# **EFFECT OF SPACING AND VARIETY ON BIOMASS PRODUCTION AND QUALITY OF** *Moringa oleifera* L.

# **MD. HANIF HOSSAIN**



# DEPARTMENT OF AGROFORESTRY AND ENVIRONMENTAL SCIENCE SHER-E-BANGLA AGRICULTURAL UNIVERSITY DHAKA-1207

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# **EFFECT OF SPACING AND VARIETY ON BIOMASS PRODUCTION AND QUALITY OF** *Moringa oleifera* L.

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# MD. HANIF HOSSAIN REGISTRATION NO. 19-10043

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**Approved by:** 

-----

Dr. Md. Kausar Hossain Professor Supervisor Dr. Nasrin Sultana Director (Research) Co-Supervisor

-----

-----

Dr. Jubayer-Al-Mahmud Chairman Examination Committee



#### Dr. Md. Kausar Hossain

Professor

Department of Agroforestry and Environmental Science Sher-e-Bangla Agricultural University Sher-e-Bangla Nagar, Dhaka-1207 Cell No. +880 1701777941 E-mail: kausar@sau.edu.bd Website: www.sau.edu.bd

Memo No.: SAU/Agroforestry & Environmental Science Date:

# CERTIFICATE

This is to certify that thesis entitled, "EFFECT OF PLANTING DENSITY AND VARIETY ON BIOMASS PRODUCTION AND QUALITY OF Moringa oleifera L." submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of OF SCIENCE IN AGROFORESTRY AND MASTER ENVIRONMENTAL SCIENCE, embodies the result of a piece of bona fide research work carried out by MD. HANIF HOSSAIN No.19-10043 under my supervision Registration and guidance. No part of the thesis has been submitted for any other degree or diploma.

I further certify that such help or source of information, as has been availed of during the course of this investigation has duly been acknowledged.

Dated: June, 2021 Dhaka, Bangladesh

Dr. Kausar Hossain Professor Supervisor

# ΟΕΦΙΓΟΑΤΕΦ ΤΟ ΜΥ ΒΕΙΟΥΕΦ ΡΑΓΕΝΤΟ ΜΑ ΓΕΓΟΛΕΦ ΓΑΚΕΛΙΖ

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The author

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Moringa oleifera L.

#### ABSTRACT

The experiment was conducted at the Cattle Research Farm, Pachutia, Bangladesh Livestock Research Institute (BLRI), Savar, Dhaka-1341, Bangladesh, with different types of available varieties of *Moringa oelifera* during the period from August 2019 to August 2020 to evaluate the foliage production of Moringa as a fodder. The experiment consisted of four varieties and four spacing V1 (PKM-1), V2 (PKM-2), V3 (Paraynal), V4 (Black), and D1 ( $1.0 \times 1.0$  feet), D2 ( $1.0 \times 1.5$  feet), D3 ( $1.5 \times 1.5$  feet), D4 ( $2.0 \times 2.0$ feet). The treatment combinations are  $T_1 = V_1D_1$ ;  $T_2 = V_2D_1$ ;  $T_3 = V_3D_1$ ;  $T_4 = V_4D_1$ ;  $T_5 =$ V1D2; T6= V2D2; T7= V3D2; T8= V4D2; T9= V1D3; T10= V2D3; T11= V3D3; T12= V4D3; T13= V1D4; T14= V2D4; T15= V3D4; T16= V4D4. The experiment was laid out in two factorial Randomized Complete Block Design (RCBD) comprising four replications. Data were collected on plant height (cm), the number of leaves per plant, stem girth (cm), biomass weight (g), dry weight (g), and crude protein (%). The highest plant height (291.3 cm) was found in T1 treatment combination at 220 DAT. At 260 DAT, significantly highest leaves per plant (45.95) were observed in T7. At 220 DAT the stem girth was observed the highest in T1 treatment combination (7.8 cm). Maximum (96.5 %) in T11 treatment combination was recorded at first harvest (180 DAT). At 220 DAT, a significantly highest biomass yield was observed in T1. Significantly highest dry matter per plant (24.21%) was observed in T1 at 260 DAT. The plants under T11 treatment combination were found maximum in crude protein (19.865) at 260 DAT. Black variety shows the highest survival rate. T1 (PKM-1 variety with 1×1 feet spacing) treatment combination exhibited the highest performance in respect of growth and yield of Moringa followed by T4 (Black variety with  $1 \times 1$  feet spacing).

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# **ABBREVIATIONS AND ACRONYMS**

SAU	=	Sher-e-Bangla Agricultural University
BLRI	=	Bangladesh Livestock Research Institute
BBS	=	Bangladesh Bureau of Statistics
AEZ	=	Agro-Ecological Zone
USDA	=	United States Department of Agriculture
WHO	=	World Health Organization
FAO	=	Food and Agriculture Organization
MS	=	Master of Science
RCBD	=	Randomized Complete Block Design
et al.,	=	And others
viz.,	=	Namely
J.	=	Journal
e.g.	=	exempli gratia (L), for example
etc.	=	Etcetera
i.e.	=	id est (L), that is
%	=	Percentage
°C	=	Degree Celsius
cm	=	Centimeter
mm	=	Millimetre
g	=	Gram (s)
LSD	=	Least Significant Difference
No.	=	Number
CV	=	Coefficient of Variation
DAT	=	Days After Transplantation
DM	=	Dry Mater
СР	=	Crude Protein
Т	=	Treatment
MPTS	=	Multi-Purpose Tree Species
CCFB	=	Cut and Carry Fodder Banks
m	=	Meter
М.	=	Moringa

#### **CHAPTER I**

#### **INTRODUCTION**

Moringa, a multi-purpose tree (MPTS) has gained a lot of publicity because of its many varied uses, which range from medicinal applications, industrial, and sanitary to nutritional. The growing interest in *Moringa oleifera* Lam., commonly known as "**Sajna**" in Bangladesh and by several names elsewhere, has resulted in its extensive cultivation worldwide. The multiple uses of moringa are being explored and investigated by researchers and development workers around the world. Moringa has very high nutritional properties that would be useful as a food supplement, especially in those marginalized communities. Besides its nutritional and medicinal applications, *M. oleifera* is very useful as an alley crop in the agroforestry industry DATh and Gupta (2009), to reduce soil erosion and improve soil conservation. Besides *M. oleifera* being processed into medicine, it contains acetone which can be prepared into an herbal formulation which is an effective anti-malaria bioagent (Patel *et al.*, 2010).

As demand for moringa fresh and dry biomass increases there is great potential for increasing supply. One approach for increased production and supply would be to cultivate moringa at high planting density which means closer plant spacing. The choice of spacing and plant density depends on the purpose of moringa cultivation and the intended use of the different plant parts. If the goal is to produce a high amount of leaf biomass, closer spacing and high plant density are required. If seed production is the primary objective, wider spacing should be used. However, if the objective is leaf and seed production, plant spacing should be intermediate.

Fuglie (1999), described many uses of the Moringa tree-like biomass production, animal forage, biogas, domestic cleaning agent, in feeding programs to fight against

malnutrition especially in Africa like countries, as a blue dye, for fencing purposes, as fertilizer, green manure, for gum extraction and honey juice-clarifier, various medicines, as ornamental plantation, as bio-pesticide against seedling damping off, rope making, tannin purpose for tanning hides and water purification.

Because of these diversified significant applications of M. oleifera and its impact on improved livelihoods and health, proper and viable agronomic practices for increased productivity of M. oleifera must be identified and established. Mahn et al. (2005); studied different spacing of Moringa on sulfate acidic soils and the results suggest that Moringa can develop in the sulfate acid soil but the young plant did not adapt well to waterlogged conditions and biomass harvests of up to 52 tons/ha were achieved. Many researchers have shown an increasing interest in trees and shrubs as alternative fodder to cattle in tropical regions. There are different practices in agroforestry systems to produce fodder for cattle such as alley farming, live fences, windbreak lines, tree browsing, and cut and carry fodder banks (CCFB). According to Pezo and Ibrahim (1998), a CCFB is an area where forage ligneous plants are cultivated in a compact block with high planting densities to maximize high-quality biomass production. Jime'nez and Muschler (2001), reported CCFB as a successful practice in livestock systems because improves the diet of the animals mainly during the dry season when the availability of biomass is generally not sufficient to satisfy the nutritional requirements of livestock. Although high densities are positively correlated with high DM yields, the spatial arrangement in the field, the high amount of labor needed, and difficulties during harvesting make high densities impractical for small and mediumscale farmers. Therefore, studies on lower densities more adapted to the practical needs of small and medium-sized farms are still needed.

The main form of cultivation of moringa is in agroforestry practices by smallholder farmers and whose promise for sustainable development lies in the ability to continue producing feed materials for animals and streams of income for farmers over several years. The envisaged demand for moringa plant parts can only be met when production is based on improved agronomic recommendations. One component of improved farming practices that affects crop output is plant population as determined by spacing, plant geometry, and stand density because it influences growth, yield, and yield components (Lauer, 1994).

This study was conducted to determine the leaf biomass yield of moringa as influenced by plant spacing and different varieties. This study established minimum, optimum, and maximum plant spacings for leaf biomass production. Specifically, this study was conducted to describe the influence of plant density and harvest frequency on plant growth parameters including plant height, stem diameter, and the number of side branches concerning leaf biomass yield and leaf quality. It was also done to determine the most appropriate harvest frequency required for maintaining M. oleifera fields meant for leaf production as a continuous crop. Therefore, the present study was undertaken with the following objectives:

- 1. To investigate the effect of variety on seed germination of M. oleifera
- 2. To assess the effect of spacing and variety on biomass production of Moringa
- To evaluate the effect of variety and spacing on chemical components of Moringa at the different cutting interval

#### **CHAPTER II**

#### **REVIEW OF LITERATURE**

This chapter reviews the literature of other scholars and their past studies on agroforestry that is related to the current experiment collected through reviewing of journals, thesis, internet browsing, reports, newspapers, periodicals, and other forms of publications presented.

#### 2.1. Botanical classification of Moringa

Kingdom	: Plantae
Division	: Magnoliophyta
Class	: Magnoliopsida
Order	: Brassicales
Family	: Moringaceae
Genus	: Moringa
Species	: Oelifera
Scientific name	: Moringa oelifera

#### 2.2. Origin and Geographical Distribution of Moringa

In the Monogeneric genus Moringa of the Moringaceae family, there are 13 species *M*. *oleifera* indigenous to sub-Himalayan tracts of Northern India (Paliwal and Sharma, 2015), among which *M. oleifera* has so far become the most used and studied. It is

native to Northern India, Pakistan, and Nepal but has become naturalized well beyond its native range, including many countries of Southeast Asia, the Arabian Peninsula, Tropical Africa, Central America, the Caribbean, and Tropical America (Francis *et al.*, 1991). This species has been dubbed "miracle tree", "natural gift", or "mother's best friend". *M. oleifera* grows in any tropical and subtropical country with peculiar environmental features, namely, dry to moist tropical or subtropical clime, with annual precipitation of 760 to 2500 mm (it requires less than 800 mm irrigation) and temperature between 18 and 28 °C. It grows in any soil type, but in heavy clay and waterlogged, with pH between 4.5 and 8, at an altitude up to 2000 m (Palada, 1996, Nouman *et al.*, 2013).

A study on local uses and geographical distribution of *M. oleifera* (Popoola and Obembe, 2013) that covers the major agro-ecological region in Nigeria, clearly established that "though considered a not indigenous species, *M. oleifera* has found wide acceptance among various ethnic Nigeria, who have exploited different uses (e.g., food, medicine, fodder, etc.). Nowadays, *M. oleifera* and its derivatives are distributed mainly in the Middle East, African and Asian countries (Ojiako, 2011) and are still spreading to other areas.

