



THE INFLUENCE OF VERMICOMPOST ON GROWTH, YIELDS, AND
DISEASE OCCURANCES OF CUCUMBER PLANT (*CUCUMIS SATIVUS* L.) IN
THE GREENHOUSE CONDITION



By
Mr. Sopheak TITH

A Thesis Submitted in Partial Fulfillment of the Requirements
for Master of Science (BIOSCIENCE FOR SUSTAINABLE AGRICULTURE)
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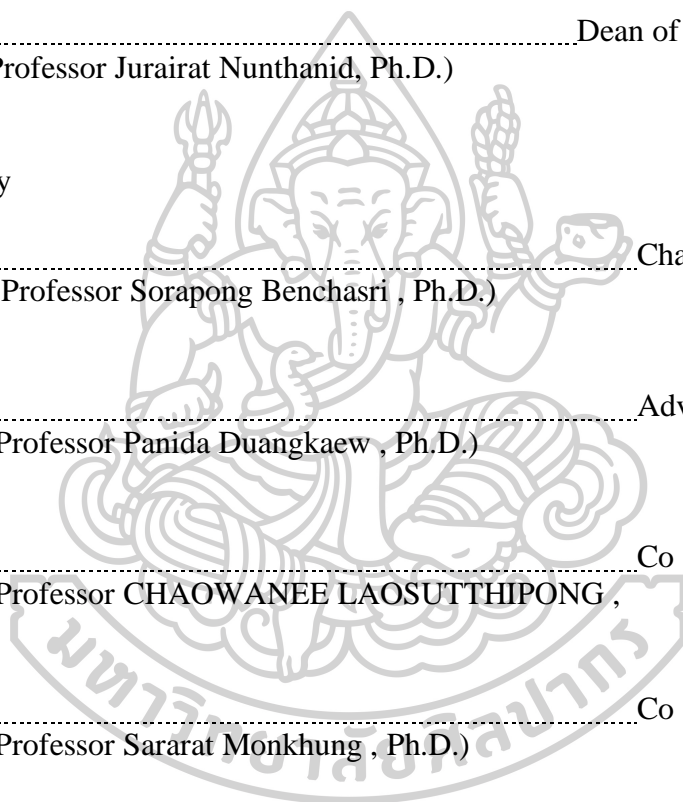
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MR. SOPHEAK TITH : THE INFLUENCE OF VERMICOMPOST ON GROWTH, YIELDS, AND DISEASE OCCURANCES OF CUCUMBER PLANT (*CUCUMIS SATIVUS* L.) IN THE GREENHOUSE CONDITION THESIS ADVISOR : ASSISTANT PROFESSOR PANIDA DUANGKAEW, Ph.D.

Vermicompost is an organic residual derived from the bio-oxidative process of wastes by the action of earthworms. Vermicompost has been known as a good material to boost soil nutrients, improve soil structure, promote plant growth, and suppress plant diseases. This study aimed to investigate the efficacy of vermicompost on the productivity of cucumber (*Cucumis sativus* L.), improvement of soil fertility, and enhancement of soil microbial population. The experiment was laid out in the Complete Randomized Design with 5 treatments including soil control (T1), soil with chemical fertilizer application (T2), and soil mixed with 10%, 20%, and 30% of vermicompost (T3, T4, and T5). Plant growth, yield, disease incidence, soil chemical analysis, and bacterial population were measured. The results revealed that the application of vermicompost had a significant effect on promoting plant height, leaf area, number of leaves, fruit characteristics, fruit yields, and suppressing leaf spot disease incidence ($p \leq 0.05$). From the chemical analysis, soil mixed with vermicompost improved in pH, electrical conductivity, total nitrogen, available nitrogen, total phosphorus, available phosphorus, total potassium, and available potassium. In addition, soil's total phosphorus was increased after planting in the vermicompost treatment. Analysis of the bacterial population demonstrated that vermicompost significantly increased the population of bacteria in the soil ($p \leq 0.05$). Based on the results, the application of 30% vermicompost showed the best positive effect on improving soil fertility together with promoting cucumber growth and yield in poor soil planting conditions. Therefore, the application of vermicompost can be a vital choice in cucumber production for sustainable agriculture as well as improvements in the food products in the world.

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CHAPTER I

INTRODUCTION

1.1 Introduction to the research problem and its significance

The uncontrolled use of chemical fertilizers contributes largely to the deterioration of the environment in the world (Khan and Ishaq, 2011). After the green revolution, the use of chemical fertilizers increased and achieved self-sufficiency in agriculture, but inappropriate use of chemical fertilizers in combination with not using organic fertilizer gave negative results including soil damage, water pollution and air pollution (Zhao et al., 2008). There have been negative effects on the human health and animals due to the residues of these agrochemicals in food products (Sharma and Singhvi, 2017). The harmful effects of chemical fertilizers and pesticides have shifted the interests of researchers towards organic amendments such as vermicompost, which are made from the conversion of organic wastes or residues into organic fertilizer by earthworms (Wani and Lee, 1992).

Vermicompost technology could be considered as an effective way of solid waste management. It is a faster process than traditional composting and landfilling since the material passes through the earthworm's gut (Sharma and Garg, 2018). The decomposition process of organic waste into nutrient-rich vermicast occurred through the combined action of earthworm enzymes and microorganisms' action by which the earthworms also increase in number, size, and weight (Pattnaik and Reddy, 2010; Ramnarain et al., 2019). Vermicompost is considered as an excellent product since it is homogeneous, has desirable esthetics, low chemical contaminants, contains rich nutrients, plant growth hormones, beneficial microbial population, and tends to hold more nutrients over a longer period thus it can increase the production of crops without adversely impacting the environment (Tajbakhsh et al., 2011). Azarmi et al. (2009) reported that vermicompost significantly increased leaf number, plant height, and chlorophyll content. Vermicompost can prevent the plant from insect pests without polluting the environment (Edwards et al., 2007). Also, Gopal et al. (2009)

reported the use of vermicompost as biological control for *Fusarium* wilt of chickpea through isolated actinomycete from herbal vermicompost. The result showed that 33 actinomycete isolates obtained from vermicompost had the antagonistic potential against *Fusarium oxysporum* f. sp. Cicero. Moreover, in the greenhouse experiment, five actinomycetes showed 45–76% reduction in *Fusarium* wilt incidence at 29 days over the FOC-inoculated control, where 100% disease incidence was observed within 20 days.

This vermicomposting technique is widely promoted in many countries such as India, Canada, Japan, Malaysia, Philippines, Spain and Thailand (Aalok et al., 2008). Unfortunately, implementation of this technique in Cambodia and some countries are still in laboratory and pilot scales. Therefore, the suitability of this technique for Cambodia is questioned since there is little information on the feasibility of vermicomposting in Cambodia (Chattopadhyay, 2012). Introducing vermicomposting in farms comes with its own set of challenges. The most important inputs, earthworms and organic wastes, have to be considered and handled with some tutelage and the function must be managed carefully in order to transform organic wastes into environmentally fertilizers that could maintain or build soil organic matter pools.

Cucumber (*Cucumis sativus* L.), one of an important commercial vegetable, is grown worldwide (Alsadon et al., 2016). Cucumber is one of economic crops in Thailand which is widely grown throughout Thailand for fresh and processed consumption as well as for the pharmaceutical industry (Tantasawat et al., 2015). Because cucumbers are cultivated in greenhouses as well as in the field, and also in a variety of climates, they offer many possibilities for the attack of a large number of pathogens and animal pests (Tatlioglu, 1993). Vermicompost was reported to suppress the occurrence of soil-borne disease of cucumber seedling about 96.1% and improved the mean of fresh weights and plant height (Hu et al., 2002). In general, the farm-scale production of cucumber depended on the use of chemical fertilizer in order to obtain high yields (Sallam et al., 2021; Thi Da et al., 2020). Thus, replacement of chemical fertilizer to environmentally safe vermicompost which still provides the same productivity or higher will be the better option for cucumber production.

Experiments had been conducted to evaluate the suitable level of vermicompost application to obtain the best effect. Atiyeh et al. (2001) displayed that the application of vermicompost improved plant growth and increase the tomato yield. Meanwhile, Yardim et al. (2006) indicated that 50 and 60% by volume of cow manure vermicompost mixture with soil resulted in a low yield of cucumber in greenhouse condition. Besides, the application of cow-dung vermicompost at 3 t/ha⁻¹ as the basal fertilizer indicated a significant improvement in the basic soil physicochemical properties, mineral nutrients, and biological properties, and it also increased the yield and fruit quality of cucumbers (Zhao et al., 2017). Therefore, the suitable amount of vermicompost application has to be considered in order to obtain the beneficial effect of vermicompost on plant growth and disease prevention, especially in different soil conditions.

In summary, to explore the potential level of a local produced vermicompost on cucumber plant production, the vermicompost from the farmers in Phetchaburi, Thailand was characterized for its chemical characteristics and was applied at different levels to the cucumber planted in poor soil conditions in the greenhouse. The parameters such as plant growth, yield, fruit characteristics, disease occurrences, and soil nutrients were determined compared to chemical fertilizer treatment.

1.2 Goal and objective of study

1. To determine the chemical nutrient properties of vermicompost produced from cow-dung by the local farmers in Phetchaburi province, Thailand.
2. To compare the effect of vermicompost application against chemical fertilizer on plant growth, yield, fruit characteristics, disease occurrences, and soil nutrients in cucumbers (*Cucumis sativus* L.) grown in the greenhouse.

1.3 Hypothesis of the study

Vermicompost has a positive effect on plant growth, yield, fruit characteristics, disease occurrences, and soil nutrients in cucumber (*Cucumis sativus* L.) plantation in greenhouses.

CHAPTER II

LITERATURE REVIEW

2.1 Vermicompost

2.1.1 Vermicompost and its significance

In recent years, the application of vermicompost has been receiving increased attention due to its remarkable physicochemical and biological features (Huang et al., 2014). Moreover, the application of vermicompost facilities is in operation in several countries such as Canada, the USA, Italy, and Japan (in both residential and industrial areas) (Ghosh, 2004). Additionally, vermicomposting is a simple biotechnological process of composting in which certain species of earthworms are used to enhance the process of waste conversion and produce a better end product for plants (Gandhi, 1997; Ghasem et al., 2014). It is now well known that vermicomposting is the most promising bio-fertilizer which besides increasing plant growth and productivity by nutrient supply, is economical and ecofriendly as illuminated in (Figure 1) (Panda, 2011). Besides, vermicompost produced from the farm wastes improves soil health and growth, enhances quality and crop yield and helps in pollution control (Karmegam and Daniel, 2000). As a result of the degrading activity of earthworms, the mineralization of nutrients is enhanced that increases crop productivity as well as it has been widely applied in traditional agriculture and horticulture and its beneficial effects have been proven on soil biota and soil structure (Bhadauria and Saxena, 2010). Moreover, vermicompost can be used for all types of crops, including agricultural, horticultural, ornamental and vegetables and at any stage (Aynehband et al., 2017). Practicing vermicompost for the disposal of fruits and vegetable wastes will thus reduce the requirement for more land in the near future, thereby creating better environments and reducing ecological risk (Mane and Raskar Smita, 2012). However, raw materials such as organic wastes, agro-residue and worm species are the main components of making vermicompost as shown in Figure 1.

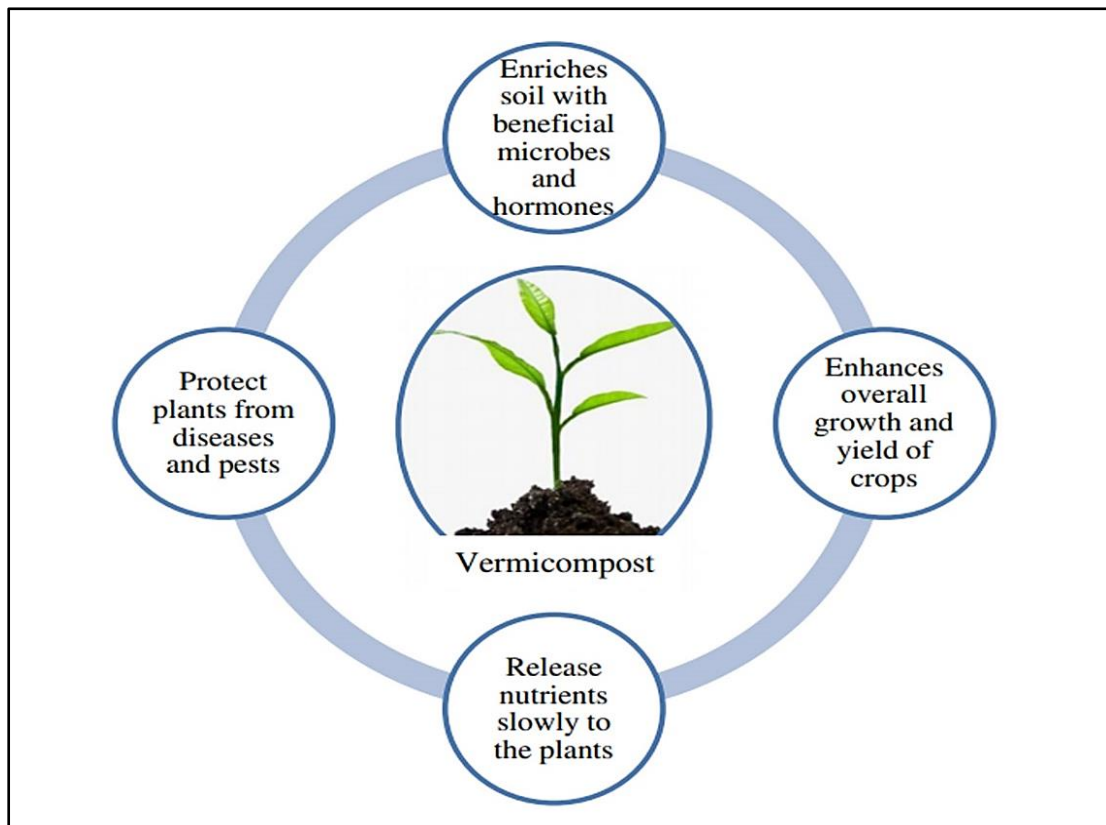


Figure 1 The addition of vermicompost to crops enriches the soil with beneficial plant growth hormones, essential nutrients, and beneficial microbes that suppress diseases and pests and enhance the overall growth and productivity of crops. Reference: (Yatoo et al., 2021).

2.1.2 Earthworm and its function

Earthworm species and organic wastes are the main components for making vermicompost. The decomposition process of organic waste into nutrient-rich vermicompost occurs through the earthworm. Earthworms improve soil fertility and significantly support agricultural productivity (Sinha et al., 2010). Earthworms consume various organic wastes and reduce the volume by 40% to 60%. Each earthworm weighs about 0.5 to 0.6 g, eats waste equivalent to its body weight and produces casting equivalent to about 50% of the waste it consumes in a day. The moisture content of castings ranges between 32% to 66% and the pH is around 7.0 (Adhikary, 2012). Earthworms are natural invertebrates of the agro-ecosystem that

belong to the Lumbricidae family and are found in temperate and tropical soils. They are hermaphrodites, both male and female reproductive organs are present in every single earthworm, but self-fertilization does not generally occur. At the time of laying eggs, the sexually mature worms have a distinctive epidermal ring-shaped area called the clitellum that has gland cells that secrete material to form a viscid, girdle-like structure known as a cocoon (Velando et al., 2008). Cocoon production starts at the age of 6 weeks and continues until the end of 6 months. Under good conditions, one pair of earthworms can produce 100 cocoons in 6 weeks to 6 months. In temperate worms, it ranges between 3-30 weeks and 1-8 weeks in tropical worms (Kaviraj and Sharma, 2003). Earthworms voraciously feed on organic wastes and while utilizing only a small part for their body synthesis they eject a large part of these consumed waste materials in a half-digested form. Since the intestines of earthworms harbor a wide range of microorganisms, enzymes, hormones, etc., these half-digested materials are transformed into a form of vermicompost within a short time (Pajon, 2009). However, earthworms are classified into *epigeic*, *endogeic* and *aneic* species based on their ecological functions (Bhatnagar and Palta, 1996; Brown, 1995). There are nearly 3600 types of earthworms in the world, which can be divided into two types; (i) burrowing, which is a type of *Pertima elongate* and *Pertima asiatica* which live deep in the soil. They are pale, 20 to 30 cm long and live for 15 years. (ii) non-burrowing: *Eisenia fetida* and *Eudrilus eugeniae* live in the upper layer of the soil surface. It has a red or purple color and is 10 to 15 cm long, but its life span is only 28 months. The non-burrowing earthworm eats 10% soil and 90% organic waste material. It converted organic waste faster than burrowing. The burrowing type of earthworm comes onto the soil surface only at night. These make holes in the soil up to a depth of 3.5 meters and produce 5.6 kg casts by ingesting 90% soil and 10% organic waste (Nagavallemma et al., 2004). Epigeic earthworms can be raised at several levels of production, from backyard bins to large-scale composting of agricultural, municipal and industrial biosolids (Appelhof et al., 1996). Earthworms occur in diverse habitats. Organic materials like manures, litter, compost is highly attractive to earthworms, but they are also found in very hydrophilic environments close to both fresh and brackish water, and some species can survive under snow. Most earthworms are omnivorous that can survive on both plant and animal matter.

