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RESEARCH ARTICLE

BJRI Mesta 4: A newly released improved vegetable Mesta variety of *Hibiscus sabdariffa* L.

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Abstract

Vegetable mesta, also known as roselle, is a common species in many countries for confectioneries and a good source of nutrients and antioxidants. BJRI Mesta 4 is a nutrient-rich, climate-smart and widely adaptable vegetable Mesta cultivar that was developed by the Bangladesh Jute Research Institute (BJRI) and released by the National Seed Board (NSB) in 2022. To explore the superiority of the new variety, a study was conducted on yield and yield components using multivariate analysis over two planting seasons compared with the well-known variety BJRI Mesta 2. Significant differences ($p \leq 0.01$ or 0.05) were recorded among the qualitative and quantitative traits. It also possesses in Multi-location Trials during 2018 and 2019, the average yield of leaf, fruit and calyx of BJRI Mesta 4 was found 7.25%, 11.51% and 10.49% higher in BJRI regional stations respectively compared to the check variety. Plant height, branches per plant, fruit yield and leaf yield were taken into consideration during selection to create vegetable mesta types with appropriate yield. However, based on yield capacity, adaptability and nutraceutical properties, the newly released cultivar BJRI Mesta 4 can be recommended and promoted to the herbal food product industry in addition to being a substitute for leafy vegetables. With these views kept in mind, the study that has been conducted now may help in creating trustworthy selection criteria for programs to improve vegetable mesta breeding in the future.

Keywords: Vegetable mesta; roselle; leaf; calyx; yield; herbal

Introduction

Vegetable mesta or Roselle (*Hibiscus sabdariffa* L.) is commonly known as "Mesta" or "chukur" in Bangladesh and the Indian subcontinent. It is a kind of *Hibiscus* from the "Malvaceae" family, most likely from West Africa (Shoosh, 1993), and it is widely available in tropical regions, especially in African nations (Abu-Tarboush et al., 1997). Mesta has two unique botanical classes: the fiber-producing var. *altissima* and the edible var. *sabdariffa* (Sobhan, 1993). The edible variety is a short bushy plant (1 - 2 m) bearing profuse smooth fruits popularly known as roselle or rosella (Dempsey, 1975). Its fleshy calyx and epicalyx is used for jelly making and the dried form is processed into other confections like cordial drinks, jams, sauces, liqueurs, wines, and food preserves (Mostofa, et al., 2021).

Mesta is known for its adaptability to poor soils and exhibits a fair tolerance to drought. Traditionally, it is grown in Bangladesh on relatively less fertile marginal uplands where jute or other major field crops cannot be grown profitably in the Kharif (Monsoon) season (Mostofa et al., 2021). The production cycle of this plant lasts around six months. There are more than 300 roselle species that are found all over the world (Balarabe, 2019). Mesta is a time-bound plant which means the flowering time is fixed from November to December (Mostofa et al., 2021). The tropics provide three separate color groups: green, crimson, and dark red (Purseglove 1977). The leaves of the green forms are eaten as vegetables, while the calyx of the red and dark red varieties is used to extract juice for a fresh drink (Islam et al., 2016). Its attractive color and unique sour taste along with its multi-purpose uses made it popular around the world (Tareq et al., 2021).

Several recipes use the seeds, leaves, fruits, and roots of vegetables as well as other components of the plant. Tender leaves of Mesta are used for preparing curry (or) chutney in several parts of the country. The fruit (capsule), which is surrounded by a fleshy calyx (sepal) is used to make wine, juice, jam, jelly, syrup, gelatin, pudding, cakes, ice cream, pickles, tea, drink, and other confections (Mohamed et al., 2012), is the component of the plant that has the greatest commercial significance. The green leaves and fruit pericarp are used like a spicy version of spinach. Leaves can also be used as tea/beverage (Islam et. al, 2021). According to scientific evidence, vegetable mesta extract has a high level of antioxidant activity, is anti-proliferative, anti-carcinogenic, anti-hypersensitive, anti-hyper lipidomics, hepato-protective, diuretic and many other properties (Islam et. al, 2016). BJRI Mesta 4 (VM-2) is a high-producing, prickly-free, and very adaptable vegetable mesta cultivar that was recently developed by the Bangladesh Jute Research Institute. It was previously selected through an exclusive screening from collected Mesta germplasms of BJRI Gene Bank and trialed against a popular cultivar to assess its agricultural suitability and adaptability to sustainable cultivation conditions. Vegetable Mesta or Roselle is gaining popularity for its vegetable properties only, whereas information on its nutritional and medicinal values is barely recognized (Sanders et al., 2023). Given this information, an effort was made in the year 2020 to compare the calyx and leaves of the recently introduced variety BJRI Mesta 4 to the widely used variety BJRI Mesta 2.

The new vegetable mesta variety bearing higher yield potentials, adaptability and a considerable number of antioxidants may improve its acceptance to consumers, which would eventually strengthen the nation's economy. Utilization of marginal and fallow land to produce BJRI Mesta 4 increases women's involvement in agriculture in Bangladesh by selling fresh vegetables and confectionaries around the year. Similar to this, only thorough information and research should be used to identify and suggest a plant for commercial cultivation (Al-Mamun et al., 2022). Only by assessing the crop varieties that are now available can one gain this knowledge. The identification of potential genotypes for commercial cultivation is therefore made more challenging.

Materials and Methods

Mesta strains (branching type) collected from different parts of the country and preserved in the Gene Bank of BJRI were thoroughly screened and tested against the check variety BJRI Mesta 2 under preliminary yield trial, advanced yield trial and zonal yield trial for assessment of yield performance and other agronomic traits. Based on the results, one advanced line L-911 (ME-4) was selected as the best and upgraded for two years (2018 and 2019) under different topography at Manikganj, Kishanganj, Rangpur, Faridpur, Jessore and Dinajpur. The trials were carried out using an RCB design (Chowdhury et.al, 2024), with 3 replications in each plot's 60 m² effective area. The advanced line ME-4 (BJRI Mesta 4) and the check variety BJRI Mesta 2 were each given an equal piece of a 10-decimal plot in six different Jute Research Regional Stations. Agronomic trials were also conducted in different locations. The seeds of advanced line ME-4 and check variety BJRI Mesta 2 were sown from mid-

August to the end of August during the tested years (2018 and 2019). Intercultural operations such as thinning, weeding, agronomic techniques, and supplemental irrigations were completed simultaneously for entire plots as recommended by Chowdhury and Hassan (2013). It was raised using only the recommended cultural methods. The crop was harvested from mid-January to the end of January and necessary records were kept. Leaves data were recorded at 60, 90 and 120 days of crop age. Fruit-relevant data were collected from fifteen randomly selected plants. Data were recorded on plant population, plant height, number of leaves/plant, the weight of leaves/plant, number of branches/plant, and number of fruits/plant, weight per fruit and weight of calyces/fruit.

Data collection

Eleven quantitative and four qualitative features in total were observed. Fifteen randomly chosen plants were used for each genotype in each replication to collect data for each characteristic. At the seedling and growth stages in the field, the qualitative characteristics including stem color and leaf shape were observed visually. Visual observations of flower color, fruit shape, and calyx color were made at the reproductive and mature stages. Quantitative data collected include plant population, plant height, leaves per plant, leaf weight per plant, branches per plant, and fruits per plant, single fruit weight, single calyx weight, leaf yield and fruit yield (Table 1). Among the quantitative characters studied, plant population, leaves/plant, and fruits/plant were verified in the field and the residual characters were measured at the reproductive and mature stages (Tareq et al., 2021; Al-Mamun et al., 2022a).

Table 1. Quantitative characters studied from vegetable mesta genotypes

Traits	Denotation	Description
Plant population (Lha ⁻¹)	PP	Number of plants of 5 decimal or 200 square metres were counted at the time of harvest.
Plant height (m)	PH	At the time of harvest, the height in meters from the soil's surface to the tips of 15 randomly chosen plants was measured.
Leaves/plant	L	The total numbers of leaves were counted at 60, 90 and 120 days of crop age. The average number of leaves per plant was recorded.
Leaves weight/plant	LWt	The weight of leaves from the randomly selected 15 plants of each replication was taken on electric balance and per plant basis weight was calculated.
Branches/plant	Br	To determine the average branch number per plant, the number of branches on 15 randomly chosen plants was counted.
Fruits/plant	Fr	The total number of fruits of 15 plants from each replication was counted and an average number of fruits per plant was recorded.
Single Fruit weight (gm)	SFrW	The average weight of randomly selected fruit from the 15 selected plants of each replication was weighted on electric balance and the reading.
Single Calyx wt. (gm)	SCw	The average weight of calyx from randomly selected fruit of each replication was weighted on electric balance and the reading.
Leaf yield (tha ⁻¹)	LY	The leaf yield data were calculated as follows: Weight of leaves/plant × Plant population/ha
Fruit yield (tha ⁻¹)	FrY	The fruit yield data were calculated as follows: Weight of fruits/plant × No. of fruits/plant × Plant population/ha
Calyx yield (tha ⁻¹)	CY	The calyx yield data were calculated as follows: Weight of calyces/fruit × No. of fruits/plant × Plant population/ha

Data analysis

Using SAS 9.4 (SAS Institute, Inc., Cary, N.C., USA), all morphological and yield data were treated for an analysis of variance. Least Significant Difference (LSD), a statistical method, was used to calculate the means comparison. The PROC VARCOMP with limited maximum likelihood (REML) approach in SAS was used to estimate the variance components such as genotypic (σ^2_g), phenotypic (σ^2_p) and error (σ^2_e) from the relevant mean squares for all examined traits. Following Oladosu et al. (2014) methodology, the genetic advance, broad-sense heritability, genotypic coefficient of variation (GCV), and phenotypic coefficient of variation (PCV) were all evaluated.

Results

Qualitative variation

The new variety BJRI Mesta 4 is bushy and shorter than the check variety BJRI Mesta 2 with a higher number of branches and higher biomass (Mostofa, et al., 2022). The new variety is comparatively dwarf and branching type and is almost resistant to diseases and pests (Mostofa, et al., 2022). The variety BJRI Mesta 4 is morphologically different from the check one by its stem (Figure 1b) and fruit color (Figure 1c). It possesses a larger fruit size with fleshy calyces (Figure 1d). It has a green stem with purple nodes and its fruit calyx is green at the early stage but slightly pinkish pigmented at maturity, whereas the check variety (BJRI Mesta 2) has a purple color stem and dark red fruit calyx (Figure 2). Both cultivars produce semi-lobed leaves, smooth stems and cream-colored flowers.

Characteristics	BJRI Mesta 4 (ME-4)	BJRI Mesta 2 (VM-1)
(a) Plant type and leaf shape	 Dwarf and bushy plants with a higher number of semi-lobed leaves. Leaf and calyx taste sour.	 Dwarf and bushy plants with a higher number of semi-lobed leaves. Leaf and calyx taste sour.

(b) Stem color				
	It has a green and smooth stem with purple nodes		BJRI Mesta 2 has a smooth and purple stem	
				
(c) Flower color	Cream color with red center		Cream-colored flowers	
				
	Fruit capsule (pointed) and reddish green in color. Its fruit calyx is green at the early stage but slightly pinkish pigmented at maturity.			
(d) Fruit and fruit calyx				
	Fruit capsule (pointed) and reddish green in color. Its fruit calyx is green at the early stage but slightly pinkish pigmented at maturity.		Fruit capsule (pointed) and red. Dark red fruit calyx.	
				

Figure 1. Some qualitative Characters' of BJRI vegetable mesta genotypes (a) Plant type and leaf shape variation, (b) Stem color variation, (c) Flower color variation, (d) Fruit and fruit calyx variation.



Figure 2. Field view, fruits and calyces of the new variety BJRI Mesta 4 (green) and BJRI Mesta 2 (red).

Morpho-physiological and yield characters

The analysis of variance for the six locations showed significant differences ($p \leq 0.01$ or 0.05) for all the evaluated traits except for leaves weight per plant, non-significant differences were observed in Table 2. The range of 11.97 to 48.70 in the CV% for yield and yield-related components indicates that the traits under study are highly variable.

Table 2. Vegetable mesta genotypes for 11 traits were analyzed for variance over two crop seasons

S.O.V	Replication	Locations	Genotypes	Gen*Location	Error	Mean±Std error	CV (%)
DF	2	5	1	5	22		
PP	0.47*	0.54**	0.00	0.02	0.12	2.02±0.07	21.11
PH	0.25	0.95*	0.17	0.01	0.31	1.94±0.10	30.50
L	34.61	523.08*	32.51	7.98	165.70	27.77±2.25	48.70
LWt	393.42	468.96	4.98	19.86	237.08	53.59±2.59	29.00
Br	48.89**	25.52**	3.74	1.17	4.39	6.82±0.51	45.15
Fr	193.72	260.63*	70.28	1.52	92.10	26.46±1.74	39.35
SFrW	0.05	2.54**	0.21	0.15	0.46	6.90±0.14	11.97
SCw	0.04	0.63**	0.20	0.05	0.15	3.43±0.07	12.96
LY	6.20	31.31**	0.59	0.57	6.08	10.59±0.49	27.93
FrY	139.05	751.26*	205.35	8.04	188.71	36.85±2.59	42.12
CY	28.59	176.67*	71.60	2.51	48.20	18.35±1.29	42.07

Note: *Significant at 0.05 probability level; **highly significant at 0.01 probability level; df, degrees of freedom; SOV, Source of variation; CV, Coefficient of variation; Std error, standard error; PP, Plant population (Lha-1); PH, Plant height (m); L, No. of Leaves/plant; LWt, Leaf weight/plant (gm); Br, No. of Branches/plant; Fr, No. of Fruits/plant; SFrW, Single fruit weight (gm); SCw, Single calyx weight (gm); LY, Leaf Yield (tha^{-1}); FrY, Fruit yield (tha^{-1}); CY, Calyx yield (tha^{-1}).

Data on yield and yield contributing traits of the new variety ME-4 and check variety VM-1 at six BJRI stations namely Manikganj, Kishoreganj, Rangpur, Faridpur, Jessore, and Dinajpur during 2018 and 2019 are summarized in Table 3. The highest leaf yield (14.0 tha⁻¹), fruit yield (51.21 tha⁻¹) and calyx yield (25.42 tha⁻¹) were obtained at Manikganj, Jessore and Jessore respectively. The mean yield performance of the two years showed that the variety ME-4 gave a higher 7.25% leaf yield, 11.51% fruit yield and 10.49% calyx yield than the check variety (Table 4). Overall, the yield performance of BJRI Mesta 4 was outstanding in all BJRI stations.

Table 3. Mean performance for yield traits of individual location

Trait	Locations						Mean	LSD
	Manikganj	Kishanganj	Rangpur	Faridpur	Jessore	Dinajpur		
PP	1.83 ^b	1.72 ^b	1.77 ^b	2.39 ^a	2.37 ^a	2.06 ^{ab}	2.02	0.4159
PH	2.38 ^a	1.35 ^b	2.38 ^a	1.73 ^{ab}	1.93 ^{ab}	1.87 ^{ab}	1.94	0.6668
L	21.33 ^b	39.83 ^a	23.50 ^b	39.45 ^a	19.05 ^b	23.48 ^b	27.77	15.413
LWt	61.05 ^{ab}	42.13 ^c	62.98 ^a	58.27 ^{abc}	53.17 ^{abc}	43.92 ^{bc}	53.59	18.436
Br	7.67 ^{ab}	3.28 ^c	6.35 ^b	7.57 ^{ab}	9.50 ^a	6.53 ^b	6.82	2.5079
Fr	33.50 ^a	17.32 ^b	32.90 ^a	20.13 ^b	27.97 ^{ab}	26.97 ^{ab}	26.46	11.491
SFrW	6.28 ^c	6.16 ^c	7.09 ^a	6.617 ^{bc}	7.46 ^a	7.76 ^a	6.90	0.8111
SCw	3.04 ^d	3.11 ^{cd}	3.50 ^{abc}	3.39 ^{bcd}	3.70 ^{ab}	3.86 ^a	3.43	0.4612
LY	11.18 ^{ab}	7.05 ^c	10.92 ^{ab}	13.27 ^a	12.29 ^a	8.85 ^{bc}	10.59	2.9533
FrY	38.82 ^{ab}	17.49 ^c	40.93 ^{ab}	31.31 ^{bc}	49.62 ^a	42.93 ^{ab}	36.85	16.448
CY	19.03 ^{ab}	8.84 ^c	20.22 ^{ab}	16.09 ^{bc}	24.59 ^a	21.30 ^{ab}	18.35	8.3125

Note: LSD, Least Significant Difference; PP, Plant population (Lha-1); PH, Plant height (m); L, No. of Leaves/plant.; LWt, Leaf weight/plant (gm); Br, No. of Branches/plant; Fr, No. of Fruits/plant; SFrW, Single fruit weight (gm); SCw, Single calyx weight (gm); LY, Leaf Yield (tha⁻¹); FrY, Fruit yield (tha⁻¹); CY, Calyx yield (tha⁻¹); Mean values are explained for each trait and upper-case letter show significance.

Table 4. Yield performance (t/ha) of two varieties at different agro-ecological zones during 2018-2019

Locations	Yield components	2018		2019		Mean over years	
		BJRI Mesta 4	BJRI Mesta 2	BJRI Mesta 4	BJRI Mesta 2	BJRI Mesta 4	BJRI Mesta 2
Manikganj	Leaf	12.89	9.15	11.93	10.76	12.41	9.96
	Fruit	49.39	37.92	35.23	32.74	42.31	35.33
	Calyx	24.73	18.64	19.14	14.18	21.94	16.41
Kishoreganj	Leaf	7.00	5.59	8.90	6.72	7.95	6.16
	Fruit	21.5	16.91	17.28	14.25	19.39	15.58
	Calyx	11.27	8.87	9.42	6.37	10.35	7.62
Rangpur	Leaf	12.36	11.07	9.86	10.39	11.11	10.73
	Fruit	52.02	45.50	35.78	30.42	43.90	37.96
	Calyx	19.4	21.39	17.42	15.43	18.41	18.41
Faridpur	Leaf	15.72	12.45	14.22	10.69	14.97	11.57
	Fruit	37.87	29.14	35.45	22.78	36.66	25.96
	Calyx	19.9	14.80	16.92	12.74	18.41	13.77

Jashore	Leaf	14.79	12.61	11.61	10.15	13.20	11.38
	Fruit	52.79	52.07	45.21	48.41	49.00	50.24
	Calyx	25.88	16.23	22.04	34.21	23.96	25.22
Dinajpur	Leaf	10.12	9.17	8.30	7.81	9.21	8.49
	Fruit	48.35	40.53	37.69	37.69	43.02	39.11
	Calyx	24.23	21.22	21.25	18.50	22.74	19.86
Mean over stations	Leaf	12.15	10.01	10.80	9.42	11.48	9.71
	Fruit	43.65	37.01	34.44	31.05	39.05	34.03
	Calyx	20.90	16.86	17.70	16.91	19.30	16.88

Note: Yield increased in leaf (18.14%), fruit (14.74%) and calyx (14.33%) compared to the check variety.

Broad-sense heritability, genetic advance, and genetic analysis

Estimates of genotypic (σ^2_g), phenotypic (σ^2_p), and error (σ^2_e) variances ranging from 0.15 to 295.76, 0.25 to 504.08, and 0.10 to 258.64, respectively, are presented in Table 5.

Table 5. Means squares of sources of variance for 11 quantitative parameters of the mesta genotypes of vegetables

Traits	Mean	σ^2_p	σ^2_g	σ^2_e	PCV (%)	GCV (%)	h^2_B (%)	GA (%)
Plant population (Lha-1)	2.02	0.25	0.15	0.10	24.52	19.03	60	30.44
Plant height (m)	1.94	0.48	0.23	0.25	35.83	24.83	48	35.46
Leaves/plant	27.77	11.03	7.24	3.79	48.73	39.48	66	65.91
Leaves wt./plant	53.59	262.87	130.11	132.77	58.37	41.06	49	59.52
Branches/plant	6.82	383.37	124.73	258.64	35.79	20.42	33	23.99
Fruits/plant	26.46	206.08	105.19	100.89	53.68	38.35	51	56.45
Single Fruit wt. (gm)	6.90	1.11	0.72	0.39	15.28	12.27	65	20.31
Single Calyx wt. (gm)	3.43	0.30	0.16	0.13	15.88	11.80	55	18.06
Leaf yield (tha-1)	10.59	17.34	8.84	8.49	38.49	27.49	51	40.45
Fruit yield (tha-1)	36.85	504.08	295.76	208.32	59.96	45.93	59	72.47
Calyx yield (tha-1)	18.35	122.62	69.27	53.34	59.44	44.67	57	69.17

Note: σ^2_p , phenotypic variation; σ^2_g , genotypic variation; σ^2_e , error variation; PCV, phenotypic coefficient variation; GCV, genotypic coefficient variation; h^2_B , broad-sense heritability; GA, genetic advance.

The number of leaves per plant, the number of branches per plant, the number of fruits per plant, the yield of fruits, and the yield of calyces all showed high phenotypic, genotypic, and error variance. Plant population,

height, the number of leaves per plant, the weight of a single fruit or calyx, and leaf production all showed low genotypic and phenotypic diversity. Estimates of the phenotypic (PCV) and genotypic coefficients of variation (GCV) ranged from 15.28% to 59.96% and 11.80% to 45.93%, respectively. The highest PCV was recorded in the fruit yield (59.96%), followed by calyx yield (59.44%) and leaves weight per plant (58.37%). Similarly, the highest GCV was observed in fruit yield (45.93%) followed by calyx yield (44.67%) and leaves weight per plant (41.06%), highlighting the significant genotypic diversity of these traits and the potential for additional selection. In this study, broad-sense heritability ranged from 33% (branches per plant) to 66% (leaves per plant). The three traits that showed high heritability ($\leq 60\%$) are plant population, leaves per plant, and single fruit weight. The rest of the traits are observed in moderate heritability (30–60%) values. In terms of the 11 quantitative features, fruit yield had the largest genetic advance (72.47%), followed by calyx yield (69.17%). Single calyx weight (18.06%) and single fruit (20.31%) had the lowest values.

Relationship between Characteristics

The yield and yield component traits' straightforward Pearson phenotypic correlation coefficients were examined using the proc corr SAS software version 9.4 as shown in Table 6. Most characteristics don't exist in isolation; rather, they are linked to one another in intricate relationships that eventually affect the yield. This connection might be favorable or unfavorable. By identifying an association, the correlation coefficient (r-value) provides an understanding of the link between two different traits. Characters' phenotypic correlation coefficients ranged from 0.05 to 1.00, indicating greater phenotypic variation. There is no linear relationship, a perfect positive linear relationship, and a negative linear relationship, respectively, according to the r-values of 1, 0, and -1. According to Oladosu et al. (2018), the values between 0.7 and 1, 0.3 and 0.7, and 0 to 0.3 represent strong, moderate, and low positive linear relationships, respectively. In contrast, the values between -0.7 and -1, -0.3 and -0.7, and -0 to -0.3 represent strong, moderate, and low negative linear relationships, respectively. There was a significant to moderately favorable phenotypic connection between leaf yield and leaf weight per plant, fruit production and fruits per plant, and calyx yield and fruit yield. However, there was a discrepancy between the number of leaves per plant and the production of fruit and calyx. In Table 6, additional correlation coefficients between trait pairs of interest to vegetable mesta plant breeders are shown.

Table 6. Analysis of 11 quantitative variables from the vegetable mesta genotypes combined across two crop seasons to determine the correlation coefficient

Traits	PH	L	LWt	Br	Fr	SFrW	SCw	LY	FrY	CY
PP	-0.352*	0.168 ^{ns}	-0.385*	0.186 ^{ns}	-0.231 ^{ns}	0.078 ^{ns}	0.106 ^{ns}	0.310 ^{ns}	0.272 ^{ns}	0.285 ^{ns}
PH		-0.356*	0.822**	0.348*	0.625**	0.168 ^{ns}	0.131 ^{ns}	0.558**	0.377*	0.368*
L			-0.054 ^{ns}	-0.426*	-0.310 ^{ns}	-0.103 ^{ns}	0.061 ^{ns}	0.088 ^{ns}	-0.280 ^{ns}	-0.238 ^{ns}
LWt				0.364*	0.460**	0.184 ^{ns}	0.180 ^{ns}	0.749**	0.213 ^{ns}	0.215 ^{ns}
Br					0.340*	0.182 ^{ns}	0.112 ^{ns}	0.504**	0.436**	0.419**
Fr						0.318 ^{ns}	0.287 ^{ns}	0.283 ^{ns}	0.827**	0.818**
SFrW							0.966**	0.245 ^{ns}	0.575**	0.582**
SCw								0.259 ^{ns}	0.540**	0.567**
LY									0.388*	0.400*
FrY										0.997**

Legend: PP, Plant population (Lha-1); PH, Plant height (m); L, No. of Leaves/plant.; LWt, Leaf weight/plant (gm); Br, No. of Branches/plant; Fr; No. of Fruits/plant; SFrW, Single fruit weight (gm); SCw, Single calyx

weight (gm); LY, Leaf Yield (tha^{-1}); FrY, Fruit yield (tha^{-1}); CY, Calyx yield (tha^{-1}); **Highly significant at 1% probability level; *Significant at 5% probability level; ns, not significant.;

Socio-economic Importance

The fleshy calyx (sepal), which is used to make various confectionaries including juice, jam, jelly, syrup, drinks, gelatin, ice cream, tastes, etc., is the most significant plant component in terms of commerce (Figure 3). Tender leaves are used for preparing curry (or) chutney. The overall attractive color and unique sour taste of this crop along with its multi-purpose uses made it popular around the world. These findings serve as a foundation for recommending the new variety for commercial cultivation in the herbal food product industry as well as adding to the human diet for boosting immunity.



Figure 3. BJRI Mesta 4 multipurpose uses with fresh vegetables and confectionery foods

Discussion

Major differences in the features can speed up identifying and classifying separate lines during breeding operations (Akinrotimi and Okocha 2018). Plant breeders can choose genotypes for future breeding programs with the use of the knowledge produced through qualitative traits-based variability. For various morphological features in this investigation, there was significant diversity between the two types. Wide variation was observed in stem and fruit calyx, where green stem with purple nodes was observed in BJRI Mesta 4, whereas purple color stem was observed in BJRI Mesta 2. The fruit calyx of BJRI Mesta 4 is green at the early stage but slightly pinkish pigmented at maturity, whereas the check variety (BJRI Mesta 2) has dark red fruit calyx. It was also

mentioned that both vegetable mesta varieties differ in plant type and leaf shape, flower, and fruit color (Figure 1). The 11 separate morpho-physiological variables from the combined quantitative data of the two years showed a significant difference (Table 2). Ibrahim and Hussein (2006) observed genotypic variations in the yield components of *H. cannabinus* and *H. sabdariffa*. Differences between the characteristics' minimum and maximum values demonstrated the presence of variability between the two kinds. In their 2006 report, Ibrahim and Hussein discussed the variation in *H. sabdariffa* L. plant height and the number of capsules produced per plant. Nearly all the characteristics had large coefficients of variation (CV%), a measure of population variance. The findings of the univariate analysis show that the analyzed rosella types have a great deal of variability. Yield is a complex character in any crop which is governed by several factors. The development of high-yielding variety is essential for migrating the total production of vegetable mesta and also for its cultivation in the marginal land of Bangladesh. Out of six BJRI stations in six different AEZs of the country, the yield of fruit and calyx of the new variety BJRI Mesta 4 was found higher in all the locations. The highest fruit yield (51.21 tha^{-1}) and calyx yield (25.42 tha^{-1}) of BJRI Mesta 4 were found in Jessore station followed by Manikganj and Dinajpur station, respectively. The leaf yield of BJRI Mesta 4 was found higher in almost all the stations but lower in Dinajpur. The mean yield performance of BJRI Mesta 4 over locations and years was 11.16 tha^{-1} leaf yield, 35.93 tha^{-1} fruit yield and 18.03 tha^{-1} calyx yield which was 2.86%, 11.20% and 14.04% higher than that of the check variety respectively. Most genotypes had GCV values greater than 20%, except for plant population (19.03), single fruit weight (12.27), and single calyx weight (11.80). Low GCV indicated that the environment had an impact on how those phenotypes were expressed. The fact that PCV in every instance was larger than GCV suggests that the degree to which environmental factors affect phenotypic expression varies. As a result, there is a solid foundation for phenotypic selection that focuses on traits that are less impacted by the environment. Indicating the presence of a significant influence of additive gene action on the inheritance of these traits, high genetic progress together with high heritability was seen for plant population, leaves per plant, and single fruit weight. For plant height, leaf weight per plant, branches per plant, fruits per plant, and yields of leaves, fruits, and calyxes, however, considerable genetic progress and moderate heritability were found. Selecting characteristics with high GCV, GA, and heritability is crucial. Plant height, leaf weight per plant, and the number of branches per plant all significantly and positively correlated with the leaf yield. This study proved that branches per plant, fruits per plant, single fruit weight, and single calyx weight were highly significantly correlated with fruit yield and calyx yield. Moreover, fruit yield and calyx yield both showed a positive and significant correlation with plant height and leaf yield. Fruit yield and the calyx yield exhibited a strong positive connection (1.0). According to Webber et al. (2002), early maturity may reduce production per plant because early flowering and maturity result in short plants since early flowering inhibits vegetative development. This is supported by the negative association between the number of leaves per plant and fruit production and calyx yield. Typically, genotypes with early flowering produce more fruit than genotypes with late flowering. In terms of leaf, fruit, and calyx yield, the findings demonstrate that BJRI Mesta 4 beat the control variety BJRI Mesta 2. Vegetable Mesta production is appropriate for tropical and sub-tropical climates that are warm and humid (Ansari et al., 2013). For developing the new variety, a dedicated screening of germplasms was carried out for their suitability to be grown in the climate context of Bangladesh to select the most promising accession in terms of production. The yield trial showed a remarkable yield increase in leaf, fruit and calyx. Moreover, the new cultivar performed better than the check in most of the locations which implicit higher adaptability. The mean yield performance of the years 2018 and 2019 showed that the variety BJRI Mesta 4 gave 11.16 tha^{-1} leaf yield, 35.93 tha^{-1} fruit yield and 18.03 tha^{-1} calyx yield which is respectively 2.86%, 11.20% and 14.04% higher than that of the check variety. Therefore, it can be said that BJRI Mesta 4 having higher yield potential and adaptability, can be recommended for growing all over the country. Except for the plant population (15.45%),

the weight of a single fruit (12.27%), and the weight of a single calyx (11.80%), most genotypes displayed GCV values greater than 20%. Low GCV indicated that the environment had an impact on how they expressed their phenotype. Every time, PCV was larger than GCV, demonstrating that different environmental conditions have various degrees of influence on how phenotypic expression is expressed (Al-Mamun et al., 2022a). As a result, there is a solid foundation for phenotypic selection that focuses on traits that are less impacted by the environment. Plant population, leaves per plant, and single fruit weight all showed strong genetic progress and heritability, indicating a significant role for additive gene activity in the inheritance of these traits. However, most of the variables, except for single calyx weight, show substantial genetic progress and moderate heritability. Choosing traits with a high GCV, GA, and heritability (Al-Mamun et al., 2022b). Total four antioxidant components e.g., total phenol content, total flavonoid content, proanthocyanidin content, and anthocyanin content were measured from the calyx sample indicating possible usefulness as a beneficial vegetable in the human diet (Almajid et al., 2023; Mollah et al., 2020). Therefore, based on the nutritional and medicinal properties studied, both cultivars can be used to promote the nutraceutical and pharmaceutical industries (Sanders et al., 2023). However, several investigations claimed that the vegetable Mesta genotypes show changes in vitamins and mineral levels along with agro-climatic conditions and soil type (Atta et al., 2013; Islam 2019). The method used to analyze the study's contents might also be a factor for this variation. This issue could be the subject of further research. Different agroecological zones suggested that the line is better than the check variety concerning yield. Moreover, it is well recognized that branches per plant are one of the important parameters for leaf, fruit and calyx yield. The newly released cultivar BJRI Mesta 4 produced a higher plant height than the check variety BJRI Mesta 2 in all stations. The finding suggested that the BJRI Mesta-4 has the potential to get a higher yield.

Conclusion

BJRI Mesta 4 is a highly productive, adaptable, and nutritive multipurpose crop that has the potential to increase the income of small farmers. One of the most underutilized crops, its importance in our nation is still underappreciated. The combined examination of the qualitative data revealed four characteristics that set the genotypes apart from the control variety. The selection of a better vegetable mesta genotype will be aided by the highly significant and favorable relationships of the key features with other agronomic traits, such as leaf yield, fruit yield, and calyx yield. The global trade roadmap predicts that the market for vegetable mesta will triple over the next five years as people become more aware of natural herbal products. It is therefore time to recognize the potential of vegetable mesta in Bangladesh and to encourage its varied application. Both BJRI cultivars investigated and compared were discovered to have promising yield capacity, varied confectionery uses, and antioxidant qualities with a wide variety of health advantages. The novel variety BJRI Mesta 4 can be suggested for commercial production and contribute to the human diet for food security, however, considering the adaptability and yield potentiality. This evaluation might help create trustworthy selection indices for favorable agronomic features in vegetable mesta. It is recommended that vegetable Mesta genotypes show changes in vitamins and mineral levels along with agro-climatic conditions and soil type should be explored in future research to establish the study's contents might also be a factor for this variation.

Declaration

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References

- Abu-Tarboush, H. M., Ahmed S. A. B. and Al Kahtani H. A. Some nutritional and functional properties of karkade (Hibiscus sabdariffa) seed products. *Cereal chemistry*. (1997). 74(3): 352-355. doi.org/10.1094/CCHEM.1997.74.3.352.
- Akinrotimi, C. A. and Okocha P. I. Evaluations of genetic divergence in Kenaf (Hibiscus Cannabinus L.) genotypes using agro-morphological characteristics. *Journal of Plant Sciences and Agricultural Research*. (2018). 2(12): 2167–2412. doi:10.4172/2167-0412.1000246.
- Almajid, A., Bazroon, A., AlAhmed, A., & Bakhurji, O. Exploring the Health Benefits and Therapeutic Potential of Roselle (Hibiscus sabdariffa) in Human Studies: A Comprehensive Review. *Cureus*. (2023).15(11).
- Al-Mamun, M., Rafii, M., Oladosu, Y., Misran, A. B., Berahim, Z., Ahmad, Z., Arolu F., and Khan, M. H. Genetic diversity among kenaf mutants as revealed by qualitative and quantitative traits. *Journal of Natural Fibers*. (2022a). 19(11): 4170-4187. doi.org/10.1080/15440478.2020.1856268.
- Al-Mamun, M., Rafii, M. Y., Misran, A. B., Berahim, Z., Ahmad, Z., Khan, M. M. H., & Oladosu, Y. Estimating Genetic Analysis Using Half Diallel Cross Underlying Kenaf (Hibiscus cannabinus L.) Fibre Yield in Tropical Climates. *BioMed Research International*, (2022b). 1, 1532987. doi.org/10.1155/2022/1532987.
- Alshoosh, W. G. A. Chemical composition of some Roselle (Hibiscus sabdariffa) genotypes. (1997). http://inis.iaea.org/search/search.aspx?orig_q=RN:30018790.
- Ansari, M., Eslaminejad, T., Sarhadynejad Z., and Eslaminejad, T. An overview of the roselle plant with particular reference to its cultivation, diseases and usages. *European Journal of medicinal plants*. (2013). 3(1): 135-145. doi.org/10.9734/EJMP/2013/1889.
- Atta, S., Sarr, B., Diallo, A. B., Bakasso, Y., Lona, I. and Saadou. M. Nutrients composition of calyces and seeds of three Roselle (Hibiscus sabdariffa L.) ecotypes from Niger. *African Journal of Biotechnology*. (2013). 12(26). doi: 10.5897/AJB12.2634.

- Balarabe, M. A. Nutritional analysis of *Hibiscus sabdariffa* L. (Roselle) leaves and calyces. *Plant Journal*. (2019). 7(4): 62-65. doi: 10.11648/j.plant.20190704.11
- Chowdhury, M.I.H., Rakib, M.H., Das, C., Hossain, M.Z. Tree Species Germination: A Comprehensive Meta-Analysis and its Implications for Pre-Sowing Treatment in Bangladesh. *Journal of Soil, Plant and Environment*. (2024). 3(1): 24–40. doi.org/10.56946/jspae.v3i1.397
- Chowdhury, M. A. H. and Hassan, M. S. Hand book of agricultural technology. Bangladesh Agricultural Research Council, Farmgate, Dhaka. (2013), 230.
- Dempsey, J. Fiber Crops. The University Presses of Florida, Gainesville. Rose Printing Co. Tallahassee. (1975).
- Ibrahim, M. and Hussein, R. Variability, heritability and genetic advance in some genotypes of roselle (*Hibiscus sabdariffa* L.). *World Journal of Agricultural Science*. (2006). 2(3): 340-345. doi: 10.5923/j.ijaf.20130307.02
- Islam, A. A., Osman, M. B., Mohamad, M. B., and Islam, A. M. Vegetable Mesta (*Hibiscus sabdariffa* L. var *sabdariffa*): A Potential Industrial Crop for Southeast Asia. In *Roselle*. Academic Press. (2021). 25-42.
- Islam, A. A., Jamini, T. S., Islam, A. M., and Yeasmin, S. Roselle: a functional food with high nutritional and medicinal values. *Fundamental and Applied Agriculture*. (2016). 1(2): 44-49.
- Islam, M. M. Food and medicinal values of Roselle (*Hibiscus sabdariffa* L. Linne Malvaceae) plant parts: A review. *Open J Nutr Food Sci*. (2019). 1(1): 1003.
- Mohamed, B. B., Sulaiman, A. A. and Dahab, A. A. Roselle (*Hibiscus sabdariffa* L.) in Sudan, cultivation and their uses. *Bull. Environ. Pharmacol. Life Sci*. (2012). 1(6): 48-54.
- Mollah, M. A. F., Tareq, M. Z., Bashar, K. K., Hoque, A. Z., Karim, M. M., and Zahid-Al-Rafiq, M. Antioxidant properties of BJRI vegetable mesta-1 (*Hibiscus sabdariffa* L.). *Plant Science Today*. (2020). 7(2): 154-156. doi.org/10.14719/pst.2020.7.2.664
- Mostofa, M. G., Al-Mamun, M., Biswas, S. K., and Nur. I. J. BJRI Mesta 4 (Sobji Mesta) (Leaflet), Agriculture Wing, Bangladesh Jute Research Institute, Manik Mia Avenue, Dhaka, Bangladesh. (2022).
- Mostofa, M. G., Al-Mamun, M., Nur I. J. and Akter, N. BJRI Mesta-3: A newly released improved variety of *Hibiscus sabdariffa* L. *International Journal of Agricultural and Applied Sciences*. (2021). 2(2): 82-86. doi : 10.52804/ijaas2021.2213
- Oladosu, Y., Rafii, M. Y., Abdullah, N., Abdul Malek, M., Rahim, H. A., Hussin G., and Kareem, I. Genetic variability and selection criteria in rice mutant lines as revealed by quantitative traits. *The Scientific World Journal*. (2014). doi.org/10.1155/2014/190531
- Oladosu, Y., Rafii, M. Y., Magaji, U., Abdullah, N., Miah, G., Chukwu S. C., and Kareem, I. Genotypic and phenotypic relationship among yield components in rice under tropical conditions. *BioMed research international*. (2018). doi: 10.1155/2018/8936767
- Purseglove, J. W. Tropical crops dicotyledons (Volumes 1 and 2 combined). (1977).
- Sanders, M., Ayeni, A. O., and Simon, J. E. (Comparison of yield and nutrition of roselle (*Hibiscus sabdariffa*) genotypes in central New Jersey. *Journal of Medicinally Active Plants*. 2020). 9(4).
- Sobhan, M. A. Heritability of fibre, fruit and seed yield in *Hibiscus sabdariffa* L (Doctoral dissertation, Doctoral dissertation, PhD Thesis. Department of Botany, Dhaka University. Dhaka, Bangladesh). (1993).
- Tareq, Z., Mollah, A. F., Sarker, S. A., Bashar, K. K., Sarker, D. H., Moniruzzaman, S.N., Islam, Al Rafiq Z., and Sadat, A. Nutritive value of BJRI mesta-2 (*Hibiscus sabdariffa* L.) leaves. *Acta Agrobotanica*. (2021). 74(1). doi.org/: 10.5586/aa.749
- Webber III, C. L., Bhardwaj H. L., and Bledsoe. V. K. Kenaf production: fiber, feed, and seed. *Trends in new crops and new uses*. (2002). 13, 327-339.

RESEARCH ARTICLE

Optimization of Amaranthus production under irrigation and poultry manure application using grey relational analysis

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Abstract

This study was carried out to optimize Amaranthus production under varying irrigation levels and poultry manure applications using Grey Relational Analysis. The experimental design was split-plot, where irrigation treatment was the main plot, while poultry manure was the subplot treatment. The irrigation levels are FIT50, FIT75, FIT100 and FIT135 which correspond to 50% Full irrigation, 75% full irrigation, 100% full irrigation and 135% full irrigation treatments, in combination with the poultry manure application rates M0t/ha, M5t/ha, M10t/ha and M20t/ha. Sixteen experimental treatments were arranged in a 4 x 4 factorial experimental design and replicated 3 times. Sprinkler irrigation system was adopted for the study. Results showed that there was corresponding increase in the response of Amaranthus growth characteristics and yield to the combination of irrigation and poultry manure treatments. The Crop Water-use efficiency initially showed a linear corresponding increase to an average value of 473kg/ha/mm but later exhibits a diminishing return beyond FIT100 irrigation. While production cost showed continual increase to average value of \$377/ha as the inputs treatment level also increased to FIT135 irrigation combined with the poultry manure M20t/ha. Grey Relational Grades (GRG) calculated were 0.854, 0.790, 0.787, 0.765, 0.740, 0.629, 0.621, 0.620, 0.546, 0.543, 0.515, 0.475, 0.458, 0.443, 0.417 and 0.403 respectively. Amaranthus cultivated under FIT100_M20t/ha treatment matched with the highest GRG value 0.854, making FIT100_M20t/ha treatment the optimum. Analysis of variance for the GRG showed that irrigation and poultry manure are highly significant input variables for Amaranthus production. The findings from this study guide farmers on the optimal combination of water and organic fertilizer to maximize Amaranthus yield and minimize resource waste, thereby enhancing sustainability in crop production.

Keywords: Amaranthus; Grey relational grades; Water; Treatments; Yield; Irrigation

Introduction

Amaranthus, commonly known as amaranth, is a vital leafy vegetable crop recognized for its high nutritional content, including proteins, vitamins and minerals (Umakanta, et al., 2020). It holds significant agricultural and economic potential, especially in regions where its cultivation is widespread (Yahia et al., 2019). Being one the

most demanded leafy vegetables, it is majorly grown in Southern Western Nigeria because of their leaves (Ogwu, 2020, Akinboye et al., 2024). The harvested leaves when boiled briefly and mixed with other condiments are eaten with pounded yam, Eba, Amala and other local dishes. The leaves have been reported to contain significant quantities of vitamins, crude fibre, phytochemicals and mineral (Umakanta, et al., 2024). The optimization of *Amaranthus* production is critical to enhance its yield and quality under varying environmental conditions, particularly under irrigation regimes and organic amendments like poultry manure. Proper management of these factors is essential for sustainable food production and improved crop performance. Irrigation plays a pivotal role in ensuring the successful cultivation of *Amaranthus*, especially in regions experiencing irregular rainfall or water scarcity (Egbebi et al., 2024). It influences not only the yield but also the overall quality of the crop. Studies have shown that water availability significantly affects plant physiological processes, including photosynthesis, transpiration, and nutrient uptake (Olufayo et al., 1996, Fasinmirin and Olufayo, 2009, Roberto and Roberto, 2022). Optimizing irrigation is crucial during the period of dry season when there is complete cessation of rainfall when water is a limiting factor. Besides, the need to efficiently manage water as a natural resource is becoming one of the major news headlines as tensions and clashes that are arising from natives due to conflicting demands is becoming frequent in sub-Sahara Africa. Freshwater is gradually becoming scarce as per head consumption increases due to global population explosion. Also, climate change signatures is taking its toll on food production as crop water demands also increase (Geraldo and Henrique, 2023). Practice of wild application of water to the field from surface irrigation is no longer acceptable in the environment. Therefore efficient water management strategies for crop production are keenly sought for by policy makers.

Irrigation scheduling and guided application rates offer a promising water management strategy, ensuring optimal water-use efficiency while preventing water stress and reducing unnecessary waste (FAO, 2021). However, in tropical regions, where water scarcity often coincides with poor soil fertility, it is crucial to manage both resources in tandem. Most African soils are inherently low in organic carbon, slightly acidic, and relatively sandy (Lal, 2009; Lehmann and Joseph, 2009, Sophie et al., 2021, Odebiri et al., 2024), making nutrient input as vital as water management for crop productivity. While efficient irrigation maximizes water use, nutrient availability particularly through organic amendments like poultry manure helps improve soil health and sustain crop yields (Hussain et al., 2023). Therefore, a balanced approach to both water and organic nutrient management is essential to overcoming the dual challenges of water and soil limitations in tropical regions, ultimately optimizing crop production. Organic amendments, particularly poultry manure have gained significant attention in recent years due to their ability to improve soil structure, enhance nutrient availability and promote sustainable agriculture (Emeghara, 2023). When documentation on comparing performance of inorganic and organic fertilizers were considered on soil, findings shown that inorganic fertilizers have not been helpful in maintaining soil health (Ozlu and Kumar 2018, Rashmi et al., 2020). They disrupt the natural balance of nutrients in the soil, leading to nutrient imbalances and reduced soil quality. The combined effects of these have been proven to result in the loss of soil organic matter, decreased soil fertility and increased susceptibility to erosion. This was also confirmed by Farmerline (2023). Poultry manure is rich in essential macro and micronutrients like nitrogen, phosphorus, and potassium, which are key for *Amaranthus* growth. Applying poultry manure as a soil amendment has been proven to increase soil organic matter, improve soil microbial activity, enhance water retention capacity and ultimately improving crop yield and quality (Ramadevi et al., 2023). The application rate of poultry manure, however, requires optimization, as excessive use can lead to nutrient imbalances and environmental concerns such as nitrate leaching (Mary et al., 2022). Thus, determining the ideal combination of irrigation levels and poultry manure application can maximize *Amaranthus* production while minimizing environmental impact. Optimization techniques, such as Grey Relational Analysis (GRA), have emerged as powerful tools to handle multi-response problems (Deng, 1982;

Sonja et al., 2015). GRA is part of Grey System Theory, which is applied when information is incomplete or uncertain. It is a multi-criteria decision-making (MCDM) technique that assesses the similarity between a reference series and alternative options (Chia-Chen et al., 2019). It helps identify which alternative is closest to an ideal solution based on various performance indicators. The process involves several key steps: Normalization which involves standardization of initial data to allow for fair comparison across different criteria. Then calculation of Grey relational coefficient, the coefficient quantifies the degree of similarity between the reference series and alternatives. Thereafter, alternatives ranking based on the grey relational grades to identify the most suitable option. It is being integrated in different fields of science and engineering (Huai et al., 2021; Dmitrovic et al., 2022, Dumitru et al., 2023, Shivakumar and Murali, 2024). In agricultural systems, integration of Grey Relational Analysis (GRA) is particularly useful for addressing multi-response optimization because it will allow for the evaluation and comparison of multiple performance criteria simultaneously. In agricultural studies, various factors such as yield, water-use efficiency, nutrient uptake and crop quality need to be optimized, which often have conflicting or interdependent relationships. GRA simplifies this complexity by providing a systematic method to rank and prioritize different outcomes based on their relative performance, even when the units or scales of the responses vary (Li et al., 2022). It is also effective in dealing with uncertainty which is common in agriculture due to environmental variability and fluctuating conditions (Xie et al., 2020). In addition, it allows flexibility by integrating diverse factors and offering a holistic approach to finding the optimal combination of inputs for enhanced agricultural productivity. By using GRA, agricultural researchers and practitioners can make data-driven decisions to improve overall crop performance across multiple criteria. In agricultural management, GRA can also be applied to various aspects such as resource allocation, crop selection and risk assessment. It can aggregate insights from multiple stakeholders, reducing individual biases and enhancing overall decision quality (Xu, 2024). Also the ability to evaluate numerous factors simultaneously makes it suitable for complex agricultural decisions where trade-offs are common. Research has demonstrated GRA's effectiveness in agricultural contexts, such as evaluating sustainable farming practices or optimizing input use (Edinam et al., 2022). For instance, studies have shown that using GRA can significantly improve decision accuracy in selecting crops based on environmental conditions and market demand (Oduniyi and Chagwiza, 2021; Lamptey, 2022). The integration of Grey Relational Analysis in Amaranthus production enables the identification of the best irrigation and poultry manure combinations to maximize multiple growth parameters including leaf area index, plant height, crop yield, water use efficiency and quality. By analyzing the interaction effects of water and nutrient inputs, GRA provides a quantitative framework for decision-making in agricultural management (Romero-Gelve et al., 2020). It allows for the simultaneous evaluation of multiple performance indicators, such as plant height, leaf greenness and crop water use efficiency, making it a suitable tool for optimizing complex agricultural systems.

The objective of this study is to optimize Amaranthus production under varying irrigation levels and poultry manure applications rates using Grey Relational Analysis. By applying this method, the study aims to determine the most favorable treatment combinations that enhance crop productivity while maintaining resource efficiency and environmental sustainability.

Materials and methods

The field experiment was conducted at the Teaching and Research Farm of the Department of Agricultural Education, Federal College of Education (Technical), Akoka, Lagos, longitude 6.3167°N, latitude 3.2250°E, with an altitude of 10m above sea level on coastal area of Southwestern Nigeria as shown in Figure 1. Lagos

state has a landmass of 356,861 hectares, of which 169,613 hectares are designated for agriculture (Lagos State Government, 2018).

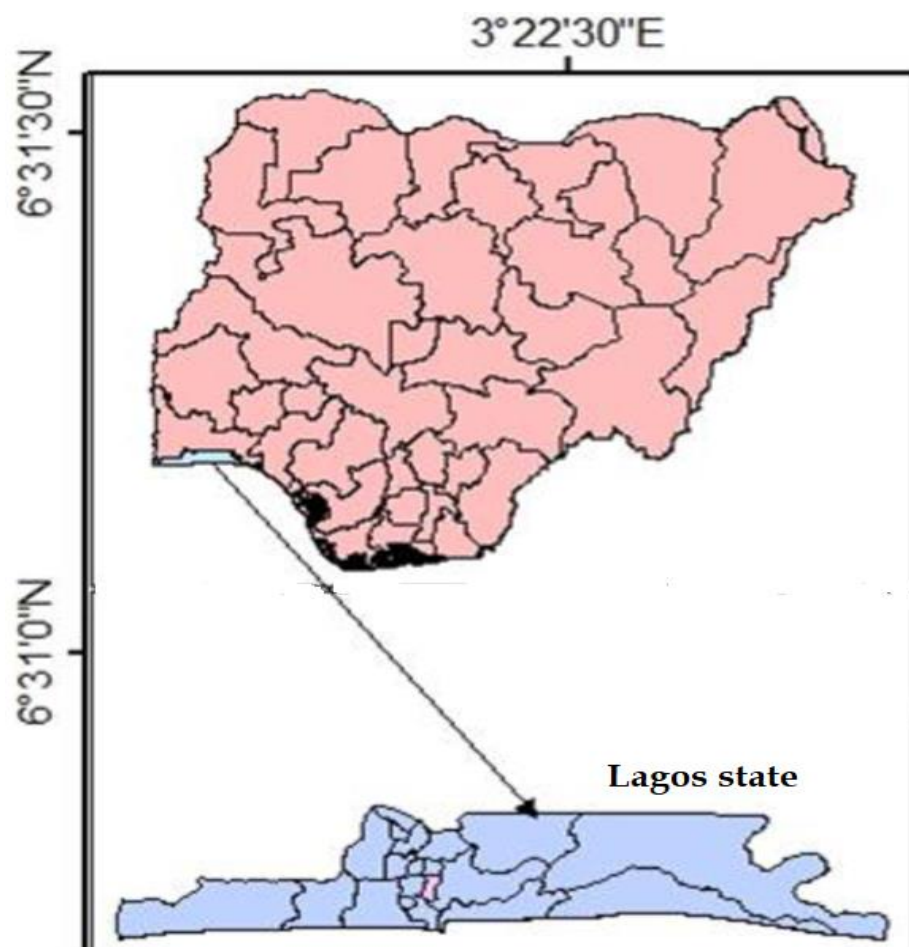


Figure 1: Map of Nigeria showing geographical location of Lagos State

The experiment was carried out between the months of January to February 2020 during the dry season. The averages of the weather parameters over the study site at the period of the experiments are recorded in Table 1.