#### 2.3. Botany of Moringa Tree

*M. oleifera* is fast growing perennial tree, which can reach a maximum height of 7-12m and a diameter of 20-60cm at chest height. The stem is normally straight, the tree grows with a short, straight stem that reaches a height of 1.5-2.0m before it begins branching and it can reach up to 3.0m. The extended branches grow in a disorganized manner as stated by (Morton, 1991) and the canopy is an umbrella shape. The leaves are alternate,

twice or thrice pinnate leaves grow mostly at the branch tips, they are 20-70m grayish downy when young, long petiole with 8-10 pair of pinnate each bearing two pairs of opposite, elliptic or obviate leaflet and at the apex, all 1-2cm with glands at bases of the petiole and pinnate. The flowers, which are pleasantly fragrant, and 2.5cm wide are produced profusely in maxillary, drooping panides 10-25cm long. They are white or cream colored and yellow dotted the base. The five reflexes' sepals are linear lanceolate. The five (5) petals are slender spatulate, surround the five (5) stamen and five (5) nodes, and are flexed except for the lowest as mentioned by (Morton, 1991). The fruits are three-lobed pods that hang down from the branches and are 20-60cm in length when they are dry open into 3 parts, each pod contains between 12-35 seeds. The seeds are round with a brownish semi-permeable seed hull. The hull itself has three (3) wings that run from top to the bottom at, 120-degree intervals. Each tree can produce between 15,000 and 25,000 seeds per year. According to Makker and Becker, (1997) the average weight per seed is 150.3g and the kernel to hull ratio is 7.2cm.

#### 2.4. Moringa as a Potential Miracle Tree

All parts of *M. oleifera* can be used in a variety of ways. Moringa leaves are full of nutrients and vitamins *M. oleifera* leaves were reported to have 25.1% crude protein, 0.50% methionine, and a metabolizable energy value of 227lkcal/kg as mentioned by (Makker and Becker, 1997). A survey of over 120 species of tropical and subtropical edible plants for nutrient content, antioxidant activity, and crop trait indicated that *M. oleifera* is one of the promising crops which could contribute to increased intake of micronutrient and antioxidants as discussed (Morton, 1991). *M. oleifera* leaves can be excellent sources of calcium, potassium and protein. It was also reported that the leaves

of *M. oleifera* plant are an excellent source of vitamins, minerals, and protein perhaps more than any other tropical vegetable. Moringa leaves extract has been reported to exhibit antimicrobial activities including inhibition for the growth of Staphylococcus areas that are commonly isolated from food and animal intestines, it also has a medicinal use among the natives (Morton, 1991). In view of the above importance of Moringa oleifera, and some that are yet to be discovered, it's important to have a comprehensive study of the Plant as a whole as stated by (Morton, 1991).

#### 2.5. General Uses of Moringa

According to Odee, (1998) *M. oleifera* is a perennial tree, having well-developed roots, stem, and leaves; it can be used in many ways. Research on *M. oleifera* tree has accumulated different information on the uses of the plant in human consumption, medicinal uses, animal fodder, water purification, fertilizer, living fence, alley cropping, natural pesticide, fuel wood, and growth hormone for plant.

#### 2.5.1. Human consumption

*M. oleifera* tree has probably been one of the most underutilized tropical crops. Leaves of *M. oleifera* could serve as a valuable source of nutrients for all age groups. In some parts of the world for example Senegal and Haiti, health workers have treated malnutrition in small children, pregnant and nursing women with *M. oleifera* leave powder. The leaves are known as a great source of vitamins and minerals being served raw when cooked or dried as discussed by (Price ,2000). Fuglie, (1999) reported that 8g serving of dried leaves powder will satisfy a child within the age of 13 years with 14% protein, 40% calcium, 23% iron, and nearly all the vitamin A that the child needs

in a day. 100g of leaves could provide women with over a third of her daily need of calcium and give her important quantities of iron, protein, copper, sulfur, and vitamin B. Flowers can be cooked and mixed with other food or fried in butter. They can also be placed in hot water for five minutes to make a kind of tea for drinking. They are also a good source of nectar for honey producing bees. The pod can be eaten from the time they first appear to when they become too woody to snap easily (up to 30cm long). They are cooked like other green beans and have a similar flavor to asparagus. The root is medicinal and similar to horseradish sources can be made from the roots when the seedling is only 60cm tall. According to Fuglie, (1999), the root bark should be completely removed as it contains harmful substances, the root is grinded up and vinegar salt is added.

The World Declaration and the Plan of Action on Nutrition, adopted by 159 countries, at the International Conference on Nutrition organized by the United Nation's Food and Agriculture Organization (FAO) and World Health Organization (WHO) in 1992, states that strategies to combat micronutrient malnutrition should: "Ensure that sustainable food-based strategies are given first priority particularly for populations deficient in vitamin A and iron, favoring locally available foods and taking into account local food habits". Studies have shown Moringa can be a cheap, all year-round, high-quality food for both humans and animals. It is also rich in health promoting phytochemicals such as carotenoids, phenolics (chlorogenic acids), flavonoids (quercitin and kaempherol glycosides), and various vitamins and minerals (Foidl *et al.*, 2001; Becker and Siddhuraju, 2003; Bennett *et al.*, 2003).

#### 2.5.2. Agricultural uses

Fertilizer the seed cake, which is produced by processing the seeds to extract oil, cannot be eaten as it contains harmful substances. However, it contains high levels of proteins and makes a good fertilizer for use in agriculture as stated by Fuglie, (1999). Using Moringa shoot as green manure can significantly enrich agriculture and in this process, the land is first tilled. Moringa seed is then planted 2cm deep at a spacing of 10 x 10 cm (a density of 1 million seeds per hectare). The density can be greater, the only limit to plant density is the availability of seed, water, and fertilizer, after 25 days. As mentioned by Fuglie, (2008). the seedlings are plowed into the soil to a depth of 15 cm and then prepared for the desired crop. Moringa has a large tap root and few lateral roots so it will not compete for nutrients with the crops, it will also add to the nutrients available as it produces many proteins rich leaves. They grow very quickly but do not provide too much shade due to the structure of their leaves. They are also very good as reclaiming, marginal land as stated by (Fuglie ,1999). Fuel wood and other uses, the wood is light but provides fairly good fuel for cooking, it has a density of 0.5 to 0.7 and yields approximately 4,600 kcal/kg. However, Fuglie, (1999) mentioned that it is not suitable for building; the bark fiber is used in making rope, mats and the wood produce a blue dye, chipping of wood can be used to make a good quality paper, the trees also produce viscose resin that is used in the textile industries.

#### 2.5.3. Medicinal Uses

*M. oleifera* is a small tree that is native to North India. It passes by an assortment of names, for example, drumstick tree, horse radish tree, or ben oil tree. All parts of the *M. oleifera* tree can be eaten or utilized as ingredients as a part of customary home-

grown drugs. The leaves and pods are commonly eaten in parts of India and Africa as stated by (Arnarsan, 2015). Around the world, every part of the Moringa tree has been used effectively against varying ailments. The leaves rubbed against the temple can relieve headaches. To stop bleeding from a shallow cut, apply a poultice of fresh leaves, there is an antibacterial and anti-inflammatory effect when applied to a wound or insect bite. Extracts can be used against bacterial or fungal skin complaints. Fuglie, (1999) stated that eating Moringa as a food product is good for those suffering from malnutrition due to the high protein and fiber content. The bark is boiled with potash to treat toothache and the seeds are grinded and taken orally for (H.I.V). The seeds are used for their antibiotic and anti-inflammatory properties to treat arthritis. According to Arnarsan, (2015) rheumatism, gout, cramp, sexually transmitted diseases, and boils. The seed is roasted, pounded, mixed with coconut oil, and applied to the problem area. Seed oil can be used for the same ailments (Arnarsan, 2015). Its multiple uses and potential attracted the attention of farmers and researchers in past historical eras. Ayurvedic traditional medicine says that *M. oleifera* can prevent 300 diseases and its leaves have been exploited both for preventive and curative purposes (Ganguly, 2013). Moreover, a study in the Virudhunagar district of Tamil Nadu India reports Moringa is among the species utilized by traditional Siddha healers (Mutheeswaran et al., 2011). Ancient Egyptians used *M. oleifera* oil for its cosmetic value and skin preparation (Mahmood et al., 2010), even if the species never became popular among Greeks and Romans, they were aware of its medical properties (Fahey, 2005).

Most of the parts of the plant contain some remarkable properties including medicinal and pharmaceutical etc. Due to this reason, irrespective of the era and area plants have always been important to humankind since the commencement of life (Singh *et al.*, 2012). All these properties make it unique biomaterials for food and allied uses.

Different preparation from Moringa leaves, flowers, and fruits are used in the Indian subcontinent for various purposes. Due to its high nutritional value, it is a popular vital food source against PEM, which is quite common in underdeveloped and developing countries.

*M. oleifera* is a highly valued plant due to its exceptional nutritional content (Foidl *et al.*, 2001). It grows in many tropical and subtropical regions (Moyo *et al.*, 2011, Anwar *et al.*, 2007). Countries and areas such as India, the Philippines, Hawaii, and countries in Africa use the tree, mainly because it has a wide range of medicinal uses and due to the relatively high nutritional value of its leaves, fruit, flowers, and immature pods compared with other food crops (Anwar *et al.*, 2007, Asante *et al.*, 2014). Nouman *et al.* (2014); reported that moringa can be grown under harsh conditions in hot, humid, and dry tropical and subtropical regions. The tree can produce substantial nutritional quality even under marginal conditions. The great potential benefits mean that there is a need to critically assess harvesting practices that optimize biomass production of moringa under different agroecological conditions. Planting density and frequency of cutting have been identified as critical management practices that affect biomass yield and leaf quality (Gadzirayi *et al.*, 2013).

#### 2.5.4. Animal Feed (Moringa leaves and seeds)

The *M. oleifera* plant can be recommended as a protein supplement in animal feed for livestock based on recent studies conducted by goats and broiler chickens. A study was conducted at the University of Fort Hare's Honeydale farm on cross-bred Xhosa lopeared goats to determine the effect of feeding *M. oleifera* leaf meal on the physiochemical characteristics of goat meat (Moyo *et al.*, 2014). The study findings and results indicated that the cross-bred Xhosa lop-eared goats that were fed a meal supplement with *M. oleifera* leaf produced goat meat of comparable quality to cross-bred Xhosa lop-eared goats fed sunflower seed cake meal.

At the University of Limpopo, a study was conducted to determine the effects of an *M*. *oleifera* seed-supplemented diet on the productivity of Ross 308 broiler chickens. The study findings and results indicated that the *M. oleifera* seed diet improved the growth rate in Ross 308 broiler chickens aged 1 to 21 days and reduced the mortality rate as no deaths were observed over the period of the study (Molepo, 2014). Therefore, both studies provided evidence of positive effects to animals fed a Moringa leaf and seed supplemented diet.

Farmers rear livestock and mainly depend on natural grazing land because of their inability to purchase feed supplements or to grow fodder crops under intensive management systems where water and fertilizer inputs are unaffordable. However, the prospect of climate change poses a serious threat to food security and feed supply, whereby some farmers will lose their livestock owing to a shortage of fodder.

In agroforestry and specifically in cut a carry system, where high amounts of nutrients are removed from the plantation area at harvest, fertilization in a general sense seems to be the only way to maintain sustainable production (Szott and Kass, 1993). This was very evident in the results from a two-year study presented by Reyes-Sa'nchez *et al.* (2006b), where DM yield of unfertilized Moringa decreased by about 60 % during the second year of production. In spite of this, few fertilization experiments have been performed with Moringa under field conditions (DATh and Gupta, 2009; Oliveira *et al.* 2009; Pamo *et al.*, 2005).

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#### 2.6. Effect of spacing on biomass production of moringa

An experiment was conducted to study the "Standardization of Ultra high-density planting system for organic leaf production in Moringa at Tamil Nadu Agricultural University during 2015-2016. The experiment was laid out in split plot design with five main plot treatment combinations (spacing) viz., M1 - 10 x 15cm (6.66 lakh plants/ha), M2 - 15 x 15cm (4.44 lakh plants/ha), M3 -20 x 10cm (5 lakh plants/ha), M4 - 20 x 20cm (2.5 lakh plants/ha), M5 - 40 x 20cm (1.25lakh plants/ha). Spacing had a significant difference among the treatment combinations for fresh leaf yield per plot. The main plot treatment combination M5 (40 x 20cm) registered the maximum leaf yield per plot of 62.07kg followed by the treatment combination M1 (10 x 15cm) of 39.27kg per plot (Ponnuswami and Rani, 2019).