Also, digestion of carbohydrates, protein, fat, and fiber and metabolize the nutrients and energy of the source absorbed. However, *Agastrodrilus*: a carnivorous genus of earthworms from the Ivory Coast of Africa has been reported to feed upon other earthworms of the family Eudrilidae (Lavelle, 1983). In addition, earthworms have an effect on soil microbials. Earthworms significantly increased the ratio of bacteria to fungi on an area basis (per m²) by more than two times in mid-summer and early autumn (Dempsey et al., 2011). Earthworms can be cultured and put to various uses such as improving and maintaining soil fertility, converting organic waste into manure, producing earthworm-based protein food (earthworm meal) for livestock, as a drug and vitamin source, or being used as a natural detoxicant. Therefore, the selection of the correct earthworm species for vermiculture application is important because different species of earthworms show variation in nutrient composition in vermicompost (Appelhof et al., 1996). Generally, the earthworm species (or composting worms) most often used are red wigglers (*Eisenia fetida* or *Eisenia andrei*), though European nightcrawlers (*Eisenia hortensis*, synonym *Dendrobaena veneta*) and red earthworm (*Lumbricus rubellus*) could also be used (Edwards et al., 2019). Here are some examples of vermicomposting using different earthworms with different sources of food raw materials as illuminated in Table 1.

2.1.3 Raw materials

Vermes is the Latin word that is used for earthworms and vermicomposting. In recent years vermicomposting has received more attention all over the world, as it is widely utilized for the management of different types of organic wastes (Bhat et al., 2018; Chauhan and Singh, 2013). Raw materials are the main food consumed by earthworms. Due to when the earthworm consumes organic wastes, the substrate passes through earthworm's gut and gets digested in the intestine of earthworm with the aid of beneficial microbes. In the intestinal tract, mucus or chemical secretions, enzymes, and antibiotics help in the breakdown of substrate to finely divided peat like material called vermicompost, which is readily available to plants (Adhikary, 2012; Naidoo et al., 2017; Pathma and Sakthivel, 2014) shown in Figure 2. Previous scientific research found that the chemical composition and autochthonous microbial communities in vermicompost are influenced by their

parental wastes (Fernández-Gómez et al., 2011). Organic waste can be treated by composting, vermicomposting, anaerobic digestion, or a combination of these methods. A great variety of organic waste materials have been used with vermicompost including agricultural residues. According to Manna et al. (2003) has been reported that *Tectona grandis* litter may have been the most appropriate food material for earthworms because it contained elevated reserves of mineral nutrients. Also, neem leaves by showing that vermicompost originated from neem leaves had a considerable positive impact on brinjal development and fruiting. Gajalakshmi and Abbasi (2004) reported that water hyacinth (WH) with *Eudrilus eugeniae* has a high population of earthworm, and produced 5-6 times more vermicompost as compared to the vermicompost weed, which has a low earthworm population. Weed and animal waste are valuable resource that provide both macro and micronutrients for plants (Gajalakshmi et al., 2002; Nasiru et al., 2013). Borges et al. (2017) have found an increase in earthworm growth rate and biomass in a mixture of swine manure and cattle dung. The author concluded that animal manure is an excellent waste for the vermicompost process under controlled temperature and moisture conditions. Besides, industrial wastes are also beneficial for making vermicompost by recycling and decomposing. These organic wastes are converted into nutrients and avoid environmental pollution (Bhat et al., 2018; Julka, 2008). More than that, there are some raw materials such as paper-pulp industry waste (Tripathi, 2014), sugar industry waste (Bhat et al., 2014), textile industry waste (Garg and Kaushik, 2005), and food industry waste (Garg et al., 2012) that were used for vermicompost production. Vermicompost can be a low-cost, effective technology, and a better option for the management of different types of organic solid waste. However, some organic wastes may have mixed with other sources to improve the mechanisms of microorganisms such as enzymes, and the availability of nutrients in vermicompost, as shown in Figure 2.

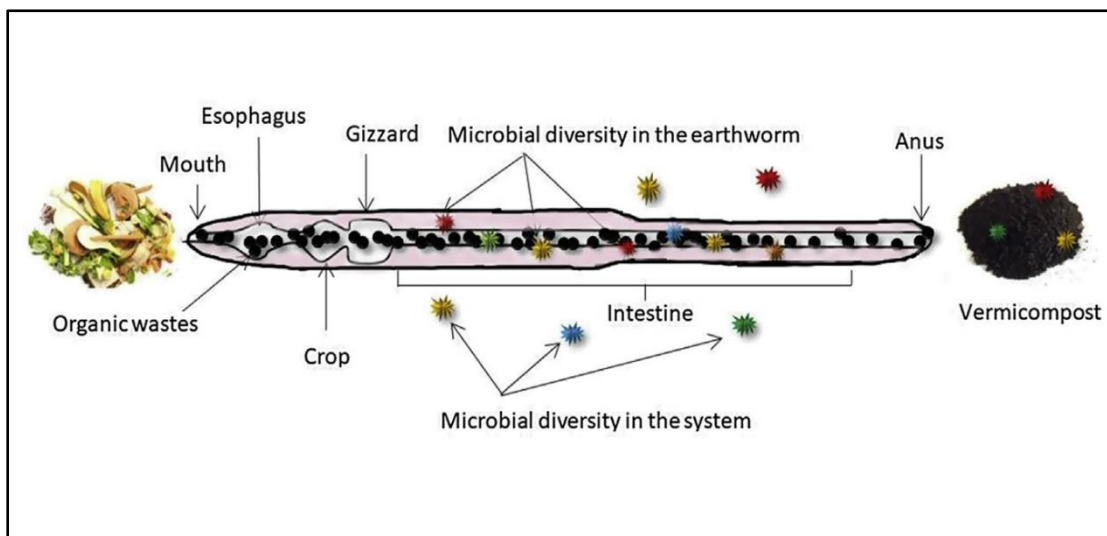


Figure 2 The illustration of the interaction between earthworm and microorganisms in earthworm's gut. Reference; (Sulaiman and Mohamad, 2020).

Table 1. Various types of raw materials and earthworm species used in vermicomposting

No.	Raw materials	Earthworm species
1	Agricultural residues	<i>Eudrilus eugeniae</i>
2	Agriculture waste and sugarcane thrash	<i>Eudrilus eugeniae</i>
		<i>Perionyx excavates</i>
3	Board mill sludge	<i>Lumbricus terrestris</i>
4	Canteen waste and vegetable waste	<i>Eisenia foetida</i>
5	Cattle manure	<i>Eudrilus eugeniae</i>
6	Deciduous forest waste, cow-dung	<i>Eisenia foetida</i>
		<i>Perionyx excavatus</i>
		<i>Dicogaster bolau</i>
7	Different mammalian animal waste	<i>Eisenia foetida</i>
8	Domestic waste + cow-dung	<i>Perionyx excavates</i>
		<i>Perionyx sansibaricus</i>
9	Fly ash + cow dung	<i>Eisenia foetida</i>
10	Gaur gum	<i>Eudrilus eugeniae</i>
11	Grass clippings, cow dung	<i>Eisenia foetida</i> ,

No.	Raw materials	Earthworm species
12	Green waste	<i>Eisenia andrei</i>
13	<i>Imperata cylindrica</i> grass	<i>Perionyx excavates</i> <i>Eisenia foetida</i>
14	Municipal solid waste	<i>Eisenia foetida</i> <i>Eudrilus eugeniae</i>
15	Municipal, agricultural and mixed solid waste	<i>Eudrilus eugeniae</i> <i>Perionyx excavates</i>
16	Onion residue/waste	<i>Eisenia foetida</i> <i>Eudrilus eugeniae</i>
17	Organic matter, moistened peat moss, crushed leaves, dried yard waste	<i>Eisenia foetida</i> <i>Lumbricus rubellis</i>
18	Organic wastes	<i>Lumbricus rubellus</i> <i>Eisenia jetida</i> <i>Eisenia Andrei</i> <i>Dendrobdena rubida</i> <i>Eudrilus eugeniae</i> , <i>Perionyx excavates</i> <i>Eiseniella tetraedra</i> .
19	Paper mill sludge	<i>Lumbricus rubellus</i> <i>Eisenia foetida</i>
20	Pig manure, food wastes, leaf wastes, yard wastes, bark wastes, chicken manure	<i>Eisenia foetida</i>
21	Potato peels	<i>Pheretima elongate</i>
22	Press mud	<i>Pheretima elongate</i> <i>Eudrilus eugeniae</i> , <i>Eisenia foetida</i> <i>Megascolex megascolex</i> , <i>Perionyx ceylanensis</i> <i>Drawida willsi</i>
23	Bagasse, sugar cane trash	<i>Drawida willsi</i>

No.	Raw materials	Earthworm species
24	Sago waste	<i>Lampito mauritii</i> <i>Eisenia foetida</i>
25	Sericulture waste	<i>Perionyx excavates</i>
26	Sheep manure + cotton industrial waste	<i>Eisenia foetida</i>
27	Shredded paper or newspaper, coir (coconut husk fiber)	<i>Perionyx excavatus</i>
28	Source separated from human	<i>Eisenia foetida</i>
29	Sugar cane residues	<i>Pheretima elongate</i>
30	Vegetable waste + floral waste	<i>Eudrilus eugeniae</i> <i>Eisenia foetida</i> <i>Perionyx excavates</i>
31	Wooden or plastic	<i>Eisenia foetida</i> <i>Eudrilus eugeniae</i> <i>Perionyx Excavates</i>

Reference; (Gupta and Prakash, 2009)

2.1.4 Vermicompost preparation

Vermicompost is a new innovative technology that requires proper technical adaptation involving the use of earthworms and other microorganisms to digest organic wastes (Selden et al., 2005). In addition, the application of vermicomposting is an easy way to have a positive environmental impact by reducing the amount of green-waste that finds its way into landfills and incinerators. Application of vermicompost resulting in nutrient-rich compost end-product is an environmentally sound amendment to enrich soil for plant growth. However, to produce good vermicompost simply requires the appropriate knowledge for vermiculture processes. Besides raw materials and earthworms, vermicompost has been developed and designed with its structure based on the scale of the farmer or the owner of vermiculture.

Nowadays, there is no design or operation for vermicompost industrial. However, the principle has been developed for vermiculture, especially in terms of biotic and abiotic factors which influence vermicast production, earthworm growth and fecundity (Aira et al., 2003; Ismail, 1997). Vermicompost has been practicing in different aspects based on the model of farms on a large and small scale. In addition, the technologies available for proper vermicomposting include beds (i), windrows (ii), and container systems (iii), and each system has a different design. However, vermicompost containers should be constructed with a concrete base or raised above the ground and covered. Some type of container is needed to house compost worms for vermicomposting. Systems can be as simple as a stack of plastic food-storage containers or as complex as an automated unit capable of processing hundreds of pounds of organic matter daily. According to Qi (2012), it has been reported that there are three basic types of vermicomposting systems of interest to farmers: windrows, beds or bins, and flow-through reactors. Each type has a number of variants. Windrows and bins can be either batch or continuous-flow systems, while all flow-through systems, as the name suggests, are of the continuous-flow variety. Beds or Bins: A top-fed bed works like a top-fed windrow used for feeding earthworms. It consists of different layers based on the small farmers' holdings for passing from layer to layer. (Edwards et al., 1972). On the other hand, Rostami (2011) has been reported that there are two major methods of vermicomposting; vermicomposting in bins and vermicomposting in vermicompost piles. The bin method is prepared to use in small scale such as home composting, in kitchen or garage and so on. The bin can be made of various materials, but wood and plastic ones are popular. Plastic bins, because of their lightness, are preferred for in-home composting. A vermicompost bin may be in different sizes and shapes, but its height should be no more than 30 cm. Bins with a height of 30-50 cm, and not much more than that, are perfect. Draining some holes in the bottom, sides and cap of the bin is so helpful to aeration and drainage. Around 10 holes with 1-1.5 cm in diameter is a good choice. Before feeding the worms using wastes, it needed to apply a worm's bed. A height of 20-25 cm. in bedding is appropriate. It may be a mixture of shredded paper, mature compost, old cow or horse manure with some soil. The pile method is mostly used for vermicomposting on a larger scale than the bin method. Whatever vermicompost is chosen as the way to

process a large amount of waste, the application of piles is cost- beneficial. The piles can be made in a porch place like a greenhouse or on a floor with some facilities for drainage in warm climates. Although the pile size may be so various in width and length, however, it cannot be so high and is better to follow the height of bin method.

Overall, vermicompost involves ingestion of the substrate by the earthworm, physical size-reduction of the ingested particles by the action of the earthworm gizzard, which is located next to the worm mouth, digestion of the substrate as it passes through the earthworm body and is acted upon by the microorganism and enzyme present in the earthworm gut.

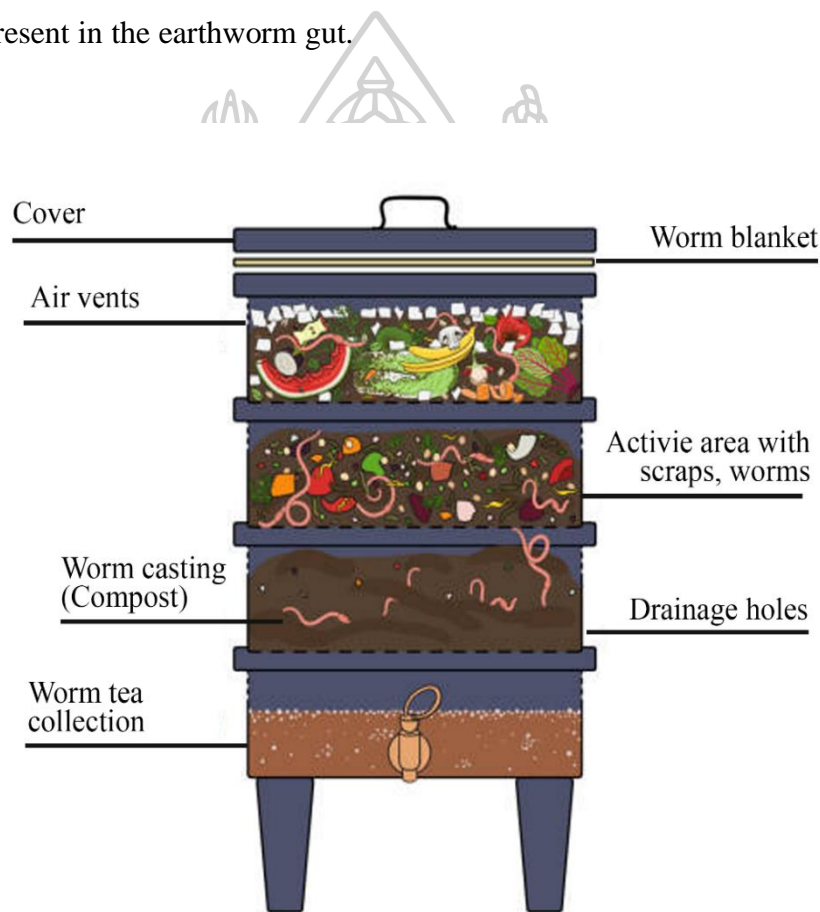


Figure 3. The infographic about vermicomposting shows the components of a vermicomposter. Vermicomposter schematic design. Worm composting. Recycling organic waste into organic fertilizer. The illustration was created by hand.

2.1.5 Vermicompost and its substances (Plant Hormones)

The presence of plant hormones in vermicompost may contribute to an increase in crop yield as well as plant growth. Furthermore, hormonelike substances (indole acetic acid, gibberellins, cytokinin) with significant effects on plant growth have been isolated from vermicompost during the early studies with vermicompost (Chen et al., 2010; Tomati et al., 1988). The presence of plant hormones is the association of plant growth-stimulating activity in vermicompost with microorganism-derived hormone substances. This is provided by data on the correlation between a number of certain groups of microorganisms and plant growth promotion (Grantina-Ievina et al., 2013). More than that, there has been reported from experiments that particular microbiological isolates from vermicompost have growth-stimulating activity on plants (Gopalakrishnan et al., 2015; Sreevidya et al., 2016). Ravindran et al. (2016) investigated the difference between composting and vermicomposting. Interestingly, the observation of phytohormones in both compost and vermicompost showed the maximum phytohormones (mg/kg) were detected as indole-3-acetic acid (7.37), kinetin (2.8), and gibberellic acid (5.7) in vermicompost and the minimum were recorded as IAA (5.84), kinetin (2.7), and GA (3.4) in compost. Hence, his report showed vermicompost was richer in phytohormones than compost. The results revealed that the maximum amount of phytohormones present in vermicompost was due to the mutual action of microbes and earthworms (Yatoo et al., 2021). Apart from this, Tomati et al. (1988) also recorded high values of hormones like auxins, cytokinins, and gibberellins in sewage sludge-based vermicompost. On the other hand, Pathma and Sakthivel (2013) have been reported based on the molecular and functional characteristics of bacteria isolated from vermicompost. He found that bacteria belonged to three major genera, viz., *Pseudomonas* (15%), *Bacillus* (57%), *Microbacterium* (12%) and the remaining bacteria comprised of the genera *Acinetobacter* (5%), *Chryseobacterium* (3%), *Arthrobacter*, *Pseudoxanthomonas*, *Stenotrophomonas*, *Paenibacillus*, *Rhodococcus*, *Enterobacter*, *Rheinheimera* and *Cellulomonas*. In addition, based on the functional characterization of the bacteria was assessed by the production of protease, cellulase, lipase, xylanase, chitinase, amylase, gelatinase, 1-aminocyclopropane-1-carboxylate deaminase, indole-3-acetic

acid (IAA), phosphate solubilization, nitrate reduction, and assimilation of different carbon source. Therefore, besides availability, nutrient application of vermicompost also enriches phytohormones, which are the mechanism for promoting plant growth due to the fact that vermicompost may be rich in beneficial micrograms such as bacillus bacteria and others.