Table 1: Average weather parameters over the study site for 2020

Months	Maximum Temperature °C	Minimum Temperature °C	Relative Humidity %	Wind Speed m/sec	Solar Radiation MJ/m ² .day	ET _o mm/day	Rainfall mm
January	35.64	23.87	70.74	2.24	17.91	3.69	15.40
February	31.57	22.60	71.15	2.52	17.33	3.51	0.00

The study site has been on continual use for crop production for over 10 years. An area of 15m x 10m portion of the field was cleared and appropriate land preparation was carried out to permit effective seed bed formation. Soil samples were collected randomly from twelve points to a depth of 0.45m and analyzed using standard soil analysis procedure prior to the starting of the experiment. The soil texture within the depth

considered is sandy loam according to Soil Survey Staff (2006). Other results of the soil sample analysis are given in Table 2.

Table 2: Soil particle size analysis and the textural classification of the study site

Particle size fractions	Minimum (%)	Maximum (%)	Standard Deviation	Coefficient of Variation (%)
Sand (%)	80.9	88.4	3.1	3.7
Silt (%)	6.1	10.6	1.7	21.6
Clay (%)	4.4	10.4	2.3	27.6
Textural Class	Sandy loam			

The experimental design was split-plot, with irrigation treatments as main plots and poultry manure as random treatments within the subplots. The treatments consisted of four irrigation levels based on water requirement. They were FIT50, FIT175, FIT100 and FIT135. The implication of this irrigation levels is explained thus: 50% Full irrigation represents a significant reduction in water availability, simulating drought or water stress conditions, which is important to understand how crops respond under water scarcity, 75% full irrigation reflects a moderate water stress due to water deficit, 100% full irrigation is the baseline treatment, representing ideal irrigation conditions where the crop receives the full amount of water required for optimal growth and FIT135 was considered excessive water application helps to assess the effects of over-irrigation. These levels correspond to realistic agricultural scenarios. Four levels of poultry manure were 0t/ha, 5t/ha, 10t/ha and 20t/ha. 0t/ha represents typical farming without manure application, 5t/ha this represent low manure application rate, scenarios where limited resources are available, 10t/ha this also represent moderate manure application rate, balancing nutrient supply with economic and environmental sustainability) and 20t/ha this mimics a high application rate, which can help explore the upper limits of nutrient input and its effects on plant growth, yield and soil properties. The experimental design was 4 by 4 full factorial combination of irrigation and poultry manure replicated 3 times.

Table 3: Chemical analysis of the soil in the study area and poultry manure applied

Chemical Properties	Concentration	
	Soil	Poultry Manure
pH	6.72 $\pm$ 0.32	6.22 $\pm$ 0.83
Organic matter (%)	5.48 $\pm$ 1.76	36.87 $\pm$ 1.21
Total N (g/kg)	8.56 $\pm$ 1.10	10.18 $\pm$ 1.42
P (mg/kg)	25.32 $\pm$ 0.55	4.86 $\pm$ 0.35
K (cmol/kg)	0.51 $\pm$ 0.08	6.97 $\pm$ 0.78
Ca (cmol/kg)	0.29 $\pm$ 0.06	24.91 $\pm$ 0.34
Mg (cmol/kg)	0.29 $\pm$ 0.53	3.79 $\pm$ 0.42
Na (cmol/kg)	0.53 $\pm$ 0.06	2.35 $\pm$ 0.42
Fe (g/kg)	0.52 $\pm$ 0.58	2.30 $\pm$ 0.28
Al (g/kg)	0.97 $\pm$ 0.65	3.12 $\pm$ 0.31
Cu (mg/kg)	21.59 $\pm$ 0.11	34.96 $\pm$ 0.35
EC (mS/m)	36.41 $\pm$ 6.72	58.56 $\pm$ 7.03

The poultry manure that was used for the experiment was collected from the Poultry Section of the Teaching Farm of the Agricultural Education Department, Federal College of Education (Technical), Akoka. The poultry manure were divided into five samples, later air dried, grounded into its fineness and was analyzed for its chemical properties. The soil samples collected were from twelve points and were analyzed for their chemical properties using the standard procedure as recommended by the Soil Science Society of America (SSSA) shown in Table 3. The manure was incorporated into the top 0.25m and was evenly mixed manually with the soil within each subplot at different rates aforementioned. These application rates were modified from the previous works of Enujeke (2013) and were studied from other related literatures to be appropriate for boosting vegetable production in soils with low fertility status (Xiang et al., 2022). Seeds of *Amaranthus Hybridus* (cultivar-NHAC₃ purchased from Lagos State Ministry of Agriculture, Alausa, Ikeja, Nigeria. The seeds were mixed thoroughly with sand at the rate 10g seed/ 100g of dry sand to ensure even distribution. Seeding by broadcasting was carried out on 18th January, 2020.

Irrigation water source was from the centralized Industrial Overhead Reservoir meant for the FCET Waterworks. The water was directed through an existing PVC pipe connection into a 3000 litres capacity tank placed at a height of 5m above the ground. The tank was connected to a 25.4mm diameter PVC pipe main line which delivers water under gravity to the submains and finally to the networks of 25.4mm diameter flexible HDPE (High Density Polyethelyn) hoses which run along the furrows as lateral with closed ends. Plastic hose risers accompanying the micro-sprinklers were connected to the laterals through the connecting accessories (barbs). The micro-sprinklers erected with the aid of its plastic spikes within the subplots. The arrangement of the micro sprinkler was carried out such that the radius of their throws overlap. The sprinklers discharge was measured using catch cans using the method reported by Oluwagbayide et al.,(2021). The pressure created from the 3000litres capacity over head tank made addition of water pump to power the micro sprinklers unnecessary. The irrigation treatments were imposed on the field by controlling the duration of water application usually in the evening by 6pm. The 135% full irrigation usually receives water late into the night. Prior to seeding, the field was adequately watered through unrestricted irrigation. This continued in the first week of planting to facilitate higher proportion of seeds germination and early establishment. Thereafter, different irrigation level treatments were initiated. Plants stands within each seed bed were thinned to 50 plant stands per 1m² subplot giving a planting density of 50,000 stands /ha.

Since the experiment was to commence in January (one of driest months of the year in Nigeria), the amount of water required to prepare the land to facilitate higher proportion of seed germination, early crop establishment and meet crop water requirement was computed from the guidelines given by Raveendra et al, (2017) and Luca et al., (2020). For the land wetting or soaking, water required was calculated using Equation 1;

$$WR_{LS} = W_s + k. ET_o + (P - P_e).....(1)$$

WR_{LS} = land wetting or soaking water requirement (mm), W_s = depth of water required to saturate the soil (mm), k = Evaporation coefficient ($k = 0.90$), ET_o = Reference crop evapotranspiration for the growing period computed from Daily reference evapotranspiration obtained using the FAO 56 Penman-Monteith method (Allen et al., 1998), P_e = Effective rainfall (mm), P = Deep percolation loss (mm), The Net Irrigation Water Requirement during the growing period of the *Amaranthus Hybridus* was calculated using Equation 2:

$$NIR = \sum_{i=1}^n (K_{c,i} ET_{o,i} + P_i) - (P_{e,i} + GW_{c,i}).....(2)$$

K_c = Crop coefficient, K_c for the *Amaranthus* at the various growth stages was estimated from the previous work of Ufoegbune et al., (2016), GW =groundwater contribution in terms of capillary rise (mm)

The Total Net Irrigation Demand was calculated using Equation 3:

$$NIWR = NIR + WR_{LS} \dots \dots \dots (3)$$

The Gross Irrigation Water Requirement was determined from using equation 4:

$$GIWR = \frac{NIWR}{E_a \times E_c} \dots \dots \dots (4)$$

E_a = Irrigation application efficiency, E_c = Field application efficiency

In order to determine the duration of irrigation (T_d) at each irrigation event, the gross irrigation ($GIWR_g$) was determined by dividing the irrigation amount (NIWR) by the irrigation application efficiency (E_a). The irrigation amount i.e the volume of irrigation water applied in litres (L) per irrigation events was computed from Equation 5

$$NIWR = W_a \times I \dots \dots \dots (5)$$

W_a is the wetted area (m^2) and I is the irrigation (mm). I is irrigation depth (mm). Irrigation application efficiency of 70% was used (ASAE, 1990).

The total crop evapotranspiration (ET) was estimated in each of the treatments using the soil water balance equation in Equation 6.

$$ET = P + I - D - R - [S_{i+1} - S_i] \dots \dots \dots (6)$$

where; ET is total crop evapotranspiration (mm), P = precipitation (mm) measured using installed manual rain gauges, I = irrigation amount (mm) which was applied to bring the soil water to field capacity. D = Deep percolation (mm), R = runoff (mm). Deep percolation was assumed to be negligible in treatments with deficit irrigation since the experiment was carried out in the dry season characterized by little rainfall. However, it was computed after deduction of other components in the treatments with surplus irrigation. Runoff from the over irrigated plots were collected using runoff collectors installed and was measured. $S_{i+1} - S_i$ is the change in soil water storage (mm) determined based on the difference in soil water content between two successive measurements within the soil depth during the first and next successive soil moisture contents.

Agronomic measurements

The average plant height, number (N) of leaves per stand, length (L) and width (W) of leaves from 4 selected Amaranthus plants per plot, were monitored on weekly basis at the center of the middle row in each plot until when the vegetable was matured for market standard. The leaf area (LA) was calculated following the procedure described by Blanco and Folegatti (2003).

Leaf area

The average leaf area was calculated by multiplying the length of leaf by the widest width and by a factor 0.851 given in Equation 7.

$$\text{Leaf Area (cm}^2\text{)} = L \times W \times 0.851 \dots \dots \dots (7)$$

Leaf area index (LAI)

The Leaf Area Index (LAI) was computed by dividing the total Leaf Area (LA) of the Amaranthus plants by the land area they occupied (equal plot size of each replicate) given in Equation 8;

$$\text{Leaf Area Index} = \frac{\text{Total Leaf Area per plant} \times \text{total number of plants}}{\text{Plot total Area}} \dots\dots\dots(8)$$

Canopy cover

The active plant canopy cover was measured using Canopeo application version for android smart phone. Canopeo is automatic colour threshold image analysis tool developed in the MATLAB programming language (Mathworks, Inc., Natick, M.A) using colour values in the red-green-blue (RGB) system. It analyses image based on the selection of pixels according to the ratios of Red/Green, Blue/Green and excess green index (Liang et al., 2012, Paruelo et al., 2000, Biró et al., 2024) The results of the analysis is converted into a binary image where white pixels correspond to the pixels that satisfied the selection criteria (green canopy) and black pixels correspond to the pixels that did not meet the selection criteria (not green canopy). Fractional green canopy cover ranges from 0 (no green canopy cover) to 1 (100% green canopy cover) (Andres and Tyson, 2015). The application automatically records geographical coordinates, date and time so that the identity of each image can be traced. The relationship connecting the total leaf area per plant and the canopy cover was established using the regression model given in Equation 9

$$\text{Active Canopy cover (\%)} = 0.0113 \text{ Total Leaf Area} + 37.584 \dots\dots\dots(9)$$

Leaf greenness index

The leaf greenness measurement was taken using Colour Analyser, a digital imaging application on Android smart phone. Images of Amaranthus leaves were taken using the smart phone camera between the hours of 10 am and 11 am. The image was processed by the MATLAB based program developed into colour analyser application. The application converts the image into colour values of red-green-blue (RGB) system given in Equation 10.

$$f(x, y) = p00 + p10 \cdot x + p01 \cdot y \dots\dots\dots(10)$$

Where p00, p10 and p01 are modal parameters or constants. According to Amar et al., (2016), the values obtained for the modal parameters are 775.4, -1218 and -827.3 respectively. In equation, the variables x and y are the mean brightness ratios r and g respectively. The three primary colors (Red, Green and Blue) are converted into mean brightness ratios (r, g, b) to get better results. Where r, g and b are given in Equations 11, 12 and 13 respectively:

$$r = \frac{R}{(R+G+B)} \dots\dots\dots(11)$$

$$g = \frac{G}{(R+G+B)} \dots\dots\dots(12)$$

$$b = \frac{B}{(R+G+B)} \dots\dots\dots(13)$$

Yield

The harvesting of the vegetable was carried out at the optimum age of 4 weeks after sowing when their stems and leaves were fully developed. At the age of 4 weeks the vegetable has not lignified. Therefore, suitable for market standard. Harvesting was carried out by uprooting the shoots. Thereafter, root depth were measured and severed manually using knives. The yield was determined by weighing using Equation 14 below.

$$\text{Fresh Harvestable Yield} = \frac{\text{Weight of fresh Amaranthus (tons)}}{\text{Area Harvested (m}^2\text{)}} \dots\dots\dots(14)$$

Water-use efficiency

The water use efficiency was determined from the ratio of the yield to irrigation depth given in Equation 15 below

$$\text{Crop water use efficiency} = \frac{\text{crop yield (kg/ha)}}{\text{total irrigation depth (mm)}} \dots\dots\dots(15)$$

Grey relational analysis

The raw experimental data from the physical characteristics expressed as vegetative, yields, water use efficiency, and production cost were first normalized using Equations 16 and 17

Highly desired plant qualities such as plant height, number of leaves, leaf area, leaf area index, stem girth, canopy cover, greenness index, harvestable yield and water use efficiency (the higher-the-better) is normalized using:

$$x_{ij} = \frac{y_{ij} - \min(y_{ij})}{\max(y_{ij}) - \min(y_{ij})} \dots\dots\dots(16)$$

While the undesired qualities such as production cost (the lower-the-better) is expressed as:

$$x_{ij} = \frac{\max(y_{ij}) - y_{ij}}{\max(y_{ij}) - \min(y_{ij})} \dots\dots\dots(17)$$

Where y_{ij} are the raw experimental data. x_{ij} is the reference value for performance of experiment i for j response. $\min y_{ij}$ is the minimum value of y_{ij} for the j th response, and $\max y_{ij}$ is the maximum value of y_{ij} for the j th response (Sonja et al., 2015).

The grey relational coefficient was calculated from the normalized experimental data using the Equation 18.

$$\gamma(x_{0j}, x_{ij}) = \frac{(\Delta_{\min} + \xi \Delta_{\max})}{(\Delta_{ij} + \xi \Delta_{\max})} \text{ for } i = 1, 2, \dots, m \text{ and } j = 1, 2, \dots, n \dots\dots\dots(18)$$

Where $\gamma(x_{0j}, x_{ij})$ is the grey relational coefficient between x_{0j} and x_{ij}

The deviation sequence is calculated using Equation 19

$$\Delta_{ij} = |x_{0j} - x_{ij}|, \dots\dots\dots(19)$$

$$\Delta_{\min} = \min\{\Delta_{ij}, i = 1, 2, \dots, m; j = 1, 2, \dots, n\}$$

$$\Delta_{\max} = \max\{\Delta_{ij}, i = 1, 2, \dots, m; j = 1, 2, \dots, n\}$$

ξ is the distinguishing coefficient; it can assume range between 0 and 1. It is the index of distinguishability. The lesser it is, the higher its distinguishability. Therefore, ξ is assumed as 0.5 in this present study. The Grey Relational Grade $\Gamma(X_0, X_i)$ is computed using Equation 20

$$\Gamma(X_0, X_i) = \sum_{j=1}^n w_j \gamma(x_{0j}, x_{ij}) \quad \text{for } i = 1, 2, \dots, m \quad \dots\dots\dots(20)$$

$\sum_{j=1}^n w_j$: is the weighting of response w_j

Overall ranking of the amaranthus physical characteristics was based on the grey relational grade. In this way, optimization of the various physical characteristics can be converted into optimization of a single grey relational grade. The highest grey relational grade is the best input interactions that yield the physical characteristics with the best optimization.

Statistical analysis

The Grey relational grade sequence was subjected to Two-way ANOVA to determine which input factors (irrigation and manure) significantly affect the optimum performance characteristic of the Amaranthus. The percentage contribution by each input factors to the performance characteristic of the Amaranthus at the optimal level was determined.

Results and discussions

Effect of irrigation levels and poultry manure application rates on amaranthus consumptive use

Table 4 provides the water balance components and evapotranspiration (ET) results for various treatments of Amaranthus during the growing. Irrigation levels ranged from 60.5 mm for FIT50 treatments to 160.5 mm for FIT135 treatments. Rainfall was constant at 15.4 mm across all treatments. Observation from the results revealed that higher irrigation levels result in increased values of ET. ET increases from 95.3 mm in FIT50_M0t/ha to 217.1 mm in FIT135_M0t/ha.

This is as more available water leads to higher plant transpiration and evaporation from the soil surface. The increase in ET from FIT50 to FIT100 suggests that Amaranthus responds positively to increased water availability, with a greater proportion of the available water being used for growth. The range of water use (ET) obtained during the amaranthus growing season closer to the range of 70mm –160mm reported by irrigation in the same ecological zone (Fasimirin et al.,2009; Fasina et al., 2015; Ufoegbune et al., 2016; Egbebi et al., 2024). Beyond FIT100, the increase in ET becomes excessive, indicating that FIT135 represents over-irrigation, as not all water is being effectively used by the plant. Runoff and deep percolation were only observed in the FIT135 treatments, with runoff ranging from -10.7mm to -19.9 mm and deep percolation ranging from -12.4mm to -16.8 mm. This is not unconnected moisture content of the soil under these treatments that were above the field capacity range due to excess water application. This indicates that much of the additional water is lost and does not contribute to crop water use after the soil field capacity is exceeded. Irrigation at this level suggests inefficiency and potential water wastage (Ogunlela and Sadiku, 2017, Ajayi et al., 2023). However, other treatments ranging from FIT50 to FIT100 had no runoff or deep percolation. Evapotranspiration increased with higher irrigation levels, ranging from 95.3 mm for FIT50 to 217.1 mm for FIT135. Negative ΔS values across all treatments indicate that soil moisture was depleted to meet the crop's

water requirements. As irrigation increases, the depletion of soil water storage becomes less severe. Treatments FIT50 and FIT135 had ΔS -19.4 mm and -4.5 mm respectively, showing that higher irrigation levels provide sufficient water, reducing the need to deplete soil moisture.

Table 4: Water balance component data and evapo-transpiration for the Amaranthus treatments during the growing season

Treatments	Irrigation (mm)	Rainfall (mm)	Runoff (mm)	Deep percolation (mm)	Change in soil water storage (ΔS) (mm)	ET (mm)
FIT50_M0 t/ha	60.5	15.4	0.0	0.0	-19.4	95.3
FIT50_M5 t/ha	60.5	15.4	0.0	0.0	-18.2	94.1
FIT50_M10 t/ha	60.5	15.4	0.0	0.0	-14.4	90.3
FIT50_M20 t/ha	60.5	15.4	0.0	0.0	-11.4	87.3
FIT75_M0 t/ha	90.5	15.4	0.0	0.0	-14.5	120.4
FIT75_M5 t/ha	90.5	15.4	0.0	0.0	-11.7	117.6
FIT75_M10 t/ha	90.5	15.4	0.0	0.0	-15.8	121.7
FIT75_M20 t/ha	90.5	15.4	0.0	0.0	-10.5	116.4
FIT100_M0 t/ha	120.5	15.4	0.0	0.0	-8.4	144.3
FIT100_M5 t/ha	120.5	15.4	0.0	0.0	-9.4	145.3
FIT100_M10 t/ha	120.5	15.4	0.0	0.0	-7.5	143.4
FIT100_M20 t/ha	120.5	15.4	0.0	0.0	-8.6	144.5
FIT135_M0 t/ha	160.5	15.4	-19.9	-16.8	-4.5	217.1
FIT135_M5 t/ha	160.5	15.4	-12.6	-13.5	-2.5	204.5
FIT135_M10 t/ha	160.5	15.4	-10.7	-12.4	-1.8	200.8
FIT135_M20 t/ha	160.5	15.4	-14.6	-14.6	-1.6	206.7

Under each irrigation regime, higher manure application rates (M5t/ha, M10t/ha, M20t/ha) slightly reduce ET. For instance, in the FIT50 treatment, ET decreases from 95.3 mm under M0t/ha treatment to 87.3 mm M20t/ha treatment, showing that manure improves soil moisture retention, reducing the need for water loss through transpiration. Manure application helps boost soil organic matter, which improves water-holding capacity, thus reducing the overall ET required by the crop. Are et al., (2017) found that organic amendments like poultry manure improve soil physical properties, including bulk density and porosity, enhancing water retention and reducing soil evaporation. This explains the lower ET values observed in manure-treated plots.

Growth parameters

The plant height, stem girth, number of leaves, root depth, average leaf area, leaf area index, canopy cover and leaf greenness, are some of the important parameters that directly reflect the growth of the Amaranthus plants. From Table 5, Under Plant height, FIT50_M0t/ha has a height of 25.12cm, while FIT100_M20t/ha shows the highest height 53.46cm. This suggests that both irrigation and poultry manure significantly improve plant height. The combination of FIT75 and M10t/ha gives an intermediate height 51.25cm, indicating that beyond a certain level, adding more manure or irrigation water may plateau the height response. The stem girth improves as manure and irrigation increase. FIT100_M10t/ha has the largest stem girth 2.33cm, showing a

substantial improvement compared to the treatment FIT100_M0t/ha that recorded girth 1.51cm. The percentage increase from FIT100_M0t/ha treatment to FIT100_M10t/ha is around 54.3%. The notable enhancement in girth at higher manure levels may point to increased nutrient availability facilitating stem thickening.

The number of leaves increases with both irrigation and manure application, peaking at treatment FIT75_M20t/ha with 35.67leaves, while treatment FIT100_M0t/ha has 31.67leaves. This corresponds to a modest 12.65% increase from the performance of FIT100_M0t/ha. However, the highest manure application rate M20t/ha enhances leaf count at each irrigation level. As for the leaf wideness, FIT100_M20t/ha exhibits the highest average leaf area 48.29cm², which is an increase of 41.5% compared to FIT100_M0t/ha that has 34.12cm². Higher leaf area improves the photosynthetic capacity of the plant, contributing to better growth (Sokoto and Victor, 2017). The leaf area index (LAI) improves with increasing irrigation and manure. Treatment FIT135_M10t/ha has the highest LAI 0.155, compared to FIT50_M0t/ha 0.027. A substantial increase, this reflects the improved plant density and canopy spread (Luka et al., 2023).

The canopy cover rises with both irrigation and manure, with FIT100_M20t/ha almost completely covering the area at 99.7%. This is a 30.6% increase over FIT100_M0t/ha, demonstrating the importance of combining both treatments to optimize plant coverage. Leaf greenness data is a proxy for chlorophyll content. It also improves with both irrigation and manure, with FIT100_M20t/ha yielding the greenest leaves (46.45%). This is crucial for photosynthesis and ultimately productivity. In all, the least growth response parameters were obtained under FIT50_M0t/ha treatment. This may be linked with the combined effect of water stress due to deficit irrigation imposed and zero soil fertility boost from no manure application (Emeghara, 2023)

Yield, water-use efficiency and production cost

At harvest, under the FIT50 Series, it is observed that as poultry manure application increases from 0 to 10 t/ha, there is a notable increase in yield (from 6.49 to 9.91 t/ha), suggesting that manure significantly enhances yield at lower irrigation levels. The highest yield at this irrigation level is achieved with M10 t/ha. Under FIT75 irrigation series, the yield increases dramatically with both irrigation and manure application; the highest yield is observed at M20 t/ha 37.58 t/ha, which is a 143.7% increase from M0t/ha at the same irrigation level FIT75. Under the scenario of over irrigation, FIT135 treatments series, increasing trends are observed, the maximum yield at M20t/ha is 42.41t/ha, which is only a slight improvement over M10t/ha. The highest yield is observed at FIT100_M20t/ha

Crop Water Use Efficiency (CWUE) increases under FIT50 irrigation treatment series with increasing manure application, peaking at M10 t/ha with 198.23 kg/ha/mm. This is similar to the findings of Egbebi et al., (2024) in their experiment on *Amaranthus viridis* during the dry season irrigation and fertilizer micro-dosing on water application in Isan Ekiti, Nigeria. However the most efficient water use observed in this study is at FIT75_M20 t/ha with 521.38 kg/ha/mm

The average cost incurred was highest under FIT135_M20t/ha treatment level with average value of 377.8\$/ha. Lowest cost was incurred under FIT50_M0t/ha treatment level. This is because FIT135_M20t/ha treatment level received the highest quantity of production inputs in terms of water application and manure applications.

Grey relational analysis

The normalized values of *Amaranthus* growth characteristics, yield characteristics and production cost are shown in Table 6. Plant height, Stem girth, number of leaves, leaf area index and canopy cover gave normalized value of 1.000 at FIT135_M20t/ha treatment. This shows that these growth characteristics are in their best response under this treatment. While root depth, average leave area and leaf greenness had

normalized values less than 1.000 under the treatment. The fresh harvestable yield and water use efficiency had their best performance under different treatments. Production cost gave normalized value of 1.000 under FIT50_M0t/ha treatment. This is because the lower the production cost the better it is for production cycle.

Table 7 shows the deviation sequence. The deviation sequence measures the values of comparable sequences how far away to the values of reference sequence. If the value of deviation is close to 1.000, it is commented that comparable sequence is remote to reference sequence. On the other hand, if the value of deviation is close to 0, they are close to each other (Irfan et al., 2016). Table 8 shows the grey relational coefficients and grade for each treatment. The highest achievable (GRG) grey relational grade is 1.000. The larger the GRG, the closer is the amaranthus quality to the objective value (Bimal et al., 2023). The treatment number 12 which is FIT100_M20t/ha treatment, highlighted in green is the acceptable and closer to the reference sequence (ideal sequence), in which the highest GRG 0.841 was obtained. Next to treatment number 12 is number 11 (FIT100_M10t/ha) highlighted in yellow colour.

Table 5: Means and standard deviations of the Amaranthus growth parameters, yield, water-use-efficiency and production cost

Treatments	Plant height (cm)	Stem Girth (cm)	Number of Leaves	Root Depth (cm)	Average Leaf Area (cm ²)	Leaf Area Index	Canopy Cover (%)	Leaf Greenness	Fresh Harvestable Yield (t/ha)	Crop Water Use Efficiency (kg/ha/mm)	Expenditure (\$/ha)
FIT50_M0t/ha	25.12±2.45	0.92±0.04	11.67±3.89	12.03±2.98	23.51±4.55	0.027±0.01	75.51±1.98	35.09±7.76	6.49±1.87	129.75±23.76	113.61
FIT50_M5t/ha	27.21±3.76	0.90±0.15	14.00±4.76	12.49±3.34	24.53±3.67	0.034±0.01	80.33±3.98	36.91±5.87	9.10±2.91	182.03±10.56	120.87
FIT50_M10t/ha	35.74±3.99	0.97±0.28	15.67±3.82	11.39±3.23	26.47±4.71	0.042±0.02	82.07±4.76	38.48±5.11	9.91±3.99	198.23±32.90	124.44
FIT50_M20t/ha	39.44±7.74	1.15±0.40	19.00±4.91	12.45±2.45	26.85±2.09	0.051±0.02	84.56±4.09	39.23±2.89	9.11±3.71	182.25±13.06	133.33
FIT75_M0t/ha	48.18±9.98	1.05±0.11	22.33±2.88	11.76±2.56	29.56±1.90	0.066±0.02	87.11±7.94	38.91±3.97	15.43±3.82	214.32±35.93	162.22
FIT75_M5t/ha	48.71±1.24	1.24±0.26	25.67±5.87	12.82±4.78	31.85±4.89	0.082±0.03	90.40±4.70	43.36±3.99	19.98±2.89	277.47±28.90	168.89
FIT75_M10t/ha	51.25±1.04	1.81±0.29	27.33±4.80	11.69±3.54	31.54±4.63	0.086±0.02	91.21±4.50	43.27±7.67	25.33±3.21	351.82±45.91	173.33
FIT75_M20t/ha	54.84±8.72	1.91±0.43	35.67±5.76	11.39±2.45	32.55±2.75	0.133±0.04	94.54±3.05	42.43±4.98	37.58±4.31	521.38±30.67	182.22
FIT100_M0t/ha	54.49±1.05	1.51±0.37	31.67±3.96	13.86±4.67	34.12±2.98	0.108±0.06	96.27±5.87	39.50±3.72	17.04±4.90	177.54±13.07	215.56
FIT100_M5t/ha	42.78±8.89	1.77±0.43	31.67±5.90	11.19±3.76	41.56±4.91	0.131±0.09	98.70±0.62	44.95±5.87	30.17±5.04	314.26±54.09	222.22
FIT100_M10t/ha	52.19±7.31	2.33±0.54	33.00±6.02	9.89±4.10	43.54±4.10	0.143±0.08	99.20±0.03	45.51±4.87	42.61±6.12	443.88±89.55	226.67
FIT100_M20t/ha	53.46±9.55	2.29±0.73	31.67±4.88	9.86±2.12	48.29±4.65	0.153±0.09	99.70±0.03	46.45±4.99	44.43±5.32	462.76±70.54	235.56
FIT135_M0t/ha	47.20±8.02	1.17±0.45	31.33±5.12	8.53±2.19	30.55±3.99	0.096±0.03	97.90±0.08	36.10±5.31	21.38±8.55	133.62±13.78	357.78
FIT135_M5t/ha	49.81±9.18	1.96±0.66	34.00±4.89	9.52±2.15	38.81±4.13	0.132±0.08	99.80±0.01	38.05±2.90	32.77±6.42	204.80±25.88	364.44
FIT135_M10t/ha	53.18±7.41	2.01±0.34	36.65±3.84	11.55±4.78	43.21±3.77	0.155±0.06	99.87±0.05	45.12±3.98	40.29±5.67	251.59±28.33	368.89
FIT135_M20t/ha	53.26±1.03	2.19±0.98	33.33±9.58	11.26±3.45	43.58±5.89	0.152±0.05	99.80±0.01	42.29±5.09	42.41±4.08	265.83±19.76	377.78

Table 6: Normalized sequence of Amaranthus growth parameters, yield, water-use-efficiency and production cost

Plant height (cm)	Stem Girth (cm)	Number of Leaves	Root Depth (cm)	Average Leaf Area (cm ²)	Leaf Area Index	Canopy Cover (%)	Leaf Greenness	Fresh Harvestable Yield (t/ha)	Water Use Efficiency (kg/ha/mm)	Expenditure (\$)
0.000	0.009	0.000	0.343	0.000	0.000	0.000	0.000	0.000	0.000	1.000
0.068	0.000	0.091	0.274	0.041	0.048	0.267	0.161	0.064	0.152	0.975
0.346	0.047	0.156	0.463	0.119	0.098	0.394	0.298	0.084	0.199	0.958
0.466	0.172	0.286	0.264	0.135	0.165	0.479	0.365	0.162	0.386	0.924
0.750	0.100	0.416	0.394	0.244	0.270	0.567	0.337	0.219	0.246	0.815
0.767	0.235	0.545	0.195	0.337	0.381	0.679	0.728	0.330	0.430	0.739
0.850	0.634	0.844	0.424	0.728	0.412	0.809	0.720	0.460	0.647	0.790
0.967	0.984	0.935	0.464	0.768	0.746	0.922	0.910	0.613	0.899	0.773
0.955	0.427	0.779	0.000	0.428	0.565	0.879	0.388	0.258	0.139	0.613
0.574	0.608	0.779	0.501	0.728	0.730	0.962	0.868	0.579	0.537	0.588
0.880	1.000	0.831	0.745	0.808	0.814	0.979	0.918	0.883	0.915	0.571
0.922	0.970	0.779	0.751	1.000	0.879	0.996	1.000	0.951	1.000	0.538
0.718	0.189	0.766	1.000	0.284	0.478	0.935	0.089	0.364	0.011	0.076
0.803	0.737	0.870	0.815	0.617	0.732	1.000	0.261	0.642	0.219	0.050
0.978	0.774	0.948	0.434	0.795	0.898	1.000	0.883	0.973	0.465	0.034
1.000	0.967	1.000	0.488	0.891	1.000	1.000	0.634	1.000	0.485	0.000

Table 7: Deviation sequence of Amaranthus growth parameters, yield, water-use-efficiency and production cost

plant height (cm)	Stem Girth (cm)	Number of Leaves	Root Depth (cm)	Average Leaf Area (cm ²)	Leaf Area Index	Canopy Cover (%)	Leaf Greenness	Fresh Harvestable Yield (t/ha)	Water Use Efficiency (kg/ha/mm)	Expenditure (\$)
1.000	0.991	1.000	0.657	1.000	1.000	1.000	1.000	1.000	1.000	0.000
0.932	1.000	0.909	0.726	0.959	0.952	0.733	0.839	0.936	0.848	0.025
0.654	0.953	0.844	0.537	0.881	0.902	0.606	0.702	0.916	0.801	0.042
0.534	0.828	0.714	0.736	0.865	0.835	0.521	0.635	0.838	0.614	0.076
0.250	0.900	0.584	0.606	0.756	0.730	0.433	0.663	0.781	0.754	0.185
0.233	0.765	0.455	0.805	0.663	0.619	0.321	0.272	0.670	0.570	0.261
0.150	0.366	0.156	0.576	0.272	0.588	0.191	0.280	0.540	0.353	0.210
0.033	0.016	0.065	0.536	0.232	0.254	0.078	0.090	0.387	0.101	0.227
0.045	0.573	0.221	1.000	0.572	0.435	0.121	0.612	0.742	0.861	0.387
0.426	0.392	0.221	0.499	0.272	0.270	0.038	0.132	0.421	0.463	0.412
0.120	0.000	0.169	0.255	0.192	0.186	0.021	0.082	0.117	0.085	0.429
0.078	0.030	0.221	0.249	0.000	0.121	0.004	0.000	0.049	0.000	0.462
0.282	0.811	0.234	0.000	0.716	0.522	0.065	0.911	0.636	0.989	0.924
0.197	0.263	0.130	0.185	0.383	0.268	0.000	0.739	0.358	0.781	0.950
0.022	0.226	0.052	0.566	0.205	0.102	0.000	0.117	0.027	0.535	0.966
0.000	0.033	0.000	0.512	0.109	0.000	0.000	0.366	0.000	0.515	1.000

Table 8. Grey relational coefficient of Amaranthus growth parameters, yield, water-use-efficiency, production cost , Grey Relational Grades and their ranks

plant height (cm)	Stem Girth (cm)	Number of Leaves	Root Depth (cm)	Average Leaf Area (cm ²)	Leaf Area Index	Canopy Cover (%)	Leaf Greenness	Fresh Harvestable Yield (t/ha)	Water Use Efficiency (kg/ha/m)	Expenditure (\$)	Grey Relational Grade	Rank
0.333	0.33	0.333	0.43	0.333	0.33	0.333	0.333	0.333	0.333	1.000	0.403	16
0.349	0.33	0.355	0.40	0.343	0.34	0.405	0.373	0.348	0.371	0.952	0.417	15
0.433	0.34	0.372	0.48	0.362	0.35	0.452	0.416	0.353	0.384	0.922	0.443	14
0.483	0.37	0.412	0.40	0.366	0.37	0.490	0.440	0.374	0.449	0.869	0.458	13
0.667	0.35	0.461	0.45	0.398	0.40	0.536	0.430	0.390	0.399	0.730	0.475	12
0.682	0.39	0.524	0.38	0.430	0.44	0.609	0.648	0.427	0.467	0.657	0.515	11
0.769	0.57	0.762	0.46	0.647	0.45	0.723	0.641	0.481	0.586	0.704	0.620	8
0.938	0.96	0.885	0.48	0.683	0.66	0.866	0.848	0.564	0.832	0.688	0.765	4
0.918	0.46	0.694	0.33	0.466	0.53	0.805	0.450	0.403	0.367	0.564	0.546	9
0.540	0.56	0.694	0.50	0.648	0.64	0.930	0.791	0.543	0.519	0.548	0.629	6
0.807	1.00	0.748	0.66	0.723	0.72	0.960	0.858	0.810	0.854	0.538	0.790	2
0.865	0.94	0.694	0.66	1.000	0.80	0.993	1.000	0.911	1.000	0.520	0.854	1
0.639	0.38	0.681	1.00	0.411	0.48	0.885	0.354	0.440	0.336	0.351	0.543	10
0.718	0.65	0.794	0.73	0.567	0.65	0.999	0.404	0.583	0.390	0.345	0.621	7
0.958	0.68	0.906	0.46	0.709	0.83	0.999	0.811	0.948	0.483	0.341	0.740	5
1.000	0.93	1.000	0.49	0.821	1.00	1.000	0.577	1.000	0.493	0.333	0.787	3

Main effects of irrigation and manure on the grey relational grade

The main effects plots for the grey relational grade are shown in Figure 2. The graph provides valuable insights into the balance between nutrient management and water use for enhancing amaranthus yield and quality. Irrigation plot shows that the grey relational grades continue to increase as the irrigation level increase until it reached the optimal point at FIT100. Beyond this irrigation level, slight diminishing returns sets in. This indicates that further addition of irrigation water beyond FIT100 will not positively affect the growth and yield characteristics of the amaranthus. The water use efficiency diminishes beyond FIT100, which is the optimal irrigation level. This is because crop will not absorb beyond its water requirement. On the other hand, further

addition of water beyond that peak will imply over-irrigation. This is a pathway to waterlogging, nutrient leaching, or other stress factors, which hamper growth. In the long run it will only lead to unhealthy environment, wastage of resources and increase in the production cost which is a major negative desire of the consumers.

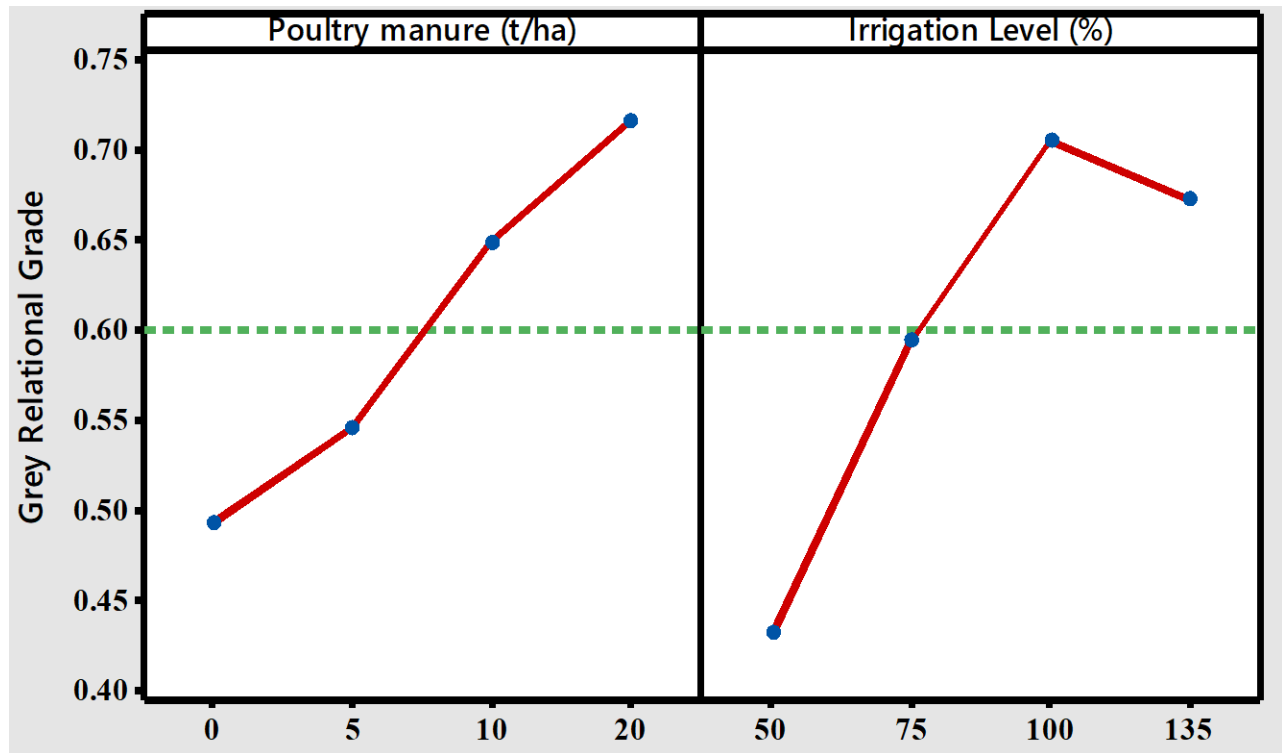


Figure 2: Grey relational grades main effects plot

Manure level increase was shown to increase the grey relational grade. The more the increase in manure levels, the closer the grey relational grade approaches unity (1.000), indicating that the more the performance response of the amaranthus. However, the trend seems to approaching plateau between 10 and 20 t/ha, indicating diminishing returns or a near-optimal range for manure application. Higher levels of manure likely enhance soil fertility, improve soil structure and increase organic matter content, all conducive to plant growth. The dotted green line at a GRG value of 0.60 represent a benchmark for satisfactory or average performance. In both panels, values above this threshold indicate better performance with increasing poultry manure and adequate irrigation. This agrees with the findings of Oworu et al., (2010). Similar findings were also reported by Zingore and Giller (2012) in Zimbabwe on Soyabean crop, a yield increase ratio of 1.18 to that of the control was obtained using cattle manure at 14t/ha. Chipomho et al., (2018) reported tomato yield increase approximately 2.7 times more than control in the same country using goat manure at 10t/ha. While Ndung'u et al., (2021) reported maize yield increase of 4times more than control at 16t/ha in Kenya. The optimal levels obtained for this study were found at 10t/ha and 20t/ha. Therefore, the optimal level of the input parameters is FIT100 irrigation level and 20t/ha manures treatment.

Analysis of variance of Grey relational grade

Table 9 presents the results of an Analysis of Variance (ANOVA) conducted on the Grey Relational Grade (GRG) values. Different irrigation levels contributed 55%, while poultry manure application rates contributes 35% of the total variability to the Grey Relational Grade values. Comparison between the two percentage

contributions shows that irrigation is the most influential factor in determining the performance of the amaranthus under each treatment as measured by GRG. The F-value for irrigation and poultry manure is 15.77 and 9.82 respectively. This further suggests that irrigation has a more significant impact on the Grey Relational Grade. The P-value for the duo is 0.001 and 0.003 respectively. They are both far below the significance threshold (typically 0.05). The P-values confirm that both production factors are statistically significant.

Table 9: Analysis of variance for the Grey relational grade

Source	DF	Adj SS	Adj MS	F-Value	P-Value	Contribution
Irrigation (%)	3	0.17713	0.059045	15.77	0.001	55%
Manure (t/ha)	3	0.11025	0.036751	9.82	0.003	35%
Error	9	0.0337	0.003744			10%
Total	15	0.32108				

Conclusions

It can be concluded that increase in the combination of irrigation and poultry manure treatment levels gave a corresponding increase in the response of Amaranthus growth characteristics. The crop water-use efficiency initially showed a linear corresponding increase but later exhibits a diminishing return beyond FIT100 irrigation level. So, the optimal levels of production input parameters for the desired performance characteristics of Amaranthus is obtained within FIT100_M20t/ha treatment. With this combination it is possible to decrease irrigation water, and maximize the growth characteristics yield, water use efficiency and minimize production cost of Amaranthus. Based on the ANOVA of the GRG results, it is observed that irrigation and manure exerted a significant influence on the amaranthus multiple response parameters. However, while the FIT100_M20t/ha treatment is highly effective for optimizing the growth, yield, and water-use efficiency of Amaranthus in the tropical rainforest agro ecological zone, its application should be carefully managed to account for environmental variability, long-term soil health and potential economic constraints. By addressing these factors, the treatment can be refined and adapted to suit a wide range of farming systems and contribute to sustainable crop production practices.

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References

- Akinboye, O.E., Makinde, A. I, Odusote, T.A., Afodu, O.J., Akinwole, O.T and Shobo, B.A.(2024). Optimizing nutrient levels for early growth of african spinach (*amaranthus* spp.) in a hydroponic system. *Nigerian Journal of Agriculture and Agricultural Technology*. Volume 4, Number 3, ISSN (Online): 2811-1893
- Allen, R.G., Pereira, L.S., Raes, D. and Smith, M. (1998). *Crop Evapotranspiration Guidelines for Computing Crop Water Requirements – FAO Irrigation and Drainage Paper 56*. Rome, FAO Food and Agriculture Organization of the United Nations, 1–13.
- Andres P and Tyson E. O. (2015). Canopeo: A Powerful New Tool for Measuring Fractional Green Canopy Cover. *Agronomy Journal*. 107:2312–2320. DOI: 10.2134/agronj15.0150
- Ajayi, E. O; Adebisi, C. R and Agaku, T. D.(2023): Growth and yield of amaranth (*Amaranthus Cruentus*) as affected by different sources of irrigation water in Ibadan, South-Western Nigeria. *Agricultural Society of Nigeria (ASN)/Proceeding of the 57th Annual Conference / 23 – 27 Oct. 2023 / Fulafia Nasarawa State*
- Are K. S, Adelana, A.O, Fademi, I.O and Aina, O.A (2017). Improving physical properties of degraded soil: Potential of poultry manure and biochar. *Agriculture and Natural Resources* 51:454-462. <https://doi.org/10.1016/j.anres.2018.03.009>
- ASAE, (1990). ASAE EP405.1. *Irrigation Systems Design*. Extension Service Bimal S · Prasenjit S · Suman D · Binod C.T and Prasanna P (2023): Grey Relational Analysis–Based MADM Strategy Under Possibility Environment and Its Application in the Identification of Most Important Parameter Affecting Climate Change and the Impact of Urbanization on Hydropower Plants. *Process Integration and Optimization for Sustainability* 7:599–608. <https://doi.org/10.1007/s41660-023-00319-w>
- Biró L, Kozma-Bognár V, Berke J.(2024) Comparison of RGB Indices used for Vegetation Studies based on Structured Similarity Index (SSIM) *J Plant Sci Phytopathol.*; 8: 007-012. DOI: 10.29328/journal.jpsp.1001124
- Blanco F.F and Folegatti M.A (2003). A New Method for Estimating the Leaf Area Index of Cucumber and Tomato Plants. *Horticultura Brasileira*, Brasília, v. 21, n. 4, p. 666-669
- Chia-Chen Y, Shang-Ling O and Li-Chang, H (2019); A Hybrid Multi-Criteria Decision-Making Model for Evaluating Companies' Green Credit Rating: Sustainability, 11, 1506; doi:10.3390/su11061506
- Chipomho, J., T. Tauro, L. Mtali-Chafadza, C. Chipomho, and A.B. Mashigaidze. (2018). Influence of organic and inorganic amendments on rape (*Brassica napus* L.) leaf yield and weed dynamics in a highly degraded sandy soil at Marondera, Zimbabwe. *Journal of Applied Science in Southern Africa* 24 (1): 50–68
- Deng, J. (1982). Control problems of grey systems. *Systems and Control Letters*, 1(5), 288-294
- Dmitrović, S. Pajčin, I. Lukić, N. Vlajkov, V.; Grahovac, M. Grahovac, J. Jokić, A (2022). Taguchi Grey Relational Analysis for multi response optimization of bacillus bacteria flocculation recovery from fermented broth by chitosan to enhance biocontrol efficiency. *Polymers*, 14, 3282. <https://doi.org/10.3390/polym14163282>
- Dumitru, T. Petrescu, M.G. Tanase, M. Ilincă, C.N.(2023) Multi-Response Optimization Analysis of the Milling Process of Asphalt Layer Based on the Numerical Evaluation of Cutting Regime Parameters. *Processes* 2023, 11, 2401. <https://doi.org/10.3390/pr11082401>

- Edinam D.S , Wanglin M and Alan R (2022): Effects of sustainable agricultural practices on farm income and food security in northern Ghana. *Agricultural and Food Economics*. <https://doi.org/10.1186/s40100-022-00216-9>
- Egbebi, I.A. Awopegba, T.M. Akinola, M.O. Egbebi, O.E. and Olakayode, A.O (2024): Effects of dry season irrigation and fertilizer micro-dosing on water application efficiency, agronomic performance and water use efficiency of *Amaranthus viridis*. *Journal of Agriculture and Environment*. Vol. 20 No. 1: 227-237
- Emeghara, U. U (2023): Effects of planting method and different nutrient sources on the yield, growth and quality of *Amaranthus* species in Morthern Guinea Savanna zone of Nigeria. *Russian Journal of Agricultural and Socio-Economic Sciences*. Issue 9(141). doi10.18551/rjoas.2023-09.06
- Enujeke, E.C. 2013. Effects of Poultry Manure on Growth and Yield of Improved Maize in Asaba Area of Delta State. *IOSR Journal of Agriculture and Veterinary Science*. e-ISSN: 2319-2380, p-ISSN: 2319-2372.
- FAO, (2021): Food and Agricultural Organisation; Agricultural Water Management| Land and water.
- Farmerline, (2023): The impact of fertilizers on the environment: Inorganic vs. Organic.
- Fasina, A. S.,Awe, G.O.and Aruleba, J. O. (2015): Irrigation suitability evaluation and crop yield an example with *Amaranthus cruentus* in Southwestern Nigeria. *International Journal of Irrigation and Water Management*. Vol. 2 (9), pp. 001-006
- Fasinmirin, J.T and Olufayo, A.A. (2009). Yield and Water Use Efficiency of Jute Mallow *Corchorus Olitorius* under Varying Soil Water Management Strategies. *Journal of Medicinal Plants Research* Vol. 3(4), pp. 186-191 <http://www.academicjournals.org/JMPR>.
- Fasinmirin, J. T. Olufayo, A. A. Oguntunde, P. G. and Oguntuase, A. M. (2009): Parametrizing Simple Model between Yield and Evapotranspiration for *Amaranthus cruentus* under Drip and Sprinkler Irrigations. *International Journal of Plant Production* 3 (1) pp 75-90
- Geraldo, C and Henrique, P. S (2023). Plant water relations: Absorption, Transport and Control Mechanisms. *Advances in Selected Plant Physiology Aspects*.pp108-132
- Huai,W.L, Chao, C.H, Bo,C.C, James J and Liou, H (2021). Building a grey-based multi-criteria decision-making model for offshore wind farm site selection. *Sustainable Energy Technologies and Assessments* (43). <https://doi.org/10.1016/j.seta.2020.100935>
- Hussain, A., Bashir, H., Zafar, S., Rehman, R.,Khalid, M., Awais, M., Sadiq, M., Amjad, I., (2023). The importance of soil organic matter (SOM) on soil productivity and plant growth. *Biol. Agricult. Sci. Res. J.* 2023 (1), 11-11.
- Irfan E. Tayfun O. Abdullah O. R. and Gulin Z. O. (2016). Grey Relational Analysis Approach In Academic Performance Comparison Of University: A Case Study Of Turkish Universities. *European Scientific Journal*. ISSN: 1857 – 7881 (Print). e - ISSN 1857- 7431.
- Lagos State Government. (2018). Agricultural sector deal book 2018. Lagos State Government.
- Lampteym S (2022) Agronomic practices in soil water management for sustainable crop production under rain fed agriculture of drylands in sub-Sahara Africa. *Afr J Agric Res* 18:18–26. <https://doi.org/10.5897/AJAR2021.15822>
- [Li, B.](#), [Zhang, S.](#), [Li, W.](#) and [Zhang, Y.](#) (2022), "Application progress of grey model technology in agricultural science", [Grey Systems: Theory and Application](#), Vol. 12 No. 4, pp. 744-784. <https://doi.org/10.1108/GS-05-2022-0045>
- Liang, L., M.D. Schwartz, and S. Fei. (2012). Photographic Assessment of Temperate Forest Understory Phenology in Relation to Springtime Meteorological Drivers. *Int. J. Biometeorol.* 56: 343– 355. doi:<https://doi.org/10.1007/s00484-011-0438-1>
- Luca I, Rodney B. T, Fernandez-Fernandezd, M. D, Stefania D. P , Alberto P, Cecilia S , Youssef R (2020); Irrigation management of European greenhouse vegetable crops. *Agricultural Water Management*. <https://doi.org/10.1016/j.agwat.2020.106393>

