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ARTICLE

Organic Hydroponics Production

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ABSTRACT

Hydroponic culture is a controlled systems use a soilless growing media, supply all of the plant's nutrition in the plant's solutions (water with dissolved fertilizers), result in higher yields of vegetables, flowers, herbs and others crops. Hydroponic systems derive in many various forms and types. Most traditional hydroponic systems are extremely specialized, controlled-environment production systems. Organic hydroponics is a system that is arranged based on organic agriculture of culture. Different approaches are used for controlling of plant pathogens such as physical, chemical, biological controls, biofertilizers, bioremediators and integrated pest management. All the required nutrients are supplied in controlled amounts, including organic crops. This article discuss the way for promoting organic hydroponics systems and to help the small-scale producer make decisions about follow this markets, production methods, and disease control.

1. Introduction

Hydroponics is basically plants that grow without traditional soil [1]. It is a more impact to provide increase productivity of plants. Plants do not use traditional soil - they use food and water in the soil. The function of the soil is just to supply plants with nutrients and to root plants. In the water park, provide plants with a complete nutritional formula and an inert growth medium to anchor the roots of plants so that food and water are easily accessible.

The management of organic hydroponics it needs much more attention than conventional hydroponics. Not in terms of elapsed time, but in terms of "visual attention". In fact, are organic hydroponics administered by eye the same as with pH and EC, although they are still important references [1].

Hydroponic solution is developed by National Agriculture and Food Research Organization (NARO), in Japan, in 2005 [1]. Originally, 'organic culture' it is means nutrients that promote a healthy plants is developed by European regulation 834/2007 on organic agriculture. The National Organic Standards Board defines organic farming for an aquaculture system as "an environmental production management system that promotes and enhances biodiversity, biological cycles and soil biological activity." [2]. The National Organic Standards Board has made recommendations on organic hydroponics for plants and fish. In November 2017, the US is arranged a rule of organic hydroponics with label and certifies. In spite of the opposition of many organic stakeholders, some accredited certifying agents are approving hydroponic process. The

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United States is joining with 24 other countries, including England, the Netherlands, and Mexico, in prohibiting organic certification for hydroponic produce [2]. Organic and conventional production systems run on the same rules of supplying nutrients in solution to support growth of plants. Studies for promoting organic hydroponics have been processed for a long time. The main variation between the two systems "conventional systems supply chemical components for nutrient supply" and "organic hydroponics" is the fertilizers and pesticides that will dissolve in water in the conventional system but in other one "organic hydroponics" the use of chemical fertilizers and pesticides not allow to do that. The first feasible organic hydroponics systems that organic fertilizer and microorganisms can be added immediately during cultivation in Japan that [2] invented the new method to originate organic hydroponic system which consists of three handling: (1) inoculation of soil beneficial microorganisms, (2) adding of small amounts of organic fertilizer, and (3) aeration. In this system, terrestrial plant roots are not in soil but sort in air, water, or a solid medium, such as vermiculite, coconut, or perlite. The roots are periodically dipped in water or bathed in a nourishing and organic solution [2].

2. Beneficial Microorganisms in Hydroponic Systems

The beneficial microorganisms can be used in the hydroponic and aquaponic systems which play a major role in the system to give clear and safe production of fruits. In hydroponic systems, most of the microbial work focuses on the some special kind of bacteria [3]. Microorganisms can be used in an organic hydroponic system, by organic fertilizer decomposing into inorganic nutrients in aqueous solution by treatment with ammonia and nitration [1,4]. Organic hydroponics systems works through a symbiotic relationship between microorganisms and plants which is necessary to provide additional microbial habitat. Microorganisms are provide a base for beneficial to reproduce and from which they can colonize plants.

3. Biofertilizers Strategies in Hydroponic

Growth promoting root bacteria (PGPR) strains and fungi have already been successfully tested in growing vegetables and fruits in aquatic systems, with positive effects on growth, yield, product quality and safety [3]. These differences, and especially the vital nutrient fraction, could investigate several factors and play a critical role in contributing to plant nutrition [5]. The different mechanisms are known by PGPRs by increasing the bioavailability of mineral nutrients in the root zone (i.e. fixation of N2 in the atmosphere, dissolution of P, production of iron acid for Fe3 + chelation) [6]. Interestingly, the application of specific PGPRs, such as *B. sphaericus* UPMB10 and *A. Brasilense Sp*7, nitrogen fixation (N2), reduced the external input of nitrogen sources into the aqueous solution used to grow banana plants without soil [7]. A mixture of PGPRs cultured in hydroponic systems of food crops such as wheat, potatoes, and soybeans was introduced for a complete life cycle, and the formation of these microbial communities was related to their roots, rhizomes, and endosphere. A recycled nutrient solution was described via the 16S - sequence [8].

There are technical difficulties, such as the management of chemicals and root by-products, which hinder the implementation of more closed aquaculture. Limited studies have been conducted on the use of materials of organic origin such as muddy manure and compost sap but cannot be replaced with a chemical solution [9-13]. With complementary organic matter, there is a need to analyze organic matter to avoid any damage from increased nitrogen level and the system remains dependent on the continuous input of external organic matter.

Organic hydroponics aims to create a mixed technology between the ancient known soil culture and the new concept of hydroponics to create a sustainable and environmentally sound production system [13]. Many researchers are trying to create a hybrid technology that can have more benefits with fewer problems compared to conventional soil and hydroponics. The goal was to make it as completely sustainable as a micro-environmental cycle. By extracting nutrients from the soil and exposing them to plant roots in liquid form, plants can absorb the minerals they need and the roots are re-secreted into the soil in a closed system, where they can interact and release excess minerals from the soil. The result will be a hydroponic system, which will be sustainable, easy to manage, cost-effective and environmentally friendly system.

Some work on organic hydroponics has already been implemented with supplementary feeding solutions but a fully sustainable system has not been introduced. Creating an organic self-sufficiency system for horticultural production not only responds to consumers' demand for these naturally managed crops, but also reduces the cost of production and simplifies the production process as preferred by the producers and can be considered a response to public sector demand in an environmentally friendly way to produce a greenhouse. Furthermore, expect a decrease in the incidence of disease in this system due to the great impact which can create a balance

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between all opposing parties giving the opportunity for plant growth. It can be concluded that the organic approach can extend to the organic hydroponic system with acceptable yield and quality. Using this system especially for food crops could be more attractive due to strict legislation on chemical waste and hazards. The following elements define the benefits and potential of a sustainable organic hydroponic system; in custom biodynamic systems, plants are given a pre-defined formula, which is ideal based on past experiences. Given that environmental factors actually change in the long term and at diurnal scales, in reality, plants cannot get what they want with the required consistency. In a sustainable organic water system, the plant can interact with the medium and can influence the concentration of certain nutrients by managing both the concentration and diversity of root exudates. Therefore, plants in this system can react to environmental stimuli and stresses through active interaction with the soil component of the system. The system is much simpler in terms of preparation and management compared to the traditional water system, so the related tool and labor costs are greatly reduced.

Supporting plants with better nutritional status identified through adaptive plant physiology can add value to the product through better product quality and post-harvest performance [13]. The use of sustainable and organic methods to produce hydroponics is a significant achievement for the protected horticulture industry. Increasing consumer demand for health products resulting from lower environmental costs in the future supports such initiatives. This system can be easily adapted with organic principles to respond to the sector's demand for greenhouse products. Creating a self-sustaining production method is the key to the great success of human survival in outer space. For example, plant production units on the Moon could be established using local ore mineral sources through this approach [12]. As a new area of research, the appropriate type of soil, the appropriate volume required for each plant, the function of the nitrogen fixation unit and the improvement of conditions to obtain the best results are the main areas that should be investigated. In addition, the stability of the system and the mode of action induced by the plant root should be examined with soil effects and soil nutrient release in the section. The role of soil organic matter in this system and the benefits of adding an appropriate type of external organic matter could be another area of research. The emergence of organic hydroponics came in the 1990s when mineral nitrogen mineralization to nitrates became possible through various steps [14,15], despite the emergence of some challenges such as having to use different bioreactors for ammonia and for nitrification [16], little success in achieving this [17], until [4], used microbes from various sources to mineralize organic nitrogen to nitrates in the same media, which are potentially organic hydroponics has been fully achieved. Nitrogenous bacteria are cultivable for compost mineralization in various conditions such as maintaining a pH at 7.7 - 8.4, a temperature of 35 ° C, 6.5 mg / l of dissolved oxygen, or some parameters such as temperature at 25 ° are different. The resulting nutritional solutions to crop production have already demonstrated benefits such as the effectiveness of both controlling soil and airborne diseases in vegetables such as lettuce and cucumber [18]. This development comes as a relief for societies with less financial ability to access hydroponics through organic sources of plant nutrition requirements. This considers that the use of traditional fertilizers for agriculture is relatively expensive worldwide [19], especially in Africa [20].

4. Bio-Control Strategies in Hydroponic

Plant pathogens occurring in hydroponics systems are generally found in soilless systems especially of hydroponic plant culture is the continuous presence of water in the system. This environment is suitable for most types of fungi and plant pathogenic bacteria. Soil-borne fungi, such as *Fusarium spp.*, *Phytophthora spp. Rhizoctonia, Macrophomina* (Figure 1), and *Pythium spp.* Verticillium and Didymella are among the most problematic pathogens due to their preference for moist environment conditions that cause especially heavy losses in leafy vegetable crops [21]. Bacteria, such as Ralstonia, Xanthomonas, Clavibacter, Erwinia, and Pseudomonas are also found in Hydroponics' irrigation system that cause damage to stem, leaves and / or fruit [22]. In the hydroponic system, the control methods are still chemical fungicides that cause toxicity to fish and beneficial bacteria (such as nitrifying bacteria in the biofilter (Figure 2). Moreover, the development of biological control agents for use in hydroponics is still in its infancy. Through the different methods used to control plant pathogens using physical, chemical and biological methods, we focus on biological controls, especially plant growth promoting root bacteria (PGPR) which are used in bio-fertilizers, biological control agents, bio-stress and biological treatments. Experiments introducing microorganisms into hydroponic systems have focused on increasing nitrification by using nitrifying or plant growth-stimulating root bacteria such as *Azospirillum brasilense* and *Bacillus spp.* To increase plant performance and productivity [23]. In hydroponics, various anti-microorganisms are used to control plant pathogens. There is now a rapid need to introduce bio

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control agents (BCA) for plant pathogens into hydroponics and reduce the use of chemical pesticides. In general, bio control agents are easier to introduce into soilless culture systems which are more accessible than soil [26]. Several "bio control agents" BCAAs are effective against plant pathogens (Figure 3) [24-26]. A number of microorganisms useful for this study, such as *Pseudomonas spp*., *Bacillus spp*., *Streptomyces spp*., *Gliocladium spp*. and *Trichoderma spp*. [27]. Recently, hydroponic system have begun using Arbuscular mycorrhizal (AM) fungi inoculum for increasing yields and provide sustainable growing conditions in organic production [28]. *Arbuscular mycorrhizal* (AM) fungi have been shown to be able of making nutrients ready to crop plants and providing best transplant achievement by offering higher root and shoot fresh weight, biomass, growth rate as well as protect plant from diseases caused by root pathogens in hydroponic production systems [28]. However, Growth promoting root bacteria PGPM-treated plants were shown to be the most effective and more stable over time of most research on hydroponic systems [8].

Figure 1. Infected vegetables grown under hydroponic system without control system in Bohera governorate (Cultivar Station of National Research Centre during 2020)

Figure 2. Vegetables grown under hydroponic system with chemical control system in Bohera governorate (Cultivar Station of National Research Centre during 2020)

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Figure 3. Vegetables grown under hydroponic system with biological control system in Bohera governorate (Cultivar Station of National Research Centre during 2020)

5. Integrated Pest Management

Diseases and insect are a main challenge in hydroponic production systems [28]. Integrated Pest Management (IPM) play an important role in the management of these diseases and insect and organic food production pesticides [29]. Diseases and insect are a main challenge in hydroponic production systems. Integrated Pest Management (IPM) play an important role in the management of these diseases and insect and organic food production pesticides [29]. IPM include the use of integration system from cultural, physical, biological, and natural chemical to cultivate crops with low use of chemical fungicides and. Cultural Control through an IPM program starts via prevent pathogens and insect pests from growing in the production area . Physical control methods are main to prevent pests to build it, where possible. Biological control of pathogens and pests via an IPM program include the release of natural beneficial microorganisms and natural enemies [29].

6. Future Perspective

The object of this review aimed to give an effective methods and future possibilities to use organic agriculture in hydroponic using beneficial microorganisms and natural products. The use of organic matters in this system is an supporting for making use of organic

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fertilizers, organic plant media or organic amendments. The use of biological control means supply of beneficial microorganisms by manipulating and managing water composition i.e. C/N ratio, nutrients and gases and pH value which is importance to sustain good nitrification and keep healthy fish. In addition, biological control by releasing of beneficial microorganisms to produce safe food and environment is needed. Integrated plant pest management (IPM) is use to management the system and reduced development and spread of plant pathogens and decreases the use of chemical pesticide. In generally, plants growing in hydroponic systems are healthy with less vulnerable to pathogens and pest rapid and attacks. In hydroponic systems, most of farms use minimal chemicals pesticides and fungicides, they may even be considered organic for safe food.

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ARTICLE

Adaptation Strategies to Mitigate Impact of Climate Change on Food Crops Farming in Oyo State, Nigeria

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ABSTRACT

The research investigated the adaptation strategies to mitigate consequence of climate change on food crops farming in Oyo State. 120 respondents were selected for this study using multi- stage sampling procedures. Primary data was collected through interview schedule and analyzed using both descriptive and inferential statistics. Available results indicated that 84.2% of the respondents were male, 93.3% of them were married and maize (45.8%), cassava (37.5%) are the mainly crops cultivated. Results also revealed that 70.0% of the respondents have knowledge of climate change with majority (84.17%) of them regularly accessed information on climate change through radio and 88.3% of them claimed to adopt planting crops favorable for the present weather condition as an adaptation techniques to mitigate the consequences of climate change more frequently. Chi-square and Correlation results revealed a significant relationship existed between farmers educational levels (X2= 4.861; p= 0.003); household size (r= -0.089; p=0.002); knowledge (r= -0.157; p= 0.002), and adaptation strategies to reduce the consequences of climate change on the food crops farming. It was recommended that food crop farmers should be provided with better education and sensitized in order for them to be acquainted with adaptation techniques and coping mechanisms that are currently been offered by research.

1. Introduction

Climate change shows multiple stresses on the biophysical, social and institutional environments that corroborate agricultural production [9]. A modification in the vegetation type, distribution and coverage may occur given a change in the climate. Some modifications in climate may lead to an increased in precipitation, warmth, improved plant growth and the subsequent sequestration of airborne carbon dioxide. Gradual increase in warmth in a region will result to alteration in the timing of life cycles of dependent organisms, earlier fruiting and flowering times, however, cold will cause plant bio-cycles too late. Larger, faster or more radical changes conversely may result in vegetation stress, rapid crops loss and desertification in certain circumstances.

Agricultural crops are drastically vulnerable to climate change, higher temperatures reduces the yields of desirable's crop, encouraging weeds and pests

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proliferation. Changes in patterns of precipitation increases the likelihood of short-run crop failures and long-run production declines, however, there may be gained in some crops in some regions of the world but the overall effects of climate change on agriculture are expected to be negative. According to [4], assessment of the consequences of climate change on crop yields is frequently negative for the tropics. [11] emphasized some of the direct consequences of climate change on agricultural system as seasonal changes in rainfall and temperature which could influence agro-climatic conditions, changing growing seasons, planting and harvesting periods, water availability, pests and diseases infestation, weeds, alteration in land suitability for crops production, evapo-transpiration and photosynthesis. The most devastating consequences of climate change in Nigeria and other subtropical countries includes environmental damage, pests and diseases infestation of crops, drought and biodiversity loss [1]. [12] reported that variations in rainfall pattern affect s crop development and phonology thereby resulted to yields loss.