2.1.6 Effect of vermicompost on plant growth

Besides providing of mineral nutrients, vermicompost have additional beneficial effects on plant growth. The optimum plant growth and development are the most important for getting yields. To achieve this, sufficient amounts of nutrients should be applied to the soil through inorganic and organic sources. Vermicompost is an organic source of plant nutrients that contains a higher percentage of nutrients necessary for plant growth in readily available forms (Theunissen et al., 2010). As a result, vermicompost has the potential to improve plant growth and dry matter yield when added to the soil. According to some studies, the results have shown that vermicompost plays a vital role in plant growth and yield of different field crops, including vegetables, flowers and fruit crops. For example, the application of vermicompost in combination with paper mill sludge and cow-dung has given a higher percentage of seed germination about 86-98 percent for seed maize and 84-98 percent for seed cowpea (GI and GP) (Karmegam et al., 2019). Apart from plant growth, different aspects of seed germination and seedling establishment are usually positively affected by low doses of vermicompost application. These include total seed germination, germination index, speed of seedling emergence, and seedling vigor. In addition, some researchers have indicated only the positive effect of vermicompost on seed germination. As an example, there was a 16% increase in germination percentage of *Pinus pinaster* (Lazcano et al., 2008). It is possible that one of the reasons for the high genetic diversity of germination responses to vermicompost treatment is related to differences in seed endosperm reserves: in seeds with a high potential to sustain prolonged seedling growth, biologically active substances in vermicompost can more likely result in inhibitory effects (Meghvansi et al., 2020). Besides of seed germination, Arancon et al. (2004) have been studied on the effects of vermicompost on strawberry growth and yields, showing that

vermicompost from food wastes and paper wastes increased strawberry growth and yields significantly, including increases of up to 37% in leaf areas, 37% in plant shoot biomass, 40% in numbers of flowers, 36% in numbers of plant runners and 35% in marketable fruit weights. Vermicompost from waste corn pulp blended with cow-dung and paper for 30 days exhibited 30%, 40%, and 67% increase in the soil nitrogen, phosphorus and potassium content, respectively. Furthermore, zinc, copper, manganese and iron were increased by 91%, 67%, 56%, and 10%, respectively (Manyuchi et al., 2014). Vermicompost was derived from pig manure applied at a rate of 100% significantly improved leaf numbers, shoot lengths, shoot and root dry mass of tomato seedlings compared with those grown in commercial potting media (MM360) after 21 days (Atiyeh et al., 2002). Zuo et al. (2018) have been reported that vermicompost not only increases plant growth but also improves fruit yield and the contents of soluble sugar and vitamin C. However, the rate or amount of vermicompost application has to be concerned with the agro-climatic conditions of the growing environment since it could affect plant growth and development.

Moreover, the plant growth regulator can be produced by microorganisms in the earthworm gut during vermicomposting (Moradi et al., 2014). Apart from this, humic acids play a vital role in terms of regulators of plant growth, mainly through changes in root architecture and growth dynamics, which result in increased root size, branching, and/or greater density of root hair with a larger surface area (Canellas and Olivares, 2014). Furthermore, humic acid is formed by the biodegradation of dead organic matter and is a major component of soil humus (Stevenson, 1994). Also, some studies indicated that humic acids abstained from Leonardite and peat were found to increase root mass of Kentucky bluegrass (*Poa pratensis L.*) by 73% and root strength by 34% (Ervin et al., 2008). Humic acids were reported as a component in vermicompost (Ramnarain et al., 2019). Humic acids from vermicompost have been reported to stimulate an increase in the number of roots, giving plants the ability to scavenge nutrients from the growing environment for growth and development (Calvo et al., 2014). According to Arancon et al. (2003), humic acids derived from cattle and paper waste vermicompost displayed a significant effect on the increment of root growth and number of strawberry fruits. Leaf areas, plant heights, and above-ground dry matter weights all increased considerably in plants grown in pots containing

humic acids. Vermicompost may also have beneficial and significant effects on other crops besides cucumbers such as sweet corns, strawberries, and lettuces (Hernández et al., 2010).

As the previous study has been reported, application of vermicompost had a positive effect on growth, including seed germination, plant vegetative growth, mineral nutrient uptake, photosynthesis and increasing the crop yield (Hosseinzadeh et al., 2016; Pii et al., 2015). The advantages of using vermicompost are based on both direct and indirect effects on plants, as well as the additional improvement of soil properties with potential long-term benefits and soil sustainability.

2.1.7 Effect of vermicompost on photosynthesis

Photosynthesis is a process which provides the energy necessary for plant growth and reproduction. Chlorophyll represents the principal class of pigments responsible for light absorption and photosynthesis in plant cells (Chaplin and Westwood, 1980). Moreover, photosynthesis is a complex process that is sensitive to environmental factors such as macronutrients and micronutrients (Marschner, 1995). Nutrients such as N, P, K, Mg, Fe and Cu, which are readily available through vermicompost, are used in the formation of chlorophyll, which is required for light harvesting and subsequent conversion into chemical energy via photo-assimilation (Tanaka et al., 1998). Magnesium (Mg) is bound to chlorophyll in amounts ranging from 6 to 35% of the total Mg and plays an important role in photosynthetic CO₂ fixation (Fischer and Bremer, 1993). Iron affects the synthesis of chlorophyll precursor S-aminolevulinic acid, thus, playing an important role in chlorophyll biosynthesis (Pushnik and Miller, 1989). Similar to copper, it is part of the plastocyanin protein responsible for electron transmission during the photosynthesis process (Ayala and Sandmann, 1988). Phosphorus and K are also essential in several biochemical activities, including photosynthetic CO₂ fixation, respiration, cell division, maintenance of high pH of the chloroplast stroma, stomata conductance, water regulation and transport as well as protein synthesis (Humble and Raschke, 1971). Deficiency of these plant nutrients may deter the formation of chlorophyll, resulting in chlorotic leaves (Kapur and Sharma, 2020). Such chlorotic leaves are not able to harvest and convert light energy into the chemical energy required by the plant

for the photosynthesis process. Since vermicompost contains plant nutrients including N, P, K, Ca, Mg, S, Fe, Mn, Zn, Cu, and B, the uptake of which has a positive effect on plant nutrition, photosynthesis, the chlorophyll content thus improves the nutrient content of the different plant components (roots, shoots and the fruits) (Fageria, 2001). A plant-available form of nitrogen (nitrate) is found in vermicompost. Available N is greater in vermicompost than conventionally composted manure (Taleshi et al., 2011). N fertilization increased growth and leaf area index of the plant, which in turn increased absorption of light, leading to more dry matter and yield (Ravi et al., 2008). All of the biochemical processes of photosynthesis depend on nitrogenous compounds which provide the basis for all the reactions that take place inside the chloroplast, including chlorophyll, proteins and enzymes required for the photosynthesis process (Isaychev et al., 2020). There are different opinions as to whether vermicompost actually increases the chlorophyll content of the leaves. For example, previous studies have been reported that application of sheep-manure vermicompost on cucumbers (*Cucumis sativus* L.) grown in greenhouses (up to 30 tons per ha) indicated that leaf number, plant height, and chlorophyll content increased significantly ($P < 0.05$) (Azarmi et al., 2009). Also, according to Fan et al. (2014), the net photosynthesis rate, the chlorophyll fluorescence, the content of chlorophyll and the chloroplast ultrastructure of chrysanthemum were improved obviously after foliar application of humic acid compared with those of the control and the NPK fertilizer. In addition, the results from another study showed that perennial ryegrass (*Lolium perenne* L.) grown in soils amended with 10 to 20% of vermicompost showed an increased in chlorophyll content compared with plants from un-amended soils (Cheng et al., 2007). On the other hand, there has been found that application of 20% vermicompost in the bedding had the greatest effect on the increase of photosynthetic pigments (chlorophyll a, total chlorophyll and carotenoids) compared with 40% and 60%. However, it has been reported that applying up to 40% vermicompost with a spray of 0.5% mM putrescine can be effective for greenhouse cucumber production (Soltani et al., 2019). It is therefore worth establishing if improvement of soil nutrients via vermicompost at different rates will enhance the chlorophyll production and photosynthetic activity in selected vegetable crops.

2.1.8 Effect of vermicompost on preventing plant diseases

Plant diseases, weeds, and insects reduce crop yields by 36% globally, and diseases alone have been reported to reduce crop yields by 14% (Agrios, 2005). On the other hand, the application of chemicals against such diseases mostly gives good results. However, the misuse of these inorganic substances has been an issue of public concern as they have destroyed the natural fertility of soil, killed beneficial soil organisms, reduced the natural resistance ability in crops, and also led to environmental pollution (Bisen et al., 2015; Keswani et al., 2014). Therefore, to overcome these problems, it is imperative to apply biological control like vermicompost and vermicompost tea as a safe and efficient alternative against diseases, which have been considered important in the last few years for the control of many soil-borne phytopathogens (Abada and Hassan, 2017; Devi and Das, 2016; Ragab et al., 2015). Interest in the application of vermicompost for the suppression of crop diseases has grown significantly within the last two decades, and there is evidence that the microbiota of vermicompost helps in biocontrol to suppress pathogens (Ersahin et al., 2009). Due to vermicompost is a finely-divided mature peat-like material which is produced by a non-thermophilic process involving interactions between earthworms and microorganisms (Dash, 2019) leading to biooxidation and stabilization of organic material (Aira et al., 2002). Furthermore, vermicompost is an effective biofertilizer and bio-control agent (Edwards et al., 2004). In the term of biocontrol, other living organisms are used to control pests. Biological control provides an eco-friendly management of crop pests by their antagonists. Besides, soil with low organic matter and microbial activity are conducive to plant root diseases (Stone et al., 2004). The addition of organic amendments can effectively help fertilize soil with more organic and nutrient matter, thus suppressing plant disease occurrences (Blok et al., 2000). Microbial antagonism might be one of the possible reasons for disease suppression as organic amendments enhance the microbial population and diversity. Traditional thermophilic composts promote only selected microbes, while non-thermophilic vermicompost is a rich source of microbial diversity and activity and harbors a wide variety of antagonistic bacteria, which act as effective bio-control agents, aiding in the suppression of

diseases caused by soil-borne phytopathogenic fungi (Scheuerell et al., 2005). Solid vermicompost in the laboratory, greenhouse, and field has proven to have great potential in the suppression of disease incidences caused by pathogens such as *Pythium*, *Phytophthora*, *Fusarium*, *Rhizoctonia*, and *Verticillium*. According to Szczech and Smolińska (2001), they have been demonstrated that the disease suppression ability of vermicompost produced from various waste streams, i.e., animal manures, etc. Because vermicompost harbors a wide variety of antagonistic bacteria, they act as effective biocontrol agents in the suppression of diseases caused by soil-borne phytopathogenic agents (Sinha et al., 2008). According to Edwards et al. (2010) have been illuminated that the soluble nutrients, free enzymes, soluble phenolic compounds, and a wide range of microorganisms pass into the tissues of plants from aqueous extracts of vermicompost, which may be the reason for the suppression of pest and diseases. Also, disease suppression mechanisms termed as “general suppression” have been defined by nutrient competition, antibiosis, in which beneficial organisms secrete antibiotics that directly inhibit the pathogen, hyperparasitism/direct parasitism (one organism feeding on another), and possibly induced systemic plant resistance (Hoitink and Grebus, 1994). Besides, You et al. (2019) to examine the influence of bamboo waste-based vermicompost on the damping-off disease of cucumber. It was found that damping-off caused by each strain of *Pythium aphanidermatum*, *P. ultimum* var. *ultimum*, and *Rhizoctonia solani* AG1-IB was substantially decreased by vermicompost as compared to an autoclaved vermicompost and a commercial nursery medium. Interestingly, this was attributed to the higher diversity and activity of microbes in vermicompost as compared to autoclaved vermicompost and commercial medium. Other authors have also reported that when organic amendments, including vermicompost, were applied, the incidence of diseases like late blight in potatoes and fusarium wilt in cucumbers was reduced significantly (Zhang et al., 2020). Besides vermicompost, application of vermicompost tea as a bio-control agent has increased significantly. An investigation was conducted by Singh et al. (2003) to test the effect of aqueous extract of vermicast by spraying it directly over the leaves against powdery mildews of pea and balsam caused by *Erysiphe pisi* and *Erysiphe cichoracearum*, respectively. Apart from this, Zaller (2006) investigated the influence of vermicast tea on the growth, yield, fruit

quality and the potential disease inhibition against *Phytophthora infestans* on three varieties of tomatoes. He noted a reduced vulnerability of leaves, stems, and fruits of tomato plants against disease caused by *Phytophthora infestans* as compared to water sprinkled control plants. This was attributed to the higher diversity and activity of microbes in vermicompost as compared to autoclaved vermicompost and commercial medium. In addition, Chaoui et al. (2002) also examined a significant reduction in disease attacks by the fungus *Pythium* in cucumbers and *Rhizoctonia* in radishes in greenhouses. Substitution of small quantities into sterilized MM360 was adequate to induce *Pythium* suppression, and that was attributed to the presence of less aeration in the soil that might lead to a greater competition between *Pythium* and beneficial microorganisms for resources. Small vermicompost substitution rates (10% by volume) may reduce *Pythium* disease, whereas the highest substitution rates (40% by volume) may suppress *Rhizoctonia* disease. They indicated the presence of *Trichoderma* spp. in the vermicompost, known as a bio-control agent for *Rhizoctonia*. They also achieved efficient suppression levels on disease incidences caused by *Verticillium* in strawberries and *Phomopsis* and *S. fuliginosa* in grapes in field conditions. They stated that the ability of pathogen suppression disappeared when the vermicompost was sterilized, convincingly indicating that the biological mechanism of disease suppression involved was “microbial antagonism”. Their results supported the statement earlier made by Edwards (1998) that earthworms promote microbial activity and diversity in organic wastes to levels even greater than those in thermophilic composts. As a result, vermicompost demonstrated an even greater potential for plant disease suppression than aerobic composts, most likely due to the stimulatory effects of earthworms on soil microbial activity (thereby encouraging competing microorganisms). Ascituo et al. (2006) evaluated the growth and suppression effect of vermicompost mixed with substrate at 75, 50, and 25% by volume on the disease incidence of damping-off of patience plants (*Impatiens walleriana*) caused by *R. solani*. Treatments with 100-75% of vermicompost showed important increases in leaf area, plant height, and fresh and dry weight of aerial and subterranean organs. As vermicompost is rich in microbial activity and contains antagonistic organisms that control plant pathogens, it is an effective biocontrol agent.

There are so many bacteria species that have been reported in vermicompost produced by different species of earthworm, as shown in Table 2.

Table 2 Different types of bacteria in different species of earthworms

Vermicompost Earthworm	Bacterial types	Beneficial traits	References
<i>Eudrilus sp.</i>	<i>Free-living N₂ fixers</i> <i>Azospirillum,</i> <i>Nitrosomonas,</i> <i>Nitrobacter,</i> <i>Ammonifying</i> <i>bacteria, Phosphate</i> <i>solubilizers,</i> <i>Fluorescent</i> <i>pseudomonas</i>	Plant growth promotion by nitrification, phosphate solubilization and plant disease suppression	(Gopal et al., 2009)
<i>Eisenia fetida</i>	<i>Proteibacteria,</i> <i>Bacteroidetes,</i> <i>Verrucomicrobia,</i> <i>Actinobacteria,</i> <i>Firmicutes</i>	Antifungal activity against <i>Colletotrichum</i> <i>coccodes, R. solani, P.</i> <i>ultimum, P. capsica</i> <i>and Fusarium</i> <i>moniliforme</i>	(Yasir et al., 2009)
<i>Lumbricus</i> <i>terrestris</i>	<i>Florescent</i> <i>pseudomonas,</i> <i>Filamentous</i> <i>actinomycetes</i>	Suppress <i>Fusarium</i> <i>oxysporum f. sp.</i> asparagi and <i>Fusarium</i> <i>proliferatum</i> in asparagus, <i>Verticillium</i> <i>dahliae</i> in eggplant and <i>Fusarium. oxysporum</i> f. sp. <i>Lycopersici</i> Race 1 in tomato	(Elmer, 2009)
<i>E. fetida</i>	<i>Bacillus spp. Bacillus.</i>	Antimicrobial activity	(Vaz-Moreira

Vermicompost Earthworm	Bacterial types	Beneficial traits	References
	<i>megaterium, Bacillus</i>	against <i>Enterococcus</i>	et al., 2008)
	<i>pumillus, Bacillus</i>	<i>faecalis</i> DSM 2570,	
	<i>subtilis</i>	<i>Staphylococcus aureus</i> DSM 1104	
<i>Aporrectodea</i>	<i>Pseudomonas</i>	Suppress	(Doube et al.,
<i>trapezoides</i>	<i>corrugata 214OR</i>	<i>Gaeumannomyces</i>	1994)
<i>Aporrectodea</i>		<i>graminis</i> var. <i>Tritid</i> in	
<i>rosea</i>		wheat	
<i>A. trapezoides</i>	<i>Rhizobium meliloti L5-</i>	Increased root	(Stephens et
<i>Microscolex</i>	<i>30R</i>	nodulation and	al., 1994)
<i>dubius</i>		nitrogen fixation in	
		legumes	
<i>Pheretima sp.</i>	<i>Pseudomonas</i>	Oxalate degradation	(Khambata
	<i>oxalaticus</i>		and Bhat,
			1953)
<i>Lumbricus</i>	<i>Rhizobium japonicum,</i>	Plant growth	(Madsen and
<i>rubellus</i>	<i>Pseudomonas putida</i>	promotion	Alexander,
			1982)

As the table showed about earthworm and bacterial species, recent studies on vermicompost using a paper mixture with cow-dung through the earthworm *E. fetida* showed the enrichment of the population of microbes such as bacteria, fungi, and actinomycetes which are involved in controlling plant disease (Singh and Suthar, 2012). More than that, it has been reported that vermicompost by promoting the growth of *Flavobacterium*, *Acidobacterium*, and *Planctomycetes* bacteria can inhibit the growth of pathogenic bacteria (Lv et al., 2018). In addition, earthworm feeding reduced the survival of plant pathogens such as *Fusarium* sp. and *Verticillium dahlia* but increased the densities of antagonistic fluorescent pseudomonads and filamentous actinomycetes, while population densities of *Bacilli* and *Trichoderma* spp. remained unaltered shown in Table 2. It has been reported that

substitution of vermicompost in the growth media reduced the fungal diseases caused by *R. solani*, *P. drechsleri* and *F. oxysporum* in gerbera (Rodríguez-Navarro et al., 2000). Apart from this, solid vermicompost mostly produced from animal manure could inhibit fungal root pathogens. The results from Szczech et al. (2002) showed that the addition of vermicompost from cattle manure to the container media significantly reduced infections of tomato plants by *P. nicotianae* and *F. axysporum*. In another report, vermicompost from cattle manure, sewage sludge, and a mixture of vegetable wastes demonstrated a great potential for the suppression of disease incidences caused by *Pythium*, *Phytophthora*, *Fusarium*, *Rhizoctonia*, and *Verticillium* (Kannangara et al., 2000). It is not the only vermicompost from cattle manure, but it is also from household waste and other organic wastes that have been reported by the researchers for vermicompost application in plant disease and pest control.

