Candidatus* Liberibacter *asiaticus*", the agent of citrus Huanglongbing (ex-greening) in Bhutan. *Plant disease, 87*(4), 448-448. doi:10.1094/PDIS.2003.87.4.448A

Dorji, K., & Lakey, L. (2015). *Citrus nursery managemnt- a technical guide*. Thimphu: Department of Agriculture, Ministry of Agriculture & Forests, Royal Government of Bhutan

Dorji, K., Lakey, L., Chophel, S., Dorji, S. D., & Tamang, B. (2016). Adoption of improved citrus orchard management practices: a micro study from Drujegang growers, Dagana, Bhutan. *Agriculture & Food Security, 5*(1), 1-8. doi:10.1186/s40066-016-0050-z

Halbert, S. E., & Núñez, C. A. (2004). Distribution of the Asian citrus psyllid, *Diaphorina citri* Kuwayama (Rhynchota: Psyllidae) in the Caribbean basin. *Florida Entomologist, 87*(3), 401-402. doi:10.1653/0015-4040(2004)087[0401:DOTACP]2.0.CO;2

Li, S., Wu, F., Duan, Y., Singerman, A., & Guan, Z. (2020). Citrus greening: management strategies and their economic impact. *HortScience, 55*(5), 604-612. doi:10.21273/HORTSCI14696-19

Li, S., Zhang, J., Liu, H., Liu, N., Shen, G., Zhuang, H., & Wu, J. (2020). Dodder-transmitted mobile signals prime host plants for enhanced salt tolerance. *Journal of Experimental Botany, 71*(3), 1171-1184. doi:10.1093/jxb/erz481

Manandhar, R., Malla, S., & Sah, D. N. (2004). Population dynamics of citrus psylla (Diaphorina citri Kuwayama) in the western hills of Nepal and spread of citrus greening disease. *Agricultural research for enhancing livelihood of Nepalese people, 30*, 178.

Navarro, L., Roistacher, C., & Murashige, T. (1975). Improvement of shoot-tip grafting in vitro for virus-free citrus. *Journal of the American Society for Horticultural Science, 100*(5), 471-479.

Razi, M. F., Khana, I. A., Jaskania, M. J., & Basrab, S. M. (2012). Graft transmission and biological indexing of huanglongbing on local citrus germplasm. *Sci Intl, 24*(2), 153-157.

RSD. (2020). *Bhutan RNR Statistics 2019*. Thimphu: Renewable Natural Resources Statistics Division (RSD), Ministry of Agriculture and Forests, Royal Government of Bhutan.

Silva-Stenico, M. E., Pacheco, F. T. H., Pereira-Filho, E. R., Rodrigues, J. L. M., Souza, A. N., Etchegaray, A., Gomes, J. E., & Tsai, S. M. (2009). Nutritional deficiency in citrus with symptoms of citrus variegated chlorosis disease. *Brazilian Journal of Biology, 69*, 859-864.

Starrantino, A., & Caruso, A. (1988). The shoot-tip grafting technique applied in citriculture. *Acta Horticulture*(227), 101-103. doi:10.17660/ActaHortic.1988.227.13

Vidalakis, G., Garnsey, S. M., Bash, J. A., Greer, G. D., & Gumpf, D. J. (2004). Efficacy of bioindexing for graft-transmissible citrus pathogens in mixed infections. *Plant disease, 88*(12), 1328-1334. doi:10.1094/PDIS.2004.88.12.1328

Effect of Irrigation Intervals and Manure Rates on Agronomic Parameters of Dill at Different Agro-ecological Zones of Bhutan

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ABSTRACT

The effect of different irrigation intervals and manure rates on agronomic parameters of dill were evaluated under three agro-ecological zones of Bhutan: ARDC-Bajo located at 1200 m asl, ARDC–Wengkhar at 1,700 m asl, and NCOA–Yusipang at 2700 m asl. The split-plot design arranged in randomized complete blocks with three replications was employed. The four levels of irrigation interval (control, every 3 days, every 7 days, and every 14 days) were allotted to main plots and four levels of manure (Control, 4 tons/acre, 8 tons/acre and 12 tons/acre) were allocated to subplots. The results showed that dill can be successfully grown in all agro-ecological zones under study. Fresh yield obtained at ARDC-Wengkhar (1.92 tons/acre) was significantly higher compared to that at NCOA-Yusipang (0.52 tons/acre) but not significant to that at ARDC-Bajo (1.41 tons/acre). Interaction of irrigation interval and manure rate significantly influenced plant height (P=0.02) and stem diameter (P=0.03) at NCOA–Yusipang, and stem per plant (P=0.02) at ARDC-Wengkhar. The irrigation interval of 3 days produced significantly better results except for the fresh to dry weight ratio at NCOA-Yusipang. Irrigation interval of 7 days yielded better results at ARDC-Bajo and ARDC-Wengkhar except for FDR at ARDC-Wengkhar. The increase in irrigation interval beyond 7 days resulted in 62% and 25% lower fresh yields at ARDC-Bajo and NCOA-Yusipang respectively. In contrast, irrigation at 3 days interval at ARDC–Wengkhar decreased the yield by 55% compared to 7 days interval. The manure treatments did not influence any of the parameters assessed at all sites. However, MR 8 tons/acre at NCOA–Yusipang, MR Control at ARDC-Wengkhar and MR 12 tons/acre at ARDC-Bajo yielded higher fresh yields compared to other manure treatments. We recommend the combined application of IR 7 days x MR 12 tons/acre, IR 7 days x MR 4 tons/acre, and IR 3 days x 8 tons/acre at ARDC–Bajo, ARDC-Wengkhar and NCOA–Yusipang respectively. Further, this research provides a reference guide for agricultural extensionists during promotional, advocacy and capacity development programs.

Keywords: *Anethum graveolens; Irrigation Interval; Manure Rate; Split-plot Design*

1. Introduction

Dill (*Anethum graveolens* L.) is an erect, annual aromatic herb belonging to the Apiaceae family which grows up to 50 to 150 cm tall. It is grown in the Mediterranean region, Europe,

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central and southern Asia (Kaur & Arora, 2010). The herb contains essential oils (2-4%), carbohydrates (36%), protein (15.68%), fiber (14.80%), ash (9.8%), fatty oil, and minerals (Carrubba, Catalano, & Bontempo, 2010; Kaur & Arora, 2010). The herb is reported to have a wide range of medicinal uses such as antispasmodic, carminative, diuretic, stimulant and stomachic (Charles, 2012). It is also used to add flavour to different Egyptian foods, bread, pickles, and soup (Charles, Simon, & Widrlechner, 1995; Elsayed, Glala, Abdalla, El-Sayed, & Darwish, 2020; Hassan, 2015).

In the Bhutanese traditional food and farming system, people usually grow and consume only a few herbs such as coriander, chives, and spring onions. The cultivation of western culinary herbs including dill is very limited and the small domestic requirements were met through imports from Thailand and India. The increased tourist inflow, pizza huts and pastry in urban towns and awareness of the health benefits of herbs amongst Bhutanese have increased the demand for herbs in the country. As there were no officially released varieties, it was very challenging for the Department of Agriculture to promote the commercial cultivation of herbs in the country. The need for the introduction and evaluation of varieties of herbs including dill has become urgent and necessary.

There are many factors affecting the successful cultivation of crops such as seed variety and quality, planting time, planting methods, spacing, intercultural operations including plant nutrients and water requirements. In the context of climate change, plant nutrient and water availability are the most important factors to achieve good plant growth, development, and potential yield (Gerami, Moghaddam, Ghorbani, & Hassani, 2016) that facilitates dissolutions and uptake of micronutrients by the plants (Ghassemi-Golezani, Rezaeipour, & Alizadeh-Salteh, 2016; Naomi, Mwanarusi, & Musyoka, 2014). Delfine, Tognetti, Desiderio, and Alvino (2005) estimated that on average, the decrease of yield due to water shortage is more than 25% in the world. Also, water deficiency induces various physiological and metabolic responses resulting in stomatal closure, a decline in growth rate, accumulation of solute and antioxidants, reduction in photosynthesis and transpiration, and the expression of stress-specific genes (Jones & Tardieu, 1998; Tsamaidia, Dafererab, Karapanosa, & Passama, 2017). In terms of aromatic herbs, water scarcity directly contributes to decreased plant height, fresh and dry weight, reduce leave area, oil yield and composition (Baher, Mirza, Ghorbanli, & Bagher Rezaii, 2002; Charles et al., 1995; Simon, Reiss-Bubenheim, Joly, & Charles, 1992; Zehtab-Salmasi, Javanshir, Omidbaigi, Alyari, & Ghassemi-Golezani, 2001).

In addition, fertilization is reported to be one of the major factors affecting growth and development including yield, and quality of dill (Elsayed et al., 2020). The excessive use of chemical fertilizer in the recent past has resulted in health and environmental hazards (Sharma & Singhvi, 2017). He also argued that the use of organic alternatives could be the answer to reducing such destruction to nature and human health. Organic manure improves soil's physical, chemical, and biological properties and enhances the moisture retention capacity and nutrient uptake (Rostaei, Fallah, Lorigooini, & Abbasi Surki, 2018). Studies have also shown that the application of organic manure can increase quantity as well as the quality of crops. However, studies related to enhanced use of irrigation water and organic manure for sustainable herb farming are largely missing particularly in the Bhutanese context. Further, Bhutan with its development philosophies deeply rooted in love for nature and sustainable development goals, it has become necessary to develop alternative solutions in terms of efficient water use and organic manure without compromising the final output. Hence, the present study investigated i) optimum irrigation interval and manure rate for production of dill, ii) study their respective as well as combined influence on agronomic parameters, and iii) study the performance of dill under three agro-ecological zones of Bhutan.

2. Materials and Method

2.1 Study sites

The study was conducted from 11 March 2021 to 15 July 2021 in three agro-ecological zones of Bhutan. The Agriculture Research and Development Centre, Bajo (ARDC-Bajo) represents low altitude (1200 masl) located at latitude of 27°29'25.0' N and longitude of 89°53'51'' E. The Agriculture Research and Development Centre, Wengkhar (ARDC-Wengkhar) represents mid-altitude (1700 masl) located at latitude of 27°16'25" N and longitude of 91°16'13" E. The National Centre for Organic Agriculture, Yusipang (NCOA Yusipang) representing high altitude (2,700 masl) located at latitude of 27°27'49.1" N and longitude of 89°42'41.3" E. ARDC Bajo falls under the humid subtropical region with clay type of soils, while ARDC Wengkhar and NCOA Yusipang fall under the dry sub-tropical region with loamy clay soils and warm temperate region with sandy clay soils respectively.

2.2 Study Design

The performance of dill was evaluated using four levels of irrigation intervals: i) No irrigation, ii) at sowing and every after three days, iii) at sowing and every after seven days, iv) at sowing and every after 14 days and four levels of manure rate i) 0 tons/acre, ii) 4 tons/acre, iii) 8 tons/acre, iv) 12 tons/acre using split plot based on randomized complete block design with

three replicates. The irrigation interval treatments were allotted as main plots and manure rates as the sub-plots. The field was ploughed, levelled, and divided into plots measuring 2×1 m with buffer distance between treatments and replications at 25 cm and 50 cm respectively. The manure treatments were then administered as per the result of randomization of treatments in STAR Software (Statistical Tool for Agriculture Research) version 2.0.1. The manure was thoroughly incorporated into the soils before sowing the seeds. The dill seed (2 seeds each) introduced from Corona seeds Inc., USA was sown at 1 to 2 cm depth with the plant to plant and row to row distance of 30 cm. The first irrigation was uniformly provided at field capacity for all the treatment plots to facilitate seed germination. A total of 10 plants in each treatment plot were maintained by randomly removing one of the plants between 10-15 days after the seed germination. The crop was harvested when the leaves were still tender. The details of trial management including intercultural operations are provided in Table 1. Weeding was done depending upon the weed pressure, while the irrigation was given at field capacity at prescribed treatment intervals.

Table 1. Record of trial management and intercultural operations at three study sites

Activities	ARDC Bajo	ARDC Wengkhar	NCOA Yusipang
Sowing Date	11 March, 2021	27 April, 2021	9 April, 2021
Germination (DAS*)	13	23	13
Crop duration (DAS)	76	80	75
Number of hand weeding	3	3	2
Number of irrigations applied for each treatment			
a) Control	0	0	0
b) 3 days interval	25	27	25
c) 7 days interval	11	12	10
d) 14 days interval	6	6	6

*DAS: Days After Sowing

2.3 Data collection and preparation

The data for different parameters like plant height, stem diameter, number of stems per plant, and fresh weight were recorded at the time of harvest. The data collection was done from 50 % of the total plant population from all the treatment plots and in the case where the plant population is less than 50%, data were recorded from all the live/standing plants. The plant height was measured from the ground surface to the tip of the main shoot using a measuring tape, while the stem diameter was measured just above the collar region using a vernier calliper. Similarly, the fresh weight of the sample plants was measured using a digital weighing balance. The sample plants from all the treatment plots were tagged and dried for a week using a solar dryer after which the dry weights were measured. However, the dry weight of the sample plants

could not be measured at ARDC-Bajo due to technical issues with the dryer and was therefore excluded from statistical analysis and inference in this article. The fresh yield per plot (kg/plot) and per acre (ton/acre) were calculated using the formula (i) and ii) respectively. The same formulae were used to obtain dry yield per plot (kg/plot) and per acre (ton/acre). The mean value and standard deviation of the mean in parenthesis are presented.

$$py = \frac{w * n}{1000} \qquad \text{(i)}$$

Whereas,
  py is the plot yield in kg,
  w is the average fresh or dry weight of sample plants in grams,
  n is the total number of live/standing plants in respective treatment plots.

$$y = \frac{py * 4047}{2 * 1000} \qquad \text{(ii)}$$

Whereas,
  y is the fresh or dry yield per acre in tons
  py is the fresh or dry plot yield in kg

2.4   Data analysis

Data were statistically analyzed in R statistical software (R Core Team, 2021) using a two-way analysis of variance (ANOVA) for split-plot experimental designs provided by the r-package 'agricolae' (de Mendiburu, 2019) followed by mean separation using Tukey Honest Significance Difference test. The differences in the agronomic parameters between study sites were analyzed using one-way analysis of variance (ANOVA) taking sites as treatments. The alpha value was set at $P = 0.05$ to detect statistically significant differences between all the comparisons made in this article. The graphical representations of the statistical data were implemented using r-package 'ggplot2' (Valero-Mora, 2010) and seaborn library in Python (Bisong, 2019). A correlation analysis between agronomic parameters was investigated using the Pearson Correlation algorithm in pandas library in Python (McKinney, 2011).

3. Results and Discussion

3.1 Effect of irrigation interval, manure rate and their interaction on agronomic parameters of dill at ARDC-Bajo

The result presented in Table 2 showed that both the irrigation interval (IR) and manure rates (MR) did not significantly influence any of the agronomic parameters. However, in the case of irrigation interval treatments, 7 days irrigation interval treatment yielded better outputs compared to other treatments while the least was obtained from the control indicating irrigation did influence though not significantly. All the parameters followed a similar trend where the yields increased with longer intervals of irrigation up to 7 days and then decreased.

The overall decrease in all the parameters of dill plants with a decrease of irrigation interval beyond 7 days could be due to a decrease in the cell enlargement and leaf senescence resulting from reduced turgor pressure. It could also be due to reduced photosynthesis activity resulting from alteration in crop canopy when irrigation was applied at a lower interval. Similar findings were reported in oregano and parsley where the decreased interval of irrigation resulted in a decrease in yield and other agronomic parameters (Gerami et al., 2016; Petropoulos, Daferera, Polissiou, & Passam, 2008). On the other hand, the poor performance of dill plants under no irrigation treatment could be due to limited water availability at the rhizosphere to facilitate nutrient uptake (Shao, Chu, Jaleel, & Zhao, 2008). It could also be due to the mechanism where the plant biomass was significantly reduced to increase water uptake under water-stressed conditions (Gerami et al., 2016; Tsamaidia et al., 2017). Shao et al. (2008) also reported that the reduction in the plant biomass and agronomic traits under the water-stressed condition is due to inhibition of physiological and biochemical processes, such as photosynthesis, respiration, ion uptake, carbohydrates, nutrient metabolism, and hormones. While our findings conform with other reports (Gerami et al., 2016; Ghassemi-Golezani et al., 2016; Gwari, Gambo, & Kabura, 2014; Petropoulos et al., 2008), such studies require precise control over various climatic to draw concrete conclusions.

In the case of manures, taller plant height (35.77 cm), bigger stem diameter (14.36 mm), a higher number of stems per plant (11.87) and higher fresh yield (2.03 tons/acre) were recorded in 12 tons/acre treatment. The least values were obtained from the control treatment. This implies that comparably higher available nitrogen in 12 tons/acre treatment has resulted in higher fresh yield and better results in almost all the agronomic parameters. The better overall results in the 12 tons/acre treatment could be due to which the application of more organic

manure created better conditions for biological processes in the root zone which facilitated balanced nutrient availability for plants in addition to better water holding capacity, better soil physical and chemical properties in comparison to other treatments. An interesting finding was reported by Gerami et al. (2016) where the yield and agronomic traits of oregano were reported to increase with an increase in organic manure up to 20 tons/hectare, and decrease when it is applied beyond 30 tons/hectare.

Further, the ANOVA (data not presented) showed non-significant interaction effects of treatments (IR x MR) on all the agronomic parameters recorded. However, the treatment combination of IR 7 days with MR 12 tons/acre yielded the highest fresh yield (3.79 tons/acre) in comparison to all other treatment combinations. The lowest fresh yield (0.43 tons/acre) was obtained from a treatment combination of IR Control and MR 8 tons/acre. The higher fresh yield in IR 7 days with MR 12 tons/acre treatment combination could be due to the combined effect of treatments enhancing the availability of water and nutrient uptake for better growth compared to other treatment combinations. The poor yield in IR Control and MR 8 tons/acre treatment plot could be due to the poor availability of dissolved nutrients for plant uptake as no irrigation was applied. Our results contrast with the research of Heidarian, Rokhzadi, and Mirahmadi (2018) who reported significant combined effects of irrigation and manure. The finding suggests that the combined application of irrigation at 7 days interval and manure rate at 12 tons/acre could be recommended for cultivation of dill under ARDC–Bajo condition.

Table 2. Effect of irrigation interval, and manure rate on yield and agronomic parameters of dill at ARDC-Bajo

Treatments	Plant height (cm)	Stem diameter (mm)	Stems per plant (No.)	Fresh Yield (ton/acre)
Irrigation Interval (IR)				
Control	30.22 (6.39)	11.26 (3.44)	9.65 (1.84)	0.76 (0.63)
3 Days	31.27 (5.09)	12.71 (3.04)	10.40 (1.83)	2.04 (0.90)
7 Days	39.15 (8.96)	14.73 (4.04)	13.15 (3.91)	2.06 (1.89)
14 Days	31.107 (6.67)	10.98 (3.10)	9.75 (1.75)	0.78 (0.57)
P-value	*0.27*	*0.37*	*0.23*	*0.15*
Manure Rate (MR)				
Control	30.99 (2.57)	11.57 (1.19)	10.15 (0.95)	1.10 (0.22)
4 tons/acre	33.28 (2.28)	11.89 (0.89)	10.63 (0.64)	1.27 (0.30)
8 tons/acre	31.66 (1.66)	11.85 (0.86)	10.30 (0.59)	1.24 (0.30)
12 tons/acre	35.77 (2.22)	14.36 (1.14)	11.87 (1.01)	2.03 (0.54)
P-value	*0.24*	*0.07*	*0.12*	*0.07*

Values in parentheses indicate standard deviation of the mean

3.2 Effect of irrigation interval, manure rate and their interaction on agronomic parameters of dill at ARDC-Wengkhar

The results (Table 3) showed that the irrigation interval (IR) and manure rate (MR) did not significantly affect any of the agronomic parameters. In terms of irrigation intervals, IR 7 days yielded better results in comparison to other irrigation treatments except for the fresh to dry weight ratio (FDR), while the least values were obtained from IR 3 days except for FDR. It was observed that all the parameters followed a similar trend. Their values increased from IR 3 days to IR 7 days and then decreased with the application of IR at 14 days. The decrease in the yield at IR 3 days could be due to the combined effect of rainfall and excessive/frequent irrigation resulting in waterlogged conditions affecting the yield and parameters negatively. A comparatively better result in IR 7 days implies that the combination of rainfall and irrigation at 7-days interval is the best practice of irrigation under ARDC–Wengkhar condition, while lower values obtained under no irrigation condition (control) could mean that the rainfall alone is not enough to enhance nutrient dissolution and uptake for proper growth and development of dill plants.

Although the literature on dill is scarce, past reports on beans and anise (Hassan, Abou El-kasem, & El-kassas, 2021), sugar beet (Heidarian et al., 2018) and oregano (Gerami et al., 2016) have reported the influence of soil type and texture on the soil microbial diversity (Obayomi et al., 2021).

Surprisingly, MR Control yielded better results (although not statistically significant) in all the agronomic parameters except for FDR compared to other treatments. This could be due to abundant residual nutrients from the previous crop cultivated in the trial site. In addition, the agronomic parameters are negatively affected by an increasing amount of manure application. This could be due to the presence of residual nutrients in the soil, and the addition of organic manures could have crossed the threshold for optimal growth and development. Similar findings were reported in past research on oregano and onion (Gerami et al., 2016; Lee, 2012).

The statistical model did not detect any significant interaction effect of the treatments (data not shown) except for the number of stems per plant at $P = 0.02$ (Figure 1). The result showed that the combined application of IR 14 days and MR 4 tons/acre obtained taller plant height (74.70 cm), higher fresh yield/acre (3.47 tons/acre) and higher FDR (13.20), while the combined application of IR 3 days and MR 8 tons/acre obtained lowest values for stem diameter (6.21 mm), stem numbers (5.63) and dry yield (0.04 tons/acre). The poor results in the treatment

combination of IR 3 days and MR 8 tons/acre could be due to frequent irrigation (every 3 days) at field capacity in addition to natural rain resulting in waterlogged conditions creating unfavourable conditions at the rhizosphere for optimal plant growth. The result also suggests that the combined application of IR 14 days and MR 4 tons/acre could be the best practice for dill cultivation under ARDC-Wengkhar region.

Table 3. Effect of irrigation interval and manure rate on agronomic parameters of dill under ARDC-Wengkhar

Treatments	Plant height (cm)	Stem diameter (mm)	Stems per plant (No.)	Fresh yield (ton/acre)	Dry yield (ton/acre)	FDR[#]
Irrigation Interval (IR)						
Control	55.43 (16.31)	10.98 (3.10)	8.49 (1.46)	1.87 (1.33)	0.24 (0.18)	7.77 (0.80)
3 Days	48.18 (21.30)	10.28 (4.62)	7.85 (2.48)	1.13 (1.07)	0.14 (0.15)	9.18 (3.04)
7 Days	65.21 (22.82)	12.23 (3.79)	9.50 (2.23)	2.51 (1.90)	0.31 (0.22)	8.00 (0.98)
14 Days	60.13 (24.90)	11.08 (3.26)	8.68 (1.99)	2.40 (1.79)	0.25 (0.19)	9.74 (4.29)
P-value	*0.57*	*0.65*	*0.51*	*0.35*	*0.64*	*0.26*
Manure Rate (MR)						
Control	60.64 (16.30)	11.76 (3.75)	9.20 (1.76)	2.36 (1.16)	0.30 (0.14)	8.08 (0.87)
4 tons/acre	57.90 (21.78)	11.57 (3.08)	9.03 (2.08)	1.91 (1.57)	0.21 (0.17)	9.60 (4.41)
8 tons/acre	52.95 (27.50)	10.35 (4.80)	8.16 (2.79)	2.08 (2.12)	0.26 (0.26)	8.96 (3.05)
12 tons/acre	57.45 (22.42)	10.89 (3.19)	8.12 (1.56)	1.56 (1.56)	0.19 (0.18)	8.05 (0.89)
P-value	*0.76*	*0.69*	*0.21*	*0.44*	*0.65*	*0.51*

[#] *Fresh weight to Dry weight Ratio; Values in parentheses indicate standard deviation of the mean*
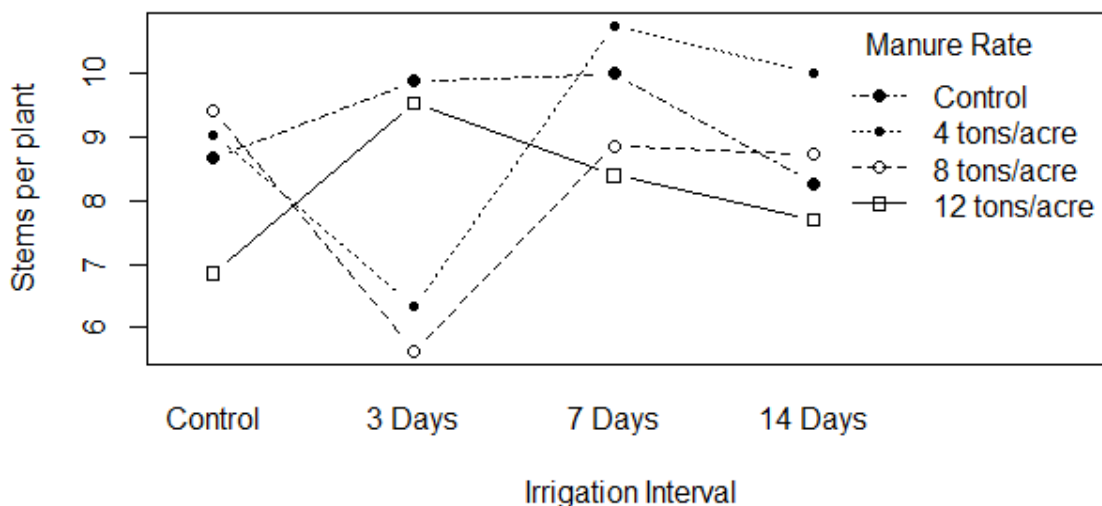

Figure 1. Interaction effect of irrigation interval and manure rate on the number of stems per plant at ARDC-Wengkhar

3.3 Effect of irrigation interval, manure rate and their interaction on agronomic parameters of dill at NCOA-Yusipang

The results (Table 4) showed significant effects of irrigation intervals on all the parameters assessed except for stem diameter under NCOA-Yusipang condition. The plant height, stems per plant, fresh yield and dry yield were significantly better in the IR 3 days treatment compared to control treatment but not significantly different from other treatments. Further, the control treatment yielded comparable outcomes to IR 14 days treatment. Although not statistically significant, IR 3 days obtained a larger stem diameter followed by IR 14 days and IR 7 days. The least stem diameter was obtained in the control treatment. The agronomic parameters followed a similar trend in which they increased to a maximum at IR 3 days and decreased with decreasing irrigation interval. The better performance of dill with the application of irrigation at 3 days interval could be due to meeting the optimal moisture requirement in addition to rainfall during the growing period under NCOA–Yusipang condition. Our results agree with the past reports on potato (Abou El-Khair, E.E., Nawar, & Ismail, 2011), onion (Gwari et al., 2014) and safflower (Khalil, Dagash, & Yagoub, 2013; Mohamed, Mariod, Yagoub, & Dagash, 2013).

The results (Table 4) also showed that plants treated with 8 tons/acre performed comparably better than other treatments (although not statistically significant) in all the parameters except for the number of stems per plant and FDR, while 4 tons/acre treatment yielded minimum values in almost all the parameters except fresh and dry yield. The highest number of stems per plant was obtained from the control treatment. All the parameters followed a similar trend in the order 8 tons/acre > control > 12 tons/acre > 4 tons/acre except for the stems per plant and fresh and dry yield. The non-significant results between all the manure treatments could be because the residual nutrient in the soil is already optimum for proper growth and development and the incorporation of additional manure did not make any difference. The effect of residual nutrients from organic manure applied in the previous year even at a very low rate is documented by Riley (2015). The higher stems per plant in the control treatment could be explained by shorter plants with slimmer diameters resulting in higher lateral shoots in the plants.

The ANOVA showed significant interaction effects of treatments (Figure 2) only for plant height *(P=0.02)* and stem diameter *(P=0.03)* and not for other parameters (data not shown). A significantly taller plant was obtained from the treatment combination of IR 3 days and MR 12

tons/acre (59.70 cm) compared to other treatment combinations. Plants of wider stem diameter were recorded in the treatment combination of IR 3 days and MR 8 tons/acre (10.80 mm) which were significantly different from other treatments while the least was obtained from the treatment combination of IR control and MR 4 tons/acre (4.85 mm). Although not statistically significant, the highest fresh yield of 0.856 tons/acre was obtained from IR 7days and MR 12 tons/acre treatment combinations, while the lowest was from IR control and MR 4 tons/acre treatment combinations (0.13 tons/acre).

Similarly, the maximum dry yield was obtained from the treatment combination of IR 3 days and MR 8 tons/acre (0.18 tons/acre), while the lowest dry yield was obtained from IR control and MR 4 tons/acre (0.02 tons/acre) treatment combination. The results imply that the best practices in terms of irrigation and nutrient management for dill under NCOA-Yusipang conditions could be the application of irrigation at field capacity at 3 days interval in combination with 8 tons/acre organic manure.

Table 4. Effect of irrigation interval, and manure rate on agronomic parameters of dill under NCOA-Yusipang

Treatments	Plant height (cm)	Stem diameter (mm)	Stems per plant (No.)	Fresh yield (ton/acre)	Dry yield (ton/acre)	FDR [#]
Irrigation Interval (IR)						
Control	40.70 (7.33) [b]	6.50 (2.23)	6.42 (1.03) [b]	0.26 (0.19) [b]	0.05 (0.03) [b]	5.59 (0.45) [a]
3 Days	57.20 (4.49) [a]	10.20 (1.62)	7.80 (0.69) [a]	0.73 (0.30) [a]	0.14 (0.06) [a]	5.09 (0.45) [ab]
7 Days	55.90 (6.38) [a]	8.96 (1.79)	7.73 (0.92) [a]	0.63 (0.25) [a]	0.12 (0.05) [a]	4.86 (0.42) [b]
14 Days	47.20 (7.40) [ab]	9.13 (1.75)	7.25 (0.77) [ab]	0.47 (0.28) [ab]	0.09 (0.05) [ab]	5.30 (0.70) [ab]
P-value	*0.02*	*0.05*	*0.03*	*0.02*	*0.01*	*0.04*
Manure Rate (MR)						
Control	50.31 (8.12)	8.77 (1.19)	7.37 (0.79)	0.50 (0.18)	0.10 (0.03)	5.15 (0.41)
4 tons/acre	48.84 (10.08)	8.37 (2.61)	7.37 (1.11)	0.52 (0.39)	0.10 (0.07)	5.33 (0.49)
8 tons/acre	52.72 (7.75)	9.08 (2.49)	7.26 (0.98)	0.56 (0.33)	0.12 (0.07)	4.96 (0.73)
12 tons/acre	49.07 (11.42)	8.54 (2.66)	7.22 (1.19)	0.51 (0.33)	0.10 (0.07)	5.39 (0.58)
P-value	*0.18*	*0.62*	*0.94*	*0.96*	*0.81*	*0.11*

[#] *Fresh weight to Dry weight Ratio; Different lower-case letters in the superscript indicate statistically significant differences following the Tukey's HSD post hoc analysis at P=0.05; values in parentheses indicate standard deviation of the mean*
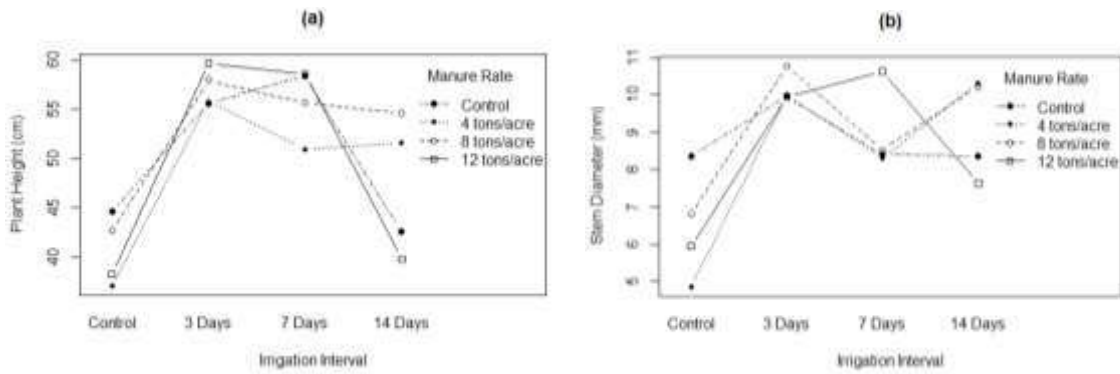
Figure 2. Interaction effect of irrigation interval and manure rate on plant height (a) and stem diameter (b) of dill at NCOA–Yusipang

3.4 Comparison of agronomic parameters of dill between three study sites

The crop gestation period was recorded highest at ARDC-Wengkhar (80 DAS) followed by ARDC-Bajo (76 DAS) and NCOA-Yusipang (75 DAS). The results of the one-way ANOVA and Tukey multiple comparison test (Table 5) showed that all the parameters under study significantly differed amongst study sites except for the survival rate. The result showed that the mean plant height was significantly taller at ARDC–Wengkhar (56.00 cm) and NCOA–Yusipang (50.24 cm) compared to that recorded at ARDC–Bajo (32.93 cm).

However, the mean plant height recorded at ARDC–Wengkhar and NCOA–Yusipang was comparable to each other. The mean stem diameter recorded at ARDC–Bajo (12.42 mm) was significantly wider compared to that at NCOA–Yusipang (8.69 mm), while it did not differ from that at ARDC-Wengkhar (10.87 mm). Similarly, a significantly higher number of stems per plant was obtained at ARDC–Bajo (10.73) in comparison to that at ARDC–Wengkhar (8.43) and NCOA–Yusipang (7.30). The stems per plant obtained at ARDC–Wengkhar was significantly higher compared to that recorded at NCOA–Yusipang. The triangulation of results suggests that the yield is directly linked to the survival percent, plant height, stem diameters and stems per plant.

The significantly higher yield at ARDC–Wengkhar (1.92 ton//acre) and ARDC–Bajo (1.41 ton/acre) is due to better results in all the parameters measured compared to NCOA–Yusipang (0.52 ton/acre). It could also be due to the geographical setting of the study sites, where the colder temperature (both day and night) during the growing season at NCOA-Yusipang limited the growth and development of dill compared to other sites which are located at comparatively lower altitudes with warmer temperature during the growing season.

Table 5. Comparison of yield and agronomic parameters of dill between study sites.

Location	Plant height (cm)	Stem diameter (mm)	Stems per plant (no)	Survival percent (%)	Fresh yield (ton/acre)
ARDC-Bajo	32.93 (7.63) [b]	12.42 (3.64) [a]	10.73 (2.82) [a]	57.15 (33.18)	1.41 (1.27) [a]
ARDC- Wengkhar	56.00 (23.31) [a]	10.87 (4.01) [a]	8.43 (2.43) [b]	72.71 (35.41)	1.92 (1.63) [a]
NCOA-Yusipang	50.24 (9.28) [a]	8.69 (2.26) [b]	7.30 (1.00) [c]	65.63 (20.62)	0.52 (0.31) [b]
P-value	<0.001	<0.001	<0.001	0.05	<0.001

Different lower-case letters in the superscript indicate statistically significant differences following Tukey's HSD post hoc analysis at P=0.05; values in parentheses indicate standard deviation of the mean

### 3.5 Correlation between variables under study

The correlation between different variables under study was computed using the combined dataset from all study sites. The result of Pearson correlation tests is given in Figure 3. The result suggests that all the parameters under study are positively correlated with each other with the strongest correlation between stem diameter and stems per plant ($r = 0.87$) and the weakest correlation between plant height and stems per plant ($r = 0.034$). The result showed that the fresh yield is strongly correlated with fresh weight of the plant ($r = 0.81$), followed by stem diameter ($r = 0.70$), stems per plant and survival percentage ($r = 0.63$) and plant height ($r = 0.46$). This implies that all the parameters need to be given due importance during further varietal breeding, selection, or introduction programs.
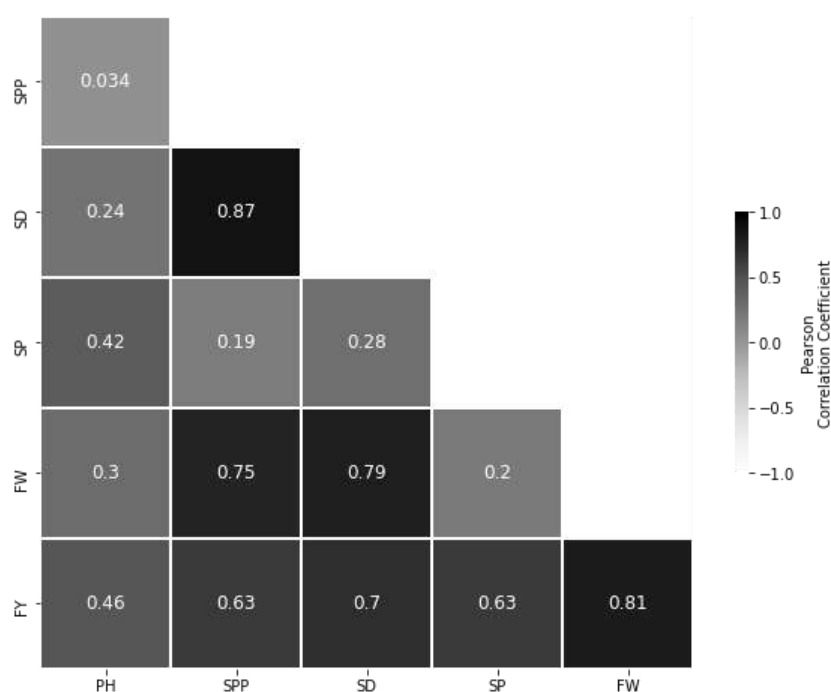
Figure 3. Correlation matrix of dill parameters of dill. FY = fresh yield, FW = fresh weight, SP = survival Percent, SD = stem diameter, SPP = stems per plant

4. Conclusion

Our findings suggest that the dill can be successfully grown under three different agro-ecological zones of Bhutan under summer conditions, while we recommend similar research under winter conditions to document response under different irrigation and nutrition regimes. Overall, the highest yield was obtained at ARDC-Wengkhar while NCOA-Yusipang obtained significantly low fresh yield compared to ARDC-Bajo and ARDC-Wengkhar.

Significant interaction effects of irrigation intervals and manure rate were observed under NCOA–Yusipang condition and not under ARDC–Bajo and Wengkhar condition. The irrigation at 3 days interval produced significantly better yield under NCOA Yusipang conditions in comparison to other irrigation treatments, while the irrigation at 7 days interval under ARDC Bajo and ARDC Wengkhar obtained better results. The decrease in the irrigation interval beyond 7 days had a negative influence on the fresh and dry yield at all study sites and irrigation at 3 days interval under ARDC – Wengkhar condition decreased the yield of dill.

The manure application did not influence any of the parameters assessed at all study sites. It was observed that manure application at 8 tons/acre yielded higher under NCOA–Yusipang condition, while the control (no application of manure) yielded better results compared to other manure treatments under ARDC–Wengkhar condition. Application of manure at 12 tons/acre under ARDC-Bajo yielded better results.

Based on the findings of our research, we recommend the combined application of IR 7 days x MR 12 tons/acre, IR 7 days x MR 4 tons/acre, and IR 3 days x 8 tons/acre under ARDC–Bajo, ARDC-Wengkhar and NCOA–Yusipang conditions, respectively. As our findings could have been affected by many other factors such as rainfall and temperature, more precise results could be obtained if such research can be conducted under controlled environment conditions such as greenhouse or climate-controlled chambers. Our findings and recommendations can be used as a reference document by agriculture extension officers for promotional and advocacy programs under three agro-ecological zones.

References

Abou El-Khair, E.E., Nawar, D. A. S., & Ismail, H. M. E. (2011). Effect of irrigation water quantity and farmyard manure on potato plant grown in sandy soil. *Egyptian Journal of Agricultural Research, 89*(1), 317-334. doi:http://10.21608/ejar.2011.174286.

Baher, Z. F., Mirza, M., Ghorbanli, M., & Bagher Rezaii, M. (2002). The influence of water stress on plant height, herbal and essential oil yield and composition in *Satureja*

hortensis L. *Flavour and Fragrance Journal, 17*(4), 275-277. doi:http://10.1002/ffj.1097.

Bisong, E. (2019). Matplotlib and Seaborn. In *Building machine learning and deep learning models on google cloud platform* (pp. 151-165). Apress, Berkeley, CA.

Carrubba, A., Catalano, C., & Bontempo, R. (2010). Cultivation of dill (*Anethum graveolens* L.) with different row arrangements. In *Proceedings of the XXVIII International Horticultural Congress on Science and Horticulture for People (IHC2010): A New Look at Medicinal and Aromatic Plants, 925* (pp. 205-211).

Charles, D. J. (2012). Dill. In *Antioxidant Properties of Spices, Herbs and Other Sources* (pp. 281-286). New York, NY. Springer.

Charles, D. J., Simon, J. E., & Widrlechner, M. P. (1995). Characterization of Essential Oil of Dill (*Anethum graveolens* L.). *Journal of Essential Oil Research, 7*(1), 11-20. doi:http://10.1080/10412905.1995.9698456.

de Mendiburu, F. (2019). Agricolae: A statistical analysis tool for agricultural research. R Package version 1.3-1. Retrieved from https://CRAN.R-project.org/package=agricolae

Delfine, S., Tognetti, R., Desiderio, E., & Alvino, A. (2005). Effect of foliar application of N and humic acids on growth and yield of durum wheat. *Agronomy for Sustainable Development, 25*(2), 183-191. doi:http://10.1051/agro:2005017.

Elsayed, S. I. M., Glala, A. A., Abdalla, A. M., El-Sayed, A. E. G. A., & Darwish, M. A. (2020). Effect of biofertilizer and organic fertilization on growth, nutrient contents and fresh yield of dill (*Anethum graveolens*L.). *Bulletin of the National Research Centre, 44*(1), 1-10. doi:http://10.1186/s42269-020-00375-z.

Gerami, F., Moghaddam, P. R., Ghorbani, R., & Hassani, A. (2016). Effects of irrigation intervals and organic manure on morphological traits, essential oil content and yield of oregano (*Origanum vulgare* L.). *Annals of the Brazilian Academy of Science, 88*(4), 2375-2385. doi:http://10.1590/0001-3765201620160208.

Ghassemi-Golezani, K., Rezaeipour, L., & Alizadeh-Salteh, S. (2016). Effects of water stress on seed yield and essential oil content of dill genotypes. *Journal of Biodiversity and Environmental Sciences, 9*(1), 420-425.

Gwari, E. Y., Gambo, B. A., & Kabura, B. H. (2014). Effect of organic manures and irrigation intervals on the growth and yield of onion (*Allium cepa* L.) in Central and Southern Borno State, Nigeria. *International Journal of Advanced Agricultural Research, 2*(7), 106-111. Retrieved from https://www.innspub.net/

Hassan, E. A. (2015). Influence of mixed minerals ores and seaweed liquid extract on growth, yield and chemical constituents of dill (*Anethum graveolens* L.) plants. *Middle East Journal of Applied Sciences, 5*(3), 751-758.

Hassan, H. M. S., Abou El-kasem, S. A. A., & El-kassas, M. S. A. (2021). Influence of intercropping system, water intervals and their interaction on growth, yield, and some

competitive indices of broad bean and anise plants. *Plant Archives, 21*(1). doi:http://10.51470/PLANTARCHIVES.2021.v21.no1.165.

Heidarian, F., Rokhzadi, A., & Mirahmadi, F. (2018). Response of sugar beet to irrigation interval, harvesting time and integrated use of farmyard manure and nitrogen fertilizer. *Environmental and Experimental Biology, 16*(3), 169-175. doi:http://10.22364/eeb.16.16.

Jones, H. G., & Tardieu, F. (1998). Modelling water relations of horticultural crops: a review. *Scientia Horticulturae, 74*(2), 21-46.

Kaur, G. J., & Arora, D. S. (2010). Bioactive potential of *Anethum graveolens,Foeniculum vulgare* and *Trachyspermum ammi* belonging to the family Umbelliferae-Current status. *Journal of Medicinal Plants Research, 4*(2), 087-094. doi:http://10.5897/JMPR09.018.

Khalil, N. A. A., Dagash, Y. M., & Yagoub, S. O. (2013). Effect of sowing date, irrigation intervals and fertilizers on safflower (*Carthamus tinctorius* L.) yield. *Discourse Journal of Agriculture and Food Sciences, 1*(5), 97-102.

Lee, J. (2012). Evaluation of composted cattle manure rate on bulb onion grown with reduced rates of chemical fertilizer. *HortTechnology, 22*(6), 798-803. doi:https://doi.org/10.21273/HORTTECH.22.6.798.

McKinney, W. (2011). pandas: a foundational Python library for data analysis and statistics. *Python for high performance and scientific computing, 14*(9), 1-9.

Mohamed, N., Mariod, A., Yagoub, S., & Dagash, Y. (2013). Effect of irrigation intervals and fertilizers on chemical composition, minerals and fatty acids of safflower (*Carthamus tinctorius* L.) seed. *Acta Agronomica Hungarica, 61*(3), 227-236. doi:https://doi.org/10.1556/AAgr.61.2013.3.6.

Naomi, R. B., Mwanarusi, S., & Musyoka, I. F. (2014). Effects of nitrogen, phosphorus and irrigation regimes on growth and leaf productivity of sage (*Salvia officinalis* L.) in Kenya. *Annals of Biological Research, 5*(2), 84-91.

Obayomi, O., Seyoum, M. M., Ghazaryan, L., Tebbe, C. C., Murase, J., Bernstein, N., & Gillor, O. (2021). Soil texture and properties rather than irrigation water type shape the diversity and composition of soil microbial communities. *Applied Soil Ecology, 161*, 103834. doi:http://10.1016/j.apsoil.2020.103834.

Petropoulos, S. A., Daferera, D., Polissiou, M. G., & Passam, H. C. (2008). The effect of water deficit stress on the growth, yield and composition of essential oils of parsley. *Scientia Horticulturae, 115*(4), 393-397. doi:http://10.1016/j.scienta.2007.10.008.

R Core Team. (2021). R: A Language and Environment for Statistical Computing. Vienna, Austria: R Foundation for Statistical Computing. Retrieved from https://www.R-project.org/

Riley, H. (2015). Residual value of inorganic fertilizer and farmyard manure for crop yields and soil fertility after long-term use on a loam soil in Norway. *Nutrient Cycling in Agroecosystems, 104*(1), 25-37. doi:http://10.1007/s10705-015-9756-8.

Rostaei, M., Fallah, S., Lorigooini, Z., & Abbasi Surki, A. (2018). The effect of organic manure and chemical fertilizer on essential oil, chemical compositions and antioxidant activity of dill (*Anethum graveolens*) in sole and intercropped with soybean (*Glycine max*). *Journal of Cleaner Production, 199*, 18-26. doi:http://10.1016/j.jclepro.2018.07.141.

Shao, H. B., Chu, L. Y., Jaleel, C. A., & Zhao, C. X. (2008). Water-deficit stress-induced anatomical changes in higher plants. *C R Biol, 331*(3), 215-225. doi:http://10.1016/j.crvi.2008.01.002.

Sharma, N., & Singhvi, R. (2017). Effects of chemical fertilizers and pesticides on human health and environment: a review. *International Journal of Agriculture, Environment and Biotechnology, 10*(6). doi:http://10.5958/2230-732x.2017.00083.3.

Simon, J. E., Reiss-Bubenheim, D., Joly, R. J., & Charles, D. J. (1992). Water stress-induced alterations in essential oil content and composition of sweet basil. *Journal of Essential Oil Research, 4*(1), 71-75. doi:http://10.1080/10412905.1992.9698013.

Tsamaidia, D., Dafererab, D., Karapanosa, I. C., & Passama, H. C. (2017). The effect of water deficiency and salinity on the growth and quality of fresh dill (*Anethum graveolens* L.) during autumn and spring cultivation. *International Journal of Plant Production, 11*(1), 33-46.

Valero-Mora, P. M. (2010). ggplot2: Elegant Graphics for Data Analysis. *Journal of Statistical Software, Book Reviews, 35*(1), 1-3. doi:https://doi.org/10.18637/jss.v035.b01.

Zehtab-Salmasi, S., Javanshir, A., Omidbaigi, R., Alyari, H., & Ghassemi-Golezani, K. (2001). Effects of water supply and sowing date on performance and essential oil production of anise (*Pimpinella anisum* L.). *Acta Agronomica Hungarica, 49*(1), 75-81. doi:https://doi.org/10.1556/AAgr.49.2001.1.9.

Assessment of Seedling Rate per Hill for Irrigated Rice in a Wet Sub-Tropical Condition of Bhutan

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ABSTRACT

A field experiment was conducted at the research farm of the Agriculture Research and Development Centre (ARDC) Samtenling in Sarpang district of Bhutan in 2020 to assess the effects of transplanting different numbers of seedlings per hill on grain yield and yield components of rice under irrigated conditions. A widely adopted rice variety, Bhur Kambja-1, was used for the study with one to seven seedlings per hill transplanted at 20×20 cm spacing. The experiment was laid out in a randomized complete block design with three replications. The results of the experiment revealed that the number of seedlings per hill had a significant effect on number of tillers per hill and grain yield. In terms of rice grain yield, a significant difference was observed only between three and seven seedlings per hill with a recorded yield of 3.9 and 2.7 t ha^{-1}, respectively. However, no significant effect was established on other yield components, such as the plant height, panicle length, number of grains per panicle, and unfilled grains per panicle. Therefore, this study recommends transplanting three seedlings hill^{-1} for cultivation of Bhur Kambja-1 rice variety in the wet-subtropical condition of Bhutan as three seedlings per hill yielded significantly higher productivity and could potentially reduce the seed rate.

Keywords: *Bhur Kambja-1; Grain yield; Seedlings hill^{-1}; Rice; Yield components*

1. Introduction

Rice (*Oryza sativa*) is the world's most important food for some 4 billion people (IRRI, 2019) and more than 90% of rice is produced and consumed in Asia (Global Rice Science Partnership (GRiSP, 2013). In Bhutan rice is the staple food crop, indispensable to Bhutanese culture, tradition, religion, and livelihoods (Ghimiray et al., 2008) which is often equated to food security (Chhogyel, Ghimiray, Wangdue, & Bajgai, 2015). It is grown in a wide range of altitudes, starting from tropical lowlands (150 m) in the south to elevations as high as 2600 m in the north (Ghimiray et al., 2008).

However, the rice sector in Bhutan is challenged by shrinking rice areas, often losing to urbanization and other infrastructural development activities. Fallowing of the wetland is also one of the prominent reasons for reduced rice production area. Rice area in Bhutan in 2010 was

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22,550 hectares while it dwindled to 14,668 hectares in 2019 (RSD, 2019), accounting for about 35% reduction over the 2010-2019 period. Despite these impediments, rice in southern Bhutan is grown annually for food in small pockets mostly applying farmers' traditional cultivation techniques. One of the prevalent techniques is the use of a thick bunch of seedlings hill[-1] at planting primarily to save the seedlings from pest damages and also for higher yield under irrigated conditions. It is seen during the field visits that this technique is used in both traditional and improved varieties by the farmers.

According to IRRI (2007), the number of seedlings transplanted hill[-1] depends on three factors which are the traditional practices of the farmers, quality of seedling, and the price of seed. Hybrid seed, which is costlier, is often transplanted as one seedling hill[-1] while traditional varieties are sometimes planted with up to six seedlings hill[-1]. In most countries, the farmers plant two to three seedlings per hill (IRRI, 2007). The use of more seedlings hill[-1] not only adds to cost but is also a waste of natural resources (Saker & Nahar, 2016). Hence, finding a genotype-based appropriate number of seedlings hill[-1] or planting density is necessary for optimum rice production under limited production resources. The number of seedlings hill[-1] is an important factor for higher production as it influences the number of tillers, which ultimately determines the yield, because the tillering ability is an inherent trait that varies among rice varieties (Chowhan, Imdadu, Rani, & M., 2019). Hence, extra seedlings are generally desired based on the tillering capacity of the variety and high-tillering rice varieties are desirable for transplanted or direct-seeded rice (De Datta, 1981). However, the use of extra seedlings is an expensive method of ensuring an increased number of tillers (Liu et al., 2021). Therefore, this study was carried out with the objectives to: (a) determine the effect of the number of seedlings hill[-1] on yield components and grain yield of Bhur Kambja-1, and (b) to recommend the optimum number of seedlings hill[-1] to the rice farmers of southern Bhutan.

2. Materials and Method

2.1 Experimental site and design

The study was carried out from June to December of 2020 at the research farm of the ARDC-Samtenling, Sarpang Dzongkhag (26° 54' 17" N latitude and 90° 25' 51" E longitude) which is located in southern Bhutan. The study site falls under wet-humid sub-tropical agroecology with an elevation of 375 meters above sea level (Figure 1). Treatments included seven different numbers of seedlings viz. 1, 2, 3, 4, 5, 6, or 7 hill[-1] of popular improved variety of rice in the region (Bhur Kambja-1). The experiment was carried out in a randomized complete block design with three replications. Each plot size measured 10 m^2 (5 m × 2 m) and uniform spacing

of 20 cm × 20 cm row to plant was maintained for the study. The experimental land was prepared using a tractor and fertilized with urea, single super phosphate (SSP), and muriate of potash (MoP) at the recommended rate of 150, 250, and 50 kg ha^{-1}, respectively. Half dose of nitrogen was applied as basal dose along with full doses of phosphorus and potassium. The remaining half dose of nitrogen was applied in two equal splits at active tillering and panicle initiation stages. Intercultural operations like weeding and irrigation were carried out manually for maintaining the normal growth and development of the crop. And these intercultural operations were uniformly carried out across the seven treatments.
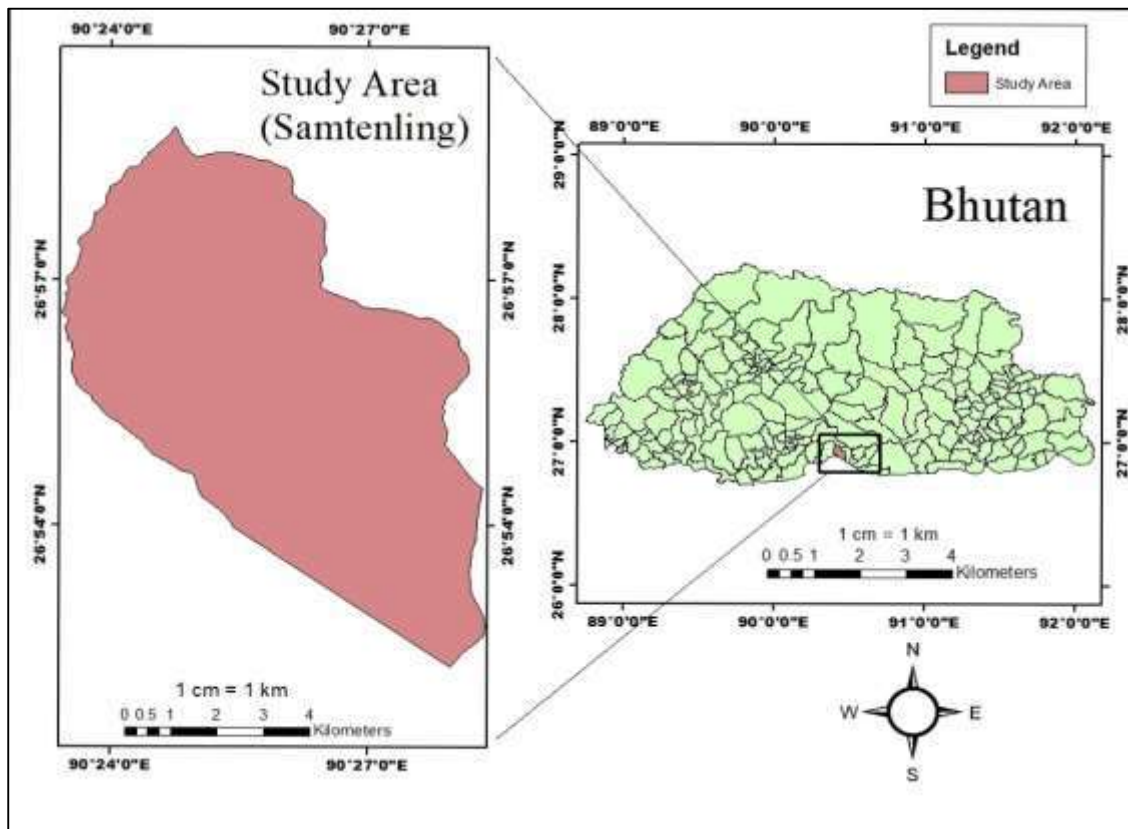
Figure 1. Location of the experimental site in Bhutan

2.2 Data compilation and analysis

Data from the experiment were gathered following the Standard Evaluation System for Rice developed by the International Rice Research Institute (IRRI, 2002) and every care had been taken to minimize errors. The experimental plots were monitored at regular intervals and data for plant height were gathered immediately after flowering, while agronomic parameters such as number of productive tillers, number of filled grains panicle^{-1}, and number of unfilled grains panicle^{-1}, panicle length, and yields were collected during the harvesting stage as per the methods described by (IRRI, 2002).

The crop was harvested at full maturity and five hills per treatment were randomly selected to gather the required data.   All data was collection was performed after discarding two rows of plants from each side of the experimental unit to avoid biases through border effect. The counting of the number of productive tillers, number of filled grains panicle[-1] and number of unfilled grains panicle[-1] were done manually and panicle length was measured using a scale. The grain yield was determined from the harvested area of 5.04 m^2 adjusting moisture content to 14 % using the standard formula.

$$Grain\ yield\ (t/ha)\ \ = \frac{Adjusted\ moisture\ x\ Plot\ yield\ (kg)\ \ x\ 10000}{Plot\ size\ x\ 1000}$$

$$Where\ Adjusted\ moisture\ \ \ = \frac{100 - MC}{100 - 86}, and\ MC = grain\ moisture\ content\ at\ harvest$$

The compilation of data was carried out using Microsoft Excel spreadsheet while analysis of variance (ANOVA) was conducted using statistical software Statistical Tool for Agriculture Research (STAR version 2.0.1) and Statistical Package for the Social Sciences (SPSS version 22). Multiple comparisons among the means were conducted using Tukey's HSD Test. All effects were declared significant at P-value $\leq$ 0.05 unless otherwise stated.

3. Results and Discussion

3.1 Plant height

Rice seedlings transplanted with one to seven seedlings hill[-1] did not show any significant differences in plant height *(P*-value=0.489). The plant heights varied from 98.8 to 103.5 cm, without showing any significant relation to the different seedling rates (Table 1). Though there were no significant differences, transplanting two to three seedlings hill[-1] resulted in the numerically tallest plants in this experiment.

