Utilization of slurry and mushroom baglog to improve growth and yield on strawberry on degraded volcanic soils

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Abstract

Utilization of slurry under the combination with mushroom baglog could be used a potential source for replacing nitrogen fertilizer and improve soil fertility. The purpose of this study was to evaluate the different combination and the level of slurry application combining with mushroom baglog on total soil nitrogen and organic contents, total soil bacteria and fungi, along with the improvement of growth and yield of strawberry (Fragaria x ananassa). The experiment used a completely randomized design with five treatments and five replications. The treatments were A1 (100% inorganic fertilizer application as control), A2 (50% composted slurry and mushroom baglog + 50% in-organic fertilizer), A3 (100% composted slurry and mushroom baglog), A4 (150% composted slurry and mushroom baglog), A5 (50% composted slurry and mushroom baglog + 50% in-organic fertilizer), A6 (100% fresh slurry (uncomposted)), and A7 (150% uncomposted fresh slurry). Results showed that the treatments significantly affected total soil nitrogen, total soil bacteria and fungi, along with the growth and yield of strawberry. The A6 treatment, which used 100% uncomposted fresh slurry, showed the highest total nitrogen in the soil (0.23%) and the total population of bacteria (7.1 log CFU/g) and the greatest number of strawberry stolons. In term of the number of leaves and total soil fungi, the A3 treatment was the greatest, resulted in number of leaves and total soil fungi at 19.7 per plant and 4.8 log cfu/g x 10⁴. However, the best yield was obtained from the A7 treatment (150% of fresh slurry) at 15.1 kg/ha.

Keywords:
contaminant
crop management
nutrient balance
organic manure
soil fertility

Introduction

Organic farming is a common agricultural practice to sustain crop production, especially on fruits and vegetables, avoiding the use of pesticides, herbicides, chemical fertilizers (Asami et al., 2003). Pollution is becoming a serious problem in many soils, affected by the rising of their intensive use of pesticides that lead to polluted agricultural products, whilst in the long period, it will cause a serious health problem in humans (Abu-Zahra and Tahboub, 2008). Nowadays, many customers prefer to consume less residue of pesticide on fruits and vegetable products. Strawberry (Fragaria virginiana), which belongs to the family Rosaceae, is the most delicious, delicate flavoured, refreshing, and attractive red fruit of the world, attracted many consumers, and it has been used for many beneficial products or as food ingredients (Sharma, 2002; LaMondia et al., 2002). It is native to most of the Northern hemisphere, including Europe and Great Britain. It is nutritious, quick growing and becoming one of the exotic fruits in Malang, and it has
been suitable for cultivating in many districts. However, to achieve a high yield and maintain optimum productions, farmers likely depend on the high rate application of inorganic fertilizer such as NPK, and it is almost impossible not to use high doses of pesticide and chemicals to avoid the pest and disease.

The area of the southern slope of Arjuno-Welirang in Malang is dominated by volcanic soils, mainly Andisols and Inceptisols. The soils are mainly used for cultivating horticultural crops, including strawberry, which those productions are distributed to local and regional markets. As a result of the frequent use of pesticides and inorganic fertilizers, the soil nutrients become unbalance, and some of the nutrients are lower than their sufficient levels for plant growth. The soils are also vulnerable to erosion as the slope is in a steep position, and the intensity of rainfall is high in a certain period of months. Both processes will reduce soil fertility and potentially leach nutrients to the deeper soil layer causing degradation of soil chemical, physical and biological properties. For anticipating this problem, the use of organic material is essential to reduce the effects of those residues (Grandy et al., 2002). Unlike inorganic fertilizers, organic manures have sustained effects on soil chemical properties and consequently crop yield. It is an excellent source of nutrients, and it could maintain high microbial population activities (Gutser et al., 2005). The sweetness of strawberry is a major problem in Malang. Organically grown strawberry increase total soluble solids content, hence produce high-quality fruit with a sweeter taste, longer shelf life (by thickening fruit peel) and better flavour (Reganold et al., 2010). Several research workers reported that nitrogen application could increase the population of soil microorganisms and nutrients uptake on strawberry plants (Di and Lin, 2014; Onwosi et al., 2017). The combined effect of strawberry cultivars and organic manures has been reported to have a significant effect on all the vegetative growth parameters such as plant height, number of leaves and runners per plant (Rashid, 2018). Organic material has been reported to improve degraded soil (Prayogo and Ihsan, 2018) and soil fertility (Arfarita et al., 2019).

