

Chemical and Biological Properties of Potted-Soil for Strawberry Cultivation

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INFO ARTIKEL	ABSTRACT/ABSTRAK
Diterima: 12-07-2022 Direvisi: 15-02-2023 Dipublikasi: 30-04-2023	Sifat Kimia dan Biologi Tanah pada Budidaya Strawberry dalam Pot
Keywords: Kimia tanah, Makronutrisi, Pupuk, Populasi mikroba	Tanaman stroberi yang ditanam di daerah Jawa Barat umumnya dibudidayakan secara intensif dalam pot, akan tetapi tanah yang menjadi media tanam kurang mendapat perhatian. Tujuan dari penelitian ini adalah untuk mengevaluasi profil kimia dan kandungan makronutrisi pada tanah yang digunakan untuk budidaya stroberi, dan untuk mengetahui populasi mikroba menguntungkan yang terdapat dalam tanah dan rizosfir stroberi. Sampel tanah dikumpulkan dari tanaman stroberi yang telah ditanam selama 12 bulan dan satu bulan, keduanya merupakan tanah inceptisol yang masing-masing telah diberi pupuk kandang 10 t/ha dan 25 t/ha. Analisis proksimat dilakukan dengan menggunakan metode Association of Official Analytical Chemist. Penghitungan populasi mikroba dilakukan dengan menggunakan metode pengenceran berseri pada media spesifik. Hasil penelitian menunjukkan bahwa tanah dalam pot sebagai media penanaman stroberi bersifat masam, rendah kandungan karbon organik dan memiliki kapasitas tukar kation yang tinggi. Reaksi tanah cocok bagi tanaman stroberi, akan tetapi nilai konduktivitas elektrik terlalu rendah. Tanah media tanam stroberi mengandung CN ratio yang rendah hingga sangat rendah, akan tetapi mengandung nitrogen total yang tinggi dan P ₂ O ₅ dan K ₂ O yang sangat tinggi. Kandungan bakteri pelarut fosfat, <i>Azotobacter</i> pemfiksasi nitrogen, jumlah total bakteri dan jamur pada media tanam sangat rendah, akan tetapi jumlahnya meningkat pada tanah yang diperlakukan dengan pupuk kandang. Perbandingan jumlah mikroba pada tanah dan rizosfir hanya sekitar 1,0-1,25. Hasil penelitian ini memvalidasi bahwa pemberian bahan organik perlu ditambahkan, sementara pupuk kimia perlu diturunkan.
Kata Kunci: Chemical properties, Macronutrients, Manure, Microbial count	Strawberries in tropical West Java are cultivated intensively in potted soil but the substrate condition received less attention. The objective of this descriptive research was to evaluate the chemical and macronutrient profile in potted-soils for strawberry cultivation, and to determine beneficial microbes' population in root-free bulk soil and rhizosphere of potted-strawberry. The soil samples collected from 12-months and one-month-old potted strawberry; both contained Inceptisols soil order with 10 t/ha and 25 t/ha manure respectively. The proximate analysis was performed using Association of Official Analytical Chemists Method. The microbial enumeration was performed by Serial Dilution Plate Method in specific medium. The results showed that potted soils

were slightly acid, low in organic carbon and high in cation exchange capacity. The soil reactions were suitable for strawberry but the electrical conductivity was too low for strawberry growth. The potted soils have low to very low carbon to nitrogen ratio, but high in total nitrogen and very high in potential P_2O_5 and K_2O . The population of phosphate-solubilizing bacteria, nitrogen-fixing *Azotobacter*, total bacteria and total fungi in old potted soil were low; but its counts were increased in manure-treated potted soil. The ratios of bulk soil to rhizosphere microbial count were only 1.0-1.25. The study verified that the doses of organic matter should be increased, while decreasing the chemical fertilizer level.

INTRODUCTION

Subtropical-native strawberry (*Fragaria x Ananassa* Duch) introduced and cultivated in tropical area mainly in higher altitude (Asadpoor & Tavallali, 2015). Farmers in upland area of Bandung Regency grow strawberries in Inceptisols. In Indonesia, Inceptisols are developed from volcanic ash and basaltic andesite parent material; and has diverse texture from clay to sandy/silty loam (Muslim *et al.*, 2020). In general, Inceptisols have low organic Carbon (C), Nitrogen (N) and Phosphorous (P) concentration (Syamsiyah *et al.*, 2018; Yuniarti *et al.*, 2019; Muslim *et al.*, 2020). It is a common practice for farmers to cultivate strawberries in potted-soil with chemical fertilizer and animal manure in the open field. In order to reduce production cost, farmers utilized potted-soil for more than a year before they cultivate strawberry in another new potted soil.