- Luka, G.L. Brahim U and Rabo M.(2023); Performance of amaranth (*amaranthus cruentus*) as influenced by NPK fertilizer and organic liquid fertilizer at Samaru, Northern Guinea Savanna of Nigeria. FUDMA Journal of Sciences. doi.org/10.33003/fjs-2023-0704-1915
- Mary, K, Miranda, M and Tom, S (2022): Environmental implications of excess fertilizer and manure on water quality. North Dakota State University Extension Service publications. www.ndsu.edu/extension
- Ndung'u, M., Ngatia, L.W., R.N. Onwonga, M.W. Mucheru-Muna, R. Fu, D.N. Moriasi, and K.F. Ngetich. (2021). The influence of organic and inorganic nutrient inputs on soil organic carbon functional groups content and maize yields. *Heliyon* 7. <https://doi.org/10.1016/j.heliyon.2021.e07881>
- Oduniyi O.S and Chagwiza C (2021) Impact of adoption of sustainable land management practices on food security of smallholder farmers in Mpumalanga province of South Africa. *GeoJournal*. <https://doi.org/10.1007/s10708-021-10497-0>
- Ogunlela, A.O and Sadiku, I.B.S. (2017). Yield and water use efficiency of *Amaranthus cruentus* grown under sms-based irrigation system. *Journal of Research in Forestry, Wildlife & Environment* Vol. 9(3)pp 47-57
- Ogwu, M.C (2020) Value of *amaranthus* [L.] species in Nigeria. <http://dx.doi.org/10.5772/intechopen.86990>
- Olufayo, A.A., Baldy, C. and Ruelle, P. (1996). Sorghum Yield, Water Use and Canopy Temperatures under Different Levels of Irrigation. *Agric. Water Manage.* 30, 77–90.
- Oluwagbayide, S.D, Fasanu,O and Oloruntade, A.J (2021). Evaluation of Sprinkler Irrigation Evaporation Losses in Ilaro, Ogun State, South Western Nigeria. *Turkish Journal of Agricultural Engineering Research. (TURKAGER)*, 2(1): 47-59.<https://doi.org/10.46592/turkager.2021.v02i01.004>
- Odebiri, O, Onesimo M, John O, Rob S, Paramu M, Romano, L, Rowan N, Trylee, N. M and Mthembeni, M (2024). Mapping sub-surface distribution of soil organic carbon stocks in South Africa's arid and semi-arid landscapes: Implications for land management and climate change mitigation. *Geoderma Regional* 37 e00817 <https://doi.org/10.1016/j.geodrs.2024.e00817>
- Oworu O. O., Dada O. A., Majekodunmi O. E. (2010). Influence of Compost on Growth, Nutrient Uptake and Dry Matter Partitioning of Grain Amaranths (*Amaranthus hypochondriacus* L.). *Libyan Agriculture Research Center Journal International* 1(6): 375-383.
- Ozlu, E and Kumar, S. (2018). Response of soil organic carbon, ph, electrical conductivity, and water stable aggregates to long-term annual manure and inorganic fertilizer. *Soil Science Society of America Journal*. Volume 82, issue 5 <https://doi.org/10.2136/sssaj2018.02.0082>
- Paruelo, J.M., W.K. Lauenroth, and P.A. Roset. (2000). Technical note: Estimating Aboveground Plant Biomass Using A Photo-Graphic Technique. *J. Range Manage.* 53: 190 – 193. doi:<https://doi.org/10.2307/4003281>
- Rashmi, I, Trisha Roy, , Kartika, K.S, Rama P, Vassanda, C, Kala, S and Shinoji, K.C (2020): Organic and inorganic fertilizer contaminants in agriculture: Impact on soil and water resources; Contaminants in Agriculture. Springer Nature doi: [10.1007/978-3-030-41552-5_1](https://doi.org/10.1007/978-3-030-41552-5_1)
- Raveendra, K.I, Vijar, P.S and Alka, U. (2017). Planning and Evaluation of Irrigation Projects, Methods and implementation. *Irrigation Methods - ScienceDirect*. Pages 353-363
- Ramadevi, S. Sivaranjani ,S. Vijaya S.S. Ramabhai, V. Arulnangai R and Murugesan R (2023). Comparative study of organic manure (cow dung, goat dung and chicken manure) on the vegetative growth of *Amaranthus dubius* L. *An International Journal on Agricultural and Biological Research. AGBIR* Vol.39 No.4
- Roberto, C and Roberto, B (2022): Plants and water in a changing world: a physiological and ecological perspective. *Rendiconti Lincei. Scienze Fisiche e Naturali* 33:479–487
- Romero-Gelvez, J. I Beltrán-Fernández, S. V Aristizabal, A. J Zapata, S and Castañeda, M. (2020). Precision Agriculture Technology Evaluation using Combined AHP and GRA for Data Acquisition in Apiculture. Workshops at the Third International Conference on Applied Informatics 2020, October 29–31, 2020, Ota, Nigeria

- Shivakumar, M.R. and Murali K. P (2024); Multi-response optimization of reinforcement parameters of aluminum alloy composites by Taguchi method and grey relational analysis; *Heliyon* (10) <https://doi.org/10.1016/j.heliyon.2024.e30183>
- Soil Survey Staff, (2006). *Keys to Soil Taxonomy*, ninth ed. NRCS, USDA
- Sonja, J. Drazen, B. and Luka, C. (2015). Application of compressed cold air cooling: achieving multiple performance characteristics in end milling process. *Journal of Cleaner Production* 100 (2015) 325-332. [www. http://dx.doi.org/10.1016/j.jclepro.2015.03.095](http://dx.doi.org/10.1016/j.jclepro.2015.03.095)
- Sokoto, M.B. and Victor, O.(2017); Growth and yield of amaranth (*Amaranthus* spp.) as influenced by seed rate and method of planting in Sokoto, Nigeria. *Archives of Agriculture and Environmental Science* 2 (1): 29-35
- Sophie F.F, Alison M. H, Markus L, Gifty E. A, Ermias A, Asmeret, A. B, Stephan, M. H, Steve P. M, Keith D. S, Andrew. M. S, Johan, S, Erick K. Towett, S. E. Trumbore, T.G. Vågen, E W, Leigh, A. W and Sebastian D (2021): Continental-scale controls on soil organic carbon across sub-Saharan Africa. *SOIL*, 7, 305–332. doi.org/10.5194/soil-7-305-2021
- Ufoegbune G.C, Adebisi, G.A and Adekunle A.A (2016). Determination of water use of three vegetables; amaranthus (*amaranthus cruentus*), jute mallow (*corchorus olitorius*) and celosia (*celosia argentea*) at Abeokuta, Nigeria. *Journal of Environmental and Analytical Toxicology*. 6:3. <http://dx.doi.org/10.4172/2161-0525.1000374>
- Umakanta, S, Motaher, H and Shinya, O (2020): Nutritional and antioxidant components and antioxidant capacity in green morph *Amaranthus* leafy vegetable. *Scientific Reports*. 10:1336. doi.org/10.1038/s41598-020-57687-3
- Umakanta, S, Shinya, O, Riaz, U, Ahmed, B, Sezai, E, Sona, S, Anna, A, Magdalena, Z and Jiri, M (2024): Nutritional and bioactive properties and antioxidant potential of *Amaranthus tricolor*, *A. lividus*, *A. viridis*, and *A. spinosus* leafy vegetables. *Heliyon* 10 e30453 <https://doi.org/10.1016/j.heliyon.2024.e30453>
- Xiang Y, Li Y, Luo X, Liu Y, Yue X, Yao B, Xue J, Zhang L, Fan J, Xu X and Li Y (2022) Manure properties, soil conditions and managerial factors regulate greenhouse vegetable yield with organic fertilizer application across China. *Front. Plant Sci*. 13:1009631. doi: 10.3389/fpls.2022.1009631
- Xie W M, Ma Y, Li S J, Zhang S S, Ruan, L, Yang M Y, Shi W M, Zhang H L, Zhang L M. (2020). Optimizing soil dissolved organic matter extraction by grey relational analysis. *Pedosphere*. 30(5): 589–596.
- Xu, D (2024). ‘Lagrange-enhanced GRA Framework for probabilistic simplified neutrosophic multiple-attributes decision-making and applications to painting teaching quality evaluation’. *Journal of Intelligent and Fuzzy Systems*, vol. 46, no. 1, pp. 2147-2159, 2024: 2147 – 2159.
- Yahia, E. M., García-Solís, P, and Celis, M. E. M. (2019). Contribution of fruits and vegetables to human nutrition and health,” in *Postharvest physiology and biochemistry of fruits and vegetables*. Ed. Yahia, E. M. (Woodhead Publishing), 19–45.
- Zingore, S., and K.E. Giller. (2012). Optimizing phosphorus and manure application in maize soybean rotations in Zimbabwe. *Better Crops*. 96: 23–25.

RESEARCH ARTICLE

Effects of Pre-Sowing Treatments on Seed Germination and Seedling Growth Attributes of the Endangered (*Anisoptera scaphula* Roxb.) Species

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Abstract

Anisoptera scaphula (Boilam), one of Bangladesh's tallest native tree species, plays a vital role in the Asian forest ecosystem. It is critically endangered and at high risk of extinction. This research, conducted from April 2022 to February 2023, examined the effects of pre-sowing treatments on seed germination and seedling growth. Seeds collected from a mother tree in Cox's Bazar were subjected to four treatments: untreated (T0), soaked in water for 12h (T1), soaked and air-dried for 12h (T2), and soaked and air-dried for 24h (T3). Results showed T2 had the highest germination rate (84.2%) and fastest germination, while T0 had the lowest (52.3%). Seedlings from T2 also exhibited the greatest height (46.4 cm) and collar diameter (0.76 cm). Around 2000 seedlings are now ready for planting. These findings support large-scale seed production and regeneration efforts to mitigate the extinction risk of *A. scaphula* and restore its ecological role.

Keywords: *Anisoptera scaphula*; Seed germination; Pre-sowing treatments; DMRT; Biodiversity conservation; Endangered

Introduction

Bangladesh is a subtropical monsoon climatic region located between 20°34' and 26°38' North latitude and 88°01' and 92°41' East longitude (Alam et al., 2023; Miah et al., 2023; Roy et al., 2015). It is bordered by India on its western, northern, and eastern sides, Myanmar to the southeast, and the Bay of Bengal to the south. The climate is humid and warm, influenced by pre-monsoon, monsoon, and post-monsoon circulations, experiencing six seasons annually (Cord et al., 2009; Masum et al., 2022; Mukul et al., 2017; Nazrul et al., 2021; South et al., 2013). January is the coolest month with average temperatures around 26°C (78°F), while April is the warmest, with temperatures ranging from 33°C to 36°C (91°F to 96°F). Most rainfall occurs during the monsoon season (June-September) and is minimal in winter (November-February), making it one of the wettest climates in the world. Most areas receive over 1,525 mm of rain annually, with regions near the hills receiving up to 5,080 mm (Benito et al., 2013; Islam et al., 2021; Tanu et al., 2021; Yudaputra, 2021). The high precipitation and hot, humid temperatures support continuous high forests in Chattogram, the Chattogram Hill Tract, and Cox's Bazar, rich in biodiversity. Bangladesh covers a total area of 147,630 sq. km (57,000 sq. miles), with forest land constituting

about 14.47% (BBS, 2022), although only 8-10% has good canopy cover. The forests, primarily located in Chattogram, the Chattogram Hill Tract, Cox's Bazar, and Sylhet, comprise a wide range of timber species.

A total of 224 timber-yielding plant species grows in Bangladesh. Important species include sal (*Shorea robusta*), gamar (*Gmelina arborea*), telsur (*Hopea odorata*), boilam (*Anisoptera scaphula*), jarul (*Lagestroemia speciosa*), teak (*Tectona grandis*), garjan (*Dipterocarpus turbinatus*), and sundari (*Heritiera fomes*) (Bangladesh Bureau of Statistics, 1996). *Anisoptera* is a genus under the Dipterocarpaceae family (Dutta & Hossain, 2016; Miah et al., 2023; Rahman et al., 2016; Reza & Perry, 2015), consisting of ten species distributed from Chattogram (Bangladesh) to New Guinea. Eight of these species are listed in the IUCN Red List, with four critically endangered and the others classified as endangered. *Anisoptera scaphula*, critically endangered and native to Bangladesh, occurs in small patches in the forested areas of Chattogram, the Chattogram Hill Tract, and Cox's Bazar. This species also thrives in Malaysia, Myanmar, Thailand, and Vietnam.

Dipterocarpaceae, a family of 16 genera and about 695 known species (Dutta & Hossain, 2016; Głowacka & Flis-Olszewska, 2022; Rahman et al., 2010; Miah et al., 2023; Rahman et al., 2016; Reza & Perry, 2015; Uddin et al., 2020), includes *A. scaphula*, one of the tallest tree species in Bangladesh, reaching 30-45 m in height with a diameter of 1.0-1.5 m. *A. scaphula* is a resinous tree with grey bark, thick simple leaves, and terminal racemes. Its flowers are sub-globose, and the fruiting calyx tube is constricted at the mouth, with two large calyx lobes. It is an evergreen species, partially flowering from December to January, with fruits ripening during April-May. Native to Bangladesh, Myanmar, and Thailand, it is found in low altitudes and deep valleys in seasonal forests (Chowdhury et al., 2019; de Souza Valente et al., 2020; Dutta & Hossain, 2016; Głowacka & Flis-Olszewska, 2022; Rahman et al., 2010; Majumdar et al., 2014; Miah et al., 2023; Mohd-Taib et al., 2020; Rahman et al., 2016, 2017; Reza & Perry, 2015; Scherer et al., 2021; Uddin et al., 2020), tending to be gregarious in more seasonal forests.

A. scaphula is a flagship species in Bangladesh. It plays a dominant role in Asian forest ecology, providing shelter for avian fauna and orchids. Its timber is hard, rigid, durable, and suitable for various construction purposes. The wood is easy to work with and takes polish well, extensively used for general light construction, furniture, boats, and plywood. A small amount of resin is obtained from the tree, used locally for torches and caulking boats (Hossain et al., 2006; Kalanzi & Mwanja, 2023; Ojha et al., 2009; Ray et al., 2018). However, biodiversity in Bangladesh is depleting due to ineffective implementation of biodiversity safeguards in political and market systems. *A. scaphula*, despite being a flagship species, is on the verge of extinction and serves as an ambassador for conservation efforts. Scattered plantations are raised in the Chittagong Forest Division and Chittagong University campus, with 38 Boilam trees identified. Natural regeneration is hindered by the lack of mother trees and seeds, ground vegetation competition, and bad seed production. The seeds are recalcitrant (Chowdhury et al., 2023), complicating collection and storage. Techniques for seed germination need development for successful artificial regeneration. Pre-sowing treatments and proper germination techniques are crucial for effective plantation programs.

The study aimed to evaluate the germination potential of *A. scaphula* for large-scale plantations, determining the effects of pre-sowing treatments (Devi & Sharma, 2014; Hossain et al., 2006; Kalanzi & Mwanja, 2023; Ojha et al., 2009; Ray et al., 2018; Sahoo et al., 2020), and investigating the initial growth performance of seedlings under control and different treatments. These objectives aim to optimize seedling development and enhance plantation efforts.

Materials and methods

Description of the study area

The study was conducted at the nursery of the Institute of Forestry and Environmental Sciences, University of Chittagong, Bangladesh, over one year from April 2022 to February 2023. The nursery is situated in an area with about 60% hilly terrain, with elevations ranging from 14 to 87 meters above mean sea level (Mia et al., 2015). The soil in this region is characterized as moderately coarse to fine-textured sandy loam to sandy clay loam, with moderate to strong acidity and poor fertility. Specifically, the soil has a pH of 5.5, organic matter content of 2.0%, base saturation percentage (BSP) of less than 40%, and cation exchange capacity (CEC) of less than 10 me/100g (Mehta et al., 2021). The nursery benefits from a tropical monsoon climate, marked by hot, humid summers and mild, dry winters. The region receives an average annual rainfall of 2,500 to 3,000 mm, with the majority occurring between June and September. The mean monthly minimum temperature is 21.24°C. May, the warmest month, has an average daily high temperature of 33°C, with maximum temperatures reaching up to 29.75°C. The period from May 2 to May 24, 2015, was notably warm. July experiences the highest percentage of days with recorded precipitation, around 84%. In contrast, May typically records the highest temperatures, averaging 32.60°C, with a minimum temperature of 14.10°C (El-Keltawi & Abdel-Rahman, 2010; Sahoo et al., 2020). This climate and soil profile provides a specific environmental context for the nursery, which plays a crucial role in understanding and managing the conditions under which seed germination and seedling growth are studied.

Seed collection

Seeds of *Anisoptera scaphula* were collected from a mother tree in Saplapur, Teknaf Upazila, Cox's Bazar, with GPS coordinates used to precisely locate the source (El-Kassaby et al., 2008; Lúcio & Sari, 2017; Ranal et al., 2009; Sultana et al., 2021) (Figure 1).

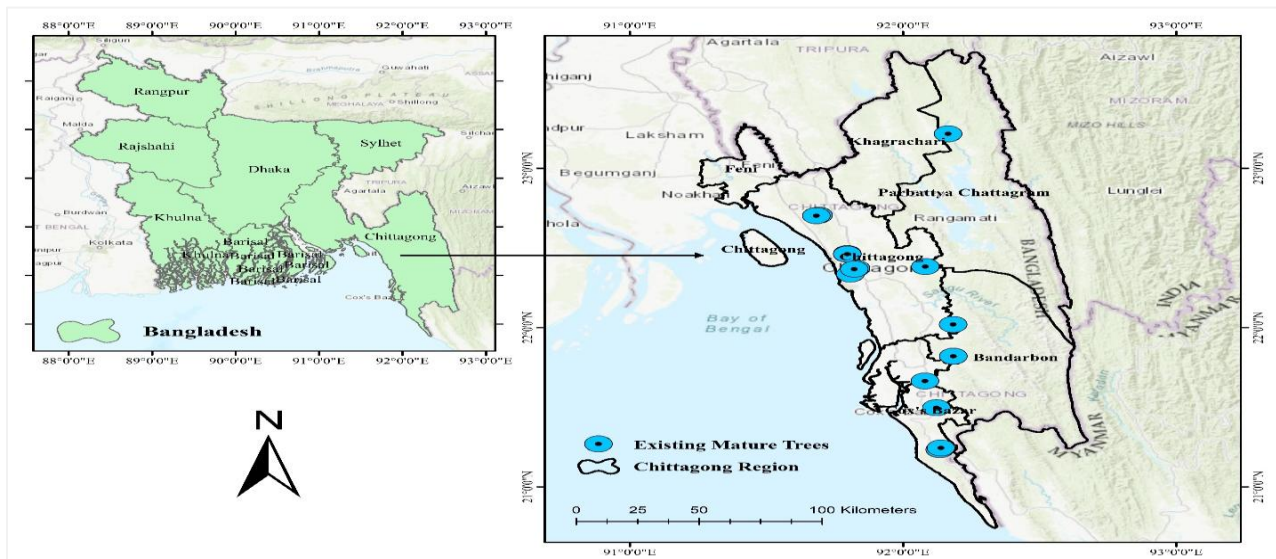


Figure 1. The map shows the geographic location of the recorded only few existing mature tree species of *A. scaphula* all over Bangladesh.

Seed purity was assessed by measuring the weight of pure seeds in the sample and calculating the percentage of pure seed. To ensure consistency in germination trials, freshly collected seeds of uniform size were selected, as seed size is positively correlated with germination success and seedling vigor (Chowdhury et al., 2024). The trials involved dewinged seeds subjected to four pre-sowing treatments: no treatment (T0), soaking in water for 12 hours (T1), soaking in water for 12 hours followed by 12 hours of air drying (T2), and soaking in water for 12 hours followed by 24 hours of air drying (T3). Each treatment was applied to 300 seeds per replicate, with three replicates per treatment, totaling 3,600 seeds. The seeds were sown inverted in polybags filled with a mixture of forest soil and decomposed cow dung at the IFESCU nursery (Sultana et al., 2021), following the methodology described by Hossain et al. (2014). This approach aimed to evaluate the effects of different pre-sowing treatments on seedling growth parameters such as germination rate, height, and collar diameter. The findings are expected to provide insights into optimizing seedling development and enhance reforestation and conservation efforts.

Statistical analysis

Germination percentage was estimated using the following formula,

Germination percentage (%) = germinated seeds /total seeds sown $\times$ 100

Mean germination time is a measure of the rate and spread of germination over time (Ranal et al., 2009). To analyze the significant differences among control and treatments, mean germination time, biomass of seedlings were tested with Duncan's multiple range test at 95% confidence intervals.

Results and discussions

Seed germination performance

Germination percentage of Boilam (*A. scaphula*) seeds was significantly affected by pre-sowing treatments. The germination percentage of dewinged Boilam seeds sown invertedly in polybags (Hossain et al., 2014) varied from 52.3% to 84.2% among the treatments. The highest germination percentage (84.2%) was obtained in T2 (Dewinged seeds soaked in water for 12 hours followed by air drying for another 12 hours), followed by 74.7% in T1 (Dewinged seeds soaked in water for 12 hours). The lowest seed germination percentage (52.3%) was observed in T0 (control; dewinged seeds without any treatment) (Figure 2). In a similar study on *Scaphula glabra*, Laurie (1940) reported 66% germination when seeds were sown superficially at 0.64 cm depth invertedly, i.e., with the apex downward and the stalk end upward. Hossain et al. (2014) reported a maximum germination percentage (65%) when dewinged *A. scaphula* seeds were sown invertedly (apex, wing side down) without soaking in water. Therefore, it has been shown clearly that the germination percentage of Boilam seeds significantly enhanced with soaking in water followed by air drying for 12 hours (Hossain et al., 2020, 2020). Expanding on these findings, several factors can be identified as influential in the successful germination of Boilam seeds. Pre-sowing treatments such as soaking in water and air drying appear to activate certain physiological and biochemical processes within the seeds (Dwivedi & Chopra, 2014). Soaking seeds in water likely facilitates the softening of the seed coat, allowing for better water absorption and subsequent activation of metabolic pathways essential for germination. This process, followed by air drying, could be contributing to the stabilization of the seed's internal moisture content, which is crucial for maintaining the viability of the seeds during the initial stages of germination (Hossain et al., 2018). Additionally, the practice of dewinging seeds, as noted in the study, removes the seed's natural dispersal apparatus, which might otherwise act as a physical barrier

to water uptake (Golev et al., 2020; Wallertz et al., 2018; Ward et al., 2000). By removing the wings, the seeds are more uniformly exposed to the soaking treatment, leading to more consistent germination results (Figure 3). The inversion of seeds during sowing, with the apex downward and the stalk end upward, is another critical factor. This orientation may promote more efficient penetration of water into the seed and facilitate the emergence of the radicle, the first part of the seedling to emerge during germination. Comparing these findings with Laurie's study on *Scaphula glabra*, it is evident that seed orientation and sowing depth also play a significant role in germination success (Golev et al., 2020; Johansson et al., 2013; Lashley et al., 2017; Mickaël et al., 2007a; Wallertz et al., 2018; Ward et al., 2000). Sowing seeds at a shallow depth of 0.64 cm ensures that the seeds remain close to the soil surface, where they can receive adequate moisture and oxygen, both of which are essential for germination. The inverted orientation aligns with the natural tendency of the radicle to grow downward, potentially reducing the energy expenditure required for the seedling to establish itself.

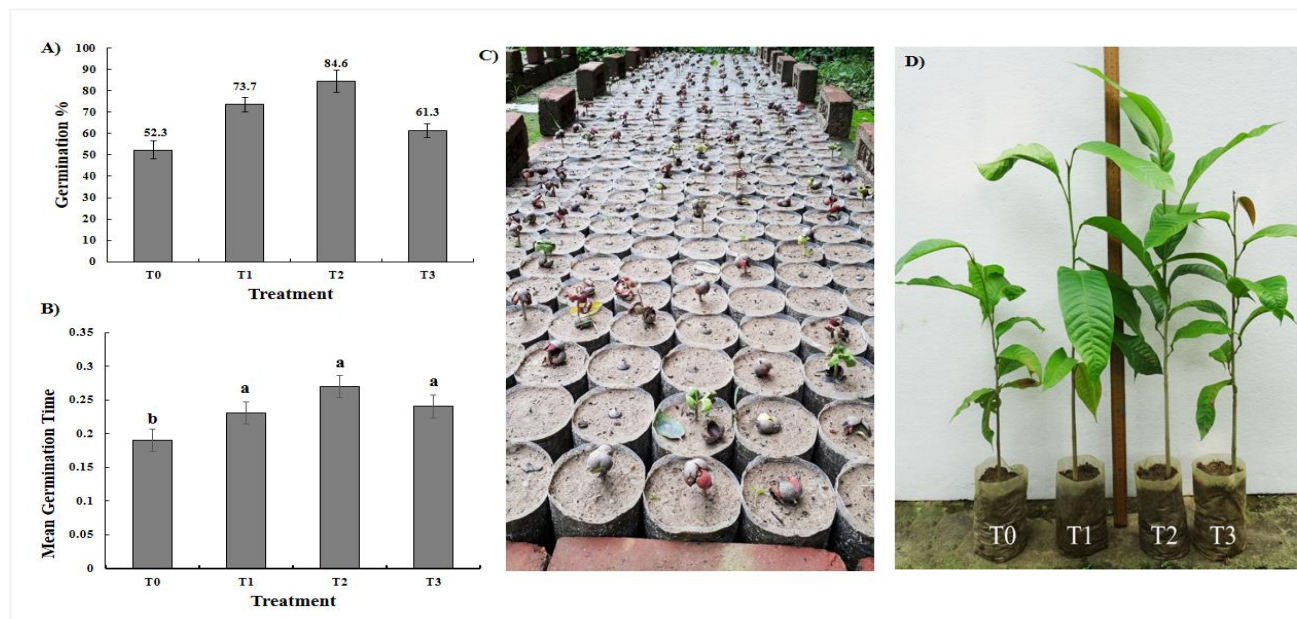


Figure 2. Effects of various pre-sowing treatments on germination potentials of the seeds of *A. scaphula* are shown in Figure 2A and 2B, the error bar indicates the standard error of means, and different alphabets (a, b) in the bar chart (Figure 2B) indicate the significant differences among different methods of pre-sowing treatment of *A. scaphula*. Figure (2D) shows the different physiological conditions of *A. scaphula* at the sapling stage, where, T0 is controlled, T1 is the pre-sowing treatment where soaking of seeds in water for approximately 12 hours, T2 is the soaking of seeds in water for 12 hours followed by air drying for 12h, and T3 is the soaking of seeds in water for 12 hours followed by air drying for 24 hours.

Moreover, the data suggests that the absence of pre-sowing treatments results in significantly lower germination percentages. The control group in the study, which did not receive any pre-sowing treatment, exhibited the lowest germination rate of 52.3%. This stark contrast highlights the importance of pre-sowing treatments in enhancing germination efficiency (Cuesta et al., 2010; Häggström et al., 2021; McKee et al., 1984).

In practical applications, these findings can be utilized to improve the propagation techniques for Boilam seeds, especially in reforestation and conservation projects where high germination rates are critical for the successful establishment of plant populations. Implementing pre-sowing treatments such as soaking and air drying can significantly increase the germination success of Boilam seeds, thereby ensuring a more robust and resilient population of this critically endangered species (Garnett et al., 2004; Parker et al., 2004, 2006).



Figure 3. Physiological condition and growth performance of *A. scaphula* seedlings from various pre-sowing treatment (T0, T1, T2, T3), where, seedling from T0 is control, seedling from T1 is soaking of seeds in water for 12 hours, seedling in T2 is soaking of seeds in water for 12 hours followed by air drying for 12 hours, and seedling from T3 is soaking of seeds in water for 12 hours followed by air drying for 24 hours.

Overall, the study underscores the significance of carefully considered pre-sowing treatments and seed-handling techniques in maximizing the germination potential of Boilam seeds. By adopting these methods, forestry practitioners and conservationists can enhance the success rates of their planting efforts, contributing to the preservation and restoration of Boilam populations in their natural habitats (Häggström et al., 2021). This research adds valuable knowledge to the field of seed science and offers practical solutions for improving seed germination outcomes in forestry and conservation contexts.

Seed germination period

Dewinged seeds sown invertedly were found to start germination one week after sowing them in the polybag and continued for up to 20 days (Parker et al., 2006). The fastest germination (least imbibition period; 6 days) was observed in T2 (seeds soaked in water for 12h followed by air drying for 12 h) and T1 (seeds soaked in water for 12h) and delayed (highest imbibition period, 20 days) was observed in T0 (control). Germination started with the development of a radicle from the winged side of the seed. Highest mean germination time (0.27) was found in T2, and lowest Mean Germination Time was in T0. There is no significant difference at $p < 0.005$, Duncan's Multiple Range Test (DMRT) (Figure 2).

Seedling Growth Performance

Seedling growth parameters such as height and collar diameter were significantly influenced by the seed treatments. The maximum average height (46.4 cm) of the seedlings was observed in T2, followed by 37.9 cm in T1, with the lowest height (28.8 cm) recorded in T0 (control). Similarly, the collar diameter of the seedlings was also greatest in T2 (0.76 cm), followed by T1 (0.64 cm), and the smallest diameter was found in T0 (0.54 cm) (Figure 4). These results suggest that the pre-sowing treatment involving soaking seeds in water for 12 hours followed by air drying for another 12 hours (T2) is highly effective in promoting seedling growth. This treatment appears to provide an optimal balance of moisture absorption and subsequent drying (Dwivedi & Chopra, 2014; Kuniyal et al., 2013), which may activate and stabilize the physiological processes necessary for robust seedling development. The increased height and collar diameter of seedlings in T2 indicate a healthier and potentially more resilient plant, which is crucial for successful establishment in the field (Mickaël et al., 2007b). The substantial difference in growth parameters between treated and untreated seeds (T0) highlights the importance of appropriate pre-sowing treatments in forestry practices. The untreated seeds (T0) produced seedlings with significantly lower growth metrics, which may affect their survival and competitiveness in natural settings (Figure 3) (Dey et al., 2022; King & He, 2005; Matilla, 2019; Van Assen et al., 2015). This underscores the value of implementing evidence-based pre-sowing treatments to enhance seedling performance and ensure the success of reforestation and conservation efforts.

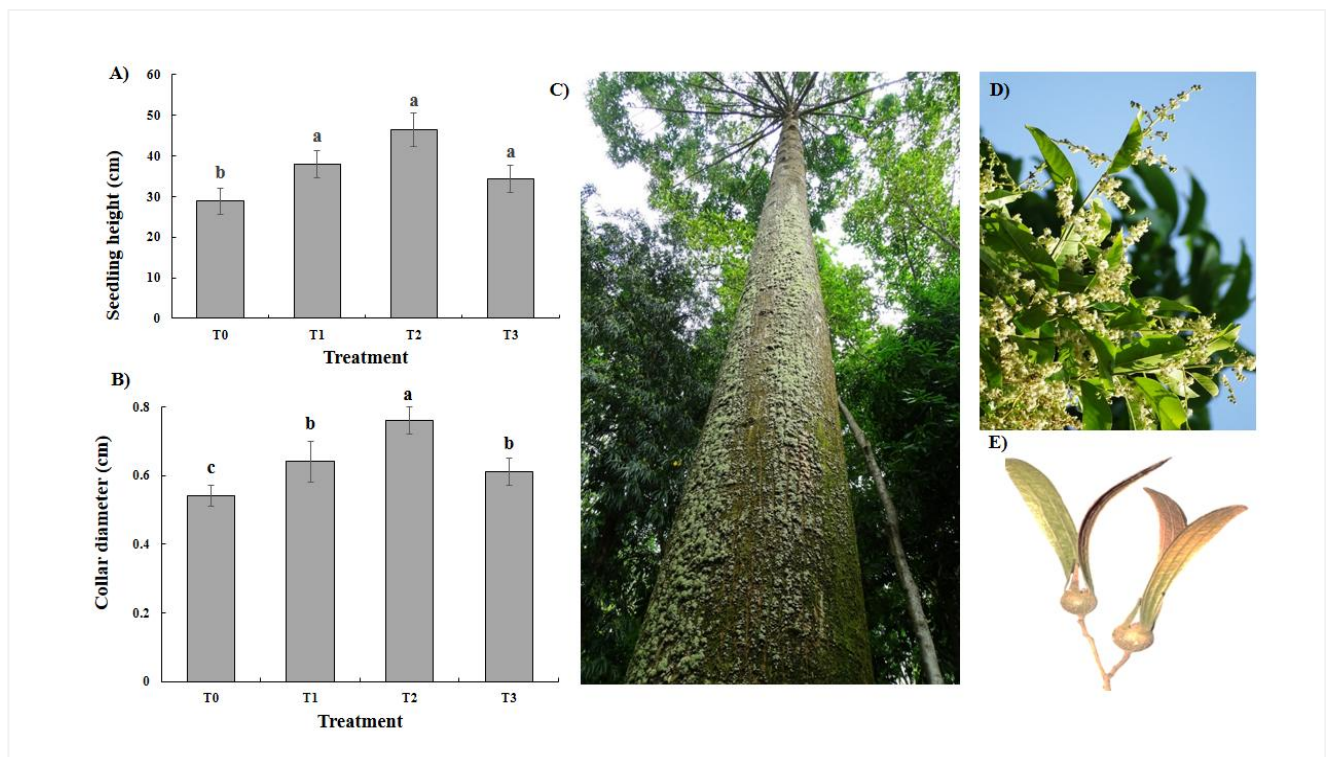


Figure 4. Height and diameter growth of Boilam seedlings develop through various pre-sowing treatment six months after sowing the seeds. T0: Control, T1: Soaking of seeds in water for 12h, T2: Soaking of seeds in water for 12h followed by air drying for 12h and T3: Soaking of seeds in water for 12h followed by air drying for 24h. Note: the same letter(s) indicates no significant difference at $p \leq 0.05$, according to DMRT. I (error bar) indicates the standard error of means. C) Mother tree, D) Flowers, E) Seeds of Boilam.

Consequently, approximately 2,000 Boilam seedlings are ready for planting in the next planting season (June-July 2023) as part of the germplasm establishment and restoration program. These seedlings, having undergone effective pre-sowing treatments, are expected to exhibit better growth and higher survival rates once planted. This initiative is a crucial step towards the conservation of the critically endangered Boilam species, contributing to the restoration of its natural habitats and the maintenance of biodiversity. In practical terms, the preparation of 2,000 seedlings demonstrates a significant scaling up of efforts to establish a sustainable population of Boilam trees (Hasnat et al., 2019; Moïse et al., 2005; Shinde & Chavan, 2017). The insights gained from this study on pre-sowing treatments can be applied to other species with similar ecological requirements, thereby broadening the impact of these findings on forest restoration practices. By ensuring that seedlings are well-prepared before planting, forestry managers can optimize resource use and improve the outcomes of their restoration projects. Overall, the data strongly support the use of specific pre-sowing treatments to enhance seedling growth parameters, such as height and collar diameter (Mariam & Alamgir, 2022). These treatments are not only beneficial for the immediate growth of the seedlings but also for their long-term survival and establishment in reforestation and conservation programs. The successful propagation and planting of Boilam seedlings will contribute to the genetic diversity and resilience of forest ecosystems, aiding in the overall goal of environmental sustainability and biodiversity conservation.

Biomass of seedlings

The study revealed that leaf fresh weight was heterogeneous, with Treatment 2 (T2) ($0.83 > p = 0.05$) significantly differing from Treatments T0, T1, and T3 ($\text{Alpha} < p = 0.05$). Homogeneity was observed in Treatment T0, which showed no significant difference with T2 ($0.83 > p = 0.05$).

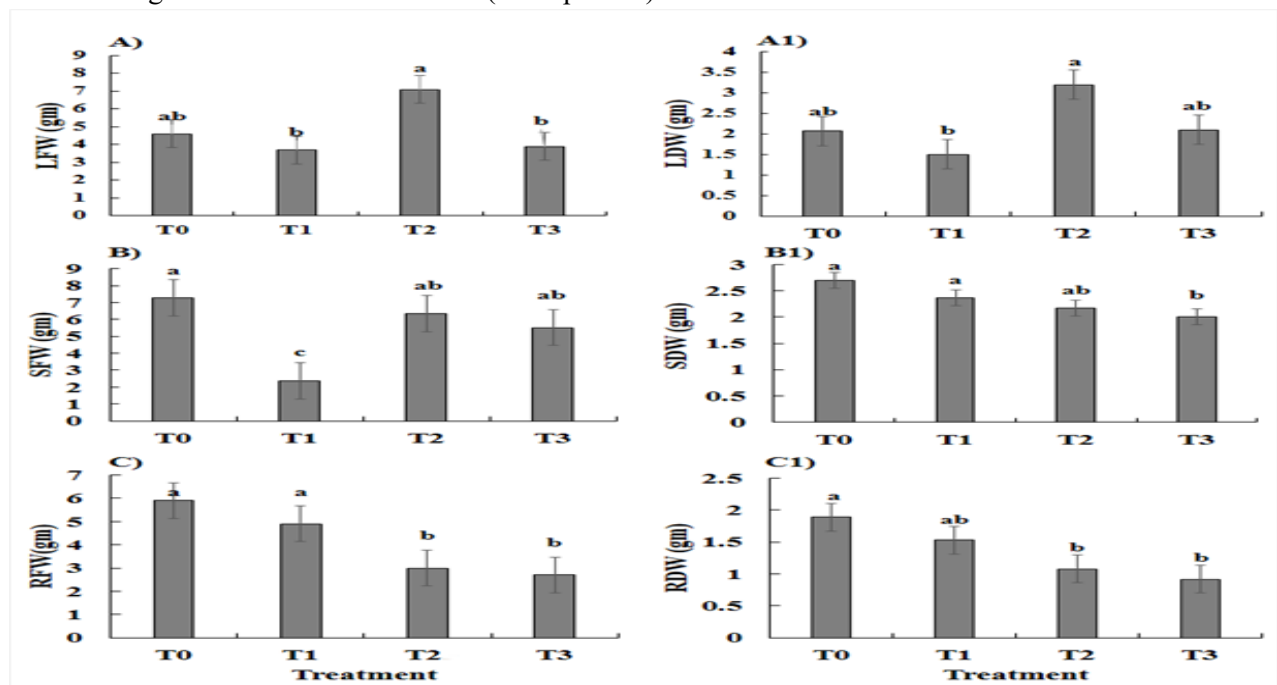


Figure 5. Effects of different pre-sowing treatments on Leaf, Shoot and Root fresh and Dry weight of *Anisoptera scaphula* seedling in Nursery, LFW(g)= Leaf Fresh Weight, LDW(g)= Leaf Dry Weight, SFW(g)= Shoot Fresh Weight, SDW(g)=Shoot Dry Weight, RFW(g)=Root Fresh Weight, RDW(g)=Root Dry Weight. Note: the same letter(s) indicates no significant difference at $p \leq 0.05$, according to DMRT. I (error bar) indicates the standard error of means.

The highest leaf dry weight was recorded in T2 (3.2g), significantly differing ($p < 0.005$) from T1, T0, and T3. For shoot fresh weight, the highest was recorded in T0 (7.28g), which significantly differed from T1, T2, and T3. Similarly, the highest shoot dry weight was recorded in T0 (2.70g) with no significant difference from T2, T1, and T3. Root fresh weight was highest in T0 (5.6g), differing significantly ($p < 0.005$) from Treatments T1, T2, and T3, and the highest root dry weight was also in T0 (1.89g), significantly differing ($p < 0.005$) from T1, T2, and T3 (Figure 5). Based on these findings, it is recommended to adopt T2 for leaf production due to its superior leaf dry weight, while T0 is suggested for maximizing root and shoot biomass, balancing the overall plant growth for optimal reforestation and conservation efforts (Lúcio & Sari, 2017; Ranal et al., 2009; Sultana et al., 2021).

Conclusion and Recommendations

The study investigated the germination performance and initial growth of seedlings, focusing on the effects of various pre-sowing treatments. The findings underscore that pre-sowing treatments significantly enhance seed germination and seedling growth, offering a cost-effective and time-efficient method for producing quality seedlings of the native tree species *Anisoptera scaphula*. The results showed that pre-sowing treatments notably influenced seed germination potential. Specifically, dewinged seeds sown invertedly began germination one week after sowing and continued for up to 20 days. The fastest germination, with an imbibition period of six days, was observed in T2 (seeds soaked in water for 12 hours followed by air drying for 12 hours) and T1 (seeds soaked in water for 12 hours). In contrast, the control group (T0) exhibited the slowest germination, with an imbibition period of 20 days. The germination percentage was highest in T2 at 84.2%, followed by T1 at 74.7%, with the lowest germination percentage of 52.3% in T0. Furthermore, the mean maximum height of seedlings was recorded as 46.4 cm in T2, 37.9 cm in T1, and 28.8 cm in T0. Similarly, the collar diameter of seedlings was greatest in T2 at 0.76 cm, followed by 0.64 cm in T1, and 0.54 cm in T0. T2 also exhibited the highest leaf fresh weight, while T0 had the highest root and shoot fresh and dry weight. The outcomes of this research have significant implications for mass-scale seed production and conservation efforts. By adopting the T2 treatment, the species stands a better chance of being removed from the endangered list and can potentially reestablish its dominance in the ecosystem, providing habitat for various other flora and fauna. Based on the study's findings, several recommendations can be made: Adoption of T2 Treatment for Large-Scale Plantations: The T2 treatment (seeds soaked in water for 12 hours followed by air drying for 12 hours) should be utilized for large-scale plantation efforts. This method has demonstrated the highest germination rates and optimal seedling growth, making it the most effective for conservation and reforestation projects.

Declaration

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Conflict of Interest: The authors declare that there are no conflicts of interest regarding the publication of this paper. Any potential conflicts have been disclosed to the journal and are detailed within the manuscript as required.

Ethics approval/declaration: Required ethics were followed.

Consent to participate: All the authors gave consent for participation.

Consent for publication: All the authors gave their consent for the paper to be published

Data Availability Statement: The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request, included within the article and its supplementary materials, etc. This statement ensures transparency and reproducibility of the research findings.

Authors' Contribution: Author 1: Atia Hoque Subah, Data collection, data curation, formal analysis, investigation, methodology, writing original draft. Author 2: Md. Aktar Hossain, conceptualization and supervision, validation. Author 3: Mohd Imran Hossain Chowdhury, research design, project administration, validation, manuscript writing. Author 4: Sumya Asma Cynthia, formal analysis, data visualization, data collection, manuscript editing and writing. Author 5: Mehedi Hasan Rakib, manuscript writing and editing." All authors have read and agreed to the published version of the manuscript.

References

- Alam, E., Hridoy, A. E. E., Tusher, S. M. S. H., Islam, A. R. M. T., & Islam, M. K. (2023). Climate change in Bangladesh: Temperature and rainfall climatology of Bangladesh for 1949-2013 and its implication on rice yield. *PLoS ONE*, 18(10 October), 1–26. <https://doi.org/10.1371/journal.pone.0292668>
- Benito, B., Cayuela, L., & Albuquerque, F. (2013). The impact of modelling choices in the predictive performance of richness maps derived from species-distribution models: Guidelines to build better diversity models. *Methods in Ecology and Evolution*, 4. <https://doi.org/10.1111/2041-210x.12022>
- Chowdhury, M. A., Islam, K. N., Hafiz, N., & Islam, K. (2019). Diversity of trees in a community managed forest: the case of Komolchori VCF, Khagrachari, Bangladesh. *Geology, Ecology, and Landscapes*, 3(2), 95–103. <https://doi.org/10.1080/24749508.2018.1508980>
- Chowdhury, M.I.H., Rakib, M. H., & Das, C. (2023). Intra and Interspecific Relation of Tree Species Aspects of Seed Biometry , Phenology , Seed Dispersion , and Germination. *Journal of Agriculture Sustainability and Environment*, 2(2), 57–69. <https://doi.org/https://doi.org/10.56556/jase.v2i2.877>
- Chowdhury, M.I.H., Rakib, M. H., Das, C., & Hossain, Z. (2024). Tree Species Germination : A Comprehensive Meta-Analysis and its Implications for Pre-Sowing Treatment in Bangladesh. *Journal Of Soil, Plant and Environment.*, 3(1), 24–40.
- Cord, A., Colditz, R. R., Schmidt, M., & Dech, S. (2009). Species Distribution and Forest Type Mapping in Mexico Species Distribution and Forest Type Mapping in Mexico. February 2014. <https://doi.org/10.1109/IGARSS.2009.5417681>
- Cuesta, B., Vega, J., Villar-Salvador, P., & Rey-Benayas, J. M. (2010). Root growth dynamics of Aleppo pine (*Pinus halepensis* Mill.) seedlings in relation to shoot elongation, plant size and tissue nitrogen

- concentration. *Trees - Structure and Function*, 24(5), 899–908. <https://doi.org/10.1007/s00468-010-0459-0>
- de Souza Valente, C., Rodiles, A., Freire Marques, M. R., & Merrifield, D. L. (2020). White spot syndrome virus (WSSV) disturbs the intestinal microbiota of shrimp (*Penaeus vannamei*) reared in biofloc and clear seawater. *Applied Microbiology and Biotechnology*, 104(18), 8007–8023. <https://doi.org/10.1007/s00253-020-10816-4>
- Devi, W. S., & Sharma, G. J. (2014). In Vitro Propagation of *Arundinaria callosa* Munro- an Edible Bamboo from Nodal Explants of Mature Plants. *The Open Plant Science Journal*, 3(1), 35–39. <https://doi.org/10.2174/1874294700903010035>
- Dey, S., Hossain, M. K., Nandi, R., & Saifullah, M. (2022). Effect of Pre-sowing Treatments on Germination and Initial Growth of *Terminalia citrina*: A Medicinal Tree Species in Bangladesh. *Journal of Tropical Forestry and Environment*, 11(02). <https://doi.org/10.31357/jtfe.v11i02.5595>
- Dutta, S., & Hossain, M. K. (2016). Infestation of *imperata cylindrica* L. and its impacts on local communities in secondary forests of sitakunda botanical garden and eco-park, chittagong, Bangladesh. *International Journal of Conservation Science*, 7(1), 167–180.
- Dwivedi, S., & Chopra, D. (2014). Revisiting *terminalia arjuna*-an ancient cardiovascular drug. *Journal of Traditional and Complementary Medicine*, 4(4), 224–231. <https://doi.org/10.4103/2225-4110.139103>
- El-Keltawi, E.M.N., & Abdel-Rahman, S.A.S. (2010). Propagation of Bamboo (*Dendrocalamus giganteus*, Munro) Through Culm-Branch Cuttings in Egypt. *Assiut Journal of Agricultural Sciences*, 41(1), 1–22. <https://doi.org/10.21608/ajas.2010.267818>
- El-Kassaby, Y. A., Moss, I., Kolotelo, D., & Stoehr, M. (2008). Seed germination: Mathematical representation and parameters extraction. *Forest Science*, 54(2), 220–227.
- Garnett, E., Jonsson, L. M., Dighton, J., & Murnen, K. (2004). Control of pitch pine seed germination and initial growth exerted by leaf litters and polyphenolic compounds. *Biology and Fertility of Soils*, 40(6), 421–426. <https://doi.org/10.1007/s00374-004-0801-z>
- Głowacka, A., & Flis-Olszewska, E. (2022). The biodiversity of weed communities of dent maize, narrow-leaved lupin and oat in relation to cropping system and weed control. *Agronomy Science*, 77(3), 123–137. <https://doi.org/10.24326/as.2022.3.9>
- Golev, A. D., Bukhtoyarov, V. N., & Ivannikov, V. A. (2020). The practice of using a sieveless separator for pre-sowing treatment of common pine seeds (*Pinus sylvestris*). *IOP Conference Series: Earth and Environmental Science*, 595(1). <https://doi.org/10.1088/1755-1315/595/1/012056>
- Häggström, B., Domevscik, M., Öhlund, J., & Nordin, A. (2021). Survival and growth of Scots pine (*Pinus sylvestris*) seedlings in north Sweden: effects of planting position and arginine phosphate addition. *Scandinavian Journal of Forest Research*, 36(6), 423–433. <https://doi.org/10.1080/02827581.2021.1957999>
- Hasnat, G. N. T., Hossain, M. K., Alam, M. S., Bhuiyan, M. K., & Hossain, M. A. (2019). Germination and initial seedling growth performance of *Vitex peduncularis* Wall. ex Schauer – A threatened native tree species of Bangladesh. *Journal of Bioscience and Agriculture Research*, 20(2), 1700–1708. <https://doi.org/10.18801/jbar.200219.207>
- Hasnat, T., & Hossain, M. K. (2018). Pre-sowing treatment effects on gutgutiya (*Protium serratum*)- a threatened tree species of Bangladesh. *Horticulture International Journal*, 2(6), 352–356. <https://doi.org/10.15406/hij.2018.02.00076>

- Hossain, M. A., Jewel, M. E. U., Sen, M., & Serajuddoula, M. (2006). Rooting ability of *Bambusa vulgaris* var. *striata* branch cuttings as influenced by cutting type and rooting hormones. *Journal of Bamboo and Rattan*, 5(3–4), 117–126.
- Hossain, M. A., Kumar, S. M., Seca, G., Maheran, A. A., & Nor-Aini, A. S. (2018). Mass propagation of *dendrocalamus asper* by branch cutting. *Journal of Tropical Forest Science*, 30(1), 82–88. <https://doi.org/10.26525/jtfs2018.30.1.8288>
- Hossain, M. K., & Uddin, M. M. (2019). Effect of Pre-Sowing Treatments on Germination and Initial Seedling Growth of *Castanopsis Indica* - An Endangered Tree Species in. December. <https://doi.org/10.7747/JFES.2019.35.4.223>
- Hossain, M. K., Alim, A., Hossen, S., Hossain, A., & Rahman, A. (2020). Diversity and conservation status of tree species in Hazarikhil Wildlife Sanctuary (HWS) of Chittagong, Bangladesh. *Geology, Ecology, and Landscapes*, 4(4), 298–305. <https://doi.org/10.1080/24749508.2019.1694131>
- Islam, S., Shetu, S. S., Rahim, M. A., & Khan, S. A. (2021). Floristic composition and plant species diversity in and around the brickfield areas of Savar-Dhamrai region of Dhaka district. *Jahangirnagar University Journal of Biological Sciences*, 9(1–2), 91–108. <https://doi.org/10.3329/jujbs.v9i1-2.53710>
- Johansson, K., Ring, E., & Högbom, L. (2013). Effects of pre-harvest fertilization and subsequent soil scarification on the growth of planted *Pinus sylvestris* seedlings and ground vegetation after clear-felling. *Silva Fennica*, 47(4), 1–18. <https://doi.org/10.14214/sf.1016>
- Kalanzi, F., & Mwanja, C. K. (2023). Effect of nodal cutting position and plant growth regulator on bud sprouting of *Dendrocalamus giganteus* Wall. Ex Munro in Uganda. *Advances in Bamboo Science*, 2(January), 100016. <https://doi.org/10.1016/j.bamboo.2023.100016>
- King, W. R., & He, J. (2005). Understanding the Role and Methods of Meta-Analysis in IS Research. *Communications of the Association for Information Systems*, 16(March). <https://doi.org/10.17705/1cais.01632>
- Kuniyal, C. P., Purohit, V., Butola, J. S., & Sundriyal, R. C. (2013). South African Journal of Botany Seed size correlates seedling emergence in *Terminalia bellerica*. *South African Journal of Botany*, 87, 92–94. <https://doi.org/10.1016/j.sajb.2013.03.016>
- Lashley, M. A., Chitwood, M. C., Nanney, J. S., Deperno, C. S., & Moorman, C. E. (2017). Regenerating White Pine (*Pinus strobus*) in the South: Seedling Position is More Important than Herbivory Protection. *Castanea*, 82(2), 156–162. <https://doi.org/10.2179/17-138>
- Lúcio, A. D., & Sari, B. G. (2017). Planejamento e condução de experimentos e análise de dados experimentais em hortaliças: Problemas e soluções. *Horticultura Brasileira*, 35(3), 316–327. <https://doi.org/10.1590/S0102-053620170302>
- Majumdar, K., Shankar, U., & Datta, B. K. (2014). Trends in Tree Diversity and Stand Structure during Restoration: A Case Study in Fragmented Moist Deciduous Forest Ecosystems of Northeast India. *Journal of Ecosystems*, 2014, 1–10. <https://doi.org/10.1155/2014/845142>
- Mariam, H., & Alamgir, A. N. M. (2022). Flowering and Fruiting Behavior, Seed Germination, and Survival Status of *Heritiera fomes* Buch-Ham and *Excoecaria agallocha* L. Species in the Sundarban Mangrove Forest of Bangladesh. *Open Access Library Journal*, 09(12), 1–11. <https://doi.org/10.4236/oalib.1109506>
- Masum, S. M., Halim, A., Shamim, M., & Mandal, H. (2022). Predicting Current and Future Potential Distributions of *Parthenium hysterophorus* in Bangladesh Using Maximum Entropy Ecological Niche