Slurry is one of the major sources of liquid organic materials resulted from anaerobic fermentation in the biogas process as a residue which is unfortunately rarely used for replacing solid manure (Kumar et al., 2015a; Kumar et al., 2015b). Hu et al. (2015) reported that slurry comprises 24.6% organic C; C/N at about 7.4; pH at about 7.13; and total nitrogen of 3.49%. According to those properties, slurry can be used as a source of organic and nitrogen fertilizer, either directly or composted (Awasthi et al., 2017). The utilization of slurry as a potential source of nitrogen is rarely informed, and it has been flushed directly to the river or stream after biogas processes being finished. On the other hand, mushroom baglog as a solid material which resulted from the excess of mushroom cultivation media is neglected since it may contain anti-fungal agent or chemicals, but it has been successful in improving sweet corn production (Sihaloho and Meriaty, 2019). Mushroom baglog can be used as a compost mixture. This material contains high levels of carbon, nitrogen, phosphorus and potassium (Meng et al., 2018a, Meng et al., 2017). The results of research by Meng et al. (2018b) indicated that baglog contains 38.942% organic C, 2.041% N, 19.08 C/N, 2.383% P, and 2.406% K. Baglog contains low water content, so it is used as a composting mixture for bio-slurry which has high humidity (90%). In addition, baglog has enzymes such as cellulases, hemicellulases, proteases and laccases, which can accelerate the degradation of organic matter in composting (Fang et al., 2016; Meng et al., 2017). According to research by Meng et al. (2018b), composting with a mixture of slurry and mushroom media waste products (baglog) can reduce nitrogen loss and improve the quality of compost products.

The combination of the use of slurry and mushroom baglog as organic sources to improve Strawberry production has never been tested. Application of slurry and mushroom baglog are hypothesized to affect total soil nitrogen and, soil bacteria and fungi along with improving crop growth and yield. The application of slurry under a combination of mushroom baglog can be an alternative farming practice in degraded volcanic soils to recover and maintain soil fertility in a sustainable manner. However, limited work has been done on the single and combined application of organic manures under different strawberry cultivars. The present study was, therefore, undertaken to elucidate the optimization of growth, yield and quality of strawberry cultivars through organic farming in degraded volcanic soils.

**Materials and Methods**

The research was conducted at the experimental field of the Faculty of Agriculture, Brawijaya University in Donowarih-Karangploso, Malang, from August to December 2019, followed by laboratory analysis at the Soil Science Department of Brawijaya University for the next following year. Different analytical methods were used to determine soil pH, soil N, soil C and organic carbon, soil P and K, CEC and total soil bacteria and fungi. Soil pH was determined with 1:1 soil water suspension. Organic matter reaction was determined with 1:2.5 compost water suspension (Fieldes and Perrott, 1996). Nitrogen total was analyzed using the Kjeldahl method. The samples were distilled and titrated with H$_2$SO$_4$, 0.01 N (Kjeldahl, 1883). Organic carbon content was determined using the Walkley and Black method. The samples were titrated with FeSO$_4$ $\cdot$ 7H$_2$O (USDA, 1982). Analysis of soil potassium was carried out using NH$_4$OAc 1N at pH 7 solution. The value was recorded by a flame photometer. Analysis of
exchangeable potassium was carried out using 5 mL HNO₃ + 5 mL HClO₄ solution. The value was recorded by using a flame photometer (Chhabra et al., 1975). Soil phosphorus content was determined using 5 mL HNO₃ + 5 mL HClO₄ solution, and the value was recorded by using a spectrophotometer at a wavelength of 410 nm (Bray and Kurtz, 1945). CEC was determined using NH₄OAc 1N at pH 7 solution (Houba et al., 1988). The soil used for this study has the following characteristics: pH = 6, total N = 0.11%, total K = 1.05%, organic C = 1.84%, soil organic matter = 1.84%, exchangeable K = 0.77 me/100 g, P₂O₅ = 12.18 ppm, and CEC = 45.08 me/100 g. The fresh slurry used for this study has the following characteristics: pH = 8, organic C = 3%, soil organic matter = 5.19%, total N = 1.05%, K₂O = 12%, and P₂O₅ = 1.57%. The characteristics of the composted slurry and mushroom baglog are as follows, pH = 7, organic-C = 19.07, organic matter = 32.8%, total N = 1.26%, K₂O = 0.6%, and P₂O₅ = 0.037%.