Sufficient organic matter increased the soil quality and microbial population that is necessary for long term strawberry production (Medeiros *et al.*, 2015; Lovaisa *et al.*, 2017). Soil organic matters support the proliferation of heterotrophic microbes in soil and their function in macronutrient nutrient cycle (Hoorman *et al.*, 2010). Compost in strawberry cultivation increased plant height, canopy area and root weight in vegetative and generative stages (Madhavi *et al.*, 2020). Otherwise, soil bacteria or fungi promote C cycle and storage, soil organic matter accumulation (Gougoulas *et al.*, 2014; Malik *et al.*, 2016) and interact with plant exudate in the rhizosphere to provide plant nutrient (Jacoby *et al.*, 2017).

Diversity and population of soil microbe determine the soil health. The heterotrophic microbes have a prominent role in nutrient cycle mainly nitrogen and phosphorous as well as provide growth factor for plant development (Jacoby *et al.*, 2017). The N-fixing *Azotobacter* and P-solubilizing

bacteria (PSB) are well known soil beneficial microbe widely used as biofertilizer (Hindersah *et al.*, 2020; Nosheen *et al.*, 2021). The role of soil microbes on strawberry growth has been studied recently, and showed to improve plant growth, yield and quality (Rahman *et al.*, 2018; Todeschini *et al.*, 2018).

In West Java, strawberry productions were in general performed in potted soil containing soil and organic matter for 12 months. The farmer amended organic matter below recommended dose meanwhile they applied chemical fertilizer more than the recommended dose. Small group of farmers used potted soil with the same soil but with recommended dose of organic matter. However, those farmers start producing strawberries 1 month ago and will last up to 12 months. In order to improve the soil quality for strawberry production, the introduction study of soil properties related to the strawberry cultivation in both potted strawberry soil is needed. The objective of this quantitative descriptive research was to evaluate some important soil chemical characteristics related to Nitrogen (N), Phosphorus (P) and Potassium (K) availability in potted-soil of strawberries. Moreover, this study was aimed to count the population of soil beneficial microbes include *Azotobacter*, PSB as well as total bacteria and fungi in the soil samples.

MATERIALS AND METHODS

This study was conducted in Sugihmukti Village, Pasir Jambu District, Bandung Regency, West Java Province, Indonesia. Farmers grown strawberry cv. California in the outdoor potted-soil. The village located at 1,292 m above sea level with geographical position of 07°11'71.2" S and 107°25'84.1" E. The average of daily temperature and humidity were 20 °C and 80 % respectively. The soil sampling was carried out in August 2018. Soil chemistry and

microbiology data were analyzed by quantitative descriptive method.

Soil Samples

The soil samples were collected by composited method from strawberry potted soil in the study area. The potted soil were 12-months old potted soil and one-month old potted soil; the pot dimension was 50-cm in height and 35 cm in diameter (Fig 1.). The soil in all pot was Inceptisols taken up from the adjacent field. The 12-months old potted soil received approximately 100 g/pot of chicken manure

approximately 10 t/ha; while the one-month old pot incorporated by composted chicken manure equal to 25 t/ha. Four strawberry seedlings were grown in a pot. The chicken manure provided by local chicken farm contained 1.70% N, C:N 11.8, total P 2.52% and total K 1.95%. The farmer added the NPK compound fertilizer of 2-3 g/plant once in a month to old potted soil, while the newest potted soil received the same dose of NPK at the transplanting time. The 12-months old potted strawberry was still used for fruit production.



Figure 1. The 12-months old (left) and one month old (right) potted strawberry in local farmers area of Pasir Jambu District, Bandung Regency.

Root-free bulk soil and rhizosphere were taken up for beneficial microbe enumeration. In this study, the research subjects were 50 potted strawberries of 12-months old as well as one-month old potted strawberry, and the sampling pots were five for each age of potted strawberry. The farmers grown four strawberry plants in each pot so that a 25 g of bulk soil was collected from 5-cm depth of 4 sampling points near the plants in each pot and then mixed evenly. The samples from five pots were mixed and put in a sealed plastic bag; 100 g of bulk soil were stored at 4°C before microbial enumeration; while 400 g of bulk soil were put in room temperature for chemical analysis. The rhizosphere soil was collected from four plant roots in each sampling pots; all rhizosphere soil from 5 pots was then mixed and put in 4°C prior to microbiological analysis. The soil sample from each age of potted strawberry was subjected to chemical and microbiological analysis in triplicate.