As previous studies have been revealed, the application of vermicompost can be used as biocontrol for suppressed plant diseases and pest control (Table 4). The application also plays a vital role in sustainable agriculture, which maintains the environment and is low-cost.

Table 3 Diseases and pest control by vermicompost

No	Disease/Pet	Crops	Treatment	References
1	Jassid (<i>Empoasca</i> verri), alphid (<i>Aphis craccivora</i>)	Groundnut	Vermicompost	(Rao et al., 2001)
2	Damping off and root rot	Cucumbers	Vermicompost	(Simsek Ersahin et al., 2009)
3	Damping-off	Tomatoes	Vermicompost	(Rivera et al., 2004)
4	Damping-off	Patience plant (<i>Impatiens walleriana</i>)	Vermicompost	(Asciutto et al., 2006)
5	<i>Tetranychus urticae</i> ,	Bush beans,	Vermicompost	(Arancon et al.,

	Pseudococcus sp.	Eggplant,		2007)
	Myzus persicae	tomato, Cucumber, and Cabbage		
6	Collar rot	Chickpea	Vermicompost	(Sahni et al., 2008)
7	Fusarium wilt	Chick pea	Vermicompost	(Gopalakrishnan et al., 2011)
8	Helicoverpa Zea and Pieris rapae	Cabbage	Vermicompost	(Little and Cardoza, 2011)
9	Earworm (elicoverpa zea)	Corn plant	Vermicompost	(Cardoza and Buhler, 2012)
10	Aphid (Lipaphis erysimi)	Mustard	Vermicompost	(Gorakh et al., 2011)
11	Fusarium wilt	Tomato	Vermicompost	(Basco et al., 2017)
12	Damping-off	Cucumber	Vermicompost	(You et al., 2019)
13	Polyphagotarsonemu latus	Chili	Vermicompost	(Jangra et al., 2019)
14	Late blight disease	Potato	Vermicompost	(Peerzada et al., 2020)
15	Fusarium wilt	Cucumber	Vermicompost	(Zhang et al., 2020)
16	Powdery mildew	Pea and Balsam	Vermicompost tea	(Singh et al., 2003)
17	Late blight	Tomatoes	Vermicompost tea	(Zaller, 2006)
18	Foot rot	Rice	Vermicompost tea	(Manandhar and Yami, 2008)
19	Root-Knot Nematode	Cucumber and Tomato	Vermicompost tea	(Mishra et al., 2017)
20	Fusarium wilt	Brinjal	Vermicompost tea	(Barman and Kalita, 2013)
21	Reniform nematode	Zucchini	Vermicompost tea	(Wang et al., 2014)

22	<i>Sclerotium cepivorum</i>	Onion	Vermicompost tea	(Amin et al., 2016)
23	Root-knot	Cucumber	Vermicompost tea	(Mishra et al., 2018)
24	Meloidogyne incognita and Rotylenchulus reniformis	Cucumber	Vermicompost tea	(You et al., 2018)
25	Meloidogyne incognita	Banana plant	Vermicompost tea/Vermicompost	(Awad-Allah and Khalil, 2019)
26	Meloidogyne incognita	Tomato	Vermicompost tea	(Liu et al., 2019)
27	Meloidogyne incognita	Tomato and bell pepper	Vermicompost tea	(Dos Santos Pereira et al., 2020)

2.2 Cucumber

2.2.1 Cucumber and its Signification

The production of vegetables, particularly cucumber in greenhouses has become an important agricultural pattern all over the world because of the growing consumption rate of vegetables and the limited cultivated lands (Liu et al., 2020). Cucumber (*Cucumis sativus* L.) is also a widely cultivated vegetable crop grown in open fields and greenhouses as well. Furthermore, it is one of the most economically important vegetable crops in Thailand (Plapung et al., 2014). Besides Thailand, cucumbers are also an important crop which plays a vital role in Cambodia. According to Ali (2002), the top eight vegetables in the five major vegetable-growing provinces are cucumber, cabbage, cauliflower, eggplant, yardlong bean, tomato, Chinese cabbage and mustard. These provinces include Kampong Cham, Kampot, Siem Reap, Kandal and Battambang. Cucumber is a member of the *Cucurbitaceae* family, which comprises 90 general and 750 species (Sitterly, 1972). *Cucumis sativus* L. belongs to the genus *Cucumis* which comprises about 30 different species distributed over two geographically distinct areas, the African group and the Asiatic

group (Esquinas-Alcazar and Gulik, 1983). The cucumber is one of the oldest cultivated vegetable crops because it has been known in history for over five thousand years and probably originated in India (Whitaker and Davis, 1962). After that, it seems to have spread from India to China and westward to Asia Minor, North Africa, and Southern Europe long before written history. The cucumber was grown by the ancient Greeks and Romans in about 300 BC (Before Christ.). Furthermore, it has been cultivated almost throughout Europe in the Middle Ages. In France, cucumber was common in the ninth century and England in 1327 (Paris et al., 2012). There are three main varieties of cucumber such as slicing, pickling and burpless and the plant has large leaves that form a canopy over the fruit (Mariod et al., 2017). The fruit of the cucumber is roughly cylindrical, elongated with tapered ends, and may be as large as 60 cm long and 10 cm in diameter. Having an enclosed seed and developing from a flower, botanically speaking, cucumber is classified as pipes, a type of botanical berry (Nonnecke, 1989). The optimum temperature range for the growth and development of cucumbers is 18 °C-24 °C (Singh et al., 2008).

Cucumber fruit is a very good source of vitamins A, C, K, B6, potassium, pantothenic acid, magnesium, phosphorus, copper and manganese (Vimala et al., 1999). As well as The ascorbic acid and caffeine contained in cucumber helps to reduce skin irritation (Okonmah, 2011). According to USDA (2011) and Ersahin et al. (2009), the appropriate nutritive value per 100g of fresh edible cucumber contains energy 12 cal, vitamin A 45 IU, protein 0.6g, vitamin B1 0.03g, Fat 0.1g, Vitamin B2 0.02g, carbohydrate 2.2-3.6g, niacin (Vitamin B3) 0.3g, dietary fiber 0.5g, vitamin C 12mg, calcium 14mg, magnesium 15mg, iron 0.3 mg, potassium 124mg, sodium 5mg, phosphorus 24mg and zinc 0.2 mg. Also, the high-water content makes cucumber a diuretic and it also has a cleansing action within the body by removing accumulated pockets of old waste material and chemical toxins. The green leaves are used as potherbs and seeds and seed oils are also edible (Hazara et al., 2011). Furthermore, cucumbers help eliminate uric acid, which is beneficial for those who have arthritis, and their fiber-rich skin and high levels of potassium and magnesium help regulate blood pressure and promote nutrient functions. The magnesium content in cucumber also relaxes nerves and muscles and keeps blood circulating smoothly (USDA, 2012). Also, it is a common ingredient in salads, being valued mainly for its crisp texture and

juiciness and usually contains 90% water, which is consumed fresh (Sotiroudis et al., 2010). Because of the small quantity of sugar present in the cucumber fruits, which helps in the burning of excess fat in the body, they are very good for diabetic patients (Kumar et al., 2010). It has ascorbic and caffeic acid. Both compounds help smooth skin irritation and reduce swelling. It was shown that the skin of the cucumber fruit has chlorophyll and silica (Duke, 1997). The seeds are useful for quitting burning sensations, constipation, tonics, and intermittent fevers (Warrier, 1993). In addition, cucumber contains a wide variety of biologically active, nonnutritive compounds known as phytochemicals, such as alkaloids, flavonoids, tannins, phlobatannins, steroids, and saponins, among others (Zieliński et al., 2017). Some bioactive compounds that have been reported from this plant belong to different chemical groups (Altemimi et al., 2017). The Cucurbitacins that can be isolated from members of the family Cucurbitaceae, such as cucumber (*Cucumis sativus* L.) and melon (*Cucumis melo* L.), which provides a bitter principle characteristic exhibited cytotoxicity and anticancer activity (Kaushik et al., 2015). The polyphenol contents have also been reported in cucumber such as tannins, cardiac glycosides, terpenoids, carbohydrates, resins, saponins, and phytosterols (Sahu and Sahu, 2015). They also suggested that the plant could be a source of useful drugs, but they recommended further studies to isolate the active component of the crude plant extract for proper drug development (Sood et al., 2012).

2.2.2 Cucumber Productivity

The production of cucumbers in greenhouses has become an important agricultural pattern all over the world because of the growing consumption rate of vegetables and the limited cultivated lands (Kuswardhani et al., 2013). The estimated total world production of cucumbers in 2017 was 83,753,861 metric tons, up 3.9% from the 2016 total of 80,616,692 tons shown in Table 3. Furthermore, China has been standing in the number one spot as the world's largest producer of cucumbers, followed by Iran, Russia, Turkey, the United States, Mexico, Ukraine, Uzbekistan, Spain, and Japan as the top 10 producers. Thailand is also stranded on the topic of 30th

in the world of cucumber production from 2017 to 2016. World ranked cucumber production in 2016 and 2017 is shown in Table 4.

Table 4 Production of cucumbers in the world (tons)

Rank	Country/Region	2016	2017	2018
01	China	61,899,582	64,824,643	56,240,428
02	Iran	1,707,190	1,981,130	2,283,750
03	Turkey	1,992,968	1,940,010	1,848,273
04	Russia	1,811,681	1,827,782	1,604,346
05	Mexico	802,220	1,012,378	1,072,048
06	Ukraine	886,270	956,005	1,072,048
07	Uzbekistan	948,900	896,280	857,076
08	United stated	933,310	813,591	700,819
09	Spain	770,704	634,824	643,661
10	Japan	550,300	559,500	550,000
11	Poland	538,057	543,726	538,676
12	Kazakhstan	519,858	488,723	460,110
13	Egypt	430,218	424,933	457,795
14	Indonesia	404,028	409,700	433,931
15	Netherlands	370,000	400,000	410,000
16	South Korea	333,760	341,364	333,233
17	Germany	260,915	256,689	267,589
18	Cameroon	282,773	248,632	257,211
19	Sudan	248,800	237,316	240,405
20	Belarus	248,690	236,618	226,443
21	Azerbaijan	217,843	220,903	223,790
22	Tajikistan	161,390	178,035	211,612
23	Jordan	280,157	122,247	209,362
24	Romania	186,471	201,001	208,585
25	India	186,361	191,064	195,768
26	Algeria	138,481	171,610	193,647
27	Palestine	167,791	169,079	170,367

Rank	Country/Region	2016	2017	2018
28	Greece	152,799	142,289	155,000
29	Lebanon	151,832	151,695	151,558
30	Thailand	178,527	165,192	150,570

Source: (Mahmood et al., 2019).

Cucumbers are adapted to a wide variety of soil types that have good drainage and adequate soil-holding capacity. Climatic conditions, which include temperature, moisture, sunlight and others, influence the development of the crop (Ingestad, 1974). It grows best in sandy loam with a well-drained and adequate amount of organic manure and moisture with an optimum pH of 5.5-7.0 range and temperatures between 18-24 °C (Norman, 1992). Low humidity and high temperatures result in poor fruit set due to the dropping of flower buds. High humidity reduces yields and tends to promote excessive vegetable growth. Temperature also affects crop yield by influencing the rate of photosynthesis and respiration. For nutrient requirements, cucumbers require low nitrogen, but they need high potassium and high phosphorus levels (Ingestad, 1973). Anyway, cucumber is a rapid-growing culture with high nutrition requirements. In addition, the cucumber plant is highly sensitive to the salt concentration of the nutrient solution and very sensitive to the environment (biotic and abiotic) as well (Shopova et al., 2019).

2.2.3 Disease problems in cucumber production

Cucumber growing is very sensitive to environmental factors such as biotics and abiotic. Also, there are several common phytopathogens and diseases-which can cause damage and a decline in the yield of greenhouse cucumber such as Fusarium wilt, Powdery mildew, Downy mildew and Alternaria blight (Migocka and Papierniak, 2011). Termed plant pathogens, they include bacteria, fungi, viruses and nematodes. Besides, greenhouse cucumbers are considered one of the main limiting factors during cucumber production (Punja et al., 2019).

2.2.3.1 Root and stem diseases

Root and stem rot of cucumber (*Cucumis sativus* L.) caused by *Fusarium oxysporum* f. sp. *radicis-cucumerinum*, is a new catastrophic disease of greenhouse cucumber crops in Greece and some countries in the world (Pavlou et al., 2002). Early symptoms in cucumber plants appear as pale-yellow lesions at the stem base in weeks 6 and 8 after sowing (Rose et al., 2003).

Furthermore, Fusarium wilt is also a common vascular wilt fungal disease, exhibiting symptoms similar to Verticillium wilt and this disease has been investigated extensively since the early years of this century (Gordon, 2017). The pathogen that causes Fusarium wilt is *F. oxysporum* (Snyder and Hansen, 1940). Fusarium wilt of cucumber caused by a fungal pathogen. *Fusarium oxysporum* f. sp. *cucumerinum* is one of the most destructive soil-borne diseases throughout the world (Han et al., 2019). *Fusarium oxysporum* f. sp. *cubense* (FOC) has been considered a necrotrophic or hemibiotrophic pathogen. Apparently, FOC needs to interact during part of its life cycle with living plant cells. In addition to the strategies described in the previous paragraph, FOC evolved to have a diverse array of proteins that determine infection capacity in bananas (Takken and Rep, 2010).

The bacterial wilt is caused by *Ralstonia solanacearum*. Over 4,000 papers have been published on this major plant disease (Elphinstone et al., 2005). Also, bacterial wilt disease, caused by *Ralstonia solanacearum*, is one of the most important pathogens in banana plantation (Addy et al., 2016). *Ralstonia solanacearum* is a soil-borne pathogen, very diverse in its species, and belongs to the group of *Ralstonia-species complex* (Seal et al., 1993). Bacterial wilt is a common, often destructive, disease of cucurbits. This disease can cause near complete losses of a planting before the first harvest. Bacterial wilt primarily affects cucumber and muskmelon (cantaloupe). While squash and pumpkin are also susceptible, the damage to these hosts is usually less severe. Initially, individual leaves or groups of leaves wilt on vines, followed by rapid wilting of entire runners or whole plants (Seebold and Bessin, 2014).

Another potential problem for numerous vegetables or cucumber is Phytophthora crown and root which caused by the soilborne

fungus, *Phytophthora capsica* (Maleki et al., 2011). The symptoms of root and crown rot were observed on cucumbers (*Cucumis sativus* L.) in a greenhouse. Cucumbers were the only crop in the greenhouse that used rockwool as a growing substrate in a hydroponically system. The first symptoms were detected in propagation material. One week after planting, symptoms of root and crown rot were observed and approximately 10% of the plants died. Later, losses of 50% in some greenhouses were observed. A yield reduction of as much as 65% was estimated (Herrero et al., 2008). It is spread by splashing water and air currents. The disease typically occurs following periods of heavy rain or irrigation and in low or poorly drained areas of the field that remain wet for several days. Disease development is favored by soil temperatures above 65°F, air temperatures between 75° and 85°F, and extended wet periods (Pivonia et al., 2002).

Downy mildew caused by fungus (*Pseudoperonospora cubensis*). Furthermore, there are a variety of fungal species that cause this disease; some specialize in one type of plant, while others can infect multiple plant types. Downy mildew favors shade and moisture. The fungi cannot survive extremely cold winters (like those in the Northeastern U.S.), but in temperate regions, they can overwinter in plant debris. Moreover, its symptoms appear light green to yellow, angular spots on the leaves. You will also find fuzzy, dark gray spots with a purplish tint (spores) on the underside of the leaves, a tell-tale sign of downy mildew. As the disease progresses, leaves will dry out, become brown, and fall off. However, visible symptoms are not always consistent (Ishii et al., 2001)

2.2.3.2 Fruit and Leaves Diseases

Cucumbers are generally infected by fungi, bacteria, phytoplasmas, nematodes, and viruses (Nazarov et al., 2020). However, cucumber mosaic virus (CMV) is one of the plant pathogenic viruses in the family of *Bromoviridae* which causes yield losses of as high as 40 to 60% (Walkey and Payne, 1990). On the other hand, cucumber mosaic virus (CMV) is one of the widest spreading viruses in the world, infecting over 1200 species belonging to more than 100 families of dicotyledonous and monocotyledonous plants (Edwardson and Christie, 1991). Its symptoms seem like yellow, wilting leaves that appear to be drying out. Besides, it

affects the stem and leaves. CMV also affects fruits and actually affects over 1200 plant species, including cucumbers and related squash, melons and curcubits, tomatoes, peppers and related plants and nearly all commonly grown vegetable and ornamental garden plants. In fact, it has the widest host range of any known plant virus.