The present study effect of different planting densities on leaf yield of moringa (*M. oleifera* Lam.) was carried out during (Feb. - Dec.) 2015-16 in the vegetable field unit, Department of Horticulture, Faculty of Agriculture, Annamalai University, Annamalai Nagar. The treatment combination consisted of five different planting density viz., T1 (45 x 45 cm), T2 (60 x 60 cm), T3 (75 x 75 cm), T4 (90 x 90 cm) and T5 (120 x 120 cm). The treatment combination T5 (120 x120 cm) recorded the highest value for leaf weight, herbage yield per plant, and chlorophyll content (Ramkumar and Anuja, 2017).

A study by Zheng *et al.* (2016); showed the interactive effect between planting density and cutting height on leaf biomass yield. The highest planting density of 20 x 20 cm in combination with a cutting height of 30 cm produced the greatest total leaf dry matter yield of 8.5 tons ha<sup>1</sup>.

Basra *et al.* (2015); reported the significant interactive effect of spacing and cutting interval on biomass yield production. Planting spacing of 15 x 30 cm with a prolonged

cutting interval of 30 days gave the highest fresh matter yield of 6.40 tons ha<sup>1</sup> and dry matter yield of 1.48 tons ha<sup>1</sup> in harvest one while in harvest two the fresh biomass yield was 6.20 tons ha<sup>1</sup> and dry biomass was 1.29 tons ha<sup>1</sup>.

A study by Patricio *et al.* (2015) was conducted to determine the leaf biomass of moringa as influenced by plant density and frequency of pruning. Moringa plants were grown at four plant spacings and densities of  $1 \times 1$  m (10,000 plants ha<sup>1</sup>);  $1 \times 0.5$  m (20,000 plants ha<sup>1</sup>);  $0.9 \times 0.37$  m (30,000 plants ha<sup>1</sup>) and  $0.5 \times 0.5$  m (40,000 plants ha<sup>1</sup>. The dry biomass yield production ranged from 2.19 - 7.26 tons ha<sup>1</sup>. The highest plant population  $0.5 \times 0.5$  m (40,000 plants ha<sup>1</sup>) gave the highest total dry leaf biomass yield (Patricio *et al.*, 2015).

Effects of different plant spacing on the biomass production of *M. oleifera* were studied in Akwanga Northern Nigeria. Yield parameters such as Fresh and biomass dry matter weight (g) were evaluated under three spacing methods namely;  $0.5 \ge 0.5$ m,  $1.0 \ge 1.0$ m, and  $1.5 \ge 1.5$ m respectively. The average values of the spacing methods with the highest yield were  $1.5 \ge 1.5$ m,  $1.0 \ge 1.0$ m, and  $0.5 \ge 0.5$ m respectively.  $1.5 \ge 1.5$ m spacing exhibited the highest (25.22g),  $1.0 \ge 1.0$ m spacing showed the lowest (4.94g) mean significant effect, while  $1.5 \ge 1.5$ m spacing showed the highest (Popoola *et al.*, 2015).

Seeds bags of *M. oleifera* were transplanted into a field on the Teaching and Research Farm of Ekiti State University, Ado- Ekiti, Nigeria at 30 x 40cm (81,833 plants ha<sup>1</sup>), 40 x 60cm (41,667 plant ha<sup>1</sup>), 60 x 80cm (20, 833 plant ha<sup>1</sup>) and 100 x 100cm (10,000 plants ha<sup>1</sup>). The 30 x 40cm spacing produced the highest fodder yield (14.89 ton ha<sup>1</sup>) which was significantly higher than other spacings (Adegun *et al.*, 2013).

Newton *et al.* (2006); conducted a series of moringa cultivation experiments in Kumasi, Ghana to study the optimum planting density and harvesting interval for obtaining maximum foliage. Maximum biomass was obtained when the seeds were sown at 5x5cm plant spacing.

An experiment was performed between 19 May 2004 and 21 March 2005. The spacing treatment combinations (5 x 5 cm, 5 x 10 cm, and 5 x 15 cm) were arranged in a 3 x 2 factorial Randomized Complete Block Design (RCBD). The results showed that the fresh and dry shoot yield per hectare was significantly different (P<0.05) with the closest spacing giving the highest yields of 101.52 and 31.32 tons of fresh and dry shoots respectively. The medium spacing gave 55.84 tons of fresh shoots and 15.73 tons of dry shoots yield per hectare. The least shoot yield per hectare was from the widest spacing, which gave a 38.47 and 11.71 tons of fresh and dry shoots yield respectively (Amaglo *et al.*, 2006).

#### 2.7. Effect of plant density (plant population) on biomass production of moringa

An experiment was established as a randomized complete block design and replicated eight times. This study was conducted to evaluate the effect of planting density on biomass and nutritional composition of *M. oleifera* planted under the semi-arid conditions of the Limpopo Province. The study was conducted at Eiland (NBef Organic Farm) over two consecutive years, 2014–15 and 2015–16. The treatment combinations included planting densities of 5000, 2500, 1667, and 1250 plants ha<sup>1</sup>. The study showed that a population of 5000 plants ha<sup>1</sup> produced the highest biomass yield of more than 1.5 tons ha<sup>1</sup> (Bopape-Mabapa *et al.*, 2020).

The study on *M. oleifera* was conducted over twelve months during 2014-2015 to evaluate the impact of the growing season and varying planting densities on biomass yield and physiological at-tributes under dryland conditions. The trial was established

at densities of 5000, 2500,1667, and 1250 plants ha-1, with eight replicates. The increase in planting density led to an increase in biomass production (Mabapa *et al.*, 2018).

Completely Randomized Design which comprised three plant populations: 1,000,000 plant/ha, 250000 plants/ha and 111,111 plants/ha replicated thrice. Results showed a significant treatment combination effect on parameters evaluated at p<0.05. Fresh leaf weight and dry leaf weight were highest at 250 000 plants/ha while meaning stem girth and stem weight were highest at 111,111plants/ha. Hence, the population density of 250,000 plants/ha could be considered the optimum for sustainable growth and high leaf yield in an intensive mono-cropping system (Adekola and Abdulrahaman, 2017).

In another study where moringa was planted at a population density of 100 000 and 167 000 plants ha<sup>1</sup>. Population density of 167 000 plants ha<sup>1</sup> gave the best results in terms of biomass production as compared to a density of 10 000 plants ha<sup>1</sup>. The total dry matter yield produced was 21.2 and 11.6 tons ha<sup>1</sup> in respect of planting density (Mendieta-Araica *et al.*, 2013).

Similar findings were reported by Foidl *et al.* (2001); where the dry matter yield was 3.33, 5.05, 8.94, 13.26, 16.56, and 44.03 metric tons ha<sup>1</sup> at a plant population of 95 000, 350 000, 900 000, 1 000 000, 4 000 000 and 16 000 000 plants ha<sup>1</sup>, respectively. These results all concur with increasing biomass production as density increases.

#### 2.8. Effect of plant density on the chemical composition of moringa

A study investigated the effect of different planting spacing and cutting frequencies on biomass production and the nutritional quality of moringa. Seeds of moringa were sown on beds at plant spacing of 15 cm  $\times$  30 cm (narrow) and 15  $\times$  60 cm (broad). A significant variation in the mineral composition of moringa leaves was observed during this research. Nitrogen (6.11%), potassium (9.14%), and ascorbate (89.73  $\mu$ g g<sup>1</sup>) were recorded at broad spacing, while phosphorous (3.40%) was recorded at 20 d cutting interval, whilst maximum calcium content (2.53%) was recorded when the crop was harvested at narrow planting spacing. In conclusion, for maximum biomass production with better nutritional composition, moringa should be established as fodder purpose at narrow spacing (15 cm × 30 cm) (Basra *et al.*, 2015).

A study was conducted to investigate the effect of spacing on growth performance and nutrient quality of Moringa under the semi-arid conditions of Sokoto, Nigeria. Completely Randomized Design (CRD) was used and replicated three times with 15 x 15 cm and 20 cm x 20 cm. 20x20 cm plant spacing indicated higher concentrations of both micronutrients (Mg, Na, P, S) and macronutrients (Cr, Fe, Mn, and Sr) (Abdullahi and Maishanu, 2021).

Black Seed Moringa (BSM-L) and White Seed Moringa (WSM), the two local cultivars, and Black Seed Moringa (BSM-T) cultivar of Thailand origin of *M. oleifera* were cultivated in the fodder research field of the Bangladesh Livestock Research Institute (BLRI) during the period of 19 August 2014 to 23 December 2015. The cultivar response to biomass production performances, chemical composition, and nutritional values was analyzed in an ANOVA of a Randomized Block Design (RBD), while the differences in the rate and extent of the DM degradability in-sacco determined using three rumina cannulated bulls were analyzed in an ANOVA of 3x3 Latin Square Design. No significant difference in chemical composition (224.9, 222.4, and 223.8 gkg<sup>1</sup> DM of crude protein (CP), respectively, and 450.9, 455.3, and 435.4 gkg<sup>1</sup> DM of neutral detergent fiber, respectively) or nutritional value (47.4, 46.7 and 45.3% of

potential, and 62.8, 64.2 and 63.6% of effective degradability of dry matter) was found for the cultivars (Ahmed *et al.*, 2020).

The effect of different planting densities (100,000 and 167,000 plants ha<sup>1</sup>) chemical composition of *M. oleifera* was studied in a split-plot design with four randomized complete blocks over 2 years with eight cuts year<sup>1</sup> at the National Agrarian University farm in Managua, Nicaragua. The chemical composition of fractions showed no significant differences between planting densities (Mendieta-Araica, 2013).

This study was conducted to evaluate the effect of planting density on biomass and nutritional composition of *M. oleifera* planted under the semi-arid conditions of the Limpopo Province. The study was conducted at Eiland (NBef Organic Farm) over two consecutive years, 2014–15 and 2015–16. Data collection included total dry matter yield, leaf yield (kg ha<sup>1</sup>), and leaf nutritional composition. Planting densities did not affect moringa leaf nutrient composition. This study also revealed that moringa contains a high level of leaf nutrients even under marginal production conditions, irrespective of the planting density (Bopape-Mabapa, 2020).

An experiment was conducted to study the "Standardization of Ultra high-density planting system for organic leaf production in Moringa at Tamil Nadu Agricultural University during 2015-2016. The experiment was laid out in split plot design with five main plot treatment combinations (spacing) viz., M1 - 10 x 15cm (6.66 lakh plants/ha), M2 - 15 x 15cm (4.44 lakh plants/ha), M3 -20 x 10cm (5 lakh plants/ha), M4 - 20 x 20cm (2.5 lakh plants/ha), M5 - 40 x 20cm (1.25lakh plants/ha) and five subplot treatment combinations (organics) S1 - FYM 25t/ha, S2 - Vermicompost 12.5 t/ha, S3 - Sheep manure 25t/ha, S4-Humic acid 20 kg/ha, S5 - Control with three replications. The enhanced quality parameters also viz., Ascorbic acid, crude fiber, beta–carotene,

iron, calcium, magnesium, manganese, and zinc content were observed in the treatment combination of M5 S4 (40 x 20cm with humic acid 20kg/ha). The treatment combination of 40 x 20cm with humic acid 20kg/ha recorded better yield and quality parameters under a high-density planting system (Ponnuswami and Rani, 2019).

#### 2.9. Effect of Cutting frequencies and intervals on biomass production of moringa

A study investigated the effect of different planting spacing and cutting frequencies on biomass production and the nutritional quality of moringa. Seeds of moringa were sown on beds at plant spacing of 15 cm × 30 cm (narrow) and 15 × 60 cm (broad). The cutting frequencies for fresh biomass were (i) 15 d (ii) 20 d and (iii) 30 d. Fresh matter yield in the 1st and 2nd year was recorded as 6.40 and 7.57 t ha<sup>1</sup>, respectively when moringa crop was planted at narrow plant spacing with 30 days cutting interval followed by cutting interval of 15 and 20 d at the same planting spacing in year 1 and 2, respectively. However, the growth rate was highest when moringa plants were harvested at 15 day cutting interval. In conclusion, for maximum biomass production with better nutritional composition, moringa should be established for fodder purposes at narrow spacing (15 cm × 30 cm) with an optimum cutting interval time of 30 days (Basra *et al.*, 2015).