In an attempt to address food crops loss and insecurity, adaptation techniques adopted by food crops farmers have become necessary particularly in Nigeria where farmers are drastically affected. Adaptation to climate change is modification made to human, ecological or physical system in response to vulnerability [2]. Climate change adaptation through the modifications or improvement of agricultural practices become imperative to continue meeting the growing food demands of modern society [13]. Adaptation is one of the policy options that helps farmers achieved their food, livelihood security and income objectives in the face of changing climatic conditions [10]. Farmers especially food crops farmers can reduce the potential damage by making tactical responses to climate effects such as irrigation methods, use of hybrids crop, mixed cropping, changing of planting dates and diversification of crops [5,8]. Analyzing adaptation strategies is therefore important for finding ways to help food crops farmers in rural economies of Nigeria, hence, the study sought information on the socioeconomic characteristics of the respondents, their enterprise characteristics, sources of information on climate change and degree of accessibility, knowledge of climate change and adaptation strategies to reduce the consequences of climate change on food crops farming. It was hypothesized that no significant relationship existed between respondent's socioeconomic characteristics, knowledge of climate change and adaptation strategies mitigate consequences of climate change.

2. Methodology

The study was carried out in Oyo State, Nigeria and the state lies between latitude 7.0^{o}N and 9.3^{o}N of the equator and between latitude 2.5^{o}E to 5.0^{o}E of the prime meridian. 120 respondents were selected using multi-stage sampling procedure. The first stage involved the categorization of local government areas of Oyo state into rural and urban. The second stage involved simple random selection of 4 rural local governments out of 21 rural local governments areas which are Ido , Egbeda , Afijio , and Akinyele local governments which represented 20%. The third stage involved simple random selection of eight (8) wards in selected four rural local governments which represented 20% of the wards in each of the local government areas namely Egbeda local government area(11 wards), Ido local government area (10 wards),Akinyele local government area (12 wards), Afijio local government area (10 wards). The fourth stage involved simple random selection of 2 communities from each selected wards giving a total of sixteen (16) communities. The fifth stage involved simple random selection of 8 food crops farmers from the selected communities which gives a total sample size of 128 food crops farmers; however, sampling rate was 94% which represented 120 farmers. Data for this study were obtained through the use of interview schedule and analyzed through the use of descriptive statistics that involved frequency, percentage and inferential statistics (Chi-square and PPMC - Pearson Product Moment Correlation) at 0.05 level of significance.

3. Result and Discussion

3.1 Socio-economic Characteristics of the Respondents

Available statistics in Table 1 revealed that the respondents have a mean age of 44.4 years, this suffices to say that food crop farmers in Oyo state are in their active age, this is in agreement with [3] that population within this age group are energetic and constitute active work force. 84.2% of the respondents were male and this may be due to the fact that women are more involved in off-farm activities as postulated by [7] that women are more involved in off-farm activities than men, especially in the area of transportation of farm produce, fire wood fetching, processing of farm produce, feeding of household members and reproductive functions. Also, 93.3% of the respondents were married and they were Christians (68.3%).In terms of their level of education a sizeable proportion (45.0%) had primary education, 37.5% had secondary education which signified their low level of educational attainment which likely to prevent them from climate change information access, this view is supported by [15] that higher levels of educational

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attainment give farmers the advantage of awareness of innovation on agriculture via communication channels (radio, television of print media). Also revealed is income of the respondents, it is shocking to know that a chunk (51%) of the respondents still earned as low as ₦ 100,000 per cropping season, this suggests that majority are still into subsistence production. Only a few (15.7%) earned above #350,000 per cropping season, suggesting that more efforts should be made to intensify production with mean income of N235, 725.00k which implies that majority of the respondents earned low from this venture. Other income generating activities farmers engaged in include poultry production (26.67%), transportation service (21.66%), bee keeping (14.17%), fish production (13.33%),petty trading (11.67%),artisan services (8.33%) and fish processing (4.17%).It is viewed that crop farmers engaged in these income generating activities at their leisure and obtained income from these venture to augment income got from crop farming. With regards to household size respondents have a fairly large household size of between 4-6, this shows that rural household heads have fairly large household size to cater for, hence, these household size can be deployed as work force. Household size has a great role to play in family labour provision in the agricultural sector [14].

Table 1. Distribution of the respondents based on their socio-economic characteristics

Characteristics	Frequency	Percentage
Age (mean=44 years)		
21-30	15	12.5
31-40	34	28.3
41-50	33	27.5
51-60	30	25.0
61-70	8	6.7
Gender		
Female	19	15.8
Male	101	84.2
Marital status		
Married	112	93.3
Single	3	2.5
Divorced	2	1.7
Widowed	3	2.5
Religion		
Islam	36	30.0
Christianity	82	68.3
Traditional	2	1.7
Educational status		
Adult education	7	5.8
No formal education	8	6.7
Primary education	54	45.0
Secondary education	45	37.5
Tertiary education	6	5.0
Income (per cropping Season) mean=235,725.0		
(#) <50,000	24	21.2
50,001-100,000	41	29.8
100,001-150,000	13	10.7
150,001-200,000	6	5.0
200,001-250,000	8	6.7
250,001-300,000	7	5.9
300,001-350,000	6	5.0
Above 350,000	15	15.7
Other Income generating activities		
Fish processing	5	4.17
Petty trading	14	11.67
Artisan	10	8.33
Bee keeping	17	14.17
Poultry Production	32	26.67
Fish production	16	13.33
Transportation	26	21.66
Household size(mean=5.25)		
1-3	14	11.7
4-6	81	67.5
7-9	24	20.0
Above 9	1	0.8
Total	**120**	**100**

3.2 Production Characteristics

Available data in table 2 revealed that 60.0% of the respondents have farm size between 1-5 acres and 26.7 % of the respondents have between 6-10 acres which signified that most crop farmers are involved in subsistence production with a mean farm size of 6.98 acres cultivated. Half (50%) of the respondents practice mixed cropping system and close to half 40.8 are into mono cropping while none of the respondents practice taungya cropping system. A substantial proportion of the respondents 61.7% engaged in crop rotation, this is an indication of food crop farmers acquainted with the benefits accrued from engaging in crop rotation, an ample proportion of the respondents 47.5% had betwwen1-10 years of farming experience while 27.5% had between 11-20 years of farming experience and 17.5% had between 21-30 years of experience in food crops activities, with an average value of 14.04 years of food crops farming it portrays that the farmers are not new in this business and have ample experience of the consequences of climate change. In addition, 51.7% of crop farmers made use of inorganic manure while 48.3% made use of organic manure as a means of replenishing soil nutrients, it suggests that crop farmers made use of inorganic to reduce cost of production as majority are small scale producers. Source of finance used by respondents include personal savings (63.3%), cooperative societies (27.5%), microfinance banks (5.0%) and from family members

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(4.2%), it is evident crop farmers plow in the funds got from other income generating activities into their main venture (crop production) and have not fully explored the benefits attached to sourcing funds from cooperative societies. Ample proportion of the respondents cultivated maize (45.8%) and cassava (37.5%) other crops cultivated include yam, rice, cowpea with 14.4%, 1.67% and 0.83% respectively, this is a reflection of the crops we have relative advantage in growing in this state.

Table 2. Distribution of respondents based on their production characteristics

Characteristics	Frequency	Percentage
Size of the farms (Acres)(mean=6.98)		
1-5	72	60.0
6-10	32	26.7
11-15	5	4.1
16-20	2	1.7
Above 20	9	7.5
Cropping system		
Mono cropping	49	40.8
Continuous cropping	11	9.2
Mixed cropping	60	50.0
Taungya system	-	-
Farming system		
Shifting cultivation	4	3.3
Crop rotation	74	61.7
Mixed farming	42	35.0
Farming experience (mean=14.04 years)		
1-10	57	47.5
11-20	33	27.5
21-30	21	17.5
31-40	8	6.7
Above 40	1	0.8
Main means of replenishing soil nutrient		
Organic manure	58	48.3
Inorganic manure	62	51.7
Main Source of labour		
Family member	74	61.7
Paid labour	19	15.8
Friends	2	1.7
Self	25	20.8
Main Source of finance		
Personal savings	76	63.3
Family member	5	4.2
Micro finance banks	6	5.0
Cooperative society	33	27.5
Crops cultivated		
Cassava	45	37.5
Rice	2	1.67
Cowpea	1	0.83
Maize	55	45.8
Yam	17	14.2
Total	120	100.0

3.3 Climate Change Information Sources Accessible to Farmers

Available data in table 3 revealed that 84.17% of the respondents regularly accessed information on climate change through radio, this is in consonance with the study of [6] which emphasized on radio as the most frequently accessed source of information by rural dwellers .A sizable proportion of the farmers (68.33%) depend on agricultural extension agents as their source of information on climate change and this can be adduced to the increase in sensitization carried out by agricultural extension agents to expose farmers to the vagaries of weather, its effects and adaptation strategies to be deployed in mitigating it, this is corroborated by the findings of Yahaya [16] which highlighted that extension agents is the second most readily accessible source of agricultural information to farmers. A notable proportion (62.50%) of the farmers accessed information on climate change from farmers association, it should be appreciated that farmers regularly share information and experiences of climate change during meetings, it is also noted that formidable groups like farmers association are usually identified with and receive sensitization and capacity building at regular interval from individual NGOs and government agencies

Table 3. Distribution of respondents according to their sources and degree of accessibility to information on climate change

Sources of Information	Degree of Accessibility					
	Regularly accessible		Occasionally accessible		Not accessible	
	F	%	F	%	F	%
Radio	101	84.17	19	15.83	_	_
Friends	15	12.50	60	50	45	37.50
Agricultural extension agents	82	68.33	15	12.50	23	19.17
Television	45	37.50	20	16.67	55	45.83
Farmers Association	75	62.50	16	13.33	29	24.17
Newspapers	40	33.33	10	8.33	70	58.33
NGOs	_	_	25	20.83	95	79.17
Internet	25	20.83	15	12.50	80	66.67

3.4 Knowledge of Climate Change

Available data in table 4 revealed that a substantial proportion of crop farmers view climate change as change in the timing of rains or period of rains (98.3%), it is associated with unpredictable seasons and instability in temperature when it is compared with previous years

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(93.3%), climate change results in global warming (90.8%), it results in changes in the timing of sunshine and that long or an extension in the number of hot days during dry season is attributed to climate change (90.0%). Crop farmers' knowledge about climate change can be attributed to the wide enlightenment and knowledge sharing on consequences of climate change and adaptation strategies to mitigate its effects that is relayed via print and electronic media, extension agents, farmers groups and associations, cooperative societies, co-farmers.

Table 4. Distribution of respondents based on their knowledge of climate change

KNOWLEDGE STATEMENTS ABOUT CLIMATE CHANGE	Freq	Percent
Increase in the number of days of dry spell indicates climate change	97	80.8
A change in the timing of rains is an indication of climate change	118	98.3
Delay in rainfall reflects climate change	76	63.3
Changes in the timing of sunshine is attributed to climate change	108	90.0
A shorter rainfall season is attributed to climate change	108	90.0
Longer periods or extension of numbers of hot days during dry season is attributed to climate	105	87.5
Reduction in the number of hot days during rainy season is attributed to climate change	99	82.5
Unpredictable seasons is a manifestation of climate change	112	93.3
Climate change is about shift in rainfall patterns	83	62.2
Climate change results instability temperature when it is compared with previous years	112	93.3
Climate change results in global warming	109	90.8
Climate change brings is about abnormal rainy season and dry season	82	68.3
Change in the elements of weather is attributed to climate change	75	62.5

3.5 Level of Knowledge on Climate Change among Respondents

Respondents level of knowledge on climate change was determined by using the mean criterion, respondents below the mean were categorized as those having low knowledge while those above the mean were categorized as those having high knowledge of climate change.

Result in table 5 revealed that 70% of respondents have high knowledge of climate change while 30% of them have low knowledge of climate change. Appreciating that a significant proportion of the crop farmers have high knowledge about climate change, with this statistics it shows that there is still need to intensify efforts towards increasing the knowledge base of the respondents, some potent means that can be deployed include translating information about climate change in local language as nuggets in news letter, handbook, bulletins etc., group sensitization, running of programmes in the media (radio) on the topic, information dissemination through various groups and associations across crop lines etc. on a frequent basis, as these will increase farmers capacity on the subject.

Table 5. Distribution of respondents based on their level of knowledge on climate change

Knowledge	Frequency	Percentage	Minimum	Maximum	Mean
Low	36	30.0	6.00	13.00	10.08
High	84	70.0			

3.6 Adaptation Techniques Adopted to Mitigate Effects of Climate Change

Table 6 indicated the adaptation techniques or coping mechanisms employed to ameliorate the impact of climate change. 88.3%, 87.5%, 83.3% of the respondents claimed to adopt planting crops favorable for the present weather condition, use of hybrid seedlings and planting different crops as adaptation techniques to reduce the impact of climate change more frequently. Also, 64.2% and 63.3% of the respondents also claimed to adopt changing planting date and use of chemical as other techniques or coping mechanisms adopted to mitigate impact of climate changes more frequently..

3.7 Relationship between Respondents' Socioeconomic Characteristics and Adaptation Strategies

Result of correlation analysis in table 7 shows that respondent's income (r= -0.096; p=0.299); age (r= -0.172; p=0.374) were not related to adaptation techniques adopted to mitigate impact of climate change on food crops farming. However, significant relationship existed between household size (r= -0.089; p=0.002) and adaptation techniques to reduce the impact of climate change on food crops, this suggests that farmers household size influence the adoption of adaptation strategies to mitigate effects of climate change on food crop production because the numbers of household size determine the farm labours in the application and the use of strategies which consequently may influence crop production outputs. Chi-square analysis also reveals that educational level of farmers (X^2= 4.861; p= 0.003) was related to adaptation strategies because education influences farmers access to climate information particularly adaptation strategies that are mostly concern to farmers for better outputs.

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Table 6. Distribution of respondents according to their adaptation strategies adopted to militate impact of climate change

Strategies Adopted F (%)	More frequently F (%)	Moderately frequently F (%)	Frequently F (%)	Not frequently	weighted score	Rank
Increased water conservation	66 (55.0)	35 (29.2)	6 (%.0)	13 (1.08)	228	6th
Planting of different crops	100 (83.3)	13(10.8)	4 (3.3)	3 (2.5)	266	3rd
Changing planting date	77 (64.2)	37 (30.8)	5 (4.2)	1 (0.8)	258	4th
Irrigation	20 (16.7)	38 (31.7)	28 (23.3)	34 (28.3)	137	12th
Use of chemical	54 (45.0)	26 (21.7)	32 (26.7)	8 (6.7)	205	7th
Use of fertilizer	76 (63.3)	21 (17.5)	18 (15.0)	5 (4.2)	240	5th
Use of hybrid seedlings	105(87.5)	7 (5.8)	5 (4.2)	3 (2.5)	278	2nd
Mulching	64 (53.3)	11(9.2)	7 (5.8)	38 (31.7)	184	8th
Relocation to another site	50 (41.7)	9 (7.5)	10 (8.3)	51 (42.5)	148	11th
Agro-forestry product	16 (13.3)	12 (10.0)	13 (10.8)	79 (65.8)	71	13th
Mixed farming	55 (45.8)	11 (9.2)	24 (20.0)	30 (25.0)	176	9th
Planting a crop favorable for the present whether condition	106 (88.3)	7 (5.8)	3 (2.5)	4 (3.3)	279	1st

Table 7. Relationship between socioeconomic characteristics of the respondents and adaptation strategies to reduce impact of climate change

Variable	X^2	df	r-value	CC	p-value
Sex	2.729	1		0.149	0.099
Marital status	2.238	3		0.135	0.524
Level of education	4.861	4		0.197	0.003
Religion Age	0.059	2	 -0.172	0.022	0.971 0.374
Income			-0.096		0.299
Household size			-0.089		0.002

3.8 Relationship between Respondent's Knowledge of Climate Change and the Adaptation Strategies

The result of the analysis in Table 8 shows that there is significant relationship between the respondents knowledge of climate change and the adaptation techniques ($r= -0.157; p= 0.002$), it implies that knowledge of farmers on climate change determines the coping strategies farmers adopts to cushion the climate change impacts on food crops, farmers that are knowledgeable embarked on practices that suitable and appropriate to cushion the climate effects on food crops production.

