ASSESSMENT OF GREEN MANURING POTENTIAL OF LEGUMINOUS WEEDS OF MIZORAM

A THESIS SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF DOCTOR OF PHILOSOPHY

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ASSESSMENT OF GREEN MANURING POTENTIAL OF LEGUMINOUS WEEDS OF MIZORAM

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Submitted

In partial fulfillment of the requirement of the Degree of Doctor of Philosophy in Forestry of Mizoram University, Aizawl.



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CERTIFICATE

This is to certify that the thesis entitled "Assessment of green manuring potential of Leguminous weeds of Mizoram" submitted by Miss Jyoti Jopir for the award of degree of Doctor of Philosophy in Forestry of Mizoram University, Aizawl, embodies the record of original investigation carried out by her under my supervision. She has duly registered and the thesis presented is worth of being considered for the award of the Ph.D. degree. The work has not been submitted for any degree to any other University.

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DECLARATION

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JULY, 2024

I **JYOTI JOPIR**, hereby declare that the subject matter of this thesis is the record of work done by me, that the contents of this thesis did not form basis of the award of any previous degree to me or to the best of my knowledge to anybody else, and that the thesis has not been submitted by me for any research degree in any other University/ Institute.

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Acronym/symbol	Full form/meaning
°C	Degree Celsius
%	percentage
<,>	less and greater than
1	minutes
"	seconds
μg	milligram
AOAC	Association of Official Analytical Collaboration
BCR	Benefit Cost Ratio
BNF	Biological nitrogen fixation
C/N	Carbon Nitrogen ratio
C/P	Carbon phosphorous ratio
С	Carbon
Ca	Calcium
CD	Critical difference
CFU	Colony forming unit
cm	centimetre
CO2	Carbon dioxide
Cu	Copper
DAP	Days after planting
DAS	Days after sowing
day ⁻¹	per day
E	East
et al	and others
etc.	and other similar things
Fe	Iron
FYM	Farmyard manure
g	gram
GHG	Green House Gas

List of abbreviations and symbols

CDT	
GRT	Green Revolution Technology
H_2SO_4	Sulphuric acid
ha ⁻¹	per hectare
HCN	Hydrogen cyanide
HNO ₃	Nitric acid
HClO ₄	Perchloric acid
ISFR	Indian State Forest Report
k	Decay constant
Κ	potassium
kg N ha ⁻¹	kilogram of nitrogen per hectare
kg	kilogram
kg ⁻¹	per kilogram
k _K	decay coefficient of potassium
km ²	square kilometre
$k_{ m N}$	Decay coefficient of nitrogen
k _P	decay coefficient of phosphorous
LSD	Least Significant difference
L/N	Lignin Nitrogen ratio
-ln	Natural logarithm
m^2	square meter
m^{-2}	per square meter
m ⁻³	cubic meter
MBC	Microbial biomass Carbon
MBP	Microbial biomass Phosphorous
ml	millilitre
mm	millimetre
Mn	Manganese
N/P	Nitrogen phosphorous ratio
Ν	Nitrogen
Ν	North
Na	Sodium

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	WUE	Water use efficiency
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	Zn	Zinc

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CHAPTER 1

GENERAL INTRODUCTION

1.1. Agriculture production - the challenge

Agriculture forms a bridge between man and nature. However, throughout the past century, global agricultural practices have altered in terms of their farm size, marketing tactics, economic and biological variety, climate change, vulnerability to urbanization, choppy international markets, and increasing dependency on external non-renewable resources (Cherr et al., 2006). The production of high-quality food grains in sufficient quantities to feed the world's permanently expanding population without compromising the fertility, quality, or productivity of the soil is a vital issue in the agriculture environment (Sulieman and Tran, 2016). The world's population is currently 7.3 billion, and by the end of the 21st century, it is predicted to stabilize at 11.2 billion after rising to 8.5 and 9.7 billion in 2030 and 2050, respectively. (UNDESA 2015). The population increase, competition for available natural resources, and meeting global food demands will be difficult tasks by the year 2050 (Meena et al., 2018). The target becomes more challenging when we include climate change and livelihood problems for farming people. According to Yadav et al. (2021), the food security and ecosystem services are in danger in various countries due to the degradation of land, increase in population and urbanization. The recent approach of intensive land use for food grain production has resulted in the progressive degradation of soil organic matter because of the dissolution of solid soil aggregates and the decomposition of organic matter which consequently deteriorates the soil health (Gill et al., 2008; Meena, 2013). Causes of land degradation also include the overuse of pesticides in agriculture and food production, which is hastening the decline of ecosystem services that regulate and support human well-being and has a negative impact on environmental sustainability. All of these make it harder to reach the sustainable development goals set out by the United Nations.

With the increasing population, farmers explore intensive agricultural practices, such as the use of chemical fertilizer, pesticides, and other chemical inputs, to boost their productivity as a result of the rising population strain on the limited agricultural area for food production and other demand (Deshar, 2013). To meet the growing demand for food by increasing agricultural productivity has become a significant problem in terms of safeguarding agricultural systems and minimizing the environmental effects (Pittelkow et al., 2015; Fischer, 2014). One of the biggest environmental issues in the world is the decline in the quality of soil (Tejada et al., 2008a; Chimouriya et al., 2018). This is also seen as a significant obstacle to obtaining the necessary level and volume of agricultural production. Global issues with soil degradation include overly intensive tillage, excessive grazing or crop residue clearance, deforestation, improper agricultural rotations, mining, building, and urban extension (Singh et al., 2021; Karlen and Rice, 2015; Lal, 2015; Komatsuzaki and Ohta, 2007). Continuous use of chemical fertilizers and pesticides to increase crop yields has been identified as the main cause of soil deterioration, health risks, and other environmental pollution (Gangwar et al., 2017; Kalu et al., 2015; Hou et al., 2010)

In developing and populous nations like India, restoring the fertility of the agricultural land soil is a great challenge. In this context, several of the so-called Green Revolution Technologies (GRTs) may be recalled, which improved the atom-level resilience of Indian agriculture but also had a significant negative impact on the agroecosystem by acerbic biodiversity and polluting the environment. The nation has seen both the advantages of Green Revolution Technologies (GRTs) in increasing grain yield and their negative effects over the past few decades, including a decline in soil fertility, deprivation of the land, genetic erosion, ecological unbalance, loss of soil flora and fauna, stagnation of yields, and associated insecurity in farmers' livelihoods (Maitra *et al.*, 2018). The Eastern Himalayan Region of India, which is home to almost 50 million people, is experiencing low agricultural yields as a result of the immense strain of land degradation (Singh *et al.*, 2021). Low farm income, inefficient use of resources, and stagnant yields are common outcomes of widespread mono-cropping (Babu *et al.*, 2020; Ansari *et al.*, 2022). Maintaining the soil fertility without reducing crop output has become increasingly difficult, and one of the main challenges in this regard is stabilizing soil quality (Babu *et al.*, 2020). In the North Eastern Himalayan (NEH) region, characterized by significant annual rainfall, farming on the steep slopes frequently leads to the depletion of organic carbon, nutrients, and binding agents of the soil. This phenomenon results in a reduction in soil aggregation and the deterioration of its structure, ultimately leading to soil erosion and environmental degradation (Choudhury *et al.*, 2022).

1.2. Improving soil health and crop yield- a sustainable approach

Soil degradation severely limits plant output and growth. Agrochemicals, such as pesticides and fertilizers, are the leading cause of soil degradation. The accumulation of agrochemicals endangers human health and harms the ecosystem. Soil deterioration can result in the loss of organic matter, chemical accumulation, diminished fertility, and lower agricultural productivity. Excessive tillage, continuous cropping, and reliance on agrochemicals threaten agriculture's future. Ensuring soil fertility at a sufficient level is crucial for optimizing the crop yield. This goal can be accomplished by implementing suitable soil and crop management techniques.

However, in order to reduce environmental issues and improve crop yield and soil health, innovative alternative methods are being developed to replace chemical fertilizers (Hwang *et al.*, 2015), and sustainable approaches are being practiced (Singh *et al.*, 2019). Organic farming is thought of as a blueprint for improving current agriculture's sustainability while reducing its negative environmental effects (Darnhofer *et al.*, 2010; Jensen *et al.*, 2015; Arlauskiene *et al.*, 2017) and is grabbing farmers' attention globally due to its advantages over modern farming techniques (Letourneau and Goldstein, 2001; Aulakh *et al.*, 2022). Its main features involve promoting long-term soil fertility through enhancing soil biological activities, managing disease and pests through biodiversity and resistant varieties, and utilizing biological nitrogen fixation (Chhonkar, 2002). The use of organic manures to increase soil fertility by preserving the balance between soil organic matter and soil microflora, eventually enhancing the soil's physico-chemical and microbiological characteristics (Gregory *et al.*, 2015; Bulluck *et al.*, 2002). The soil's physical characteristics and

nutrient status can be improved by adding organic amendments to it (Brar *et al.*, 2015). However, it is also impossible for organics such as farmyard manure and compost to be made available in substantial quantities. In this regard, the use of organic manures, including green manuring is a crucial tactic for preserving and enhancing soil fertility for long-term crop production.

1.3. Green manuring and green manure crops

Green manuring has been widely practiced in India, but in recent decades, interest in it has declined due to increased pressure on food production, the presence of different competitive and profitable crops, and the accessibility of both cheap and expensive chemical fertilizers. However, it has recently regained importance for both organic growers and low-input farms using conventional agronomic methods due to access to soil issues, depletion of soil fertility, and public concern about abuse and energy conservation. Where farmers cannot purchase artificial fertilizers because of financial restrictions, it is imperative to employ farm-grown green manures effectively and rationally (Meena *et al.*, 2014). Therefore, to increase future food grain output while protecting soil health, farmers and researchers have chosen to adopt conservation agriculture practices, resource conservation, and the usage of green manuring in the farming system (Meena and Majumdar, 2016; Meena *et al.*, 2016).

Green manuring is the process of incorporating a crop into the soil while it is still green to enhance the soil's fertility and structure. Green manuring as defined by Pieters 1927, "is the practice of incorporating fresh or dried, undecomposed plant matter into soils, either grown in place or brought from elsewhere". Adding green manure to the soil is an alternative method for preserving the productivity of the soil and agricultural environment. The incorporation of the entire plant of green manure which contains more nutrients than crop straw, into the soil increases soil fertility while also adding organic matter to the soil (Khan *et al.*, 2020; Yang *et al.*, 2019) and also increases the activity of soil enzymes, which fosters plant growth and controls nutrient fluxes (Chavarri *et al.*, 2016). Incorporating green manure into agriculture fields significantly improved crop growth and productivity while lowering the incidence of plant diseases and insect pests (Yang *et al.*, 2019; Li *et al.*, 2015; Larkin and Griffin,

2007). The activities of soil enzyme increase with the incorporation of green manure and these enzymes are considered a crucial marker for the quality and fertility of the soil, which helps in promoting the growth of the plant and also in regulating the nutrient cycling (Dai *et al.*, 2019; Chavaria *et al.*, 2016).

Green manure crops used for enriching the soil N can be leguminous as well as non-leguminous (Singh and Kumar, 2009), however, the choice of the green manure crop depends on several variables, such as the prevailing climatic condition, cropping method, seed availability and other variables such as local customs and prejudices (Meena et al., 2018). Any crop cultivated to be incorporated beneath when green or shortly after maturation for soil improvement is referred to as a "green manure crop" (Soil Science Society of America, 1997). The Green manure crops not only add nutrients to the soil but can also develop deep roots that can absorb minerals from deeper soil that is less readily available, increasing the concentration of plant nutrients in the surface soil (Noordwijk et al., 2015) and lower the use of fertilizer (Yang et al., 2019). Legume crops have an advantage over non-leguminous crops due to their ability to fix the atmospheric nitrogen (Vyn et al., 2000; Rao, 2014; Lee et al., 2010) which tends to result in higher N content in the legumes (Asghar and Kataoka, 2022; Kim et al., 2007) and thus increasing the soil's nitrogen pool (Mayer et al., 2003; Carlsson and Huss-Danell, 2003). Leguminous green manure crops are widely known to play a significant part in managing soil health (Whitbread et al., 2000) and have recently attracted more attention in improving soil fertility and agricultural sustainability (Ray and Gupta, 2001; Fageria, 2007).

A vast array of legumes has the potential to be used as green manures. Tropical legumes come in several hundred species, however, only a small portion of the potentiality of these species as green manure has been investigated (Fageria, 2007). In temperate regions also legume crops, which can be utilized as green manure crops, are widespread. The legumes that are grown most frequently are *Crotolaria juncea*, *Sesbania aculeate*, *Sesbania rostrata*, *Trifolium alexandrinum*, and *Vigna radiata* (Meena *et al.*, 2018). In addition to BNF, a green manure crop should have the following desirable traits: fast-growing habit, high rate of Nitrogen accumulation, high tolerance to biotic stresses and abiotic stresses, a wide range of ecological adaptability,

timely release of nutrients, photoperiod insensitivity, high seed production, higher seed viability, and fast decomposability (Meena *et al.*, 2015; Irin and Biswas, 2021). Legume-based systems increase the availability of nitrogen and phosphorus, as well as the organic carbon and humus content of the soil (Kumar *et al.*, 2020; Jensen *et al.*, 2012). Therefore, using legumes in cropping systems is crucial for improving soil health and production. (Binder *et al.*, 2010; Dhakal *et al.*, 2016).

1.4. Scope of the study

Organic farming used to be mostly a grassroots movement, but in recent years it has caught the attention of governments, and the majority of nations have created organic farming legislation. Concern over the long-term viability of agricultural systems as well as increased public knowledge of ecologically friendly agriculture practises and wholesome diets are becoming more widespread. To address these issues agricultural production with minimum or no inorganic and maximum organic inputs for maintaining soil health is being continuously advocated. Besides the recent trends in the global climatic phenomenon have compelled policymakers and the scientific community all over the world to adopt appropriate land management practices including agricultural systems which can be beneficial in reducing GHG emissions and in increasing carbon sequestration. The Govt. of India has adopted the entire North Eastern region as a potential organic production hub. Therefore, sustaining agricultural production under organic farming in this region is anticipated to increase the requirement for more organic amendment sources.

Mizoram is rich in biodiversity and harbors many plant species which are otherwise considered weeds that may have the potential of being utilized as green manure crops. Moreover, one of the readily available sources of organic matter and plant nutrients is weed biomass (Parbhankar and Mogle, 2017). Many weed species like *Cassia*, *Crotolaria*, and *Aeschynomene* are used as green manure (Chamle, 2007). In this context, using locally available plant resources such as green manuring crops may help maintain overall soil health with improved soil fertility, in situ water conservation, weed control, and management of soil-borne pathogens. These potential green manure crops can be used as additional input sources for soil amendments towards organic production systems. The information generated can help in the selection of new locally available plant species which can be utilized as green manure crops to be recommended to the farmers of the region.

1.5. Research hypothesis

The study tested the following hypothesis:

- 1. The selected leguminous weed species produces high amount of biomass and accumulate significant amount of nutrients in their shoot and root biomass.
- 2. The leguminous weed species decompose and release nutrient at faster rate.
- 3. The leguminous species have the potential to fix high amount of nitrogen.
- 4. The leguminous species have significant influence on soil physical, chemical and biological properties, and thus improve soil health and increase the yield of agriculture crops.

1.6. Objectives

Considering the above facts in view, the present study entitled "Assessment of Green Manuring Potential of Leguminous weeds of Mizoram" was conducted with the following objectives:

- 1. To estimate the biomass production and nutrient content of the selected species as green manure plants.
- 2. To assess the decomposition rate and nutrient release pattern of the selected species.
- 3. To assess the nitrogen-fixing ability of the selected species.
- To determine the impact of the selected weeds on the soil properties and growth & yield of some agriculture crops.

The outcome of the study is also expected to enhance our understanding on new approach of weed management and utilising weed biomass for improving soil health and crop yield while also contributing to soil carbon sequestration and mitigation of climate change.

CHAPTER 2

REVIEW OF LITERATURE

In an era marked by increasing environmental concerns and the urgent need for sustainable agricultural practices, green manure has emerged as a powerful tool in the hands of farmers and land stewards worldwide. Green manuring is an old practice that dates to the beginning of the Christian era and has been studied in India for a long time (Allison, 1973). Green manures' positive impacts are mainly attributable to their rapid growth, greater accumulation of biomass, nitrogen fixation, maintenance of nutrients in their green tissues, and mineralization of nutrients, which increases nutrient uptake by crops (Bhayal *et al.*, 2018) besides various other soil health benefits.

In this chapter, an attempt has been made to review the research works carried out regarding green manuring and its various aspects which have been presented in different headings and subheadings as follows:

2.1. Biomass production and nutrient accumulation of green manure

Biomass production and nutrient addition by green manure crops mainly depend on the type of green manure species, climatic conditions, and duration of green manure crop. Leguminous species can produce large amounts of biomass and accumulate large concentrations of nutrients, and the production of biomass is influenced by changes in temperature, altitude, and the direction of the region towards the sun (Matos *et al.*, 2008). When predicting the suitability of a legume for green manuring, biomass production, and N accumulation are frequently taken into consideration (Yoshida and Kitou, 1995).

Stopes *et al.* (1996) reported that over a year of green manuring, white clover accumulates the highest dry matter (12.2 t DM ha⁻¹ year⁻¹) and red clover the most N above ground (371 kg N ha⁻¹ year⁻¹). Vimala *et al.* (1999) reported fresh and dry biomass of 29, 31, 37 and 39 t ha⁻¹ and 6.1, 7.0, 6.4 and 8.7 ha⁻¹ for *Centrosema pubescence, Calopogonium caeruleum, Pueraria javanica* and *Calopogonium mucunoides* respectively.

Another study by Sakala *et al.* (2003) inferred *Mucuna pruriens*, *Crotalaria juncea*, and *Lablab purpureus* produced 6.7, 4.9, and 4.9 t ha⁻¹ of biomass from early integrated residues, respectively, while the same legumes produced 5.9, 5.2, and 4.1 t ha⁻¹ of biomass from late incorporated residues respectively.

Turgut *et al.* (2005) observed that on a three-year average, the aboveground dry matter and root dry matter yield by *Pisum sativum* L., *Vicia sativa* L., and *Vicia faba* L. was 3065 kg ha⁻¹, 2647 kg ha⁻¹ and 1307 kg ha⁻¹; 141 kg ha⁻¹, 138 kg ha⁻¹, and 261 kg ha⁻¹ respectively. The N content accumulated in the aboveground was 68.7 kg ha⁻¹, 54.0 kg ha⁻¹ and 30.0 kg ha⁻¹ while in the root was 2.00 kg ha⁻¹, 1.88 kg ha⁻¹ and 4.12 kg ha⁻¹ for *Pisum sativum*, *Vicia sativa*, and *Vicia faba* respectively.

According to Balana *et al.* (2010) the aerial biomass produced by Mucuna sp, *C. juncea* and *Canavalia ensiformis* was 26.1, 50.8 and 22.5 t ha⁻¹ respectively. Also further confirmed to the finding that the Nitrogen accumulation from the three legumes ranged from 144 to 349 kg ha⁻¹ over the evaluation period, with *Crotalaria juncea* being the most significant contribution.

Odhiambo *et al.* (2010) reported after 4 months of planting, the biomass produced by five green manure legumes (Cow pea, Sunhemmp, Lablab, Butterfly pea and Mucuna) ranged from 2.1 mg ha⁻¹ to 13.6 mg ha⁻¹ in the first year and 0.8 to 2.9 mg/ha⁻¹ in the second year of which Sunhemmp produce the highest biomass in both the year, and further reported that N content ranged between 73 and 279 kg ha⁻¹ and 10 -51 kg ha⁻¹ in the first and second year respectively.

Pooniya *et al.* (2012) carried out a field experiment with summer green manuring crops, namely, green gram, cowpea, and dhaincha of which the highest crop residue was added by dhaincha, or 38.56 mg ha⁻¹. This further resulted in recycling 180.5, 22.6, and 267.8 kg ha⁻¹ of N, P, and K, respectively.

According to Irin *et al.* (2019) *Crotolaria juncea* significantly produced the highest fresh and dry biomass (35.00 t ha⁻¹ and 5.25 t ha⁻¹respectively) followed by Sesbania rostrata (29.33 t ha⁻¹ and 5.12 t ha⁻¹ respectively) and Sesbania aculeata (28.12 t ha⁻¹ and 4.35 t ha⁻¹ respectively). In another study Chand *et al.*, (2015) revealed

Sesbania aculeata significantly produced the highest fresh and dry biomass accumulation among the summer green manuring crops studied.

Fernandes *et al.* (2021) studied that the biomass produced by showy crotolaria and velvet bean was 8.6 and 6.1 Mg ha⁻¹ respectively and the nutrient accumulated by Crotolaria and velvet beans were respectively 211 and 159 kg N, 18.3 and 9.8 kg P, 156 and 101 kg K, 58 and 44 kg Ca, 27 and 14 kg Mg, 129 and 80 g Cu, 2386 and 2320 g Fe, 474 and 537 g Mn, and 466 and 265 g Zn. Similarly, a study by Neelima *et al.* (2007) reported a total biomass of 35 t ha⁻¹ (29.37 t ha⁻¹ of shoot and 5.9 t ha⁻¹ of root) by *Crotolaria juncea*.

Eberle *et al.* (2021) reported studies in rainfed and irrigated *Crotolaria juncea* as green manure accumulates biomass ranging from 0.2 to 5.1 Mg ha⁻¹ and 0.2 to 4.2 Mg ha⁻¹ respectively after 60 days of planting Likewise, Schomberg *et al.* (2007) observed *C. micans* after 60 DAP yielded biomass of 4.6 and 5.0 Mg ha⁻¹. Garzon *et al.* (2020) reported biomass 1.0 to 3.3 Mg/ha biomass at 60 DAP. Cherr *et al.* (2006) documented sunnhemp yields of 4.0 Mg ha⁻¹ at 60 days after planting (DAP).

In a study by Prikhodko *et al.* (2021) significant amount of dry matter yield was produced by the various green manure crops where the average dry matter yield obtained was about 4.9 t ha⁻¹. The study further revealed that the green manure crops Melilot and Sainfoin had the highest nitrogen content in their biomass, with 156 and 142 kg ha⁻¹, respectively.

A field experiment conducted by Najan *et al.* (2022) during the kharif season and the results revealed that the highest fresh and dry biomass was produced by dhaincha (40.98 t ha⁻¹) which was at par with Sunnhemp (40.40 t ha⁻¹). The study also revealed that dhaincha recorded the highest content of Nitrogen and Potassium with 3.90 % and 2.10 % respectively while the total phosphorous content was the highest in soybean (0.44 %).

Assessment on the nutrient accumulation of nine leguminous green manure crops revealed that *D. lablab* shown superior ability in accumulating phosphorus, potassium, and calcium in 1 Mg dry matter. However, *C. juncea*, *C. cajan*, *M. cinerea*,

and *M. aterrima* exhibited exceptional nutrient accumulation per hectare (De Jesus Avila-Escobedo *et al.*, 2022).

2.2. Decomposition and nutrient release pattern of green manure crop

Green manure when incorporated into the soil, undergoes decomposition and mineralization (Fageria, 2007; Meena *et al.*, 2018). Decomposition mainly undergoes two processes namely mineralization and immobilization (Gill and Fick, 2001; Dinnes *et al.*, 2002). Soil conditions, the chemical composition of plant residues, and the prevailing climatic all have an impact on the decomposition and mineralization of green manures (Thonnissen *et al.*, 2000 Trinsoutrot *et al.*, 2000; Fageria and Baligar, 2005; Reddy, 2016).

Green manure's source of nutrients comes from mineralization after mass decomposition when it is cut or mowed (Tei *et al.*, 2017). In the early days of decomposition, the chemical composition of green manure biomass such as the contents of C, N and C/N ratios influences the decomposition and mineralization of Nitrogen (Trinsoutrol *et al.*, 2000; Radicetti *et al.*, 2017). However, at the later stages the presence of more resistant components, including lignin, cellulose and lignin/N affects the breakdown of C and N (Vahdat *et al.*, 2011; Hadas *et al.*, 2004). Havstad *et al.* (2010); Ha *et al.* (2008) added the breakdown of C and N depends on the climatic and soil conditions such as moisture, texture, pH, temperature and biological activity of the soil.

The soil variables that had an impact on the breakdown and mineralization of green manure are soil texture, structure, response, microbial activity, and nutritional condition (Thonnissen *et al.*, 2000; Dhakal *et al.*, 2015). Verbene *et al.* (1990) claimed that the rate of decomposition of green manure legume was faster in the sandy soils as compared to fine-textured soils. Three major elements govern the processes of decomposition and nutrient release from organic residues applied to soil: (i) Soil properties and climate of the environment; (ii) Species composition; (iii) decomposition community (Perin *et al.*, 2010). The factors that affect the decomposition process under similar climate and management settings, are the soil

decomposition community and the chemical quality of plant wastes supplied to the soil (Brito, 2003).

Thonnissen *et al.* (2000) observed that *Glycine max* and *Indigofera tinctoria* degraded quickly, losing 30 to 70 % of their biomass after 5 weeks of inclusion. Likewise, Broder and Wagner (1988) claimed the incorporation of soybean residues lost about 68 % of the biomass in the first 32 days. Cobo *et al.* (2002) reported that the leaves of *Indigofera constricta* and *Tithonia diversifolia* had the highest rate of decomposition among all the other green manure species. And also, the release of potassium was the highest among all the nutrients for all the green manure species.

Ruiz- Vega *et al.* (2010) claimed that *Vigna radiata* and *Dolichus lablab* reported the highest decomposition rate with 8.8 and 9.1 g ha⁻¹ day⁻¹, respectively while common beans and Sunhemp had the decay rate of 5.7 and 4.1 g ha⁻¹. Also clamied that the C/N ratios of the plant material and the soil temperature and water content had a significant effect on the green manures rate of decomposition. According to the findings by Odhiambo (2010), all three green manure legumes can contribute large amounts of N for plant uptake, with sunhemp releasing N at a higher rate than lablab and mucuna.

Matos *et al.* (2011) revealed in coffee agroforestry systems under different climates, *Arachis pintoi*, *Calopogonium mucunoides*, *Stizolobium aterrimum, and Stylosanthes guianensis* loss about 25 % of its weight in the initial 15 days and also, further claimed that 32 % total N content in the plant residues of the green manure crops was released in the initial 15 days. Phosphorous was the fastest to be released among all other nutrients with *k* value ranging from 0.017 - 0.039 day⁻¹.

Matheus *et al.* (2018) reported *C. usaramoensis*, *P. lunatus*, *M. pruriens*, and *C. pubescens* decomposed at a rate of 2.76 year ⁻¹, 2.41 year ⁻¹, 1.71 year ⁻¹ and 0.95 year ⁻¹ respectively. More than 50 % of N, P, K, and Ca was released in 20 days by *C. usaramoensis* and *P. lunatus* while *M. pruriens* and *C. pubescens* released 50 % at 30 days.

Pereiria et al. (2016) reported that Crotalaria spectabilis and Canavalia ensiformes had higher decomposition and nutrient release rates among all the six leguminous green manure legumes, making them the most promising for the region. However, *Crotalaria juncea*, on the other hand, is more recommended for greater residue persistence in the soil.

Cardoso *et al.* (2018) evaluated the rate of decomposition and Nitrogen mineralization of *Canavalia ensiformis* and *Dolichus lablab* used as green manure and intercropped with coffee trees. The study revealed that the release of N was faster in *Canavalia ensiformis* than *Dolichus lablab* at 60 days while the mass loss in both the species was greater than the mineralization of Nitrogen. The average period for 50 % mass breakdown for both the green manure species was between 9 - 24 days.

Watthier *et al.* (2020) reported faster rate of decomposition in green manure mulch when mixed with higher proportion of legumes and P mineralization is faster than N mineralization in green manure mulch containing more than 50 % legumes. Also further added that the chemical composition of jack bean and millet mulch used as green manure influences mass decomposition and N mineralization.

Mangaravite *et al.* (2023) carried out a study on the decomposition and nutrient release dynamics of four tropical green manure plant residues, where it was concluded that the decomposition and release of N, C, K and Mg was lower in *Crotolaria micans* and *Cajanas cajan* as compared to *Canavalia ensiformis* and *Mucuna deeringiana*. It also further concluded that for supplying nutrients in shorter periods, jack bean and dwarf mucuna are recommended, However, pigeon pea and Sunn hemp, on the other hand, are recommended for better mulch persistence on the soil.

2.3. Biological Nitrogen Fixation by green manure crops

As a result of their symbiotic association with Rhizobium bacteria, legumes might contribute to 40 % of the total N fixation in the world and can satisfy 50–80 % of plant N requirements through biological nitrogen fixation (Meena *et al.*, 2018; Meena *et al.*, 2015; Ladha *et al.*, 1992).

Ladha *et al.* (1998) reported 2.6 kg of N ha⁻¹ day⁻¹ on average under various legume green manure crops. Further also reported that 45-60 days old dhaincha species fixed about 200 kg N ha⁻¹ and 55 days old fixed about 303 and 383 kg N ha⁻¹ without

and with inoculation of *Azorhizobium* bacteria respectively. Thus, it is evident that different legume species fix different amounts of nitrogen, and that these amounts vary depending on the species of legume, its variety, the number of functional root nodules, the type of soil, agronomic practices, water management practices, and current climatic conditions as well as their interactions with other factors (Fageria *et al.*, 2005).

Ramos *et al.* (2001) revealed *Crotalaria juncea* fixed N on an average of 40% which was higher than *Mucuna aterrima* and *Cannavalia ensiformis*. A study by Espindola *et al.*, 2006 studied the BNF of tropical leguminous *Pueraria phaseoloides*, *Arachis pintoi*, *Macroptilium atropurpureum* where it contributed about 33.7, 40.5, and 24.2 % respectively of the N in banana leaf tissues. Mueller and Kristensen (2001) reported Hairy vetch, crimson clover and Persian clover green manure fixed more than 100 kg N ha⁻¹.

Gathumbi *et al.* (2002) demonstrated that *Crotolaria grahamiana* fixed approximately 150 kg N ha⁻¹ compared to *Arachis hypogaea*, *Macroptilium* atropurpureum, *Calliandra calothyrsus*, *Sesbania sesban*, *Cajanus cajan* and *T. vogelii* which fixed an average of 8, 64, 24, 52, 91 and 100 kg N ha⁻¹ respectively.

According to Espindola *et al.* (2006), *Pueraria phaseoloides*, *Arachis pintoi* and *Macroptilium atropurpureum* supplied about 33.7, 40.5, and 24.2 % of the nitrogen found in banana leaf tissue, respectively. Also, According to Paulino *et al.* (2009), Crotalaria transfers 22.5 % of N-BNF to soursop trees.

Nezomba *et al.* (2008) revealed that Crotolaria species fixed the highest amount of N where about 58-105 kg N ha⁻¹ was fixed by *C. juncea*. Samba *et al.* (2002) found that *Crotalaria ochroleuca* obtained 47–53 % of its nitrogen from nitrogen fixation. Mapfumo *et al.* (1999) found that pigeon pea (*Cajanus cajan*) obtained 81 % of its nitrogen from nitrogen fixation.

Tauro *et al.* (2009) affirmed that the Indigenous legumes obtained 61-90 % of their nitrogen via fixation in all studied areas and also confirmed that after 3 and 6 months, *Crotolaria pallida* in all the study areas, fixed the highest amount of nitrogen resuting to about 140 - 193 kg N ha⁻¹.

Mendonca *et al.* (2017) reported green manure legumes *C. mucunoides*, *S. guainensis*, *C. cajan*, and *D. lablab* had the highest percentage of BNF, at 46.1, 45.9, 44.4, and 42.9 %, respectively and also further claimed the amount of N from biological fixation that was transferred to the coffee plants by *C. mucunoides C. spectabilis* and *C. cajan*, was 48.1 %, 48.8 %, and 55.8 % respectively. Similarly, For the Asian leguminous shrub Flemingia (*Flemingia macrophylla*), 360 days after planting, 76 % of the N which contributes about 57 kg ha⁻¹ of N in was taken up from BNF (Salmi *et al.*, 2013).

According to Meena *et al.* (2018) legumes could contribute 40% of total N fixation in the world due to their symbiotic interaction with Rhizobium bacteria, and biological nitrogen fixation can meet 50 - 80 % of plant N requirements.

2.4. Effect of green manure on the soil properties

The impact of green manure application on soil characteristics relies on the quality of the green manure crop due to their different chemical compositions. The major goal of applying green manure is to improve the organic matter and nitrogen content of the soil, which immediately increases soil health and plant productivity (Sharma *et al.*, 2017). In addition to enhancing soil health, Legume green manure aids in the management of weeds, insects, and diseases (Kumar *et al.*, 2014), serves as a binding agent for soil and enhances soil structure (Schutter and Dick, 2001). The use of green manures between alternating crops was to access the organic matter in the soil (Pung *et al.*, 2004) which promotes the mineralization of plant nutrients and soil microbial activity (Eriksen, 2005) and therefore enhances the quality and fertility of soil.

2.4.1. Soil physical properties

According to Boparai *et al.* (1992), the application of green manure increased the water-stable aggregates between 0.1- 0.5 mm by 62 % and infiltration rate and reduced the soil bulk density.

Mandal *et al.* (2003) carried out an experiment where according to the study, the magnitude of reduction in bulk density due to green manure (*Sesbania rostrata*,

Sesbania aculeata and *Vigna radiata*) over fallow was 0.03-0.07 Mg m⁻³ and 0.05-0.09 Mg m⁻³ in 0-15 cm and 15-30 cm soil layer respectively. Similarly, Sultani *et al.* (2017) reported that Sesbania, cluster bean and rice bean on average reduced the soil bulk density by 5 %, increased total porosity by 8% and macropores and large mesopores by 28 %.

Singh *et al.* (2006) claimed treating the soil with various treatments in addition to green manure considerably increases the content of soil organic carbon which in turn improves the infiltration rate, soil's aggregation state and reduces the dispersion ratio, bulk density and soil strength.

Tejada *et al.* (2008a) reported when Beet vinasse was constituted with green manure (Red clover) the soil structural stability was increased by 10.5 % and soil bulk density decreased by 13.5 % concerning the control soil. Jeon *et al.*, 2008 stated green manure cropping decreases the soil temperature and there was an improvement in the soil porosity and bulk density at the topsoil.

Selvi and Kalpana (2009) also reported that reduced bulk density, greater pore space, water intake, and water retention are the consequences of incorporating green manuring.

Salahin *et al.* (2013) revealed a significant increase in the soil moisture and porosity concerning *Sesbania aculeata*, *Mimosa invisa*, and *Vigna radiata* green manuring as compared to the control soil. Similar studies also found that moisture content and infiltration rate increased after the addition of green manure (Triplett *et al.*, 1968; Zerega *et al.*, 1995; Pradit *et al.*, 1993).

In another experiment by Hafifah *et al.* (2016) noted that the incorporation bulk density was reduced by 27 % and porosity of soil increased by 15 % after incorporating the soil with *Tithonia diversifolia* as green manure when compared to control (initial soil). Additionally, it was reported that the addition of sunnhemp to the Rabi sorghum-sunflower yearly rotation cropping system reduced the bulk density of the soil from 1.18 Mg m⁻³ - 1.0 Mg m⁻³ and enhanced porosity by 6.5 % (Guled *et al.*, 2010). Also, an experiment carried out by Chikowo *et al.* (2004) in Zimbabwe, found that adding

woody legumes to the soil decreases bulk density while increasing soil porosity and granulation.

Mujdeci and Uzumcu (2017a) observed an increase in the aggregation of soil and the size of aggregates after green manure and farmyard manure application. Several other studies have also claimed an increase in the soil water retention capacities with the application of organic matter (Tadesse *et al.*, 2013; Alaboz *et al.*, 2017; Mujdeci *et al.*, 2017b).