This study was conducted to determine the leaf biomass of moringa as influenced by plant density and frequency of pruning. Moringa plants were grown at four plant spacings and densities: a)  $1 \times 1$  m (10,000 plants ha<sup>1</sup> - pph); b)  $1 \times 0.5$  m (20,000 pph); c)  $0.9 \times 0.37$  m (30,000 pph) and d)  $0.5 \times 0.5$  m (40,000 pph), and harvested at three frequencies: 4, 6, and 8 weeks after first pruning for all treatment combinations. Results on leaf biomass indicated no increasing trend in leaf fresh biomass with increasing plant density except at 8 week harvest frequency. The highest fresh biomass of 30 t ha<sup>1</sup> was

obtained from 40,000 pph harvested at the 8 week interval. The lowest fresh leaf biomass was obtained at 10,000 pph. Data on fresh leaf biomass from plants harvested at 4 and 6 week intervals at high density (40,000 pph) (Patricio *et al.*, 2015).

Seeds bags of *M. oleifera* were transplanted into a field on the Teaching and Research Farm of Ekiti State University, Ado- Ekiti, Nigeria at 30 x 40cm (81,833 plants ha<sup>1</sup>), 40 x 60cm (41,667 plant ha<sup>1</sup>), 60 x 80cm (20, 833 plant ha<sup>1</sup>) and 100 x 100cm (10,000 plants ha<sup>1</sup>). Fodder was harvested from the top to tender stems 60 days after transplanting and fresh and dry weights were measured. Regrowth was harvested at 30-day intervals during the wet (60, 90, 120, and 150 days after transplanting) and dry (210, 240, 270, and 300 days after transplanting) seasons. Regrowth height was measured on each cutting day and harvesting involved the tender portion of the stem. The 30 x 40cm spacing produced the highest fodder yield (14.89 ton ha<sup>1</sup>) which was significantly higher than other spacings. Fodder yield in the wet season (7.6MT.ha<sup>1</sup>) was significantly higher than that in the dry season (2.79MT.ha<sup>1</sup>) (Adegun *et al.*, 2013).

Two provenances of *M. oleifera* grown in Zimbabwe, namely Malawi and Mutoko were evaluated to determine the effect of plant spacing and cutting interval on plant growth. A split-split plot experimental design was set up with provenance as the main plot factor, plant spacing the as sub-plot factor, and cutting interval sub-sub plot factor at Bindura University of Science Education's research unit. The provenances were tested at two plant spacings of 15 cm x 15 cm and 20 cm x 20 cm, and two cutting intervals of 60 days and 75 days. The 60 days cutting interval had the highest (p<0.05) plant growth performance for Mutoko provenance at each subsequent cutting to most of the growth parameters. At 75 day cutting interval, Mutoko provenance demonstrated a higher (p<0.05) mean growth of sprout height (Gadzirayi *et al.*, 2013a). In Ghana, maximum biomass production was obtained when Moringa leaves were harvested at 40 days intervals (Newton *et al.*, 2006). He also reported that Moringa plants gave higher yields when harvested at cutting intervals of 35 - 40 days.

In Nicaragua, Sanchez *et al.* (2006) reported a maximum Moringa fresh and dry biomass yield of 100,700 and 24,700 kg/ha using the cutting frequency of 75 days during the first year of the experiment.

#### **CHAPTER III**

#### **MATERIALS AND METHOD**

The experiment was conducted to evaluate the biomass production of different varieties of *M. oleifera* based on different planting densities. As well as the nutrient content of a different variety of *M. oleifera* was examined in this study. This chapter describes the materials and methods that were used in experimenting.

#### 3.1. Location of Experimental Site

This experiment was carried out at the Cattle Research Farm, Pachutia, Bangladesh Livestock Research Institute (BLRI), Savar, Dhaka-1341, Bangladesh, *M. oelifera* during the period from August 2019 to August 2020. The experimental site was located at 23°89'0" N, 90°27'9" E at an altitude of 5 m above sea level.

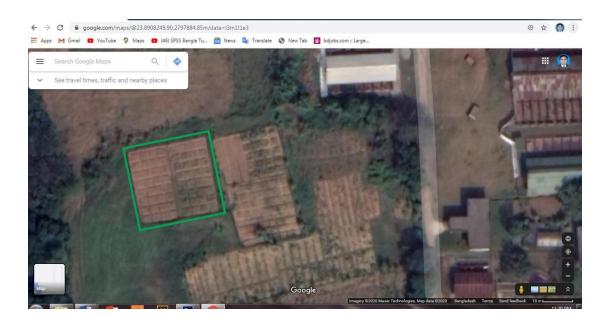


Figure 1. Google map location of the experimental site

#### **3.2. Soil Characteristics**

The experimental site was belonging to the Madhupur Tract Agro-ecological Zone (AEZ-28) of Bangladesh. The physicochemical properties of the soil were analyzed before the start of experimentation using standard procedures. Soil samples were collected from the depth of 0-15 cm of the experimental field. The clayey textured soil of the station is strongly acidic (pH 4.5-5.7) containing very little (<1.5%) organic matter. The analytical data of the soil sample collected from the experimental area were determined in the SRDI, Soil Testing Laboratory, Khamarbari, Dhaka-1215 have been presented in appendix I.

### 3.3. Agro-climate of The Experimental Site

The climate of the experimental site is subtropical, characterized by heavy rainfall during the months from April to September (Kharif season) and scanty rainfall during the rest of the year (Rabi season). The highest rainfall at the experimental site was in July 373.1 mm and the lowest January 7.7 mm during the period of the experiment. The average maximum and minimum temperatures were 33.7°C and 10.2° C respectively and humidity ranged from 37% to 74% during the experimental period. The maximum and minimum temperature, humidity, rainfall, and sunshine during the study period were collected from the Bangladesh Meteorological Department (climate division) and have been presented (Appendix II).

#### **3.4. Planting Materials used**

A total of four varieties of Moringa namely PKM1, PKM-2, Paraynal, and Black were used as planting material for this experiment. As planting material seeds of these varieties were shown for seedling raising. Seeds of PKM1, PKM-2, and Paraynal were imported from India, and Black seeds were collected from locally cultivated plants in BLRI. A total of 4120 seeds were used for germination, 1030 seeds of PKM-1, 1030 seeds of PKM-2, 1030 seeds of Paraynal, and 1030 seeds of the Black variety.  $15 \times 15$ cm polybags were used for seed seedling growth and establishment.

#### 3.5. Seedling establishment

For the seedling establishment, 1225 square feet of land was taken in the nursery shade beside the mainland. The total area was divided into four plots (according to variety) spaced at 1 foot respectively which ultimately was used as a walk.  $32 \times 32$  feet four plots were prescribed in the nursery bed.



Plate 1. Seedling establishment at nursery for transplanting in the main field

#### 3.6. Soil Preparation for seed sowing

The soil was collected from the nearby research field of BLRI, Saver, Dhaka-1341. All inert materials namely stone, bricks, crop residue, etc. were removed from the soil. Besides, that cow dung was collected from nearby cattle shade. Then cow dung and soil were mixed properly at 1:2 ratios and were used as a germination medium for seed germination of *M. oleifera*. The mixture of soil and cow dung was left for seven days before using it as a germinating media.

## 3.7. Pre-germination Treatment combination

Collected seeds were soaked in tap water for 12 hours. After that, the seeds were sown into the germination medium. Gunny bags and tissue paper were used as a germination medium. At first, one layer of gunny bag was placed, then it was soaked by water and then two layers of tissue paper were placed over it. At that time soaked seeds were placed on it and covered with two layers of tissue paper and one layer of gunny bag. Watering cane was used to give water regularly for keeping the optimum moisture level for sprouting the seeds for 3 days. After 3 days the seeds were sprouted and were ready for sowing into the polybag.

## 3.8. Growth medium for seedling establishment

The prepared soil was filled into a 15 cm  $\times$  15 cm polybag. The polybags were filled properly with soil and cow dung mixture. The pre-germinated seeds were placed into the polybag by making a hole. One seed per polybag was sown. After that, it was covered by soil and was given. Germination percentage and survival rate were measured. Seedlings were watered regularly for the first month and two days intervals for the second month. For watering the seedling, a water splash was used to prevent the decay of the seed. After observing and caring for the seeding it was planted in the main field for further study.

#### **3.9.** Preparation of experimental plots

## 3.9.1. The treatment combination of the experiment

A total of 16 treatment combination combinations were comprising four varieties marked as factorial A (V<sub>1</sub>=PKM-1, V<sub>2</sub>=PKM-2, V<sub>3</sub>=Paraynal, and V<sub>4</sub>=Black). Besides that, four spacing was used in this study as factorial B (D<sub>1</sub>=1.0×1.0 feet spacing, D<sub>2</sub>=1.0×1.5 feet spacing, D<sub>3</sub>=1.5×1.5 feet spacing, and D<sub>4</sub>=2.0×2.0 feet spacing). The treatment combinations are T<sub>1</sub>= V<sub>1</sub>D<sub>1</sub>; T<sub>2</sub>= V<sub>2</sub>D<sub>1</sub>; T<sub>3</sub>= V<sub>3</sub>D<sub>1</sub>; T<sub>4</sub>= V<sub>4</sub>D<sub>1</sub>; T<sub>5</sub>= V<sub>1</sub>D<sub>2</sub>; T<sub>6</sub>= V<sub>2</sub>D<sub>2</sub>; T<sub>7</sub>= V<sub>3</sub>D<sub>2</sub>; T<sub>8</sub>= V<sub>4</sub>D<sub>2</sub>; T<sub>9</sub>= V<sub>1</sub>D<sub>3</sub>; T<sub>10</sub>= V<sub>2</sub>D<sub>3</sub>; T<sub>11</sub>= V<sub>3</sub>D<sub>3</sub>; T<sub>12</sub>= V<sub>4</sub>D<sub>3</sub>; T<sub>13</sub>= V<sub>1</sub>D<sub>4</sub>; T<sub>14</sub>= V<sub>2</sub>D<sub>4</sub>; T<sub>15</sub>= V<sub>3</sub>D<sub>4</sub>; T<sub>16</sub>= V<sub>4</sub>D<sub>4</sub>.