Apart from this, powdery mildew in cucumbers (*Cucumis sativus* L.) can be caused by two fungi such as *Erysiphe cichoracearum* and *Sphaerotheca fuliginea*. They are *Erysiphe cichoracearum* DC and *Sphaerotheca fuliginea*, which are distinct from one another in intralia by the fruit bodies of the sexual stage, the *cleistothecia*. *Sphaerotheca fuliginea* has *cleistothecia* with one ascus, while those of *E. cichoracearum* have more asci. Furthermore, it is very common in nearly all cucumber-growing areas. Moreover, its symptoms seems like white, powdery spots or layers on the leaves and stems and fruits can also be affected (Kooistra, 1968).

Phytophthora root rot, crown rot, leaf and stem blight, and fruit rot of cucumber can cause serious losses, and are difficult to control. Furthermore, Phytophthora blight and root rot are caused by fungal-like organisms belonging to the genus *Phytophthora*. They are more commonly referred to as water-molds due to their ability to produce asexual, swimming spores in the presence of water. Several species of *Phytophthora* are responsible for diseases in ornamental plants in greenhouses, nurseries, and landscapes in North Carolina, including *P. nicotianae*, *P. cactorum*, *P. cryptogea*, *P. drechsleri*, *P. palmivora*, and *P. tropicalis* (Foster and Hausbeck, 2010) Therefore, cucumber plant diseases are mostly caused by pathogens, which include fungi, bacterial and virus diseases. However, cucumber plants can be infected by diseases that affect the leaves, roots, and stems.

CHAPTER III

MATERIALS AND METHODS

3.1 Materials

Cucumber seeds (East-West Seed International Ltd, Thailand) were purchased from a local store in Phetchaburi province. Cow dung vermicompost fertilizer was purchased from a local farm in Nakorn Pathom province. It was made by the heap method using the earthworm *Eisenia foetida* and cow manure as the parent raw materials. Chemical fertilizer (20:20:20) (Esteem Intertrade Co., Ltd, Thailand) was bought from a local store in Phetchaburi, Thailand. The soil used in this study was local non-fertilized soil purchased from a local supplier in Phetchaburi province, Thailand. Plastic pots (25 cm height x 18 cm in diameter) were bought from a local store in Phetchaburi province, Thailand.

3.2 Treatments and Experimental Design

The experiment was laid out in a Complete Randomized Design (CRD) consisting of 5 treatments with 8 replications including soil control (T1), soil with chemical fertilizer (T2), and soil mixed with 10, 20 and 30% vermicompost (T3, T4, and T5) as shown in Table 5.

Table 5. The composition of the treatments

Treatment	Composition
T1	Soil only
T2	Soil + Inorganic fertilizer (N-P-K = 20-20-20)
T3	Soil + cow-dunk vermicompost 10% w/w
T4	Soil + cow-dunk vermicompost 20% w/w
T5	Soil + cow-dunk vermicompost 30% w/w

3.3 Cucumber Plantation

The cucumbers were planted in the greenhouse at the Faculty of Animal Sciences and Agricultural Technology, Silpakorn University, Phetchaburi, Thailand from August, 10th 2020 to January 21st, 2021. Seedlings were prepared in the seed tray in the laboratory room on 10th July 2020. Seven-day-old seedlings were transferred into a pot containing the desired planting materials for each treatment and kept in the greenhouse (Chen et al., 2017). Application of chemical fertilizer was foliar applied by spraying to the appropriate pots from week 2nd until week 6th of planation (Rahbar et al., 2018). During cucumber plantations, water was applied by pouring two times per day in the early morning and evening, depending on the rainfall condition. Weeds and pests were monitored every day and were removed by hand. No pesticide was applied during the experiment.

3.4 Plant Growth and Yield Measurement

Plant height, number of leaves, and leaf area data were collected for each week during plantation weeks 1 to 6. Leave areas were recorded by Portable Leaf Area Meter serial 470-010/01 from (CID Bio-Science, Inc., United States). All fertilized flowers were counted cumulatively. Mature fruits were counted in week 8 and week 9 to assess the total yields and fruit characteristics for each treatment. Fruit length, fruit weight, fruit circumference, and fresh fruit thickness were measured in three randomly selected fruits from each treatment.

3.5 Evaluation of Cucumber Disease Occurrences

To evaluate the effect of vermicompost on controlling plant diseases, disease indexes were recorded during plantation by measuring disease incidence and disease severity. During the week 9th late harvesting period, the disease occurrence on cucumber leaves was evaluated. Disease incidence was calculated using the following formula:

$$\% \text{ Disease incidence} = \frac{\text{Total number of infected leaves}}{\text{Total number of leaves per plant}} \times 100$$

Disease severity is represented as a percentage of the infected area of cucumber leaves during the late harvesting period of week 9th using the formula as shown below. According to the percentage of infected area, infected leaves will be classified into the following five categories (Table 6) and the following formula will be used (Mridha et al., 2007).

$$\% \text{ Diseases severity} = \frac{\text{sum of all numerical ratings}}{\text{total number of leaves per plant} \times \text{maximum rating}} \times 100$$

Table 6. The description of the rating of disease severity based on spot leaf

Numerical rating	Description of rating
0	0 Healthy leaves
1	1-10% infected area of leaf
2	11-25% infected area of leaf
3	26-40% infected area of leaf
4	41-60% infected area of leaf
5	>60% infected area of leaf

Reference: Modified from Mridha et al. (2007).

3.6 Soil and Vermicompost Chemical Nutrient Analysis

The soil was collected after mixing and after plantation. Soil, vermicompost, and mixtures of soil and vermicompost in each ratio were measured for pH, electric conductivity (EC), and macronutrients including total nitrogen, total phosphorus, and total potassium as well as available nitrogen, available phosphorus, and available

potassium. All samples were oven-dried at 40 °C and sieved through 2 mm-sieves before being subjected to macronutrient analysis. The details of the methods are described as below.

3.6.1 Soil pH and soil electric conductivity

Soil pH was measured by using a pH meter (Adwa, Hungary) following the method of (McLean, 1983). Soil samples (10g fresh weight) were weighed and suspended in the deionized water in the soil: water of 1:2. After shaking for a few seconds, the pH was measured and the data were recorded. Whereas, soil electric conductivity was measured following the method of (Rayment and Higginson, 1992). The soil solution was prepared in a 1:3 (soil: water) ratio. The sample was shaken for one hour, then the conductivity was determined by EC meter (AZ Instrument, Taiwan).

3.6.2 Determination of total nitrogen

Analysis of total nitrogen was carried out by using the method of (Kjeldahl, 1883). The Kjeldahl method was used to determine the total nitrogen and available nitrogen content in organic and inorganic samples, which involved three main steps: *(i)* digestion, *(ii)* distillation and *(iii)* titration.

In the digestion process, dried soil weigh 2-5g was transferred to the digestion tube volume of 250 ml. The tube was added the concentrated of sulfuric acid 98% (20 ml) into each digestion tube with a half-spoon of catalyst that consist of (Potassium sulfate 20g + copper sulfate 1g). Then the digestion tubes were loaded into a digester and the heat block was heated by starting the temperature at 200 °C and increasing it to 420 °C. After completed digestion, the tube was changed from color to colorless and kept until it cooled down and placed in the distillation machine for the second step of determination.

In the distillation process, a volume of 50 ml of 30% NaOH (sodium hydroxide) solution was automatically added to the sample while distilling. The digested sample was heated by passing steam at a steady rate and the liberated ammonia was absorbed in 50 ml of 4% boric acid that contained an indicator solution (0.066g methyl red + 0.099g bromocresol green) dissolved in 100ml of 95% alcohol. After the pinkish color of the boric solution turned green, the green color distillate

was subjected to titration with 0.02 N H₂SO₄ (sulfuric acid) until the color changed to its original shade. The amount of ammonium ions as a percentage of nitrogen was calculated by the formula:

$$(\%) \text{ Nitrogen} = \frac{(\text{sample titer (ml)} - \text{blank titer(ml)}) \times \text{Normality of acid} \times \text{Atomic weight of nitrogen} \times 100}{\text{Sample weight (g)} \times 100}$$

3.6.3 Determination of available nitrogen

Available nitrogen analysis was carried out by the alkaline permanganate method, that which consists of two main steps, including digestion and titration. Soil was weighted (2–5g) and transferred to the digestion tube (size 250 ml) with 25 ml of 0.32% KMnO₄ (alkaline potassium permanganate) solution and 25 ml of 2.5% NaOH (sodium hydroxide) solution in the distillation unit. Samples were heated steadily by a passing stream, and the liberated ammonia was absorbed for a few drops in 20 ml of 2% boric acid containing mixed indicator solution (0.066g methyl red + 0.099g bromocresol green dissolved in 100 ml of 95% ethanol). Nearly 150 ml of distillate was collected after being heated. The pinkish color turned green once the ammonium was absorbed. The green distillate was subjected to titration with 0.02 N sulfuric acid until the color changed to its original pinkish color. Available nitrogen was calculated by the following formula (Subbaiah and Asija, 1956):

$$(\%) \text{ Available Nitrogen} = \frac{(\text{sample titer (ml)} - \text{blank titer(ml)}) \times 0.00028 \times 100}{\text{sample weight (g)} \times 100}$$

3.6.4 Determination of total phosphorus

Phosphorus is an essential plant nutrient and it occurs in many different forms such as phosphate ion and orthophosphate. Soil phosphorus is found in two forms, namely organic and inorganic and phosphate. Furthermore, total soil

phosphorus concentrations are generally high, with up to 80% of this phosphorus being immobile and unavailable for plant uptake. Therefore, a reliable procedure for measuring the total amount of phosphorus is needed.

The determination of total phosphorus has been done by the ashing colorimetric and ascorbic acid methods. In the ashing step, soil was weighted (2-5g) and transferred into a crucible cup, then completely oxidized in a furnace at 500 to 550 °C for 24 hours. The cool crucible was placed with 10 ml of 10 M of hydrochloric acid and heated for 20 minutes on the hot plate. Then the crucible cup was transferred to filtration with Whatman filter paper 42, and added deionized water to a volume of 100 ml in the volumetric flask. The liquid samples were kept in the plastic bottle for total phosphorus measurement.

For the measurement with ascorbic acid method, one milliliter of each sample solution was transferred into the dilution tube and combined with reagent A and B (Appendix 2). The intensity of blue color was read on spectrophotometer (Spectro SC) at 880 nm wavelengths. The standard curve was generated by using the phosphorus standard of KH_2PO_4 . Phosphorus concentration was calculated using a standard equation to obtain the F value. The percentage of total phosphorus was calculated by using the following formula:

$$(\%) \text{ Total P} = \frac{F \times 100 \text{ ml of total volume of filtrated} \times 100 \times 10}{1000 \times 1000 \times \text{weight of soil sample}}$$

Where F = concentration of phosphorus (ppm) was calculated using the standard equation (Olsen, 1954).

3.6.5 Determination of available phosphorus

The available phosphorus was determined by following the procedure of Bray No 1 (Bray, 1929), designed to extract absorbed forms of phosphate with 0.025 N HCl (pH <7.5) followed by the ascorbic acid method.

For soil extraction, a sample (2-5g) was placed into 45 ml of centrifuge tube. Twenty milliliters of Bray-I extracting reagent was added into the centrifuge tube that contained the sample, then vortexed for 5 minutes. Samples were filtrated by

Whatman filter paper 42. Then, the sample was made to volume up with the extraction solution to 100 ml in volumetric flask.

One milliliter of filtrate was added with reagent A+B (Appendix 3) for recording data from the spectrophotometer after 10 minutes of incubation. The intensity of the blue color was read on a spectrophotometer by using 880 nm of wavelengths on the spectrophotometer. A serial amount of standard KH_2PO_4 was mixed with reagent A and B for standard curve generation (Appendix 3, Figure 5). Phosphorus concentration was calculated using a standard equation to obtain the F value. The percentage of total phosphorus was calculated by using the same formula as in 3.6.4.

3.6.6 Determination of total potassium

Potassium is the major nutrient for plant growth (Lalitha and Dhakshinamoorthy, 2014). The determination of total potassium in soil samples employed the ashing method and the extracted potassium was measured by the Atomic Absorption Spectrometer AA240 following the method of (Jackson, 2005).

Briefly, soil was weighted 2 to 5g and transferred into a crucible cup, then completely oxidized in a furnace at 500 to 550 °C for 24 hours. The cool crucible was filled with 10 ml of 10 M hydrochloric acid (HCl) and heated on the hot plate for 20 minutes. Then the crucible cup was transferred to filtration using No. 40 filter paper, and added deionized water to a volume of 100 ml in the 100 ml volumetric flask and kept the solution in the plastic bottle for total potassium analysis.

Standard potassium was prepared by using KNO_3 (Appendix 4). Sample measurement was done by mixing 0.01 ml of samples with the calcium chloride reagent for analysis in the absorption spectrometer. Calculation of the percentage of total potassium was done using the formula:

$$(\%) \text{ Total K} = \frac{R \times 100 \text{ ml of total volume of filtrated} \times 100 \times 100}{1000 \times 1000 \times \text{Sample weight}}$$

Where R (ppm) = concentration of potassium calculated from standard equation

3.6.7 Determination of available potassium

Potassium is an essential and major nutrient for crop development. The available potassium was determined by extracting the soil with neutral ammonium acetate and measuring it using the Atomic Absorption Spectrometer.

Briefly, soil was weighted with 2 to 5 g of soil sample and placed in a plastic centrifuge tube. Then 25 ml of ammonium acetate reagent was added into the centrifuge tube. The plastic centrifuge tube that contained the sample with reagent was shaken for 5 minutes, then filtered through Whatman No. 1 filter paper and made up the volume to 25 ml in the plastic bottle.

A standard curve was set up using the standard solution of HNO₃. Sample filtrate (0.1 ml) was taken and combined with reagents for reading in the atomic absorption spectrometer. Reading data were calculated using the following formula (Black and Black, 1965).

$$(\%) \text{ Available K} = \frac{R \times 25 \text{ ml of total volume of extraction} \times 1000 \times 100}{1000 * 1000 * \text{sample weight}}$$

Where R (ppm) = concentration of potassium calculated from standard equation

3.7 Microbial Analysis

Soil and soil mixed with vermicompost samples were collected before and after planting. Serial dilutions of each sample were prepared with 0.85% sodium chloride (NaCl). One hundred microliter aliquots of each dilution were plated onto a nutrient agar (NA) plate in triplicate. After incubation at 27 °C for 24 hours, colonies were count and calculated in CFU/ml unit.

3.8 Statistical Analysis

Data were analyzed using variant (ANOVA) in the R program version 4.0.3. Differences among means were tested by Duncan's multiple range tests ($p \leq 0.05$).



CHAPER IV

RESULTS

4.1 Effect of Vermicompost on Plant Growth

4.1.1 Plant Heights

Plant heights were recorded using a meter and represented in a centimeter (cm) unit as shown in Table 7. Application of vermicompost showed a positive effect on plant height in greenhouse conditions ($p \leq 0.05$). The results demonstrated that, after 2 weeks of plantation, 30% of the vermicompost treatment group showed the highest plant-height followed by 20% and 10% of the vermicompost, respectively. This may be due to the enrichment of soil with essential nutrients for plant growth provided by vermicompost.

In the poor soil condition of this experiment, control soil and chemical treatment showed significantly low values of plant height during week 6th plantation. Foliar application of chemical fertilizer showed a positive effect on plant height at week 5th; however, the effect was less than application of vermicompost treatment.

4.1.2 Leave Number

The main function of a leaf is to produce food for the plant by photosynthesis, due to photosynthesis takes place mainly in a plant's leaves. Therefore, the number of leaves had an effect on cucumber growth. Similar to plant heigh, application of vermicompost significantly increased the leave number. As demonstrated in Table 8, vermicompost treatment provided a higher number of leaves compared to control soil and chemical fertilizer. The results revealed that 30% of vermicompost significantly promoted the production of leaves at the highest levels (21.25 leaves) at week 6th of plantation compared to other treatments.

Table 7. Plant height (means¹ ± SD, cm) of cucumber planted in different potting material treatments.

Treatment ²	1 st week	2 nd week	3 rd week	4 th week	5 th week	6 th week
T1	13.50 ^d ± 0.41	22.08 ^d ± 0.80	34.34 ^d ± 2.19	45.59 ^d ± 1.41	47.20 ^e ± 1.91	54.80 ^e ± 4.07
T2	12.40 ^e ± 0.65	12.30 ^e ± 0.45	25.33 ^e ± 0.14	43.32 ^d ± 1.57	60.30 ^d ± 0.62	68.10 ^d ± 0.22
T3	16.65 ^c ± 0.60	41.68 ^c ± 0.68	62.98 ^c ± 0.48	82.39 ^c ± 2.48	106.54 ^c ± 0.58	127.25 ^c ± 0.50
T4	19.18 ^a ± 0.35	46.87 ^b ± 1.92	74.60 ^b ± 3.95	107.47 ^b ± 0.45	114.15 ^b ± 1.26	148.57 ^b ± 2.79
T5	18.20 ^b ± 0.27	53.46 ^a ± 0.29	115.38 ^a ± 0.10	126.22 ^a ± 0.83	185.67 ^a ± 0.58	205.69 ^a ± 1.08
CV value	3.01	3.00	2.76	1.74	1.02	1.49

Note: ¹ Means in each column with different superscript letters were significantly different by Duncan's Multiple Range Test ($p \leq 0.05$).

² The treatment including soil control (T1), soil with chemical fertilizer application (T2), and soil mixed with 10, 20, and 30% vermicompost (T3, T4, and T5).