Ahmed *et al.* (2020) reported that the green manuring practiced considerably increased the soil physical characteristics and nutrient availability and also further added that green manuring under dhaincha enhanced the soil's physical environment, softened the soil as evidenced by a decrease in bulk density, an increase in porosity, and the accessibility of important nutrients which ultimately increased the yield. Luo *et al.* (2020) observed that the application of green manure in the long term has been reported to reduce the bulk density and increase the soil porosity.

Ma *et al.* (2022) confirmed to the findings that in the 0-20 cm soil layer, green manure treatment reduced bulk density by 5.6 %, and in the > 20 cm layer, it was 4.0 %, compared to the fallow control. According to various reports by Sultani *et al.* (2007); Sanchez de Cima *et al.* (2016); Bassouny and Chen (2016), the root bioturbation that occurs during the growing stage of green manure and the ensuing increase in soil organic matter due to the decomposition of leftovers are the main causes of this drop in soil bulk density.

Naz *et al.* (2023) claimed that during three years of testing, adding green manure to the soil enhanced the soil's physical qualities and also further added that the breakdown of green manure root increased soil porosity and decreased bulk density because soil organic matter loosens the soil and increases its ability to store water (Ashworth *et al.*, 2020; Austin *et al.*, 2017).

2.4.2. Soil chemical properties

Due to their different chemical compositions, the impacts of green manure application on soil chemical characteristics rely on the quality of the green manure crop (Sumiahadi *et al.*, 2020). Sharma *et al.* (2017) added that the major goal of applying green manure is to improve the organic matter and nitrogen content of the soil, which immediately increases soil and plant productivity. Further, Fanish (2017) added green manure recycles nutrients into the soil while adding organic matter to the soil and aids in preventing nutrient loss from the soil.

Soil organic matter is the source of nitrogen and all other nutrients (Karakut 2009). Nitrogen is essential for the growth of plants and the production of high-quality crops (Rosenfeld and Ryans, 2011; Talgre *et al.*, 2012).

Yaduvanshi (2003) also noted a decrease in soil pH with the addition of green manure or farmyard manure to alkaline soils. Likewise, Nagar *et al.*, (2016) reported a reduction in pH and electrical conductivity in pigeon pea-black gram and pigeon pea-green gram legume-based intercropping systems.

Astier *et al.* (2006) reported that the addition of green manure also had an impact on soil pH, which was noticeably lower under oat and vetch than under the control. The formation of CO_2 and organic acids during the decomposition of incorporation of green manure may be responsible for the slightly lower soil pH in plots with green manure compared to the control (Agbede *et al.*, 2018b). Studies by Kumar *et al.* (2010); Subehia *et al.* (2013) stated that the incorporation of green manure on a long-term basis reduced the soil pH.

According to Ziblim *et al.* (2013) the soil incorporated with *Mucuna pruriens* and *Crotolaria juncea* increased the soil phosphorous, potassium, organic carbon and soil pH. Shoko and Tagwira, (2005) also noted that *C. juncea* and *M. pruriens* have the potential to improve the soil pH and the organic matter exchangeable availability. Likewise, Vanlauwe *et al.* (2000) reported that legumes *M. pruriens* and *C. juncea* increase the availability of phosphorous in soil after its incorporation.

Ehsan *et al.* (2014) reported Sesbania that was grown and rotavated in-place was claimed to have boosted the amount of organic matter and accessible P and K. Similar study by Abera and Gerkabo, (2021) also recorded that the incorporation of green manure legumes at respective times significantly increases the soil available P, exchangeable K, organic carbon and Total N.

Dey and Nath, (2015) claimed green manure addition raises soil organic carbon, which lowers soil pH and prevents phosphate from being fixed with iron and aluminium in the soil, thus increasing soil P availability. Additionally, continuous applications of green manure crops result in increased soil aggregation and wetness as well as decreased bulk density of the soil that has the potential to boost P uptake by subsequent crops due to their impacts on increased root and mycorrhizal growth (MacRae and Mehuys, 1985).

Sihi *et al.* (2017) reported the long-term inclusion of FYM, neem cake, and green manuring increased cation exchange capacity (CEC), and micronutrients such as Mn, Fe, Zn, and Cu levels in soil

Borthakur *et al.* (2018) reported organic carbon and nitrogen content improved by 0.25 % and 12.7 % respectively under *Sesbania aculeata* green manuring. Similarly, Oliveira *et al.* (2018) claimed, green leaf manure from *G. sepium* considerably improved post-harvest maize soil chemical characteristics over control, particularly nitrate-N, ammonium-N, total inorganic N, and K. In a different experiment, Yadav *et al.* (2019) stated that the addition of *G. sepium* green leaf manure under the ricerapeseed cropping system led to a relatively greater SOC pool, sequestration rate, and carbon retention efficiency throughout a four-year experiment.

Adekiya *et al.* (2019b) claimed that the incorporation of green manure such as Papaya, Moringa, Neem, and Mesquite leaves increased the soil organic matter, N, P, K, Ca, and Mg concentrations as compared to the control, and further added that these nutrients are abundant in green manures and that the decomposition of green manures releases these nutrients into the soil. Several other studies also reported increased soil organic matter, N, P, K, Ca, and Mg contents due to the addition of green manure in the soil (Mandal *et al.*, 2003; Herrera-Arreola *et al.*, 2007). Irin *et al.* (2019) reported the incorporation of *Sesbania rostrata* and *Sesbania aculeata* residues improved the soil by introducing more organic matter and nitrogen to the soil as compared. Similarly. Chanda and Sarwar, (2017) revealed as a result of ongoing continuous cultivation of green manure crops, soil organic matter increased, ranging from 2.30 % to 2.95 % at the early stage to 2.71 % to 2.98 % at the post-harvest stage.

Leite *et al.* (2021) showed that leguminous crops increase the soil's total carbon and total nitrogen and decrease the soil C/N ratio. Following the intercropping of the three green manure crops, the NH_4^+ - N concentration dramatically rose which may be because of the ability of the leguminous rhizobia to fix nitrogen (Zhong *et al.*, 2018). Similarly, Ding *et al.* (2021) reported that intercropping *Dactylis glomerata* in apple orchards increases the soil pH and reduces the available potassium in soil.

Hu *et al.* (2022) claimed that the amount of fractions of soil organic carbon considerably increased with the application of green manure, and high levels of green manure input increased the amount of organic carbon fractions that moved and accumulated in the soil. Several other studies added that the cultivation of green manure over an extended period can greatly enhance soil organic matter (Castellano-Hinojosa and Strauss, 2020; Rupullo-Ruiberriz de Torres *et al.*, 2021).

Yuan *et al.* (2023) observed that *Desmodium ovalifolium*, *Grona heterocarpos* and *Stylosanthes guianensis* increased the soil total N, ammonium nitrogen and available phosphorous by 7.93 %, 558.85 % and 1207.34 % respectively and also reported 47.29 % and 58.48 % reduction of available potassium by *D. ovalifolium* and *S. guianensis* respectively.

2.4.3. Soil biological properties

Green manure serves as a source of food, for various soil organisms and microorganisms, thus improving the soil's physical qualities and enhancing subsequent and succeeding crop growth (Irin and Biswas, 2022; Sultani *et al.*, 2007). Soil microbial biomass carbon plays a significant role in the nutrient cycling, stabilization of organic matter (Jedidi *et al.*, 2004) and is the living constituent of soil organic matter (Kumar *et al.*, 2020) and change in it may affect the cycling of soil organic matter (Shahriari *et al.*, 2011; Tajik *et al.*, 2012). Also, microbial biomass can be an early indicator of changes in the total organic carbon in soil (Wiesmeier *et al.*, 2019).

The breakdown of green manures performs crucial roles for microorganisms by providing C and energy for their growth and formation of new cells, which multiply their colony by depending on the decomposing organic matter (Ye *et al.*, 2014). Incorporating different green manure legumes in crop rotation and cropping systems can increase microbial diversity. (Schutter and Dick, 2001; Eriksen, 2005). Soil bacteria proliferate in great numbers as long as there is a C source of energy (Kumar *et al.*, 2014).

According to Chirinda *et al.* (2008), cropping systems that use green manure legumes had greater MBN and nitrification rates than systems that only used inputs from manure and mineral fertilizer.

Tejada *et al.* (2008a) reported that soil treated with *Trifolium pratense* green manures at a rate of 25 t ha⁻¹ had a 79.2 % increase in soil microbial biomass-C as compared to the control soil. Additionally, increased in soil microbial biomass C and N was claimed by (Stark *et al.*, 2007; Kautz *et al.*, 2007; Balota *et al.*, 2010). The increase in soil microbial biomass carbon and respiration can be because of the quick degradable organic materials that promote microbial activity (Tejada *et al.*, 2008b).

According to Shah *et al.* (2010), the average gains from the application of green manuring and N fertilizers over summer fallow were 1.79 higher microbial activity, 1.70 higher microbial biomass carbon, 1.49 times higher microbial biomass-N, 3.36 higher bacterial population, and 1.46 higher fungal population. Likewise, Pooniya *et al.* (2012) claimed *Sesbania aculeata* green manuring had the maximum amount of

soil microbial biomass carbon, followed by cowpea and mung bean. In comparison to cowpea, mung bean, and summer fallow, the microbial biomass carbon in soil rises with *S. aculeata* by 8.5, 15.9, and 27.3 %, respectively.

Surucu *et al.* (2014) reported application of the faba plant's above-ground sections resulted in higher urease and dehydrogenase activity than the application of the plant's underground stubbles and control soil.

Carvalho *et al.* (2015) reported that compared to soil without green manure, the addition of green manure encouraged an increase in MBC where *Mucuna pruriens* showed the highest increase in soil MBC as compared to the other green manure. Likewise, several studies also claimed the use of legumes as green manure improves the soil microbial biomass population (Biederbeck *et al.*, 2005; Liu *et al.*, 2006; Shah *et al.*, 2010).

Kataoka *et al.* (2017) stated that when compared to chemical fertilizer and nongreen manure, hairy vetch increased the biomass and diversity of soil fungi, hence promoting their activity. In addition, the inclusion of hairy vetch boosted phosphatase activity and Cladosporium *sp.* abundance in soil.

Mambu *et al.* (2018) claimed that *Crotolaria juncea* and *Sesbania cannabina* increased β -glucosidase and phosphomonoesterase soil enzymes. In another study, Bedi *et al.* (2009) also observed significant higher microbial biomass carbon, dehydrogenase, and phosphates activity by *S. aculeata* green manuring in combination with N fertilizers.

Dai *et al.* (2019) regarded soil enzyme activities as key markers of soil quality and fertility because it helps in reflecting the soil nutrient cycle. Similarly, Chavarria *et al.* (2016) inferred the incorporation of green manure increases the activity of soil enzymes, which foster plant growth and control nutrient fluxes. Actinomycetes, Azotobacter, and Azospirillum population in the soil was found to be increased when *Sesbania aculeata* was utilized as green manure with nitrogen at a rate of 90 kg ha⁻¹, according to Krishnaprabhu (2019). The soil microbes play a critical role in the functioning of soil ecosystems by promoting organic matter decomposition, nutrient cycling and plant growth promotion (Bardgett and Van Der Putten, 2014; Burton *et al.*, 2010). The addition of green manure to soil increased soil enzyme activity and, directly or indirectly, altered the makeup of the microbial population (Zhao *et al.*, 2016; Piotrowska and Wilczewski, 2012). Stark *et al.* (2007) also noted that during the early stages of their experiment, green manure significantly impacted the soil microbial population and altered soil microbial activity and biomass.

According to Kumar *et al.* (2020), changes in the microbial biomass may affect the cycling of soil organic matter (Shahriari *et al.*, 2011; Tajik *et al.*, 2012. Also, microbial biomass can be an early indicator of changes in the total organic carbon in soil (Weismeier *et al.*, 2019).

According to Nima *et al.* (2020), basmati rice-wheat cropping systems using farmyard manure and green manuring considerably enhanced basal soil respiration and dehydrogenase activity by 55.6 and 50.3 %, respectively, compared to systems using chemicals.

Zhou *et al.* (2021) found that green manuring treatment increased the abundance of gram-negative bacteria, fungi, and actinomycetes in post-harvest paddy soil. Thapa *et al.* (2021) also showed the addition of green manure promotes fungal biomass and microbial rhizosphere colonization. Likewise, Asghar and Kataoka, (2021) reported the incorporation of Hairy vetch and *Brassica juncea* increased the soil enzyme activities and also altered the soil fungal community.

Soil biological characteristics were significantly improved by green manure, according to study by Ma *et al.* (2021) reported that the green manure application resulted in increment of 28 % of the microbial biomass compared to control fields that were kept barren throughout the fallow period. They also further conclude that as result of applying green manure, the soil phosphatase, urease, catalase, and invertase activities were shown to rise by 14 % to 39 %.

2.5. Effect of green manuring on growth and yield of crops

Growth characteristics are crucial for higher crop yields since they are strongly connected with early and better crop growth due to the prior build-up of photo-assimilates and their partitioning towards the economic component. Increased yield from green manuring annual crops relies on the type of crop, the environment, and the management strategies used for both the green manuring crops and the succeeding field crops (Fageria, 2007). Crop production is a good indicator of the beneficial effects of legume green manuring on SOM and other soil characteristics linked to an increase in nutrition to growing crops (Meena *et al.*, 2018).

Turgut *et al.* (2005) observed average sweet corn ear yields on plots receiving green manuring were considerably higher (15127 kg ha⁻¹) as compared to those without the application of green manuring (13826 kg ha⁻¹) of which *Vicia sativa* green manure produced the highest ear yield (15764 kg ha⁻¹), plant height (129.5 cm), ear height (53.0 cm) and ear diameter (42. 8 mm).

Mehra and Singh, (2007) revealed that under the rice-wheat cropping system, *Sesbania aculeata* green manuring with inorganic fertilization led to greater growth indices of succeeding wheat crops. Likewise, Mandal and Pal, (2009) revealed the considerable residual effect of *S. aculeata* green manuring on the root length, root volume, and root dry weight of the next wheat crop under a rice-wheat cropping system. In another experiment, Hemaltha *et al.* (2000) noted that 12.0 t ha⁻¹ of dhaincha incorporated in in situ recorded a greater number of tillers per hill, taller plants, the output of dry matter, straw and grain yield and leaf area index. Similarly, Mandal *et al.* (2003) reported sesbania green manures increase the yield of rice and succeeding wheat.

According to Sharma and Behera, (2009) in a maize-wheat cropping system, growth and development of wheat were superior under the residual influence of summer legumes to summer fallow treatments. Likewise, Premi *et al.* (2013) observed, that *Sesbania aculeata* green manuring had a notable residual influence on plant height, the number of branches, the number of siliquae, and ultimately seed yield

increased by 42.3 %, and the oil yield of mustard as compared to fallow-mustard practice.

According to Ali *et al.* (2012), leguminous cropping patterns and green manuring increased paddy yield in comparison to rice-wheat cropping patterns. Another study by Egbe *et al.*, 2012 assessed the influence of green manure + fertilizer on the production of maize, where it was observed that Calliandra + fertilizer provided the highest grain yield (4696 kg ha⁻¹) and the least in control (3332 kg ha⁻¹).

Caliskan *et al.* (2014) claimed addition of green manure and its combination were found to be more successful in promoting lettuce plant development and yield than the standard production system's use of mineral fertilizer.

Tao *et al.* (2017) investigated the effects of four green manure fertilization regimes on the growth of maize, of which the most promising crop for increasing crop production was milk vetch, which also enhanced maize yield by 31.3 %.

Bhayal *et al.* (2018) revealed that green manuring and intercropping increased the plant height, nodulation, branching, seeds per pod and pods per plant of soybean by 7–11 %, 9–18 %, 11–34 %, and 7–12 %, respectively and also reported maize grains per cob were 20-23 % greater under green manuring and 20 % higher under maize + soybean intercropping.

Agbede (2018a) revealed that the use of Gliricidia as green manure resulted in an enhancement in cassava productivity, whereas Moringa contributed to the improvement of root quality and also added that green manure crops such as Neem, Moringa, Gliricidia and Leucaena were observed to enhance the mineral and starch levels while decreasing the HCN content in cassava tuber roots when compared to the control group.

Adekiya *et al.* (2019b) confirmed that among the several green manures tested, Moringa demonstrated the most significant improvement in the nutritional composition of Okra, particularly in terms of potassium, calcium, iron, zinc, copper, and vitamin C content. They also suggested that using Moringa as green manure improves the quality of okra fruits and Mesquite to increase the quantity. Alagoz *et al.* (2020) recorded the highest leaf chlorophyll concentration (45.6 CCI), stomatal conductance (74.8 mmol m⁻² s⁻¹), and yield (2.24 kg plant⁻¹) of tomato recorded in the raised bed with faba beans green manure incorporated at flowering stage. Additionally, according to Manici *et al.* (2018), the incorporation of hairy vetch into the soil, increased the growth of zucchini and tomatoes as a result of the improved nutrient availability and microbial growth promotion associated with leguminous crops.

Idham *et al.* (2021) demonstrated that maize's growth and productivity increased by the types and dosage of green manure, as measured by the plant's leaf area, stem diameter, cob length, weight of 100 dry-shelled seeds, and production per hectare of the dried weight of shelled maize. The application of *C. pubescens* green manure at a dose of 10 t ha⁻¹ resulted in the maximum maize production.

Kumar *et al.* (2021a) studied a two-year field experiment on the effect of summer green manuring on transplanted rice on sandy clay loam soil. The study found that *S. aculeata* over summer fallow resulted in significantly higher growth parameters, including plant height, tillers m⁻², and dry matter accumulation. According to a different study by Kumar *et al.* (2021b), transplanted rice that was green-manured with *S. aculeata* had greater crop growth indices like Crop growth rate, relative growth rate, and leaf area index compared to summer fallow.

A study by Liang *et al.* (2022) affirmed that the use of LGM resulted in a substantial increase in the production of the three main grain crops. Compared to crops without LGM treatment, the overall yield increased by 12.60 %. Specifically, the yield of wheat, maize, and rice increased by 9.49 %, 16.70 %, and 19.22 %, respectively.

Study by Liang *et al.* (2023) conclude that when compared to control, intercropping with apple with ryegrass and spring - rape treatments resulted in an 11.7 % and 5.7 % increase in apple yield and a 14.4 % and 7.5 % increase in water-use efficiency (WUE), respectively.

The results of Saquee *et al.* (2023) indicated that plots treated with a mixture of *Calopogonium* and *Pueraria* showed significant improvements in various growth parameters, including leaf number, leaf area, stem girth, plant height, ear height (ranging from 64.6 to 78.5 cm), cob yield (ranging from 1.2 to 1.4 t ha⁻¹), ear yield (ranging from 1.8 to 2.1 t ha⁻¹), and dry grain yield (ranging from 0.5 to 0.7 t ha⁻¹).

The necessity to develop sustainable agricultural systems has led to a recent upsurge in interest for green manuring. The aforementioned literature review reveals that there is a severe lack of data regarding the practice of green manuring in the northeastern region and in Mizoram in particular. People in the north-eastern hill's region avoid using chemical fertilizers due to concerns about their environmental impact. Hilly soils are not only 'hungry', but also 'thirsty', which is a severe issue with 'jhum' fields. Besides forest fires deplete organic matter, nutrients, and microbial biota, causing soil heating and degradation. Growing green-manuring crops can significantly enhance soil organic matter and micronutrients, leading to more sustainable agricultural crop output. This study is significant not only for filling a knowledge gap on the role of green manure in soil management and nutrient cycling, but also for utilizing the abundantly available weeds as potential resources for soil health management for sustainable crop production while developing ecorestoration strategies for degraded areas in various land use systems in the state of Mizoram and in North East hilly region as well.

STUDY SITE AND SPECIES DESCRIPTION

3.1. Study site

The present investigation entitled "Assessment of the Green Manuring Potential of Leguminous Weeds of Mizoram" was carried out from the year 2020 to 2023 at Mizoram University, Tanhril located in Aizawl district of Mizoram, India and is about 20 km from the capital city (Figure 3.1).

3.1.1. Mizoram

3.1.1.1. Geographical location

Mizoram, one of the seven sister states of North-East India, is the southernmost landlocked part of the region covering an area of 21,087 km². Nestled amidst the picturesque hills of the Mizo Hills, it is known for its breathtaking landscapes, rich cultural heritage, and vibrant tribal population. The state lies between 21°56'N to 24°31'N, and 92°16'E to 93°26'E long sharing inter-state borders with Assam and Manipur in the north and Tripura in the west. The state also shares an international border with Myanmar to the east and Bangladesh to the west and south. The tropic of cancer passes through the state of Mizoram in North Lungsai. The state's topography is characterized by steep hills and an interspersed range of valleys, undulating rugged terrain with an elevation range of 50 to slightly above 2000 m above sea level.

3.1.1.2. Climate

Mizoram has a pleasant climate, with an average of 11°C to 21°C in the winter and 20 °C to 30 °C in the summer. The state's climate pattern is moist tropical to moist sub-tropical. Monsoons impact the area, bringing heavy rain from May to September and minimal rain in the winter, making this time of year cold and dry. The state receives heavy rainfall with an average of 2500 mm to 3000 mm every year. The state's northwestern region experiences the most precipitation i.e., more than 3500 mm annually. The rainfall also rises southward with an increase in humidity.

3.1.1.3. Characteristics of the Soil

The state soils are young and immature and are sandy in nature and primarily loose sedimentary (Pachuau, 1994). The soil of Mizoram is entisols, ultisols and inceptisols (Singh and Dutta, 1989). The soils have high organic carbon richness, low in available phosphorous content, and moderately rich in available potash. The surface soils in the hilly region of Mizoram are characterized by their black color, leaching, low base content, high iron content, and predominantly acidic nature, with a pH ranging from 4.5 to 6.0. The surface layer of soil is composed of loam to clay loam, with the clay percentage becoming more as the soil depth increases. The fertility of the soil is significantly affected by cultivation practices, soil erosions, and landslides associated with heavy rainfall.

3.1.1.4. Vegetation and forest

The most prevalent type of forest is the tropical wet-evergreen forest and also includes other forest types like semi-evergreen forests and tropical-moist deciduous forests with the occurrence of bamboo forest in pockets. The state has a forest cover that spans 17,820 km², accounting for 84.5 % of its overall geographical area. The state's forest cover has declined from 85.41 % (ISFR, 2019) to 84.53 % (ISFR, 2021) resulting in a loss of approximately 186 km² compared to the previous ISFR (2019) assessment. The forest cover assessment as per the ISFR (2021) is shown in the following Table 3.1.

The most common species found in this forest are *Schima wallichii*, *Castanopsis purpurella*, *Quercus acitissima*, *Duabanga grandiflora*, *Quercus semiserrata*, *Elaeocarpus serratus*, Garcinia spp, *Michelia champaca*, *Rhus semialata* etc. Bamboo is prevalent in the state, usually found alongside other forest vegetation types. Approximately 9,245 km² (44 %) of the state's total area is covered by bamboo. Bamboos often thrive as an undergrowth beneath tree species in tropical evergreen and sub-tropical mixed-deciduous forests. However, *Melocanna baccifera* creates dense or pure forests in specific regions within the State. Bamboo forests primarily occur along river banks and on abandoned jhumland, where they serve as the dominating secondary vegetation. Tree ferns, aroids, palms, ferns, orchids, bryophytes, and lichens are also found in the state's forest.

Forest cover	Area (km ²)	Geographical area (%)				
Very Dense Forest	156.79	0.74				
Moderately Dense Forest	5,715.24	27.11				
Open Forest	11,948	56.68				
Total	17,820.00	84.53				
Scrub	1	0.00				

Table 3.1. The forest cover of Mizoram (As per ISFR, 2021)

3.1.1.5. Agriculture

In Mizoram, agriculture has always been a livelihood primarily focused on meeting one's own basic needs, with over 50 % of the population engaged in this occupation. Out of the entire land, 21 % is dedicated to paddy/seasonal crops. Up to 63 % of the entire crop area is dedicated to shifting agriculture. Despite the utilization of the same method of shifting cultivation, which involves cutting down trees and burning them to prepare the land for farming, the same plot of land has been repeatedly used instead of moving to a different location. This is due to the scarcity of available land. The crops cultivated in the jhum are intermingled. The primary crop cultivated is paddy, with other crops including maize, cucumber, beans, ginger, mustard, sesame etc. In addition to that, the state also cultivates tapioca, sugarcane, cotton, legumes, and oilseeds. The oilseed crops, namely sesame, mustard, and soybean, are thriving. Paddy accounts for nearly 50 % of the overall crop area, and food grains occupy more than 88 % of the total crop area.

3.1.1.6. Demography

As per the 2011 Census of India (COI), Mizoram's population is 1,097,206, with 555,339 (50.61 %) being males and 541,867 (49.39 %) being females. In 2011, the population of Mizoram accounted for 0.09 % of the total population of India. The population experienced a growth rate of 47.89 % and had a population density of 52 individuals km². The state's sex ratio was 976, above the national average of 940. In Mizoram, there are 5,25,435 individuals (47.89 %) residing in rural regions and 5,71,771 individuals (52.11 %) residing in urban areas. The literacy rates of Mizoram stand at 91.33 %. The male literacy rate is 93.35 %, while the female literacy rate is 89.27 %. Mizoram has a total of 8,48,175 literate individuals, with males being 4,38,529 and females comprising 4,09,646 (Statistical handbook, Mizoram 2022).

3.1.2. Aizawl

3.1.2.1. Geographical location

Aizawl (21°58'N to 21°85'N and 90°30'E to 90°60'E), one of the state's eight districts as well as the largest city and the capital of Mizoram, is situated in the north of the Tropic of Cancer in the northern part of the state. The district is bordered to the east by Champhai district and Manipur state, to the west by Mamit district and Kolasib district, to the north by Cachar district of Assam state, and to the south by Serchhip district. The Aizawl district spans a total geographical area of 3576 km², which represents 16.96 % of the entire geographical area of the state. It is nestled among the Mizo Hills, which are part of the Eastern Himalayas and is perched on a ridge 1,132 meters above sea level, with the valleys of the Tlawng and Tuirial rivers to its west and east, respectively. Aizawl stands as the political, cultural, and economic heart of the state of Mizoram. This city, with its unique blend of modernity and tradition, offers a captivating introduction to the beauty and diversity of Mizoram.

3.1.2.2. Climate

Aizawl experiences a pleasant and moderate climate throughout the year greatly influenced by its location in the Eastern Himalayas, which contributes to its moderate temperatures and lush green landscapes. In summer and winter, the ambient air temperature ranges from 20 to 30° C and 11 to 21° C, respectively (Rai and Chutia, 2014). The climatograph of the study area during the study period is given in the following Figure 3.2.

3.1.2.3. Soil characteristics

The soils of Aizawl are extremely deep, dark yellowish to dark brown, with a surface that is highly acidic and a subsurface that is also highly acidic, clay loam to clay, well-drained, hillside slopes that have significant erosion, and a variety of other characteristics (Colney and Nautiyal, 2013).

3.1.2.4. Forest and vegetation

The district has a forest cover that spans 3064.9 km², accounting for 85.7 % of its overall geographical area. The forest cover of Aizawl districts is shown in the following Table 3.2. The predominant forest cover type in the district of Aizawl is mostly classified as Tropical Wet Evergreen Forest and Montane Subtropical Forest, with bamboo scattered throughout. The Tropical Wet Evergreen Forest accounts for 45.2 % of the total forest cover in Aizawl, while the Mixed Forest covers 31.3 %. A significant portion of the Aizawl forests consists of bamboo, with the Bamboo Forest occupying 16.7 % of the total area.

The most common species found are *Michelia champaca*, *Trema orientalis*, *Albizzia chinensis*, *Emblica officinales*, *Gmelina arborea*, *Bombax ceiba*, *Terminilia myriocarpa*, *Mesua ferrea*, *Toona ciliata*, *Schima wallichii*, *Acrocarpus fraxinifolius* etc. and bamboo species like *Melocanna bacciferra*, Dendrocalamus spp are also found.

Forest cover	Area (km ²)	Geographical area (%)				
Very Dense Forest	30.3	0.84				
Moderately Dense Forest	1062.7	29.7				
Open Forest	1971.9	55.11				
Total	3064.9	85.65				

Table 3.2. The forest cover of Aizawl (As per ISFR, 2021)

3.1.2.5. Demography

As per to the 2011 Census of India, the population of Aizawl was 4,00,309. Women comprise 50.22 % of the total population, while men account for 49.78 % of the population. The majority of the population is comprised of Mizos from several tribes. The majority of the city's population, approximately 93.63 %, adheres to Christianity. Additional minority religions include Hinduism with a population percentage of 4.14, Islam with 1.52, Buddhism with 0.45, Others with 0.09 %, Sikhism with 0.03 %, and Jainism with 0.02 % while 0.11 % of individuals did not disclose their religious affiliation. The literacy rate of the city was 97.89 %.

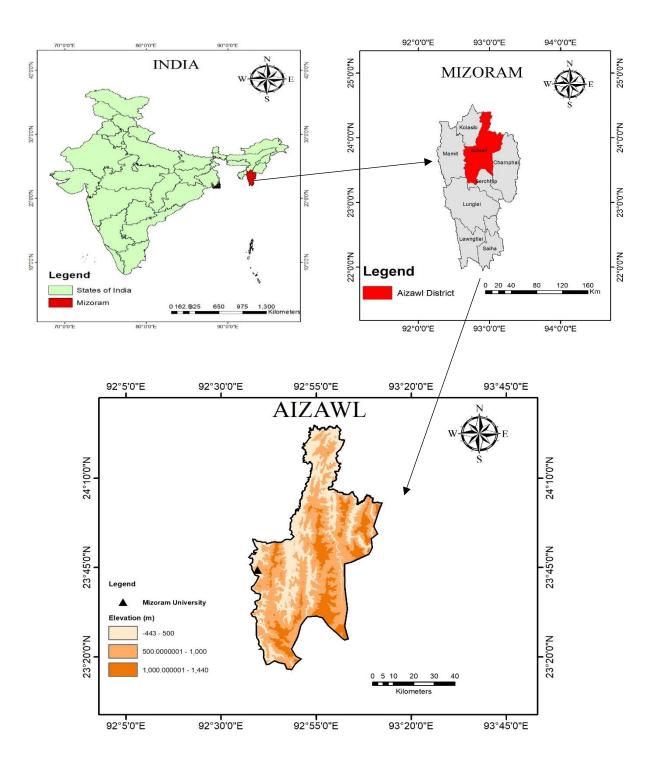


Figure 3.1. Map of the Study area.

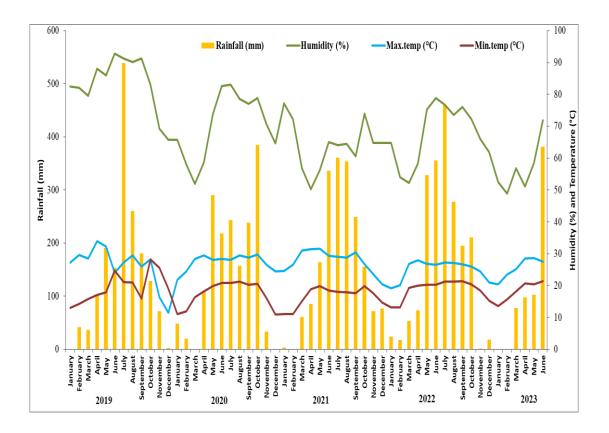


Figure 3.2. Climatograph of the study site during the study period (Source: Department of Science and Technology, Aizawl, Mizoram)

3.2. Species Description

Three abundantly found leguminous weed species viz: *Crotolaria micans*, *Aeschynomene indica*, and *Calopogonium mucunoides* were selected to evaluate their green manuring potential. The weed species selected were cross-checked with their accession number from the Botanical Survey of India, Shillong. The description of the species is given below:

3.5.1. Crotolaria micans Link.

Kingdom: Plantae Phylum: Spermatophyta Class: Dicotyledonae Order: Fabales Family: Fabaceae Genus: *Crotolaria* Species: *micans* Link.



Crotolaria micans Link. commonly known as Shining rattlepod, is an erect perennial shrub belonging to the family Fabaceae. It can grow to a height of 3 - 4 m and have angular branchlets. The leaves are alternate-spiral, trifoliate, oblong-oblanceolate in shape. Inflorescence racemes, yellow petals twice as long as the calyx, and flowering starts from July-August. Fruits are oblong-terete pod and transversely nerved, 3-5 seeds, and they fruit throughout the year. The shrub exhibits a high level of tolerance against a diverse array of climatic and soil conditions. This plant is indigenous to tropical America but has now become naturalized and is commonly spotted growing alongside roadways. The shrub is used as a green manure and cover crop in coffee, tea, tobacco, and rice farms and is suitable for cultivating to prevent erosion.

3.5.2. Calopogonium mucunoides Desv.

Kingdom: Plantae Phylum: Spermatophyta Class: Dicotyledonae Order: Fabales Family: Fabaceae Genus: *Calopogonium* Species: *mucunoides* Desv.



Calopogonium mucunoides Desv. commonly known as Calopo is a flowering species belonging to the Fabaceae family. It is a vigorous, hairy annual, creeping, short-lived perennial herb that attains up to 3-5 m tall and they usually tangled into a mass of foliage. The leaves are alternate and trifoliate; leaflets are 4-10 cm x 2-5 cm, elliptical and ovate, densely pubescent, the base is round or oblique. Axillary pseudo-racemes inflorescence; blue-purplish flower, small caducous bracts, and bracteoles, unequal 5-lobed calyx. The fruits are oblong-linear, flattened; 3-8 quadrangular seeds (3mm long), reddish brown. *C. mucunoides* primarily thrives in humid tropical regions, ranging from sea level to an elevation of 2000 meters above sea level. It is particularly prevalent in regions that have been disrupted, such as disturbed areas, forest borders, roadsides, and streams. It is also found in agricultural grounds where it thrives as a weed (Cook *et al.*, 2005).

3.5.3. Aeschynomene indica L.

Kingdom: Plantae Phylum: Spermatophyta Class: Dicotyledonae Order: Fabales Family: Fabaceae Genus: *Aeschynomene* Species: *indica* L.



Aeschynomene indica L. commonly known as Joint vetch is a branched, erect herbaceous or shrubby plant belonging to the family Fabaceae and can grow up to 80-90 cm tall. Fine tuberculate-based hairs cover the immature twigs. The leaves are alternate and evenly compound, narrow, with 8-40 pairs of oblongs, glabrous, and sensitive leaflets. The inflorescence is an axillary, short raceme (2-6 flowers), and flowers are pale yellow- with reddish longitudinal striations. The fruits are long linear to slightly curved legumes (3-5 cm long) with transverse articulations and 8-12 articles; seeds are kidney-shaped. It is a freely nodulating nitrogen-fixing species and is utilized as green manure. The plant commonly thrives in moist and marshy environments, such as floodplains and swamps, and is also recognized as being found on terrestrial surfaces. It thrives in environments that have been disrupted, such as roadside ditches, and often exhibits characteristics of a weed. It has medicinal uses and is frequently employed for the treatment of cold, cough, and fever, is used as an antidote for snake bites, jaundice, and leprosy (Kavitha and Yasodamma, 2023).

CHAPTER 4

BIOMASS PRODUCTION AND NUTRIENT ACCUMULATION OF THE LEGUMINOUS SPECIES

4.1. Introduction

In the quest for sustainable agricultural practices that promote soil health and reduce the dependency on synthetic fertilizers, green manure has emerged as a valuable ally for farmers worldwide. Green manuring is a technique that involves adding a significant amount of plant material to the soil, which gradually enhances the nutrient levels and organic matter. This process promotes nutrient cycling and prepares the soil for future harvests. Nevertheless, Cherr *et al.* (2006) assert that while the use of green manures in agriculture is economically viable and helps mitigate environmental damage, its potential is hindered by a lack of understanding regarding its intricate nature and the interplay between green manures, management practices, and various soil conditions.