Table 8. Respondents knowledge of climate change and the adaptation techniques

Variable	r-value	p-value
Knowledge	-0.157	0.002

4. Conclusion and Recommendations

The findings established that majority of food crop farmers were male, active in their age, attained below secondary education and earned an average of #235,725.00k per cropping season which was low. It further established that they cultivated an average of 6.98 acres of land, have an average of 14.04 year of farming experience with majority cultivated maize. Majority have knowledge on climate change effect and radio was regularly accessed as source of information on climate change. Majority claimed to adopt planting crops favorable for the present weather condition, use of hybrid seedlings and planting different crops as an adaptation techniques and coping

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mechanisms to reduce the climate change impact on food crops more frequently. Significant relationship existed between household size, educational level, knowledge and adaptation techniques and coping mechanisms to alleviate climate change impact on food crops. It was recommended that Private extension particularly NGOs should intensify more efforts and incorporate alleviation of climate change impact on crops as part of their programme component and food crop farmers should be better educated and sensitized in order for them to be well furnished with coping mechanisms and adaptation techniques that are currently been offered by research. There is a need to deviate from total reliance on rain-fed food crops production and utilization of irrigation system should be given proper attention, therefore, there is need for adequate provision of irrigation and drainage infrastructures to absorb climate change impact on food crops production.

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ARTICLE

Biofumigation Efficacy of Spider Plant (*Cleome gynandra* L.) Accessions on Nematode Control in Tuberose (*Polianthes tuberosa* L.)

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ABSTRACT

Tuberose (*Polianthes tuberosa L.*) is a perennial summer flower grown by smallholders in Kenya for export. However, its production and export volumes have declined drastically due to nematodes infestation. This study evaluated the effect of *Cleome gynandra* accessions on nematode management on tuberose. Experiment was carried out at Egerton University, Kenya using a randomized complete block design with four replications. The treatments were *C. gynandra* namely "Simlaw", "Egerton", "Taastrup", "PS" and "IP8", applied at 6 kg/m^2 and compared with *Brassica napus*, solarization and untreated control. Data was collected on growth and yield parameters, nematode infestation and quality of tuberose. Data collected was subjected to analysis of variance at $p \leq 0.05$ and means separated using Tukey's test. Biofumigation with *Cleome gynandra* accessions helped to reduce nematode population by 34%, gall numbers by 83% and galling index by 96% when compared with the control. Use of biofumigation helped to improve plant height and leaf number of tuberoses by 16% and 87%, respectively, when compared with the control. Use of biofumigation helped to improve spike length by 32%, marketable spikes by 80%, and flower yield by 90% and reduced nonmarketable spikes by 95% when compared with the control. Based on the above results, use of *Cleome gynandra* accessions and other biofumigants such as rape seed can be used to manage nematodes and improve growth, yield and quality of tuberose.

1. Introduction

Floriculture is one of the fastest growing sub-sectors of agriculture in Kenya. The sub-sector is one of the Kenya's main foreign exchange earners [13]. However, the quantity of cut-flower production has been on the decline since 2012 [17]. This has been mainly attributed to pest attacks and high costs involved due to compliance to standards such as Global Good Agricultural Practices (Gobal GAP), hence being a challenge to smallholder farmers. Use of pesticides to control pests impact heavily on flower industry since they affect global market access due to interceptions during exportations caused by non-compliance to phyto-sanitary requirements such as pesticide residues [13].

Tuberose (*Polianthes tuberosa L.*) is a cut-flower which originated from Mexico, belonging to the family Amaryllis and Genus Polianthes [31]. It is grown in the

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tropical and sub-tropical areas as a summer flower [14]. In Kenya, summer flowers comprise of 2.9% of the total production of cut-flowers and are mainly done by smallholder's growers with tuberose contributing to 0.09% of the 2.9% [13].

In Kenya, Kiambu County has the highest hectares under tuberose (MOALD, 2002)[24] which is grown primarily for the export market. The main export destination is the Netherlands with negligible amounts to other countries [13]. Tuberose is graded according to stem length, namely: Grade I (≥70 cm), Grade II (69-60 cm), Grade III (59-50 cm) and Grade IV (49-40 cm) [3]. The flowers that do not meet the export standard are sold to the local market at lower prices. Tuberose export volumes and values have been declining since 1997 (HCD, 2017) [13]. This has been mainly due to pest attack including nematodes [9].

Nematode (*Meloidogyne spp*) belong to the order Tylenchida and family Heteroderidae. They are parasitic on a wide range of flowers causing up to 5% yield losses [8,26]. The most common nematodes in tuberose is root-knot nematode (RNK) [9]. The RKN form disease complex with plant pathogenic bacteria and fungi, thus their management is important [9]. Although nematicides are available for the control of nematodes in tuberose, most of the synthetic nematicides are too expensive for small-scale farmers who account for the majority of tuberose growers to afford, besides they are unfriendly to the environment [26]. For instance, most nematicides belong to toxicity class 1 or 2 which are either prohibited or restricted for use in horticultural crops in Kenya. However, concerns about the environment and the general public health has led to re-evaluation of these products [2,12,32]. Environmentally sound alternative methods including organic amendments such as biofumigation have been suggested for use in the management of nematodes [1,11,26,33,34].

Biofumigation is the suppression of soil-borne pests and pathogens by biocidal compounds released when plant materials especially brassicaceous residues are crushed and incorporated into the soil [1,21]. The use of biofumigants is a safer alternative method of soil-borne pest management which can be applied on plants containing significant quantities of thioglucoside compounds known as glucosinolates (GSLs). Brassicaceae plant suppress nematodes through volatile toxic compound, such as isothiocyanates generated from glucosinolates which are also known to possess broad activity against weeds, bacteria and fungi [22]. Synthetic commercial nematicides, such as dazomet and metham sodium also contain isothiocyanates especially methyl isothiocyanates (MITIC) which is the active ingredient to manage nematodes [10,23].

Spider plant (*Cleome gynandra L*. syn. *Gynandropsis gynandra L.*) belongs to the family Cleomoideae, formerly Capparidaceae, which is a phylogenetic relative of Brassicas family [7]. According to [28], homogenized leaves of spider plant emit significant quantities of biologically active isothiocyanates including methyl isothiocyanate and their levels of methyl isothiocyanate vary among *C. gynandra* accessions. However, its use in nematode management has not been extensively studied. This study therefore evaluated the efficacy of biofumigation with different *Cleome gynandra* accessions on nematode infestation, growth, yield and quality of tuberose.

2. Materials and Methods

2.1 Experimental Site

The study was done at the experimental field at Egerton University, Kenya. The field is within the latitude of 0.23° South and longitudes 35.35° East in the Lower Highland III Agro Ecological (LH3) at 2,238 meters above sea level. Average maximum and minimum temperature ranges between 19 °C to 22 °C and 5 °C to 8 °C respectively, with a mean annual rainfall of 1,000 mm. The soil are predominately sandy-vitric mollic andosols [15].

2.2 Planting Materials

Tuberose bulbs was bought from PJ Dave Flora Limited and thoroughly cleaned and treated with Bavistin (0.2%) for 30 minutes and dipped in thiourea solution to break dormancy. *Brassicas napus* (rape seed) and *Cleome gynandra* accession "Simlaw" seeds was obtained from Kenya Seed Company, Nakuru. Seeds of other *C. gynandra* accessions were obtained from previous collections: "Egerton", from Egerton University; "Taastrup" from Denmark; "PS" and "IP8" from AVRDC, Arusha, Tanzania.

Brassica napus and *C. gynandra* seeds was planted and all agronomic and maintenance practices was done following the technical recommendation for the respective crops. The leafy twigs at flowering were harvested. The two are known to contain glucosinolates which has nematicidal properties [18]. At flowering stage, the *C. gynandra* plants were uprooted, chopped into small pieces of equal or less than three centimeters (≤ 3 cm) and applied immediately to the experimental plots.

2.3 Nematode Collection, Augmentation and Inoculation

Nematodes were extracted from infested tomato plants and augmented in two weeks old tomato seedlings established outdoor [30]. Specifically, galls were extracted from

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the roots of infested tomato seedlings, chopped and mixed with native soil. To augment the nematode inoculum, the mixture was added to the soil with 2 weeks old tomato seedlings. The nematode inoculum was allowed to infest, develop and multiply on the tomato plants for 8 weeks. After augmentation, nematode egg masses were extracted from the galled tomato roots to prepare inoculum. Galled root tissues were chopped to a length of 0.5 cm and macerated to release the egg masses. This was placed in 15 cm diameter sieves of 1 mm pore size, lined with cross-layered tissues paper and placed for hatching in a glass petri-dishes containing distilled water and incubated at 27 °C. After hatching, it was transferred into a 2 L conical flask. Quantification of the juveniles was done under light microscope with gridded petri dishes. Ten 1 ml replicate samples were drawn from the well mixed suspension to establish the average number of juveniles per ml. Nematodes suspension sample was adjusted to contain approximately 596 juveniles in 1000 ml of distilled water.

2.4 Experimental Design and Treatment Application

The experiment was laid in a randomized complete block design with 4 replications. There were 9 treatments consisting of chopped *C. gynandra* accessions: "Simlaw", "Egerton", "Taastrup", "PS" and "IP8"; *Brassica napus;* fumigant, solarization and; untreated control. Plots of 1.2 m × 1.2 m was dug to a depth of 20 cm, lined with polyethylene sheet and filled with the top soil. The second instar juvenile stage of *Meloidogyne spp*. inoculum suspension was added to each experimental unit ensuring uniform distribution. The chopped plant materials (*C. gynandra*) were incorporated into the soil up to 0.3 m depth and plots covered with 0.14 mm thick clear polyethylene sheet. At the same time, plots treated with Basamid® were also re-dug and fumigated at the rate of 0.029 kg m^{-2} and covered. The edges of the polyethylene sheet were buried 0.15 m into the soil to ensure air tight conditions for four weeks. The untreated plots (negative control) were re-dug and left without incorporating *Cleome gynandra* or Basamid® application. After four weeks; the treated plots were uncovered and left to aerate for 14 days.

2.5 Tuberose Establishment

A plot measuring 12.4 m × 6.3 m was marked, cleared, ploughed, harrowed and demarcated into 36 plots each measuring 1.2 m × 1.2 m. Spacing of the tuberose was carried out at 20 cm by 20 cm. There was a spacing of 0.5 m between blocks and 0.2 m between plots. Each plot had 5 rows each having 5 plants each. Diammonium phosphate (DAP; 18% N, 46% P_2O_5) fertilizer was applied at the rate of 240 kg ha^{-1} (approximately 10 g per hole) (HCD, 2017)[13] and a thorough mixed with soil prior to planting the bulbs. Soil was sampled before application of the nematode suspension and incorporation of amendments to check moisture content and plant parasitic nematode population.

2.6 Data Collection

Data was collected and recorded on the following parameters:

(a) Nematode population

To determine the nematode population in the soil, soil was extracted from 100 cm^3 soil from each experimental unit, using the method described by [17]. At 80 days after planting, the soil was sampled by taking 100 $cm^3$of sample. The sample was placed in a plastic mesh with pore diameter of 1 mm lined with double layered tissue paper. The sieves were half immersed in plastic beakers containing 250 ml of distilled water to allow nematode migration into the water underneath for 24 hours. Nematode counts were determined in 3 replicate sample of 1 ml for each.

(b) Gall numbers

For gall assessment, 6 plants were gently uprooted, at the end of the experiment and their roots washed under tap water to remove excess adhering soil. The plant material containing nematode was chopped to small pieces (≤ 1 cm length), placed in a folded muslin cloth to enclose the material and gently submerged into the water in the funnel. Nematode emerged from the tissues and sunk to the bottom of the funnel stem. After 24 to 48 hours, the clamp was fully opened and 5 to 10 ml of water containing nematodes was rapidly withdrawn and transferred in to a shallow viewing dish for examination.

(c) Galling index

Galling was determined by counting the number of galls of 1 mm diameter and above. The index score was in a scale 1 to 10. Where 0:no gall, 1:10-50 galls, 2:51-100 galls, 3:101-150 galls, 4:151-200 galls, 5:201-250 galls, 6:251-300 galls, 7:301-350 galls, 8:351-400 galls, 9:401-450 galls, 10:451 and above [17]. The scores were converted into numerical entries and their mean determined.

(d) Number of leaves

Leaf count data was collected from the 6 plants in each plot. Leaf count data collection commenced 21 days after planting and continued at an interval of 14 days up to 100 days after planting. At each instance of the data collection the mean number per leaves per plant from each replicate was computed. The mean number of leaves per plant was determined by computing the means of the replicate mean.

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(e) Plant height

Plant height data was collected from 6 plants in each plot. This started at 21 days after planting and continued for 14 days interval up to 100 days. The plant height was measured using a tape measure. At each instance of data collection, the mean height per plant from each replicate was computed.

(f) Yield and quality variables

Weekly harvesting at bud-burst stage from the 6 tagged plants per treatment was done. At each harvest, stems were sorted into marketable and non-marketable categories. Tuberose is graded according to stem length, namely: Grade I (≥ 70 cm), Grade II (69 - 60 cm), Grade III (59 - 50 cm) and Grade IV (49 - 40 cm) [3]. The height was measured and stem length of 50 cm and above was considered marketable stems. Spike length and flower yield was also determined.

2.7 Data Analysis

Data was analysed using SAS statistical package (version 9.1). The proc univariate procedure was used to check for normality of the data before analysis, and data transformation was done. All numerical data was subjected to analysis of variance (ANOVA) at $P \leq 0.05$ and significant means were compared using Tukey's test at $P \leq 0.05$.

3. Results

3.1 Efficacy of Biofumigation with Different *Cleome gynandra* Accessions on Nematode Infestation during Production of Tuberose

Cleome gynandra accessions had significant effect on nematode population. Untreated (control) soil had the highest nematode population followed by solarization when compared with the rest of the treatments which were not significantly different from each other (Figure 1).

Similar to nematode population, gall number was significantly affected by the treatments. Gall numbers was the highest in the untreated tuberose roots, followed by the solarization, when compared with the rest of the treatments that were not significantly different (Figure 2).

Galling index was significantly affected by the treatments. Galling index was the highest in the untreated tuberose roots followed by solarization when compared with the rest of the treatments that were not significantly different (Figure 3). Untreated control was significantly different from solarization for the galling index, nematode population and gall numbers.

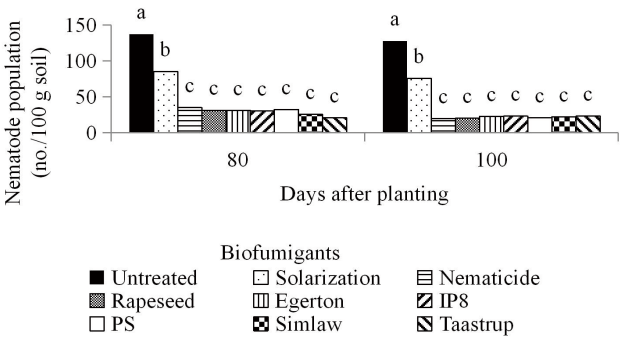

Figure 1. Effect of biofumigation with different *Cleome gynandra* accessions on nematode population during tuberose production. Means followed by the same letter within an evalutation period is not significantly different according to Tukey's test at 5% level of significance

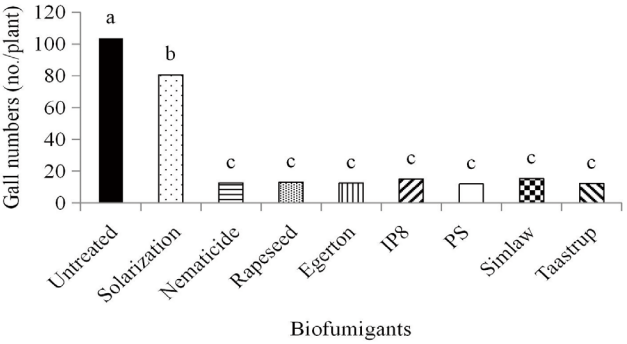

Figure 2. Effect of biofumigation with different *Cleome gynandra* accessions on gall numbers on tuberose roots during production. Means followed by the same letter is not significantly different according to Tukey's test at 5% level of significance

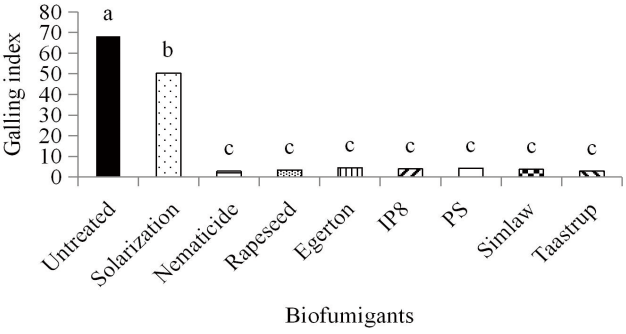

Figure 3. Effect of biofumigation with different *Cleome gynandra* accessions on galling index during tuberose production. Means followed by the same letter is not significantly different according to Tukey's test at 5% level of significance

3.2 Effect of Biofumigation with Different *Cleome gynandra* Accessions on Growth of Tuberose

Use of *Cleome gynandra* accessions had significant influence on the height of tuberose plants. Taastrup and

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nematicide had the tallest tuberose plants throughout the study when compared with the untreated, solarisation and rapeseed. In most of the evaluation days all the *Cleome gynandra* accessions had similar effect on plant height compared with use of nematicide and solarization (Table 1).