Our finding agrees with those of Mahamud, Haque, and Hasanuzzaman (2013) and Faruk, Rahman, and Hasan (2009) who also reported having taller plants when three rice seedlings were transplanted per hill. Plant height is an important trait in rice and has greater bearings on the grain yield and harvest index (IRRI, 2006). The tallest plant height recorded in this study for three seedlings hill[-1] is similar to the findings of Faruk et al. (2009), who reported the tallest plant height from two seedlings hill[-1] density in BRRI Dhan-33 variety of rice. The Bhutanese farmers use rice straw as cattle feed, and therefore it is important to assess the optimum seedling rate that will give good grain yield without compromising the straw potential of crop varieties.

In an earlier study, the rice variety, Bajo Maap, gave good grain as well as straw yields when transplanted with three to four seedlings hill[-1] in Wangdi valley of Bhutan (Ghaley, Høgh-Jensen, & Christiansen, 2010).

3.2 Panicle length

Panicle length is an important characteristic in rice because together with spikelet number and density, and seed setting rate determines the grain number per panicle which elucidates the productivity or yield in a given situation. The longest panicle was recorded in one seedling hill[-1] corresponding to 22.5 cm while the shortest was found in the treatment which had three seedlings hill[-1] (20.8 cm) through the values did not differ significantly (Table 1). A similar finding was also reported by Mahamud et al. (2013) and (Chowhan et al., 2019) who recorded the longest panicle from one seedling hill[-1]. In contrast, Ninad, Bahadur, Hasan, Alam, and Rana (2017) reported the longest panicle length from four seedlings hill[-1] while they found the shortest panicle length under one seedling hill[-1] and this could be possibly related to varietal traits. Yao et al. (2015) reported that the panicle length is a quantitative trait controlled by multiple genes which are greatly affected by the environment. However, non-significant differences among treatments could be related to Bhur Kambja-1 and the effect on other varieties may differ.

3.3 No. of tillers hill[-1]

Tillering ability is one of the yield-determining parameters in a rice crop and it makes agronomic denotation to evaluate the ability in Bhur Kambja-1. The number of tillers hill[-1] was significantly affected by seedling rates (*P*-value=0.017). Transplanting of rice at three seedlings hill[-1] showed the highest number of productive tillers (11). Treatments with one, two and four seedlings hill[-1] resulted in the lowest number of productive tillers of eight each, whereas the use of five, six and seven seedlings hill[-1] produced nine tillers hill[-1] (Table 1).

The significant variation in terms of the tillering ability of Bhur Kambja-1 under different numbers of seedlings at transplantation corroborates the findings of Dejen (2018), Mahamud et al. (2013), Promsomboon et al. (2019), Imran, Inamullah, Naeem, and Khan (2015), and Islam and Salam (2017) who reported significant differences in the number of tillers under different number of seedlings hill[-1]. Tillering capacity may be attributed to the variety and is also affected by cultivation methods such as fertilizer application (Nuruzzaman, Yamamoto, Nitta, Yoshida, & Miyazaki, 2000). However, Faruk et al. (2009) reported the highest number of tillers from four seedlings hill[-1] while Saker and Nahar (2016) reported insignificant

differences in tillering due to differences in the number of seedling hill[-1], and so the literature needs to be consulted with reference to varieties and crop management practices, not just the number of tillers per hill. In general, high-tillering varieties have a higher number of panicles and thus higher yield in comparison to that of the low-tillering varieties.

3.4 Number of filled grains panicle-1

The result revealed no significant differences in the case of the number of filled grains panicle[-1]. All the treatments responded with a similar number of filled grains with the highest being 136 filled grains recorded in one seedling hill[-1] at transplantation (Table 1). The lowest number of grains was observed for six seedlings hill[-1] with only 121 filled grains. This study saw a non-uniform pattern in the number of grains across the treatments with different numbers of seedlings.

Chowhan et al. (2019) reported the maximum number of grains from one seedling hill[-1] and their finding is in agreement with our observations. However, the findings of this study are not consistent with the results of Dejen (2018) and Hasanuzzaman, Rahman, Roy, Ahmed, and Zobaer (2009) who found a decreasing trend in the number of grains per panicle with increasing number of seedlings hill[-1]. This result also disagrees with the finding of Ninad et al. (2017) who reported an increased number of grains panicle[-1] with an increase in the number of seedlings hill[-1] in rice variety BRRI Dhan-48. Further, the number of grains panicle[-1] accounts for higher grain yield (Zhang, Li, Ashraf, Liu, & Li, 2019) while it is determined by panicle architecture like panicle length and length of panicle branch-lets (Kovi, Bai, Mao, & Xing, 2011). Further, the seed setting rate often determines the number of grains panicle[-1] which is largely affected by external environment (Zhang et al., 2019). Therefore, the effect of seedlings number in a hill in terms of the number of grains panicle[-1] could be location- and genotype-specific under the influence of different environmental conditions. Further, the study showed unfilled grains panicle[-1] ranging from 16 to 23 numbers among the treatments. Higher numbers of unfilled grains were observed in six and seven seedlings hill[-1] with 23 and 21 unfilled grains, respectively. Thus, as stated by Kobata, Yoshida, Masiko, and Honda (2013) and Fu et al. (2021), halving plant density during flowering increased spikelet fertility by 1.3 to 1.5 times, and therefore use of the right number of seedlings hill[-1] is more economic.

3.5 Grain Yield

In terms of rice grain yield, the significant difference (P-value=0.013) was recorded only between three and seven seedlings hill[-1] with 3.9 and 2.7 t ha^{-1}, respectively (Table 1). In other

words, transplanting with one to seven seedlings hill[-1] produces no significant yield differences except for the three seedlings hill[-1] treatment.

The lower grain yield in seven seedlings hill[-1] may be attributed to lower number of tillers (9) and higher number of unfilled grains (21) (Table 1) in comparison to the three seedlings hill[-1] treatment. This is also true for other treatments concerning the number of unfilled grains (17 to 23), but the numbers of tillers hill[-1] showed an inconsistent pattern. The maximum grain yield of rice using three seedlings hill[-1] was also obtained by Bhowmik, Sarkar, and Zaman (2012) and Dejen (2018). The highest grain yield might be due to optimum density suitable for Bhur Kambja-1 to tiller adequately. A higher number of tillers, especially fertile tillers, was found as the determinant of higher yield in rice (Dejen, 2018; Yumnam et al., 2021). Pruneddu and Spanu (2001) revealed that higher grain yield in hybrid rice was due to a higher number of effective tillers and a higher number of filled grains. However, it must be restated that our findings only represent Bhur Kambja-1 in wet sub-tropics and cannot be generalized to other varieties and locations.

Table 1. Effects of different numbers of seedlings hill[-1] at transplanting on yield and yield component of Bhur Kamjha-1 rice variety

Treatment	Plant height (cm)	Panicle length (cm)	No. of Tillers hill[-1]	No. of grains panicle[-1]	No. of unfilled grains panicle-1	Yield tha[-1]
One seedling hill[-1]	101.9	22.5	8b	136	18	3.3ab
Two seedling hill[-1]	103.5	21.6	8b	122	17	3.1ab
Three seedling hill[-1]	101.4	20.8	11a	130	16	3.9a
Four seedling hill[-1]	102.7	21.5	8b	132	18	3.4ab
Five seedling hill[-1]	102	21.6	9ab	123	18	3.3ab
Six seedling hill[-1]	98.8	20.9	9ab	121	23	3.2ab
Seven seedling hill[-1]	102.2	20.9	9ab	128	21	2.7b
P-value	0.489	0.06	0.017	0.388	0.09	0.013
CV (%)	2.69	2.85	8.77	6.85	16.4	9

Means with the same letter(s) in the same column are not significant at $P \leq 0.05$

4. Conclusion

Information on seedling rates for all released varieties under the given set of environments is highly useful in exploiting their yield potential. To this end, this is the first study to document the optimum seedling rate hill[-1] for a popular rice variety, Bhur Kambja-1 in a field experiment conducted at ARDC Samtenling in Sarpang. The study found that the use of three or four seedlings hill[-1] produced the highest number of productive tillers hill[-1], the lowest number of unfilled grains panicle[-1,] and the highest when planted during the main season under the wet-subtropical condition of Southern Bhutan. This research also shows that the number of

seedlings hill^{-1} did not have significant effects on other yield attributes, such as plant height and the number of filled grains panicle^{-1}. The findings of this study are in agreement with the research conducted earlier, both in Bhutan as well as in other parts of Asia. Thus, this study recommends the use of three seedlings hill^{-1} for Bhur Khamja-1 rice variety as this seedling rate not only gives the highest grain yield but also could reduce the seed quantity as compared with the more than three seedling hill^{-1}.

References

Bhowmik, S. K., Sarkar, M. A. R., & Zaman, F. (2012). Effect of spacing and number of seedlings per hill on the performance of aus rice cv. NERICA 1 under dry direct seeded rice (DDSR) system of cultivation. *Journal of the Bangladesh Agricultural University, 10*(452-2016-35674), 191-195. doi:10.22004/ag.econ.209319

Chhogyel, N., Ghimiray, M., Wangdue, K., & Bajgai, Y. (2015). Enhancing Bhutanese Rice in the Domestic Market through increased Production and Favorable Pricing Regime. *Journal of Bhutan Studies*(32), 26-44.

Chowhan, S., Imdadu, H. M., Rani, G. S., Islam, , & M., B. A. M. (2019). Effect of variety and seedling number on the growth and yield of Boro Rice in Bangladesh. Journal of Experimental Agriculture International. *41*(6), 1-15. doi:https://doi.org/10.9734/jeai/2019/v41i630436

De Datta, S. K. (1981). *Principles and practices of rice production*. Los Baños (Philippines): International Rice Research Institute (IRRI).

Dejen, T. (2018). Effect of Plant Spacing and Number of Seedlings per Hill toTransplanted Rice (Oryza Sativa X Oryza Glaberrima) under Irrigation in Middle Awash, Ethiopia. *Journal of Applied Life Sciences International*, 1-9. doi: 10.9734/JALSI/2018/42081

Faruk, M. O., Rahman, M. A., & Hasan, M. A. (2009). Effect of seedling age and number of seedling per hill on the yield and yield contributing characters of BRRI Dhan 33. *International journal of sustainable crop production, 4*(1), 58-61.

Fu, Y.-q., Zhong, X.-h., Zeng, J.-h., Liang, K.-m., Pan, J.-f., Xin, Y.-f., . . . Chen, R.-b. (2021). Improving grain yield, nitrogen use efficiency and radiation use efficiency by dense planting, with delayed and reduced nitrogen application, in double cropping rice in South China. *Journal of Integrative Agriculture, 20*(2), 565-580. doi:https://doi.org/10.1016/S2095-3119(20)63380-9

Ghaley, B. B., Høgh-Jensen, H., & Christiansen, J. L. (2010). Recovery of nitrogen fertilizer by traditional and improved rice cultivars in the Bhutan Highlands. *Plant and soil, 332*(1), 233-246. doi:https://doi.org/10.1007/s11104-010-0288-3

Ghimiray, M., Dorji, K. D., Katwal, T. B., Penjore, U., Dorji, S., Pem, S., . . . Pradhan, K. (2008). *Rice in Bhutan - A Source Book*. Thimphu: Council for RNR Research of Bhutan (CoRRB). Ministry of Agriculture, Bhutan.

GRiSP. (2013). *Rice almanac* (4 ed.). Los Baños (Philippines): Global Rice Science Partnership (GRiSP), International Rice Research Institute (IRRI).

Hasanuzzaman, M., Rahman, M. L., Roy, T. S., Ahmed, J. U., & Zobaer, A. S. M. (2009). Plant characters, yield components and yield of late transplanted Aman rice as affected by plant spacing and number of seedling per hill. *Advances in biological research, 3*(5-6), 201-207.

Imran, A. A. K., Inamullah, F. A. S., Naeem, L. Z. M., & Khan, M. N. (2015). Phenological traits of rice as influenced by seedling age and number of seedling per hill under temperate region. *J Biol Agri Healthcare, 5*, 145-149.

IRRI. (2002). *Standard Evaluation System for Rice (SES)*. Los Baños (Philippines): International Rice Research Institute (IRRI).

IRRI. (2006). Rice Knowledge Bank. Retrieved from http://www.knowledgebank.irri.org/ricebreedingcourse/Grain_quality.htm

IRRI. (2007). Rice Knowledge Bank: Planting the rice. Retrieved from http://www.knowledgebank.irri.org/ericeproduction/II.6_Seedling_and_plant_rate.htm

IRRI. (2019). *Annual Report 2019. Race for Impact*. Los Baños (Philippines): International Rice Research Institute (IRRI).

Islam, T., & Salam, M. A. (2017). Effect of number of seedlings hill-1 on the yield and yield contributing characters of short duration Aman rice cultivars. *Progressive Agriculture, 28*(4), 279-286. doi:https://doi.org/10.3329/pa.v28i4.36367

Kobata, T., Yoshida, H., Masiko, U., & Honda, T. (2013). Spikelet Sterility is Associated with a Lack of Assimilate in High-Spikelet-Number Rice. *Agronomy Journal, 105*(6), 1821-1831. doi: https://doi.org/10.2134/agronj2013.0115

Kovi, M. R., Bai, X., Mao, D., & Xing, Y. (2011). Impact of seasonal changes on spikelets per panicle, panicle length and plant height in rice (Oryza sativa L.). *Euphytica, 179*(2), 319-331. doi:https://doi.org/10.1007/s10681-010-0332-7.

Liu, J., He, L., Luo, H., Wu, T., Zhang, T., Wen, R., . . . Tang, X. (2021). Different seedling managements and fertilizer rates at tillering stage influences the yield formation and aroma content in fragrant rice. *Chilean journal of agricultural research, 81*(2), 128-137. doi:http://dx.doi.org/10.4067/S0718-58392021000200128

Mahamud, J. A., Haque, M. M., & Hasanuzzaman, M. (2013). Growth, dry matter production and yield performance of transplanted aman rice varieties influenced by seedling densities per hill. *International Journal of Sustainable Agriculture, 5*(1), 16-24.

Ninad, T. A., Bahadur, M. M., Hasan, M. A., Alam, M. M., & Rana, M. S. (2017). Effect of Spacing and Seedling Per Hill on the Performance of Aus Rice var. BRRI dhan48. *Bangladesh Agronomy Journal, 20*(2), 17-26. doi:https://doi.org/10.3329/baj.v20i2.37084

Nuruzzaman, M., Yamamoto, Y., Nitta, Y., Yoshida, T., & Miyazaki, A. (2000). Varietal differences in tillering ability of fourteen japonica and indica rice varieties. *Soil Science and Plant Nutrition, 46*(2), 381-391.

Promsomboon, P., Sennoi, R., Puthmee, T., Marubodee, R., Ruanpan, W., & Promsomboon, S. (2019). Effect of seedlings numbers per hill on the growth and yield of Kum Bangpra RiceVariety (Oryza sativa L.). *International Journal of Agricultural Technology, 15*(1), 103-112.

Pruneddu, G., & Spanu, A. (2001). Varietal comparison of rice in Sardinia. *Informatore Agrario, 57*(5), 47-49.

RSD. (2019). *RNR Census of Bhutan*. Thimphu: Renewable Natural Resources Statistics Division (RSD), Ministry of Agriculture and Forests, Royal Government of Bhutan.

Saker, B. C., & Nahar, N. (2016). Effect of number of seedlings per hill and spacing on growth and yield of transplanted Aman rice (Var. Ranisalute). *Khulna University Studies, 14*, 27-38.

Yao, X. Y., Li, Q., Liu, J., Jiang, S. K., Yang, S. L., Wang, J. Y., & Xu, Z. J. (2015). Dissection of QTLs for plant height and panicle length traits in rice under different environment. *Sci Agric Sin, 48*(3), 407-414. doi:https://doi.org/10.3864/j.issn.0578-1752.2015.03.01

Yumnam, L., Sorokhaibam, S., Laishram, B., Hajarimayum, S. S., Yambem, S., & Newmai, Z. K. (2021). Effect of planting date and spacing on growth and yield of black aromatic rice (*Oryza sativa* L.) cultivar chakhao poireiton. *The Pharma Innovation Journal 2021, 10*(3), 382-387.

Zhang, J., Li, M., Ashraf, U., Liu, S., & Li, R. (2019). Exploring the relationships between yield and yield-related traits for rice varieties released in China from 1978 to 2017. *Front. in Plant Sci., 10*, 1-12. doi:https://doi.org/10.3389/fpls.2019.00543

Effect of Gibberellic Acid and Germination Medium on Kiwifruit Seed Germination

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ABSTRACT

In Bhutan, cultivation of exotic commercial Kiwifruit fruit is new though wild kiwifruit is found aplenty. There is a high scope for kiwifruit cultivation as it is suitable in a wide range of areas in the country. It can also be grown organically due to the lack of any known serious pest or disease. Fittingly, kiwifruit is counted among the most potential high-value fruit crops in the country. Its cultivation is gaining popularity in the current times and it would consequently increase the demand for kiwifruit seedlings. Kiwifruit seeds exhibit physiological seed dormancy which hampers kiwifruit seedling production. Hence, a study was conducted in 2019 and 2020 at ARDC, Wengkhar. In the study, the combined effect of GA3 and growth medium on kiwifruit seed germination was studied using eight treatments with three replications laid out in a Completely Randomized Design. The study aimed to find out the germination parameters like germination percentage, relativized percentage, mean germination time, time taken for 10% germination, and time taken for 25% germination. According to the study, Cocopeat + 1500 ppm GA3 gave the best germination percentage at 51% followed by Biochar + 1500 PPM at 39% while rice bran and compost exhibited a low germination percentage. Overall, the mean germination time was found to be 50 days. The highest relativized percentage of 63 % was observed in Cocopeat + 1500 ppm GA3. As for the time taken to achieve 10% and 20% germination, those treatments with 1500 GA3 application had an advantage of approximately eight days over the others. Overall, considering the average weighted effect of germination percentage and time, Cocopeat + GA3 1500 PPM gave the best result followed by Biochar + GA3 1500 PPM and Cocopeat + GA3 0 PPM. Hence, the study indicates that GA3 treatment in combination with an appropriate germination medium can significantly enhance seed germination in kiwifruit.

1. Introduction

Kiwifruit (*Actinidia deliciosa*) is an edible berry of a woody vine. It is native to southwestern China. China is the global leader in kiwifruit production accounting for half of the total worldwide production (Ferguson, 2015). In Bhutan, cultivation of exotic kiwifruit is

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comparatively recent (Gyeltshen, 2011) even though wild kiwifruit is found aplenty. Kiwifruit can be grown at an altitude ranging from 1400 to 2500 m above sea level. Its cultivation is suitable for a vast range of areas in the country and it can be grown under organic management (Phuntsho, 2010) as incidences of major pests and diseases are yet to be observed. In recent times, kiwifruit cultivation is gaining attraction in the country due to its high value in the market. It can cost up to Nu.250 (3.38 USD) for six medium-sized kiwifruits. Consequently, the demand for kiwifruit seedlings is increasing. Seedling production through grafting and cuttings are two modes of propagation for kiwifruit (Luh & Wang, 1984). Grafting on seedling rootstocks is preferred over propagation through cuttings for its better root system (Celik, Zenginbal, & Özcan, 2006). However, seed dormancy is prevalent in kiwifruit seed germination. Seed dormancy can be defined as the inability of a viable seed to germinate in the given environment (Bewley, 1997). Physiological germination is common in seeds of angiosperms and it is the predominant form of dormancy in seed banks of temperate fruits (Finch-Savage & Leubner-Metzger, 2006). For that reason, the germination rate of kiwifruit seeds is low (Maghdouri et al., 2021). These contribute to the difficulty in kiwifruit seedling production, and thus the issue of higher prices.

Various approaches such as GA3 application, stratification, and scarification can help in obtaining greater seed germination (Sekhukhune, Nikolova, & Maila, 2016). Likewise, other growth hormones play an important role in seed germination. Dormant seeds can germinate under the influence of ethylene by impeding the adverse effect of ABA on seed germination; radical growth in seeds is affected by ethylene and gibberellins where gibberellins are considered to have the most significant influence (Miransari & Smith, 2014). IAA priming was also found to stimulate seed germination by aiding in the production of useful endogenous phytohormones and hindering the production of inhibitory phytohormones (Zhao et al., 2020).

Numerous other studies showed positive influence of GA3 on seed germination. GA hormone works to trigger the transition from embryo to seedling; GA signaling aids in auxin-induced root growth and development of seeds (Hauvermale & Steber, 2020). GA interacts with ABA and environmental conditions to break dormancy (Kucera, Cohn, & Leubner-Metzger, 2005). Hence GA3 treatment alleviates seed germination and shortens the mean germination time (Bishwas et al.).

Similarly, growing media plays an important role in germination through effective root establishment. Growing media provides nutrients and influences the quality of roots produced

(Shanker, Misra, Topwal, & Singh, 2019). The use of appropriate rooting medium facilitates proper aeration and drainage while the use of inappropriate substrate hinders the efficacy of water absorption (Miri-Nargesi, Sedaghathoor, & Environment, 2015). Currently, the use of growing media like biochar is picking up speed in Bhutan (Tamang, Dorji, & Dorji, 2020) and it has been released as a technology by the Technology Release Committee (TRC) of the Department of Agriculture, Ministry of Agriculture and Forests on 29 May 2020.

Therefore, this study assessed the effect of different growing media and gibberellic acid application in enhancing the germination percentage of kiwifruit seeds.

The experiment was carried out under greenhouse condition at ARDC Wengkhar which is located at 1708 m above sea level (27°16'14'' N; 091°16'19''E), Mongar, Bhutan. Seeds were manually extracted from well-ripened fruits of Wengkhar green cultivar harvested from the research block of ARDC Wengkhar. Extracted seeds were washed and air-dried.

2. Materials and Method

2.1 GA3 treatment

Seeds were soaked in 1500 ppm GA3 solution (7.3 pH) for 24 hours before the sowing time. As for the control treatments, seeds were soaked in distilled water (7.3 pH) for the same duration and kept in the laboratory at room temperature.

2.2 Experimental design

A two-factor factorial Completely Randomized Design with eight treatments and three replications was used. The factors comprised four growth mediums (Coco peat, Biochar, Rice bran, and Compost) and Gibberellic acid applications (0 ppm and 1500 ppm). In each treatment, 100 kiwifruit seeds were used as a unit of study. The treatments used were as follows:

1. T_1- Biochar + GA3 0 ppm
2. T_2- Biochar + GA3 1500 ppm
3. T_3- Coco peat + GA3 0 ppm
4. T_4- Coco peat + GA3 1500 ppm
5. T_5- Compost + GA3 0 ppm
6. T_6- Compost + GA3 1500 ppm
7. T_7- Rice bran + GA3 0 ppm
8. T_8- Rice bran + GA3 1500 ppm

2.3 Germination medium and Seed sowing

Flat germination trays of 60 cm x 30 cm were used. Each tray was divided into two halves to constitute two plots. As for the germination medium, easily available growth mediums like

biochar, coco peat, compost, and rice bran were used. Seed sowing was done on 14th February in 2019 and 2020. Data recording was done on daily basis after the first germination on 18th March 2019 and 16th March 2020 till 4th May in both years. Irrigation was done on alternate days using a knapsack sprayer to avoid displacement of seeds from the media.

2.4 Data collection

Data were recorded in Microsoft Excel sheet. Advanced seed germination measurements excel tool developed by Dr. Frahan Khalid (Khalid, 2021) was used to calculate attributes of seed germination measurements like germination percentage, relativized percentage, mean germination time, time taken for 10% germination, and time taken for 25% germination.

2.4.1 Germination percentage

Germination percentage gives the total seed germinated out of the entire sample taken in an experiment (Labouriau & Viladares, 1976). It was calculated using the following formula:

$$G\% = \frac{\sum_{i=1}^{k} n_i}{N} \times 100$$

Where;

n_i = number of seeds germinated in the ith time

N = total number of seeds used

2.4.2 Relativized percentage

It standardizes the assessment within the comparable treatments when there is a difference in the quantity of dormancy broken and it is calculated using the following formula (Fitch, Walck, Hidayati, & Ecology, 2007):

$$R(\%) = \frac{AP}{HP} \times 100$$

Where;

AP = acutual percentage

HP = highest percentage amongst group of data

2.4.3 Mean germination time

Mean germination time shows the time taken for the seed to emerge. The following formula was used to calculate it:

$$\bar{t} = \frac{\sum_{i=1}^{k} n_i t_i}{\sum_{i=1}^{k} n_i}$$

Where;

$n_i t_i =$ The product of seeds germinated at the ith time with the corresponding time interval

$n_i =$ number of seeds germinated in the ith time

2.4.4 Time to 10% germination

Time to 10% germination gives the time taken for 10% of the seeds to germinate. T10 was calculated using the following formula:

$$T10 = \frac{t_i + (\frac{\sum_{i=1}^{k} n_i}{10} - n_i)(t_j - t_i)}{n_j - n_i}$$

To find out the value of n_i and n_j, we take the cumulative number of seeds germinated for which the condition is given below:

$$n_i < \frac{\sum_{i=1}^{k} n_i}{10} < n_j$$

Where;

$n_i =$ nearest cumulative number of seeds germinated $(Cn_i) < \frac{\sum_{i=1}^{k} n_i}{10}$

$n_J =$ nearest cumulative number of seeds germinated $(Cn_J) > \frac{\sum_{i=1}^{k} n_i}{10}$

$t_i =$ the time interval corresponding to n_i

$t_j =$ the time interval corresponding to n_j

We can calculate $T25$ by replacing $\frac{\sum_{i=1}^{k} n_i}{10}$ with $\frac{\sum_{i=1}^{k} n_i}{4}$

2.5 Data analysis

The Statistical Tool for Agricultural Research (STAR) 2.1.0 software was used to analyze pooled data for two years using Analysis of Variance (ANOVA) model. Data were tested for assumption for ANOVA, and data were found to be normal and variance homogenous, thus satisfying the assumption of ANOVA. Tukey's Honest Significant Difference (HSD) Test was used for the Pairwise Mean Comparison of treatments since it is a robust method. Graphs and tables were constructed using MS Excel.

3. Results and Discussion

Effect of GA3 application and use of different substrates on kiwifruit seed germination are discussed in terms of seed germination measurement parameters like germination percentage, relativized percentage, mean germination time, time to 10% germination, and time to 25% germination.

3.1 Germination percentage

Actinidia species have exhibited high resistance to seed germination hence resulting in reduced seed emergence but the use of GA3 in Actinidia species was found to significantly ($P \leq .05$) affect germination percentage (Sekhukhune et al., 2016). Germination percentage was found to be considerably influenced by GA3 application and growing medium (Sharmaa et al., 2021). Our study found that the germination percentage was significantly affected by the use of various substrates and GA3 application ($P\text{-}value < .004$). Amidst the treatments, Coco peat + GA3 1500 ppm had the highest germination percentage of 51% followed by Biochar + GA3 1500 ppm, Coco peat + GA3 0 ppm and Biochar + GA3 0 ppm at 39%, 30% and 23% respectively. While the treatments with the lowest germination rates were rice bran (0 ppm and 1500 ppm) and compost (0 ppm and 1500 ppm) at 5% and 10% correspondingly. As per our study, the best germination percentage was acquired in treatments with cocopeat + GA3 1500 ppm as shown in Table 1. A similar study by Sekhukhune et al., (2016) found that GA3 treatment of 1435 to 1565 ppm was found to be preeminent for seed germination in kiwifruit. Likewise, cocopeat was found to considerably affect growth, germination, and development parameters due to its suitable physical, chemical, and biological properties (Bhardwaj, 2014). Biochar supplementation also offers a considerable influence on seed germination, shoot, and root growth (Bu, Xue, Wu, & Ma, 2020).

Table 1. Germination percentage of kiwifruit seeds under various treatments

Treatment	Mean
Biochar + GA3 0 ppm	23[ab]
Biochar + GA3 1500 ppm	39[ab]
Coco peat + GA3 0 ppm	30[ab]
Coco peat + GA3 1500 ppm	51[a]
Compost + GA3 0 ppm	10[b]
Compost + GA3 1500 ppm	10[b]
Rice bran + GA3 0 ppm	5[b]
Rice bran + GA3 1500 ppm	5[b]
P- value	< .004

Means with the same letter are not significantly different

GA3 treatment and medium combination had a paramount influence on seed germination, whereas a control treatment without GA3 treatment combined with a medium showed the least germination percentage (Celik et al., 2006).

3.2 Relativized percentage

The statistical value for relativized percentage of germination for various treatments was found to be significantly different (*P-value* < .004). The highest value was seen in T4 which is Cocopeat + GA3 1500 ppm followed by T2 (Biochar + GA3 1500 ppm) as given in Figure 1. As per Bhardwaj (2014) favourable physical and biological conditions in cocopeat have a good impact on germination and root development. Biochar provides good conditions for germination. Biochar is a conducive medium for earthworms (Raza et al., 2021). This could help in soil aeration and improve soil conditions.

The lowest value was observed in treatments with rice bran followed by compost. Low germination percentage in rice bran might be because of the presence of numerous antioxidant substances which cannot be decomposed easily (Nozoe et al., 2021). Whereas, compost performs better as a soil amendment. Supplementing the soil with compost enables the seed to germinate and establish itself in the soil (Paradelo et al., 2012) as compost acts as a fertilizer source by increasing soil nitrogen and decreasing C: N ratio (Raza et al., 2021).
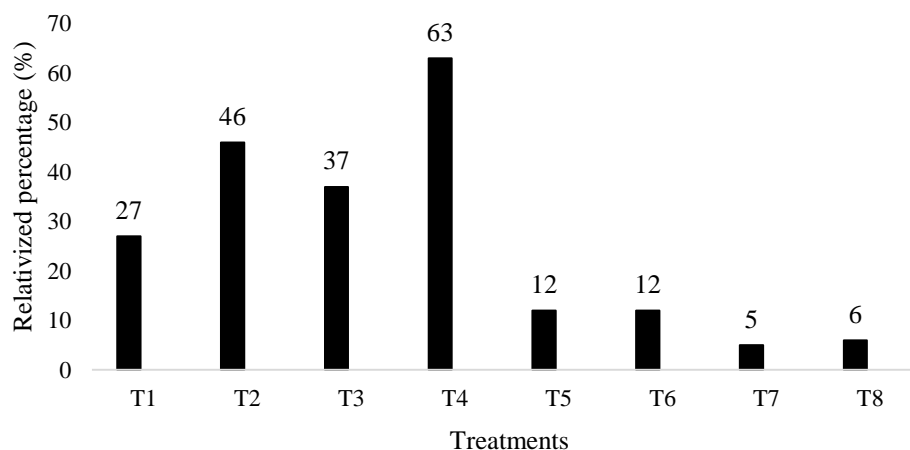
Figure 1. Relativized percentage for various treatments

3.3 Mean germination time

The overall germination time for our study was 50 days. The shortest germination time was observed in coco peat + GA3 0 ppm at 45 days and the longest germination time was taken in compost + GA3 0 ppm (Table 2). Varying germination time for each treatment was observed. The combined effect of GA3 was significant, on top of it, different mediums influenced the whole germination time separately (Celik et al., 2006).

Table 2. Average time taken for seed germination

Treatment	Mean germination time (Days)
Biochar + GA3 0 ppm	55
Biochar + GA3 1500 ppm	51
Cocopeat + GA3 0 ppm	45
Cocopeat + GA3 1500 ppm	46
Compost + GA3 0 ppm	56
Compost + GA3 1500 ppm	47
Rice bran + GA3 0 ppm	53
Rice bran + GA3 1500 ppm	47
Grand mean	50

3.4 Time to 10% germination and Time to 25% germination

Overall, there is not much variation in the time taken for 10% germination and 25% germination for individual treatment. As for time taken to germinate 10% and 25% of seeds, T2 which is Biochar + GA3 1500 ppm took the minimum time (42 days and 44 days respectively) while T7 which is Rice bran + GA3 0 ppm took the maximum time (60 days and 61 days respectively) as shown in Figure 2.

	T1	T2	T3	T4	T5	T6	T7	T8
Time to 10% germination	49	42	51	46	56	46	60	52
Time to 25% germination	50	44	54	48	57	48	61	51
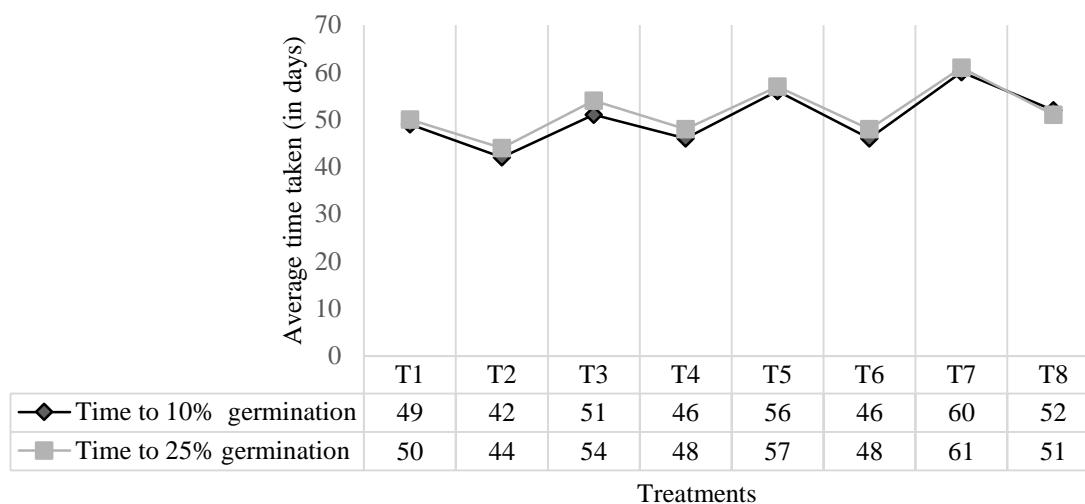
Treatments

Figure 2. Time taken to germinate 10 and 25% seeds

GA3 treated seeds germinate faster and their mean germination time is lesser than water-soaked seeds (Adhikari, Dhital, Ranabhat, & Poudel, 2021). Similarly, in our study, control treatments without GA3 treatment and other treatments with GA3 treatment showed considerable differences in their germination time. GA3 treated treatments took comparatively less time to acquire 10% and 25% germination. It can infer that the physiological dormancy of seeds can be prevented with GA treatment (Finch-Savage & Leubner-Metzger, 2006).

3.5 Overall ranking of growing media

To determine the most effective treatment combinations, the weighted average rank of mean germination percentage, germination time, relativized germination percentage, 10% and 25% germination percentage was calculated:

$$WA = \frac{\sum_{i=1}^{n} w_i X_i}{\sum_{i=1}^{n} w_i}$$

Where, WA is weighted average

w_i is weightage assigned to each parameter

X_i: list of parameters (germination percentage, germination time, relativized germination percentage, 10%, and 25% germination percentage)

Based on mean germination percentage, germination time, relativized percentage, 10% germination time, and 25% germination, the weighted average value for each treatment was estimated. Since the objective of the study is to study the effect of treatments on germination, the highest weightage was assigned to germination percentage, followed by relativized percentage, 10 and 25 percentage and time of germination. Overall, Cocopeat + GA3 1500 ppm received the highest weighted average score and rank as number one making it the most suitable treatment to enhance kiwifruit seeds germination followed by Biochar + GA3 1500 ppm and Cocopeat + GA3 0 ppm (Table 3).

Table 3. Overall ranking of weighted average

Treatments	Weighted average rank
Coco peat + GA3 1500 ppm	1
Biochar + GA3 1500 ppm	2
Coco peat + GA3 0 ppm	3
Biochar + GA3 0 ppm	4
Compost + GA3 0 ppm	5
Compost + GA3 1500 ppm	5
Rice bran + GA3 1500 ppm	7
Rice bran + GA3 0 ppm	8

4. Conclusion

In conclusion, the presented result showed that the combined effect of GA3 and different growth mediums can significantly enhance germination percentage in kiwifruit seeds. Based on the final weighted average of germination parameters, we can deduce that the use of growing

mediums like cocopeat and biochar along with 1500 ppm GA3 seed treatment can help in obtaining satisfactory seed germination.

The combined effect of GA3 and the use of suitable mediums like cocopeat and biochar gave the highest germination percentage of 51 % and 39 % respectively. GA3 treatment also helped in accelerating mean seed germination time. GA3 treatment in combination with all the germination mediums reduced the time taken to achieve 10% and 25% germination to an average of 47 and 48 days, respectively.

In our study, we observed that cocopeat enhanced seedling emergence but it remained stunted throughout the study period while emerged seedlings in biochar medium grew to a considerable height. Therefore, we would also like to recommend further study to assess the parameters of kiwifruit growth and development on top of seed germination through the combined use of germination medium and GA3 application.

Acknowledgement

References

Adhikari, B., Dhital, P. R., Ranabhat, S., & Poudel, H. (2021). Effect of seed hydro-priming durations on germination and seedling growth of bitter gourd (*Momordica charantia*). *PloS one, 16*(8), e0255258.

Bewley, J. D. (1997). Seed germination and dormancy. *The plant cell, 9*(7), 1055.

Bhardwaj, R. J. A. j. o. p. s. (2014). Effect of growing media on seed germination and seedling growth of papaya cv. Red lady. *8*(4), 178-184.

Bishwas, K., Amit, K., Santosh, S., Raj Kumar, K., & Dipendra, R. Effect of GA3 on Germination Parameters of Different Varieties of Kiwi. *Current Investigations in Agriculture and Current Research, 4*(3), 338-341. doi:10.32474/CIACR. 2018.04. 000186.

Bu, X., Xue, J., Wu, Y., & Ma, W. (2020). Effect of Biochar on Seed Germination and Seedling Growth of *Robinia pseudoacacia* L. In Karst Calcareous Soils. *Communications in Soil Science and Plant Analysis, 51*(3), 352-363.

Celik, H., Zenginbal, H., & Özcan, M. (2006). Enhancing germination of kiwifruit seeds with temperature, medium and gibberellic acid. *Horticultural Science, 33*(1), 39-45.

Ferguson, A. R. (2015). *Kiwifruit in the World*. Paper presented at the VIII International Symposium on Kiwifruit 1096, Dujiangyan (Chengdu), China

Finch-Savage, W., E., & Leubner-Metzger, G. (2006). Seed dormancy and the control of germination. *New phytologist, 171*(3), 501-523.

Fitch, E., Walck, J., Hidayati, S., & Ecology. (2007). Temporal Fulfilment of the Light Requirement for Seed Germination: an Example of its Use in Management of Rare Species. In S. W. Adkins, S. E. Ashmore, & S. C. Navie (Eds.), *Seeds: Biology, Development* (pp. 365). Cambridge, MA: CAB International.

Gyeltshen, S., Tshering, K., Phuntsho, L., Khampa, Penjor, T., Tshering, G., & Tomiyasu, Y. (2011). Kiwifruit - A potential fruit crop for fruit diversification. *Journal of Renewable Natural Resources Bhutan, 7*(11), 11-20.

Hauvermale, A. L., & Steber, C. M. (2020). GA signaling is essential for the embryo-to-seedling transition during Arabidopsis seed germination, a ghost story. *Plant signaling, 15*(1), 1705028.

Khalid, F. (2021). Advanced seed germination measurements excel tool. *Excel Tools: AgriStats Lecture*. http://agronexcel.blogspot.com/2018/06/this-tutorial-is-about-advanced-seed.html

Kucera, B., Cohn, M. A., & Leubner-Metzger, G. J. S. S. R. (2005). Plant hormone interactions during seed dormancy release and germination. *15*(4), 281-307.

Labouriau, L. G., & Viladares, M. E. B. (1976). On the germination of seeds of *Calotropis procera* (Ait.) Ait. f. *Annals of the Brazilian Science Academy, 48*(2), 263-284.

Luh, B. S., & Wang, Z. (1984). Kiwifruit. *Advances in food research, 29*, 279-309. doi:https://doi.org/10.1016/S0065-2628(08)60059-2

Maghdouri, M., Ghasemnezhad, M., Rabiei, B., Golmohammadi, M., & Atak, A. (2021). Optimizing Seed Germination and Seedling Growth in Different Kiwifruit Genotypes. *Horticulturae, 7*(9), 314. doi:https://doi.org/10.3390/horticulturae7090314

Miransari, M., & Smith, D. L. (2014). Plant hormones and seed germination. *Environmental and experimental botany, 99*, 110-121.

Miri-Nargesi, S., Sedaghathoor, S., & Environment. (2015). Study on rooting of kiwifruit cultivars (*Actinidia chinensis*) in different substrates and rooting hormones. *South-Western Journal of Horticulture Biology & Environment, 6*(1), 33-42.

Nozoe, T., Miura, S., Tazawa, J., Uchino, A., & Usui, Y. (2021). Factors destabilizing the control of *Monochoria vaginalis* by rice bran: its conflicting powers influence both suppression and promotion of germination in paddy soil. *Plant Production Science, 24*(1), 83-93.

Paradelo, R., Devesa-Rey, R., Cancelo-González, J., Basanta, R., Pena, M., Díaz-Fierros, F., & Barral, M. (2012). Effect of a compost mulch on seed germination and plant growth

in a burnt forest soil from NW Spain. *Journal of soil science plant nutrition, 12*(1), 73-86.

Phuntsho, L. G., S. Khampa, Tenzin K. Norbu U. Penjor, T. Tshering, G. Rai N.B. Phuntsho, Tashi S. Tshering, K. Tshering, K. & Tomiyasu, Y. (2010). *Horticulture Research Trial Report*. Retrieved from Mongar:

Raza, S. T., Wu, J., Ali, Z., Anjum, R., Bazai, N. A., Feyissa, A., & Chen, Z. (2021). Differential Effects of Organic Amendments on Maize Biomass and Nutrient Availability in Upland Calcareous Soil. *Atmosphere, 12*(8), 1034.

Sekhukhune, M. K., Nikolova, R. V., & Maila, M. Y. (2016). *Effect of cold stratification and gibberellic acid on in vitro germination of Actinidia arguta and Actinidia chinensis.* Paper presented at the VII International Symposium on Seed, Transplant and Stand Establishment of Horticultural Crops, Pretoria, South Africa.

Shanker, K., Misra, S., Topwal, M., & Singh, V. (2019). Research review on use of different rooting media in fruit crops. *Pharma Phytochem, 5*, 258-261.

Sharmaa, A., Devkotab, D., Thapaa, S., Sapkotaa, M., & Bistaa, B. (2021). Improving germination and stand establishment of kiwifruit (Actinidia deliciosa cv. hayward) seed through media selection and hormonal use in Dolakha, Nepal. *Tropical Agrobiodiversity (TRAB), 2*(1), 16-21. doi:http://doi.org/10.26480/trab.01.2021.16.21

Tamang, B., Dorji, T., & Dorji, L. (2020). Effect of Wood-derived Biochar as Growth and Development Medium for Kiwifruit Seedlings. *Bhutanese Journal of Agriculture, 4*(1), 28-37.

Zhao, T., Deng, X., Xiao, Q., Han, Y., Zhu, S., & Chen, J. (2020). IAA priming improves the germination and seedling growth in cotton (*Gossypium hirsutum* L.) via regulating the endogenous phytohormones and enhancing the sucrose metabolism. *Industrial crops and products, 155*, 1-9. doi:https://doi.org/10.1016/j.indcrop.2020.112788

Impacts of COVID-19 on Agri-Food Value Chains in the Global South

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ABSTRACT

This paper reviewed the impact of COVID-19 on the agri-food value chains in the Global South. The COVID-19 pandemic significantly affected the food value chain, one of the most critical of activities in an economy. The pandemic resulted in restrictive food trade policies, a curb on the movement of labourers, and increased financial pressure on food value chains, thereby, worsening the food security status in the Global South. Countries imposed trade restrictions to secure food for their domestic consumers. Small-scale firms and poor workers in the import-driven countries bore the major brunt. Food production and distribution networks were put to test, and labour markets shrunk with the OECD estimating a loss of over 500 million full-time jobs as well as an overall projected contraction in the economy by 4.9% in 2020. Bhutan, an import-driven country is no exception. However, most government and media reports hailed Bhutan's success in containing the pandemic through its strategic planning and effective implementation. The Health Ministry's statistics show a minimum number of COVID-19 confirmed cases (2,641) and a high recovery rate (99.39%) with a death rate of merely 0.11%. Due to the lack of robust empirical research, the impact on the agri-food value chain is ambiguous. Overall, considering the impact of COVID-19 in the Global South countries there is a need for a more resilient and responsive agri-food value chain to combat hunger and malnutrition.

Keywords: *COVID-19 Pandemic; Food Value Chain; Food Security; Global South; Lockdown.*

1. Introduction

The coronavirus (COVID-19) is an ongoing global concern that has affected the safety and welfare of billions of individuals worldwide (WHO, 2020a). After the 1918 influenza virus (H1N1), 1957 influenza virus (H2N2), 1968 influenza virus (H3N2), and the 2009 Pandemic flu, COVID-19 is the 5th pandemic (H1N) (Liu, Kuo, & Shih, 2020). However, we know much less about the character of this virus and its effect, and we are yet to precisely assess the extent and severity of its impact on the health and wellbeing of people around the globe. According to the WHO (2020b), there have been about 263,563,622 confirmed cases globally and 5,232,562 deaths as of 3 December 2021. Towards early 2020 when COVID-19 spread rapidly, many countries were forced to enforce public health emergencies. On March 11, 2020, the

WHO acknowledged the rapid spread of the virus and alerted countries to take preparatory and appropriate response actions (WHO, 2020b).

The WHO response plan includes strict public health measures to contain the pandemic. These caused the temporary closure of institutions, schools, tourism, restaurants, and restrictions on social gatherings and travel (WHO, 2020a). As a result of the stringent health regulations and the varied responses across governments, the impact spilt over into a complex food crisis globally. Food value chains faced significant disruptions, and widespread hunger has been recorded in early 2020 (UN, 2020). According to Khorsandi (2020), the World Food Programme (WFP) predicted that the crisis would lead to an additional 130 million people facing acute hunger. Experts have affirmed the breakdown of food access for vulnerable communities and the disruption of the supply chain on a global scale (Moseley & Battersby, 2020; Power, Doherty, Pybus, & Pickett, 2020).

The Global South countries, in general, are referred to as underdeveloped or developing nations, and the UN (2020) forecast that many people in the Global South may bear the long-term effects of COVID-19. The pandemic endangers marginalized actors along the value chain, including marginalized women, physically challenged people, and youths. The effect size may vary depending on the locations, pandemic response policies, and virus infection rate. Due to the complex effect of the crisis, generalizing its overall impact on food security is challenging.

Clapp and Moseley (2020) explain that the general contours of these effects can be traced by assessing disrupted food supplies, reduced incomes, price hikes, and levels of food insecurity for many people. Thus, this paper collated and analyzed how the COVID-19 pandemic has impacted the agri-food value chain in the Global South and what policy intervention approaches could be drawn from this experience. The information and data presented are based on secondary sources. First, the assessment of likely implications for a trade route for food supply was carried out, followed by the impact on the input supply chain. Furthermore, this paper analyzed how labour supply was disrupted along the value chain, affecting both producers and workers. The analysis also includes how consumers are affected due to loss in income and increased food prices. Finally, the paper discusses the policy response to the COVID-19 pandemic and attempts to recommend relevant policy measures to build a more resilient food value chain in the Global South.

2. Materials and Method

This paper used a secondary research approach to collate and analyze the impact of COVID-19 on the agri-food value chain. The majority of the information is obtained from published peered reviewed journals, research articles, and survey and newspaper reports. They were sourced from international journals such as Agricultural Economics, Global Food Security, and scientific databases like EBSCO host and Science Direct, including Google Scholar. Survey reports from the websites of the International Food Policy Research Institute (IFRI), Food and Agriculture Organisation (FAO), World Trade Organisation (WTO), and Organisation for Economic Co-operation and Development (OECD) were referred. Country reports were also examined to understand and compare results. Some of the research papers that were not readily accessible were requested from the authors via emails.

2.1 Conceptual framework

To adequately capture the impact of the COVID-19 pandemic on the food value chain, the review is split into four segments: 1. trade route and processing, 2. input supply and services, 3. producers, and 4. consumers (Figure 1). When countries have declared a state of health emergency and lockdowns were enforced to curb the further spread of the virus, the immediate impact was observed on international trade routes and food processing industries. Therefore, firstly, the impact on the trade routes was reviewed as in how lockdown protocols affected transportation and food processing. Due to the closure and restriction in transportation and the resulting hoarding of food commodities, there was an uneven increase in food prices. So, the impact of food prices on producers and consumers along the value chain was assessed. In the second stage, the effect on input supply and services was analyzed, followed by the impact on the producers. Lastly, this study delves into how consumers, in general, are affected, and how COVID-19 has impacted food security.
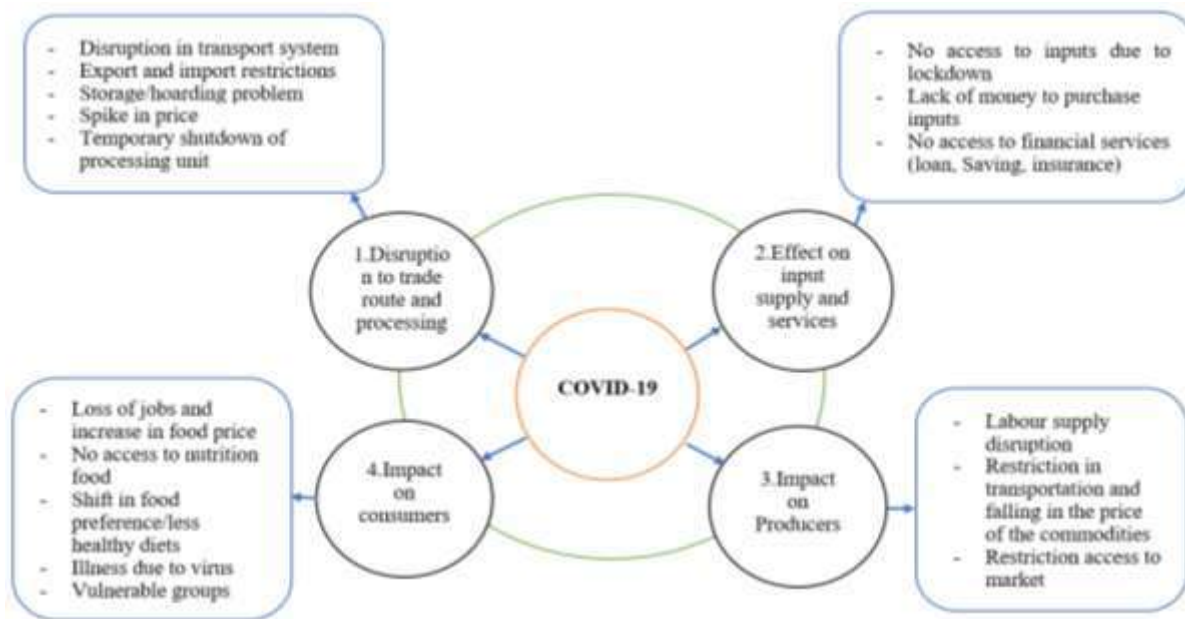
Figure 1: Conceptual framework for the impact of COVID-19 on the food value chain

3. Results and Discussion

3.1 Covid-19 effect on trade route and processing in Global South

COVID-19 crisis was not the first pandemic to have impacted the food value chain, particularly international trade. Several past pandemics and calamities have had major effects (Liu et al., 2020). According to Clapp and Moseley (2020), policy responses to past crises have somehow rendered countries' food systems vulnerable to Covid-19. Food policies have enabled food systems to be highly interconnected and economically efficient, yet largely exposed and highly vulnerable to such disruptions. It is estimated that about 80% of the world's population depends on food imports, and the expenditures on food imports in 2019 was three times the amount spent ten years ago (Economist, 2020). Nearly 20% of the dietary energy supply (DES) of a country's population comes from imported food (FAO et al., 2020). These figures suggest that many countries rely on both the export and import of significant quantities of food. The COVID-19 pandemic interrupted trade due to the closure of the international borders and restrictions on imports and exports. For example, Ukraine and Russia controlled the export of wheat and other cereal grains, which affected around 5% of global food calories (Laborde, 2020). In some countries, due to the shortage in the food supply, the ensuing demand spike led to an increase in the price of several food items. Further, the delay in transport and differences in the timing of COVID-19 lockdowns across different countries led to interruptions in food delivery as a direct result of missing intermediates (Bacchetta et al., 2021). According to

Laborde, Martin, and Vos (2020), the economic consequence in the United States, China, and Europe due to the pandemic has severely affected emerging countries through declines in trade and remittances, causing higher economic costs and hunger in these countries. Qualitative research in Latin America and the Caribbean shows that food loss and waste due to disruption in the transport system by COVID-19 was 49%, and an additional 18% through interruption in food trade (FAO & ECLAC, 2020). In the Bolivarian Republic of Venezuela, about 5 000 tonnes of vegetables that are shipped weekly remained in the fields due to lack of transport.

Several countries have imposed trade restrictions to secure food for their domestic consumers. Intuitively, the restrictions of export on staple foods by exporting countries will cause world prices to rise.
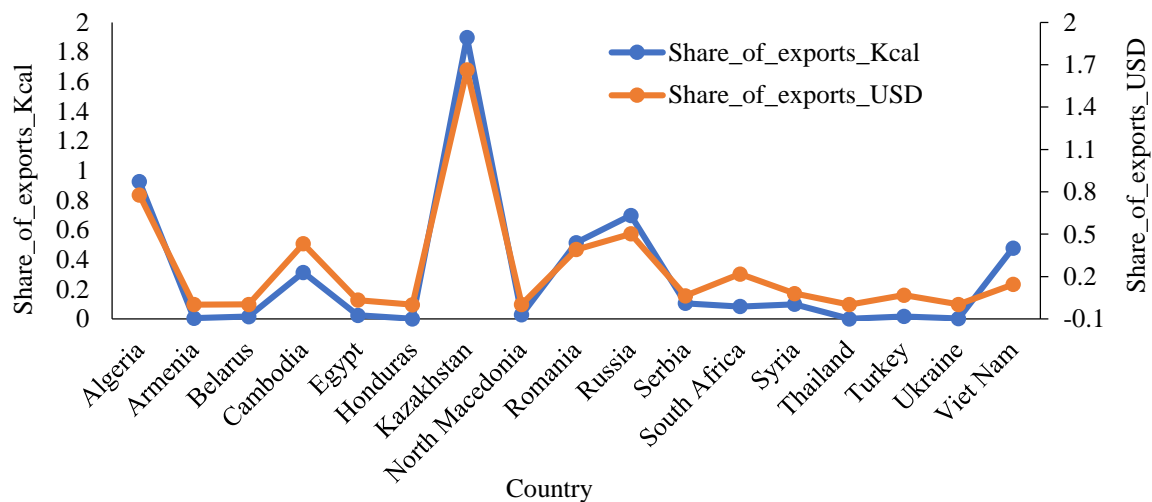
Figure 2. Share of restricted product in the country food exports measured in Kcal and USD. *Source*: *Adapted from Laborde et al. (2020).*

However, the global markets for staple crops remained good as they were well-stocked before the pandemic (UN, 2020a) except for the increase in market prices for wheat and rice. The world market prices for rice increased by 20% between January and May 2020 (Aday & Aday, 2020). This was because of the export restrictions imposed by major exporting countries and panic buying (Aday & Aday, 2020). According to IFPRI (2020), about 19 countries took measures to restrict exports of 27 food products due to the COVID-19 outbreak in the beginning. These restrictions adversely affected the food calories of the importing countries like Tajikistan, Uzbekistan, Afghanistan, and Azerbaijan by 79%, 70%, 61%, and 54%, respectively (IFPRI, 2020). Kyrgyzstan was also affected by the ban on the export of wheat and wheat flour by Kazakhstan.

The COVID-19 pandemic has a heterogeneous effect on food prices. In some countries, the disruptions in international trade triggered an increase in local prices, limiting the affordability of foods (Devereux, Béné, & Hoddinott, 2020). For example, Hirvonen, Abate, and de Brauw (2020) found that the pandemic led to significant increases in vegetable prices in Ethiopia, thereby, impacting the livelihoods and the diets of urban households. According to Espitia, Rocha, and Ruta (2020), countries that depend on imports experienced the highest rise in food prices. For instance, between February 2020 to July 2020, countries like Guyana, Venezuela, and Zambia saw a considerable increase (29%, 47%, and 49.8%, respectively) in local food prices, compared to developed nations such as Switzerland, the United Kingdom, and Canada that experienced stable or only moderate increases (Figure 2). In contrast, global cereal stocks were high, and generally, prices decreased at the start of the pandemic (FAO, 2020). Similarly, meat, dairy, sugar, and cooking oil prices also decreased towards the end of August 2020 (FAO, 2020).

The COVID-19 crisis has affected food security for households in Papua New Guinea both through disruptions in trade and an increase in the price of food commodities (Schmidt, Dorosh, & Gilbert, 2021). Since Papua New Guinea is a rice importing country with as much as 95% of its rice coming from imports, trade restrictions by major rice exporters led to a surge in the price in the domestic market. Between December 2019 and September 2020, Thailand and Vietnamese rice prices have increased by 25% on average (Schmidt et al., 2021). Schmidt et al. (2020) estimate that the rise in the rice price might lead to a fall in rice consumption by 17% for the poor. On the other hand, when restrictive export policies were applied, local producers could not sell, leading to wasting as well as economic losses (Arianina & Morris, 2020).

In Ghana, the demand for tilapia fish has drastically reduced due to closures in the tourism and hospitality industries. Most fish farmers had difficulty selling their fish products because of low demand and higher transportation costs during the COVID-19 pandemic. The crisis has diminished incomes for most actors along the aquaculture value chain and this could also affect future production (Ragasa, Amewu, & Asante, 2021). Likewise, the horticulture produces in Kenya too suffered. Kenya export about 80% of its horticulture produces to the European Union (Roussi, 2020). Due to the pandemic regulations, export contracts were cancelled, leaving products on the farm to rot (FAO, 2020a; Roussi, 2020).
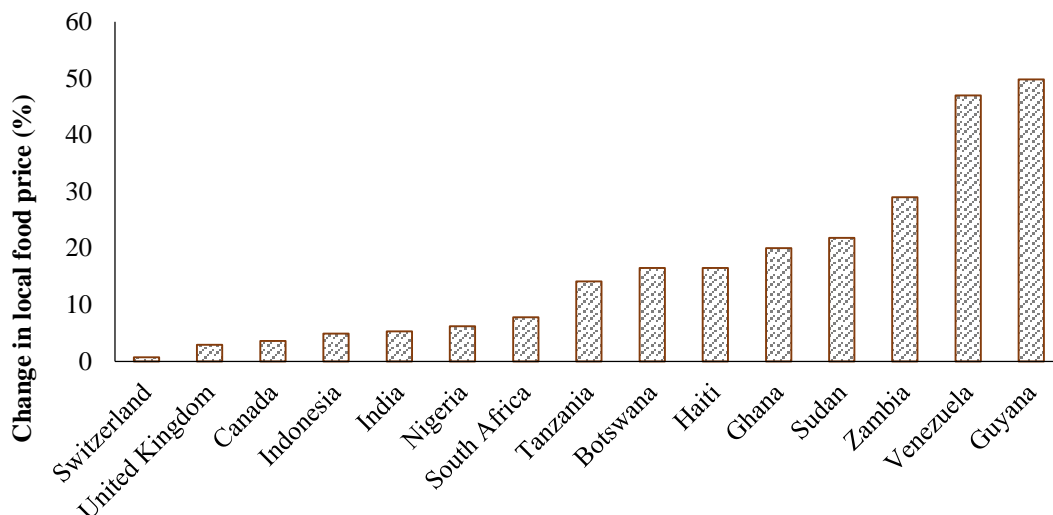
Figure 3. Variation in local food prices in the wake of COVID-19 (Feb. 14, 2020, to July 09, 2020) in some developed and Global South countries. *Source: Adapted from Clapp and Moseley (2020)*

Similarly, in East Africa and India, most horticulture produces rotted on the way because drivers found themselves having to spend days at border crossings for COVID screening, while the price in the city markets soared. In May 2020, exporters of mangoes from India to US markets faced increased freight costs of about 300% (Parkin and Rodrigues 2020), rendering such small businesses altogether unprofitable. Mango exports from Guatemala to the US too suffered shipment cancellations (FAO & ECLAC, 2020). In India, the nationwide lockdown forced the closure of feed mills, hatcheries, and processing plants. A decrease in demand from Europe and the United States reduced exports of frozen shrimp, which account for 70% of Indian seafood exports (Parkin & Rodrigues 2020). Similar impacts have been reported in Myanmar and Bangladesh (Mamun, Shieh, & Belton, 2020). The pandemic has disrupted international trade, leading to food crises affecting the livelihoods of a large proportion of humanity, especially the Global South countries.