Shoot biomass and nutrient content play a crucial role in maintaining the nutritional balance within the soil-plant system throughout nutrient cycling. The success of green manuring depends on several factors, including the quality of residues produced, the amount of nutrients released during decomposition, and the timing of nutrient release in relation to the crop's needs (Matos *et al.*, 2008).

Leguminous species vary greatly in their biomass yield and uptake of nutrients, which must be related to each species' unique characteristics, how crop residues are handled, and the soil and climate conditions at the time (Pereira *et al.*, 2016). The biomass produced by the legumes, which in turn depends on how well the legumes have adapted to their surroundings, determines how much system improvement there was after the introduction of legumes (Douxchamps *et al.*, 2012). Since dry mass production is connected with the capacity to increase nutrients through symbiosis with microbes, soil cover, and nutrient recycling, it stands out among the optimal characteristics for species selection for green manure (Teodora *et al.*, 2015). The amount of biomass, N accumulation, and rate of nitrogen release into the accessible forms are significantly influenced by the selection of an appropriate species of legume

and the age at termination (Hirpa *et al.*, 2009). However, the timing of green manure crop integration influences the amount of plant biomass and N if the green manure responds to variations in the weather (Cherr *et al.*, 2006).

Crotolaria juncea, Arachis pintoi, Stylosanthes guianensis, Calopogonium mucunoides, and *Stizolobium materrimum* are just a few examples of the many species of leguminous plants that have an exceptional capacity to produce large amounts of biomass and accumulate high nutrient concentrations that become available to crops after litter decomposition (Matos *et al.,* 2008). In 50-60 days, the most prolific green manure crops produced 4-5 t ha⁻¹ of dry biomass (Irin and Biswas, 2021). Many leguminous plants growing as weeds in forest and agricultural lands can be significant sources of nutrients trapped in their biomass which can be effectively utilized for improving soil health. The present chapter highlights biomass production and nutrient accumulation by three leguminous such weed species viz: *Crotolaria micans, Aeschynomene indica* and *Calopogonium mucunoides* having potential to be utilized as green manure crops.

4.2. Materials and Methods

4.2.1. Experimental details

In order to assess the biomass production and nutrient accumulation of the leguminous weed species, seeds of the selected species were collected from the wild, roadside, and in and around the Mizoram University Campus. The study was carried out for consecutive two-year field experiment (April – June of 2021 and 2022) at the Horticulture Research Farm, Mizoram University. The experiment was conducted using a Randomized Block design, with five replications for each species. The soil was prepared for sowing of the leguminous seeds using typical agricultural procedures, which including ploughing and mild harrowing and preparing a raised bed for sowing the seeds. However, no fertilizer was applied. The total number of experimental plots was 15 (each plot is a replication for each species), and each plot had dimensions of 1.5 m^2 , and the replications were spaced 0.5 m apart from each other.

Seeds of the selected legumes were treated before sowing viz: (a) *C. micans* and *A. indica* seeds were treated with 95 % H_2SO_4 for 5 minutes and (b) *C. mucunoides* seeds were soaked in cold water for 24 hours. Seeds were sown manually in line spaced 30 x 10 cm at a depth of 1-2 cm during the last week of March.

4.2.1. Biomass sampling

Sampling of biomass per plot of the leguminous plants was done at 90 days after sowing (DAS). The leguminous plants from each plot were uprooted carefully in order to ensure the least amount of root loss. The plants were cut posteriorly at the base of the stem to separate the shoot from the root system, and each component was weighed separately and recorded as fresh weight. Sub-samples from each plot were collected and kept in a hot air oven at 60° C until constant weight was obtained to determine the dry weight for determining shoot and root dry biomass. the biomass production for each species was represented on per hectare basis.

4.2.2. Growth parameters

To evaluate the growth performance, 10 individual plants harvested as above from each plot were selected randomly. The parameters viz., shoot length, root length, basal diameter, and number of primary branches per plant were recorded. Shoot and root length were measured using a measuring tape and the basal diameter was recorded using a calliper. The growth parameters were measured and represented on per-plant basis.

4.2.3. Nutrient analysis

The oven-dried shoot and root of each species mentioned above were milled and passed through a 1mm sieve for nutrient analysis. The samples were pooled together and made into triplicates separately for the shoot and root and further analysis were carried out. The macronutrient content in both shoot and root biomass was expressed in g kg⁻¹ and the micronutrient content was expressed in mg kg⁻¹ basis. The following procedures to determine the nutrient contents in both the shoot and root of the green manure legumes are described below:

4.2.3.1. Total nitrogen

The total nitrogen content in plants was determined by digesting 0.5 g of the plant samples with H₂SO₄ by the micro Kjeldahl method (AOAC, 1995).

4.2.3.2. Total phosphorous

The total P contents in the plant samples were determined by digesting the plant samples with a Diacid (3:1: Nitric acid: Perchloric acid) mixture (Koenig and Johnson, 1942). In this, 1 g of plant sample was taken in a conical flask where 10-15 ml of the diacid mixture was added. The flask was then put on a hot plate for digestion till the sample turned colorless, and the solution was filtered several times with hot water and the volume was made up. The P content was determined using a spectrophotometer at 660 nm.

4.2.3.3. Total potassium

The total potassium content was determined by digesting the plant samples in a triacid mixture (9:2:1) of HNO₃, H₂SO₄, and HCLO₄. The potassium content was read on a flame photometer.

4.2.3.4. Calcium, Magnesium and micronutrients (Zinc, Copper, Iron and Sodium)

The plant samples were digested by using a diacid mixture as prepared for the estimation of P mentioned above and the reading was taken using Atomic Absorption Spectrophotometer (Pinnacle 900 F).

4.2.3.5. Nutrient accumulation

Accumulation of macro and micronutrients was estimated by multiplying the nutrient content with the dry biomass and was expressed in kg ha⁻¹ and g ha⁻¹ for macronutrient and micronutrient respectively.

4.2.3.6. Statistical Analysis

The data obtained were subjected to Analysis of Variance (ANOVA) using the IBM-SPSS statistics, version 25 and the means were compared by Duncan's Multiple Range Test at a significance threshold of 5 %.

4.3. Results

4.3.1. Growth parameters of the legume species

C. micans recorded the highest shoot length (91.29 cm) among the three species, however, the shoot length of *C. mucunoides* (58.50 cm) and *A. indica* (47.51 cm) were at par (Table 4.1). There was no significant difference in the root length (Table 4.1). The maximum collar diameter was recorded from *C. micans* (7.09 mm), followed by *A. indica* (3.83 mm). and *C. mucunoides* (2.56 mm). Number of branches plant⁻¹ was maximum in *C. micans* followed by *A. indica* and *C. mucunoides* with value 27.00, 16.38 and 11.86 respectively.

While the shoot length varied significantly (p < 0.05) among the species but showed no significant changes in the first and second year (Figure 4.1). However, for the root length there was no difference among the species as well as the cropping year (Figure 4.1). While the collar diameter was found to vary significantly among the species and the cropping year. Nonetheless, in case of no. of branches, there was no significant difference between the cropping year, but a significant variation (p < 0.05) was observed among the species (Figure 4.2).

Table 4.1. Growth parameters of the leguminous weed species (Average pooled data of two consecutive years); n=10.

Species	Shoot length	Root length	No. of branches	Collar		
	(cm)	(cm)	plant ⁻¹	diameter (mm)		
C. micans	91.29ª	29.04ª	27.00ª	7.09 ^a		
C. mucunoides	58.50 ^b	24.29 ^{ab}	11.86 ^b	2.56 ^b		
A. indica	47.51 ^b	22.77 ^{bc}	16.38°	3.83 ^b		

Values that share the same alphabets along the column are not significantly different (p < 0.05).

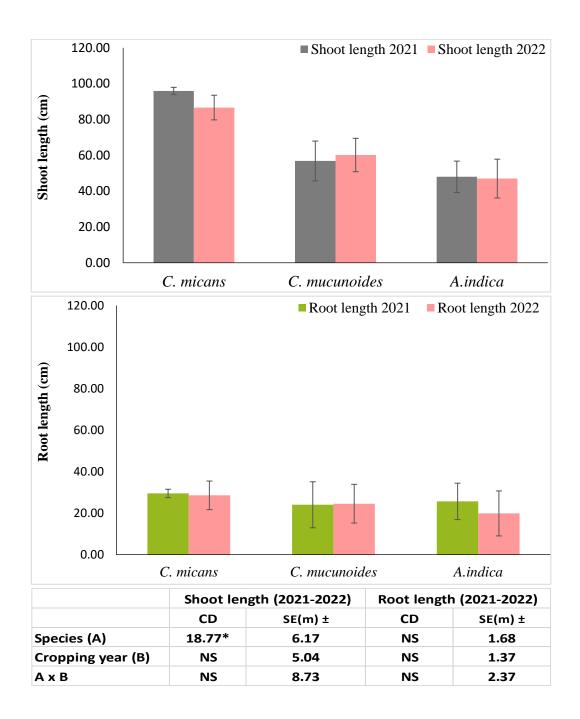


Figure 4.1. Shoot and root length of the leguminous species during the first and second year.

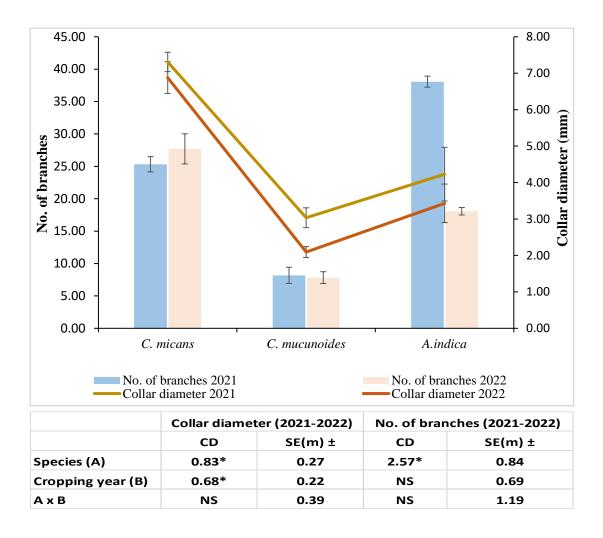


Figure 4.2. Collar diameter and no. of branches produced by the leguminous species during the first and second year.

4.3.2. Fresh and dry biomass of shoot and root

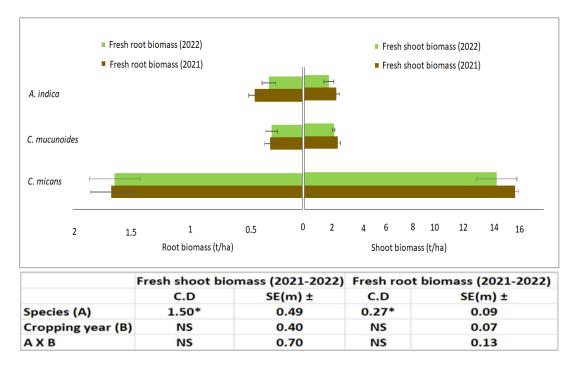
The fresh and dry biomass of the shoot and root showed significant difference among the species. However, the fresh and dry biomass of the shoot and root of *C*. *mucunoides* and *A*. *indica* were at par (Table 4.2). The fresh and dry biomass of shoot ranged from 2.1 to 15.16 t ha⁻¹ and 0.58 to 2.90 t ha⁻¹ respectively which was in the *C*. *micans* > *C*. *mucunoides* > *A*. *indica*. However, the fresh and dry biomass of the root was in the order *C*. *micans* > *A*. *indica* > *C*. *mucunoides*, value ranging from 0.58 to 2.90 t ha⁻¹ and 0.14 to 0.58 t ha⁻¹ respectively.

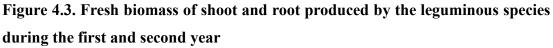
The fresh and dry shoot and root biomass was observed to vary significantly among the species, however showed no variation in the cropping year (Figure 4.3 and 4.4). Furthermore, first year biomass was higher than second year biomass.

	Fresh bioma	ass (t ha ⁻¹)	Dry biomass (t ha ⁻¹)				
Species	Shoot	Root	Shoot	Root			
C. micans	15.16 ^a	1.65ª	2.90 ^a	0.58ª			
C. mucunoides	2.36 ^b	0.28 ^b	0.68 ^b	0.14 ^b			
A. indica	2.10 ^b	0.36 ^b	0.58 ^b	0.16 ^b			

Table 4.2. Fresh and dry biomass of the leguminous weed species for two consecutive years.

Values that share the same alphabets along the column are not significantly different (p < 0.05)





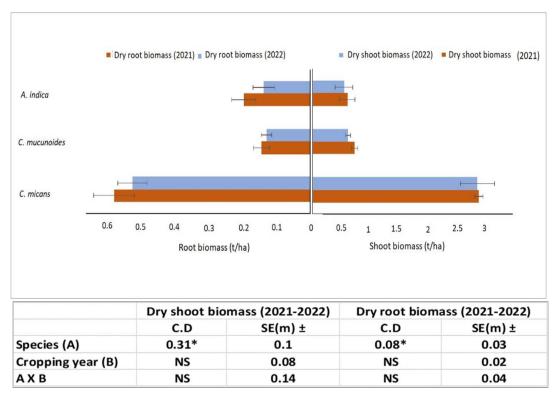


Figure 4.4. Dry biomass of shoot and root produced by the leguminous species during the first (2021) and second year (2022).

4.3.3. Macronutrient content

Table 4.3 shows the macronutrient contents of the leguminous species for consecutive two year as well as the pooled data. *C. micans* demonstrated significantly higher nitrogen concentration in both the shoot (28.25 g kg⁻¹) and root (21.46 g kg⁻¹), in both the cropping year.

C. micans exhibited significantly higher phosphorus content in the shoot and root tissues with a value of 5.19 g kg⁻¹ and 4.46 g kg⁻¹ respectively (pooled data). However, the pooled data for the phosphorous content in the shoot biomass showed that the P content of *C. mucunoides* and *A. indica* were at par. The phosphorous content in the root biomass of *C. micans* and *C. mucunoides* were at par in the first and second year (Table 4.3).

The shoot of *C. micans* demonstrated the highest K concentration, measuring 21.85 g kg⁻¹ (pooled data). Contrarywise, *C. mucunoides* revealed the highest K content in the root, measuring 16.79 g kg⁻¹ (pooled data).

The shoot biomass of the plants had a Ca content ranging from 5.68 to 6.77 g kg⁻¹ (pooled data) with *A. indica* displaying the highest concentration. Conversely, the root showed a Ca content ranging from 0.32 to 2.17 g kg⁻¹ (pooled data) with *C. mucunoides* exhibiting the highest concentration.

Regarding the magnesium, it was observed that *C. mucunoides* exhibited the highest in the shoot with value of 0.22 g kg⁻¹ (pooled data) whereas *C. micans* displayed the highest concentration in the root biomass with value of 1.83 g kg⁻¹ (pooled data).

Macronutrient content (g kg ⁻¹)															
Species		Р			Ν			K			Ca			Mg	
	Shoot														
	2021	2022	Pooled	2021	2022	Pooled	2021	2022	Pooled	2021	2022	Pooled	2021	2022	Pooled
C. micans	5.24 ^a	5.13ª	5.19ª	29.40ª	27.10 ^a	28.25ª	22.18ª	21.51ª	21.85ª	6.62ª	6.06 ^b	6.34	0.14 ^c	0.11°	0.12°
C. mucunoides	4.87 ^b	4.18°	4.52 ^b	15.18°	13.61°	14.40°	19.76 ^b	18.83 ^b	19.30 ^b	5.83 ^b	5.53°	5.68	0.23 ^b	0.21 ^b	0.22 ^b
A. indica	4.56 ^c	4.57 ^b	4.57 ^b	21.52 ^b	21.04 ^b	21.28 ^b	16.89°	15.44 ^c	16.17°	6.92 ^a	6.63ª	6.77	0.17 ^a	0.16 ^a	0.17 ^a
								Root							
C. micans	4.53 ^a	4.40 ^a	4.46 ^a	21.91ª	21.01 ^a	21.46 ^a	11.07 ^b	12.58 ^b	11.83 ^b	0.34 ^c	0.31°	0.32 ^c	1.83 ^a	1.77 ^a	1.80 ^a
C. mucunoides	4.29 ^a	4.11ª	4.20 ^b	10.33°	10.76°	10.55°	17.60ª	15.98ª	16.79ª	2.23ª	2.12ª	2.17ª	0.65°	0.62°	0.63°
A. indica	3.48 ^b	3.37 ^b	3.43°	15.15 ^b	13.86 ^b	14.51 ^b	11.23 ^b	11.34°	11.29 ^b	1.91 ^b	1.83 ^b	1.87 ^b	1.64 ^b	1.59 ^b	1.62 ^b

Table 4.3. Macronutrient content in the shoot and root biomass of the leguminous species; n=3.

Values that share the same alphabets along the column are not significantly different (p < 0.05)

4.3.4. Micronutrient content

There was considerable variation in the micronutrient concentrations among the leguminous weed species in both the cropping year except for the Zn content in the shoot biomass showed that the Zn content of *C. micans* and *C. mucunoides* were at par in both the cropping year and the Zn content of *C. micans* and *A. indica* in the root biomass were at par (Table 4.4). Interestingly it was observed that for all the species the micronutrient contents were higher in the root biomass when compared to the shoot biomass.

The pooled data for the Na content varied from 123.22 mg kg⁻¹ in *C. micans* to 211.08 mg kg⁻¹ in *C. mucunoides* in the shoot biomass and 638.91 mg kg⁻¹ in *C. mucunoides* to 820.65 mg kg⁻¹ in *C. micans*.

C. mucunoides displayed notably elevated levels of Fe content both in the shoot and root biomass, with value of 974.28 and 1971.33 mg kg⁻¹ respectively (pooled data).

The Zn content in the shoot and root biomass varied from 25.09 to 38.53 mg kg⁻¹ and 58.74 to 78.58 mg kg⁻¹ respectively (pooled data).

C. micans exhibited the maximum Cu content in the shoot whereas *C. mucunoides* recorded the highest content in the root biomass.

				Mic	ronutrient c	ontent (mg	kg ⁻¹)					
		Na			Fe			Zn			Cu	
						Shoot						
Species	2021	2022	Pooled	2021	2022	Pooled	2021	2022	Pooled	2021	2022	Pooled
C. micans	130.07°	116.37°	123.22°	521.78 ^b	513.28 ^b	517.53 ^b	26.23 ^b	25.30 ^b	25.77 ^b	16.13ª	14.87ª	15.50ª
C. mucunoides	216.80ª	205.37ª	211.09ª	991.90ª	956.67ª	974.28ª	27.55 ^b	22.63 ^b	25.09 ^b	12.65 ^b	13.58ª	13.12 ^b
A. indica	166.37 ^b	160.60 ^b	163.48 ^b	481.28°	467.27°	474.28°	40.67ª	36.38ª	38.53ª	10.28°	9.93 ^b	10.11°
						Root						
C. micans	833.17ª	808.13ª	820.65ª	1634.93°	1658.83°	1646.88°	57.95°	59.53°	58.74°	32.10 ^c	37.92 ^b	35.01°
C. mucunoides	648.68°	629.13°	638.91°	1978.55ª	1964.10ª	1971.33ª	84.47ª	72.68ª	78.58ª	65.85ª	64.57ª	65.21ª
A. indica	688.17 ^b	650.05 ^b	669.11 ^b	1851.13 ^b	1817.27 ^b	1834.20 ^b	63.78 ^b	57.80 ^b	60.79 ^b	36.62 ^b	38.62 ^b	37.62 ^b

Table 4.4. Micronutrient content in the shoot and root biomass of the leguminous species; n=3

Values that share the same alphabets along the column are not significantly different (p < 0.05)

4.3.5. Nutrient accumulation

C. micans demonstrated statistical superiority (p < 0.05) in terms of macronutrient accumulation (Table 4.5), specifically for N, P, K, Ca and Mg with values of 81.86, 15.03, 63.31, 18.37, 0.37 kg ha⁻¹ respectively and also exhibited the maximum micronutrient accumulation (Na, Fe, Zn and Cu) with values 357.03, 1499.89, 72.49, 44.92 g ha⁻¹ respectively, in the shoot biomass (Table 4.6). Furthermore, *C. micans* had a greater capacity for N (12.42 kg ha⁻¹), P (2.56 kg ha⁻¹), K (6.84 kg ha⁻¹), Mg (1.04 kg ha⁻¹), Na (474.88 g ha⁻¹), Fe (952.94 g ha⁻¹), Zn (33.99 g ha⁻¹) and Cu (20.25 g ha⁻¹) accumulation in the root biomass. Nevertheless, *C. mucunoides* exhibited a notable increase in the accumulation of Ca in the root biomass with a recorded value of 0.30 kg ha⁻¹ (Table 4.5). The Ca content in the shoot biomass of *C. mucunoides* and *A. indica* were at par therefore no significant difference was observed (Table 4.5).

Macronutrients accumulation in the shoot and root biomass differed significantly among species and cropping years, with the exception of potassium accumulation in the root biomass, which did not show significant variation among cropping years (Figure 4.5 and 4.6).

Figures 4.7 and 4.8 demonstrated that the accumulation of micronutrients differed among species and over different years of cropping. Nevertheless, the buildup of Cu did not differ significantly among the species or cropping years.

		Macronutrie	nt (kg ha ⁻¹)		
Species	Ν	Р	K	Ca	Mg
			Shoot		
C. micans	81.86 ^a	15.03ª	63.31ª	18.37ª	0.37ª
C. mucunoides	9.86°	3.10 ^b	13.18 ^b	3.88 ^b	0.15 ^b
A. indica	12.41 ^b	2.69°	9.44°	3.95 ^b	0.10 ^c
			Root		
C. micans	12.42 ^a	2.56ª	6.84ª	0.19°	1.04ª
C. mucunoides	1.43°	0.58 ^b	2.28 ^b	0.29 ^b	0.09°
A. indica	2.36 ^b	0.56 ^b	1.82°	0.30 ^a	0.26 ^b

 Table 4.5. Macronutrient accumulation in the shoot and root biomass of the leguminous

 species (pooled data of two consecutive years)

Values that share the same alphabets along the column are not significantly different (p < 0.05)

 Table 4.6. Micronutrient accumulation in the shoot and root biomass of the leguminous

 species (pooled data of two consecutive years)

Micronutrient (g ha ⁻¹)				
Species	Na	Fe	Zn	Cı
			Shoot	
C. micans	357.03 ^a	1499.89ª	72.49ª	44.92 ^a
C. mucunoides	144.19 ^b	665.02 ^b	18.28°	8.91 ^b
A. indica	95.33°	276.52°	23.08 ^b	5.89°
			Root	
C. micans	474.88 ^a	952.94ª	33.99 ^a	20.25ª
C. mucunoides	86.50°	266.74°	11.63 ^b	9.04 ^b
A. indica	108.69 ^b	296.77 ^b	10.67°	6.51°

Values that share the same alphabets along the column are not significantly different (p < 0.05)

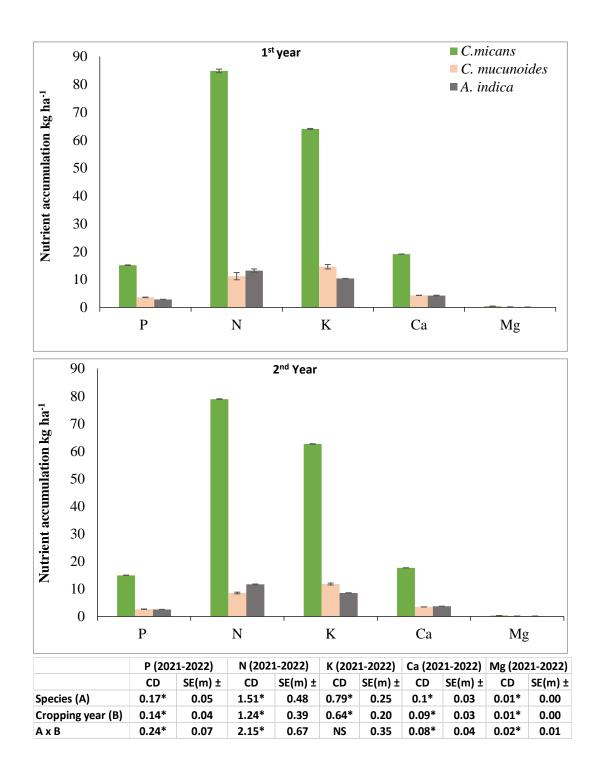


Figure 4.5. Macronutrient accumulation in the shoot biomass of the three leguminous species during the first and second year.

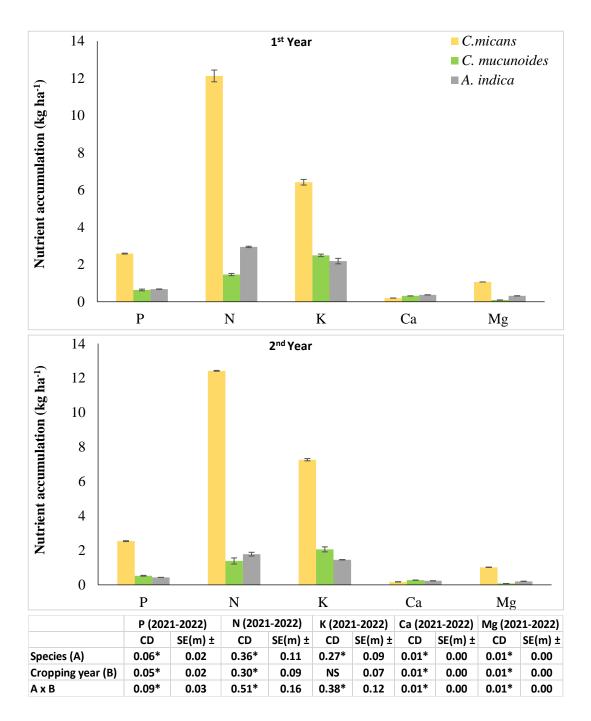


Figure 4.6. Macronutrient accumulation in the root biomass of the three leguminous species during the first and second year.

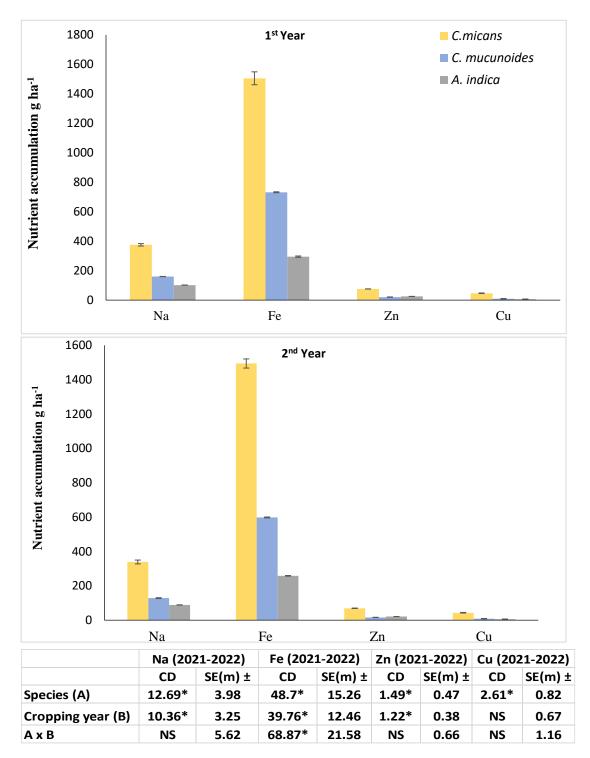
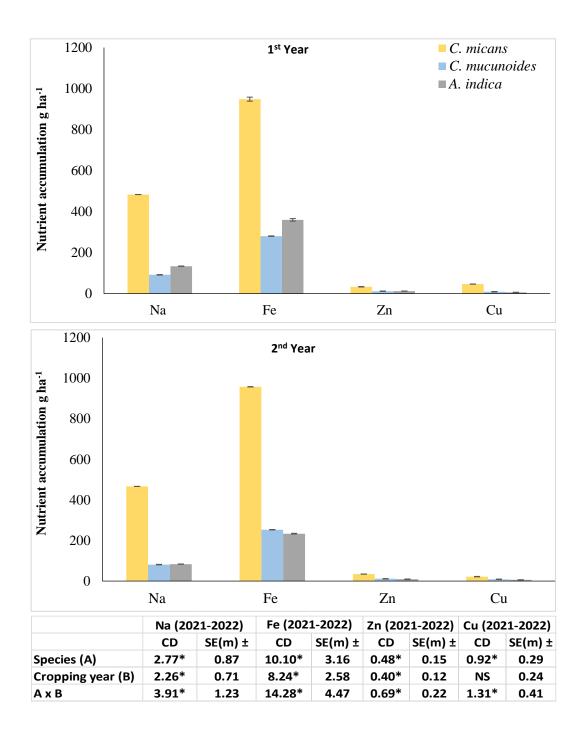
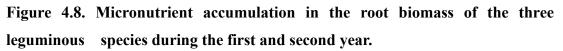


Figure 4.7. Micronutrient accumulation in the shoot biomass of the three leguminous species during the first and second year





4.4. Discussion

4.4.1. Growth parameters

Plant growth and development can be determined by major morphological traits such as plant heights, number of leaves, leaf length and width, stem diameter, and number of branches. Several variables constrain the growth and development of a crop. The optimal environmental conditions and continuous soil fertility are the primary factors that contribute to achieving the maximum potential of the crop in terms of enhanced growth and production. Variations in the growth parameters of the leguminous species during the study period likely resulted from the unique genetic composition of the species. Various environmental elements, such as soil moisture, temperature, and the physical, chemical, and biological properties of the soil, can influence the growth of roots (Fageria, 2009). The data pertaining to the growth factors, namely shoot and root length, collar diameter and number of branches revealed that C. micans recorded the maximum in terms of growth parameters. In their study, Duarte et al. (2013) examined various green manuring crops and observed that Crotolaria juncea exhibited a greater height (129.3 cm) compared to the other crops throughout its flowering stage. Similarly, Irin and Biswas (2021) reported that C. juncea exhibited the greatest height compared to the others. Similarly, Nitisha and Girjesh (2013) stated that the maximum plant height of 111.60 cm was observed in Sesbania spp. at a density of 50 plants pot⁻¹ at 45 DAS. Pramanik *et al.* (2009) obtained similar results, reporting that Sesbania had the tallest plants among different green manuring crops, with S. rostrata being the tallest, followed by S. aculeata and C. juncea.

4.4.2. Biomass production

There was a substantial disparity in the biomass of the shoots and roots and our finding revealed that *C. micans* exhibited more biomass accumulation, likely due to its rapid and predictable development pattern, resulting in enhanced soil fertility. This can also be related to its distinct morphological traits, as indicated in Table 4.1. *Crotolaria* genus is usually believed to produce substantial biomass, typically exceeding 8 Mg ha⁻¹, that makes them to be utilized in farming as a component of crop rotation systems (Menezes *et al.*, 2009; Perin *et al.*, 2010; Soratto *et al.*, 2012). There have been several previous studies that have provided evidence that *Crotolaria* species have the potential to produce dry matter (Torres *et al.*, 2005; Mattar *et al.*, 2015; Chilagne *et al.*, 2018). dos Santos Nascimento *et al.* 2021 reported in their study that *Crotolaria ensiformis* had the highest values for shoot dry biomass production (1645.8 kg ha⁻¹) compared to the other plant species studied and also confirmed that plants belonging to the Fabaceae family exhibit substantial biomass production due to their capacity for nitrogen fixation.

According to Manuhuttu *et al.* (2014), plant fresh weight (canopy) is determined by the growth and accumulation of plant tissue, including factors such as the number of leaves, leaf area, and plant height. These factors are regulated by the water and nutrient content in the cells of the plant tissue. Duarte *et al.* 2013 confirmed that the shoot fresh biomass reported in *C. ensiformis* and *C. juncea* legumes can be attributed to variables such as nodulation and variations in root development among plants, which likely contributed to the observed variation in the results. Nevertheless, Pramanik *et al.* (2009) and Chand *et al.* (2015) found that *Sesbania* had a notably greater fresh shoot biomass in comparison to *Crotolaria*.

The biomass production of crops is influenced by several factors including genotype, sowing date, management strategies, soil and meteorological conditions, growing season, and plant population (Da Silva *et al.*, 2020). The variations in biomass among the examined species can also be attributed to the region's light availability, temperature, and climatic conditions. Odhiambo *et al.* (2010) supported the aforesaid statement. Environmental conditions significantly impact crop growth and biomass

production. Temperature and water availability are two environmental factors that have a substantial impact on several physiological, biochemical, and molecular processes, ultimately affecting the buildup of biomass (Hasanuzzaman et al., 2013; Rahman et al., 2020). In their study, Thomas and Palaniappan (2012) noted that certain leguminous green manure crops, such as Crotolaria juncea, are affected by changes in day length. They also noticed that these crops have a limited vegetative development period due to shorter days and colder temperatures. And also, additionally in the absence of competition, plant populations under normal conditions will experience an increase in plant biomass (Feichtinger et al., 2004; Daudu et al., 2006). The presence of other weeds may have also impacted the biomass production of the legumes, particularly due to their cultivation during the rainy season, when competition with weeds is more pronounced. However, it may not certainly have affected the overall biomass yield. Favero et al. (2000) observed that the presence of weeds had no impact on the yield of shoot dry weight, indicating that the total biomass output was not reduced by the presence of weeds. Hansch and Mendel (2009) and Pereira et al. (2016) proposed that assessing total plant biomass is a reliable criterion that can serve as an indication of plant growth.

4.4.3. Nutrient content and accumulation

The differences in nutrient content in the above-ground and below-ground biomass of the leguminous weed species seen in this study, can be related to genetic changes within the species, as well as the time of harvest of the crop. The higher N values seen in *C. micans* can be ascribed to the plant's exceptional effectiveness in fixing biological nitrogen through its root nodules. However, the N content in the shoot biomass of *C. micans* (28.25 g kg⁻¹), *C. mucunoides* (14.40 g kg⁻¹) and *A. indica* (21.28 g kg⁻¹) were below what was observed by Duarte *et al.*, 2013 for the N content among the green manure crops. Similarly, Pereira *et al.* (2007) also discovered a significant difference (p < 0.05) in the levels of N, P, K, Ca, and Mg among the cover crops. The phosphorus content in the legume under investigation was found to be greater compared to the legumes tested by Duarte *et al.* (2013) and Cazetta *et al.* (2005). The total N, P and K content (Shoot + root) of the species in our study was much higher

than the findings observed by Mauad *et al.* (2019) who observed 23.08, 3.04 and 27.38 g kg⁻¹ of N, P and K content in *Crotolaria spectabilis* after 98 days after emergence.

The micronutrient content among the species was as follows: Fe > Na > Zn > Cu (Table 4.5). Fageria *et al.* (2002) and Fageria *et al.* (2016) both reported that the order of microelements concentration in crops was in the order Fe > Mn > Zn > Cu. Duarte *et al.* (2013) found that the concentration of Fe was higher and the concentration of Cu was the lowest in the aboveground parts of the green manure plants studied. Zandavakili *et al.* (2017) observed a range of 186–284 mg kg⁻¹, 21-23 mg kg⁻¹ and 13-38 mg kg⁻¹ for Fe, cu and Zn respectively in the root biomass of four green manure crops which was lower than the range observed in our study. The primary component of utmost significance is iron (Fe), predominantly found in the roots. This occurrence arises from the adsorption of oxide particles onto the surface of the roots, making it challenging to eliminate during the sampling procedure (Mauad *et al.*, 2019) and this might be the reason of higher Fe content in the root biomass when compared to the shoot biomass in the present study.

The leguminous species exhibited notable variations in nutrient accumulation in their shoot and root biomass, illustrated in Table 4.6. *C. micans* exhibited significant superiority in the accumulation of all the nutrients except for Ca in the root biomass which may be because of its highest dry biomass output that leads to the highest nutritional accumulation among the legumes studied. The study conducted by Castro *et al.* (2004) found that *C. juncea* had the highest total N levels of 126 kg ha⁻¹ before eggplant cultivation. Pereira *et al.* (2016) recorded a nitrogen accumulation of approximately 377 kg ha⁻¹ at 92 days, while Duarte *et al.* (2013) observed an accumulation of around 175.8 kg ha⁻¹ N during the complete flowering season.