Biofumigation with different *Cleome gynandra* accessions had effect on leaf number of tuberose plants. Taastrup and nematicide had the highest number of leaves when compared with the untreated which had the lowest number of leaves, followed by rapeseed and then solarization(Table 2). *Cleome gynandra* accessions were not different with the highest treatments. However, in the remaining evaluation period, untreated tuberose had the lowest number of leaves compared with the rest of the treatments which were not significantly different from each other.

Table 1. Effect of biofumigation with different *Cleome gynandra* accessions on plant height (cm) of tuberose during production

Treatments	49 days	63 days	77 days	91 days
Untreated	30.1^{d}	35.9^{d}	40.2^{d}	53.5^{c}
Solarization	34.8^{c}	38.8^{c}	42.1^{c}	62.3^{b}
Nematicide	41.2^{a}	50.5^{a}	63.4^{a}	84.5^{a}
Rapeseed	39.2^{b}	42.1^{b}	50.5^{b}	71.7^{ab}
Egerton	38.7^{ab}	48.2^{ab}	60.9^{ab}	84.5^{a}
IP8	37.0^{ab}	46.9^{ab}	60.0^{ab}	82.3^{a}
PS	36.6^{ab}	45.2^{ab}	58.6^{ab}	81.1^{a}
Simlaw	36.4^{ab}	46.4^{ab}	59.6^{ab}	79.7^{a}
Taastrup	41.3^{a}	51.2^{a}	65.5^{a}	86.0^{a}

Means followed by the same letter within a column is not significantly different according to Tukey's test at 5% level of significance

Table 2. Effect of biofumigation with different *Cleome gynandra* accessions on leaf number (no./plant) of tuberose during production

Treatments	49 days	63 days	77 days	91 days
Untreated	12.7^{d}	14.8^{c}	18.0^{c}	19.8^{c}
Solarization	20.2^{c}	23.5^{b}	25.0^{b}	28.1^{b}
Nematicide	30.1^{a}	30.8^{a}	34.0^{a}	36.8^{a}
Rapeseed	22.9^{b}	24.2^{b}	26.1^{b}	30.1^{b}
Egerton	27.6^{ab}	30.9^{a}	34.5^{a}	39.7^{a}
IP8	27.4^{ab}	31.1^{a}	33.3^{a}	36.5^{a}
PS	26.6^{ab}	31.6^{a}	34.2^{a}	37.5^{a}
Simlaw	24.1^{ab}	27.1^{a}	29.7^{a}	32.2^{a}
Taastrup	32.1^{a}	34.4^{a}	38.3^{a}	41.2^{a}

Means followed by the same letter within a column is not significantly different according to Tukey's test at 5% level of significance

3.3 Effect of Biofumigation with Different *Cleome gynandra* Accessions on Yield and Quality of Tuberose

Spike length was significantly affected by the treatments. All the biofumigants used produced the longest spike (best quality) when compared with the untreated (control) but was not different with the solarization (Figure 4).

Similar to the trend on spike length, *Cleome gynandra* accessions had significant effect on marketable spikes (>50 cm). At the treatments had the highest number of marketable spikes compared with the untreated and solarization (Figure 5).

Cleome gynandra accessions had significant effect on nonmarketable spikes (<50 cm). Untreated tuberose had the highest number of nonmarketable spikes followed by solarisation when compared with the rest of the treatments (Figure 6).

Cleome gynandra accessions had significant effect on yield of tuberose flowers. *Cleome gynandra* accessions namely. All the treatments had the highest tuberose yield when compared with the untreated which had the lowest followed by solarization (Figure 7).

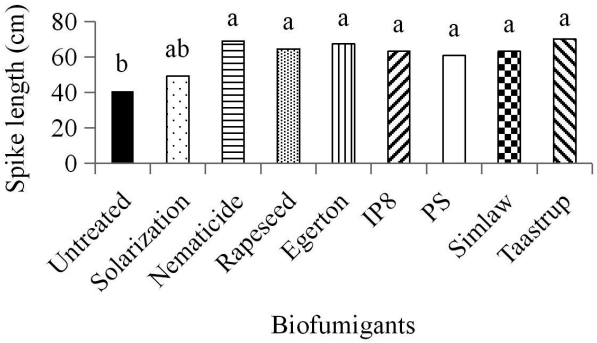

Figure 4. Effect of biofumigation with different Cleome gynandra accessions on spike length of tuberose production. Means followed by the same letter is not significantly different according to Tukey's test at 5% level of significance

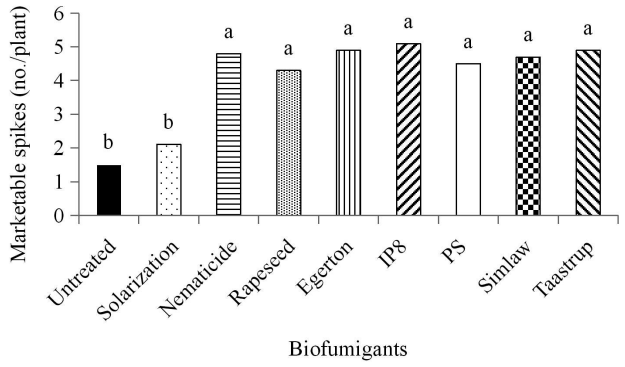

Figure 5. Effect of biofumigation with different Cleome gynandra accessions on marketable spikes of tuberose during production. Means followed by the same letter is not significantly different according to Tukey's test at 5% level of significance

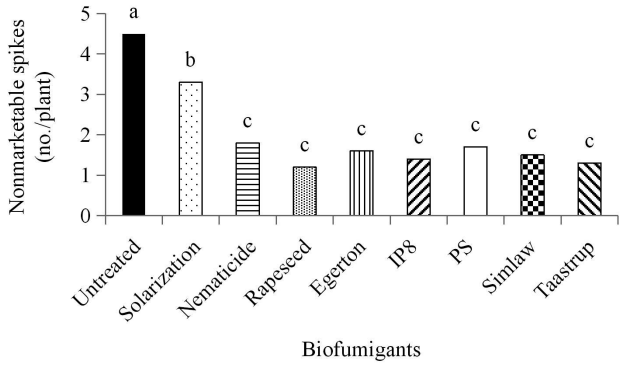

Figure 6. Effect of biofumigation with different Cleome gynandra accessions on nonmarketable spikes of tuberose during production. Means followed by the same letter is not significantly different according to Tukey's test at 5% level of significance

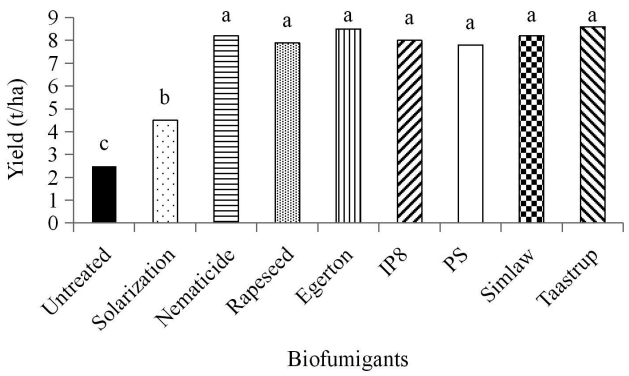

Figure 7. Effect of biofumigation with different Cleome gynandra accessions on flower yield of tuberose during production. Means followed by the same letter is not significantly different according to Tukey's test at 5% level of significance

4. Discussion

Biofumigation helped to reduce nematode population by 34%, gall numbers by 83% and galling index by 96% when compared with the control, with the best effect observed when *Cleome gynandra* accessions, mainly Taastrup was used. This was comparable with the use of nematicide. The results of the study is corroborated by the findings of Argento *et al.* [4] Who observed that application of different concentrations of *Brassica macrocarpa* helped to reduce nematode population and galling index in root-knot nematodes (*Meloidogyne* spp.) on greenhouse tomato plants. Use of biofumigants has been observed to result in enhanced soil properties and nematicidal activities [4]. Biofumigants incorporated into the soil decompose thereby releasing various mineral elements and also improve soil organic matter content. This helps in enhancing soil biological, physical and chemical properties. This therefore helps in better crop development as observed in the study. Similarly, biofumigants release volatile compounds with nematicidal properties such as isothiocyanates. The promotion of plant yield parameters may be as a result of increased availability of nutrients in the root zone and inducing root growth that led to nutrient uptake. Biofumigation was also observed to reduce pest population, disease incidences, and increased crop yield by 30% compared with untreated controls [30]. The enhanced yield may also due to the suppression of nematodes in the soil by the volatile compounds released during decomposition of the studied biofumigants

In another study, Lwande *et al.* [20] were unable to test the efficacy of methyl isothiocyanate, one of the biochemical compounds found in *C. gynandra* on ticks because of the practical difficulties caused by its toxicity, but suggested that essential oil of *C. gynandra* could be contributing to the tick repellency effect. Lwande *et al.* [20] reported that terpenes and their derivatives as the main constituents of the essential oil of *C. gynandra* extracted by hydro distillation, and two of them, β-cyclocitral and β-ionone. *Cleome gynandra* has been observed also to have traces of methyl isothiocyanate that possess miticidal properties as a result of the volatile emissions from entire plants and their detached leaves [27]. It has also been found that hydrolysates of *Cleome spinosa,* a related species to *C. gynandra* contain methyl isothiocyanate and have similar active ingredient of pesticides such as metam sodium, metam potassium and dazomet which are effective biofumigants against fungi and nematodes [23,29].

Barros *et al.* [6] also observed that sulfur-containing compounds, mostly isothiocyanates, were found in mustard, a close relative of rape seed used in the present

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study. Barros *et al.* [6] further demonstrated that irrigation applied directly after biofumigation may trap the volatile organic compounds in soil water thus keeping nematode toxicity longer. *Brassica juncea* eco-types namely Nemfix, Fumus, and ISCI99 were observed to contain high contents of 2-propenyl glucosinolate and resulted in more than 95% mortality of encysted eggs of *G. pallida* in soils covered with polyethylene [19]. Aydınlı & Mennan [5] observed that galling index and nematode eggs were on tomatoes biofumigatated with *E. sativa* and *R. sativus*. Therefore, it is possible that the effect of the studied biofumigants on nematode population and galling index could be associated with the repellence and toxicity effects of the biochemicals that they produce.

Use of biofumigation helped to improve growth of tuberose, such as plant height and leaf number of tuberose by 16% and 87%, respectively, when compared with the control. The results were comparable with the use of nematicide. Similar results were observed by Argento *et al*. [4] when *Brassica macrocarpa* was used as a biofumigant against root-knot nematodes (*Meloidogyne* spp.) on greenhouse tomato plants. According to Argento *et al*. [4] most crop residues incorporated into the soil tend to release nutrients to crops, that can also improve the physical, chemical and biological properties of the soil. This results in better physiological development of tuberose plants. In addition, biofumigant also release volatile compounds against various insect pests, including nematodes [1]. Argento *et al.* [4] observed that biofumigation with *Brassica macrocarpa* in the open field was better than in the greenhouse since it allows for natural decomposition process and better chemical changes to reactive volatile molecules which are released in a more controlled manner, resulting in a longer residual nematicidal activity. Improved growth of tuberose plants may be attributed to improved soil physico-chemical and nematicidal properties of the biofumigants.

Use of biofumigation helped to improve spike length by 32%, marketable spikes by 80%, and flower yield by 90% and reduced nonmarketable spikes by 95% when compared with the control. The results were similar to the use of nematicide. Argento *et al.* [4] observed increase in marketable yield and dry matter on greenhouse tomatoes when *Brassica macrocarpa* was used as a biofummigant against root-knot nematodes (*Meloidogyne* spp.). Hassan *et al.* [12] also observed that biofumigation with radish crushed leaves or seed meal induced high inhibition activity against and *Meloidogyne* spp resulting in higher eggplant yield. Use of biofumigants have been observed to result in enhanced soil properties and nematicidal activities [4]. Biofumigants incorporated into the soil decompose releasing various mineral elements and also improve soil organic matter content. This helps in enhancing soil biological, physical and chemical properties which result in better leaf development as observed in the study. Similarly, biofumigants release volatile compounds with nematicidal properties. The enhanced yield may also be due to the suppression of nematodes in the soil by the volatile compounds released during decomposition of the studied biofumigants.

5. Conclusions and Recommendations

Biofumigation helped to reduce nematode population by 34% and galling index by 96% when compared with the control, with the best effect observed when *Cleome gynandra* accessions. Use of biofumigation helped to improve growth of tuberose, such as plant height and leaf number of tuberose by 16% and 87%, respectively, when compared with the control. Use of biofumigation helped to improve spike length by 32%, marketable spikes by 80%, and flower yield by 90% and reduced nonmarketable spikes by 95% when compared with the control. This was comparable with the use of nematicide. Based on the above results, use of *Cleome gynandra* accessions and other biofumigants such as rape seed can be used to manage nematodes and improve growth, yield and quality of tuberose. However, further studies on the optimization of the rates of the studied biofumigants against nematode management and improving growth and yield of tuberose is necessary.

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ARTICLE

Postharvest Quality and Safety of Potted Greenhouse Tomato Grown on Forest Soil-Biosolids substrate, Blended with NPK Fertilizer

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ABSTRACT

Studies on the effects of biosolids (BS) amended substrate on food quality and safety in tomato production have not been adequately addressed. The objective of this study was to investigate the influence of composted BS and NPK fertilizer on post-harvest quality and safety of potted greenhouse tomato *Solanum lycopersicum* L. Potted tomatoes "Maxim F1" were grown in a randomized complete block design with four replications. Inorganic fertilizer NPK (17:17:17 fertilizer was applied at 0, 100 kg ha^{-1} (5g per pot) and 200 kg ha^{-1}; (10g per pot), BS was applied at 0%, 10%, 20%, 30%, and 40% v/v, in all possible combinations. Tomato were harvested and analyzed for ascorbic acid, chlorophylls, carotenoids and total phenolic compounds; weight loss, fruit firmness, titratable acidity and total soluble solids, as well as heavy metals and microbial contaminants. Results revealed that tomato fruit at 10% BS in combination with NPK fertilizer at 100 kg ha^{-1} had the highest β-carotene (6.1 mg 100 g^{-1}), lycopene (26.1 mg 100 g^{-1}), ascorbic acids (128.0 mg 100 g^{-1}), total phenolic acids (13.2 mg 100 g^{-1}), total soluble solids (17%). However, the same rates produced tomato fruit with lower titratable acidity (2.2%) and had heavy metal residues within the permissible level, according to International EPA standards on biosolids utilization for food crops production. Similarly, no trace of pathogenic bacteria; *Salmonella, Escherichia coli, Staphylococcus* was observed on the harvested tomato. This study reveals at BS 10% with NPK fertilizer at 100 kg ha^{-1} substrate as a better option of plant nutrient source for quality and safe greenhouse tomato production.