Soil Chemical Properties Analysis

Organic C in soil was examined by Walkley and Black method with dichromate in an acidic condition as oxidizer. The Kjeldahl method with sulfuric acid was utilized to determine the total N

content that includes organic and inorganic N. Total P_2O_5 and K_2O in soil were determined by using HCl 25%. Exchangeable cations (Ca^{+2} , Mg^{+2} , H^+ , K^+ and Na^+) were determined by Ammonium Acetate method using flame photometer and atomic adsorption spectrophotometer. The cation exchange capacity (CEC) was determined by ammonium saturation method, using a 1 M solution of Ammonium Acetate at pH 7. Soil acidity (pH) was determined to measure the acidity of a soil suspension in ion-free water (1:1; v/v) using pH meter by potentiometric method. The determination of electrical conductivity (EC) was conducted with a conductivity cell in EC meter by measuring the electrical resistance of a 1:5 soil and water suspension. All the chemical analysis methods were referred to the Association of Official Analytical Chemists (AOAC) method for proximate analysis (AOAC, 2012).

Enumeration of Soil Microbes

Population of *Azotobacter*, P-solubilizing bacteria (PSB), total bacteria (TB) and total fungi (TF) were analyzed by Serial Dilution Method (Ben-David & Davidson, 2014) with specific medium (Table 1). All media were sterilized in the autoclave for 20 min

at 121 °C (1.02 atm pressure). Soil samples were diluted using sterilized 0.85% of sodium chloride p.a in aseptic environment. The count of viable microbes was estimated based on number of colonies in certain

dilution plate with three replication. Data average and standard error were then calculated from three replications of chemical as well as microbiological analysis.

Table 1. Composition of specific medium for soil microbial enumeration

Microbes	Specific Media	Composition in one liter
Azotobacter	Nitrogen-free	10 g mannitol, 0.2 g KH ₂ PO ₄ , 0.2 g MgSO ₄ .7H ₂ O, 0.2 g NaCl,
	Ashby's agar	0.1 g CaCO ₃ , 10 mg Na ₂ MoO ₄ 15 g agar, pH 7.0 ±0,2
Phosphate Solubilizing bacteria	Pikovskaya agar	10 g glucose, 5 g Ca ₃ (PO ₄) ₂ , 0.5 g (NH ₄) ₂ SO ₄ , 0.2 g NaCl, 0.1 g MgSO ₄ .7H ₂ O, 0.2 g KCl; 0.5 g yeast extract, 0.002 g MnSO ₄ .H ₂ O, 0.001 g FeSO ₄ , pH 7.0 ±0,2
Total bacteria	Nutrient agar	5 g peptone, 2 g yeast extract, 5 g sodium chloride, 15 g agar, pH 7.0 ±0,2
Total Fungi	Potato dextrose agar	Infusion from 200 g potato, 20 g dextrose, 0.1 g chloramphenicol, 15 g agar, pH 5.6±0,2

RESULTS AND DISCUSSION

Soil Chemical Properties

All soils were slightly acid in pH, low in EC and average in CEC (Table 2). The acidity of newest potted soil that treated with higher dose of manure application increased up to 6.5 compared to old potted soil. All soil had EC below 1 dS/m (Table 2) indicated that they were non-saline soil. However, the EC of both potted soils were low. The CEC of 12-months old potted-soil was slightly lower than CEC of new potted soil. Higher level of chicken manure amendment in newest potted soil was slightly increased the CEC compared to the old potted soil with low doses of chicken manure.

The pH of all soils was slightly acid which is in accordance with strawberry requirement. The strawberry prefers to grow in pH level of 5.5-6.8. Optimal pH for strawberry is 6.2 that ensure the optimal uptake of P, but they enable to grow in soil with pH of 5.4 (Trejo-Téllez and Gómez-Merino, 2014). The result showed that organic matter amendment increased soil reaction to near neutral. This agrees with the soil pH enhancement after mixing the soil with poultry manure (Islam *et al.*, 2021). Organic acid from organic matter fermentation can chelate Aluminum and then increase of soil reaction (Opala *et al.*, 2012).

Table 2. The acidity, electrical conductivity and cation exchange capacity of potted soil as substrate for strawberry production.

Soil samples	pH	EC (dS/m)*	CEC (cmol/kg)
12-months old potted soil	5.64 ± 0.03	0.37 ± 0.051	24.43 ± 0.26
One-month old potted soil	6.55 ± 0.02	0.61 ± 0.056	28.44 ± 0.23

Mean ± standard errors were calculated from three replications. The soil nutrient status based on Indonesian Soil Research Centre (PPT, 1995). *0-2 ds/m = non saline, 2-4 ds/m = low salinity, 4-8 ds/m = mild salinity, 8-16 ds/m = high salinity and > 16 = severe salinity.