The varying nutrient accumulation in the shoot and root biomass of the leguminous species in this study and other studies may be attributed to genetic variations in the species. The study determined that *Crotolaria micans* had the most favourable growth performance, biomass production, and nutrient accumulation throughout a 90-day period after being sown.

4.5. Conclusion

In the light of results obtained from the findings it can be concluded that, the legume species exhibited a significant accumulation of biomass and nutrients, highlighting their capacity for nutrient cycling within the agricultural system. The growth parameters revealed significant differences among the studied species, with *C. micans* displaying superior performance in terms of shoot length, root length, collar diameter, and number of branches. Biomass production assessments and nutrient analysis underscored *C. micans* exceptional capacity to accumulate substantial amounts of fresh and dry biomass, contributing significantly to soil organic matter and nutrient.

CHAPTER 5

DECOMPOSITION AND NUTRIENT RELEASE PATTERN OF THE LEGUMINOUS WEED SPECIES

5.1. Introduction

Decomposition is a physiochemical and biological process that replenishes the soil with nutrients and organic matter from plant vegetative components that act as a source of nutrients for plant growth. In terrestrial contexts, more than 50% of net primary production is recycled into the soil through decomposition (Wardle *et al.*, 2004). Plant residues include a substantial amount of organic materials and nutrients, however, the benefits are only realized after decomposition and subsequent nutrient release, which is controlled by their biochemical quality (Anguria *et al.*, 2017).

Due to its capacity to absorb sizeable amounts of nutrients into the soil through decomposition and nutrient release from biomass, the use of leguminous green manure can be an alternative for the agricultural systems in the area (Pereira *et al.*, 2016). Legumes employed as green manure break down their biomass to release vital chemical compounds into the soil (Da Costa *et al.*, 2019; Lalremsang *et al.*, 2022). Green manure's source of nutrients comes from mineralization after mass decomposition when it is chopped or mowed (Wattier *et al.*, 2019). The chemical properties of the biomass influence the breakdown and nitrogen mineralization processes (Radicetti *et al.*, 2017) such as the content of C and N notably during the first week of degradation (Trinsoutrot *et al.*, 2000). However, long-term mineralization of C and N is influenced by the presence of more resistant components, such as cellulose (Hadas *et al.*, 2004), the content of lignin (Deb *et al.*, 2005) and lignin:N ratio (Vahdat *et al.*, 2011).

The breakdown of legumes is affected by several elements, such as the prevailing climate and the soil condition (Perin *et al.*, 2010; Matos *et al.*, 2011; Kumar *et al.*, 2021), the specific legume species and its chemical composition (Talgre, 2017; Karki *et al.*, 2022; Li *et al.*, 2022; Mangaravite *et al.*, 2023;) and the type and amount of decomposing micro-organisms (Garcia-Palacios *et al.*, 2016; Berg *et al.*, 2010; Thonnissen *et al.*, 2000). Legumes offer biomass with a low C:N ratio, resulting in

quick N mineralization after incorporation and good impacts on N nutrition even in the early stages of growth of the subsequent crop (Radicetti *et al.*, 2017). Furthermore, leguminous plants have a high rate of mass breakdown and N mineralization if the C:N ratio is approximately 25 and the lignin concentration is less than 15% (Palm *et al.*, 2001). Temperature, moisture content, microbial activity, soil pH, aeration status, and texture all influence the rates of C and N mineralization (Havstad *et al.*, 2010; Ha *et al.*, 2008;) and also depend on the organic matter (Chaves *et al.*, 2004). Therefore, the present chapter aims to ascertain the decomposition and nutrient release pattern of three commonly available leguminous weed species of Mizoram, in order to evaluate their suitability as green manure crops.

5.2. Materials and Methods

5.2.1. Collection and preparation of plant samples

The decomposition study of the leguminous weed species was undertaken using litterbag technique (Bocock and Gilbert, 1957). Fresh plant samples of the leguminous weed species were collected by harvesting the plants from the wild at their peak vegetative stage along with the roots. The collected plant samples were air-dried for about a week and then chopped into small pieces. 10 g of oven dry equivalent airdried samples were measured with the help of a weighing balance and placed in nylon mesh bags of size 20 cm x 20 cm with a mesh size of 1 mm x 1 mm. 60 bags were prepared for each species and placed randomly in 15 different plots (5 plots for each species) on the experimental site. And in each plot 13 bags were enclosed. The bags were buried at 10 cm soil layer in the month of March 2020 and the decomposition process was monitored. On a random basis, one bag from each plot for each species were retrieved at monthly intervals. After recovery, the litter bags were transported to the laboratory where, adhering residual materials and the contents of each bag were carefully brushed and rinsed to remove soil and other extraneous materials, and the remaining material was oven dried at 60° C to a constant weight and the dry weight was recorded.

5.2.2. Chemical analysis of the plant samples

The chemical analysis of the samples was undertaken before placing the plant samples in the litter bag and after retrieval of the litter samples from the experimental plots. The oven- dried samples were powdered and sieved for chemical analysis. The dry mass from the recovered litter bags per species were pooled together for chemical analysis. All the analysis for the monthly nutrient contents were carried out in triplicates.

5.2.2.1. Total phosphorous and Potassium: Total P contents in the plant samples were determined by digesting the plant samples with a Diacid (3:1: Nitric acid: Perchloric acid) mixture (Koenig and Johnson, 1942) and the P content was determined using a spectrophotometer at 660 nm. The total potassium content was determined by digesting the plant samples in a triacid mixture (9:2:1) of HNO₃, H₂SO₄, and HCLO₄. The potassium content was read on a flame photometer.

5.2.2.2. Nitrogen and Carbon: C and N contents of the plant samples were determined by using CHNS analyzer (Elementar vario Marco cube).

5.2.2.3. Lignin, Cellulose and Hemicellulose: The initial lignin, cellulose and hemicellulose contents of the plant samples for each species was determined by the Van Soest method (Van Soest *et al.*, 1991).

5.2.2.4. Total Ash: The initial ash content of the plant samples was determined by taking 3 g of the grounded samples into a pre-weighed silica crucible. The silica crucible along with the samples undergoes decarbonization by heating on flame till no smoke is emitted, then the decarbonized samples are transferred to a muffle furnace and ignited at 500 - 600 °C for 3 hours till the sample turned whitish in color. Then the crucibles with ash was cooled in a desiccator and the weight was recorded. The ash % was calculated by the formula:

Ash $\% = \frac{(Weight of silica crucible with ash-weight of empty crucible)}{weight of sample} \ge 100$

5.2.2.6. Total phenol: Analysis of the plant samples for total phenol content s was carried out by Folin- Ciocalteu reagent and the phenolic content was measured at 650 nm colorimetrically (Bray and Thorpe, 1954).

5.2.2.7. Crude fibre: The crude fibre content was determined using the method described by Maynard (1970).

5.2.3. Computation

The decay constant of the selected species was determined by using the negative exponential decay model (Olson, 1963): $k = -\ln (X/X_0)/t$, where, X is weight remaining in the end of the study period, X₀ is initial dry weight of the sample, k is decay rate constant; and t is the time period. The time required to achieve 50 % (t₅₀) and 99 % (t₉₉) decay of the samples were calculated as t₅₀ = 0.693/k and t₉₉= 5/k.

The monthly mass loss was calculated by the formula: $(X/X_0) \times 100$, where X is the weight remaining in the end of the study period, X_0 is initial dry weight of the sample.

The nutrient content of the decomposing species was derived by the formula: % Nutrient remaining = $(C/C_0) \times (DM/DM_0) \times 100$, where C is the concentration of nutrients in the time of sampling, C₀ is the initial nutrient concentration, DM is the mass of plant sample at the time of sampling, DM₀ is the initial mass that was kept for decomposition (Bockheim *et al.*, 1991). The nutrient release constant (k_N, k_P and k_K) was calculated using same exponential decay model $k = -\ln (X/X_0)/t$ where, X is nutrient remaining at the end of the study period, X₀ is initial nutrient stock of the sample, k is decay rate constant; and t is the time period.

5.2.4. Statistical analysis

A one-way ANOVA was conducted to compare the means of the initial plant chemistry and decay rates and the time required to decompose 50 % and 99 % of the initial mass using IBM-SPSS statistics, version 25 and the means were compared by Duncan's Multiple Range Test. Pearson Correlation between the initial chemical compositions of the plant with the decay rate and nutrient release and also the decay rate with the climatic factors was carried out.

5.3. Results

5.3.1. Initial chemical composition of the plant samples

The initial composition varied among the leguminous species (Table 5.1). The initial total ash content ranged from 10.90 to 20.10 %, the highest was recorded in C. mucunoides and the lowest was in C. micans. For crude fiber, C. micans (16.22 %) recorded the lowest and A. indica the highest (21. 17 %). Initial lignin contents, was the highest in A. indica (16.98 %) and the lowest in C. mucunoides (7.55 %). For cellulose and hemicellulose, C. mucunoides showed the maximum with 32.19 % and 29.19 % respectively. The highest initial N content was observed in A. indica (3.11%), followed by C. micans (3.04 %), and the lowest in C. mucunoides (1.96 %). As for P the maximum content was observed C. micans (0.27 %), while the minimum in C. mucunoides (0.17 %). For C content ranged from 33.67 to 38.12 %. The K content was the highest in C. micans (1.55 %) and the lowest was found in C. mucunoides (0.88 %). The total phenolic content of A. indica was found to be the highest (1.8 mg/100g). The C/N ratio was in the order C. mucunoides (17.22) > C. micans (12.52) > A. indica (11.41). The L/N ratio ranged from 2.45 to 6.5. The N/P ratio was found to be in the order A. indica (12.33) > C. mucuinoides (11.74) > C. micans (11.17) however, C/P ratio was in the order C. mucunoides (200.23) > A. indica (140.68) > C. micans (139.79). Initial (L + TP)/N ratio ranged 4.24 to 6.03.

Initial Components	C. micans	C. mucunoides	A. indica
Total ash (%)	$10.90^{\rm c}\pm0.01$	$20.10^{\mathtt{a}}\pm0.01$	$15.36^{\text{b}}\pm0.00$
Crude Fiber (%)	$16.22^{\text{b}}\pm0.00$	$17.18^{\circ}\pm0.00$	$21.74^{\rm a}\pm0.00$
Lignin (%)	$12.71^{b} \pm 0.01$	$7.55^{\rm c}\pm0.04$	$16.98^{\rm a}\pm0.00$
Cellulose (%)	$18.39^{\rm c}\pm0.21$	$32.19^{\mathtt{a}}\pm0.01$	$20.22^{\text{b}}\pm0.01$
Hemicellulose (%)	$16.29^{\rm c}\pm0.00$	$29.19^{\mathrm{a}}\pm0.00$	$19.70^b\pm0.00$
N (%)	$3.04^{\text{b}}\pm0.01$	$1.95^{\rm c}\pm0.01$	$3.11^{a} \pm 0.00$
P (%)	$0.27^{\rm a}\pm0.00$	$0.17^b\pm0.00$	$0.25^{\rm a}\pm0.00$
K (%)	$1.55^{\rm a}\pm0.00$	$0.88^{\rm c}\pm0.00$	$1.46^{\text{b}}\pm0.00$
C (%)	$38.12^{\mathrm{a}}\pm0.00$	$33.67^{\rm c}\pm0.01$	$35.50^{b} \pm 0.01$
Total phenol (mg/100g)	$0.68^{\text{b}}\pm0.01$	$0.73^{b} \pm 0.01$	$1.80^{\rm a}\pm0.03$
C/N	$12.52^{\text{b}}\pm0.03$	17.22 ^a ±0.06	$11.41^{\rm c}\pm0.01$
L/N	4.17 ^b 0.01	$3.86^{\rm b}\pm0.01$	$5.45^{\mathrm{a}} \pm 0.00$
N/P	$11.17^{\rm c}\pm0.12$	$11.74^{\text{b}}\pm0.13$	$12.33^{a} \pm 0.10$
C/P	$139.79^{b} \pm 1.37$	$200.23^{\mathtt{a}}\pm3.91$	$140.68^{b}\pm 1.01$
(L + TP)/N	$4.40^{\text{b}}\pm0.01$	$4.24^{b}\pm0.01$	$6.03^{\mathrm{a}}\pm0.01$

Table 5.1. Initial chemical composition of the leguminous species before the decomposition period ($n=5; \pm SE$).

Values across the rows that share the same alphabet are not significantly different at p < 0.05.

5.3.2. Decomposition pattern and Decay rate constant

The decomposition pattern of all the species exhibited significant variation, as shown in Figure 5.1. During the first month (30 days), it was observed that *C. micans* showed a reduction of around 45 % in its dry mass. During the 3^{rd} month, approximately 90 % of the initial mass was gradually lost. This was followed by a rather slow phase of disintegration until the last incubation period, as depicted in Figure 5.1a. However, during the first month, *C. mucunoides* and *A. indica* recorded a loss of approximately 38.16 % and 35 % of their dry mass, respectively. In the second month, it was observed that 50 % of the dry mass of *C. mucunoides* and *A. indica* was lost, as depicted in Figure 5.1 b and c. *C. mucunoides* experiences a reduction of around 90% in its dry mass by the fifth month, but A. indica exhibited a gradual decomposition pattern after the second month. At the end of the decomposition period (180 days), approximately 1.8 % (0.18 g), 2.85 % (0.25 g), and 6.64 % (0.66 g) of the initial mass of *C. mucunoides*, and *A. indica*, respectively, remained as undecomposed dry material (Table 5.2).

The decay rate constant varied significantly among the three leguminous species and the values ranged from 0.015 to 0.021 k day⁻¹ (5.50 to 8.2 k year ⁻¹) with *C. micans* having the highest decay rate followed by *C. mucunoides* and *A. indica*. The maximum days required for 50 % and 99 % of the dry mass to decay was recorded in *A. indica* with the value of 46 and 331.89 days respectively and the minimum period was taken by *C. micans* with 32. 99 and 238.04 days respectively (Table 5.3).

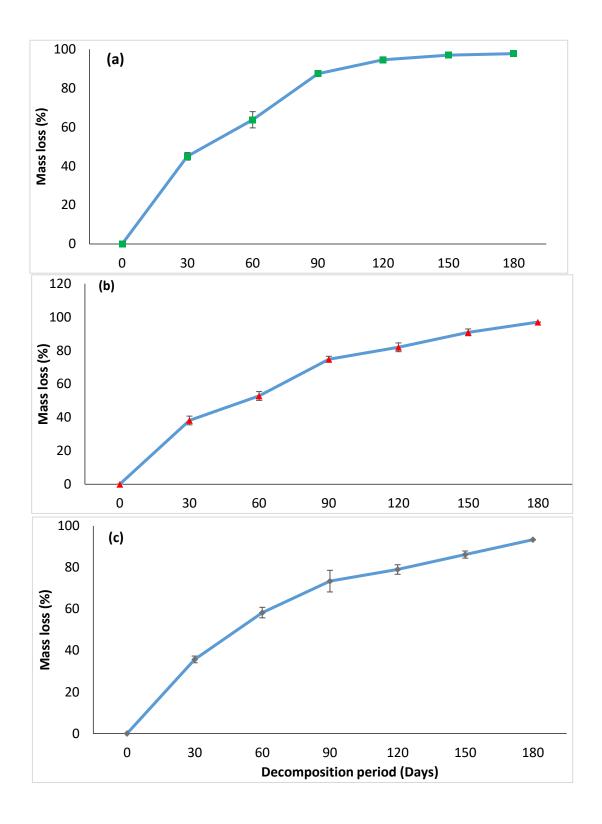


Figure 5.1. Mass loss (%) of (a) *C. micans*, (b) *C. mucunoides* and (c) *A. indica* during the decomposition period.

Decomposition Days	C. micans	C. mucunoides	A. indica
0	10	10	10
30	5.39 ± 0.19	$\boldsymbol{6.18\pm0.26}$	6.03 ± 0.13
60	3.72 ± 0.42	4.72 ± 0.27	4.17 ± 0.25
90	1.25 ± 0.13	2.51 ± 0.18	2.66 ± 0.52
120	0.54 ± 0.15	1.80 ± 0.26	2.10 ± 0.23
150	0.30 ± 0.08	0.91 ± 0.20	1.38 ± 0.17
180	0.18 ± 0.02	0.30 ± 0.00	0.66 ± 0.02

Table 5.2. Dry mass remaining (g) of the three leguminous species during the decomposition period; $n=5, \pm$ (SE).

Table 5.3. Decay rate constant and the days required for 50 % and 99 % mass loss of the three leguminous weed plants. (n = 5).

	Decay parameter					
Species	k day-1	k year-1	t ₅₀ (days)	t99 (days)		
C. micans	0.021ª	8.21ª	32.99°	238.04°		
C. mucunoides	0.019 ^b	7.15 ^b	34.96 ^b	252.24 ^b		
A. indica	0.015°	5.50°	46.00ª	331.89ª		

Value that shares the same alphabets along the column are not significantly different (p < 0.05).

5.3.3. Nutrient release pattern

5.3.3.1. N Dynamics

The concentration of N in the decomposing plant samples of *C. mucunoides* decreased gradually with time (Figure 5.2a). The concentration of N in *C. micans* and *A. indica* decreased rapidly in the initial (0-30 days) and then increased with time (60-120 days) and then decreased again. The study revealed that the liberation of N from the three legumes commenced during the initial portion of the incubation period (Figure 5.3a). The release of N from *A. indica* ($k_N = 0.005 \text{ day}^{-1}$) was comparatively slower than *C. micans* (0.006 day⁻¹) and *C. mucunoides* (0.006 day⁻¹) as shown in Table 5.3. The N mass remaining (% of initial) during the decomposition period are provided in Figure 5.3a. At the conclusion of the incubation period N mass remaining (%) was 0.69 %, 1.50 % and 3.33 % for *C. micans*, *C. mucunoides* and *A. indica* respectively (Figure 5.3a).

5.3.3.2. P dynamics

The observed trend in Figure 5.2b indicates a reduction in the concentration of phosphorus (P) in the plant residues of *A. indica* as time progressed. In the case of *C. mucunoides*, the concentration of phosphorus (%) exhibited an initial increase until 60 days, followed by a subsequent drop during the course of 60-180 days. The P concentration (%) of *C. micans* exhibited a decline during the first month, followed by an upward trend in the second month. Subsequently, the concentration continued to drop until the final day of the decomposition period (Figure 5.2b). The release of P in *C. micans* was faster as compared to the other two species (Figure 5.3b). The k_P value was 0.007 *k* day⁻¹ (2.45 *k* year⁻¹), 0.003 *k* day⁻¹ (0.95 *k* year⁻¹) and 0.005 *k* day⁻¹ (1.88 *k* year⁻¹) for *C. micans*, *C. mucunoides* and *A. indica* respectively (Table 5.4). The P mass remaining (%) were 0.72 %, 2.72 % and 3.41 % for *C. micans*, *C. mucunoides* and *A. indica* respectively (Figure 5.3b).

5.3.3.3. K Dynamics

The K concentration decreased rapidly after 30 days of incubation till 90 days across all three species. Subsequently, there was an increase in the K concentration for *C. micans* at 120 days and then decreased again, and in the case of *C. mucunoides* and *A. indica* the concentration of K increased till 150 days followed by a gradual reduction until the conclusion of the incubation period. The potassium concentration (%) in the plant residues at the conclusion of the decomposition period, was found to be 0.23 %, 0.211 %, and 0.245 % for *C. micans*, *C. mucunoides* and *A. indica* respectively (Figure 5.2c). The percentage of K mass remaining in the plant residues for *C. micans*, *C. mucunoides*, and *A. indica* were found to be 0.32 %, 1 %, and 1.31 % correspondingly, as shown in Figure 5.3c. The $k_{\rm K}$ value ranged from 0.008 (2.81 $k_{\rm K}$ year⁻¹) to 0.01 $k_{\rm K}$ day⁻¹ (3.86 $k_{\rm K}$ year⁻¹) as shown in Table 5.3.

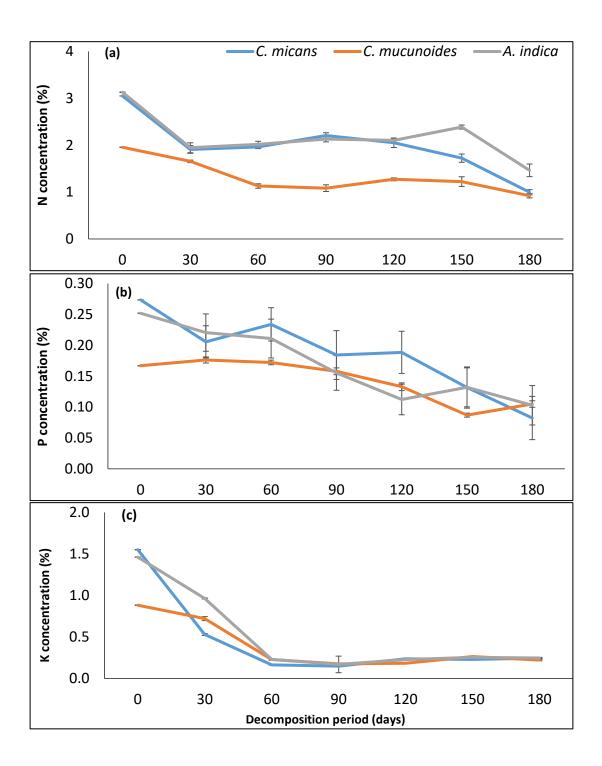


Figure 5.2. N (a), P (b) and K (c) concentration (%) during the decomposition period of the three leguminous weed species.

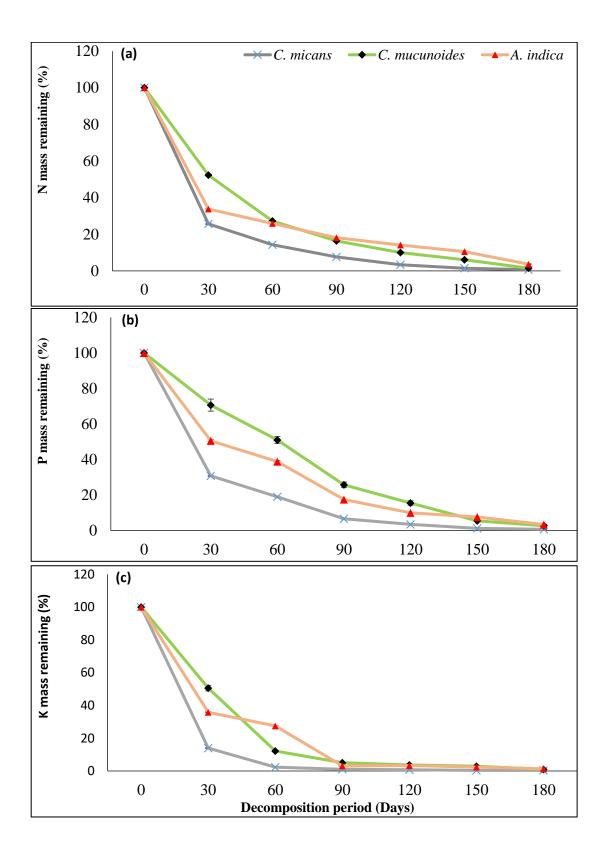


Figure 5.3. Variations in the percentage of N (a), P (b) and K (c) mass remaining (%) of the three leguminous weed species during the decomposition period.

Species	$k (\mathrm{day}^{-1})$	<i>k</i> (year ⁻¹)	t ₅₀ (days)	t99 (days)
			Ν	
C. micans	0.006ª	2.31ª	109.71°	791.6°
C. mucunoides	0.006 ^b	2.01 ^b	126.64 ^b	913.68 ^b
A. indica	0.005°	1.74°	145.95 ^a	1053.02ª
			Р	
C. micans	0.007ª	2.45 ^a	104.19 ^b	751.76 ^b
C. mucunoides	0.003°	0.95°	274.03ª	1977.13ª
A. indica	0.005 ^b	1.88 ^b	134.57 ^b	970.94 ^b
			K	
C. micans	0.011ª	3.86ª	65.58 ^b	473.15 ^b
C. mucunoides	0.008 ^b	2.81°	90.07ª	649.86ª
A. indica	0.01°	3.62 ^b	69.91 ^b	504.43 ^b

The value that shares the same alphabets along the column are not significantly different (p < 0.05).

5.4. Correlation between decay rate and nutrient release rates (N, P and K) with the initial chemical compositions of the species.

Decay rates of the three species showed significant negative correlation with crude fiber, lignin, total phenol and with the ratios of N/P, L/N and (L+Total phenol)/N (Table 5.3).

N release rate showed a significant positive correlation with lignin, N, P, K, total phenol and the ratio of L/N and (L+TP)/N while negative correlation with C/N and C/P ratio and the cellulose and hemicellulose contents. However, a strong negative correlation was observed between hemicellulose, cellulose and the ratio of C/N and C/P and a significant positive correlation between C, N, P and K with k_P and k_K respectively (Table 5.5).

Chemical composition	Decay rate	k N	<i>k</i> _P	<i>k</i> _K
Crude fibre	-0.93**	0.58	-0.07	0.15
Lignin (%)	-0.69**	0.93**	0.59	0.81*
Cellulose (%)	0.17	-0.86**	-0.93**	-0.98**
Hemicellulose (%)	0.03	-0.79*	-0.95**	-0.97**
Carbon (%)	0.33	0.56	0.94**	0.89**
Nitrogen (%)	-0.33	0.91**	0.85**	0.96**
Phosphorous %	-0.10	0.84**	0.94**	0.97**
Potassium %	-0.16	0.85**	0.92**	0.97**
Total phenol (mg/100g)	-0.98**	0.74*	0.14	0.38
C/P	0.09	-0.89**	-0.88**	-0.95**
C/N	0.45	-0.93**	-0.78*	-0.93**
N/P	-0.87**	0.23	-0.41	-0.10
L/N	-0.91**	0.80**	0.26	0.51
(L+TP)/N	-0.95**	0.76*	0.188	0.431

Table 5.5. Correlation between decay rate and the nutrient release rates of N, P and K with the initial chemical compositions of the species.

**Correlation is significant at the 0.01 level; *Correlation is significant at the 0.05 level.

5.5. Relationship between mass loss of the three leguminous species with the abiotic factor.

An analysis was conducted to examine the correlation between the remaining mass of different forms of litter of the three leguminous species and the abiotic factors (Table 5.5). The decay rate of *C. micans* was positively correlated with the rainfall, however the *C. mucunoides* and *A. indica* showed no relation with the rainfall. Humidity and the temperature showed positive correlation with all the mass loss of the litters of the three species.

 Table 5.6. Correlation between the mass loss of the three leguminous species with the abiotic factors.

Abiotic factors	C. micans	C. mucunoides	A. indica	
Rainfall (mm)	0.83*	0.74	0.75	
Humidity (%)	0.98**	0.95**	0.94**	
Temperature (°c)	0.89*	0.91*	0.94**	

**Correlation is significant at the 0.01 level; *Correlation is significant at the 0.05 level.

5.5. Discussion

5.5.1. Initial chemical characteristics of the plant residues

Plant residues that contain more than 2 % and 0.2 % of N and P, respectively, can be classified as high quality (Mafongoya *et al.*, 1998; Hoorman *et al.*, 2010). Considering the N and P levels, *C. micans* and *A. indica* in our study fall within the acceptable range and are thus classified as excellent quality. The initial N and P contents in our findings fall within the range reported by Mangaravite *et al.* (2023) for four green manure legumes. However, certain substances as lignin and polyphenols can regulate the accessibility of these nutrients (Gentile *et al.*, 2008). Residues that have initial C/N ratios of less than 20 and C/P ratios of less than 200 are classified as good quality (Horman *et al.*, 2010) and in our study, all three species fall in the range reported by Pereiria *et al.*, 2016 for 6 leguminous green manure plants. Green manure crops that have a lignin concentration below 15%, is believed to undergo rapid mass breakdown and N mineralization (Palm *et al.*, 2001).

5.5.2. Decomposition pattern

The decomposition pattern varied significantly across the species. Microorganisms play a crucial role in various ecological processes, including the decomposition of organic matter, nutrient cycling, preservation of soil structure and functionality, and facilitation of nutrient availability to plants via microbial succession. The pattern of mass loss for all three species varied significantly. The plant samples were interred in late February, when the soil temperature gradually rises following a period of severe cold, thereby providing optimal circumstances for fast decomposition. The presence of soil bacteria leads to colonization and hence an increased rate of degradation during the initial days of the incubation period following winter was observed. The decomposition process across the species were higher during the initial phase, followed by a subsequent slow period. Several other authors have reported this phenomenon where (Pandey *et al.*, 2007; Mubarak *et al.*, 2008; Das and Mondal, 2016; Bohara *et al.*, 2019; Hou *et al.*, 2021). Additional research has also documented an accelerated rate of decomposition during the early stages, primarily attributed to the

presence of sugars, amino acids, and proteins, which are classified as easily decomposable components. Earlier studies suggest that plant residues with a low biochemical quality tend to decompose slowly. This is because nutrients, particularly N, get trapped and unavailable during the initial stages of decomposition (Baggie *et al.*, 2005; Fosu *et al.*, 2007). However, in the later phases, the rate of decay decreases as more resistant substances such as lignin, tannins, and cellulose accumulate (Lupwayi *et al.*, 2004; Thonnissen *et al.*, 2000; Anguria *et al.*, 2017). It has been confirmed that the labile parts of litter provide a readily available source of energy for the decomposer; moreover, the nutrients are easily leached. Therefore, the nutrient concentration ought to be the crucial factor in determining the rate of breakdown during the early stages (Siqueira *et al.*, 2022).

The decomposition rates of plant residues vary due to the influence of climate. However, the chemical composition of the residue, including elements such as C, N, cellulose, hemicellulose, polyphenols, and lignin, is the most accurate indicator of decomposition at a local level (Adl, 2003). The findings of the study revealed a negative relationship between the mass loss and the initial levels of lignin, as well as the ratios of L/N (Table 5.5). The aforementioned factors were considered to be the most reliable indicators of litter decomposition rates, which aligns with the findings of Isaac and Nair (2006) and Upadhyaya et al. (2012). The lignin content of plant residues has traditionally been utilized as an indicator of organic matter quality. Litter with a lower concentration of lignin promotes the degradation process, as opposed to litter with higher lignin content (Lalremsang et al., 2022). The rate of decomposition can be attributed to the lignin content of the plant alone or the ratio of L/N (Butenschoen et al., 2014). The lignin concentration (Table 5.1) of C. micans (12.71 %) and C. *mucunoides* (7.56 %) was found to be lower than that of A. *indica* (16.98 %) in our study, which consequently accelerates their breakdown rate. In a study by Carvalho et al. (2010) confirmed that the lower rate of decomposition of Sunhemmp and pigeon pea was probably due to the higher content of Cellulose and lignin respectively when compared among the other Fabaceae species. Sharma et al., (2018) found that despite greater nitrogen content, lignin was identified as the primary factor limiting the degradation rate of Perilla leaf litter.

The overall phenolic concentrations in the plant material also function as an indicator of alterations in the litter's quality, as they inhibit microbial activity and cause protein to precipitate from the litter (Palm and Sanchez, 1990). The phenolic concentrations as well as the (L + TP)/N ratio in *C. micans* and *C. mucunoides* were found to be lower than *A. indica* (Table 5.1). This disparity in phenolic contents may explain the slower breakdown rate seen in *A. indica* as compared to the other two species investigated. The phenolic content has a notable impact on the breakdown of biomass and the accessibility of nitrogen for soil microorganisms, as they can rapidly combine with nitrogen (Hattenschwiler and Vitousek, 2000). Our study also revealed that the correlation coefficient ($R^2 = 0.98$) between the decomposition rate and the phenolic content were comparatively higher than those between the decomposition rate.

The initial concentration of N, C/N and C/P ratios significantly influence the decomposition of plant residues (Garcia- Palacios *et al.*, 2013; Ahirwal *et al.*, 2021). However, our study found no relation between the decomposition rate and the abovementioned compositions (Table 5.3). Frank *et al.* (2004) and Gorisen and Cotrufo (2000) also suggested that the ratio of C/N in residue does not significantly influence the rate of decomposition, instead, it serves as an indicator of the quality of the residue.

The decomposition of litter is significantly influenced by abiotic conditions such as rainfall, humidity, and temperature (Grzyp *et al.*, 2020; Giweta M. 2020; Boyero *et al.*, 2016). The current study found that litter decomposition of the three species was mostly influenced by humidity and temperature rather than rainfall. The study conducted by Wallenstein *et al.* (2010) and Conant *et al.* (2011) reveals that temperature plays a crucial role in regulating the rate of decomposition of plant debris. However, the faster breakdown seen in the litters of *C. micans* could also be attributed to the abundant rainfall, which likely promoted increased microbial activity, resulting in a quicker rate of decomposition. Across geographical gradients, research has shown that precipitation has a substantial impact on litter decomposition. It has been observed that wet locations have faster rates of litter decomposition compared to dry areas (Huang and Li, 2017). Decreased rainfall is also expected to decrease the activity and

composition of soil microbes (Kardol *et al.*, 2011; Zhang *et al.*, 2014; Santonja *et al.*, 2022), which negatively impacts decomposition.

5.5.3. Nutrient release pattern

Throughout the process of decomposition, there was significant variation in the release of nutrients from the decaying plant species. Plant residues that have lower C/N ratios, which are linked to lower levels of resistant organic compounds such as lignin, and have greater rates of nutrient mineralization, can supply significant amounts of nutrients for future crops (Swift *et al.*, 1979; Monteiro and Gama-Rodrigues, 2004).

N Dynamics

The decline in the nitrogen concentration from its original level in decaying plant residues can be due to leaching. The rapid decline in N concentration during the final stages of decomposition can be attributed to an increased demand for nitrogen due to strong microbial activity. The rapid release or immobilization of nitrogen may be directly associated with the chemical composition of different forms of litter, as demonstrated by Semwal et al. (2003) in various tree species found in the middle Himalayas, India. Legume residues typically exhibit a lower carbon-to-nitrogen (C/N) ratio in comparison to other tree species. A low C/N ratio is frequently linked to higher rates of mineralization (Brunetto et al., 2011). Moreover, it is easy for the Fabaceae plant residues to release N from biomass unless they contain substantial amounts of lignin and polyphenols (Palm and Sanchez, 1991; Constantinides and Fownes, 1994; Cobo et al., 2002). The N mineralization process of the various green manures showed a positive correlation with the overall N content in the mass, and a negative correlation with the cellulose and hemicellulose content as well as the C/N and C/P ratio (Table 5.5). The findings align with prior research conducted by Tosti et al. (2012), Halde and Entz (2016) and Watthier et al. (2020). At the end of incubation in the present study C. micans (0.99%) and C. mucunoides (0.92%) had a much lower percentage of nitrogen than A.indica (1.33 %), whereas A.indica had a much higher content of N remaining (3.33 %) than other plant residues (Figure 5.3a). This suggests that the plant residue of

C. micans and *C. mucunoides* degraded and released nitrogen more quickly than *A. indica* throughout the incubation period.

P Dynamics

The loss of phosphorus shown by three legume species during the early stages of decomposition could be due to the release of soluble P- containing compounds (Isaac and Nair, 2005). Palm et al. (2001) recommended that high nitrogen (> 2.5 %) and phosphorus (> 0.25 %) during decomposition enhance the mineralization of P, and in the present study the phosphorus content in *C. micans* was higher than *A. indica* and *C. mucunoides* (Table 5.1) which could be the reason for the faster release of phosphorus in *C. micans* as compared to the other two species. The fast mineralization of phosphorus (P) is linked to the depletion of soluble P that has accumulated in the vacuoles of plant tissues (Watthier *et al.*, 2020). The overall inorganic phosphorus content and soluble phosphorus in the residues, as well as the effective activity of microbes in the organic fractions directly influence the release of phosphorus (Giacomini *et al.*, 2003).