1. Introduction

Biosolids are organic materials that result from the treatment of domestic sewage in a wastewater treatment [28]. World production of biosolids is ever increasing due to global population rise and social progress, particularly in developing countries [31]. Although technologies to manage, transform, or reuse biosolids are continuously being developed, their safe agricultural use is also considered a sustainable option. They are, not only rich in nutrients but also contain significant level of contaminants such as pathogens and pollutants [10]. However, for use in crop production, they are stabilized to reduce or eliminate pathogens and manage volatile organic solid. Thus, application of biosolids as a fertilizer in crop cultivation is a common practice in many countries [28]. When applied to land, biosolids can improve crop

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yields through fertilization, increase soil water storage, improve soil quality, avert greenhouse gas emissions, and accelerate carbon sequestration by improving the capacity of the soil to store carbon [8].

The use of biosolids has been reported to enhance secondary metabolites in tomatoes *Solanum lycopersicum* L especially carotenoids, of which lycopene is the most abundant in the ripe fruit, accounting for approximately 80-90% of the total pigments [22]. Lycopene is an antioxidant which protects cells from oxidative damage and helps to decrease the risk of chronic diseases such as coronary heart diseases and cancer. Environmental conditions and growing medium fertility can affect fruit lycopene content [19]. Similarly, α-, β-, γ-, δ-carotene, zeaxanthin, lutein, neurosporene, phytoene, and phytofluene, which has clinically been proven as natural anti-carcinogenic and health related compounds in tomatoes, is directly dependent on nutritional application on tomato[9]. Previous studies have shown that tomato plants accumulate phenolic compounds as defense mechanism under certain stress conditions, such as low N availability. Ribas-Agustí *et al.*[41] also suggested that fruit quality in terms of weight, diameter, brix and phenolic content can be achieved by partially replacing mineral N fertilizer with composted biosolids. An increase in tomato phenolic content was observed after organic amendment, biosolids, was added to the substrate[7]. In a different study, Tzortzakis *et al.*[48], working on greenhouse pepper *Capsicum annum* production reported that use of biosolids substrate, increased total phenols and fruit lightness. However, it reduced fruit acidity but did not affect fruit dry matter content, firmness, green colour, total soluble sugars and EC of peppers. In another study by Perez-Espinosa *et al.*[40], titrable acidity, and soluble solids in fresh fruit seemed to increase with sewage sludge application. Mostly these organic fertilizers have been known to supply both macro and micronutrients. Among the micronutrients, boron and zinc play an important role in improving the yield and quality of tomato in terms of secondary metabolites and chlorophyll content [40].

In another hand, product safety in the use of biosolids is of great concern and especially on production of fresh produce. The rates of biosolids application in agriculture have been guided by the amount of trace element in the samples used [35]. Trace elements are of particular concern in regard to their effects on human and animal health. United States EPA (2002) has analyzed the risks of heavy metals and trace elements to humans, animals, plants, and soil organisms from exposure to pollutants in biosolids via different pathways for applied biosolids. The application of biosolids ceases, if it is estimated that the cumulative loading limit is exceeded [13].

Microbial contamination in biosolids is mainly of faecal origin [35]. However, as most of the contaminating micro-organisms are heat-sensitive, they are eliminated during composting, leading to a faecal pathogen-free end product. For persistent microbes, D'Addabbo *et al.* [11] proposed sanitization, through employing solarization of the media. Strauch [42] quantified the pathogens of concern and their evolution during composting. Pathogens found in biosolids can be viruses, bacteria, protozoa or helminths. These micro-organisms are potential contaminants because of their pathogenicity and are indicators of faecal contamination. Decrease in faecal contamination indicators and elimination of faecal pathogens was reported by De´portes *et al.* [12], when biosolid were left before use over period of one year and all were eliminated. The objective of this work was to determine the effect of biosolids blended with forest soil and NPK fertilizer on post-harvest quality and safety of greenhouse tomato.

2. Materials and Methods

2.1 Experimental Site

This study was conducted for two growing seasons (season 1: January to May 2018 and season 2: June to November 2018) at the Horticulture Research and Teaching Field, Egerton University, Njoro, Kenya. The site is located on latitude 0 23' S and longitude 35 35' E in the lower highland III (LH3) agro ecological zone at an altitude of 2238 m above sea level [25]. A tunnel-shaped greenhouse measuring 8 m by 60 m by with a height of 3 m and covered with UV stabilized polyethlene sheet gauge 150 µm from Amiran Kenya Ltd Nairobi, Kenya. The air temperatures inside the greenhouse during the experiment were 24.5 ± 0.9 °C and 13.3 ± 4 °C during the day and night, respectively. The average day and night relative humidity inside the greenhouse were 55.6 ± 9.6% and 80.8 ± 3.6%, respectively.

2.2 Biosolids and Forest Soil Sample Collection, Substrate Preparation and Analysis

Biosolids (BS) were collected from a lagoon pond at Egerton University Wastewater Treatment Plant and forest soil (typically tropical forest soil), within the same locality. Before blending with NPK (17:17:17) rates, the substrates for tomato production were prepared by mixing the biosolids and forest soil (BS: FS at rates of 0, 10, 20, 30, and 40% (v/v). Samples from each rate, were comprehensively analysed in Laboratory at Kenya Plant Health Inspectorate Services (KEPHIS), Kitale, Kenya. This was to determine the physico-chemical characteristics and nu-

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trient levels of the substrates (Table 1).

Table 1. Physico-chemical characteristics of the substrate used for tomato transplant production

Characterization/ substrates	FS	BS 10%	BS 20%	BS 30%	BS 40%
Bulk density (g cm^{-3})	1.7	1.6	1.5	1.3	1.3
Porosity (%)	35.9	39.6	43.4	50.9	50.9
Moisture content (%)	25.8	34	40.8	42.8	44.5
EC (mS m^{-1})	2.6	3.2	3.6	4.4	5.1
pH	7.4	6.2	6.6	6.5	6.4
Organic matter (g kg^{-1})	157.7	197.8	196.7	210	209.8
C:N	21.3	19.7	15.4	9.6	12.7
Total Carbon (mg g^{-1})	91.7	115.0	114.4	122.1	122.0
Total N (g kg^{-1})	4.3	5.9	7.4	12.9	9.6
Total P (mg k g^{-1})	69.1	83	90.3	101	95.9
K (mg kg^{-1})	132.5	412.3	419.9	427.8	422.4
Ca (mg kg^{-1})	21.9	24	22.8	29.5	27
Mg (mg kg^{-1})	131.1	126.1	117.7	119.1	113.8
Na (mg kg^{-1})	62.9	254.8	342.1	252.8	348.3
Mn (mg kg^{-1})	69.6	530.4	524.8	539.4	553.9
Fe (mg kg^{-1})	27	2490	2473.9	2479.1	2471.5
Zn (mg kg^{-1})	4.7	47.4	44	44	45.9
Cu (mg kg^{-1})	4.4	12.2	12.7	10.3	12.7
Cd (mg kg^{-1})	0.0023	0.0128	0.0115	0.0117	0.0122
Pb (mg kg^{-1})	109.6	2.8	2.1	5.1	3.1

Key: FS (Forest soil), BS (Boisolids).

2.3 Experimental Design and Treatments

The experiment was as arranged in a split-plot in a randomized complete block design, replicated four times. Inorganic fertilizer NPK (17:17:17) at three levels: 0, 100 kg ha^{-1} (5g per pot) and 200 kg ha^{-1}; (10g per pot) was the main plot factor while Biosolids mixing rates in forest soil (FS) at five levels: 0, 10%, 20%, 30% and 40% (v/v) constituted the subplot factor. Four blocks each with 15 treatment combinations were separated by a 0.7 m path. Each experimental unit had 10 potted tomato plants in two rows. Using khaki paper bags, tomato samples for post-harvest analysis were harvested at breaker stage from the six pots in the middle of each plot. They were arranged in the laboratory according to the design in the field.

2.4 Postharvest Analysis

After harvesting tomato from the greenhouse, thirty fruits were randomly selected from each treatment, for further postharvest analysis. Fruits were placed on the shelf for ten days to ripen uniformly. Within the period of those ten days, data for various postharvest data were collected at interval of 0, 5 and 10 days after harvest. The average temperatures inside the laboratories shelves at Egerton were 22 ± 4 °C and 21 ± 0.9 °C for day and night respectively. The average relative humidity inside the laboratory was 55 ± 2.3% and 60 ± 3.6%, for day and night, respectively.

Tomato secondary metabolites and other quality aspects were done in the Molecular Biotechnology laboratory at Egerton University, Kenya. For extraction, 4 tomatoes were randomly picked from each substrate treatments, cut into small pieces, blended into a paste using pestle and mortar and duplicated 3 times.

2.4.1 Ascorbic Acid Content

Ascorbic acid was determined by titration with 2, 6-dichlorophenolindophenol dye. Ten grams of fresh tomato fruit sample were extracted in 30 mL of 5% oxalic acid using a pestle and mortar, and then filtered (Whatman No.1 filter paper). Standard indophenol solution was prepared by dissolving 0.05 g of 2, 6-dichlorophenol-indophenol in distilled water then diluted to 100 mL and filtered. Ascorbic acid standard solution was prepared by dissolving 0.05 g of pure ascorbic acid in a small volume of 5% oxalic acid solution and then diluted to 250 mL with the same oxalic acid solution. Ten milliliter of the ascorbic acid standard solution was then titrated with the indophenol solution to a slight pink end point. Ten milliliters of oxalic acid were titrated as a blank. The amount of ascorbic acid corresponding to one milliliter of indophenol solution was then calculated. Ten milliliters of the filtered sample extract were pippeted into a 50 mL flask and made to the mark with the 5% oxalic acid solution. The standard indophenol solution was used for titrating 10 mL of the filtrate. The ascorbic acid content was expressed in mg per 100 g sample. Using the formula;

Ascorbic acid = C×V× (DF/WT)

Where C = ascorbic acid (mg); V= Volume of dye used for titration of diluted samples (mL)

DF = dilution factor, WT= sample weight (g).

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2.4.2 Total Phenolic

The total phenolic content (TPC) was measured according to Genovese *et al*. (2008)[18]. The samples were extracted in proportions of 1:20 (m/v) with methanol, using a homogenizer for 1 minute. The sample was re-extracted in the same ratios. The supernatants were filtered using filter paper No. 1 and the volume made up to 50 mL. An aliquot (1 mL) of supernatant was oxidized with 1 mL of Folin-Ciocalteu's reagent and neutralized by 1 mL of 20% sodium carbonate. The reaction mixture was incubated for 30 minutes at ambient temperature and absorbance was measured at 745 nm using a UV/Vis Spectrophotometer Model: U-T6, (Shanzhai) Co. Ltd, and China. Total phenolic content was obtained using a calibration curve of garlic acid (1 mg/mL) as standard. Total phenolics were expressed in mg equivalents of garlic acid per 100 grams of the sample (mg GAE/100 g).

2.4.3 Determination of Lycopene, β-carotene and Chlorophylls

Lycopene and β-carotene and chlorophyll were extracted as described by Fish *et al*. [15] using acetone/hexane (4:5). The samples were analysed by UV-Vis spectrophotometer, for β-carotene (453 nm), lycopene (505 nm), and chlorophyll a (663 nm) and b (645 nm). Carotenoids and chlorophylls were calculated and expressed as µg/g DM.

For calculating β-Carotene and Lycopene, the formula for their content was = *(Ex x V)/FW,*

Where Ex; absorbance depending on the carotenoids, V; volume of the solution (25 ml) and FW; the fresh weight of the sample.

Total carotenoids = (450 x V x 4)/FW

Where E450 is the absorbance at 450nm, Vis the volume of the solution (25ml), 4 is a constant and FW is the fresh weight of the sample.

Chlorophyll a = {(10.1 x E663) - (10.1 x E645) x V}/FW

Chlorophyll b = {(16.4 x E645 - 1.01 x E663) x V}/FW

Where E663 and E645; the absorbance of chlorophyll a and b respectively, V; the volume of the solution and FW; the fresh weight of the sample done according to Gogo et al.[20].

2.4.4 Weight Loss

Weight loss was determined from harvested tomato fruits on the individual biosolids treatments. A random sample of five fruits was drawn from each treatment lot and weighed from harvest. The same fruits were kept open at room temperature and re-weighed after every two days until the tenth day and results presented as percent weight loss based on the initial weight.

2.4.5 Fruit Firmness

The determination of tomato fruit firmness was done from harvested tomato fruits. Using a random sampling, ten fruits drawn from each treatment lot and data collected on a period of 0, 5 and 10 days. Fruit firmness (kgF cm^{-2}) was taken from each fruit using a hand-held penetrometer (FT327; Shangai Precision and Scientific Instrument Co., China), with bore size no. 12 and the means were computed for each treatment lot according to Otieno *et al.* [39].

2.4.6 Total Soluble Solids

Fruits total soluble solids (TSS) was determined by extracting juice by squeezing and then centrifuged to obtain a homogenized sample. TSS was determined using a hand held refractometer (RHB; Shangai Precision and Scientific Instrument Co., China) as per the procedure described by Tigchelaar [47]. Results from the treatments were reported as % TSS.

2.4.7 Titratable Acidity

Five milliliters of tomato juice was diluted with 50 mL of distilled water and titrated against 0.1 N NaOH solution using phenolphthalene indicator The volumes of NaOH titre required to change the indicator from colourless to pink were recorded and multiplied by a factor of 0.064, the acid factor for the predominant acid in tomato (citric acid), to estimate the TA levels according to the formula of Tigchelaar [47]:

$$Acid\ (\%) = Titer \times acid\ factor \times 100\ 10\ mL\ of\ tomato\ juice$$

2.4.8 Heavy Metal Analysis

Determination of heavy metal contaminants on tomato fruits was done in the soil and tissue analysis Laboratory at Kenya Plant Health Inspectorate Services (KEPHIS), Kitale, Kenya. This was to assess the presence of Pb Cd, Cu, Zn and Ni. Ripe and ready to eat tomato were blended and wet digestion method was employed. Five ml of blended tomato juice sample was taken and transferred to 100mL conical flask. An aliquot of 4 mL HNO_3, and 0.5 mL H_2O_2 was added and special containers packed and placed into a microwave for digestion. The resultant solution was transferred to 50 mL volumetric flask diluted by Internal Standard Solution (ISTD) Ge, Rh, T1 at 50 ppb for nitric acid digestion. The Inductively Coupled Plasma Mass Spectrometry (ICP-MS) conditions were adjusted, calibrated by blank solution. Then for better operating conditions, the ICP-MS was adjusted to nebulizer gas flow

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0.91 L/min, radio frequency (RF) 1200 W, lens voltage 1.6V, cool gas 13.0L/min, and auxiliary gas 0.70 L/min and finally metals contents of the fruits were analysed by ICP-MS, method adopted by Musa and Lal [36].

2.4.9 Microbial Load

Microbial contamination study was done in the Food Science Microbiology laboratory at, Egerton University, Kenya. This study was based on safety aspect of the tomato fruits ready for consumption, targeting the existence of *Salmonella* sp, *Escherichia coli, Staphylococcus* spp as contaminants. This was done to determine specific microbes mentioned, in the fruit when biosolids is used as a plant nutrient. Ripe tomatoes from different rates of BS were used for microbial load analysis. Using random selection, four tomatoes samples were picked from each plot, washed thoroughly in running water, and then rinsed in double distilled water. The tomatoes fruits were cut in to small pieces and 25 g of the pieces were blended using a juice blender into a tomato paste. Samples were tested for the presence/absence of biological contaminants such as *Salmonella* spp, *Escherichia coli, Staphylococcus* sp. Using manufacturers procedure, three types of selective media were prepared for the inoculation of the specific bacteria of concern: *Salmonella spp in* Salmonella Shigella Agar, *Escherichia coli* in Eosin Methylene Blue Aga and *Staphylococcus* in Baivd Parker Agar. For the counting of bacteria, 1 mL of homogenate was aseptically transferred onto plate count nutrient agar (Oxoid, England) in triplicates. The plates were incubated at 37 °C for 24 hours under aerobic atmosphere. After incubation and isolation, the number of colonies were determined by a colony counter and recorded as colony-forming unit (CFU) g^{-1} and total viable count (TVC) g^{-1} of the growing medium. The evaluation of microbial cellular content in samples were determined by counting of plates. The existence of pathogenic microbial organisms like *Salmonella sp*., *Escherichia coli* and *Staphylococcus sp.* was determined in each sample in triplicates, method used by Otieno *et al*. [38].