The study revealed that soil EC are < 1. The growth substrate with EC < 1 decreased the fruit production (Depardieu *et al.*, 2016) which is supposed due to low nutrient sufficiency for optimal strawberry growth and their productivity. Increased of EC was clear in potted soil that received intensive fertilizer due to increase of ionic nutrient (do Carmo *et al.*, 2016). The effect of EC on marketable yield of strawberry has been studied (Suarez & Grieve, 2013). They found that yield of strawberry cv. Camarosa

decreased up to 43% per plant once the EC of irrigation water was higher than 0.6 dS/m; but Ventana variety yield started to decreased only when the irrigation water salinity was up to 1.71 dS/m.

The CEC of manure-amendment potted soil was slightly increased which confirm that organic matter contributes to the amount of negative charge of soil. These anionic charges hold and exchange the cations in soil solution. Soil reaction, EC and CEC are major soil physicochemical indicators that influence

nutrients solubility. Soil pH and EC can predict nutrient availability for root uptake and biological activities within the soil. The CEC is important soil indicator since it represents cation supply such as Ca^{+2} , Mg^{+2} , K^+ , Na^+ and NH_4^+ for plant and prevent cationic nutrients (Sidi *et al.*, 2015). In order to increase the CEC, intensive organic matter application is needed.

Macro-nutrients profile

The study demonstrated a different macronutrient properties between two potted soils (Table 3). The organic-C in 12-months-old potted soil was clearly lower than potted with higher dose of manure. The total N in one-month old potted soil was higher than the old pots. The ratio of C to N (C:N) of both potted soil was low but C:N was enhanced in soil with 25 kg/ha of chicken manure. The clear increase of organic C in new potted soil resulted in the increase of C:N. The total N, potential P_2O_5 as well as K_2O in both potted soil were comparable.

Inadequate organic matter amendment practice resulted in low organic C (Table 2). The C:N

of potted soil increased after manure amendment although it was still lower than C:N of normal soil. The C:N of all soil also less than 7 showed insufficient organic C for microbial metabolisms. High N is caused by the accumulation of fertilizer residue during previous strawberry cultivations. However, low C:N in soil limits proliferation of bacteria and fungi. The average ratio of C to N of bacteria as described elsewhere is 4:1 – 8:1 and C:N fungi is 17.23-18.71 (Zhang & Elser, 2017). The farmers in study area applied 2-3 g NPK fertilizer per plant once in a month by adding the fertilizer in a hole near the stem. Intensive chemical fertilizer (NPK or/urea application) during strawberry production in potted soil resulted in the higher rate of total N, P and K in soil. Chemical fertilization without proper amount of organic fertilizer reduced organic C content and the C:N ratio become very low. Organic matter application in strawberry significantly increased plant growth and fruit number in individual plant (Madhavi *et al.*, 2021).

Table 3. Some nutrient profile of potted soil for strawberry production

Soil samples		Organic-C (%)	Total-N (%)	C:N	Potential P_2O_5 (mg/kg)	Potential K_2O (mg/kg)
12-months old potted soil	old	1.19 ± 0.04 Low*	0.55 ± 0.03 High	2.16 ± 0.15 Very low	57.38 ± 0.82 High	85.24 ± 8.11 Very High
One-month old potted soil	old	4.65 ± 0.05 High	0.72 ± 0.07 High	6.65 ± 1.49 Average	50.21 ± 0.42 High	83.00 ± 7.89 Very High

Mean $\pm$ standard errors were calculated from three replications. The soil nutrient status based on Indonesian Soil Research Centre (PPT, 1995).

Microbial Population

The microbial populations were varied depend on microbial group and source of soil samples (Table 4). Microbial population in the rhizosphere is slightly higher than in root-free bulk soil. The population of N-fixer *Azotobacter* in any soil samples was clearly lower than phosphate solubilizing bacteria and the count of total bacteria was higher than total fungi. The population of PSB in old potted soil was lower than one-month old one but the total fungi was vice versa (Table 4). The ratio of rhizosphere to bulk soil microbial population (R:S) was >1 (Table 5). It showed that the development of microbes in the rhizosphere was slightly higher than in bulk soil. This study showed that the R:S of total bacteria and fungi in manure-amendment potted soil were enhanced.