Factorial A (Variety)	<b>Factorial B (Spacing)</b>
V1 (PKM-1)	$D_1 \ (1.0 \times 1.0 \ feet)$
V <sub>2</sub> (PKM-2)	$D_2 ((1.0 \times 1.5 \text{ feet})$
V <sub>3</sub> (Paraynal)	$D_3 (1.5 \times 1.5 \text{ feet})$
V <sub>4</sub> (Black)	$D_4 (2.0 \times 2.0 \text{ feet})$

Table 1. Design for the treatment combination of <i>M. oleijera</i> for experimental plo
setup

	$\mathbf{V}_1$	$\mathbf{V}_2$	$V_3$	$\mathbf{V}_4$
<b>D</b> 1	$T_1 = V_1 D_1$	$T_2 = V_2 D_1$	$T_3 = V_3 D_1$	$T_4 = V_4 D_1$
<b>D</b> 2	$T_5 = V_1 D_2$	$T_6 = V_2 D_2$	$T_7 = V_3 D_2$	$T_8 = V_4 D_2$
<b>D</b> 3	$T_9 = V_1 D_3$	$T_{10} = V_2 D_3$	$T_{11} = V_3 D_3$	$T_{12} = V_4 D_3$
<b>D</b> 4	$T_{13} = V_1 D_4$	$T_{14} = V_2 D_4$	$T_{15} = V_3 D_4$	$T_{16} = V_4 D b_4$

## Table 2. The treatment combination of *M. oleifera*

Treatment combinations	Treatment combination
$T_1 = V_1 D_1$	PKM-1 (1×1 feet)
$T_2 = V_2 D_1$	PKM-2 (1×1 feet)
$T_3 = V_3 D_1$	Paraynal (1×1 feet)
$T_4 = V_4 D_1$	Black (1×1 feet)
$T_5 = V_1 D_2$	PKM-1 (1×1.5 feet)
$T_6 = V_2 D_2$	PKM-2 (1×1.5 feet)
$T_{7}=V_{3}D_{2}$	Paraynal (1×1.5 feet)
$T_8 = V_4 D_2$	Black (1×1.5 feet)
$T_{9}=V_{1}D_{3}$	PKM-1 (1.5×1.5 feet)
$T_{10} = V_2 D_3$	PKM-2 (1.5×1.5 feet)
$T_{11} = V_3 D_3$	Paraynal (1.5×1.5 feet)
$T_{12} = V_4 D_3$	Black (1.5×1.5 feet)
$T_{13} = V_1 D_4$	PKM-1 (2×12 feet)
$T_{14} = V_2 D_4$	PKM-2 (2×2 feet)
$T_{15} = V_3 D_4$	Paraynal (2×2 feet)
$T_{16} = V_4 D_4$	Black (2×2 feet)

## **3.9.2.** Experimental layout design

A uniformly plain land area of 6006.25 square feet was selected. Selected land was plowed by a tractor for five-time to allow accurate seeding placement. The experiment was laid out in 2 factorial Randomized Complete Block Design (RCBD) with four replications. Total land was divided into four blocks; each block was again divided into sixteen experimental sub-plots. The size of each plot was  $8 \times 8$  feet and plots were accordingly to the treatment combination which was ultimately used as a drain.

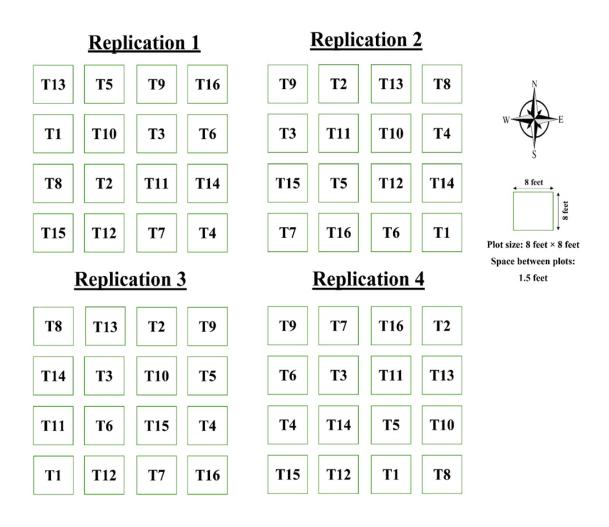


Figure 2. Field layout of the two factors experiment in the Randomized Complete Block Design (RCBD)

## 3.10. Cultivation techniques of moringa

### **3.10.1. Land preparation**

Selected land was prepared for planting the *M. oleifera* seedlings. The land was cleared manually to remove existing vegetation and soil was worked thoroughly to remove pebbles and hard objects. The land of the experimental field was first opened on 10 October 2019 with a power tiller. Then it was exposed to the sun for 24 hours before the next plowing. Thereafter, the land was deeply plowed and cross-plowed to obtain

good tilth. To get a better yield of *M. oleifera*, leveling was done to break the soil clods followed by final plowing by mowing. Manuring and fertilization were done at the time of final land preparation. After final land preparation, the experimental plot was laid out and the edge around each unit plot was raised to check run out of nutrients. A15×15 cm pits were made for planting the seedling in the main field.



Plate 2. Preparation of experimental field for transplanting of moringa saplings

## 3.10.2. Manuring and fertilization

A blanket dose of 10 t/ha cow dung, 150 kg /ha nitrogen (N), 25 kg/ha Phosphorous (P), and 30 kg/ha potash (K) was used (Haque *et al.*, 2016). All chemical fertilizers were used in three installments. A general application of Cow dung @ 262 kg/ha and half dose of nitrogen was applied during final land preparation. 1/3 dose of phosphorous and potash was applied at the time of transplantation. The rest of the nitrogen, phosphorus, and potash was used at 90 DAT and 180 DAT.

### **3.10.3.** Transplantation of the seedlings

After land preparation, the prepared seedlings were transplanted into the main field. 60 days old uniform *M. oleifera* seedlings were transplanted in the early morning into each subplot (64 square feet). The transplanting was done on 15 October 2019 to 19 October 2019. Recommended spacing was maintained at the time of transplanting. Planting dates of four varieties with recommended density were followed.

A total of 2320 seedlings of *M. oleifera* were transplanted in the main field. In different spacing, 580 seedlings of each variety were transplanted. The polybag forms the seedling was removed properly by using a sharp knife. The seedlings were placed into the pit carefully and fill the pit with extra soil. Sticking was done to support the seedlings. After planting the *M. oleifera* seedling, watering was done as necessary.

#### **3.11. Intercultural operations**

## 3.11.1. Gap filling

Gap filling was done two times, the first one in 30 DAT and the second one in 50 DAT.

#### **3.11.2.** Weeding

Weeding was done at 50 DAT, 100 DAT, 150 DAT, 200 DAT, and 250 DAT.

## 3.11.3. Irrigation

Splinker irrigation was done at 30 DAT, 60 DAT, 90 DAT, 120 DAT, and 180 DAT.

## 3.11.4. Stalking

Bamboo sticks were used for stalking at the time of transplanting of seedlings.



Plate 3. Irrigation practice at the experimental plot

## 3.11.5. Disease and pest management

Cutworms attacked in the experimental field. To control this insect 50 kg/ ha Furadan 3G was used in three installments. Mainly it was applied at the time of fertilizer application. Furadan 3G 1kg/ ha was applied at the time of final land preparation, 90 DAT, and 180 DAT. Besides that, Actara® (Thiamethoxam) 3 ml/L, Ripcord 400 EC 1 ml/L were applied at 50 DAT, 65 DAT, 120 DAT, and 135 DAT.

#### 3.12. Harvesting

Harvesting of the moringa plant was done after 180 DAT, 220 DAT, and 260 DAT when the plant height reached above 60 cm height. Above 60 cm from ground level, the stem was cut for measurement of the biomass Harvesting was done very carefully for avoiding the plants from any injury.



Plate 4. Harvesting of Moringa foliage from the experimental plot

## 3.13. Data Collection

Five plants were selected randomly and tagged in each plot for recording various morphological observations at 180 DAT, 220 DAT, and 260 DAT. The cultivated *M. oleifera* seedlings were allowed to grow for 180 days while monitoring growth and development. The parameters studied included plant height, number of leaves, and stem girth at 40 days intervals. The following observations were recorded at different stages of crop growth and the average was computed.



Plate 5. Tagging and data collection for different parameter

#### **3.14.** Growth parameters

## 3.14.1. Germination rate (%)

The germination rate of *M. oleifera* seedlings was calculated by the following formula.

Germination rate= (Total germinated seeds)/ (Total number of seeds)  $\times 100$ 

#### **3.14.2.** Plant height (cm)

Plant height was measured at 180 DAT, 220 DAT, and 260 DAT at the time of harvest. The height of the plant was determined by measuring the distance from the soil surface to the tip of the fully opened leaf on the main shoot by measuring tape and the mean plant height was expressed in centimeters.

#### 3.14.3. The number of leaves per plant

The number of leaves arising on the main stem in the five randomly selected and tagged leaf numbers was recorded at different growth stages. The mean number of leaves per plant was worked out and expressed in a number.

#### **3.14.4. Stem girth (cm)**

The diameter of the stem was measured at the top part, middle part, and bottom part then its average value was recorded.

### **3.14.5.** Survival rate of the plant (%)

The survival rate of the plant in the plot was calculated by the following formula.

Survival rate= (Total survived seeds)/ (Total number of seeds) ×100

## 3.15. Yield determination

After a post-transplantation growth period of 180 DAT, 220 DAT, and 260 DAT, branch tops with leaves were harvested at a height from the ground of 60 cm. The plants were allowed to grow after each cut and fertilized. The fresh weight of shoots harvested per plot was determined using a weighing scale in the field. Five randomly selected plants were taken from each plot after harvesting and their fresh weights were taken using an electronic beam balance.



Plate 6. Preparing of sample for weighting and chemical analysis

## 3.16. Analysis of data

All the data were subjected to analysis of variance (ANOVA) and tested for significance

by Least Significant Difference (LSD) using Statistix 10.0.

## **CHAPTER IV**

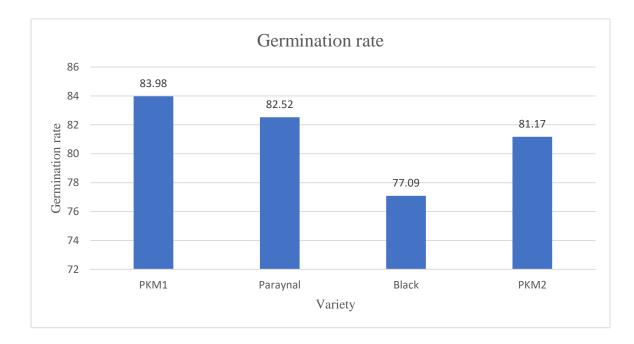
## **RESULTS AND DISCUSSION**

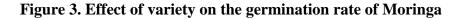
In this chapter the presentation and discussion of results from the current experiment are carried out to study and analyze the effect of variety and spacing on the growth, yield, yield attributing parameters, and chemical composition of *M. oleifera*. The results of the experiment are presented and interpreted with the following headings and subheadings.

## 4.1. Result

## 4.1.1. Effect of variety on the germination rate of moringa

The highest germination rate was found with the PKM-1 variety and it was 83.98%. Paraynal gave the second highest seed germination rate about 82.52%. The seed germination rate in PKM-2 was quite similar to Paraynal it was about 81.17%. Black seed gave the lowest germination rate it was 77.09%.





## 4.1.2. Effect of variety and spacing on the growth of Moringa oleifera

## 4.1.2.1. Plant Height

Plant height of different varieties of moringa was found significantly at different sampling dates (Table 2). The plant height was different due to regrowth after harvesting at 60 cm above the soil surface. At 180 DAT, T<sub>3</sub> exhibited the highest plant height 225.75 cm followed by T<sub>6</sub> (214.93 cm) and T<sub>2</sub> (212.92cm). Besides that, low plant height was found in T<sub>8</sub> (115.30 cm) followed by T<sub>13</sub> (119.33 cm) and T<sub>16</sub> (120.93 cm). At 220 DAT, significantly highest plant height (291.18 cm) was observed in T<sub>1</sub> followed by T<sub>7</sub> (286.13 cm) and T<sub>8</sub> (273.85cm). But low plant height was found in T<sub>13</sub> (179.75 cm). At 260 DAT, the highest plant height was observed in T<sub>7</sub> (213.43 cm) followed by T<sub>15</sub> (211.25 cm) and T<sub>1</sub>/T<sub>16</sub> (206.83 cm) treatment combinations, and plants belonging to the T<sub>13</sub> treatment combination were the shortest in height.