- Predicting Current and Future Potential Distributions of *Parthenium hysterophorus* in Bangladesh Using Maximum Entropy Ec. June. <https://doi.org/10.3390/agronomy12071592>
- Matilla, A. J. (2019). Seed coat formation: Its evolution and regulation. *Seed Science Research*, 29(4), 215–226. <https://doi.org/10.1017/S0960258519000254>
- McKee, W. H., Hook, D. D., DeBell, D. S., & Askew, J. L. (1984). Growth and Nutrient Status of Loblolly Pine Seedlings in Relation to Flooding and Phosphorus. *Soil Science Society of America Journal*, 48(6), 1438–1442. <https://doi.org/10.2136/sssaj1984.03615995004800060046x>
- Mehta, B., Nagar, B., Patel, B., Chaklashiya, P., Shah, M., Verma, P., & Shah, M. B. (2021). A review on a Lesser Known Indian Mangrove: *Avicennia officinalis* L. (Family: Acanthaceae). *International Journal of Green Pharmacy*, 15(1), 1–10.
- Miah, M. D., Hasnat, G. N. T., Nath, B., Saeem, M. G. U., & Rahman, M. M. (2023). Spatial and Temporal Changes in the Urban Green Spaces and Land Surface Temperature in the Chittagong City Corporation of Bangladesh Between 2000 and 2020. *Forestist*, 73(2), 171–182. <https://doi.org/10.5152/forestist.2022.22013>
- Miah, R., Hasan, M., Parisha, J. T., & Sayok, A. K. (2023). A Framework on Biodiversity Conservation Related Policy Analysis. January. <https://doi.org/10.5923/j.ajee.20231301.01>
- Mickaël, H., Michaël, A., Fabrice, B., Pierre, M., & Thibaud, D. (2007a). Soil detritivore macro-invertebrate assemblages throughout a managed beech rotation. *Annals of Forest Science*, 64(March 2004), 219–228. <https://doi.org/10.1051/forest>
- Mickaël, H., Michaël, A., Fabrice, B., Pierre, M., & Thibaud, D. (2007b). Soil detritivore macro-invertebrate assemblages throughout a managed beech rotation. *Annals of Forest Science*, 64, 219–228. <https://doi.org/10.1051/forest>
- Mohd-Taib, F. S., Mohd-Saleh, W., Asyikha, R., Mansor, M. S., Ahmad-Mustapha, M., Mustafa-Bakray, N. A., Mod-Husin, S., Md-Shukor, A., Amat-Darbis, N. D., & Sulaiman, N. (2020). Effects of anthropogenic disturbance on the species assemblages of birds in the back mangrove forests. *Wetlands Ecology and Management*, 28(3), 479–494. <https://doi.org/10.1007/s11273-020-09726-z>
- Moïse, J. A., Han, S., Gudynaite-Savitch, L., Johnson, D. A., & Miki, B. L. A. (2005). Seed coats: Structure, development, composition, and biotechnology. *In Vitro Cellular and Developmental Biology - Plant*, 41(5), 620–644. <https://doi.org/10.1079/IVP2005686>
- Mukul, S. A., Biswas, S. R., & Rashid, A. Z. M. M. (2017). Biodiversity of Bangladesh. February, 1–15. <https://doi.org/10.20944/preprints201702.0045.v1>
- Nazrul, K., Rahman, L., Rana, S., Islam, K., & Hossain, S. (2021). Climate change and the distribution of two *Ficus* spp . in Bangladesh – predicting the spatial shifts Trees , Forests and People Climate change and the distribution of two *Ficus* spp . in Bangladesh – predicting the spatial shifts. March. <https://doi.org/10.1016/j.tfp.2021.100086>
- Ojha, A., Verma, N., & Kumar, A. (2009). In vitro micropropagation of economically important edible bamboo (*Dendrocalamus asper*) through somatic embryos from root, leaves and nodal segments explants. *Research on Crops*, 10(2), 430–436.
- Parker, W. C., Noland, T. L., & Morneau, A. E. (2004). Effect of seed mass on early seedling growth of five eastern white pine (*Pinus strobus* L.) families under contrasting light environments. *Canadian Journal of Botany*, 82(11), 1645–1655. <https://doi.org/10.1139/B04-129>
- Parker, W. C., Noland, T. L., & Morneau, A. E. (2006). The effects of seed mass on germination, seedling emergence, and early seedling growth of eastern white pine (*Pinus strobus* L.). *New Forests*, 32(1), 33–49. <https://doi.org/10.1007/s11056-005-3391-1>

- Rahman, M. M., Al Mahmud, M. A., Ahmed, F. U., & Deb, R. (2017). Developing alternative income generation activities reduces forest dependency of the poor and enhances their livelihoods: The case of the chunati wildlife sanctuary, Bangladesh. *Forests Trees and Livelihoods*, 26(4), 256–270. <https://doi.org/10.1080/14728028.2017.1320590>
- Rahman, M. M., Mahmud, M. A. Al, Shahidullah, M., Nath, T. K., & Jashimuddin, M. (2016). The competitiveness of the phytosociological attributes of the protected areas in Bangladesh with that in the other tropical countries. *Journal of Sustainable Forestry*, 35(6), 431–450. <https://doi.org/10.1080/10549811.2016.1202841>
- Rahman, M. H., Abu Sayed Arfin Khan, M., Jannatul F. M., & Roy, B. (2010). Status, Distribution and Diversity of Invasive Forest Undergrowth Species in the Tropics: a Study from Northeastern Bangladesh. *Journal of Forest Science*, 26(3), 149–159.
- Ranal, M. A., de Santana, D. G., Ferreira, W. R., & Mendes-Rodrigues, C. (2009). Calculating germination measurements and organizing spreadsheets. *Brazilian Journal of Botany*. 32(4), 849–855. <https://doi.org/10.1590/S0100-84042009000400022>
- Ray, S. S., Ali, M. N., Banerjee, M., & Yeasmin, L. (2018). In vitro and in vivo assessment of Thidiazuron mediated micro-clones of *Dendrocalamus asper*, an ornamental bamboo species. *J. Crop and Weed*, 14(1), 150–157.
- Reza, A. H. M. A., & Perry, G. (2015). Herpetofaunal species richness in the tropical forests of Bangladesh. *Asian Journal of Conservation Biology*, 4(2), 59.
- Roy, M., Biswas, B., & Ghosh, S. (2015). Trend Analysis of Climate Change in Chittagong Station in Bangladesh. *International Letters of Natural Sciences*, 47(1), 42–53. <https://doi.org/10.18052/www.scipress.com/ilns.47.42>
- Sahoo, S., Kar, R. K., & Mohanty, T. L. (2020). Effect of Plant Growth Hormones on Sprouting and Rooting Behaviour of Clones of Various Bamboo Species in coastal Odisha. *International Journal of Current Microbiology and Applied Sciences*, 9(7), 3521–3532. <https://doi.org/10.20546/ijcmas.2020.907.411>
- Scherer, L., De Laurentiis, V., Marques, A., Michelsen, O., Alejandre, E. M., Pfister, S., Rosa, F., & Rugani, B. (2021). Linking land use inventories to biodiversity impact assessment methods. *International Journal of Life Cycle Assessment*, 26(12), 2315–2320. <https://doi.org/10.1007/s11367-021-02003-y>
- Shinde, A. S., & Chavan, N. S. (2017). Effect of pre-sowing treatment on seed germination of *Ixora coccinea* L. *Current Botany*, 8(September). <https://doi.org/10.19071/cb.2017.v8.3270>
- South, C., Division, F., Hossain, M. A., Hossain, M. K., Salam, M. A., & Rahman, S. (2013). Composition and Diversity of Tree Species in Dudhpukuria-Dhopachori Wildlife Research Journal of Pharmaceutical , Biological and Chemical Sciences Composition and Diversity of Tree Species in Dudhpukuria-Dhopachori Wildlife Sanctuary of Chittagong (South. May.
- Sultana, N., Limon, S. H., Rahman, M. S., Akther, A., Salekin, S., Meason, D. F., & Bloomberg, M. (2021). Germination and growth responses to water stress of three agroforestry tree species from Bangladesh. *Environmental Challenges*, 5(August), 100256. <https://doi.org/10.1016/j.envc.2021.100256>
- Tanu, M. B., Barman, A. C., Siddique, M. F., Sku, S., Hossen, M. N., Rayhan, A., & Mahmud, Y. (2021). Determining of suitable size of paraffin image for image pearl production in freshwater mussel (*Lamellidens marginalis*) in Bangladesh. *International Journal of Fisheries and Aquatic Studies*, 9(6), 222–226. <https://doi.org/10.22271/fish.2021.v9.i6c.2605>
- Uddin, M., Chowdhury, F. I., & Hossain, M. K. (2020). Assessment of tree species diversity, composition and structure of Medha Kachhania National Park, Cox's Bazar, Bangladesh. *Asian Journal of Forestry*, 4(1), 15–21. <https://doi.org/10.13057/asianjfor/r040104>

- Van Assen, M. A. L. M., Van Aert, R. C. M., & Wicherts, J. M. (2015). Meta-analysis using effect size distributions of only statistically significant studies. *Psychological Methods*, 20(3), 293–309. <https://doi.org/10.1037/met0000025>
- Wallertz, K., Björklund, N., Hjelm, K., Petersson, M., & Sundblad, L. G. (2018). Comparison of different site preparation techniques: quality of planting spots, seedling growth and pine weevil damage. *New Forests*, 49(6), 705–722. <https://doi.org/10.1007/s11056-018-9634-8>
- Ward, J. S., Gent, M. P. N., & Stephens, G. R. (2000). Effects of planting stock quality and browse protection-type on height growth of northern red oak and eastern white pine. *Forest Ecology and Management*, 127(1–3), 205–216. [https://doi.org/10.1016/S0378-1127\(99\)00132-2](https://doi.org/10.1016/S0378-1127(99)00132-2)
- Yudaputra, A. (2021). Predicting habitat suitability of critically endangered *Nepenthes sumatrana*. *IOP Conference Series: Earth and Environmental Science*, 948, 12020. <https://doi.org/10.1088/1755-1315/948/1/012020>

RESEARCH ARTICLE

Optimum limestone rates for improved soil fertility and Potato (*Solanum tuberosum* L.) yield in two agroecological zones of Cameroon

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Abstract

Potato (*Solanum tuberosum* L.) is a high demanding crop in terms of nutrient requirements. In Cameroon soil fertility is poorly managed and lead to very low yield. This study estimates the limestone rate required for sustainable soil management and improved potato production in two main potatoes producing agroecological zones (AEZ). Two field experiments carried out in two villages namely Wassande (Guinean high savannah zone (AEZII)) and Bangou (Western Highlands zone (AEZ III)) consisted to plant potato genotypes Dosa and Jacob2005 on a split plot design including unfertilized, NPK-fertilized and NPK+manure-fertilized soils in which limestone was applied 4 weeks before, at 1, 2, 3, 4 and 5 t. ha⁻¹ rates. Data were collected on soil physico-chemical properties and potato yield. Limestone rate increased soil pH (hydrogen potential), organic matter (MO) content, cation exchangeable capacity (CEC) as well as sum of exchangeable bases (SEB), and decreased the Al (aluminum) content. P(phosphorus) and Ca (calcium) content should be increased with limestone rate but decreased at 5 t. ha⁻¹. Potato yield increased with limestone rate on unfertilized soil in both sites. On fertilized soils, the optimum limestone rate was 3 t. ha⁻¹ in Wassande and 4 t. ha⁻¹ in Bangou. There was no significant difference between NPK and NPK+manure fertilized soils at higher limestone rates (3-5 t. ha⁻¹) in Wassande, whereas in Bangou, the highest yield was obtained with NPK+manure. Genotype Dosa performed better than Jacob2005 in Wassande but less in Bangou than Jacob2005. The best performance was obtained with 3 t. ha⁻¹ of limestone in Wassande and 4 t. ha⁻¹ in Bangou.

Keywords: Soil amendment; Potato yield; Limestone rate; Soil fertility; Cameroon

Introduction

Potato (*Solanum tuberosum* L.) is one of the most widely grown vegetable crops in the world (Zhang et al., 2017). Due to its high content in vitamins and minerals (Ducharme, 2016), potato is considered as a vital crop for food security worldwide (Devaux et al., 2020). It is the world's leading non-cereal foodstuff. Global production of this commodity was estimated at 370.4 million tons in 2019 (FAO, 2020). In Cameroon, potato production contributes to improve the livelihood of farmers. It is grown mainly in three of the five agro-ecological zones of the country including: the Sudano-Sahelian zone, the western highlands and the Guinean high savannah zone. Despite its nutritional and socio-economic importance, potato yield in Cameroon is low (9 t.ha⁻¹), with a yield gap of 60% between the current yield and the potential yield of 25 t.ha⁻¹ (Yengoh and Ardo, 2014). The low productivity of agricultural sector in many parts of Sub-Saharan Africa is largely attributed to low soil fertility. Low soil fertility is related to acid rain, erosion, continuous cropping, soil acidity, organic matter depletion and deterioration of

soil biological properties (Nduwumuremyi et al., 2013 ; Getachew, 2019). In Cameroon, most of the soils have a high leaching potential. This leaching is accompanied by nutrient deficiencies (N, P and K) and soil acidification (Djoukeng et al., 2014). In order to increase soil fertility, the use of chemical fertilizers is generally recommended, sometimes in combination with manure (Djoukeng et al., 2016). However, recent studies showed that, the use of chemical fertilizers alone even at higher rates do not improve yield when organic matter (OM) is low (Oshunsanya, 2017). Chemical fertilizers contribute to a decline of soil fertility and crop yields through the destruction of soil microflora and microfauna, as well as the accumulation of Al^{3+} (aluminum ions) ions and nitrates (Oshunsanya, 2017). Chemical fertilizers act on the clay-humus complex, resulting in the desaturation of exchangeable Ca into phytotoxic minerals (Tsozué et al., 2015 ; Tamfuh et al., 2019). According to Brady and Weil (2004)), accumulation of Al^{3+} and H^+ (hydrogen ions) inhibits plant root development and the absorption of essential nutrients. This accumulation gradually becomes toxic and can reduce yields by 3 to 67% (Opala et al., 2015 ; Bekele et al., 2018). Demographic pressure on land has reduced fallow time, exposing soils to erosion and nutrient depletion (Getachew et al., 2014). However, the combination of NPK and manure improves crop yields. Žydelis et al. (2019) showed that the combined use of manure and mineral fertilizer increases maize yield compared to the application of organic or mineral fertilizer alone. According to Žydelis et al. (2019), manure has great potential for improving soil nutrient availability.

Solutions have been proposed to manage soil acidity and improve soil fertility; this includes addition of dolomite or limestone to the soil in order to reduce acidity (Basak et al., 2016), reducing Al and H toxicity, hence improving soil pH as well as releasing nutrients retained in the soil colloidal system (Nduwumuremyi et al., 2013). Calcium released by the applied lime forms rigid bonds with the pectic chains and favors the resistance of plant cell walls to enzymatic degradation from pathogens (Fageria and Baligar, 2008). Charles et al. (1992) reported that in Rwanda, application of limestone doses of up to 4.5 t.ha^{-1} on acid soils resulted in significant increase of wheat, bean and potato production. (Tamfuh et al., 2019) used dolomite in the highlands of Cameroon, to improve the yield of green bean (*Phaseolus vulgaris*). There are many sources of lime (wood ash, dolomite and calcium carbonate) in Cameroon. Cameroon's soils are highly acidic, with a pH below that required for potato cultivation ($6 < \text{pH} < 6.5$). This acidic character contributes to low potato yields. The addition of limestone corrects the pH and therefore breaks the bonds created between nutrients, making them available for plant nutrition and optimal potato production. Calcium carbonate is frequently used by farmers to restore and maintain soil quality. However, each farmer applies limestone at its own rate and this is not efficient for high crop production. There is a need to optimize limestone rate tailored on AEZ for efficient use and high potato production. The aim of this study was to determine the limestone rate required for sustainable soil management and improved potato production under different fertilization types in two agroecological zones of Cameroon.

Material and methods

Study sites

Two field experiments were conducted in Cameroon under different environmental conditions to study the effect of limestone rate on potato genotypes. Experiment 1 was carried out in Wassande and experiment 2 in Bangou. All the experiments were carried out from April to July 2021. The experiments were repeated on different plots during the same period in 2022 in order to appreciate the effect of the limestone that occurs in the previous year. Wassande is a village in the Guinean high savannah agro-ecological zone (AEZ II), located between latitudes $7^{\circ}03'$ and $7^{\circ}32'$ North and longitudes $13^{\circ}20'$ and $13^{\circ}54'$ East (Sy et al., 2008). The Guinean high savannah zone is characterized by a humid tropical Sudanian climate, with a monomodal rainfall pattern (Sy et al., 2008).

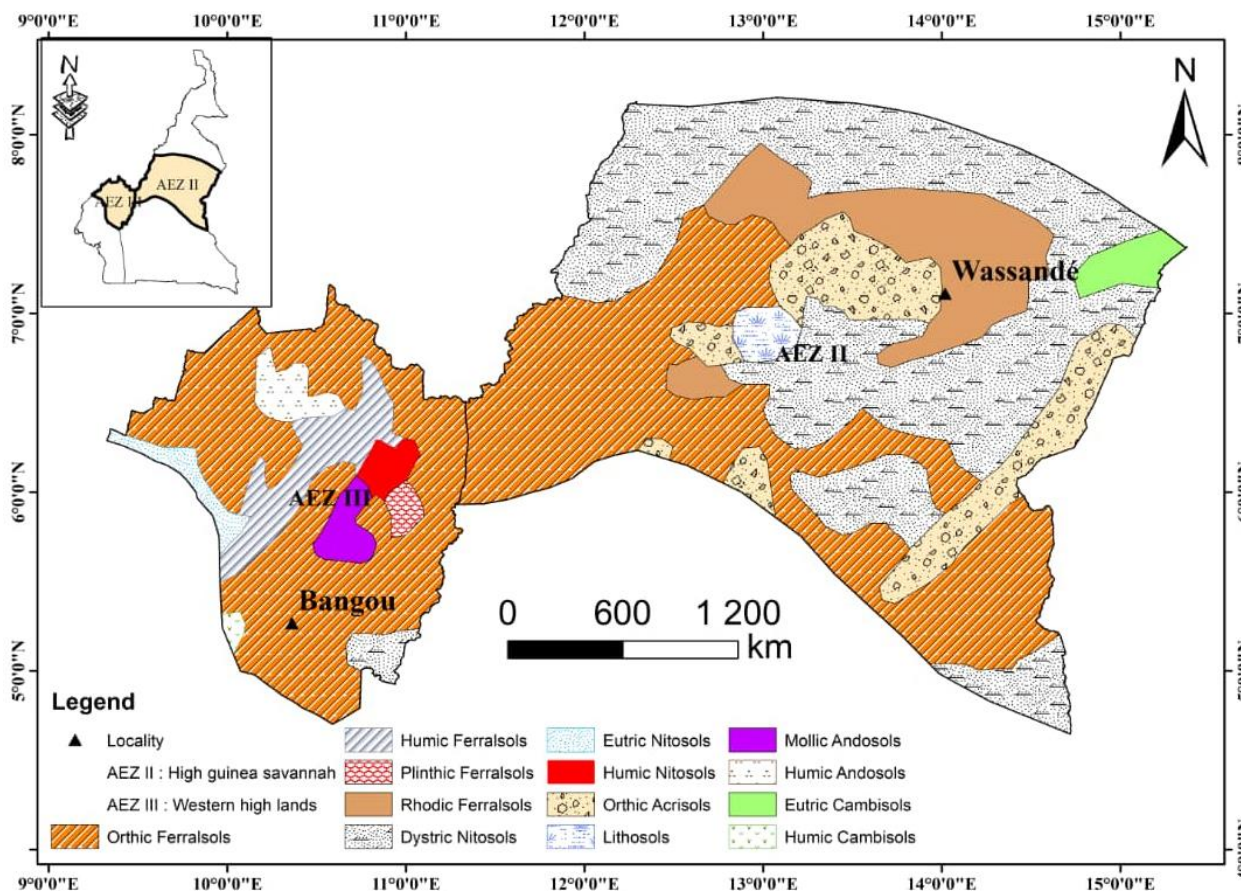


Figure 1. Presentation of the study sites

Average rainfall is around 1.500 mm. The average temperature is 22°C. The following soil types are encountered: ferralsols, lithosols, acrisols and cambisols (Ngachie, 1992), ferralsols and acrisols are found in Wassandé (Figure 1). Bangou is a village of Western Highlands agro-ecological zone, located between latitudes 4°54" and 6°36" North and longitudes 9°18" and 11°24" East. The Western Highlands zone is characterized by a "Cameroonian highland" climate, exhibiting low average temperatures (19°C), with abundant rainfall (1500-2000 mm) following a monomodal pattern. The landscape is characterized by medium-sized mountains, savannah vegetation, depressed basins and plains crossed by gallery forests. The following soil types are encountered: ferralsols, andosols, nitosols and cambisols (Ngachie, 1992), Bangou shows ferralsols (Figure 1).

Plant material

Plant material consisted of healthy potato (*Solanum tuberosum* L.) tubers of similar physiological age provided by the TOWA structure of seed production. Two genotypes Dosa and Jacob2005 were used.

Analysis of the soil physicochemical characteristics

Soil samples were collected at 20 cm depth for analysis before limestone application and 16 weeks after. After air-drying and crushing, samples were analyzed using the international methods recommended by (Pauwels et al., 1992).

General aspects of the field experiments

The experimental fields were cleared, ploughed and ridged. For each experimental site, a field of 2.10 m x 57.5 m was divided into 18 plots of 2.10 m x 2.25 m replicated three times. Limestone was applied 4 weeks before planting (Fageria and Baligar, 2008) at different rates 1, 2, 3, 4 or 5 t.ha⁻¹ for each soil fertilization type including unfertilized, NPK-fertilized and NPK+manure-fertilized soils. Non-limed plots of each fertilization type were considered as control plots. Potato seeds were planted in a split-plot design, where soil fertilization type + limestone rate was the plot and potato genotype, the subplot giving a total of 36 experimental units [(3 soil fertilization types) x (5 limestone rates + 1 control) x 2 genotypes] with 3 replicates. Limestone applied had the following characteristics: 8.1 pH_{H2O}; 40 meq/100g cation exchange capacity (CEC); 22.8 meq/100g Ca content; 6.16 meq/100g Mg content and 29.26 exchangeable bases (SEB). Seeds of each potato genotype were planted into each subplot measuring 2.10 m x 2.25 m. Each subplot consisted of 4 rows; spacing were maintained as 75 cm between rows and 30 cm between plants of the same row, giving 8 stems per row and 32 stems per subplot. Each plot and block were separated from each other by a buffer zone of 1.5 m. Fertilization was done at planting time using 400 kg.ha⁻¹ NPK (12 6 20) alone or supplemented with 5 t.ha⁻¹ chicken manure (MINADER, 2008; Sanon and Ouattara, 2019). Plot management consisted of weeding the field, hilling the plants and controlling diseases. Fungicide (Mancozeb 80 wp + Cymoxanil (6 %) + Chlorothalonil (30 %)) was sprayed at one-week intervals using the manufacturer recommended rates, to control late blight. Cypermethrin (360 g.l⁻¹) was sprayed at two-week intervals to control insects.

Data collection and statistical analysis

In each experimental unit, data were collected on 10 plants selected in the center rows following the "W" pattern on 2 yield parameters including the total number of tubers per plant, and the yield of marketable tubers (larger than 30 mm). Analysis of variance (ANOVA) was used to compare means of the different treatments for each parameter. Means were separated using Fisher's LSD (Low Significant Different) test when the ANOVA test showed significant probability ($p < 0.05$) of difference between means. All analyses were performed using GenStat Edition 12 software (VNS International, 2012).

Results

Data of both first and second seasons were merged as there was no significant difference for season with regard to soil characteristics and yield parameters ($p > 0.05$).

Physico-chemical characteristics of the soil before liming

The quantities of sand, silt and clay contained in the soils are 33%, 35% and 32% in Wassande and 43.5%, 24.5% and 32.0% in Bangou respectively. On the basis of these quantities and according to the USDA (United States Department of Agriculture) textural triangle, soil has a silty-clay texture in Wassande and a sandy-clay texture in Bangou. With a 4.9 pH_{water}, 3.8 pH_{KCl} in Wassande and a 5.2 pH_{water}, 4.4 pH_{KCl} in Bangou, these soils are acidic. OM is high in Wassande (3.1 %) and low in Bangou (1.2 %). Nitrogen content is moderate in Wassande (1.33 g/kg) and Bangou (1.54 g/kg); Ca and Mg are low in Wassande (2.4 meq/100g and 1.8 meq/100g respectively) and higher in Bangou (6.7 meq/100g and 3.60 meq/100g respectively), P content is very low in Wassande (1.7 mg/kg) and low in Bangou (9.7 mg/kg). Aluminum content is high in Wassande (1.4 mg/kg) than in Bangou (0.2 mg/kg), K (potassium) content is low in both sites (between 0.1 and 0.3 meq/100g). Sodium content is moderate in Wassande (0.7 meq/100g) and low in Bangou (0.2 meq/100g). The SEB is moderate in Wassande (5.1) and

high in Bangou (10.8) and the CEC is high in Wassande (31.6 meq/100g) and moderate and in Bangou (21.6 meq/100g).

Soil chemical characteristics after liming

Table 1 shows the chemical properties of different soil types as affected by limestone rate in both sites. On unfertilized soil, the pH increased with limestone rate. Higher limestone rates contributed to higher N content. P, Ca and K content increased with the limestone rate, reached a pick at 3 and 4 t. ha⁻¹ of limestone in Wassande and Bangou respectively and started decreasing at higher rates. Na content increased with 1 t. ha⁻¹ of limestone and formed a plateau at higher rates. The same trend was observed on fertilized soils for P and Ca content. However, K content reached the pick at 2 and 3 t. ha⁻¹ on NPK+manure fertilized soil. Mg increased with limestone rate in both sites, but with higher slopes in Wassande. This trend was observed on fertilized soils. Al content decreased with limestone rate and reached 0.0 at 3 and 5 t. ha⁻¹ in Wassande and Bangou respectively. This same trend was observed on fertilized soils, with slight difference in Bangou where Al content reach 0.0 at 3 t. ha⁻¹ on NPK+manure fertilized soil. CEC increased with limestone rate at both sites for all soil fertilization types.

Table 1. Soil chemical properties of the different soil types as affected by limestone rate at both sites

Soil fertilizations types	Parameters	Limestone rate (t/ha) by locality											
		Wassande						Bangou					
		0	1	2	3	4	5	0	1	2	3	4	5
Unfertilized	pH _{water}	4.9	5.7	6.2	6.4	6.6	6.9	5.2	5.5	5.9	6.5	6.5	6.7
	pH _{kcl}	3.8	5.1	5.7	5.8	5.9	6.1	4.4	4.4	4.9	5.1	5.8	6.1
	CO (%)	1.79	2.49	3.38	2.95	2.41	2.41	1.10	2.37	2.54	3.31	3.03	2.76
	MO (%)	3.1	4.3	5.1	5.8	4.2	4.2	1.2	4.1	4.4	4.8	5.2	5.7
	CEC (meq/100g)	31.6	31.2	47	61	68	65	21.6	29.2	30.3	34	46.8	32.8
	N (g/kg)	1.33	1.34	1.36	1.37	1.43	2.45	1.54	1.79	2.10	2.14	2.32	2.52
	P (mg/kg)	1.7	3.1	5.5	10.3	6.3	4.6	9.7	10.9	11.8	13	9.9	9.6
	Al (mg/kg)	1.41	0.1	0.02	0	0	0	0.2	0.19	0.07	0.03	0.01	0
	C/N	14	18	22	18	17	13	7	17	10	13	10	16
	Ca (meq/100g)	2.4	15.4	27.2	28.8	28.5	27.6	6.7	15.7	19	23	25.7	18.2
	Mg (meq/100g)	1.8	4.6	4.9	8	8.1	10	3.6	4.7	5	5	5.5	6
	K (meq/100g)	0.2	0.4	0.4	0.5	0.6	0.5	0.3	0.7	0.7	1.9	3.3	1
	Na (meq/100g)	0.7	0.8	0.9	0.9	0.8	0.8	0.2	1.1	0.9	0.7	1.1	0.7
	SEB	5.1	21.2	33.4	38.2	38	39	10.8	22.2	25.6	30.7	35.6	25.9
NPK fertilized	pH _{water}	4.8	5.4	6.2	6.3	6.6	6.9	4.9	5.4	5.6	6.3	6.6	6.8
	pH _{kcl}	4.1	4.5	5.5	5.6	5.7	5.8	3.9	4.6	4.6	4.7	5.9	6.3
	CO (%)	1.32	1.63	2.33	2.72	2.49	2.17	2.21	2.76	3.26	3.59	3.53	3.31
	MO (%)	2.8	2.3	4	4.7	4.3	3.8	3.8	4.8	5.6	6.2	6.1	5.7
	CEC (meq/100g)	28.2	35	50	54	62.3	67	24	26.4	32	34	42.6	43.6
	N (g/kg)	1.03	1.25	1.32	1.49	1.56	1.71	1.67	1.79	2.00	2.14	2.35	2.95
	P (mg/kg)	7.5	10.3	11.8	12.7	9.8	7.5	13	15.3	15.7	19	18.7	15.1
	Al (mg/kg)	1.3	0.7	0.3	0	0	0	1.5	0.7	0.4	0.05	0.01	0
	C/N	16	11	23	8	19	17	12	12	11	18	21	15
	Ca (meq/100g)	3.4	13.6	25.8	26.2	28.2	26.6	7.1	10.8	16.2	27	27.1	13.8
	Mg (meq/100g)	3.1	3.1	4.2	5.4	7	8.7	4	4	4.9	5	6	7.2
	K (meq/100g)	0.3	0.4	0.5	0.6	0.4	0.4	0.3	1.3	1.4	1.4	1.5	0.8
	Na (meq/100g)	0.2	0.4	0.7	0.9	1	0.9	0.2	0.8	0.9	0.7	0.9	0.6
	SEB	7	17.5	31.1	33.1	36.6	36.6	11.6	16.9	23.4	34.1	35.5	22.4
NPK+manure fertilized	pH _{water}	5.2	6.2	6.4	6.4	6.5	6.8	4.8	5.6	5.7	6.4	6.6	6.6
	pH _{kcl}	4.6	5.6	5.8	5.9	6.1	6.2	4	4.7	4.7	5.7	5.9	6.1
	CO (%)	1.94	2.25	2.56	3.34	3.18	2.87	2.48	2.98	3.86	3.64	3.92	3.70

MO (%)	3.4	3.9	4.4	5	5.8	5.5	4.3	5.1	6.7	6.3	6.8	6.4
CEC (meq/100g)	30	40.7	52	58	62	60.1	29.6	33.6	39	44	49.6	50
N (g/kg)	1.50	1.62	1.60	1.77	1.78	2.16	1.65	1.96	2.38	2.80	3.00	3.12
P (mg/kg)	16.5	25.3	29.4	43.6	41.2	31.3	17.8	32.1	37.5	41	36.2	34.2
Al (mg/kg)	1	0.4	0.1	0	0	0	0.9	0.3	0.17	0	0	0
C/N	17	21	13	13	20	11	13	13	14	12	13	22
Ca (meq/100g)	5.8	19.4	27.3	27.8	27.4	27.1	10.6	11.3	13	20	26.2	18.6
Mg (meq/100g)	4.6	6.8	8.2	8.5	10.9	11.7	2.6	2.9	3.9	5.3	5.4	8.2
K (meq/100g)	0.5	0.5	0.8	0.8	0.5	0.5	0.5	1.1	1.9	5.2	4.5	4.3
Na (meq/100g)	0.9	0.9	1	1	1.2	1.1	0.8	1	1.1	1.3	1.4	1.2
SEB	11.7	27.6	37.3	38	40	40.4	14.5	16.3	19.9	31.8	37.5	32.3

Legend: CO, organic carbon; MO, organic matter; CEC, cation exchangeable capacity; N, Nitrogen; P, Phosphorus; Al, aluminum; C/N, carbon nitrogen ratio; Ca, calcium; Mg, magnesium; K, Potassium, Na, sodium, SEB, sum of exchangeable bases.

Influence of limestone rate on potato yield parameters

The yield parameters evaluated were the number of tubers and the marketable yield. Table 2 shows the mean of square values of potato yield parameters on different soil fertilization types as affected by limestone rate and genotype in Wassande and Bangou. Limestone rates and soil fertilization type affected both yield parameters at both sites. Genotype also influenced marketable yield at both sites. This factor also influenced the number of tubers in Wassande. There was no significant influence of genotype on the number of tubers in Bangou. Interaction between limestone rate and genotypes for all parameters in both sites was not significant. Similarly, there was no interaction between limestone rate and Soil fertilization types for the number of tubers in both sites. However, interaction effect was observed between limestone rate and soil fertilization types for the marketable yield at both sites. Interaction effect was observed between genotypes and soil fertilization types for the marketable yield at Wassande but not at Bangou.

Table 2. Mean squares values of potato yield parameters on different soil fertilization types as affected by limestone rate and genotype in both experimental sites

Source of variance	Df	Number of tubers		Marketable yield	
		Wassande	Bangou	Wassande	Bangou
Limestone rate (L)	5	13.69***	28.16***	137.58***	211.43***
Genotype (G)	1	22.59***	5.07	178***	83.9***
Soil fertilization type (SFT)	2	75.77***	267.01***	3398.64***	8602.93***
LxG	5	1.01	1.88	6.20	5.90
LxSFT	10	0.33	0.99	16.37*	16.46*
GxSFT	2	2.05	4.35	19.06*	10.71

Note:***: Difference is significant ($p < 0.001$); *: Difference is significant ($p < 0.05$)

Number of tubers

Figure 2 shows the influence of the limestone rate on the number of tubers on different soil types. In Wassande, the number of tubers increased with limestone rate on all fertilization types; it reached higher values (6.80, 8.45 and 9.35 tubers on unfertilized, NPK-fertilized and NPK+manure-fertilized soils respectively) at 4 t. ha⁻¹, and form a plateau at higher rate (Figure 2a). This parameter also increased in Bangou with limestone rate on all fertilization types and reached a peak (5.70, 8.87 and 12.33 tubers on unfertilized, NPK-fertilized and

NPK+manure-fertilized soils respectively) at 4 t. ha⁻¹ (Figure 2b). The highest number of tubers was obtained on NPK+manure fertilized soil, followed by NPK-fertilized in both sites.

With regards to the effect of genotype on the number of potato tubers, Dosa produced more tubers (7.71) than Jacob2005 (6.80) in Wassande (Figure 3a). There was no significant difference between both genotypes in Bangou (Figure 3b).

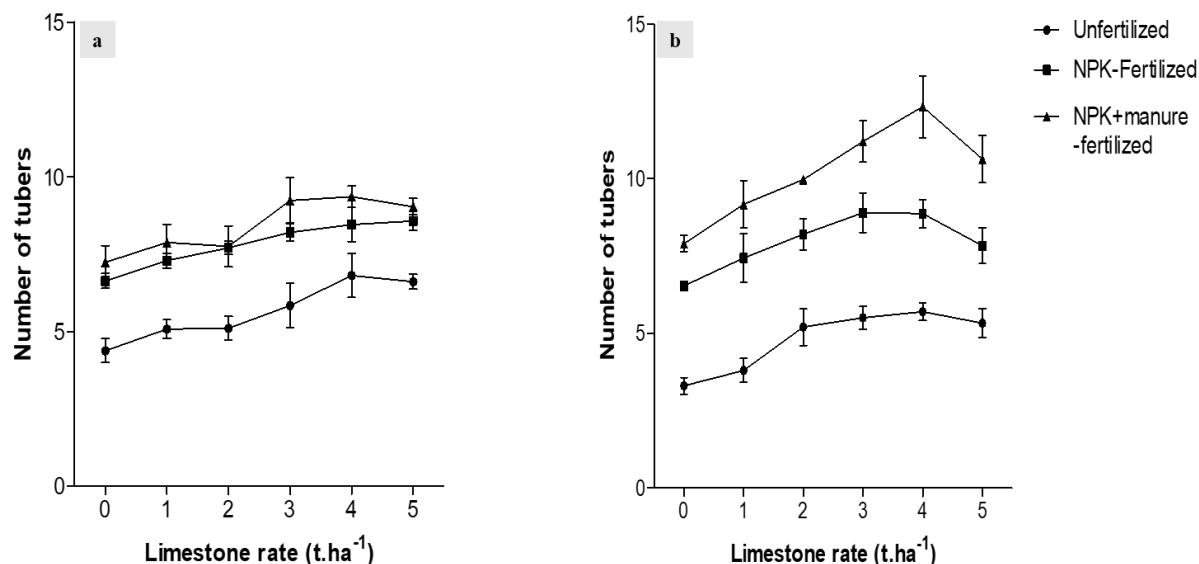


Figure 2. Number of potato tubers per plant as affected by limestone rate on different soil fertilization types in Wassande (a) and Bangou (b)

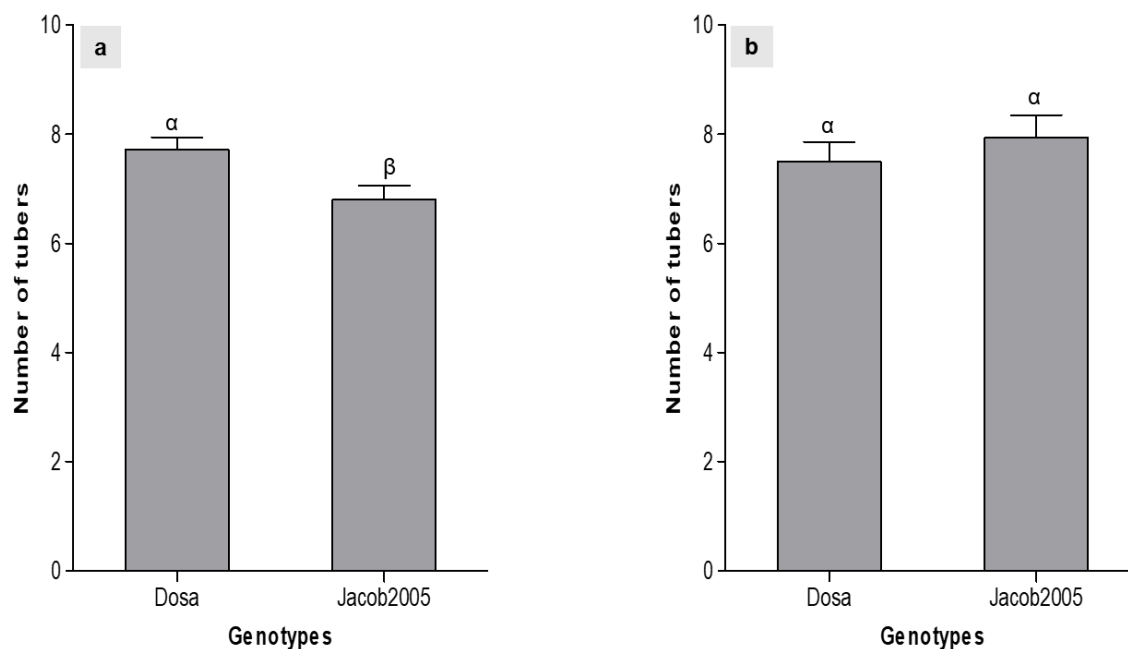


Figure 3. Number of tubers per plant as affected by potato genotypes in Wassande (a) and Bangou (b)

Marketable yield

On unfertilized soil, Marketable yield increased with limestone rate and reached the highest values (15.14 and 8.20 t. ha⁻¹ in Wassande and Bangou respectively) at 5 t. ha⁻¹ of limestone (Figure 4). With regard to fertilized soils, marketable yield increased with limestone rate and reached a peak (28.92 t. ha⁻¹ and 31.07 t. ha⁻¹ on NPK-fertilized and NPK+manure-fertilized soils respectively) at 3 t. ha⁻¹ of limestone in Wassande (Figure 4a). The same trend was observed in Bangou where the peak (29.18 t. ha⁻¹ and 41.41 t. ha⁻¹ on NPK-fertilized and NPK+manure-fertilized soils respectively) was reached at 4 t. ha⁻¹ of limestone (Figure 4b). The highest values of marketable yield were obtained on NPK+manure-fertilized soil, followed by NPK-fertilized in both sites (Figure 4). With regards to the effect of genotype on the marketable yield, Dosa had higher yield (22.37 t. ha⁻¹) than Jacob2005 (19.80 t. ha⁻¹) in Wassande (Figure 5a), whereas Jacob had higher yield (22.51 t. ha⁻¹) than Dosa (20.74 t. ha⁻¹) in Bangou (Figure 5b).

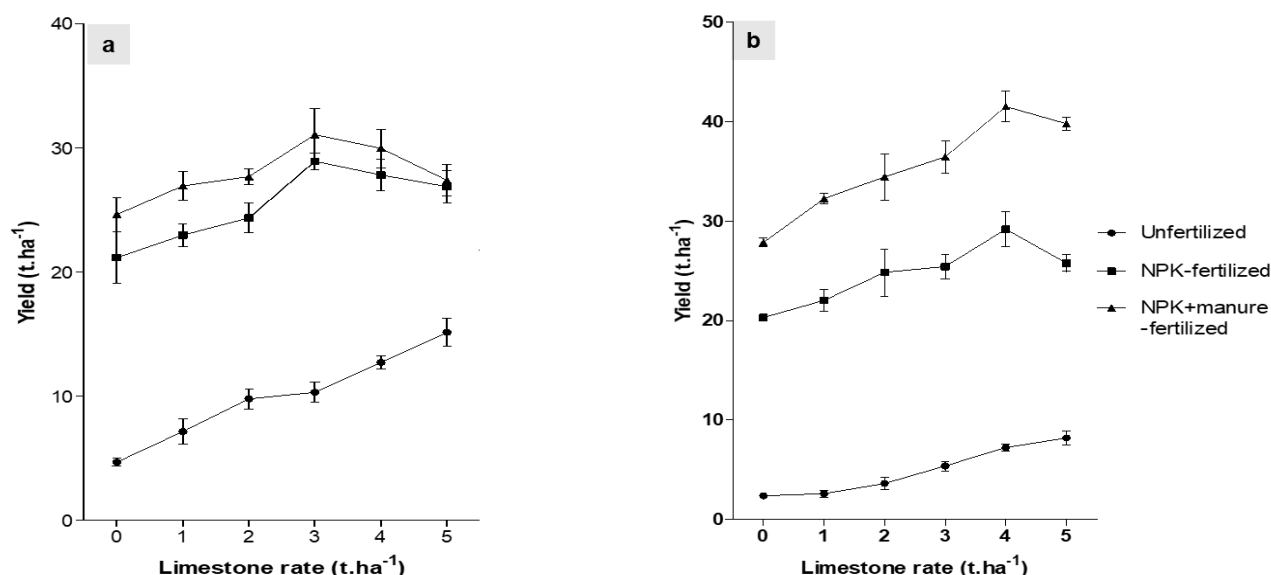


Figure 4. Marketable yield as affected by limestone rate on different soil fertilization types in Wassande (a) and Bangou (b)

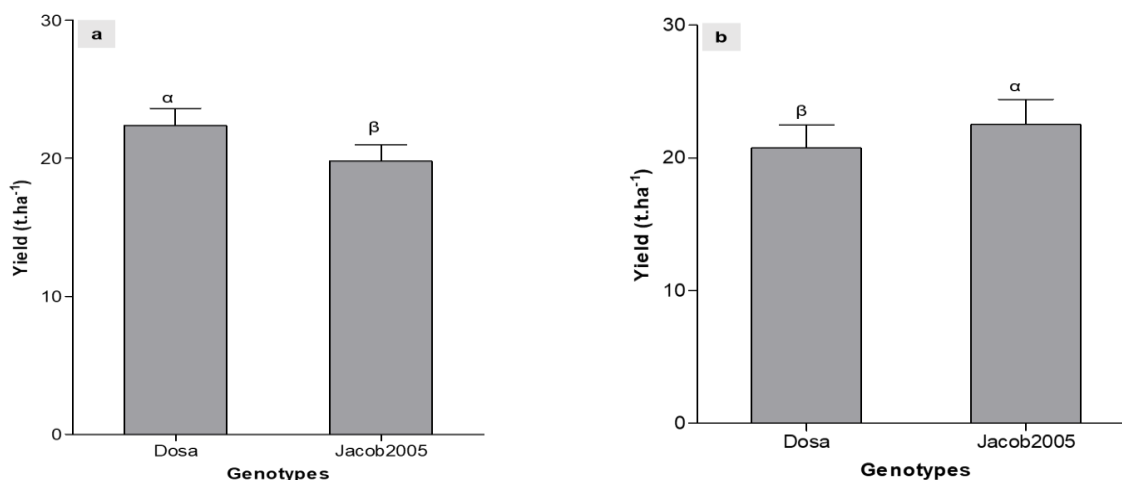


Figure 5. Marketable yield as affected by potato genotypes in Wassande (a) and Bangou (b)

Discussion

Limestone application modified the physico-chemical properties of the soil, whether fertilized or not. Majority of nutrients were higher on fertilized soils than on unfertilized soil. Fertilizer addition increased the availability of some nutrients in the soil as previously reported (Ali et al., 2014). Limestone application significantly increased the pH of the soil, both in terms of pH_{water} and pH_{KCl} . pH_{water} is higher than pH_{KCl} at all limestone rates. Although pH_{water} is used to determine key soil characteristics, pH_{KCl} is more suitable for estimating exchangeable acidity and knowing the level of exchangeable Al associated with infertility in acidic soils (Antoniadis et al., 2015). Increase in soil pH is due to the release of OH^- ions, and basic cations from calcium carbonate into the soil solution. This release helps to neutralize soil acidity (Antoniadis et al., 2015). The increase in pH with increasing rates of limestone could be explained by the fact that, calcium carbonate has a high pH (8.1). Limestone application contributed to increase organic matter and N in the soil. According to Grover et al. (2017) increase in soil pH contributes to increase microbial activity, which leads to greater decomposition of OM and consequently increases N content. Mkhonza (2020) reported that the large quantities of carbon act as an energy source for microorganisms in mineralization leading to increased N production. In this way, the high levels of OM may have likely contributed to N production through decomposition. Limestone addition increased Ca and P content in the soil. Acidic soils have a high phosphorus fixation capacity due to the high Al and Fe content, hence reducing its availability. The content of P and Ca increased with the limestone rate up to 3 or 4 t. ha^{-1} respectively and decreased with higher rates. Decrease in Ca and P content may be related to the formation of Ca-P complex which increases with limestone rate as previously reported by Mkhonza (2020). Al content decreased with increasing rate of limestone on either fertilized soil or not. According to Ramarosan (2017), decrease in the exchangeable Al^{3+} content of soils associated with an increase of calcium carbonate rate is due to the replacement of Al^{3+} ions bound in the colloidal complexes in soil by Ca^{2+} ions. Al content decreased weakly for low limestone rates. On the other hand, a rapid decrease in Al was observed for high limestone rate. The weak reduction in Al^{3+} content could correspond to an exchange phase without neutralization of protons, as reported by Sanchez and Uehara (2015). The second phase, linked to the rapid decrease in the Al^{3+} content of soils, could correspond to the neutralization of protons by the OH^- ions provided by the amendments. This may mean that Al^{3+} exchange reactions in the colloidal complexes happens before hydrolysis and proton neutralization (Djondo, 1995). Changes of soil properties in the present study is associated with improvement potato yield parameters. Results showed that the dose of limestone had a positive influence on the number of potato tubers as compared to the control. This may be due to the increase in P content, which have favored rooting and stolon formation, and consequently tubers formation as previously reported by Cr  mer et al. (2008). Yields were better with the combinations of limestone, NPK and manure. This may be due to the fact that the combined application of NPK and manure provides more nutrients to the plant, unlike the application of any of both fertilizers. These results are in agreement with those obtained by   ydelis et al. (2019) who showed that the combined use of poultry manure and mineral provided better yield in maize as compared to the application of organic or mineral fertilizer alone. Moreover, (Bayu and Gebeyehu, 2013) reported that neither mineral nor organic fertilizers used alone can ensure optimum crop productivity. Integrated use of lime and chemical fertilizers is a good approach for sustainable crop production in acid soils as far as it may build ecologically healthy and sustainable farming systems (Bekele et al., 2018). The high yield obtained on soils with high limestone rate is probably due to the positive effects of liming on soil properties. This results are in agreement with those obtained by (Nduwumuremyi, 2013). The decrease in yield at higher limestone rate may be related to the decrease in many nutrients specially Ca and P which has been reported to be due to the increase of Ca-P complex at higher limestone rates. It has been reported that liming, through optimization of soil pH and improved nutrient availability, improves potato yields under acid soil conditions (Caires et al., 2015). According to Jovovic et al. (2021),

increased crop yields due to lime application have been attributed to reduced exchangeable aluminum, increased soil pH and higher concentrations of basic nutrients such as Ca, Mg and K. The addition of lime has also been reported to increase phosphorus uptake, which would otherwise be unavailable to crops due to phosphorus fixation in acidic soils (Ejigu et al., 2023)

Conclusion

This study aimed to determine the optimal limestone rate for sustainable soil management and improved potato production under different fertilization treatments in two agroecological zones of Cameroon. This agro-ecological zones included AEZ II (Wassande) and AEZ III (Bangou). The application of limestone improved the pH and consequently the availability of minerals in the soil at both sites. Increasing concentrations of limestone improved the yields of the two potato varieties studied. The best performance was obtained with 3 t. ha⁻¹ of limestone in Wassande which was associated to 5.9 pH and 4 t. ha⁻¹ of limestone in Bangou which was associated with 5.9 pH. Policymakers will be able to encourage investment in the processing of agricultural limestone to make it more available to farmers in the various production zones. Dosa had the best performances in Wassande, while Jacob2005 had the best performances in Bangou.

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Consent for publication: All the authors agreed that the manuscript can be publish

Data availability: All relevant data are included in the manuscript

Author contributions: Anoumaa Mariette and Djomo Sime Hervé designed the project Leumassi Mbotchak Michel d'Aquin and Pomo Kamptoum Dieudonné Marcaire performed the field work and collected the data. Leumassi Mbotchak Michel d'Aquin and Anoumaa Mariette analysed the data.. The first draft of the manuscript was written by Leumassi Mbotchak Michel d'Aquin and all authors commented on previous drafts of the manuscript. All authors read and approved the final manuscript.