Low organic C in soil resulted in low population of beneficial microbes in both bulk soil

and rhizosphere although organic manure increased their population. The average population of bacteria and fungi in soil are 10^5 – 10^6 and 10^8 – 10^9 CFU/g respectively (Hoorman & Islam, 2010). *Azotobacter* population in soil was reported between 10^3 – 10^4 CFU/g (Maurya *et al.*, 2012; Widiastuti *et al.*, 2016) while PSB count in soil was ranging from 10^3 – 10^5 CFU/g (Ndung'u-Magiroy *et al.*, 2012; Rfaki *et al.*, 2018). Nonetheless, fungal count in manure-amendment soil was average. Heterotrophic microbes are depending on soil organic matter for their energy and C. The low rate of organic matter application is supposed to relate with low beneficial microbes count in soil.

Soil microbes depend on nutrient availability in soil for their metabolic process. Despite high N, P and K content in soil, a prominent increase of microbial count in the rhizosphere over bulk soil was

not found. The imbalance composition of C, N, P and K in soil might inhibit the root and shoot growth, and reduced the quality and quantity of root exudates in the rhizosphere. Low C:N can reduce the population of heterotrophs since the microbes needs sufficient

content of organic matter for their proliferation. The low organic C and C:N ratio, and excess N, P and K in soil possibly cause improper strawberry growth and yield.

Table 4. Soil beneficial microbial counts of bulk soil and rhizosphere of strawberry grown in potted soil

Soil samples	Microbial population (log10 of CFU/g)			
	Azotobacter	PSB*	Total Bacteria	Total Fungi
12-months old potted soil				
Bulk soil	2.53 ± 0.07	3.47 ± 0.12	5.66 ± 0.10	3.48 ± 0.05
Rhizosphere	2.92 ± 0.03	3.94 ± 0.22	5.73 ± 0.06	3.51 ± 0.07
One-month old potted soil				
Bulk soil	2.56 ± 0.03	4.55 ± 0.19	4.78 ± 0.10	4.67 ± 0.04
Rhizosphere	2.74 ± 0.06	4.81 ± 0.04	6.0 ± 0.03	5.75 ± 0.09

*PSB, Phosphate Solubilizing Microbes. Mean ± standard errors were calculated from three replications.

Table 5. The microbial population ratio in the rhizosphere to bulk soil (R:S) of strawberry potted soil

Soil samples	<i>Azotobacter</i>	PSB*	Total bacteria	Total fungi
12-months old potted soil	1.16 ± 0.04	1.12 ± 0.03	1.01 ± 0.009	1.01 ± 0.03
One-month old potted soil	1.07 ± 0.03	1.06 ± 0.02	1.25 ± 0.008	1.23 ± 0.03

*PSB, Phosphate Solubilizing Microbes. Mean ± standard errors were calculated from three replications.

The farmers in study area applied approximately 2-3 g NPK fertilizer/plant every month. In total, the 12-months old potted soil have been treated with 24-36 g/plant NPK. Suppose that in the field, the planting distance is 30 x 30 cm; so NPK dose for one plant is 2.7 g. The Urea 200 kg/ha recommended by Indonesian Agricultural Ministry is used in soilless strawberry cultivation (Hindersah et al., 2021). A total of 2 g/plant of NPK (225 kg/ha of NPK) is applied during vegetative stage (Palupi et al., 2017). Meanwhile, Astuti *et al.*, (2015) suggested 12 g/plant of NPK (16:16:16) in four times application. In comparison, the research in India verified that the best dose of N, P and K fertilizer to increase the strawberry yield is N,P, and K of 70, 80 and 80 kg/ha respectively. It is clear that farmer's potted soil in study area received too much fertilizer. In current study, the acidity, organic-C, total-N and C:N of 1-months potted soil were higher than 12-months old potted soil. The increase of organic-C is prominent for the activity of N-fixer bacteria and PSB which is in turn enhance the availability of N and P for plant uptake.

CONCLUSION

The study verified that the reaction of potted soil was slightly acid which is agree with the strawberry requirement. All soils were non saline and have medium to high cation exchange capacity. The increase of organic C was only showed by potted soil received significant amount of chicken manure. High level NPK compound fertilizer resulted in high N, P, K level in soil. Low population of *Azotobacter*, P-solubilizing bacteria, total bacteria total and fungi were recorded in old potted soil due to low C:N. Nonetheless, manure amendment increased the total bacteria and fungi. Microbial population in the rhizosphere were likely similar to their density in the rhizosphere, resulted in the ratios of bulk soil to rhizosphere microbial count (R:S) about 1.0-1.25. This study showed that potted soil treated with 25 t/ha chicken manure had better nutrient profile than 12-months old soil especially the pH, organic-C, total-N and C:N. In order to improve the chemical properties of potted soil quality, the farmers need to raise the level of organic matter amendment and reduce the dose as well as timing and frequency of chemical fertilizers.

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