The disruption in the supply chain has also affected the processing sector. Processing units were temporarily shut down due to either the outbreak of the disease or as precautionary measures. In Ghana, for example, about 534 employees contacted the virus at a fish-processing factory (Aday and Aday, 2020). In general, the UN (2020) predicted that the jobs in food processing, food services, and distribution would struggle by 60 % (around 650 million jobs) due to the pandemic. These include small-scale producers, youth, women, seasonal workers, migrants, and distributors, including informal food sellers (UN, 2020).

3.2   Covid-19 impact on input and services in the food value chain

Agricultural activities are season-bound with proper and largely fixed schedules. Since all processes and stages in an agri-food value chain are firmly connected, a small delay or disruption in one stage can affect production in other stages, ultimately affecting the whole value chain. A study carried out by Love et al. (2021) and Ragasa et al. (2021) found that most fish farmers in China, Ecuador, Norway, and Ghana were affected by disruptions in input supplies. Studies show that about 54% of fish farmers experienced difficulties accessing inputs, mainly fish feeds. For instance, fish farming in China saw a decrease of 40% - 50% in early 2020 as compared to 2019 (Clavelle, 2020). Shortage of farm inputs and a surge in input prices were also reported in many countries due to border closings. Ethiopia, Myanmar, Latin America, and the Caribbean experienced a shortage of farm inputs and increased prices for vegetable production. The price of fungicides, insecticides, herbicides, fertilizers, and improved seeds increased (Diao et al., 2020; FAO & ECLAC, 2020; Hirvonen, Minten, Mohammed, & Tamru, 2021). Pesticides production in China declined sharply due to lockdown measures affecting the producers (OECD, 2020). In West Africa, the Mali government has rolled back subsidies on fertilizers, further raising the cost of production (Diarra, 2020).

In Myanmar, some rural communities are at risk of losing their land because they used their landholdings as collateral for loans. Due to Covid-19-enforced lockdowns, economic activities took a backseat, leaving communities with little means to repay loans (Oxfam, 2020). An increase in the price of agricultural inputs, including seeds and fertilizers, creates challenges for farmers. Farmers are not able to buy inputs due to a loss in income. According to Oxfam (2020), closures of Myanmar's borders with China have hindered the supply of raw materials from the beginning of 2020, even before the virus was reported in Myanmar. Another study in Myanmar found that about two-thirds of the financial services in communities were disrupted, and banks were closed for several months. There were shortages of money in the ATMs, and further, people faced challenges in meeting credit officials from banks (MAPSA, 2021). The unavailability of such facilities could have largely hampered the farmers in Myanmar from purchasing agriculture inputs. Therefore, COVID-19 impact on the input and services could potentially disrupt agricultural production over the following season and further into the future.

3.3   Impact on the producers

The COVID-19 pandemic regulations have hampered production capacities, mainly through the disruption of labour supply. Farm production in developing countries is generally labour-intensive with a string of processes such as ploughing, planting, harvesting, post-harvest

handling, and transportation that usually entails workers working in close coordination. Prohibition of mass gatherings as part of the COVID-19 safety protocol directly hindered food production. Amare, Abay, Tiberti, and Chamberlin (2021) reported that Nigeria saw a significant reduction in labour market activities. The study observed a positive relationship between the number of confirmed cases and the reduction in major economic activities. About 72 % of households reported a decline in farming income, while 84 % reported a reduction in income from non-farm businesses. Poor households living in remote areas and with high rates of infection experienced deterioration in food security (Amare et al., 2021). On the other hand, urban households did not suffer much from food insecurity, although economic activities were significantly reduced. This could be most likely attributed to better underlying food security and improved market access in urban areas.

Studies carried out in India, Thailand, Ecuador, and Ghana on seafood companies found that most of the employees lost their jobs, exposing them to substantial food security risks (Havice, Marschke, & Vandergeest, 2020; Marschke et al., 2021; Reardon & Swinnen, 2020). According to Wang et al. (2021), the restrictions on labour mobility were reported as the primary cause of substantial income declines in rural livelihoods in Hubei Province, China, the initial epicentre of the pandemic. The findings confirm that about 74% of respondents reported that villagers had stopped working due to closures of workplaces. These figures rise further if rural workers who are unable to reach workplaces due to travel restrictions are also included. Consequently, most villagers (92%) had to forego substantial income.

The effect of COVID-19 on production companies varies due to the differences in their economies of scale. Van Hoyweghen et al. (2021) found that large fresh fruits and vegetable companies in Senegal were barely affected compared to the smaller firms. These large firms are vertically integrated into the value chain as well as export-oriented. They managed to limit labour supply disruptions during the COVID-19 outbreak by supplying their workers with health-protective gears, expanding the number of shifts, and providing safer transport facilities. In contrast, small domestic companies showed little capacity to adjust and respond to the shock. They were severely affected by a lack of access to credits and cold chain facilities (Van Hoyweghen et al., 2021). At the same time, Minten, Mohammed, and Tamru (2020) found that small vegetable farms in Ethiopia are less affected than medium-sized farms by the pandemic-induced labour disruptions because smaller farms relied less on hired labour. This finding is consistent with that of Reardon and Swinnen (2020) who report that labour disruption due to

COVID-19 restrictions shows an inverted U-shaped relationship with economy of scale; that is, medium-sized firms relatively show high dependence on hired labour.

Smallholder farmers were adversely impacted due to restrictions in transportation and a slump in the selling price. For instance, cocoa prices in Ghana, Côte d'Ivoire, and Nigeria declined by 25% between the end of February and mid-July 2020 (Clapp & Moseley, 2020). Interestingly, it is reported that the pandemic did not severely affect some rural agriculture production areas because they were geographically located far away from urban densities (Moseley & Battersby, 2020), and were mostly insulated from rising COVID-19 cases in the urban areas.

3.4 COVID-19 impact on consumers

During pandemics, the cost of food becomes more relevant to consumers. The situation turns worst when the prices of the essential food items increase and their income shrink. However, Clapp and Moseley (2020) observed that the current COVID-19 crisis did not cause a dramatic surge in food prices in the global market but instead had an unforeseen impact on food systems that are often difficult to disengage. At first, food supplies were disrupted due to lockdown measures, and people working in the food system got infected with the virus. This was followed by massive job loss caused by the pandemic, leading to increased hunger (Clapp and Moseley, 2020). Several combining factors have been attributed to unequal food prices at local and global scales that have worsened hunger in many developing countries. People's ability to buy food was hugely affected (Clapp and Moseley, 2020). The International Monetary Fund (IMF) projected a 4.9% overall economic contraction by the end of 2020 (OECD, 2020), and developing economies are expected to fall by around 3% on average (IMF, 2020). The global economic contraction is primarily the result of jobs lost due to lockdown measures during the pandemic. According to the International Labour Organization (ILO, 2020), more than 500 million full-time jobs have been lost since the start of the pandemic. The loss of jobs and income is bound to severely affect food security because of the direct implications on their ability to purchase food.

A survey in Myanmar between January and June 2020 (n =1,072) showed that 51% of respondents lost jobs in various livelihood activities. People without land were mostly affected more by the crisis, mainly because of loss in farm incomes and the absence of non-farm employment (Ragasa et al., 2021). The authors also found that women and men in these landless households were equally affected by lower wages and lack of farm work during COVID-19. The loss of jobs and increase in the food prices in the market put people in

worsening situations. For instance, FAO estimated that undernourishment overall could rise between 83 and 132 million (FAO et al. 2020) with a drop in income. According to the UN (2020), about 45 million people became acutely food insecure, of which the majority (33 million) are in South and Southeast Asia, followed by sub-Saharan Africa South (12 million).

Dietary preferences are also influenced by food shortages, increases in food prices, and lack of money. COVID-19 disrupted markets, mainly the perishable food commodities compared to staple foods. The disruptions in perishable food items supply aggravated income-related issues, causing poor households to shift their consumption and reduce dietary diversity (Clapp and Moseley, 2020). This is consistent with the study conducted in Ethiopia, India, and Guatemalan (Ceballos, Hernandez, & Paz, 2021; Harris et al., 2020; Hirvonen et al., 2021) that discounts in household food consumption primarily reduced purchases of nutritious foods such as fruit, meat, eggs, and dairy. Such poor dietary patterns may potentially affect human health and development, especially the growing children and women. The ability to cope with income shock due to the COVID-19 pandemic varies among low-income households. Hirvonen et al. (2021) found that a rundown of savings was the first recourse as a coping strategy in Ethiopia, but fewer households possessed enough savings to meet their food needs for a month or more.

Aday and Aday (2020) found that the closure of the hotels and open area services for eating affected the purchasing habits and shifted the demand for food from retail and supermarkets. He claims that almost 100% of the customers purchase food from supermarkets during the COVID-19 pandemic. They buy food items that can be stored for an extended period and stock them at home due to misinformation. Food stocking led to increasing prices and a shortage of stocks in supermarkets, affecting most consumers in developing countries (Aday and Aday, 2020).

In many parts of Asia, Sub-Saharan Africa, and Latin America, most workers are engaged under informal arrangements, where their rights are unclear (FAO, 2020b). These workers have suffered from COVID-19 infections due to a lack of clarity on their rights and limited access to healthcare and social support services at their workplaces (FAO, 2020b). Such unfavourable circumstances could increase the extent of malnutrition of breadwinners and their dependents in a household. Another study in China found that labour movement restrictions caused a decrease in rural income, and many people were separated from their work in urban areas (Swinnen & Vos, 2021). Similarly, Guatemala, India, Myanmar, and Nepal reported a significant reduction in remittance incomes (Ceballos et al., 2021; Diao et al., 2020; Nicola et

al., 2020). This decline in remittance might have exposed those households more vulnerable to food insecurity and reduced diet due to lack of money. The diets of the school students from low-income family backgrounds were also affected by the pandemic. According to the survey, ten million Latin American and Caribbean students were put at risk because of school closure, whose almost only food source was school meals (FAO & ECLAC, 2020). Introspectively, economically vulnerable food consumers are the most affected group in the food value chain.

3.5   The case of Bhutan – a summary

Bhutan - a small mountainous landlocked country seems to be coming out of the crisis in a different way. It may appear to be an outlier in many ways, such as its geographical location, governance, economy, and social-cultural attributes. Although located in between two global giants - China and India, where COVID-19 cases are recorded the highest in the region, Bhutan wisely planned and implemented its strategies to safeguard her countrymen and everyone within from the pandemic. At the time of preparing this manuscript, Bhutan recorded only 2,641 COVID-19 confirmed cases with a high recovery rate (99.39%) and the lowest death rate of 0.11% in the world, clearly indicating the achievements Bhutan has made in successfully containing the pandemic (MoH, 2021).

However, due to the lack of empirical studies, the impact on the agri-food value chain is ambiguous. While detailed and proper research in the coming years will significantly contribute to our understanding of the impacts of the pandemic, a cursory look at the developments in the aftermath of the pandemic does indicate some form of minor disruption in the country's food system as a result of the restrictions imposed to contain the pandemic. Supplies of food commodities were affected by panic buying and hoarding (Yuden, 2020a) while imported fruits at the country's central market nearly doubled. The pandemic also exposed the country's lack of a reliable and efficient marketing and distribution system when as the first lockdown came into effect in August 2020, both the government and the public were caught unprepared (Yuden, 2020b). Collection and distribution of fresh food commodities like vegetables amongst the consumers suffered. Although exports did not suffer altogether, border restrictions and associated COVID-19 measures that neighbouring India enforced slowed down Bhutanese exports. Perishable commodities like cole crops that make up the major export volume in summer for farmers were affected when regulated markets in West Bengal closed due to the pandemic (Giri, 2020).

On the flip side, the pandemic provided an opportunity to raise domestic food production. A

few reported incidences of disturbance in farm inputs supply did not have major adverse effects on production, and the nationwide lockdown probably did not cause serious unsettling disruption in farm labour set-up. On the contrary, the 2020 production figure for most agricultural commodities in Bhutan shows an increase over the previous year's figures (RSD, 2021). This can be largely attributed to the many measures the government put in place to offset the effects of the pandemic – of relevance is the Economic Contingency Plan (ECP) prepared and launched in May 2020 (Pem, 2020). The ECP put in a series of coping measures in the three important sectors of labour, tourism and agriculture. The first series of the ECP for agriculture with an outlay of Nu. 200 million targeted boosting the production of cereals, lentils, oilseeds, and vegetables through a range of support across the value chain that include technology, marketing, value-addition, and year-round accessibility. As of October 2021, the intervention directly helped produce a little over 30,974 Mt of cereals, oilseeds, grain legumes and priority vegetables such as chilli, onion, and tomatoes. Farmers sold around half of the produce which generated total revenue of Nu. 385.78 million (DoA, 2021).

3.6 Policy discussion

The current pandemic has forced policymakers and analysts to debate the need for interventions that help restructure food value chains to mitigate stress and shocks on essential food trade and improve the resilience of value chains. For example, certain factions in the US (Lighthizer, 2020) declare an end to what they claim as the era of offshoring US jobs while some call for larger autonomy in the US food value chain system, asserting that principles of food autonomy and support for local markets should gain centre stage. On the contrary, some economists are not convinced about the need for a restructuring of value chains as the best policy response. Freund (2020) and Miroudot (2020) argue that taking supply chains back home would challenge economic wisdom and that self-sufficiency-centred production will not benefit global value chains. Since most of the Global South are import-driven countries, the strategy of restructuring value chains to limit dependency on imports could further deteriorate the existing hunger and malnutrition status.

The effect of the COVID-19 pandemic on the food value chain varies across places due to its infection rate and the type of response measures adopted. For instance, poor households in urban areas experienced more challenges to food insecurity due to more infection rates and lockdown. Similarly, small food producers in remote areas and infection zones are more vulnerable to food insecurity because of lockdowns. At the individual or community level,

people strived to string together their different mechanisms to cope with the situation. A study in Myanmar showed that people resorted to borrowing from friends or selling their household assets such as gold, land, or jewellery (Ragasa et al., 2021). However, these alternatives typically were not sufficient to prevent increased food insecurity for extended periods.

Political governments, global leaders, and various international organizations can play a significant role in coping mechanisms against this pandemic. On the international trade front, world leaders could come together and facilitate trade routes to remain open for essential food transport following hygienic practices. Stringent regulations like lockdowns in the early phase of the pandemic significantly affected the food value chain. Countries must find ways to lift export bans and import taxes to make food supplies available and affordable. Yet, such measures including working from home were the only effective response to temper the overall impact on the health and wellbeing of the people.

On the consumer's side, governments can link producers and consumers without intermediaries during such pandemics. Furthermore, governments could introduce support systems like buying back schemes. For example, Ceballos et al. (2021) found that wheat farmers in India were guaranteed ready markets through buying-back system at fixed prices. This has averted wheat price declines and offered income protection to wheat growers, and made wheat available to consumers.

While COVID-19 has revealed food value chain vulnerabilities, it has also prompted new ideas and opportunities in using technology and organizing food value chains. In a major crisis, value chain experts tend to reinvent business procedures (Reardon & Swinnen, 2020), and likewise, the COVID-19 pandemic has triggered a rise in the usage of digital platforms for organizing food delivery and food services. Although digital platforms have been in use, they were primarily limited to developed countries. The pandemic brought digital technology to the fore, thereby, reinforcing its importance and role, and its subsequent diffusion in developing countries became faster (Reardon & Swinnen, 2020). Businesses found themselves increasingly shifting to e-commerce. Government and policymakers should hence emphasize the extension of modern technologies to facilitate the food value chain to preempt such eventualities in times of crisis.

The pandemic also provided opportunities for businesses all over in building reliable and long-term workforce through skilling initiatives for local employees to prevent meltdowns as a direct consequence of relying heavily on foreign labour. Smallholder farmers have faced difficulties

in exporting their produces to the market due to travel restrictions. Building agricultural production collection centres or improved storage structures in the communities can facilitate marketing and minimize food waste. Governments could also support interest-free loans to needy farmers so that production could resume and ensure that the food supply chain remains uninterrupted. Further, the facilitation of timely support in input supply is essential so that the food production in the coming season is not affected. Establishing proper biosecurity is also seen as one of the necessary arrangements to ensure health and food safety throughout the value chain.

4. Conclusion

The ongoing COVID-19 pandemic has impacted the lives of billions of people throughout the world, putting all critical value chain agents at risk. Besides food producers and consumers, migrant farm labour and food processing workers were also affected, leading to food supply disruptions. The global economic recession as an immediate fallout from the pandemic resulted in massive job losses and worsened access to food due to increasing food prices. Export restrictions and increases in freight costs gave rise to highly uneven food prices, thus highlighting the risks in import-dependent Global South countries. On the other hand, smaller farmers in rural areas could not sell their produce due to restrictions in movement, resulting in their produces getting damaged in the field and storage. Bhutan, an import-driven country is no exception to this. Although national newspaper and country reports mentioned successful stories in containing the virus, the implication on its food value chain needs further empirical research and analysis for future policy intervention.

Overall, producing excess food for global value chains does not promise markets for poor producers or access to food for those who have their incomes compromised. Instead, more diverse production and enhanced trading platforms need to be prioritized. Global rules of trade may require a relook to assure affordable food for all in such times of distress, and move away from restrictive policies like export bans. Furthermore, smallholder farmers and vulnerable workers should be supported financially. The use of modern technology, e-commerce, artificial intelligence, and robotics could be explored to complement human labour to reduce disease spread and ensure uninterrupted food distribution during such crises. Considering the extent of food import dependence in many Global South countries, keeping the trade routes open is very important to reduce hunger and malnutrition.

Reference

Aday, S., & Aday, M. S. (2020). Impact of COVID-19 on the food supply chain. *Food Quality and Safety, 4*(4), 167-180. doi:10.1093/fqsafe/fyaa024

Amare, M., Abay, K. A., Tiberti, L., & Chamberlin, J. (2021). COVID-19 and food security: Panel data evidence from Nigeria. *Food Policy, 101*, 102099. doi:10.1016/j.foodpol.2021.102099

Arianina, K., & Morris, P. (2020). COVID-19 Export Restrictions Threaten Global Food Supply. Retrieved from Law360 website:https://www.squirepattonboggs.com/-/media/files/insights/publications/2020/05/covid-19-export-restrictions-threaten-globalglobal-food%20supply/law360covid19exportrestrictionsthreatenglobal

foodsupply.pdf.

Bacchetta, M., Bekkers, E., Piermartini, R., Rubinova, S., Stolzenburg, V., & Xu, A. (2021). COVID-19 and global value chains: A discussion of arguments on value chain organization and the role of the WTO *WTO Staff Working Paper No. ERSD-2021-3*. doi:10.30875/40db0106-en

Ceballos, F., Hernandez, M. A., & Paz, C. (2021). Short-term impacts of COVID-19 on food security and nutrition in rural Guatemala: Phone-based farm household survey evidence. *Agricultural Economics, 52*(3), 477-494. doi:10.1111/agec.12629

Clapp, J., & Moseley, W. G. (2020). This food crisis is different: COVID-19 and the fragility of the neoliberal food security order. *The Journal of Peasant Studies, 47*(7), 1393-1417. doi:10.1080/03066150.2020.1823838

Clavelle, T. (2020). Global fisheries during COVID-19. Retrieved from Global Fishing Watch website: https://globalfishingwatch.org/data/global-fisheries-during-covid-19/

Devereux, S., Béné, C., & Hoddinott, J. (2020). Conceptualising COVID-19's impacts on household food security. *Food Security, 12*(4), 769-772. doi:10.1007/s12571-020-01085-0

Diao, X., Aung, N., Lwin, W. Y., Zone, P. P., Nyunt, K. M., & Thurlow, J. (2020). Assessing the impacts of COVID-19 on Myanmar's economy: A Social Accounting Matrix (SAM) multiplier approach. *Myanmar SSP Working Paper 1, 1*.

Diarra, S. (2020). As Mali fights coronavirus, cotton farmers fear loss of climate aid. *Reuters*. Retrieved from https://www.reuters.com/article/us-health-coronavirus-mali-climatechange/as-mali-fights-coronavirus-cotton-farmers-fear-loss-of-climate-aid-idUSKBN23N1SR

DoA. (2021). *Progress Report on ECP (Agriculture) - Unpublished Report*: Department of Agriculture, Ministry of Agriculture & Forests.

Espitia, A., Rocha, N., & Ruta, M. (2020). Covid-19 and food protectionism: The impact of the pandemic and export restrictions on world food markets. *World Bank Policy Research Working Paper*(9253).

FAO. (2020). Food Outlook – Biannual Report on Global Food Markets. Retrieved from Food and Agriculture Organization (FAO) website: http://www.fao.org/3/ca9509en/ca9509en.pdf doi:10.4060/ca9509en

FAO. (2020a). COVID-19 and the risk to food supply chains: How to respond? Retrieved from Food and Agriculture Organization (FAO) website: http://www.fao.org/3/ca8388en/CA8388EN.pdf doi:10.4060/ca8388en

FAO. (2020b). Policy Responses to keep input markets flowing in times of COVID-19. *Policy Brief*. Retrieved from Food and Agriculture Organization (FAO) website: http://www.fao.org/3/ca8979en/CA8979EN.pdf doi:10.4060/ca8979en

FAO, & ECLAC. (2020). Food systems and COVID-19 in Latin America and the Caribbean: Health risks; safety of workers and food safety. (Bulletin 4). https://www.fao.org/3/ca9112en/CA9112EN.pdf doi:10.4060/ca9112en

FAO, IFAD, UNICEF, WFP, & WHO. (2020). *The State of Food Security and Nutrition in the World. Transforming food systems for affordable healthy diets*. Rome: FAO, IFAD, UNICEF, WFP and WHO.

Freund, C. (2020, May 1, 2020). Governments could bring supply chains home. It would defy economic rationality. *Barron's*.

Giri, P. (2020, 23 June 2020). With Covid-19 cases spiking, traders suspend business in north Bengal's biggest town. *Hindustan Times*. Retrieved from https://www.hindustantimes.com/india-news/with-covid-19-cases-spiking-traders-suspend-business-in-north-bengal-s-biggest-town/story-SXq1KXK8zI0SVuGb2w5hfM.html

Harris, J., Depenbusch, L., Pal, A. A., Nair, R. M., & Ramasamy, S. (2020). Food system disruption: initial livelihood and dietary effects of COVID-19 on vegetable producers in India. *Food Security, 12*(4), 841-851. doi:10.1007/s12571-020-01064-5

Havice, E., Marschke, M., & Vandergeest, P. (2020). Industrial seafood systems in the immobilizing COVID-19 moment. *Agriculture and Human Values, 37*(3), 655-656. doi:10.1007/s10460-020-10117-6

Hirvonen, K., Abate, G. T., & de Brauw, A. (2020). Survey suggests rising risk of food and nutrition insecurity in Addis Ababa, Ethiopia, as COVID-19 restrictions continue. In J. Swinnen & J. McDermott (Eds.), COVID-19 and Global Food Security (pp. 46-49). Washington, DC: International Food Policy Research Institute (IFPRI). Retrieved from https://www.ifpri.org/publication/survey-suggests-rising-risk-food-and-nutrition-insecurity-addis-ababa-ethiopia-covid-19. doi:10.2499/p15738coll2.133762_10

Hirvonen, K., Minten, B., Mohammed, B., & Tamru, S. (2021). Food prices and marketing margins during the COVID-19 pandemic: Evidence from vegetable value chains in Ethiopia. *Agricultural Economics, 52*(3), 407-421. doi:10.1111/agec.12626

IFPRI. (2020). COVID-19 Food Trade Policy Tracker [Dataset]. (on May 23, 2021). Retrieved from International Food Policy Research Institute (IFPRI) website: https://www.ifpri.org/project/covid-19-food-trade-policy-tracker

ILO. (2020). COVID-19 and the impact on agriculture and food security. *International Labour Organization (ILO) Sectoral Brief.* Retrieved from International Labour Organization (ILO) website: https://www.ilo.org/wcmsp5/groups/public/---ed_dialogue/---sector/documents/briefingnote/wcms_742023.pdf

IMF. (2020). World economic outlook, April 2020: the great lockdown. Retrieved from International Monetary Fund website: https://www.imf.org/en/Publications/WEO/Issues/2020/04/14/weo-april-2020

Khorsandi, P. (2020). WFP chief warns of 'hunger pandemic' as Global Food Crises Report launched. *World Food Programme Insight, 22.* https://www.wfp.org/stories/wfp-chief-warns-hunger-pandemic-global-food-crises-report-launched

Laborde, D. (2020). Food export restrictions during the Covid-19 crisis. *International Food Policy Research Institute (IFPRI).* Retrieved from International Food Policy Research Institute (IFPRI) website:

Laborde, D., Martin, W., & Vos, R. (2020). Poverty and food insecurity could grow dramatically as COVID-19 spreads. In J. Swinnen & J. McDermott (Eds.), COVID-19 and Global Food Security (pp. 16-19). Washington, DC: International Food Policy Research Institute (IFPRI). Retrieved from https://www.ifpri.org/blog/poverty-and-food-insecurity-could-grow-dramatically-covid-19-spreads. doi:10.2499/p15738coll2.133762_02

Lighthizer, R. E. (2020, May 11). The Era of Offshoring U.S. Jobs Is Over. *The New York Times.* Retrieved from https://www.nytimes.com/2020/05/11/opinion/coronavirus-jobs-offshoring

Liu, Y., Kuo, R., & Shih, S. (2020). COVID-19: The first documented coronavirus pandemic in history. *Biomedical Journal, 43*(4), 328-333. doi:10.1016/j.bj.2020.04.007

Love, D. C., Allison, E. H., Asche, F., Belton, B., Cottrell, R. S., Froehlich, H. E., . . . Zhang, W. (2021). Emerging COVID-19 impacts, responses, and lessons for building resilience in the seafood system. *Global Food Security, 28*, 100494. doi:10.1016/j.gfs.2021.100494

Mamun, A., Shieh, J., & Belton, B. (2020). *Qualitative assessment of COVID-19 impacts on aquatic food value chains in Bangladesh.* Dhaka: CGIAR Research Program on Fish Agri-Food Systems (FISH).

MAPSA. (2021). *Community perceptions of changes in rural livelihoods since the onset of COVID-19 in Myanmar: Insights from Round 7 of the National COVID-19 Community Survey (NCCS)* Washington, DC: Myanmar Agriculture Policy Support Activity (MAPSA), International Food Policy Research Institute (IFPRI).

Marschke, M., Vandergeest, P., Havice, E., Kadfak, A., Duker, P., Isopescu, I., & MacDonnell, M. (2021). COVID-19, instability and migrant fish workers in Asia. *Maritime Studies, 20*(1), 87-99. doi:10.1007/s40152-020-00205-y

Minten, B., Mohammed, B., & Tamru, S. (2020). Emerging medium-scale tenant farming, gig economies, and the COVID-19 disruption: The case of commercial vegetable clusters in Ethiopia. *The European Journal of Development Research, 32*(5), 1402-1429. doi:10.1057/s41287-020-00315-7

Miroudot, S. (2020). Resilience versus robustness in global value chains: Some policy implications. In R. E. Baldwin & S. J. Evenett (Eds.), COVID-19 and trade policy: Why turning inward won't work (pp. 117-130). London: CEPR Press.

MoH. (2021). National Situation Update on COVID-19. Retrieved from https://www.moh.gov.bt/

Moseley, W. G., & Battersby, J. (2020). The vulnerability and resilience of African food systems, food security, and nutrition in the context of the COVID-19 pandemic. *African Studies Review, 63*(3), 449-461. doi:10.1017/asr.2020.72

Nicola, M., Alsafi, Z., Sohrabi, C., Kerwan, A., Al-Jabir, A., Iosifidis, C., . . . Agha, R. (2020). The socio-economic implications of the coronavirus pandemic (COVID-19): A review. *International Journal of Surgery, 78*, 185-193. doi:10.1016/j.ijsu.2020.04.018

OECD. (2020). Food supply chains and COVID-19: impacts and policy lessons. Retrieved from OECD - Better Policies for Better Lives website: http://www.oecd.org/coronavirus/policy-responses/food-supply-chains-and-covid-19-impacts-and-policy-lessons-71b57aea/

Oxfam. (2020). The hunger virus: How COVID-19 is fuelling hunger in a hungry world. Retrieved from Oxfam website: https://oxfamilibrary.openrepository.com/bitstream/handle/10546/621023/mb-the-hunger-virus-090720-en.pdf

Parkin, B., & Rodrigues , A. (2020, May 9). India's lockdown puts squeeze on mango harvest. *The Financial Times*. Retrieved from https://www.ft.com/content/5fce816c-ecb1-406a-adf8-fa4fd8f11029

Pem, S. (2020, 26 July 2020). Nu 200 M for Agriculture Economic Contingency Plan. *BBS*. Retrieved from http://www.bbs.bt/news/?p=135040

Power, M., Doherty, B., Pybus, K., & Pickett, K. (2020). How COVID-19 has exposed inequalities in the UK food system: The case of UK food and poverty. *Emerald Open Research, 2*, 11. doi:10.35241/emeraldopenres.13539.2

Ragasa, C., Amewu, S., & Asante, S. (2021). Immediate impacts of COVID-19 on the aquaculture value chain in Ghana. *GSSP Working Paper 54*. doi:10.2499/p15738coll2.134374

Reardon, T., & Swinnen, J. (2020). COVID-19 and resilience innovations in food supply chains. In J. Swinnen & J. McDermott (Eds.), COVID-19 and Global Food Security

(pp. 132-136). Washington, DC: International Food Policy Research Institute (IFPRI). Retrieved from https://www.ifpri.org/publication/covid-19-and-resilience-innovations-food-supply-chains. doi:10.2499/p15738coll2.133762_30

Roussi, A. (2020, June 4). Kenya Farmers Face Uncertain Future as COVID-19 Cuts Exports to EU. *Financial Times*. Retrieved from https://www.ft.com/content/05284de8-c19f-46de-9fe7-482689be364b

RSD. (2021). *Agriculture Statistics 2020*. Thimphu: Renewable Natural Resources Statistics Division (RSD), Ministry of Agriculture and Forests, Royal Government of Bhutan

Schmidt, E., Dorosh, P., & Gilbert, R. (2021). Impacts of COVID-19 induced income and rice price shocks on household welfare in Papua New Guinea: Household model estimates. *Agricultural Economics, 52*(3), 391-406. doi:https://doi.org/10.1111/agec.12625

Swinnen, J., & Vos, R. (2021). COVID-19 and impacts on global food systems and household welfare: Introduction to a special issue. *Agricultural Economics, 52*(3), 365-374. doi:https://doi.org/10.1111/agec.12623

UN. (2020). COVID-19 dampens the initially positive shrimp forecast for 2020. *GLOBEFISH - Information and Analysis on World Fish Trade*. Retrieved from Food and Agriculture Organization (FAO) website: https://www.fao.org/in-action/globefish/market-reports/resource-detail/en/c/1296667/

Van Hoyweghen, K., Fabry, A., Feyaerts, H., Wade, I., & Maertens, M. (2021). Resilience of global and local value chains to the Covid-19 pandemic: Survey evidence from vegetable value chains in Senegal. *Agricultural Economics, 52*(3), 423-440. doi:https://doi.org/10.1111/agec.12627

Wang, H., Dill, S.-E., Zhou, H., Ma, Y., Xue, H., Sylvia, S., . . . Rozelle, S. (2021). Health, economic, and social implications of COVID-19 for China's rural population. *Agricultural Economics, 52*(3), 495-504. doi:https://doi.org/10.1111/agec.12630

WHO. (2020a). *2019 Novel Coronavirus (2019-nCoV): Strategic preparedness and response plan*. Geneva: World Health Organization (WHO).

WHO. (2020b). WHO Coronavirus (COVID-19) Dashboard. Retrieved July 9, 2021, from World Health Organization (WHO) https://covid19.who.int/

Yuden, K. (2020a, 4 April 2020). Chilli prices soar amid panic buying. *The Bhutanese*. Retrieved from https://thebhutanese.bt/chilli-prices-soar-amid-panic-buying/

Yuden, K. (2020b, 9 December 2020). Very poor marketing, distribution and post-harvest management: Agriculture minister. *The Bhutanese*. Retrieved from https://thebhutanese.bt/very-poor-marketing-distribution-and-post-harvest-management-agriculture-minister/

Effect of Different Pruning Systems on Yield and Quality of Tomato Grown Under Greenhouse

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ABSTRACT

An experiment was conducted to assess the effect of different pruning systems on the yield and quality of tomatoes grown under greenhouse at the National Centre for Organic Agriculture, Yusipang for the growing period of March to November 2019. The experiment was conducted with a randomized complete block design with a single factor at three different levels viz. single leader system (T1), double leader system (T2), and unpruned plants (T3) which were treated as the control for the experiment. All the leaves and axillary shoots below the first flower cluster were pruned off. For treatment T1, only the main stem was encouraged to grow. As for the treatment T2, the sucker growing just below the cluster was also encouraged to grow along with the main stem. This sucker served as the second leader. No pruning of leaves or axillary shoots was carried out in the control plot (T3) at all times. The plants that were pruned started fruiting and maturing earlier than that of unpruned plants. It was also observed that the vegetative and reproductive growth of plants was lengthened by pruning. Although the plants pruned into a single leader system yielded higher than the double leader and the unpruned plants, no statistically significant differences were observed amongst the means of the total yield. The difference in the individual fruit weight, length and diameter amongst the treatments were also found to be statistically not significant. The findings from this research suggest that although pruning gives a higher yield than control there is no significant difference in the yield of tomatoes grown in greenhouse till the 6th harvest. Pruning the plants to a single leader system proved to improve the fruit quality substantially for tomatoes grown in the greenhouse.

Keywords: *Tomato Pruning; Yield; Fruit Quality*

1. Introduction

Tomato (*Solanum lycopersicon*) cultivars are annuals belonging to the Solanaceae family and are also sometimes grown as biennials. Tomatoes are among the most important and popularly grown vegetables in terms of economic, nutritional, and culinary values. The flowering habit of tomatoes ranges from highly indeterminate to highly determinate. Owing to its ability to self-pollinate, the crop is suitable for cultivation in greenhouse.

In Bhutan, tomatoes are grown both under greenhouses and in open fields. Although the total cultivated land under tomato production has gradually increased over the years, the productivity is still considerably low in the country. The Agriculture Statistics 2017 (RSD, 2018a) reports 383 tons of tomatoes produced from 320 acres. This translates to a yield of 1.19 tons from an acre of land which is substantially low. Surprisingly, there is a stark decrease in both the cultivated area as well as production according to RSD (2020). The report highlights a total production of only 232.66 tons from an area of 148.05 acres. This production from within the country was unable to meet the consumer demand, so 2978.46 tons worth Nu.70.14 million of tomatoes was imported from India (RSD, 2018b), and a slightly lesser quantity worth Nu. 62.27 million were imported in 2019 (RSD, 2020).

Various confounding factors influenced by management practices and environmental conditions affect the yield. Ali and Moniruzzaman (2017) stated in their paper on the "Effect of Stem Pruning and Staking on Growth and Yield of Tomato" that yield which is also a genotypic expression is mainly governed by environment and other management factors. As is reflected in RSD (2018b) and RSD (2020) the yield of tomatoes varied amongst dzongkhags. The yield variation could have occurred due to different cultural practices and climatic conditions. The farmers usually grow tomatoes with no or minimal intercultural practices such as stem pruning. The yield, quality, and fruit size of tomatoes are influenced by many factors, including fruit pruning (Saglam & Yazgan, 1999), as well as stem pruning and cultivar selection (Maboko & Du Plooy, 2008). Several reports affirm the benefits of pruning on tomato yields under controlled conditions. Nonetheless, pruning needs and effects on yield depend on cultivar and place. Some literature recommends that tomato plants be pruned to one stem by removing all side shoots (Snyder, 2007). On the other hand, some reported an increase in yield when pruned to two stems (Aung, 1999). Borisoy, Borisova, and Belik (1978) recorded an increase in the yield/area by 10% to 15% when pruned to two stems rather than one.

These literatures confirm that the efficacy of the pruning systems differs by place and cultivar. In Bhutan, no such studies were carried out and farmers usually grow tomatoes without any pruning or management practices. Therefore, this experimental trial was conducted to determine the effect of pruning systems on the yield and quality of indeterminate tomatoes (Cosmic cultivar) under protected conditions in the temperate agroecology of Bhutan.

2. Materials and Method

This experimental research was conducted from March to October 2019 at the National Centre for Organic Agriculture (NCOA), Yusipang. The trial was conducted in a 20 x 5m² greenhouse. Cosmic hybrid, which is an indeterminate variety was used as the test material. The trial was laid out in a Randomized Complete Blocking Design (RCBD) with three replications and three treatments. The three treatments comprised different pruning systems: single leader system (one stem), double leader system (two stems), and unpruned (with multiple leaders). Each replication block measured 16.5 m in length and 1.2m in width comprising randomly distributed treatment plots of sizes 6.6 m². A spacing of 50 cm between each plot was kept while maintaining a spacing of 40 cm between two blocks. The plant to plant and row to row spacing was maintained at 50 cm; each treatment plot accommodated 24 plants.

Seeds were sown on 7th March 2019 under a polytunnel. The seedlings were transplanted after 47 days of sowing on 23rd April. After a month of transplanting all the plants were trained on strings tied down vertically from an overhead horizontal trellis placed at a height of 2m. Undesirable lateral branches and axillary shoots were pruned off depending on the treatments. For treatment 1, all the leaves and axillary shoots below the first flower cluster were pruned off as a result of which a single vine-like leader with no side shoots developed. As for treatment 2, all the leaves and suckers on the main stem were removed up to the first flower cluster except for the sucker growing just below the cluster. This sucker was encouraged to grow as the second leader. No pruning of leaves or axillary shoots was carried out in the control plot at all times. Irrigation and other intercultural operations were carried out as and when required. During the whole period of the trial, suckers were clipped off in treatment 1 and treatment 2 whenever required.

The first harvest was carried out on 26th July, 99 days after transplanting. Ten plants were randomly selected from each treatment plot and marked for data collection at each harvest. During each harvest, the individual fruit weight, fruit diameter, and fruit length were measured from 10 randomly selected fruits from the tagged plants. The total yield of each tagged plant and the treatment plot was also recorded during each harvest. Data were recorded from the first harvest to the sixth harvest. The harvest period spanned for approximately 3 months.

Although more than six harvests have been carried out, data was collected for six harvests only. This was due to the lack of adequate samples from control plots after the sixth harvest. Yield and yield parameters like fruit weight, length, and diameter were treated as dependent variables

while the three treatments were treated as independent variables. The significance in difference between group means was evaluated using ANOVA while LSD *post hoc* test was conducted for multiple means comparison. Data were analyzed using Statistical Package for Social Sciences (SPSS). Data representation and graphs were carried out using Microsoft Excel.

3. Results and Discussion

3.1 Yield

3.1.1 Yield trend

There are considerable differences between yields of different treatments during all harvests except the third harvest where the differences appear negligible (Figure 1). From the total of six harvests that were assessed, plants pruned into single leader system yielded more than the other two systems for four harvests (Figure 1). During the fourth and sixth harvest, the unpruned plants with multiple leaders (control) yielded higher than the other two treatments. From Figure 1, it can be inferred that it is either the single leader system or the unpruned plants that yielded higher. The plants pruned to double leader system yielded slightly higher than unpruned multiple leader plants for three harvests while yielding lower for the other three harvests.
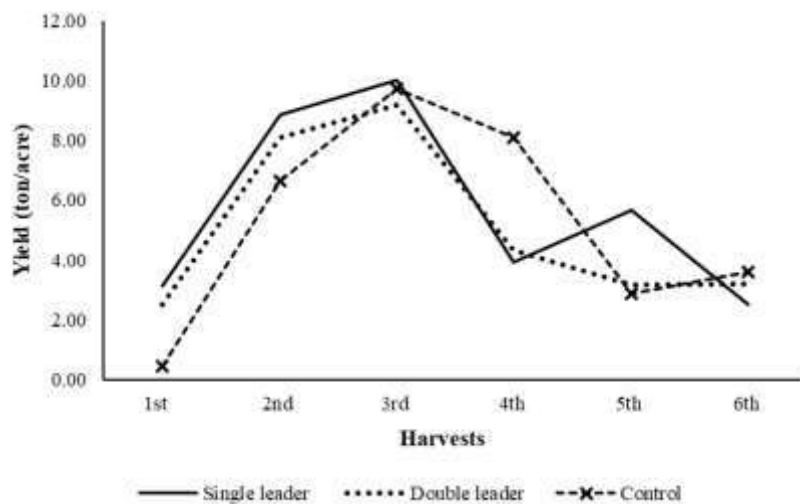

Figure 1. Yield /harvest from each treatment

The yield from pruned plants (single leader and double leader) was significantly higher than the yield from unpruned plants during the first and second harvests although there was no significant difference between the yields of single leader and double leader system plants (Table 1). A similar preliminary study conducted in 2018, published in *Sanam Drupdrey*

(MoAF 2020), revealed that the productivity of the plants grown with plastic mulching and pruned to single leader system gave the highest yield.

Table 1. Tomato yield from the three treatments during each harvest

Treatments	1st Harvest	2nd Harvest	3rd Harvest	4th Harvest	5th Harvest	6th Harvest
Single Leader	3.15±0.35[a]	8.86±0.42[a]	10.02±2.78[a]	3.93±1.93[a]	5.66±2.38[a]	2.52±0.57[a]
Double Leader	2.52±0.83[a]	8.08±0.82[a]	9.17±3.56[a]	4.33±2.19[a]	3.15±2.23[a]	3.19±0.14[a]
Control	0.44±0.11[b]	6.64±0.55[b]	9.73±2.29[a]	8.09±2.18[a]	2.88±0.74[a]	3.62±0.48[a]
P-value	0.002	0.013	0.938	0.095	0.230	0.056

Values with different letters are significantly different according to LSD test at $P<0.01$. The yield means are in tons per acre.

The pruned plants started bearing fruits earlier than the unpruned plants. Apart from that, it was observed that the fruits in pruned plants started maturing earlier compared to those in unpruned plants. This onset of early fruit-bearing and maturity, according to Preece and Read (2005), can be the probable reason for the yields from the two systems being significantly higher than that of non-pruned plants during the initial harvests. Preece and Read (2005) further supplemented that pruning off excessive vegetative growth increases photosynthetic efficiency which results in earlier fruit maturity. This is in agreement with the findings of Mbonihankuye, Kusolwa, and Msogoya (2012) who observed that the fruits pruned into single-stemmed plants matured earlier than those with more stems. In a similar study carried out in Bangladesh, Ali and Moniruzzaman (2017) also observed that pruned plants matured earlier (79.27 days) than unpruned plants (90.27days).

Apart from delayed fruit maturity, it was also observed that the unpruned plants bore fruits later than the pruned plants. Attributing to this, Goda, Mohamed, Helaly, and El-Zeiny (2014) reported a higher early yield per plot due to the pruning treatments. Davis and Estes (1993) reasoned that fruiting is delayed in unpruned plants due to carbohydrates being partitioned to vegetative growth instead of reproductive growth for a longer period. Further, Mbonihankuye et al. (2012) stated that in addition to efficient carbohydrate partitioning, pruning also accelerates an increase in photosynthetic efficiency which results in earlier fruiting and maturity. On the other hand, yields from unpruned plants were significantly higher than that of single leader plants during the fourth and sixth harvests (Table 1). This is an indication that earlier fruit-bearing and maturity contributed substantially to the initial yields of pruned plants.

The result presented in Table 1 is in accordance with a report by Mondal, Puteh, and Razzaque (2016), which explains that single leader plants resulted in higher yield due to earlier fruit-bearing and maturity owing to increased reproductive efficiency caused by pruning. The study by Mbonihankuye et al. (2012) revealed that pruned and staked tomato plants produce flowers two to three weeks earlier than non-pruned plants, resulting in earlier fruit set and maturity.

Though data on the number of flowers were not collected, it was observed that the number of flowers borne by plants pruned to single stems was comparatively lesser than the plants with two stems and no pruning. This is in agreement with Mbinga (1983) who also found that the more severe the pruning, the lower the number of flowers per plant but the fruits were bigger. This led to the single leader system bearing comparatively lesser numbers but heavier and larger fruits. Single stem plants produced the highest fruit set followed by two stem and non-pruned plants according to Mbonihankuye et al. (2012). This is attributed to the improved fruit formation as a result of a better leaf-fruit ratio and correspondingly lesser competition in pruned plants. Ara, Bashar, Begum, and Kakon (2007) found the same results where they observed that the highest proportion of flowers formed fruits under a single stem system.

With the sixth observed harvest, the reproductive period of unpruned plants ended earlier than pruned plants. The sixth harvest was carried out on 18th September, 2019. Two more subsequent harvests that extended till 22nd October, 2019 were carried out in the single and double leader plants after the sixth harvest. It is indicative from the late onset of fruits and earlier cessation of the reproductive phase, that the reproductive period of unpruned plants is shorter by a month than pruned plants. The unlimited vegetative growth in unpruned plants can be a plausible cause for the short reproductive period. When the vegetative growth is excessive, the available assimilates for the reproductive growth is decreased which results in a shortened reproductive life of plants. Mbonihankuye et al. (2012) also reported a similar inference where the increase in available assimilates for fruit set will lead to an increase in the length of both the vegetative and productive periods. Mangal and Jasim (1987) showed that pruning of axillary shoots help in diverting the flow of nutrients towards the apical growing point. This improves plant growth which ultimately enhances the assimilation of materials like carbohydrates and proteins resulting in higher fruit yield and a longer reproductive phase. To supplement further, Hesami, Khorami, and Hosseini (2012) wrote that the lengthened growth of tomato plants may be attributed to the increase in the availability of nutrients, water, and light to plants enabled by pruning.

3.1.2 Total Yield

The overall yield produced by single leader plants is higher than the yields from other two treatments (Table 2).

Table 2. Yield/acre

Treatments	Yield (tons/acre)
Single Leader	35.68 ± 5.17^a
Double Leader	32.88 ± 3.10^a
Control	32.25 ± 2.07^a
P-value	*0.517*

Although a difference in the yield was observed between single leader and the other two treatments, the difference is not found to be statistically significant (Table 2). Similar studies conducted in other places also reported higher yields from pruned plants than from unpruned plants. However, these studies observed plants pruned into two stems yielding higher than plants pruned into single stem unlike the current study where single stemmed plants yielded higher. For instance, from similar research conducted in Dhaka by Sultana, Dilruba, Parveen, Kulsum, and Parvin (2016) a minimum yield (33.97kg/plot) from plants pruned to one stem and the highest yield (36.57 kg/plot) from double leader plants was reported. An experiment carried out in Bangladesh by Ali and Moniruzzaman (2017) also exhibited a higher yield (29.57 tons/ac) from the double leader system followed by triple leader system (27.99 tons/ac) with the lowest yield from single leader system (24 tons /ac). Another study carried out by Ara et al. (2007) in Bangladesh reported the highest yield from two stem pruning (38.35 tons /ac) and the lowest from no pruning (26.14 tons /ac).

Though no significant difference was observed from this study regarding total yield, the noticeable superiority of the single leader plants over the other two treatments is probable due to the subsequent reason. The other two treatments yielded lower than single leader plants due to competition between the stems within plants for water and nutrients, as well as root system limitations to cope with water and nutrient demand (Mourão, Brito, Moura, Ferreira, & Costa, 2017). As is indicative from Table 2, this resulted in single leader plants producing larger fruits and heavier fruits which ultimately increased the overall yield from plants pruned to a single stem. Nganga (1984, as cited in Mbonihankuye et al., 2012) reported a similar finding in his experiment that pruning resulted in a higher flower-fruit ratio in general, and it is suggested that a higher fruit: leaf ratio achieved on fewer leaves through pruning may enhance fruit yield production in a plant Similarly, Mbonihankuye et al. (2012) based on the study by Mnzava

(1984) argue that pruning to a single stem had the effect of increasing fruit set. Thus, a higher yield from the pruned plants during the initial stages eventually increased the overall yield of the single stemmed plants.

3.2 Fruit Quality

To assess fruit quality, parameters like fruit weight, length and diameter were taken into consideration.

3.2.1 Fruit Weight

Regarding fruit characteristics, it can be gathered from Table 3 that single leader plants produced bigger fruits that weighed higher than the ones from double leader and multiple leaders or unpruned (control) plants. However, the differences were not significant between the treatments in terms of individual fruit weight (Table 3). The comparatively higher fruit weight in plants pruned to single stem corroborates with the findings of Ara et al. (2007) who reported the highest fruit weight from plants pruned to one stem.

In a similar study conducted in Bangladesh by Ali and Moniruzzaman (2017), an observation that corroborates this result was made. They reported a maximum individual fruit weight (108.40g) from one stemmed plant (combined with staking) whereas the minimum fruit weight of 69.13g was observed in unpruned plants (with staking).

Hence, it can be inferred from these observations that pruning takes a vital part in enhancing the fruit size and weight. It is unmistakable that pruning a plant into a single stem reduces the number of potential fruiting branches, thus leading to a reduced number of fruits on a plant. This reduction in the number of fruits in single leader plants conceivably reduced the competition between fruits for assimilates which resulted in comparatively heavier fruits. Hence, it can be understood that the significantly heavier fruits compensated for the lesser number of fruits per plant in single leader plants. An observation made by Veliath and Ferguson (1972) validates this statement where it was observed that an increase in the total number of flowers and fruits has been shown to increase the competition for photosynthates which ultimately led to a decrease in fruit size and weight. The improved reproductive efficiency due to reduced competition and increased assimilates translocation to the sink (fruit) resulted in larger and good quality fruits in pruned plants than in unpruned plants (Mondal et al., 2016).

Apart from increased fruit size, as a response to the reduced fruit loads, fruit quality is also improved (Hesami et al., 2012). Muhammad and Singh (2007) also elucidated that unpruned

plants produced smaller sized fruits that are of inferior quality because of the continuous partition of carbohydrates between vegetative and reproductive growth. Thus, results from research conducted in various places prove that proper balance between vegetative and reproductive growth which is an outcome of pruning could improve fruit quantity and quality (Arzani, Bahadori, & Piri, 2009).

3.2.2 Fruit Length

There are no apparent significant differences in the fruit length between all the treatments (Table 3), though plants pruned to single leader yielded a comparatively higher length of fruits than the two other treatments.

3.2.3 Fruit Diameter

In terms of individual fruit diameter, plants pruned to a single stem produced fruits of comparatively higher diameter followed by plants pruned to double and multiple leaders. The differences were found to be non-significant between the treatments, however.

Table 3. Fruit quality characteristics

Treatments	Fruit weight	Fruit length	Fruit Diameter
Single Leader	83.21 ± 19.85^a	57.25 ± 5.56^a	49.77 ± 4.64^a
Double Leader	74.51 ± 15.46^a	55.82 ± 4.34^a	47.80 ± 3.97^a
Control	69.45 ± 21.69^a	54.20 ± 6.40^a	46.29 ± 5.23^a
P- value	*0.104*	*0.260*	*0.088*

As per the Agricultural and Processed Food Products Export Development Authority (APEDA)(APEDA, 2017), India, tomatoes from all three treatments fall under the small category (fruit weight = <100g) for the international market. In the case of size, single leader and double leader plants produced tomatoes that fall under the size code 4 (Diameter: 47-56mm) while those from multiple leader plots fall under size code 3 (Diameter:40-46mm). According to these standards, all three treatments produced fruits that fulfilled the international market standards.

In addition, it was observed that unpruned plants were severely susceptible to blight. Similarly, according to Kanyomeka and Shivute (2005), tomato plants that were not pruned were attacked by early blight disease while the pruned ones were not affected at all. This can be attributed to the dense vegetative growth preventing efficient air movement and light penetration around the plants, thus making it favourable for the fungus to thrive and spread profusely. In conjunction with the observations made, a paper on Pruning and Staking Tomatoes by Chen and Lal (1999)

states that Pruning (removal of side shoots and lower shoots) allows for efficient air circulation, thereby reducing the incidence of blight.

4. Conclusion

Although the pruned plants produced higher yields compared to unpruned, the difference is not statistically significant till the 6th harvest. Thus, till the 6th harvest, there is no significant effect of pruning on the yield of tomatoes as per this study. However, pruned plants produced comparatively bigger and heavier fruits, thus improving quality in terms of size and weight. In addition to enhancing the fruit quality, the results indicate that pruning enables earlier fruit set and maturity, and lengthens both the vegetative and reproductive growths of tomato plants.

Overall, this study indicates that pruning contributes to a slight increase in the yield compared to non-pruning, and it significantly enhances fruit quality and extends the harvest period.

References

Ali, M. K., & Moniruzzaman, M. (2017). Effect of Stem Pruning and Staking on Growth and Yield of Tomato (*Lycopersicon esculentum* L.). *Journal of Agroecology and Natural Resource Management, 4*, 1-4.

APEDA. (2017). Tomato. http://apeda.in/agriexchange/Market%20Profile/one/TOMATO. aspx

Ara, N., Bashar, M. K., Begum, S., & Kakon, S. S. (2007). Effect of spacing and stem pruning on the growth and yield of tomato. *International journal of sustainable crop production, 2*(3), 35-39.

Arzani, K., Bahadori, F., & Piri, S. (2009). Paclobutrazol reduces vegetative growth and enhances flowering and fruiting of mature 'JH Hale'and 'Red Skin'peach trees. *Horticulture Environment and Biotechnology, 50*(2), 84-93.

Aung, M. (1999). *Effect of pruning and spacing on performance of fresh market tomato*. Taiwan: AVRDC: The World Vegetable Centre.

Borisoy, V. Y., Borisova, R. L., & Belik, V. T. (1978). The dependence of tomato yield on spacing and plant training. *Puti Povysheniya Urozhainosti Plodov i Ovoshchnykh Kul'tur*, 54-59.

Chen, J. T., & Lal, G. (1999). *Pruning and staking tomatoes*. Tainan, Taiwang: AVRDC.

Goda, Y., Mohamed, A. A., Helaly, A. A., & El-Zeiny, O. A. H. (2014). Effect of shoot pruning on growth, yield, and fruit quality of husk tomato (*Physalis pubescens* L.). *Journal of American Science, 10*(1), 5-10.

Hesami, A., Khorami, S. S., & Hosseini, S. S. (2012). Effect of shoot pruning and flower thinning on quality and quantity of semi-determinate tomato (*Lycopersicon esculentum* Mill.). *Notulae Scientia Biologicae, 4*(1), 108-111. doi:https://doi.org/10.15835/nsb417179

Kanyomeka, L., & Shivute, B. (2005). Influence of pruning on tomato production under controlled environments. *Agricultura Tropica Et Subtropica, 38*(2), 79-83.

Maboko, M. M., & Du Plooy, C. P. (2008). Effect of pruning on yield and quality of hydroponically grown cherry tomato (*Lycopersicon esculentum*). *South African Journal of Plant and Soil, 25*(3), 178-181.

Mangal, J. L., & Jasim, A. M. (1987). Response of tomato varieties to pruning and plant spacing under plastic house. *Haryana Journal of Horticultural Science, 16*(3-4), 248-252.

Mbinga, E. W. (1983). *Pruning and spacing effect on tomato var*. Thailand: Asian Regional Centre (ARC).

Mbonihankuye, C., Kusolwa, P., & Msogoya, T. J. (2012). *Assessment of the effect of pruning systems on plant developmental cycle-yield and quality of selected indeterminate tomato lines.* Paper presented at the 2nd All Africa Horticulture Congress, Kruger National Park (South Africa).

Mnzava, N. A. (1984). *Influence of cluster and spacing on set, growth, yield and quality of early fruit of the West Virginia 63 tomato (Lycopersicon esculentum Mill).* (M.Sc. Dissertation), Sokoine University of Agriculture, Morogo, Tanzania.

Mondal, M. M. A., Puteh, A. B., & Razzaque, A. H. M. (2016). Debranching improves morpho-physiological characters, fruit quality and yield of tomato. *Pakistan J. Bot, 46*, 2249-2253.

Mourão, I., Brito, L. M., Moura, L., Ferreira, M. E., & Costa, S. R. (2017). The effect of pruning systems on yield and fruit quality of grafted tomato. *Horticultura Brasileira, 35*, 247-251. doi:https://doi.org/10.1590/S0102-053620170215

Muhammad, A., & Singh, A. (2007). Yield of tomato as influenced by training and pruning in the Sudan savanna of Nigeria. *Journal of Plant Sciences, 2*(3), 310-317.

Preece, J. E., & Read, P. E. (2005). *The Biology of Horticulture* (2nd ed.). New York: John Wiley and Sons.

RSD. (2018a). *Agriculture Statistics 2017*. Thimphu: Renewable Natural Resources Statistics Division (RSD), Ministry of Agriculture and Forests, Royal Government of Bhutan

RSD. (2018b). *Bhutan RNR Statistics 2017*. Thimphu: Renewable Natural Resources Statistics Division (RSD), Ministry of Agriculture and Forests, Royal Government of Bhutan.

RSD. (2020). *Agriculture Statistics 2019*. Thimphu: Renewable Natural Resources Statistics Division (RSD), Ministry of Agriculture and Forests, Royal Government of Bhutan

Saglam, N., & Yazgan, A. (1999). Effect of fruit number per truss on yield and quality in tomato. In Y. Tüzel, S.W. Burrage, B.J. Bailey, A.R. Smith, & O. Tuncay (Eds.), *International Symposium Greenhouse Management for Better Yield & Quality in Mild Winter Climates* (pp. 261-264). Antalya, Turkey Acta Horticultarae.

Snyder, R. G. (2007). *Greenhouse Tomato Handbook*. Crystal Springs, MS: Mississippi State University Extension.

Sultana, R., Dilruba, S., Parveen, K., Kulsum, U., & Parvin, N. (2016). Effect of Pruning on Growth and Yield of Tomato *(Lycopersiconesculentum* M). *European International Journal of Science and Technology, 5*(9).