References

Ali, A., Sharif, M., Wahid, F., Zhang, Z., Shah, S.N.M., Zaheer, S., Khan, F., Rehman, F. Effect of Composted Rock Phosphate with Organic Materials on Yield and Phosphorus Uptake of Berseem and Maize. American Journal of Plant Sciences. (2014). 05, 975–984. <https://doi.org/10.4236/ajps.2014.57110>

- Antoniadis, V., Hatzis, F., Bachtsevanidis, D., and Koutroubas, S.D. Phosphorus Availability in Low-P and Acidic Soils as Affected by Liming and P Addition. *Communications in Soil Science and Plant Analysis*. (2015). 46, 1288–1298. <https://doi.org/10.1080/00103624.2015.1033539>
- Basak, N., Datta, A., Biswas, S., Mitran, T., Mandal, B. Organic Amendment Influences Soil Quality in Farmers' Field under Rice-based Cropping Systems in Indo-Gangetic Plains of India. *Journal of the Indian Society of Soil Science*. (2016). 64, 138–147. <https://doi.org/10.5958/0974-0228.2016.00018.9>
- Bayu, W. and Gebeyehu, S. Effect of Farmyard Manure and Inorganic Fertilizers on the Growth, Yield and Moisture Stress Tolerance of Rain-fed Lowland Rice. *American Journal of Research Communication*. (2013). 1, 275–301.
- Bekele, A., Kibret, K., Bedadi, B., Balemi, T., Yli-halla, M. Effects of lime, vermicompost and chemical P fertilizer on yield of maize in Ebantu District, Western highlands of Ethiopia. *African Journal of Agricultural Research*. (2018). 13, 477–489. <https://doi.org/10.5897/AJAR2017.12949>
- Brady, N.C. and Weil, R.R. Elements of the Nature and Properties of Soils, *Journal of Chemical Information and Modeling*. (2004).
- Caires, E.F., Haliski, A., Bini, A.R., and Scharr, D.A. Surface liming and nitrogen fertilization for crop grain production under no-till management in Brazil. *European Journal of Agronomy* [Internet], (2015). 66, 41–53. Available from: <http://dx.doi.org/10.1016/j.eja.2015.02.008>
- Charles, B., Burleigh, F.Y.J.R., and Eylands, V.J. Correction of acid infertility in rwandan oxisols with lime from an indigenous source for sustainable cropping. *International Agricultural Programs*. (1992). 28, 417–424.
- Devaux, A., Goffart, J., Petsakos, A., and Kromann, P. Global Food Security, Contributions from Sustainable Potato Agri-Food Chapter 1 Global Food Security, Contributions from Sustainable Potato Agri-Food Systems. (2020). <https://doi.org/10.1007/978-3-030-28683-5>
- Djondo, M.-Y. Propriétés d'échange ionique des sols ferrallitiques argileux de la vallée du Niari et sableux du plateau de Mbe-Bateke au Congo: application à la correction de leur acidité. Centre de Montpellier. (1995). 259.
- Djoukeng, H.G., Tankou, C.M., Degré, A. Pollution and siltation of rivers in the western highlands of Cameroon: A consequence of farmland erosion by runoff. 21st Century Watershed Technology Conference and Workshop 2014: Improving Water Quality and the Environment. (2014). 3, 95–101. <https://doi.org/10.13031/wtcw.2014-012>
- Djoukeng, H.G. Dogot, T. Tankou, C. M. & Degré, A. Contraintes socio-économiques de répartition des terres et impacts sur la conservation des sols dans les hauts plateaux de l'Ouest du Cameroun. (2016).
- Ducharme, A. Augmentation de la résistance à la gale commune de cultivars de pomme de terre par l'habitation et la sélection de cellules somatiques. (2016).
- Ejigu, W., Selassie, Y.G., Elias, E., and Molla, E. Effect of lime rates and method of application on soil properties of acidic Luvisols and wheat (*Triticum aestivum*, L.) yields in northwest Ethiopia. *Heliyon* [Internet], (2023). 9(3), e13988. Available from: <https://doi.org/10.1016/j.heliyon.2023.e13988>
- Fageria, N.K. and Baligar, V.C. Ameliorating Soil Acidity of Tropical Oxisols by Liming for Sustainable Crop Production. *Advances in agronomy*. (2008). 99, 345–399. [https://doi.org/10.1016/S0065-2113\(08\)00407-0](https://doi.org/10.1016/S0065-2113(08)00407-0)
- FAO. L'actualité agricole en Méditerranée. Ed. Ciheam. (2020).
- Getachew, E., Mohammed, A., Tesfaye, A., and Nebiyu, A. Growth and yield response of green beans (*Phaseolus vulgaris* L.) In relation to time of sowing and plant spacing in the humid tropics of Jimma, southwest. *International Journal of Soil and Crop Science*. (2014). 2, 61–67.

- Getachew, T. Health Extension Workers' diagnostic accuracy for common childhood illnesses in four regions of Ethiopia: a cross-sectional study. *Acta Pædiatrica*. (2019). 1–7. <https://doi.org/10.1111/apa.14888>
- Grover, S.P., Butterly, C.R., Wang, X., and Tang, C. The short-term effects of liming on organic carbon mineralisation in two acidic soils as affected by different rates and application depths of lime. *Biology and Fertility of Soils*. (2017) .53, 431–443. <https://doi.org/10.1007/s00374-017-1196-y>
- Jovovic, Z., Dolijanovic, Z., Spalevic, V., Dudic, B., Przulj, N., and Velimirovic, A. Effects of liming and nutrient management on yield and other parameters of potato productivity on acid soils in montenegro. *Agronomy*. (2021) .11(5), 1–13.
- MINADER. Programme d'appui à la Relance de la Filière Pomme de Terre (PRFPT), choix du matériel végétal et du site, aménagement de la parcelle, plantation et entretien. MINADER Yaoundé, Cameroun. (2008).15.
- Mkhonza, N. P. Buthelez-Dube, N. N., and Muchaonyerwa, P. Effects of lime application on nitrogen and phosphorus availability in humic soils. *Scientific Reports*. (2020).1–12. <https://doi.org/10.1038/s41598-020-65501-3>
- Nduwumuremyi, A. Effects of different limes on soil properties and yield of irish potatoes (*solanum tuberosum*. l) in burera district, rwanda. (2013).89.
- Nduwumuremyi, A., Ruganzu, V., Mugwe, J.N., and Rusanganwa, A.C. Effects of Unburned Lime on Soil pH and Base Cations in Acidic Soil. *International Scholarly Research Notices*. (2013).
- Ngachie, V. A general assessment of soil ressources and soil fertility constraints in Cameroon on the basis of FAO-UNESCO soil map analysis. *Tropicultura*. (1992).
- Opala, P.A., Kisinyo, P.O., and Nyambati, R.O. Effects of Tithonia diversifolia, farmyard manure and urea, and phosphate fertiliser application methods on maize yields in western Kenya. *Journal of Agriculture and Rural Development in the Tropics and Subtropics* (2015). 116, 1–9.
- Oshunsanya, S. O. K., Oluwasemire, O., and Taiwo, O. J. Use of GIS to Delineate Site-Specific Management Zone for Precision Agriculture. *Communications in Soil Science and Plant Analysis*. (2017). 3624, 30. <https://doi.org/10.1080/00103624.2016.1270298>
- Pauwels, J.M., Van Ranst, E., Verloo, M. and Mvondo Ze, A. Manuel de laboratoire de pédologie. Méthodes d'analyses des sols et des plantes, équipement, gestion de stocks de verrerie et de produits chimiques, Publicatio. ed. Publications Agricoles 28, Bruxelles. (1992).
- Ramaroson Volaniaina Henintsoa. La spectrométrie infrarouge comme outil de caractérisation des constituants et des propriétés physico- chimiques des sols ferrallitiques des Hautes Terres de Madagascar. (2017). 194.
- Sanchez, P.A. and Uehara, G. Management Considerations for Acid Soils with High Phosphorus Fixation Capacity. *The Role of Phosphorus in Agriculture*. (2015).471–514. <https://doi.org/10.2134/1980.roleofphosphorus.c18>
- Sanon, A. and Ouattara, S. Effets à court terme de l' application des fientes de volaille sur le rendement du maïs (*Zea mays* L.) et les caractéristiques chimiques d ' un sol ferrallitique dans la zone sud-soudanienne du Burkina Faso Effects on short term application of poultry m. *International Journal of Biological and Chemical Sciences*. (2019). 13, 2041–2052.
- Sy, A.A. and Houssou, M. Diagnostic du système national de recherche et de vulgarisation agricoles du Cameroun. (2008).
- Tamfuh, P.A., Pangop, C.K., Tapindje, D.D., Boukong, A., Tabi, F., Cho-Ngwa, F., and Biton, D. Effect of dolomite amendment of acid Andosols on the performance of two green beans varieties in the Cameroon

- Western Highlands. *International Journal of Advanced Geosciences* (2019). 7, 1. <https://doi.org/10.14419/ijag.v7i1.18372>
- VSN International. *Genstat for Windows 21st Edition*. (2022).
- Tsozué, D., Nghonda, J.P., and Mekem, D.L. Impact of land management system on crop yields and soil fertility in Cameroon. *Solid Earth*. (2015). 6, 1087–1101. <https://doi.org/10.5194/se-6-1087-2015>
- Yengoh, G.T. and Ardo, J. Crop Yield Gaps in Cameroon. (2014). 175–190. <https://doi.org/10.1007/s13280-013-0428-0>
- Zhang, H., Xu, F., Wu, Y., Hu, H. and Dai, X. Progress of potato staple food research and industry development in China. *Journal of Integrative Agriculture*. (2017). 16, 2924–2932. [https://doi.org/10.1016/S2095-3119\(17\)61736-2](https://doi.org/10.1016/S2095-3119(17)61736-2)
- Žydelis, R., Lazauskas, S., Volungevičius, J., Povilaitis, V. Effect of organic and mineral fertilisers on maize nitrogen nutrition indicators and grain yield. *Zemdirbyste-Agriculture*. (2019). 106, 15–20. <https://doi.org/10.13080/z-a.2019.106.002>

REVIEW ARTICLE

Peptides, Modern Technology for Plant Defense Response for Sustainable Green Agriculture for Increasing worldwide populations

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Abstract

Peptides are short chains of amino acids that are the building blocks of chosen proteins. Peptides are a secure and efficient alternative to existing synthetic pesticides and insecticides. A constant and reasonable food supply is critical to the foundation of economic prosperity and growth in several countries. Innovations in fertilizers, pesticides, plant breeding, trait development, and improved farming methods are the main drivers of productivity and growth. India's per capita food prices are among the lowest in the world among healthcare, pharmaceuticals, animal nutrition, and plant nutrition. In current years, peptides have become widespread research targets in crop protection as antibacterial agents, immune inducers, plant growth regulators, insecticides, and herbicides due to their rich raw material sources, excellent activity, and supreme environmental compatibility. The present manuscript briefly introduces the advances in peptide research, provides an overview of peptide research in crop protection, and summarizes the utilization of peptides in crop protection and the prospects of peptides as green pesticides for agriculture and sustainable development for increasing populations.

Keywords: Peptide; Plant Protection; Antimicrobial; Plant Growth Regulator; Insecticide

Introduction

Pesticides is a significant tool for plant protection and play a vital role in agriculture sector and food security (Zhang *et al.*, 2023). Without the utilization of pesticides, there would be 78% loss in fruit production, 54% loss in vegetable production, and 32% loss in cereal production (Tudi *et al.*, 2021). Therefore, pesticides contribute to increasing crop yields around the world but require updating to meet agricultural development needs and environmental safety requirements. In the era of organic agriculture with an emphasis on sustainable development, there is an urgent need for effective and environmentally friendly green pesticides that are effective against pests while posing a low risk to non-target organisms. As of late, peptides arose as a rising new star in the field of plant protection because of the wide accessibility of natural substance (Mita and Sato 2019; Hedges and Ryan 2020; Troyano *et al.*, 2021; Patel *et al.*, 2024) excellent activity (Wang *et al.*, 2021) and ideal environmental compatibility (Bomgardner 2017; Zhou *et al.*, 2022; Tang *et al.*, 2023). They have been applied as antimicrobials and immune inducers, plant growth regulators, insecticides, and herbicides to protect plants from bacteria, viruses, pests, and weeds. Currently 18 peptides have been commercialized as green agriculture for plant protection.

The bioinsecticide Spear®, derived from a neuropeptide of spider toxin (the Blue Mountains funnel-web spider bug), won the Presidential Green Science Challenge Award in 2020 and Best New Biological Agent Award in 2021 in the US. Peptides with excellent quality and success stories, such as Spear®, are considered important new technology for crop protection and are therefore very attractive for green pesticide research and development. Understanding the studies and uses of current peptides is necessary for the development of novel peptide insecticides. Pesticides, important crop protection tools, play an important role in agriculture and food security. Production use of pesticides, fruit, vegetable, and grains production would be dramatically reduced. Therefore, pesticides contribute to increasing crop yields around the world but require updating to meet agricultural development needs and environmental safety requirements.

Peptides are short-chain biomolecules composed of two to fifty amino acids connected by peptide bonds. They can also be obtained from intermediate products of protein hydrolysis. Peptides can be classified into homomers and heteromers based on their composition, with the former consisting only of amino acids and the latter containing amino acids and non-amino acids such as glycopeptides. All peptides except cyclic peptides have N-terminal (amine group) and C-terminal (carboxyl group) residues and are classified into natural peptides and artificially synthesized peptides depending on their origin. Most natural peptides are derived from animals, plants, and microorganisms. Natural and synthetic peptides can be produced by chemical synthesis, biological fermentation, genetic recombination, and other methods. Peptides are ubiquitous in living organisms and regulate many physiological processes, making them common subjects of research in medicine, cosmetics, agriculture, and more. This article briefly introduces the advances in peptide research, reviews peptides in crop protection, and summarizes the applications of peptides in crop protection for sustainable agriculture. Peptides are a useful tool for sustainable agriculture since they are safe for the environment, effective, and have multiple applications in plant protection.

Utilization of Green Peptides in Plant Protection for Sustainable Agriculture

Peptides are advantageous for effective and environmentally friendly pest management in sustainable agriculture. They are employed as insecticides, herbicides, growth regulators, inducers of plant immunity, and antibacterials. Figure 1 illustrates the accomplishments in the history of peptide development.

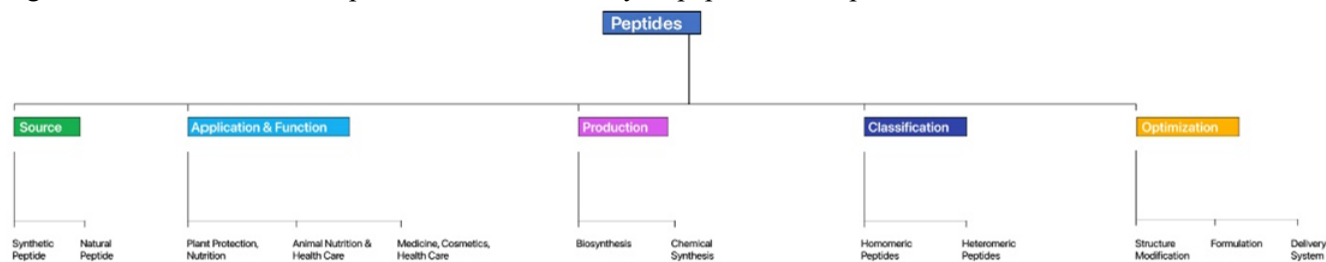


Figure1: Diagrammatic Skeleton of Peptide Research on sources, Classification, Optimization, Production, Application

Antimicrobial and Immune-inducing Peptides for Agriculture Applications

Plant pathogens attack crops and have serious negative effects on their growth and development. Conventional chemical fungicides provide effective prevention of crop pathogens. A selection of animal antimicrobial peptides (AMPs) carrying out agricultural activities is shown in Table 1.

Table 1. Classification and Agricultural Bioactivity of Animal Antimicrobial Peptides

Peptide	Source	Function	Species effectiveness	References
Abaecin	Apis mellifera	Antibacterial	Agrobacterium tumefaciens Erwinia salicis, Pseudomonas syringae Xanthomonas campestris	Casteels et al., 1990
Apidaecins	Apis melifera	Antibacterial	A. tumefaciens, E. salicis, P. syringae Rhizobium meliloti	Casteels et al., 1989, Casteels et al., 1994
Cecropin B	Hyalophora cecropia	Antibacterial, Antifungal	P. syringae pv. Tomato, P. syringae pv. Syringae, P. syringae pv. Tabaci X. campestris pv. Vesicatoria, Clavibacter michiganensis subsp. Michiganensis, Erwinia carotovora subsp. Carotovora, E. carotovora subsp. Chrysanthemi, A. tumefaciens, Penicillium digitatum Phytophthora infestans	Alan and Earle 2002
Dermaseptin	Rhacophorus	Antibacterial	Xylella fastidiosa	Kuzina et al., 2006
Drosomycin	Drosophila melanogaster	Antifungal	Botrytis cinerea, Fusarium culmorum F. oxysporum, Nectria haematococca Alternaria brassicola, A. longipes Trichoderma viride, Ascochyta pisi	Fehlbaum et al., 1994
Indolicidin	Bovine	Antibacterial	X. fastidiosa	Kuzina et al., 2006
LfcinB	Bovine	Antifungal	P. digitatum., P. Italicum, P. expansum, Penicillium sp. Alternaria sp., Aspergillus nidulans B. cinerea, F. oxysporum	Mun-oz et al., 2006
Magainin II	Xenopus laevis	Antibacterial, Antifungal	P. syringae pv. Tomato P. syringae pv. Syringae P. syringae pv. Tabaci X. campestris pv. Vesicatoria C. michiganensis subsp. Michiganensis P. digitatum X. fastidiosa	Alan and Earle, 2002
Penetratin	Drosophilid	Antibacterial	Bacillus megaterium	Palm et al., 2006
PGQ	X. laevis	Antibacterial	X. fastidiosa	Kuzina et al., 2006
pVEC	Mammalian	Antibacterial	B. megaterium	Palm et al., 2006
Spodopsin Ia	Spodoptera litura	Antibacterial	B. megaterium	Choi et al., 1997

Table 2 mentioned agriculture bioactivities antimicrobial peptides obtained from plants. Classification and agricultural bioactivity of microbes and some other antimicrobial peptides describe in table 3 and 4. Synthetic peptides accounted for 49.33% of all peptides listed, followed by his AMPs derived from plants (29.33%), animals (16.00%), and microorganisms (5.33%). Microbial and some other sources of important bioactive antimicrobial peptides carrying out agriculture activities are shown in Tables 3 and 4. Natural AMPs are produced by animals, plants, and microbes (Sharma *et al.*, 2000; Li *et al.*, 2021a).

Table 2. Classification and Agricultural Bioactivity of Plants Antimicrobial Peptides

Peptide	Source	Function	Species Effectiveness	References
α 1-purothionin	Triticum aestivum	Antibacterial	Xanthomonas	Caleya et al., 1972
BLAD	Lupinus albus	Antifungal	Erwinia B. cinerea Erysiphales	Pinheiro et al., 2018
Ca-AFP	Capsicum annum	Antifungal	F. oxysporum Phytophthora capsici	Capella et al., 2001
Ca-LTP1	C. annum L.	Antifungal	F. oxysporum Colletotrichum lindemuthianum	Cruz et al., 2010
J1	C. annum	Antifungal	C. gloeosporioide, C. musae F. oxysporum	Diz et al., 2006; Seo et al., 2014
maSAMP	Citrus australasica F. Muell	Antibacterial	Liberobacter asianticum	Jagoueix Wang et al, 2021
NaD1	Nicotiana glauca	Antibacterial, Antifungal	B. cinerea, F. oxysporum F. oxysporum f. Sp. vasinfectum Thielaviopsis basicola Verticillium dahlia A. nidulans, F. graminearum	Van der et al., 2008; Van der et al., 2010. Kerenga et al., 2019
Pa-AFP1	Passiflora alata Curtis	Antifungal	C. gloeosporioide	Ribeiro et al., 2011
Pe-AFP1	Passiflora edulis	Antifungal	A. fumigatus, F. oxysporum	Pelegrini et al., 2006
Peptide-1	Oryza sativa	Antifungal	Magnaporthe oryzae	Sagehashi et al., 2017
Pf2	Passiflora edulis f. Flavicarpa	Antifungal	F. oxysporum C. musae C. lindemuthianum	Agizzio et al., 2003
PhD1	Petunia hybrida	Antifungal	B. cinerea F. oxysporum	Lay et al., 2003; Segonzac and Monaghan 2019
PhD2	P. hybrida	Antifungal	B. cinerea	Lay et al., 2003. Jenssen et al., 2006
PvD1	Phaseolus vulgaris	Antifungal	F. oxysporum F. solani, F. lateritium	Mello et al., 2011
SD2	Helianthus annuus	Antifungal	Sclerotinia sclerotiorum	Urdangarin et al., 2000
Snakin-1	Solanum tuberosum	Antibacterial, Antifungal	B. cinerea, F. solani F. culmorum, F. oxysporum Plectosphaerella cucumerina Colletotrichum lagenarium C. graminicola, Bipolaris maydis A. flavus, C. michiganensis, Ralstonia solanacearum	Segura et al., 1999; Berrocal-Lobo et al., 2002

Snakin-2	S. tuberosum	Antibacterial, Antifungal	C. michiganensis R. solanacearum (rfa-) R. meliloti, B. Cinerea, F. Solani, F. culmorum, F. oxysporum f. Sp. Conglutinans, F. oxysporum f. Sp. Lycopersici, P. Cucumerina, C. graminicola, C. lagenarium, B. maydis, A. flavus	Berrocal-Lobo et al., 2002
Snakin-Z	Ziziphus jujuba	Antifungal	Pythium ultimum, A. niger	Daneshmand and Gill 2013
Thi2.1	Arabidopsis thaliana	Antifungal	F. oxysporum	Epple et al., 1995
Tn-AFP1	Coconut water Trapa natans	Antifungal	F. oxysporum, Mycosphaerella arachidicola	Wang and Ng 2005
ZmD32	Zea mays	Antibacterial, Antifungal	F. graminearum	Kerenga et al., 2019
ZmPep1	Z. mays	Antifungal	Helminthosporium, Pythium spp. Fusarium	Huffaker et al., 2011

Cathelicidins and defensins represent two major AMP families in mammals (Ouellette *et al.*, 2006). Based on their sequence and structure, plant AMPs are commonly referred to as thionins, defensins, and hevein type peptides, knottins, stability-like peptides, lipid transfer proteins, snakins and cyclotides (Li *et al.*, 2021b). Bacteria and fungi are also his own AMP reservoirs (Huan *et al.*, 2020). AMPs from bacteria are not produced to prevent infection, but as a competitive strategy to kill other microbes present in the body. Food and food compete for the same ecological niche, ensuring the survival of a single bacterial cell (Jenssen *et al.*, 2006).

Synthetic peptides can be synthesized using chemical methods and screened from combinatorial libraries like bacterial two-hybrid system screening peptide library (Xu *et al.*, 2019b), yeast-based two-hybrid system screening peptide library (Xu *et al.*, 2019a). Hybrid libraries, (Colombo *et al.*, 2020) and phage display (Lee *et al.*, 2019) are based on affinity and specificity for key target proteins. AMPs have various inhibitory effects on bacteria, fungi, parasites, and viruses. Microbial NAF exhibits antimicrobial action against *Aspergillus flavus*, *Fusarium solani*, and *Penicillium italicum*. Animal PVEC is active against *Bacillus megaterium*, whereas plant PhD2 is active against *B. cinerea*. *Verticillium dahlia* can be effectively combated with Synthetic Alf-AFP. Table 2 listed several AMPs in development. BLAD (Pinheiro *et al.*, 2018), a peptide produced during germination of *Lupine albus*, was developed as two products. ProBlad® Verde from Sym-Agro in the USA and Problad Plus™ from Consume em Verde in Portugal. Problad Plus™ (<https://www.cev.com.pt/>) is a biofungicides containing 20% BLAD that acts on susceptible fungal pathogens by damaging cell walls and internal membranes.

It is effective against pathogens such as powdery mildew and gray mold, and is recommended for crops such as strawberries, grapes, tomatoes, and stone fruits. Currently, it is used to control coffee leaf rust, white mold, gray mold, powdery mildew, anthracnose, bluegrass leaf rust, leaf blight, and Rhizopus from grapes, herbs, coffee, and leafy vegetables.

Table 3. Classification and Agricultural Bioactivity of Microbe's Antimicrobial Peptides

Peptide	Source	Function	Species Effectiveness	References
AFP	Aspergillus giganteus	Antifungal	F. culmorum, F. equiseti, F. Lini, F. moniliforme, F. oxysporum, F. poae, F. Proliferatum, F. solani, F. sporotrichoides, F. vasinfectum, Magnaporthe grisea, P. infestans	Marx 2004
ANAFP	A. niger	Antifungal	A. fumigatus, A. flavus, F. oxysporum, F. solani	Marx 2004
NAF	Penicillium nalgiovense	Antifungal	A. flavus, F. solani, P. italicum	Marx 2004
PAF	Penicillium chrysogenum	Antifungal	A. fumigatus, A. flavus, A. niger, B. cinerea Cochliobolus carbonum F. oxysporum, Blumeria graminis f. Sp. Hordei Puccinia recondita f. sp. tritici	Kaiserer et al., 2003; Marx 2004; Barna et al., 2008

Table 4. Classification and Agricultural Bioactivity of some other Important Antimicrobial Peptides

Peptide	Source	Function	Species effectiveness	References
α _P2	Synthesized	Antifungal	P. capsici	Lee et al., 2019
Alf- AFP	Recombinant expression	Antifungal	Verticillium dahliae	Gao et al., 2000
CAMEL	Rational designed	Antibacterial	Pectobacterium carotovorum P. chrysanthemi	Kamysz et al., 2005
Cecropin P1	Recombinant expression	Antibacterial	P. syringae Pseudomonas marginata E. carotovora	Zakharchenko et al., 2005; Yevtushenko et al., 2005
CEMA	Rational-designed	Antifungal	F. solani	Yevtushenko et al., 2005
Dm-AMP1	Recombinant expression	Antifungal	B. cinerea Verticillium alboatrum	Turrini et al., 2004
DS01-THA	Rational designed	Antifungal	Phakopsora pachyrhizi	Duman-Scheel 2019; Schwinges et al., 2019
D4E1	Rational-designed	Antibacterial, Antifungal	Colletotrichum destructivum A. tumefaciens Xanthomonas populi	Cary et al., 2000; Rajasekaran et al., 2005 Vila-Perello et al., 2003
D32R	Rational-designed	Antibacterial, Antifungal	F. oxysporum, P. cucumerina B. cinerea, X. campestris pv. Translucens, C. michiganensis	Vila-Perello et al., 2003
ESF12	Rational-designed	Antifungal	Septoria musiva	Liang et al., 2002
Isegaran	Rational-designed	Antibacterial	P. carotovorum, P. chrysanthemi	Kamysz et al., 2005
KYE28	Rational-designed	Antibacterial	X. vesicatoria X. oryzae	Datta et al., 2016 Huang et al., 1997; Liu et al., 2001
MB39	Rational-designed	Antibacterial	P. syringae Erwinia amylovora	Schaefer et al., 2005

Mj- AMP1	Recombinant expression	Antifungal	Alternaria solani	Schaefer et al., 2005
MSI-99	Rational-designed	Antibacterial, Antifungal	F. oxysporum, MycosphaerellaChakrabarti et al., 2003; musicola, P. syringae pv. Tomato Alan et al., 2004; P. syringae pv. Syringae Mun-oz et al., 2006 P. syringae pv. Tabaci, X.Osusky et al., 2000 campestris pv. Vesicatoria C. michiganensis subsp. Michiganensis, E. carotovora subsp. Carotovora, E. carotovora subsp. Chrysanthemi, A. tumefaciens, P. digitatum	
MSrA1	Rational-designed	Antibacterial, Antifungal	E. carotovora Phytophthora cactorum F. solani	Osusky et al., 2005; Yevtushenko and Misra 2007
MsrA2	Rational-designed	Antibacterial, Antifungal	E. carotovora, Pythium irregulare P. paroecandrum, F. Solani, Rhizoctonia, Phytophthora P. sp. f. oxysporum, A. Alternata, B. cinerea, P. carotovorum	Osusky et al., 2004
MsrA3	Rational designed	Antibacterial, Antifungal	E. carotovora P. infestans Phytophthora erythroseptica	Osusky et al., 2004
Myp30	Rational designed	Antibacterial, Antifungal	E. carotovora Peronospora tabacina	Li et al., 2001
NCR044	Medicago truncatula	Antifungal	B. cinerea Fusarium spp.	Colombo et al., 2020
NoPv1	Peptide aptamer library	Antifungal	P. viticola P. infestans P. digitatum	Colombo et al., 2020
O3TR, C12O3TR	Rational designed	Antifungal		Li et al., 2019
Peptaibol	Chemically modified	Antifungal	B. cinerea, Bipolaris sorokiniana F. graminearum, P. expansum	De Zotti et al., 2020
Pep11	Rational-designed	Antifungal	P. infestans	Li et al., 2019
Pexiganan	Rational designed	Antibacterial	P. carotovorum, P. chrysanthemi	Kamysz et al., 2005
PV5	Rational designed	Antibacterial, Antifungal, Antiviral	E. carotovora, TMV	Toth et al., 2020
Γ NFAP-opt, Γ NFAP-Rational designed	Rationally designed	Antifungal	C.herbarum E. carotovora	Toth et al., 2020 Arce et al., 1999
optGZ SB-37				
Shiva-1	Rational designed	Antibacterial	Pseudomonas solanacearum E. carotovora Phytoplasma	Jaynes et al., 1993; Yi et al., 2004; Du et al., 2005
SNP-D4	Peptide aptamer library	Antifungal	M.oryzae	Xy et al., 2016; Xu et al., 2019b
Tachyplesin I	Recombinant expression	Antibacterial, Antifungal	V. dahliae	Allefs et al., 1996

TK VI	Trichoderma pseudokoningii strain SMF2	Antifungal	E. carotovora, B. cinerea F. oxysporum, Ascochyta citrulline	Shi et al., 1912; Zhao et al., 2018
VG16KRKP	Rational designed	Antibacterial	Phytophthora parasitica V. dahliae; X. oryzae	Datta et al., 2015
10 R,11 R	Rational designed	Antibacterial, Antifungal, Antiviral	X. campestris E. carotovora Fungi TMV	Bhargava et al., 2007

Immune Inducing Peptides (IIPs) for Sustainable Agriculture

Because of their function in host innate defence (HID), AMPs can act as immune inducers in plants, trigger defence signals, and enhance innate immunity (Bende et al., 2014; Bende et al., 2015). Three types of peptides have been investigated as commercial immune inducers. Invaio Sciences' peptide maSAMP (<https://www.invaio.com/>) is being used to combat the destructive citrus disease. This peptide kills *Liberobacter asianticum jagoueix*, the causative agent of Huanglongbin disease, and activates the plant's immune system to prevent subsequent infections. The helical structure of maSAMP quickly penetrates bacterial membranes and lyses them within 30 minutes. Considering the lack of effective products to combat this disease, it is hoped that maSAMP can become an effective tool. This product is used as a seed treatment against Asian soybean rust. Phytotech's FLG22 (<https://phytotechlab.com/>) induces a natural immune response. Its sequence is derived from the highly conserved N-terminal region of *Pseudomonas aeruginosa* flagellin. FLG22 and its derivatives induce defense responses and exhibit elicitor activity in *Lycopersicon esculentum* and *Arabidopsis thaliana*. Numerous IIPs are under development. PIP1 and PIP2 enhance immune responses and pathogen resistance in *Arabidopsis* (Hou et al., 2016). *Nicotiana tabacum* NbPPI1 stimulates the immune response and increases plant resistance to *Phytophthora* (Wen et al., 2021). The maize immune signaling peptide Zip1 reduces the virulence of the maize smut fungus (Stegmann et al., 2017). Cowpea and kidney bean defense causes an increase in the defense-related plant hormones salicylic acid and jasmonic acid (Schmelz et al., 2006; Schmelz et al., 2012). Treatment of plants with inceptin produces volatile organic molecules such as indole and methyl salicylate, which are natural enemies of armyworm moths. Therefore, it provides indirect protection.

Plant Growth Regulating Peptides (PGRPs)

PG and development are influenced by plant hormones such as auxins, cytokinins, and gibberellins that mediate intercellular communication during development. However, recent studies have shown that peptide signaling molecules (PSMs) also play important roles in various developmental processes and environmental responses in plants (Olsen et al., 2002; Ryan et al., 2002; Boller 2005; Germain et al., 2006; Matsubayashi and Sakagami 2006; Farrokhi et al., 2008; Hou et al., 2014). PGRPs are a new class of plant hormones (Matsubayashi and Sakagami, 2006) with signaling properties and hormonal actions. They exhibit significant biological activity at very low concentrations (10⁻⁷-10⁻⁹ M). These findings highlight the importance of peptides in regulating plant growth. Approximately 30 families of peptide phytohormones have been identified in plants, and many more interact with plants, including bacterial and fungal pathogens, plant-parasitic nematodes, and symbiotic and plant beneficial bacteria and fungi. It exists in a variety of living things (Dodueva et al., 2021). Classes of plant hormones (CLE, CEP, RALF, IDA, PSK, PSY, and PEP) have been found in bacteria, fungi, and nematodes that interact with plants (Dodueva et al., 2021). These peptide precursors are processed into mature peptides in plants, which then interact. It binds to plant receptors and activates downstream signaling pathways, triggering growth responses. Peptides

that are effective in regulating PG are listed in table 5, 6 and 7, along with their functions in plants. Functions Peptides and their functions as Plant Growth Regulators are mentioned in table 8. The sources of these PGRPs are showing that plant peptides account for 58.93%, followed by synthetic 19. 64%, microbial 14. 29%, and animals 7.14%.

Table 5. PGR Peptides for Animal and their Functions as Plant Growth Regulators

Peptide	Source	Function	References
CLE	Heterodera spp, Globodera spp, Rotylenchulus spp. Meloidogyne spp.16D10 Meloidogyne spp. MAP	Activate downstream signaling pathway leading to growth response	Dodueva <i>et al.</i> , 2021
CEP	<i>Rotylenchulus</i> spp. Meloidogyne spp.	Activate downstream signaling pathway leading to growth response	Dodueva <i>et al.</i> , 2021
Hicure®	Animal protein hydrolysates	Improve plant quality and enhance resistance to environmental stresses	Carillo <i>et al.</i> , 2022
IDA	<i>Meloidogyne</i> spp.	Activate downstream signaling pathway leading to growth response	Dodueva <i>et al.</i> , 2021

Table 6. PGR Peptides for Plants and their Functions as Plant Growth Regulators

Peptide	Source	Function	References
AtRALF1	<i>A. thaliana</i>	Overexpression causes semi-dwarfism, exogenous peptide	Haruta <i>et al.</i> , 2008; Mingossi <i>et al.</i> , 2010
AtRALF23	<i>A. thaliana</i>	Overexpression impairs brassinolide-induced hypocotyl elongation and causes semi-dwarfism	Srivastava <i>et al.</i> , 2009
CEPs	<i>A. thaliana</i>	N-demand signaling, lateral growth, nodulation	Yoshii <i>et al.</i> , 2014; Mohd-Radzam <i>et al.</i> , 2016
CIFs	<i>A. thaliana</i>	Casparian strip formation	Doblas <i>et al.</i> , 2017; Nakayama <i>et al.</i> , 2017
CLE19	<i>A. thaliana</i>	Root apical meristem size	Nakayama <i>et al.</i> , 2017
CLE25	<i>A. thaliana</i>	Improvement of ABA level	Takahashi <i>et al.</i> , 2018
CLE41/44 (TDIF)	<i>A. thaliana</i>	Inhibition of xylem differentiation	Fisher and Turner 2007; Hirakawa <i>et al.</i> , 2008
CLE40	<i>A. thaliana</i>	Cell differentiation	Stahl <i>et al.</i> , 2013; Nikonorova <i>et al.</i> , 2018
CLV3, CLE2	<i>A. thaliana</i>	Stem cell renewal and differentiation	Clark <i>et al.</i> , 1993; Kayes and Clark 1998; DeYoung <i>et al.</i> , 2006; Muller <i>et al.</i> , 2008;

EPF2	A. thaliana	Stomata development	Kinoshita et al., 2010; Hu et al., 2018
GmCLE40	A. thaliana	Stem cell differentiation	Lee et al., 2015
GRI	A. thaliana	Cell death control	Stuhrwohldt and Schaller 2019
IDA	A. thaliana	Floral organ abscission	Wrzaczek et al., 2015
			Cho et al., 2008; Stenvik et al., 2008; Santiago et al., 2016
LRPP	Plants	Improve the resistance to environmental stresses	https://www.hello-nature.com/us/
MtCLE12	A. thaliana	Regulation of nodulation	Kassaw et al., 2017
MtRALFL1	Medicago trunculata	Overexpression causes reduced number and abnormal nodule development, regulated by bacterial nod factors	Combiér et al., 2008
NaRALF	Nicotiana attenuata	RNAi downregulation causes long roots, abnormal root hairs	Wu et al., 2007
Phytosulfokine (PSK)	A. thaliana	Root and hypocotyl cell elongation	Matsubayashi et al., 2002; Matsubayashi et al., 2010; Wang et al., 2015
PSY	A. thaliana	Cell proliferation and expansion	Amano et al., 2007; Fuglsang et al., 2014
PtdRALF1, PtdRALF2	Hybrid Populus	Exogenous peptide causes alkalinisation of cell culture growth medium	Haruta and Constabel, 2003
RALF1	A. thaliana	Extracellular alkalinisation	Haruta et al., 2014
RALF23	A. thaliana	Extracellular alkalinisation	Stegmann et al., 2017
RGFs	A. thaliana	Root meristem activity, gravitropism	Hidefumi et al., 2016; Ou et al., 2016; Song et al., 2016
SacRALF1	Saccharum spp	Exogenous peptide causes inhibition of microcalli development	Mingossi et al., 2010
SIPRALF	Solanum lycopersicum	Exogenous peptide causes inhibition of pollen tube growth	Covey et al., 2010
SIRALF	Solanum lycopersicum	Exogenous peptide causes alkalinisation of growth medium and inhibition of tomato and Arabidopsis root growth	Pearce et al., 2001
Systemin	A. thaliana	Wound response	Wang et al., 2018

TobHypSys, TomHypSy	A. thaliana	Defence signaling	Stuhrwohltd Schaller 2019	and
Tomato CLV3	A. thaliana	Stem cell renewal	Cao et al., 2015	

Table 7. PGR Peptides for Microorganisms and their Functions as Plant Growth Regulators

Peptide	Source	Function	References
CLE	Actinobacteria sp. Thiotrichales sp. Acidimicrobiaceae sp. Gemmatimonadetes sp. Actinobacteria sp. Rhizophagus irregularis, R. diaphanous R. cerebriforme, R. clarus	Mimic peptide phytohormones	Dodueva et al., 2021
CEP	Ralstonia syzygii	Mimic peptide phytohormones	Dodueva et al., 2021
Eapeptide 91,938	E. amylovora	Stimulate crop growth and enhance defense ability and stress resistance	https://www.planthealthcare.com/
IDA	Elampsora larici-populina Colletotrichum fructicola	Mimic peptide phytohormones	Dodueva et al., 2021
PEP	Metschnikowia sp. JCM33374 Mycolicibacterium conceptionense	Mimic peptide phytohormones	Dodueva et al., 2021
PSK	Tilletia sp., Diplodia sp. Macrophomina phaseolina Cercospora sp., Pseudocercospora sp. Zymosentoria spo Proteobacteria s	Mimic peptide phytohormones	Dodueva et al., 2021
PSY	X. oryzae pv. Oryza other Xanthomonas species	Mimic peptide phytohormones	Dodueva et al., 2021
RALF	F. oxysporum Other 26 fungal species Stretomyces acidiscabies and 8 species of Actinobacteria	Extracellular alkalinisation	Dodueva et al., 2021

Plant growth regulating peptide (GRP) has a wide range of functions in PG and development. For example, the functional peptide PY91, discovered by TIBO Crop Science in 2021, disrupts plant growth. CLAVATA3 peptide regulates meristem size (Lay and Anderson 2005). The S cysteine-rich (SCR) peptide is a self-incompatibility recognition factor in Brassicaceae pollen (Fletcher *et al.*, 1999). RALF is a family of peptides that play a key role in plant cell growth (Okuda *et al.*, 2009). The root derived CLE25 peptide allows plants to cope with self-incompatibility. Cope with drought stress by regulating the expression of NCED (Takahashi *et al.*, 2018; Mita and Sato 2019). NCED increases abscisic acid (ABA) levels (Endo *et al.*, 2008; Seo *et al.*, 2000) to induce stomatal closure and maintain water balance. The effects of traditional plant hormones can be amplified or deactivated using

peptide-based hormone products. CLE41/TDIF, BR peptide, and auxin jointly control root formation. In contrast, CLE27 expression is inhibited by auxin. Four types of peptides are used as commercially available plant growth regulators (PGRs). The plant peptide LRPP (<https://www.hello-nature.com/us/>) is also a biostimulant that is the active ingredient in the commercial product Tandem developed by the Italian company Hello Nature. It is a powerful biostimulant that improves tolerance to environmental stresses such as drought, low and high temperatures, or poor soils. This product is used during the sowing stage to establish a closer and mutually beneficial relationship with the seeds. As a growth regulator, it protects against fungi, bacterial pathogens, and nematodes by stimulating growth, the plant's natural defenses and metabolism. Recommended uses include seed treatments and foliar applications. Hicure® (<https://www.syngenta.com/en>) is a natural biostimulant with excellent potency and flexibility, containing easily absorbed peptides and amino acids, which enhances the quality of botanicals. It has been proven to improve and increase resistance to environmental stress. The product is applied as a conventional spray or macerating solution for best results during critical developmental stages, before replanting or transplanting, environmental stress, or before transportation.

Table 8. Other PGR Peptides and their Functions as Plant Growth Regulators

Peptide	Source	Function	References
CAMEL	Rational designed	Inhibiting the growth of different species of <i>Pectobacterium</i>	Kamysz <i>et al.</i> , 2005
CEP1	Rational designed	Increases nutrient uptake rates along plant roots	Roy <i>et al.</i> , 2022
KEYLAN Ca	Rational designed	Optimizing Calcium uptake and boosting plant metabolism	https://www.hello-nature.com/us/
KEYLAN Combi	Rational designed	Optimizing nutrient uptake and boosting plant metabolism	https://www.hello-nature.com/us/
KEYLAN Fe	Rational designed	Optimizing Iron uptake and boosting plant metabolism	https://www.hello-nature.com/us/
KEYLAN Fe	Rational designed	Optimizing Iron uptake and boosting plant metabolism	https://www.hello-nature.com/us/
KEYLAN Max	Rational designed	Optimizing nutrient uptake and boosting plant metabolism	https://www.hello-nature.com/us/
KEYLAN Mn	Rational designed	Optimizing Manganese uptake and boosting plant metabolism	https://www.hello-nature.com/us/
KEYLAN Zn	Rational designed	Optimizing Zinc uptake and boosting plant metabolism	https://www.hello-nature.com/us/
NOP-1	Rational designed	Inhibiting plant senescence	Hofmann <i>et al.</i> , 2021
PEP6-32	Rational designed	Plant seedlings presented longer hypocotyls and diminished cotyledon expansion when grown under red light.	Shuipys <i>et al.</i> , 2019
PSK- α	Rational designed	Promote cell growth and proliferation	Yu <i>et al.</i> , 2016

Insecticidal Peptides (ISPs)

Pest control is an important concern for agriculture, as pests can cause approximately 13-16% crop losses (Rivera-de-Torre *et al.*, 2021). Pests are mainly controlled by chemical insecticides. Inappropriately, the widespread use of these products has resulted in decreased resistance to pests as well as negative effects on human health (HH) and the environment (Windley *et al.*, 2012; Maienfisch and Koerber 2024). Therefore, there is a need to develop biopesticides as an alternative approach for pest control (Park *et al.*, 2011). ISPs with standard properties are currently being considered as possible alternatives. ISPs act on a variety of pests, including Lepidoptera, Diptera, and Hemiptera. Some reported ISPs and their functions are summarized in Table 9-11. ISPs were mostly derived from animals (63.1%), followed by plants (19.64%) and synthetic peptides (11%). There are very few ISPs derived from microbes, except for some fungi and bacteria, such as destruxin A (DA) secreted by *Metarhizium anisopliae* and longibrachinA-I from *Trichoderma longibrachiatum* RIFAI. Natural ISPs are mainly derived from the venoms of arthropods (spiders, scorpions, ants, etc.) and marine animals (jellyfish) (Arntzen 1972; Yu *et al.*, 2005; Yu *et al.* 2021). Sea anemone (Liu *et al.*, 2010; Yan *et al.*, 2014) Cone snail (Bruce *et al.*, 2011; Lebbe *et al.*, 2014; Gao *et al.*, 2017; Chen *et al.*, 2018). These studies pave the way for the development of novel insect control agents based on cyclic peptidomimetics. Antagonists in the backbone of insect neuropeptides (Elakkiya *et al.*, 2019). Vestaron, a recent Green Chemistry award winner, is leading the peptide-based revolution in crop protection. GS- ω/κ -HXTX-Hv1a peptide was used as the active ingredient to launch two Spear® products ((<https://www.vestaron.com/>): 1) Spear® -T is effective against thrips, whiteflies, fruit flies and spider mites found in greenhouses, Spear®-Lep is effective against lepidopteran pests such as caterpillars, nematodes, and raptors in indoor and outdoor crops.

Table 9. Classification and Agricultural Bioactivity of Plants Insecticidal Peptides and their Function

Peptide	Source	Function	Species effectiveness	References
BrD1	Brassica rapa	Insecticidal	Nilaparvata lugens	Choi et al., 2009
Cter M	Clitorea ternatea	Insecticidal	H. armigera	Poth et al., 2011
CycloviolacinH3,	Viola odorata	Insecticidal	M. persicae	Colgrave et al., 2008;
CycloviolacinO1,			Pomacea	Plan et al., 2008;
CycloviolacinO2,			canaliculate	Colgrave et al., 2009
CycloviolacinO3,			Trichostrongylus	
CycloviolacinO8,			Colubriformis	
CycloviolacinO12,			Hemonchus	
CycloviolacinO13,			contortus	
CycloviolacinO14,				
CycloviolacinO15,				
CycloviolacinO16,				
CycloviolacinO19,				
CycloviolacinO24,				
CycloviolacinY1,				
CycloviolacinY4,				
CycloviolacinY5				

Hypa A	Hybanthus parviflorus	Insecticidal	Ceratitidis capitata	Mulinari et al., 2007
JaburetoX-2Ec	Canavalia ensiformis	Insecticidal	Dysdercus peruvianus Callosobruchus maculatus S. frugiperda	Mulinari et al., 2007
KalataB1, KalataB2, KalataB6, KalataB7, KalataB	Oldenlandia affinis	Insecticidal	H. armigera P. canaliculate H. contortus T. colubriformis	Jennings et al., 2005; Plan et al., 2008. Poth et al., 2011
Parigidin-Br1	Palicourea rigida	Insecticidal	Diatraea saccharalis S. frugiperda	Pinto et al., 2012
PA1b	Pisum sativum	Insecticidal	Sitophilus oryzae S. granarius S. zeamays, Culex pipiens. A. pisum Aedes aegypti	Gressent et al., 2007; Eyraud et al., 2013; Eyraud et al., 2017
Sero-X®	C. ternatea	Insecticidal	H. armigera Bemisia tabaci Nezara viridula	Eyraud et al., 2017
Varv A, Varv E, Kalata S	V. odorata	Insecticidal	H. contortus T. colubriformis	Segonzac and Monaghan 2019
Vhl-1	V. hederacea	Insecticidal	H. contortus T. colubriformis	Colgrave et al., 2008
VrCRP	Vigna radiata	Insecticidal	Callosobruchus chinensis	Chen et al., 2002
VrD1	V. radiata	Insecticidal	Bruchidae	Liu et al., 2006

Maximum plant-based ISPs, such as cyclic peptides (CPs), *pea albumin*, defense and recombinant peptides (Grover *et al.*, 2021), are derived from the Rubiaceae, Fabaceae, Violaceae, Solanaceae, and Cucurbitaceae families (Craik 2010). More than 47 CPs from *Clitoria ternatea* (Gilding *et al.*, 2016) showed insecticidal effects. PA1b (*Pea albumin 1 subunit b*) is a 37 amino acid peptide isolated from the seeds of *Pisum sativum* and exhibits significant insecticidal activity against insects such as cereals beetles, *Culex mosquitoes*, and *Aedes aegypti* (Gressent *et al.*, 2011). and certain species of aphids. This toxin binds to subunits c and e of the plasma membrane H-ATPase (V-ATPase) in the insect midgut (Eyraud *et al.*, 2017). In 2017, Sero-X®, the world's first plant-based cyclopeptide bioinsecticide, was developed by his Innovate Ag in Australia (Muttenthaler *et al.*, 2021).

Due to its non-toxic and bee-friendly properties, it is approved for use on Australian cotton and macadamia nut plants to control *Helicoverpa armigera*, *Bemisia tabaci* and *Nezara viridula*. A series of mimic peptides with favorable insecticidal activity (Horodyski *et al.*, 2011; Hardy *et al.*, 2013; Hou *et al.*, 2014; Haruta *et al.*, 2014; Hidefumi *et al.*, 2016; Herzig *et al.*, 2016; Doblas *et al.*, 2017; Heep *et al.*, 2019; Fuminori *et al.*, 2019; Herzig *et al.*, 2020; Hofmann *et al.*, 2021; He *et al.*, 2021) were obtained by modifying natural peptides to overcome bio instability.

Table 10. Classification and Agricultural Bioactivity of Microbes Insecticidal Peptides and their Function

Peptide	Source	Function	Species Effectiveness	References
Beauveriolide I	Beauveria bassiana	Insecticidal	<i>S. litura</i> <i>C. chinensis</i>	Mochizuki et al., 1993
Cyclic depsipeptides	Marine fungi	Insecticidal	<i>S. litura</i> <i>C. chinensis</i>	Wang et al., 2018
Cyclodipeptides	Marine fungi	Insecticidal	<i>Helicoverpa zea</i>	Wang et al., 2017
Destruxins	Metarhizium anisopliae	Insecticidal	Lepidoptera Homoptera Diptera Orthoptera	Hu et al., 2004
Iso-isariin D	B. bassiana	Insecticidal	<i>Artemia salina</i>	Hu et al., 2004
Longibrachin A-I, Longibrachin A-II–b	Trichoderma longibrachiatum RIFAI	Insecticidal	<i>Calliphora vomitoria</i>	Du et al., 2014
Pumilacidin C	Marine bacteria	Insecticidal	<i>A. aegypti</i>	Moreira et al., 2022
SLP1	Streptomyces laindensis H008	Insecticidal	<i>Lipaphis erysimi</i>	Xu et al., 2016

Table 11. Classification and Agricultural Bioactivity of some Others Insecticidal Peptides and their Function

Peptide	Source	Function	Species Effectiveness	References
CAPA-PK analogue (1895 p 2315)	CAPA-PK analogue	Insecticidal	<i>M. persicae</i>	Shi et al., 2022
GS- ω /K-HXTX-Hv1a	Genetic engineering	Insecticidal	<i>Aphid Hrips</i> , <i>Delphacidae</i>	Bomgardner 2017
H17	Allatostatin mimic	Insecticidal	<i>D. punctata</i>	Kai et al., 2009
K-Aib-1	Kinin mimic	Insecticidal	<i>A. pisum</i>	Smagghe et al., 2010
K15, K24, P5, B1, II12, A6	Allatostatin mimic	Insecticidal	<i>D. punctata</i>	Kai et al., 2010; Wang et al., 2018; Wang et al., 2019
Manse-AT	Allatotropin	Insecticidal	<i>M. sexta</i>	Horodyski et al., 2011
Manse-AT (10–13)	Allatotropin	Insecticidal	<i>M. sexta</i>	Kai et al., 2018
PPK-Jo Insecticidal	Kinin analogues	Insecticidal	<i>Moths</i>	Kaczmarek et al., 2021
II-1, IV-3, M1, L25, L7	Kinin mimic	Insecticidal	<i>Aphis glycines</i>	Zhou et al., 2006; Zhang et al., 2015; Zhang et al., 2020; Zhou et al., 2022
2460 analogue	Kinin analogues	Insecticidal	<i>M. persicae</i>	Shi et al., 2022
1963 analogue	Diapause hormone analogue	Insecticidal	<i>H. zea</i>	Reynolds et al., 2019

By introducing unnatural amino acids into the enzyme site, several insect kinin mimetics have been obtained that produce products that are significantly resistant to enzymatic degradation (Nachman *et al.*, 2002). Zhou *et al.* (2022) recently discovered insect kinin analogs L25 and M1 with excellent aphicidal activity and low toxicity to honeybees. The discovery of these peptide-like compounds provides a new strategy for developing new environmentally friendly insecticides.