K Dynamics

During the decomposition period, *C. micans* and *A. indica* exhibited a nutrient release pattern of K > P > N, while *C. mucunoides* followed a pattern of K > N > P. Hasanuzzam and Hossain (2014) reported a comparable pattern of nutrient release (K > N > P) from the litter of agroforest tree species. After 180 days of decomposition, the highest proportion of released nutrients was observed for K in all species (Table 5.2). Prior research studies, including Lalremsang *et al.* (2022), Das and Mondal (2016), Brunetto *et al.* (2011), and Gomez-Munoz *et al.* (2014), have seen a similar release pattern, with K exhibiting the highest release rate. Additional studies undertaken in different soil and climatic conditions have also observed a significant release of K from biomass in various cover crops belonging to the Fabaceae family (Gama-rodriques *et al.*, 2007; Pereira *et al.*, 2016). The significant release in K concentration observed during the decomposition phase can be attributed to its high mobility and the leaching of water-soluble minerals (Jeong *et al.*, 2015; Patricio *et al.*, 2012). Furthermore, K is the predominant ion in plant cells, and its quick release can

be attributed to its presence in an ionic state, rather than being bound to any structural component of plant tissue (Mangaravite *et al.*, 2023).

5.6. Conclusion

The study provides valuable insights into the decomposition and nutrient release patterns of three leguminous weeds in Mizoram, with potential for use as green manure crops. Each species exhibited distinct decomposition patterns over the 180 days incubation period. *C. micans* decomposed most rapidly, followed by *C. mucunoid*es and *A. indica. C. micans* and *A. indica* released nutrients in the order of K > P > N, while *C. mucunoides* released them in the order of K > N > P. In summary, this study recommends the utilization of leguminous plants that are readily accessible in the local area as green manure, owing to their fast rate of decomposition.

CHAPTER 6

NODULATION BEHAVIOUR AND NITROGEN FIXATION OF THE LEGUMINOUS WEED SPECIES

6.1. Introduction

Nitrogen, an essential element for plant growth and development, often poses a limiting factor in agricultural productivity and to restore the nitrogen levels in the soil, methods such as organic amendments, incorporating agricultural residues, green manure, cover crops, and biological nitrogen fixation (BNF) can provide substantial amounts of usable soil nitrogen (De Oliveira *et al.*, 2017; Du *et al.*, 2020; Tang *et al.*, 2018; Xiao *et al.*, 2004).

Legumes are a notable reservoir of nitrogen (Quilbe *et al.*, 2021) commonly cultivated as green manuring and the purpose of these practices is to promote nutrient cycling through biological nitrogen fixation. Legumes are important in agroecosystems because they can form a symbiotic relationship with soil bacteria called rhizobia. This relationship allows the legumes to develop root nodules, which fix atmospheric nitrogen. This process is a key contributor to the global nitrogen cycle (Moulin *et al.*, 2001; Mortier *et al.*, 2012; Abd-Alla *et al.*, 2014). In addition to agricultural legumes, wild legumes (herbaceous and arboreal) form nodules that can perform nitrogen fixation, facilitate reforestation, and mitigate soil erosion (Ahmad *et al.*, 2020).

Only a small fraction, specifically 20 %, of all legume species and around half of legume genera have been scrutinized for nodulation (Ezrin *et al.*, 2010; Onyango *et al.*, 2011). In addition, an even smaller number of legumes have been tested for their real nitrogen fixing capacity. Members of the three sub-families exhibit varying abilities to facilitate nodulation. Among the species that have been studied, 97 % of the Papilionideae, 90 % of the Mimosoideae, and 23 % of the Caesalpinoideae have been seen to form nodules (Al-Fredan, 2011). In recent decades, several studies have investigated the process of nodule organogenesis at the molecular, cellular, and organ levels (Stacey *et al.*, 2006; Stougaard, 2000; Crespi and Galvez, 2000). However, there is a scarcity of data on the dynamics of nodule number and biomass throughout the growth cycle (Voisin *et al.*, 2010). Assessing the presence of nodules at different stages of development in a species can help determine the optimal timing for applying fertilizer, particularly because the size and quantity of nodules are closely linked to the process of atmospheric nitrogen fixation (Kashyap *et al.*, 2012). The promotion of legume production and the implementation of crop engineering techniques to enhance nitrogen fixation will decrease the reliance on synthetic nitrogen fertilizers and support the goals of sustainable agriculture in both the short and long term (Charpentier and Oldroyd, 2010). Therefore, an endeavour was undertaken to examine the growth and nodulation patterns of three commonly available leguminous weeds in Mizoram.

6.2. Materials and methods

6.2.1. Experimental details

A polypot experiment was carried out to determine the nitrogen fixation and nodulation behavior of the leguminous weed species. The experiment was laid out in a Completely Randomized design with 7 replications each for every interval for species. During the first week of March, seeds of the leguminous species were sown in polypots of 30 X 30 cm size filled with well sieved potting mixture. The nodule number and fresh and dry biomass of the nodules were observed in the first (April), second (May), third (June) and sixth month (September). However, the nitrogen fixation by the legumes was studied in the last month (September).

6.2.2. Potting mixture

The potting mixtures to be filled in the polypot consist of well sieved soil, sand and FYM in a ratio of 3:1:1 respectively. The soil used to fill the polypots were collected from the forest and a small portion of the samples were kept separately and air dried. The air-dried samples were grounded and sieved and analysed for the nitrogen content that was estimated by the micro-Kjeldahl method (Subbiah and Asija, 1957), soil pH (Jackson, 1973), soil organic carbon (Walkey and Black method, 1934), soil texture was determined by the hydrometer method (Bouyoucos, 1962). The characteristics of the soil used for the potting mixture are represented in the following Table 6.1.

Soil parameters			
Soil texture	Sandy loam (65.28 % Sand, 15.83 % Silt and 16.78 % Clay)		
Soil pH	5.10 ± 0.08		
Soil organic carbon (%)	1.93 ± 0.05		
Available N kg ha ⁻¹	328.91±2.29		

Table 6.1. Characteristics of the soil used for filling the polybags, ± SE (n=3).

6.2.3. Number of nodules

The number of nodules for each species was observed after 1st month, 2nd month, 3rd month and 6th month. To observe the number of nodules, the individual plants for each species were carefully removed from the polybag and the plant samples along with the soil surrounding the root system were transferred to a tub full of water. After a while, the soil surrounding the plant's roots was detached and settle down to the bottom of the tub. The plant roots were then again cleaned under running water and the rootlets were separated into category viz., primary, secondary, and tertiary. The nodules under each of the root category were counted individually for each species. The Total number of nodules for each individual plant for each species were obtained by pooling all the root nodules from each category.

6.2.4. Fresh biomass of nodules and Dry biomass of nodules

The nodules counted from different root category was then weighed with the help of electronic digital balance and fresh biomass recorded. The fresh biomass obtained from each root category were pooled to obtain the total fresh biomass of the nodules. After the fresh biomass for each category was noted, the nodules were oven dried at 60 °C for 24 hours to determine the dry weight. Dry biomass from each root category was pooled together to obtain the total dry biomass of the nodules.

6.2.5. Shoot and root length

The shoot and root length were observed at 1st month, 2nd month, 3rd month and 6th month. The measurement of shot length was conducted by extending from the collar area to the tip of the shoot using a measuring tape. Similarly, the length of the root was determined by measuring it from the collar to the tip using a scale.

6.2.6. Shoot and root biomass

The measurement of fresh biomass from both the root and shoot was conducted using an electronic digital balance. The fresh biomass samples, consisting of the root and shoot, were subsequently subjected to oven-drying in a hot air oven at a temperature of $65 \pm 5^{\circ}$ C until a consistent weight was achieved. This weight was then utilized to ascertain the dry biomass of both the root and shoot.

6.2.7. Nitrogen Content of the plant and root nodules

The total nitrogen content of the plant (both shoot and root) and the root nodules were assessed at the end of the study period (at 6th month). The plants were harvested and data for the fresh and dry weight of the shoot and root were recorded separately. The oven dried samples for each plant parts were grounded and sieved through 1mm sieve and analysis for the total nitrogen content was carried out. The total nitrogen content of the composite samples of shoot, roots and nodules was estimated by the CHNS Analyzer (Perkin Elmer, 2400 Series 2).

6.2.8. Nitrogen Content of soil

Soil samples were analyzed both before sowing of the legume seeds and also at the end of the study period. The samples for each species were collected at 6 months from top (0-5 cm) of the polypot and from near the root system after removing the plants from the polybags. The collected samples from each polypot for different species were air dried and composite samples of each species was analyzed to determine the available nitrogen content by the micro-kjeldhal method (Subbiah and Asija 1957).

6.2.9. Nitrogen fixation

At the conclusion of the experiment, when the plants were six months old, the nitrogen contents of the soil and plant samples (both root and shoot) were analysed to determine the species' capability for fixing nitrogen. The Nitrogen fixed by each plant was estimated by difference method (Kashyap *et al.*, 2012) as the sum of the enrichment of available nitrogen in soil and the utilization (uptake) of nitrogen by the plant. The figures were expressed in terms of kg/ha/yr. Cross-section of the polythene bag was used to determine the nitrogen fixation capacity of each species and the total amount of nitrogen fixed over a hectare (10,000 m²) was then calculated as follow

Nitrogen fixed (kg ha⁻¹ yr⁻¹)

$$= 12 x \left\{ \frac{\text{Nitrogen enrichment in soil}\left(\frac{\text{kg}}{\text{ha}}\right)}{\text{Age of the legumes (months)}} + \frac{\text{Nitrogen uptake by plant (mg)}}{\text{Area of cross section of polybag (m2)}} x \frac{10000 \times 10^{-6}}{\text{Age of the Legumes (Months)}} \right\}$$

The nitrogen uptake (mg plant⁻¹)

<u>= (Root dry biomass X Root N %) + (Shoot dry biomass X Shoot N %)</u> X 1000 100

Nitrogen enrichment in soil (kg ha⁻¹) = Available nitrogen (kg ha⁻¹) at 6^{th} month – Available nitrogen (kg ha⁻¹) before seed sowing.

6.2.10. Statistical analysis

Analysis of variance was done for the data recorded on the nodules produced, the weight of the nodules, nitrogen content of the various parts of the plants and the nitrogen fixed using the OP-STAT (Online statistical package: <u>http://14.139.232.166/opstat/</u>). The critical difference and standard error of the means were computed to assess the significance between the species means.

6.3. Results

6.3.1. Number of nodules

The total number of nodules/plants produced irrespective of the different root categories were significantly different (p < 0.05) for all the species at different intervals (Table 6.1).

Nodulation at 1st month

In the first month the highest total number of nodules was obtained from A. *indica* followed by C. *micans* with value of 9.43 and 1.14 respectively (Table 6.2). However, the root nodules were absent in 1st month old seedlings of C. *mucunoides*. Nodulation began on the primary roots for both A. *indica* and C. *micans* with 6.57 and 2.71 respectively (Figure 6.1a). Growth for nodules in secondary root for A. *indica* was 2.86 and 1.14 for C. *micans*. However, there was no nodulation in the tertiary roots for all the species.

Nodulation at 2nd month

Total number of nodules produced in the 2^{nd} month were significantly higher for *A. indica* (37.43) followed by *C. mucunoides* (9.14) and the least by *C. micans* with a value of 6.57 (Table 6.2). *A. indica* obtained the maximum number of nodules from the primary root with a value of 20.57. However, in the case of the other two legumes, tertiary roots produced the most nodules, with values of 3.286 and 5 for *C. micans* and *C. mucunoides*, respectively (Figure 6.1b).

Nodulation at 3rd month

Data pertaining to the total number of nodules produced in the third month has been shown in Table 6.2. The nodulation produced by the species was in the order of *A. indica* (501) > *C. micans* (28) > *C. mucunoides* (17.86). With respect to the root categories, the maximum number of nodules was obtained from the secondary roots for *A. indica* (306.74) and tertiary roots for *C. micans* (18.14) and *C. mucunoides* (12.43) (Figure 6.1c)

Nodulation at 6th month

During the 6th month, there were differences in the total number of nodules among the legume seedlings, ranging from 90.86 to 1869.43. *A. indica* had the highest number of nodules, while *C. micans* had the lowest (Table 6.2).

In *A. indica*, nodulation followed the same pattern as in the 3^{rd} month, with maximal nodulation seen in the secondary roots. *C. micans* and *C. mucunoides* showed that the tertiary roots produced the highest number of nodules, with 74 and 135.43 respectively, whereas the primary roots produced the fewest (Fig 6.1d).

Table 6.2. Total number of nodules plant⁻¹ (irrespective of the root categories) produced by the three leguminous species at different intervals.

Number of nodules plants ⁻¹				
Species	1 st month	2 nd month	3 rd month	6 th month
A. indica	9.43	37.43	501	1,869.43
C. micans	1.14	6.57	28	90.86
C. mucunoides	0	9.14	17.86	160.714
CD _{0.05}	1.52	12.88	79.22	582.49
$SE(m) \pm$	0.51	4.30	26.46	194.54

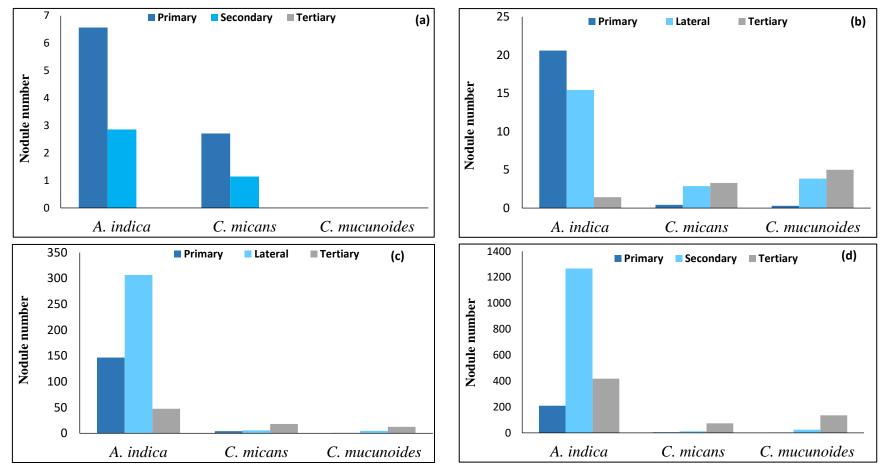


Figure 6.1. Numbers of nodules produced on different root categories (primary, secondary and tertiary) of the leguminous species at (a) 1st month, (b) 2nd month, (c) 3rd month and (d) 6th month

6.3.2. Fresh biomass of the nodules

Fresh biomass of nodules at 1st month

There was significant difference in the total fresh biomass of the nodules produced among the species (Table 6.3). *C. micans* recorded the maximum fresh biomass followed by *A. indica*. Initially *C. mucunoides* did not produce any nodules on the first month. With respect to the root category, *A. indica* and *C. micans* recorded the highest fresh biomass on the primary roots with value of 3.7 mg and 8.89 mg respectively as shown in Figure 6.2a. The secondary fresh biomass for *A. indica* and *C. micans* were 1.69 and 3.54 mg respectively

Fresh biomass of nodules at 2nd month

At 2^{nd} month stage, the total fresh biomass obtained did not vary across the species (Table 6.3). The fresh biomass of the nodules from *A. indica* reduces from primary to tertiary roots as shown in Figure 6.2b. The fresh biomass of *C. micans* and *C. mucunoides* rises when roots progress from primary to tertiary. *C. micans* exhibited the highest fresh biomass of nodules on the secondary roots (16.97 mg) and tertiary roots (27.53 mg) as depicted in Figure 6.2 b.

Fresh biomass of nodules at 3rd month

The total fresh biomass obtained varied significantly among all the species examined. *A. indica* had the highest total fresh biomass of 358.04 mg, followed by *C. micans* with 285.42 mg and *C. mucunoides* of 189.55 as shown in Table 6.3. The fresh biomass in the primary roots of *A. indica* was 117.7 mg, and in the secondary roots, it was 211.36 mg, which was the highest among the other species (Fig 6.2c). *C. micans* had the maximum fresh biomass on the tertiary roots (158.41 mg).

Fresh biomass of nodules at 6th month

At 6th month, *C. micans* had the highest total fresh biomass with 2506.60 mg, whereas C. mucunoides had the least with 189.55 mg (Table 6.3). When examining the several root categories. It was observed that *A. indica*, recorded the highest fresh biomass on the primary (159.12 mg) and secondary (853.76 mg) roots (Figure 6.2d). The fresh biomass of the tertiary root was highest in *C. micans* (2173.20 mg), followed by *A. indica* (264.47 mg) and *C. mucunoides* (156.21 mg).

Table 6.3. Total fresh biomass of the nodules/plants produced by the three leguminous species at different intervals (irrespective of the root categories)

Fresh biomass (mg)				
Species	1 st month	2 nd month	3 rd month	6 th month
A. indica	5.39	21.1	358.04	1,090.10
C. micans	13.44	45.31	285.42	2,506.60
C. mucunoides	0	13.03	35.76	189.55
CD _{0.05}	7.24	NS	135.63	979.33
SE(m)±	2.42	9.01	45.1	327.07

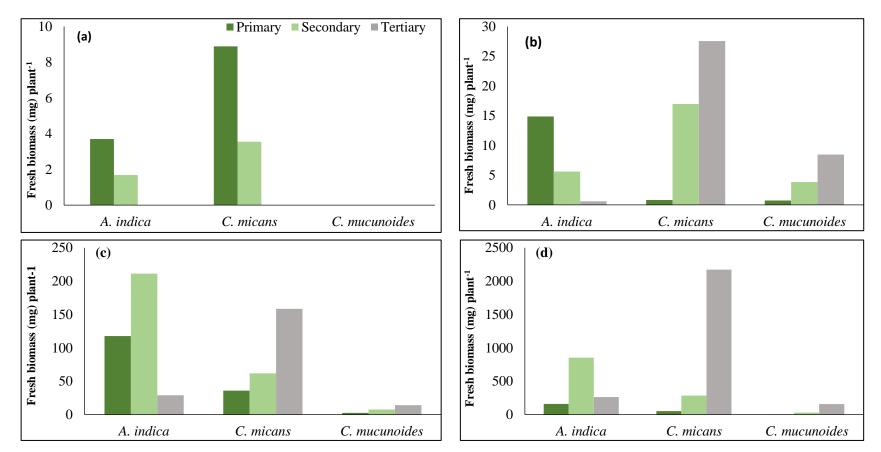


Figure 6.2. Fresh biomass of nodules produced on different root categories (primary, secondary and tertiary) of the leguminous species at (a) 1st month, (b) 2nd month, (c) 3rd month and (d) 6th month.

6.3.3. Dry biomass of the nodules

The total dry biomass of the nodules varied significantly at each interval, with *C. micans* recording the maximum dry biomass at the first month (4.6 mg), second month (21.38 mg), and sixth month (915.8 mg) (Table 6.2). In the third month, we noticed that *A. indica* obtained the greatest dry biomass of 176.1 mg. The nodules of *C. mucunoides* had the lowest dry biomass throughout the study period, as shown in Table 6.4.

During the first month, a notable variation in dry biomass of nodules was noted in the primary root category, ranging from 0.21 to 0.68 mg. There was no notable variation in the dry biomass of nodules between the secondary and tertiary roots. *C. micans* had the highest dry biomass in both categories, with values of 3.12 mg and 0.31 mg for the secondary and tertiary roots, respectively (Figure 6.3a).

In the 2nd month the dry weight of the nodules on the primary roots ranked as follows, *A. india* (5.82 mg) > *C. micans* (0.43 mg) and *C. mucunoides* (0.28 mg). The ranking of the secondary root samples based on weight was *C. micans* (7.29 mg) > *A. indica* (2.38 mg) > *C. mucunoides* (1.20 mg). The dry biomass values for tertiary roots were 0.26 mg for *A. indica*, 3.1 mg for *C. mucunoides*, and 13.65 mg for *C. micans*.

In the 3^{rd} month there was significant difference in the dry biomass of the root nodules observed at various categories. *A. indica* accorded the highest dry biomass on the primary and secondary roots with value of 50.63 and 21.93 mg respectively. Whereas, the highest amount in the tertiary root was observed in *C. micans* (40.74 mg), with *A. indica* following at 10.53 mg and *C. mucunoides* at 3.19 mg (Figure 6.3c).

Results pertaining to the dry biomass at 6th month from different root category vary among the species (Figure 6.2d). Thy dry biomass of primary and secondary root nodules was highest with *A. indica* (58.1 and 272.94 mg respectively) followed by *C. micans* (21.5 and 101.14 mg respectively) and minimum with *C. mucunoides* (0.5 and 9.49 mg respectively). However, the dry biomass of the tertiary nodules was highest wit C. micans (795.03 mg), *A. indica* (84.39 mg) and 8.6 mg by *C. mucunoides* (Figure 6.3d).

		Dry biomass (mg)		
Species	1 st month	2 nd month	3 rd month	6 th month
A. indica	2.1	8.46	176.16	353.429
C. micans	4.68	21.38	68.08	915.876
C. mucunoides	0	4.59	6.62	72.56
CD _{0.05}	2.49	13.65	51.77	337.81
SE(m)±	0.83	4.56	17.29	112.823

Table 6.4. Total dry biomass of the nodules/plant produced by the three leguminous species at different intervals (irrespective of the root categories).

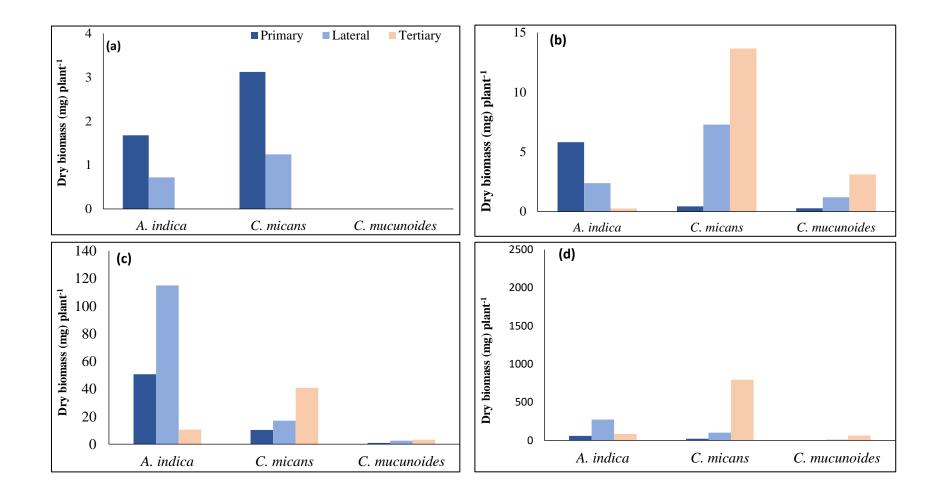


Figure 6.3. Dry biomass of nodules produced on different root categories (primary, secondary and tertiary) of the leguminous species at (a) 1st month, (b) 2nd month, (c) 3rd month and (d) 6th month.

6.3.4. Shoot and root length

At different stages of the seedling growth, the shoot and root length were recorded and are represented in Table 6.5. At the age of 1 month, there was significant difference in the shoot and root length among the three species. The maximum shoot and root length was recorded by *A. indica* with value 16.76 cm and 8.41 cm respectively and the least growth was recorded by *C. mucunoides* (11.30 cm and 5.66 cm respectively).

As the seedling grows, the shoot and root length varied significantly among the three species at various growth stages (Table 6.5). In the second month the shoot and root length range from 29.24 -40.43 cm and 15.34 -17.99 cm, with *A. indica* recording the highest and the lowest growth rate was observed by *C. micans*.

In the third month, the growth of *C. micans* was slower when compared to the other two species. The shoot length ranges from 45.28-104.40 cm and root length ranges from 18.14-35.19 cm.

However, in the 6th month a rapid increase in the growth was observed by *C*. *micans* and *A. indica*. *C. mucunoides* recorded the minimum shoot and root length with 86.70 cm and 33.36 cm respectively.

		Shoot length	n (cm) plant ⁻¹	
Species	1 st month	2 nd month	3 rd month	6 th month
C. micans	13.61	29.24	45.29	103.73
C. mucunoides	11.30	32.74	70.20	86.70
A. indica	16.76	40.43	104.40	120.60
CD _{0.05}	0.94	2.24	18.01	23.59
$SE(m) \pm$	0.31	0.75	6.02	7.88
		Root length	(cm) plant ⁻¹	
C. micans	5.97	15.34	18.14	36.03
C. mucunoides	5.66	17.99	25.81	33.36
A. indica	8.41	19.93	35.19	56.34
CD _{0.05}	0.53	0.71	5.67	11.76
$SE(m) \pm$	0.18	0.24	1.89	3.93

Table 6.5. Shoot and root length (cm) of the three leguminous species at different intervals (per plant).

6.3.5. Shoot and root biomass

Fresh and dry biomass accumulation of shoot and root (Table 6.6 and Table 6.7) have been observed to vary among the species with age. During the initial stage (1st month), *A. indica* observed to have significantly higher fresh and dry biomass of shoot biomass, while the root biomass tends to be higher in *C. micans*. The fresh and dry biomass of shoot range from 1.21-3.35 g and 0.27-1.18 g respectively, while the root accumulates biomass ranging from 0.17-0.40 g (fresh root biomass) and 0.08-0.17 g (dry root biomass).

As the day increases, *A. indica* tends to accumulates higher shoot and root biomass. As in Table 6.6 and 6.7, it is observed that *A. indica* record for higher shoot biomass with value 16.46 and 5.25 g (fresh and dry shoot biomass respectively) and 49.10 and 13.00 g (fresh and dry shoot biomass respectively) in the 2nd and 3rd month respectively. Likewise, even for the root biomass, *A. indica* record for the highest fresh and dry root biomass.

During the 6^{th} month stage of growth, it was observed that although the shoot biomass was higher in *A. indica*, the root biomass tends to accumulates more when compared among the other two species with value of 12.59 g (fresh root biomass) and 3.89 g (dry root biomass). The least biomass was observed by *C. mucunoides*.

Fresh Biomass (g)					
		Shoot			
Species	1 st month	2 nd month	3 rd month	6 th month	
C. micans	3.00	9.14	19.13	122.20	
C. mucunoides	1.21	4.90	16.39	29.84	
A. indica	3.35	16.46	49.10	132.42	
CD _{0.05}	0.36	0.48	11.24	37.62	
$SE(m) \pm$	0.12	0.16	3.76	12.57	
		1	Root		
C. micans	0.40	1.12	1.84	12.59	
C. mucunoides	0.17	1.33	1.78	3.77	
A. indica	0.23	2.18	2.41	7.48	
CD _{0.05}	0.09	0.17	NS	3.91	
$SE(m) \pm$	0.03	0.06	0.27	1.31	

Table 6.6. Fresh biomass (g) of the shoot and root of the three leguminous species at different intervals (Per plant).

Dry Biomass (g)					
		Shoot			
Species	1 st month	2 nd month	3 rd month	6 th month	
C. micans	0.71	2.11	3.97	26.05	
C. mucunoides	0.27	1.70	3.36	5.34	
A. indica	1.18	5.25	13.00	43.44	
CD _{0.05}	0.23	0.28	2.94	12.58	
$SE(m) \pm$	0.08	0.09	0.98	4.20	
		Ro	ot		
C. micans	0.17	0.67	0.86	3.89	
C. mucunoides	0.08	1.09	0.79	1.46	
A. indica	0.12	1.24	1.07	2.7	
CD _{0.05}	0.02	0.19	NS	1.45	
$SE(m) \pm$	0.01	0.06	0.11	0.49	

Table 6.7. Dry biomass (g) of the shoot and root of the three leguminous species at different intervals (per plant).

6.3.6. Nitrogen content and nitrogen uptake

The results presented in Table 6.8 clearly shows that there were substantial differences in the total nitrogen content of the roots among the species studied at 6 months of age. *Calopogonium mucunoides* recorded the highest nitrogen content with value of 1.96 %, followed by *Crotolaria micans* (1.81 %) and the least by *Aeschynomene indica* (0.91 %).

The nitrogen content in the shoots varied significantly among the species (Table 6.8). The highest nitrogen level in the shoot was found in *C. micanss* at 2.07 %, followed by *C. mucunoides* at 1.78 % and *A. indica* at 1.05 %.

The nitrogen levels in the nodules varied significantly amongst the species. The highest percentage was recorded for *C. micans* at 4.78 %, followed by *A. indica* at 3.25 % and *C. mucunoides* at 3.03 % (Table 6.8).

Significant variations in nitrogen uptake among the species were observed (Table 6.3). In *C. micans*, the highest nitrogen uptake was 550.97 mg, followed by *A. indica* with 500.58 mg and *C. mucunoides* with 165.76 mg.

Species	N % (shoot)	N % (root)	N % (nodules)	N uptake (mg plant ⁻¹)
C. micans	2.07	1.81	4.78	609.77
A. indica	1.05	0.91	3.25	480.24
C. mucunoides	1.78	1.96	3.03	123.62
CD _{0.05}	0.07	0.15	0.24	16.46
$SE(m) \pm$	0.02	0.04	0.07	4.67

Table 6.8. Nitrogen content and nitrogen uptake plants⁻¹ by the leguminous species at 6 months.

6.3.7. Available nitrogen in soil

After the 6th month of experiment, it was observed that the soil available nitrogen content increased from the initial stage (Table 6.9). The soil nitrogen availability was significantly different for all the three species with *C. micans* observed for the maximum (360.24 kg ha⁻¹) and the minimum by *A. indica* (338.03 kg ha⁻¹). The soil nitrogen enrichment was maximum in *C. micans* (31.34 kg ha⁻¹) and the minimum was obtained by *C. mucunoides* (14.60 kg ha⁻¹)

Table 6.9. Available nitrogen content of the soil (after 6th month).

Species	Available nitrogen (kg ha ⁻¹)	Nitrogen enrichment (kg ha ⁻¹)	
C. micans	360.24	31.34	
A. indica	343.51	14.60	
C. mucunoides	351.37	22.47	
CD _{0.05}	6.81	8.21	
$SE(m) \pm$	1.93	2.33	

6.3.8. Nitrogen fixed per year

At 6 months of age, the leguminous species were evaluated for total nitrogen fixed through soil enrichment and plant uptake. It was observed that *C. micans* fixed the highest amount of nitrogen at 227.36 kg ha⁻¹ yr⁻¹, followed by *A. indica* at 174.73 kg ha⁻¹ yr⁻¹, and *C. mucunoides* at 62.46 kg ha⁻¹ yr⁻¹ (Figure 6.4).

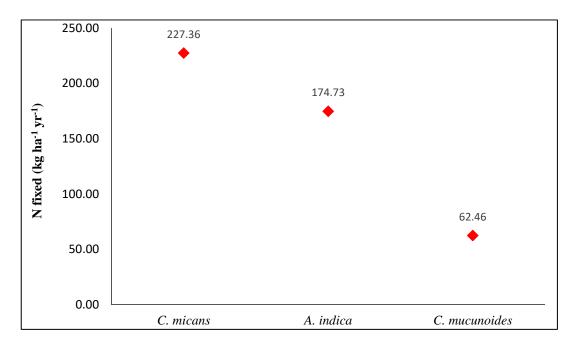


Figure 6.4. Nitrogen fixed (kg ha⁻¹ yr⁻¹) by the leguminous weed species.

6.4. Discussion

6.4.1. Nodulation behavior

The total number of nodules was determined by counting the nodules on the primary, secondary, and tertiary rootlets. It has been observed that the number of nodules increased with the increase in time. It has also been observed that the number of nodules, irrespective of different rootlet nodulation was maximum in *A. indica* as compared to the other two species. However, there was no nodulation formed by *C. mucunoides* in the first month which may be attributed to a combination of environmental conditions. Likewise, a study by Rejili *et al.* (2009) also confirmed the findings that limited nodulation in *Ononis natrix* was attributed to various environmental factors.

Differences in nodule quantity could be attributed to genetic variability within the species. Genetic characteristics of various species and environmental factors influence root nodulation (Gibson and Jordon, 1983; Pokhriyal *et al.*, 1991). This may also be associated with the adaptability of bacteria and plant species (Kucuck and Cevheri, 2014). In the field, the quantity of nodules per plant exhibited variability contingent upon factors such as growth conditions, sample site, and plant species. (Gehlot *et al.*, 2012).

Initially, the nodulation started from the primary root in all three species and gradually growth of nodules increased in the secondary and tertiary root in the later stages of plant growth. The observed decrease in nodulation per rootlet compared to its parent root can potentially be due to factors such as reduced surface area, smaller infection site, and less enzymatic activity. Improved nodulation of the sample plants in the later growth stages can be attributed to the increased photosynthetic activity and root growth which could positively impact nodulation (Kashyap *et al.*, 2012; Gordon and Wheeler, 1978). Kashyap *et al.* (2012) noted a comparable pattern in nodule development and growth among three nitrogen-fixing tree species: *Alnus nitida, Acacia catechu,* and *Albizia chinensis.* According to Mendonca and Schiavinato (2005) study, the application of mineral nitrogen to *Crotalaria juncea* plants led to increased growth and also it was noted that all treatments resulted in the presence of

nodules. The total nodule produced (irrespective of rootlets) in the present study was within the range that was higher than the study reported by Ranjan *et al.* (2007) who studied on the nodulation behaviour of *Acacia mangium*, *Albizia lebbek*, *Gliricidia sepium*, *Leucaena diversifolia* and *Leucaena leucocephala*. Irin and Biswas (2021) observed that *S. rost*rata had the highest nodulation rate of 36.53 plant⁻¹ after 30 days of seeding, while *L. leucocephala* had the lowest rate of 11.00 per plant. Pramanik *et al.*, (2009), found *that S. aculeata*, *C. juncea*, and *P. mungo* generated the highest number of nodules per plant after 30 days of seeding, with values of 21.99, 17.69, and 17.44 respectively. Similarly, in a study by de Castro Piers *et al.* (2018), Mimosa species were found to produce nodules ranging from 0.0 to 120.6 after 4 months of harvest, which was lower than our results.

In the present study, there was observed proportionality between the nodule biomass and the number of nodules for each species. Similar study was observed where relationship between nodule biomass and the number and size of nodules on the main, secondary, and tertiary roots was shown to be directly proportional (Kashyap *et al.*, 2012).

6.4.2. Nitrogen content and uptake

The plant nitrogen content is a measure of the efficiency with which the host plants utilize available nitrogen. Plants that fix nitrogen exhibit a higher nitrogen content compared to plants that do not fix nitrogen. In the present study, it was observed that the nitrogen content in the various parts of the plant was significantly different among the three species. In the present study, the nitrogen content in the shoot and root was higher in *C. micans* and *C. mucunoides*. The enhanced nitrogen dynamics observed in the shoots and roots of *C. micans* and *C. mucunoides* can be attributable to the higher nitrogen content in these plant parts (Beniwal *et al.*, 1995).

The nodules of *C. micans* exhibited the highest nitrogen content while having a much lower number of nodules as compared to *A. indica*. Purohit *et al.* (1997) reported a comparable association in which nodules of *Dalbergia sericea* exhibited elevated nitrogen content, while the fresh biomass generated was comparatively reduced in comparison to *Albizia chine*nsis and *Dalbergia sissoo*. According to Barbosa (2020) findings, *C. juncea* exhibited a notable capacity for nitrogen accumulation in its shoots, with the stem serving as the primary site for nutrient buildup. A similar correlation between nodule biomass and nitrogen concentration was also discovered by Kashyap *et al.*, (2012) in *Albizia chinensis*, *Acacia catechu*, and *Alnus nitida*.