Veliath, J. A., & Ferguson, A. C. (1972). effect of deblossoming on fruit size, yield, and earliness in tomato. *HortScience, 7*, 278 - 279.

Management of Litchi Fruit and Shoot Borer (*Conomorpha sinensis*) in Three Litchi Varieties in Samtenling, Bhutan

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ABSTRACT

In Bhutan, litchi production is constrained by insect pest damage. Among insect pests, the litchi fruit and shoot borer (Conopomorpha sinensis Bradley) is an emerging pest of economic importance in the subtropical regions of Bhutan. Up until now, there is documented research on its biology and management in Bhutan. Therefore, this study was undertaken to study the efficacy of two different management methods against litchi fruit borer infestation in three litchi varieties (Bhur litchi 1, Bhur litchi 2 & Hong houy). The management methods consisted of three treatments; T1-Azadirachtin followed by Imidacloprid, Azadirachtin and Cypermethrin; T2-Azadirachtin followed by Imidacloprid, Azadirachtin and Azadirachtin; T3-Untreated control. The result showed that the fruit infestation at harvest was significantly lower in management method T1 (34.74%) as compared to T2 (78.07%) and untreated control (89.98%). In terms of the variety, the lowest fruit infestation at harvest was observed for Hong houy (56.02%) as compared to Bhur Litchi 1 (73.28%) and Bhur Litchi 2 (73.45%). There were no significant differences in fruit drop percent and percent infestation of dropped fruits between the treatments and varieties tested. Heavy fruit drop was observed in the third week, due to unknown reasons, which then decreased over the fruit development stages. Fruit borer infestation seems to increase rapidly after the fifth and eighth weeks indicating that most of the fruit borer damage occurs at the fruit maturity stage after fruit colour break. Therefore, the last application of cypermethrin 10% EC at the fruit colour break stage was found necessary to reduce fruit infestation during this vulnerable pest infestation period besides scheduled application of Azadirachtin and Imidacloprid at the early fruit development period.

Keywords: *Litchi fruit and shoot borer; Emerging pest; Pest management*

1. Introduction

Litchi (*Litchi chinensis Sonn.*) is an important subtropical evergreen fruit crop belonging to the family Sapindaceae. Litchi fruits are prized on the world market for their flavour, semi-translucent white aril, and attractive red skin (Yang, Wang, Prasad, Pan et al., 2011). About 95% of litchi production is in Southeast Asia, with China, Vietnam, Thailand, India, Bangladesh, and Nepal being the most important producers in the world (Menzel & Waite,

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2005). It contributes significantly to the livelihood of several million people throughout Southeast Asia (Menzel & Waite, 2005).

In Bhutan, litchi is grown in fifteen districts with a total production of 384.69 metric tons (RSD, 2020). Sarpang, Samtse, Samdrup Jongkhar, Dagana and Chhukha are the top five producers of litchi, contributing about 95% of the total production. Currently, there are two released varieties (Bhur Litchi 1 and Bhur Litchi 2) and 3 notified varieties (Bhur Selection 1, Shahi and Early Bedena) in Bhutan (DoA, 2021). Although litchi production is gaining momentum in the subtropics of Bhutan (BBSC, 2012; Rai, 2017), the incidence of insect pests and diseases have become a major constrain while growing this crop.

Litchi fruit and shoot borer (*Conopomorpha sinensis* as described by Bradley (1986) is becoming one of the major pests of economic importance in Bhutan (ARDC, 2018). In India, litchi fruit borer damage is estimated between 48% to 74% in West Bengal (Chakraborti & Samanta, 2005), 13.6% to 64.9% in Himachal Pradesh and 24% to 32% in Bihar (Alam, Patra, & Samanta, 2019). The insect lays eggs on the calyx end of the fruit and the developing larva bores into the fruit and feeds on it. The larvae also damage newly emerged shoots from September to October resulting in the failure of the shoots to bloom. Further, it punctures the fruits from April to May resulting in severe losses through fruit drop and deteriorates the fruit quality due to the presence of larva feeding in fruit and frass in funicle while eating (Kumar, Kumar, & Nath, 2011; Srivastava, Patel, Kumar, Pandey et al., 2017).

According to Meng, Hu, Li, Dai et al. (2018), *C. sinensis* is difficult to manage with chemical control due to its cryptic feeding behaviour and overlapping generations. The vulnerable period between egg hatching and fruit penetration, where the larvae are exposed to chemical treatments outside the fruits, is very short to attain the required control (Schulte, Martin, & Sauerborn, 2007). However, Upadhyay, Aryal, Bhusal, and Chaudhary (2020) tested five insecticides applied at ten-day interval against *C. sinensis* and found that all the insecticides were effective as compared to the control. Schulte et al. (2007) suggested weekly applications of insecticides such as permethrin for effective control of fruit borer. However, heavy reliance on insecticides can lead to several adverse effects such as development of insecticide resistance, outbreak of secondary pests, harmful effects on beneficial organisms and problems of pesticide residue. Kumar et al. (2011) recommend integrated pest management methods such as the use of pheromone trap, biocontrol agents, removal, and destruction of dropped fruits,

prophylactic spray of neem-based insecticides and need-based application of chemical insecticides for controlling litchi fruit and shoot borer infestation.

With limited knowledge about this pest and its management in Bhutan, this study was conducted to evaluate the efficacy of two spray regimes by combining different insecticides in three litchi varieties to effectively manage this pest in litchi crop.

2. Materials and Method

2.1 Experimental site

This experiment was conducted in the litchi germplasm block of the Agriculture Research and Development Centre (ARDC), Samtenling (26°54'14" N, 90°26'20" E), Sarpang during the litchi fruiting season (March to May) of 2021. The site is located at an altitude of 375 meters above sea level (masl) which falls under the "wet sub-tropical" agro-ecological zone. The experiment was laid out in a split-plot design. The main plot consisted of three litchi varieties (Bhur Litchi 1, Bhur Litchi 2 and Hong houy) and each variety had three trees per replication. The subplot consisted of three management methods including untreated control (see Section 2.3 below) which were replicated three times per variety making a total of nine trees per replication. The age of litchi plants used in this study was 15 years with full fruit-bearing potential.

2.2 Treatments

The management methods were: (T1) One foliar application of Azadirachtin 300 ppm (@ 6 mL/L of water) during the time of panicle emergence before flowering (33 days before flowering) followed by second spray with Imidacloprid 20% SL (@1 mL/L of water) at clove sized fruit stage (14 days after flowering (DAF)), third spray of Azadirachtin 300 ppm (@ 6 mL/L of water) when the fruits attained about 50% of the final size (45 DAF) and fourth spray of Cypermethrin 10% EC (@1 mL/L) at fruit colour break stage (61 DAF) as described by Wei et al. (2013); (T2) with a foliar spray of Azadirachtin 300 ppm (@ 6 mL/L of water) during the time of panicle emergence before flowering (33 days before flowering) followed by second spray of Imidacloprid 20% SL (@1 mL/L of water) at clove sized fruit growth stage (14 DAF) followed by third and fourth spray of Azadirachtin 300 ppm (@ 6 mL/L of water) when the fruits attained about 50% of the final size (31 DAF) and at fruit colour break stage (61 DAF); (T3) Untreated control. Each treatment was applied using a separate battery-operated knapsack sprayer (Neptune, NF-708).

Table 2. Description of different insecticides used in the experiment; Source: (IRAC, 2021)

S.N.	Common name	Trade name	Mode of action	Formulation	Dose
1.	Azadirachtin	Neem Baan	UN*	300 ppm	6 mL/L
2.	Imidacloprid	Termitox	Nicotinic Acetylcholine Receptor (NACHR) competitive modulators	20% SL	1 mL/L
3.	Cypermethrin	Cypermethrin	Sodium channel modulator	10% EC	1 mL/L
4.	Wetting agent	Anuvit	-	-	2 mL/L

*Compounds of unknown or uncertain mode of action

2.3 Data collection

Data were collected from all the nine trees selected for each of the treatments making a total of 27 trees. From each tree, 120 fruits were randomly harvested during the fruit maturity stage (27th May) and examined for fruit borer infestation damage. To determine the fruit drop rate and borer infestation over the growing period, all the dropped fruits per tree were collected weekly over the fruit development stages (7 weeks) and dissected with the help of a sharp knife. The fruits were assigned as infested if larvae were present inside the fruit, or if there were the presence of entry and exit holes or insect excreta in the fruits. Percent fruit infestation at harvest, percent fruit drop and percent infestation of dropped fruit were calculated as;

$$\text{Percent fruit infestation at harvest (\%)} = \frac{\text{Number of infested fruit per tree}}{\text{Number of fruits examined per tree (120)}} * 100 \quad \dots\dots\dots (1)$$

$$\text{Percent fruit drop (\%)} = \frac{\text{Number of dropped fruit per tree per week}}{\text{Total number of fruits per tree}} * 100 \quad \dots\dots\dots\dots\dots\dots\dots\dots (2)$$

Where; Total number of fruits per tree was calculated as sum of total fruit bearing at harvest and total number of fruits dropped

$$\text{Dropped fruit infestation percent (\%)} = \frac{\text{Number of infested dropped fruits per week}}{\text{Total number of dropped fruit per week}} * 100 \quad \dots\dots\dots (3)$$

In addition, the length and width of the fruits were measured at each sampling point, using a standard vernier calliper to correlate fruit infestation rate with fruit size over the fruiting season.

2.4 Statistical analysis

The distribution of the variables was determined using the Shapiro-Wilk test and the data on percent fruit drop, percent infestation of dropped fruit and percent fruit infestation at harvest were largely normal and variance were homogenous. The data were subjected to a two-way

ANOVA using the Statistical Tool for Agriculture Research (STAR) (Version: 2.0.1) software package. The relationship between percent fruit drop, dropped fruit infestation percent and fruit size amongst the varieties was tested using Pearson correlation.

3. Results and Discussion

3.1 Fruit drop percent

The result showed no significant difference in percent fruit drop between the three management methods tested on three varieties during the entire fruit development period (Table 2). It was observed that 70% to 78% of the fruits per tree dropped during the entire fruiting season and only 21% to 30% of the fruits were retained on the tree at harvest. Among the three varieties, the lowest percent fruit drop was recorded in Hong houy variety (71.35%) with the lowest fruit drop observed for management method T1 (70.35%). The growth of litchi fruit (length and width) followed a sigmoid curve. A weekly observation showed the highest fruit drop percent (56.8%) at the third week after the fruit set when the fruit attained a size of 1.15 cm in length and 0.61 cm in width (Figure 1). Fruit drop percent decreased to 2.7% by the fourth and fifth week and increased gradually to 7.4% in the sixth week and subsequently decreased towards the last two weeks when the fruits t matured.

Table 3. Fruit drop percent in three varieties of litchi with three treatments during fruiting period in 2021

Factors	Percent fruit drop (%)							Total fruit drop (%)	Fruits retained on tree (%)	Total No. of fruits/ tree
Main Plot (Variety)	Weeks after fruit set (WAFS)									
	3	4	5	6	7	8	9			
Bhur Litchi 1	64.22	5.88	12.35	29.65	4.47	2.26	10.08	78.28	21.72	3500
Bhur Litchi 2	55.32	17.01	14.26	32.57	6.77	2.27	8.24	74.80	25.20	3233
Hong houy	50.82	11.24	19.31	26.75	7.65	2.08	8.20	71.35	28.65	3161
Sub plot (Treatments)										
T1	52.97	2.02	3.47	7.97	1.79	0.46	1.67	70.35	29.65	3011
T2	56.93	2.93	4.36	7.01	1.49	0.49	2.04	75.26	24.74	3570
T3	60.46	3.15	3.44	7.22	1.48	0.59	2.47	78.82	21.18	3314
P>F Variety	0.21	0.12	0.35	0.59	0.14	0.93	0.98	0.13	0.13	0.73
Treatments	0.42	0.56	0.74	0.86	0.85	0.75	0.11	0.06	0.06	0.74
Variety*Treatment	0.17	0.12	0.32	0.21	0.37	0.35	0.25	0.43	0.43	0.15
Std. Error Mean	2.57	0.49	0.59	0.73	0.23	0.07	0.18	1.42	84.62	279.05

3.2 Percent infestation in dropped fruits

There was no significant difference in percent infestation of dropped fruits among the management methods as well as the tested varieties throughout the fruiting season except at the third week after fruit set. In the third week, a significant difference ($P = 0.03$, F = 5.00, df = 2) was observed in percent infestation of dropped fruit between the two management methods and the control. There was zero percent infestation of dropped fruit in both the treatments T1 (00.00 %) and T2 (0.00%), but significantly higher percent infestation of dropped fruits in the untreated control T3 (0.56 %) (Table 3). Dropped fruit infestation was recorded as early as the third week after fruit set (12th April) with an infestation rate of 0.19%. The trend increased gradually following sigmoid curve with the highest infestation rate at fruit maturity (69.09 %) (Figure 1 & 3).
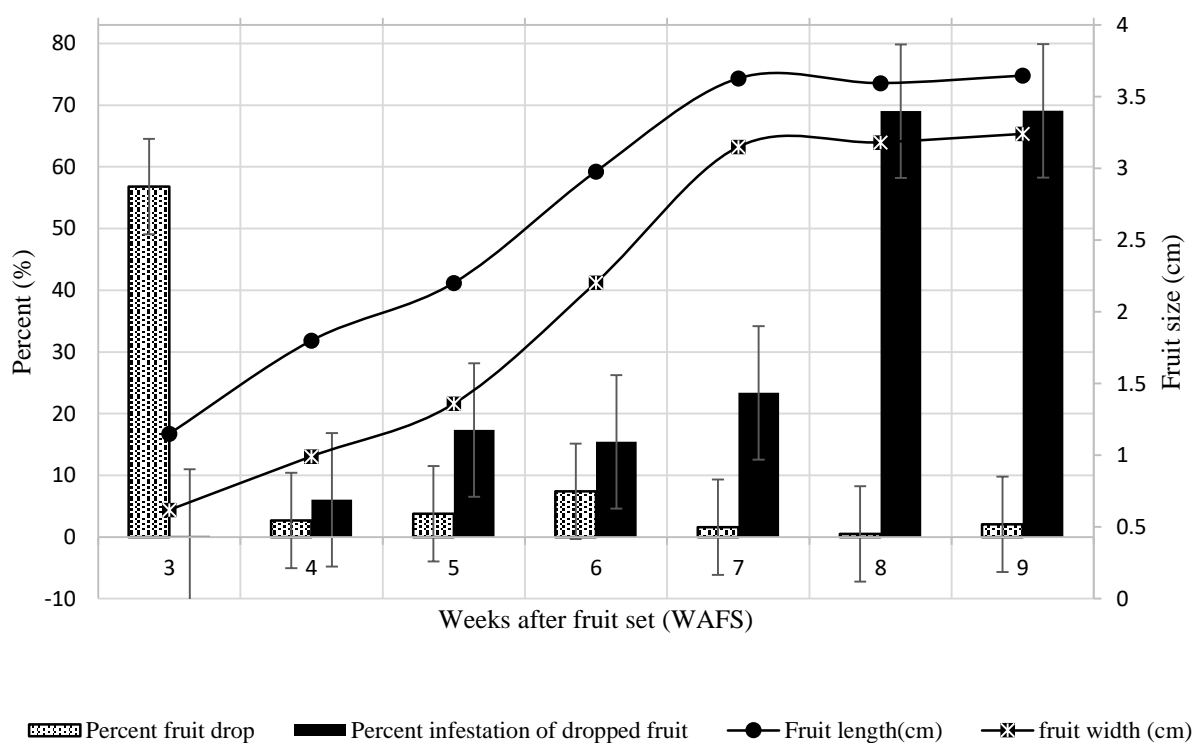

Figure 3. Percent fruit drop (%) and percent infestation of dropped fruits by *C. sinensis* during the fruit development period from 12th April to 24th May 2021

Table 4. Percent infestation of dropped fruit in three varieties of litchi with three treatments during fruiting period of 2021

Factors	Percent infestation of dropped fruit (%)						
	Weeks after fruit set (WAFS)						
Main Plot (Variety)	3	4	5	6	7	8	9
Bhur Litchi 1	0.11	7.55	14.18	18.17	24.72	62.04	63.62
Bhur Litchi 2	0.00	4.55	24.69	15.49	30.68	84.56	73.23
Hong houy	0.44	6.04	13.23	12.63	14.73	60.52	70.42
Sub Plot (Treatments)							
T1	0.00 b	5.59	14.70	14.44	21.63	65.18	72.25
T2	0.00 b	5.02	15.35	17.01	19.99	64.87	66.78
T3	0.56 a	7.53	22.05	14.84	28.51	77.08	68.24
P>F							
Variety	0.145	0.570	0.296	0.504	0.199	0.291	0.757
Treatments	0.026	0.476	0.056	0.841	0.062	0.610	0.864
Variety*Treatments	0.090	0.701	0.023	0.944	0.613	0.530	0.073
Std. Error Mean (±)	0.11	0.83	2.19	1.65	2.22	5.61	4.68

Means with the same letter are not significantly different at $P \leq 0.05$

3.3 Correlation analysis

There was no positive correlation observed between percent fruit drop and fruit size ($r = -0.71$ & $r = -0.62$, $P \leq 0.05$). Similarly, no positive correlation was observed between percent fruit drop and percent dropped fruit infestation level ($r = -0.49$, $P \leq 0.26$). However, there was a strong positive correlation ($r = 0.79$, $P \leq 0.03$ & $r = 0.82$, $P \leq 0.02$) between percent dropped fruit infestation and fruit size (length, width) (Table 4).

Table 5. Pearson correlation analysis between fruit drop rate, percent infestation of dropped fruit and fruit size

	Percent fruit drop (PFD)	Dropped fruit infestation percent (DFIP)	Fruit length (FL)	fruit width (FW)
(PFD)	1	-0.49	-0.71	-0.62
(DFIP)	-0.49	1	0.79*	0.82*
(FL)	-0.71	0.79	1	0.99
(FW)	-0.62	0.82	0.99	1

*Correlation is significant at the 0.05 level (2- tailed)

3.4 Fruit infestation at harvest

Percent fruit infestation at harvest was significantly lower ($P \leq 0.001$, F = 34.06, df = 2) in management method T1 (34.74%) as compared to T2 (78.07%) and untreated control (89.98%). Although no significant difference was observed among the varieties, the lowest fruit infestation percent at harvest was found in variety Hong houy (56.02%) as compared to Bhur Litchi 1 (73.28%) and Bhur Litchi 2 (73.45%) (Figure 2).
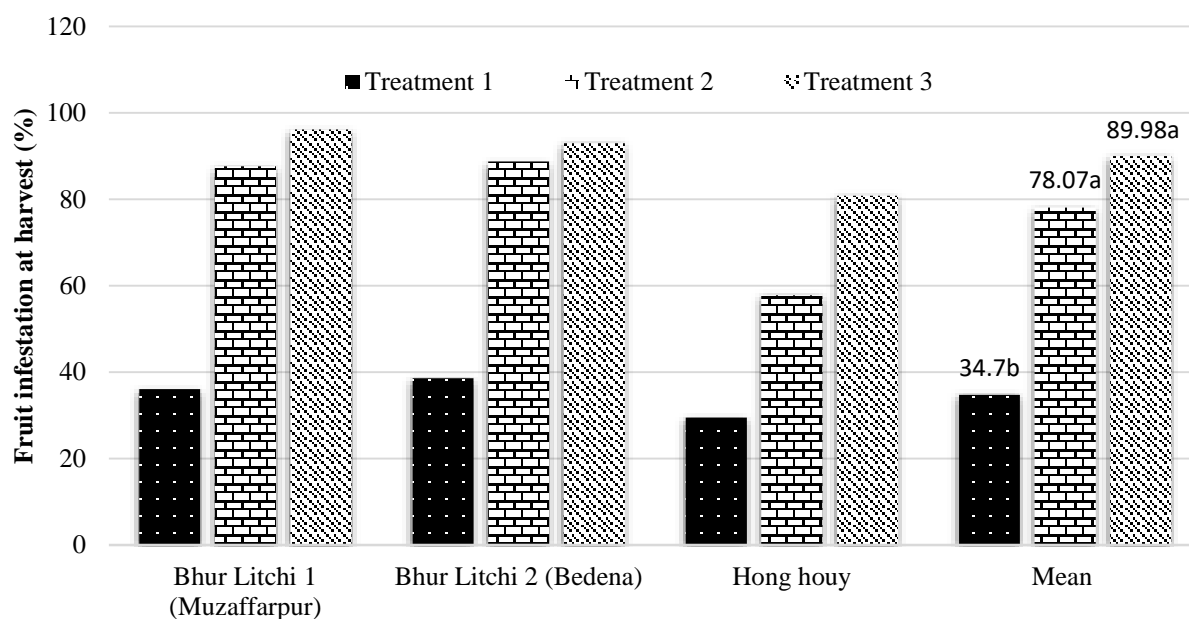

Figure 2. Fruit infestation percent at harvest by *Conopomorpha sinensis* in three varieties of litchi treated with three management methods (Treatments) during the fruiting period in 2021 at ARDC- Samtenling

Table 6. Analysis of variance of the fruit infestation percent at harvest between three varieties tested with three management methods (Treatments)

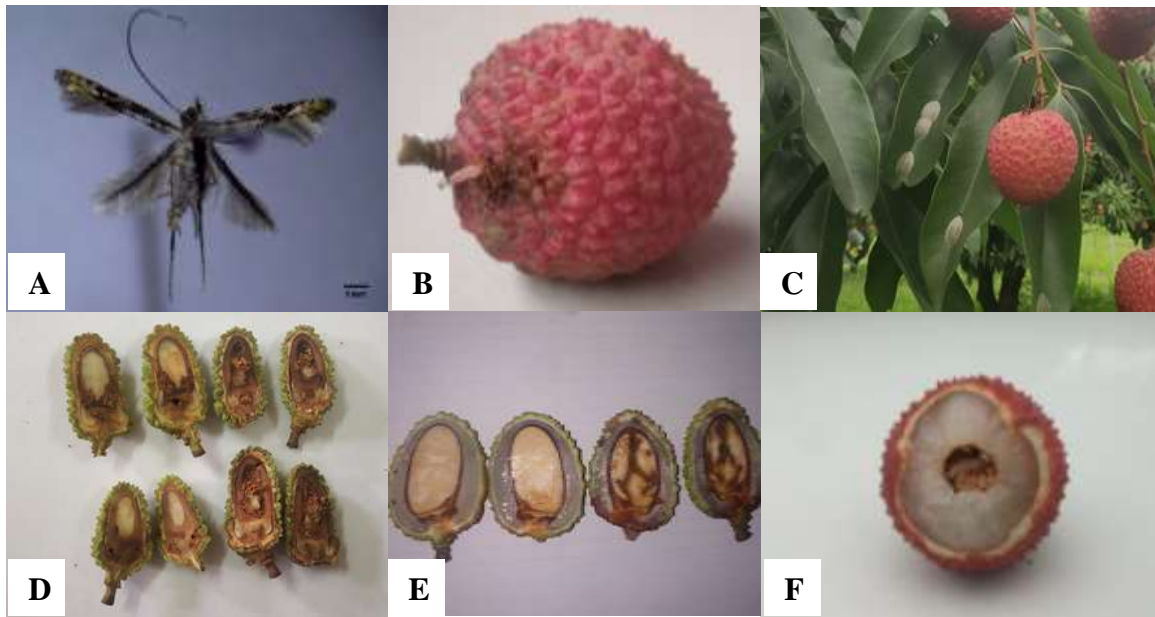| Source of variation		DF	SS	MS	F	p
Main plot	Rep	2	570.56	285.28	0.87	0.485
	Variety	2	1805.20	902.60	2.76	0.177
	Main plot Error	4	1309.95	327.49		
Sub Plot						
	Treatment	2	15232.83	7616.41	34.06	<0.0001
	Variety*Treatment	4	574.76	143.69	0.64	0.643
	Sub plot Error	12	2683.79	223.65		
	Total	26	22177.09			

Figure 3. (A) Adult moth of Litchi Fruit borer, (B) Larva exiting from exit hole at the base of fruit, (C) Pupation on leaves after exiting from the fruit, (D) Fruit infestation at early fruit development stage, (E) Fruit infestation and damage at fruit maturity, (F) Poor fruit quality due to insect feeding frass at the base of fruit

A study by Schulte et al. (2007) demonstrated that litchi fruit infested by fruit borer at an early fruit growth stage shows higher fruit drop. In this study, although heavy fruit drop was observed in the third week after fruit set, fruit borer infestation level was observed to be very low. Studies have shown physiological fruit drop in litchi during the early fruit growth stage (Wei, Zhang, Li, Xie et al., 2013). Therefore, the early fruit drop, as observed in this study, might primarily be some form of physiological fruit drop than that caused by fruit borer infestation.

Research by Ramakrishnaiah, Damodaram, Rai, Rajendra et al. (2017) showed a positive correlation between the number of dropped fruits and fruits infested with *C. sinensis*, suggesting that borer incidences lead to heavy fruit drop. However, in contrast, there was no strong correlation observed between the percent fruit drop and percent infestation of dropped fruit in this study. This suggests that fruit borer incidence might not be the main factor causing fruit drop. Instead, other factors such as physiological fruit drop and infestation by other pests such as fruit borer (*Deudorix epijarbas*) larva, litchi bugs, birds and other abiotic factors might also have contributed to the observed fruit drop.

Ramakrishnaiah et al. (2017) also found that about 48.4% of the dropped fruit was infested by *C. sinensis* and the infestation decreased as the fruit matured. However, in our study the infestation of dropped fruits increased exponentially up to 69% as the fruit matured. A strong

positive correlation between the dropped fruit infestation rate and fruit size was observed which indicates that the percent dropped fruit infestation rate increased with an increase in fruit size. Most of the fruit borer damage occurred at fruit maturity after the fruit colour break stage. Therefore, the results of this study align with the findings of Schulte et al. (2007), where a similar trend between the fruit infestation rate and fruit size was reported.

Schulte et al. (2007) also showed the ineffectiveness of insecticides such as ilorbac, imidacloprid and the three different concentrations of spinosad against *C. sinensis*. This was primarily attributed to the short exposure period between the egg hatching and fruit penetration by the larvae, where the larvae are not fully exposed to the insecticides to attain effective control. Finally, this study found that scheduled and combined application of Azadirachtin, Imidacloprid, Azadirachtin and Cypermethrin were effective in reducing litchi fruit borer infestation.

4. Conclusion

This study reveals the presence of heavy litchi fruit borer infestation regardless of the varieties grown in the sub-tropical region of Sarpang, Bhutan. Based on the result, the management method T1 was effective in managing *C. sinensis* as the lowest fruit infestation at harvest was observed in T1 (34.74%) as compared to T2 (78.07%) and the untreated control (89.98%). In terms of the variety, the lowest fruit infestation at harvest was observed in Hong houy (56.02%) as compared to Bhur Litchi 1 (73.28%) and Bhur Litchi 2 (73.45%).

Although fruit borer infestation in dropped fruit was observed as early as the third week after fruit set, infestation level was found to increase rapidly after the fifth and eighth week indicating that most of the fruit borer damage occurs at the fruit maturity stage after fruit colour break. The initial combined applications of biopesticide (Azadirachtin 300 ppm) and insecticide (Imidacloprid 20% SL) seem to have significantly suppressed the successive generations of litchi fruit borer. Further, the last application of Cypermethrin 10% EC at fruit colour break stage is found necessary to reduce fruit infestation during this vulnerable pest infestation period in addition to the scheduled application of control measures right from the early fruit development period. Further study needs to be carried out on the pest's biology and the effect of these combinations on natural enemies so that such management methods can be effectively used for managing this pest species in litchi.

References

Alam, S. K. F., Patra, B., & Samanta, A. (2019). Evaluation of some new insecticide mixtures for management of litchi fruit borer. *Journal of Entomology Zoology Studies, 7*(1), 1541-1546.

ARDC. (2018). *Annual Report 2017-2018*. Gelephu, Bhutan: Agriculture Research & Development Centre (ARDC), Samtenling, Department of Agriculture, Ministry of Agriculture and Forests.

BBSC. (2012, May 26th). Farmers experiment litchi cultivation. *Bhutan Broadcasting Service Corporation (BBSC)*. Retrieved from http://www.bbs.bt/news/?p=13138

Bradley, J. D. (1986). Identity of the South-East Asian cocoa moth, *Conopomorpha cramerella* (Snellen)(Lepidoptera: Gracillariidae), with descriptions of three allied new species. *Bulletin of Entomological Research, 76*(1), 41-51. doi:10.1017/S000748530001525X

Chakraborti, K., & Samanta, A. (2005). Evaluation of litchi germplasms based on biochemical parameters alongwith incidence of leaf roller (*Platypepla aprobola* Meyr) and fruit borer (*Conopomorpha cramerella* Snellen). *Annals of Plant Protection Sciences, 13*(2), 338-342.

DoA. (2021). *Inventory of Released and De-notified Crops in Bhutan (1988-2021)*. Thimphu, Bhutan: Department of Agriculture, Ministry of Agriculture and Forests, Royal Government of Bhutan.

IRAC. (2021). *IRAC Mode of Action Classification Scheme*: Insecticide Resistance Action Committee (IRAC).

Kumar, V., Kumar, A., & Nath, V. (2011). Emerging pests and diseases of litchi (*Litchi chinensis* Sonn.). *Pest Management in Horticultural Ecosystems, 17*(1), 11-13.

Meng, X., Hu, J., Li, Y., Dai, J., Guo, M., & Ouyang, G. (2018). The preference choices of *Conopomorpha sinensis* Bradley (Lepidoptera: Gracilariidae) for litchi based on its host surface characteristics and volatiles. *Scientific Reports, 8*(1), 1-8. doi:10.1038/s41598-018-20383-4

Menzel, C. M., & Waite, G. K. (2005). *Litchi and longan: botany, production and uses*. Wallingford, UK: CABI Publishing.

Rai, R. (2017, July 8th). Dumseygang farmers try lychee farming. *Kuensel*. Retrieved from https://kuenselonline.com/dumseygang-farmers-try-lychee-farming/

Ramakrishnaiah, J. M. B., Damodaram, K. J. P., Rai, S. P., Rajendra, S. P., Subramaniam, S., Thimmappa, R., & Thimmappa, N. (2017). Occurrence of *Conopomorpha sinensis* Bradley, 1986 (Lepidoptera: Gracillariidae) on litchi (*Litchi chinensis*) in India. *The Pan-Pacific Entomologist, 93*(4), 199-203. doi:10.3956/2017-93.4.199

RSD. (2020). *Agriculture Statistics 2019*. Thimphu: Renewable Natural Resources Statistics Division (RSD), Ministry of Agriculture and Forests, Royal Government of Bhutan.

Schulte, M. J., Martin, K., & Sauerborn, J. (2007). Biology and control of the fruit borer, *Conopomorpha sinensis* Bradley on litchi (*Litchi chinensis* Sonn.) in northern Thailand. *Insect Science, 14*(6), 525-529. doi:10.1111/j.1744-7917.2007.00182.x

Srivastava, K., Patel, R. K., Kumar, A., Pandey, S. D., Reddy, P. V. R., & Nath, V. (2017). Integrated management of litchi fruit and shoot borer (*Conopomorpha sinensis*) using insect growth regulators under subtropics of Bihar. *Indian Journal of Agricultural Sciences, 87*(11), 1515–1518.

Upadhyay, S. K., Aryal, S., Bhusal, B., & Chaudhary, B. (2020). Evaluation of Insecticides for the Management of Litchi Fruit and Shoot Borer. *Journal of Nepal Agricultural Research Council, 6*, 85-91. doi:doi.org/10.3126/jnarc.v6i0.28119

Wei, Y. Z., Zhang, H. N., Li, W. C., Xie, J. H., Wang, Y. C., Liu, L. Q., & Shi, S. Y. (2013). Phenological growth stages of lychee (*Litchi chinensis* Sonn.) using the extended BBCH-scale. *Scientia horticulturae, 161*, 273-277. doi:10.1016/j.scienta.2013.07.017

Yang, B., Wang, H., Prasad, N., Pan, Y., & Jiang, Y. (2011). Use of Litchi (*Litchi sinensis* Sonn.) seeds in health. In V. R. Preedy, R. R. Watson, & V. B. Patel (Eds.), *Nuts and Seeds in Health and Disease Prevention* (pp. 699-703). San Diego: Academic Press.

Automation of Hydroponics System using Open-source Hardware and Software with Remote Monitoring and Control

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ABSTRACT

This study aimed to develop and install an open-source hardware and application software for the automation of different actuators and sensors in the hydroponics system established at ARDC-Wengkhar. A prototype automation system was developed using Raspberry Pi 3 installed with open-source hydroponics application software called Mycodo which acted as a main computing hub for the automation. The automation features included the schedule or timer-based switching of different pumps, conditional switching of the ventilation fans based on temperature/humidity, alarm and notifications via email when certain parameters exceed the normal value, data logging and remote access to the system. The prototype was installed in the existing hydroponics structures containing nutrient film technique, deep water culture and vertical tower. The prototype was found efficient, reliable, useful, affordable and expandable as it offers more flexibility and advanced features for any automated hydroponics system.

Keywords: *Automation; Hydroponics; Mycodo; Open-Source; Raspberry Pi*

1. Introduction

Hydroponics is the process of growing plants in the absence of soil with the help of added nutrient solutions (El-Kazzaz & El-Kazzaz, 2017). Hydroponics has become popular especially in urban areas to grow plants without soil and several studies have shown that plants grown with hydroponics are of high quality and consume fewer resources than traditional growing methods (Kularbphettong, Ampant, & Kongrodj, 2019). In Bhutan, the concept of hydroponics farming was recently introduced by the Department of Agriculture first being established at the Agriculture Research and Development Centre (ARDC) Wengkhar in 2019 and later, different models of hydroponics systems were developed in other research centres (DoA, 2021).

In the hydroponics system, there are several parameters such as air temperature and humidity, lights, water temperature, nutrient EC and pH which are difficult to be controlled or maintained

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precisely by human intervention. These parameters are important for healthy and faster plant growth (DoA, 2021). Therefore, automation is necessary to maintain these parameters within the optimum level to provide ambient conditions for plant growth (Dudwadkar, Das, Suryawanshi, Dolas et al., 2020). Also, it is reported that labour cost is one of the biggest recurring costs for any hydroponics system (DoA, 2021). Automation of hydroponics systems can reduce the labour required for crop production and reduce labour costs. Besides reducing labour costs, automation can also improve the efficient use of inputs such as plant nutrients, water and electricity which further reduces the overhead cost for the operation of the hydroponics system (Sharma, Acharya, Kumar, Singh et al., 2018). Although various types of automation devices for hydroponics systems are available in the market many are expensive, lack local technical expertise for installation, usage and maintenance under Bhutanese conditions. Therefore, a locally designed automation for hydroponics system was developed with the following objectives:

- Develop a prototype of an internet of things (IoT) based automated hydroponics system using open-source resources (hardware and software)
- Deploy and test the prototype for automation of existing hydroponics at the research centre

The basic architecture of the prototype is given in Figure 1. The primary functions of the prototype are to monitor the different parameters of plant growth which is done by different sensors and to control the parameters within the optimum level with the help of actuators present in the system. The secondary functions of the prototype are for remote monitoring, control and data acquisition. All these operations and controls were carried out by Raspberry Pi installed with open-source automation software called Mycodo.
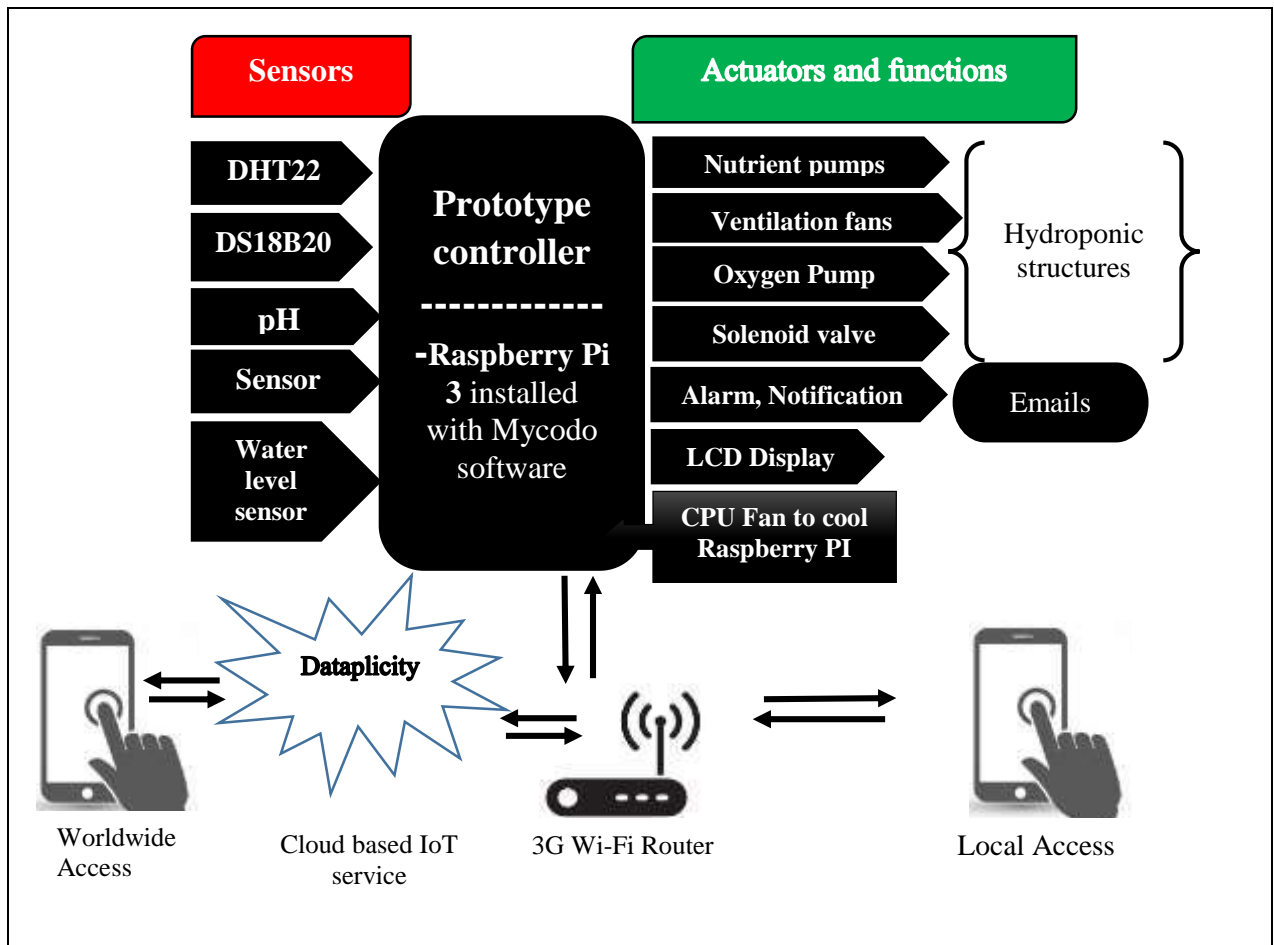
Figure 1. Basic architecture of the prototype automation of the hydroponics system

2. Materials and Method

2.1 Hardware:

2.1.1 Raspberry Pi

In this study, we used Raspberry Pi 3 model B+ (Figure 2) installed with the latest Rasbian OS Lite version downloaded from the Raspberry Pi Foundation website on 32GB micro-SD card. Raspberry Pi is a credit-card size single-board computers (SBCs) developed in the UK by the Raspberry Pi Foundation in association with Broadcom (Jolles, 2021). It is widely used in many areas as they are low cost with an open design that provides a set of general-purpose

Figure 2. Raspberry Pi 3 Model B+

input/output (GPIO) pins that allow to control electronic components for physical computing and explore the IoT.

2.1.2 DHT22 Environment Sensor

The DHT22 (Figure 3) is a low-cost digital temperature and relative humidity sensor with a single wire digital interface. It uses a capacitive humidity sensor and a thermistor to measure the surrounding air and spits out a digital signal on the data pin (Mihai, 2016). The sensor is factory pre-calibrated and does not require extra components to measure humidity and temperature. The temperature measuring range is from -40 to +125 degrees Celsius with ±0.5 degrees accuracy and humidity measuring range from 0 to 100% with 2-5% accuracy. The sensor is connected to GPIO-24 of the Raspberry Pi and is used for monitoring the temperature and humidity inside the hydroponics structure.
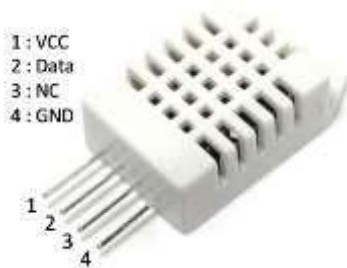

Figure 3. DHT22 Humidity and temperature sensor

2.1.3 DS18B20 Digital Temperature Sensor

DS18B20 is a 1-wire interface temperature sensor manufactured by Dallas Semiconductor Corp (Figure 4). The 1-wire interface requires only one digital pin for two-way communication with a microcontroller (Papoutsidakis, Chatzopoulos, & Piromalis, 2019). The sensor is quite accurate and does not require any external components to operate. It can measure temperatures from -55 ° C to +125 ° C with an accuracy of ± 0.5 ° C. This sensor is connected to the GPIO-15 of Raspberry Pi and used for monitoring the temperature of hydroponics nutrient solutions or water temperature.

Figure 4. DS18B20 digital Temperature Sensor

2.1.4 LCD Display

We used LCD1602 which can display 2 rows with 16 column characters with an integrated serial I^2C interface and green backlight (Boloor, 2015). The display is connected to the Raspberry Pi through I^2C interface GPIO pins and powered by a 5V power supply (Figure 5). It was used mainly for displaying critical data such as the temperature, humidity and IP address of the Raspberry Pi. It is necessary to know the IP address of the Raspberry Pi to access the system when connected to the local area network. In most home wifi routers, it is configured as Dynamic *Host Configuration Protocol* (*DHCP*) protocol which assigns IP addresses automatically and keeps changing daily.
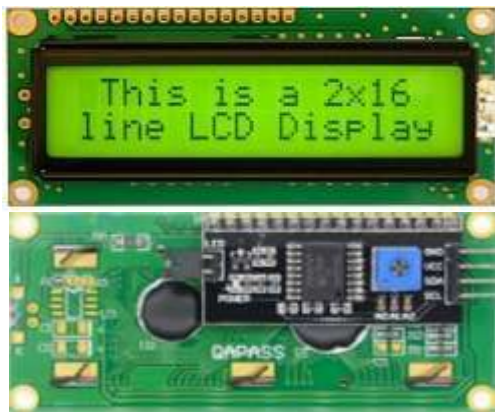

Figure 5. LCD1602 display integrated with I^2C serial interface

2.1.5 Actuators

An actuator is a device that is operated or activated based on the sensor values that are pre-set in the Mycodo software. The following actuators were used in the hydroponics structure at ARDC-Wengkhar.

- Nutrient circulation pump: Two centrifugal pumps (1HP and 0.5HP) for circulation of nutrient solutions in nutrient film technique and vertical tower
- Exhaust and ceiling fan: Two exhaust fans and three ceiling fans were installed for cooling and ventilation of the hydroponic systems
- Oxygen pump: Three oxygen pumps were installed in the DWC system for providing oxygen in the nutrient solution

2.1.6 Electronic components

Several other electronic components such as transistors, diode, resistors and relay modules were used for prototyping printed circuit board (PCB) for interfacing Raspberry Pi with sensors, actuators, and power supply (Table 1).

Table 1. Electronic components used in the study

Items		Specifications/module
1.	Relay module	10Amp, 12V relay
2.	Transistor	NPN, BC547
3.	Resistors	1 K Ohm resistor
4.	Screw Terminals	2 way, 3.5 mm pitch
5.	Male Pin header	20 pin, 2.54 pitch
6.	Female pin header	20 pin, 2.54 pitch
7.	PCB board	12X16 mm, double-sided
8.	Water pump	240V AC, 1 HP centrifugal pump
9.	Exhaust fan	240V AC, 70 watts
10.	Ceiling fan	240V AC, 80 watts
11.	Oxygen pump	240V AC, 18 watts
12.	Water filter	100 micron, flow rate $12m^3$/hr
13.	Wifi router	3G or 4G wifi router
14.	SIM card	Tashi cell SIM with data package
15.	Enclosure box	30X45 mm plastic or steel box

2.2 Software

2.2.1 Automation software- Mycodo

The automation software installed on Raspberry Pi 3 is called Mycodo, which is open-source software for environmental monitoring and regulation systems developed and maintained by Gabriel (2021). The software was installed on the Raspberry Pi 3 by following instructions provided by Gabriel (2021).

2.2.2 Application software- Electronic Design Automation (EDA) software- KiCad

For drawing the electronic schematics and developing them into PCB, KiCad software was used, which is also free and open-source software for EDA which facilitates the design of schematics for electronic circuits and their conversion to PCB designs (Hill, 2010). KiCad version 5.1.5 was downloaded and installed on the Microsoft Windows 10 PC.

3. Results and Discussion

3.1 Schematic and prototype PCB development

The detailed circuit schematic and PCB were developed in collaboration with a local IT firm on KiCad software. Based on the circuit schematic, two prototype PCB boards were developed

(Figures 6 & 7) and printed on a 12 X 16 mm double-sided copper PCB board using HP laser printer. The printed PCB board was then itched with Ferric Chloride solution and all electronic components were soldered on the PCB board. These tasks were carried out in the local IT firm's fabrication workshop. The final prototype PCB board contained the following peripheral connectors:

- Raspberry Pi 3 or Raspberry Zero W model
- Sensor connectors for pH sensor, EC sensor, temperature and humidity sensor (DHT22 and DS18B20).
- Display connector pins for LCD 16X2 and 16X4 display through I2C bus. The other sensors and actuators which support the I2C protocol can be connected to the same connector
- Eight output connectors for actuators through relays boards
- One USB webcam connection through Raspberry Pi USB port
- Two input power supply connectors 12V and 5V DC
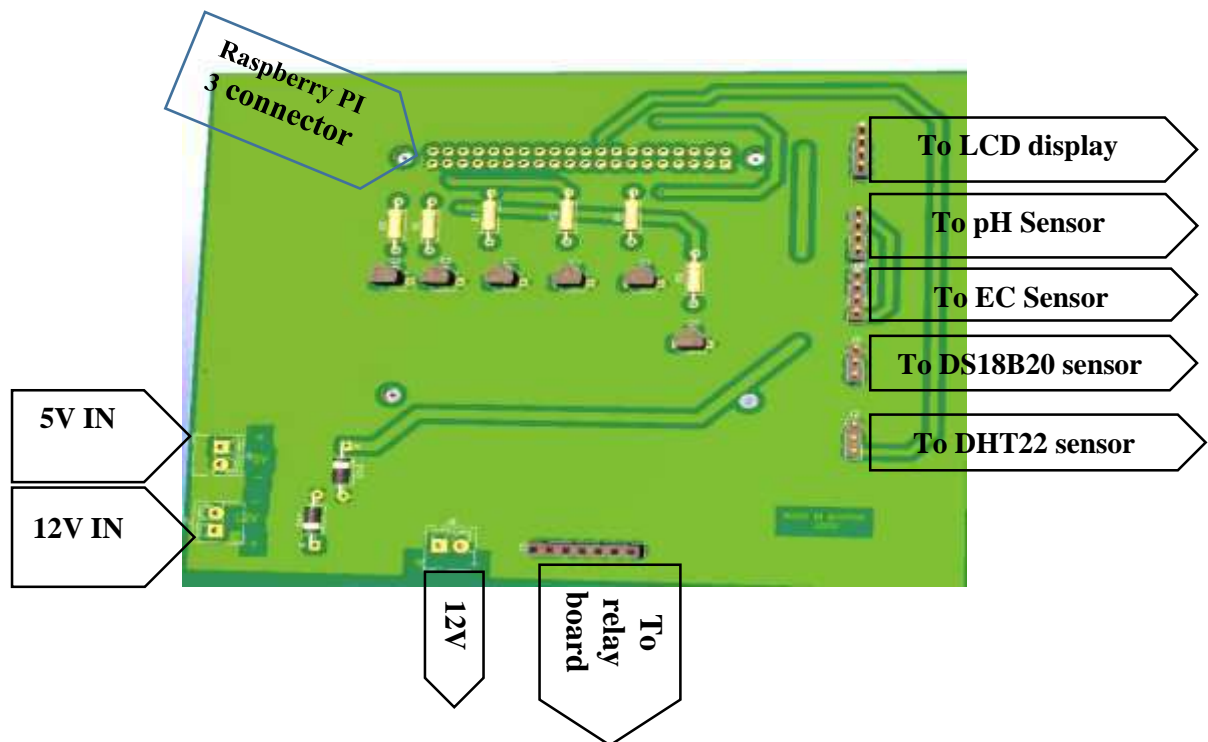- One 12V DC output power supply

Figure 6. Front view of the double-sided PCB board for interfacing with Raspberry Pi 3, sensors, actuator and power supply
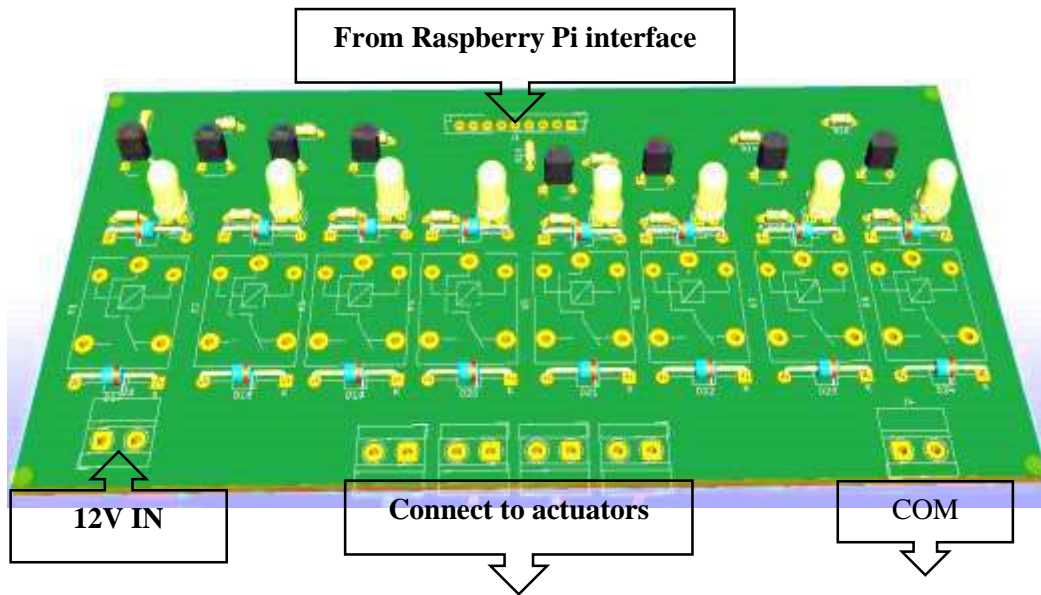
Figure 7. PCB board for relay module

3.2 Test results on the hardware configuration

The prototype PCBs were connected with sensors and actuators as shown in Figures 7 and 8 connector pins layout. Only two temperature and humidity sensors (DHT22 and DS18B20) were installed in the system. Other sensors such as pH and EC were not tested as they are not readily available for online purchase but connectors were made available on the PCB for future usage. The actuators included two numbers of AC pumps that circulated nutrient solutions in the NFT and vertical tower. For cooling and ventilation, two exhaust and ceiling fans were installed in the hydroponics structure and six numbers of oxygen pumps for providing oxygen in DWC.

Figure 8. Prototype automation box- inside view (*left*) & outside view (*Right*)

The system was also supported with a USB webcam for taking time-lapse photography and remote real-time monitoring of the hydroponics operation. The PCB boards and power supply modules were fitted into the weather-proof 35X48 mm size steel box (Figure. 8) and installed in the hydroponics structure containing NFT, DWC and the vertical tower that were already growing crops like lettuce, strawberry and medicinal herbs.

3.3 Test results on software configuration

Mycodo software installed on a Raspberry Pi acted as the main computing hub of the automation. The system consisted of two parts: a backend (daemon) and a frontend (web server). The backend performed tasks such as acquiring measurements from sensors and devices and coordinated a diverse set of responses to those measurements, including the ability to modulate outputs switch, relays, generate PWM signals, operate pumps, switch wireless outlets, publish/subscribe to MQTT, regulate environmental conditions with PID control, schedule timers, capture photos and stream video, and triggered actions when measurements meet certain conditions. The frontend of Mycodo hosted a web interface that enabled viewing and configuration of the system from any browsers using device IP address. The web interface supported an authentication system with user/password credentials, user roles that grant/deny access to parts of the system, and SSL for encrypted browsing (Gabriel, 2021). After logging into Mycodo system, the INPUT and OUTPUT features of the system were configured as follows:

3.3.1 Input device

Three inputs device were selected and activated in the system (Figure 9)

- DHT22 temperature and humidity sensor for measuring and monitoring the temperature/humidity inside the hydroponic structures
- DS18B20 temperature sensor for periodic measurement of the water temperature in the tank
- CPU temperature for monitoring the Raspberry Pi internal CPU temperature

3.3.2 Output device

Five GPIO pins of the Raspberry Pi were configured as outputs which were connected to the relay module (Figure 9). The relay module is connected to the following actuators:

- 12V CPU fan for cooling the Raspberry Pi
- Four numbers of exhaust and ceiling fans with parallel connections from one relay
- Two numbers of oxygen pump parallel connection from one relay
- One nutrient Pump (1HP) that circulates nutrient solutions in NFT

- 0.5 HP nutrient Pump which circulates nutrient solutions in the vertical tower

Figure 9. Configuration of INPUT (*Left*) and OUTPUT (*Right*) on Mycodo system

3.3.3 Configuring Functions

In the Mycodo software, the automation was mainly achieved by the interaction of various sensors and actuators located under the FUNCTION setting. The following functions were tested for automation of the system.

- Nutrient Pumps: The two nutrient pumps which circulate nutrient solution were configured to run for two minutes every 10 minutes interval on a timer-based module under the FUNCTION setting.

- Oxygen Pumps: Also, timer-based which runs for five minutes at every 30 minutes interval.

- Exhaust and CPU fans: Exhaust and CPU fans were configured as conditional functions which operate when certain conditions such as high and low set-point temperature of the greenhouse were met. These conditions were fed to the system in a few lines of python script under Function settings.

3.3.4 Configuring the LCD

On the Setup -> LCD page, 16×2 LCD" was set up to display the temperature, humidity and IP address of the device. 16×2 LCD has 16 Columns and 2 Rows and displays 32 characters on the screen.

3.3.5 Dashboard

Dashboard is an information management tool used to track, analyze, and display key performance indicators, metrics, and data points to monitor the overall health of the automation system. Dashboards in Mycodo are user-friendly and easy to set up.

On the Data -> Dashboard page, a custom dashboard was created as shown in Figure 10. The dashboard provided real-time detailed information on all sensors data and the status of various

actuators present in the system. Based on this information, decisions were made on whether or not to turn on the air pump, water pump and lamp remotely according to the acquired information.

Figure 10. Dashboard for real-time monitoring of sensor data and the status of the actuators

3.3.6 Remote access

The remote access to the Mycodo system was done by installing a small piece of software called *dataplicity* on the Raspberry Pi that allowed to access Mycodo system which is behind firewalls and NAT (Bepery, Baral, Khashkel, & Hossain, 2019). Dataplicity's main function is similar to the port forwarding method for remote access, but it is easier, efficient and more secure than the port forwarding method. After installation, the Mycodo automation system was made accessible worldwide and can be accessed by any device (laptop, desktop, smartphone) connected to different internet networks like mobile data, broadband and lease line internet services.

3.3.7 Temperature and humidity test data

The Mycodo application showed that it received the data from sensors timely and made proper control of the actuators. It provided detailed information on temperature, relative humidity and dewpoint inside the hydroponics. Each log was updated constantly and showed the progression of temperature and humidity from day to night (Figure 11). The other sensors and actuators data (data not shown) such as water temperature, the temperature of CPU, the amount of time and duration of the different actuators (like pumps, lights, fans) ON-OFF state were also recorded and automatically logged into the system.
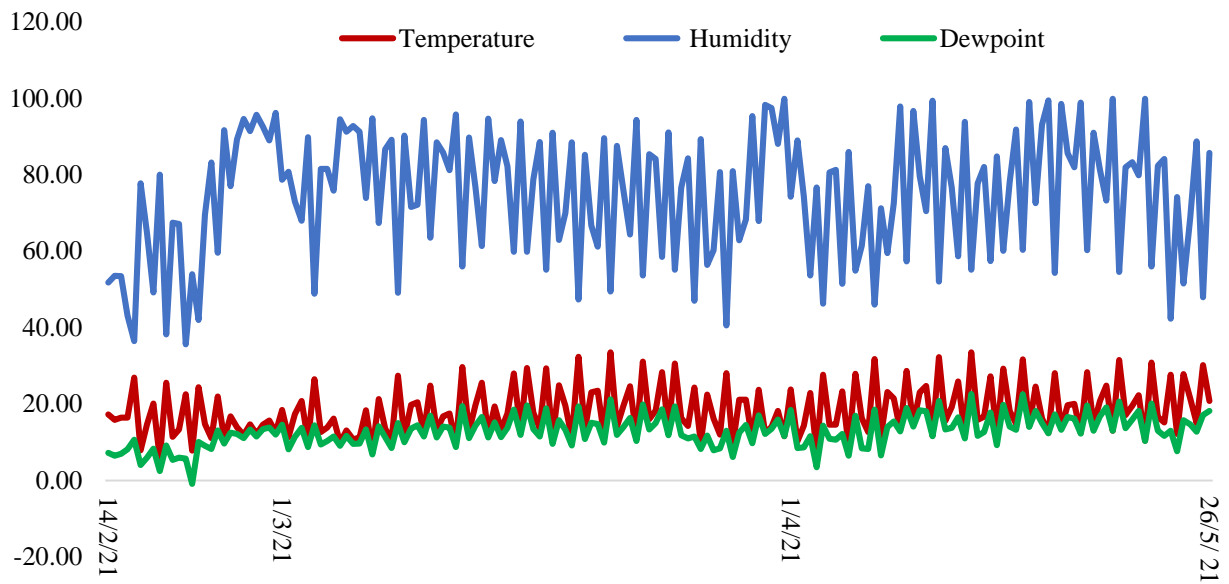
Figure 11. Log data on air temperature and humidity in the automated hydroponics system

3.3.8 Comparison of run time and energy consumption of different devices

The automation system was continuously tested for 70 days in the hydroponics structures that had been planted with different vegetable crops. The runtime and energy consumption of different devices in the automated and non-automated hydroponics system were compared using the logged data set in the prototype automation device. Figure 12 shows that the run time (hours) and energy consumption (kWatt) of the devices in the non-automated systems (manual operation) were more than 50% higher than in the automated hydroponics system.
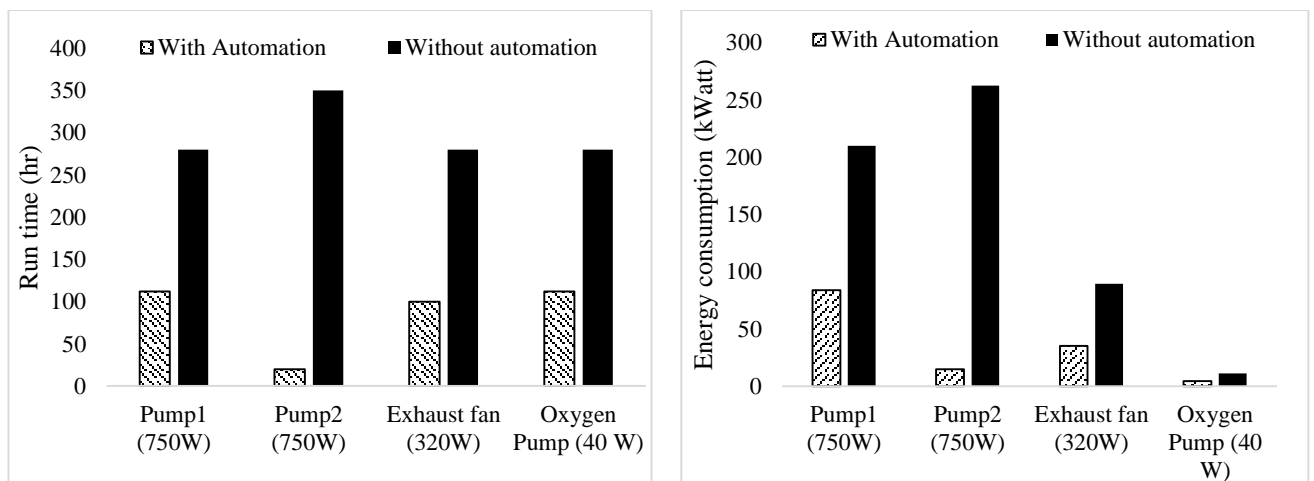

Figure 12. Comparison of runtime and energy consumption of different actuators during the testing period

4. Conclusion

Recently, hydroponics is seen as a promising technology for growing different crops as it is possible to grow short duration crops like vegetables around the year in a limited space and with limited water as it has water-saving efficiency of 70-90% compared to soil farming (Sharma et al., 2018). In Bhutan, hydroponics technology is expected to develop and be promoted as one of the means to attract youths into agriculture farming. Therefore, it is important to develop a suitable hydroponics automation system that reduces dependence on human labour and lower overall operational costs. The hydroponics automation prototype that we developed at ARDC-Wengkhar was deployed in real-field conditions for more than eight months and is tested to be efficient, reliable and useful. The prototype automation system is found to be expandable as it offers more flexibility and advanced features for any automated hydroponic systems for future upgrades and upscaling.