Herbicidal Peptides (HPs)

In general, weeds cause the highest potential losses (34%) compared to pests and pathogens (18% and 16% losses). Weed control is mechanical or chemical and therefore more effective than controlling animal pests and diseases (Oerke 2006). Traditional herbicides help maintain crop yields, but the heavy reliance on herbicides has resulted in negative impacts such as residues on crops and the environment. Therefore, there is a growing need for new environmentally friendly herbicides (Shi *et al.*, 2020). Tables 12 and 13 present an overview of the functions and classifications of various herbicidal peptides. They are mainly derived from microbes (43.75%), followed by plants (37.50%), and synthetic substances (18.75%). Natural and synthetic peptides are considered promising herbicides for crop protection applications. Bialaphos is a tripeptide isolated and purified from the fermentation broth of *Streptomyces hygroscopicus* (Shi *et al.*, 2020). Thamatomycin A is derived from *Streptomyces acidiscabies*. Several herbicidal peptides are found in plants, such as five dipeptides and one pentapeptide from corn gluten meal hydrolysate (Liu and Christians 1996; Unruh *et al.*, 1997). As shown in Tables 12 and 13, these peptides are effective against a variety of weeds. For example, Romidepsin is active against *Amaranthus palmeri* L. and *Sinapsis arvensis* L. Ala-Ala is active against *Lolium perenne* L. Tentoxin, a cyclic tetrapeptide from *Alternaria tenuis*, inhibits cyclic photosynthetic phosphorylation period and energy transfer, leading to chlorosis in seedlings while chlorosis had no effect on soybeans and corn. Peptides can also be used with herbicides to encourage weeds' stomata to open under unfavorable circumstances, thereby improving the weeds' ability to absorb the drug and causing them to die back more quickly. Thaxomine A, the active ingredient in Opportune™ (<https://marronebio.com/>) developed by Marrone Bio Innovation, is a unique cellulose synthesis inhibitor. In 2013, it was approved by the U.S. Environmental Protection Agency as a non-polluting organic herbicide for weed control in rice and other grain fields.

Table 12. Classification and Agricultural Bioactivity of Plant Herbicidal Peptides

Peptide	Source	Function	Species Effectiveness	References
Ala-Ala	<i>Z. mays</i> L.	Herbicidal	<i>Lolium perenne</i> L.	Unruh et al., 1997
Ala-Asn	<i>Z. mays</i> L	Herbicidal	<i>L. perenne</i> L.	Liu and Christians 1994
Ala-Gln	<i>Z. mays</i> L	Herbicidal	<i>L. perenne</i> L	Liu and Christians 1994
Gly-Ala	<i>Z. mays</i> L	Herbicidal	<i>L. perenne</i> L.	Liu and Christians 1994
Gln-Gln	<i>Z. mays</i> L	Herbicidal	<i>L. perenne</i> L.	Liu and Christians 1994
Leu-Ser-Pro-Ala-Gln	<i>Z. mays</i> L	Herbicidal	<i>L. perenne</i> L	Liu and Christians 1996

Table 13. Classification and Agricultural Bioactivity of Microbes Herbicidal Peptides

Peptide	Source	Function	Species effectiveness	References
AMPB-Ala-Ala-AMPB	Actinomycetes	Herbicidal	Weed	Shi et al., 2020
AMPB-Gly-Ala	Actinomycetes	Herbicidal	Weed	Shi et al., 2020
Basta	Streptomyces viridochromogenes	Herbicidal	Weed	Schwartz et al., 2004
Bialaphos	Actinomycetes	Herbicidal	weed	Shi et al., 2020
Compounds 1-7	Bacillus clausii	Herbicidal	Poa annua L	Guo et al., 2015
DTM1				
des-N2 methylthaxtomin C	- S. scabies	Herbicidal	Agrotis palustri	Shi et al., 2020
Herbiace	S. viridochromogenes	Herbicidal	Weed	Schwartz et al., 2004
Hydroxythaxtomin A	S. scabies	Herbicidal	Lemna minor	Shi et al., 2020
Hydroxythaxtomin C	S. scabies	Herbicidal	L. minor	Shi et al., 2020
Phosalacine	Actinomycetes	Herbicidal	Weed	Shi et al., 2020
Plumebmycin A	Actinomycetes	Herbicidal	Weed	Shi et al., 2020
Plumebmycin B	Actinomycetes	Herbicidal	Weed	Shi et al., 2020
Resormycin	Streptomyces platensis MJ953-SF5	Herbicidal	Dicotyledonous weeds	Shi et al., 2020
Romidepsin	Burkholderia rinojensis	Herbicidal	Amaranthus palmeri L. Sinapsis arvensis L., Amaranthus tuberculatus Trifolium repens L. Conyza canadensis L. Bassia scoparia L. Stellaria media (L.) Abutilon theophrasti Medik. Convolvulus arvensis L. P. annua L. Avena fatua L. Echinochloa crus-galli(L.) P. Beauv. Commelina virginica L. Setaria faberi Herm. Cyperus dif formis L	Owens et al., 2020
Tentoxin	Alternaria tenuis	Herbicidal	weed	Arntzen 1972

Thaxtomin A	S. scabies	Herbicidal	L. minor A. palustris	Shi et al., 2020
Thaxtomin A o-isomer	S. scabies	Herbicidal	L. minor, A. palustris	Shi et al., 2020
Thaxtomin A p-isomer	S. scabies	Herbicidal	A. palustris	Shi et al., 2020
Thaxtomin B	S. scabies	Herbicidal	A. palustris	Shi et al., 2020
Thaxtomin C	S. scabies	Herbicidal	L. minor	Shi et al., 2020
Triaphos	Actinomycetes	Herbicidal	Weed	Shi et al., 2020
Compounds 14,15	Synthesis	Herbicidal	Weed	Dai and Chen 1999
Compound 5a	Synthesis	Herbicidal	Barnyard Crabgrass	Grass Chen et al., 1993

Pest Resistance Peptides (PRPs)

Insects develop resistance to pesticides over time (just as disease-causing bacteria can become resistant to antibiotics), extending the effective lifespan of individual pesticides. For example, the fall armyworm, an annual corn pest that can destroy entire crops, has developed resistance to pesticides (Gutierrez-Moreno *et al.*, 2019; Barber *et al.*, 2023). The peptides used for controlling plant pathogens and their scale of testing are mentioned in Table 14. The development of pest resistance can be minimized with integrated pest management solutions, such as rotating different pesticides through the seasons. However, in many cases, the lack of new pesticides leaves farmers with few options.

Table 14. The Peptides used for Controlling Plant Pathogens and their Scale of Testing

Peptides	Origin	Target Pathogens	Testing Method	Testing Scale	References
Snakin-1	Potato	Clavibacter michiganensis, Botrytis cinerea	Transgenic expression	Lab scale	Segura et al., 1999
MSI-99	Synthetic	F.oxysporum, Sclerotinia sclerotiorum, A. alternata, B. cinerea	Transgenic expression	lab and Green house scale	Chakrabarti et al., 2003
Thionin	Arabidopsis thaliana	Ralstonia solanacearum, Fusarium oxysporum	Transgenic expression	Lab scale	Chan et al., 2005
NmDef02	Nicotiana megalosiphon	Peronospora hyoscyami	In-vitro killing assay, Transgenic expression	Lab, Green house and Field scale	Portieles et al., 2010
Thanatin	Podisus maculiventris	Fusarium graminearum, B. cinerea	In-vitro killing assay	Lab scale	Koch et al., 2012
MsrA-1	Synthetic	Phytophthora cactorum, F. solani	Foliar spray Transgenic expression	lab and Green house scale	Osusky et al., 2000; Rustagi et al., 2014

Cecropin A	Hyalophora cecropia	F. oxysporum, Dickeya dadantii, F. verticillioideis	In-vitro killing assay, Transgenic expression	lab and Green house scale	Cavallarin et al., 1998; Bundo et al., 2014; Montesinos et al., 2016
Melittin	Apis mellifera	X. oryzae	In-vitro killing assay	Lab scale	Shi et al., 2016
Tachyplesin I	Horseshoe crab	Pectobacterium carotovorum	In-vitro killing assay, Transgenic expression	Lab scale	Lipsky et al., 2016
GTannins	Sapium baccatum	Ralstonia solanacearum	In-vitro killing assay, Foliar spray	Lab and greenhouse scale	Vu et al., 2017
BP/BPC serial peptides	Synthetic	Erwinia amylovora, X. vesicatoria, Pseudomonas syringae, F. oxysporum, Penicillium expansum, A. niger, Rhizopus stolonifer, Stemphylium vesicarium	In-vitro killing assay, Foliar spray, Transgenic expression	Lab, Green house and Field scale	Monroc et al., 2006; Badosa et al., 2007; Badosa et al., 2009; Puig et al., 2014; Puig et al., 2015; Montesinos et al., 2017; Montesinos et al., 2021
hCAP18/LL-37	Human	Pectobacterium carotovorum	In-vitro killing assay, Transgenic expression	Lab scale	Jung et al., 2012; Holaskova et al., 2018
PAF26	Synthetic	P. digitatum	In-vitro killing assay	Lab scale	Wang et al., 2018
Random peptides mixture	Synthetic	X. perforans, X. campestris	In-vitro killing assay, Foliar spray	Lab and greenhouse scale	Topman et al., 2018
alfAFP	alfalfa	Verticillium dahliae	Transgenic expression	Lab, Green house and Field scale	Gao et al., 2000
NoPv1	Synthetic	Plasmopara viticola, Phytophthora infestans	In-vitro killing assay, Foliar spray	Lab and greenhouse scale	Colombo et al., 2020
ε-poly-L-lysine	Synthetic	Botrytis cinerea	In-vitro killing assay, Foliar spray	Lab, Green house and Field scale	Sun et al., 2017; Shu et al., 2021
SAMP	Microcitrus australiasica	Liberibacter crescens	In-vitro Killing assay,	Lab scale	Huang et al., 2021

Ac-AMP2	Amaranthus caudatus	Penicillium expansum	Transgenic expression, Foliar spray In-vitro killing assay, Transgenic expression	Lab scale	Huang et al., 2021
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Advantages of Peptide Biologics for Crop Protection

The food supply is at risk when innovation moves more slowly because pests get resistant to current pesticides. The dual challenges are that sustainability and the demand for new products are required, yet environmental safety cannot be compromised. Biologics peptide designs environmentally friendly pesticides that work as effectively as standard synthetic agrochemicals, while also resetting the resistance clock through rapid introduction of more products quickly and more safely. Biopesticides make up a small portion of the \$58 billion crop protection market but are growing more than 15% per year and are expected to rival the synthetic market over the next two decades (Damalas and Koutroubas 2017). The promising potential of biologics as biopesticides has been known for several decades, but challenges in production and distribution prevented their widespread commercialization until recently. Large protein biologics such as antibodies or enzymes have driven a biotechnological revolution in the pharmaceutical industry due to their high efficacy, predictable safety, and ease of development. However, in crop protection, they are less successful as biopesticides because they generally have limited stability in the field, require a cold chain, and have difficulty penetrating the cuticle of insects to achieve essential goals. To ensure a sustainable future and move away from synthetic agricultural chemicals, these challenges must be addressed. Smaller versions of proteins called peptides can overcome this stability and transport issues and target the same receptors as synthetic pesticides. In nature, insect-specific peptide neurotoxins are used by many species of spiders, scorpions, and millipedes to immobilize and kill their prey. This provides a model for the development of safe and effective pesticides (King 2019). Other advantages include the small size of the peptides. This allows for easier passage through external barriers, allows for more efficient manufacturing, and reduces input costs for farmers. Another advantage in manufacturing and delivery is the fact that peptides can be highly stabilized by cross-linking, thus ensuring sustained field performance and stability throughout the supply chain. Stable peptides do not require a cold chain, thus removing a problematic and costly barrier to the use of biologics in crop protection. Importantly, peptide insecticides have low environmental toxicity because they are amino acid building blocks and do not contain toxic metabolites.

Because of their great selectivity for target pests and receptors, there is no chance that vertebrates or helpful insects like bees will unintentionally become harmed.

Challenges and Opportunities for a Peptide Biological Future

Three factors play a role in the widespread commercialization of peptide biopesticides: 1. Bioavailability, 2. Manufacturing cost, and 3. Regulation.

Bioavailability of Peptide

For biologics that target insect receptors, bioavailability remains a significant obstacle to their commercialization (Windley *et al.*, 2012). This challenge can be conceptualized by imagining the pest's external structures such as exoskeleton or intestinal mucosa as a filter that discriminates by size. Large molecules such as proteins and nucleic acids are mostly prevented from entering, but small molecules such as synthetic chemicals can pass through relatively easily. Peptides have an intermediate ability to overcome these barriers because they are the same size as synthetic pesticides and protein biologics. The inherent bioavailability of peptides may be sufficient to directly attack internal receptors such as those in the nervous system. For example, the recently approved peptide GS-w/k-Hctx-Hv1a targets the same receptors as two major classes of synthetic pesticides, and commercially available formulations can kill insects on contact.

Performance Optimization on Structure and Formulation

Improving the stability and bioavailability of natural peptides is an important goal in the discovery of new peptide-based drugs and pesticides. Optimization of the structure of natural peptides and proper formulation can result in more acceptable peptides or their mimetics, and optimization of delivery systems can also result in peptide products with better bioavailability.

Structural Optimization

To overcome the hurdles of low stability and weak activity of natural peptides, various structure optimization methods have been developed, including amino acid substitutions, cyclization strategies, mimetic designs, etc. (Yao *et al.*, 2018; Muttenthaler *et al.*, 2021). Natural peptides can be modified by genetic engineering to obtain new peptides with desirable properties. Such as, the bioinsecticide Spear® was developed using genetic engineering to add a glycine-serine dipeptide to the natural spider venom peptide ω /κ-HCTX-Hv1a. This product has higher activity, lower risk, and longer shelf life than natural products, so it is considered a sustainable and effective green pest control tool (GPCT) in agriculture and public health (Tan and Tong, 2022).

Formulation Development

The development of various formulations such as suspensions, microemulsions, and capsule suspensions can prevent the degradation of peptide molecules due to environmental factors such as water, UV light, temperature, and metabolic enzymes. This not only increases stability but also improves bioavailability as these formulations can easily enter the body through the epidermis and reach target sites (Tan and Tong 2022). Likewise, the rainproof amphiphilic peptide thanatin (THA) AMP dermaseptin 01 (DS01) is tightly anchored to the surface of soybean leaves when sprayed. A fusion of the antimicrobial peptide DS01 and THA (DS01-THA) inhibits the germination of *P. pachyrhizi* spores and alleviates Asian soybean rust in vitro (Schwinges *et al.*, 2019). Moreover, peptide formulations mixed with chemical insecticides through various mechanisms can extend the spectrum of activity and delay resistance (Gonzalez *et al.*, 2002; Lopez-Garcia *et al.*, 2003). Hexapeptide PAFs can inhibit fungi (*Alternaria* sp.) that are not affected by commonly used fungicides (Lopez-Garcia *et al.*, 2003). Hexapeptide 66-10 and heptapeptide derivatives 77-3 and 77-12 can act synergistically with thiabendazole (TBZ) to delay the resistance of *Fusarium sambucinum* to TBZ (Gonzalez *et al.*, 2002).

Delivery Systems (DSs)

Drug delivery systems (DDSs) can deliver the appropriate amount of drug to the target through controlled release technologies such as hydrogels, cubosomes, and nanocarriers, increasing the efficiency of drug utilization (Nordstrom and Malmsten 2017; Martin- Serrano *et al.*, 2019). Easily degraded peptides can be used in combination with new DDSs strategies for precision agriculture. Inject insecticidal peptides into plant lectins or viral coat proteins to improve utilization and insecticidal activity (Nakasu *et al.*, 2014). Herzig *et al.* (2014) delivered insecticidal toxins released by transgenic insect pathogens such as baculovirus, the soil bacterium *Bacillus thuringiensis*, and the fungus *Metarhizium*. These pathogens can infect insects and simultaneously express insecticidal toxins, thereby achieving a synergistic insecticidal effect (Herzig *et al.*, 2014).

Biosynthesis of Peptides (BSPs)

Peptides are generally prepared by chemical synthesis methods called "liquid phase synthesis" and "solid phase synthesis". However, chemical synthesis is expensive and there are limits to large-scale production. Therefore, many studies are aimed at obtaining peptides more economically. Peptide biosynthesis using enzymes, fermentation, and genetic engineering methods is preferred due to its advantages, such as widely available raw materials and low cost (Akbarian *et al.*, 2022). Heterologous systems currently used for peptide production include bacteria such as *Escherichia coli* and *A. subtilis* and fungi such as *Pichia pastoris* and *Saccharomyces cerevisiae*, plants (cell and tissue culture), and related strategies to achieve higher-performance peptide production (Parachin *et al.*, 2012; Narayani *et al.*, 2020). Cyclic peptides are obtained through biosynthesis in fungi, bacteria, and plants (Narayani *et al.*, 2020). Therefore, the production of peptide pesticides by biosynthesis is worthy of further study. Advantages and disadvantages of using antimicrobial peptides to treat plant pathogens described in table 15.

Table 15. Advantages and Disadvantages of using Antimicrobial Peptides to Treat Plant Pathogens

Advantages	Disadvantages
AMPs rapidly kills both fungal and bacterial pathogens at all stages, including spores	The systemic effects of AMPs are weak and cannot eliminate pathogens within the host plant
AMP slowly selects resistant pathogens	Because AMP has a strong inoculating effect, the concentration used must be carefully determined.
AMPs can be easily incorporated into plants through transgenic transformation	Expression of AMPs in plants depletes resources used for growth and reduces yield
AMPs act synergistically with many other antimicrobial agents	Manufacturing AMPs remains relatively expensive
AMPs manipulate symbionts within plants and promote plant growth and development	Long-term exposure to AMPs can lead to the selection of resistant pathogens

Future Concerns

Create modern functional peptides utilizing target-oriented approaches to move forward selectivity against plant pathogens, optimize soundness within the plant environment, and provide low poisonous quality. Design and

validate peptide details to extend the adequacy and shelf life of the peptides. Provide appropriate strategies for conveying the peptide definitions to the plant, particularly with endotherapy devices, to protect trees against endophytic bacterial plant pathogens. Establish a large-scale production processor that is more affordable, maintainable, and capable of fulfilling the demands of producers for plant protection. Conduct field experiments using the most relevant diseases and crops to evaluate and approve the effectiveness and application of plant-protection products made of functional peptides.

Conclusion

The past 120 years have seen the emergence of peptides and the rapid development of peptide-based drugs and pesticides development for human and agriculture development. Global trends are for products with high efficiency, low toxicity, and environmental safety. The field of peptide, pesticides are maturing in several aspects, including long-term and extensive research into the utilization, production, and discovery of numerous agricultural peptides as potential candidates in agriculture. Several promising technologies in structure optimization, formulation, delivery systems, and biosynthesis will continue to contribute to the growth of peptide base pesticides. Peptides that meet high efficiency and safety requirements are useful for crop protection in organic agriculture. The industrialization of peptides is quickened by quick advancements in molecular biology, organic chemistry, synthetic biology, and hereditary building. In addition to X-ray crystallography, AlphaFold2 and RoseTTAFold can now be used to obtain 3D structures of proteins, and other new methods (phage peptide libraries, mRNA display, virtual screening) can be used to identify useful peptides. The developed peptides have high target affinity, meaning that very small amounts of peptides are sufficient to control weeds, pathogens, and insects. Due to the lower dosage of fully or partially fermented peptides, the cost is more competitive than existing pesticides. Peptides can probably become the most important for crop protection. It is not possible in this manuscript to discuss all aspects of peptides as new tools for plant protection in organic agriculture. However, I would like to draw the attention of biochemists, molecular biologists, and agronomists to this new field and promote more detailed and thorough research of peptide-based pesticides for the sustainable development of green agriculture. Synthetic pesticides are too expensive, too slow to develop, and unsustainable for the environment. Biopesticides address these deficiencies. Biopesticides are faster to develop, cheaper, and environmentally sustainable. Peptides offer the safety profile of microbial biopesticides combined with the efficacy of synthetic pesticides. Taken together, these properties make peptides a sustainable alternative to synthetic pesticides. The biotech revolution in agriculture is getting closer and closer. In crop protection, there is a strong desire to move away from synthetic pesticides and toward more sustainable pesticides, but failure to maintain pesticide effectiveness can increase risks to food security. The dual properties of sustainability and efficacy make peptide-based biologics formulation an ideal candidate to replace synthetic drugs and form the backbone of a green future in crop protection.

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References

- Agizzio AP, Machado OLT and Alves EW (2003). A 2S albumin-homologous protein from passion fruit seeds inhibits the fungal growth and acidification of the medium by *Fusarium oxysporum*. *Arch Biochem Biophys*. 416(2):188-195. doi.10.1016/S0003-9861(03)00313-8.
- Akbarian M, Khani A and Eghbalpour S (2022). Bioactive peptides: synthesis, sources, applications, and proposed mechanisms of action. *Int J Mol Sci*. 23(3):1445. doi.10.3390/ijms23031445
- Alan AR and Earle ED (2002). Sensitivity of bacterial and fungal plant pathogens to the lytic peptides, MSI-99, magainin II, and cecropin B. *Mol Plant Microbe Interact*. 15(7):701-708.
- Alan AR, Blowers A and Earle ED (2004). Expression of a magainin-type antimicrobial peptide gene (MSI-99) in tomato enhances resistance to bacterial speck disease. *Plant Cell Rep*. 22(6):388-396.
- Allefs S, Jong E and Florack DEA (1996). *Erwinia* soft rot resistance of potato cultivars expressing antimicrobial peptide tachyplesin I. *Mol Breed*. 2(2):97-105.
- Arce P, Moreno M and Gutierrez M (1999). Enhanced resistance to bacterial infection by *Erwinia carotovora subsp. atroseptica* in transgenic potato plants expressing the attacin or the cecropin SB-37 genes. *Am J Potato Res*. 76(3):169.
- Arntzen CJ (1972). Inhibition of photophosphorylation by tentoxin, a cyclic tetrapeptide. *Biochim Biophys Acta*. 283(3):539-542.
- Badosa E, Ferre R, Frances J, Bardaji E, Feliu L and Planas M (2009). Sporocidal activity of synthetic antifungal undecapeptides and control of penicillium rot of apples. *Appl. Environ. Microbiol*. 75:5563-569. doi: 10.1128/AEM.00711-09.
- Barber DM, Jackson VE, Ling KB (2023). Innovations in crop protection towards sustainable agriculture. *Pest Management Science*. doi.10.1002/ps.7749.
- Badosa E, Ferre R, Planas M, Feliu L, Besalu E and Cabrefiga J (2007). A library of linear undecapeptides with bactericidal activity against phytopathogenic bacteria. *Peptides* 28:2276–2285. doi: 10.1016/j.peptides.2007.09.010.
- Barna B, Leiter E and Hegedus N (2008). Effect of the *Penicillium chrysogenum* antifungal protein (PAF) on barley powdery mildew and wheat leaf rust pathogens: antifungal protein of *P. chrysogenum*. *J Basic Microbiol*. 48(6):516-520. doi. 10.1002/jobm.200800197.
- Bende NS, Dziemborowicz S and Herzig V (2015). The insecticidal spider toxin SF11 is a knottin peptide that blocks the pore of insect voltage-gated sodium channels via a large β -hairpin loop. *FEBS J*. 282(5):904-920. doi. 10.1111/febs.13189
- Bende NS, Dziemborowicz S and Mobli M (2014). A distinct sodium channel voltage- sensor locus determines insect selectivity of the spider toxin Dc1a. *Nat Commun*. 5:4350. doi.10.1104/pp.010685.

- Berrocal-Lobo M, Segura A and Moreno M (2002). Snakin-2, an antimicrobial peptide from potato whose gene is locally induced by wounding and responds to pathogen infection. *Plant Physiol.* 128(3):951-961. doi.10.1104/pp.010685.
- Bhargava A, Osusky M and Hancock RE (2007). Antiviral indolicidin variant peptides: Evaluation for broad-spectrum disease resistance in transgenic *Nicotiana tabacum*. *Plant Sci.* 172(3):515-523. doi.10.1016/j.plantsci.2006.10.016
- Boller T (2005). Peptide signalling in plant development and self/non-self-perception. *Curr Opin Cell Biol.* 17(2):116-122.
- Bomgardner MM (2017). Spider venom: an insecticide whose time has come? *C&EN.* 95(11):30-31.
- Bruce C, Fitches EC and Chougule N (2011). Recombinant conotoxin, TxVIA, produced in yeast has insecticidal activity. *Toxicon.* 58(1):93-100. doi.10.1016/j.toxicon.2011.05.009
- Bundo M, Montesinos L, Izquierdo E, Campo S, Mieulet D and Guiderdoni E (2014). Production of cecropin, an antimicrobial peptide, in rice seed endosperm. *BMC Plant Biol.* 14: 102. . doi: 10.1186/1471-2229-14-102
- Caleya RFD, Gonzalez-Pascual B and Garcia-Olmedo F (1972). Susceptibility of phytopathogenic bacteria to wheat purothionins in vitro. *Appl Microbiol.* 23(5):998-1000.
- Cao X, Liberatore KL, Macalister CA (2015). A cascade of arabinosyltransferases controls shoot meristem size in tomato. *Nat Genet.* 47(7):784-792.
- Capella AN, Menossi M and Arruda P (2001). COI1 affects myrosinase activity and controls the expression of two flower-specific myrosinase-binding protein homologues in Arabidopsis. *Planta.* 213(5):691-699.
- Carillo P, Pannico A and Cirillo C (2022). Protein Hydrolysates from Animal or Vegetal Sources Affect Morpho-Physiological Traits, Ornamental Quality, Mineral Composition, and Shelf-Life of Chrysanthemum in a Distinctive Manner. *Plants.* 11(17):2321. doi.10.3390/plants11172321
- Cary JW, Rajasekaran K and Jaynes JM (2000). Transgenic expression of a gene encoding a synthetic antimicrobial peptide results in inhibition of fungal growth in vitro and in planta. *Plant Sci.* 154(2):171-181. doi.10.1016/S0168-9452(00)00189-8
- Casteels P, Ampe C and Riviere L (1990). Isolation and characterization of abaecin, a major antibacterial response peptide in the honeybee (*Apis mellifera*). *Eur. J. Biochem.* 187(2):381-386. doi.10.1111/j.1432-1033.1990.tb15315.x
- Cavallarin L, Andreu D and San Segundo B. (1998). Cecropin a-derived peptides are potent inhibitors of fungal plant pathogens. *Mol. Plant Microbe Interact.* 11:218-227. doi: 10.1094/MPMI.1998.11.3.218
- Chakrabarti A, Ganapathi TR and Mukherjee PK (2003). MSI-99, a magainin analogue, imparts enhanced disease resistance in transgenic tobacco and banana. *Planta.* 216(4):587-596. doi: 10.1007/s00425-002-0918-y
- Chan Y, Prasad V, Sanjaya, Chen KH, Liu PC and Chan M (2005). Transgenic tomato plants expressing an arabidopsis thionin (thi2.1) driven by fruit-inactive promoter battle against phytopathogenic attack. *Planta* 221:386-393. doi: 10.1007/s00425-004-1459-3
- Chen J, Liu XM and Zhang Y (2018). Venom based neural modulators. *Exp Ther Med.* 15(1):615-619. doi:10.1001/jama.2017.20288
- Chen KC, Lin CY and Kuan CC (2002). A novel defensin encoded by a mungbean cDNA exhibits insecticidal activity against bruchid. *J Agric Food Chem.* 50(25):7258-7263.
- Chen RY, Zhang YH and Chen MR (1993). The Synthesis of Novel Phosphonodipeptides and Their Herbicidal Activity. *Heteroat Chem.* 4(1):1-5. doi.10.1002/hc.520040102
- Cho SK, Larue CT and Chevalier D (2008). Regulation of floral organ abscission in Arabidopsis thaliana. *Procnatl Acadsciusa.* 105(40):15629-15634. doi.10.1073/pnas.0805539105

- Choi CS, Yoe SM and Kim ES (1997). Purification and characterization of antibacterial peptides, spodopsin ia and ib induced in the larval haemolymph of the common cutworm, *Spodoptera litura*. Anim Cell Syst. 1(3):457-462.
- Choi MS, Kim YH and Park HM (2009). Expression of BrD1, a plant defensin from *Brassica rapa*, confers resistance against brown planthopper (*Nilaparvata lugens*) in transgenic Rices. Mol Cell. 28(2):131-137. doi.10.1007/s10059-009-0117-9
- Clark SE, Running MP and Meyerowitz EM (1993). CLAVATA1, a regulator of meristem and flower development in Arabidopsis. Development. 119(2):397-418. doi.10.1242/dev.119.2.397
- Colgrave ML, Kotze AC and Huang YH (2008). Cyclotides: natural, circular plant peptides that possess significant activity against gastrointestinal nematode parasites of sheep. Biochemistry. 47(20):5581-5589. doi.10.1021/bi800223y
- Colgrave ML, Kotze AC and Kopp S (2009). Anthelmintic activity of cyclotides: In vitro studies with canine and human hookworms. Acta Trop. 109(2):163-166. doi.10.1016/j.actatropica.2008.11.003
- Colombo M, Masiero S, Rosa S, Caporali E, Toffolatti SL and Mizzotti S (2020). Nopv1: a synthetic antimicrobial peptide aptamer targeting the causal agents of grapevine downy mildew and potato late blight. Sci. Rep. 10:17574. doi: 10.1038/s41598-020-73027-x
- Combier JP, Kuster H and Journet EP (2008). Evidence for the involvement in nodulation of the two small putative regulatory peptide-encoding genes MtRALFL1 and MtDVL1. Mol Plant-Microbe Interact MPMI. 21(8):1118-1127.
- Covey PA, Subbaiah CC and Parsons RL (2010). A pollen-specific RALF from tomato that regulates pollen tube elongation. Plant Physiol. 153(2):703-715. doi.10.1104/pp.110.155457
- Craik DJ (2010). Discovery and applications of plant cyclotides. Toxicon. 56(7):1092-1102. doi.10.1016/j.toxicon.2010.02.021
- Cruz L, Ribeiro S and Carvalho A (2010). Isolation and partial characterization of a novel lipid transfer protein (LTP) and antifungal activity of peptides from chilli pepper seeds. Protein Pept Lett. 17(3):311-318. doi. 10.2174/092986610790780305
- Dai Q and Chen RY (1999). A Novel Synthesis of (n-Arylsulfonyl)-Phosphonodipeptides Derivatives. Phosphorus Sulfur Silicon Relat Elem. 149(1):237-244. doi.10.1080/10426509908037035
- Damalas C and Koutroubas S (2017). Current status and recent developments in biopesticide use. Agriculture 8(1):13-8010013.
- Datta A, Bhattacharyya D and Singh S (2016). Role of Aromatic Amino Acids in Lipopolysaccharide and Membrane Interactions of Antimicrobial Peptides for Use in Plant Disease Control. J Biol Chem. 291(25):13301-13317. doi.10.1074/jbc.M116.719575
- Datta A, Ghosh A and Airolidi C (2015). Antimicrobial Peptides: Insights into Membrane Permeabilization, Lipopolysaccharide Fragmentation and Application in Plant Disease Control. Sci Rep. 5:11951.
- De Zotti M, Sella L and Bolzonello A (2020). Targeted Amino Acid Substitutions in a Trichoderma Peptaibol Confer Activity against Fungal Plant Pathogens and Protect Host Tissues from Botrytis cinerea Infection. Int J Mol Sci. 21(20):7521. doi.10.3390/ijms21207521
- DeYoung BJ, Bickle KL and Schrage KJ (2006). The CLAVATA1-related BAM1, BAM2 and BAM3 receptor kinase-like proteins are required for meristem function in Arabidopsis. Plant J Cell Mol Biol. 45(1):1-16.
- Diz MSS, Carvalho AO and Rodrigues R (2006). Antimicrobial peptides from chilli pepper seeds causes yeast plasma membrane permeabilization and inhibits the acidification of the medium by yeast cells. Biochim Biophys Acta BBA - Gen Subj. 1760(9):1323-1332. doi.10.1016/j.bbagen.2006.04.010

- Doblas VG, Elwira SL and Fujita S (2017). Root diffusion barrier control by a vasculature- derived peptide binding to the SGN3 receptor. *Science*. 355(6322):280-284. doi.10.1126/science.aaj1562
- Dodueva I, Lebedeva M and Lutova L (2021). Dialog between kingdoms: enemies, allies and peptide phytohormones. *Plants Basel Switz*. 10(11):2243. doi.10.3390/plants10112243
- Du FY, Li XM and Zhang P (2014). Cyclodepsipeptides and Other O-Containing Heterocyclic Metabolites from *Beauveria felina* EN-135, a Marine-Derived Entomopathogenic Fungus. *Mar Drugs*. 12(5):2816-2826. doi.10.3390/md12052816.
- Daneshmand S, Gill IS (2013). Minimally Invasive Post Chemotherapy Retroperitoneal Lymph Node Dissection: Caution and Prudence. *Eur Urol* 2013; 63:1018–9.
- Du T, Wang Y and Hu QX (2005). Transgenic Paulownia Expressing shiva-1 Gene Has Increased Resistance to Paulownia Witches' Broom Disease. *J Integr Plant Biol*. 47(12):1500-1506. doi.10.1111/j.1744-7909.2005.00168.x
- Duman-Scheel M (2019). *Saccharomyces cerevisiae* (Baker's Yeast) as an Interfering RNA Expression and Delivery System. *Curr Drug Targets*. 20(9):942-952. Doi. 10.2174/1389450120666181126123538
- Elakkiya K, Yasodha PB and Justin CGL (2019). Neuropeptides as novel insecticidal agents. *Int J Curr Microbiol Appl Sci*. 8(2):869-878. doi.10.20546/ijcmas.2019.802.098
- Endo A, Sawada Y and Takahashi H (2008). Drought induction of arabidopsis 9-cis- Epoxycarotenoid dioxygenase occurs in vascular parenchyma cells. *Plant Physiol*. 147(4):1984-1993. doi.10.1104/pp.108.116632
- Epple P, Apel K and Bohlmann H (1995). An arabidopsis-thaliana thionin gene is inducible via a signal-transduction pathway different from that for pathogenesis-related proteins. *Plant Physiol*. 109(3):813-820. doi.10.1104/pp.109.3.813
- Eyraud V, Balmand S and Karaki L (2017). The interaction of the bioinsecticide PA1b (*Pea Albumin 1 subunit b*) with the insect V-ATPase triggers apoptosis. *Sci Rep*. 7(1):4902.
- Eyraud V, Karaki L and Rahioui I (2013). Expression and biological activity of the cystine knot bioinsecticide PA1b (*Pea Albumin 1 Subunit b*). *PLoS One*. 8(12): e81619. doi.10.1371/journal.pone.0081619
- Farrokhi N, Whitelegge JP and Brusslan JA (2008). Plant peptides and peptidomics. *Plant Biotechnol J*. 6(2):105-134. doi.10.1111/j.1467-7652.2007. 00315.x
- Fehlbaum P, Bulet P and Michaut L (1994). Insect immunity. Septic injury of *Drosophila* induces the synthesis of a potent antifungal peptide with sequence homology to plant antifungal peptides. *J Biol Chem*. 269(52):33159-33163.
- Fisher K and Turner S (2007). PXY, a receptor-like kinase essential for maintaining polarity during plant vascular-tissue development. *Curr Biol*. 17(12):1061-1066. doi.10.1016/j.cub.2007.05.049
- Fletcher JC, Brand U and Running MP (1999). Signaling of cell fate decisions by CLAVATA3 in Arabidopsis shoot meristems. *Science*. 283(5409):1911-1914.
- Fuglsang AT, Kristensen A and Cuin TA (2014). Receptor kinase-mediated control of primary active proton pumping at the plasma membrane. *Plant J*. 80(6):951-964. doi.10.1111/tpj.12680
- Fuminori, Takahashi and Kousuke (2019). Hormone-like peptides and small coding genes in plant stress signaling and development. *Curr Opin Plant Biol*. 51:88-95. doi.10.1016/j.pbi.2019.05.011
- Gao A, Hakimi SM, Mittanck CA, Wu Y, Woerner BM and Stark DM (2000). Fungal pathogen protection in potato by expression of a plant defensin peptide. *Nat. Biotechnol*. 18:1307-1310. Doi. 10.1038/82436
- Gao BM, Peng C and Lin B (2017). Screening and validation of highly-efficient insecticidal conotoxins from a transcriptome-based dataset of Chinese tubular cone snail. *Toxins*. 9(7):E214. doi.10.3390/toxins9070214.

- Guo Y, Dong J, Zhou T, Auxillos J, Li T, Zhang W, Wang L, Shen Y, Luo Y, Zheng Y, Lin J, Chen GQ, Wu Q, Cai Y, Dai J (2015). eastFab: the design and construction of standard biological parts for metabolic engineering in *Saccharomyces cerevisiae*. *Nucleic Acids Res* 43(13):e88
- Germain H, Chevalier E and Matton DP (2006). Plant bioactive peptides: an expanding class of signaling molecules. *Can J Bot*. 84(1):1-19. doi.10.1139/b05-162
- Gilding EK, Jackson MA and Poth AG (2016). Gene coevolution and regulation lock cyclic plant defence peptides to their targets. *New Phytol*. 210(2):717-730. doi.10.1111/nph.13789
- Gonzalez CF, Provin EM and Zhu L (2002). Independent and synergistic activity of synthetic peptides against thiabendazole-resistant *Fusarium sambucinum*. *Phytopathology*. 92(8):917-924.
- Gressent F, Da Silva P and Eyraud V (2011). *Pea albumin I subunit b* (PA1b), a promising bioinsecticide of plant origin. *Toxins*. 3(12):1502-1517. doi.10.3390/toxins3121502
- Gressent F, Duport G and Rahioui I (2007). Biological activity and binding site characteristics of the PA1b Entomotoxin on insects from different orders. *J Insect Sci*. 7:1-10. doi.10.1673/031.007.1201
- Grover T, Mishra R and Bushra (2021). An insight into biological activities of native cyclotides for potential applications in agriculture and pharmaceuticals. *Peptides*. 135:170430. doi.10.1016/j.peptides.2020.170430
- Gutiérrez-Moreno R, Mota-Sanchez D, Blanco CA, Whalon ME, Teran-Santofimio H, Rodriguez-Maciel JC and DiFonzo C (2019). Field-evolved resistance of the fall armyworm (*Lepidoptera: Noctuidae*) to synthetic insecticides in Puerto Rico and Mexico. *Journal of Economic Entomology* 112(2):792-802.
- Hardy MC, Daly NL and Mobli M (2013). Isolation of an orally active insecticidal toxin from the venom of an *Australian tarantula*. *PLoS One*. 8(9): e73136. doi.10.1371/journal.pone.0073136
- Haruta M, Monshausen G and Gilroy S (2008). A cytoplasmic Ca²⁺ functional assay for identifying and purifying endogenous cell signaling peptides in arabidopsis seedlings: identification of AtRALF1 peptide. *Biochemistry*. 47(24):6311-6321. doi.10.1021/bi8001488
- Haruta M, Sabat G and Stecker K (2014). A peptide hormone and its receptor protein kinase regulate plant cell expansion. *Science*. 343(6169):408-411. doi.10.1126/science.1244454
- He CB, Wang XL and Zhang J (2021). MCRT, a multifunctional ligand of opioid and neuropeptide FF receptors, attenuates neuropathic pain in spared nerve injury model. *Basic Clin Pharmacol Toxicol*. 128(6):731-740. doi.10.1111/bcpt.13566
- Hedges JB and Ryan KS (2020). Biosynthetic pathways to nonproteinogenic alpha-amino acids. *Chem Rev*. 120(6):3161-3209. doi.10.1021/acs.chemrev.9b00408
- Heep J, Skaljic M and Grotmann J (2019). Identification and functional characterization of a novel insecticidal decapeptide from the myrmicine ant *manica rubida*. *Toxins*. 11(10):562. doi.10.3390/toxins11100562
- Herzig V, Bende NS, Alam MS, Kennedy RM and King GF (2014). Chapter eight - methods for deployment of spider venom peptides as bioinsecticides. In: Dhadialla TS, Gill SS, eds. *Advances in Insect Physiology*. Academic Press; 389-411. doi.10.1016/B978-0-12-800197-4.00008-7
- Herzig V, Bende NS, Alam MS, Kennedy RM and King GF (2020). Chapter eight - methods for deployment of spider venom peptides as bioinsecticides. In: Dhadialla TS, Herzig V, Cristofori-Armstrong B, Israel MR, *et al*. *Animal toxins - nature's evolutionary-refined toolkit for basic research and drug discovery*. *Biochem Pharmacol*. 181:114096. doi.10.1016/j.bcp.2020.114096
- Herzig V, Ikonopoulou M and Smith JJ (2016). Molecular basis of the remarkable species selectivity of an insecticidal sodium channel toxin from the African spider *Augacephalus ezendami*. *Sci Rep*. 6:29538.
- Hidefumi, Shinohara and Ayaka (2016). Identification of three LRR-RKs involved in perception of root meristem growth factor in *Arabidopsis*. *Proc Natl Acad Sci USA*. 113(14):3897-3902. doi.10.1073/pnas.1522639113

- Hirakawa Y, Shinohara H and Kondo Y (2008). Non-cell-autonomous control of vascular stem cell fate by a CLE peptide/receptor system. *Proc Natl Acad Sci USA*. 105(39):15208-15213. doi.10.1073/pnas.0808444105d
- Hofmann A, Mingos A and Groth G (2021). Interfering peptides targeting protein-protein interactions in the ethylene plant hormone signaling pathway as tools to delay plant senescence. *Methods Mol Biol Clifton NJ*. 2213:71-85. doi. 10.1007/978-1-0716-0954-5_7
- Holaskova E, Galuszka P, Micuchova A, Sebela M, Oz MT and Frebort I (2018). Molecular farming in barley: development of a novel production platform to produce human antimicrobial peptide LL-37. *Biotechnol. J*. 13:1700628.
- Horodyski FM, Verlinden H and Filkin N (2011). Isolation and functional characterization of an allatotropin receptor from *Manduca sexta*. *Insect Biochem Mol Biol*. 41(10):804-814. doi.10.1016/j.ibmb.2011.06.002
- Hou L, Guo L, Chunling Wang C and Wang C (2016). Genome sequence of *Candida versatilis* and comparative analysis with other yeast. *J Ind Microbiol Biotechnol* 43(8):1131-8.
- Hou SG, Wang X and Chen DH (2014). The secreted peptide PIP1 amplifies immunity through receptor-like kinase 7. *PLoS Pathog*. 10(9):e1004331. doi.10.1371/journal.ppat.1004710
- Hu C, Zhu YF and Cui YW (2018). A group of receptor kinases are essential for CLAVATA signalling to maintain stem cell homeostasis. *Native Plants*. 4(4):205-211.
- Hu QB and Ren SX (2004). Review of Destruixins of *Metarhizium anisopliae* Sorokin. *Chin J Biol Control*. 20(4):234.
- Huan YC, Kong Q and Mou HJ (2020). Antimicrobial peptides: classification, design, application and research progress in multiple fields. *Front Microbiol*. 11:582779.
- Huang C, Araujo K, Sanchez JN, Kund G, Trumble J and Roper S (2021). A stable antimicrobial peptide with dual functions of treating and preventing citrus huanglongbing. *Proc. Natl. Acad. Sci. U.S.A*. 118: e2019628118.
- Huang Y, Nordeen RO and Di M (1997). Expression of an Engineered Cecropin Gene Cassette in Transgenic Tobacco Plants Confers Disease Resistance to *Pseudomonas syringae* pv. *tabaci*. *Phytopathology*. 87(5):494-499.
- Huffaker A, Dafoe NJ and Schmelz EA (2011). ZmPep1, an ortholog of Arabidopsis elicitor peptide 1, regulates maize innate immunity and enhances disease resistance. *Plant Physiol*. 155(3):1325-1338. doi.10.1104/pp.110.166710
- Jaynes J, Nagpala P and DeStefano BL (1993). Expression of a Cecropin-B Lytic Peptide Analog in Transgenic Tobacco Confers Enhanced Resistance to Bacterial Wilt Caused by *Pseudomonas-solanacearum*. *Plant Sci*. 89(1):43-53. doi.10.1016/0168-9452(93)90169-Z
- Jennings CV, Rosengren KJ and Daly NL (2005). Isolation, solution structure, and insecticidal activity of kalata B2, a circular protein with a twist: do Moebius strips exist in nature? *Biochemistry*. 44(3):851-860. doi.10.1021/bi047837h
- Jenssen H, Hamill P and Hancock REW (2006). Peptide antimicrobial agents. *Clin Microbiol Rev*. 19(3):491-511.
- Jung Y, Lee S, Moon Y and Kang K. (2012). Enhanced resistance to bacterial and fungal pathogens by overexpression of a human cathelicidin antimicrobial peptide (hCAP18/LL-37) in Chinese cabbage. *Plant Biotechnol. Rep*. 6:39-46.
- Kaczmarek K, Pacholczyk-Sienicka B and Albrecht L (2021). Solid-Phase Synthesis of an Insect Pyrokinin Analog Incorporating an Imidazoline Ring as Isosteric Replacement of a trans Peptide Bond. *Molecules*. 26(11):3271. doi.10.3390/molecules26113271
- Kai ZP, Huang J and Tobe SS (2009). A potential insect growth regulator: Synthesis and bioactivity of an allatostatin mimic. *Peptides*. 30(7):1249-1253. doi.10.1016/j.peptides.2009.03.010

- Kai ZP, Huang J and Xie Y (2010). Synthesis, Biological Activity, and Hologram Quantitative Structure Activity Relationships of Novel Allatostatin Analogues. *J Agric Food Chem.* 58(5):2652-2658. doi.10.1021/jf902156k
- Kai ZP, Zhu JJ and Deng XL (2018). Discovery of a *Manduca sexta* Allatotropin Antagonist from a *Manduca sexta* Allatotropin Receptor Homology Model. *Molecules.* 23(4):817. doi.10.3390/molecules23040817
- Kamysz W, Krollicka A and Bogucka K (2005). Antibacterial activity of synthetic peptides against plant pathogenic *Pectobacterium* species. *J Phytopathol.* 153(6):313-317. doi.10.1111/j.1439-0434.2005.00976.x
- Kassaw T, Nowak S and Schnabel E (2017). Root determined nodulation1 is required for *M. truncatula* CLE12, but not CLE13 peptide signaling through the Sunn receptor kinase. *Plant Physiol.* 00278. doi.10.1104/pp.17.00278
- Kayes JM and Clark SE (1998). CLAVATA2, a regulator of meristem and organ development in *Arabidopsis*. *Dev Camb Engl.* 125(19):3843-3851. doi.10.1242/dev.125.19.3843
- Kerenga BK, McKenna JA and Harvey PJ (2019). Salt-tolerant antifungal and antibacterial activities of the corn defensin ZmD32. *Front Microbiol.* 10:795.
- King GF (2019). Tying pest insects in knots: The deployment of spider-venom-derived knottins as bioinsecticides. *Pest Management Science* 75(9):2437-2445.
- Kinoshita A, Betsuyaku S and Osakabe Y (2010). RPK2 is an essential receptor-like kinase that transmits the CLV3 signal in *Arabidopsis*. *Dev Camb Engl.* 137(22):3911-3920.
- Koch A, Khalifa W, Langen G, Vilcinskis A, Kogel K and Imani J (2012). The antimicrobial peptide thanatin reduces fungal infections in *Arabidopsis*. *J. Phytopathol.* 160:606-610. doi: 10.1111/j.1439-0434.2012.01946.x
- Kuzina LV, Miller TA and Cooksey DA (2006). In vitro activities of antibiotics and antimicrobial peptides against the plant pathogenic bacterium *Xylella fastidiosa*. *Lett Appl Microbiol.* 42(5):514-520.
- Lay FT and Anderson MA (2005). Defensins—components of the innate immune system in plants. *Curr Protein Pept Sci.* 6(1):85-101. Doi. 10.2174/1389203053027575
- Lay FT, Brugliera F and Anderson MA (2003). Isolation and properties of floral defensins from ornamental tobacco and petunia. *Plant Physiol.* 131(3):1283-1293. doi.10.1104/pp.102.016626
- Lebbe EKM, Peigneur S and Wijesekara I (2014). Conotoxins targeting nicotinic acetylcholine receptors: an overview. *Mar Drugs.* 12(5):2970-3004. doi. 10.3390/md12052970
- Lee SC, Kim SH and Hoffmeister RA (2019). Novel peptide-based inhibitors for microtubule polymerization in *Phytophthora capsici*. *Int J Mol Sci.* 20(11): E2641
- Li J, Kolbasov VG, Lee D, Pang Z, Huang Y and Collins N (2021a). Residue dynamics of streptomycin in citrus delivered by foliar spray and trunk injection and effect on '*Candidatus liberibacter asiaticus*' titer. *Phytopathology* 111:1095-1103.
- Li JP, Hu SP and Jian W (2021b). Plant antimicrobial peptides: structures, functions, and applications. *Botanical Studies.* 62(1):5. doi. 10.1145/3422337.344783
- Li QS, Lawrence CB and Xing HY (2001). Enhanced disease resistance conferred by expression of an antimicrobial magainin analog in transgenic tobacco. *Planta.* 212(4):635-639.
- Li XD, Wang WJ and Liu S (2019). Effects of the peptide H-OOWW-NH₂ and its derived lipopeptide C12-OOWW-NH₂ on controlling of citrus postharvest green mold. *Postharvest Biol Technol.* 158:110979. doi.10.1016/j.postharvbio.2019.110979
- Liang HY, Catranis CM and Maynard CA (2002). Enhanced resistance to the poplar fungal pathogen, *Septoria musiva*, in hybrid poplar clones transformed with genes encoding antimicrobial peptides. *Biotechnol Lett.* 24(5):383-389.