2.5 Data Analysis

Data analysis was carried out using SAS statistical package version 9.1 (SAS Institute, Cary Inc., 2001). Numerical data were subjected to analysis of variance (ANOVA) at $p \leq 0.05$. Means for significantly different treatments were separated using Tukey's test at $p \leq 0.05$. Data are presented in table and graphs as means ± standard deviations.

3. Results

3.1 Ascorbic Acid

The ascorbic acid content of tomato fruits was significantly influenced by substrates used for production. Generally, ascorbic acid content started to reduce from the time of harvest. The interactive effect of BS at the rates of BS 10% in combination with 5g per plant of NPK showed a consistency in high ascorbic acid content throughout the 10 days storage period (Figure 1A). However, beyond BS 20%, combining the substrate with NPK fertilizer did not improve ascorbic acid content in the tomato fruit.

3.2 Total Phenolic Acids

Total phenolic acids of the fruits generally decreased during the storage period. Regarding responses to the substrate rates, phenolic acids was significantly higher in NPK 5g per plant with BS 10% compared to the control and other substrates observed before 5 Days after harvest (DAH). At 5 DAH, similar trend was observed with NPK 100 kg ha^{-1} and BS 10% registering higher phenolic acids compared to the control and the higher rates both BS and NPK combined. This was consistent throughout 10-day period (Figure 1B).

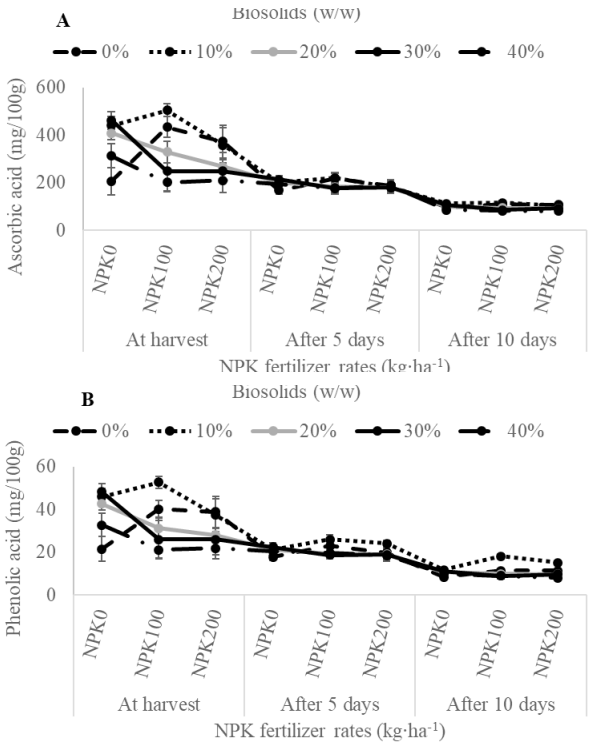

Figure 1. Effect of NPK and biosolids rates on tomato ascorbic acid and phenolic acid contents during room temperature storage

Note:
**Values represent means ± standard deviations.*

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3.3 Beta β-carotene Content

The substrate significantly influenced the fruit tomato β-carotene. Generally, tomato β-carotene content increase with days in the shelf. The results showed the substrate significantly influenced the fruit tomato β-carotene from 5 DAH. At both 5 DAH and 10 DAH, interactive effect of BS 10% and NPK 5g per plant registered higher fruit β-carotene content than control and other higher rates but not significantly different from BS 30% in for forest soil (Figure 2A).

3.4 Tomato Lycopene Content

Tomato lycopene content generally increased during the 10-days storage period. Fruit lycopene content generally increased with shelf-life. For the fruits produced in BS without NPK fertilizer, 10%, 20% and 30% were significantly higher in lycopene content than control and BS at 40%. However, the interactive effect between BS 10% with fertilizer 5g per plant, registered higher lycopene content consistently throughout the 10 DAH. Any rate of BS above 10% did not increase lycopene content of the fruits in combination with fertilizer (Figure 2B).

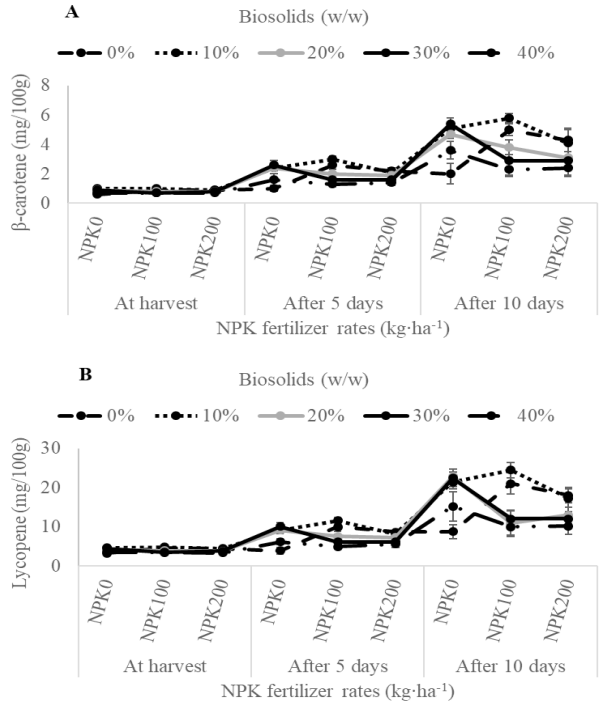

Figure 2. Effect of NPK and biosolids rates on tomato β-carotene and lycopene contents during room temperature storage

Notes:
**Values represent means ± standard deviations.*

3.5 Tomato Chlorophyll a and b

Generally, the total chlorophyll of tomato decreased with storage days as were significantly influenced by BS rates and when combined with NPK fertilizer. From BS at 10% to 30%, chlorophyll a content of the fruits were higher than control and BS at 40%. While on substrate combined with 5g per plant of NPK, only BS at 10% showed significantly higher chlorophyll a (Figure 3A). A similar response was observed in chlorophyll b content of the tomato fruits during the ten days period (Figure 3B). Similarly, any rate of BS above 10% with NPK fertilizer did not show a significant increase in both chlorophyll a and b.

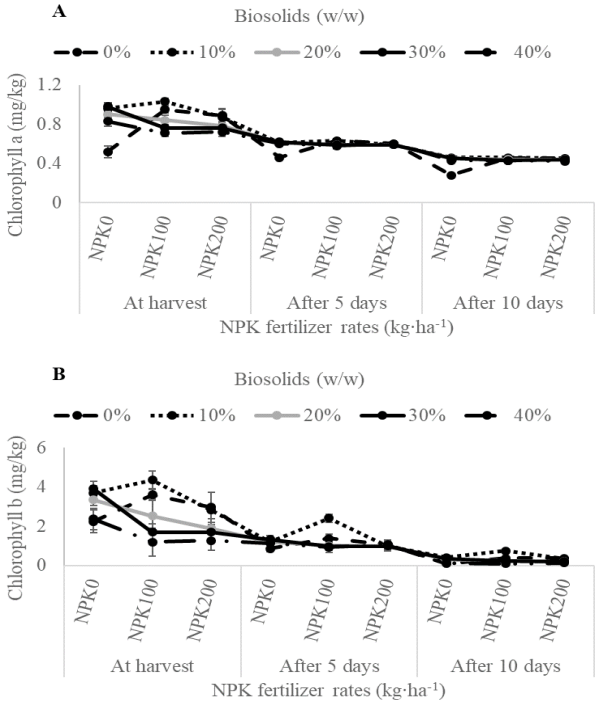

Figure 3. Effect of NPK and biosolids rates on tomato chlorophyll a and b contents during room temperature storage

Note:
**Values represent means ± standard deviations.*

3.6 Fruit Weight Loss

Tomato weight loss was significantly influenced by various rates of biosolids and fertilizer. Forest soil (control) showed significantly ($p<0.05$) higher percentage in rate of weight loss within the 10 days of storage. The same trend was observed in BS at 20% and 30% with higher percentage in weight loss compared to the rest of the substrates (Figure 4A). Interactive effect of the BS and NPK fertilizer was evident as from day after harvest where lowest rate of weight loss was observed on BS 10% combined with 5g per plant.

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3.7 Fruit Firmness

Generally, fruit firmness decreases with increase in days after harvest. The interactive effect of BS at 10% and NPK at 5g per plant was observed as the best rate of the substrate on tomato fruit firmness and this was consistent within the 10-days period in the shelf (Figure 4B).

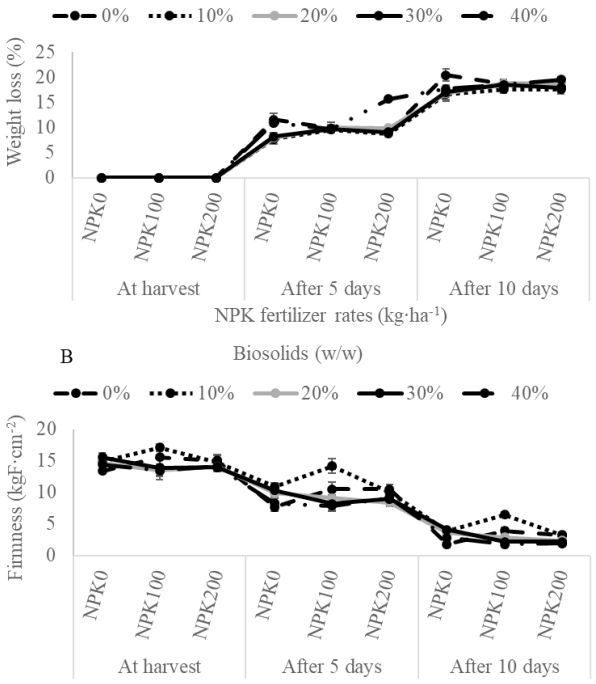

Figure 4. Effect of NPK and biosolids rates on tomato weight loss and fruit firmness during storage at room temperature

Note:
**Values represent means ± standard deviations.*

3.8 Total Soluble Solids

The TSS generally increased within the 10-days in the shelf (Figure 5A). The combination of BS at 10% and NPK at 5g per plant was significantly higher than the control. While BS rate alone were best at 30%, although not significantly different from the combination of BS and fertilizes aforementioned above.

3.9 Titratable Acidity

The results showed a general decrease in fruit Titratable acidity (TA) from different substrate rates. Titratable acidity was significantly higher on the fruits produced from substrate biosolids 20 to 40% and those blended with NPK 10g per pot compared to the rest of the substrates (Figure 5B). This trend was consistent throughout the 10-days of tomato postharvest.

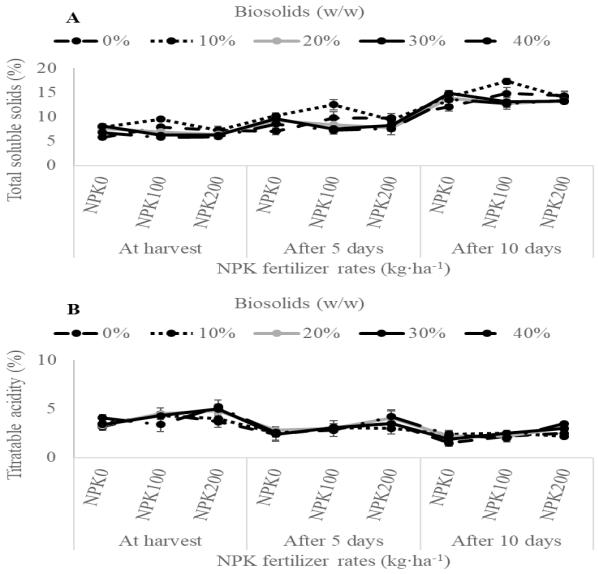

Figure 5. Effect of NPK and biosolids rates on tomato total soluble solids and titratable acidity during room temperature storage

Note:
**Values represent means ± standard deviations.*

3.10 Heavy Metal

The rates of biosolids substrate differently influenced the presence of heavy metals in the fruits. Rates of BS at 20% to 40% showed higher significance Zn, Ni, Cu, Cd, and Pb compared to the control, and BS at 10%. Nevertheless, all were still below the standard stipulated by the EPA (Table 2). The interactive effect of BS and NPK fertilizer reduce the amount of BS required for most of the post-harvest quality to boisolids rate of 10% and NPK 5g per pot.

3.11 Microbial Load on the Tomato Fruits

In the tomato fruit extracts the targeted bacteria; were *Salmonella, Escherichia coli, Staphylococcus* as food poisoning main contaminants associated with wastewater. As was observed in the biosolids substrates rates, the absence of the aforementioned bacteria, similar observation was made on the fruits grown on the biosolids substrates used in the greenhouse production; there was absence of those specified bacterial contaminants in the tomato (Table 3).

4. Discussion

Consumers have become increasingly concerned about the safety and quality of food products they are consume. The present study evaluated the effect of blended biosolids (BS) on postharvest quality of tomato. Various pre-harvest factors including plant nutrition are known to influence the phytochemical properties, which may improve post-harvest quality of horticultural crops including tomato.

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Table 2. Effect of NPK and biosolids rates on tomato heavy metal content during production

		µg·kg^{-1}				
NPK	**Biosolid**	**Zn**	**Ni**	**Cu**	**Cd**	**Pb**
NPK_0	0%	3.5±0.3^{c}*	3.1±0.1^{c}	3.5±0.3^{b}	2.3±0.1^{b}	5.0±0.3^{b}
	10%	3.9±0.2^{bc}	3.4±0.1^{bc}	3.7±0.4^{b}	2.4±0.3^{b}	5.2±0.4^{b}
	20%	4.7±0.2^{ab}	4.3±0.1^{ab}	4.8±0.5^{a}	3.2±0.3^{a}	6.0±0.3^{a}
	30%	4.8±0.2^{ab}	4.5±0.2^{ab}	5.1±0.3a	3.4±0.2^{a}	6.4±0.3^{a}
	40%	5.1±0.2^{a}	4.9±0.2^{a}	5.3±0.5^{a}	3.5±0.4^{a}	6.6±0.2^{a}
NPK_5	0%	3.5±0.2^{c}	3.4±0.1^{bc}	3.8±0.7^{ab}	2.4±0.3^{b}	5.3±0.5^{b}
	10%	3.7±0.4^{bc}	3.6±0.4^{bc}	3.8±0.5^{ab}	2.5±0.3^{b}	5.3±0.6^{b}
	20%	4.1±0.3^{b}	3.6±0.4^{bc}	4.3±0.1^{ab}	2.5±0.4^{b}	5.6±0.1^{ab}
	30%	4.2±0.2^{b}	4.0±0.2^{ab}	4.2±0.2^{ab}	2.8±0.1^{ab}	5.4±0.3^{ab}
	40%	4.7±0.2^{ab}	4.5±0.3^{ab}	4.9±0.6^{a}	3.3±0.4^{a}	6.5±0.3^{a}
NPK_{10}	0%	3.5±0.3^{c}	3.5±0.2^{bc}	3.6±0.4^{b}	2.3±0.2^{b}	5.6±0.1^{ab}
	10%	3.7±0.3^{bc}	3.6±0.4^{bc}	4.2±0.3^{ab}	2.8±0.2^{ab}	5.7±0.2^{ab}
	20%	3.8±0.5^{bc}	3.9±0.4^{ab}	3.9±0.4^{ab}	2.6±0.2^{ab}	5.6±0.2^{ab}
	30%	4.3±0.4^{b}	4.1±0.1^{ab}	4.7±0.4^{a}	3.1±0.2^{a}	5.8±0.3^{ab}
	40%	4.4±0.4^{b}	4.2±0.1^{ab}	4.9±0.4^{a}	3.3±0.3^{a}	6.0±0.1^{a}
	EPA^{y}	200.0	60.0	100.0	1.0	150.0
	WHO/FAO^{Z}	100.0	67.0	73.0	0.2	0.3

Notes:
**Means ± standard deviation followed by the same letter within a column are not significantly different according to Tukey's test at $p \leq 0.05$. ^{y}Maximum ceiling values of heavy metals(mg kg^{-1}) for agricultural land application according to New South Wales EPA [13]. ^{Z}Source = Codex Alimentarius Commission FAO/WHO (mg kg^{-1}) WHO [51].*

Table 3. Effect of NPK and biosolids rates on microbial contaminant after harvest

NPK	**Biosolid**	**TVC (PCA)**	**CFU (MAC)**	***E. coli* (EMB)**	***Salmonella* (BPA)**	***Staphylococcus* (BPA)**
NPK_0	0%	TNTC	4.5	-ve	-ve	-ve
	10%	121x 10^{-6}	TNTC	-ve	-ve	-ve
	20%	NG	NG	-ve	-ve	-ve
	30%	TNTC	TNTC	-ve	-ve	-ve
	40%	77x10^{-6}	107.5	-ve	-ve	-ve
NPK_5	0%	67x10^{-6}	20	-ve	-ve	-ve
	10%	TNTC	48.5	-ve	-ve	-ve
	20%	6.5x 10^{-6}	6.5	-ve	-ve	-ve
	30%	5x10-6	5	-ve	-ve	-ve
	40%	16x10^{-6}	113	-ve	-ve	-ve
NPK_{10}	0%	TNTC	TNTC	-ve	-ve	-ve
	10%	TNTC	51	-ve	-ve	-ve
	20%	TNTC	28.5	-ve	-ve	-ve
	30%	88x10^{-6}	44.5	-ve	-ve	-ve
	40%	TNTC	6	-ve	-ve	-ve

Key:
TVC- Total Viable Count, CFU- Colony Forming Units, EMB- Eosin Methylene Blue (E. coli), Mac- MaCconkey Agar, BPA- Bairvd Parker Agar (Staphylococcus sp), BPA- Bairvd Parker Agar (Salmonella sp). TNTC- Too Numerous to Count, No Growth, -ve = Absent.