Reference

Bepery, C., Baral, S., Khashkel, A., & Hossain, F. (2019). Advanced home automation system using raspberry-pi and arduino. *International journal of computer science and engineering, 8*(2), 1-10.

Boloor, A. J. (2015). *Arduino by example*. Birmingham, UK: Packt Publishing Ltd.

DoA. (2021). *Hydroponics farming technology- a skilling program (training manual)*. Thimphu, Bhutan: Department of Agriculture, Ministry of Agriculture & Forests, Royal Government of Bhutan.

Dudwadkar, A., Das, T., Suryawanshi, S., Dolas, R., & Kothawade, T. (2020). Automated Hydroponics with Remote Monitoring and Control Using IoT. *International Journal of Engineering Research and Technology (IJERT), 6*(9).

El-Kazzaz, K. A., & El-Kazzaz, A. A. (2017). Soilless agriculture a new and advanced method for agriculture development: an introduction. *Agricultural Research & Technology, 3*(2), 63-72. doi:10.19080/ARTOAJ.2017.03.555610

Gabriel, K. T. (2021). Mycodo: Environmental monitoring and regulation system, Version 8.12.9. Retrieved from Mycodo website: https://kizniche.github.io/Mycodo/mycodo-manual.pdf

Hill, J. (2010, June). *Schematic Capture And Technical Drawing Software For Computer Engineering*. Paper presented at the 2010 Annual Conference & Exposition, Louisville, Kentucky.

Jolles, J. W. (2021). Broad-scale applications of the Raspberry Pi: A review and guide for biologists. *Methods in Ecology and Evolution, 12*(9), 1562-1579. doi:https://doi.org/10.1111/2041-210X.13652

Kularbphettong, K., Ampant, U., & Kongrodj, N. (2019). An automated hydroponics system based on mobile application. *International Journal of Information and Education Technology, 9*(8), 548-552. doi:10.18178/ijiet.2019.9.8.1264

Mihai, B. (2016). How to use the DHT22 sensor for measuring temperature and humidity with the arduino board. *Acta Uiversitatis Cibiniensis–Technical Series, 68*, 22-25. doi:10.1515/aucts-2016-0005

Papoutsidakis, M., Chatzopoulos, A., & Piromalis, D. (2019). Distance control of water temperature via android devices. *Journal of Multidisciplinary Engineering Science and Technology, 6*(12), 11240-11244.

Sharma, N., Acharya, S., Kumar, K., Singh, N., & Chaurasia, O. P. (2018). Hydroponics as an advanced technique for vegetable production: An overview. *Journal of soil and water conservation, 17*(4), 364-371. doi:10.5958/2455-7145.2018.00056.5

Evaluation of Extension Device on Kubota Power Tiller for its Operational Maneuverability on Steep Terrain in Bhutan

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ABSTRACT

Bhutanese agriculture has been at the subsistence level and the adoption of mechanization started only in late 1986 with the establishment of the Agriculture Machinery Centre. One of the most popular farm machines is the power tiller, the use of which is fast replacing the old tradition of land preparation using oxen. However, the adoption of farm machinery for land preparation is limited to the narrow plain areas along the border with India. Most of the country is mountainous where the use of power tillers has become unsafe and less efficient due to high gradients. Further, farmers have been stretching the use of the power tillers for ploughing on the mountain slopes even though it is less labour-efficient and involves risk-taking. In many cases, power tillers are used on land with gradients exceeding 15 degrees with additional labour to prevent the machine from toppling. The experiment was carried out to determine the critical safe angle of the sideways tilt while ploughing, and also to determine the critical angle that is practically possible to increase by using an extension device that was designed and fabricated to increase the wheel tread. This research compares the three treatments, i.e., rubber wheel with normal axle, rubber wheel with the extension device, and paddy wheel for required machine stability. It was observed that the rubber wheel with the normal axle was only feasible up to 9.93 degrees slope, the paddy wheel and rubber wheel with extension device were feasible up to 20 degrees slope. There was a significant (P<0.05) difference between the rubber wheel with normal axle and the rubber wheel with the extension device in terms of stability and no significant difference was observed between the extension device and the paddy wheel. This was achieved by decreasing the centre of gravity in a high slope gradient land and this could help farmers bring more agricultural land with high gradient into meaningful cultivation, thereby, enhancing food production.

Keywords: *Gradient; Toppling; Critical angle; Extension Device; Machine stability*

1. Introduction

Bhutan is a landlocked country in the eastern Himalayas between China and India. Geographically, it lies between 26°40' and 28°15'N and 88°45' and 92°10'E with an area of 38,393 km^2 (Dendup, 2018) Agriculture is the main occupation of Bhutanese people. Generally, traditional subsistence or semi-subsistence farming practice and 57% of the Bhutanese population is dependent on agriculture for their livelihood (Chhogyel & Kumar,

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2018). However, there has been rapid adoption of mechanization, mainly for land preparation and post-harvest processing. Among the farm machinery, power tillers in land preparation are the most popular. The use of power tillers has substantially reduced farm drudgery besides reducing farm labour requirements (JICA, 2016). Farm mechanization has provided some solace in the face of decreasing farm labour in rural areas, particularly fueled by rural-urban migration.

However, the use of a power tiller for land preparation is severely limited by the gradient of the agricultural land (Kinga & Chetem, 2019; Norbu, 2017). With the decreasing animal draught power, the use of power tillers has been put to their extremes with the machine being used even in fields with slopes that are much beyond what is considered technically safe to operate. Further, 49.5% of Bhutan's geographical area has slopes greater than 50 degrees (Dendup, 2018). According to the Monitoring and Evaluation Report on Government Hiring Service, 26% of the respondents reported power tiller accidents from rolling over caused by inadequate measures to stabilize the machines (AMC, 2019). The existing method employed by the farmers to maintain the balance in the power tillers on extreme slopes is manual, involving one or two extra farm labourers supporting the machine to prevent it from rolling over the slope. This manual process continues throughout ploughing on extreme slopes and often requires one or two farm labours to exert constant force in pulling the machine upwards throughout the operation. Thus, the risk of accident is not only high but also the efficiency is drastically reduced. In the world, more than 50% of accident that involves death occurs due to lateral overturning and backward rollover of agricultural machines (Hwang, Jang, & Nam, 2021). Even though the demand for power tiller is high but due to the lack of its manoeuvrability on the slope, the usage is reduced (AMC, 2019). Therefore, there is a need to determine the way around utilizing power tillers safely and effectively on the slope of agricultural land.

Thus, this paper attempts to determine the maximum slope on which a power tiller can safely operate. There were three experimental treatments: a normal rubber wheel, a rubber wheel with an extension device, and paddy wheel attached to the power tiller. According to agriculture statistics, farm labour shortage is a major constraint in agriculture (DoA, 2016), with this research we plan to mitigate the labour shortage by improving the safe maneuverability of power tillers to operate at the higher gradient. The field experiment was conducted in three regions to ensure the validity of the findings across the regions.

2. Materials and Method

2.1 Test area

The common and the most popular power tiller in Bhutan, the Kubota model RT125 (Japanese made) was used to test the safe manoeuvrability of the power tiller at the maximum slope. The experiment was conducted in three regional districts of Pemagatshel, Paro and Trashigang. Pemagatshel is located in the southeastern part of Bhutan and is characterized by highly dissected mountain ranges, steep slopes, and narrow valleys with little flat land. Paro is situated in the north-western part of the country and is characterized by more gentle slopes and flat land as compared with Pemagatshel. Paro is located at an altitude of 2,250 m above sea level. Trashigang is located in the eastern part of Bhutan and has a topography similar to Pemagatshel. Trashigang is at an elevation of about 1174 m above sea level.

2.2 Tools and equipment

Kubota model RT125 (Japanese made) is used in the experiment as it is the commonly used power tiller and would represent power tiller used for land preparation in Bhutan. The detailed specification on physical make, engine and tyre as presented in Table 1 is based on a test report prepared by the AMC (AMC, 2015).

Table 1. Machine specification was used in the experiment (AMC, 2015)

Serial No	Particular	Specification
1	Power tiller	
	a) Model:	RT125
	b) Make:	Kubota
	c) Overall dimensions (mm)	
	• Length:	2145 mm
	• Width:	866 mm
	• Height:	1210 mm
2	Engine	
	a) Type:	Diesel, horizontal
	b) Number of cylinders:	1
	c) Type of combustion:	Internal
	d) Make:	Kubota
	e) Model:	RT125
	h) Rated engine power:	12.5HP/2400 rpm
3	Tyre size (inch):	6-12 inches

Three treatments were used: a normal rubber wheel with normal axle, rubber wheel with extension device, and paddy wheel attached to the power tiller model as provided above (Table 2). The wheel-tread distance of the experimental treatments ranged from 700 to 970 mm.

Table 2. Three experiment treatments with different wheel tread distances.

Serial No.	Treatments	Wheel tread distance
1	Rubber wheel with normal axle	700 mm
2	Rubber wheel with an extension device	970 mm
3	Paddy wheel	950 mm

The extensions used for the experiment were designed and fabricated at the AMC research workshop using mild steel plates. The fabricated parts were hexagonal sockets into which the hexagonal wheel axle could be inserted. Using the extensions, the wheel tread could be increased in steps of 40 mm from 970 mm to a maximum wheel tread of 1050 mm. The fabricated parts are shown in Fig.1 with their specification in Table 3.

Table 3. Specification of extension devices

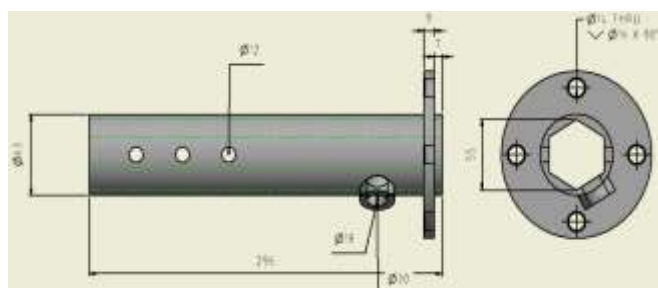| Model	Extension device
Length	296 mm
Outer diameter	130 mm
Inner across the corner	55 mm
Inner across the flat	47.5
Machine weight	3.5 kg
Cost	Nu.2500

Figure 1. Extension device

2.3 Measurement procedures

The main objective of the experiment was to observe the stability of the power tiller operating on different gradients by using rubber wheels, rubber wheels with an extension of the axle, and the paddy wheels as different treatments during ploughing operation. The tests were conducted to determine the angles of tilts up to which the power tiller under different wheel treads successfully operates without toppling over. The experiment was conducted following a completely randomized design (CRD). In each of the three locations, the three treatments were replicated three times to increase the accuracy of the experiment.

In each location, a suitable experiment field with slopes ranging from 8 to 25° was selected using a Japanese-made inclinometer. Then the fields were ploughed using the three sets of treatments. The first set of data was collected using the rubber wheels with the normal axle,

followed by the rubber wheel with the extension, and finally with the paddy wheel. The slope of the portion of the test plot was noted. The distance between the start and the end for one run was marked. The time taken for each run and the critical angle of tilt was recorded.

The experiment recorded two types of inclination, i.e., land gradient which is the initial inclination of the land before ploughing, and machine gradient which is measured when one part of the wheel falls on the furrow during the ploughing operation. The tests were repeated for varying slopes until the machine tended to topple. The data on the type of soil, depth and width of the furrow, speed of ploughing, and slope of the land were collected.

2.4 Laboratory test

For the stability analysis of the power tiller, the factor used is the sideways overturning angle (Kang et al., 2019). The experiment was conducted in the laboratory by making an inclined plane on which the power tiller is placed. The angle of inclination of the inclined plane is variable and was increased until the power tiller tended to topple. This meant that the centre of gravity is more than the power tiller's base stability, and the side rollovers depend upon the centre of gravity and centrifugal force of the power tiller.

2.5 Theoretical calculation of the centre of gravity of power tiller

The centre of gravity of the power tiller could be determined using similar formulae which are used to determine the centre of gravity of the tractor. The point where all the parts of a physical object balance one another is referred to as the centre of gravity (Prabhat, Ashish, & Kunal, 2014).

The terminologies used in the calculation of the centre of gravity and the tilting angle are:

Wheelbase (L),
Front tread (T1),
Rear tread (T2),
Left front load (Wl1),
Right front load (Wr1),
Left rear-load (Wl2),
Right rear-load (Wr2),
Total load (W),
Height in an inclined position (h),
Right rear load in an inclined position (W'r2),
Left rear load in an inclined position (W'l2) and

113

Effective radius (R)

2.5.1 Calculation of longitudinal length from a stand (Macmillan, 2002)

The location of the centre of gravity in a longitudinal direction could be found by measuring the weight on the front and rear wheel as:

$$Lo= (W2*L)/W\ldots\ldots\ldots\ldots\ldots\ldots\ldots (1)$$

Where Lo= longitudinal distance of support from the stand; W2= Wr2+Wl2

2.5.2 Calculation of lateral distance from centre to right as:

$$\text{Lateral distance from centre to right (M)} = \frac{(Wr1-Wl1)T1+(Wr2-Wl2)T2}{2W}\ldots\ldots\ldots\ldots (2)$$

2.5.3 Calculation of vertical height (Macmillan, 2002)

This method could be calculated by lifting the front or rear wheel of the power tiller and measuring the weight in raised conditions.

$$V = R + \frac{F*L}{W*Tan\Phi}\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots.. (3)$$

where V= Vertical height; Φ= Tilting angle and F= difference between the weight of the front wheel

2.5.4 Calculation of tilting angle (Ciuffoli, 2019)

The maximum tilting slope could be obtained using the equation:

$$\text{Tilting angle}= \tan^{-1}(T2/2)/V\ldots\ldots\ldots\ldots(4)$$

where T2= Rear tread and V= Vertical height

2.6 Data analysis

The research data was compiled in Microsoft Excel and were analyzed using Excel ToolPak. Data generated using the completely randomized experimental design were subjected to the one-way analysis of variance (ANOVA) followed by post-hoc analysis with Bonferroni correction at $P <0.05$ level of significance for multiple comparisons of the treatment means.

3. Results and Discussion

3.1 Laboratory result

During the sideways overturning test, it was found that the point of no return (critical angle) using the rubber wheel was 30 degrees. As it can be seen, the power tiller would not topple as long as the moment by the centre of gravity is less than the counter moment acting upwards by the reactions through the wheels. This will remain so if the line of action of the centre of gravity is within the wheel tread. Therefore, the stability can be increased by the extension of the axle length to contain the action of the centre of gravity within the wheel tread.

114

Based on the theoretical calculation, the tilting angle of Kubota RT 120 was 28.5 degrees. It was found that with the increase in the wheel tread, the tilting angle also increased. This explains why the stability of the power tiller with the extension device and paddy wheel is higher than that of the rubber wheel. The tilting angle with the rubber wheel calculated in the laboratory and that calculated theoretically were almost the same, i.e., 30 and 28.5 degrees, respectively.

In summary, the maximum tilting angle can be obtained with the maximum rear tread and minimum vertical height of the centre of gravity. Any condition that modifies these two factors will change the tilting angle and thus, the stability of the tractor changes, which is also similar to the findings described by (Guzzomi, Rondelli, Guarnieri, Molari, & Molari, 2009).

3.2 Field result

The laboratory experiments were followed by the field experiments. The soil texture was analysed at the end of the rainy season and was found to be friable. The soil structure in Paro (Pangbesa) was sandy loam, while that in Pemagatshel (Chimong) and Trashigang (Kanglung) were observed to be clay loam.

During the field experimentation, the average speed of the plough was maintained at 1.32km/hr with a furrow depth of 150 mm. Table 4 shows the results of an experiment conducted on a rubber wheel, extension device, and paddy wheel. Based on the data, it was found that the feasible angle of inclination of land (land gradient) for the rubber wheel was 9.93 degrees. At this degree (9.93) the machines were found to be stable and completed the ploughing operation without any difficulty and once we increased the gradient, the machines tended to topple. The feasible angle of inclination of land (land gradient) for the paddy wheel and the rubber wheel with extension device was ≥20 degrees. A similar observation was made during the ploughing operation on the rubber wheel with extension and paddy wheel, i.e., the machines ran in a stable condition but would face some difficulty while turning around, as we increased the gradient the machine was stable but not able to plough due to the downward sliding moment.

Table 4. Critical land gradient observed among the three treatments

Regions	Rubber Wheel	Extension Device	Paddy Wheel
Pemagatshel	10.2 ±0.26	20.50 ±0.62	20.5 ±0.40
Paro	10 ±0.2	20.00 ±0.43	20 ±0.20
Trashigang	9.6 ±0.17	20.00 ±0.20	19.5 ±0.36
Mean	9.93 ±0.31[a]	20.17 ±0.29[b]	20.00 ±0.5[b]

Values are mean ± standard deviations of the means. Mean values within the rows with different superscripts are significantly different amongst the treatments at $P<0.05$ by Bonferroni correction.

Similarly, it was found that the feasible mean angle of inclination of the machine (machine gradient) was 23.33, 30.71, and 29.18 degrees for the rubber wheel, extension device, and paddy wheel, respectively (Table 5).

Table 5. Critical machine gradient observed among the three treatments

Regions	Rubber Wheel	Extension Device	Paddy Wheel
Pemagatshel	22.33 ±2.52	29.67 ±2.31	29.67 ±0.58
Paro	24.67 ±1.53	32.33 ±1.15	28.67 ±1.53
Trashigang	23.00±3.46	30.12 ±1.26	29.2 ±0.58
Mean	23.33 ±1.21[a]	30.71 ±1.42[b]	29.18 ±0.50[b]

Values are mean ± standard deviations of the means. Mean values within the rows with different superscripts are significantly different (amongst the three treatments) at $P<0.05$ by Bonferroni correction.

During the analysis (by one-way ANOVA), it was found that there is a significant difference among the three treatments. Further segregation of means of the treatment was done using post-hoc analysis by Bonferroni correction. There was a significant ($P<0.05$) difference between the use of the rubber wheel with normal axle and rubber wheel with the extension device on the stability and no significant difference between the rubber wheel with extension device and the paddy wheel. However, there was no significant difference among the treatments in a particular location. Figure 2 shows the feasible gradient in the three different districts.
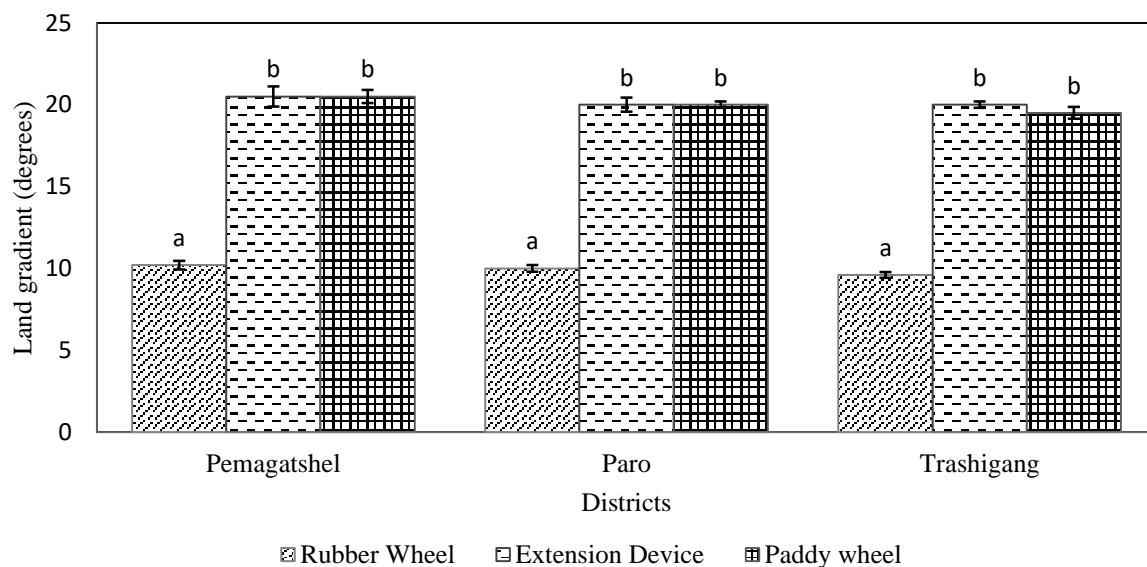

Figure 2. Feasible land gradient in three districts with the three treatments.

Values are mean ± standard deviations of the means. Mean values within a district with different letters indicate a significant difference at $P<0.05$.

Slope manoeuvrability is a very difficult test as the data collected varies from place to place due to geographical diversity. Therefore, we conducted the test in three different districts so

that the data collected was generally valid on a gentle to a somewhat steep slope. The usage of extension and paddy wheel decreases the centre of gravity eliminating the risk of toppling and compared to the normal rubber wheel they can go up to 20 degrees. However, if we go beyond 22 degrees, even though the machine remained stable, the power tiller was not able to plough the field due to the downward sliding motion.

The government is encouraging farm mechanization as it makes farming easier by reducing farm drudgery and making farming more productive. Implementation of mechanized farming will create job opportunities and enhance national food self-sufficiency. Since a large percentage of the agricultural land is on slopes beyond the critical angle, the development and consolidation of the lands with slopes greater than the critical slopes will greatly enhance labour efficiency and safety.

3.3   Limitation

Although the experiment was conducted efficiently and data collected successfully, certain limitations should be considered before using/interpreting these findings. The key limitations are: (1) the result is applicable for the most common model of the power tiller, Kubota RK125/K120. For other power tillers, the centre of gravity may differ and therefore, the stability might vary. However, the experiment and the procedures to test other machines will remain the same, (2) the precision of the critical angle of the slope determined is limited and may vary with soil structures, moisture content, the skill of the operator, and the ploughing speed, and (3) in stony fields the safety limit of the slope will tend to be lower and will depend on the skill of the operator in stabilizing the machine.

4. Conclusion

Farm mechanization starts with land preparation using the power tiller, a machine that is key to preparation. However, due to the rough topography, most agricultural lands in the country are left fallow. The finding of this research indicates that the stability of the power tiller across varying slopes is improved by increasing the wheel tread (using the extension device and paddy wheel). This was achieved by decreasing the centre of gravity in a high slope gradient land, and this could help farmers bring more agricultural land with higher gradients into meaningful cultivation, which ultimately can enhance food production.

The use of power tiller is limited by the topography of our country. Initially, the farmers could not plough above 9 degrees due to machine toppling and the risk associated. Henceforth, Bhutanese farmers can plough land up to a 20-degree gradient easily using the extension device

or paddy wheel. This study found that the normal rubber will plough only up to 9 degrees and by using the extension device or the paddy wheel, the power tiller could plough up to 20 degrees. The finding can serve as a way forward for the modification of different types of power tillers to make them suitable for mechanizing land with higher gradients.

Acknowledgement

References

AMC. (2015). *Power tiller (Kubota-RT125) -Test Report*. Retrieved from Paro, Bhutan

AMC. (2019). *Monitoring and evaluation report on government hiring service*. Paro, Bhutan: Agriculture Machinery Centre, Department of Agriculture, Ministry of Agriculture and Forests

Chhogyel, N., & Kumar, L. (2018). Climate change and potential impacts on agriculture in Bhutan: a discussion of pertinent issues. *Agriculture & Food Security, 7*(1), 1-13. doi:10.1186/s40066-018-0229-6

Ciuffoli, A. (2019). *Stability assessment of agricultural tractors and self-propelled sprayers.* (PhD Thesis), Università di Bologna Bologna, Italy.

Dendup, T. (2018). Agricultural transformation in Bhutan: From peasants to entrepreneurial farmers. *Asian Journal of Agricultural Extension, Economics & Sociology, 23*(3), 1-8.

DoA. (2016). *Agriculture Statistics 2016*. Thimphu: Department of Agriculture (DoA), Ministry of Agriculture and Forests, Royal Government of Bhutan

Guzzomi, A. L., Rondelli, V., Guarnieri, A., Molari, G., & Molari, P. G. (2009). Available energy during the rollover of narrow-track wheeled agricultural tractors. *Biosystems Engineering, 104*(3), 318-323. doi:10.1016/j.biosystemseng.2009.07.005

Hwang, S.-J., Jang, M.-K., & Nam, J.-S. (2021). Application of Lateral Overturning and Backward Rollover Analysis in a Multi-Purpose Agricultural Machine Developed in South Korea. *Agronomy, 11*(2), 297. doi:10.3390/agronomy11020297

JICA. (2016). *The preparatory survey report on the project for improvement of farm machinery for hiring services in kingdom of Bhutan*. Thimphu, Bhutan: JICA - Bhutan.

Kang, N. R., Choi, I. S., Lee, W. J., Woo, J. K., Kim, Y. K., Choi, Y., . . . Yoo, S. N. (2019). Sideways Overturning and Overturning Angle Test for a Three-Wheel Riding-Type

Cultivator. *Journal of Biosystems Engineering, 44*(1), 12-17. doi:10.1007/s42853-019-00008-y

Kinga, N., & Chetem, W. (2019). An economic analysis of government custom hiring services for different farm machineries in Bhutan. *SAARC Journal of Agriculture, 17*(2), 93-101. doi:10.3329/sja.v17i2.45297

Macmillan, R. H. (2002). *The mechanics of tractor-implement performance: theory and worked examples: a textbook for students and engineers*. Melbourne: RH Macmillan.

Norbu, K. (2017). Present situation and future prospect for farm mechanization in Bhutan. *AMA-Agricultural Mechanization in Asia, Africa and latin America, 48*(2), 40-43.

Prabhat, G., Ashish, D., & Kunal, S. (2014). To increase the stability of a tractor by lowering down the centre of gravity. *International Journal of Analytical, Experimental and Finite Element Analysis, 1*(3), 164–168.

Storage Losses of Maize in Four Different Storage Methods in Thangrong Gewog, Mongar Dzongkhag

Karma Dorji[o], Kinley Wangmo[p], Sujan Pradhan[o], Dorji Rinchen[q]

ABSTRACT

Several maize storage technologies are made available and recommended to the farmers in the villages by different agencies to reduce post-harvest losses. This study aimed to evaluate the storage losses of maize mainly to insect and fungal damages in four different storage methods for a maximum of five months storage period in Thangrong, Mongar dzongkhag (district). Freshly harvested maize was shelled, dried, weighed and stored under four different storage methods and replicated three times for each storage method. Moisture content, physiological weight loss, insect and fungal damages were the storage quality parameters assessed monthly until five months of storage. All the storage methods maintained the moisture content within the range recommended for the safe storage of maize (12-14 %). The physiological loss of weight (PLW) was the lowest at 0.10 % for the maize grain stored in super bag and the highest at 0.30 % was observed in grains stored in curing and storage shed over the four-month storage period. Traditional storage and silos resulted in PLW of 0.20 % and 0.16 %, respectively. Overall insect damage (sum for five months) of 2.54 % in the traditional storage was the highest reported among the storage methods followed by 1.79 % for grains stored in curing and storage sheds. Insect damage of 0.64 % for super bag and 0.27 % for silo stored maize grains were observed. Fungal damaged grain ranged from a low of 0.19 % for curing and storage shed to a high of 0.31 % for super bag stored maize grains. Overall storage losses (sum for five months) to insects and fungal diseases were in the range of 0.51-2.80 % with the highest being observed in traditional storage and the lowest in silo storage. All the storage methods evaluated maintained good quality maize grains with minimal damage till five months of storage. The existing improved storage technologies could help in the safe storage of maize grains if stored after proper drying. It is recommended to conduct a similar comparative study for the maize harvested and stored in the summer months and also at a different location to validate the results.

Keywords: *Maize; Storage methods; Storage losses; Insect damages; Fungal damages*

1. Introduction

Maize (*Zea mays*) also known as corn is the staple food of many Bhutanese, especially in the eastern and southern dzongkhags. Maize dominated the total cereals cultivation area of Bhutan

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with 46.3 % for the year 2015 (PPD, 2015). In the same year, the share of maize production among cereal crops was 45 %. Mongar, Tashigang, Dagana, Samdrup Jongkhar, Sarpang, Pemagatshel, Tsirang and Zhemgang are major maize producing dzongkhags in Bhutan (PPD, 2015). It is also cultivated in small quantities in other dzongkhags of Bhutan for self-consumption and as a cash crop in recent years. In 2017, total maize production was recorded at 94,051 Mt from 66,043 acres of land with the highest production in the dzongkhags of Mongar, Tashigang, Samdrup Jongkhar, Samtse, Tsirang, Dagana and Pema Gatshel (NSB, 2018). Maize production was reported to be 55,259 Mt and 46,235 Mt in the year 2018 and 2019, respectively (RSD, 2020). Despite the huge production of maize in the country, the loss at the post-production storage phase is estimated to be high according to informal sources and an unpublished report from the field (NPHC, 2016). The major reason cited was the lack of proper and appropriate storage technologies for long term storage of maize and the easy attack of maize by insects and fungal infections due to improper storage technology. Additionally, poor knowledge of safe moisture content requirements for storage, improper drying and not sorting the grains before storage result in poor storage quality.

The major post-harvest losses of crops are due to poor methods of harvesting and handling, use of inappropriate containers while packaging, poor storage conditions, and poor transportation and distribution system (Kiaya, 2014). According to Basappa (2004), the important factors leading to storage losses are long-duration storage using traditional methods, inadequate knowledge, poor storage structures, non-availability of separate godowns and damage by rodents, insects, and dampness of storage environment and storage of improperly dried grains. Among the several factors affecting the storage life of maize, the use of an improved storage system that is clean with good ventilation for proper circulation of air and additional features to protect from rodents and wild animals will significantly reduce storage losses in maize. The growers should also ensure that only the properly dried maize with a recommended moisture content of 14 % or below should be stored to deter fungal and mould growth.

The post-harvest loss of cereals in Sub-Saharan Africa ranged between 5-40 % with an estimated worth of around $4 billion (Zorya, Morgan, Diaz Rios, Hodges et al., 2011). Losses of cereal crops in developing countries are estimated to be as high as 25% of the total production. The maximum losses of 50-60% of cereal grain occur during the storage period due to the lack of proper storage management and structures (Kumar & Kalita, 2017). Meronuck's 1987 report (as cited in Suleiman & Kurt, 2015) indicates that losses of maize in various storage facilities in undeveloped tropical countries are in the range of 15-25%. As per

the findings from the farm survey on maize impact study by Shrestra, Katwal, and Ghalley (2006), the overall post-harvest loss in maize in Bhutan was reported to be 20%. Additionally, the post-harvest loss was 26% higher among the farmers growing traditional varieties compared to those who adopted modern varieties. This annual post-harvest loss in monetary terms was valued at Nu.181 million for Nu.10/kg.

The storage losses are classified into two main categories namely, biotic and abiotic factors. Biotic factors are insects, rodents, pests and fungi while abiotic factors refer to temperature, humidity and other environmental factors. However, the storage losses are caused due to the combined effect of both biotic and abiotic factors. Temperature, humidity and moisture content of the crops are the driving force behind the cause of many storage problems such as insect infestation and fungal growth in the stored crops. Most of the storage moulds grow rapidly at a temperature between 20-40 °C and relative humidity of 70 % and above (Kumar & Kalita., 2017). Minimizing the post-harvest losses would increase the amount of food available for human consumption and enhance global food security. In addition, crop production contributes a significant proportion of typical incomes in certain regions of the world and reducing food loss can directly increase the real incomes of producers.

The preliminary study to determine the post-harvest storage losses of maize was conducted in three Dzongkhags of Bhutan; Chhukha (west), Dagana (south-central) and Mongar (east) in the year 2019. That study was conducted to obtain the baseline data on post-harvest storage losses of maize in Bhutan and at the end of six months of storage, the total mean storage losses were 16.18%, 38.21% and 23.83% for Chhukha, Dagana and Mongar, respectively (Dorji, Tshering, & Lhamo, 2020). A survey study carried out in Nepal reported insects as the major cause of maize storage loss with 62% of the respondents' expressing insects as its cause while 39% of the respondents presumed the storage losses to be between 10-20% (Bhandari, Achhami, Karki, Bhandari et al., 2015). In continuation to the above study, it was found to be of utmost importance to compare the storage losses of maize in different storage methods disseminated by different agencies amongst the communities - traditional storage method, super bags, silos and NPHC curing and storage shed. This study was conducted to compare the storage losses of maize in different existing storage methods in Bawcholing, Thangrong Gewog, Mongar.

2. Materials and Method

2.1 Experimental design

The experiment was set up at Bawcholing village, Thangrong Gewog under Mongar Dzongkhag. Thangrong (Mongar), Karmaling (Dagana) and Sampheling (Chhukha) are the places that were initially selected by the Food and Agriculture Organizations (FAO) as a pilot sites for carrying out agriculture and post-harvest related programs under one of their projects. Accordingly, the improved curing and storage shed that was designed by National Post Harvest Centre (NPHC) was constructed at these sites. The sites at Chhukha and Dagana could not be included in the current study due to the COVID-19 situation that resulted in the two sites being labelled under red zones. Thangrong Gewog was selected as the site for study since maize is abundantly cultivated in the gewog and also as all the storage methods were readily available for the study. The study site is located at an altitude of 1361 masl.

Maize was harvested in February 2021, shelled and it was dried in the sun as practised by the farmers till the average moisture content of 12 % was achieved. The experiment was immediately started after the desired moisture level of 12 % for safe storage was achieved in February. The moisture content of maize for storage is recommended to be between 12-14% (FAO, 1992). The data collection was carried out monthly and continued till August 2021.

The following four different storage methods were evaluated.

Storage method 1: A metal silo designed and fabricated by the Agriculture Machinery Centre (AMC) was borrowed for the study. The silos were made out of a galvanized iron sheet and had a capacity of 500 kg. It has an opening from the top to put in the grains with a lid to close the storage. Twenty kilograms of maize grains were weighed and put into three silos. It was covered with the lid and the silos were placed in the basement of the house.

Storage method 2: Super grain bags are airtight plastic bag imported from Grainpro which comes with a zipper. Super grain bags were collected from National Plant Protection Centre. The capacity of the bag was 25 kg. The three replicated bags were filled with 20 kg each of maize grains, tied with a zip lock and kept in the basement of the house.

Storage method 3: Maize curing and storage shed was designed and constructed by the NPHC. The dimensions of the structure are 5m × 4m and raised by 2m from the ground. The sides of the structure have a mesh to protect from insects and pests with full aeration for good airflow.

The roof is covered with a corrugated galvanized iron sheet. Twenty kilograms of maize grains were put into the aerated gunny bags and placed in different corners of the shed.

Storage method 4: This is the farmer's traditional method which was used as one of the treatments. This method consists of filling the shelled maize grains in the gunny bags and keeping them in the corners of rooms, basement and attic of the house. Gunny bags were filled with 20 kg each of maize grains and stored in the basement of the house as practised. Grains that were free of insect and fungal damage were used for the storage study. For uniformity, maize grains were checked for moisture content and maize grains from the same lot with equal moisture content were stored in the storage methods. Each storage type had three replications.

A separate lot of 5 kg of maize in each replication for the four storage methods was stored to determine the change in physiological weight loss of the maize grains during the storage period. Traditional maize variety *Ashom barmu* was used during the storage study.

2.2 Determination of moisture content of maize grains

The moisture content of maize grain was measured using the portable digital moisture tester (A-Grain, India). The hopper of the moisture meter was filled with the maize sample and then the handle was released to transfer the samples into the sample holder. The measuring button was pressed for a few seconds and the moisture content displayed on the screen was recorded. The moisture of grain was checked before storage and from there on the moisture content of grains was recorded from all the samples during the monthly data collection.

2.3 Determination of physiological weight loss of grains

A separate lot of maize kept in each of the storage methods for determining the physiological weight loss was measured monthly. Weight was measured using the digital weighing balance. After the measurement of weight, the samples were put back in their respective storage methods for checking weight in the subsequent months. The physiological weight loss of the grain was converted into percentage as follows;

$$\text{Physiological weight loss (\%)} = \frac{\text{Weight at the time of monitoring}}{\text{Initial weight of the grains}} * 100 \qquad \ldots\ldots\ldots\ldots\ldots (1)$$

2.4 Determination of storage damages/losses

One kilogram of maize grains was sampled for all the replications (three replications) from different corners of the traditional bag, super bag, silos, and improved shed. Damages by insects and fungi were recorded as the main grain loss parameters in different storage methods. The number of grains that were damaged by insects and fungal diseases from the sampled lot

was counted from the sample grains and these numbers were converted into loss percentages. The monitoring and assessment of storage loss were continued for 150 days at an interval of one month each. The post-harvest storage losses of maize in each category were expressed in percentage;

$$\text{Losses to Insects damage } [A](\%) = \frac{\text{No. of insects damaged grains}}{\text{Total number of grains [C]}} * 100 \text{ …………..…..…… (2)}$$

$$\text{Losses to fungal damages } [B](\%) = \frac{\text{No. of fungal damaged grains}}{\text{Total number of grains [C]}} * 100 \text{ ………………..…… (3)}$$

$$\text{Overall storage losses } (OSL \text{ in } \%) = \frac{A+B}{C} * 100 \text{…………………..…………………... (4)}$$

Where;

OSL=Overall storage losses (%)

A= Number of insects damaged grains

B=Number of disease damaged grains

C=Total number of grains

2.5 Statistical analysis

Microsoft Excel 2016 was used to arrange the data, do basic calculations and plot figures. The post-harvest storage losses of maize in different storage methods were statistically analyzed using STAR (Statistical Tools for Agricultural Research) software. The data was arranged as per the format in STAR software and it was imported into the software. One-way ANOVA test was carried out to see if there is a significant difference among the means. Duncan's Multiple Range Test was the post-hoc test carried out to determine the significance between storage means ($P<0.05$).

3. Results and Discussion

3.1 Moisture content, physiological weight loss and percent of insect and fungal damaged grains after one month in storage

At the end of one month in storage, the moisture content of maize grains from different storage methods ranged from 10.53 to 11.30 % and there was no significant difference (Table 1). The average moisture content of the grain was 12.3 % at the time of storage but was observed to be reduced in all the storage methods after one month which is due to the loss of moisture and other substrates during the storage period. A significantly lower physiological loss of weight (PLW) of 0.1 % was recorded for the maize samples stored in the super bags compared to grains from other storage methods after one month in storage. The lower weight loss from the

super bag was obviously due to the low permeability of the bag that minimized the air and water vapour movement thus retaining its weight. Traditional, silos and sheds had more free air movement and resulted in the physiological loss of weight in the range of 0.36-0.47 %. Insect infested grain of 0.1 % in the traditional storage method was the highest recorded among the grain samples among four storage methods compared to 0.03-0.05 % for grains from the other storage methods but not statistically significant. Fungal infected grains were 0.03 %, 0.07 % and 0.05 % for grains stored in traditional methods, super bag and improved shed, respectively while no fungal infection was observed in the silo stored grains. The overall damage (sum of insect damage and fungal damage) of 0.13 % in the traditional storage method was the highest followed by 0.1 % for super bag and curing and storage shed while overall damage was just reported 0.03 % for silo stored grains. At the end of one month, the overall damage was very minimum from all the storage methods.

Table 1. Moisture content, physiological weight loss and percent of insect and fungal damaged grains after one month in storage

Storage method	Moisture content (%)	Physiological weight loss (%)	Insect damages (%)	Fungal damages (%)	Overall storage losses (%)
Traditional	10.53	0.36a	0.1	0.03	0.13
Super bag	11.30	0.10b	0.03	0.07	0.1
Silo	10.93	0.47a	0.03	0.0	0.03
Curing & storage shed	10.90	0.45a	0.05	0.05	0.1
S.E.	0.12	0.05	0.01	0.01	-
P- value	0.17	0.006	0.39	0.48	

Means in the column with different letters are significantly different between storage methods at $P < 0.05$ by Duncan's post-hoc test.

3.2 Moisture content, physiological weight loss and percent of insect and fungal damaged grains after two months in storage

After two months in storage, the moisture content was significantly higher in grains from silos (12.6 %) and super bag (12.5 %) followed by 11.87 % for traditional stored grains and 11.30 % for curing and storage shed grains (Table 2). The enclosed nature of silo and super bag probably retained slightly higher moisture content while higher moisture loss in traditional storage and curing and storage shed resulted in higher physiological loss of weight. There was no statistically significant difference in insect and fungal damaged grains with an overall storage loss in the range of 0.06-0.2 %. Similar to one month after storage, overall storage loss was highest in traditional storage methods after two months of storage at 0.2 %.

Table 2. Moisture content, physiological weight loss and percent of insect and fungal damaged grains after two months in storage

Storage method	Moisture content (%)	Physiological weight loss (%)	Insect damages (%)	Fungal damages (%)	Overall storage losses (%)
Traditional	11.87b	0.4a	0.1	0.1	0.2
Super bag	12.50a	0.1b	0.03	0.03	0.06
Silo	12.60a	0.0b	0.03	0.1	0.13
Curing & storage shed	11.30c	0.6a	0.07	0.03	0.1
S.E.	0.16	0.07	0.01	0.01	-
P value	0.00	0.006	0.36	0.11	

Means in the column with different letters are significantly different between storage methods at $P < 0.05$ by Duncan's post-hoc test.

3.3 Moisture content, physiological weight loss and percent of insect and fungal damaged grains after three months in storage

There was no significant difference in the moisture content of maize grains after three months of storage in four storage methods (12.03-12.97 %). Physiological loss of weight was 0.03 % for grains stored in traditional, and curing and storage shed while the physiological weight loss was nil for the other two methods of storage. The curing and storage shed has free air movement within its environment since it is a structure constructed outside and protected only by a wire mesh from the sides. This probably caused some weight losses from the grains stored in the curing and storage shed while physiological weight losses were not recorded from the other storage methods due to the enclosed status of the product in the basement room. In the third month, no insect damage was reported from the grains stored in traditional and silo storage while super bag, curing and storage shed reported insect damage of 0.03 % and 0.07 %, respectively. Grains stored in the curing and storage shed reported no fungal damage while other methods resulted in grain damage of 0.07-0.13 %. The overall storage damage was the highest at 0.16 % in super bag followed by 0.1 % for traditional storage. Except for grains from the super bag the traditional storage method still resulted in a slightly higher overall storage damage of grains. Silo storage and curing and storage shed resulted in an overall storage loss of 0.07 % (Table 3).

Table 3. Moisture content, physiological weight loss and percent of insect and fungal damaged grains after three months in storage

Storage method	Moisture content (%)	Physiological weight loss (%)	Insect damages (%)	Fungal damages (%)	Overall storage losses (%)
Traditional	12.17	0.03	0.0	0.1a	0.1
Super bag	12.53	0.00	0.03	0.13a	0.16
Silo	12.97	0.00	0.0	0.07ab	0.07
Curing & storage shed	12.03	0.03	0.07	0.0b	0.07
S.E.	0.15	0.01	0.01	0.01	-
P-value	0.12	0.59	0.56	0.02	

3.4 Moisture content, physiological weight loss and percent of insect and fungal damaged grains after four months of storage

There was a significant difference in moisture content of maize grains after four months in storage. The lowest moisture content of 12.8 % was recorded for grains stored in silos. The fourth month of storage was in the months of June-July and due to the increased summer temperature, the silo which is made up of galvanized sheets could have absorbed heat and caused the moisture content to reduce slightly. The moisture content in general increased for grains from all storage types due to the wet season that has set in. Physiological loss of weight ranged from 0.13 %-0.20 % for three storage methods while it was nil for traditional stored grains without any significant differences. Gunny bags that were used in the traditional storage method helped absorb moisture easily and could have led to zero physiological weight loss. Silos did not result in any insect damage while the highest damage of 0.2 % was reported from grains stored in super bags followed by 0.1 % from traditional and curing and storage sheds. Fungal damage was reported only from traditional storage in the fourth month of storage. Overall damage in the fourth month of storage was 0.2 % for super bag storage, 0.13 % for traditional storage and 0.1 % for curing and storage shed. No damage was reported for the maize stored in silos in the fourth month of storage. Similar to the first three months of storage, the overall storage loss was slightly higher in the traditional storage method except for super bag grains.

Table 4. Moisture content, physiological weight loss and percent of insect and fungal damaged grains after four months of storage

Storage method	Moisture content (%)	Physiological weight loss (%)	Insect damages (%)	Fungal damages (%)	Overall storage losses (%)
Traditional	13.77a	0.00	0.1b	0.03	0.13
Super bag	13.43ab	0.20	0.2a	0.00	0.2
Silo	12.80c	0.17	0.0c	0.00	0.0
Curing & storage shed	13.23bc	0.13	0.1b	0.00	0.1
S.E.	0.12	0.04	0.02	0.01	-
P-value	0.01	0.36	0.008	0.44	

Means in the column with different letters are significantly different between storage methods at $P < 0.05$ by Duncan's post-hoc test.

3.5 Moisture content and percent of insect and fungal damaged grains after five months in storage

At the end of five months in storage, the moisture content of maize increased between 12.77 to 14 % with significant differences among the storage methods but within the recommended range for safe storage of maize (Table 5). The wet monsoon season in August could have increased the humidity around the atmosphere and led to an increase in moisture content. Similarly, the wet monsoon season possibly increased the humidity in the air and resulted in grains absorbing more moisture to have zero physiological weight losses. The highest degree of insect infestation was observed in maize from traditional storage samples at 2.24 % followed by 1.5 % in curing and storage shed. The insect infestation of 0.21% and 0.35 % for silo and super bag was the lowest among the storage treatments. Fungal damaged grains were in the range of 0.07 % to 0.11 % for super bag, silo and curing and storage shed while it was not observed for traditional storage samples. The overall storage losses to insect and fungal infestation are between 0.28 % to 2.24 %. After five months of storage, the overall storage losses were highest at 2.24 % for the traditional storage method. From the data, it can be seen that all three storage types resulted in lower overall storage losses compared to the traditional method and if stored after proper drying, grains could be stored well without significant quantity losses for five months.

Table 5. Moisture content and percent of insect and fungal infected grains after five months under different storage methods

Storage method	Moisture content (%)	Insect infested grains (%)	Fungal infected grains (%)	Overall storage losses (%)
Traditional	13.93a	2.24a	0.0b	2.24
Super bag	13.60b	0.35c	0.08a	0.43
Silo	12.77c	0.21c	0.07a	0.28
Curing & storage shed	14.00a	1.50b	0.11a	1.61
S.E.	0.15	0.25	0.0.01	-
P- value	0.000	0.000	0.01	

Means in the column with different letters are significantly different between storage methods at $P < 0.05$ by Duncan's post-hoc test.

3.6 Average moisture content of maize grains over five months storage period

The average moisture content of maize grains over the five months storage period in different storage methods is shown in Figure 1. The highest moisture content of 12.67 % was recorded in grains from super bag followed by 12.45 % for the traditional method and 12.41 % for silo storage. It was observed that the more enclosed the storage type, the higher the moisture content. The lowest average moisture content during the five months storage period was in

curing and storage shed maize grains at 12.29 %. The low moisture content in the curing and storage shed is due to the higher ventilation of the shed leading to higher moisture removal from the grains. The moisture content in general increased during the storage period for the maize from all types of storage methods due to the increase in relative humidity during the monsoon season. The moisture content of maize remained with minimal change in some of the hermetic containers while it decreased in the woven bag and aerated containers over seven months of storage (Baributsa, Bakoye, Ibrahim, & Murdock, 2020). It was observed that all the storage methods maintained the moisture level at the recommended range during the five months of storage duration. The moisture content of maize for storage is recommended to be between 12-14% (FAO, 1992). The moisture content of 13 % is also recommended for maize storage in the extension manual on quality maize seed production through the community-based seed production approach in Bhutan (Katwal, Dorji, & Wangdi, 2009).
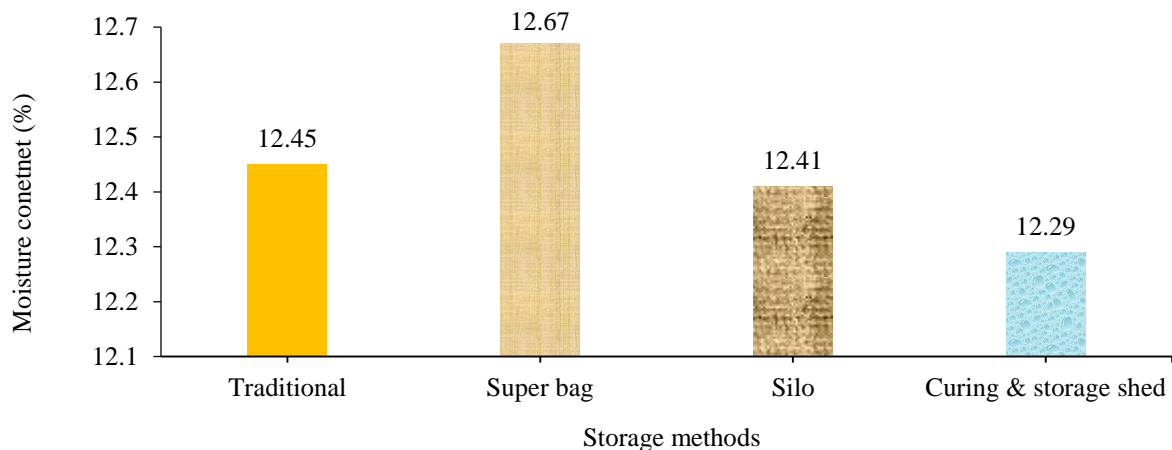

Figure 1. Average (mean) moisture content (%) of maize grains stored under different storage methods over the five months storage period

3.7 Average physiological weight loss over four months storage period

The four-month average physiological loss of weight was the lowest at 0.10 % for the maize grain stored in the super bag (Figure 2). The lowest physiological weight loss from the super bag is directly linked to the high moisture content of grains reported for this grain sample. The barrier property of the super bag probably led to minimal water loss from the grains resulting in lower physiological weight loss. The highest PLW of 0.30 % was observed from the grain stored in the curing and storage shed due to adequate ventilation that caused moisture removal. Traditional storage and silos resulted in average PLW of 0.20 % and 0.16 %, respectively during the four-month storage period.
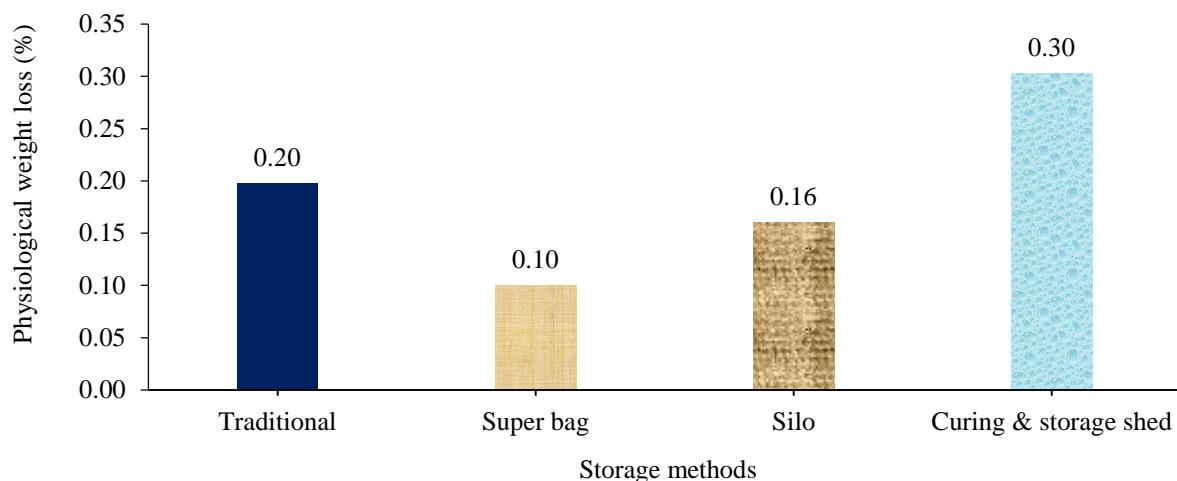
Figure 2. Mean physiological loss of weight (%) of maize grains stored under different storage methods over the four months of the storage period

3.8 Overall storage losses

After five months in storage, the highest overall insect damage of 2.54 % **was** observed in the maize stored in the traditional storage method followed by 1.79 % for the maize grains from curing and storage sheds (Figure 3). The insect damages were 0.64 % and 0.27 % for the super bag and silo storage, respectively. The higher damages in traditional, curing and storage shed could be due to the good airflow that is required for insects to thrive while the slightly enclosed nature of maize in the silo and super bag deterred insects to an extent. Fungal damages, in general, were quite low for maize grains from all the storage methods (0.19 to 0.31 %). It is believed that proper drying of maize and bringing the moisture to recommended levels before storage helped in keeping the fungal and mould growth to a very minimal level. The overall storage losses to insect and fungal damages were highest for traditional storage method grains at 2.80 % followed by 1.98 % for curing and storage shed while super bag and silo stored grains recorded 0.95 % and 0.51 %, respectively after five months of storage. In general, the overall damage of maize grain was observed quite low irrespective of storage methods for the five-month storage period. Based on the findings from this study, maize grains harvested in February (late winter to early spring) could be stored without maximum losses to insect and fungal damages in all kinds of storage methods if grains are harvested at the right maturity stage and properly dried before storage and if only good quality grains are put into storage.
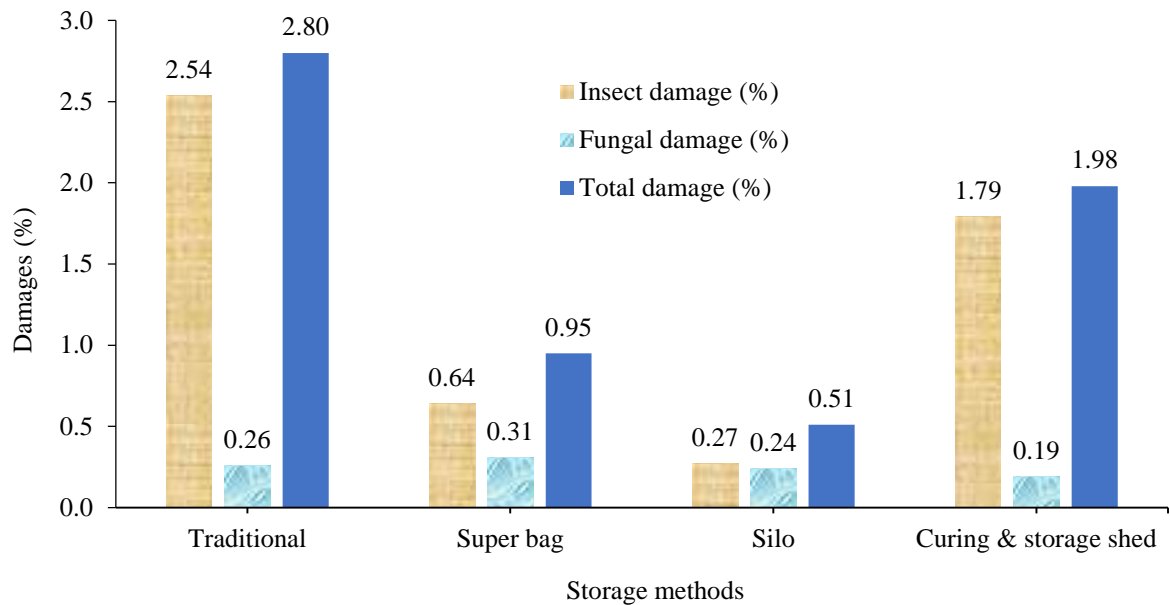
Figure 3. Total maize grain damage by insects and fungi throughout the five-month storage period under different storage methods

The trend for overall storage losses of grains over five months storage period is shown in figure 4. The overall storage losses of grain were slightly higher from the traditional storage method except for months three and four where super bag recorded slightly higher overall storage loss. Silo storage recorded the least overall storage loss during the storage period.
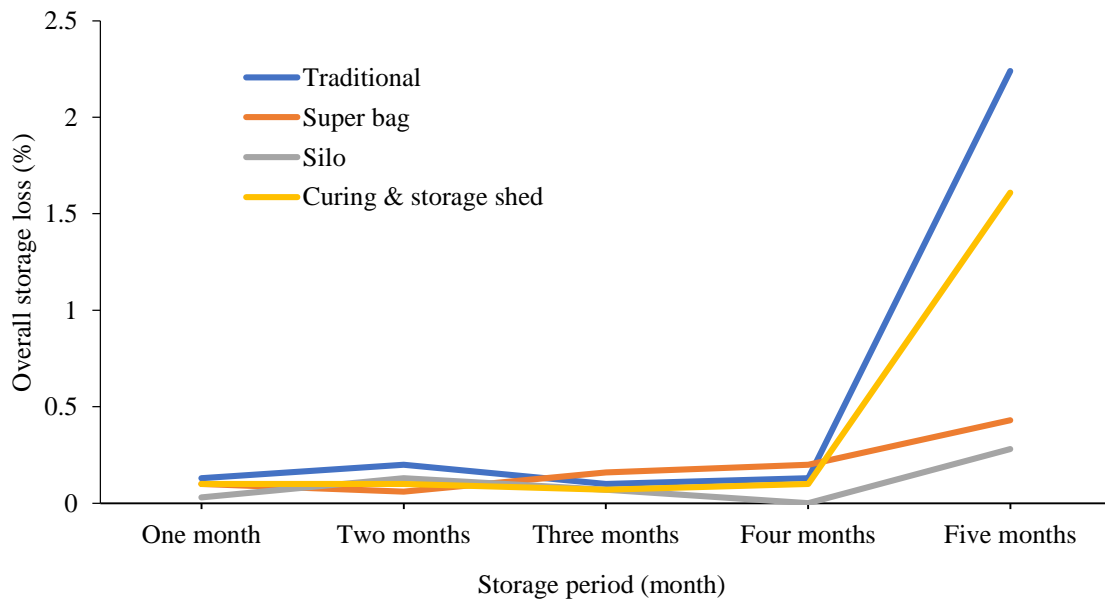

Figure 4. Overall storage losses of grains under different storage methods over the five months storage period

4. Conclusion

The moisture content of the grains remained within the recommended range for safe storage in all the storage methods over the five month storage period. Physiological weight loss was slightly higher for grains stored in curing and storage shed and traditional method while lower physiological weight loss was observed in grains stored in the super bag. Generally, insect damages were higher than damages from fungal diseases except for silo storage in some months. The lowest insect damage was reported in grains from silos while curing and storage shed reported the lowest fungal damage. The minimum overall storage losses were observed in the grain from silo storage compared to the highest that was recorded for the traditional method of storage after five months in storage. Silo could be useful for maize storage if placed under cool conditions away from direct sunlight and heat and grains. All the storage methods performed similarly in storing the grains for five months without alarming differences in losses. The storage losses in maize could be significantly reduced by adopting the four different storage methods. However, adequate awareness programs have to be instituted on these approaches like proper drying of maize grains before storage and keeping the storage bags, silos and sheds in the cool condition away from direct heat and sunlight. To validate these findings, it is recommended that a repeat of the study be carried out to evaluate these storage methods for the maize crops harvested and stored during summer.