The nutrient composition of fresh and composted slurry and mushroom baglog fulfilled all minimum requirements for the basic nutrient composition of materials for producing organic fertilizer (Ministry of Agriculture, 2019). The total population of bacteria and fungi analysis was carried out using the pour plate method. Nutrient agar (NA) medium was used for bacteria, and potato dextrose agar (PDA) medium was used for fungi (Pochon et al., 1969). Crop parameters observed were plant length, number of leaves, number of stolons, fruit weight, yield, fruit brix, and nitrogen uptake that were collected after 12 weeks after planting. Plant length was measured using a metal ruler, number of leaves and number stolon were counted using a hand counter. Fruit weight was measured using digital balance, and yield per hectare was estimated from the total production per plots, whilst nitrogen uptake was determined using a leaf sample from each treatment before analyzing it using the Kjeldahl method (Kjeldahl, 1883).

Experimental design and data analysis

The treatments tested were A1 (100% inorganic fertilizer), A2 (50% compost + 50% inorganic fertilizer), A3 (100% composted slurry and mushroom baglog), A4 (150% composted slurry and mushroom baglog), A5 (50% composted slurry and mushroom baglog + 50% inorganic fertilizer), A6 (100% fresh slurry), A7 (150% fresh slurry). The seven treatments were arranged in a randomized block design with four replications. Healthy seedlings of strawberry cultivars that were collected from a nursery in the Indonesian Citrus and Subtropical Fruits Research Institute (ICSFRI) of Tlekung-Malang were acclimatized for a week before transplanting to the field. The seedlings of uniform size were transplanted on 14 September 2019 at a spacing of 40 cm × 50 cm. Each unit plot was 2.5 m × 3.2 m in size, which consisted of 35 plants each. The spacing of plot to plot was 50 cm, and block to block was 50 cm. Different organic manure treatments (inorganic fertilizers, composted slurry + mushroom baglog, and fresh slurry) were applied to each plot at 28 days before planting of seedlings and then it was carefully incorporated into the soils. After transplantation, irrigation, weeding and plastic mulching were done when necessary. Data obtained were subjected to the analysis of variance (ANOVA) using licenced Genstat version 18 followed by the Duncan's Multiple Range Test at 5% significant level. Multivariate analysis Biplot was adopted to discover the relationship amongst parameter observed (Prayogo et al., 2021).

Results and Discussion

Total soil nitrogen

Applications of fresh slurry, composted slurry, and mushroom baglog significantly affected total soil nitrogen (Figure 1). The highest total soil nitrogen of 0.22% was detected at the A6 treatment (100% fresh slurry). This value was significantly different from other treatments except with the A2 treatment (50% of composted slurry and mushroom baglog + 50% of inorganic fertilizer).

![Figure 1. Effect of the application of fertilizer, slurry, mushroom baglog on total soil nitrogen. A1 = 100% inorganic fertilizer, A2 = 50% composted slurry and mushroom baglog + 50% inorganic fertilizer, A3 = 100% composted slurry and mushroom baglog, A4 = 150% composted slurry and mushroom baglog, A5 = 50% fresh slurry and 50% inorganic fertilizer, A6 = 100% fresh slurry, A7 = 150% fresh slurry. The lowest total soil nitrogen of 0.16% was discovered at the A1 treatment (100% inorganic fertilizer). Liu (2015) revealed that the application of organic material could increase total soil nitrogen and microbial activity, which is related to the nitrogen mineralization process, which, by organic matter, produces organic acid and organic nitrogen being released to soil (Fließbach et al., 2007). The ability of soil to provide nitrogen can be relied on by the condition and the amount of organic matter which is related to the rate of
mineralization of organic matter. According to the research of Tagliavini et al. (2005), nitrogen in strawberry plants (Fragaria x ananassa) can increase leaf area, plant growth and affect the fruit ripening process. Application of nitrogen fertilization of 91 kg N/ha resulted in nitrogen uptake in strawberry plants of 40% per plant (Tagliavini et al., 2005). Meanwhile, Muramoto et al. (2014) reported that nitrogen uptake in strawberry plants occurs in the mid-vegetative phase at 36 to 120 days after sowing. Nitrogen application of 255 kg/ha resulted in nitrogen uptake of 120 grams per plant and strawberry plant production of 38.7 t/ha. The application of nitrogen to strawberry plants in accordance with the plant needs can increase the growth and yield of strawberry plants. If the nitrogen applied to strawberry plants is insufficient, it will obstruct the strawberry plant growth process. This nitrogen deficiency causes stunted growth, the appearance of chlorosis on old leaves and red and purple spots on the leaves, limiting lateral shoot growth. However, excessive nitrogen application will have a negative impact on plant growth, and the leaves will turn dark green. The need for nitrogen in strawberry plants (Fragaria x ananassa) is in accordance with the research of Hart et al. (2000) about 67 kg N/ha.