Table 8. Leave number (means $^1 \pm$ SD) of cucumber planted in different potting material treatments.

Treatment ²	1 st week	2 nd week	3 rd week	4 th week	5 th week	6 th week
T1	1.00 ^c \pm 0.63	3.50 ^b \pm 0.53	5.00 ^c \pm 0.00	6.80 ^c \pm 0.45	6.20 ^c \pm 0.45	6.00 ^d \pm 0.00
T2	1.00 ^c \pm 0.76	3.16 ^b \pm 0.35	4.83 ^c \pm 0.41	5.75 ^c \pm 0.46	7.80 ^d \pm 0.45	8.00 ^d \pm 0.00
T3	2.00 ^b \pm 0.84	5.00 ^a \pm 0.63	8.40 ^b \pm 0.55	10.75 ^b \pm 0.96	10.71 ^c \pm 0.49	12.86 ^c \pm 0.38
T4	2.38 ^{ab} \pm 0.52	5.40 ^a \pm 0.55	8.40 ^b \pm 0.55	12.50 ^b \pm 0.58	13.17 ^b \pm 0.41	16.17 ^b \pm 0.98
T5	2.63 ^a \pm 0.52	5.72 ^a \pm 0.49	10.80 ^a \pm 0.45	16.75 ^a \pm 1.26	18.25 ^a \pm 1.49	21.25 ^a \pm 2.43
CV value	1.17	11.54	5.81	7.68	7.12	9.86

Note: 1 Means in each column with different superscript letters were significantly different by Duncan's Multiple Range Test ($p \leq 0.05$).

2 The treatment including soil control (T1), soil with chemical fertilizer application (T2), and soil mixed with 10, 20, and 30% vermicompost (T3, T4, and T5).

4.1.3 Leave Area

The leave area was recorded by using a leaf area meter (Portable Leaf Area Meter serial 470-010/01) and represented as centimeter square (cm²). As shown in Table 9, vermicompost has a positive impact on the leaf area. The cucumber plant showed larger leaves in 10%, 20%, and 30% vermicompost treatments compared to control and chemical treatment. In the first week, the application of vermicompost 10%, 20% and 30% were not significantly different from each other. However, after 2 weeks of plantation, 30% vermicompost treatment showed the best results with the highest leave area at week 6 of 166.85cm², followed by 20% vermicompost (117.06 cm²) and 10% vermicompost (117.32cm²).

4.1.4 Fertilized Flower and Fruit Yield

The fertilized flowers were recorded in weeks 8th and 9th by counting them as cumulative fertilized flowers in each treatment. Based on this study, cucumber plants started producing flowers at around week 5th in 20% of vermicompost and became fertilized. The cumulative number of cucumber fruits is shown in Table 10. The control treatment showed the lowest number of fertilized flowers, but pots consisting of vermicompost produced a greater number which were 95, 87, and 83 flowers for 30%, 20%, and 10% vermicompost, respectively (Table 10).

Mature fruits were harvested from week 8th to week 9th. However, not all fruits can be harvested; only some of them grow until maturity. Many fertilized flowers fell since there was heavy rain during the plantation. However, as shown in Table 10, 30% vermicompost promoted the production of fruits better than other treatments with the highest total yield of 1108.27g, followed by 20% vermicompost (689.41g), 10% vermicompost (232.51g), and chemical fertilizer (159.52g). Unfortunately, there is no fruit that can be harvested in the control treatment.

Table 9. Leave area (means \pm SD, cm²) of cucumber planted in different potting material treatments

Treatment ²	1 st week	2 nd week	3 rd week	4 th week	5 th week	6 th week
T1	15.54 ^c \pm 1.66	67.06 ^d \pm 2.84	84.14 ^d \pm 1.04	87.96 ^d \pm 0.62	87.25 ^d \pm 1.50	95.42 ^d \pm 1.50
T2	7.80 ^b \pm 0.13	56.28 ^e \pm 0.43	75.50 ^e \pm 3.81	85.65 ^d \pm 0.92	95.67 ^c \pm 2.89	104.08 ^c \pm 2.89
T3	27.30 ^a \pm 6.69	126.68 ^c \pm 0.15	136.69 ^c \pm 2.51	146.20 ^b \pm 2.17	106.25 ^b \pm 0.50	117.32 ^b \pm 0.50
T4	28.25 ^a \pm 0.30	128.29 ^b \pm 0.35	143.87 ^b \pm 1.62	152.69 ^c \pm 2.39	107.25 ^b \pm 1.89	117.06 ^b \pm 2.89
T5	27.05 ^a \pm 1.96	134.92 ^a \pm 0.39	193.82 ^a \pm 0.94	227.16 ^a \pm 2.85	177.25 ^a \pm 2.06	166.85 ^a \pm 2.06
CV value	14.76	1.45	1.60	1.36	1.50	1.64

Note: ¹ Means in each column with different superscript letters were significantly different by Duncan's Multiple Range Test ($p \leq 0.05$).

² The treatment including soil control (T1), soil with chemical fertilizer application (T2), and soil mixed with 10, 20, and 30% vermicompost (T3, T4, and T5).

Table 10. Cumulative numbers of fertilized flowers and fruit yields in each treatment

Treatment ¹	Total number of fertilized flowers	Total number of harvested fruits	Total yield (g)
T1	18	0	0
T2	70	5	159.52
T3	83	6	232.51
T4	87	11	689.41
T5	95	13	1108.27

Note: ¹ The treatment code is soil control (T1), soil with chemical fertilizer application (T2), and soil mixed with 10, 20, and 30% vermicompost (T3, T4, and T5).

4.1.5 Fruit Characteristics

The results of the physical characteristics of cucumber fruits were measured for average fruit weight, fruit length, fruit circumference, and fresh fruit thickness. As shown in Table 11, 30% vermicompost showed the highest fruit length (11.24 cm). Cucumber fruits from 20% and 30% vermicompost treatment had a significantly higher fruit weight (138.53 g and 137.88 g for 20% and 30% vermicompost, respectively). Similar results in which 20 and 30% vermicompost showed the significantly highest values were found in fruit circumference and fresh fruit thickness (Table 11). Fruit circumference and fresh fruit thickness of 30% and 20% vermicompost treatment showed the best values compared to 10% vermicompost and chemical fertilizer. The results showed that 30% vermicompost has a great fruit circumference of 13.57 cm with 20% at 12.45 cm. Likewise, the fresh fruit thickness in the 30% vermicompost group was 0.86 cm and the 20% group was 0.92cm. Cucumber fruit shapes in a longitudinal cut were shown in Figure 4.

Table 11. Average fruit weight, fruit length, fruit circumference and fresh fruit thickness (means $^1 \pm$ SD) of each treatment.

Treat Ment ²	Fruit weight (g)	Fruit length (cm.)	Fruit circumference (cm.)	Fresh fruit thickness (cm.)
T1	n. a.	n.a.	n.a.	n.a.
T2	31.90 ^b \pm 6.30	6.17 ^c \pm 0.76	10.48 ^b \pm 0.47	0.53 ^b \pm 0.01
T3	58.13 ^b \pm 8.69	6.61 ^c \pm 0.19	10.75 ^b \pm 0.40	0.60 ^b \pm 0.02
T4	137.88 ^a \pm 32.84	8.82 ^b \pm 0.31	12.45 ^a \pm 0.37	0.92 ^a \pm 0.04
T5	158.32 ^a \pm 32.80	11.24 ^a \pm 1.26	13.57 ^a \pm 0.62	0.86 ^a \pm 0.07
CV Value	24.56	9.30	4.03	5.79

Note: ¹ Means in each column with different superscript letters were significantly different by Duncan's Multiple Range Test ($p \leq 0.05$).

² The treatment including soil control (T1), soil with chemical fertilizer application (T2), and soil mixed with 10, 20, and 30% vermicompost (T3, T4, and T5).

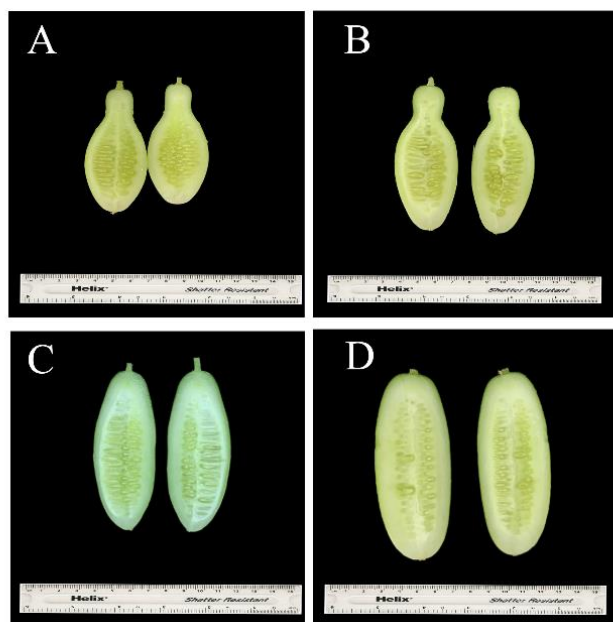


Figure 4. Longitudinal cut of cucumber fruit of (A) T2: chemical treatment, (B) T3: vermicompost 10%, (C) T4: vermicompost 20%, and (D) T5: vermicompost 30% treatment.

4.1.5 Effect of Vermicompost on Leaf Spot Disease Incidence and Severity

As the results are shown in Table 12, leaf spot incidence was significantly different among treatments. It was shown that the highest incidence was found in control (82.11%) and in the application of chemical fertilizer (83.81%), while vermicompost treatment had significantly lower incidences (62.24%, 51.67%, and 66.34% for 10%, 20% and 30% of vermicompost, (respectively). As a result, vermicompost 20% treatment showed the lowest disease incidence at 51.67%. However, the disease severity showed no significant difference among all treatment groups.

Table 12. Leaf spot disease incidence and severity (means \pm SD) of cucumber plants at 9th week after plantation ¹

Treatment ²	Disease incidence (%)	Disease severity (%)
T1	82.11 ^a \pm 1.83	50.00 \pm 0.00
T2	83.81 ^a \pm 3.30	48.78 \pm 2.92
T3	62.24 ^b \pm 1.21	48.50 \pm 2.60
T4	51.67 ^c \pm 2.89	47.41 \pm 2.63
T5	66.34 ^b \pm 1.80	45.38 \pm 3.87
CV Value	3.39	5.68

Note: ¹ Means in each column with different superscript letters were significantly different by Duncan's Multiple Range Test ($p \leq 0.05$).

² The treatment including soil control (T1), soil with chemical fertilizer application (T2), and soil mixed with 10, 20, and 30% vermicompost (T3, T4, and T5).

4.2 Macronutrient Chemical Analysis

4.2.1 Chemical Characteristics of Soil and Vermicompost

Chemical nutrients in soil and vermicompost were determined and presented in Table 13. Soil showed lower values in pH, EC, and all nutrients, including total nitrogen, available nitrogen, total phosphorus, available phosphorus, total potassium, and available potassium compared to vermicompost. These results demonstrated that

the soil used in this study was very poor in nutrients, while vermicompost has much more enriched nutrients than the soil used in this study.

Table 13. Chemical characteristics of soil and vermicompost (means \pm SD).

Parameters	Soil	Vermicompost
pH	5.09 \pm 0.03	6.25 \pm 0.01
EC (mS)	0.06 \pm 0.00	4.56 \pm 0.02
Available N (%)	0.01 \pm 0.00	0.06 \pm 0.00
Total N (%)	0.10 \pm 0.01	1.85 \pm 0.02
Available P (%)	< 0.01 \pm 0.00	0.08 \pm 0.00
Total P (%)	0.20 \pm 0.00	0.45 \pm 0.00
Available K (%)	0.06 \pm 0.00	1.39 \pm 0.00
Total K (%)	0.23 \pm 0.00	0.47 \pm 0.01

4.2.2 Comparison of Chemical Parameters and Nutrients

Soil or a mixture of soil and vermicompost was collected from each treatment and analyzed for pH, electrical conductivity (EC), and chemical macronutrients before and after plantation (Table 14). The results demonstrated that the pH values were increased after mixing the soil with vermicompost and slightly increased after plantation, except for the application of chemical fertilizer (NPK). The pH changed from the acidic range of 4.88 to 5.26 in control soil and chemical fertilizer to the more neutral pH in range of 6.00 to 6.59 in vermicompost treatment. Similar to the effect on pH, mixing with vermicompost made the EC values higher based on the ratio of mixing. However, the electric conductivity (EC) values were decreased in all treatments after plantation.

As shown in Table 14, the levels of both available and total nitrogen and potassium levels were increased when vermicompost was mixed with soil. In all treatments, total nitrogen and phosphorus were reduced after plantation. While available nitrogen and potassium levels in the control and chemical fertilizer groups showed no change after plantation, the 10% 20% and 30% vermicompost groups showed a decrease of these nutrients after plantation. The reduction of nitrogen and

potassium content after plantation might be due to plants absorbing nutrients for their growth.

The levels of total and available phosphorus were found to slightly increase when mixed with vermicompost (Table 14). However, when comparing the change in phosphorus levels during plantation, the levels of available phosphorus were too low to clearly assess the usage of available phosphorus during plantation. Surprisingly, the total phosphorus levels were found to increase after plantation when the soil was mixed with 10%, 20%, and 30% vermicompost but not the control soil and chemical fertilizer shown Table 14.

4.3 The effect of Vermicompost on Bacterial Population

Serial dilutions of soil mixture with vermicompost were prepared in 0.85% sodium chloride (NaCl) and 0.1 ml aliquot of each dilution was spread onto Potato Dextrose Agar (PDA) then incubation at 27 °C for 24 hours. The results demonstrated that the application of vermicompost had positive effects on the microbial population by showing a significant increase in total bacteria count when vermicompost was mixed with the soil ($p \leq 0.05$) (Table 15). The bacteria population increased in a positive relationship to the volume of vermicompost that has been mixed in the soil pot. The highest number of bacterial colonies was found in the 30% vermicompost group, while the lowest levels were in the control soil and with the application of chemicals (Table 15). Therefore, based on the results, besides being low in nutrients, soil used in this study was also low in microbial population.

In addition, the bacteria isolation was clearly increased after plantation. The magnitude of the increment was approximately 3 to 4 times greater from before plantation to after plantation (Table 15). The highest value of bacteria population was observed in a pot treated with 30% vermicompost and the lowest was observed in control and chemical treatment similar to that before plantation (Table 15).

Table 14. Comparison of chemical parameters and nutrients (means¹ ± SD) among each treatment² before and after plantation.

Parameter	Time	T1	T2	T3	T4	T5
pH	Before	5.09 ^c ± 0.03	5.09 ^c ± 0.03	6.00 ^b ± 0.08	6.14 ^a ± 0.02	6.22 ^a ± 0.03
	After	5.26 ^b ± 0.12	4.88 ^c ± 0.05	6.46 ^a ± 0.01	6.51 ^a ± 0.02	6.59 ^a ± 0.01
EC (mS)	Before	0.06 ^d ± 0.00	0.06 ^d ± 0.00	0.75 ^c ± 0.05	1.26 ^b ± 0.00	2.10 ^a ± 0.03
	After	0.04 ^c ± 0.00	0.03 ^c ± 0.00	0.12 ^d ± 0.00	0.18 ^b ± 0.00	0.27 ^a ± 0.00
AN (%)	Before	0.01 ^b ± 0.00	0.01 ^b ± 0.00	0.02 ^b ± 0.00	0.02 ^b ± 0.00	0.03 ^a ± 0.00
	After	0.01 ^b ± 0.00	0.01 ^c ± 0.00	0.01 ^c ± 0.00	0.01 ^b ± 0.00	0.01 ^a ± 0.00
TN (%)	Before	0.10 ^d ± 0.01	0.10 ^d ± 0.01	0.23 ^c ± 0.01	0.32 ^b ± 0.01	0.67 ^a ± 0.01
	After	0.08 ^d ± 0.00	0.07 ^d ± 0.00	0.17 ^c ± 0.00	0.23 ^b ± 0.00	0.36 ^a ± 0.01
AP (%)	Before	0.00 ^d ± 0.00	0.00 ^d ± 0.00	0.01 ^c ± 0.00	0.02 ^b ± 0.00	0.02 ^a ± 0.00
	After	0.00 ^c ± 0.00	0.00 ^d ± 0.00	0.01 ^c ± 0.00	0.01 ^b ± 0.00	0.02 ^a ± 0.00
TP (%)	Before	0.20 ^b ± 0.00	0.20 ^b ± 0.00	0.19 ^b ± 0.01	0.20 ^b ± 0.00	0.23 ^a ± 0.00
	After	0.03 ^d ± 0.00	0.03 ^d ± 0.00	0.20 ^c ± 0.00	0.65 ^b ± 0.01	1.17 ^a ± 0.03
AK (%)	Before	0.06 ^d ± 0.00	0.06 ^d ± 0.00	0.11 ^{cd} ± 0.00	0.16 ^b ± 0.01	0.32 ^a ± 0.00
	After	0.07 ^d ± 0.00	0.06 ^d ± 0.00	0.09 ^{cd} ± 0.00	0.10 ^b ± 0.00	0.15 ^a ± 0.00
TK (%),	Before	0.23 ^c ± 0.00	0.23 ^c ± 0.00	0.26 ^b ± 0.01	0.26 ^b ± 0.01	0.37 ^a ± 0.01
	After	0.17 ^c ± 0.01	0.17 ^c ± 0.00	0.22 ^b ± 0.01	0.21 ^b ± 0.01	0.24 ^a ± 0.01

Note: ¹ Means in each row with different superscript were significantly different by Duncan's Multiple Range Test ($p \leq 0.05$).