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Ilupeju *et al.* [23] noted that pre-harvest factors including soil fertility influenced ascorbic acid content in tomato. Results from the present study have demonstrated that application of BS 10% combined with NPK 5g per pot produced fruits with higher ascorbic acid content. This was a contribution of the available nutrient and organic matter in both BS independently and also when NPK added to make a blended BS substrate. Similar to this study, Taghavi *et al.* [43] reported in their findings the impact of fertilization and composts, on water and nutrient supply to the plant and its influence in the nutritional composition, higher ascorbic acid content on tomato fruit. Among the elements found in the substrates as mentioned by Otieno *et al.,* [38], boron and zinc were significantly higher in the FS and BS in combination as a substrate at different rates, which subsequently enhanced quality to tomato fruits. Boron is among the very essential trace elements that plays a big role in the synthesis of one of the bases for RNA formation. Boron has been shown to influence ascorbic acid content of tomato fruit [44].

The current study demonstrates that the use of BS 10% and NPK 5g per pot demonstrated higher ascorbic acid content in tomato fruits. However, at BS 10% when the rate of NPK were doubled in the substrate, ascorbic acid content reduced. This could have been due to the antagonistic effect of these elements in excess in the BS substrate. A different study has shown that K, Mg and Ca can antagonize in nutrient solution when in excess [6]. The antagonistic effect of increased Mg levels on the K uptake was reported to be due to differences in their ionic mobility [26]. Similarly, high K concentrations in the nutrient solution may result in Mg deficiencies in the plant tissue and vice versa. In the current results, the reduction in ascorbic acid with increased rate to up to10g of NPK per plant may have resulted from the above-mentioned antagonistic effects of those elements at higher concentration. In a similar argument, whenever Na dominates the media, K is normally over taken by the latter element and therefore may not be available for the growing crops. Reduction of ascorbic acid in the present work especially at higher rate of BS and NPK could also be related to higher N in the substrate, which is associated with more vegetative growth, which led to shading effect of tomato fruits. Development of ascorbic acid in tomato fruit also depends on exposure to light for its accumulation in fruits. In the current study this phenomenon was probably the reason for low content of ascorbic acid registered on fruits in higher rates of BS and NPK observed.

The content of fruit phenolic acid reflected nutritional characteristics of the different substrates tested. As common with many plant secondary metabolites fruit phenolic acids are affected by different growth environments including soil properties such mineral nutrients, salinity and drought [2]. The result of the current study demonstrates a wide range phenolic acid content in tomato within the 10 days of storage period. The significant presence of phenolic acids in fruits raised in BS rates from 10 to 30% without NPK and BS at 10% with NPK 5g per plant, was not only due to availability of mineral nutrients in the substrates but also favored by a suitable pH (6.2 to 6.5 and EC range (3.2 to 4.4 dS m^{-1}), as earlier reported by Otieno et al. [38].

The present study reported higher content of β-carotene in BS at 10% with combined NPK at 5g per pot. Carotenoids have been reported to possess provitamin A activity Tang [45], a precursor of vitamin A essential for promotion of general growth, maintenance of visual function [19], and N being one of the essential elements for their production. In regards to β-carotene, Bojović and Stojanovic [5] demonstrated that carotenoid content depended on the presence and ratio of macronutrients especially N as one of the most essential elements and its deficiency decreases its accumulation. Whereas in the present study, β-carotene content reduced with further increasing rates of BS, and when rates of NPK was doubled. Salt stress in the substrate has been indicated as one of the factors leading to N deficiency [6]. Similarly, lower content of β carotene in fruits from BS at 10% to 40% in without NPK was due to lower N, P and K in different rates of substrate as was reported by Otieno *et al.*[38]. This result is in conformity with those reported Khavari-Nejad *et al.* [29] on the reduction of β carotene in tomato due to deficiency of N and P deficiency. However, López-Ráez and Bouwmeester [32] reported another scenario that P starvation can induce changes in gene expression of some carotenoids including β carotenes and compounds derived from them in tomato roots.

In the current study, tomato fruit lycopene content generally increased in all the substrates with days on the shelf, indicating that the fruit accumulated more of this important secondary metabolite, with shelf-life. Lycopene represents 60-74% of the tomato fruit carotenoids and of other tomato products and content is affected by many pre-harvest factors such as plant nutrition[39]. Biosolids consist of humic acids after degradation, and is able to release plant nutrients by decomposition. Lycopene content plays a key role in tomato appearance and attractiveness to consumer, a part from its immense health benefits[30]. Color is a key quality indicator of tomato fruit. Lycopene in tomato is responsible for the redness, and β-carotene for orange coloration. Deeper red color in tomato is an indication of predominant existence of lycopene which an important antioxidative compound. Nutrition as a pre-

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harvest factor has been known for carotenoid development leading to high lycopene content in tomatoes. In the current study, BS at 10% to 30%, and interactive effect of BS at 10% and 5g of NPK per plant resulted in increase of lycopene content, which consistently increased within 10 days of tomato post-harvest period. Higher nutrient content in the tested biosolids probably enhanced soil moisture status and nutrient availability such as N, P, K, Mg, B Zn, Cu and Mo[38], leading to biosynthesis of carotenoids which are responsible for tomato fruit color especially lycopene.

Chlorophyll content in a tomato fruit is normally affected by pre-harvest factors, of which the main one is nutrition. At post-harvest the chloroplast content may not increase but degrade into chromoplast which, eventually turn into carotenoids [21]. Thus, in the present study, generally, the total chlorophyll of tomato decreased with storage days as were significantly influenced by biosolids rate as well as NPK fertilizer. The present results demonstrated that at harvest, there was higher chlorophyll in fruits produced at BS rates of at 20% to 40%, than FS. This implied there was higher N in organic in the substrate rates as reported by Otieno *et al*. [38]. While on substrate with 5g of NPK, only BS at 10% showed significantly higher chlorophyll a and b. It was demonstrated that combination of forest soil and biosolids as substrate may not only contribute to organic matter but also Mg, Fe, Cu and Zn in the nutritional pool. Magnesium and iron are essential mineral elements for plant growth and also the development of cell structural component. Iron in particular ranges at the upper limit of the micronutrient category with approximately 2 μmol/g plant dry weight and plays an important role as an activator of many biochemical and enzymatic processes [21]. In regards to the current study, Cu and Zn were found to be in sufficient quantity in BS at 10% combined with NPK at 100 kg ha^{-1}. This probably resulted to the higher plant chlorophyll content as observed in the study. Our results are in agreement with Alves *et al.* [1] who demonstrated that concentration of organic fertilizer in the soil and their association with doses of biofertilizer influenced the gas exchange and SPAD chlorophyll content in tomato plants. However, the higher dose of biofertilizer reduced chlorophyll content of tomato plants, which led to the reduction of gas exchange hence reduced photosynthesis.

Weight loss and fruit firmness were influenced by use of BS and NPK fertilizer. Calcium plays a crucial role in cell division and the maintenance of cell permeability and cell wall integrity, all of which directly influence factors such as firmness and shelf-life [16]. Calcium has also been shown to improve cell membrane integrity [37], thus lead to reduction in fruit water loss and consequently weight loss and quality. The present study has demonstrated the benefits of using BS at 10% and with 100 kg ha^{-1} of NPK fertilizer in improving the post-harvest quality of tomato fruits in terms of weight loss reduction as influenced by the use of BS. Fruit firmness may be considered side by side with fruit weight loss since they are affected by plant cell wall integrity. This may have been due to the presence of Ca as indicated to be within the range of pH 6.2 to 7 [44]. Calcium plays a crucial role in cell division and the maintenance of cell permeability and cell wall integrity, all of which directly influence factors such as firmness and shelf-life [16], also known to improve cell membrane integrity [37]. The results of the current study are in conformity with study by Otieno *et al*. [39], who reported that higher rates soil *Lippia kituensis* Vatke and *Ocimum gratissimum* L. used as substrate organic amendment produced tomato with firm fruits compared to the lower rates of organic amendments used. Firmness of fruits goes together with water loss and nutrients. In the present study, weight loss was reduced by use of biosolids at higher rates.

Biosolids are rich in N and organic matter which holds the mineralized plant nutrients together in a substrate. Addition of NPK availed N in the current study which played a significant role in photosynthesis, hence production of carbohydrates and subsequently sugar formation during ripening. Chlorophyll and carotenoid content formation at the vegetative stage of the crop may contribute to the quality of tomato at post-harvest. This is from the carbohydrates production [4], leading to higher TSS in fruits. This concurs with the present study regarding the inverse relationship between soil nitrogen concentration and fruit TSS [4]. Nitrogen seems to increase not only vegetative growth but also yield in expense of fruit quality. Wang *et al.* [50], confirms decrease of TSS with increasing N concentration in the substrate due to salt stress situation and antagonism among the major plant element especially in the higher BS rates blended with double NPK fertilizer.

Application of BS at 20% to 40% blended with 100 kg ha^{-1} of NPK resulted in increased TA, and this was indicative of salt stress with elevated EC of the substrate. Higher EC was observed in the substrate with biosolids at 20% and higher, blended with NPK at 200 kg ha^{-1}. Mardi et al. [34] made similar observations. Thus, higher TA observed in fruits produced from BS at higher rates of BS blended with NPK is attributed to this scenario. Contrary to the findings of the present study, Tzortzakis and Economakis [49] observed a reduction of TA in tomato production with use of perlite, pumice and their mixtures compared with pure soil treatments. This scenario may be alluded to the fact that soilless substrate normally has narrow range of buffering capacity as opposed to soil-based substrate, and this may lead to essential and trace

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element deficiency for crop quality. In a different study, Fathy *et al.* [15] reported that when humic acid was applied to an apricot crop, which led to an increase in the content of TSS and decreasing fruit acidity, a positive constituent of biosolids. Under high available soil moisture, the root may absorb more water, resulting in a reduction in the TSS by water dilution as observed in BS at 40%.

The heavy metal concentration in fresh commodities has been a subject of great concern to consumers. Several factors such as metal concentrations in soils, soil pH, cation exchange capacity, organic matter content, types and varieties of crops have been reported to affect the uptake of heavy metals [33]. It is generally accepted that the metal concentration in soil is the dominant factor as far as residues are concerned [27]. The concentration of heavy metals in plants is often positively correlated with the abundance of these elements in soils or substrate. In the present study, it was observed that at BS at 10% with 100 kg ha^{-1} of NPK is a suitable rate of production, since it registered heavy metals concentration below the maximum allowable limit, according to standards of New South Wales, EPA[13]. These were Zn (3.7 µg kg^{-1}), Ni (3.6 µg kg^{-1}), Cu (3.8 µg kg^{-1}), Cd (2.5 µg kg^{-1}) and Pb (5.3 µg kg^{-1}). In this regard, other authors have also reported the presence of these micro-nutrient concentration in biosolids[24]. Our results are similar to those of Bagdatlioglu, et al.[3], who demonstrated that the concentrations of Cu, Zn, Fe, Pb, and Cd, which were within safety baseline levels for human consumption (in reference to permissible limits of heavy metals as per FAO/WHO [51], These were as follows: Cd (0.1 mg g^{-1}), Pb (0.2 mg g^{-1}), As (0.1 mg g^{-1}), Hg (0.03 mgg^{-1}), Cu (40 mg kg^{-1}), Zn (0.60 mg kg^{-1}), Fe (5.0 mg kg^{-1}.Biosolids therefore are among the most important organic fertilizer rich in micronutrients (trace elements) such as, Zn, Mn, Cu, Fe, B, Cl, Mo and Ni, which are essential to crop growth, yield and quality. As various studies have shown, not all heavy metals pose risks in crop production systems. Some heavy metals are useful in plant physiological processes especially Zn, Cu and Ni [3]. Many enzymes in plant need Zn for their activity and it may also be required for chlorophyll biosynthesis in other plants [44]. Copper, like Fe is also associated with enzymes involved in redox reactions such as plastocyanin, which is involved in electron transfer during the light reaction of photosynthesis [46]. Nickel is another heavy metal that plays a key role in the production of secondary plant metabolites that influence resistance to diseases [17]. In another study, it has been shown that tomato respond well with Ni at 30 mg kg^{-1} and this level may increase quality of fruit; auxin and gibberellin content [17]. Others are non-essential metals; arsenic (As), cadmium (Cd), lead (Pb), and mercury (Hg), not required for normal biological function [44].

The results on microbial contamination of the tomato fruits produced in this study indicated the absence of the targeted microbes. This was not surprising because previous analyses had shown that targeted microbes *Salmonella sp., Escherichia coli* and *Staphylococcus* were absent in all the dry BS substrates [38]. The absence of microbial contaminants can be explained from the fact that microbe community in biosolids are faecal in nature, most being parasite on animals, therefore survival in the plant system was not possible except by external contamination. Additionally, they are heat-sensitive, and their survival may have been reduced during decomposition, solarization and storage of the substrate for six months before planting crops. Strauch [42] also reported reduction of *Salmonella, Shigella, Streptococci* and *Escherichia coli*) in sludge during decomposition. Concerning decrease in faecal contamination, De´portes *et al.* [12] reported disappearance of faecal pathogens, when biosolids were left before use over period of one year. This probably rendered the BS substrates used in this study free from targeted pathogen in the tomato fruits produced by the use of substrate.

5. Conclusion

Based on data from the present study, use of biosolids blended in forest soil and 5g per plant of NPK, is a viable means of improving greenhouse tomato postharvest quality through improving substrate status; a practice, which may also be used in integrated nutrient management strategies. This work has revealed that for safe production of quality tomato fruit, forest soil blended with BS 10% and 5g per plant of NPK resulted in fruits with higher postharvest quality. While the study lays a good foundation on improving greenhouse tomato performance, further study using other organic sources as amendments and various greenhouse tomato varieties could be studied to ascertain the outcome of our results on product quality and safety.