Microbial nitrogen cycle processes play roles in organic and inorganic nitrogen stocks affecting plant uptake, leaching, immobilization of N microbes and denitrification. Some of the nitrogen is lost from the biological cycle by means of a denitrification process. Denitification is a process of reducing nitrate to nitrogen gas. Nitrate (NO3−), which is used as an alternative electron acceptor in anaerobic respiration, is reduced to nitrogen gases (N2, NO, or N2O). The bacteria that play roles are Bacillus, Paracoccus, and Pseudomonas (Robertson et al., 2009).

Application of fresh slurry or composted slurry and mushroom baglog showed no significant effect (P>0.05) on nitrogen uptake in strawberry (Figure 2). This result was in the opposite finding by Yavari et al. (2009), who reported that the application of organic matter such as residual organic waste and inorganic fertilizers could increase the nitrogen uptake in strawberry plants. According to Nestby et al. (2005), factors affecting nitrogen uptake by plants are root density, root distribution, soil pH. Plants usually absorb nitrogen in the form of N-NH4+ and N-NO3− to support respiration, photosynthesis and crop metabolism (Leghari et al., 2016).

**Total population of soil bacteria and fungi**

Unlike nitrogen uptake, the application of fresh or even composted slurry and mushroom baglog significantly affected the total population of bacteria. The highest total soil bacteria population was found at the A5 treatment (50% of fresh slurry and 50% inorganic fertilizer), which was not significantly different to other treatments except with the A1 treatment (100% inorganic fertilizer) (Figure 3a).

This result was similar to that has been reported by Nasseri et al. (2010) that the application of organic materials in the form of slurry compost increased total soil bacteria. The treatments also significantly affected (p<0.05) the total population of soil fungi. The total fungi population in the soil was 4.4 log cfu/g under the treatment of A1 (100% inorganic fertilizer) as the lowest. This value was significantly lower (p<0.05) than other treatments, whilst the highest soil fungi population was detected at A3 (100% composted slurry and mushroom baglog) to about 4.8 log cfu/g (Figure 3b).

![Figure 2](image)

**Treatments**

Figure 2. Effect of the application of fertilizer, slurry, mushroom baglog on nitrogen uptake by plant. A1 = 100% inorganic fertilizer, A2 = 50% compost slurry and mushroom baglog + 50% inorganic fertilizer, A3 = 100% composted slurry and mushroom baglog, A4 = 150% composted slurry and mushroom baglog, A5 = 50% fresh slurry and 50% inorganic fertilizer, A6 = 100% fresh slurry, A7 = 150% fresh slurry.

According to Feng et al. (2011), microbes contained in slurry are cellulolytic microbes that are useful for composting, nitrogen-fixing microbes, phosphate-solubilizing microbes, Lactobacillus sp microbes which play a role in controlling soil-borne disease attacks. Whereas in the study of Hong-Yan et al. (2013), slurry contains several microbes such as Bacillus sp, Clostridium sp, Psychrobacter sp, Pseudomonas sp, Acetomicrobium sp, Sphingobacterium composti and Clostridium sp. Microbes Bacillus sp. is a microbe that plays a role in composting. Clostridium sp. and Psychrobacter sp is a role in the decomposition of cellulose. Composting processes could improve nutrients on decomposable organic matters which have benefits to improve crop yield (Sanchez et al., 2017). In addition, the research of Nassium et al. (2010) showed that slurry application could increase total bacteria by 7 × 107 cfu/mL, which has the potential as bioremediation in removing PAH (polycyclic aromatic hydrocarbons) from contaminated soil. This value was higher than the total bacteria or fungi in this study. In addition, the study
by Liu et al. (2015) indicated that the application of bioorganic (compost with a slurry mixture) could increase soil pH, electrical conductivity, organic carbon, NH$_4^+$, NO$_3^-$ and available K content, microbial activity and microbial carbon biomass. In this study, the application of organic input successfully increased total soil bacteria and fungi, which play an important role in supporting biochemicals processes in soil.