References

Baributsa, D., Bakoye, O. N., Ibrahim, B., & Murdock, L. L. (2020). Performance of five postharvest storage methods for maize preservation in Northern Benin. *Insects, 11*(8), 541. doi:https://doi.org/10.3390/insects11080541

Basappa, G. D. (2004). *Post-Harvest Losses of Maize Crop in Karnataka An Economic Analysis.* (PhD Dissertation), University Of Agriculture Sciences GKVK, Dharwad, Dharward.

Bhandari, G., Achhami, B. B., Karki, T. B., Bhandari, B., & Bhandari, G. (2015). Survey on maize post-harvest losses and its management practices in the western hills of Nepal. *Journal of Maize Research and Development, 1*(1), 98-105. doi:https://doi.org/10.3126/jmrd.v1i1.14247

Dorji, K., Tshering, D., & Lhamo, S. (2020). Assessment of Storage Losses of Maize in Three Districts of Bhutan. *Bhutanese Journal of Agriculture, 2*(1), 127-137.

FAO. (1992). *Maize in Human Nutrition*. Rome, Italy: Food and Agriculture Organization (FAO) of the United Nations.

Katwal, T. B., Dorji, L., & Wangdi, N. (2009). *Quality Maize Seed Production through Community Based Seed Production Approach: An Extension Manual*. Wengkhar, Mongar: Council for RNR Research of Bhutan, Ministry of Agriculture, Bhutan.

Kiaya, V. (2014). Post-harvest losses and strategies to reduce them. *ACF (Action Contre la Faim (ACF) Technical Paper on Postharvest Losses, 25*, 2-26.

Kumar, D., & Kalita, P. (2017). Reducing postharvest losses during storage of grain crops to strengthen food security in developing countries. *Foods, 6*(1), 8. doi:https://doi.org/10.3390/foods6010008

NPHC. (2016). *Study on post-harvest storage losses of maize*. Paro, Bhutan: National Post Harvest Centre (NPHC), Department of Agriculture, Ministry of Agriculture & Forests.

NSB. (2018). *Statistical Yearbook of Bhutan 2018*. Thimphu: National Statistics Bureau (NSB), Royal Government of Bhutan.

PPD. (2015). *Bhutan RNR statistics 2015*. Thimphu: Policy and Planning Division (PPD), Ministry of Agriculture and Forests, Royal Government of Bhutan

RSD. (2020). *Bhutan RNR Statistics 2019*. Thimphu: Renewable Natural Resources Statistics Division (RSD), Ministry of Agriculture and Forests, Royal Government of Bhutan.

Shrestra, S., Katwal, T. B., & Ghalley, B. B. (2006). *Adoption and impact assessment of improved maize technologies in Bhutan*. Thimphu: Council of RNR Research of Bhutan, Ministry of Agriculture.

Suleiman, R., A.,, & Kurt, R., A.,. (2015). *Current Maize Production, Postharvest Losses and The Risk Of Mycotoxins Contamination In Tanzania*. Paper presented at the 2015 ASABE Annual International Meeting, St. Joseph, MI. https://elibrary.asabe.org/abstract.asp?aid=46136&t=5

Zorya, S., Morgan, N., Diaz Rios, L., Hodges, R., Bennett, B., Stathers, T., Mwebaze, P., & Lamb, J. (2011). *Missing food: the case of postharvest grain losses in sub-Saharan Africa*. Washington, D.C.: The International Bank for Reconstruction and Development/The World Bank.

Effect of Planting Methods on Grain Yield and Crop Management in Finger Millet

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ABSTRACT

A field experiment on two planting methods (direct seeding and transplanting) in finger millet was conducted using two cultivars; Samtenling Memja 1 (SM1) and IE4425 at the Agriculture Research and Development Centre (ARDC), Samtenling from July to December 2020. The study aimed to determine the best planting method that gave better yield and economic advantage in crop production. The results of the experiment showed that the transplanted finger millet recorded the highest yield (0.81 Mt ha^{-1}) compared to directed seeded (0.65 Mt ha^{-1}) but statistically not significant. There was a percent yield difference of 19.7 % between the two methods. Similarly, there was no significant effect between planting methods and yield components such as plant height, productive tillers, and finger numbers; but there were varietal significant effects on plant height and length of the fingers (P=0.00). However, a significantly shorter maturity duration was observed in direct-seeded millet (120 days) as compared to transplanted millet (126.5 days). Moreover, economic analysis indicates that net returns for direct seeding were considerably greater (>25%) than that of the transplanting method. Based on the study, direct-seeded finger millet could be promoted in farmers' fields considering the economic advantage and early maturity.

Keywords: *Finger millet, Planting methods; Days to maturity; Grain yield; Economic analysis*

1. Introduction

Finger millet (*Eleusine coracana* L) is generally a small-seeded cereal known for its high nutritive value. It is a staple food for the tribal and lower-income groups (Kumar, Tomer, Kaur, Kumar et al., 2018). Generally, finger millet is the only millet that occupies the largest area under cultivation among other small millets. The global millet production was estimated at 28.4 million metric tons in 2019, with India being the largest global producer with 41.0% global market share followed by Africa (FAO, 2019). In Bhutan, millets are cultivated over an area of 7,313.45 hectares (ha) with a production of 1,240.45 Mt and average productivity of 0.0058 Mt ha^{-1} (RSD, 2019). The global recorded accession of finger millet is about 25,707 and Bhutan has around 84, which constitutes 0.33 % of the total accessions (Sood et al., 2016). Even though

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finger millet is categorized as a neglected and underutilized species (NUS) (Chadha & Olouch, 2006; Kahane, Hodgkin, Jaenicke, Hoogendoorn et al., 2013) and it is grown for its superior nutritional properties and minimal inputs (Gupta et al., 2017). A unique feature of finger millet is that it has the highest productivity among millets due to its resilience and adjustability to adverse agro-climatic conditions (Seetharam, 2006) as a result of its C4 photosynthetic pathway (Wafula, Korir, Ojulong, Siambi et al., 2016). It tolerates salinity better than most cereals (CABI, 2003). Finger millets are cultivated for strengthening nutritional security rather than for food security purposes directly (Puranik, Kam, Sahu, Yadav et al., 2017). Globally, finger millets are cultivated both in irrigated and dry land. However, only dryland cultivation is practised in Bhutan hitherto.

Bhutan is a mountainous country where the arable land is only about 2.83%, i.e., 1,08,534 ha from a total area of 38,394 km^2 (RSD, 2019). Farm mechanization is limited and traditional farming still predominates. Traditional farming is laborious and entails more drudgery. Moreover, labour shortage is one of the main farming constraints in the country. According to the report by the Department of Agriculture (DoA, 2016) and Dendup and Chhogyel (2018), about 53% of the farming constraint in Bhutan is accounted for by farm labour shortage. Planting methods vary among farmers according to their choice of where the crop is cultivated. The most common practice is to transplant where nurseries are raised by broadcasting the seeds. The transplanting method is the dominant method of finger millet establishment in all of Asia (Pandey, 1995). Despite transplanting being a major traditional method for raising millets, the economic factors and recent changes in millet production technology have provided an increased impetus to direct seeding methods (Pandey & Velasco, 2005).

The direct seeding method is defined as the seeding method which involves the sowing of seeds directly into the soil where the plants are let to eventually mature (Bareja, 2021). Plants bear no transplanting stresses, and the crop is seen to develop faster. The primary economic motives for switching to direct seeding are to reduce the labour cost and explore the possibility of crop intensification. According to Mortimer, Riches, Mazid, Pandey et al. (2008), the major forces driving the spread of direct seeding methods were the rising cost of agricultural labour, the need for intensifying crop production, the development of high-yielding short-duration modern varieties, and the availability of chemical weed control methods that largely promoted this change as evidenced in Malaysia and Thailand in the late 1980s and 1990s. Direct seeding offers the advantage of faster and easier planting, reduced labour and less drudgery, and 7–10 days' earlier crop maturity (Balasubramanian & Hill, 2002). Moreover, Pandey (1995) has also

reported that in developing countries, direct seeding is adopted because of the migration of farm labour to nonfarm jobs and the consequent shortage of labour and high wages. Accordingly, the rise in population pressure, scarcity of water and agricultural land, and the continuing shortages of labour will continue to pressure a shift toward direct-seeding methods.

De Datta and Flinn (1986) reported that Asian farmers are shifting to direct seeding given the primary advantage that the crop can be established in time so that better crop stands can be achieved for higher productivity. With the same available farm power and labour, much larger acreage can be brought under cultivation in much less time through direct seeding. A similar advantage was reported by Balasubramanian and Hill (2002). The productivity of the direct-seeded crop is found at par with those raised through the transplanting method while the net profit obtained was higher (Y. Singh, Singh, Johnson, & Mortimer, 2005). However, the drawbacks of the direct-seeded method are the lack of uniformity in crop density and the difficulty at times in undertaking intercultural operations. One of the most critical challenges reported in the direct-seeded method is weed stresses. Yet, if the weed pressure is managed well, farmers could still obtain yields comparable to the transplanting method (Rana, Al Mamun, Zahan, Ahmed et al., 2014).

Among agronomic practices, planting methods are one of the important operations to realize higher productivity. Suitable planting methods and selection of improved cultivars play a critical role in exploiting the yield potential of the crop under agro-climatic conditions. Among the possible technological options in finger millet farming, direct seeding has immense potential, and this could be promoted. In Bhutan, finger millet is mostly transplanted, and direct seeding is not popular. This could be largely due to a lack of knowledge and the absence of proper scientific documentation on the different planting methods available. Therefore, it is important to study the effect of planting methods on finger millet varieties under Bhutan's sub-tropical conditions. The objectives of this study were to compare grain yield from different planting methods with two varieties for promotion as new technology and to assess the comparative advantage of the two methods in terms of labour input and cost.

2. Materials and Method

2.1 Evaluation/trial site

Field experiment was conducted at ARDC, Samtenling, in Sarpang (Figure 1). The site is located at 26^0 54'-14' N latitude and $90^0 26'$-20' E longitude. It falls under the wet sub-tropical agro-ecological zone of Bhutan by latitude between the elevations (375 <600masl),

temperature (max 35°C, min 12°C), and rainfall (2,500-5,500mm). The average monthly precipitation is 1,032mm with a relative humidity of 86.13% (NCHM, 2020). The texture of the soil at the experimental site is sandy loam. The soil is inherently infertile with gentle slopes and good drainage for crop production. Crop grown includes rice, maize, groundnuts, sesame, millets, and oilseeds.
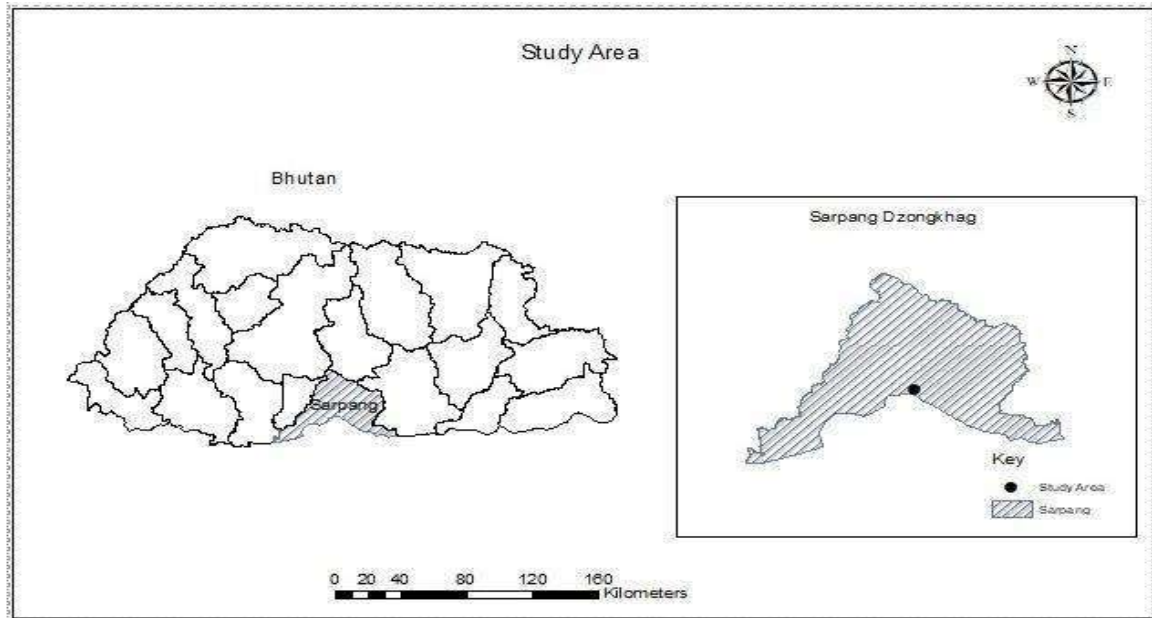

Figure 1. Study site (ARDC research station) in Sarpang

2.2 Evaluation methods

The experiment was laid out in a split-plot design with three replications. The main plot factor consisted of two planting methods (direct-seeded and transplanting) and the sub-plot factor consisted of two varieties *viz.,* SM 1 and IE 4425. This experiment had a total of twelve plots covering an area of 75 m^2. Slope/vertical gradient was a blocking factor. The plot size of 2.25 m x 3 m per treatment was used. The distance between blocks and interspacing for the individual plot was 0.5 m. A spacing of 0.2 m row to row and 0.1 m plant to plant were maintained. Farmyard manure (FYM) at the rate of 5 t ha^{-1} was applied to all the experimental plots uniformly. A fertilizer dose of 40:20:20 NPK kg ha^{-1} was applied. Nitrogen was applied in a split dose (0.352 kg) as basal along with 1.012 kg of phosphorus and 0.27 kg of potassium. Another 0.352 kg of nitrogen was applied 35 days after sowing (DAS) in direct-seeded plots and 30 DAT in transplanted plots. The nursery was raised during the first week of August and transplanted in the third week of August 2020. To suppress the weed pressure, two hand-weeding for direct-seeded plots were performed at 25 DAS and 40 DAS, and for the

138

transplanted plot at 25 DAT and 40 DAT, respectively. The crop received a total recorded rainfall of 2,645 mm during the study period.

2.3 Data collection

The data were recorded adopting standard procedure using UPOV (International Union for the Protection of New Varieties of Plants) guidelines for finger millet. The data observations on yield attribute *viz.* plant height, number of tillers, days to 50% flowering, number of fingers, leaf shape, length of flag leaf, days to maturity, plot yield, and harvest index (%) were recorded. Five plants were randomly selected to measure plant height which was recorded before harvesting while maturity days were calculated from the day of sowing until the grains attended physiological maturity. The harvested plant samples were dried for a week before threshing and threshed grains were weighed using a digital weighing balance. Grain and straw yields were calculated based on the yield obtained from each net plot and converted to t ha^{-1}. Before recording the straw yield, the plot-wise bundles of straw were sun-dried for a week to remove excess moisture.

1. Harvest index was calculated as per the formula given below, (Huhn, 2008)

$$H.I. = Economic\ yield\ /\ biological\ yield \times 100$$

Where, Economic yield = grain weight (g)
 Biological yield = Total plant yield (g)

2.4 Cost of Production

Cost of production and economic returns were estimated as per the existing farmer's wages. The cost for millet grains and straw per kg was taken at the existing farmer's rate. A man-day or working hours was assumed to be eight hours a day, based on which the calculations were made. Cost of cultivation was done considering the management practices including land preparation, weeding, thinning, and transplanting.

2.5 Statistical analysis

Data were collected and entered in MS Excel and analyzed at a 5% level of probability and were subjected to analysis of variance (ANOVA) using STAR (Statistical Tool for Agricultural Research) version 2.0.1 and IBM SPSS Statistics version 22. The results were expressed in tabular form by generating response surfaces.

3. Results and Discussion

3.1 Effect of planting methods on grain yield

The results (Table 1) show that planting methods had no significant effects on most of the growth attributes of finger millet such as plant height, number of fingers, flag leaf length, and number of productive tillers. However, planting methods had a significant effect on finger length and days to maturity. Finger millets raised from the transplanting method recorded a higher yield at 0.81 t ha^{-1} whereas finger millet from direct seeding showed a low yield (0.65 t ha^{-1}) with a percent yield difference of 19.7 % between the two methods. The SM 1 produced an average grain yield of 0.83 t ha^{-1} compared to IE 4425 with 0.63 t ha^{-1}. The percent yield difference between the two varieties was 24%. Similarly, the highest straw yield was found in SM 1 under the transplanted method (1.60 t ha^{-1}) and the lowest in the direct seeded method in the variety IE 4425 (1.3 t ha^{-1}). The higher yield obtained in the transplanted method could be attributed to the cumulative effect of increased tiller production due to the combined effects of abiotic factors such as light, temperature, relative humidity, bright sunshine hours coupled with optimum day length that possibly led to increased photosynthesis efficiency. This in turn could have contributed to increased dry matter production.

Gavit, Rajemahadik, Bahure, Jadhav et al. (2017) have reported that the increased yield attributes in the transplanted method might be due to increased growth and plant development. Nigade, Bagade, and Bhilare (2020) and Hebbal, Ramachandrappa, and Thimmegouda (2018) have reported higher millet yield in the transplanted method as compared to the direct-seeded method. The higher yield could be ascribed to the involvement of increased weed and other management practices, which also agree with the findings of Rana et al. (2014). In addition, Fayisa, Welbira, and Bekele (2016) reported similar results where the increase in spacing led to an increase in tillering, panicle number, and grain yield. On the contrary, the low yield in the direct-seeded crop may be due to the slow initial growth rate as seeds were broadcast, followed by weed stresses later. This might also be due to the adverse effect of competition between plants associated with closer spacing, as well as the poor yield from those that ended in scattered stands. Nonetheless, the broadcast method is considered one of the best planting options compared to drill and spot planting as it returns a higher yield. These results align with those of Adeyeye, Ahuchaogwu, Shingu, Ibirinde et al. (2014) and Shinggu and Gani (2012).

The interaction effects between the planting method and varieties were found to be non-significant (P=0.48). This indicates that there was no combined effect of planting methods and

varieties on grain and straw yield. The non-significant differences in other yield attributes between the planting methods further reveal that these methods have the potential for sustainable millet production by skipping nursery to transplanting operations that involve cost. If the management practices are applied correctly, any of these methods can produce a credible millet crop. Therefore, a direct-seeded method can be adopted and promoted for a sustainable millet production system.

Table 1. Effect of different planting methods on finger millet grain yield

Treatments	Grain yield (tha^{-1})	Straw yield (tha^{-1})	Days to maturity	Harvest Index (%)
Planting methods (Main plot)				
Direct seeding	0.65	1.30	120.00	30.90
Transplanted	0.81	1.60	126.50	49.43
S. E*	0.06	0.10	0.45	2.67
CD* (*P*=0.05)	0.25	0.02	0.00	0.00
Varieties (Sub plot)				
SM 1	0.83	1.57	123.50	43.81
IE 4425	0.63	1.33	123.00	36.52
S. E	0.09	0.15	0.64	3.77
CD (*P*=0.05)	0.17	0.76	0.59	0.20
Interaction (Main x Sub plot)				
S. E	0.13	0.21	0.91	5.34
CD (*P*=0.05)	0.48	0.19	0.13	0.75

Note: S. E*: Standard Error, CD*: Critical Difference (P-value at 5% probability)

3.2 Effect of planting methods on days to maturity

Crop phenology and maturity duration are some of the most important agronomic parameters in all millet growing ecosystems. Transplanted millet involves uprooting and replanting seedlings that directly expose them to physical and mechanical stresses, which in turn requires days to recover before the plants can perform normal physiological functions like any other growing plant. Transplanting therefore is associated with transplanting injury, hardening, and increased crop period leading to a longer duration for crop maturity.

The results (Table 1) of the study reveal that planting methods had a significant effect on days to maturity (*P*=0.00). However, there was no significant effect of variety on days to maturity and no statistical interaction effects for both the treatments. The direct seeded record (120 days) compared to transplanted millet (126.5 days). This may be due to the ability of the plant in a direct-seeded method to germinate easily and establish earlier as they are devoid of the stresses that transplanted millets are subjected to. This result agrees with the findings of Dendup and Chhogyel (2018). It could also be due to various factors such as root depth, nutrient use

efficiency, weed pressure, and inter-crop competition. A difference of 6.5 days in maturity days is observed between the two planting methods.

Alterations in crop maturity and plant stature offer new cropping system opportunities. Early maturity is an excellent drought escape mechanism in the drought-prone finger millet growing areas. Many farmers around the world prefer crops with environmental adaptability that demonstrate yield stability with early maturing traits in cultivars that help mitigate erratic rainfalls and abiotic stresses (Asrat, Yesuf, Carlsson, & Wale, 2010). Thus, the direct-seeded millet can be harvested early, thereby providing a sufficient time window for the following crop. Generally, direct-seeded finger millets suffer from intensive weed pressure if intercultural operation is delayed. Fufa and Mariam (2016) have reported that there was an 82% yield reduction from weedy plots which in turn delayed the physiological growth and maturity period. In a mountainous country like Bhutan where cropping periods are rather short, maturity duration is more important since the crop has to fit in within a single growing period (Dendup & Chhogyel, 2018). Hence, the crop should be sown in time to optimize its maximum yield potential.

3.3 Effect of planting methods on finger millet plant height

Our analysis (Table 2) revealed that the plants did not show any significant differences in their heights under the different planting methods (P=0.24). However, a significant height difference was observed between the two varieties (P=0.02). The mean plant heights recorded in transplanted and direct-seeded millets were 82.85 cm and 79.15 cm, respectively. Taller plant height in transplanted millet could be due to the deeper root system as seedlings were planted into the soil directly, while the direct-seeded millet is sown at a surface leading to a shallow and reduced root system. This also conforms to the findings of Naresh, Misra, and Singh (2013) who reported taller plant heights in transplanted rice compared to those directly seeded. The transplanting method is always associated with better moisture utilization, nutrient supply, and optimum growth condition during nursery leading to better performance in growth parameters, including taller plant heights. Similar results were also reported by Hebbal et al. (2018).

Plant height is a central part of the plant ecological system that strongly correlates with its life span, seed mass, and time to maturity. It is also a major determinant of a plant's ability to compete for light and is directly correlated to the yield of a crop (Moles, Warton, Warman, Swenson et al., 2009). An increase in plant height is always advantageous as it intercepts more light resulting in increased dry matter production per unit area. Transplanted millet is

associated with uniform and wider spacing compared to the direct-seeded method, and its interaction effect with desirable genotype results in taller plant growth. The results of this present investigation are further substantiated by similar findings by Kalaraju (2007) and Nandini and Sridhara (2019) in foxtail millet. Kalaraju, Kumar, Nagaraja, and Ningappa (2009) have also noticed that the increased plant height and larger number of tillers lead to more leaves, thereby resulting in increased straw yield in pearl millet. Further, the interaction effect between planting methods and varieties was also found to be non-significant (P=0.91). This indicated that there was no combined effect on plant height. The taller plant height, however, could be attributed to the varietal effect where the height differences were also observed between the two planting methods.

Table 2. Effect of different planting methods on plant height and growth parameters

Treatments	Plant Height (cm)	Productive tiller hill^{-1}	No. of finger earhead^{-1}	Length of finger (cm)	Length of flag leaf (cm)
Planting methods (Main plot)					
Direct seeding	79.15	2.65	4.00	6.83	22.66
Transplanted	82.85	3.16	4.33	8.00	25.16
S. E	1.47	0.18	0.14	0.20	0.75
CD (P=0.05)	0.24	0.21	0.28	0.02	0.13
Varieties (Sub plot)					
SM 1	85.00	3.16	4.66	8.16	24.16
IE 4425	77.00	2.66	3.67	6.67	23.66
S. E	2.07	0.26	0.20	0.28	1.07
CD (P=0.05)	0.02	0.21	0.00	0.02	0.75
Interaction (Main x Sub plot)					
S. E	2.93	0.37	0.28	0.40	1.15
CD (P=0.05)	0.91	0.66	0.28	0.06	0.46

3.4 Effect of planting methods on productive tillers per plant

The results (Table 2) of this study revealed no significant difference (P=0.21) between the planting methods on the productive tillers hill^{-1}. Nevertheless, a greater number of productive tillers was observed in the transplanted method (3.16) compared to the direct-seeded method (2.65). Statistically, the interaction effects between planting methods and varieties on productive tiller numbers were non-significant (P=0.66). The varietal effect on productive tillers per hill was greater compared to the planting methods. The transplanting method usually involves a larger space, distributed uniformly in that it helps in the effective utilization of available resources such as land, light, and nutrients. The greater number of productive tillers

in the transplanted method could be attributed to the larger space between the plants. Wider spacing and more nutrient availability in the transplanting method contribute to a greater number of tillers per plant as also observed by Kalaraju (2007). Sarawale, Rajemahadik, Shendage, and Mane (2016) reported that tillering in cereal grains could be induced by transplanting shock as well as through wider spacing between the individual plants. This is consistent with the results obtained by Maobe, Nyang'au, Basweti, Getabu et al. (2014) where transplanted finger millet had higher tiller formation which in turn influenced productive panicle formation and increased the overall grain yield. Similar results were obtained by Awan, Ali, Safdar, Ashraf et al. (2007), and Sakadzo, Bvekwa, and Makaza (2019).

On the other hand, direct-seeded millet recorded a lower number of productive tillers (2.65 tillers/hill) which could be due to higher weed pressure during the initial growth stages. This conforms with the earlier works of V. P. Singh, Singh, Singh, Kumar et al. (2008). The higher number of productive tillers in transplanted finger millet could be due to proper spacing and uniformity which was not the case in direct-seeded millet. In transplanted millet, there was no overcrowding of seedlings, and weed pressure was considerably less. Hebbal et al. (2018) also reported similar findings. Therefore, we can safely conclude that germinated seeds when sown directly perform well and are comparable to transplanted millet which normally produces a higher number of productive tillers.

3.5   Effect of planting methods on finger number per earhead of finger millet

Among the two planting methods used, transplanted finger millet resulted in a greater number of fingers per earhead than the direct-seeded millet, although the results were not statistically significant (P=0.28). A higher number of fingers were produced in the transplanting method (4.33) as compared while direct-seeded millets (4.00). However, there was a significant effect (P=0.00) on the number of fingers per earhead between the varieties (Table 2). Variety SM 1 produced a higher number of fingers per earhead (4.66) than IE 4425 (3.67). These results are also corroborated by earlier findings of Kumari and Singh (2015) who reported that the greater the productive tillering, the more would be the finger numbers. Further, similar results were also observed in the research work of Gavit et al. (2017) who evaluated the effect of establishment techniques and sowing time on yield and yield attributes of Proso millet. This is also supported by the results obtained by Hebbal et al. (2018) in India where they showed that transplanted finger millet recorded higher finger numbers as compared to the direct-seeded method.

3.6 Effect of planting methods on finger length per earhead of finger millet

Finger length is one of the important yield parameters in finger millet. The panicle length is positively correlated to the number of grains per plant, and ultimately to the grain yield of the crop (Chandan, 2018). Our results (Table 2) showed that the interaction effects between the planting method and the length of fingers were non-significant *(P=0.06)*. Longer finger lengths were observed in transplanted millets (8.00 cm), while direct-seeded recorded a mean finger length of 6.86 cm. Moreover, SM 1 (8.16 cm) recorded a greater number of finger lengths than IE 4425 (6.67 cm). Statistically, the planting method and variety had a significant effect on the finger length *(P=0.02)*, but their combined interaction effect did not. The main effect of planting methods on the finger length was more in transplanted SM 1 millets than in direct-seeded SM 1 millets. Similar results were obtained for the IE 4425 variety as well. As indicated earlier, this can be due to the higher moisture and larger space available in the transplanted method that aid in the efficient use of water, air, and nutrients, resulting in better growth and development. This is supported by the findings of Sakadzo et al. (2019) and Michaelraj and Shanmugam (2013) where mean finger length was recorded as the highest in transplanted millets as against their broadcast counterparts. A study by Tamilmozhi, Karthikeyan, Sakthivel, and V Ravichandran (2020) on the influence of seedling age, planting pattern, and the number of seedlings per hill on the growth and yield of finger millet under Tamil Nadu conditions in India also supports these results.

3.7 Labour requirement and cost of production

The field operations considered for this study include nursery development, field preparation, planting, weeding, and thinning. These all incur costs. The ultimate objective of any agricultural technology is to realize the maximum returns per Ngultrum (Nu.) invested. Any farming technology to be adopted under farmer conditions should be economically viable.

Table 3. Man-day's requirement and cost of labour for different planting methods per hectare

Cultivation practice	Unit	Planting methods of Finger millet	
		Direct Seeded	Transplanting method
Nursery development	(Man-days)	0	6
Seedling Uprooting	(Man-days)	0	5
Field preparation	(Man-days)	6	6
Seed sowing	(Man-days)	1.5	0
Transplanting (manual)	(Man-days)	0	24
Seedling thinning	(Man-days)	4	0
Weeding (Hand weeding)	(Man-days)	16	10

Total labour required	(Man-days)	27.5	51
Labour cost incurred	(Nu/head)	450	450
Total costs	(Nu)	12,375/-	22,950/-
Cost percent advantage against transplanting method	%	46%	

Table 4. Gross return, net return, and cost-benefit ratio analysis for different planting methods per hectare

Particulars	Planting methods	Quantity (t ha^{-1})	Rate kg^{-1} (Nu.)	Amount (Nu.)
Produce/Grains (t ha^{-1})	1	0.6	30/-	18,000
	2	0.8	30/-	24,000
Straw (t ha^{-1})	1	1.3	5/-	6,500
	2	1.6	5/-	8,000
Gross returns (A)	1			24,500
(Nu.)	2			32,000
Cost of production (B)	1			12,375
(Nu.)	2			22,950
Net returns (A-B)	1			12,125
(Nu.)	2			9,050
C: B ratio	1			1:1.98
	2			1:1.39
Profitability (%) against transplanting				25%

Note: 1* = Direct seeded, 2*= Transplanted method

To examine the labour requirement and undertake cost analysis, the main field operations such as seedling thinning, and weeding was considered. Management practices other than seedling preparation and transplanting were the same for both methods. The study results showed that the transplanting method in finger millet requires 51 man-days due to the additional manpower required for nursery, seedling uprooting, and transplanting operations (Table 3). Transplanting alone required 24 man-days, while these activities were not necessary for the direct-seeded crop. Thus, direct seeding showed a drastic reduction in labour requirements. The transplanting method required higher labour resulting in a higher cost of cultivation. The results (Table 4) show that the gross returns (Nu. 32,000/-) and total cost of production (Nu.22,950/-) was higher in the transplanted method whereas net returns (Nu. 12,125/-) and cost-benefit ratio (1: 1.98) was higher in the direct-seeded method. This confirms the findings of Hebbal et al. (2018) who reported that transplanting methods of planting recorded the highest cost of production and gross returns compared to the direct-seeded method in finger millet under Bangaluru conditions in India.

There was a labour cost difference of Nu. 10,575/- per ha between transplanted and direct-seeded methods. Additionally, the cost advantage of the direct-seeded method against the transplanting method was as high as 46%. The direct seeded method again recorded a higher net profit of 25%. Similar findings were made by (Y. Singh et al., 2005) who argued that the productivity of the direct-seeded crop is at par with the transplanting method and the net profit remains higher as well. Gill, Walia, and Gill (2014) also reported that direct seeding reduced the cost of production by about 9%. Many farmers still adopt the transplanting method since the high labour inputs were often offset with higher yields. Many studies have reported that the direct seeded method is more economical than the transplanted method (Bhardwaj, Singh, Singh, & Singh, 2018; Jaiswal, Pradhan, Kumar, Sharma et al., 2020; Naresh et al., 2013) due to the minimal costs involved. Direct-seeding method of planting, therefore, offers potential for millet production with fewer labour requirements.

4. Conclusion

The study indicated that the direct seeding method in millet is a potential alternative approach to millet cultivation that could be promoted in the country. Though the growth parameters of millet *viz.,* plant height, tiller number per hill, number of fingers per plant, and grain yield were found to be insignificant between the planting methods, it has a numerical difference in grain yield and other yield attributes. Based on the yield and yield attributes, the transplanting method is worth adopting. However, based on the cost advantage and number of labourers required for different cultivation practices, direct-seeding was found to be better since it required less labour. This contributed to the reduction in labour cost, and in turn, enhanced the profitability (25%) of millet farming. Direct-seeded millet required 27.5 man-days compared to 51 man-days in transplanted millet. Thus, there is an additional cost difference of Nu. 10,575/- in transplanted millet. Based on the overall results, the transplanted method which is labour-intensive and associated with higher cost can be replaced by direct seeding without compromising the productivity of the crop. The direct-seeded method could be, therefore, recommended for finger millet production for optimum growth, higher yield, and early maturity at a lower cost of production. This study recommends the direct-seeding method in millet cultivation as a potential technology to be incorporated as a part of the strategy to overcome farm labour shortage and reduce costs in finger millet farming.

References

Adeyeye, A., Ahuchaogwu, C., Shingu, C., Ibirinde, D., Musa, G., & Technoledge. (2014). Germination and establishment of finger millet variety (*Eleusine coracana*) as affected by planting method. *The International Journal of Science, 2*(9), 110.

Asrat, S., Yesuf, M., Carlsson, F., & Wale, E. (2010). Farmers' Preferences for Crop Variety Traits: Lessons for On-farm Conservation and Technology Adoption. *Ecological Economics, 69*(12), 2394-2401. doi:https://doi.org/10.1016/j.ecolecon.2010.07.006

Awan, T. H., Ali, I., Safdar, M. E., Ashraf, M. M., & Yaqub, M. (2007). Economic effect of different plant establishment techniques on rice, *Oryza sativa* production. *Journal of Agricultural Research, 45*(1), 73-81.

Balasubramanian, V., & Hill, J. (2002). Direct Seeding of Rice in Asia: Emerging Issues and Strategic Research Needs for the 21st Century. In S. Pandey, M. Mortimer, L. Wade, T. P. Tuong, & H. B. Lopez K (Eds.), *Direct Seeding: Research Strategies and Opportunities* (pp. 15-39). Los Banõs: International Rice Research Institute (IRRI), pp. 15-39.

Bareja, B. G. (2021). Methods of planting crops in the farm. *Crop Farming Review*. http://www.cropsreview.com/planting-crops.html

Bhardwaj, R., Singh, M., Singh, C., & Singh, A. (2018). Efficacy of Crop Establishment Methods on Yield and Economics of Rice (*Oryza sativa L.*) under Puddled Condition. *International Journal of Current Microbiology and Applied Sciences, 7*(7), 3625-3631.

CABI. (2003). *Crop protection compendium*. Oxfordshire: CAB International.

Chadha, M., & Olouch, M. (2006). *Healthy Diet Gardening Kit-For Better Health and Income.* Paper presented at the I International Conference on Indigenous Vegetables and Legumes. Prospectus for Fighting Poverty, Hunger and Malnutrition 752.

Chandan, K. (2018). *Performance of Finger Millet (Eleusine coracana L.Gaertn) Under Different Crop Establishment Methods*. Ranchi, India: Birsa University.

De Datta, S. K., & Flinn, J. C. (1986). *Technology and Economics of Weed control in Broadcast-seeded Flooded Tropical Rice.* Paper presented at the Weeds and the environment in the tropics. Proceedings of the 10th conference of the Asian–Pacific Weed Science Society, Chiang Mai, Thailand.

Dendup, C., & Chhogyel, N. (2018). Ngawang. Effects of Different Planting Methods on Rice (*Oryza sativa L.*) Crop Performance and Cost of Production. *Bhutanese Journal of Agriculture, 1*(1), 13-22.

DoA. (2016). *Agriculture Statistics 2016*. Thimphu: Department of Agriculture (DoA), Ministry of Agriculture and Forests, Royal Government of Bhutan

FAO. (2019). *FAOStat: Agriculture Data*. Rome: Food and Agriculture Organization of the United Nations Statistics Division.

Fayisa, B. A., Welbira, G. D., & Bekele, D. A. (2016). Determination of optimum rates of nitrogen and phosphorus fertilization for finger millet (*Eleusine coracana* L. Gaertn) production at Assosa Zone. *Benishangul-Gumuz Region of Ethiopia. Advances in Sciences and Humanities, 2*(1), 1-6.

Fufa, A., & Mariam, E. G. (2016). Weed Control Practices and Inter-Row Spacing Influences on Weed Density and Grain Yield of Finger Millet (*Eleusine coracana* L. Gaertn) in the Central Rift Valley of Ethiopia. *International Journal of Research in Agriculture and Forestry, 3*(9), 1-7.

Gavit, H., Rajemahadik, V., Bahure, G., Jadhav, M., Thorat, T., & Kasture, M. (2017). Effect of Establishment Techniques and Sowing Time on Yield and Yield Attributes of Proso Millet (*Panicum miliaceum* L.). *International Journal of Current Microbiology and Applied Sciences, 6*(5), 1523-1528. doi:https://doi.org/10.20546/ijcmas.2017.605.166.

Gill, J. S., Walia, S. S., & Gill, R. S. (2014). Direct seeded rice: An alternative rice establishment technique in north-west India–A review. *International Journal of Advanced Research, 2*(3), 375-386.

Gupta, S. M., Arora, S., Mirza, N., Pande, A., Lata, C., Puranik, S., . . . Kumar, A. (2017). Finger Millet: a "Certain" Crop for an "Uncertain" Future and a Solution to Food Insecurity and Hidden Hunger Under Stressful Environments. *Frontiers in Plant Science, 8*, 643. doi:https://doi.org/10.3389/fpls.2017.00643

Hebbal, N., Ramachandrappa, B., & Thimmegouda, M. (2018). Yield and Economics of Finger Millet with Establishment Methods under Different Planting Geometry and Nutrient Source. *Indian Journal of Dryland Agricultural Research and Development, 33*(1), 54-58. doi:10.5958/2231-6701.2018.00010.6

Jaiswal, P., Pradhan, A., Kumar, M., Sharma, G., & Singh, D. (2020). Effect of Crop Establishment Methods on Yield of Rice (*Oryza sativa* L.) during Summer under Lowland Farming Situation. *Int.J.Curr.Microbiol.App.Sci, 9*(10), 581-590. doi:https://doi.org/10.20546/ijcmas.2020.910.069

Kahane, R., Hodgkin, T., Jaenicke, H., Hoogendoorn, C., Hermann, M., Hughes, J. d. A., Padulosi, S., & Looney, N. (2013). Agrobiodiversity for Food Security, Health and Income. *Agronomy for Sustainable Development volume, 33*(4), 671-693. doi:https://doi.org/10.1007/s13593-013-0147-8

Kalaraju, K. (2007). *Response of Finger Millet Genotypes to Methods of Planting under Organic Farming.* (M.Sc Thesis), University of Agricultural Sciences GKV, Bangalore, Retrieved from http://krishikosh.egranth.ac.in/handle/1/5810097872

Kalaraju, K., Kumar, N., Nagaraja, N., & Ningappa, K. B. (2009). Effect of Methods of Planting on Growth and Yield of Finger Millet Genotypes under Organic Farming. *Research on Crops, 10*(1), 20-24.

Kumar, A., Tomer, V., Kaur, A., Kumar, V., & Gupta, K. (2018). Millets: a Solution to Agrarian and Nutritional Challenges. *Agriculture & Food Security, 7*(1), 1-15. doi:https://doi.org/10.1186/s40066-018-0183-3

Kumari S, Singh KS (2015) Assessment of genetic diversity in promising nger millet [Eleusine coracana (L.) Gaertn] genotypes. Suppl Genet Plant Breed 10(2):825-830

Maobe, S., Nyang'au, M. K., Basweti, E., Getabu, A., Mwangi, T., & Ondicho, A. (2014). Effect of Plant Density on Growth and Grain Yield of Finger Millet (*Eleusine coracana*) Under High Potential Conditions of Southwest Kenya. *World Journal of Agricultural Sciences, 10*(6), 261-268. doi: 10.5829/idosi.wjas.2014.10.6.1743

Michaelraj, P., & Shanmugam, A. (2013). A Study on Millets Based Cultivation and Consumption in India. *International Journal of Marketing, Financial Services & Management Research, 2*(4), 49-58.

Moles, A. T., Warton, D. I., Warman, L., Swenson, N. G., Laffan, S. W., Zanne, A. E., Pitman, A., Hemmings, F. A., & Leishman, M. R. (2009). Global Patterns in Plant Height. *Journal of Ecology, 97*(5), 923-932. doi:https://doi.org/10.1111/j.1365-2745.2009.01526.x

Mortimer, A., Riches, C., Mazid, M., Pandey, S., & Johnson, D. (2008). *Direct Seeding of Rice and Weed Management in the Irrigated RiceWheat Cropping System of the Indo-Gangetic Plains*. Los Baños, Philippines: International Rice Research Institute.

Nandini, K., & Sridhara, S. (2019). Response of Growth Yield and Quality parameters of Foxtail Millet Genotypes to Different Planting Density. *Int.J.Curr.Microbiol.App.Sci., 8*(2), 1765-1773. doi: https://doi.org/10.20546/ijcmas.2019.802.208

Naresh, R., Misra, A., & Singh, S. (2013). Assessment of Direct Seeded and Transplanting Methods of Rice Cultivars in the Western Part of Uttar Pradesh. *International Journal of Pharmaceutical Sciences and Business Management, 1*(1), 1-8.

Nigade, R. D., Bagade, S. V., & Bhilare, R. L. (2020). Ensuring Economic Gains due to Different Dates of Sowing on Finger Millet Varieties (*Eleusine coracana* L.). *International Journal of Current Microbiology and Applied Sciences, 9*(5), 2487-2492. doi:https://doi.org/10.20546/ijcmas.2020.905.284

Pandey, S. (1995). Socio-economic research issues on wet seeding In K. Moody (Ed.), *Constraints, opportunities and innovations for wet-seeded rice* (pp. 73-84). Los Baños: International Rice Research Institute (IRRI).

Pandey, S., & Velasco, L. (2005). Trends in Crop Establishment Methods in Asia and Research Issues. In K. Toriyama, K. L. Heong, & B. Hardy (Eds.), *Rice Is Life: Scientific Perspectives for the 21st Century*. Los Baños: International Rice Research Institute (IRRI).

Puranik, S., Kam, J., Sahu, P. P., Yadav, R., Srivastava, R. K., Ojulong, H., & Yadav, R. (2017). Harnessing Finger Millet to Combat Calcium Deficiency in Humans: Challenges and Prospects. *Frontiers in Plant Science, 8*, 1311. doi: https://doi.org/10.3389/fpls.2017.01311

Rana, M. M., Al Mamun, M. A., Zahan, A., Ahmed, M. N., & Mridha, M. A. J. (2014). Effect of Planting Methods on the Yield and Yield Attributes of Short Duration Aman Rice. *American Journal of Plant Sciences, 5*(3), 251-255. doi:10.4236/ajps.2014.53033

RSD. (2019). *RNR Census of Bhutan*. Thimphu: Renewable Natural Resources Statistics Division (RSD), Ministry of Agriculture and Forests, Royal Government of Bhutan.

Sakadzo, N., Bvekwa, G., & Makaza, K. (2019). Influence of Establishment Methods on Yield and Yield Parameters of Finger Millet (*Eleusine Coracana* L. Gaertn) in Ward 24, Zaka District. *Agricultural Science, 1*(1), 47-56. doi:https://doi.org/10.30560/as.v1n1p47

Sarawale, P., Rajemahadik, V., Shendage, G., & Mane, S. (2016). Effect of Different Varieties and Establishment Methods on Growth and Yield of Finger Millet [*Eleusine Coracana (L.) Gaertn.*] under Condition. *J. Indian Soc. Coastal Agric. Res., 34*(2), 22-26.

Seetharam, A. (2006). *Millets: Handbook of Agriculture*. New Delhi: Indian Council of Agricultural Research (ICAR).

Shinggu, C., & Gani, M. (2012). Effects of Planting Methods, Sowing Dates and Spacing on Weed and the Productivity of Finger Millet (*Eleusine corocana* L. Gaertn) in the Northern Guinea Savanna of Nigeria. *Global Journal of Bio-Science & Biotechnology 1*(2), 160-162.

Singh, V. P., Singh, G., Singh, S. P., Kumar, A., Singh, Y., Johnson, D. E., & Mortimer, A. M. (Eds.). (2008). *Direct seeding and weed management in the irrigated rice-wheat production system*: International Rice Research Institute/Directorate of Experiment Station, GB ….

Singh, Y., Singh, G., Johnson, D. E., & Mortimer, M. (2005). Trends in Crop Establishment Methods in Asia and Research Issues. In K. Toriyama, K. L. Heong, & B. Hardy (Eds.), *Rice is Life: Scientific Perspectives for the 21st Century*. Los Baños: International Rice Research Institute (IRRI).

Sood, S., Kumar, A., Babu, B. K., Gaur, V. S., Pandey, D., Kant, L., & Pattnayak, A. (2016). Gene discovery and advances in finger millet [*Eleusine coracana* (L.) Gaertn.] genomics—an important nutri-cereal of future. *Frontiers in Plant Science, 7*, 1634. doi:https://doi.org/10.3389/fpls.2016.01634

Tamilmozhi, R., Karthikeyan, R., Sakthivel, N., & V Ravichandran, V. (2020). Influence of seedling age, planting pattern and number of seedlings per hill on the growth and yield of finger millet. *International Journal of Chemical Studies, 8*(5), 1126-1130. doi:10.22271/chemi.2020.v8.i5p.10445

Wafula, W. N., Korir, K., Ojulong, H., Siambi, M., & Gweyi-Onyango, J. P. (2016). Finger Millet (*Eleusine coracana* L.) Grain Yield and Yield Components as Influenced by Phosphorus Application and Variety in Western Kenya. *Tropical Plant Research, 3*(3), 673-680. doi:10.22271/tpr.2016.v3.i3.088

Analysis of Head Rice Recovery using Different Types of Rice Mills for Two Rice Varieties Grown in Two Extreme Rice Growing Altitudes

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ABSTRACT

There have been issues of increased broken rice when farmers finally sell their rice crop, especially in the higher altitudes, and this has always been attributed to the quality of milling machines used. In this study, the head rice recovery of two rice varieties grown in high and low altitudes in Bhutan was assessed in both pre-and post-milling through manually peeling and milling in four different types of rice milling machines, respectively. The head rice recoveries on manual peeling of high and low altitude rice varieties were 54.00±0.41% and 83.68± 0.45%, respectively, and were significantly different at P<0.05. Grain crack percentages were 29.44±0.45% and 5.37±0.45%, respectively, indicating that the climatic conditions had some influence on crack development and head recovery of rice. For machine milling, the rice head recoveries were statistically significant both between varieties and among the milling machines used. The head yield was higher in low attitude variety compared to that of the high altitude one. The friction type machine with 3.32 m/s peripheral velocity gave lower head yield compared to friction type of 1.2 m/s and rubber roller type I and II milling machines. This study recommends improving the drying method presently practised in high altitudes by not laying the paddy on the ground after harvest to avoid exposure of harvested paddy to extreme day and night temperature fluctuation. The use of lower peripheral speed rice milling machines and rubber rollers is recommended to increase head yield.

1. Introduction

Bhutan is located in the eastern Himalayas between China in the north and India to its east south and west. It has altitudes ranging from 200 masl in the south to more than 7500 masl in the north. Rice (*Oryza sativa* L.) is the main staple crop of the Bhutanese with a per capita available for consumption computed at 130.51 kg/year as of 2019 (Bajgai, Lakey, Phuntsho, Om et al., 2021). It is grown in altitudes from 200 to 2500 masl, thus having two extreme rice cultivation altitudes in the country. The rice self-sufficiency in the country for the year 2019 is estimated at 34.71% (Bajgai et al., 2021). Rice in Bhutan is cultivated under both irrigated and

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rain-fed systems (Chhogyel, Ghimiray, Wangdue, & Bajgai, 2015). Rice production in Bhutan was recorded at 83,332 t and 86,385 t in the year 2016 and 2017 respectively (RSD, 2018). To achieve 60% rice self-sufficiency in the 12th Five Year Plan (FYP), the Department of Agriculture is assertively promoting rice cultivation in high altitude areas and during spring seasons across the country (Karchung, 2017). Most rice varieties are composed of roughly 20% rice hull, 11% bran layers and 69% starchy endosperm, also referred to as the total milled rice (Das, Saha, & Alam, 2016).

The use of machines both during paddy cultivation and processing of harvested rice is important. The mechanization in rice cultivation has progressed more than for any other crops with the maximum coverage in milling. There are comparatively very high benefits of the machine mills over the traditional method using mortar and pestle, in terms of the capacity, drudgery, total milling and in reducing the operation cost. As a result, milling rice using machines have almost completely replaced the traditional method. More than 3000 rice mill sets of the Engelberg screw type and a few de-husker cum polisher have been supplied to different parts of Bhutan. Engelberg brand machine imported from India is dominant in the country due to their low cost and the same model has prevailed for the last four decades. However, the rice-growing countries that used the Engleberg hullers previously, have now shifted to more sophisticated milling systems due to the high breakage of paddy grain, resulting in very poor rice recovery. However, even today these single-pass mills are found widely in operation in countries like Bangladesh and other African nations (Alam, 2005).

The milling losses were not considered by farmers as actual losses since the by-products constituting the husk, bran and the broken rice exiting the milling machine together, can be ultimately used as animal feed. Therefore, the necessity to introduce a milling machine with a higher head rice recovery was not considered. It is estimated that the 9 % of the total post-harvest losses in rice crops which range from 15 to 16 % is primarily due to the use of old and outdated methods of drying and milling, improper and unscientific methods of storage, transport and handling (Mejía, 2004). Of late, higher head rice recovery capacity of milling machines is given added importance due to the local rice fetching a premium price. The higher head rice recovery has become important, especially for the export market. Increasing efficiency in processing as an important aspect of post-production technology in field crops has been emphasised in the Renewal Nature Research policy of Bhutan (CoRRB, 2012). Many rice growers have also concluded machines as the primary cause behind the higher rates of broken rice without clear studies ever being conducted.

The Agriculture Machinery Centre (AMC) has been putting in concerted effort to assess the milling efficiencies of different rice milling machines from different countries to identify and recommend suitable ones for the farmers. One of the efficiency parameters being looked into is the head rice recoveries during milling. Using the same machines, the head rice recoveries vary considerably from region to region as well as among the rice varieties. The causes of more broken rice in the colder region compared to that in the warmer parts need to be explored using the same machine types. Although no studies have been conducted until recently to determine the causes for the low percentage of head rice and overall low head yield recoveries, these have been prejudicially attributed to the qualities of the machines.

The atmospheric temperature during the harvesting season of paddy varies a lot between the two altitudes. In colder areas, especially in Paro, the temperature fluctuations between warm days and chill nights are quite high. The average maximum temperatures in Paro for October and November are $18.7^{o}C$ and $13.9^{o}C$, respectively and the mean minimum temperatures for the same months are $7.4^{o}C$ and $1.4^{o}C$, respectively. Observations reveal that traditionally after harvesting, paddy is left out in the field for a few days for natural drying. The dried paddy is then stacked in the field and threshing is done a few months later. It is also noticed that rice kernels are exposed to the temperatures stress of fluctuating hot days and chill nights which leads to the development of cracks in the rice kernels during natural drying in the field, and these kernels with cracks get easily broken during milling. On the other hand, there is no report on such cases of broken rice from southern Bhutan. Therefore, this study was taken up to investigate the various causes of broken rice.

2. Materials and Method

2.1 Paddy sample and pre-milling recovery

Two rice varieties from two extreme rice growing altitudes in Bhutan were collected for the experiment. One variety (Yusi Ray Kaap) from high altitude area of Paro Dzongkhag at an elevation of 2250 masl and another variety (Kalo Bhog) from the low elevation area of Sarpang dzongkhag located at 350 masl were used in this study. The dried samples collected from farmers were further sun-dried and brought to a milling moisture content of 12.0%. To check the physical conditions of the grains before milling, the samples comprising 900 grains each were selected randomly, and their husks manually peeled off. The brown rice was then examined for cracks, broken and yellowish colour formation with the help of a grain inspector. As a standard practice, the broken brown grains of lengths two-thirds and above are considered whole brown rice (BSB, 2018). Three replications were taken for each variety.

2.2   Milling machines and milling recovery

In the present study, four different rice milling machines were evaluated to estimate rice head yield. They were two imported friction type rice mills named Friction Type I and Friction Type II and rice mill with 2 and 3 rubber rollers and polishers together named Roller Type I and Roller Type II (Figure 1-3). The experimental design used was a randomized complete block design (RCBD) with three replications. The complete specification and dimensions of all rice mills were recorded by disassembling the machine parts.

Figure 1. Fiction type rice miller type I Type I & II

Figure 2. Rubber roller rice machine

Figure 3. Friction type rice milling type II

For each replication, 5 kg paddy samples were used and the time taken to complete milling was recorded. The samples for the laboratory tests were collected randomly during the replication operation by placing an empty plastic bag at regular intervals during the milling process.

Broken rice grains were observed using the grain grader (Sataka Co., Ltd) in the laboratory. The parameters of the milling process were calculated using the following equations:

$$Milling\ Capacity\ (\frac{kg}{h}) = \frac{Weight\ of\ sample\ paddy\ (kg)}{Milling\ time\ (h)} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(1)$$

$$Milling\ Recovery\ (\%) = \frac{Weight\ of\ Milled\ rice\ (kg)}{Weight\ of\ Sample\ paddy\ (kg)} \times 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(2)$$

$$Head\ rice\ recovery\ (\%) = \frac{Weight\ of\ milled\ head\ rice\ (kg)}{Weight\ of\ milled\ rice\ (kg)} \times 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots(3)$$

$$Head\ yield\ (\%) = \frac{Head\ rice\ recovery\ (\%) \times Milling\ recovery\ (\%)}{100} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots(4)$$

$$Milling\ degree\ (\%) = \frac{Weight\ of\ brown\ rice\ (1000\ nos) - Weight\ of\ milled\ rice\ (1000\ nos)}{Weight\ of\ brown\ rice\ (1000\ nos)} \times 100 \ldots\ldots(5)$$

2.3 Statistical analysis

Microsoft Excel spreadsheet (Microsoft Office 2010) was used to calculate the mean and standard deviation for both the pre-milling and post-milling conditions. An unpaired t-test was conducted between the two varieties of two altitudes for pre-milling parameter evaluation. A two-way ANOVA was run for four different machines and the two varieties. All the analyses for t-test and ANOVA were carried out using the data analysis tool package in MS Excel. A post-hoc test for separation of means was conducted after the ANOVA test.

3. Results and Discussion

3.1 Pre-milling recoveries

The head brown rice recovery for high and low altitude varieties were $54.00\pm0.41\%$ and $83.68\pm 0.45\%$, respectively on manual peeling. There was a significant difference at $P<0.05$ between the varieties. The crack percentage was $29.44\pm0.45\%$ and $5.37\pm0.45\%$ for high and low varieties, respectively as shown in Figure 4.

The above result showed that the rice grains had gone through too much stress and hence had huge cracks and were broken before milling in the machine, especially for the high-altitude variety. Factors like weather during harvesting and drying methods might have played a significant role in lowering the head rice recovery. Especially in Paro, located at a higher altitude, the crop was harvested in late October, and the paddy was laid out in the paddy field for natural drying, thereafter, exposing them to extreme weather conditions. It is a normal practice for harvesting and threshing paddy in Paro. Kunze (2008) found that the low moisture-containing grain (dried) reabsorbs moisture from any source having moisture to which it is exposed, and when compressive stresses at the surface exceed the tensile strength of the grain at the centre, a fissure develops. Most breakage in rice processing can be attributed to grains that were fissured before milling according to his study. Earlier studies also prove that the major factors responsible for fissuring are rice variety, management of post-harvest operations

and drying conditions which include drying methodology (Ban, 1971; Bautista, Siebenmorgen, & Cnossen, 2000; Cnossen & Siebenmorgen, 2000; Kunze, 1979; Kunze & Choudhury, 1972).
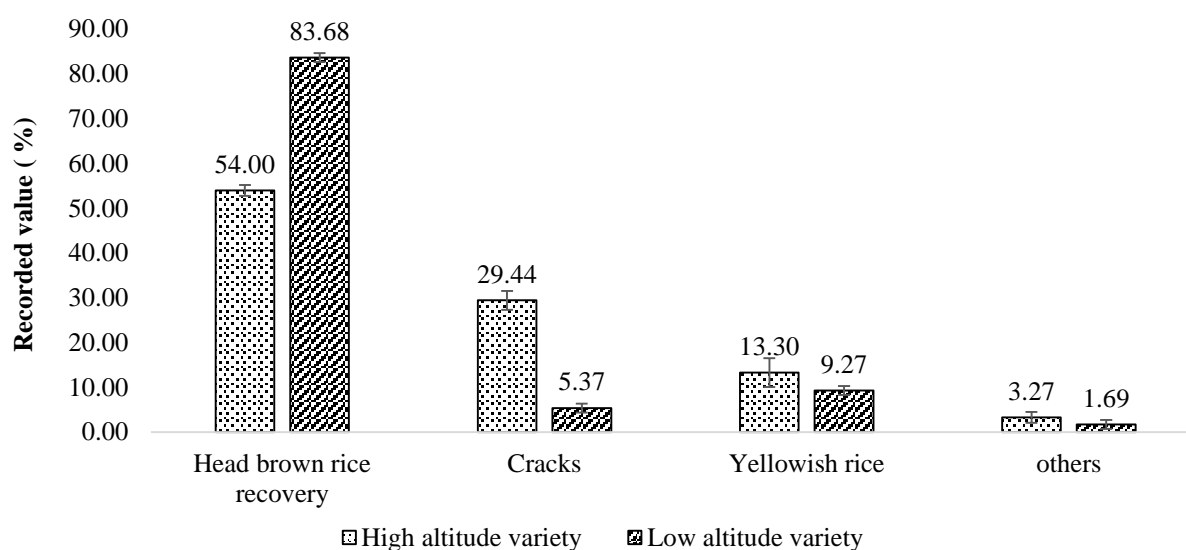

Figure 4. Characteristics of pre-milled rice on manual peeling. Error bars represent standard error of means.