Plant Height (cm)				
Treatment	180 DAT	220DAT	260 DAT	
combination				
$T_1 = V_1 D_1$	210.22abcb	291.18a	206.63ab	
$T_2 = V_2 D_1$	212.92ab	273.30bc	201.85bc	
$T_3 = V_3 D_1$	225.75a	264.45cd	179.50f	
$T_4 = V_4 D_1$	142.30h	225.67hi	179.25f	
$T_5 = V_1 D_2$	198.22bcd	233.80fgh	191.30de	
$T_6 = V_2 D_2$	214.93ab	251.38de	190.67de	
$T_7 = V_3 D_2$	181.33de	286.13ab	213.43a	
$T_8 = V_4 D_2$	115.30i	273.85bc	180.80f	
$T_9 = V_1 D_3$	160.95fg	247.55ef	184.60ef	
$T_{10} = V_2 D_3$	192.00cd	241.77efg	195.10cd	
$T_{11} = V_3 D_3$	173.70ef	207.55j	186.38ef	
$T_{12} = V_4 D_3$	131.07hi	241.55efg	201.13bc	
$T_{13} = V_1 D_4$	119.33i	179.75k	169.00g	
$T_{14} = V_2 D_4$	146.63gh	231.75gh	191.55de	
$T_{15} = V_3 D_4$	161.43fg	212.80ij	211.25a	
$T_{16} = V_4 D_4$	120.93i	253.50de	206.83ab	
LSD	18.28	15.45	7.69	
CV%	4.22	2.46	1.56	
Significance	**	**	**	
Level				

Table 3. Effect of variety and spacing on plant height (cm) of M. oleifera at different measurement dates

 $T_1 = PKM-1$  (1×1 feet);  $T_2 = PKM-2$  (1×1 feet);  $T_3 = Paraynal$  (1×1 feet);  $T_4 = Black$  (1×1 feet);  $T_5 = PKM-1$  (1×1.5 feet);  $T_6 = PKM-2$  (1×1.5 feet);  $T_7 = Paraynal$  (1×1.5 feet);  $T_8 = Black$  (1×1.5 feet);  $T_{9}$ = PKM-1 (1.5×1.5 feet);  $T_{10}$ = PKM-2 (1.5×1.5 feet);  $T_{11}$ = Paraynal (1.5×1.5 feet);  $T_{12}$ = Black (1.5×1.5 feet);  $T_{13}$ = PKM-1 (2×12 feet);  $T_{14}$ = PKM-2 (2×2 feet);  $T_{15}$ = Paraynal (2×2 feet);  $T_{16}$ = Black (2×2 feet). Different alphabetical letters within the same column indicate significant differences among various treatment combinations according to a least significant difference test (LSD) (P < 0.01). DAT means day after transplanting.

\*\* Significant at P < 0.01

## 4.1.2.2. Number of leaves

Number of Leaves					
Treatment	180 DAT 220DAT 260 DAT				
combination					
$T_1 = V_1 D_1$	18.55abc	38.35ab	12.5g		
$T_2 = V_2 D_1$	21.2abc	22c	13.4fg		
$T_3 = V_3 D_1$	21.8ab	40.7a	13.25fg		
$T_4 \!\!= V_4 D_1$	11.9def	26.6abc	25.75cde		
$T_5 = V_1 D_2$	17.2abcd	23.5bc	31.25bcd		
$T_6 = V_2 D_2$	21.45abc 21.6c 37.6		37.65b		
$T_{7} = V_{3}D_{2}$	22a 41.15a 45.		45.95a		
$T_8 = V_4 D_2$	7.1f	7.1f 22c 24.0			
$T_9 = V_1 D_3$	19.62abc	9.62abc 23.15bc 21.			
$T_{10} = V_2 D_3$	18abcd	bcd 19.3c 22.1			
$T_{11} = V_3 D_3$	16.35abcde	35abcde 12.2c 25.15c			
$T_{12} = V_4 D_3$	8.07ef 20.4c 37.		37.05b		
$T_{13} = V_1 D_4$	15.85bcde	15.2c	12.55g		
$T_{14} = V_2 D_4$	15.65cde	15.3c	31.8bcd		
$T_{15} = V_3 D_4$	17.4abcd	38ab	32.8bc		
$T_{16} = V_4 D_4$	10.3ef	18.8c	22.05e		
LSD	6.11	15.44	8.1		
CV%	14.54	24.22	12.39		
Significance level	*	** **			

Table 4. Effect of variety and spacing on number of leaves of moringa at different measurement dates

 $T_1$ = PKM-1 (1×1 feet);  $T_2$ = PKM-2 (1×1 feet);  $T_3$ = Paraynal (1×1 feet);  $T_4$ = Black (1×1 feet);  $T_5$ = PKM-1 (1×1.5 feet);  $T_6$ = PKM-2 (1×1.5 feet);  $T_7$ = Paraynal (1×1.5 feet);  $T_8$ = Black (1×1.5 feet);  $T_9$ = PKM-1 (1.5×1.5 feet);  $T_{10}$ = PKM-2 (1.5×1.5 feet);  $T_{11}$ = Paraynal (1.5×1.5 feet);  $T_{12}$ = Black (1.5×1.5 feet);  $T_{13}$ = PKM-1 (2×12 feet);  $T_{14}$ = PKM-2 (2×2 feet);  $T_{15}$ = Paraynal (2×2 feet);  $T_{16}$ = Black (2×2 feet). Different alphabetical letters within the same column indicate significant differences among various treatment combinations according to a least significant difference test (LSD) (P < 0.01). DAT means the day after transplanting.

\*\* Significant at P < 0.01

\* Significant at P < 0.05

The number of leaves per plant exhibited different results under different treatment combinations (Table 3). The highest number of leaves per plant at harvest was found where different varieties were grown under different plant spacing.

At the early stage of growth is 180 DAT, the  $T_7$  treatment combination resulted in the highest number (22.00) of leaves compared to other combinations of treatment combinations followed by  $T_3$ ,  $T_6$ , and others. During the growth stage (180 DAT), a black variety of moringa grew at low spacing  $T_8$  showed the least no. of leaves.

At the  $2^{nd}$  harvest (220 DAT), under the T<sub>7</sub> treatment combination, the number of leaves per plant was 41.15 which was the highest, and the lowest no. of leaves was found for the T<sub>11</sub> treatment combination which was 12.2 where moringa was grown at the maximum plant-to-plant distance.

At 260 DAT, significantly highest leaves per plant (45.95) were observed in  $T_7$  followed by  $T_6$  (37.65) and  $T_{12}$  (37.05). But low leaves per plant were found in  $T_1$  (12.5).

## 4.1.2.3. Stem girth

At the early stage of growth (180 DAT), the stem girth was found maximum (4.925 cm) in the T<sub>3</sub> treatment combination and lowest (2.45 cm) in T<sub>8</sub>. At 220 DAT of the growth stage of moringa, the stem girth was observed the highest in the T<sub>1</sub> treatment combination (7.8 cm) while the lowest was recorded in the T<sub>13</sub> treatment combination (2.82 cm). During the harvesting period of moringa (260 DAT), stem girth was recorded as the highest in the T<sub>7</sub> treatment combination (6.92 cm) and the lowest in was T<sub>8</sub> treatment combination (1.75 cm).

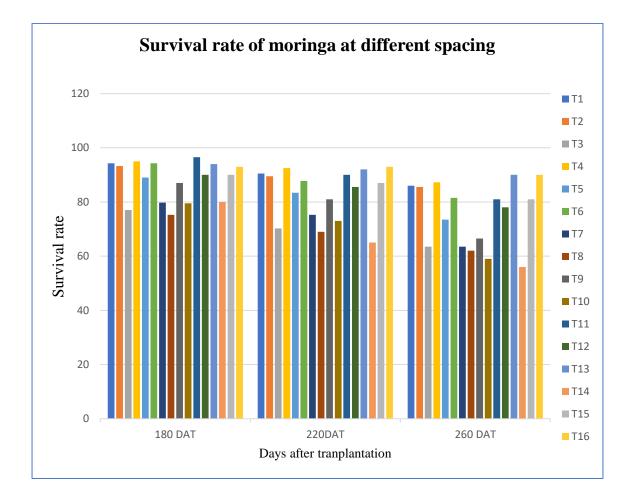
Stem Girth (cm)				
Treatment	180 DAT 220DAT 260 DAT			
combination				
$T_1 = V_1 D_1$	4.37ab	7.8a	6.5a	
$T_2 = V_2 D_1$	4.55ab	6.7bc	4.37c	
$T_3 = V_3 D_1$	4.92a	5.7de	3.67de	
$T_4 = V_4 D_1$	3.25de	5.47ef	4.15cd	
$T_5 = V_1 D_2$	4.45ab	4.1h	4.07cde	
$T_6 = V_2 D_2$	4.82a	6.6bc	3.57e	
$T_7 = V_3 D_2$	4.02bc	6.9b	6.92a	
$T_8 = V_4 D_2$	2.45f	5.5ef	1.75f	
$T_9 = V_1 D_3$	3.55cd	4.02h	1.82f	
$T_{10} = V_2 D_3$	3.72cd	6.27cd	1.82f	
$T_{11} = V_3 D_3$	3.47cd	4.82g	3.57e	
$T_{12} = V_4 D_3$	2.5f	5.02fg	4.25c	
$T_{13} = V_1 D_4$	2.62f	2.82i	3.6e	
$T_{14} = V_2 D_4$	3.45cde	5.42efg	3.87cde	
$T_{15} = V_3 D_4$	3.5cd	5.77de	6.5b	
$T_{16} = V_4 D_4$	2.85ef	6.52bc	4.15cd	
LSD	0.62	0.61	0.52	
CV%	6.62	4.32	5.19	
Significance level	**	**	**	

Table 5. Effect of variety and spacing on stem girth of moringa at different measurement dates

 $\begin{array}{l} T_{1}=\text{PKM-1} \ (1\times 1 \ \text{feet}); \ T_{2}=\text{PKM-2} \ (1\times 1 \ \text{feet}); \ T_{3}=\text{Paraynal} \ (1\times 1 \ \text{feet}); \ T_{4}=\text{Black} \ (1\times 1 \ \text{feet}); \\ T_{5}=\text{PKM-1} \ (1\times 1.5 \ \text{feet}); \ T_{6}=\text{PKM-2} \ (1\times 1.5 \ \text{feet}); \ T_{7}=\text{Paraynal} \ (1\times 1.5 \ \text{feet}); \ T_{8}=\text{Black} \ (1\times 1.5 \ \text{feet}); \\ T_{9}=\text{PKM-1} \ (1.5\times 1.5 \ \text{feet}); \ T_{10}=\text{PKM-2} \ (1.5\times 1.5 \ \text{feet}); \ T_{11}=\text{Paraynal} \ (1.5\times 1.5 \ \text{feet}); \ T_{12}=\text{Black} \ (1\times 1.5 \ \text{feet}); \ T_{13}=\text{PKM-1} \ (2\times 12 \ \text{feet}); \ T_{14}=\text{PKM-2} \ (2\times 2 \ \text{feet}); \ T_{15}=\text{Paraynal} \ (2\times 2 \ \text{feet}); \ T_{16}=\text{Black} \ (2\times 2 \ \text{feet}). \\ \text{Different alphabetical letters within the same column indicate} \\ \text{significant differences among various treatment combinations according to a least significant} \\ \text{difference test} \ (LSD) \ (P < 0.01). \ DAT \ means \ day \ after \ transplanting. \\ ** \ \text{Significant at } P < 0.01 \end{array}$ 

# 4.1.2.4. Effect of variety and spacing on the survival rate of moringa the at different cutting intervals

Survival rates exhibited different results under different treatment combinations (Figure 1). The highest result at survival rate was found where different varieties were grown under different plant spacing.



 $\begin{array}{l} T_1 = PKM-1 \ (1 \times 1 \ feet); \ T_2 = PKM-2 \ (1 \times 1 \ feet); \ T_3 = Paraynal \ (1 \times 1 \ feet); \ T_4 = Black \ (1 \times 1 \ feet); \\ T_5 = PKM-1 \ (1 \times 1.5 \ feet); \ T_6 = PKM-2 \ (1 \times 1.5 \ feet); \ T_7 = Paraynal \ (1 \times 1.5 \ feet); \ T_8 = Black \ (1 \times 1.5 \ feet); \\ T_9 = PKM-1 \ (1.5 \times 1.5 \ feet); \ T_{10} = PKM-2 \ (1.5 \times 1.5 \ feet); \ T_{11} = Paraynal \ (1.5 \times 1.5 \ feet); \ T_{12} = Black \ (1.5 \times 1.5 \ feet); \ T_{13} = PKM-1 \ (2 \times 12 \ feet); \ T_{14} = PKM-2 \ (2 \times 2 \ feet); \ T_{15} = Paraynal \ (2 \times 2 \ feet); \ T_{16} = Black \ (2 \times 2 \ feet). \end{array}$ 

Figure 4. Effect of variety and spacing on the survival rate of Moringa different measurement dates

At the early stage of growth (180 DAT), the survival rate was found maximum (96.5 %) in the  $T_{11}$  treatment combination and lowest (75.25%) in  $T_8$ . At the second harvesting (220 DAT) of the growth stage of moringa, the survival rate was observed the highest in  $T_{16}$  treatment combination (93%) while the lowest was recorded in the  $T_{14}$  treatment combination (65%). During the harvesting period of moringa (260 DAT), the survival rate was recorded as the highest in the  $T_{13}$  treatment combination (90%) and the lowest in was  $T_{14}$  treatment combination (56%).