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ARTICLE

Wheat Varietal Investigation for the Hill Region of Nepal

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ABSTRACT

Multilocation testing of the Coordinated Varietal Trial for Mid-hill and High Hill (CVT-MHH) of wheat genotypes were conducted at different hill research stations of Nepal Agricultural Research Council (NARC) during the normal planting season of 2012-13 and 2013-14. Twenty genotypes including two check varieties were included in Randomized Complete Block (RCB) design with three replications in the experiment. Data on the different yield attributing traits were recorded. Highly significant difference ($p<0.01$) among the genotypes for the days to heading, days to maturity, plant height, thousand grain weight and grain yield was observed in 2012-13. Wheat genotype BL 4061 had the highest grain yield with 3802 kg/ha followed by NL 1153 (3736 kg/ha), NL 1159 (3733 kg/ha), NL 1154 (3674 kg/ha) and NL 1156 (3462 kg/ha). In 2013-14 also a highly significant difference among the genotypes for all the recorded traits was observed and these genotypes were stable for the yield and yield attributing traits. The most promising genotype for the grain yield was NL 1153 (5816 kg/ha) followed by NL 1178 (5760 kg/ha), NL 1156 (5454 kg/ha), NL 1159 (5259 kg/ha) and NL 1179 (5075 kg/ha). From the yield and other yield attributing trait wheat genotypes NL 1055, NL 1153, NL 1159, NL 1156 and NL 1179 need to be tested under farmers' field for further confirmation and release as variety.

1. Introduction

Wheat is the third major cereal crop after rice and maize in Nepal. Wheat is grown from Terai to high mountain region and consumption of wheat in recent years is increasing day by day. In Nepal wheat has covered the area of 703992 ha with the production of 2,005,665 metric tons and productivity of 2849 kg/ha [15]. The Mid-hills and high hills represent 32% of the total production and 43% of the area. The area under wheat production in mid and high hills was 317458 ha and production was 598243 metric tons with the average productivity of 2047 kg/ha (MOAD, 2017) [14]. The low level of productivity in hills and high hills is mainly due to difficulty in the availability of improved varieties on one hand and occurrence of disease on the other. Yield gaps are generally associated with the lack of adoption of recommended technologies [8]. Since there is little scope for increasing land area under wheat, the major challenge will be to break the yield barrier by pragmatic genetic and developmental approaches [3]. It is very crucial to increase the grain yield to meet the current and future demands of food. Development of the

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high yielding varieties for the hills of Nepal is important to meet the required production and food security of that region.

Nepal has a wide variety of climatic condition due to the topographical differences within short north-south span of the country. About 70 to 90% of the rainfall occurs during the summer monsoon months (June to September) in Nepal and the rest of the months are almost dry. Wheat is cultivated during the dry winter period and therefore, supplementary irrigation plays a vital role in its cultivation. Varieties of wheat have been developed to suit the local climatic conditions. Due to the availability of improved seeds, modern cultivation practice and a supplementary irrigation; the wheat cultivation has increased substantially throughout Nepal in the recent years [16].

Improvement and selection of high input, stress responsive and wide adaptive wheat genotypes with preferred traits are necessary to increase production and productivity for ensured food security [4]. Existing varieties are not sufficient to increase sustainable production and productivity due to climate change, disease outbreak, population growth, decreasing land and other resources. Because of the fast rate of urbanization, transport system development, growing industry and changing food habits of the people, quantity and quality improvement in wheat is the urgent need of the country. Therefore, with the objective to develop high yielding varieties with other desirable traits for hilly region, Coordinated Varietal Trials were conducted at different locations representing hill environments of the country.

2. Materials and Methods

Multilocation testing of wheat genotypes was done at five different agricultural research stations. Planting was done in normal wheat season (November) of 2012-2013 and 2013-2014 in Agriculture Botany Division, Khumaltar; Horticulture Research Station, Dailekh; Agriculture Research Station, Pakhribas; Hill Crop Research Program, Dolakha and Agriculture Research Station, Jumla (Table 1). Selected genotypes from the CIMMYT nurseries and yield trials and newly developed genotypes through hybridization at Bhairahawa and Agricultural Botany Division, Khumaltar were included in the CVT-MHH. Twenty genotypes including two check varieties, *Dhaulagiri* and *WK 1204* were used in the experiment. Randomized Complete Block (RCB) design with three replications was used. Arrangement of the treatments was with four genotypes in the rows and five genotypes in the column. The distance between the rows was 25 cm. The area of each experimental unit was six square meter witheight rows of three meter length.

Data on quantitative characteristics like days to heading (DH), days to maturity (DM), plant height (PH), Grains per Spike (GPS), thousand grain weight (TGW), Spikes per meter square (SPMS) and grain yield (GY) were recorded. Data were analyzed utilizing ANOVA technique on Gen-stat program [22].

Table 1. Details of the Experiment conducted locations in 2012-13 and 2013-14

S.N.	Research Stations	Address	Geographical Position	Altitude (meters above sea level)
1	Agriculture Research Station (ARS)	Pakhribas, Dhankuta	27˚05' N 87˚14'E	1889
2	Hill Crop Research Program (HCRP)	Kabre, Dolakha	27˚38' N 86˚08'E	1733
3	Agriculture Botany Division (ABD)	Khumaltar, Lalitpur	27˚39' N 85˚19'E	1321
4	Horticulture Research Station (HRS)	Kimugaun, Dailekh	28˚50.83' N 081˚43.3' E	1255

3. Results and Discussion

A highly significant difference ($p < 0.01$) among the tested genotypes was observed for all the traits in 2012-13 and 2013-14. Location specific variation for the tested genotypes for all traits was non-significant (>0.05) in 2012-13 (Table 2). In 2013-14 also a highly significant difference among the tested genotypes was observed for all the recorded traits and the genotypes were stable for the yield and yield attributing traits (Table 3). Genotypically plant height, grains per spike and 1000-grain weight were positively and significantly correlated with grain yield while highly significantly associated phenotypically [12]

3.1 Days to Heading

The stage from partial to full appearance of spike is also called ear emergence or heading [1]. In this study, there was a significant difference among the tested genotypes for days to heading in 2012-13. The most early genotype in heading days was NL 1055 (108) followed by NL 1082 (109), NL 1153 (109), BL 4241 (110), NL 1118 (111) and NL 1117 (112) in 2012-13 (Table 2). The 12 genotypes were earlier than the mean days to heading and 10 genotypes were earlier than the check varieties Dhaulagiri and WK 1204. Similarly, a highly significant difference among the tested genotypes was observed for days to heading in 2013-14 (Table 3). Check variety Dhaulagiri was the earliest genotype with 122 days to heading which

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was followed by NL 1084 (123), NL 1055 (124), NL 1185 (125), BL 4423 (126), and NL 1120 (129) (Table 3). The result with the highly significant difference among the genotypes for days to heading is in agreement with the Subedi *et al*. [20] in the study of the genotypic performance in the hills of Nepal.

3.2 Days to Maturity

Physiological maturity is usually defined as the time when the flag leaf and spikes turn yellow [9]. Asignificant difference among the tested genotypes for days to maturity was observed in 2012-13 (Table 2). The most early maturing genotype was BL 4291 (156) followed by NL 1055 (157), NL 1082 (159), NL 1118 (160), BL 4278 (160), WK 2164 (161) and NL 1156 (161). Seven genotypes matured earlier than the mean days to maturity in 2012-13. Similarly a highly significant difference was observed in 2013-14 for the days to maturity. In 2013-14, the earliest maturity was observed in NL 1055 with 173 days followed by Dhaulagiri (175), NL 1184 (176), NL 1082 (177), WK 2183 (177), NL 1180 (178), WK 2182 (178) and NL 1179 (179) (Table 3). These observations are similar with the result obtained by Pandey et al. [18].

3.3 Plant Height

The plant height was determined by measuring the distance between the base of the stem and the top of the spike excluding awns. Both grain and plant height are important objectives for any wheat breeding program because grain provides energy, protein and dietary fiber in human nutrition while the height can increase the straw yield which becomes the important forage for livestock. Therefore, breeding for plant height as well as grain yield is the foremost challenges for a wheat breeder. Plant height, number of grains per spike, thousand grain weight, biological yield and grain yield contribute equally to average grain yield of wheat crop (Khan, 2016)[13]. Highly significant difference among the tested genotypes for plant height was observed in both years. There was non-significant GXE interaction (Table 2 and Table 3). The shortest plant height was observed in NL 1160 with 72 cm followed by WK 1204 (73 cm), NL 1117 (76 cm), and NL 1118 (76 cm). Eleven genotypes were shorter than the mean genotypes in 2013-14. Similarly, in 2013-14 the shortest plant height was in NL 1185 with 85 cm followed by WK 2123 (86), WK 1204 (87), NL 1055 (89), NL 1082 (89), WK 2182 (90) and NL 1179 (92). Eleven genotypes had lower height than the mean (Table 3). This finding is similar to the previous findings obtained by Gautam *et al*.[6].

Table 2. Combined analysis of different traits evaluated in 2012-13 in CVT-MHH at Khumaltar and Pakhribas

Genotypes	DH	DM	PH	SPMS	TGW	GY
BL 4061	114	163	85	274	42	3802
NL 1055	108	157	81	278	34	3205
NL 1078	114	162	81	195	39	3235
NL 1082	109	159	83	224	40	3457
NL 1117	112	162	76	332	33	3053
NL 1118	111	160	76	306	33	3275
NL 1120	112	163	84	253	41	3170
WK 1581	127	161	100	244	38	3110
BL 4278	112	160	85	213	35	3188
BL 4291	110	156	83	274	33	2339
NL 1153	109	164	92	224	44	3736
NL 1154	115	161	85	224	41	3674
NL 1156	113	161	84	229	41	3462
NL 1157	115	163	84	253	36	2969
NL 1159	115	163	84	235	40	3733
NL 1160	115	162	72	245	35	3186
NL 1161	112	162	79	264	36	3256
WK 2164	112	161	82	235	32	3366
Dhaulagiri (Check)	113	164	80	223	37	2868
WK 1204 (Check)	115	164	73	262	37	3287
GM	113	161	82	249	37	3269
F-test (E)	*	**	**	**	**	**
F-Test (EXL)	ns	ns	ns	ns	ns	ns
LSD (0.05)	9.44	2.64	5.57	45.82	3.76	444.04
CV (%)	8.8	1.6	6.8	14.4	10	8.6

Notes:
*Where GM= Grand Mean, E= Entry (Genotype), L= Location, LSD= Least Significant Difference, CV = Coefficient of Variation *Indicates significant difference among the tested genotypes (where, p is> 0.01 to <0.05). **indicates the highly significant difference among the tested genotypes (where, p is <0.05) ns = non-significant difference among the tested genotypes (where, p > 0.05).*

3.4 Spikes per meter Square

Highly significant difference among the tested genotypes in both years was observed for the number of spikes per meter square. There was no GXE interaction observed. Spikes per meter square indicate the tillering capacity of the crop and the plant population in the experiment. The highest number with 332 spikes per meter was observed in NL 1117 followed by NL 1118 (306), NL 1055 (278)

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and BL 4291 (274). Ten genotypes had higher spikes per square meter than the mean in 2012-13 (Table 3). In 2013-14 the highest number of spikes per square meter was observed in WK 2134 (343) followed by NL 1179 (321), WK 2123 (303), NL 1185 (299), NL 1055 (294), WK 2183 (292), WK 2180 (291). Ten genotypes had higher SPMS than the grand mean (279). The present result coincides with those of Thapa *et al.*[21].

3.5 Grains per Spike

It has generally been observed that high yield in bread wheat varieties is associated with the increasing number of grains spike (Ashebr *et al.*., 2020)[2]. The observation was not taken for the grains per spike in 2012-13 (Table 2). The tested wheat genotypes were significantly different for grains per spikewith non-significant GE interaction, indicating variation among genotypes with no effect of location. Our result for the grains per spike has been supported by the result of Ilyas and Mohammad [10]. In the 2013-14, the number grains per spike was observed in NL 1082 (53) and NL 1153 (53) which was followed by WK 2183 (52), BL 4460 (50) WK 1204 (49), NL 1178 (48), WK 2180 (47) and NL 1156 (47) (Table 3). Nine genotypes had higher grains per spike than the mean of 46 grains.

3.6 Thousand Grain Weight

The thousand grain weight was the highest in NL 1153 with 40 g followed by BL 4061 (41), NL 1120 (41), NL 1156 (41), NL 1154 (41), NL 1082 (40), NL 1159 (40) and NL 1078 (39). Nine genotypes had the higher thousand grain weight than the mean and both of the check varieties Dhaulagiri and WK 1204 (Table 2). Highly significant difference among the tested genotypes was observed and there was no GXE and the variation among the genotypes for the grain weight was due to the genotypic variation. The highest TGW was in NL 1178 (59) in 2013-14 followed by NL 1153 (58), BL 4423 (58), NL 1184 (58), Dhaulagiri (57), NL 1156 (57) and NL 1183 (57) (Table 3). Eleven genotypes had the higher TGW than the mean of 54 g. This result is similar to the result obtained by Gautam *et al.*. [6]and Pandey et *al.*., (2020).

3.7 Grain Yield

Highly significant difference among the tested genotypes for grain yield was observed. Wheat genotype BL 4061 had the highest grain yield of 3802 kg/ha followed by NL 1153 (3736 kg/ha), NL 1159 (3733 kg/ha), NL 1154 (3674 kg/ha), NL 1156 (3462 kg/ha) and NL 1082 (3457 kg/ha). Nine genotypes had higher grain yield than the grand mean (3269 kg/ha) and seven had the higher yield than the checks. The highest GY in 2013-14 was in NL 1153 (5816) followed by NL 1178 (5760), WK 2123 (5475), NL 1156 (5454), NL 1159 (5259), NL 1179 (5075) and WK 1204 (5035). Nine genotypes had the higher grain yield than the mean yield. In our experiments there were significant differences among the genotypes for grain yield which are in agreement with Sharma [19]; Kamat [11]; Ginkel et al. [7]; Dwivedi *et al.* [5] and Pandey *et al*. [17].

Table 3. Summary of combined analysis in CVT-MHH across 4 locations (Dolakha, Pakhribas, Khumaltar and Dailekh) in 2013-14

Genotypes	DH	DM	PH	GPS	SPMS	TGW	GY (kg/ha)
NL 1153	130	184	108	53	259	58	5816
NL 1178	134	181	96	48	284	59	5760
WK 2123	135	180	86	46	303	48	5475
NL 1156	133	181	94	47	277	57	5454
NL 1159	137	182	96	44	260	55	5259
NL 1179	132	179	92	43	321	54	5075
WK 1204 (Check)	137	182	87	49	287	53	5035
NL 1185	125	181	85	42	299	49	4937
WK 2182	132	178	90	45	259	56	4917
WK 2180	134	180	94	47	291	55	4897
NL 1183	131	180	89	46	262	57	4895
NL 1180	129	178	91	38	267	55	4829
NL 1184	123	176	98	47	241	58	4710
WK 2134	138	181	93	37	343	48	4605
NL 1082	125	177	89	53	258	50	4486
NL 1055	124	173	89	46	294	50	4480
BL 4460	134	181	96	50	247	51	4475
WK 2183	134	177	91	52	292	51	4441
BL 4423	126	180	92	40	254	58	4435
Dhaulagiri (Check)	122	175	95	42	283	57	4283
GM	130.6	179.3	92.53	45.72	279	53.88	4913
P value (Genotype)	**	**	**	**	**	**	**
P value (Genotype X Environment))	*	ns	ns	ns	ns	ns	ns
LSD (0.05)	3.1	2.4	8.08	5.9	41.9	5	685
CV (%)	3	1.7	10.8	14.2	13.6	11.6	14

Note:
*Where GM= Grand Mean, E= Entry (Genotype), L= Location, LSD= Least Significant difference, CV = Coefficient of Variation *Indicates significant difference among the tested genotypes (where, p is> 0.01 to <0.05). **indicates the highly significant difference among the tested genotypes (where, p is <0.05). ns = non-significant difference among the tested genotypes (where, p > 0.05).*

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4. Conclusion

There was a highly significant difference ($p < 0.01$) among the tested genotypes for days to heading, days to maturity, plant height, spikes per meter square, number of grains per spike, thousand grains weight and grain yield in both years. This indicates the presence of sufficient variability among the tested genotypes and provides a good opportunity for wheat improvement program. There was no significant GXE indicates these genotypes are stable for the observed traits. The most promising genotypes with higher grain yield, more number of grains per spike, bolder grain size, early days to heading and maturity were identified from the varietal investigation. From the analysis of yield and other yield attributing trait wheat genotypes NL 1055, NL 1153, NL 1159, NL 1156 and NL 1179 need to be tested under farmers' field for further confirmation and release as variety.

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