Unkovich and Pate (2000) have reported that the estimation of N intake and N₂ fixation is frequently underestimated due to the omission of belowground N, which comprises N present in roots, nodules, exudates, and rhizodeposition. The uptake of nitrogen by plants is influenced by various factors, including plant growth rate, the active-passive mechanism of the root surface, the availability of nitrogen, the physiological properties of the soil, and the prevailing climatic circumstances. The maximum nitrogen uptake was recorded by *C. micans* (550.97 mg plant⁻¹) as shown in Table 6.8. The observed difference can be attributed to the changing efficiency of nutrient uptake by the species which is influenced by various soil conditions, as well as the efficiency of nutrient use. Moreover, the usage of nutrients by various plant parts varies which may further depend on the microclimate of the place also. Salvagiotti *et al.* (2008) reviewed various research work and concluded that the average total nitrogen uptake was 219 kg ha⁻¹, and a maximum of 485 kg ha⁻¹.

6.4.3. Nitrogen fixation

Legumes generally contribute approximately 50-60% of their nitrogen intake through symbiotic nitrogen fixation (Vance, 2002). However, the process of nitrogen fixation in legumes depends on factors, including soil moisture, temperature, the presence of soil nutrients, both living and non-living stressors, the native rhizobia strains, as well as field conditions and management techniques (Palmer and Young, 2000; Kiers *et al.*, 2003). The optimization of nodulation and biological nitrogen fixation in legumes is a critical concern in light of the prevalent increase in soil degradation. The nitrogen fixation capacity of a legume per acre is contingent upon several factors, including the number and size of nodules, their longevity, the bacterial strain present, the state of plant growth, and the management of the crop (Rajagopalan and Sadasivan, 1964). In favor of the above statement a study by Houngnandan (2000) concluded that the impact of rhizobia on the symbiotic nitrogen fixation of *Mucuna pruriens*, a leguminous cover crop, is reliant upon the rhizobial population present in the field and the application of rhizobial inocula.

The nitrogen fixation of the species was determined by assessing their nitrogen intake and soil enrichment. Our findings indicate that *C. micans* exhibited the highest nitrogen fixation rate, with a value of 283.44, surpassing the nitrogen fixation rates of the other two species examined in our study (Figure 6.4).

The nitrogen-fixing capabilities of Crotalaria species, specifically *C*. *ochroleuca*, have been observed to be substantial, indicating their potential value in various cropping systems (Samba, 2002). Similarly, the nitrogen-fixing capacity of root nodules in many plant species was examined by Bond (1958); Harris (1958) who proposed the possibility of nitrogen fixation in *C. juncea*. Nevertheless, the nitrogen fixation rate observed during the current experiments was relatively higher than those observed previously by Kashyap *et al.* (2012) for *Alnus nitida* (14.10 kg ha⁻¹ yr⁻¹), *Albizia chinensi* (8.84 kg ha⁻¹ yr⁻¹) and *Acacia catechu* (7.41 kg ha⁻¹ yr⁻¹)

Soil acidity has been widely recognized as a factor that reduces the process of symbiotic nitrogen fixation in legumes, which has a detrimental impact on their growth and productivity. This is particularly true for plants that rely only on symbiosis to obtain nitrogen (Bekere *et al.*, 2013; Mohammadi *et al.*, 2012). However, Ferreira *et al.* (2016) demonstrates the ability of *Calopogonium mucunoides* to nodulate and fix nitrogen at a pH level as low as 5.5. Another study by Camargos and Sodek (2010) claimed *C. mucunoides* exhibits distinct and significant characteristics, including nodulation and nitrogen fixation, even when exposed to nitrates.

6.5. Conclusion

The study investigated the nodulation behavior of three leguminous weed species in Mizoram. The plant species *A. indica* demonstrated notable number of nodules, particularly in the secondary roots. The nitrogen content exhibited variation across the shoots, roots, and nodules of different species, with *C. micans* demonstrating the maximum nitrogen level in its shoot and root nodules. *C. micans* and *A. indica* fixed higher amount nitrogen comparing to *C. mucunoides*. The aforementioned findings contribute to the advancement of knowledge regarding the dynamics of legume nodulation, providing valuable insights into the particular responses of different species and the potential for promoting sustainable agriculture. Nevertheless, it is important to conduct a comprehensive examination in the field circumstances in order to comprehend the correlation between the nodulation and symbiotic nitrogen-fixing capacity of the studied species, with the aim of determining their appropriateness as green manure crops.

CHAPTER 7

EFFECT OF THE GREEN MANURE LEGUMES ON THE SOIL PROPERTIES AND THE GROWTH AND YIELD OF TOMATO (Lycopersicon esculentum Mill.)

7.1. Introduction

The usage of fertilizer has a significant impact on crop yield, soil nutrient content, agricultural production, and their corresponding environmental consequences. Reduced soil fertility and rising costs of mineral fertilizers have led to the increased popularity of legumes as organic fertilizers. Enhancing soil fertility is a major function of organic fertilizers. In this context an alternative is to utilize green manure which if properly utilized, green manures have the potential to substitute for the nitrogen and other minerals needed by the crops that follow them.

Introducing green manures as substitutes to minimize dependence on mineral fertilizers is regarded as a commendable agricultural practice. Nevertheless, the impact of each green manure on soil characteristics and crop productivity is contingent upon its chemical composition. Organic fertilizers such as manures are frequently inadequate in organic arable farming, which requires alternative sources of nitrogen to sufficiently fertilize high-yielding crops (Talgre *et al.*, 2012). Using green manures to soil is widely regarded as a beneficial management practice in all types of agricultural production systems. This is because it stimulates the growth and activity of soil microbes, leading to the release of plant nutrients through mineralization. As a result, it enhances soil fertility and quality.

Soil fertility is influenced by soil organic matter, which relies on the supply of biomass to counteract mineralization. Nitrogen is the most extensively studied nutrient, although the amounts of phosphorus and potassium are equally important. Farms, especially those practicing organic farming, rely significantly on soil organic matter. Green manure, when added to the soil, enhances its physical and chemical properties and promotes plant growth (Ding *et al.*, 2021; Dos Santos *et al.*, 2021; Seminchenko and Sukhareva, 2022). Green manure crops act as an alternative source of nitrogen not only enrich the soil with nutrients but also promote the development of

deep root systems that can absorb nutrients from deeper soil layers. This results in a reduced uptake of available nutrients, leading to an increased concentration of plant nutrients in the topsoil (Islam *et al.*, 2019).

It is necessary to investigate the potential of the plant materials as green manure and their impact on soil properties and the growth and yield of the succeeding crops grown with each green manure. Therefore, the present chapter aims to assess and compare the effects of green manure species on the growth and yield aspects of tomato and also its effect on the soil properties.

7.2. Materials and Methods

7.2.1. Experimental details

To assess the effects of leguminous weeds as green manure on the soil properties and the growth and yield of tomatoes, a consecutive two-year field experiment was carried out. Seeds of the selected weed species were collected from their natural habitat and cultivated in a specified experimental area (as described in chapter 4). After 90 days of sowing the legumes were harvested and chopped into small pieces and then ploughed and pulverized properly into the soil of respective experimental plots. 60 days after incorporating the legumes soil samples were collected again and four weeks old tomato seedlings were transplanted to the plot in line and routine cultural operation was carried out to ensure proper growth of the tomato plants. The details of the experiment are given below in Table 7.1. and Figure 7.1.

Table 7.1. Experimental details

Experiment carried out	Horticulture Research Farm, Mizoram University		
Design	Randomized Block Design		
Plot size	1.5 m x 1 m = 1.5 sq. m		
Bed type	Raised beds		
Number of treatments	4 (T ₁ = <i>C. micans</i> ; T ₂ = <i>C. mucunoides</i> ; T ₃ = <i>A. indica</i> and T ₀ = Control)		
Number of replications	5		
Crop	Tomato (Arkha Rakshak)		
Date of transplanting the tomato seedlings	September 2021 and 2022		
Spacing	$Tomato = 45 \text{ x } 45 \text{ cm}^2$		

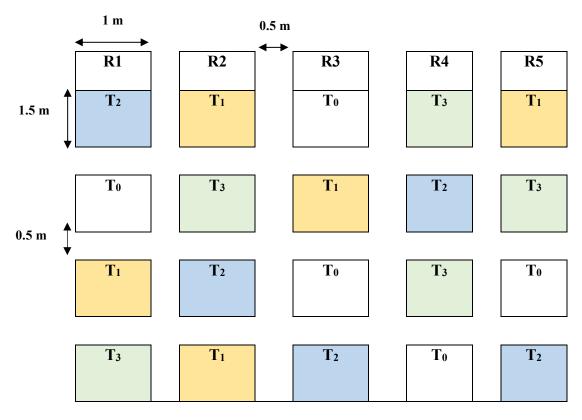


Figure 7.1. The layout plan of the experimental field.

7.2.1. Soil sampling

To determine the impact of green manuring on selected plant species, soils from each replicated plot for each treatment were taken at the beginning of the field experiment i.e. just before the sowing of the seed of the legume weeds. The soil samples were collected again after 60 days of incorporating the legume species into the soil. Using a soil corer, 3 corers were taken from each plot at two depths viz. 0-10 cm and 10-20 cm. Composite soil sample were formed by pooling the replicated soil samples treatment wise. The samples were then kept in polythene bags and brought to the laboratory. Before any analysis the soil samples were divided into two parts, one part was air-dried and the other part was kept in the refrigerator at 4° C for analysis of biological properties. The air-dried samples were the grounded and sieved through 2 mm sieve and analysis were undertaken. For the determination of soil moisture content and pH fresh samples were used.

7.2.2. Determination of soil physico-chemical properties

7.2.2.1. Soil moisture

Soil moisture content was determined by gravimetric method

Moisture content (%) = $\frac{W_1 - W_2}{W_2} \times 100$

Where, W₁= Weight of fresh soil

W₂=Weight of oven dried soil

7.2.2.2. Soil pH

Soil samples were analyzed for pH by using a glass electrode with a pH meter in 1:2.5 (Soil:water) suspension (Jackson, 1973)

7.2.2.3. Soil Organic Carbon (SOC)

It was determined by the Walkey and Black method (1934).

Organic carbon (%) = $\frac{(Blank-Sample reading) \ge 0.5 \ge 0.003 \ge 100}{Sample weight} \ge 1.3$

Where, 1.3 is the correction factor.

7.2.2.4. Available Nitrogen

It was estimated by the Alkaline permanganate method (Subbiah and Asija, 1956)

Available N (kg ha⁻¹) =
$$\frac{\text{R x Normality of acid x 0.014 x 2.24 x 10^{6}}}{\text{Sample weight}}$$

Where, R= Titration value – Blank value

7.2.2.5. Available Phosphorous

The available Phosphorous was estimated by the Bray's P-1 (Bray and Kurtz 1945).

Available P (kg ha⁻¹) =
$$\frac{Q \times V \times 2.24 \times 10^6}{A \times S \times 10^6}$$

Where, Q = quantity of P in μ g read on X-axis against a sample reading

V = volume of extracting reagent used (ml)

A = volume of aliquot

S = Weight of sample (g)

7.2.2.6. Potassium

Available potassium was determined by the Neutral Normal Ammonium Acetate method (Stanford and English 1949)

Available K (kg ha⁻¹) = $\frac{C \times V}{\text{Sample weight (g)}} \times 2.24$

Where, C = Concentration of K (mg l⁻¹) in the sample filtrate as obtained from standard curve

V = Total volume of the extractant

7.2.3. Soil biological properties

7.2.3.1. Microbial Biomass Carbon (MBC)

The study of soil microbial biomass carbon (MBC) was conducted using fresh soil samples, using the chloroform-fumigation extraction method described by Brookes *et al.* (2006).

MBC (
$$\mu$$
 g⁻¹) = $\frac{EC_F - EC_{NF}}{K_{EC}}$

Where, $EC_F = Organic C$ extracted from fumigated soil

 EC_{NF} = Organic C extracted from non-fumigated soil

 $K_{EC} = 0.25$

7.2.3.2. Microbial Biomass Phosphorous

The soil Microbial biomass phosphorous was determined by the Chloroform fumigation extraction method, followed by the Bray's P-1 solution extraction.

 $P \mu g^{-1} = Concentration of P x dilution factor$

 P_F and $P_{UF} = P \times V_s/M_s$

Where, $V_s =$ Volume of moisture + Volume of extractant

 M_s = volume of oven dried soil

MBP
$$\mu$$
 g⁻¹ = $\frac{P_F - P_{UF}}{K_{EP}}$

where, $P_F = Extracted$ phosphorous from the fumigated soil

 P_{UF} = Extracted phosphorous from the unfumigated soil

K_{EP} = Extractable phosphorous (= 0.4; Brookes *et al.*, 1982)

7.2.3.3. Microbial population (Bacteria and Fungi)

The soil microbial population was quantified by Spread-plate technique described by Taylor *et al.* (1983). The microbial population was examined and quantified as colony-forming units per ml (CFU ml⁻¹).

 $CFU ml^{-1} = \frac{No. of colonies x Dilution factor}{Volume of culture}$

7.2.4 Relative changes (%)

The relative changes of the soil properties before and after incorporation of the green manure was outlined by the formula:

Relative change (%) =

Final value (after incorporation)- Initial (before sowing of legume) Initial value x 100

7.2.5. Growth and Yield of Tomato

7.2.5.1. Biometric observation

All the plants from each plot were tagged as replication from each plot and all the parameters were recorded at successive growth stages of the tomato plants.

7.2.5.2. Growth parameters

7.2.5.2.1. Plant height

The plant height was observed at an in interval of 30, 60 and 90 days after planting. The plant height (cm) was measured using a measuring tape from the base to the tip of the uppermost latest leaf.

7.2.5.2.2. Collar diameter

The collar diameter (mm) was also recorded after 30, 60 and days after planting with the help of a digital vernier caliper.

7.2.5.2.3. Number of primary branches plant⁻¹

At each successive growth stages, the number of branches per plant was recorded and the mean was determined.

7.2.5.2.4. Number of clusters per plant

The number of clusters per plant was tallied and subsequently averaged.

7.2.5.2.5. Number of fruits per cluster

The number of fruits each cluster was tallied and subsequently the average was calculated.

7.2.5.2.6. Number of fruits per plant

The number of fruits collected at each harvest was counted, and then the total number of fruits per plant was computed. Finally, the mean was determined.

7.2.5.2.7. Fruit length and breadth (cm)

Each time the fruits from each treatment collected, the length and diameter of the fruits were recorded using a digital vernier calliper and the mean was computed.

7.2.5.2.8. Average fruit weight

The average fruit weight was calculated by dividing the total fruit weight per plot by the number of fruits gathered per plot.

7.2.5.2.9. Fruit yield

Fruit yield was calculated based on the yield of each individual plot. The yield was expressed in term of quintal ha⁻¹.

7.2.6. Economics

The different economic parameters were determined by the following formulas:

Total Cost of Cultivation (Rs) = Total input Cost (Operational cost of green manure + Tomato) + Cost of Seeds.

Gross return = Yield (kg ha⁻¹) x Price per kg (Rs)

Net Return = Gross return (Rs) – Cost of cultivation (Rs)

Benefit Cost Ratio (BCR) = Gross return (Rs) / Cost of cultivation (Rs)

7.2.7. Statistics

The data obtained were subjected to Analysis of Variance (ANOVA) using the OP-STAT (online statistical package: <u>http://14.139.232.166/opstat/</u>). The means were compared using the critical difference and standard error of the mean at a significance threshold of 0.05

7.3. Results

7.3.1. Physico-chemical properties of soil

7.3.1.1. Moisture content (%)

Throughout the experimental period, soil moisture content showed no significant variations among the treatments but varied before and after the incorporation of green manure (Table 7.2). The control plot i.e. T_0 (22.14 % and 22.21 % at 0-10 and 10-20 cm respectively) had the highest moisture content while T_1 had the lowest at both the surface (20.44 %) and sub-surface layer (18.88 %).

Figure 7.2 shows a that the soil moisture content in all the treatments was at par and hence shows a non-significant variation (p < 0.05) among the treatments in the surface as well sub-surface layer in both the cropping year. However, the soil moisture content showed variation after the incorporation of the green manure when compared from the initial (before sowing of green manure) in both the cropping year except for the surface layer in the first cropping year (Figure 7.2). Furthermore, the soil moisture content was higher in the surface (0-10 cm) than in the sub-surface layer (10-20 cm).

A decreasing trend in the soil moisture was observed in all the treatments as shown in Figure 7.3. When compared to the initial stages, T_1 had the most relative changes (25.14 % at surface and 40.96 % at the sub-surface soil layer), and the least changes was observed in T_0 with value of 16.84 % and 25.64 % in 0-10 and 10-20 cm respectively.

	Soil moist	ure content (%	()	
	0-10	cm	10-20 cm	
Treatments	Before	After	Before	After
T ₀	26.78	22.21	29.88	22.14
T_1	27.38	20.44	32.03	18.88
T ₂	26.94	21.58	30.04	20.05
T ₃	26.46	21.99	29.94	21.03
	CD _{0.05}	$SE(m) \pm$	$CD_{0.05}$ SE(m) ±	
Factor (A)	NS	± 0.45	NS	± 0.55
Factor (B)	0.92	± 0.32	1.13	± 0.39
(A X B)	1.85	± 0.63	2.25	± 0.77

 Table 7.2. Effect of green manure weed species on the soil moisture (pooled data of two consecutive years).

Factor A: indicates the Treatments where $T_1 = C$. *micans*, $T_2 = A$. *indica*, $T_3 = A$. *indica* and $T_0 = C$ ontrol.

Factor B: indicates soil moisture **before** sowing the leguminous crops and **after** incorporation of the legumes into the soil.

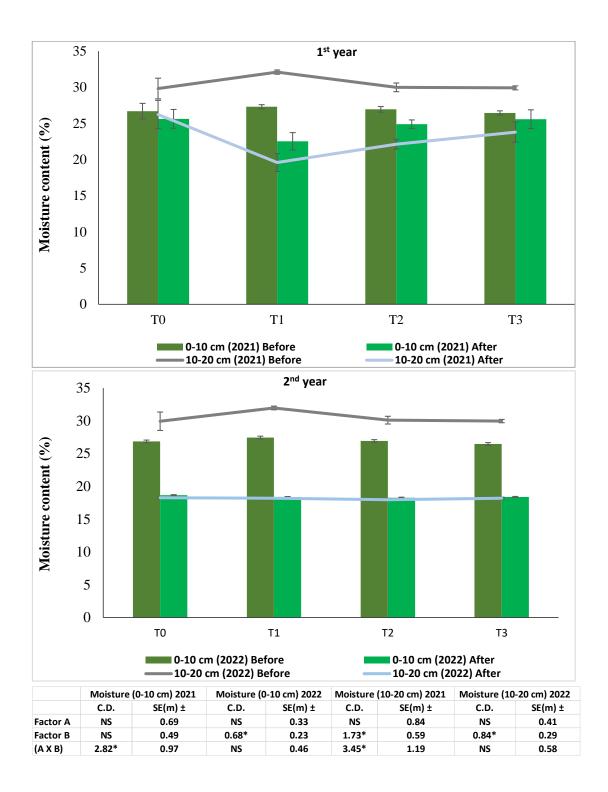


Figure 7.2. Effect of green manuring on the soil moisture during the 1st and 2nd year cropping at 0-10 and 10-20 cm (Factor A= treatments, Factor B= before sowing the leguminous crops and after incorporation of the legumes into the soil, NS= non-significant and CD= Critical difference, at 0.05 level).

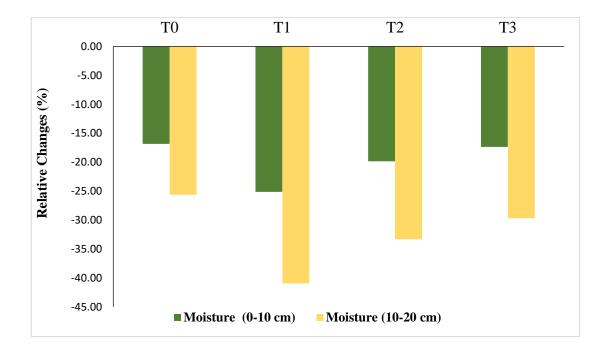


Figure 7.3. Relative changes in the soil moisture from the initial (before sowing of legume) and final (after the incorporation of the legume into the soil) at 0-10 and 10-20 cm soil depth.

7.3.1.2. Soil pH

Soil pH was determined to be statistically significant among the treatments and also the soil pH was significantly influenced by the incorporation of green manure. Before sowing the legumes, the soil pH value ranged from 5.33 to 5.43 and 4.82 to 4.98 at 0-10 cm and 10-20 cm respectively. After the incorporation of green manure, the soil pH tends to decrease while the soi pH in the plots without any green manure increases. The observed soil pH range was 5.03 to 5.46 and 4.65 to 5.05 for 0-10 cm and 10-20 cm respectively (Table 7.3). Figure 7.4 shows a significant variation (p < 0.05) among the treatments before the legume sowing and after the incorporation of the legume into the soil at 0 – 10 cm in both the cropping year. However, the soil pH at 10-20 cm in the first cropping year showed no significant difference (p < 0.05) among the treatments as well as the before and after incorporation of the legumes into the soil.

A decreasing pH trend was detected in treatments involving the incorporation of green manure (T₁, T₂, and T₃) compared to treatments without green manure (T₀) as shown in Figure 7.5. The observed range of percent decrease was 2.92 % (T₁), 6.45 (T₂) and 5.34 % (T₃); 3.61 % (T₁), 5.36 % (T₂) and 6.59 % (T₃) for 0-10 cm and 10-20 cm respectively. Without the incorporation of green manure (T₀), there was an increase in the soil pH at 0-10 and 10 -20 cm where the observed value was 2.39 % (0-10 cm) and 4.92 % (10-20 cm).

		Soil pH			
Treatments	0 -	10 cm	10-20 cm		
	Before	After	Before	After	
ТО	5.34	5.46	4.82	5.05	
T1	5.43	5.27	4.96	4.79	
T2	5.38	5.03	4.98	4.71	
Т3	5.33	5.04	4.97	4.65	
	CD _{0.05}	SE(m) ±	CD _{0.05}	SE(m) ±	
Factor (A)	0.07	± 0.02	0.08	± 0.03	
Factor (B)	0.05	± 0.03	0.05	± 0.02	
(A X B)	0.10	± 0.02	0.11	± 0.04	

Table 7.3. Effect of green manure weed species on the soil pH (pooled for two consecutive years)

Factor B: indicates soil pH **before** sowing the leguminous crops and **after** incorporation of the legumes into the soil.

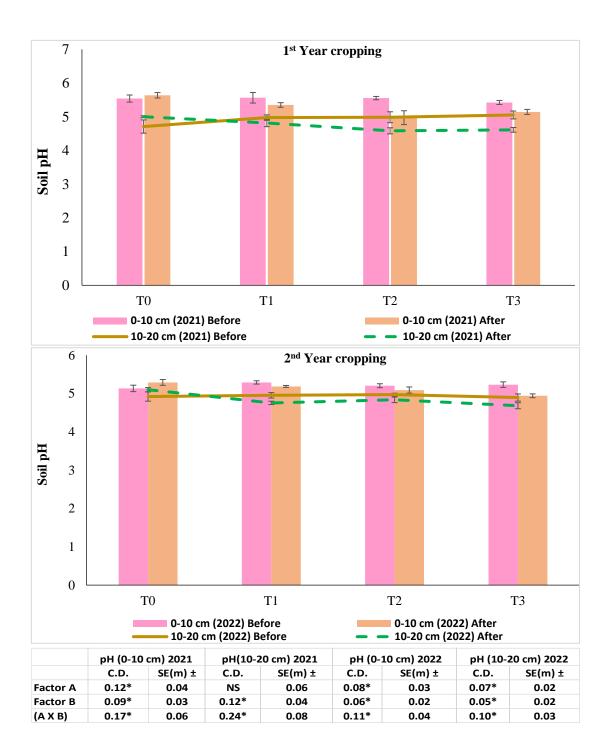


Figure 7.4. Effect of green manuring on the soil pH during the 1st and 2nd year cropping at 0-10 and 10-20 cm (Factor A= treatments, Factor B= before sowing the leguminous crops and after incorporation of the legumes into the soil and NS= non-significant and CD= Critical difference, at 0.05 level).

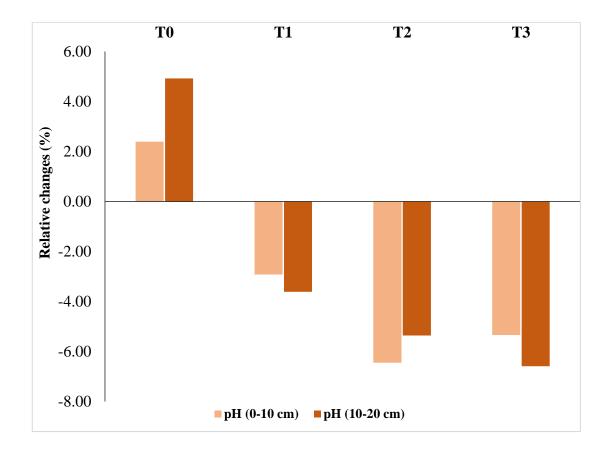


Figure 7.5. Relative changes in the soil pH from the initial (before sowing of legume) and final (after the incorporation of the legume into the soil) at 0-10 and 10-20 cm soil depth.

7.3.1.3. Organic carbon (%)

The soil organic carbon was determined to be statistically significant (p < 0.05) among the treatments and also significantly influenced by the incorporation of green manure throughout the experimental period. The initial soil organic carbon at 0-10 cm and 10-20 cm ranges from 1.12 to 1.52 % and 0.67 to 1.13 % respectively (Table 7.4). The soil organic carbon at 0-10 cm and 10-20 cm increases after the incorporation of the green manure legumes. Among the treatments the higher organic carbon was recorded from T₁ with value of 2.02 % (0-10 cm) and 1.39 % (10- 20 cm) followed by T₂ with value 0f 1.71 % (0-10 cm) and 1.20 % (10-20 cm), T₃ with value 1.59 % (0-10 cm) and 1.07 % (10-20 cm) and the least was observed from the plot without any green manure legumes (T₀) with value 1.30 % (0-10 cm) and 0.79 % (10-20 cm).

It was also observed that the soil organic carbon in both cropping years vary significantly (p < 0.05) among the treatments as well as before sowing of the legume and after incorporation of the green manure (Figure 7.6). It was also observed that the levels of soil organic carbon decreases as the depth increased from the soil surface.

The soil organic carbon exhibited a favorable and considerable increasing trend following the implementation of green manure treatments, in comparison to its initial state prior to the seeding of the legume (Figure 7.7). The relative changes varied from 16.13 to 48.04 % and 19.13 to 24.53 % at 0-10 cm and 10-20 cm respectively, where the maximum changes occurred in T_1 and the minimum changes was observed from T_0 for both the depth.

Soil organic carbon (%)							
Treatments	0 -	0 – 10 cm		10-20 cm			
	Before	After	Before	After			
To	1.12	1.30	0.67	0.79			
T_1	1.52	2.02	1.13	1.39			
T_2	1.40	1.71	0.96	1.20			
T ₃	1.37	1.59	0.87	1.07			
	CD _{0.05}	SE(m) ±	CD _{0.05}	SE(m) ±			
Factor (A)	0.04	± 0.01	0.03	± 0.01			
Factor (B)	0.02	± 0.02	0.02	± 0.01			
(A X B)	0.06	± 0.02	0.04	± 0.02			

Table 7.4. Effect of green manure weed species on the soil organic carbon (pooledfor two consecutive years)

Factor B: indicates soil organic carbon **before** sowing the leguminous crops and **after** incorporation of the legumes into the soil.

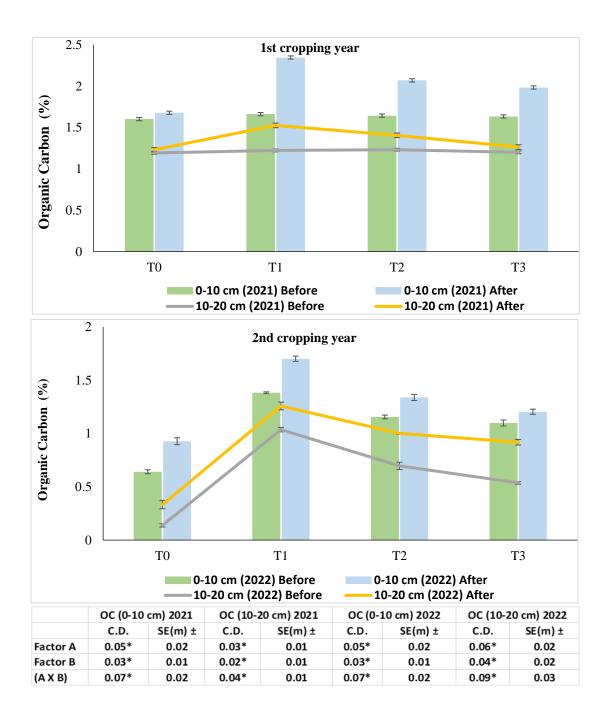


Figure 7.6. Effect of green manuring on the soil organic carbon (%) during the 1st and 2nd year cropping at 0-10 and 10-20 cm soil depth (Factor A= treatments, Factor B= before sowing the leguminous crops and after incorporation of the legumes into the soil and NS= non-significant and CD= Critical difference, at 0.05 level).

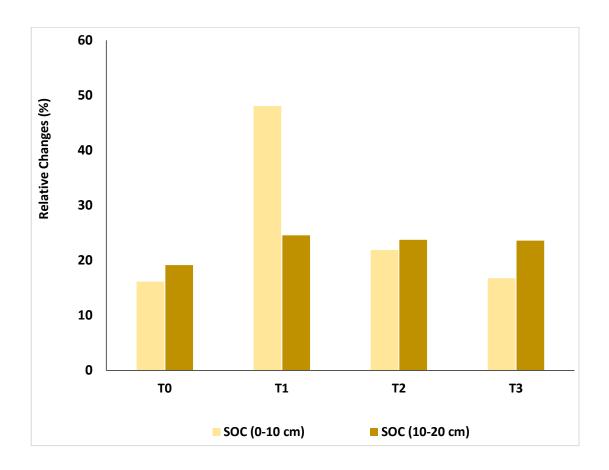


Figure 7.7. Relative changes in the soil organic carbon (%) from the initial (before sowing of legume) and final (after the incorporation of the legume into the soil) at 0-10 and 10-20 cm soil depth.

7.3.1.4. Available Nitrogen

The integration of accessions of different green manure resulted in an increase in available nitrogen (kg/ha). During both the cropping year, T_1 consistently had the highest recorded nitrogen levels both before sowing the legume seeds as well as after incorporation of the legume respectively, with values of 359.45 kg ha⁻¹ (0-10 cm) and 239.69 kg ha⁻¹ (10-20 cm) and with value of 512.35 kg ha⁻¹ (0-10 cm) and 369.27 kg ha⁻¹ (10-20 cm). On the other hand, treatment T_0 had the lowest nitrogen levels both before and after the incorporation of green manure legumes (Table 7.5).

There was a significant change (p < 0.05) in soil available nitrogen at both 0-10 cm and 10-20 cm depths between the treatments before seeding of the legumes and after incorporating the green manure legumes into the soil during the cropping year (Figure 7.6). Nevertheless, it was noted that the N content in the soil was greater during the first cropping season than that of the second season. Moreover, it was also observed that the amount of nitrogen available in the soil decreases moving from the top layer to the lower layer.

The incorporation of green manure legumes leads to considerable and favourable changes in the soil's available nitrogen compared to its initial status before sowing the legume. The inclusion of green manure led to a substantial increase in relative change compared to the control (T_0). The greatest relative changes were seen in T_1 , T_2 , and T_3 , whereas the least changes were reported in T_0 at 0-10 cm and 10-20 cm (Figure 7.7).

Available Nitrogen (kg ha ⁻¹)							
	0 – 10 cm		10-20 cm				
Treatments	Before	After	Before	After			
T ₀	327.73	257.84	204.95	128.62			
T_1	359.45	512.35	239.69	369.27			
T_2	344.68	424.49	220.50	286.49			
T ₃	329.35	358.71	215.14	271.45			
	CD _{0.05}	$CD_{0.05}$ SE(m) ±		SE(m) ±			
Factor (A)	13.9	± 4.77	15.21 ± 5.22				
Factor (B)	9.83 =	9.83 ± 3.38		± 3.69			
(A X B)	19.65 ± 6.75		21.51 ± 7.39				

Table 7.5. Effect of green manure weed species on the soil available nitrogen(pooled data of two consecutive years)

Factor B: indicates soil available N **before** sowing the leguminous crops and **after** incorporation of the legumes into the soil.

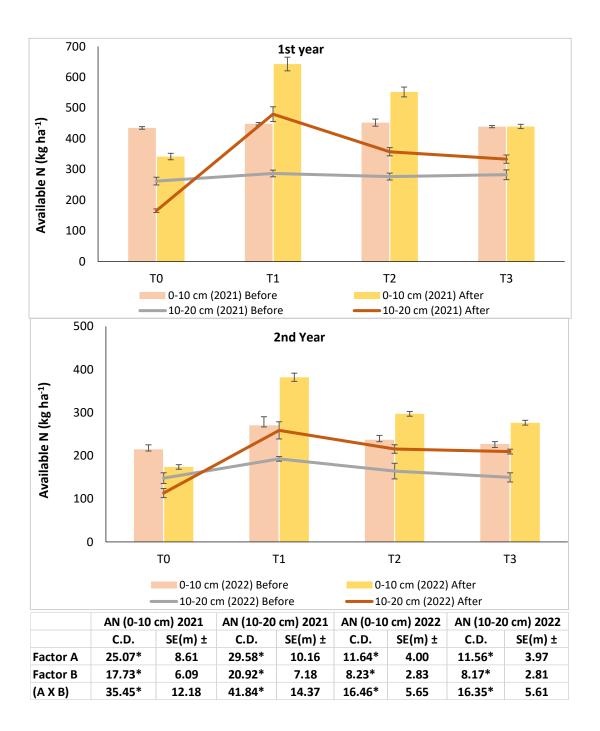


Figure 7.6. Effect of green manuring on the soil available nitrogen (Kg ha⁻¹) during the 1st and 2nd year cropping at 0-10 and 10-20 cm soil depth (Factor A= treatments, Factor B= before sowing the leguminous crops and after incorporation of the legumes into the soil, NS= non-significant and CD= Critical difference at 0.05 level).

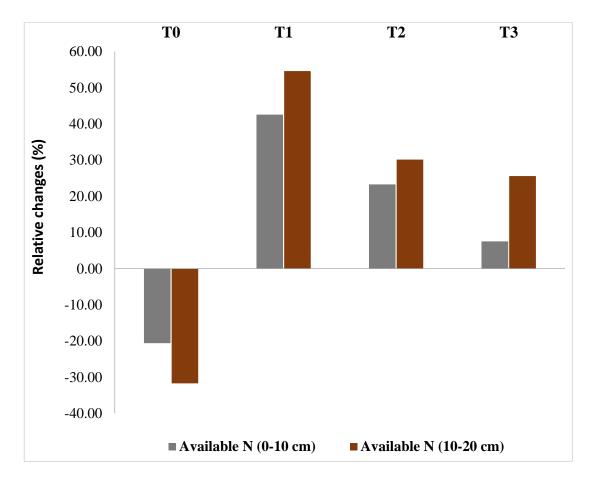


Figure 7.7. Relative changes in the soil available nitrogen (kg ha⁻¹) from the initial (before sowing of legume) and final (after the incorporation of the legume into the soil) at 0-10 and 10-20 cm soil depth.

7.3.1.5. Available Phosphorous

The incorporation of green manuring had a considerable impact on the amount of available phosphorus (kg ha⁻¹). The initial maximum availability of phosphorus (Kg ha⁻¹) in the surface and sub surface soil ranged from 24.33 to 25.91 kg ha⁻¹ and 18.00 to 19.90 kg ha⁻¹ (Table 7.6). After introducing the green manure accessions at 90 days, it was observed that there was a rise in the amounts of accessible phosphorous from 28.86 to 31.29 kg ha⁻¹ and 21.39 to 23.88 kg ha⁻¹ at the surface (0-10 cm) and sub surface layer (10-20 cm). However, the plot without the incorporation of green manure (T₀) resulted in a decrease amount of accessible phosphorous from its initial stages in surface and the sub surface layer. The highest accessible phosphorous was obtained from T₁ followed by T₃ and T₂ and the least was observed from T₀.