- Lipsky A, Joshi JR, Carmi N and Yedidia I (2016). Expression levels of antimicrobial peptide tachyplesin I in transgenic *Ornithogalum* lines affect the resistance to *Pectobacterium* infection. *J. Biotechnol.* 238:22-29.
- Liu DN and Christians NE (1994). Isolation and identification of root-inhibiting compounds from corn gluten hydrolysate. *J Plant Growth Regul.* 13(4):227-230.
- Liu DN and Christians NE (1996). Bioactivity of a pentapeptide isolated from corn gluten hydrolysate on *Lolium perenne* L. *J Plant Growth Regul.* 15(1):13.
- Liu Q, Ingersoll J and Owens L (2001). Response of transgenic Royal Gala apple (*Malus x domestica* Borkh.) shoots carrying a modified cecropin MB39 gene, to *Erwinia amylovora*. *Plant Cell Rep.* 20(4):306-312.
- Liu SH, Yang L and Zhang C (2010). Purification of peptides with insecticidal activity from the venom of sea anemone *anthopleura xanthogrammica*. *J. Zhejiang Ocean Univ., Nat. Sci.* 29(6):566-571.
- Liu YJ, Cheng CS and Lai SM (2006). Solution structure of the plant defensin VrD1 from mung bean and its possible role in insecticidal activity against bruchids. *Proteins: Struct, Funct, Bioinf.* 63(4):777-786. doi. 10.1002/prot.20962
- Lopez-Garcia B, Veyrat A and Perez-Paya E (2003). Comparison of the activity of antifungal hexapeptides and the fungicides thiabendazole and imazalil against postharvest fungal pathogens. *Int J Food Microbiol.* 89(2-3):163-170.
- Martin-Serrano A, Gomez R and Ortega P (2019). Nanosystems as vehicles for the delivery of antimicrobial peptides (AMPs). *Pharmaceutics.* 11(9):448. doi.10.3390/pharmaceutics11090448
- Marx F (2004). Small, basic antifungal proteins secreted from filamentous ascomycetes: a comparative study regarding expression, structure, function and potential application. *Appl Microbiol Biotechnol.* 65(2):46-56.
- Matsubayashi Y and Sakagami Y (2006). Peptide hormones in plants. *Annu Rev Plant Biol.* 57(1):649-674. doi.10.1146/annurev.arplant.56.032604.144204
- Matsubayashi Y, Ogawa M and Morita A (2002). An LRR receptor kinase involved in perception of a peptide plant hormone, phytosulfokine. *Science.* 296(5572):1470-1472.
- Matsubayashi Y, Shinohara H and Ogawa M (2010). Identification and functional characterization of phytosulfokine receptor using a ligand-based approach. *Chem Rec.* 6(6):356-364. doi.10.1002/tcr.20090.
- Maienfisch P and Koerber K (2024). Recent innovations in crop protection research. *Pest Management Science.* doi.10.1002/ps.8441.
- Mello EO, Ribeiro SFF, Carvalho AO, Santos IS, Da Cunha M and Santa-Catarina C (2011). Antifungal activity of pvd1 defensin involves plasma membrane permeabilization, inhibition of medium acidification, and induction of ROS in fungi cells. *Curr. Microbiol.* 62: 1209-1217. doi: 10.1007/s00284-010-9847-3
- Mingossi FB, Matos JL and Rizzato AP (2010). SacRALF1, a peptide signal from the grass sugarcane (*Saccharum* spp.), is potentially involved in the regulation of tissue expansion. *Plant Mol Biol.* 73(3):271-281. doi. 10.1007/s11103-010-9613-8
- Mita T and Sato Y (2019). Syntheses of alpha-amino acids by using CO₂ as a C1 source. *Chem Asian J.* 14(12):2038-2047. doi.10.1002/asia.201900379
- Mochizuki K, Ohmori K and Tamura H (1993). The Structures of Bioactive Cyclodepsipeptides, Beauveriolides I and II, Metabolites of Entomopathogenic Fungi *Beauveria* sp. *Bull Chem Soc Jpn.* 66(10):3041-3046. doi.10.1246/bcsj.66.3041
- Mohd-Radzman NA, Laffont C and Ivanovici A (2016). Different Pathways Act Downstream of the CEP Peptide Receptor CRA2 to Regulate Lateral Root and Nodule Development. *Plant Physiol.* 171(4):2536-2548. doi.10.1104/pp.16.00113

- Monroc S, Badosa E, Feliu L, Planas M, Montesinos E and Bardaji E (2006). *De Novo* Designed cyclic cationic peptides as inhibitors of plant pathogenic bacteria. *Peptides* 27:2567-2574. doi: 10.1016/j.peptides.2006.04.019
- Montesinos L, Bundo M, Badosa E, San SB, Coca M and Monte E (2017). Production of bp178, a derivative of the synthetic antibacterial peptide bp100, in the rice seed endosperm. *BMC Plant Biol.* 17:63-69. doi: 10.1186/s12870-017-1011-9
- Montesinos L, Bundo M, Izquierdo E, Campo S, Badosa E and Rossignol M (2016). Production of biologically active cecropin, a peptide in rice seed oil bodies. *PloS One* 11: e146919. doi: 10.1371/journal.pone.0146919
- Montesinos L, Gascon B, Ruz L, Badosa E, Planas M and Feliu L (2021). A bifunctional synthetic peptide with antimicrobial and plant elicitation properties that protect tomato plants from bacterial and fungal infections. *Front. Plant Sci.* 12:756357. doi: 10.3389/fpls.2021.756357
- Moreira EA, Rezende-Teixeira P and Albernaz LC (2022). Marine Bacteria from the Southeast Coast of Brazil as a Source of Insecticidal Compounds (September, 10.1007/s43450-022-00293-3, 2022). *Rev Bras Farmacogn.* 32(5):858-858. Doi. 10.1007/s43450-022-00293-3
- Mulinari F, Stanisçuaski F and Bertholdo-Vargas LR (2007). Jaburetox-2Ec: an insecticidal peptide derived from an isoform of urease from the plant *Canavalia ensiformis*. *Peptides.* 28(10):2042-2050. doi. 10.1016/j.peptides.2007.08.009
- Muller R, Bleckmann A and Simon R (2008). The Receptor Kinase CORYNE of Arabidopsis Transmits the Stem Cell–Limiting Signal CLAVATA3 Independently of CLAVATA1. *Plant Cell.* 20(4):934-946. doi.10.1105/tpc.107.057547
- Mun-oz A and Marcos JF (2006). Activity and mode of action against fungal phytopathogens of bovine lactoferricin-derived peptides. *J Appl Microbiol.* 101(6):1199-1207. doi.10.1111/j.1365-2672.2006.03089.x
- Muttenthaler M, King GE and Adams DJ (2021). Trends in peptide drug discovery. *Nat Rev Drug Discov.* 20(4):309-325.
- Nachman RJ, Strey A and Isaac E (2002). Enhanced in vivo activity of peptidase-resistant analogs of the insect kinin neuropeptide family. *Peptides.* 23(4):735-745. doi.10.1016/S0196-9781(01)00654-4
- Nakasu EYT, Nakasu YET and Fitches E (2014). Transgenic plants expressing ω -ACTX- Hv1a and snowdrop Lectin (GNA) fusion protein show enhanced resistance to aphids. *Front Plant Sci.* 5:673.
- Nakayama T, Shinohara H and Tanaka M (2017). A peptide hormone required for Casparian strip diffusion barrier formation in Arabidopsis roots. *Science.* 355(6322):284-286. doi.10.1126/science. aai9057
- Narayani M, Babu R, Anju C (2020). Production of bioactive cyclotides: a comprehensive overview. *Phytochemistry Rev.* 19(4):787-825. doi. 10.1007/s11101-020-09682-9
- Nikonorova N, Yue K and Beeckman T (2018). Arabidopsis research requires a critical re- evaluation of genetic tools. *J Exp Bot.* 69(15):3541-3544. doi.10.1093/jxb/ery161
- Nordstrom R and Malmsten M (2017). Delivery systems for antimicrobial peptides. *Adv Colloid Interface Sci.* 242:17-34. doi. 10.1016/j.cis.2017.01.005
- Oerke EC (2006). Crop losses to pests. *J Agric Sci.* 144:31-43.
- Okuda S, Tsutsui H and Shiina K (2009). Defensin-like polypeptide LUREs are pollen tube attractants secreted from synergid cells. *Nature.* 458(7236):357-361.
- Olsen AN, Mundy J and Skriver K (2002). Peptomics, identification of novel cationic Arabidopsis peptides with conserved sequence motifs. *Silico Biol.* 2(4):441-451.

- Osusky M, Osuska L and Hancock RE (2004). Transgenic potatoes expressing a novel cationic peptide are resistant to late blight and pink rot. *Transgenic Res.* 13(2):181-190.
- Osusky M, Osuska L and Kay W (2005). Genetic modification of potato against microbial diseases: in vitro and in planta activity of a dermaseptin B1 derivative. *MsrA2. Theor Appl Genet.* 111(4):711-722. doi. 10.1007/s00122-005-2056-y
- Osusky M, Zhou GQ and Osuska L (2000). Transgenic plants expressing cationic peptide chimeras exhibit broad-spectrum resistance to phytopathogens. *Nat Biotechnol.* 18(11):1162-1166.
- Ou Y, Lu XT and Zi Q (2016). RGF1 INSENSITIVE 1 to 5, a group of LRR receptor-like kinases, are essential for the perception of root meristem growth factor 1 in *Arabidopsis thaliana*. *Cell Res.* 26(6):686-698. . doi:10.1038/cr.2016.63.
- Ouellette AJ (2006). Paneth cell alpha-defensin synthesis and function. *Curr Top Microbiol Immunol.* 306:1-25. doi.10.1007/3-540-29916-5_1.
- Owens DK, Bajsa-Hirschel J and Duke SO (2020). The Contribution of Romidepsin to the Herbicidal Activity of *Burkholderia rinojensis* Biopesticide. *J Nat Prod.* 83(4):843-851. doi/10.1021/acs.jnatprod.9b00405
- Palm C, Netzereab S and Heallbrink M (2006). Quantitatively determined uptake of cell- penetrating peptides in non-mammalian cells with an evaluation of degradation and antimicrobial effects. *Peptides.* 27(7):1710-1716. doi.10.1016/j.peptides.2006.01.006.
- Parachin NS, Mulder KC, Americo Barbosa Viana Antonio, Dias SC and Franco OL (2012). Expression systems for heterologous production of antimicrobial peptides. *Peptides.* 38(2):446-456. doi.10.1016/j.peptides.2012.09.020.
- Park SC, Park Y and Hahm KS (2011). The role of antimicrobial peptides in preventing multidrug-resistant bacterial infections and biofilm formation. *Int J Mol Sci.* 12(9):5971-5992. doi. 10.3390/ijms12095971.
- Patel P, Benzle K, Pei D, Wang GL (2024). Cell-penetrating peptides for sustainable agriculture. *Trends in Plant Science.* 29(10):1131-1144. doi.10.1016/j.tplants.2024.05.011.
- Pearce AK, Crimmins K, Groussac E, Hewlins MJE, Dickinson JR, Francois J, Booth IR, Brown AJP *et al.* (2001) Pyruvate kinase (Pyk1) levels influence both the rate and direction of carbon flux in yeast under fermentative conditions. *Microbiology (Reading)* 147(Pt 2):391-401.
- Pinheiro AM, Carreira A and Ferreira RB (2018). Fusion proteins towards fungi and bacteria in plant protection. *Microbiology.* 164(1):11-19. doi.10.1099/mic.0.000592
- Pinto MFS, Fensterseifer ICM and Migliolo L (2012). Identification and Structural Characterization of Novel Cyclotide with Activity against an Insect Pest of Sugar Cane. *J Biol Chem.* 287(1):134-147. doi.10.1074/jbc.M111.294009
- Plan MRR, Saska I, Cagauan AG (2008). Backbone cyclised peptides from plants show molluscicidal activity against the rice pest *Pomacea canaliculata* (golden apple snail). *J Agric Food Chem.* 56(13):5237-5241. doi.org/10.1021/jf800302f
- Portieles R, Ayra C, Gonzalez E, Gallo A, Rodriguez R and Chacón O (2010). Nmdef02, a novel antimicrobial gene isolated from *Nicotiana megalosiphon* confers high-level pathogen resistance under greenhouse and field conditions. *Plant Biotechnol. J.* 8:678-690.
- Poth AG, Colgrave ML and Lyons RE (2011). Discovery of an unusual biosynthetic origin for circular proteins in legumes. *Proc Natl Acad Sci USA.* 108(25):10127-10132. doi.10.1073/pnas.1103660108
- Puig M, Moragrega C, Ruz L, Montesinos E and Llorente I (2014). Postinfection activity of synthetic antimicrobial peptides *Against Stemphylium Vesicarium* in pear. *Phytopathology* 104: 1192-1200. doi: 10.1094/PHYTO-02-14-0036-R

- Puig M, Moragrega C, Ruz L, Montesinos E and Llorente I (2015). Controlling brown spot of pear by a synthetic antimicrobial peptide under field conditions. *Plant Dis.* 99:1816-1822. doi: 10.1094/PDIS-03-15-0250-RE
- Rajasekaran K, Cary JW and Jaynes JM (2005). Disease resistance conferred by the expression of a gene encoding a synthetic peptide in transgenic cotton (*Gossypium hirsutum* L.) plants. *Plant Biotechnol J.* 3(6):545-554. doi.10.1111/j.1467-7652.2005.00145.x
- Reynolds JA, Nachman RJ and Denlinger DL (2019). Distinct microRNA and mRNA responses elicited by ecdysone, diapause hormone and a diapause hormone analog at diapause termination in pupae of the corn earworm, *Helicoverpa zea*. *Gen Comp Endocrinol.* 278:68-78. doi.10.1016/j.ygcen.2018.09.013
- Ribeiro SM, Almeida RG and Pereira CA (2011). Identification of a *Passiflora alata* Curtis dimeric peptide showing identity with 2S albumins. *Peptides.* 32(5):868-874. doi.10.1016/j.peptides.2010.10.011
- Rivera-de-Torre E, Rimbault C and Jenkins TP (2021). Strategies for heterologous expression, synthesis, and purification of animal venom toxins. *Front Bioeng Biotechnol.* 9:811905. doi.10.3389/fbioe.2021.811905
- Roy S, Griffiths M and Torres-Jerez I (2022). Application of Synthetic Peptide CEP1 Increases Nutrient Uptake Rates Along Plant Roots. *Front Plant Sci.* :12. doi.10.1215/17432197-9516897
- Rustagi A, Kumar D, Shekhar S, Yusuf MA, Misra S and Sarin BN (2014). Transgenic *Brassica juncea* plants expressing msra1, a synthetic cationic antimicrobial peptide, exhibit resistance to fungal phytopathogens. *Mol. Biotechnol.* 56:535-545. doi: 10.1007/s12033-013-9727-8
- Ryan CA, Pearce G and Scheer J (2002). Polypeptide hormones. *Plant Cell.* 14(Suppl):s251-s264. doi.10.1105/tpc.010484
- Sagehashi Y, Takaku H, Yatou O (2017). Partial peptides from rice defensin OsAFP1 exhibited antifungal activity against the rice blast pathogen *Pyricularia oryzae*. *J Pestic Sci.* 42(3-4):172-175.
- Santiago J, Brandt B and Wildhagen M (2016). Mechanistic insight into a peptide hormone signaling complex mediating floral organ abscission. *ELife Sci.* 5:e15075.
- Schaefer SC, Gasic K and Cammue B (2005). Enhanced resistance to early blight in transgenic tomato lines expressing heterologous plant defense genes. *Planta.* 222(5):858-866. doi. 10.1007/s00425-005-0026-x
- Schwartz D, Berger S, Heinzelmann E (2004). Biosynthetic gene cluster of the herbicide phosphinothricin tripeptide from *Streptomyces viridochromogenes* Tu494. *Appl Environ Microbiol.* 70(12):7093-7102.
- Schmelz EA, Carroll MJ and LeClere S (2006). Fragments of ATP synthase mediate plant perception of insect attack. *Proc Natl Acad Sci USA.* 103(23):8894-8899. doi. 10.1073/pnas.0602328103
- Schmelz EA, Huffaker A and Carroll MJ (2012). An amino acid substitution inhibits specialist herbivore production of an antagonist effector and recovers insect- induced plant defenses. *Plant Physiol.* 160(3):1468-1478. doi. 10.1104/pp.112.201061
- Schwinges P, Pariyar S and Jakob F (2019). A bifunctional dermaseptin–thanatin dipeptide functionalizes the crop surface for sustainable pest management. *Green Chem.* 21(9):2316-2325. doi.10.1039/c9gc00457b
- Segonzac C and Monaghan J (2019). Modulation of plant innate immune signaling by small peptides. *Curr Opin Plant Biol.* 51:22-28. doi. 10.1016/j.pbi.2019.03.007
- Segura A, Moreno M, Madueno F, Molina A and García-Olmedo F (1999). Snakin-1, a peptide from potato that is active against plant pathogens. *Mol. Plant-Microbe Interact.* 12:16-23. doi: 10.1094/MPMI.1999.12.1.16
- Seo M, Peeters A and Koiwai H (2000). The Arabidopsis aldehyde oxidase 3 (AAO3) gene Seo HH, Park S and Park S (2014). Overexpression of a defensin enhances resistance to a fruit-specific anthracnose fungus in pepper. *PLoS One.* 9(5):e97936. doi.10.1371/journal.pone.0097936

- Sharma A, Sharma R and Imamura M (2000). Transgenic expression of cecropin B, an antibacterial peptide from *Bombyx mori*, confers enhanced resistance to bacterial leaf blight in rice. *FEBS Lett.* 484(1):7-11. doi.10.1016/S0014-5793(00)02106-2.
- Srivastava A and Thomson SB (2009) Framework Analysis: A Qualitative Methodology for Applied Policy Research. *Journal of Administration and Governance*, 4:72-79.
- Shi LQ, Wu ZY and Zhang YN (2020). Herbicidal secondary metabolites from actinomycetes: structure diversity, modes of action, and their roles in the development of herbicides. *J Agric Food Chem.* 68(1):17-32. doi.10.1021/acs.jafc.9b06126
- Shi W, Li C, Li M, Zong X, Han D and Chen Y (2016). Antimicrobial peptide melittin against *Xanthomonas oryzae* pv. *Oryzae*, the bacterial leaf blight pathogen in rice. *Appl. Microbiol. Biotechnol.* 100:5059-5067. doi: 10.1007/s00253-016-7400-4
- Shi Y, Pandit A and Nachman RJ (2022). Transcriptome analysis of neuropeptides in the beneficial insect lacewing (*Chrysoperla carnea*) identifies kinins as a selective pesticide target: a biostable kinin analogue with activity against the peach potato aphid *Myzus persicae*. *J Pest Sci.* 1-12.
- Shu C, Cui K, Li Q, Cao J and Jiang W (2021). Epsilon-poly-l-lysine (ϵ -pl) exhibits multifaceted antifungal mechanisms of action that control postharvest alternaria rot. *Int. J. Food Microbiol.* 348:109224. doi: 10.1016/j.ijfoodmicro.2021.109224
- Shuipys T, Carvalho RF and Clancy MA (2019). A synthetic peptide encoded by a random DNA sequence inhibits discrete red-light responses. *Plant Direct.* 3(10):e00170.
- Smagghe G, Mahdian K and Zubrzak P (2010). Antifeedant activity and high mortality in the pea aphid *Acyrtosiphon pisum* (Hemiptera: Aphidae) induced by biostable insect kinin analogs. *Peptides.* 31(3):498-505. doi.10.1016/j.peptides.2009.07.001
- Song W, Liu L and Wang JZ (2016). Signature motif-guided identification of receptors for peptide hormones essential for root meristem growth. *Cell Res.* 26(6):674-685. doi:10.1038/cr.2016.6
- Stahl Y, Grabowski S and Bleckmann A (2013). Moderation of Arabidopsis root stemness by CLAVATA1 and ARABIDOPSIS CRINKLY4 receptor kinase complexes. *Curr Biol CB.* 23(5):362-371. doi.10.1016/j.cub.2013.01.045
- Stegmann M, Monaghan J and Smakowska-Luzan E (2017). The receptor kinase FER is a RALF-regulated scaffold controlling plant immune signaling. *Science.* 355(6322):287-289. doi. 10.1126/science.aal2541
- Stenvik GE, Tandstad NM and Guo YF (2008). The EPIP peptide of INFLORESCENCE DEFICIENT IN ABSCISSION is sufficient to induce abscission in arabidopsis through the receptor-like kinases HAESA and HAESA-LIKE2. *Plant Cell.* 20(7):1805-1817. doi.10.1105/tpc.108.059139
- Stuhrwoldt N and Schaller A (2019). Regulation of plant peptide hormones and growth factors by post-translational modification. *Plant Biol.* 21(S1):49-63. doi.org/10.1105/tpc.108.059139
- Sun G, Wang Y, Wang G, Xiang L and Qi J (2017). A New Anti-Aging Lysophosphatidic Acid from Arabidopsis thaliana. *Med Chem.* 13(7):641-647. doi: 10.1016/j.peptbp.2017.07.007
- Takahashi F, Suzuki T and Osakabe Y (2018). A small peptide modulates stomatal control via abscisic acid in long-distance signalling. *Nature.* 556(7700):235-238.
- Tang R, Tan H, Dai Y, Li L, Huang Y, Yao H, Cai Y and Yu G (2023). Application of antimicrobial peptides in plant protection: making use of the overlooked merits. *Front. Plant Sci.* 14:1-11. doi.10.3389/fpls.2023.1139539.
- Tan HJ and Tong YL (2022). Progress of research, development, and application on GS-omega/ kappa-HXTX-Hv1a, a new polypeptide biological insecticide. *World Pestic.* 44(7):13.

- Topman S., Tamir-Ariel D., Bochnic-Tamir H., Stern Bauer T., Shafir S., Burdman S., *et al.* (2018). Random peptide mixtures as new crop protection agents. *Microb. Biotechnol.* 11, 1027–1036. doi: 10.1111/1751-7915.13258
- Toth L, Varadi G and Boros E (2020). Biofungicidal Potential of Neosartorya (*Aspergillus*) Fischeri Antifungal Protein NFAP and Novel Synthetic γ -Core Peptides. *Front Microbiol.* 11:820.
- Troyano FJA, Merkens K and Anwar K (2021). Radical-based synthesis and modification of amino acids. *Angew Chem, Int Ed.* 60(3):1098-1115. doi.10.1002/anie.202010157
- Tudi M, Daniel Ruan H and Wang L (2021). Agriculture development, pesticide application and its impact on the environment. *Int J Environ Res Publ Health.* 18(3):1112. doi.10.3390/ijerph18031112
- Turrini A, Sbrana C and Pitto L (2004). The antifungal Dm-AMP1 protein from *Dahlia merckii* expressed in *Solanum melongena* is released in root exudates and differentially affects pathogenic fungi and mycorrhizal symbiosis. *New Phytol.* 163(2):393-403.
- Unruh JB, Christians NE and Horner HT (1997). Herbicidal effects of the dipeptide alanine- alanine on perennial ryegrass (*Lolium perenne* L.) seedlings. *Crop Sci.* 37(1): crops1997.0011183X003700010035x.
- Urdangarin MC, Norero NS and Broekaert WF (2000). A defensin gene expressed in sunflower inflorescence. *Plant Physiol Biochem.* 38(3):253-258. doi.10.1016/S0981-9428(00)00737-3
- Van der Weerden NL Hancock REW and Anderson MA (2010). Permeabilization of fungal hyphae by the plant defensin NaD1 occurs through a cell wall-dependent process. *J Biol Chem.* 285(48):37513-37520. doi. 10.1074/jbc.M110.134882
- Van der Weerden NL, Lay FT and Anderson MA. (2008). The plant defensin, nad1, enters the cytoplasm of *Fusarium oxysporum* hyphae. *J. Biol. Chem.* 283:14445-14452. Doi.10.1074/jbc.M709867200
- Vila-Perello M, Sanchez-Vallet A and García-Olmedo F (2003). Synthetic and structural studies on *Pyrularia pubera* thionin: a single-residue mutation enhances activity against Gram-negative bacteria. *FEBS Lett.* 536(1-3):215-219. doi.10.1016/S0014-5793(03)00053-X
- Vu TT, Kim H, Tran VK, Vu HD, Hoang TX, Han JW (2017). Antibacterial activity of tannins isolated from *Sapium baccatum* extract and use for control of tomato bacterial wilt. *PloS One* 12, e181499. Doi.10.1371/journal.pone.0181499
- Wang C, Steenhuyse-Vandeveld M, Lin CC and Billen J (2021). Morphology of the novel basimandibular gland in the ant genus *Strumigenys* (Hymenoptera: Formicidae). *Insects* 12:50. 10.3390/insects12010050. doi.org/10.3390/insects12010050
- Wang HX and Ng TB (2005). An antifungal peptide from the coconut. *Peptides.* 26(12):2392-2396. doi.10.1016/j.peptides.2005.05.009
- Wang JZ, Li HJ and Han ZF (2015). Allosteric receptor activation by the plant peptide hormone phytosulfokine. *Nature.* 525:265-268
- Wang MZ, Li XL and Chen MT (2019). 3D-QSAR based optimization of insect neuropeptide allatostatin analogs. *Bioorg Med Chem Lett.* 29(7):890-895. doi.10.1016/j.bmcl.2019.02.001
- Wang MZ, Zhang L and Wang XW (2018). Exploring the N-terminus region: Synthesis, bioactivity and 3D-QSAR of allatostatin analogs as novel insect growth regulators. *Chin Chem Lett.* 29(9):1375-1378. doi.10.1016/j.ccl.2017.11.022
- Wang SL, Nehring R and Mosheim R (2018). Agricultural productivity growth in the United States: 1948-2015.
- Wang XH, Gong X and Li P (2018). Structural Diversity and Biological Activities of Cyclic Dipeptides from Fungi. *Molecules.* 23(1):169. doi.10.1016/j.foodcont.2018.01.008

- Wang XH, Li YY and Zhang XP (2017). Structural Diversity and Biological Activities of the Cyclodipeptides from Fungi. *Molecules*. 22(12):2026. doi.10.3390/molecules22122026
- Wen QJ, Sun ML and Kong XL (2021). The novel peptide NbPPII identified from *Nicotiana benthamiana* triggers immune responses and enhances resistance against *Phytophthora* pathogens. *J Integr Plant Biol*. 63(5):961-976. doi.10.1111/jipb.13033
- Windley MJ, Herzig V and Dziemborowicz SA (2012). Spider-venom peptides as bioinsecticides. *Toxins*. 4(3):191-227. doi.10.3390/toxins4030191
- Wrzaczek M, Vainonen JP and Stael S (2015). GRIM REAPER peptide binds to receptor kinase PRK5 to trigger cell death in *Arabidopsis*. *EMBO J*. 34(1):55-66. doi.10.15252/embj.201488582
- Wu QH, Patocka J and Kuca K (2018). Insect antimicrobial peptides, a mini review. *Toxins*. 10(11):461. doi.10.3390/toxins10110461
- Xu C, McDowell NG, Fisher RA, Wei L, Sevanto S, Christoffersen BO, Weng E and Middleton RS (2019a). Increasing impacts of extreme droughts on vegetation productivity under climate change. *Nat. Clim. Change*. 9(12):948-953.
- Xu LJ, Liang KK and Duan BS (2016). A Novel Insecticidal Peptide SLP1 Produced by *Streptomyces laindensis* H008 against *Lipaphis erysimi*. *Molecules*. 21(8): 1101. doi.10.3390/molecules21081101
- Xu Q, Ye X and Ma X (2019b). Engineering a peptide aptamer to target calmodulin for the inhibition of *Magnaporthe oryzae*. *Fungal Biol*. 123(7):489-496. doi.10.1016/j.funbio.2019.04.005
- Yan F, Cheng X and Ding XZ (2014). Improved insecticidal toxicity by fusing Cry1Ac of *Bacillus thuringiensis* with Av3 of *Anemonia viridis*. *Curr Microbiol*. 68(5): 604-609. doi.10.1007/s00284-013-0516-1
- Yao JF, Yang H and Zhao YZ (2018). Metabolism of peptide drugs and strategies to improve their metabolic stability. *Curr Drug Metabol*. 19(11):892-901. doi. 10.2174/1389200219666180628171531
- Yevtushenko DP and Misra S (2007). Comparison of pathogen-induced expression and efficacy of two amphibian antimicrobial peptides, MsrA2 and temporin A, for engineering wide-spectrum disease resistance in tobacco. *Plant Biotechnol J*. 5(6):720-734. doi.10.1111/j.1467-7652.2007. 00277.x
- Yevtushenko DP, Romero R and Forward BS (2005). Pathogen-induced expression of a cecropin A-melittin antimicrobial peptide gene confers antifungal resistance in transgenic tobacco. *J Exp Bot*. 56(416):1685-1695. doi.10.1111/j.1467-7652.2007. 00277.x
- Yi JY, Seo HW and Yang MS (2004). Plant defense gene promoter enhances the reliability of shiva-1 gene-induced resistance to soft rot disease in potato. *Planta*. 220(1):165-171.
- Yoshii, Tomoaki and Tabata (2014). Perception of root-derived peptides by shoot LRR- RKs mediates systemic N-demand signaling. *Science*. 346:343-346.
- Yu HH, Li RF and Wang XQ (2021). Field experiment effect on citrus spider mite *panonychus citri* of venom from jellyfish *nemopilema nomurai*: the potential use of jellyfish in agriculture. *Toxins*. 13(6):411. doi.10.3390/toxins1
- Yu HH, Liu XG and Dong XL (2005). Insecticidal activity of proteinous venom from tentacle of jellyfish *Rhopilema esculentum* Kishinouye. *Bioorg Med Chem Lett*. 15(22):4949-4952. doi.10.1016/j.bmcl.2005.08.015
- Yu LL, Liu Y and Liu YM (2016). Overexpression of phytosulfokine- α induces male sterility and cell growth by regulating cell wall development in *Arabidopsis*. *Plant Cell Rep*. 35(12):2503-2512. doi.10.1007/s00299-016-2050-7
- Zakharchenko NS, Rukavtsova EB and Gudkov AT (2005). Enhanced resistance to phytopathogenic bacteria in transgenic tobacco plants with synthetic gene of antimicrobial peptide cecropin P1. *Genetika*. 41(11):1445-1452.

- Zhang CL, Li XL and Song DL (2020). Synthesis, aphicidal activity and conformation of novel insect kinin analogues as potential eco-friendly insecticides. *Pest Manag Sci.* 76(10):3432-3439. doi.10.1002/ps.5721.
- Zhang CL, Qu YY, Wu XQ (2015). Eco-Friendly Insecticide Discovery via Peptidomimetics: Design, Synthesis, and Aphicidal Activity of Novel Insect Kinin Analogues. *J Agric Food Chem.* 63(18):4527–4532. doi.10.1021/acs.jafc.5b01225.
- Zhang YM, Ye DX, Liu Y, Zhang XY, Zhou YL, Zhang L, Yang XL (2023). Peptides, new tools for plant protection in eco-agriculture. *Advanced Agrochem.* 2(1): 58-78. doi.10.1016/j.aac.2023.01.003.
- Zhao P, Ren A and Dong P (2018). Antimicrobial Peptaibols, Trichokonins, Inhibit Mycelial Growth and Sporulation and Induce Cell Apoptosis in the Pathogenic Fungus *Botrytis cinerea*. *Appl Biochem Microbiol.* 54(4):396-403. doi. 10.1134/S0003683818040154
- Zhou S, Zhao S, and Jia F (2006). A taxonomic study on the ant genus *Pheidologeton* Mayr (Hymenoptera, Formicidae, Myrmicinae) from China. *Acta Zootaxonomy Sinica* 31:870-873.
- Zhou YL, Zhang YM and Zhang YH (2022). Insect kinin mimics act as potential control agents for aphids: structural modifications of Trp (4). *J Pept Sci.* 29(1):e3444. doi.10.1002/psc.3444

REVIEW ARTICLE

Challenges and Opportunities of Bangladesh Agriculture Sector: The Role of Chemical Fertilizer and Scope of Organic Farming

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Abstract

Agriculture is the base of Bangladesh's economy, supporting livelihoods and ensuring food demand for its growing population. However, this sector is facing some challenges including soil and environmental degradation due to heavy reliance on chemical fertilizer. This paper will explore the critical role of chemical fertilizer to boost the yield of crops and to ensure food security. Simultaneously, it highlights the crying need for organic farming to ensure the long-term preservation of soil and environmental quality, including the limitations of organic farming in Bangladesh. Key opportunities are identified to improve the agriculture sector, including the implementation of balanced fertilizer management, promotion of government policies and creating awareness among the producers. The paper emphasizes the need for an integrated approach that combines the use of chemical fertilizer with organic farming to ensure the long-term sustainability of the environment in Bangladesh.

Keywords: Bangladesh's agriculture, conventional farming, organic farming, chemical fertilizer, sustainable agriculture, soil health

Introduction

Agriculture is a cornerstone of Bangladesh's economy, promoting food security, providing employment and improving the livelihoods of the nation (BBS, 2023). The fields of Bangladesh are more than soil and crops; they are the base of nations survival and success. Agriculture contributes 11.60% of total GDP (BER, 2024), along with employment sources of more than 40% of total employment (ADB, 2023). About 1.25 million individuals are directly employed by it, while 12 million rural dwellers indirectly benefit from it (Chowhan et al., 2024). Because of the nation's large proportion of impoverished and vulnerable populations, high population density, susceptibility to climate change and dependency on agriculture, food security is crucial for social stability. Additionally, at the national level, 0.82% of households are severely food insecure and 21.92% have moderate or severe food insecurity (BBS, 2023). It has been reported from long-term integrated nutritional management experiments that fertilizer accounted for 35% of the total rice yield in transplanted *aman* season and 50% of the total rice yield in *boro* season (Naher et al., 2015). However, Bangladesh has significantly developed food security over the last two decades by increasing food production, especially rice production through the increased use of chemical fertilizer and better irrigation management strategies (Shahabuddin, 2016). High population pressure significantly contributed to increases in the use fertilizer in densely populated areas. More specifically, a 1% increment in population pressure increases fertilizer use by 0.118% (Xiang et al., 2020). It is expected that the population of Bangladesh will exceed

200 million in 2050 with a population density of 1355 km², amplifying the dependency on chemical fertilizers for crop production (Karim et al., 2024). Nonetheless, long-term fertilization has detrimental effects on the environment such as soil salinity, heavy metal accumulation, eutrophication, nitrate accumulation, greenhouse gas emissions, and biomagnification. The excessive use of chemical fertilizers has also given rise to many interrelated problems including soil, water, and air pollution. In response, organic farming is a viable alternative to protect the ecosystem from long-term degradation. Organic farming, with its emphasis on the use of natural inputs and sustainable practices, offers the potential to mitigate the adverse effects of extensive use of chemical fertilizer. This review examines the role of chemical fertilizers in Bangladesh's agri sector, exploring the impact and emerging opportunities for organic farming. By analysing the current use and trends of agriculture, this review aims to provide insights into the challenges and opportunities of climate resilience farming in Bangladesh.

Materials and Methods

This review was conducted by systematically searching of academic databases based on keywords like organic farming problems and prospects, chemical fertilizer, including Google Scholar, ResearchGate, ScienceDirect, and PubMed, along with different national and international sources like the Bangladesh Bureau of Statistics (BBS), Bangladesh Economic Review (BER), Food and Agriculture Organization (FAO), and International Federation of Organic Agriculture Movements (IFOAM). Keywords such as chemical fertilizers in Bangladesh, organic farming in Bangladesh, challenges of organic farming and the role of synthetic fertilizers in food security were used to identify relevant literature. The study used statistical tools to compare the consumption pattern of chemical fertilizers with grain production trends. Environmental and economic impacts of fertilizer use assessed through existing literature.

Results and discussion

Present status of chemical fertilizer consumption and grain production

There are several factors to increase fertilizer consumption in Bangladesh. First of all, the green revolution in the 1980s, facilitated by high-yielding crop varieties, especially rice and wheat, drove fertilizer consumption remarkably. Additionally, in 1995, authorities started to provide free fertilizer among the producers along with subsidies, which further exacerbated the issue. Furthermore, the widespread fertilizer industry increment and retailer network increased the accessibility of fertilizer to the producers, which ultimately enhanced fertilizer utilization in our country. However, the government's favorable policies regarding fertilizer allocation and price regulation, along with the supply of fertilizer-responsive HYV rice to the nation in the mid-1960s, caused the demand for fertilizer to go up from 875.179 to 6825.55 metric tons (BARC, 2018; BER, 2024) between 1980-81 and 2022-23 (up to February 2023). According to BARC (2018) and BER (2024), the use of urea, Triple Super Phosphate (TSP), Muriate of Potash (MoP) and Diammonium Phosphate (DAP) fertilizers increased by 559.776 to 2286.00, 215.061 to 674.00, 452.04 to 826.00 and 417.36 to 1427 metric tons, respectively, between 1980-81 and 2022-23. The widespread accessibility of inputs that increase crop output and the rapid advancement in growing technologies over the years have made Bangladeshi agriculture more reliant on chemical fertilizers to boost agricultural productivity. In Bangladesh, fertilizer is one of the essential components of soil nutrients and is crucial to crop productivity. Increasing urbanization and population growth have caused Bangladesh to lose 0.08 million hectares of arable land annually and expanding agricultural land use will not boost the yields of food crops (BBS, 2018). As a result, increasing land utilization by undertaking the implementation of innovations that boost production, such as fertilizer, is essential to attaining food sufficiency.

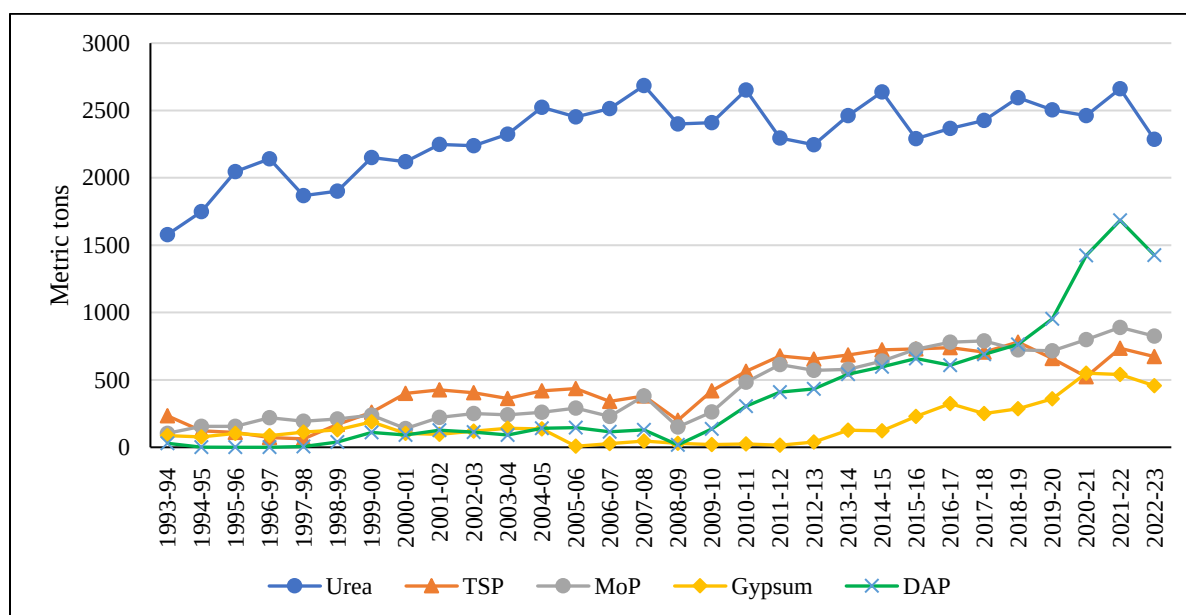


Figure 1: Major fertilizer consumption outlook in Bangladesh

Chemical fertilizers have played a significant role in increasing the major food grains (rice, wheat) production over the years, which play a critical role in ensuring food security in Bangladesh (BBS, 2023). Since their introduction, they are assisted crop production by increasing soil fertility and meeting the nutritional demand of crops. Rice production has experienced a significant growth from 250.87 lakh metric tons to 391.02 lakh metric tons from 1992-93 to 2022-23 farming year; wheat production reached 16.70 lakh metric tons from 11.71 lakh metric tons for the same period, whereas production of maize witnessed 1.49 lakh metric tons from 64.32 lakh metric tons during the period (Table 1).

Table 1. 23-year food grains production in Bangladesh

Market Year	Production (Lakh metric tons)						
	Rice	Wheat	Maize	Market Year	Rice	Wheat	Maize
2000-01	250.87	16.70	1.49	2012-13	338.33	12.55	21.78
2001-02	243.00	16.06	1.52	2013-14	343.74	13.02	25.16
2002-03	251.88	15.07	1.75	2014-15	347.10	13.48	23.61
2003-04	261.90	12.53	2.41	2015-16	347.10	13.48	27.59
2004-05	251.57	9.76	3.56	2016-17	338.06	13.12	35.78
2005-06	256.53	7.35	5.22	2017-18	362.79	11.53	38.93
2006-07	273.18	7.25	8.99	2018-19	373.63	11.48	46.99
2007-08	289.31	8.44	13.46	2019-20	386.95	12.46	54.02
2008-09	313.17	8.44	7.30	2020-21	376.08	10.85	56.63
2009-10	322.57	9.69	8.87	2021-22	391.80	10.86	56.30
2010-11	335.41	9.72	15.52	2022-23	391.02	11.71	64.32
2011-12	338.90	9.95	19.54	2023-24	434.22	12.29	66.95

Source: (BER, 2005, 2013, 2019 and 2024)

Exploring the roots of fertilizer dependence in Bangladesh

Population pressure

Bangladesh, with a total population of over 165 million and an area of approximately 147570 square kilometers, is the eighth most populated country in the world (Wikipedia, 2024). This immense population density exerts extreme pressure on the country's agriculture sector to produce sufficient food. With limited arable land, farmers are forced to adopt intensive cropping systems, often planning two to three crops annually on the same plot of land. As a result, soil fertility is depleting rapidly, making the use of chemical fertilizer crucial for replenishing nutrients and sustaining high yields. Additionally, government subsidies and the accessibility of chemical fertilizers have made them a more cost-effective and readily available solution for farmers.

Staple food

Rice and wheat are the primary food sources in Bangladesh. Rice alone supplies almost 70% of the caloric demand for the population (Bin Rahman and Zhang, 2023), with *Aus*, *Aman*, and *Boro* seasons. Wheat, though less dominant, plays a vital role in diversifying the food supply. Bangladesh has adopted high-yielding crop cultivars to meet the demands of its growing population, which require intensive nutrient replenishment. Chemical fertilizers such as urea, triple superphosphate, and muriate of potash are widely used to supply nitrogen, phosphorus, and potassium, respectively, ensuring optimal crop production. This heavy reliance on synthetic fertilizers has become a key strategy to ensure food security in the nation.

Quick and visible result

Chemical fertilizers have an immense impact on crop growth and development, which increase crop yields within a shorter period compared to organic practices. These fertilizers provide essential nutrients for crops, especially staple crops like rice and wheat, where farmers aim to maximize output to meet both their family's and country's high food demand. As a result, the need for immediate yield improvement and the pressure to sustain multiple cropping systems have made chemical fertilizers the preferred choice for producers.

Government subsidies and support

The widespread consumption of synthetic agrochemicals in Bangladesh is largely driven by government policies that make them accessible and affordable for farmers. Subsidized pricing, especially for urea, significantly reduces costs, enabling even smallholder farmers to afford fertilizers. Additionally, an extensive distribution network ensures the chemical fertilizers are readily available in rural areas. During the green revolution, the authorities actively promoted chemical fertilizers through campaigns, training programs, and financial incentives to boost agricultural productivity. This long-term support increases the adaptations of synthetic fertilizers to sustain high land for food security.

Table 2. Govt. subsidy for chemical fertilizer for different farming year in Bangladesh

Farming year	Subsidy (Crore)
2005-06 ^a	1,095
2009-10 ^b	4,009
2020-21 ^b	7,717
2021-22 ^c	14,941
2022-23 ^d	16,000
2023-24 ^d	24,000

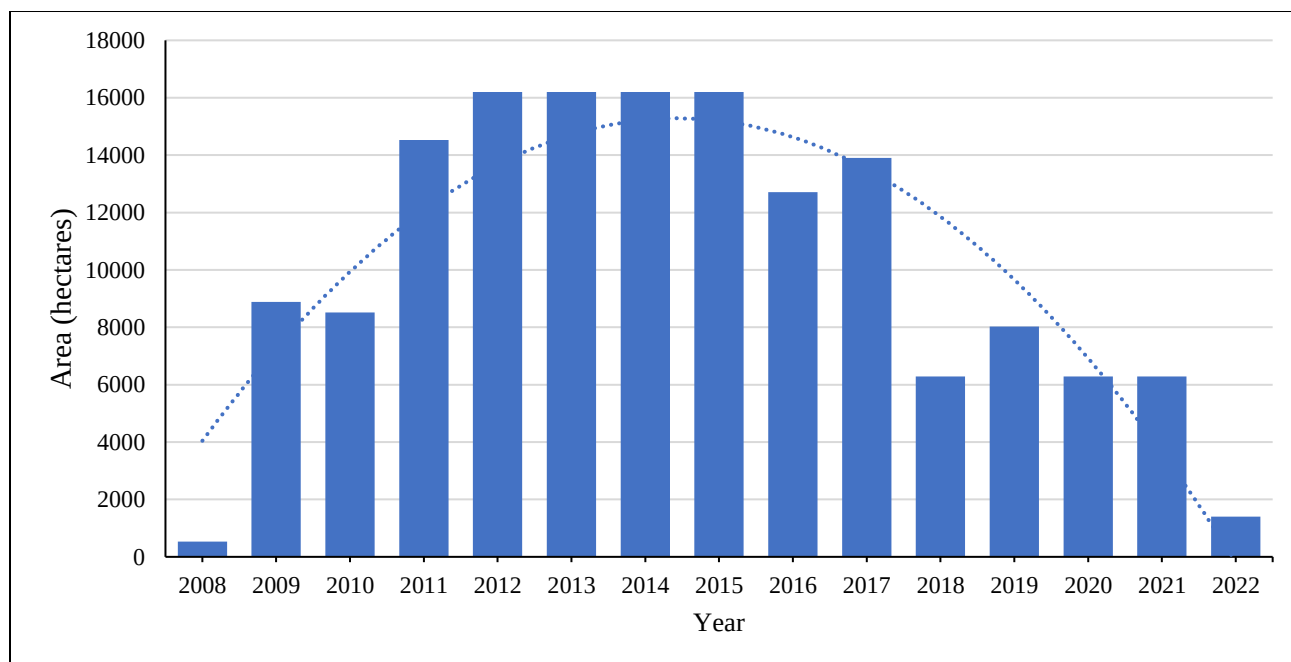
Sources: (^aBSS, 2022; ^bDhaka Tribune, 2024; ^cDhaka Tribune, 2024; ^dByron, 2023)

Lack of awareness and fragmentation of land

The preference for synthetic fertilizers in Bangladesh stems from both a lack of awareness about organic farming and the issue of fragmented land, which hinders the use of alternative practices. Many farmers lack knowledge about the profound effects of organic farming on our environment and ecosystem. Furthermore, land fragmentation, with most farmers operating on small. Scattered plots make it more difficult to execute labour-intensive methods like organic methods like composting. This small-scale farmer finds chemical fertilizers more practical and manageable, requiring less time and effort, making them a preferred choice for boosting productivity.

Present status of organic farming in Bangladesh

The Bangladeshi organic agriculture movement is being introduced and expanded across the nation by the commercial sector, which has emerged as a pioneer in this area. By raising consciousness, educating the public, and assisting the marketplace for organic products, NGOs and other private entities have stood in the forefront. In order to guarantee that the manufacturer receives a fair price, they have established a value chain for organic goods. Additionally, they provided materials for organic farming along with innovative strategies for cultivating climate resilience. Currently, the government is also participating in such attempts by putting legislation into action, offering subsidies, and creating a certification program to encourage organic farming across the nation. Together with other non-governmental organizations, Bangladesh managed 177,700 hectares, or 2% of its total area, under organic farming in 2006, making it the second-largest country in Asia (IFOAM, 2006). A few traditional growers who are not affiliated with non-governmental organizations have begun to grow organic crops after witnessing the advantages of doing so from NGO members. After analysing the data from 2008 to 22, it was found that the land used for organic farming showed increasing tendencies up until the years 2012 to 2015. Following that, the area showed dropping patterns until the present (Figure 2). Nevertheless, just 526, 16198, 8030, 6285, and 1400 hectares of land had been transformed as of 2008, 2015, 2019, 2021, and 2022, respectively (Willer et al., 2024). Organic farming is used in Bangladesh for growing fruits, vegetables, shrimp, and tea (*Camellia sinensis* L.).



Source: (Willer et al., 2011, 2015, 2019 & 2024)

Figure 2: Average organic farming area (ha) in Bangladesh across different years

Organic market in Bangladesh

Although Bangladesh's organic food business remains in its beginning stages, it is expanding quickly. Bangladeshi's limited financial resources prevent them from affording organic food. Bangladesh's organic food market is primarily composed of middle-class to upper-class, educated, health-conscious people who value the hygiene and nutritional advantages of organic products. A lot of organizations specifically focused on organic farming have been started as a result of the market demand and high cost of organic products. Supermarkets, specialty shops, internet retailers, and other retail establishments are among the places where organic items can be found. Probortana, PROSHIKA, and Meena Bazar are among the popular organic stores in Bangladesh. In rural regions, the scenario is the reverse. Producers typically grow organic food for domestic consumption, ensuring it is devoid of any artificial pesticides or fertilizers. These food items are rarely sold. Bangladesh has a lot of prospects for exporting its organic vegetable crops. Even though Bangladesh's vegetable exports account for a very small portion of the world's vegetable international trade, they are expanding quickly. Export revenues from this industry climbed from US\$ 14.5 million to US\$ 49.26 million over fifteen years from 1995-1996 to 2009-2010, representing an ordinary yearly rise of 16% (Bhuyan and Uddin, 2010). Nonetheless, the marketplace for organic goods is projected to have reached value at \$10 million or more by 2023 and is predicted to grow to \$50 million by 2025 (Rahman, 2023).

Limitations of organic farming in Bangladesh

Organic farming in Bangladesh holds immense prospects for promoting sustainability of the environment and ecosystem; however, it faces several drawbacks that hamper its widespread adaptation. Transitioning from traditional crop production practices to eco-friendly practices often involves high initial costs, which many small-scale farmers find unaffordable. By reducing chemical fertilizer demand, it helps to increase the sustainability of natural resources like water, soil, and microorganisms. This approach not only produces healthier produce for consumers but also ensures the long-term sustainability of agriculture. Realizing the full potential of organic farming in Bangladesh, successful adaptation requires addressing several challenges to transition farmers from inorganic practices to sustainable crop production. However, key limitations are summarized below:

Government and farmer perception

Policymakers think that hunger and extreme poverty would have been more severe than they are now if cereal production hadn't increased so dramatically. Organic farming is said to produce lower yields, and there isn't enough organic fertilizer to significantly increase yields. In Bangladesh, the government defines nutritional safety as having a sufficient supply of rice. Additionally, having the necessary amount of rice in his home gives a Bangladeshi farmer a sense of security regarding food (UBINIG, 2011). It has been reported that food security problems may arise as a result of the yield difference between traditional and organic agriculture (De Ponti et al., 2012; Kniss et al., 2016).

High production costs

Organic farming in Bangladesh faces a lot of challenges, making its adaptation and sustainability difficult for farmers. The high cost of organic inputs like biofertilizers, composts, and vermicompost's, along with their limited availability, significantly increases the production cost. Also, this farming method is labour-intensive, requiring more manpower for activities like weeding, pest control, and compost preparation, which has limited the farmer's perception of towards organic farming. Moreover, transitioning to organic farming involves a period of reduced yields as the soil restores its natural fertility and productivity, which raises the costs per unit of production. Organic farming also faces difficulties coping with the high price of biopesticides, which ultimately exacerbates the issues.

Low yield and productivity

In the initial stages, organic farming results in lower yields compared to chemical-based crop production. This happens due to the long period of time it requires for the soil to adjust to using organic pesticides as opposed to synthetic pesticides. Additionally, a considerable amount of time is spent encouraging the development of beneficial creatures for the farm's pollination. At the same time, the farmer loses a significant amount of yield, making organic cultivation considerably less appealing to the typical Bangladeshi producer. As a result, this has become a significant deterrent for the farmers to meet household needs or market demand, who are relying on high-yielding crop cultivation with inorganic fertilizers. The research by Uddin et al. (2016) revealed that the tomato yield was initially low at 55.45 t ha⁻¹ during the 2008-09 farming year. However, by the 2014-15 period, after continued organic farming practices, the yield increased to 65.91 t ha⁻¹.

Lack of organic inputs

Organic farming heavily depends on inputs like compost, green manure, vermicompost, farmyard manure, green manure and biofertilizers. Unfortunately, in Bangladesh, these inputs are not widely produced or distributed properly. Most farmers find it difficult to source organic alternatives, forcing them to continuously use synthetic fertilizers. Moreover, organic fertilizer production requires additional labour and resources, which may not be feasible for resource-poor farmers.

Lack of awareness and training

The lack of awareness and training facilities considerably hinders the growth of organic farming in Bangladesh. Many farmers are unfamiliar with the principles and long-term benefits of this practice, leading them to rely on conventional chemically intensive methods that provide faster results. Without adequate knowledge, they may struggle to adopt organic techniques like composting, crop rotation and biological pest control. Similarly, access to training programs and expert guidance is limited, particularly in rural areas. Farmers frequently lack the skills to manage soil fertility, productivity, pests and disease organically. The absence of proper government or institutional initiatives to educate farmers further ignites the problem. Without proper guidance, farmers are reluctant to swift away from the chemical-demand methods they are accustomed to, fearing potential crop failures or reduced incomes.