3.2 Machines' physical structures

The dimensions of all the machines and their peripheral speeds recorded in the test are as in Table 1. The peripheral speed varied from 1.30 to 3.32 m/s among the machines. The higher the peripheral speed of the machines, the higher was the percentage of broken rice on milling.

Table 1. Specification of four different rice milling machines used in the experiment

Name of Rice mill	Milling shaft		Shaft shape	Screen shape	Clearance (mm)	Motor capacity (hp)	Peripheral speed (m/s)
	Diameter (mm)	Length (mm)					
Friction Type I	70	305	3 straight beads on shaft	Round	11	7.5	3.32
Friction Type II	48	186	4 straight beads on shaft	Round	5	3	1.31
2 Rubber rollers with polisher	25	145	2 helical beads on the shaft	Hexagonal	5	1	1.62
3 Rubber rollers with polisher	25	145	2 helical beads on the shaft	Hexagonal	5	1	1.62

3.3 Milling recovery

The milling recoveries indicated that fairly 30% of the paddy was lost during the milling process as husk and bran. Statistically, there were significant differences among different machines, between varieties and also their interactions at $P<0.05$. The percentage of milling

recovery was comparatively higher for low altitude variety compared to that of higher altitude variety in all machine types. The husk percentage was lower for the low altitude variety.

3.4 Head yield evaluation

The percentage of head yield obtained on the milling is shown in Figure 5. Statistically, there was a significant difference among different machines, between varieties and also their interactions at $P<0.05$ for the head yield.

The absence of broken or cracked features in grain is very important for the quality of rice. When the intact length of rice is more than ¾ of its whole length, then it is referred to as head rice and is the main criterion for rice quality (BSB, 2018). Das et al. (2016) also found that the Engleberg rice huller commonly used in Bangladesh had a 2% higher loss in whole rice over improved rice mills. In our case, it was more in percentage due to the milling degree which was used for whitening purpose. However, the loss percentage trend observed was similar to that reported in the Engleberg rice huller.
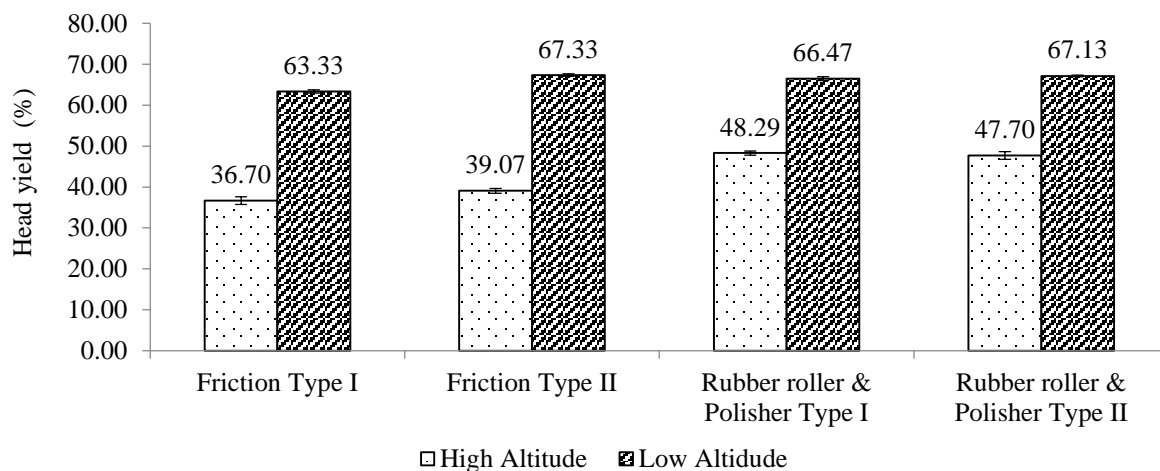
Figure 5. Rice head yield recorded in four different milling machines and two rice varieties grown at low and high-altitude areas (Error bars represent SEM)

Table 2. Unpaired t-test with Bonferroni correction with unequal means for variety and machine interactions

Treatment	Friction I x Friction II	Friction I x Roller I	Friction I x Roller II	Friction II x Roller I	Friction II x Roller II
High altitude rice variety	*	*	*	*	*
Low altitude rice variety	*	*	*	NS	NS

Each variety within the same column with single * is significantly different at $P\leq 0.05$ by unpaired t-test (n=3). NS represents statistically not significant.

Table 2 indicated that there were significant differences in high altitude variety for every milling machine tested. However, there was no significant difference at $P\leq0.05$ for low altitude rice variety among machines (Friction II x Roller I and Friction II x Roller II). This may be because the peripheral speed of Friction II which was made in Korea was less compared to Friction II which was Indian made. Higher peripheral speed affects the head rice recovery.

Table 3. Unpaired t-test with Bonferroni correction for each machine type on two varieties

Treatment	Friction rice mill Type I	Friction rice mill type II	Roller I (2 rollers with polisher)	Roller II (3 rollers with polisher)
High altitude & Low altitude rice varieties	*	*	*	*

The two varieties within rows with single * are significantly different at $P\leq 0.05$ by unpaired t-test (n=3).

There was a significant difference in the head recovery between the varieties grown in extreme altitudes. The analysis also showed that the two varieties had a significant difference in head rice recovery on each machine type tested at $P\leq0.05$. This might be that the grains were already stressed but were not observed during the pre-milling experiment and the damage was expressed only after the milling in respective machines.

4. Conclusion

The results from this study do not support our initial hypothesis of lower quality of machines being the main factor contributing to the high percentage of breakage in local rice. The head brown rice recovery for high and low altitude varieties were 54.00±0.41% and 83.68± 0.45%, respectively from the manual peeling experiment, indicating that other prominent factors resulted in this difference in head rice recovery. Statistically significant differences at $P<0.05$ between the varieties on manual peeling were observed. The crack percentages were 29.44±0.45% and 5.37±0.45% for high and low altitude varieties, respectively. Factors like weather during harvesting and drying methods might have played a significant role in lowering head rice recovery through the formation of grain fissures. At high altitudes, paddy was harvested in October and was laid out in the paddy field for natural drying, exposing them to daily extreme weather conditions. The machine with higher peripheral velocity of 3.32m/s had the lowest head yield recovery compared to other machines with lower peripheral velocities. The head yield was also higher for low altitude variety after milling in machines compared to high altitude variety. Thus, this study recommends changing the drying and threshing methods presently practised in Paro by harvesting and threshing simultaneously to avoid the adverse impact of cold weather. Further, the newly introduced and promoted combined harvesters may

have reduced the cases of broken rice which needs to be studied. The primary limitation of the present study is that only one variety of rice from colder and warmer regions was included for evaluation of broken percentage of rice and head rice recovery in the milling machines. The effect of different rice varieties on head rice recovery within the region was not evaluated. Hence, the influence of rice varieties on head rice recovery in two different altitudes needs to be studied in the future.

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References

Alam, M. M. (2005). *Identification of Agri-machinery SMEs and Service Provider, and Sub sector Supply and Value Chain Analysis*. Dhaka: South Asia Enterprise Development Facilities (SEDF).

Bajgai, Y., Lakey, L., Phuntsho, L., Om, K., ., Subba, M., ., Zangmo, C., Ghimiray, M., Zangmo, C., Tobgay, T., Yangzom, T., ., & Dukpa, W. (2021). *Self-Sufficiency and Dietary Energy Supply of Food Crops in Bhutan (A Status Report)* (L. Lakey & K. Om Eds. 1st ed.). Thimphu: Department of Agriculture, Ministry of Agriculture & Forests, Royal Government of Bhutan.

Ban, T. (1971). Rice cracking in high rate drying. *Japan Agricultural Research Quarterly, 6*(2), 113-116.

Bautista, R. C., Siebenmorgen, T. J., & Cnossen, A. G. (2000). *Fissure formation characterization in rice kernels using video microscopy.* Paper presented at the 12th International Drying Symposium, Noordwijkerhout, The Netherlands.

BSB. (2018). *Bhutan Standard Rice Mill - basic requirement (Part 1)*. Thimphu, Bhutan: Bhutan Standards Bureau (BSB)

Chhogyel, N., Ghimiray, M., Wangdue, K., & Bajgai, Y. (2015). Enhancing Bhutanese rice in the domestic market through increased production and favorable pricing regime. *Journal of Bhutan Studies, 32*, 26-44.

Cnossen, A. G., & Siebenmorgen, T. J. (2000). The glass transition temperature concept in rice drying and tempering: Effect on milling quality. *Transactions of the ASAE, 43*(6), 1661-1667. doi:10.13031/2013.3066

CoRRB. (2012). *Renewable Natural Resources Policy of Bhutan*. Thimphu, Bhutan: Council for Renewable Natural Resources Research of Bhutan (CoRRB), Ministry of Agriculture and Forests (MoAF).

Das, A. K., Saha, C. K., & Alam, M. M. (2016). Evaluation of a traditional rice husking mill. *Journal of Agricultural Machinery and Bioresources Engineering, 7*(1), 41-46.

Karchung. (2017). *High altitude paddy cultivation in Haa Dzongkhag*. Department of Agriculture, Ministry of Agriculture & Forests, Royal Government of Bhutan. Retrieved from www.moaf.gov.bt/High-altitude-paddy-cultivation-in-haa-dzongkhag

Kunze, O. R. (1979). Fissuring of the rice grain after heated air drying. *Transactions of the ASAE, 22*(5), 1197-1201. doi:https://doi.org/10.13031/2013.35183

Kunze, O. R. (2008). Effect of drying on grain quality-moisture readsorption causes fissured rice grains. *Agricultural Engineering International: CIGR EJournal, 10*, 1-17.

Kunze, O. R., & Choudhury, M. S. U. (1972). Moisture adsorption related to the tensile strength of rice. *Cereal Chemistry, 49*, 684-696.

Mejía, D. J. (2004). Rice post-harvest system: an efficient approach. In *International year of rice 2004*. Rome: Food and Agriculture Organization of the United Nations.

RSD. (2018). *Agriculture statistics 2017*. Thimphu, Bhutan: Renewable Natural Resources Statistic Division (RSD), Ministry of Agriculture and Forests, Royal Government of Bhutan.

Evaluation of Locally Fabricated Machine Attached to Tractor for Making Suitable Raised Bed for Vegetable Cultivation

Pem Lham[u], Zangmo[u], Kinga Norbu[u], Ugyen Phuntsho[u]

ABSTRACT

Growing vegetable on a raised bed improves soil physical parameters, and irrigation drainage and prevent waterlogging which ultimately increase the yield of vegetables. Making raised bed manually is not only time consuming but also a tedious job that involves much labour. A bed-making implement to be attached to a tractor machine was designed and tested to determine the dimensions of the bed formed specifically to suit the available plastic mulch width for cultivation of vegetable crops. A 34HP tractor was used as a power source for the bed making implement. The machine was tested at three different tractor forward speeds of 1.89, 2.54 and 5.04 km/h to record the width and height of the bed formed at these corresponding forward speeds. The results indicate that although the bed width increased with an increase in forward speed (76.89, 80.11 and 87.22 cm for 1.89, 2.54 and 5.04 km/h, respectively), the optimum bed width suitable for vegetable cultivation using the available plastic mulch was 80.11 cm with a bed height of 25.33 cm formed at the forward speed of 2.54 km/h. The machine field capacity recorded at the forward speed of 2.54 km/h was 3.80 acre/day, while that of a person's capacity to make the bed of the same dimension manually was 0.02 acre/day.

Keywords: *Bed making; Bed width; Bed height; Productivity*

1. Introduction

Although 49.9% of the population in Bhutan is engaged in agriculture (NSB, 2020), the country continues to import close to 30% of the food commodities from outside the country (Chhogyel & Kumar, 2018). The situation under the current Covid-19 pandemic has made it very clear that dependence on imports is not only unreliable but also highly risky. The country saw a major shortage of foods and vegetables during the lockdowns and their prices soared unrealistically. This has re-emphasized the importance of domestic food production to secure food security and self-sufficiency.

There is an urgent need to upscale the cultivation of vegetables in Bhutan as they are mostly perishable. Vegetable cultivation consists of various operations, the main one being field

preparation. Field preparation entails making beds of suitable heights to prevent waterlogging and improving aeration, thereby increasing production. The tops of the beds are wide with furrows on either side and furrows serve primarily as drains and traffic lanes for the water to flow to reduce deep drainage. Hamilton, Bakker, Houlebrook, and Spann (2000) showed that the raised beds were engineered to create a root zone that is about 30 centimetres deep with a stable structure and a low density. The dimensions, stability, and low density of raised beds ensure rapid absorption of rainwater, drainage of excess soil-water and improved aeration. Excess soil-water drains sideways from the beds into the furrows and from there it flows into catch-drains, waterways and dams preventing deep drainage. Cultivation of crops on raised beds not only facilitates proper drainage during the rainy season but also ensures better input management and improvement in soil physical conditions (Campbell, McConkey, Zentner, Selles et al., 1996).

In the bed planting system, vegetable crops are planted on raised beds (David, Jagadish, & Elizabeth, 2003). Ismail and Ismail (2013) indicated that the vegetables planted on the raised beds receive better growth conditions as it realizes low levels of groundwater and high infiltration. In the bed and furrow irrigation system, the plants are grown on raised beds which not only use irrigation water more efficiently but also ensure better crop growth (Berkout, Yasmeen, Maqsood, & Kalwij, 1997). The bed planting system facilitates weed control, allows mechanical cultivation of crops, and improves the stands of the plants (Miah, Hossain, Duxbury, & Lauren, 2015). It also provides an opportunity for easy field entry resulting from row orientation on the beds, efficient management of irrigation water, and also requires less labour (Fischer, Sayre, & Monasterio, 2005). This system has many other advantages such as reducing the seed rate, increasing crop yield, low water use, higher nitrogen use efficiency, and reduction in crop lodging as compared with the conventional tillage/sowing systems (Hobbs & Giri, 1997).

Raised bed-furrow planting method is adopted by many farmers throughout the world (Govaerts, Sayre, Lichter, Dendooven et al., 2007). In raised bed and furrow method, the fields are divided into narrow strips of raised beds separated by furrows as per the requirement of the crops. The raised bed planting system is being evaluated and advocated for many crops including cereals and vegetables in South Asia (Singh, Dwivedi, Shukla, & Mishra, 2010).

Generally, farmers in Bhutan grow vegetables on raised beds that are prepared manually without maintaining the recommended size due to the unavailability of machines. The size of

a raised bed has a significant impact on the total crop production and water consumption (Kukal, Sudhir, Humphreys, Amanpreet et al., 2010). Memon, Ullah, Siyal, Leghari et al. (2020) indicated that the size of a raised bed in the bed-furrow method has a significant effect on crop yield, water productivity, and salt distribution. The yield and crop water productivity were recorded higher under raised beds with a width of 80 cm. The optimal raised bed width for wheat in Koga (clay soil) is between 60 to 80 cm bed (Tewabe, Abebe, Enyew, & Tsige, 2020). Raised beds that are 81-91cm (32-36 inches) wide and 15-25 cm (6-10 inches) high are typically used for fresh market vegetable production (Clark & Maynard, 1992). Making raised bed manually is laborious, time-consuming, and expensive. Realizing the importance of raised beds for enhancing crop production and the difficulty in making beds of uniform size manually, this study designed and evaluated a bed-making implement attached on either side of the rotary tiller of the tractor for constructing raised beds of suitable size for the cultivation of vegetables using available plastic mulch films.

2. Materials and Method

2.1 Designing of the bed making implement

The bed making implement was designed to prepare a raised bed of heights 15-25 cm and 80 cm in width to facilitate easy access to the centre of the bed for weeding, crop and water management, and harvest. This bed width was made to suit the currently available mulching plastic film which is 120 cm wide and the provision of about 20 cm of plastic was kept for covering under the soil on either side of the bed to protect the mulch plastic against the wind. The frame and other working parts were made of a mild steel sheet of 3mm thickness and the parts were assembled with nuts and bolts to suit various dimensions. The process involved making the parts by cutting, grinding, and welding, followed by fitting and assembling. The prototype was tested in the field before this experiment.

The designed implement consisted of three units: (1) the front unit of width 110 mm connected by the shaft which is to be attached to the rotor of the tractor, (2) the rear unit (bed shaper) of width 80 mm (can be adjusted) which was made to form a raised bed of required width and height with sloping angle of 40°, and (3) a soil leveller unit attached to a roller shaft located between the two opposing sheets that slightly pressed the soil to level and compress the soil to form a flat and uniform bed top width. This unit could be adjusted according to the height and volume of the soil. The technical specification of the machine is presented in Table 1.

Table 1. Technical specifications of bed-making machine attached to tractor

S.N.	Particular	Dimension
1	Overall machine dimensions, (Length × Width × Height) (mm)	955×860 (rear) × 1070 (front) ×350
2	Power transmission	PTO (rotar) bed making attached to rotar
3	Width of furrow (mm)	500-600
4	Shaper	Flat sheet attached to roller shaft
5	Power (HP)	34 HP tractor
6	Field capacity (acre/day)	2.8 -3.8
7	Speed of operation (km/h)	1.89-2.54
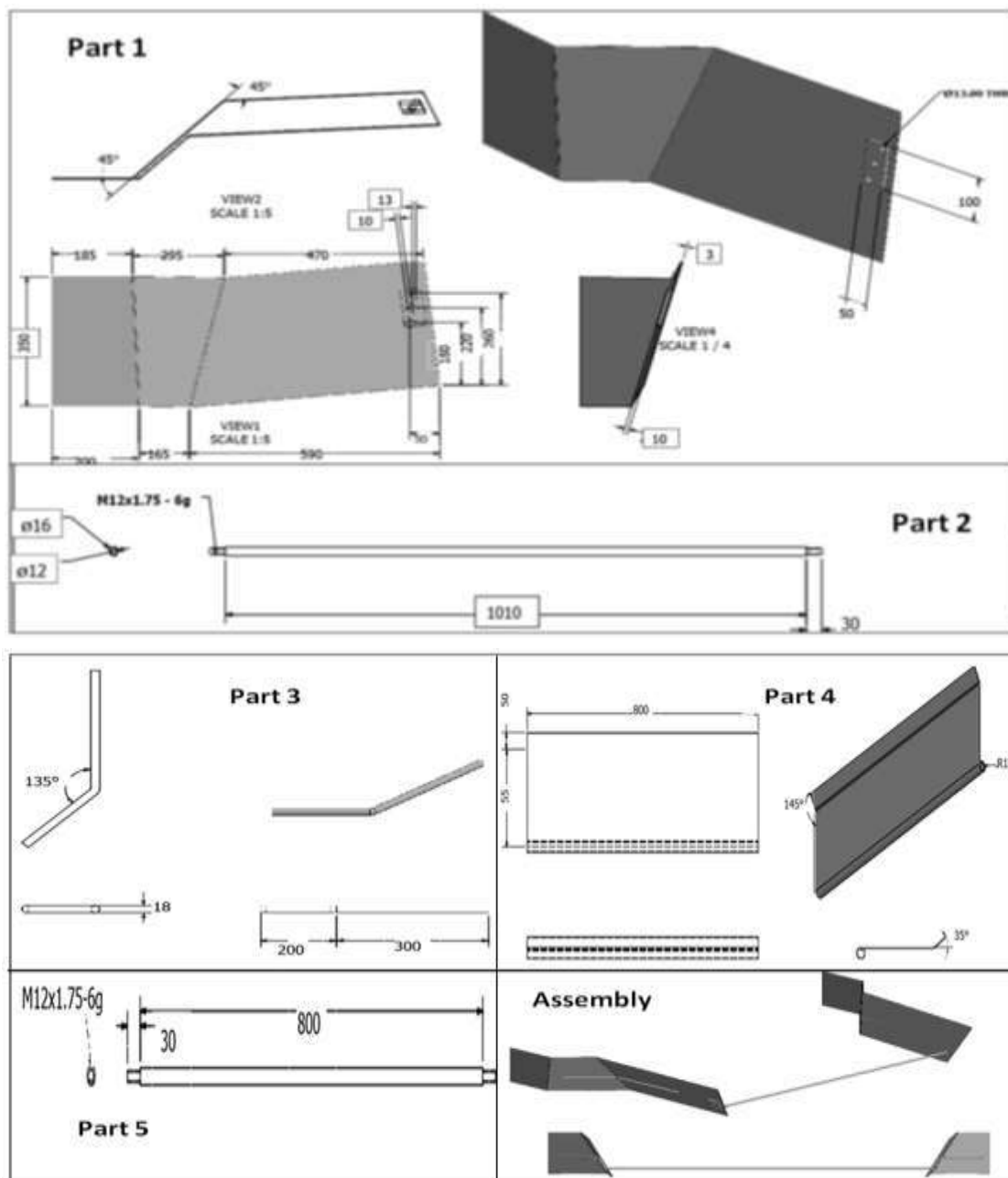
Figure 1. Schematic diagram of the machine design

2.2 Field experiment

The experiment was conducted in Genekha (27.3036° N, 89.6054° E), Thimphu, to evaluate the performance of the machine for making raised beds. The soil type at the site was sandy. The experiment was conducted adopting a completely randomized design (CRD) with three replications for each treatment. A total of nine plot units were selected for applying the treatments: i) forward speed of 1.89 km/h, ii) forward speed of 2.54k m/h, and iii) forward speed of 5.03 km/h for making the raised beds. The experiment was conducted in May 2021.

Samples of the field soil were collected to assess the soil moisture content. The machine was tested to determine the height of the bed, width of the bed and furrow width at three forward speeds which were found by recording the time taken to cover a distance of 30 m. The three forward speeds of 1.89, 2.54 and 5.03km/h at three different gear positions of 1, 2 and 3 respectively were calculated by using the following formula:

$$V = \frac{S \times 3.6}{t} \qquad (1)$$

Where;
V = forward speed (km/h); S = travelled distance (m); t = time of the experiment (sec);
3.6 = 1m/sec= 3.6 km/h

The theoretical field capacity (TFC), actual field capacity and field efficiency were also determined in testing the machine. Theoretical field capacity is the rate of field coverage of the implement, based on 100 per cent of the time at the rated speed and covering 100 per cent of its rated width. Theoretical field capacity was calculated using the following formula:

$$TFC = V \times W \qquad (2)$$

Where;
 TFC = theoretical field capacity (m^2/sec); V = forward speed (m/sec); W = rated width of implement (m)

Effective field capacity (Ef) is the total area covered/ total time taken to cover that area inclusive of the time lost time, etc.

$$Ef = \frac{A}{T} \qquad (3)$$

Where;

Ef = Effective field capacity (m²/sec); A = total area covered (m²); T = total time taken to cover that area (sec)

$$Fe = \frac{Ef}{TFC} * 100 \qquad (4)$$

Where;

Fe = Field Efficiency

2.3 Experiment with making bed manually

The separate experiment was conducted in a completely randomized design by three men with three replications each to find the time taken to make a raised bed of 25 cm in height, 80 cm wide and 10 m long in a well ploughed and pulverized field. The area coverage by each of them in a day (7 hours) was also calculated using the following formula.

$$HC = \frac{A}{T} * 7 \qquad (5)$$

Where;

HC = Human capacity (acre/day); A = Total area covered (acre); T = Total time taken to cover that area (h); 7 = Number of working hours in a day

2.4 Statistical Analysis

The data collected were subjected to analysis of variance (ANOVA) using the Statistical Package for Social Science (SPSS) version 21. Differences among treatment means were examined after statistical significance using the Duncan's Multiple Range Test (DMRT) at 5% significance level.

3. Results and Discussion

3.1 Effect of forward speed on the width of beds formed

Generally, the bed width and height were affected by the forward speed of the tractor at the same moisture content at 8%. Forward speed played an important role in the formation of optimum bed width. The result indicated that with increasing forward speed from 1.89 to 5.04 km/h, the bed width also increased from 76.89±1.35 to 87.22±3.8 cm respectively (Figure 2). There was no significant difference in bed width formed at forward speeds of 1.89 km/h and 2.54 km/h but the forward speed of 5.04 km/h was significantly different from the other two forward speeds.

A study conducted by Ismail and Ismail (2013) showed that increasing the forward speed increased the ridge profile upper width, but decreased the bed height since the increase in the

forward speed increases the collapsed soil, and consequently more quantity of soil falls on the profile sides. The bed width 76.89 cm and 80.11 cm formed at forward speeds of 1.89 and 2.54 km/h, respectively in the present study was comparable to the finding of Kukal et al. (2010) who also recommended a bed width of 80 cm for growing vegetable crops under different dry region for the long-term agricultural sustainability.
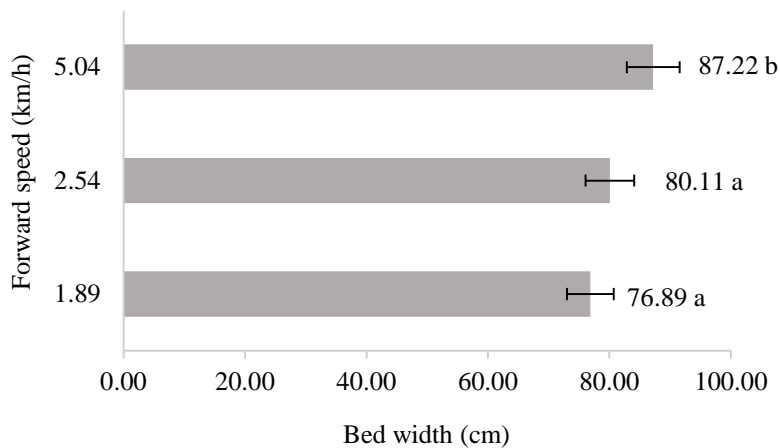

Figure 2. Effect of forward speeds on the width of planting bed formed (Error bars represent standard error of mean)

3.2 Effect of forward speeds on the height of bed formed

Bed heights of 25.33±1.86 cm and 25±2.00 cm were observed at forward speeds of 1.89 km/h and 2.54 km/h, respectively (Figure 3). The lowest bed height (15.56±1.68) cm was formed at the forward speed of 5.04 km/h which could have been due to more agitation of the soil at the higher speed of the tractor. There was no significant difference in bed heights formed by different forward speeds of the tractor.

Ismail and Ismail (2013) indicated that this trend was caused by the effect of the forward speed that prevents the forming unit share from penetrating the soil, thereby discouraging more soil layers from accumulating to form the ridge profile. In most situations, the bed height of 20 – 25 cm was found very effective (Wightman, Peries, Bluett, & Johnston, 2005). Therefore, the forward speed of 1.89 and 2.54 km/h is recommended to make a bed of optimum size which will also be suitable for laying plastic mulch film of 120 cm width available in the market currently. The higher speed forms a bed of lower height and wider width which is not suitable for plastic mulching.
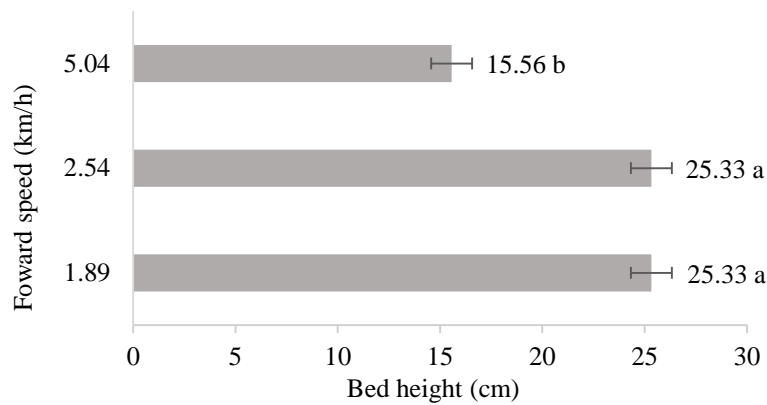
Figure 3. Effect of forward speeds on the height of bed formed (Error bars represent standard error of mean)

Figure 4. Bed-making machine attached to tractor

Figure 5. Bed making during field experiment

3.3 Bed-making machine productivity compared to bed making manually

A person took two hours to make a raised bed of height 25 cm, 80 cm wide and 30 m long covering an area of 0.02 acre/day inclusive of the time they took for breaks. There was no significant difference between the times taken among the men to make beds of similar bed dimensions. On the other hand, the field capacity of the designed bed making implement recorded was 3.8 acres/day at the forward speed of 2.54 km/h and made a bed of 80.11cm in width, 25.3 cm high and 30 m long in a minute.

4. Conclusion

To increase the productivity of vegetables and to use irrigation water efficiently, it is vital to cultivate vegetables on raised beds. Preparing beds manually is a tedious, time consuming and laborious job. Hence, the bed-making machine designed in this study offers better opportunities especially in terms of efficiency and in reducing drudgery. Based on the results obtained from the field test, the following conclusions can be drawn: i) the bed-making machine attached to

the tractor makes a bed of optimum size for vegetable cultivation at the appropriate forward speed, ii) the machine also forms a bed dimension that is suitable for laying plastic mulch film, iii) the recommended forward speed of the tractor is 1.89-2.54 km/h to form the required bed dimension and iv) the field capacity of the machine recorded was 3.8 acre/ day when operated at the forward speed of 2.54 km/h against the human capacity to prepare a bed of the same size at only 0.02 acre/day.

Acknowledgement

References

Berkout, N. M., Yasmeen, F., Maqsood, R., & Kalwij, I. M. (1997). *Farmers' Use of Basin, Furrow and Bed-and-Furrow Irrigation Systems and the Possibilities for Traditional Farmers to Adopt the Bed-and Furrow Irrigation Method*. Lahore: International Irrigation Management Institute (IIMI).

Campbell, C. A., McConkey, B. G., Zentner, R. P., Selles, F., & Curtin, D. (1996). Long-term Effects of Tillage and Crop Rotations on Soil Organic C and Total N in a Clay Soil in Southwestern Saskatchewan. *Canadian Journal of Soil Science, 76*(3), 395-401. doi:https://doi.org/10.4141/cjss96-047

Chhogyel, N., & Kumar, L. (2018). Climate Change and Potential Impacts on Agriculture in Bhutan: A Discussion of Pertinent Issues. *Agriculture & Food Security, 7*(1), 1-13. doi:https://doi.org/10.1186/s40066-018-0229-6

Clark, G. A., & Maynard, D. N. (1992). Vegetable Production on Various Bed Widths Using Drip Irrigation. *Applied Engineering in Agriculture, 8*(1), 28-32. doi:https://doi.org/10.13031/2013.26029

David, J. C., Jagadish, T., & Elizabeth, H. (2003). Prospects for Permanent Beds for the Rice-Wheat System. In Jagdish K. Ladha, James E. Hill, John M. Duxbury, Raj K. Gupta, & R. J. Buresh (Eds.), *Improving the Productivity and Sustainability of Rice-Wheat Systems: Issues and Impacts* (Vol. 65, pp. 197-210). Madison, Wisconsin: ASA Special Publications.

Fischer, R. A., Sayre, K., & Monasterio, I. O. (2005, 1–3 March). *The Effect of Raised Bed Planting on Irrigated Wheat Yield as Influenced by Variety and Row Spacing*. Paper presented at the Evaluation And Performance Of Permanent Raised Bed Cropping Systems in Asia, Australia and Mexico, Griffith, NSW, Australia.

Govaerts, B., Sayre, K. D., Lichter, K., Dendooven, L., & Deckers, J. (2007). Influence of Permanent Raised Bed Planting and Residue Management on Physical and Chemical

Soil quality in Rain Fed Maize/Wheat Systems. *Plant and Soil, 291*(1), 39-54. doi:https://doi.org/10.1007/s11104-006-9172-6

Hamilton, G., Bakker, D., Houlebrook, D., & Spann, C. (2000). Raised Beds Prevent Waterlogging and Increase Productivity. *Journal of the Department of Agriculture, Western Australia, Series 4, 41*(1), 3-9.

Hobbs, P. R., & Giri, G. S. (1997). Reduced and Zero-Tillage Options for Establishment of Wheat After Rice in South Asia. In H. J. Braun, F. Altay, W. E. Kronstad, S. P. S. Beniwal, & A. McNab (Eds.), *Wheat: Prospects for Global Improvement* (Vol. 6, pp. 455-465). Dordrecht, Netherlands: Springer Science+Business Media.

Ismail, Z. E., & Ismail, N. K. (2013). Soil Layer Deformation Model During Wide Raised Bed's Construction. *Australian Journal of Basic and Applied Sciences, 7*(10), 20-30.

Kukal, S. S., Sudhir, Y., Humphreys, E., Amanpreet, K., Yadvinder, S., Thaman, S., Singh, B., & Timsina, J. (2010). Factors Affecting Irrigation Water Savings in Raised Beds in Rice and Wheat. *Field Crops Research, 118*(1), 43-50. doi:https://doi.org/10.1016/j.fcr.2010.04.003

Memon, M. S., Ullah, K., Siyal, A. A., Leghari, N., Tagar, A. A., Ibupoto, K. A., Tahir Ata-ul-karim, S., Tahir, M., & Memon, N. (2020). The effect of different raised bed sizes under furrow irrigation method on salt distribution in soil profile and yield by Hydrus (2/3d). *Pakistan Journal of Agricultural Research, 33*(1), 113. doi:http://dx.doi.org/10.17582/journal.pjar/2020/33.1.113.125

Miah, M. A. M., Hossain, S., Duxbury, J. M., & Lauren, J. G. (2015). Adoption of Raised Bed Technology in Some Selected Locations of Rajshahi District of Bangladesh. *Bangladesh Journal of Agricultural Research, 40*(4), 551-566. doi:https://doi.org/10.3329/bjar.v40i4.26930

NSB. (2020). *The 2020 Labour Force Survey Report*. Thimphu: National Statistics Bureau (NSB), Royal Government of Bhutan.

Singh, V. K., Dwivedi, B. S., Shukla, A. K., & Mishra, R. P. (2010). Permanent Raised Bed Planting of the Pigeonpea–Wheat System On a Typic Ustochrept: Effects on Soil Fertility, Yield, and Water and Nutrient Use Efficiencies. *Field Crops Research, 116*(1-2), 127-139. doi:https://doi.org/10.1016/j.fcr.2009.12.002

Tewabe, D., Abebe, A., Enyew, A., & Tsige, A. (2020). Determination of Bed Width On Raised Bed Irrigation Technique of Wheat at Koga and Rib Irrigation Projects, North West, Ethiopia. *Cogent Food & Agriculture, 6*(1), 11. doi:https://doi.org/10.1080/23311932.2020.1712767

Wightman, B., Peries, R., Bluett, C., & Johnston, T. (2005, 1–3 March). *Permanent Raised Bed Cropping in Southern Australia: Practical Guidelines For Implementation.* Paper presented at the Evaluation And Performance Of Permanent Raised Bed Cropping Systems In Asia, Australia and Mexico, Griffith, Australia.

AMC-made Impeller-type Buckwheat Dehuller and Optimization of Impeller Speed

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ABSTRACT

The outer hull or husk of buckwheat (Fagopyrum esculentum Moench.) kernel comprises about 20-30% of the weight of the buckwheat grain. Since it is inedible it should be removed before processing into flour. While imported and expensive dehulling machines are available in Bhutan, to reduce cost and gain wider adoption by farmers and perform the operation efficiently, the Agriculture Machinery Centre (AMC) has developed a buckwheat dehuller based on the impact and shear principle for efficient dehulling of whole buckwheat with most of the machine's body parts available locally. The present study was carried out to test the machine and to evaluate the best operating impeller speed for efficient dehulling of buckwheat. The machine was tested at five different levels of impeller speed: 1,700, 1,800, 1,900, 2,000 and 2,100 revolutions per minute (rpm). Process performance of dehulling efficiency (DE), and broken percentage (B%) were measured at each rpm. There was a significant difference in DE and (B%) across the five different speeds. The best DE of 88.94 % was observed at 2,100 rpm. However, there was also a high correlation between dehulling efficiency and the proportion of broken kernels, with the highest broken percentage of 43.96% observed at 2,100 rpm.

Keywords: *Buckwheat: Dehulling efficiency; Dehuller machine*

1. Introduction

Buckwheat (*Fagopyrum esculentum* Moench.) is a pseudocereal, having gluten-free grains characterized by an excellent nutrient profile (Solanki, Mridula, Kudos, & Gupta, 2018). Buckwheat belongs to the Polygonaceae family and is taxonomically distant from the Gramineae family to which most cereals such as rice, wheat and maize belong. However, buckwheat seed has chemical and utilization characteristics like most cereal grains and thus is usually classified as a cereal (Ikeda, 2002).

There are several varieties of buckwheat species (Nagatomo, 1984; Ohnishi, 1991 as cited in Ikeda-K, 2002). The commonly cultivated species for human consumption are common buckwheat (*Fagopyrum esculentum* Moench.) also known as sweet buckwheat and Tartary or bitter buckwheat (*E tataricum* Gaertner). Buckwheat is a very versatile crop that can grow on

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most infertile land where most crops often fail. It is suitable for ecological growing, without the use of artificial fertilizers or pesticides (Krkošková & Mrazova, 2005). It is a rich source of starch and contains many valuable compounds, such as proteins, antioxidative substances, trace elements and dietary fibre (Bonafaccia, Marocchini, & Kreft, 2003; Kreft, Knapp, & Kreft, 1999). Due to the well-balanced amino acid composition buckwheat proteins have a high biological value, although their digestibility is relatively low (Yeshajahu & George, 1972). Buckwheat protein extracts may have a strong healing effect on some chronic diseases, such as diabetes, hypertension, hypercholesterolemia, and many other cardiovascular diseases (Li & Zhang, 2001). Besides high-quality proteins, buckwheat seed contains several components with healing benefits: flavonoids and flavones, phytosterols, fagopyrins and thiamin-binding proteins (Krkošková & Mrázová, 2005).

Agriculture Statistics (2019) (RSD, 2020) show that buckwheat is grown in all 20 dzongkhags of Bhutan. Amongst them, higher quantities are produced by farmers of Samdrupjongkhar, Bumthang, Haa, Trongsa, Dagana, Wangdue, Chhukha and Samtse dzongkhags. The total production was 2,350.43 Mt with an average yield of 459.389 kg/acre in the year 2019 (RSD, 2020). As per the buckwheat value chain analysis carried out by the Department of Agriculture Marketing and Cooperatives (DAMC), 44.9% of the households cultivate both bitter and sweet buckwheat; another 22.2% cultivate only bitter buckwheat and 32.9% grow sweet buckwheat (DAMC, 2019). In terms of agricultural assets owned by the buckwheat producing households, as high as 42.1% of households did not own any of the main agricultural equipment/machinery. Both males and females equally take part in buckwheat farming. While ploughing is led by males, land preparation, sowing, harvesting, and threshing is done by both males and females. It is mostly the males who engage in grinding using watermills and women who use stone grinders (DAMC, 2019). Thus the adoption of a cheap and efficient dehulling machine may lead to less drudgery, especially for women.

In Bhutan, buckwheat is mainly grown for its grains. The grains are used in the form of processed products, mainly flour. Buckwheat *Choydam* (hard dough), *Puda* (noodles), *Kontong* (cooked small balls), *Teyzey* (pancake), *Khuli* (cooked, soft Roti type), *Keptang* (unleavened circular bread) and *La zey* (flour baked with distilled alcohol); *Drengo* (flour paste); *Tshangjay* (flour paste with fermented wine); and *Hentey* (dumplings) are items that are usually prepared from buckwheat flour (Norbu & Roder, 2001). Buckwheat is also used to make *Ara* (distilled local liquor) / *Bangchang* (fermented wine). Sweet buckwheat hulls are also used to make pillows and mattresses.

Buckwheat is regaining popularity because of growing demand in the market fueled by a newfound awareness of its nutritional value (Yonten, 2018). People are realizing the nutritional value of buckwheat and the traditional food cultures of the past. Buckwheat is also identified as one of the three organic products under the National Organic Flagship Program (NOFP) in the country.

The common sweet buckwheat bears triangular seeds with a black hull that covers the light green to white kernel inside. The outer hull or husk is mainly composed of cellulose and it comprises about 20-30% of the weight of the buckwheat grain depending on the variety. Since it is inedible and cannot be digested by the human digestive system the hull is removed from the buckwheat kernel before further processing into flour (Solanki et al., 2018). The hull has a smaller density than water and this allows easier removal of the hull from the kernel. The hardness of the hull depends on the species of buckwheat (Krkošková & Mrazova, 2005). Traditionally, buckwheat grains are directly milled using a water mill, traditional stone grinder or any other crude method. After milling, the segregation of the husk from the flour is done using traditional sieving methods. This is highly inefficient in terms of the product quality as a huge portion of inseparable crushed husks remains in the flour.

While performing preliminary trials employing various grain dehulling methods used for paddy in the country, it was observed that the dehulling based on the principle of impact and shear (impeller type huller) gave better performance for these grains as compared to dehulling of grains using rubber roll sheller. The latter resulted in a high proportion of broken grains. Given this, a buckwheat dehuller machine based on the impact and shear principle was developed at the AMC (Figure 1). The present study was carried out to evaluate the machine's performance by comparing dehulling operations at five different speeds measured in revolutions per minute (rpm).

Figure 1. AMC made buckwheat dehuller

2. Materials and method

2.1 Description of the machine components

The buckwheat dehulling machine consists of three major components: frame, dehulling unit and aspiration unit. The machine has two frames, the first and the main one supports the entire dehulling unit and the second frame holds the aspiration unit. The dehulling unit is the main component of the machine responsible for removing the husk from the buckwheat groats. This unit comprises 12 blades impeller and a stationary concave casing design based on centrifugal and Coriolis forces, whereby the buckwheat grains are thrown against the liner part of the impeller housing. This unit is driven by a 1 horsepower (hp) single phase motor in conjunction with pulley drive to maintain desirable speeds. The aspiration unit is designed to separate the husk and other lighter fractions from the mixture of groats, husk and unhusked grains produced from the dehulling operation. It comprises a 0.56 hp blower attached to the separating duct.

2.2 Working of the machine

The buckwheat grains are fed through the hopper into the dehulling unit where the buckwheat grains are dehulled, and the mixture of groats, husk and unhusked grains then passes through the aspirator attached to the machine. The aspirator separates the husk from the mixture and blows them out of the husk outlet, while the mixture of groats and unhusked grains is obtained at an outlet below the machine.

2.3 Dehulling principle

The dehulling takes place based on the impact and shear principle. The grains are fed into the centre of the rotor, through the rotating impeller where they are subjected to centrifugal force and thrown towards the rubber-lined casing. The shelling takes place here due to the high-force impact that hits the grains because of the frictional force between impeller blades and the grains (Figure 2). This causes the hulls to break loose from the groats, eventually releasing the groats free.
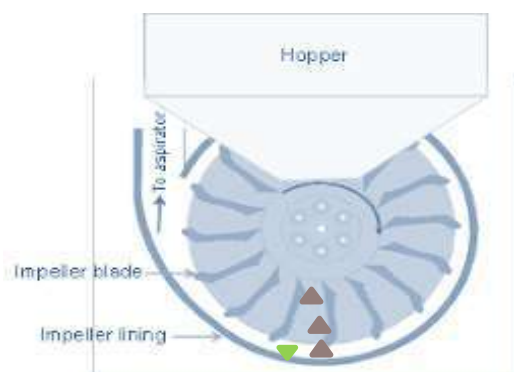

Figure 2. Schematic drawing of impact and shear principle in dehulling machine

2.4 Design consideration

The materials used for the fabrication of the machine were of adequate strength and stability. Most of the components were purchased from the local hardware stores. Consideration was given to keep the cost of the components reasonably low and yet, readily available. This will allow easy duplication by in-country manufacturers and at the same time satisfy all strength requirements.

2.5 Design support frame

The machine has two frames, the first main frame supports the entire dehulling unit and the second frame supports the aspiration unit. A frame of 550 x 265 x 530 mm size was fabricated to support the entire weight of the dehulling unit. The base of the frame was made of a C-channel of 100 x 50 x 6 mm size and an angle bar of 40 x 40 x 6 mm size to give the required rigidity. A second frame of 955 x 243 x 500 mm size that supports the aspirating unit was designed using 25 x 25 x 3 mm size angle bar.

2.6 Design of dehulling unit

The dehulling unit comprises 12 impeller blades radially arranged on an impeller hub, a liner and a casing. The impeller blades were made up of mild steel sheets of 3 mm thickness (Figure 3).
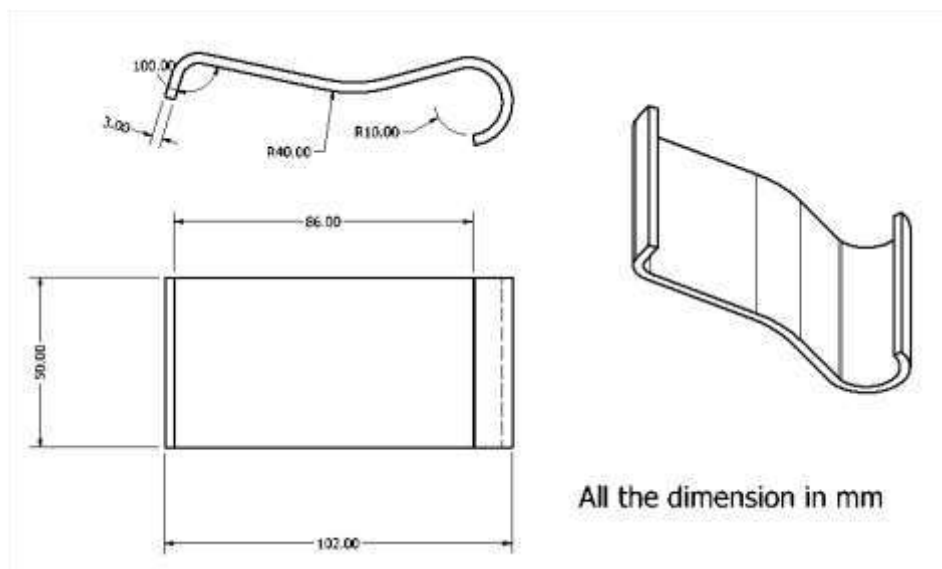

Figure 3. Impeller blades

The impeller hub (Figure 4) consists of a simple 3 mm mild steel hub with a diameter of 308 mm to accommodate twelve blades at an angular spacing of 30 degrees. It was made into two symmetrical halves for easy placement of blades in between. The thickness of each half was 3

mm; one half of the hub is slotted with a hole of a diameter of 91 mm for the entry of grains and another half with 20 mm hole for the shaft.
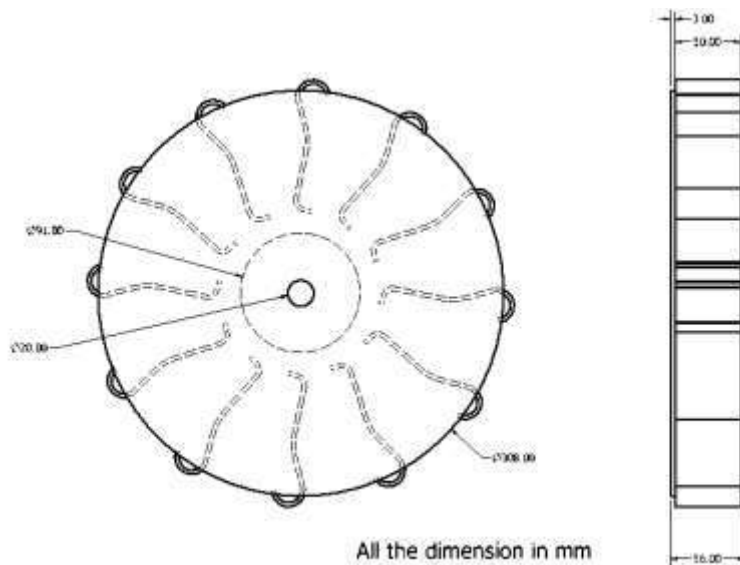

All the dimension in mm

Figure 4. Impeller hub

The liner used was a locally available polyester rice mill nylon belt. It was placed over the entire inner circumference of the casing. The casing and the outlet chute were fabricated with a mild steel sheet of size 2 mm.

The hopper is a frustum of a pyramid (pyramid portion with its upper head cut off by a plane parallel to its base). It is the medium through which grains are introduced to the dehulling unit. It is made of 2 mm thick mild steel. The volumetric capacity of the hopper is 7,930 cm^3.

2.7   Design of aspiration unit

The aspiration unit (Figure 5) is designed to separate the husk and other lighter fractions from the mixture of groats, husk and unhusked grains produced from the dehulling operation. The unit comprises a centrifugal air blower of 0.56 hp with 2800 rated rpm attached to the separating duct. The blower used has an independent power connection. The separating ducts used were made of locally available polyvinyl chloride pipes of size three inches.
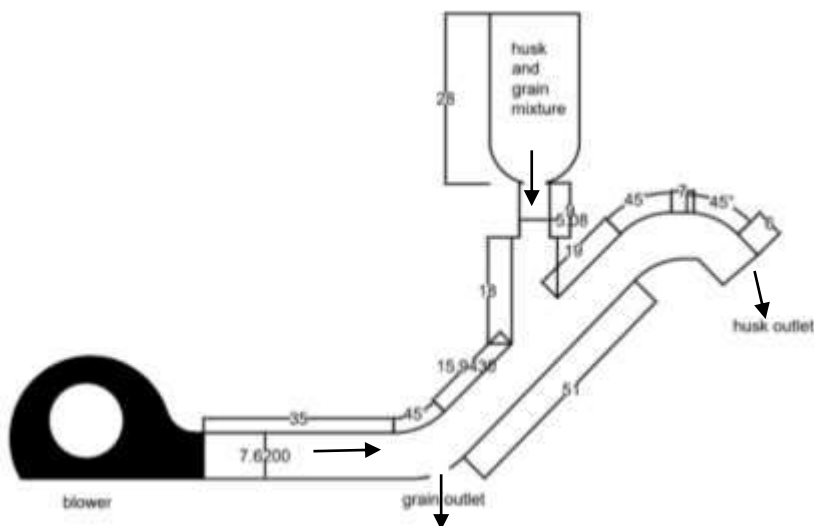
Figure 5. Aspirator duct (all dimension in cm)

2.8 Sample preparation

Common sweet buckwheat grains (*Fagopyrum esculentum*) were obtained from the National Seed Centre, Paro. The dehulling test was conducted at AMC, Paro. Grains were cleaned using a manual mechanical winnower to remove foreign matters, and broken and immature grains. The grains were then dried to an average moisture content of 8.56±0.15%. The moisture content of the grains was measured using universal grain moisture meter (INDOSAW).

2.9 Dehulling test and measurements

The buckwheat samples of dried buckwheat at 8.56±0.15% moisture content were dehulled to evaluate the performance of the AMC designed impeller dehulling machine at five different levels of speed (1,700, 1,800, 1,900, 2,000 & 2,100) rpm. The mass of grain and husk obtained at the outlet were classified as whole groats, un-hulled grains, broken grains and husk.

The experiment was carried out following a completely randomized design (CRD) with a single factor (speed) at 5 levels with three replicates each.

Dehulling efficiency (DE) was measured as follows:

$$DE = \frac{Mt - Mn}{Mt} * 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{(Solanki et al., 2018)}$$

Where:
 Mn is a mass of grain left unhusked after the dehulling operation
 Mt is the total mass of grain fed into the hopper

Broken percentage (B%) was measured as follows:

$$B\% = \frac{Ml}{Mt} * 100 \ \text{.........................} \ \text{(Singh, Saha, \& Mishra, 2010)}$$

Where:
Ml is the broken kernels loss in the form of powder and broken grain after dehulling
Mt is the total mass of grain fed into the hopper

2.10 Statistical analysis

Pearson's correlation, simple linear regression and one-way ANOVA were performed to explore relationships between impeller speed (rpm), dehulling efficiency (%) and broken percentage (%). Bartlett's test for equal variances was performed to check the data for equality of variances assumption before running ANOVA. Tukey's test was used to test for significance among treatment means.

3. Results and Discussion

Dehulling efficiency increased linearly with an increase in the speed of the impeller (Figure 6). Simple linear regression of DE and speed (rpm) was found highly significant ($P<0.000$). The average dehulling efficiency of the machine was 83.26±4.80% which is considered acceptable. In a study by Solanki et al., (2018) who used roller type dehusker set at 800 rpm, only 66.61% of DE was achieved.
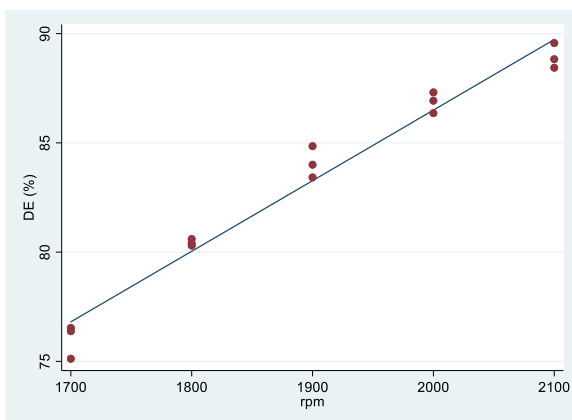

Figure 6. Scatter and line plot of DE% and impeller speed (rpm)

DE was significantly different between all the five levels of speed (Table 1). The highest dehulling efficiency was observed at 2,100 rpm (88.95%) and the lowest at 1,700 rpm (72.69%).

Similarly, the broken percentage was also found to increase linearly with increasing impeller speeds (Figure 7) although there was no significant difference between the rpms 1,700 and 1,800; 1,900 and 2,000; and 2,000 and 2,100 (Table 1). The absence of significant difference between the aforementioned pairs, despite the means being different from each other as in the case of dehulling efficiency, could be due to the higher variation in the broken proportion data for rpms of 1,800 and 2,000.

Table 1. Mean dehulling efficiency and broken percentage across the five different speeds

Speed level (rpm)	Dehulling efficiency (%)	Broken (%)
1700	76.01 ± 0.77 [e]	29.54 ± 0.54[c]
1800	80.43 ± 0.15[d]	32.94 ± 2.86[c]
1900	84.09 ± 0.72[c]	38.36 ± 1.46[b]
2000	86.87 ± 0.48[b]	39.40 ± 2.37[ab]
2100	88.95 ± 0.57[a]	43.97 ± 0.72[a]

Mean values within the columns with different superscripts are significantly different between speeds levels at $P < 0.05$ by Tukey's test (mean ± standard deviation, n=3)
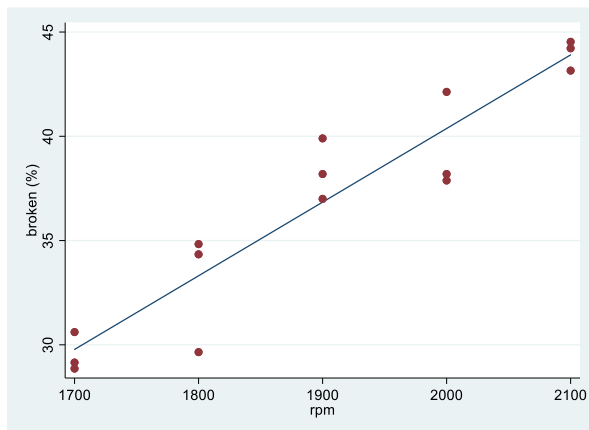

Figure 7. Scatter and line plot of broken % and impeller speed (rpm)

What could be the reason for observing a higher proportion of brokens as the impeller speed increased? Just as the higher roller speed induces rapid separation of the hull from the kernels, a similar frictional force may be induced on the body of the kernels, especially if they are quite fragile or soft. Similar observations were also made by Solanki et al. (2018), Jain (1980), Joshi (1993) and Guptaa and Das (1999) in dehulling of buckwheat, paddy, pumpkin seed and sunflower seed, respectively.

The results show a very high correlation between the machine's dehulling efficiency and the proportion of broken grains, with a Pearson's correlation coefficient of 0.95 and a highly

significant *P*-value ($P<0.000$). Is this a paradox then? In the case of buckwheat, people often grind it further into flour and then consume it in the form of various products mentioned earlier. Hence, it may be argued that the presence of broken grains is of no concern as they will be ground further along with the unbroken grains into flour at the end of the day. Thus, it can be safely reasoned that the machine can be operated at 2,100 rpm to obtain the highest dehulling efficiency without worrying about broken grains proportion.

4. Conclusion

While it was already proven through informal tests conducted at the Agriculture Machinery Centre that the locally fabricated huller machine was effective at dehulling buckwheat grains, this study substantiates this further with the empirical evidence generated. The results indicate that the best dehulling performance for this machine could be obtained if the system is operated at a roller speed of 2,100 rpm at 8.56% moisture content. Under these conditions, the values of dehulling efficiency were as high as 88.95%. However, the percentage of the broken grain was also observed to be the highest at this speed. On the other hand, since buckwheat kernels are anyway crushed further and consumed as flour, it is of no concern.

Acknowledgement

References

Bonafaccia, G., Marocchini, M., & Kreft, I. (2003). Composition and Technological Properties of the Flour and Bran from Common and Tartary Buckwheat. *Food Chemistry, 80*(1), 9-15. doi:https://doi.org/10.1016/S0308-8146(02)00228-5

DAMC. (2019). *Buckwheat Value Chain Analysis*. Thimphu: Department of Agricultural Marketing and Cooperatives (DAMC), Ministry of Agriculture and Forests (MoAF), Royal Government of Bhutan.

Guptaa, R. K., & Das, S. K. (1999). Performance of Centrifugal Dehulling System for Sunflower Seeds. *Journal of Food Engineering, 42*(4), 191-198. doi:https://doi.org/10.1016/S0260-8774(99)00119-3

Ikeda, K. (2002). Buckwheat Composition, Chemistry, and Processing. *Advances in Food and Nutrition Research, 44*, 395-434. doi:https://doi.org/10.1016/S1043-4526(02)44008-9

Jain, R., K. (1980). *Optimization of operating parameters of centrifugal sheller.* (M. Tech Thesis), Indian Institute of Technology, Kharagpur, Kharagpur, India.