Figure 7.8 shows that in both the cropping year there was a significant variation (p < 0.05) in the amount of available phosphorous among the treatments before and after the incorporation of the green manure at both the depths. Furthermore, it was also observed that the soil accessible phosphorous was higher in the second cropping and in addition, it was further observed that the soil accessible phosphorous decreases from the surface to the sub- surface.

The inclusion of green manure legumes leads to considerable and favorable changes in the soil's available phosphorus compared to the beginning phases (before sowing of the legumes). Highest increased was observed in T₁ with value of 20.91 and 19.94 % (0-10 cm and 10-20 cm respectively), followed by T₃ with 18.51 % (0-10 cm) and 16.69 % (10-20 cm). However, T₀ (control) showed a decreasing percent at both 0-10 cm (10 %) and 10-20 cm (13 %) (Figure 7.9).

Available Phosphorous (kg ha ⁻¹)							
Treatments	0 – 10 cm		10-20 cm				
	Before	After	Before	After			
T1	25.91	31.29	19.90	23.88			
T2	25.50	28.86	19.79	21.39			
T3	25.55	30.08	18.83	21.91			
Τ0	24.27	21.70	18.00	15.57			
	CD _{0.05} S	SE(m) ±	CD _{0.05} S	E(m) ±			
Factor (A)	1.13	± 0.39	0.94 ± 0.32				
Factor (B)	0.80	± 0.27	$0.27 0.66 \pm 0.23$				
(A X B)	1.59	± 0.55	1.33 ± 0.46				

Table 7.6. Effect of green manure weed species on the soil available phosphorous(pooled for two consecutive years).

Factor A: indicates the Treatments where $T_1 = C$. *micans*, $T_2 = A$. *indica*, $T_3 = A$. *indica* and $T_0 = C$ ontrol.

Factor B: indicates soil available Phosphorous **before** sowing the leguminous crops and **after** incorporation of the legumes into the soil.

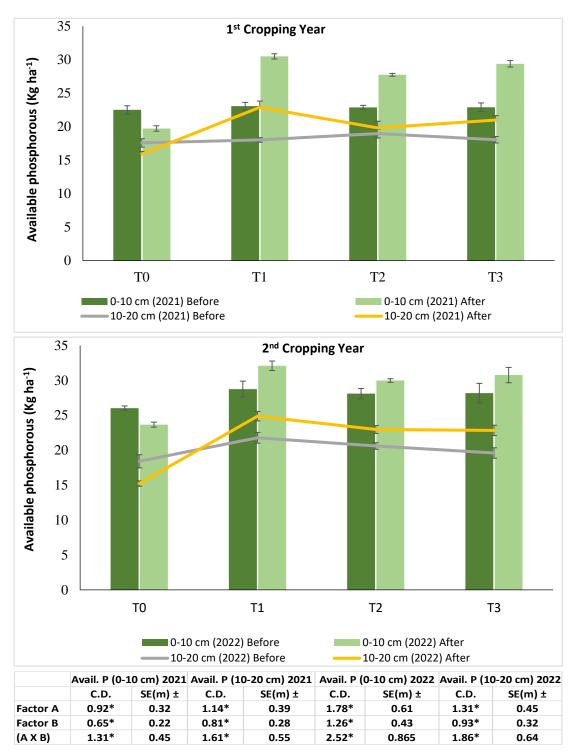


Figure 7.8. Effect of green manuring on the soil available phosphorous (Kg ha⁻¹) during the 1st and 2nd year cropping at 0-10 and 10-20 cm soil depth (Factor A= treatments, Factor B= before sowing the leguminous crops and after incorporation of the legumes into the soil, NS= non-significant and CD= Critical difference at 0.05 level).

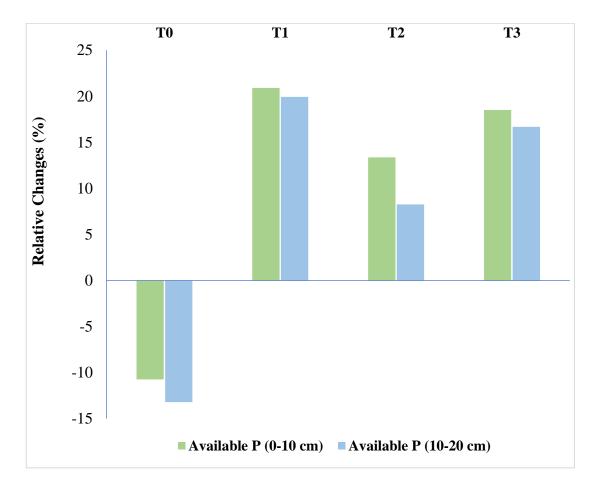


Figure 7.9. Relative changes in the soil available phosphorous (kg ha⁻¹) from the initial (before sowing of legume) and final (after the incorporation of the legume into the soil) at 0-10 and 10-20 cm soil depth.

7.3.1.6. Available Potassium

The accessible potassium in the soil surface and sub-surface layer was greatly influenced by the incorporation of different types of green manure legumes as evidenced by the results. Initial (before sowing of the legume) potassium content in the soil ranged from 298.9 to 336.20 kg ha⁻¹ (0-10 cm) and 228.74 to 248.48 kg ha⁻¹ (10-20 cm). The incorporation of *C. micans* (T₁) had the highest available potassium [474.88 kg ha⁻¹ (0-10 cm) and 307.69 (10-20 cm)] while the control (T₀) had the lowest [351.68 kg ha⁻¹ (0-10 cm) and 268.32 kg ha⁻¹ (10-20 cm)] as shown in Table 7.7.

At 0-10 cm soil depth in both the cropping year, a significant difference (p < 0.05) between the treatments before and after incorporating the green manure into the soil was observed, whereas at 10-20 cm, no difference between the treatment was observed. However, a significant difference in the second cropping year was observed among the treatments before and after incorporating the green manure into the soil (Figure 7.10). Additionally, it was discovered that the concentration of soil-available potassium in the surface soil layer was greater than that in the sub-surface layer.

The soil's available potassium exhibits favourable and significant changes in relation to the incorporation of green manure into the soil. T₁ (addition of *C. micans*) recorded the highest available phosphorous percent increase with value 38.9 % (0-10 cm) and 23.77 % (10-20 cm), followed by T₂ (addition of *C. mucunoides*) with value 35.83 % (0-10 cm) and 23.36 % (10-20 cm) and T₃ (addition of *A. indica*) with value 28.94 % (0-10 cm) and 18.43 % (10-20 cm). The least percent increase was obtained from T₀ (without any green manure) with 17.66 % and 17.32 % at 0-10 cm and 10-20 cm respectively (Figure 7.11).

	Available Potassium (kg ha ⁻¹)				
	0 – 10 cm		10-20 cm		
Treatments	Before	After	Before	After	
ТО	298.90	351.68	228.744	268.322	
T1	336.20	474.88	248.48	307.69	
T2	325.46	437.88	231.84	285.30	
Т3	318.24	410.21	237.85	281.608	
	$CD_{0.05}$ SE(m) ±		$CD_{0.05}$ SE(m) ±		
Factor (A)	18.28*	$18.28^* \pm 6.28$ $11.17^* \pm 3.23$		3 ± 3.83	
Factor (B)	$12.93^* \pm 4.49$ $7.9^* \pm 2.71$		± 2.71		
(A X B)	25.85	± 8.87	$NS \pm 5.43$		

Table 7.7. Effect of green manure weed species on the soil available potassium(pooled for two consecutive years).

Factor B: indicates soil available potassium **before** sowing the leguminous crops and **after** incorporation of the legumes into the soil.

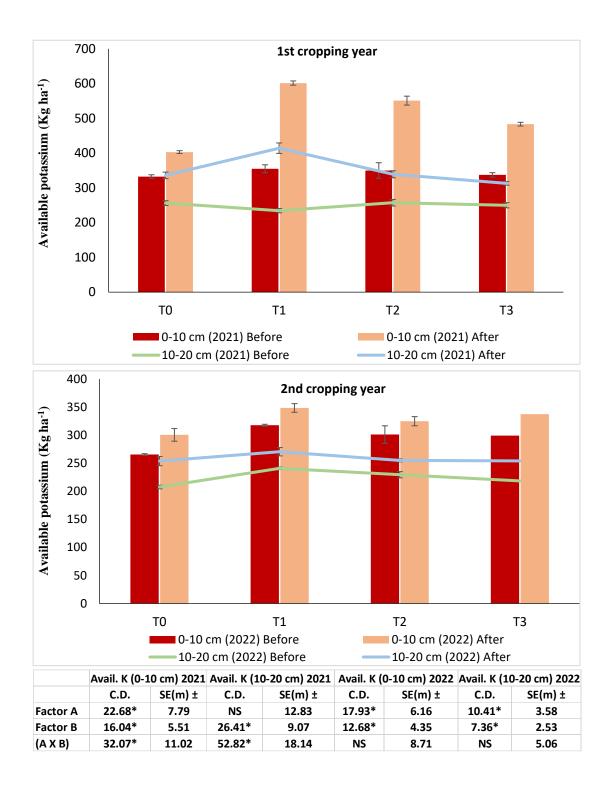


Figure 7.10. Effect of green manuring on the soil available potassium (Kg ha⁻¹) during the 1st and 2nd year cropping at 0-10 and 10-20 cm soil depth (Factor A= treatments, Factor B= before sowing the leguminous crops and after incorporation of the legumes into the soil, NS= non-significant and CD= Critical difference at 0.05 level)

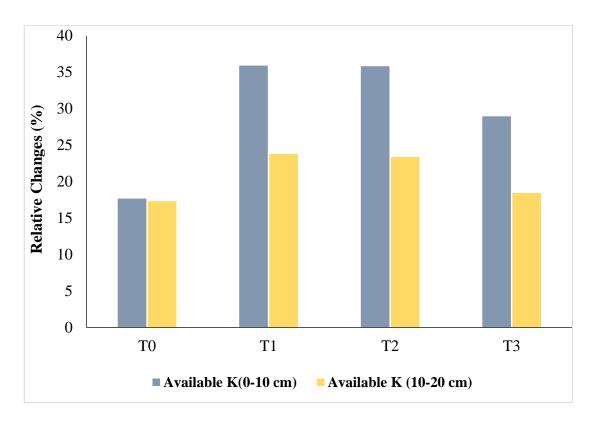


Figure 7.11. Relative changes in the soil available potassium (kg ha⁻¹) from the initial (before sowing of legume) and final (after the incorporation of the legume into the soil) at 0-10 and 10-20 cm soil depth.

7.3.2 Biological properties

7.3.2.1 Microbial biomass carbon

The soil biomass carbon content exhibited variations across different treatments, as indicated by the data presented in the Table 7.8. The incorporation of leguminous green manure into the soil had a considerable increase on the biomass carbon where T₁ had the highest soil MBC content [354.30 μ g g⁻¹ (0-10 cm) and 200.59 μ g g⁻¹ (10-20 cm)], followed by T₃ [243.98 μ g g⁻¹ (0-10 cm) and 192.31 μ g g⁻¹ (10-20 cm)], T₂ [244.83 μ g g⁻¹ (0-10 cm) and 184.09 μ g g⁻¹ (10-20 cm)] and the least by control (T₀) [212.47 μ g g⁻¹ (0-10 cm) and 157.10 μ g g⁻¹ (10-20 cm)].

Figure 7.12 depicted a significant disparity (p < 0.05) in the soil MBC at 0-10 cm across the treatments as well as before and after the incorporation of the green manure at first and second cropping year. However, at 10-20 cm in the second year there was no significant variation (p < 0.05) among the treatments but showed variation before and after incorporation of the green manure. Furthermore, the levels of soil microbial biomass carbon (MBC) exhibited a declining pattern as the depth of the soil increased.

After the incorporation of green manure, the soil MBC showed an increase trend in the surface and sub-surface layer. The change ranges from 13 to 60 % at the surface and 8 to 13 % at the sub-surface layer. T₁ had the maximum relative change, while T₀ had the minimum of 60.93 % and 13.46 % at 0-10 cm and 33.68 % and 8.56 % at 10-20 cm respectively (Figure 7.13). It was also observed the relative changes was higher in the surface layer for all the treatment than the sub-surface.

	Microbial biomass carbon (µg g ⁻¹)					
	0–10 cm		10-20 cm			
Treatments	Before	After	Before	After		
Τ0	187.72	212.47	144.61	157.10		
T1	220.13	354.30	151.43	200.59		
T2	194.89	243.98	151.04	184.09		
Т3	190.61	244.83	152.39	192.31		
	CD _{0.05}	SE(m) ±	CD _{0.05}	SE(m) ±		
Factor (A)	14.82	± 5.09	14.85	± 5.10		
Factor (B)	10.48	10.48 ± 3.60		± 3.61		
(A X B)	20.96 ± 7.20		NS	± 7.21		

 Table 7.8. Effect of green manure weed species on the soil microbial biomass

 carbon (pooled for two consecutive years)

Factor B: indicates soil MBC **before** sowing the leguminous crops and **after** incorporation of the legumes into the soil.

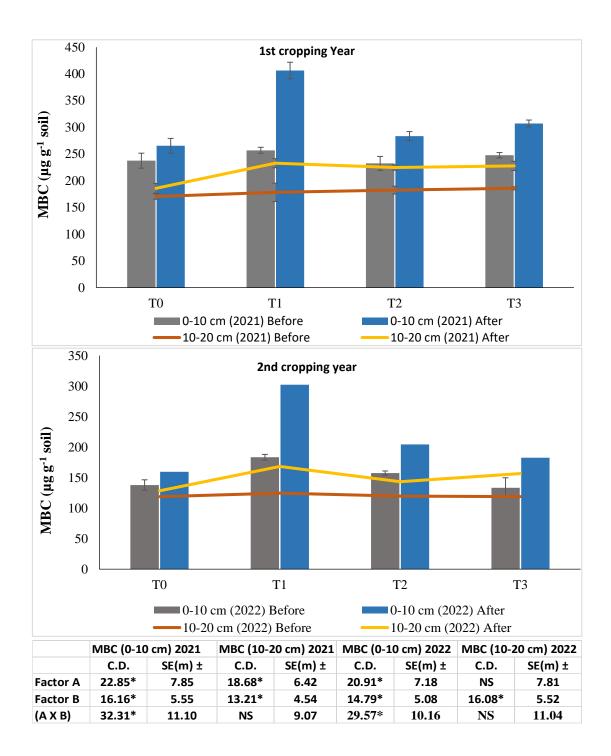


Figure 7.12. Effect of green manuring on the soil MBC (µg g⁻¹ soil) during the 1st and 2nd year cropping at 0-10 and 10-20 cm soil depth (Factor A= treatments, Factor B= before sowing the leguminous crops and after incorporation of the legumes into the soil, NS= non-significant and CD= Critical difference at 0.05 level).

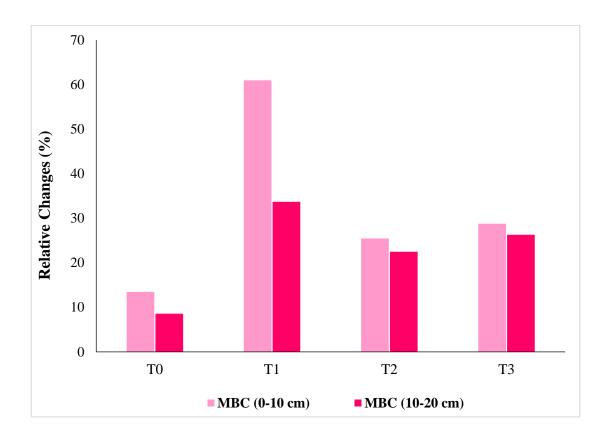


Figure 7.13. Relative changes in the soil MBC (µg g⁻¹ soil) from the initial (before sowing of legume) and final (after the incorporation of the legume into the soil) at 0-10 and 10-20 cm soil depth.

7.3.2.2 Microbial biomass phosphorous

Table 7.9 depicts the effect of the incorporation of green manure on the soil MBP at the surface and sub-surface layer. The concentration of soil MBP have increased after the addition of green manure legumes, however there was a decreased in the soil biomass phosphorous in the plot without the addition of green manure. The addition of *C. micans* (T₁) into the soil have reported the highest soil MBP with value of 13.28 μ g g⁻¹ (0-10 cm) and 8.42 μ g g⁻¹ (10-20 cm) followed by the incorporation of *C. mucunoides* (T₂) with value 10.45 μ g g⁻¹ (0-10 cm) and 8.25 μ g g⁻¹ (10-20 cm) and the lowest was obtained from control (T₀) with value 7.22 μ g g⁻¹ (0-10 cm) and 4.45 μ g g⁻¹ (10-20 cm). The soil MBP increases for all the treatments at the surface layer, however at the sub-surface the soil MBP decreases for the control.

A significant difference (p < 0.05) in the soil MBP at surface and sub-surface layer was observed across the various treatments before and after the incorporation of the green manure in both the cropping year (Figure 7.14). it was also recorded that the soil MBP was higher in the first year but gradually decreases as the cropping year increased. Furthermore, it was noted that the surface layer had the higher soil MBP than the sub-surface layer in both the cropping year.

The soil MBP content undergoes considerable changes after the integration of various green manure legumes, in contrast to the initial state (before sowing of the green manure). T₁ had the highest relative change with 47.56 % at the surface and 34.67 % at the sub-surface layer that was followed by descending order of $T_2 > T_3 > T_0$ at 0-10 and 10-20 cm (Figure 7.15).

Microbial biomass phosphorous (µg g ⁻¹)							
0–10 cm		10-20 cm					
Before	After	Before	After				
7.03	7.22	5.52	4.45				
8.73	13.28	6.25	8.42				
8.29	10.45	6.66	8.25				
7.63	9.14	6.03	7.29				
CD _{0.05} S	$CD_{0.05}$ SE(m) ±		SE(m) ±				
0.38 =	± 0.13	0.39 ± 0.14					
0.27	0.27 ± 0.1 $0.56 \pm$		± 0.1				
0.54 ± 0.19		NS ± 0.2					
	0-10 Before 7.03 8.73 8.29 7.63 CD _{0.05} S 0.38 = 0.27	0-10 cm Before After 7.03 7.22 8.73 13.28 8.29 10.45 7.63 9.14 CD _{0.05} SE(m) ± 0.38 \pm 0.13 0.27 \pm 0.1	0-10 cm 10-20 Before After Before 7.03 7.22 5.52 8.73 13.28 6.25 8.29 10.45 6.66 7.63 9.14 6.03 CD _{0.05} SE(m) \pm CD _{0.05} SE(m) \pm 0.38 \pm 0.13 0.39 0.56				

 Table 7.9. Effect of green manure weed species on the soil microbial biomass

 phosphorous (pooled for two consecutive years)

Factor B: indicates soil MBP **before** sowing the leguminous crops and **after** incorporation of the legumes into the soil.

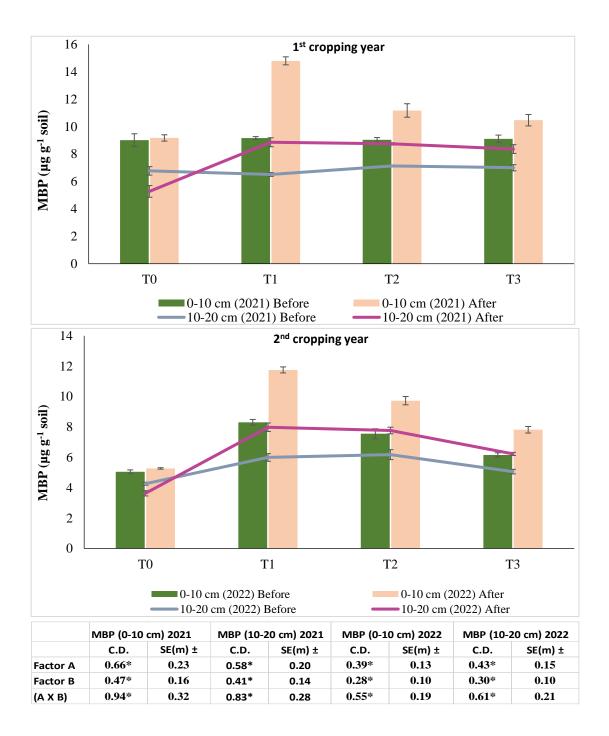


Figure 7.14. Effect of green manuring on the soil MBP (µg g⁻¹ soil) during the 1st and 2nd year cropping at 0-10 and 10-20 cm soil depth (Factor A= treatments, Factor B= before sowing the leguminous crops and after incorporation of the legumes into the soil, NS= non-significant and CD= Critical difference at 0.05 level).

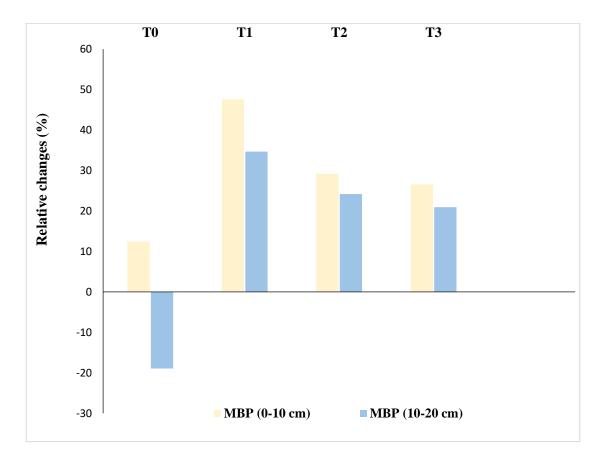


Figure 7.15. Relative changes in the soil MBP (μ g g⁻¹ soil) from the initial (before sowing of legume) and final (after the incorporation of the legume into the soil) at 0-10 and 10-20 cm soil depth.

7.3.2.3. Microbial population

The viable count of soil bacterial and fungal population in the surface and subsurface layer are depicted in the following Table 7.10. The initial bacterial population in the surface and sub-surface ranges from 1.45×10^8 to 1.92×10^8 CFU's ml⁻¹ and 1.24x 10^8 to 1.35×10^8 CFU's ml⁻¹ respectively. While the fungal population ranges from 0.67×10^6 to 0.76×10^6 CFU's ml⁻¹ and $0.60 - 0.64 \times 10^6$ CFU's ml⁻¹ at 0-10 cm and 10-20 cm respectively. The soil microbial population both fungal and bacterial population increased significantly after the incorporation of the different green manure legumes at the surface and sub-surface layer. After the incorporation of green manure, the largest bacterial population was seen in T₁ (incorporation of C. micans) at a depth of 0-10 cm, with a density of 2.44 x 108 CFU's ml⁻¹ and 1.80 x 108 CFU's ml⁻¹ at 10-20 cm which was followed by the incorporation of A. indica (T₂) (2.24 and 1.52 x 10^8 CFU's ml⁻¹ at 0-10 and 10-20 cm respectively), incorporation of C. mucunoides (T_3) with value of 2.13 and 1.34 x 10^8 CFU's ml⁻¹ and the least was depicted from the plot without the incorporation of green manure (T_0) with value of 1.73 and 1.32 at 0-10 and 10-20 cm respectively. In the case of the fungal population at the surface soil (0-10 cm) and sub-surface soil varied between 0.64 and 0.97 x 10⁶ CFU's ml⁻¹ soil and 0.55 and 0.80 x 10⁶ CFU's ml⁻¹ soil across different treatments (Table 7.10). The highest occurrence was seen after incorporating with C. micans (T_1) and the lowest in control (T_0) in 0-10 and 10-20 cm respectively.

Soil bacterial population in both the cropping year varied significantly (p < 0.05) among the treatments as well as before and after the incorporation of the green manure legume at both the soil depth (Figure 7.16). In case of the soil fungal population there was significant difference (p < 0.05) across the treatments as well as before and after the incorporation of the green manure legumes at 0-10 cm in both the cropping year. However, at 10-20 cm, no significant variation across the treatments was observed in the first cropping year but a significant variation among the treatments in the second year as well as before and after the incorporation of green manure in both the year (Figure 7.17).

Furthermore, it was noted that the bacterial population exhibited a decline as the soil depths increased across all the treatments.

When compared to the initial stages, it was observed that all the treatments have a significant relative change in the soil microbial community. The soil bacterial population depicted a positive relative change for all the treatments in both the soil depth, where the maximum change was recorded from T_1 with value of 26.89 and 36.43 % at the surface and sub-surface layer respectively and the least recorded from T_0 with value of 19.20 and 9.83 % at 0-10 cm and 10-20 cm respectively (Figure 7.18). However, in the case of the soil fungal population, a positive relative change was observed across the treatments with incorporation of green manure but showed a negative change in the treatments without any green manure incorporation (Figure 7.19), where T_0 (control) had the minimum and T_1 the maximum relative change value of-6 % and 27.67 % respectively at 0-10 cm and -5.59 % and 35.76 % respectively at 10-20 cm (Figure 7.19).

Bacteria (10 ⁸) CFU's ml ⁻¹				Fungi (10 ⁶) CFU's ml ⁻¹				
0-		10 cm 10-2		20 cm		0–10 cm		20 cm
Treatments	Before	After	Before	After	Before	After	Before	After
Τ0	1.45	1.73	1.24	1.32	0.69	0.64	0.62	0.55
T1	1.92	2.44	1.35	1.80	0.76	0.97	0.60	0.80
T2	1.81	2.24	1.26	1.52	0.67	0.85	0.64	0.78
Т3	1.77	2.13	1.24	1.34	0.68	0.82	0.60	0.68
	CD _{0.05}	SE(m) ±	CD _{0.05}	SE(m) ±	CD _{0.05}	SE(m) ±	CD _{0.05}	SE(m) ±
Factor (A)	0.08	± 0.03	0.06	± 0.02	0.05	± 0.02	0.05	± 0.02
Factor (B)	0.06	± 0.02	0.04	± 0.02	0.04	± 0.01	0.04	± 0.01
(A X B)	0.11	± 0.04	0.09	± 0.03	0.07	± 0.03	0.07	± 0.03

Table 7.10. Effect of green manure weed species on the soil microbial population (pooled for two consecutive years)

Factor B: indicates soil microbial population before sowing the leguminous crops and after incorporation of the legumes into the soil.

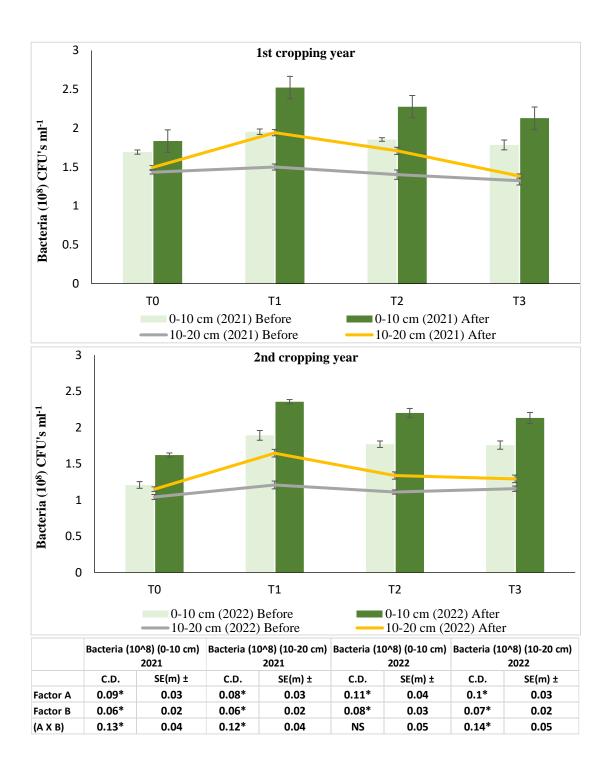


Figure 7.16. Effect of green manuring on the soil bacterial population during the 1st and 2nd year cropping at 0-10 and 10-20 cm soil depth (Factor A= treatments, Factor B= before sowing the leguminous crops and after incorporation of the legumes into the soil, NS= non-significant and CD= Critical difference, at 0.05 level).

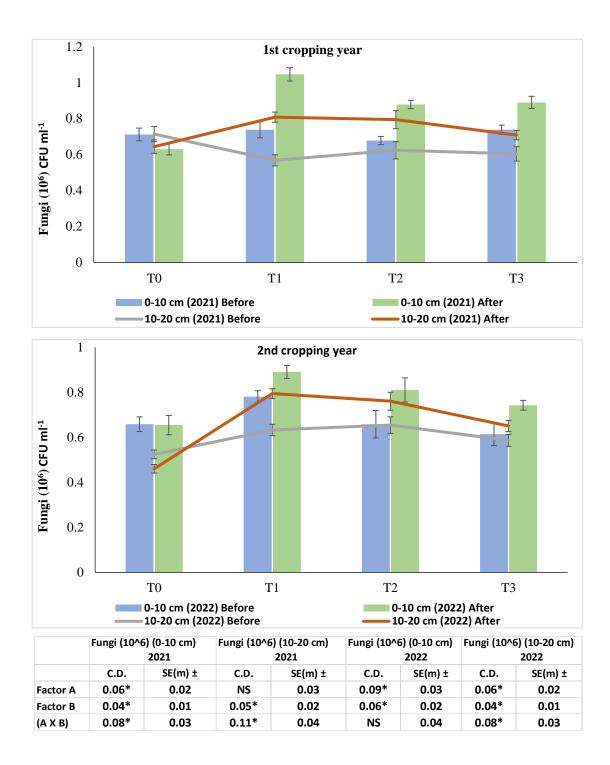


Figure 7.17. Effect of green manuring on the soil fungal population during the 1st and 2nd year cropping at 0-10 and 10-20 cm soil depth (Factor A= treatments, Factor B= before sowing the leguminous crops and after incorporation of the legumes into the soil, NS= non-significant and CD=Critical difference, at 0.05 level)).

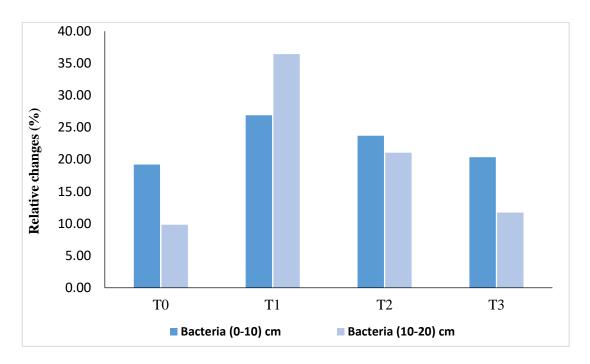


Figure 7.18. Relative changes in the soil bacterial population from the initial (before sowing of legume) and final (after the incorporation of the legume into the soil) at 0-10 and 10-20 cm soil depth.

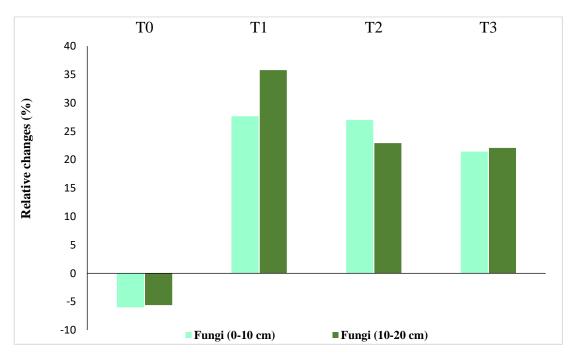


Figure 7.19. Relative changes in the soil fungal population from the initial (before sowing of legume) and final (after the incorporation of the legume into the soil) at 0-10 and 10-20 cm.

7.3.3. Effect of green manure weed species on the growth and yield of tomato

7.3.3.1. Plant height (cm)

Plant height is a crucial indicator of plant growth. It provides insight into forecasting the crop's growth rate and production. The plant heights at 30, 60, and 90 days after planting (DAT) are shown in Table 7.11. Examining data on the plant height over time shows a consistent growth in plant height as the crop ages.

Incorporation of *C. micans* (T₁) resulted in the tallest plants with values of 47.51, 75.47, and 96.39 cm at 30, 60, and 90 DAT respectively greatly surpassing the other treatments (Table 7.11). The least was recorded from T₀ with values of 40.15 cm (30 DAT), 63.85 cm (60 DAT), and 83.54 cm (90 DAT). The plant height was observed in the order of $T_1 > T_2 > T_3 > T_0$ throughout the study period. Green manuring significantly affected the height of tomato plants at all stages in both years of the research. T₁ had higher plant height in both years. However, the plant height decreased significantly (p < 0.05) from the first year to the second year (Figure 7.20) The height of the tomato plants went up steadily up to 90 days after planting, after which it stabilized till maturity, regardless of the treatments. Noticeable differences (p < 0.05) were found in the tomato plant across several treatments and cropping years at different growth stages.

Plant height (cm)				
Treatments	30 (DAT)	60 (DAT)	90 (DAT)	
T ₀	40.15	63.85	83.54	
T_1	47.51	47.51 75.47		
T ₂	44.51	68.32	89.04	
T ₃	43.94	64.14	84.64	
SE(m) ±	0.74	1.14	1.09	
CD _{0.05}	2.29	3.54	3.38	

Table 7.11. Effect of green manure weed species on the plant height in cm at 30, 60, and90 DAT (pooled data for two consecutive years)

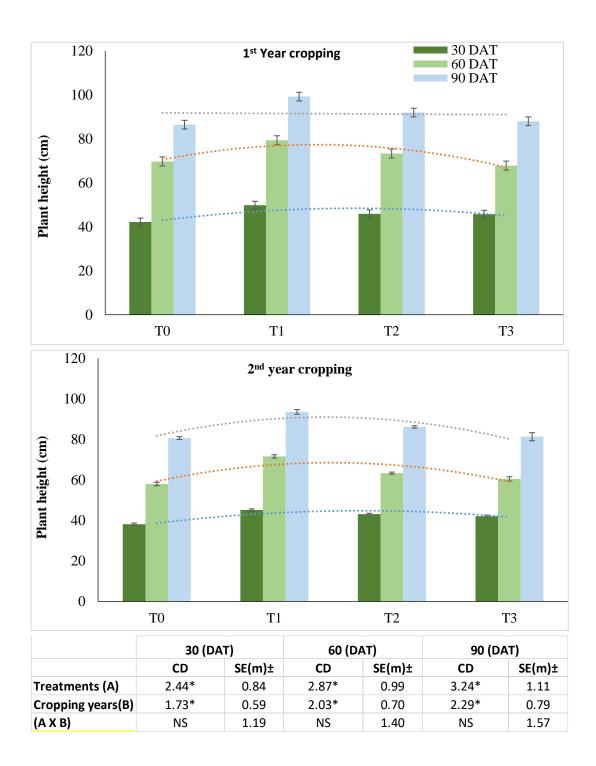


Figure 7.20. Effect of incorporation of the green manure species on the plant height (cm) during the 1^{st} and 2^{nd} year of cropping (p < 0.05).

7.3.3.2. Collar diameter (mm)

The collar diameter can serve as an indicator of the plant's general vigour and health. The average pooled data of the collar diameter at different growth stages of tomato plants are shown in Table 7.12. During the experiments, it was observed that the incorporation of *C. micans* (T₁) as green manure had the highest collar diameter (5.18, 6.35, and 7.72 mm at 30, 60 and 90 DAT) followed by the incorporation of *C. mucunoides* (T₂) and *A. indica* (T₃) with value of 4.83, 5.88 and 7.07 mm; 4.35, 5.76, 6.78 mm respectively at 30, 60 and 90 DAT. The lowest collar diameter was obtained when the tomato plants were treated without any green manure (T₀). T₁ had a higher collar diameter in both years at its growing stages. However, there was a significant (p < 0.05), decrease in the collar diameter from the first year to the second year (Figure 7.21).