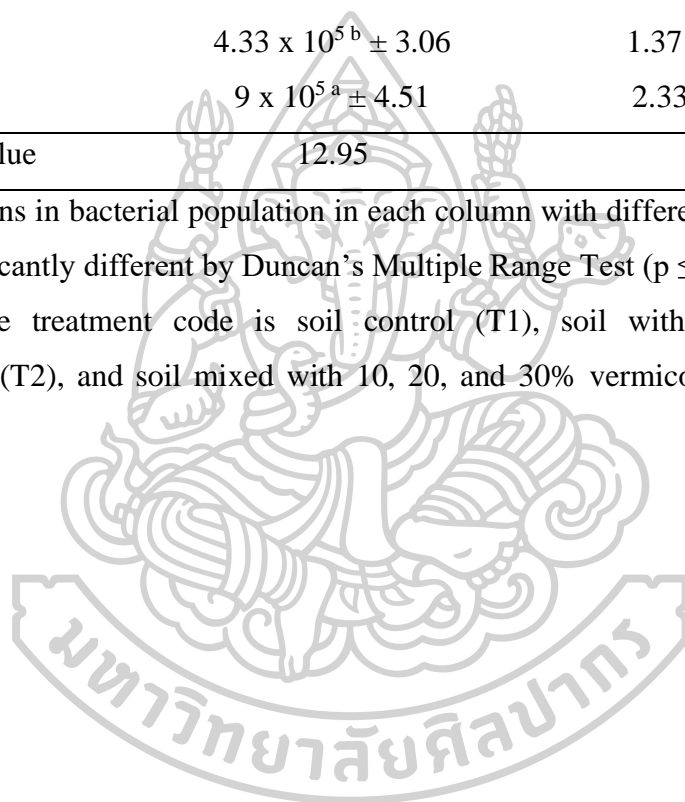
² The treatment code is soil control (T1), soil with chemical fertilizer application (T2), and soil mixed with 10, 20, and 30% vermicompost (T3, T4, and T5).

Table 15. Bacteria population ¹ (CFU/ml) of soil or soil mixed with vermicompost before and after plantation.

Treatment ²	Bacterial population (CFU/ml)	
	Before plantation	After plantation
T1	$1 \times 10^5 \text{ c} \pm 0.00$	$4 \times 10^5 \text{ c} \pm 0.00$
T2	$1 \times 10^5 \text{ c} \pm 0.00$	$4 \times 10^5 \text{ c} \pm 0.00$
T3	$2.67 \times 10^5 \text{ bc} \pm 2.31$	$1.13 \times 10^6 \text{ bc} \pm 2.08$
T4	$4.33 \times 10^5 \text{ b} \pm 3.06$	$1.37 \times 10^6 \text{ bc} \pm 5.77$
T5	$9 \times 10^5 \text{ a} \pm 4.51$	$2.33 \times 10^6 \text{ a} \pm 0.00$
CV value	12.95	28.99

Note: ¹ Means in bacterial population in each column with different superscript letters were significantly different by Duncan's Multiple Range Test ($p \leq 0.05$).

² The treatment code is soil control (T1), soil with chemical fertilizer application (T2), and soil mixed with 10, 20, and 30% vermicompost (T3, T4, and T5).



CHAPER V

DISCUSSION

In recent years, the intensive use of inorganic fertilizers and pesticides in the agricultural field has globally destroyed soil fertility, killed beneficial microorganisms, and decreased natural resistance in crops, thereby making them more vulnerable to diseases and affecting human health and environmental pollution (Yatoo et al., 2021). Due to the adverse effects of chemical fertilizers and pesticides, interest in the use of organic fertilizers as a potting medium has grown, particularly concerning their use as an environmentally friendly and efficient medium for plant cultivation. Thus the application of vermicompost as an organic fertilizer could be a proper choice in sustainable agriculture to promote plant growth, yield, improving soil properties, and increased the soil microbial populations (Anand et al., 2012).

5.1 Effects of vermicompost application on plant growth and yield

The result clearly showed that the application of vermicompost increases the plant growth. The highest plant height, leaf number, leaf area, fruit characteristics, and total fruit yield were obtained in the 30% vermicompost treatment. The improvement in the growth and productivity of plants can be due to the high amount of organic matter together with plant hormones such as auxins, gibberellins, and cytokinins of microbial origin in vermicompost, which enhance plant growth and yield (Ding et al., 2021; Emperor and Kumar, 2015; Singh et al., 2010). Several other reports also suggest that the increase in the growth and yield of plants is a result of the increased nutrient levels in the vermicompost amended soil/media (Das et al., 2017; Tognetti et al., 2005).

Application of vermicompost increased the leaf number per plant. Leaves play an important role in photosynthesis to produce starch, protein, cellulose, etc. Therefore, the number of leaves has effects on photosynthesis and nutrient accumulation. Azarmi et al. (2009) reported that the application of vermicompost at 20 and 30 ton/ha contributed to a greater leaf number compared to the untreated

control. Vermicompost made from goat manure significantly increased the total number of cucumber leaves (Yeole, 2013).

The leaf area was promoted by the application of vermicompost. The largest leaf area was observed in a pot treated with 30% of vermicompost (166.82cm²). However, the leaf area observed in this study was smaller than that observed by Opara et al. (2012). Opara et al. (2012) reported the cucumber leaf area was different among seasons. Leaves were larger during the dry season (343.77cm²) but smaller during the wet season (292.17cm²). The difference may be the contribution of the environmental stress and soil conditions used in this study. Kaciu et al. (2011) reported that vermicompost promoted the leaf area of cucumber seedlings grown under saline conditions.

In this experiment, there was no fruit to be harvested in the untreated control treatment. This could be because the cucumber plant is not strong enough to withstand environmental stress from both biotic and abiotic factors. In addition, most of the flowers fell down in all treatments since there was heavy rain during the plantation. Actually, excessive rainfall can affect crop productivity in various ways, including direct physical damage, restricted root growth, oxygen deficiency, nutrient loss, pathogen effect, and reduced flowering (Li et al., 2019). Li et al. (2019) reported that heavy rainfall reduced maize yields by up to 34% of the total yield. Thus, some researchers suggest that the dry season is more suitable for cucumber planting than the rainy season (Opara et al., 2012).

In contrast to this experiment, the application of 20% vermicompost promoted the highest plant growth and yield, while vermicompost from 30% to 60% retarded plant growth, including lower fruit yields (Akhzari and Pessarakli, 2017). The difference may be due to the differences in the original soil conditions used in the study. Nutrient-rich soil may need a lower amount of vermicompost since their soil was more enriched with nutrients compared with this experiment. Therefore, the application of vermicompost should be considered according to the soil condition in each area to set up the proper ratio for different crops and different soil conditions. It has been reported by Zhao et al. (2019) that the use of the vermicompost amendment aided cucumber fruit yield under continuous cropping conditions. Also, previous

studies have investigated whether the application of vermicompost can alleviate continuous cropping obstacles and improve crop yield and quality (Lv et al., 2020).

5.2 Effect of vermicompost application on plant diseases

As the result demonstrated, the leaf spot disease index was significantly different among treatments. Chemical fertilizer treatment and untreated control showed the highest percentage of disease incidence (82.11% for control and 83.81% in chemical treatment), while vermicompost application led to a reduction of the disease incidence (51.67% from 20% vermicompost treatment). A study on cucumber in plantation by Wylie and Punja (2021) showed that vermicompost suppressed *Rhizoctonia solani* and *Fusarium oxysporum* f. sp. in the laboratory by the action of microbial activity. Many studies have demonstrated the effectiveness of vermicompost in protecting against various plant diseases (Arancon et al., 2002). Earthworms can promote microbial activity and diversity in organic wastes to greater levels than those in thermophilic composts (Nelson et al., 2000). The possible mechanisms for disease suppression included a general competition among microorganisms for nutrients or destruction of pathogen propagules such as spores (Edwards et al., 2006). Some researchers indicate that the improved nutrient availability and the presence of antimicrobial compounds such as flavonoids, phenolics, and humic acids in the vermicompost may induce resistance to pathogens in the plants (Hussain and Abbasi, 2018; Patnaik et al., 2020). The uptake of soluble phenolic compounds from the vermicompost aqueous extracts into the plant tissues could be one of the reasons to combat with the plant pests by making plants unattractive to pests and affected to pest reproduction and survival rates (Ayneband et al., 2017; Edwards et al., 2010).

Microbial antagonism is one of the possible reasons for disease suppression as organic amendments enhance the microbial population and diversity. Thus, vermicompost can be used as biocontrol due to the presence of antibiotic bacteria. (Liu et al., 2021) showed that among 374 bacterial strains isolated from fresh vermicompost, 28 bacterial strains showed antagonistic activity against *Fusarium oxysporum* f. sp. *cucumerinum*. Eight bacterial species, including *Actinomyces*

israeli, *Azotobacter*, *Micrococcus luteus*, *Bacillus cereus*, *Bacillus subtilis*, *Pseudomonas aeruginosa*, and *Enterobacter* were identified in the vermicompost (Satpathy et al., 2020). The presence of these bacteria may play an important role in the increase in cucumber yields, and they have a positive effect on leaf spot disease occurrences.

5.3 Effect of vermicompost on soil chemical and nutrient status

5.3.1 Soil pH

Soil pH has a direct impact on the availability of soil nutrients for plant growth and microbial activity (Neina, 2019). In the experiment, soil pH significantly increased after mixing with vermicompost and slightly increasing after cultivation. However, the use of chemical fertilizer did not improve the soil pH similar to those reported by (Singh Brar et al., 2015). The soil pH obtained from application of 30% vermicompost increased from acidic pH (5.09) to slightly acidic pH (6.22) and increased to more neutral pH (6.59) after cultivation. An increase in pH values upon vermicompost treatment has been previously reported. Mahmud et al. (2020) showed that supplementation with vermicompost led to a significant increase in the soil pH when compared to the soil treated with chemical fertilizer and unfertilized (control) soils. According to Uz and Tavali (2014), the application of vermicompost at a 30 ton/ha⁻¹ dose resulted in higher soil pH values in the fourth, seventh, thirteenth, and sixteenth weeks compared to farmyard manure applied at the same dose. The increase in soil pH may be due to the release of nutrients, organic matter, and microbes from vermicompost, so the mineralizable fractions of C and N increase (Curtin et al., 1998). The consumption of H⁺ by a large number of hydroxyl groups, phenols, and other functional groups produced during the mineralization and humification of organic matter in vermicompost application could lead to the increase of pH (Cai et al., 2020).

Chemical treatment decreased the soil pH after planting. These results are in agreement with (Han et al., 2016). This might be due to the composition of chemical fertilizer with 9% ammonium (NH₄⁺) and 6% nitrate (NO₃⁻) as the source of nitrogen. The leaching of NO₃⁻ and increasing H⁺ accumulation in the soils (released from NH₄⁺) can accelerate soil acidification (Bolan and Hedley, 2003). The results of this

study were in agreement with the results of several studies that have shown vermicompost treatment increased soil pH, but chemical fertilizer treatments, such as NPK fertilizer, decreased soil pH (Altuntas et al., 2018; Liu et al., 2010; Wang et al., 2021).

5.3.2 Soil electrical conductivity (EC)

Soil EC is an important indicator of soil health. It affects to crop suitability, plant nutrient availability, and the activity of soil microorganisms which finally affects to the plant growth and crop yield. The result showed that vermicompost addition increased the soil EC, the highest value observed from 30% of vermicompost treatment. The increase in EC upon vermicompost application has been previously reported Atiyeh et al. (2001); Wang et al. (2017) said that the higher ion content in organic compost led to a higher EC value. In comparison between before and after planting, the soil EC decreased. These results were similar to those reported by Natsheh and Mousa (2014). The decrease in soil EC may be because plants obtain nutrients from the soil, thus the ion content was lower.

5.3.3 Nitrogen content

The nitrogen (N) characteristics of soil directly reflect its fertility. Similar to soil pH and electrical conductivity, results showed that adding vermicompost to the soil increased the level of total and available nitrogen. However, the nitrogen level was found to decrease after planting. This might be because the plant absorbed nutrients for growing.

Several studies elucidated the increase in total and available N upon vermicompost treatment. Zaman et al. (2015) found that the total N, available P, exchangeable K, Ca and Mg, and available S, Zn and B were increased with vermicompost application at 10 ton/ha compared to the unfertilized soils. (Yang et al., 2015) have shown that application of vermicompost increases soil mineral nutrition such as NH_4^+ -N, NO_3^- -N, and phosphorus. Esmailpour et al. (2020) also showed that the increasing of vermicompost levels in the soil significantly increased the percentage of nitrogen based on the level of vermicompost. The authors also reported that the addition of vermicompost in soil affected the content of plant nutrients such

as N, P, K, Mg, Ca, Fe, Zn, Cu, and Mn. Soil available N and P were increased by vermicompost treatment which resulted in the enhancement of soil health and productivity on a long-term basis for sustainable crop production (Yadav et al., 2016). Manivannan et al. (2009) have also been reported that vermicompost significantly increased the available nitrogen, available phosphorus, available potassium, and micronutrients such as Zn, Fe and Cu in pots treated with vermicompost.

5.3.4 Phosphorus content

Phosphorus (P) has an important role in several physiological processes in the plant such as energy storage, photosynthesis, transfer, respiration, cell enlargement, and cell division. Also, phosphorus is an important structural component such as nucleic acids (DNA, RNA), coenzymes, nucleotides, sugar phosphate, and phospholipids. More than that, it stimulates root growth, fruit setting, blooming, and seed formation (Memon, 1996). Phosphorus in the soil can be in the inorganic (orthophosphate) and organic form (phosphates bounding). Soil available phosphorus is the fraction of total phosphorus in soil that is readily available for absorption by plant roots. The dominant inorganic P form in the soil is orthophosphate (HPO_4^{2-} and H_2PO_4^- ions) that can be absorbed directly by plant and microbial cells. Polyphosphates (including pyrophosphate) are another form of inorganic P that may present in soils, generally in low concentrations relative to orthophosphate (Condrón et al., 2005; Fardeau et al., 1988).

In this study, the total phosphorus content was significantly influenced by vermicompost addition similar to those observed in the nitrogen content. The maximum total phosphorus content was observed from pot treated with 30% of vermicompost (0.23%). Similarly, the increase in phosphorus in vermicompost treatment has been previously reported (Zaman et al., 2015). However, it might be noted that total phosphorus was increased after planting. According to Arancon et al. (2006), the increase of orthophosphates could be explained by the significant correlations between the microbial biomass nitrogen and orthophosphates and the release of phosphorus was due largely to the activity of soil microorganisms. Metabolism by soil microbes may lead to the release of several organic acids such as

glucose phosphate, mostly by oxidative respiration or by fermentation when glucose is used as a carbon source (Satyaprakash et al., 2017).

For available phosphorus, the level was low and did not change after planting. This may be due to plants absorbing the phosphorus that is slowly released from vermicompost during planting. On the other hand, it may be because the plant was not able to absorb phosphorus, so the level did not change after planting. Low phosphorus absorption might be one of the reasons for the flower weakness observed in this study, since phosphorus plays an important role in reproductive organs and the initiation of flowering (Malhotra et al., 2018). The low-level of phosphorus availability might result in less resistance to disease, low photosynthesis, low fruit yield, flowers, and seed problems (Farag et al., 2010). Unsuitable soil pH may affect the absorption of phosphorus (Margenot et al., 2018).

(Gichaba et al., 2020) reported that the addition of goat manure-based vermicompost (30 t/ha) increased available phosphorus content in the soil in comparison to the untreated soil at the end of harvesting season. The gradual-release of phosphorus in the soil may be attributed to the release of humic acid during organic matter decomposition, which results in the conversion of unavailable soil phosphorus into available forms by the action of microbial activity (Mahmoud and Ibrahim, 2012). The availability of organic phosphorus is very dependent on conditions in the soil and the weather, which influence microbial activity. The mineralization of organic phosphorus to inorganic forms is favored by optimum soil pH and nutrient levels, good soil physical properties, and warm, moist conditions.

5.3.5 Potassium content

Potassium (K) plays a major role in stimulation of enzymes, photosynthesis, regulation of osmotic pressure, movement of the stomata, protein synthesis, phloem transport, transfer of energy, cation-anion balance in soil and improving resistance against stress (Alfaro et al., 2003; Khawilkar and Ramteke, 1993; Marschner, 2012). In general, potassium is available in four forms in the soil, which are K^+ ions in the soil solution, exchangeable potassium, potassium fixed by weathered micaceous minerals and potassium present in the lattice of certain K-containing primary minerals (Rehm and Sorensen, 1985).

Based on the results, the application of vermicompost increased the total and available potassium content in soil. Additionally, the maximum level of total K content was observed in the pot treated with 30% of vermicompost (0.37%) and the minimum value was observed in the control soil (0.23%). The level of total K was lower compared to Nuraini et al. (2020). The application of compost and vermicompost increased total K in comparison with the original K in the soil, with the highest level of total K occurring in vermicompost treatment (Nuraini et al., 2020). According to Angelova et al. (2013), a significantly higher value of available K was obtained after the vermicompost addition compared to compost fertilizer. This result may be related to the fact that the increase in soil organic matter resulted in decreased K fixation and a subsequent increase in K availability (Olk and Cassman, 1993).

5.4 Effect of vermicompost on bacterial population

Earthworms harbor a variety of decomposer microbes in their gut and excrete them along with nutrients in their excreta, and both are found to be mutual partners. Various enzymes and intestinal mucus in the earthworm's intestinal tract play a key role in the breakdown of organic macromolecules, which in turn results in a greater increment of the available surface area for microbial colonization, their biological activity, and higher nutrient retention (Vijayabharathi et al., 2015). Nitrogen fixing and phosphorus solubilizing bacteria in the earthworms' intestines are important in promoting plant growth by increasing the nitrogen and phosphorus uptake when used as biofertilizers (Shaarani et al., 2019). Researchers have demonstrated that vermicompost contains a higher microbial number and diversity than normal compost (Anastasi et al., 2004; Vivas et al., 2009).