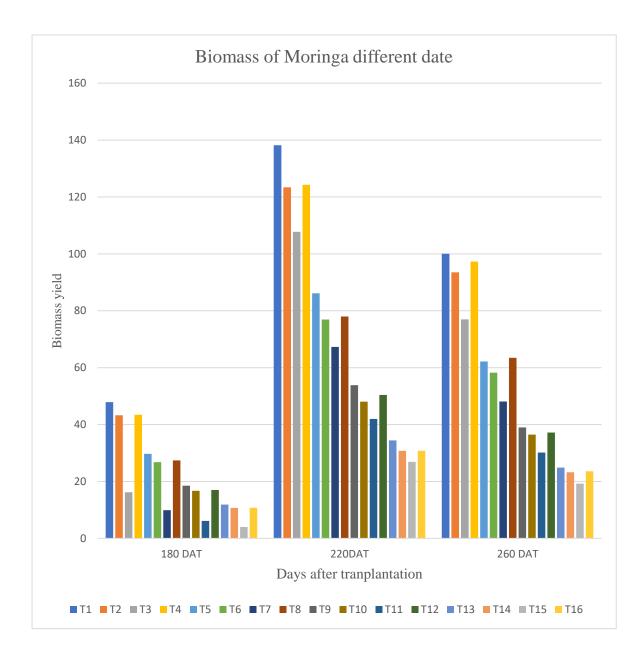
# **4.1.3.** Effect of variety and spacing on yield of moringa at the different cutting intervals

#### 4.1.3.1. Biomass

At 180 DAT,  $T_1$  exhibited the highest biomass yield 47.88 t/ha followed by  $T_4$ . Besides that, low biomass yield was found in  $T_{15}$  at 3.98 t/ha.

At 220 DAT, significantly the highest biomass yield (138.16 t/ha) was observed in  $T_1$  followed by  $T_4$ . But low plant biomass yield in  $T_{15}$  (26.88 t/ha).

At harvest (260DAT), the highest biomass yield was found in  $T_1$  (100 t/ha) followed by  $T_4$ , and the lowest plant biomass yield in the  $T_{15}$  treatment combination.



 $\begin{array}{l} T_1 = PKM-1 \ (1\times 1 \ feet); \ T_2 = PKM-2 \ (1\times 1 \ feet); \ T_3 = Paraynal \ (1\times 1 \ feet); \ T_4 = Black \ (1\times 1 \ feet); \\ T_5 = PKM-1 \ (1\times 1.5 \ feet); \ T_6 = PKM-2 \ (1\times 1.5 \ feet); \ T_7 = Paraynal \ (1\times 1.5 \ feet); \ T_8 = Black \ (1\times 1.5 \ feet); \\ T_9 = PKM-1 \ (1.5\times 1.5 \ feet); \ T_{10} = PKM-2 \ (1.5\times 1.5 \ feet); \ T_{11} = Paraynal \ (1.5\times 1.5 \ feet); \ T_{12} = Black \ (1\times 1.5 \ feet); \ T_{13} = PKM-1 \ (2\times 12 \ feet); \ T_{14} = PKM-2 \ (2\times 2 \ feet); \ T_{15} = Paraynal \ (2\times 2 \ feet); \ T_{16} = Black \ (2\times 2 \ feet). \end{array}$ 

Figure 5. Effect of variety and spacing on Biomass of Moringa different measurement dates

# **4.1.4.** Effect of variety and spacing on the chemical composition of moringa at different cutting intervals

## 4.1.4.1. Dry matter

At the early stage of growth is 180 DAT,  $T_{14}$  treatment combination resulted in the highest number (23.19%) of leaves compared to other combination of treatment combinations followed by  $T_3$ ,  $T_{15}$ , and others. During the growth stage (180 DAT),  $T_9$  showed the least dry matter.

At the 2nd harvest (220 DAT), under  $T_4$  treatment combination, the dry matter per plant was 23.73% which was the highest and the lowest dry matter was found for  $T_{10}$ treatment combination which was 19.61 % where moring was grown at 1.5 feet plant to plant distance.

At 260 DAT, significantly highest dry matter per plant (24.21%) was observed in  $T_1$  followed by  $T_4$  (24.03%) and  $T_{14}$  (23.97%). But low dry matter per plant was found in  $T_9$  (21.003%).

Dry Mater (%)				
Treatment	180 DAT 220DAT 260 DAT		260 DAT	
combination				
$T_1 = V_1 D_1$	22.14bc	22.14bc 23.37ab 24.2		
$T_2 = V_2 D_1$	21.96abcd	21.88bcde	22.82cdefg	
$T_3 = V_3 D_1$	23.09ab	21.45cde	22.45defg	
$T_4\!\!=V_4D_1$	22.37abc	23.73a	24.03ab	
$T_5 = V_1 D_2$	19.93efg	21.56cde	22.98bcdefg	
$T_6 = V_2 D_2$	22.16abc	22.84abc	23.09abcdef	
$T_7 = V_3 D_2$	20.69def	59def 20.94def 21.92fg		
$T_8 \!\!= V_4 D_2$	21.74bcd	22.4abcd 22.85cdef		
$T_{9} = V_{1}D_{3}$	19.20g	19.74f	21h	
$T_{10} = V_2 D_3$	19.56fg	19.61f	21.17h	
$T_{11} = V_3 D_3$	19.57fg	21.59cde 21.82gh		
$T_{12} = V_4 D_3$	21.16cde	21.03def	22.02efgh	
$T_{13} = V_1 D_4$	22.43abc	22.25abcd	23.12abcde	
$T_{14} = V_2 D_4$	23.19a	23.43a	23.97abc	
$T_{15} = V_3 D_4$	22.77ab	20.74ef	21.17h	
$T_{16} = V_4 D_4$	22.69ab	22.37abcd	23.37abcd	
LSD	1.42	1.5	1.17	
CV%	2.58	2.69	2.03	
Significance	**	**	**	
Level				

Table 6. Effect of variety and spacing on dry mater of moringa at different measurement dates

 $\begin{array}{l} \hline T_1 = PKM-1 \ (1 \times 1 \ feet); \ T_2 = PKM-2 \ (1 \times 1 \ feet); \ T_3 = Paraynal \ (1 \times 1 \ feet); \ T_4 = Black \ (1 \times 1 \ feet); \\ \hline T_5 = PKM-1 \ (1 \times 1.5 \ feet); \ T_6 = PKM-2 \ (1 \times 1.5 \ feet); \ T_7 = Paraynal \ (1 \times 1.5 \ feet); \ T_8 = Black \ (1 \times 1.5 \ feet); \\ \hline T_9 = PKM-1 \ (1.5 \times 1.5 \ feet); \ T_{10} = PKM-2 \ (1.5 \times 1.5 \ feet); \ T_{11} = Paraynal \ (1.5 \times 1.5 \ feet); \ T_{12} = Black \ (1.5 \times 1.5 \ feet); \ T_{13} = PKM-1 \ (2 \times 12 \ feet); \ T_{14} = PKM-2 \ (2 \times 2 \ feet); \ T_{15} = Paraynal \ (2 \times 2 \ feet); \ T_{16} = Black \ (2 \times 2 \ feet). \\ \hline Different \ alphabetical \ letters \ within \ the \ same \ column \ indicate \ significant \ difference \ test \ (LSD) \ (P < 0.01). \\ \hline Different \ alphabetical \ letters \ within \ the \ same \ column \ indicate \ significant \ difference \ test \ (LSD) \ (P < 0.01). \\ \hline DAT \ means \ day \ after \ transplanting. \\ \hline \end{array}$ 

\*\* Significant at P < 0.01

#### 4.1.4.2. Crude Protein

The chemical component of moringa was found significantly different due to different plant-to-plant spacing among different varieties at different sampling dates (Table 6). Crude protein of moringa exhibited different results in terms of different treatment combinations (Table 6) according to AOAC. 1990 Crude protein was observed highest (19.59%) on  $T_{15}$  treatment combination, where  $T_8$  treatment combination (14.6%) comprises low crude protein at 180 DAT. At second harvesting (220 DAT) crude protein was observed highest (19.52%) on  $T_{11}$  treatment combination, where  $T_{14}$  treatment combination (15.62%) comprises the lowest. The plants under  $T_{11}$  treatment combination were found maximum in crude protein (19.86) and  $T_{14}$  results (16.07) minimum crude protein at 260 DAT.

Crude Protein (%)				
Treatment	180 DAT 220DAT 260 DAT			
combination				
$T_1 = V_1 D_1$	18.95abc	16.72de	17.16def	
$T_2 = V_2 D_1$	19.33ab	19.35a	19.18a	
$T_3 = V_3 D_1$	19.3ab	17.1cd	17.34bcdef	
$T_4 \!\!= V_4 D_1$	19.42ab	16.7de	16.97f	
$T_5 = V_1 D_2$	18.45c	16.66de	16.82f	
$T_6 = V_2 D_2$	19.45ab	17.3cd	17.12ef	
$T_7 = V_3 D_2$	17.42d	17.09cd 17.21cde		
$T_8 = V_4 D_2$	14.6f	17.81bc 17.84bc		
$T_9 = V_1 D_3$	17.62d	16.11ef	16.79f	
$T_{10} = V_2 D_3$	16.32e	17.77c	17.76bcde	
$T_{11} = V_3 D_3$	17.5d	19.52a 19.86a		
$T_{12} = V_4 D_3$	18.88bc	18.7ab 19.2a		
$T_{13} = V_1 D_4$	17.29d	17.21cd 17.9b		
$T_{14} = V_2 D_4$	16.57e	15.62f	16.07g	
$T_{15} = V_3 D_4$	19.59a	17.05cd	17.89bc	
$T_{16} = V_4 D_4$	19.42ab	17.54cd	17.91bc	
LSD	0.66	0.91	0.71	
CV%	1.43	2.05	1.57	
Significance	**	**	**	
Level				

 Table 7. Effect of variety and spacing on Crude Protein of moringa at different

 measurement dates

 $T_{1}=PKM-1 (1\times1 \text{ feet}); T_{2}=PKM-2 (1\times1 \text{ feet}); T_{3}=Paraynal (1\times1 \text{ feet}); T_{4}=Black (1\times1 \text{ feet}); T_{5}=PKM-1 (1\times1.5 \text{ feet}); T_{6}=PKM-2 (1\times1.5 \text{ feet}); T_{7}=Paraynal (1\times1.5 \text{ feet}); T_{8}=Black (1\times1.5 \text{ feet}); T_{9}=PKM-1 (1.5\times1.5 \text{ feet}); T_{10}=PKM-2 (1.5\times1.5 \text{ feet}); T_{11}=Paraynal (1.5\times1.5 \text{ feet}); T_{12}=Black (1.5\times1.5 \text{ feet}); T_{13}=PKM-1 (2\times12 \text{ feet}); T_{14}=PKM-2 (2\times2 \text{ feet}); T_{15}=Paraynal (2\times2 \text{ feet}); T_{16}=Black (2\times2 \text{ feet}). Different alphabetical letters within the same column indicate significant differences among various treatment combinations according to a least significant difference test (LSD) (P < 0.01). Different alphabetical letters within the same column indicate significant difference test (LSD) (P < 0.01). DAT means day after transplanting. ** means 1% level of significance.$ 

# 4.1.5. Correlation between different planting spacing and Various growths, yield parameters, and chemical analysis of Moringa

When data were plotted in the analysis including different cutting intervals, negative relationships were found in the most case between planting distance and Various growths, yield parameters, and dry mater analysis of Moringa. A weak negative significant correlation was found between planting space and biomass yield of moringa at 180 DAT (r= .285\*). But at 220 DAT (r= .355\*\*) and 260 DAT (r= .497\*\*) a moderate positive significant correlation was found between planting space and biomass yield was found. On other hand, a negative weak relationship was found between plant spacing and crude protein and dry matter in most of the cases.