Joshi, D. C. (1993). *Mechanical Dehulling and Oil Expression of Pumpkin Seed.* (Ph.D Thesis), IIT, Kharagpur, Retrieved from http://www.idr.iitkgp.ac.in/xmlui/handle/123456789/6011

Kreft, S., Knapp, M., & Kreft, I. (1999). Extraction of Rutin from Buckwheat (*Fagopyrum esculentum Moench*) Seeds and Determination by Capillary Electrophoresis. *Journal of Agricultural Food Chemistry, 47*(11), 4649-4652. doi:https://doi.org/10.1021/jf990186p

Krkošková, B., & Mrazova, Z. (2005). Prophylactic Components of Buckwheat. *Food Research International, 38*(5), 561-568. doi:https://doi.org/10.1016/j.foodres.2004.11.009

Li, S.-q., & Zhang, Q. H. (2001). Advances in the Development of Functional Foods from Buckwheat. *Critical Reviews in Food Science Nutrition, 41*(6), 451-464. doi:https://doi.org/10.1080/20014091091887

Norbu, S., & Roder, W. (2001). Traditional Uses of Buckwheat in Bhutan. In I. Kreft, K. J. Chang, Y. S. Choi, & C. H. Park (Eds.), *Ethnobotany of Buckwheat* (pp. 34-38). Seoul: Jinsol Publishing Co.

RSD. (2020). *Agriculture Statistics 2019*. Thimphu: Renewable Natural Resources Statistics Division (RSD), Ministry of Agriculture and Forests, Royal Government of Bhutan.

Singh, K., Saha, S., & Mishra, H. (2010). Optimization of Barnyard Millet Dehulling Process using RSM. *Ama, Agricultural Mechanization in Asia, Africa Latin America, 41*(2), 15.

Solanki, C., Mridula, D., Kudos, S. K. A., & Gupta, R. K. (2018). Buckwheat Dehuller and Optimization of Dehulling Parameters. *International Journal of Current Microbiology and Applied Sciences, 7*(11), 1041-1052. doi:doi: https://doi.org/10.20546/ijcmas.2018.711.120

Yeshajahu, P., & George, S. (1972). Amino acid composition of buckwheat. *Journal of Agricultural and Food Chemistry, 20*(2), 270-274. doi:https://doi.org/10.1021/jf60180a029

Yonten, K. (2018). From Potato to Buckwheat. *The Bhutanese* Retrieved from https://thebhutanese.bt/from-potato-to-buckwheat/

Study on Clarification of Apple Juice using Enzymes

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ABSTRACT

One of the key challenges in apple juice processing is obtaining a good juice recovery and attaining a juice with good clarity. The presence of pectin and starch components inhibits the juice extraction process and leads to the formation of a cloudy haze which is undesirable in apple juice. For these purposes, maceration enzymes such as pectinase, amylase, cellulose, and hemicellulase are added both before and or after juice extraction to enhance juice recovery percentage and clarify the juice. Process parameters such as type of enzymes used, stage of enzyme addition, incubation time and temperature influence the efficiency of enzymes. The optimisation of these parameters is critically important in apple juice processing for better juice recovery and attaining the desired juice clarity. In this study, juice recovery percentage was compared amongst the control and three treatments- addition of enzyme pectinase at 0.02%, amylase at 0.02% and a combination of both at 0.01%. For the optimisation of process parameters, the type of enzymes used, stage of enzyme addition, incubation temperature, and time were studied as independent variables by comparing with the transmittance value as a dependent variable. Amylase at 0.02%, pectinase at 0.02%, and a combination of 0.01% each of both enzymes were used. The enzyme was added to the pomace and the juice. The treatments were incubated at 20^0C and 40^0C. Measurements were done after every 1, 2, 4, and 24 hours of incubation. The juice recovery percentage was not significantly different in the 3 treatments and control where the enzyme was added to the crushed apple. For clarification of apple juice, it is recommended to add the combination of 0.01% each of amylase and pectinase directly to the pomace before juice extraction and incubating the juice obtained at 40^0C for 24 hours.

Keywords: *Enzymes; Apple juice; Clarification; Juice recovery; Process parameter*

1. Introduction

Apple is one of the major cash crops grown in the country for export to Bangladesh and India. Apple is predominantly grown in Paro, Thimphu, Haa, Chhukha and Bumthang Dzongkhags. The main apple varieties grown in the country are Royal Delicious, Red Delicious and Golden Delicious (Choden & Shanawaz, 2015). Over the past 5 years from 2015 to 2019, an average of 5589 metric tonnes (t) of apples were produced in the country (RSD, 2020). Besides the export, two main agro-processing companies in the Country-Bhutan Agro Industries Limited

in Thimphu and the Fruit Processing Enterprise in Bumthang utilize local apples as raw materials for processing products such as apple juice, beverages and jams.

In 2016, the total apple production was reported to be 6587 Mt (DoA, 2016) and about 8% of the total production volume amounting to 450 Mt was procured by Bhutan Agro Industries Ltd. (DAMC, 2017) for apple juice processing. Ready to serve apple juice is processed from fresh apples by crushing the washed and sorted raw apples and subsequent pressing the crushed apples by a frame filter press. The quality of apples used for juice processing determines the end quality of the juice, thus the genetic composition and other growth factors such as nutrition, climatic condition, maturity and storage of the apples affect the quality of the juice (Pollard & Timberlake, 1971).

Juice recovery from apple and clarity of the juice are two critical factors for ensuring the economic viability of the processing units and acceptance of the product in the market. The process of juice extraction involves rupturing of cells to release juice and pressing the ruptured mass of cells to extract the juice. Thus, apple juice contains the soluble constituent present in the apple which generally has 85% water, 10-12% carbohydrate, 1% pectin, 0.5 - 1% organic acid, 0.5% potassium, phenol, amino acid and flavouring in small amounts (Ryan, 1972). Pectic substance in apple is present in the cell wall and middle lamella. Pectin is a complex long-chain polysaccharide made of multiple units of (1,4)-a-D-polygalacturonic acid. Starch is present in the fruit as a reserve food and as the fruit ripens this starch gets converted into sugar. Apple juice obtained after crushing and pressing may contain up to 1% starch which presents a problem of cloudiness in the juice (Sorrivas, Genovese, & Lozano, 2006).

During the extraction process, the juice along with suspended particles comprising both water-soluble and insoluble materials leave the extractor. The presence of pectin in the juice inhibits the juice extraction process resulting in a low juice recovery rate (Root & Barrett, 1996). The presence of hydrophilic hydroxy group gives pectin its water-binding capacity, as a result, the juice binds to the pectin present in the pulp hindering the extraction process (Shiv, 2015). This also causes an increase in the viscosity of the juice and lubricates the crushed fruit pulp which results in slippage during pressing and as a result lowers the juice recovery rate (McLellan & Padilla-Zakour, 2004).

Consumer preference for clear apple juice (Kilara & Buren, 1989) makes it imperative to remove the cloudy haze present in apple juice. This cloudy haze is due to the presence of pectin and starch (Padma, Sravani, Mishra, Sneha, & Anuradha, 2017). The cloudy haze is formed

due to the reaction between starch with protein to form a positively charged protein-carbohydrate complex which acts as a positive core and attracts the negatively charged pectin. This particle is responsible for the cloudiness in apple juice (Yamasaki, Yasui, & Arima, 1964). The protein-carbohydrate complex surrounded by the protective pectin coat prevents aggregation of the particles resulting in a stable suspension thus hindering the process of sedimentation and subsequent filtration (Sorrivas et al., 2006).

The use of maceration enzymes such as pectinase breaks down the pectin structure and lowers its water-binding capacity, thus freeing up the juice bound to the pectin structure and improving the juice recovery rate. Pectinase enzymes digest the pectin by hydrolysing or de-esterifying pectin, as a result, pectin loses its water-binding capacity, flocculates and settles down as sediments. This process is known as depectinization (Kilara & Buren, 1989). Pectinase enzymes are naturally present in fruits and help convert the insoluble protopectin in unripe fruit to soluble pectin during ripening. However, naturally occurring pectinase enzymes are not sufficient enough to achieve adequate clarification in the juice, thus pectinases mostly from microbial sources are used for commercial juice processing (Patidar, Nighojkar, Kumar, & Nighojkar, 2018).

Amylase and pectinase are used individually and in combination to clarify apple juice to improve the juice recovery percentage. Studies have reported that when pectinase and amylase are used in combination, the synergistic effect has a more desirable effect on the clarity of the juice than when used individually (Padma et al., 2017). The use of a mix of enzymes during the clarification process can bring about both depectinization and destarching. For example, amylase is a starch degrading enzyme that works by hydrolysing the glyosidic linkage in long-chain starch breaking it into smaller units in a process known as destarching (Rana, Verma, Vaidya, & Dipta, 2017). Further, the synergistic effect entails the depectinisation reaction first followed by the prevention of possible agglomeration of the starch molecules with the protein-pectin complex formed as a result of depectinisation (Dey & Banerjee, 2014).

The enzyme is added at various stages of processing. For enhancing juice recovery percentage, the addition of enzymes takes place after the crushing process and before the pressing stage. For clarification purposes, enzymes are usually added to the juice after the pressing stage. To achieve both the purpose of enhanced juice recovery and clarification, some processors add enzymes at both stages (McLellan & Padilla-Zakour, 2004). Enzymic degradation of the haze forming components brought about by the macerating enzymes is responsible for the enhanced

juice recovery and clarification. The process of enzymic degradation is influenced by parameters such as the type of enzymes, dosage used, exposure time of the substrate to the enzyme, the temperature and the pH (Singh & Singh, 2015). For this study, only the type of enzymes used, the exposure time and temperature were observed. All the treatments were done at the original pH of the naturally extracted juice. This study attempted to replicate the industrial process used in the production of natural apple juice where alteration of pH is not commercially practised (Rai, Majumdar, Dasgupta, & De, 2004), but the process parameters such as types of enzymes used, duration of treatment and incubation temperature after the addition of enzymes were observed.

It has been reported by Kilara and Buren (1989) that at a commercial level, an enzyme dosage of 0.02% calculated on the volume of juice is used. Ezugwu et al. (2014) reported that at 40^0C the enzymes gave the most favourable result. To ensure the effectiveness of enzyme treatment in enhancing both the juice recovery and clarity of juice and also to have a cost-effective process, it is imperative to know the effect of the process parameters on their own and also in combination with other parameters for better optimization of apple juice processing.

Thus, the objectives of this study were:

a) to compare the juice recovery from apple pomace treated with 0.02% amylase, 0.02% pectinase and a combination of 0.01% amylase and pectinase before juice pressing, and

b) to compare the clarity of apple juice treated with amylase, pectinase or in a combination of amylase and pectinase and the effect of the process parameters such as incubation temperature, duration of treatment and stage of addition of enzyme on clarity of the apple juice.

2. Materials and Method

The experiment was conducted in the food analysis laboratory at the National Post Harvest Centre (NPHC), Paro. Apples stored in the cold store at 4 degrees Celsius were sorted, washed and the cores removed. For the extraction process, a fruit crusher and frame filter press were used. Processing equipment and utensils such as a fruit pulper, frame filter press, autoclave, stainless steel knife, plastic buckets, and stainless-steel stirrer were used for the process. Apples were first crushed in a pulping machine and the crushed apples were pressed using a hydraulic juice press. Enzyme dosage of 0.02% amylase, pectinase and 0.01% each of amylase and pectinase were added to 7-kg each of crushed apple before the pressing process. A control without any addition of enzyme was used. The weight of juice from triplicates of each treatment

and control was recorded and the juice recovery percentage was calculated using the following formula:

Juice recovery percentage (%) = [Volume of extracted juice (litre)/Weight of crushed apple used for juice extraction (kg)] ×100

The juice obtained from crushing the three treatments was taken as the three treatments when enzyme addition is done to the pomace before juice extraction. Enzyme dosage of 0.02% amylase, pectinase and 0.01% each of amylase and pectinase were added to 3.5 litres of apple juice extracted from the pomace without prior addition of enzymes. The juice obtained from control in the previous experiments was also taken as the control sample. 400 ml of juice from triplicates of all six treatments and control were transferred to glass jars. Two similar batches were prepared whereby each batch was placed in an electric dryer with the temperature set at 20^0C and 40^0C. Samples were drawn after 1, 2, 4 and 24-hour of incubation period and filtered for transmittance measurement. The juice was filtered using Whatman 5 filter paper. The sample drawn was immediately autoclaved at 100^0C for 5 minutes to de-activate the enzyme. The transmittance of the samples was measured at 660 nanometers using an advanced microprocessor UV-VIS Single Beam Spectrophotometer (LI-295). Distilled water was used as a blank to calibrate the spectrophotometer to give a transmittance value of 100% (Berutu, Fahrurrozi, & Meryandini, 2017).

Factorial ANOVA with main and combination effects was done to study if the enzyme type had a significant effect on the juice recovery. A two-way ANOVA and post hoc test was also done to study the main and interaction effect of enzyme type, temperature and time on the clarity of the extracted juice. The analysis was carried out using the Statistical Package for Social Science software (SPSS). *P*-values ≤0.05 were considered significant in all analyses.

3. Results and Discussion

3.1 Juice Recovery Percentage

The juice recovery percentage from the treatment treated with 0.02% of enzyme pectinase gave the highest juice recovery of 66.5% followed by the 0.02% amylase added sample. The 0.01% amylase and pectinase added sample which gave similar values of 62.5% and 62.8%, respectively. The control had the lowest value of 60.4%.

An ANOVA (Table 1) revealed that there is no significant difference ($F_{(3, 8)}$ = 0.949, P = 0.461). An increase of 6-7% in juice recovery was reported upon the addition of commercially available pectinase enzyme at a dosage of 0.01-0.05% (Oszmiański, Wojdyło, & Kolniak,

2009). However, in this study, the addition of enzymes did not significantly increase the juice recovery percentage. Though some studies report that the addition of enzymes increases the juice recovery percentage, the increase in juice recovery is also dependent on the commercial grade of enzymes used (Chang, Siddiq, Sinha, & Cash, 1995).
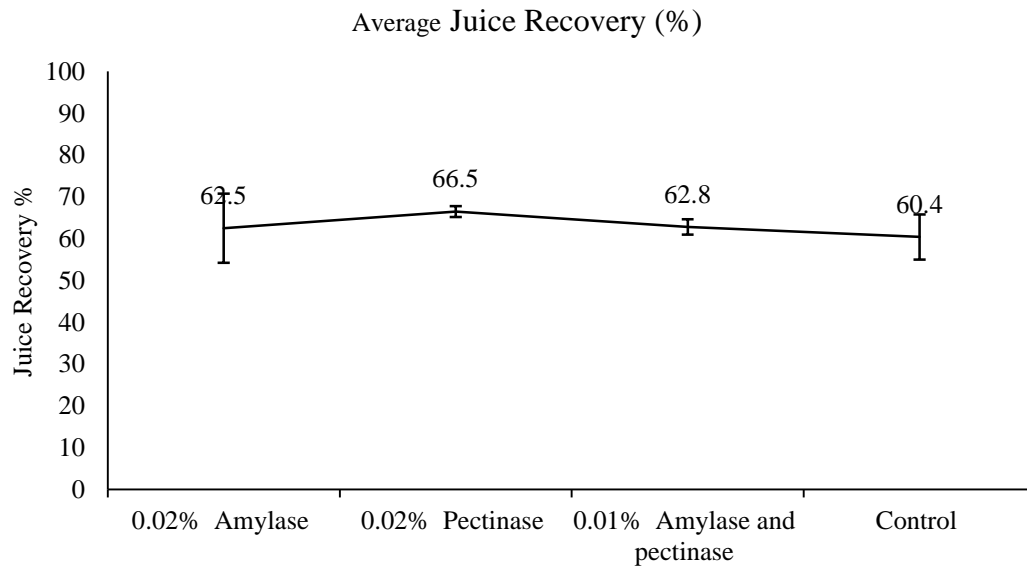
Average Juice Recovery (%)

Figure 1. Average juice recovery percentage value

3.2 Transmittance Value

Transmittance value (expressed as percent transmittance) measured in spectrophotometer is the amount of light that passes through the sample (Garner, Crisosto, Wiley, & Crisosto, 2008), hence this value was used as a measure of clarity. A factorial Between-Subjects ANOVA with the experimental design 2X4X2X4 was performed to study the main and interaction effect of process parameters such as the stage of enzyme addition, treatments, incubation temperature and time on the transmittance value (Table 2). If the P- value of the parameters on their own and in combination is less than 0.01 then the parameters or the combination of parameters are reported to have a significant impact on the clarity of the juice. The partial eta squared is the value of the magnitude of this impact. The effect of the parameters both on their own and in combination are ranked in Table 1 from highest to lowest (1-7) in superscripts in the 5th column based on the partial eta squared values. The Treatment (F $(2,112)$ = 151.788, P = 0, partial η^2 = 0.730) had the highest main effect on the transmittance value followed by incubation time (F $(3,112)$ = 85.404, P = 0, partial η^2 = 0.696). The interaction effect of treatment and incubation time (F $(6,112)$ = 37.757, P = 0, partial η^2 = 0.637) had the third-highest impact on the transmittance value. A similar finding was reported by Umsza-Guez et al. (2011) whereby the

clarity was significantly influenced by the enzyme and the incubation time. Though in total 3 main effects and six combined effects were detected, only the first 5 effects ranked according to the partial eta squared will be discussed.

Table 2. Test of between-subject effects

Source	df	F	Sig.	Partial Eta Squared
Corrected Model	55	23.316	.000	.920
Intercept	1	779.527	.000	.874
Stage of Enzyme addition	1	.019	.892	.000
Incubation Temperature	1	32.222	.000	.223[6]
Treatment	2	151.788	.000	.730[1]
Incubation time	3	85.404	.000	.696[2]
Before_after enzyme addition * Treatment	2	9.596	.000	.146[9]
Incubation temperature * Treatment	2	17.668	.000	.240[8]
Incubation temperature * Incubation time	3	13.025	.000	.259[7]
Treatment * Incubation time	6	32.757	.000	.637[3]
Before_after enzyme addition * Temperature * Treatment	2	1.790	.172	.031
Before and after enzyme * Temperature* Incubation Time	3	2.220	.090	.056
Before and after enzyme * Treatment* Incubation Time	6	8.567	.000	.315[5]
Temperature * Treatment * Incubation Time	6	15.701	.000	.457[4]
Error	112			
Total	168			
Corrected Total	167			

3.2.1 Main effect of treatment, incubation time and temperature on transmittance value

The main effect gives the stand-alone impact of a single process parameter without taking into consideration the effect of other parameters involved. The stand-alone effect of treatment type followed by incubation time had the highest main effect on the transmittance measurement. Incubation temperature also had an impact on the juice clarity. However, whether the enzyme is added to the pomace before juice extraction or to the juice after the extraction on its own without considering the other process, parameters did not seem to make much difference to the transmittance value.

Thus, a one-way ANOVA and post hoc Tuckey test was used to detect significant difference among the main effect of different treatment, incubation time and temperature. Both the ANOVA test for treatment (F (3, 164) = 15.863, P = 0.000) and incubation time F (3, 164) = 17.115, P = 0.00) revealed that there was a statistically significant difference in the mean transmittance values (Table 3 and Table 4 respectively). The post hoc test revealed that the

mix of enzyme added samples had significantly higher clarity followed by the pectinase added sample. The 0.02% amylase added sample had significantly lower clarity in comparison to the other treatment. A similar finding has been reported by (Padma et al., 2017) whereby the combination of pectinase and amylase enzyme was found to be more effective in achieving higher juice clarity than the individual enzyme on its own. In addition, studies conducted by Kothari, Kulkarini, and Baig (2013) reported that a combination of pectinase and amylase gave better transmittance value followed by the addition of pectinase on its own. They also reported that the amylase enzyme added samples gave the lowest transmittance value among the three.

The sample with the 24-hour incubation time also had a significantly higher clarity measurement than the samples incubated at 1, 2 and 4-hour. (Table 4). The ANOVA test for incubation temperature (F (1, 166) = 6.689, P = 0.011) revealed that there was a statistically significant difference in the mean transmittance values (Table 5) with the sample incubated at 40^0C having significantly higher clarity measurement than the one incubated at 20^0C.

Table 3 – One Way ANOVA & post hoc Tuckey test for different treatments

Treatment	Average Transmittance value with standard error
0.02% amylase	2.86±0.31[c]
0.02% pectinase	21.91±3.05 [ab]
0.01% amylase and pectinase	26.39±3.65[a]
Control	12.14±1.87 [b]

Means within a column with different superscripts differ significantly (P≤0.05)

Table 4 – One Way ANOVA & post hoc Tuckey test for different incubation time

Incubation Time in hours	Average Transmittance value with standard error
1	9.17±1.30 [bc]
2	18.37±2.55 [b]
4	6.25±1.09[c]
24	31.62±4.58 [a]

Means within a column with different superscripts differ significantly (p≤0.05)

Table 5 – One Way ANOVA for different incubation temperature

Incubation Temperature in degree celsius	Average Transmittance value with standard error
20	12.37±1.58 [b]
40	20.34±2.64 [a]

Means within a column with different superscripts differ significantly (P≤0.05)

3.2.2 Interaction effect of treatment, incubation time and temperature on transmittance value

As reported in the factorial ANOVA, the combination of treatment and incubation time had the highest interaction effect. The addition of a combination of enzymes incubated at 24 hours gave the highest clarity measurement and this effect is seen when the incubation temperature is 40^0C (Figures 2 & 3). The combined impact of treatment and time is more pronounced in the samples incubated at 40^0C than at 20^0C. A possible reason could be that the 40^0C incubation temperature is closer to the reported optimum temperature for the pectinase activity which is 50^0C and for amylase which is reported to be 45^0C (Khatri, Bhattarai, Shrestha, & Maharjan, 2015).
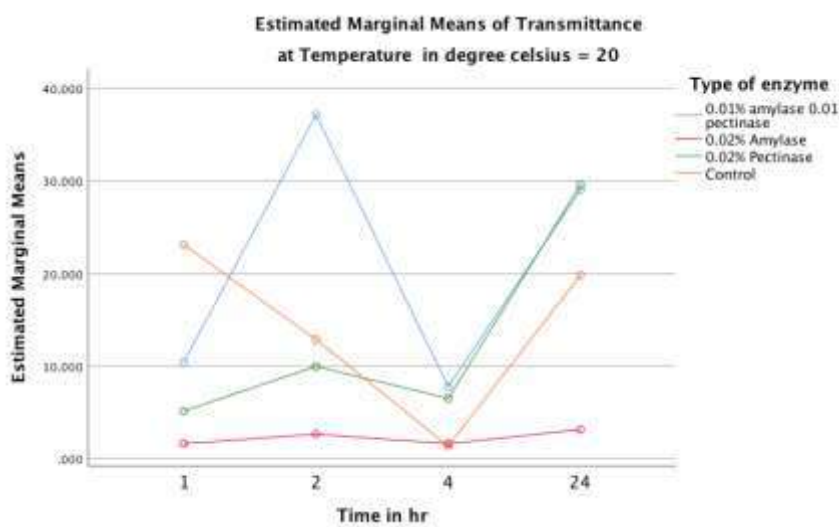

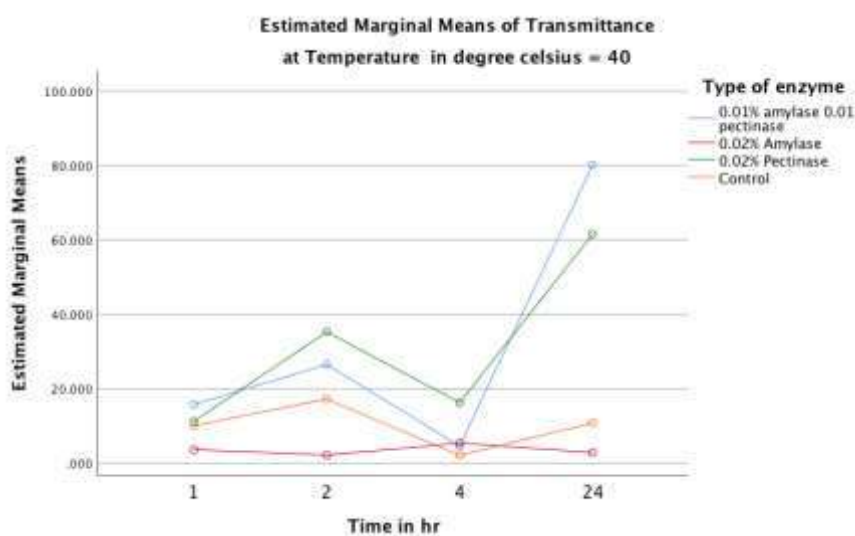Figure 2. Interaction effect plot of incubation time and different treatments at an incubation temperature of 20^0C

Figure 3. Interaction effect plot of incubation time and different treatments at an incubation temperature of 40^0C

The combination of the incubation time and different treatments when enzymes are added at different stages had the second-highest interaction effect. The mix of enzymes added to the pomace before juice extraction and incubated at 24-hour gave a comparable transmittance value to the pectinase enzyme added directly to the juice and incubated at 24-hour duration. Thus, if only the type of enzyme is to be considered in instances where maintaining incubation time and temperature would be difficult, a mix of pectinase and amylase added to the pomace before juice extraction will give juice with better clarity. This can be attributed to the synergistic mechanism whereby amylase prevents the agglomeration of starch with protein pectin complex leading to the prevention of haze and better clarity in the juice. As the mix of enzymes is added directly to the pomace, the synergistic effect starts in the pomace even before the juice is extracted and within 1 hour of incubation time, the clarity of the juice is higher as compared to the other treatment. However, with increasing incubation time and higher incubation temperature, the clarity of the sample where pectinase is added directly to the juice gave a comparable result.
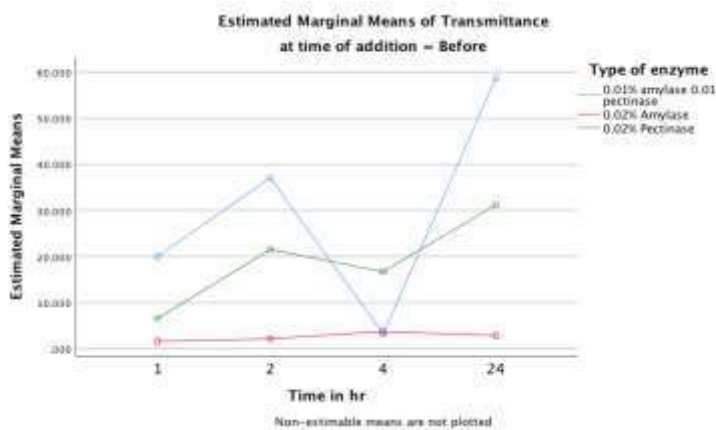

Figure 4. Interaction effect plot of incubation time and different treatment when enzyme is added to the pomace before juice extraction
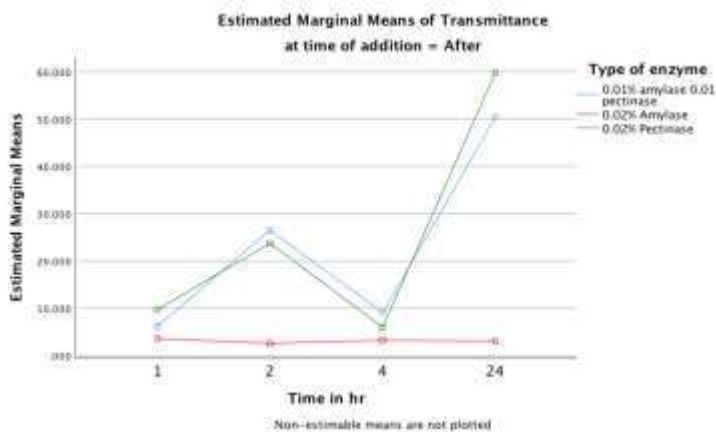

Figure 5. Interaction effect plot of incubation time and different treatments when enzyme is added to the juice

192

4. Conclusion

The study found that the use of enzymes did not achieve significantly different juice recovery percentages. For clarification of apple juice on a commercial scale, the process parameters must be optimized to achieve good quality clear apple juice. When it comes to the precedence of optimizing the process parameters, the most significant parameter is the type of enzyme to be used followed by the incubation time and temperature. However, since this result is based on the main effect, this case is only applicable when other process parameters such as temperature and incubation time are not considered. Thus, if the processor can only control one parameter, it would be prudent to give priority to the type of enzymes to be used and then the incubation time followed by temperature. In this study using a mix of amylase and pectinase enzyme gave the highest clarity measurement without taking into consideration the stage of enzyme addition, incubation temperature and time. Similarly, a 24-hour incubation time and the temperature maintained at 40^0C gave higher clarity juice. However, if various process parameters can be optimized to select a combination that can help achieve juice with significantly higher clarity, then the use of pectinase enzyme added directly to the juice and the mix of enzyme added to the pomace before juice extraction incubated for 24 hours at 40^0C is recommended.

Acknowledgement

References

Berutu, C. A. M., Fahrurrozi, F., & Meryandini, A. (2017). Pectinase production and clarification treatments of apple (*Malus domestica*) juice. *Annales Bogorienses, 21*(2), 63-68. doi:http://doi.org/10.14203/ab.v21i2.311

Chang, T. S., Siddiq, M., Sinha, N. K., & Cash, J. N. (1995). Commercial Pectinase and Yield Quality of Stanley Plum Juice 1. *Journal of Food Processing and Preservation, 19*(2), 89-101. doi:doi: 10.1111/j.1745-4549.1995.tb00280.x

Choden, J., & Shanawaz. (2015). Apple Cultivation in Bhutan - Past Trends and Future Potential. In Shanawaz & U. Thinley (Eds.), *Climate Change, Environment and Development in Bhutan : Occasional publication No.1* (pp. 65-72). Wangdue: College of Natural Resources (CNR), Royal University of Bhutan (RUB).

DAMC. (2017). *Why are growers unable to tap the raw material demand of local agro-industries?* . Thimphu: Department of Agricultural Marketing and Cooperatives (DAMC), Ministry of Agriculture and Forests (MoAF), Royal Government of Bhutan.

Dey, T. B., & Banerjee, R. (2014). Application of decolourized and partially purified polygalacturonase and α-amylase in apple juice clarification. *Brazilian journal of microbiology, 45*(1), 97-104. doi:doi:10.1590/S1517-83822014000100014

DoA. (2016). *Agriculture Statistics 2016*. Thimphu: Department of Agriculture (DoA), Ministry of Agriculture and Forests, Royal Government of Bhutan

Ezugwu, A. L., Ezike, T. C., Ibeawuchi, A. N., Nsude, C. A., Udenwobele, D. I., Eze, S. O. O., . . . Chilaka, F. C. (2014). Comparative studies on pectinases obtained from Aspergillus fumigatus and Aspergillus niger in submerged fermentation system using pectin extracted from mango, orange and pineapple peels as carbon sources. *Nigerian Journal of Biotechnology, 28*, 26-34.

Garner, D., Crisosto, C. H., Wiley, P., & Crisosto, G. M. (2008). Measurement of soluble solids content. 17(4), 2. *Central Valley Postharvest Newsletter, 17*(4), 2-3.

Khatri, B. P., Bhattarai, T., Shrestha, S., & Maharjan, J. (2015). Alkaline thermostable pectinase enzyme from Aspergillus niger strain MCAS2 isolated from Manaslu Conservation Area, Gorkha, Nepal. *SpringerPlus, 4*(1), 1-8.

Kilara, A., & Buren, J. P. V. (1989). Clarification of apple juice. In D. L. Downing (Ed.), *Processed apple products* (pp. 83-96). New York: Springer.

Kothari, M. N., Kulkarini, J. A., & Baig, M. M. V. (2013). Clarification of Apple Juice by Using Enzymes and their Mixture. *World Research Journal of Biotechnology, 1*(2), 29-31.

McLellan, M., & Padilla-Zakour, O. (2004). Juice Processing. In D. M. Barrett, L. Somogyi, & H. S. Ramaswamy (Eds.), *Processing fruits: science and technology* (2 ed.). Florida: CRC Press.

Oszmiański, J., Wojdyło, A., & Kolniak, J. (2009). Effect of enzymatic mash treatment and storage on phenolic composition, antioxidant activity, and turbidity of cloudy apple juice. *Journal of Agricultural and Food Chemistry, 57*(15), 7078-7085. doi:https://doi.org/10.1021/jf900806u

Padma, P. N., Sravani, P., Mishra, P., Sneha, N., & Anuradha, K. (2017). Synergistic effect of multiple enzymes on apple juice clarification. *Indian J. Sci. Technol, 10*(10), 1-5.

Patidar, M. K., Nighojkar, S., Kumar, A., & Nighojkar, A. (2018). Pectinolytic enzymes-solid state fermentation, assay methods and applications in fruit juice industries: a review. *3 Biotech, 8*(4), 1-24. doi:10.1007/s13205-018-1220-4

Pollard, A., & Timberlake, C. F. (1971). Fruit Juices. In A. C. Hulmes (Ed.), *The Biochemistry of Fruits and their Products* (pp. 573-621). London: Academic Press.

Rai, P., Majumdar, G. C., Dasgupta, S., & De, S. (2004). Optimizing pectinase usage in pretreatment of mosambi juice for clarification by response surface methodology. *Journal of Food Engineering, 64*(3), 397-403. doi:https://doi.org/10.1016/j.jfoodeng.2003.11.008

Rana, N., Verma, N., Vaidya, D., & Dipta, B. (2017). Application of bacterial amylase in clarification of juices and bun making. *J Pharm Phytochem, 6*(5), 859-864.

Root, W. H., & Barrett, D. M. (1996). Apples and apple processing. In D. M. B. LP Somogyi, & YH Hui (Ed.), *Processing fruits: science and technology* (Vol. 2, pp. 1-35). New York: CRC Press.

RSD. (2020). *Agriculture Statistics 2019*. Thimphu: Renewable Natural Resources Statistics Division (RSD), Ministry of Agriculture and Forests, Royal Government of Bhutan

Ryan, J. J. (1972). Chemical composition of Canadian apple juice. *Journal of the Association of Official Analytical Chemists, 55*(5), 1104-1108. doi:https://doi.org/10.1093/jaoac/55.5.1104

Shiv, K. (2015). Role of enzymes in fruit juice processing and its quality enhancement. *Advances in Applied Science Research, 6*(6), 114-124.

Singh, R., & Singh, R. K. (2015). Role of Enzymes in Fruit juices Clarification during Processing: A review. *Int.J.Biol.Technology, 6*(1), 1-12.

Sorrivas, V., Genovese, D. B., & Lozano, J. E. (2006). Effect of pectinolytic and amylolytic enzymes on apple juice turbidity. *Journal of Food Processing and Preservation, 30*(2), 118-133. doi: https://doi.org/10.1111/j.1745-4549.2006.00054.x

Umsza-Guez, M. A., Rinaldi, R., Lago-Vanzela, E. S., Martin, N., Silva, R. d., Gomes, E., & Thoméo, J. C. (2011). Effect of pectinolitic enzymes on the physical properties of caja-manga (Spondias cytherea Sonn.) pulp. *Food Science and Technology, 31*(2), 517-526. doi:doi:10.1590/S0101-20612011000200037

Yamasaki, M., Yasui, T., & Arima, K. (1964). Pectic enzymes in the clarification of apple juice: part I. Study on the clarification reaction in a simplified model. *Agricultural and Biological Chemistry, 28*(11), 779-787. doi:doi:10.1080/00021369.1964.10858304

Adaptation of New Adzuki Bean Variety for Organic Production System in Bhutan

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ABSTRACT

Adzuki bean (Vigna angularis Willd.) is a native legume that is cultivated by subsistence Bhutanese farmers as one of the many other pulses for household food security. Several scientific literatures have revealed that many Bhutanese Adzuki bean germplasm has been assessed for genetic variability confirming that Adzuki bean is native to Bhutan. The cultivation of Adzuki bean currently is confined to small areas as a marginal crop for household consumption. The drive towards a certified organic production system in Bhutan has called for an urgent need to adapt, release and promote organic technologies, particularly on organic pests and diseases, and sustainable soil fertility management. Adaptation of high-yielding Adzuki bean varieties and their commercialization as a health food, high-value crop for export, and a good source of nitrogen for sustainable soil fertility management was studied. One new Japanese variety Erimo was rapidly evaluated in the researcher-managed and farmer-managed trials under the organic production system at different locations. The seed yield in the researcher-managed trial was significantly different over locations (P<0.05) and ranged from 540 to 1215 kg ac^{-1} with a mean yield of 907.50 kg ac^{-1}. Under the farmer-managed trials in the 2020 season, seed yield ranged from 150 to1080 kg ac^{-1} with a mean of 431.40 kg ac^{-1}. The days to crop maturity ranged from 104 to 126 days which fits well into the farmers' existing cropping system. The 23rd Variety Release Committee (VRC) of the DoA endorsed the release of this variety Erimo with the local name Yusi-Adzuki. This variety will be promoted for commercial cultivation for export to Japan, and as a sustainable soil fertility management technology in the organic production system.

Keywords: *Adzuki bean; Legume; Organic system; Yield; Variety release*

1. Introduction

Adzuki bean (*Vigna angularis* Willd.*)* commonly known as adzuki bean, red bean, and red mung bean is an annual legume that belongs to the Fabaceae family (Sindhu & Manickavasagan, 2020). The scientific community has established that the Adzuki bean was domesticated in China 12,000 years ago and it is currently cultivated in more than 30 countries, especially in the East Asia region. In Bhutan, the Adzuki bean is traditionally cultivated by

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subsistence farmers as one of the many pulses for household food security. However, it has largely remained as a marginal crop that is cultivated in a small area for household consumption because of its low economic value. Several scientific literatures mention the assessment of Bhutanese Adzuki bean germplasm for genetic variability which indicates that Adzuki bean is native to the Bhutanese agriculture systems. A study conducted by Isemura et al. (2001) has reported the use of 43 Bhutanese Adzuki bean strains which demonstrated region-specific traits and distinct differences in morphological characters. In another study on geographical distribution and evolutionary relationships between cultivated and wild Adzuki bean, cultivated Adzuki bean germplasm from Bhutan and Nepal have been used indicating that Adzuki bean is indigenous to Bhutan (Xu-xiao et al., 2003). These studies confirm the cultivation of Adzuki bean in Bhutan and its role in the Bhutanese food system.

Adzuki bean is a traditional legume crop that plays an important role as a source of protein and a versatile nitrogen-fixing legume that has been used for biological nitrogen fixation for sustainable soil fertility management in agriculture (Sun, Shahrajabian, & Cheng, 2019). Further, Adzuki bean is considered to be highly nutritious that is rich in protein, minerals, carbohydrates, and fibre, and is used in various forms such as extract and paste (Kimura et al., 2004; Sindhu & Manickavasagan, 2020). According to Shahrajabian et al. (2019), Adzuki bean has considerable potential to be a global functional food for health, nutrition, and prevention of many lifestyle diseases. It has broad adaptability, high tolerance to poor soil fertility, and is a high-value rotation legume crop that contributes to the improvement of soil through nitrogen fixation under organic farming systems (Kharwal, Singh, & Bhardwaj, 2020). It has been concluded by Kimura et al. (2004), that Adzuki bean adds a significant level of nitrogen in the form of nitrate-nitrogen (NO_3-N) in the soil after its cropping cycle. The root system of Adzuki beans has a high ability for symbiotic nitrogen fixation and the crop has the special ability to accumulate nitrogen in the early reproductive stage. In an organic production system, maintaining and sustaining soil fertility remains a practical challenge since the use of synthetic fertilizers is prohibited. Amongst many other approaches for maintaining soil fertility, the cultivation of legumes for biological nitrogen fixation through suitable crop rotations and intercropping practices is a feasible option recommended to farmers (Forster et al., 2012). The other advantages of legume cultivation, apart from food security, include huge benefits to soil by way of reducing its compaction, and erosion, improving its structure, enhancing organic matter and soil microbial activity, and nitrogen content through nitrogen fixation (Kocira et al., 2020). Further, Wyngaarden, Gaudin, Deen, and Martin (2015), have concluded that the

inclusion of a suitable legume as cover crops in a cropping system increases agro-ecological resilience and crop productivity through nitrogen fixation, soil temperature, and moisture regulation; reduction of erosion, runoff, and leaching; suppresses weeds and disrupts the cycle of established pests and diseases. Adzuki bean could also be promoted as a cover crop to improve both soil fertility and productivity in the organic system.

Bhutanese agriculture represents a traditional mountain farming system that is integrated into sustaining livelihood, the health of soils, ecological processes, biodiversity, and nutrient cycle with only 37% of the farmers using agrochemicals in 19% of the cultivable land. This situation provides a strong basis to adopt organic farming (MoAF, 2018). Through the National Organic Flagship Program (NOFP), several potential landscapes have been supported for rapid conversion from natural farming to a certified organic production system. This drive and focus towards a certified organic production system have created a pressing necessity to adapt, release and promote organic technologies, particularly on organic pests and diseases, and sustainable soil fertility management.

Grain legumes are an important component of Bhutanese agriculture production systems and are popularly grown as pulses, vegetables, and fodder for household food security, animal feeds, and for sale in cases of small surpluses. The role of legumes as a contributor to soil nutrient management is poorly understood and rarely exploited by farmers. However, any crop in subsistence smallholder systems has to first contribute to household food security and income generation. The national drive to transition to Organic production systems thus provides an enormous opportunity for the integration of potential legumes that serve as a source of food, income, and soil nutrient management. In the warm temperate agroecosystem (1800 -2600 masl), apart from Peas (*Pisum sativum*), there are no suitable legumes with high market value resulting in limited diversity of legume crops, particularly for the organic production system. The introduction and adaptation of new high-yielding varieties of Adzuki bean which is a nutritious and healthy food with a very good market potential in Japan, and high nitrogen fixation ability provides Bhutanese farmers with a new opportunity. The four underlying complimentary benefits of including pulses in the food systems are food security, health, sustainable agriculture, and adaptation to climate change emanating from its large genetic diversity and climate-resilient varieties (Calles., 2016). In a study to assess the effects of large-scale conversion to organic farming in Bhutan, Feuerbacher et al. (2018) have recommended the adoption of nitrogen-fixing crops, improved animal husbandry systems to enhance animal manure production, and access to markets with a price premium for organic products as some

of the suitable adaptation strategies to bridge the average yield gap of 24% between organic and conventional production systems.

The work on the adaptation of the Japanese Adzuki bean variety *Erimo* was initially started in 2013 at the National Centre for Organic Agriculture (NCOA), Yusipang which was declared an organic farm in 2004 by the Department of Agriculture (DoA). However, the work was discontinued because the desired volume required by the Japanese entrepreneur for export could not be met by the farmers. In 2020, NCOA, Yusipang was again assigned to coordinate and undertake the fast-track evaluation and release of Adzuki bean variety with the reintroduction of the Japanese variety *Erimo*. Accordingly, NCOA Yusipang initiated the evaluation of this variety through researcher-managed trials and on-farm trials under farmers' management under an organic production system. *Erimo* variety is used to prepare bean paste and is the most preferred variety for Japanese consumers. This variety bred for the Japanese market is considered superior to Blood Wood, another high-yielding and early maturing variety highly popular in the Japanese market (Motley, McCaffery, & Lachlan 2004).

The three underlying objectives of this study were to rapidly assess the adaptability of the new Japanese Adzuki bean variety *Erimo* under an organic production system in the warm temperate agro-ecology in Bhutan; to release a new high yielding variety with a good yield potential that is acceptable for export to Japan; and to recommend and upscale the commercial cultivation of Adzuki bean as a new alternative high-value legume for food security, income, and sustainable soil fertility management under the organic production system in Bhutan.

2. Materials and Method

In the 2020 cropping season, the researcher-managed trials and on-farm trials adaptation were conducted under the organic production system. The researcher-managed trials were conducted under the direct supervision of researchers at three organic sites: Yusipang and Khariphu in Thimphu, and Khatoe in Gasa (Table 1). Adzuki beans variety *"Erimo"* supplied by the Japanese company SUN SMILE Co., Ltd. was evaluated in large observation plots. At Yusipang and Khariphu, the crop was established on the 14th and 15th of May 2020, respectively while at Khatoe the crop was planted on the 2nd of July, 2020. Seeds were sown in line with a row-to-row distance maintained at 0.65 m and plant-to-plant spacing of 0.20 m. At least three to four seeds were sown on each hill without the preparation of any raised beds. The seed rate used was 20 kg ac^{-1}. Locally produced Farm Yard Manure (FYM), vermicompost, and chicken manure were applied in sufficient quantity during the field preparation. Being an organic

production system no in-organic fertilizers and pesticides were applied. Crops were grown under rain-fed conditions. At least four times manual weeding was done to keep the crop free of weed competition.

On-farm adaptation trials managed by farmers were conducted under the organic production system in five locations (Table 3). In the on-farm trials, seed sowing started in May and continued till the end of July depending on the altitude of the location. In higher elevations, early planting was started by the first week of May and continued till mid-June. In all the trial sites, seed sowing was demonstrated by the field crops researchers from the NCOA and Agriculture Research and Development Centre (ARDC), Wengkhar, Mongar.

For both types of trials, data were collected by the researchers from NCOA, Yusipang, and ARDC, Wengkhar with assistance from the dzongkhag agriculture extension officers. At each site, three standard plots (Department of Agriculture [DoA], 2020) each measuring 6 m^2 were randomly marked for collecting the data on yield parameters. Data on morphological characteristics were collected as per the International Union for Protection of New Varieties of Plants (UPOV) guidelines for the conduct of tests for distinctness, uniformity, and stability in Adzuki bean (UPOV, 2015). Both qualitative and quantitative data were collected to evaluate its performance. Data was compiled and computed in MS Excel which was then analyzed using SPSS Version 22.

3. Results and Discussion

The rapid assessment of this new Japanese variety under the organic production system in 2020 and further adaptation in some new locations in 2021 have shown promising results. Results from the researcher-managed trials and on-farm farmer-managed trials are discussed.

3.1 Evaluation under researcher managed trial in three sites under organic production system

In 2020, the researcher-managed trials were conducted in five locations under the organic production system (Table 1). The seed yield was significantly different over locations (P<0.05). The seed yield ranged from 540 to 1215 kg ac^{-1} with a mean yield of 907.50 kg ac^{-1}. The seed yield at different locations differed significantly and the difference among the sites could be underpinned by various factors such as soil fertility, water availability, and crop management. The higher mean seed yield at Yusipang and Khariphu is attributable to a higher number of pods per plant as compared to in Khariphu. Further, the crop at Yusipang was given supplementary irrigation and optimum weed management as compared to two other sites due

200

to distance. Researchers at the CSK Himachal Pradesh Krishi Vishvavidyalaya, Palampur in India who have evaluated 15 genotypes of Adzuki bean under inorganic and organic production systems have reported the mean seed yield of 100.36 and 284.08 kg ac^{-1}, respectively (Kharwal et al., 2020). The mean seed yield of Adzuki bean in China which is considered the centre of origin of this crop is 638 kg ac^{-1} or 1595 kg ha^{-1} (Li et al., 2017). In a yield trial of 231 Chinese germplasm evaluated in Australia, a mean seed yield of 824.00 kg ac^{-1} (2,060 kg ha^{-1}) has been reported by Wang et al. (2001). The mean seed yield of Japanese varieties *Erimo* and *Bloodwood* which were included as checks in this trial was 656 kg ac^{-1} (1,640 t ha^{-1}) and 640 kg ac^{-1}(1,600 t ha^{-1}), respectively.

Table 7. Seed yield and other agronomic traits in three locations, 2020

Locations	Plant height (cm)	Length of pod (cm)	Number of pods/plant	Number of seeds/pod	Seed yield kg ac^{-1}
Yusipang	67.93	11.56	60	6	1215.00
Khatoe	37.47	9.90	34	6	540.00
Khariphu	67.89	10.57	61	7	967.50
Mean	57.76	10.68	52	6	907.50
P value	<0.001	<0.001	<0.05	<0.001	<0.05
Std. Dev (±)	17.57	0.84	15	0.58	341.48

Adzuki as a new crop has to be accepted by the Bhutanese farmers and it has to fit into their established cropping system. The days to maturity of any crop are very critical to recommend and fit this crop in the farmer's existing system. The days to maturity of Adzuki bean in three researcher-managed trial sites ranged from 104 to 126 days with a mean value of 116 days (Table 2).

Table 8. Crop maturity (days) in three locations, 2020

Sites	Date of sowing	Date of Final harvest	Days to 50 % Flowering	Days to maturity
Yusipang	14.5.2020	17.9.2020	65	126
Khariphu	15.5.2020	11.9.2020	70	119
Khatoe	2.7.2020	13.10.2020	70	104
Mean			68	116

3.2 On-Farm evaluation in five sites under organic production system

In the farmer-managed on-farm trials under organic production system in five sites in the 2020 cropping season, the seed yield ranged from 150 to1080 kg ac^{-1} with a mean of 431.40 kg ac^{-1} (Table 3). In the 2021 cropping season, Adzuki bean was again evaluated in more sites under the organic production system. The seed yield recorded in 2021 ranged from 200 to 782.61 kg ac^{-1} with a mean yield of 427.20 kg ac^{-1} (Table 4). The seed yield recorded under farmers'

management in both years is comparable to yields obtained in other countries although the mean yield is comparatively low. One of the critical yield-limiting factors observed in the farmer's field was heavy weed pressure. In the warm temperate areas, Adzuki season falls in the summer season which favours high weed competition. Yield loss from weed infestation is a major production impediment in Adzuki beans (Soltani, Brown, & Sikkema, 2020). The package of practices for Adzuki bean production needs to evaluate and recommend suitable weed control measures as the shortage of farm labour is a major constraint in Bhutan.

Table 9. Seed yield of Adzuki bean, under organic system under farmers management, 2020

Dzongkhag	Gewog	Village	Altitude (masl)	Seed yield kg ac^{-1} at 14% MC
Thimphu	Mewang	Khariphu	2280	1080
Paro	Luni	Chimkha	2700	150
Haa	Samar	Nobgang	2600	150
Gasa	Khatoe	Jabisa	2500	540
Mongar	Mongar	Mongar	640-2100	237
Mean yield kg ac^{-1}				431.40
Std. Dev (±)				396.43

Table 10. Seed yield of Adzuki bean under organic system, 2021

Dzongkhag	Gewog	Village	Seed yield kg ac^{-1} at 14% MC
Chhukha	Bongo	Boeri	782.61
Chhukha	Geling	Chanachen	225.00
Paro	Luni	Chimakha	200.00
Thimphu	Chang	Yusipang	501.20
Mean yield kg ac^{-1}			427.20
Std. Dev (±)			273.43

The time for the maturity of Adzuki beans in warm temperate zones ranged from 104 to 126 days. This result also indicates that adzuki can very well fit as a second crop in the warm temperate agro-ecology after the early harvest of potato, the first crop of vegetables, and winter wheat or barley. The most suitable rotations for Adzuki bean in the warm temperate region (1800 -2600 masl) are Adzuki bean – winter wheat; Adzuki bean-barley; Adzuki bean-vegetables (peas, carrot, radish, cole crops); potato- Adzuki bean; vegetables (cole crops)-Adzuki bean; and Adzuki bean- mustard. In the wet and dry sub-tropical region (600-1800 masl) Adzuki bean can fit into several rotations but the most suitable ones are maize- Adzuki bean; winter wheat/barley- Adzuki bean; potato- Adzuki bean; and vegetables- Adzuki bean. In the lower elevations below 600 masl, Adzuki bean can be grown as a winter crop after harvesting maize.

3.3 Phenotypic characteristics

It is important to study the crop phenotypic appearances of new crops. The basic phenotypic characteristics based on the UPOV DUS scale observed in the warm-temperate agroecology in Bhutan are presented in Table 5. The adzuki bean variety is an indeterminate type with sequential flowering and at least three harvests have to be done.

Table 11. Phenotypic characteristics of Adzuki bean variety Erimo as expressed under the Warm- temperate agroecology in Bhutan, 2020

No	Characters	Phenotypic descriptions (UPOV DUS Scale)	Observations
1	Days to flowering	50% Flowering	68
2	Plant growth type	Dwarf, climbing	Dwarf
3	Leaf colour	Very light, light, medium, dark. very dark	Light-medium green
4	Leaf shape	Triangular, circular, rhombic, triangular or rhombic, circular or rhombic	Rhombic
5	Flower bracts	Small, medium, large.	Medium
6	Flower colour	White, pinkish-white, pink, violet	Light yellow
7	Pod colour (mature)	Yellow, green, violet, brown	Brown
8	Pod shape	Concave, S-shaped, convex, cylindrical	Cylindrical
9	Seed shape	Circular, circular to elliptical, elliptic, cylindrical, kidney-shaped, rectangular	Cylindrical

4. Conclusion

NCOA Yusipang has adapted Adzuki bean as a potential legume crop for the organic production system with a high nutritional and commercial value and with an assured export market in Japan. Adzuki bean is a versatile pulse that can be successfully cultivated from humid sub-tropical (150-600 masl) to warm temperate (1800 -2600 masl) agroecology in Bhutan. Appreciable seed yields have been obtained even under farmers' management. Private entrepreneurs have shown keen interest to export Adzuki beans to Japan and have already offered a farm gate price of Nu. 250 per kg. However, production at the required scale for the export market with high-quality beans needs to be achieved. Several studies elsewhere have recommended that the inclusion of a suitable legume in the organic production system is considered one of the most sustainable options to improve and enhance soil fertility. Adzuki bean fixes atmospheric nitrogen symbiotically with special bacteria (rhizobia) that thrive in the root nodules. The promotion of Adzuki bean as a commercial pulse under the organic production systems will have two far-reaching benefits - income generation and sustainable soil fertility improvement. Competition from weeds has been observed as one of the major yield-limiting factors in Adzuki beans. It is therefore very important to evaluate and

recommend suitable weed control methods as the shortage of farm labour is a major issue in Bhutan. With the endorsement of the release of the new variety *Yusi-Adzuki* by the 23rd VRC of the DoA, and assured export market, seed production and rapid promotion of this crop need to be taken up in the organic landscapes across the country. Further, the commercial cultivation of Adzuki beans as new organic technology will contribute to household food security, income generation and serve as a pragmatic option for sustainable soil fertility management under the organic production system in Bhutan.

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References

Calles., T. (2016). *The International Year of Pulses: what are they and why are they important?* Retrieved from Rome: https://www.fao.org/3/bl797e/bl797e.pdf

Feuerbacher, A., Luckmann, J., Boysen, O., Zikeli, S., & Grethe, H. (2018). Is Bhutan destined for 100% organic? Assessing the economy-wide effects of a large-scale conversion policy. *PloS one, 13*(6), e0199025. doi:https://doi.org/10.1371/journal.pone.0199025

Forster, D., Adamtey, N., Messmer, M. M., Pfiffner, L., Baker, B., Huber, B., & Niggli, U. (2012). Organic agriculture - driving innovations in crop research. In G. S. Bhullar & N. K. Bhullar (Eds.), *Agricultural Sustainability-Progress Prospects in Crop Research* (pp. 21-46). London: Elsevier.

Isemura, T., Noda, C., Mori, S., Yamashita, M., Nakanishi, H., Inoue, M., & Kamijima, O. (2001). Genetic variation and geographical distribution of Azuki bean (*Vigna angularis*) landraces based on the electrophoregram of seed storage proteins. *Breeding Science, 51*(4), 225-230. doi:https://doi.org/10.1270/jsbbs.51.225.

Kharwal, N., Singh, N., & Bhardwaj, N. (2020). Performance of different genotypes of Adzuki bean [*Vigna angularis* (Willd.) Ohwi & Ohashi] under inorganic and organic farming conditions. *Himachal Journal of Agricultural Research, 46*(1), 84-90.

Kimura, S. D., Schmidtke, K., Tajima, R., Yoshida, K., Nakashima, H., & Rauber, R. (2004). Seasonal N uptake and N 2 fixation by common and adzuki bean at various spacings. *Plant Soil, 258*(1), 91-101. doi:https://doi.org/10.1023/B:PLSO.0000016539.73233.

Kocira, A., Staniak, M., Tomaszewska, M., Kornas, R., Cymerman, J., Panasiewicz, K., & Lipińska, H. (2020). Legume cover crops as one of the elements of strategic weed management and soil quality improvement. A Review. *Agriculture, 10*(9), 394. doi:https://doi.org/10.3390/agriculture10090394

Li, L., Yang, T., Liu, R., Redden, B., Maalouf, F., & Zong, X. (2017). Food legume production in China. *The Crop Journal, 5*(2), 115-126. doi:https://hdl.handle.net/20.500.11766/6316

MoAF. (2018). *Flagship Program on Organic Sector Development for Sustainable Food Security and Livelihood*. Thimphu: Ministry of Agriculture and Forests (MoAF), Bhutan.

Motley, K., McCaffery, D., & Lachlan , D. M. (2004). *Azuki beans: Irrigated planting guide 2004-2005*. Sydney: New South Wales Department of Primary Industries.

Shahrajabian, M. H., Sun, W., Khoshkharam, M., Zandi, P., & Cheng, Q. (2019). Adzuki beans (*Vigna angularis*), a traditional Chinese legume for sustainable agriculture and food production. *Journal of Biological and Environmental Sciences, 13*(38), 79-84.

Sindhu, & Manickavasagan. (2020). Adzuki Bean. In A. Manickavasagan & P. Thirunathan (Eds.), *Pulses: Processing and Product Development* (pp. 1-16). Cham, Switzerland: Springer.

Soltani, N., Brown, L., & Sikkema, P. H. (2020). Weed Management in Azuki Bean with Postemergence Herbicides. *American Journal of Plant Sciences, 11*(9), 1467-1477. doi:https://doi.org/10.4236/ajps.2020.119106

Sun, W., Shahrajabian, M. H., & Cheng, Q. (2019). *Adzuki bean, Soybean and Mungbean, three legumes in Traditional Chinese Medicine with Super Nitrogen-Fixing Capabilities* (First ed.). New Delhi: AkiNik Publications

UPOV. (2015). *Guidelines for the conduct of tests for Distinctness, Uniformity and Stability. TG/312/1. Adzuki Bean, UPOV Code: VIGNA_ANG*. Geneva: International Union for the Protection of New Varieties of Plants (UPOV).

Wang, S., Redden, R., Jiapeng, J. H., Desborough, P., Lawrence, P., & Usher, T. (2001). Chinese adzuki bean germplasm: 1. Evaluation of agronomic traits. *Australian journal of agricultural research, 52*(6), 671-681. doi:https://doi.org/10.1071/AR00104

Wyngaarden, S. L., Gaudin, A., Deen, W., & Martin, R. C. (2015). Expanding red clover (*Trifolium pratense*) usage in the corn–soy–wheat rotation. *Sustainability, 7*(11), 15487-15509. doi: https://doi.org/10.3390/su71115487

Xu-xiao, Z., Vaughan, D., Tomooka, N., Kaga, A., Xin-wang, W., Jian-ping, G., & Shu-min, W. (2003). Preliminary study on geographical distribution and evolutionary relationships between cultivated and wild adzuki bean (*Vigna angularis* var. *angularis* and var. *nipponensis*) by AFLP analysis. *Plant Genetic Resources, 1*(2-3), 175-183. doi:10.1079/PGR200327