From the results obtained, the application of vermicompost provided the highest number of bacterial populations. Also, soil microbes increased more after cultivation. The result was in agreement with (Sun et al., 2020) that found the maximum enrichment of the microbial communities in the soil with vermicompost mixing. Lazcano et al. (2013) demonstrated that microbial populations were higher in an organic fertilizer group (vermicompost and manure) compared to inorganic fertilizer and initial soil.

Furthermore, in the competition with chemical fertilizer, the results displayed that all treatment with vermicompost provided a large number of bacteria in comparison with chemical fertilizer. This result was similar to those reported by Zhao et al. (2020). The authors reported that microbial functional diversity was higher in the vermicompost treatment than in the application of chemical fertilizer and poultry manure compost treatments in the continuous tomato cropping in a greenhouse. Similarly, Lara-Capistrán et al. (2020) found that with a higher ratio of vermicompost use, a higher number of the bacterium CFU/ml was obtained. Moreover, Maji et al. (2017) also reported that vermicompost treatment showed the highest bacterial diversity and the lowest value was observed in soil with chemical application. According to Nakhro and Dkhar (2010), the application of organic fertilizers increased the organic carbon content of the soil, thereby increasing the microbial counts and microbial biomass carbon. The use of inorganic fertilizers resulted in low organic carbon content, microbial counts and microbial biomass carbon in the soil. This phenomenon can be attributed to the fact that humic substances present in vermicompost may serve as a source of nutrients for microorganisms such as reducing sugars, organic acids, amino acids, peptides, and amino sugars that may promote the growth of indigenous microbial communities, thus increasing overall microbial growth and population (Pathma and Sakthivel, 2012).

Previous studies have been reported on plant growth-promoting rhizobacteria (PGPR) in vermicompost. PGPR are known to improve plant growth and are used as biofertilizers due to their numerous benefits to agriculture such as phosphorus solubilization and phytohormone production (Lebrazi et al., 2020). According to Kawicha et al. (2020), PGPR from vermicompost was able to produce IAA, siderophore, and amylase. Furthermore, *Streptomyces sp.* SEF47 was identified and showed a significant effect on reduction of the disease incidence and severity. Pathma and Sakthivel (2013) reported the isolation of 193 bacteria isolates which belonged to three major genera *Pseudomonas*, *Bacillus* and *Microbacterium* and the rest belong to the genera *Acinetobacter*, *Chryseobacterium*, *Arthrobacter*, *Pseudoxanthomonas*, *Stenotrophomonas*, *Paenibacillus*, *Rhodococcus*, *Enterobacter*, *Rheinheimera* and *Cellulomonas*. Among these 193 isolates, there were up to 96 isolates that showed antagonistic potential against phytopathogenic fungi. As a result, among the bacteria

isolated in this study, some may be beneficial bacteria that improve soil fertility, nutrient availability, plant growth, and disease suppression. For this reason, the identification of isolated bacteria should be further investigated.

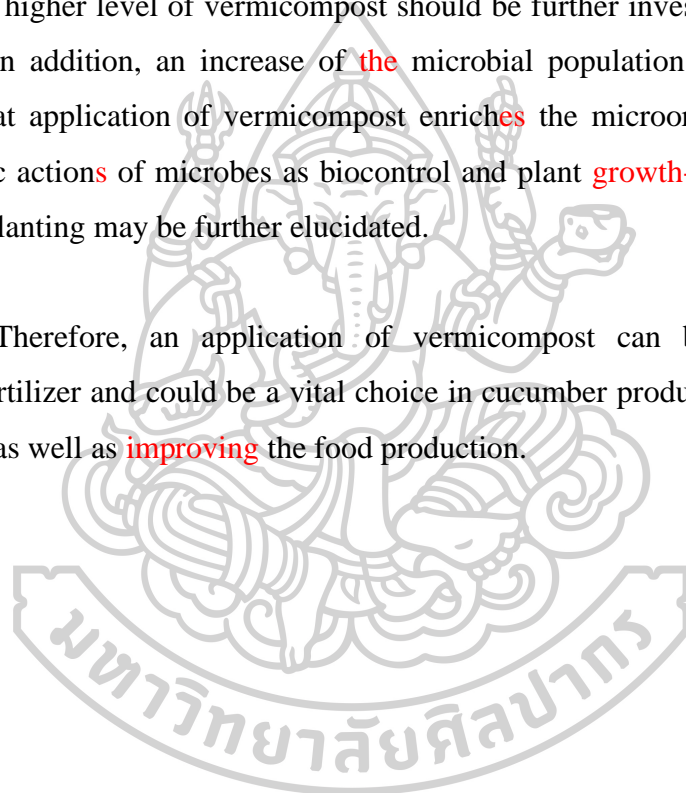


CHAPTER VI

CONCLUSION

In conclusion, vermicompost could be used as a soil amendment to improve the growth, yield and fruit quality of cucumber. In the present study, the application 30% vermicompost showed the best positive effect on improving soil fertility and promoting cucumber plant growth and yields in poor soil conditions. However, a higher level of vermicompost should be further investigated for poor soil condition. In addition, an increase of **the** microbial population in the soil strongly supports that application of vermicompost **enriches** the microorganisms in the soil. **The specific actions** of microbes as biocontrol and plant **growth-promoting agents** in cucumber planting may be further elucidated.

Therefore, an application of vermicompost can be used instead of chemical fertilizer and could be a vital choice in cucumber production for sustainable agriculture as well as **improving** the food production.



APPENDIX

Table 16 Weather parameter during the investigation in Meaung, Phetchaburi province, Thailand

Monthly	Max. temperature (°C)	Min. temperature (°C)	Average Rainfall (mm)	Average rainfall day/month	Relative humidity (RH%)
1 st Season (2019)					
September	30	25	370.3	29	78
October	30	26	369.9	28	76
November	31	24	137.1	20	73
December	30	22	27.6	8	63
2 nd Season (2020)					
January	30	24	2.50	7	65
February	30	24	2.10	8	66
March	31	26	22.4	8	70
April	32	28	89.6	13	68
May	33	29	58.4	27	68
3 rd Season (2020)					
June	31	27	112.6	30	70
July	31	27	206.0	29	72
August	30	26	176.4	28	72
September	31	27	315.2	30	75
October	28	24	633.4	27	82
November	26	24	128.8	25	76

Appendix 1. Chemical reagent for total phosphorus and standard

1. The ash was dissolved in 10 ml of hydrochloric acid and evaporated to about 5 ml on an electric hot plate.
2. Reagent A: Ascorbic acid stock solution: Fresh ascorbic acid reagent dissolved 1.76 g ascorbic acid with 100 ml of distiller water.
3. Reagent (B): Acid molybdate stock solution: Prepare 14% of H₂SO₄ (sulfuric acid solution) 35.71ml with 250 ml of distilled water, and 0.27 g of Potassium antimonyl tartrate with 100 ml distilled water and stored at 4°C in a dark, glass-stoppered bottle. And 4 % of ammonium molybdate adjusted with 100 ml of distilled water and Stored in a plastic bottle at 4°C. Reagent A and B used together for standard curve and reading Total Phosphorus.
4. Phosphorus standard solution (1000 ppm): Dissolved 0.4393 g of pre-dried (105°C for one hour) potassium phosphate monobasic (KH₂PO₄) (CASRN 7778-77-0) in reagent water and diluted to 1000 mL solution.

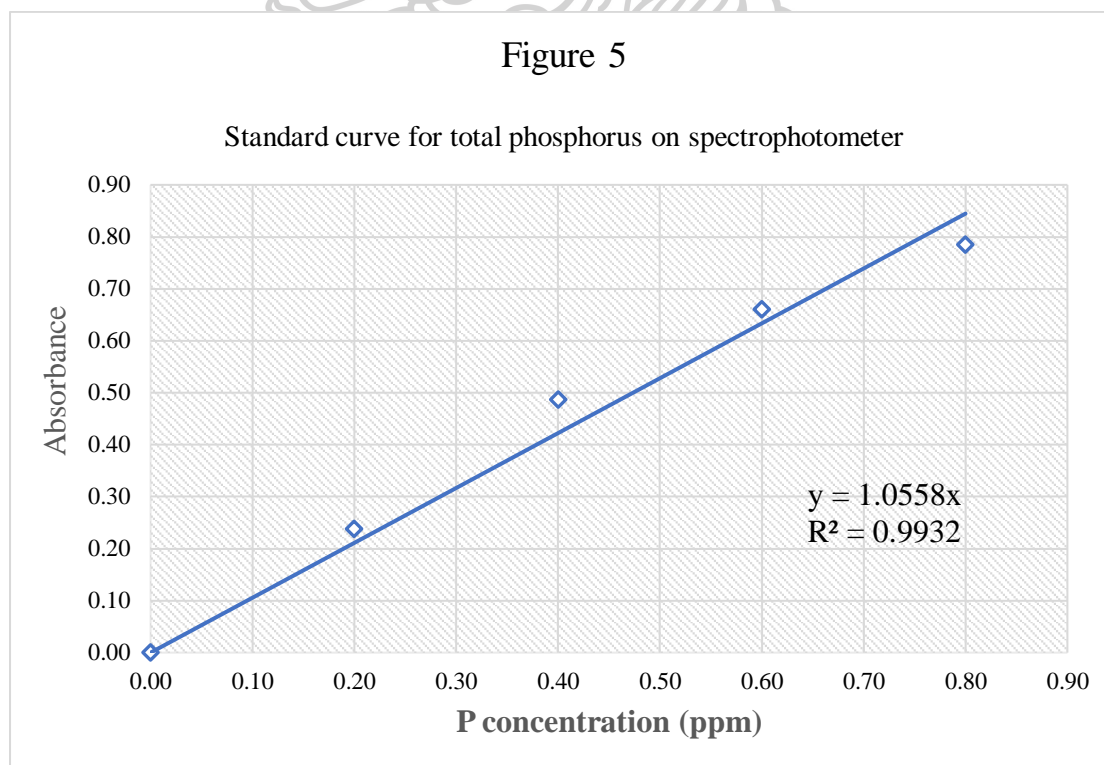


Figure 5 Standard curve for total phosphorus determined by using concentration reagent range from 0.00 ppm to 0.80 ppm of KH₂PO₄.

Appendix 2. Chemical reagent for available phosphorus and standard

1. Chemical Reagent for Soil extraction: neutral normal ammonium acetate was prepared by adding 58 ml of glacial acetic acid to 600 ml of distilled water and then added 70 ml of concentrated ammonia (sp. Gr. 0.90). Diluted the solution to one liter. Then adjusted pH of solution at 7.0 with the help of acetic acid.
2. Bray I Extracting Reagent with Soil sample: soil extraction preparations were done by weighted soil sample 2-5g and placed into centrifuge tube. Added 20 ml of Bray-I extracting reagent into centrifuge tube that contained sample, and shook with Vortex Mixer machine for 5 minutes. Plastic tube samples were filtrated by 42 filter papers. Then, made the volume up with the extraction solution to 100 ml in volumetric flask. The extract sample were kept for measurement.
3. Ascorbic acid chemical reagent and standard preparation method was prepared the same as total phosphorus analysis showed in appendix 2.

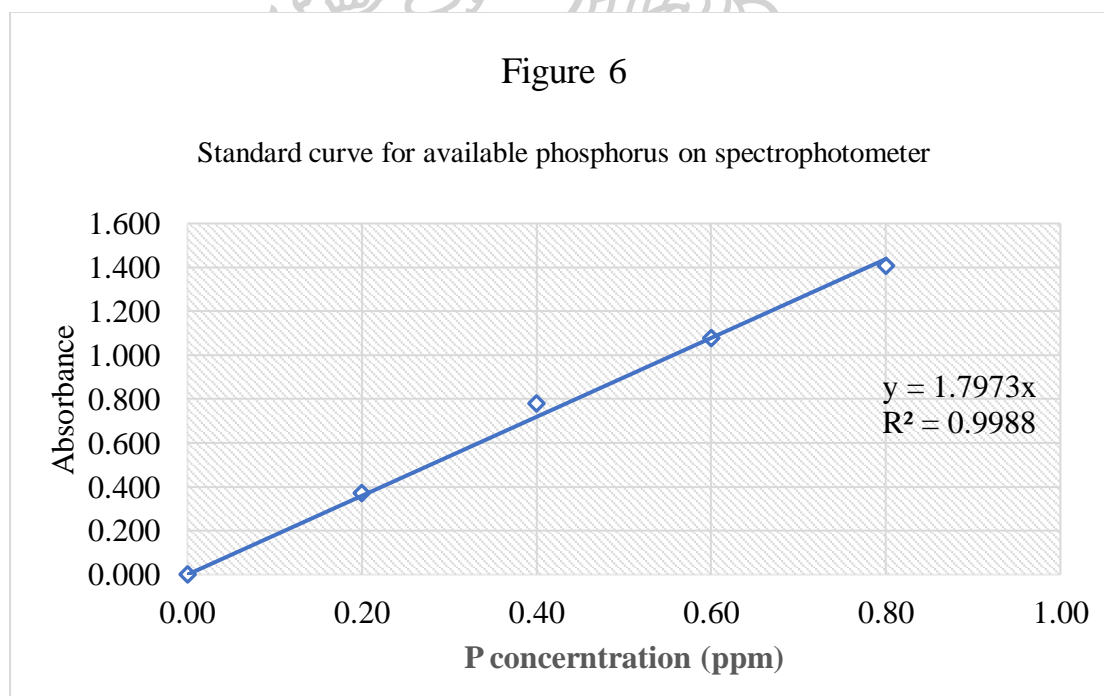


Figure 6. Standard curve for available phosphate determination in soil by using working concentration reagent range from 0.00 ppm to 0.80 ppm of KH_2PO_4 .

Appendix 3. Chemical reagent for total potassium and standard

1. The ash was dissolved in 10 ml. of hydrochloric acid and evaporated to about 5 ml on an electric hot plate the same as total P.
2. Cesium chloride solution equivalent to 5 % cesium: Dissolve 6.330 g of cesium chloride dissolved in demineralized water and adjust the volume to 100 ml.
3. Potassium reference solution at 1g/l commercial or prepared as follows: dissolve 2.5856 g KNO_3 in water, adjusted to 1 liter.
4. Diluted potassium solution at 100 mg/l: Place 10 ml of 1 g/l potassium reference solution in a 100 ml graduated flask and 1 ml of pure nitric acid; complete to volume 100 ml with pure demineralized water for analysis.

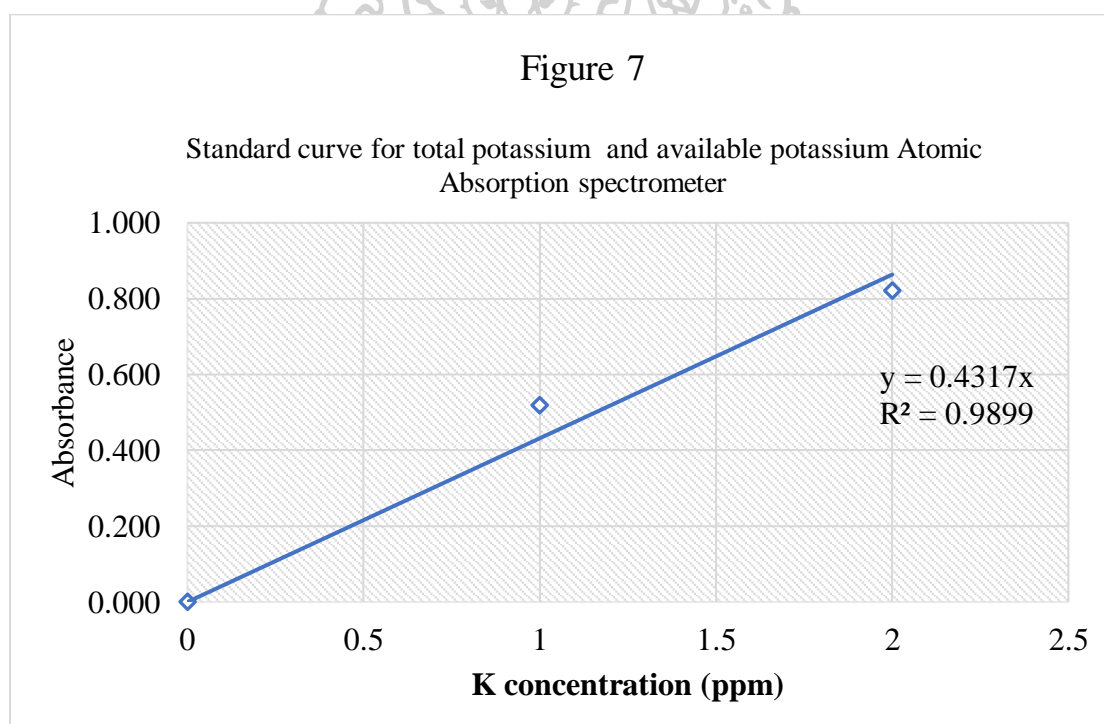


Figure 7 Standard curve for available and total potassium determination in soil from Atomic Absorption Spectrometer.

Appendix 4 Chemical reagent for available potassium and standard

1. Reagents Neutral Normal Ammonium Acetate: Added 58 ml of glacial acetic acid to about 600 ml H₂O and then added 70 ml of concentrated ammonia (sp. gr 0.90) Dilute the solution to one liter. Adjusted pH of solution at 7.0 with the help of ammonia. Acetated (CH₃COONH₄) directly in H₂O and volume to be made one liter and then adjust the pH 7.0.
2. Standard potassium solution used the same as total potassium determination from Atomic Absorption spectrometer.



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