Crop growth
The treatments significantly affected crop length (Table 1). Under the A3 treatment (100% composted slurry and mushroom baglog), the lowest crop length of 15.48 cm was 3 cm lower than the highest crop length of 18.07 cm at the A5 treatment (50% fresh slurry and 50% inorganic fertilizer). A different pattern was detected in the number of leaves, whereas the greatest number of leaves was discovered under the A3 treatment (100% composted slurry and mushroom baglog), which was significantly different from other treatments except with the A7 treatment (150% of fresh slurry) as the lowest. In term of the number of stolons, the A6 treatment showed the highest number of plant stolons. This was closely related to the greatest abundance of soil nitrogen at about 6.38 that was significantly different from the A2 and A3 treatments. The increase of plant length is usually influenced by the availability of nitrogen in the slurry (Wentzel et al., 2016), which contains amino acids, fatty acids, organic acids, humic acids, auxin, and cytokinins, which affect the vegetative growth process and plant productivity. Generally, soil nitrogen plays an important role in strawberry plants for increasing plant growth (Tagliavini et al., 2005). In this study, however, the raising of soil nitrogen just only affected the number of stolons. Andriolo et al. (2011) explained that nitrogen application to strawberry plants increased the number of leaves in strawberry plants.

Table 1. Effect of the application of fertilizer, slurry, mushroom baglog on vegetative crop growth (crop length, number of leaves, and number of stolons).

| Treatments | Crop Length (cm) | Number of Leaves | Number of Stolons |
|------------|------------------|------------------|-------------------|
| A1         | 16.52$^{b,ab}$ ± 0.24 | 14.0$^{b}$ ± 1.313 | 5.82$^{ab}$ ± 0.793 |
| A2         | 17.58$^{ab}$ ± 1.076 | 11.4$^{ab}$ ± 1.424 | 4.20$^{ab}$ ± 0.247 |
| A3         | 15.48$^{a}$ ± 0.083 | 19.7$^{b}$ ± 0.901 | 4.08$^{a}$ ± 0.370 |
| A4         | 15.98$^{ab}$ ± 0.865 | 10.7$^{ab}$ ± 1.321 | 4.35$^{ab}$ ± 0.507 |
| A5         | 18.07$^{b}$ ± 0.911 | 13.9$^{b}$ ± 2.362 | 4.78$^{ab}$ ± 0.439 |
| A6         | 17.68$^{ab}$ ± 0.996 | 14.3$^{b}$ ± 0.885 | 6.38$^{b}$ ± 0.877 |
| A7         | 17.48$^{ab}$ ± 1.217 | 12.9$^{a}$ ± 0.846 | 5.51$^{ab}$ ± 0.322 |

Remarks: A1=100% in-organic fertilizer, A2= 50% composted slurry and mushroom baglog + 50% inorganic fertilizer, A3=100% composted slurry and mushroom baglog, A4= 150% composted slurry and mushroom baglog, A5= 50% fresh slurry and 50% inorganic fertilizer, A6= 100% fresh slurry, A7= 150% fresh slurry. The numbers followed by the same letters in the same column are significantly different at the Duncan test of 5% ($p=0.05$); ± indicated of SEM (standart error of means) ($n=4$).
Crop yield