Relationship between	Diffe	Different cutting interval		
	180 DAT	220 DAT	260 DAT	
	Different planting distances (feet) and			
Plant height (cm)	605**	.373**	1	
No. of leaves/plant	.193	100	.022	
Stem girth (cm)	197	.234	.263*	
Survival rate	030	115	248*	
Biomass	285*	.355**	.497**	
Dry matter	271*	069	.121	
Crude protein	.058	088	145	

 Table 8. Correlation between different planting spacing and Various growths,

 yield parameters, and chemical analysis of Moringa

\*\* Correlation is significant at the 0.01 level (2-tailed)

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

#### 4.2. Discussion

PKM-1 variety of moringa was found highest germination rate at 83.98% followed by Perennial at 82.52%, PKM-2 at 81.17%, and black at 77.09%. Genetic and environmental factors are responsible for seed germination. Adequate moisture content with proper temperature and light is a big factor in seed germination.

In 1st and 3rd harvest Paraynal variety in highest planting height compare to other varieties. This might have occurred temperature and rainfall effect on variety. From the treatment combination,  $T_7$  was more potential than others in the highest plant height. On the other hand, low planting Black variety was found with low plant height with higher spacing.  $T_{13}$  treatment combination gives lower plant height. This might be because water with soluble nutrients provided the efficient growth condition of moringa as irrigation was done intensively (White and Short 1987). Plant height is influenced by genetic as well as environmental conditions. The increase in plant height also could be due to better availability of soil nutrients in the growing areas, especially nitrogen and phosphorus which have enhancing effect on the vegetative growth of plants by increasing cell division and elongation and the varietal variability to absorb the nutrients from the soil (El-Tohamy et al., 2006). Another reason for increased plant height at different levels might be due to the availability of nutrients that came from manuring which helped in the vegetative growth of stem amaranth. This result is similar to the results of Sharma et al. (1999).

The number of leaves under the T<sub>7</sub> treatment combination was found highest among all treatment combinations on different harvesting dates. The results indicated that the availability of water in open field conditions and well-plowed soil increased the number of leaves per plant. Water soluble nutrients and nitrogen provided by cow dung

enhanced the growth and development of moringa at different levels. As higher doses of nitrogen in cow dung increased the length of the moringa plant increased the number of leaves per plant. Research results obtained by Devi et al. (2003) are relevant to this character. So, there was no competition for the light which enhanced the number of leaves per plant. Decreased light can become a limiting factor to plant growth when shading occurs; one major effect of shade is to slow the rate of photosynthesis relative to respiration (Harper, 1977).

Paranal variety with lower plant spacing was found to maximum stem girth. Stem girth was recorded differently for different treatment combinations which might be due to the accumulation of nutrients which enhanced the overall plant growth of moringa as increased plant diameter. Nitrogen has been reported to be a major nutrient that increased basal area per plant, and the size of stems and leaves (Vallentine 1980). Our results are following the findings of Sundstrom (1984), who reported that increased available nutrients increased the stem diameter. The results also showed that the manure cow dung progressively and significantly increased the cell number and/or size in stem amaranth. Devi (2003) also reported the same results.

The black moringa variety with higher plant spacing was found highest survival rate followed by the Paraynal variety with the low plant-to-plant spacing. Black was a local variety so it was easily adopting the local environment. Edward et al. (2013) also found a similar result for moringa survival rate. The increase in survival rate also could be due to the better availability of soil nutrients in the growing areas (El-Tohamy et al., 2006). Bedside that genetic character of variety with environmental factors also responsible for plant survival rate.  $T_1$  exhibited the highest biomass weight compared to other treatment combinations. This is might be due to the availability of water and soil nutrients accumulated by plants. Available soluble nutrients enhanced the overall growth and increased the shoot length, and weight of stem and root. These observations were revealed in previous research (Wight and Black, 1979; Power, 1983; Rauzi and Fairbourn, 1983, Patricio et al., 2015,).

The maximum biomass weight of plants was possibly too long time photosynthesis which lead to more deposition of photosynthates during the vegetative growth of plants. Results obtained for biomass weight in this experiment are also comparable to the results of Talukder (1999), Adegun et al., 2013.

In most cases,  $T_4$  gives optimum dry matter. A similar result was found by Ahmed et al., (2020). The results indicated that the accumulation of nutrients increases the dry matter content of shoot and root. Diaz-ortega et al. (2004) observed that with the increasing of nutrient level (mainly Nitrogen) level biomass production of moringa increased significantly which increases the dry matter of stem and root.

Treatment combination  $T_{11}$  (Paraynal moringa variety at low 1.5\* 1.5 feet spacing) was found highest crude protein among all the treatment combinations. Ponnuswami and Rani (2019) also found that high-density plant populations give similar results.

## **CHAPTER V**

## SUMMARY AND CONCLUSION

The experiment was conducted at the Cattle Research Farm, Pachutia, Bangladesh Livestock Research Institute (BLRI), Savar, Dhaka-1341, Bangladesh, with different types of available variety of *M. oelifera* during the period from August 2019 to August 2020 to evaluate the higher foliage yield of moringa as a fodder. The results considering the effect of plant spacing on growth, biomass yield, yield attributing characters, and chemical composition of Moringa were interpreted in this experiment. The experiment consisted of 16 treatment combinations viz.,  $T_1 = PKM-1$  (1×1 feet);  $T_2 = PKM-2$  (1×1 feet); T<sub>3</sub>= Paraynal (1×1 feet); T<sub>4</sub>= Black (1×1 feet); T<sub>5</sub>= PKM-1 (1×1.5 feet); T<sub>6</sub>= PKM-2 (1×1.5 feet);  $T_7$ = Paraynal (1×1.5 feet);  $T_8$ = Black (1×1.5 feet);  $T_9$ = PKM-1  $(1.5 \times 1.5 \text{ feet}); T_{10} = PKM-2 (1.5 \times 1.5 \text{ feet}); T_{11} = Paraynal (1.5 \times 1.5 \text{ feet}); T_{12} = Black$  $(1.5 \times 1.5 \text{ feet}); T_{13} = PKM-1$  (2×12 feet);  $T_{14} = PKM-2$  (2×2 feet);  $T_{15} = Paraynal$  (2×2 feet);  $T_{16}$ = Black (2×2 feet). The experiment was laid out in two factorial Randomized Complete Block Design (RCBD) comprising four replications. The seeds of *M. oelifera* were sown in the nursery in a polybag on 25 August 2019 and then transplanted at the main field with 60 days old Moringa seedlings on 25 October 2019 which were harvested on 25 April 2020. Moringa plants were randomly selected from the respective plots. A total of 320 (5 from each replication) plants of moringa were selected from each plot for data collection. Data were collected on plant height (cm), the number of leaves per plant, stem girth (cm), biomass weight (g), dry weight (g), and crude protein (%). The collected data were analyzed statistically and the differences between the means were evaluated by the R Core test.

Germination rate was found high in the PKM-1 variety. But There was no significant difference between other varieties. In the case of the growth rate of moringa in different spacing give different results in different parameters.

The highest plant height was found in the Paraynal variety with low spacing.  $T_7$  gave the highest plant height among all the treatment combinations. Besides that, low plant height was found in the Black moringa variety with higher spacing.

The maximum number of leaves and stem girth was found in the Paraynal variety with less plant-to-plant distance. T<sub>7</sub> treatment combination gave the maximum number of leaves at the third harvest and it was about 47.

The survival rate is fully opposite to another parameter. The black variety of moringa was able to tolerate more adverse conditions and give a maximum survival rate than another variety.

Biomass yield is very important for moringa as fodder. The highest biomass yield was found in the  $T_1$  treatment combination compared to others. PKM-1 variety gives more potential yield with low spacing. In the case of crude protein and dry matter, the highest crude protein was found in Paraynal with optimum spacing but dry mater in black with the least spacing.

## CONCLUSION

The results from these experiments showed that the effect of population density on parameters evaluated was significant (p<0.05).  $T_1$  exhibited the highest performance in respect of growth and yield of moringa followed by  $T_4$ . Again, there was a negative correlation with spacing in respect of biomass yield and dry weight. It was found that for fodder and leaf production  $T_1$  is suitable for large-scale cultivation. But it was imported from India and was very costly and difficult for the farmer.  $T_4$  is easily available and the mortality rate is low. Moringa can be harvested at a height of 60 cm above ground level, which facilitates mechanical harvesting, and while the stem is still pliable. The crop can supply livestock with a satisfactory amount of crude protein which makes it a potentially valuable source of feed supplement at times when there is not enough natural fodder due to drought.

## RECOMMENDATION

- PKM-1 variety of moringa with 1×1 feet spacing give high yield individually but on an average Black variety of moringa in 1.0 × 1.0 feet should be recommended. Besides that, PKM-1 seed need to export from India and it was very costly but Black is locally produced and easily available. Considering Bangladeshi environment condition and soil and the above data of the experiment following recommendation should be taken.
- Irrigation is a big factor in moringa production manage proper irrigation for better yield.
- Avoid harvesting from December to March, in this period growth of the moringa plant is very slow.
- Be careful at the time of transplanting. Injury on the plantation is responsible for a high mortality rate.
- This experiment should be conducted in a different region of Bangladesh for a more correct result.

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## **APPENDICES**

**Appendix I.** Monthly record of year temperature, rainfall, relative humidity, soil temperature, and sunshine of the experimental site during the period from August 2019 to August 2020

Year	Months	Observations					
		*Air temperature (°c) Relative Rainfal		Rainfal	**Sunshin		
		Minimum temperatur e (°C)	Maximum temperatur e (°C)	Average temperatur e (°C)	humidit y (%)	1 (mm)	e (h/day)
	August	33.6	26.3	29.95	73	204.5	6.2
	Septem ber	32.8	25.9	29.35	71	102.4	6.5
2019	October	31.2	23.8	27.5	65	67.7	7.1
	Novemb er	29.6	12.2	20.9	53	34.4	8.0
	Decemb er	26.2	11.1	18.65	50	12.8	8.2
	January	25.4	10.2	17.8	46	7.7	6.2
	Februar y	28.1	14.9	21.5	37	28.9	7.4
	March	32.5	18	25.25	38	65.8	7
2020	April	33.7	19.1	26.4	42	156.3	6.5
	May	32.1	24.8	28.45	59	339.4	5.4
	June	33.4	27.0	30.2	72	340.4	5
	July	31.6	27.9	29.75	72	373.1	4
	August	31.6	27.6	29.6	74	316.5	5

\*Monthly average

\*\* Monthly total

Source: Bangladesh Meteorological Department (climate division), Agargoan, Dhaka-1207

Appendix II. Characteristics of Cattle Research Farm soil are analyzed by Soil Resource and Development Institute (SRDI), Khamar Bari, Farmgate, Dhaka. A. morphological characteristics of the experimental field

Morphological features	Characteristics
Location	Cattle Research Farm, Pachutia,
	Bangladesh Livestock Research Institute
	(BLRI), Savar, Dhaka-1341
AEZ	Madhupur Tract (28)
General Soil Type	The shallow red brown terrace soil
Land type	High land
Soil series	Tejgaon
Topography	Fairly leveled
Flood level	Above flood level
Drainage	Well drained
Cropping pattern	Moringa

## B. Physical and chemical properties of the field soil

Characteristics	Value
Particle size analysis	
% Sand	23
% Silt	37
% clay	40
Textural class	Clayey
Ph	4.9
Organic carbon (%)	0.45
Organic matter (%)	0.92
Total N (%)	0.13
Available P (ppm)	15.00
Exchangeable K (me/100 g soil)	0.30
Available S (ppm)	45
Source: SRDI	