Limited market access and infrastructure

There are few facilities for preservation, packaging, and transportation in the weakened market structures for organic products. Farmers' capacity to effectively meet customer demand for fresh products is impacted by this. Furthermore, the transportation and cold storage of biological goods from rural to city centres are hampered by a lack of reliable facilities.

Market demand

Organic items are costlier in Bangladesh than inorganic ones. Consumers in the country are often cost-sensitive and organic items are typically more expensive than conventional products due to higher input and maintenance costs and limited economies of scale. This lack of affordability makes it more difficult for farmers to sell their products at a profitable price. The domestic market for organic products is not well structured. Moreover, the export market for these products remains untapped due to challenges meeting international certification standards, which require substantial time as well as monetary demand and expertise. Without strong consumer demand, organic farmers face difficulties recovering their high production costs.

Soil fertility management

It is more difficult for farmers to regulate soil fertility in an organic farming system because of the tougher guidelines regarding the use of synthetic fertilizers. In order to efficient replenish of soil nutrients, a lot of farmers struggle to locate sufficient supplies of organic matter, vermicompost, compost, green manure, or other organic fertilizers in a timely manner.

Pest and disease control

Managing pests and diseases in organic farming is quite challenging in Bangladesh due to a combination of environmental, technological, and knowledge-based factors. The country's warm and humid environmental conditions foster rapid pests and disease proliferations, especially in highly valuable crops like rice, wheat, and maize. Unlike traditional farming, organic farming adheres closely to natural pest control techniques; yet, employing natural predators to eradicate insects may prove to be less efficient and require more time than utilizing chemicals. The main challenge facing farmers is the infestation of pests and diseases, which can lead to crop loss. As a result, illnesses, pest infestations, and pests become some of the barriers to large-scale dissemination of organic farming practices. Furthermore, pests may develop resistance to commonly used organic remedies, making management more difficult, which makes pests and disease control a persistent obstacle for organic farming in Bangladesh.

Policy and support issues

Policy and support issues significantly influence the adaptation of organic farming in Bangladesh, often acting as barriers due to misaligned government priorities. The emphasis on chemical fertilizer, along with a large number of subsidies, discourages farmers from transitioning to organic farming in Bangladesh. Such policies make conventional farming more attractive to farmers due to its economic viability in the short term.

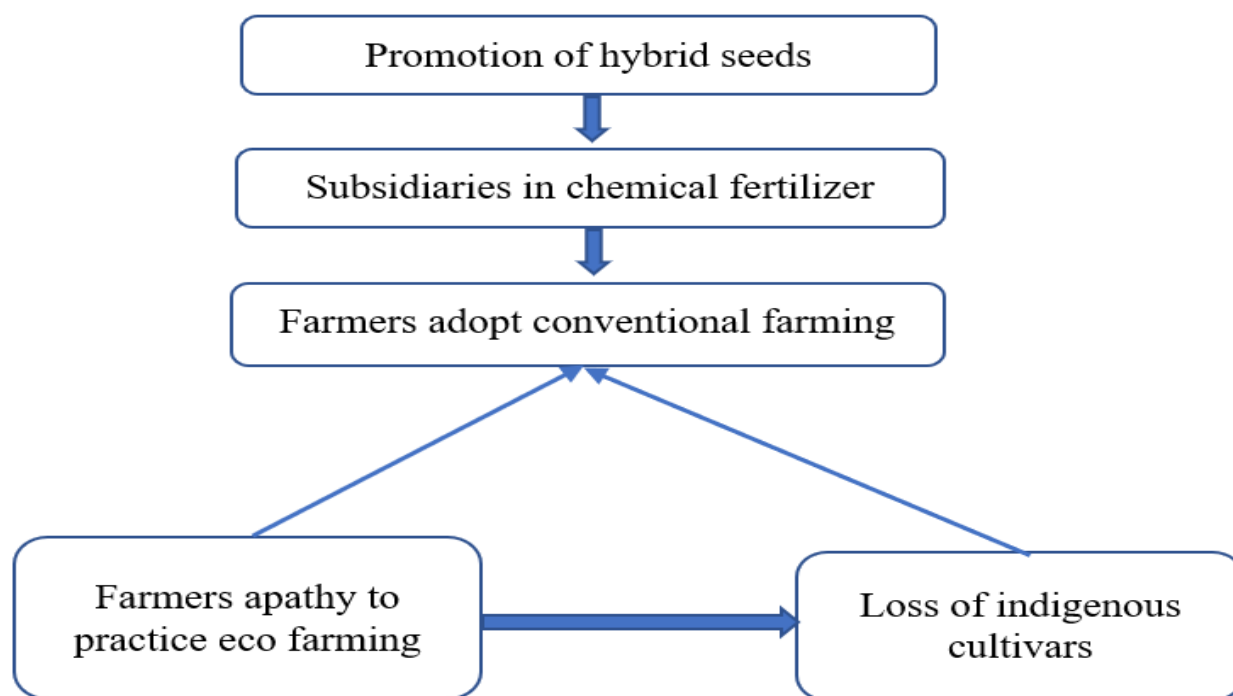


Figure 3. Influence of government policies on organic farming

Land fragmentation

Land fragmentation creates consistent challenges to organic farming in Bangladesh, stemming from a number of structural and economic determinants. The country's farming landscape is determined by small and scattered plots, a result of population pressure, stick regulations, and rapid urbanization. These fragmented lands limit the implementation of organic farming practices such as crop rotation, intercropping, and conservational crop production techniques. Additionally, farmers face further difficulties in monitoring and controlling pests and diseases, as fragmented plots create barriers to employing integrated diseases and pest management strategies. Moreover, land fragmentation reduces economies of scale, making it harder for farmers to collectively market their organic produce or share their essential infrastructure, such as compost pits and irrigation systems. The lack of cooperative farming models and government support compound these issues, discouraging farmers from transitioning to organic farming. Furthermore, land fragmentation often suffers from uneven soil fertility, which is one of the most limiting factors for organic crop production. These factors highlight the structural limitations, necessitating policy interventions to encourage land consideration and cooperative farming to support sustainable farm practices.

Prospect of organic farming in Bangladesh

Organic farming in Bangladesh holds substantial promise offering solutions to environmental, economic, and health challenges. Below is a detailed exploration of key prospects for organic farming in Bangladesh:

Diverse agro-ecological zones

Bangladesh is blessed with a diverse climate that allows the cultivation of various crops throughout the year. Thus, it is ideal for organic farming, where crop rotation and diversification are key practices. Organic farming can enhance soil health in the long term through diversified crop planting. Furthermore, the alluvial soils in Bangladesh's delta are highly fertile, providing a good foundation for organic farming.

Traditional knowledge of farmers

Many farmers in Bangladesh have a long history of traditional farming practices like floating agriculture, sorjan methods and ring methods, which align with organic principles. These existing knowledge bases can be built upon to introduce more sustainable methods, ensuring a smoother transition for farmers moving towards organic farming.

Environment sustainability

Organic farming can mitigate the adverse effects of conventional farming in several ways:

Improving soil health:

From 1980-81 to 2022-23, the use of major chemical fertilizers like urea, TSP, and MoP in Bangladesh has significantly increased (Abusiddique, 2022). This surge in fertilizer use has led to a decline in soil fertility and beneficial microorganisms, resulting in plants not receiving sufficient nutrients despite increased fertilizer application. Most arable land in Bangladesh contains less than 1.5% organic matter, while healthy soil should have at least 2.5% (Cavallito, 2023). Additionally, the nitrogen balance is negative and other essential nutrients like phosphorus and potassium are either deficient or near zero, worsening the overall soil nutrient status. Organic practices emphasize the use of compost, green manure, and crop rotation, all of which help to replenish soil health naturally. Unlike synthetic fertilizer, organic farming helps to increase soil organic matter, which improves soil structure, texture, water retention, and microbial diversity. This leads to a long-term increase in soil fertility.

Reducing pollution

The extensive use of agrochemicals has resulted in water and soil pollution in Bangladesh. Fertilizer runoff can contaminate above-ground and underground water resources, causing issues such as eutrophication, which harms aquatic ecosystems. Excessive nitrate in water used for drinking (more than 50 mg NO₃⁻/L) can cause the water to become eutrophic and cause serious health risks such as cardiac disease, blue baby syndrome, stomach cancer, goiter, and birth defects (Feigin & Halevy, 1989). Organic farming reduces the use of synthetic chemicals, thereby preventing the leaching of harmful substances into water resources and reducing the negative impacts on biodiversity. It has been reported that organic agriculture helps to considerably lower the amount of persistent and water-soluble chemical pesticides that contaminate waterways by avoiding artificial chemicals (Vincent & Fleury, 2015).

Economic opportunities

Globally, there is an increasing demand for organic food due to growing health concerns among consumers. In Bangladesh, this trend is also catching up, particularly among the urban middle-class population who are willing to pay a premium for chemical-free products. With improved consumer awareness and demand for organic products, farmers in Bangladesh can tap into new markets both domestically and internationally, creating new economic opportunities for organic products.

Employment generation

Organic farming tends to be more labour-intensive than traditional farming. Practices such as crop rotation, manual weeding and compost preparation require more human labour, which can create employment opportunities, especially in rural areas. This is crucial in Bangladesh, where agriculture is still the primary source of income for millions of people. By promoting organic farming, Bangladesh can create sustainable job opportunities that will ensure rural development.

Health benefits

Shifting towards organic farming can significantly impact public health, especially in a developing country like Bangladesh, where the population faces health risks due to the consumption of foods containing pesticide residues.

Chemical free produce

One of the most significant health benefits of organic crop production is the production of food that is free from harmful chemicals. Pesticides, herbicides, and any synthetic fertilizers commonly used in traditional farming can leave residues on crops that are detrimental to human health. Chronic exposure to these chemicals has been linked to various health issues, including cancer, neurological disorders, and reproductive problems. For instance, long-term excessive nitrate consumption has been linked to several gastrointestinal disorders, including cancer (Jones et al., 2016). Young children are primarily affected by methemoglobinemia, which is caused by consuming water that contains excessive NO₃⁻ (Johnson, 2019). Evidence suggests that consuming high-level NO₃⁻ containing drinking water increases the incidence of birth abnormalities of the eye, ear, face, neck, and nervous system (Stayner et al., 2022). However, eating organic food may lessen exposure to microorganisms that are resistant to antibiotics. Organic practices can eliminate these risks, providing consumers with safe and healthier food, as organic products have also been demonstrated to have higher levels of antioxidant activity, carotenoids, phenolics, and flavonoids (Rempelos et al., 2021; Crinnion, 2010).

Reduced health care cost

With the reduced use of chemicals in food production, there is a potential decrease in pesticide-related health problems. This reduction in chemical use can lower healthcare costs related to diseases and other health disorders caused by long-term exposure to pesticides.

Policy support and initiatives

While organic farming is still in its initial stage in Bangladesh, there are promising signs of support from the government as well as non-government organizations.

Government efforts

The Bangladesh government has begun to recognize the importance of organic farming for ensuring food security and environmental sustainability. Several pilot's projects and programs have been launched by the government to encourage farmers to adopt organic farming. The government is also promoting organic farming through the Department of Agricultural Extension (DAE) by providing training and offering support for the transition to organic agriculture. However, more policy invention is needed to scale these efforts.

Table 3. SWOT analysis for organic farming in Bangladesh

Strengths	Weaknesses
➤ Favorable agro-climatic ➤ Growing consumer awareness ➤ Fertile agricultural land ➤ Rich traditional farm practices ➤ Government initiatives ➤ Export opportunities ➤ Diverse climatic conditions	➤ Small landholdings ➤ Lack of inputs ➤ Limited knowledge of farmers ➤ High input cost and certification barriers ➤ Lower initial yields ➤ Weak marketing channels ➤ Cultural resistance
Opportunities	Threats
➤ Increasing domestic demand ➤ Export potentials ➤ Sustainability goals ➤ Increasing urbanization ➤ Government supporting programs ➤ Digital marketing platforms ➤ Youth and entrepreneurship	➤ Competition from conventional farming ➤ Vulnerable to climate change ➤ Consumer affordability ➤ Consumer awareness gap ➤ Lack of certification bodies ➤ Pests and diseases infestations ➤ Lack of proper organized market

Non-government organizations

NGOs in Bangladesh have been playing a key role in promoting organic farming, especially in rural areas. These organizations provide farmers with technical support, organic inputs, and training on organic farm practices. It also helps farmers access organic certification, which is essential for marketing. Their efforts contribute to raising awareness about the benefits of organic farming to improve the livelihoods of smallholder farmers.

SWOT analysis for organic farming in Bangladesh

The analysis was conducted based on collected responses from both farmers and consumers, providing a clear understanding of the current landscape and future perspective of organic farming in Bangladesh. Producers highlighted key challenges such as high production costs, limited access to organic inputs, and market

uncertainties. However, many also recognized the growing demand for organic products and the potential for higher profits with proper market access. In contrast, consumers were significantly aware of the benefits of such products, like chemical-free options. However, issues like affordability and availability of organic products in local markets were noted as obstacles. The analysis, based on collected responses, is summarized in table 3.

Conclusion

The agriculture sector of Bangladesh is a critical platform, where ensuring food security for an increased population along with environmental sustainability has become challenging through organic farming. The synthetic fertilizers have become a crucial factor in achieving food security for dense populations with limited arable land. Chemical fertilizer significantly increased the crop yield, resulting in meeting the food demand of a growing population. However, the dependency on chemical fertilization has become a great concern for the country. One of the primary concerns is that excess use of synthetic fertilizer degrades the soil quality through the disruption of microbial activity, reducing the water holding capacity of the soil, depletion of organic matter, increasing the soil acidification, reduction of soil infiltration capacity, increasing heavy metal contamination, and so on. As a result, fertilizer dependency is increasing day by day. On the other hand, organic farming is the platform for overcoming these challenges. Organic farming practices, which involve the use of organic residues from animals and plants, ensure the long-term development of soil, water, and air quality through the reduction of nitrate contamination in water and greenhouse gas emissions in the air. The adaptation of organic farming restores soil fertility through the increment of organic matter in soil and microbial activities and thus promotes pesticide-free food for a healthy population. However, the limited availability of inputs and lower yield have become the most important limiting factors for organic crop production in Bangladesh. Indeed, organic farming can't meet the food demand of Bangladesh, as the demand for rice is high and increasing day by day. Despite these limitations, there is great opportunity for the organic market, as the increasing domestic and global demand for organic foods is creating a lucrative market for organic produce. At present, turning fully to organic farming with limited arable land can condense food insecurity, resulting in the enhancement of the import cost of different goods from other countries. Integration of organic and inorganic farming can be the possible way to ensure food security as well as sustainable uses of natural resources in contemporary terms.

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References

- ABD (Asian Development Bank). (2023). Bangladesh's agriculture, natural resources, and rural development sector assessment and strategy. <http://dx.doi.org/10.22617/TCS230050>
- Abusiddique. (2022, December 30). Degraded soil threatens to exacerbate Bangladesh food crisis. Mongabay Environmental News. <https://news.mongabay.com/2022/12/degraded-soil-threatens-to-exacerbate-bangladesh-food-crisis/>
- BARC (Bangladesh Agricultural Research Council). 2018. Fertilizer Recommendation Guide - 2018. https://www.researchgate.net/publication/352356098_Fertilizer_Recommendation_Guide_-_2018
- BBS (Bangladesh Bureau of Statistics). (2018). Preliminary Report on Agricultural Census 2019. Bangladesh Bureau of Statistics, Stat. Div., Minis. Plan., Govt. People's Repub. Bangladesh, Dhaka. https://bbs.portal.gov.bd/sites/default/files/files/bbs.portal.gov.bd/page/b343a8b4_956b_45ca_872f_4cf9b2f1a6e0/9beb0d821f582859681d77f76e89e321.pdf
- BBS (Bangladesh Bureau of Statistics). (2023). Food Security Statistics 2023. Bangladesh Bureau of Statistics, Stat. Div., Minis. Plan., Govt. People's Repub. Bangladesh, Dhaka. https://bbs.portal.gov.bd/sites/default/files/files/bbs.portal.gov.bd/page/b343a8b4_956b_45ca_872f_4cf9b2f1a6e0/2024-03-16-15-54-15eb3ff72d71e6c5680a6b8111b197cc.pdf
- BBS (Bangladesh Bureau of Statistics). (2023). Population and Housing Census 2022: English Version. Bangladesh Bureau of Statistics, Stat. Div., Minis. Plan., Govt. People's Repub. Bangladesh, Dhaka. <https://bbs.gov.bd/site/page/47856ad0-7e1c-4aab-bd78-892733bc06eb/Population-&-Housing.2022>
- BER (Bangladesh Economic Review). (2005). Bangladesh-Economic-Review-2005. Minis. of Finance, Govt. People's Repub. Bangladesh, Dhaka. <https://mof.gov.bd/site/page/44e399b3-d378-41aa-86ff-8c4277eb0990/Bangladesh-Economic-Review-Archive>
- BER (Bangladesh Economic Review). (2013). Bangladesh-Economic-Review-2005. Minis. of Finance, Govt. People's Repub. Bangladesh, Dhaka. <https://mof.gov.bd/site/page/44e399b3-d378-41aa-86ff-8c4277eb0990/Bangladesh-Economic-Review-Archive>
- BER (Bangladesh Economic Review). (2019). Bangladesh-Economic-Review-2005. Minis. of Finance, Govt. People's Repub. Bangladesh, Dhaka. <https://mof.gov.bd/site/page/44e399b3-d378-41aa-86ff-8c4277eb0990/Bangladesh-Economic-Review-Archive>
- BER (Bangladesh Economic Review). (2024). Bangladesh-Economic-Review-2024. Minis. of Finance, Govt. People's Repub. Bangladesh, Dhaka. <https://mof.portal.gov.bd/site/page/28ba57f5-59ff-4426-970a-bf014242179e/Bangladesh-Economic-Review-2024>
- Bhuyan, M. A. J., and M. N. Uddin. (2010). "Present Status and Improvement Strategy of Vegetables Crops through Regional Trials." Regional workshop on 'Improvement of Vegetables and Adaptive Trials in SAARC Countries', Hotel Sheraton, Dhaka, Bangladesh, September 8–9.
- Bin Rahman, A. R., & Zhang, J. (2023). Trends in rice research: 2030 and beyond. Food and Energy Security, 12(2), e390. <https://doi.org/10.1002/fes3.390>
- BSS (Bangladesh Sangbad Sangstha). (2022, February 14). Fertilizer subsidy rises to Tk 28,000 cr crossing govt's budget estimate. Available at: <https://www.bssnews.net/news-flash/44576>
- Byron, R. K. (2023, May 30). Budget Subsidies: Big chunks go to power, fertiliser. The Daily Star. <https://www.thedailystar.net/news/bangladesh/news/budget-subsidies-big-chunks-go-power-fertiliser-3332821>

- Cavallito, M. (2023, January 30). Land degradation and food crisis hit Bangladesh. Re Soil Foundation. <https://resoilfoundation.org/en/agricultural-industry/land-degradation-bangladesh/>
- Chowhan, S., Rahman, M. M., Sultana, R., Rouf, M. A., Islam, M., & Jannat, S. A. (2024). Agriculture Policy and Major Areas for Research and Development in Bangladesh. *Sarhad Journal of Agriculture*, 40(3), 245-251. <https://dx.doi.org/10.17582/journal.sja/2024/40.3.819.831>
- Crinnion, W. J. (2010). Organic foods contain higher levels of certain nutrients, lower levels of pesticides, and may provide health benefits for the consumer. *Alternative Medicine Review*, 15(1), 4-12. Available at: <https://pubmed.ncbi.nlm.nih.gov/20359265/>
- De Ponti, T., Rijk, B., & Van Ittersum, M. K. (2012). The crop yield gap between organic and conventional agriculture. *Agricultural systems*, 108, 1-9. <https://doi.org/10.1016/j.agsy.2011.12.004>
- Dhaka Tribune. (2024, 21 November). Budget 2022-23: Govt to increase fertilizer subsidy to ensure food security. Available at: <https://www.dhakatribune.com/bangladesh/270790/budget-2022-23-govt-to-increase-fertilizer>
- Dhaka Tribune. (2024, 21 November). Govt provided 97,873C as subsidy for agriculture in last 13 years. Available at: <https://www.dhakatribune.com/bangladesh/agriculture/312828/razzaque-govt-provided-97-873c-as-subsidy-for>
- Feigin, A., & Halevy, J. (1989). Irrigation-fertilization-cropping management for maximum economic return and minimum pollution of ground water. In Research report, Inst. Soil Water, ARO, The Volcani Center, Bet Dagan.
- International Federation of Organic Agriculture Movements (IFOAM). (2006). The World of Organic Agriculture Statistics and emerging trends (Helga Willer and Minou Yussefi eds), 1-167. <https://orgprints.org/id/eprint/5161/2/willer-yussefi-2005-world-of-organic.pdf>
- Johnson, S. F. (2019). Methemoglobinemia: Infants at risk. Current problems in pediatric and adolescent health care, 49(3), 57-67. <https://doi.org/10.1016/j.cppeds.2019.03.002>
- Jones, R. R., Weyer, P. J., DellaValle, C. T., Inoue-Choi, M., Anderson, K. E., Cantor, K. P., & Ward, M. H. (2016). Nitrate from drinking water and diet and bladder cancer among postmenopausal women in Iowa. *Environmental health perspectives*, 124(11), 1751-1758. <https://doi.org/10.1289/EHP19>
- Karim, R., Pk, M. B., Dey, P., Akbar, M. A., & Osman, M. S. (2024). A study about the prediction of population growth and demographic transition in Bangladesh. *Journal of Umm Al-Qura University for Applied Sciences*, 1-13. <https://doi.org/10.1007/s43994-024-00150-0>
- Kniss, A. R., Savage, S. D., & Jabbour, R. (2016). Commercial crop yields reveal strengths and weaknesses for organic agriculture in the United States. *PloS one*, 11(8), e0161673. <https://doi.org/10.1371/journal.pone.0161673>
- Naher, U. A., Shah, A. L., Sarkar, M. I. U., Islam, S. M., Ahmed, M. N., Panhwa, Q. A., & Othman, R. (2015). Fertilizer consumption scenario and rice production in Bangladesh. *Advances in tropical soil science*, 3, 81-98. https://www.researchgate.net/publication/354144255_Fertilizer_Consumption_Scenario_and_Rice_Production_in_Bangladesh
- Rahman, M. M. (2023, December 2). Organic food: a global phenomenon and Bangladesh's promising future. <https://www.linkedin.com/pulse/organic-food-global-phenomenon-bangladeshs-promising-future-rahman-verwec/>
- Rempelos, L., Baranski, M., Wang, J., Adams, T. N., Adebuseyi, K., Beckman, J. J., & Leifert, C. (2021). Integrated Soil and Crop Management in Organic Agriculture: A Logical Framework to Ensure Food Quality and Human Health? *Agronomy*, 11(12), 2494. Available at: <https://doi.org/10.3390/agronomy11122494>

- Shahabuddin, Q. (2016, February 3). Food security prospects. The Daily Star. <https://www.thedailystar.net/supplements/25th-anniversary-special-part-4/food-security-prospects-212050>
- Stayner, L. T., Jensen, A. S., Schullehner, J., Coffman, V. R., Trabjerg, B. B., Olsen, J., & Sigsgaard, T. (2022). Nitrate in drinking water and risk of birth defects: Findings from a cohort study of over one million births in Denmark. *The Lancet Regional Health–Europe*, 14. <https://doi.org/10.1016/j.lanepe.2021.100286>
- Uddin, M. N., Siddiqy, M. A., Hossain, M., Islam, F., Halim, G. M. A., & Bari, M. L. (2016). Impact of organic and conventional practices on, soil health and crop yield under tropical and subtropical environment of Bangladesh. *International Journal of Environmental & Agriculture Research*, 2, 89-100. <https://www.adpublications.org/impact-organic-conventional-practices-soil-health-crop-yield-tropical-subtropical-environment-bangladesh/>
- Vincent, A., & Fleury, P. (2015). Development of organic farming for the protection of water quality: Local projects in France and their policy implications. *Land use policy*, 43, 197-206. <https://doi.org/10.1016/j.landusepol.2014.10.020>
- Wikipedia contributors. (2024, November 16). Demographics of Bangladesh. Wikipedia. https://en.wikipedia.org/wiki/Demographics_of_Bangladesh
- Willer, H., Trávníček, J., & Schlatter, S. (2011). The World of Organic Agriculture. Statistics and Emerging Trends 2011. <https://www.fibl.org/fileadmin/documents/shop/1546-organic-world-2011.pdf>
- Willer, H., Trávníček, J., & Schlatter, S. (2015). The World of Organic Agriculture. Statistics and Emerging Trends 2015. <https://www.fibl.org/fileadmin/documents/shop/1663-organic-world-2015.pdf>
- Willer, H., Trávníček, J., & Schlatter, S. (2019). The World of Organic Agriculture. Statistics and Emerging Trends 2019. <https://www.fibl.org/fileadmin/documents/shop/2020-organic-world-2019.pdf>
- Willer, H., Trávníček, J., & Schlatter, S. (2024). The World of Organic Agriculture. Statistics and Emerging Trends 2024. https://orgprints.org/id/eprint/52272/1/1747-organic-world-2024_light.pdf
- Xiang, T., Malik, T. H., & Nielsen, K. (2020). The impact of population pressure on global fertiliser use intensity, 1970–2011: An analysis of policy-induced mediation. *Technological Forecasting and Social Change*, 152, 119895. <https://doi.org/10.1016/j.techfore.2019.119895>

RESEARCH ARTICLE

Growth and Yield Performance of Carrot (*Daucus carota* L.) as Influenced by Organic Manure under Open Field Conditions

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Abstract

Carrot is an exotic vegetable in Liberia, commonly used in combination with other vegetables when making soups, stews, and salads. The study focused on assessing the growth and yield of carrots by combining NPK doses and organic manures in an open field at the Teaching and Research Farm, William V. S. Tubman University Campus, Harper City, Liberia. The randomized complete block design included soil amendments of 10 t/ha, 15 t/ha, and 20 t/ha of chicken manure, along with 300 kg/ha of NPK fertilizer (15:15:15), and a control for evaluation. The findings from the physical and chemical analysis of different macronutrients in the soil, such as soil carbon, phosphorus, potassium, calcium, magnesium, and sodium, were deemed to be moderate and satisfactory. The result of analysis of variance (ANOVA) showed that all growth and yield parameters were significantly affected ($P < 0.05$). Results indicated that the longest Plant height (19.10 cm) was recorded in the 20 ton ha⁻¹ compost application, while the lowest plant height (6.84 cm) was measure in the control. The highest leaf number (15.15) was recorded in 20 t ha⁻¹ while the lowest leaf number (10.22) was recorded in the control (0 t ha⁻¹). The result revealed that the leaf area (39.76 cm²) was recorded in the 20 ton ha⁻¹, while the minimum (13.18cm²) was recorded from the control. The next best results were seen with 20 t ha⁻¹ of chicken manure, which had root lengths of 19.31cm, while the lowest plant root length (6.84cm) was measure in the control. The highest root diameter of (7.87mm) was recorded in 20 t ha⁻¹ while the lowest root diameter (2.16 mm) was recorded in the control (0 t ha⁻¹). The result revealed that the maximum root weight (31.76 g) was recorded in the 20 ton ha⁻¹, while the minimum (10.33 g) was recorded from the control. The study found that the carrot plants showed the best growth and highest yield when treated with 20 tons per hectare of chicken manure.

Keywords: Carrot; organic manures; root length; root weight, plant nutrients

Introduction

Carrots are highly esteemed exotic vegetables in Liberia, widely used in cooking alongside other vegetables (Ahmad et al., 2005). This versatile taproot vegetable is consumed in various ways, such as raw in salads or cooked in soups and other dishes (Rahman et al., 2020). However, carrot cultivation in Liberia faces challenges

due to poor soil fertility, which significantly impacts farmers' productivity. Continuous cropping and the limited use of mineral and organic fertilizers have led to nutrient depletion in agricultural soils (Abuzar et al., 2011). Effective nutrient management is essential to improving soil fertility and enhancing crop yields, as highlighted by Silveria and Kohmann (2020). Plants primarily obtain essential minerals from the soil (Vijayprabhakar et al., 2020), and applying the right combination of nutrients is critical for achieving sustainable productivity. However, improper fertilizer application can adversely affect both soil health and crop production (Pandey et al., 2020). Although synthetic fertilizers are commonly used by carrot growers to boost yields, excessive reliance on these fertilizers has raised concerns about their negative impacts on human health and soil quality (Toor et al., 2020). Integrated nutrient management, which combines organic and mineral fertilizers, has shown promise in improving soil fertility and crop yields while minimizing environmental impacts (Singh et al., 2020). Natural soil amendments, such as chicken manure, cattle manure, and solid waste, can enhance soil quality by adding nutrients, improving water retention, and promoting microbial activity (Amjad, 2013; Goel et al., 2020). Organic manures not only improve soil structure but also serve as cost-effective alternatives to synthetic fertilizers, making them more accessible to farmers (Hafez et al., 2020). While organic fertilizers release nutrients more slowly than synthetic options, the combined use of organic and mineral fertilizers has been shown to boost crop productivity, enhance soil health, and reduce pollution risks (Hammad et al., 2020; Karmakar et al., 2020). Despite the potential benefits, limited research has been conducted on the effectiveness of integrated nutrient management in carrot cultivation. Therefore, this study aimed to evaluate the growth and yield response of carrots to combinations of NPK fertilizers and organic manures. This research is important for identifying sustainable nutrient management practices that balance productivity, soil fertility, and environmental protection. The aim of this study was to assess the effects of integrating NPK fertilizers with organic manures on the growth and yield of carrots. Specifically, the study sought to evaluate the yield response of carrots to various organic manure treatments, highlighting the potential benefits of integrated fertilizer application.

Materials and Methods

Descriptions of the study area

The experiment was carried out at the Teaching and Research Farm, William V. S. Tubman University, Harper City, Liberia from January to May 2023 cropping season under rain-fed conditions. The site is situated 4km from the capital of Harper City, in the southeastern region of the country. The study area is located at the coordinates 4°34' N and 7°40' W. It is located in the West African tropical rainforest, where the typical monthly temperatures range from a minimum of 25°C to a maximum of 30°C. The region experiences unpredictable, irregular annual rainfall totaling 1628 mm per year. The study area's natural vegetation consisted of a mix of savanna grasses and forested areas. The soil in the region consists of sandy loam.

Experimental design and treatments

The field trial included four different treatments of chicken manure: 10 tons per hectare, 15 tons per hectare, 20 tons per hectare, and no chicken manure (Control). Additionally, 300 kg per hectare of NPK (15:15:15) fertilizer was applied evenly to each bed at a rate of 0.5 kg per hectare. Each bed measured 2.5 meters in length and 1 meter in width. A distance of 0.4 meters was maintained between each bed. The study was set up using a random complete block design with three replications, as outlined in Gomez and Gomez's (2010) procedures. The experimental plots were randomly assigned treatments within a block.

Soil sample collections

The study obtained soil samples from the surface layer (0-25 cm deep) that were categorized as Latosol according to the USDA's soil classification system. Soil samples were collected from each block of the field using a stratified random sampling method. This was carried out in a randomized complete block design at various plot locations, utilizing core samplers and Dutch augers.

Laboratory Procedures

The soil samples gathered were dried at 105°C in the College of Agriculture and Food Science (CAFS) lab, then crushed in a mortar and pestle and sifted to eliminate debris such as roots and plant components.

Bulk density

The core method (Blake and Hartge, 1986) was used to measure the bulk density. The core samples were dried in an oven at 105°C in the lab and periodically weighed until a consistent weight was reached. The bulk density is defined as the ratio of the dry mass of solid material to the total volume of soil, including both solid components and pore spaces.

Moisture content

The soil moisture level was measured using the oven-drying technique for a period of 24 hours. The sample was brought to the lab and 10g each of CaCl₂, H₂O, and KCl were added. The mixture was stirred for 5 minutes each and left to settle for 30 minutes to allow for ion exchange in order to determine the soil moisture content (Ahn et al., 2014). The weighing dishes were set on a balance scale and adjusted to read zero grams to eliminate the weight of the dishes. The wet soil samples gathered in the field were moved into the containers, weighed using a scale, and the weights of the wet soil samples were documented. The wet soil samples were dried in the oven at a temperature of 105 degrees Celsius. The soil was taken out of the oven and left to cool down. The soils that had been dried in the oven were weighed again, and the weights of the dried soils were noted down. Thus, the percent soil moisture content was calculated as:

$$\% \text{ moisture content (MC)} = \frac{\text{weight of moist soil} - \text{weight of oven-dried soil}}{\text{weight of oven-dried soil}} \times 100$$

Soil pH

The pH of the soil was measured in both distilled water and 0.01 M CaCl₂ solution (with a ratio of 1 part soil to 2 parts solution) using a digital pH meter with a combined glass electrode. The measurement was taken after allowing the soil to equilibrate for 30 minutes, following the method described by Thomas (1996). The pH was measured twice to confirm the results. 10 grams of each soil sample was measured and placed into a plastic pH cup. In the initial group of cups, 10 ml of pure water was mixed, while in the second group, 20 ml of 0.01 M CaCl₂ solution was added to achieve a soil solution ratio of 1:2. The mixture was stirred right away and left to settle for half an hour before measuring the pH with a digital meter. The delta pH (pH change) is calculated as the discrepancy between the pH levels in CaCl₂ and H₂O. The above process was repeated for pH in 1 M KCl by adding 10 ml of KCl solution to 10 g of soil in a 1:1 ratio of soil to solution.

Electrical conductivity

The level of electrical conductivity determines the concentration of salt that is dissolved in the soil solution. The level of soluble salts in the liquid was assessed using the saturated paste technique as per the US Salinity Laboratory Staff's guidelines from 1954. A small quantity of de-ionized water was poured into the beaker before adding an approximately 2/3 full air-dried soil sample. De-ionized water was slowly poured into the beaker containing soil, all the while mixing it. Additional dry soil was incorporated when the mixture was too moist, while water was added when the mixture was too dry during the blending process. After blending, the sample was left to settle for one hour. The thick paste was moved to a Buchner funnel containing filter paper. A vacuum was used to extract and collect the saturated paste in a 250 ml vacuum flask. The extract's temperature was recorded and utilized for calibrating the conductivity meter. The electrical conductance of the saturated paste extract was measured using the JENWAY 4510 model conductivity meter.

Soil organic carbon determination

The soil organic carbon was analyzed using the Walkley and Black modified technique, as described by Nelson and Sommers (1982). An Erlenmeyer flask was filled with one gram of soil that had been dried. Blank samples were also provided for comparison. The samples and empty flasks were each given ten milliliters of 1 N (0.1667 M) potassium dichromate solution. Two milliliters of concentrated sulfuric acid were poured onto the soil slowly and carefully using a measuring cylinder. The mixture was then stirred and allowed to sit for 30 minutes in a fume cupboard. Next, 250ml of distilled water was mixed with 10ml of concentrated ortho-phosphoric acid. After adding 1 ml of diphenylamine indicator, the reaction mixture was titrated with a 1.0 M solution of ferrous sulphate.

Calcium and magnesium determination

The analysis of calcium and magnesium was conducted by adding 25 ml of the extract into an Erlenmeyer flask, along with 2% potassium cyanide, 2% potassium ferrocyanide, hydroxylamine hydrochloride, 10 ml ethanolamine buffer, and 0.2 ml Eriochrome Black T solution. In order to achieve a pure turquoise hue, a solution was titrated using 0.01 M EDTA (Ethylene Diamine Tetraacetic Acid).

Soil particle size distribution

The hydrometer method by Boyoucos (1962) was used to analyze soil texture. To do this, 50 grams of soil that has been dried in air were placed into a measuring cylinder, followed by the addition of 50 milliliters of sodium hexamethaphosphate. The treatment was agitated and allowed to settle. Hydrometer readings were taken and corrected at the 40-second and 5-hour marks.

Experimental materials and procedures

Orange carrot seeds were utilized as the experimental crop for this study. The soil was meticulously cultivated to a depth of 30 cm using traditional tillage methods, creating optimal conditions for planting. Following this, a field layout was created, and each treatment was allocated randomly to the experimental plots. Seeds were planted in raised beds that were 20 cm tall, with spacing determined by the treatment assigned to the plot. Carrot seeds were planted by making drilled rows that were 1 meter by 1 meter in length in every plot. Two weeks before planting, different amounts of chicken manure were applied all at once, while NPK fertilizers in granular form were applied in two stages: half during sowing and the other half during the mid-tillering crop

stage at 35 days after emergence, in between the rows. All plots received the same level of watering and other necessary cultural practices. Weeds were handpicked and gathered from the fields where crops were grown, and harvesting was done when the crops were fully mature using a manual hand tool.

Data collection

The growth indicators of plant height, number of leaves, and leaf area (length x width) were accurately measured by selecting five plants at random. The root length and diameter were determined by measuring parameters from five selected plants with a vernier caliper. The weight of the roots of each plant was measured by weighing five randomly chosen plants with a precise balance, and the average weight was then calculated and utilized for further analysis.

Statistical analysis

The data gathered was analyzed using Analysis of Variance (ANOVA) in SAS 9.1 to test any significant differences among the selected treatments. Mean comparisons were made using the Least Significant Difference (LSD) at 0.05 significant levels.

Results and Discussion

Table 4.1 displays the physical and chemical characteristics of the soil that were examined prior to and throughout the study. The soil bulk density measured at 1.43 g/cm³ indicates the soil's texture and structure. Typically, soils with low bulk densities are linked to increased total porosity, but root penetration and seed emergence may prove challenging when bulk density exceeds 1.6 g cm⁻³ (Russell, 1976). The results fall within the acceptable range for the majority of agricultural practices (Wild, 1993). Additionally, the average soil moisture level was recorded at 14.3%, which falls within the ideal range for cultivating most crops. The moisture content of soil plays a crucial role in regulating the various processes within the soil system, including its physical, chemical, and biological aspects (Buss et al., 2010). Sandy soil accounted for 83.0%, while clay made up 11.0% and fine silt accounted for 6.0% of the soil composition. Additionally, sandy soils typically have greater bulk densities than fine silts and clays due to their larger yet less numerous pore spaces. Clay soils with well-maintained soil structure have more pore space due to the small particle size, allowing for numerous small pore spaces to exist between them. The levels of different macronutrients in the soil samples, including carbon, phosphorus, potassium, calcium, magnesium, and sodium, were found to be moderate and adequate. The nitrogen levels were at a low 0.16%. The presence of Ca, Mg, Na, and K as exchangeable cations suggests a significant capacity for cation exchange. Overall, the soil had a low pH of 3.61, with its acidity likely due to factors such as high rainfall causing leaching of bases from the soil, acid rain, and crop harvesting. The soil pH falls within the highly acidic range of 4.0 - 5.5 according to Adepetu et al. (2014). The soil's pH was lower than the ideal range of 6.0 to 7.0 for most crops like cabbage. The electrical conductivity (EC) measurement was recorded at 0.032 dS m⁻¹. The recorded value fell below the acceptable salinity level of 4 dS m⁻¹ according to Adepetu et al. (2014). The soil EC levels were found to be low, with no concerns about salinity anticipated. Electrical conductivity (EC) indicates the level of soluble salts in the soil solution. The results also show that the levels of iron and copper in soil are optimal for crop growth, with most plants thriving when the copper level is at 0.6 parts per million or higher. The appropriate level of manganese in the soil for crops may differ depending on the type of crop being grown. Soil test results ranging from 1 to 5 parts per million (ppm) are

generally considered satisfactory. Manganese deficiencies tend to be found primarily in soil with a pH of 8.0 or higher. The findings indicate that manganese levels are significantly low across all the sites where samples were taken. A zinc soil test level of 1.5 ppm or higher is adequate for the majority of crops.

Table 1: Physical and chemical properties of topsoil analyzed before planting from the study area

Soil physical and chemical properties	Constituent units
pH (1:1)	3.61
N (%)	0.16
OC (%)	1.05
Sand (%)	83.0
Clay (%)	11.0
Silt (%)	6.0
Ca (cmol Kg-1)	2.26
Mg (cmol Kg-1)	1.41
K (cmol Kg-1)	0.63
Na (cmol Kg-1)	0.27
EC (dS m ⁻¹)	0.032
P (cmol Kg-1)	5.83
Mn (ppm)	0.18
Fe (ppm)	0.34
Cu (ppm)	0.91
Zn (ppm)	4.80
Bulk Density (g cm ⁻³)	1.42
Moisture Content (%)	14.6

Growth parameters of carrot variety among the selected treatments under an open field system

The mean growth analysis (number of leaves, leaf area and plant height) of carrot evaluated in an open field system is shown on table 4.2. The higher values of plant height (19.10cm), leaf number (15.15) and leaf area (39.76 cm²) were observed at the 20 ton ha⁻¹ chicken manure application, while the lowest values of plant height (6.84cm), leaf number (10.22) and leaf area (13.18cm²) were recorded in the control (0 t ha⁻¹) chicken manure. When crops have to compete with their neighboring plants for soil nutrients and sunlight, their health and growth can be negatively impacted. Poorly functioning plants will not attain their desired height or leaf area and their roots will have to compete not only for nutrients and water but also for space. According to Kiran *et al.*, (2016) found a significant increase in plant growth of carrots with the application of higher dose of compost than its lower dose. Moreover, leaf weight can be affected by the accumulation and the partitioning of synthesized food to non-photosynthetic parts Osorio (*et al.*, 2014). Leaf area size is likely to enhance photosynthesis, leading to the production of more leaves, which has been supported by the findings of Appiah *et al.*, (2017) in other plants.

Table 2. Mean plant height (cm), mean leaf number, and Leaf Area (cm²) of carrot

Treatment	Plant height (cm)	Leaf number	Leaf area (cm ²)
10 ton/ha Chicken Manure	(11.67) ^c	(11.23) ^c	(27.89) ^a
15 ton/ha Chicken Manure	(13.52) ^b	(12.51) ^b	(33.82) ^b
20 ton/ha Chicken Manure	(19.10) ^a	(15.15) ^a	(39.76) ^a
Control	(6.84) ^d	(10.22) ^d	(13.18) ^d

Mean values followed by different letter(s) are significantly different at 5% probability.

Yield evaluation of carrot variety among the selected treatments under an open field system

The study showed that the highest root length (19.31cm), root diameter (7.87mm), and root weight (31.76g) were observed with the application of 20 tons per hectare of chicken manure. The next best results were seen with 15 tons per hectare of chicken manure, with values of 17.03 cm for root length, 5.37mm for root diameter, and 22.01g for root weight. In contrast, the control treatment (0 t ha⁻¹) showed the lowest values for root length (6.84cm), root diameter (2.16mm), and root weight (10.33g) (Table 4.3). The research clearly demonstrated that using more chicken manure led to longer roots with a greater diameter, ultimately resulting in higher plant yield. This could be because the roots had enough resources to grow and develop without facing as much competition for soil nutrients. According to the research conducted by Kiran et al., (2016), compost is highly effective in enhancing soil fertility, especially when used for the cultivation of vegetable crops. Adelaide (2011) also discovered that compost is an effective fertilizer that can help sustain soil fertility levels and enhance agricultural output. The results align with the observations made by Asante et al. (2019), who noted notable differences in carrot root thickness due to the use of chemical fertilizers and organic compost.

Table 3. Root length, root diameter, and root weight comparisons among the selected treatments

Treatment	Root length (cm)	Root diameter (mm)	Root weight (g)
10 ton/ha Chicken Manure	(15.51) ^c	(3.97) ^c	(15.10) ^c
15 ton/ha Chicken Manure	(17.03) ^b	(5.37) ^b	(22.01) ^b
20 ton/ha Chicken Manure	(19.31) ^a	(7.87) ^a	(31.76) ^a
Control	(6.84) ^d	(2.16) ^d	(10.33) ^d

Note: Mean values followed by different letter(s) are significantly different at 5% probability

Conclusions

Effective agronomic practices greatly impact the growth and productivity of carrots, with soil fertility playing a crucial role in management. The experiment results indicated that the growth and yield traits, such as plant height, leaf number, leaf area, root length, root diameter, and root weight, were most favorable when using the highest rates of chicken manure. The average of these parameters was found to be greater with the highest chicken manure application rate of 20 tons per hectare. The differences observed were statistically significant ($p \leq 0.05$), probably because of variations in nutrient levels that support the plant's root length, diameter, and weight during growth and development. The minimal metrics may be due to the insufficient amount of chicken manure being used on the soil. The study results suggest that the soils at the site do not exhibit significant characteristics that could interfere with the study parameters. Hence, it is essential to work together to enhance soil fertility by utilizing a combination of organic and inorganic fertilizers as an integrated approach.

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Consent to participate: Participants were informed about their right to ask questions relating to the research.

Consent for publication: Informed consent was obtained from all subjects involved in the study.

Data availability: Data used to support the findings of this study are available from the corresponding author upon request.

Authors contribution: Paye Deekermue and Aloysius H. Tye conceived the study idea, co-wrote the paper, and Investigated data collection and analysis; Oluwatosin Mayowa Olajide conducted the soil laboratory analysis and Abubakar Shehu Yaradua revised the paper.

References

- Abuzar M.R., Sadozai G.U., Baloch M.S., Baloch A.A., Shah I.H., Javaid T., Hussain N., 2011 Effect of plant population densities on yield of maize. *J. Animal Plant Sci.*, 21(4): 692–695.
- Adelaide, M. (2011). Influence of temperature on yield and quality of carrots (*Daucus carota* var. *sativa*). November, 1–84.
- Adepetu, J. A., Adetunji, M. T. and Ige, D. V. (2014). Soil Fertility and Crop Nutrition by J. A. Adepetu (2014). Printed by Jumak Nig. Ltd. Jumak Publishers, Ringroad, Ibadan. pp. 1 – 560.
- Ahmad Z, Ali N, Ahmad M, Ulhag S, Ahmad S (2005). Yield and economics of carrot production in organic farming. *Sarhad J. Agric.*, 21(3): 357-364.
- Ahn, J. Y., Kil, D. Y., Kong, C., & Kim, B. G. (2014). Comparison of oven-drying methods for determination of moisture content in feed ingredients. *Asian-Australasian Journal of Animal Sciences*, 27(11), 1615-1622. <http://doi.org/10.5713/ajas.2014.14305>
- Almendo-Candel, María Belén, Ignacio Gómez Lucas, Jose Navarro-Pedreño, Antonis A. Zorpas. Physical properties of soils affected by the use of agricultural waste. *Agricultural waste and residues*. 2008; 2(1):77- 99
- Appiah E.A., Kugbe J.X., Rufai A.M., 2021. Maize growth and yield response to incremental rates of phosphorus in p-depleted lxisols of Nothern Ghana. *Inter. J. Agric. Envir. Biores.*, 6(3): 257275.
- Asante, K., Manu-Aduening, J., & Essilfie, M. E. (2019). Nutritional Quality Response of Carrot (*Daucus carota*) to Different Rates of Inorganic Fertilizer and Biochar. *Asian Journal of Soil Science and Plant Nutrition*, 5(2), 1–14.
- Black CA. Methods of soil analysis, Part I. Physical and mineralogical properties, including statistics of measurement and sampling. Part II. Chemical and microbiological properties. (M. ASA, Ed.) *Agronomy Journal*. 1986; 54:464-465
- Bouyoucos GJ. Hydrometer method improved for making particle size analysis of soils. *Journal of Experimental Botany*. 1962; 57.
- Busse, M. D., Shestak, C. J., Hubbert, K. R. and Knapp, E. (2010). Soil physical properties regulate lethal heating during burning of woody residue. *Soil Science Society of America Journal* 74: 947-955.
- Fallah, S., Mouguee, S., Rostaei, M., Adavi, Z., Lorigooini, Z. and Shahbazi, E. 2020. Productivity and essential oil quality of *Dracocephalum kotschyi* under organic and chemical fertilization conditions. *Journal of Cleaner Production*, 255: 120189.
- Goel, R., Debbarma, P., Kumari, P., Suyal, D.C., Kumar, S. and Mahapatra, B.S., 2021. Assessment of soil chemical quality, soil microbial population and plant growth parameters under organic and conventional rice–wheat cropping system. *Agricultural Research*, 10(2), pp.193-204
- GOMEZ K.A., GOMEZ A.A., 2010 Statistical procedures for agricultural research. Wiley and Sons Inc., New York, USA, pp. 680.

- Hafez, M., Popov, A.I. and Rashad, M. 2020. Integrated use of bio-organic fertilizers for enhancing soil fertility–plant nutrition, germination status and initial growth of corn (*Zea mays* L.). *Environmental Technology & Innovation*, 21: 101329.
- Hammad, H.M., Khaliq, A., Abbas, F., Farhad, W., Fahad, S., Aslam, M., Shah, G.M., Nasim, W., Mubeen, M. and Bakhat, H.F. 2020. Comparative effects of organic and inorganic fertilizers on soil organic carbon and wheat productivity under arid region. *Communications in Soil Science and Plant Analysis*, 51(10): 1406-1422
- Karmakar, S., Bhattacharyya, A., Ghosh, B., Roy, R., Kumar, S., Kar, B. and Saha, G. 2020. Suitability of coupling application of organic and inorganic fertilizers for crop cultivation. *Ecological and Practical Applications for Sustainable Agriculture*, Springer, pp. 149-177.
- Kiran, M., Jilani, M. S., Waseem, K., & Marwat, S. K. (2016). Response of Carrot (*Daucus carota* L.) Growth and Yields to Organic Manure and Inorganic Fertilizers. *American-Eurasian J. Agric. & Environ. Sci*, 16(6), 1211–1218.
- Nelson DW, Sommers LW. Total carbon, organic carbon and organic matter. In: Page, A.L., Miller, R.H and Keeney, D.R. (eds.). *Methods of soil analysis. Part 2. Second edition. Chemical and microbiological properties*. Wisconsin USA. American Society of Agronomy and Soil Science Society of America, 1982, 301-312
- OSORIO S., RUAN Y.L., FERNIE A.R., 2014 An update on source-to-sink carbon partitioning in tomato. *Frontiers Plant Sci.*, 5: 516.
- Pandey, M., Shrestha, J., Subedi, S. and Shah, K.K. 2020. Role of nutrients in wheat: A review, *Tropical Agrobiodiversity*, 1(1):18-23.
- Rahman, N., Uddin, M.B., Quader, M.F.B and Bakar, M.A. 2020. Optimization of mixed peels from banana, carrot and apple to develop high fiber biscuit. *International Journal of Natural and Social Sciences*, 7(1): 21-5.
- Russell, E. W. (1976). *Soil condition and plant growth*. Longman publications Limited London. 10th Edition. 80pp.
- Silveira, M.L. and Kohmann, M.M. 2020. Maintaining soil fertility and health for sustainable pastures. In: *Management Strategies for Sustainable Cattle Production in Southern Pastures*, Elsevier, pp. 35-58
- Singh, B., Singh, A., Singh, T. and Singh, N. 2007. Integrated nutrient management in carrot (*Daucus carota* L.). *Progressive Agriculture*, 7(1&2): 84-86.
- Thomas, G. W. (1996). Soil pH and soil acidity. In: *Sparks, D. L. (ed.), Methods of Soil Analysis Part 3: SSSA Book Series No. 5* Soil Science Society of America Inc., American Society of Agronomy Inc, Madison, Wisconsin, pp. 363 – 376.
- Toor, M.D., Amin, M.M., Khan, B.A., Nadeem, M.A., Usman, M., Faizan, M., Arshad, A., and Zafar, K. 2020. Consequence of surplus fertilizers and nutrients: a review on effect on plants and humans, *International Journal of Botany Studies* 5(3): 360-364.
- US Salinity Laboratory Staff, (1954). *Diagnosis and improvement of saline and alkali soils*. US Department of Agriculture Handbook, Washington, DC, 60.
- Vijayaprabhakar, A., Hemalatha, M. and Joseph, M. 2020. Utilization of paddy straw as a source of nutrients for succeeding paddy and its effect on soil available nutrients, nutrient uptake and crop yield. *International Journal of Farm Sciences*, 10(1): 53-58.
- Wild A. (1993). *Soils and environment, An introduction*. Great Britain: *University of Cambridge*.