The fruit weight of the strawberry was significantly affected by the treatments (Figure 4.). The highest yield of 15,100 kg/ha was obtained under the A7 treatment (150% of fresh slurry), which was almost two times higher than the lowest strawberry fruit weight at the A1 treatment. The yield was lower than those of the combined application of cow dung + MOC + poultry manure that gave the maximum yield of 19.14 t/ha (Rashid, 2018). Similarly, Huan-Tao et al. (2008) reported that the application of slurry could increase fruit quality, such as heavy fruit on plant strawberries of 13 grams. This is related to content in the slurry are amino acids and nitrogen, which affected the yield of fruit weight. Nestby et al. (2005) and Khalil et al. (2017) reported that the application of nitrogen nutrients through organic and inorganic fertilization affected the quality of strawberry fruit. The application of organic matter affects the growth and yield of strawberry plants. This corresponds to Kumar et al. (2018) that the application of organic materials in the form of cow dung compost of 8.5 t/ha can increase vegetative growth. The nitrogen content in cow dung compost is 1.3% (Kumar et al., 2018). While research done by Khalil and Agah (2018) showed that application of a combination of chemical fertilizer 50% + compost + biological fertilizer could increase leaf area by 529.66 cm²/plants, the number of leaves by 28, the number of fruit 34 and crop yields 367.24 g/plant. In addition, the application of a combination of 50% chemical fertilizers + compost + biological fertilizers resulted in a nitrogen absorption content of 2.36%. This content was higher than other treatments. This is due to an increase in nutrients in the root biosphere (Khalil and Agah, 2018). In line with Odongo et al. (2008), the application of cow dung compost of 5.4 t/ha increased the growth and yield of strawberry plants. According to Tagliavini et al. (2005), nitrogen in strawberry plants can increase leaf area, plant growth and affect the fruit ripening process. Increased growth and yield of strawberry plants is caused by photosynthetic production.

Figure 4. Effect of the application of fertilizer, slurry, mushroom baglog on crop yield. A1=100% inorganic fertilizer, A2=50% composted slurry and mushroom baglog + 50% inorganic fertilizer, A3=100% composted slurry and mushroom baglog, A4=150% composted slurry and mushroom baglog, A5=50% fresh slurry and 50% inorganic fertilizer, A6=100% fresh slurry, A7=150% fresh slurry.

The presence of phosphorus and nitrogen content in cow dung compost can increase shoot dormancy and increase the flowering site. In addition, the application of organic matter can increase the generative phase; this is due to the increase in the vegetative phase, which is influenced by the synthesis of chlorophyll due to photosynthesis and assimilation of carbon dioxide, which causes an increase in fruit quality. The growth performance and yield of strawberry amongst different treatment of this study are presented in Figure 5.

Figure 5. Visual growth performance and yield of strawberry cultivated on degraded volcanic soils.
Fruit brix

Application of fresh slurry or even composted slurry and mushroom baglog did not significantly affect the fruit brix on strawberries (Figure 6). Fruit brix is the level of sweetness indicated by fructose or glucose contents, representing the quality performance of the fruits. The average fruit brix observed at the A1 treatment was similar to the A5 treatment. Brix contains sucrose, glucose, fructose, vitamins, minerals, amino acids, proteins, hormones, and other solid substances in the fruit of plants (Harril, 1998). Factors affecting the value of strawberry fruit brix are cultivars and the environment, such as temperature, light, air, and plant nutrients (Arancon et al., 2004). Application of organic matter such as slurry in this study could not increase the brix value in strawberry plants except it was combined with inorganic fertilizer. This is the opposite with the finding from Federico et al. (2007), who reported that the application of organic materials increased levels of organic acids (malic and citric acids), sugar levels (fructose, glucose, and total sugar) of crops.

Relationship between parameters

Though the principal components analysis is important to determine the relationship between parameter and understanding their magnitude and direction, PCA Biplot was adopted (Figure 7). It was verified that the first principal component analysis axis (PC1) responded to 95.31% of the total observed parameter (total soil N, number of leaves, soil fungi, and fruit brix) and had similar magnitude and direction (upper position). PC1 was accounted mostly for the above parameters. The soil bacteria, number of stolons, and crop length were in the other direction and magnitude (bottom position) (Figure 7).

Conclusion

Application of fresh slurry or composted slurry and mushroom baglog affected total soil nitrogen total population of soil bacteria and fungi, crop length, number of leaves, and strawberry yield. Each treatment had a different effect on the soil environment and crop performance. However, there was an indication that the application of fresh slurry is better than the composted material in terms of crop yield performance. For sure, the addition of organic input had a better effect on soil chemical and biological properties of soil compared to the application of inorganic fertilizer alone. The relationship among parameter could be detected using multivariate analysis, which indicated that soil N had a linear relationship with number of leaves, soil fungi, and fruit brix.

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