Table 7.12. Effect of green manure weed species on the collar diameter (mm) at
30, 60, and 90 DAT (pooled data for two consecutive years)

Collar Diameter (mm)				
Treatments	30 (DAT)	60 (DAT)	90 (DAT)	
T ₀	4.20	5.42	6.948	
T_1	5.23	5.23 6.35		
T ₂	4.83	5.88	7.368	
T ₃	4.63 5.76		7.076	
SE(m) ±	0.06	0.07	0.09	
CD _{0.05}	0.18	0.23	0.31	

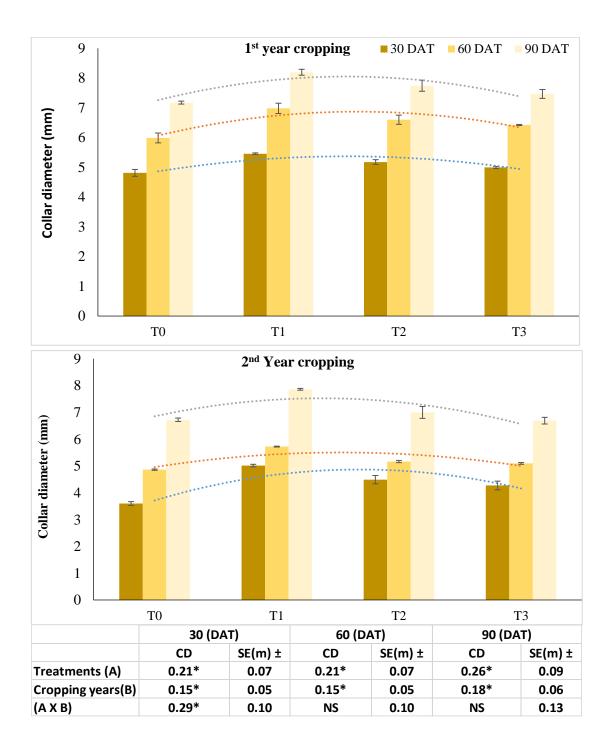


Figure 7.21. Effect of green manure species on the collar diameter (mm) of the tomato plant during the 1^{st} and 2^{nd} year of cropping (p < 0.05).

7.3.3.3. Number of primary branches plant⁻¹

The number of primary branches results at 30, 60 and 90 days after planting are displayed in Table 7.13. There were significant variations among the treatments at every growth stage of the plants. Incorporation of green manure *C. micans* (T_1) at 30, 60, and 90 DAT recorded the highest number of branches which was significantly higher than T_2 , T_3 , and control (T_0). The incorporation of green manuring considerably affected the number of branches of the tomato plants at all stages in both years of the study. The number of primary branches was higher in the first year and significantly it decreased in the second year (Figure 7.22).

Treatments	Number of primary branches plant ⁻¹			
	30 (DAT)	60 (DAT)	90 (DAT)	
T ₀	6.41	10.03	14.47	
T ₁	7.87	12.20	17.52	
T ₂	7.43	11.27	16.17	
T ₃	7.14	10.48	15.70	
SE(m) ±	0.2	0.27	0.17	
CD _{0.05}	0.61	0.84	0.54	

Table 7.13. Effect of green manure weed species on the number of branches plant⁻¹ at 30,60, and 90 DAT (pooled data for two consecutive years)

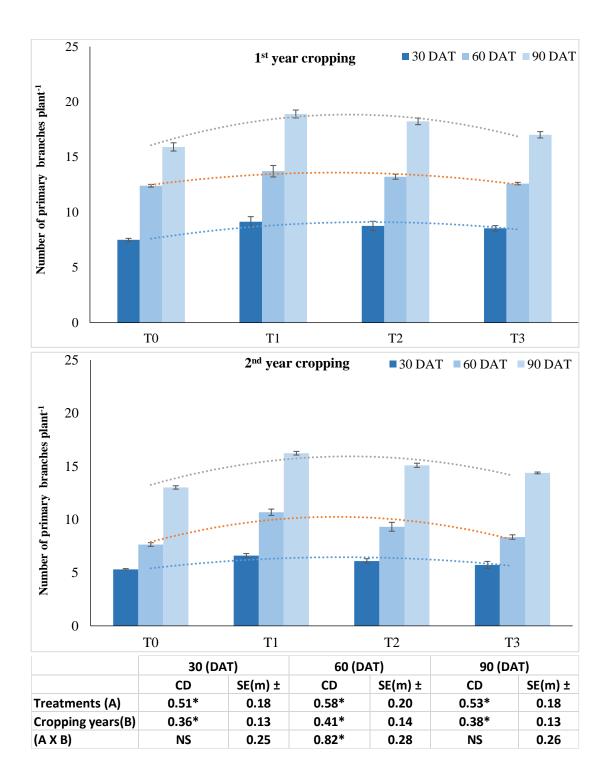


Figure 7.22. Effect of green manure species on the number of primary branches of the tomato plant during the 1^{st} and 2^{nd} year of cropping (p < 0.05).

7.3.3.4. Number of clusters plant⁻¹ and Number of fruits cluster⁻¹

The data reported in Table 7.14 demonstrates a significant effect of the application of green manures on the number of clusters per plant. The incorporation of *C. micans* (T₁) resulted in a statistically significant increase in the maximum number of clusters per plant with value of 6.56. Specifically, the recorded values after the incorporation of green manure *C. mucunoides* (T₂) and *A. indica* (T₃) were 5.53 and 4.93, which were higher than the control (T₀) values of 3.77 respectively. A notable disparity was seen in the number of clusters per plant across the various treatments during both the cropping years. Nevertheless, there was a substantial decline in the number of clusters per plant from the initial year to the subsequent year (Figure 7.23).

Based on the pooled data provided in Table 7.14, it can be observed that the incorporation of green manure legumes (T1, T2, T3) resulted in the higher number of fruits per cluster with 5.15, 4.08 and 3.45 respectively. The control group exhibited the lowest number of fruits per cluster, measuring 2.60. There was a significant difference observed in the number of fruits per cluster among the different treatments in both cropping years. However, a significant decrease in the number of fruits per cluster was observed from the first year to the next year (Figure 7.23) and in both the year the incorporation of *C. micans* recorded the maximum number of fruits per cluster when compared among the other treatments.

Treatments	Number of clusters plant ⁻¹	Number of fruits cluster ⁻¹
To	3.77	2.60
T_1	6.56	5.10
T ₂	5.53	4.08
T ₃	4.93	3.45
CD _{0.05}	0.25	0.31
SE(m) ±	0.08	0.10

Table 7.14. Effect of green manure weed species on the number of clusters plant⁻¹ and number of fruits clusters⁻¹ (pooled data for two consecutive years).

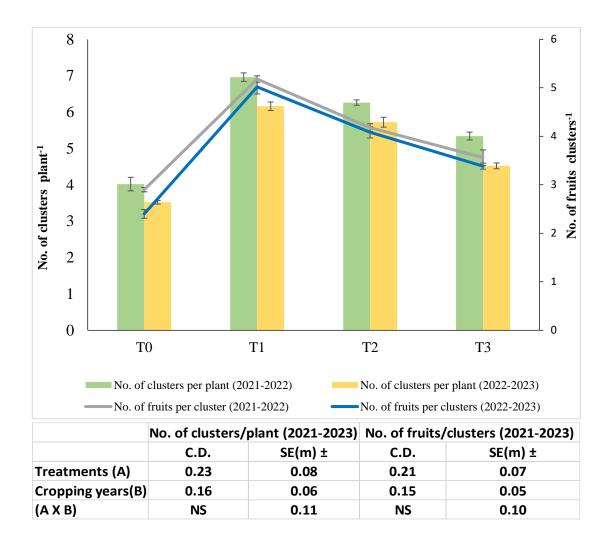


Figure 7.23. Effect of green manure species on the number of clusters plant⁻¹ and the number of fruits cluster⁻¹ during the 1st and 2nd year of cropping (p < 0.05).

7.3.3.5. Number of fruits plant⁻¹

The incorporation of green manure had a significant effect on the number of fruits plant⁻¹; a maximum of 16.13 fruits plant⁻¹ was recorded from T_1 followed by T_2 (12.20), T_3 (10.32), and the least by the control (8.68) as shown in Table 7.15. There was significant difference in the number of fruits among the treatments in both the cropping years. However, the number of fruits plant⁻¹ significantly decreased from the first year to the second year (Figure 7.24).

Table 7.15. Effect of green manure weed species on the number of fruits plant⁻¹ (pooled data for two consecutive years).

Treatments	Number of fruits plant ⁻¹
T ₀	8.68
T ₁	16.13
T ₂	12.20
T ₃	10.32
CD _{0.05}	0.83
$SE(m) \pm$	0.27

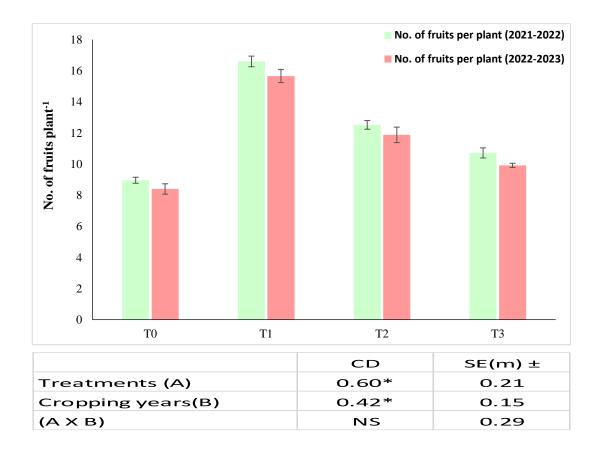


Figure 7.24. Effect of green manure weed species on the number of fruits plant⁻¹ during the 1st and 2nd year of cropping (p < 0.05).

7.3.3.6. Fruit length and breadth (cm)

The fruit length (cm) in the tomato plant differed substantially with the different treatments applied (Table 7.16). Comparatively lengthy tomato fruits (4.12 cm) were observed in the plants treated with the incorporation of *C. micans* (T₁) followed by the incorporation of *C. mucunoides* (T₂) with the value of 3.86 cm and incorporation of *A. indica* (T₃) with the value of 3.73 cm, and the least fruit length (3.47 cm) was recorded from control (T₀). The fruit length varied significantly (p < 0.05) among the various treatments and also in both the cropping years. However, there was a decrease in the fruit length from the first year to the second year (Figure 7.25).

The plot incorporated with *C. micans* raised the fruit breadth significantly (p < 0.05) as compared to the other plots treated with *C. mucunoides* and *A. indica* and also the control plot (Table 7.16). During both the cropping years the crop receiving *C. micans* (T₁) as green manure was recorded with the highest fruit breadth and the lowest was reported with control (Figure 7.25).

Treatments	Fruit Length (cm)	Fruit Breadth (cm)
T ₀	3.47	3.41
T_1	4.12	4.00
T ₂	3.86	3.71
T ₃	3.73	3.50
CD _{0.05}	0.23	0.20
$SE(m) \pm$	0.08	0.06

Table 7.16. Effect of green manure weed species on the fruit length and breadth(pooled data for two consecutive years).

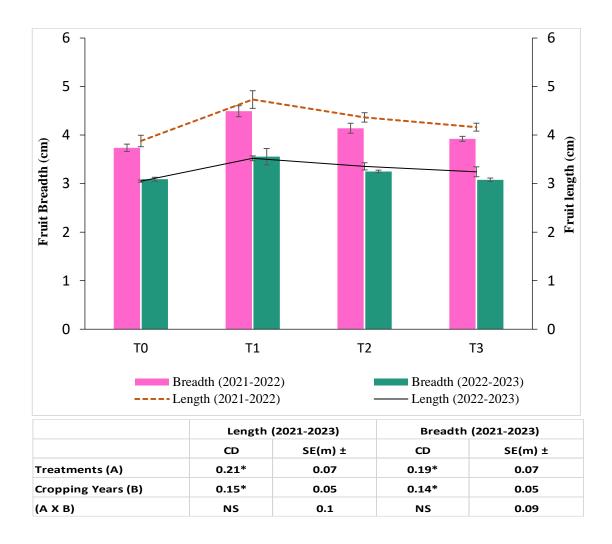


Figure 7.25. Effect of green manure weed species on the fruit length and fruit breadth during the 1^{st} and 2^{nd} year of cropping (p < 0.05).

7.3.3.7. Average weight of fruits (g) and fruit yield (q ha⁻¹)

Different green manures had a significant effect on the average fruit weight. T_1 recorded a higher fruit weight of 48.35 g in comparison to T_2 (40.75 g), T_3 (38.01 g), and T_0 (31.54 g) as seen in Table 7.17. It has also been observed that the fruit weight was significantly higher in the first year (Figure 7.26) and also the higher fruit weight was recorded from T_1 in both the cropping years.

Data pertaining to the fruit yield have been presented in Table 7.17. The fruit yield was highest where *C. micans* (T₁) was incorporated into the soil with value of 105.44 q ha⁻¹. Minimum fruit yield was obtained where there was no incorporation of green manure (T₀) with value of 54.10 q ha⁻¹. The fruit yield was significantly influenced by the incorporation of the green manure crops in the first and second year (Figure 7.27). However, the fruit yield was significantly higher in the first year.

Treatments	Average fruit weight (g)	Fruit yield (q ha ⁻¹)		
T ₀	31.54	54.10		
T_1	48.35	105.44		
T ₂	40.75	83.54		
T ₃	38.01	68.69		
CD _{0.05}	1.87	4.18		
$SE(m) \pm$	0.60	1.34		

Table 7.17. Effect of green manure weed species on the average fruit weight and fruit yield (pooled data for two consecutive years).

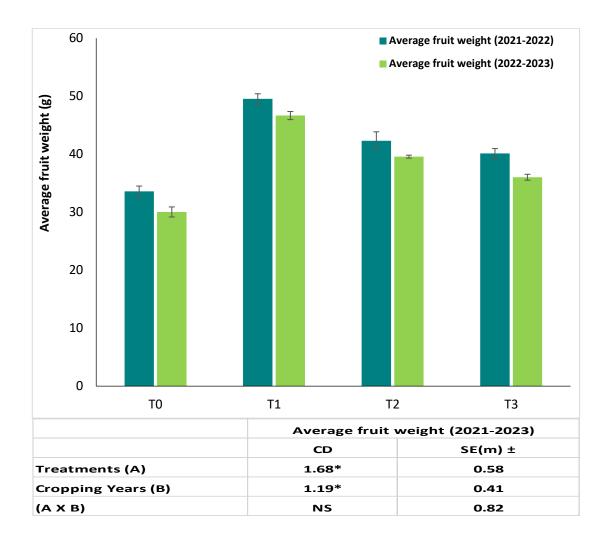


Figure 7.26. Effect of green manure weed species on the average fruit weight during the 1^{st} and 2^{nd} year of cropping (p < 0.05).

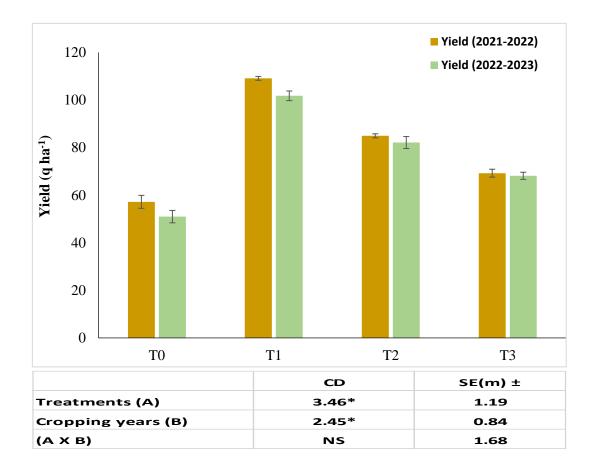


Figure 7.27. Effect of green manure weed species on the fruit yield during the 1^{st} and 2^{nd} year of cropping (p < 0.05).

7.3.4. Effect of green manure weed species on the economics.

7.3.4.1. Cost of cultivation

The examination of the data presented in the Table 7.18, indicates that the cost of cultivation exhibited significant variation mainly as a result of the utilization of different green manuring crops. The incorporation of the green manure *C. micans* (T₁) had the highest variable cost of cultivation with value of 1,74,433.67 Rs ha⁻¹ followed by *C. mucunoides* incorporated with value 1,73,962.67 Rs ha⁻¹. On the other hand, the incorporation of *A. indica* (T₃) followed by control (T₀) had the lowest cost of cultivation with value 1,72,697.47 and 114333.33 Rs ha⁻¹ respectively.

Treatments	Total Input Cost	Total Operational cost	Total Cost of cultivation
			(Rs ha ⁻¹)
To	6000	91,666.67	114333.33
T_1	7767	1,66,666.67	1,74,433.67
T ₂	7296	1,66,666.67	1,73,962.67
T ₃	6030.8	1,66,666.67	1,72,697.47

7.3.4.2. Gross, Net Return, and the Benefit Cost Ratio.

Data pertaining to the gross and net return have been presented in Table 7.19. The incorporation of *C. micans* (T₁) had the highest gross and net return with values of Rs 5,27,176.90 ha⁻¹ and Rs 3,52,743.30 ha⁻¹ respectively. The minimum was obtained from the control (T₀) with Rs 2,38,472.30 and Rs 1,24,139.00 ha⁻¹ respectively. It was noticed that there was a significant (p < 0.05) variation in the gross and net return among the various treatments as well as the cropping year (Figure 7.28). However, it was observed that the gross and net return significantly decrease from the first to the second year.

Table 7.19 displays data on the B: C ratio, showing that the treatment with *C*. *micans* incorporation (T₁) had the highest benefit-cost ratio of 3.02, greatly surpassing all other treatments. The minimum B:C was obtained from the incorporation of *A*. *indica* (T₃) with value 1.99. Figure 7.29 shows that there was significant difference (P < 0.05) in the B:C among the various green manure legumes as well as the control in both the cropping years. The order of B:C in first and second year was in the order was $T_1 < T_2 < T_0 < T_3$.

 Table 7.19. Effect of green manure weed species on the returns and Benefit cost

 ratio (pooled data for two consecutive years).

Treatments	Gross Return	Net Return	B:C
T ₀	238472.30	124139.00	2.09
T_1	527176.90	352743.30	3.02
T_2	417719.50	243756.80	2.40
T ₃	343467.30	170769.90	1.99
CD _{0.05}	19004.26	19005.66	0.13
$SE(m) \pm$	6100.04	6100.49	0.04

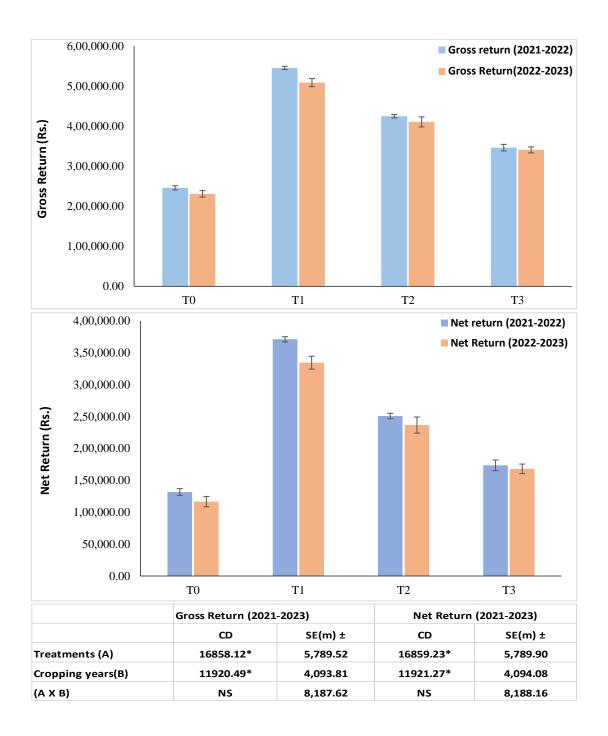


Figure 7.28. Effect of green manure weed species on the gross and net return during the 1^{st} and 2^{nd} year of cropping (p < 0.05).

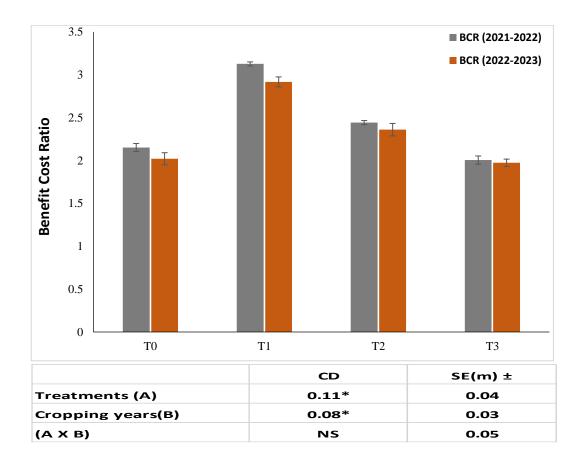


Figure 7.29. Effect of green manure weed species on the benefit cost ratio during the 1^{st} and 2^{nd} year of cropping (p < 0.05).

7.3.5. Correlation between the soil properties (physical, chemical and biological properties) and the plant parameters.

7.3.5.1. Soil physico-chemical properties with plant parameters.

The soil physical properties (moisture content) showed no significant correlation with all the plant parameters except for plant height, collar diameter and yield (Table 7.20). However, in the case of soil chemical properties, all the parameters except for the soil pH exhibited a significant positive correlation with all the plant growth and yield parameters. This indicates that green manure crops improved soil chemical characteristics, which may have increased tomato growth and productivity.

7.3.5.2. Soil biological properties with plant parameters.

The plant growth and yield parameters showed a positive significant correlation with all the soil biological properties (Table 7.21). This suggest that the soil biological properties had a significant influence on the growth and productivity of the tomato plants in both the cropping year.

Plant parameter	Moisture	рН	OC	Avail. N	Avail. P	Avail. K
Plant height	-0.48*	-0.08	0.90**	0.85**	0.59**	0.8**
Collar Diameter	-0.48*	-0.01	0.85**	0.83**	0.60**	0.76**
No. of branches	-0.31	-0.38	0.87**	0.93**	0.83**	0.87**
No. of fruits plant ⁻¹	-0.42	-0.19	0.97**	0.95**	0.**	0.87**
Fruit length	-0.25	-0.27	0.74**	0.83**	0.72**	0.83**
Fruit breadth	-0.43	-0.12	0.81**	0.82**	0.65**	0.81**
No. of clusters plant ⁻¹	0.44	-0.36	0.95**	0.98**	0.83**	0.94**
No. of fruits clusters ⁻¹	0.41	-0.27	0.91**	0.94**	0.75**	0.89**
Average fruit weight	-0.42	-0.28	0.92**	0.95**	0.83**	0.93**
Fruit yield	-0.51*	-0.26	0.96**	0.98**	0.76**	0.91**

Table 7.20. Correlation between the soil physico-chemical properties and plant parameters.

* Correlation is significant at the 0.05 level; ** Correlation is significant at the 0.01 level.

Plant parameter	MBC	MBP	Bacteria	Fungi
Plant height	0.88**	0.92**	0.77**	0.76*
Collar Diameter	0.80**	0.90**	0.61**	0.75*
No. of branches	0.76**	0.91**	0.90**	0.88*
no. of fruits plant ⁻¹	0.91**	0.95**	0.77**	0.76**
Fruit length	0.75**	0.81**	0.75**	0.70**
Fruit breadth	0.81**	0.87**	0.90**	0.76**
Average fruit weight	0.86**	0.97**	0.87**	0.90**
No. of clusters plant ⁻¹	0.82**	0.96**	0.85**	0.88**
No. fruits cluster ⁻¹	0.82**	0.97**	0.84**	0.91**
Fruit yield	0.81**	0.95**	0.83**	0.89**

Table 7.21. Correlation	hetween th	e soil biological	nronerties and	nlant narameters
Table 7.21. Correlation	Detween in	ic son biological	properties and	plant parameters

* Correlation is significant at the 0.05 level; ** Correlation is significant at the 0.01 level.

7.9. Discussion

7.9.1. Soil physico-chemical properties

7.9.1.1. Soil moisture content

Within our study, it was revealed that the application of green manure resulted in a little reduction in soil water content in the surface layer (0-10 cm) but a higher reduction in the sub-surface layer (10-20 cm). This suggests that green manure had a greater impact on deep soil water levels compared to surface soil water levels. Other research has reached similar outcomes (Alvarez *et al.*, 2018; Ma *et al.*, 2021)

Green manure has an impact on rainfall infiltration, runoff, soil water evaporation, and transpiration (Eshel *et al.*, 2015; Yu *et al.*, 2016). However, the fact that the soil moisture content depends on various other factors like the climatic conditions, topography, soil characteristics, vegetation, and land-use patterns (Rasheed *et al.*, 2022). Planting green manures during the less precipitation period, used up a significant amount of soil water compared to bared field (Xue *et al.*, 2017).

During the period of green manure growth, the water used for its growth in the top layer of soil can be replenished by rainfall. However, the amount of limited rainfall that is unlikely to penetrate deep into the soil. This could explain the variations in water content at different depths of the soil (Ma *et al.*, 2021).

7.9.1.2. Soil pH

The soil pH was significantly reduced with the addition of green manure legumes into the soil in both the cropping year compared to the control (without any green manure. These could be caused by the release of organic acid into the soil during plant decomposition, leading to a decrease in soil pH. The current outcome aligns with the findings of Adekiya *et al.* (2019a); Rani *et al.* (2020); Abera *et al.* 2021. Salahin *et al.* (2013) also observed that the application of green manure in soils led to a drop in soil pH. Furthermore, contrary to certain case studies that suggest a substantial impact of green manure on soil pH, meta-analysis by Ma *et al.* (2021) revealed that green manure treatment in northern China did not have a significant influence on soil pH.

7.9.1.3. Soil organic carbon

In our study, incorporating green manure instead of leaving the area fallow resulted in an increase in soil organic carbon content. However, the effect varied across different soil depth and types of green manure species incorporated. C. micans incorporation significantly increased the organic carbon content among all the treatments in both the cropping year (Table 7.4), which may be attributed to the higher biomass accumulation of C. micans (Chapter 4), which results in an increased organic carbon content of the soil and improved nutrient availability, as previously observed by other researchers (Dwivedi et al., 2005; Pooniya et al., 2012). This outcome is consistent with the observations made by Ehsan et al. (2014); Abera et al. 2021, who observed an augmentation in organic carbon content due to in-situ cultivation incorporation of Sesbania sp. According to Souza et al. (2018) and de Melo et al. (2019), certain plant species from the Fabaceae family, such as Crotolaria ochroleuca, Crotolaria spectabilis, and Mucuna pruriens used as green manure, have the ability to enhance the soil organic carbon. The findings also align with Pawar et al. (2019); Zhang and Fang (2007), who observed that the addition of crop residues and green manures led to an increase in organic matter.

Soil organic matter serves as both a supplier and a reservir of plant nutrients in soils. It plays a crucial role in preserving soil structure, facilitating the penetration of air and water, enhancing water retention, and minimizing erosion (Gregorich *et al.*, 1994). In their study, shown that the addition of organic manure or green manure cropping considerably enhanced the storage of the organic carbon in soil.

7.9.1.4. Available nitrogen

Our study demonstrates that the application of green manure resulted in an increase in the availability of nitrogen in the soil compared to the control plot left bare during the study period. This could perhaps be linked to the release of nitrogen by their roots and nodules during their growth. The treated plots of *C. micans* exhibited a significantly higher concentration of accessible soil nitrogen compared to the other green manure treated plots as well as the plot without any green manure, likely because of the greater biomass incorporation. The study aligned with the findings of Mandal *et*

al. (2003). The decrease in soil pH in our present study may also anticipated to have a substantial positive effect on the availability of soil nitrogen. Similar findings were reported by Abera and Gerkabo (2021).

Not only may green manure reduce surface runoff, but it can also slow down the infiltration of precipitation into the soil during the fallow season, which in turn limits the movement of nitrogen. According to Couedel *et al.* (2018), the Azotobacter that is found in the rhizosphere of legume green manure has the ability to fix huge quantities of nitrogen from the atmosphere. This results in the creation of areas that are relatively rich in nitrogen and a significant increase in the amount of nitrogen present in the soil.

7.9.1.5. Available phosphorous

The incorporation of green manure greatly enhanced the availability of P that is an essential soil element for crop growth, especially in the surface layer. The incorporation of green manure accelerated the conversion of inaccessible phosphorus into accessible P, which might have resulted in a substantial enhancement in the availability of P in the soil. This remark aligns with the findings of the study conducted by Ma *et al.* (2021). Comparing among the treatments, the available phosphorous were highest after the incorporation of *C. micans* in both the cropping year (Figure 7.8). The increased levels of soil P in the plot with Crotalaria might be as a result of the breakdown of the green manure that contains a significant amount of P that contributes to the release of these elements into the soil. Similar results were observed, where Crotolaria species added higher amounts of soil available phosphorous (Ziblim *et al.*, 2013; Carvalho *et al.*, 2015).

The availability of phosphorus in soil can also be linked to the decomposition of organic matter and the conversion of insoluble compounds into soluble forms by an increasing population of microorganisms due to the incorporation of green manure (Rani *et al.*, 2020). A recent study demonstrated that intercropping *S. guianensis* as green manure, enhances soil phosphatase activity, perhaps leading to an increase in soil accessible phosphorus content by intercropping (Zhou *et al.*, 2019).

7.9.1.6. Available potassium

The incorporation of the leguminous green manure species significantly enhanced the availability of potassium (kg ha⁻¹) in the surface and sub-surface layer in both the cropping year. The increase in soil available potassium could be due to the deep root structure of green manure crops that enhances the soil's physical condition and releases CO_2 and organic acids, that aid in the disintegration of organic potassium in the soil, thereby boosting the availability of potassium. The findings of our study were supported by several other researchers (Dhar *et al.*, 2014; Adekiya *et al.*, 2019a; Islam *et al.*, 2019) who observed increased levels of soil potassium as a result of incorporating the green manure into the soil.

7.9.2. Soil biological properties

7.9.2.1. Soil microbial biomass carbon

Microbial biomass serves as both an indication of soil quality and the primary driver of nutrient cycling that controls the availability of nutrients to crops and is influenced by factors such as moisture, temperature, and other environmental conditions (Shah *et al.*, 2010).

After incorporating green manure, the soil MBC showed an increasing trend in the surface and sub-surface layer in the present investigation. The change in the soil MBC ranges from 13 to 60 % at the surface and 8 to 13 % at the sub-surface layer. The rise in soil microbial biomass carbon can be attributed to the inclusion of readily decomposable organic components, which enhance microbial activity. This is in line with the findings by Tejada *et al.* 2008. The increase in the soil microbial biomass carbon might be associated to the fact that green manure makes a better living environment for microbes due to its high energy and nutrient content (Ochoa-Hueso *et al.*, 2017; Ju *et al.*, 2019).

7.9.2.2. Soil microbial biomass phosphorous

Application of green manure into the soil, similar to soil MBC resulted in the increase in the soil microbial biomass phosphorous content in the soil rhizosphere. Differences in the quality and quantity of organic matter input, by the plant material that went through process of decomposition after incorporating into the soil, serve as a source for organisms in the soil, which in turn affects the soil microbial biomass. According to Chen *et al.* (2005), a drop in soil organic carbon (SOC) leads to a reduction in the amount of microbial biomass in the soil. The incorporation of the green manure has enhanced the soil physico-chemical and the biological properties, and this might be the reason for the gradual build-up of the soil microbial biomass phosphorous content (Haripal and Sahoo, 2014).

7.9.2.3. Soil microbial population

The incorporation of green manure substantially enhanced the bacterial and fungal population in the soil when compared to the plot without any green manure. The rise in microbial population in the soil may be associated to the increased influx of organic matter from green manure legumes and higher cropping intensity (Stromberger *et al.*, 2007). The enhancement of fertility in organic farming systems has implications for soil biological features, such as the microbial population (Robertson *et al.*, 2000; Mader *et al.*, 2002).

Other studies have also reported a comparable enhancement in the population of bacteria and fungi due to the use of green manure legumes. For instance, Biederbeck *et al.* (2005) reported a 385 % rise in the bacterial population and a 210 % increase in the fungal population following the use of green manure compared to fallow-wheat cropping. Similarly, a study by Kataoka *et al.* 2017 observed that hairy vetch used as green manure increased the fungal biomass when compared to plots with fertilizer and non-green manure. Several other studies showed that the incorporation of green manure significantly increased the soil microbial community (Ding *et al.*, 2021; Zhu *et al.*, 2022).

7.9.3. Growth Parameters of tomato

The results of the current study revealed that the growth parameters of the tomato crop, such as plant height, number of primary branches and collar diameter at all growth stages, differed considerably across the various treatments (Figure 7.20, 7.21 and 7.22) during both years of experimentation. Further data revealed that the incorporation of C. micans (T1) recorded higher values of growth parameters for the tomato crop. In this research, tomato growth response to green manure may be attributed to increased nitrogen content, phosphorus, potassium, and other nutrients derived from green manure decomposition. Maximum growth parameters of tomato crop by T₁ may be attributed to improved availability of major and micronutrients in plots treated with biomass inclusion. This statement is in line with Makinde (2007); Rajeshwari et al. (2007). The incorporation and decomposition of biomass may aid in the mobilization of nutrients from unavailable to available forms, as described by Balyanetal (2006); Balai et al. (2011). According to Liu et al. (2007), and Tonfack et al. (2009), the amount of nitrogen available is determined by the total biomass integrated. C. micans biomass releases more nitrogen minerals than other legumes, leading to improved N uptake by tomato plants. This promotes vegetative development, resulting in increased plant height, collar diameter and number of primary branches. As stated by Muazu and Yusuf (2021), green manure provides slowrelease nutrients, leading to more efficient nutrient usage.

The enhanced development of tomato plants in the green manure plots, compared to the control plot, in this experiment can be due to the higher levels of organic matter, nitrogen, and potentially other nutrients that are released from the incorporated green manure.

7.9.4. Yield of Tomato

The total performance of the tomato plant, as measured by the growth rate, was an excellent predictor for yield throughout the research period. The treatments' positive impact on tomato yield was most likely a result of the increase in organic matter, which led to improved soil physical and chemical conditions. This statement is in line with Boparai *et al.* (1992), Mandal *et al.* (2003), Adekiya (2019a). The observed enhancements in the yield of tomato following the addition of green manure align with findings reported by other researchers (Togun *et al.*, 2004, Akanni and Ojeniyi, 2007). The timing of green manure application affects yields, which vary depending on rainfall distribution and manure content (Boffa, 1999).

Incorporating green manures into the soil led to improved tomato growth and fruit yield compared to the control. Increased tomato performance from green manure compared to control, could be due to the increase soil OM, N, P, K and soil biological biomass. The study found a significant and positive correlation between the growth and yield parameters of tomato plants and the levels of soil organic carbon, N, P and the soil biological properties (Table 7.20 and Table 7.21). These soil characteristics were found to have a major influence on the growth and yield of tomato plants in this study.

The decrease in the crop yield during the second year (Figure 7.27) compared to the first year might be attributed to the slight lower pH after the incorporation of green manure, could have inhibited the uptake of nutrients due to the acidity and the damage it caused to the roots (Undie *et al.*, 2013; Agbede *et al.*, 2018b).

7.9.5. Economics

When compared to all the other treatments, the incorporation of *C. micans* (T₁), provided larger gross return, net income and a better B:C ratio. The higher productivity of the tomato fruit that was achieved through the use of *C. micans* (T₁) led to an increase in both the return and the benefit cost ratio. In situ green manuring is a method that is both possible and inexpensive, and it has the potential to increase both production and profitability (Kumar *et al.*, 2011; Sajjad *et al.*, 2019). According to the findings of another study, green manuring led to a 16.8 percent rise in comparison to the control group. It was shown that cowpea green manure had a positive overall performance on wheat, and the benefit was Rs 10283 ha⁻¹ (Sajjad *et al.*, 2018). Another study by Bhayal *et al.* (2018) revealed that green manuring intercropping resulted in the highest net returns of Rs 21581 ha⁻¹ and a benefit cost ratio of 2.25.

7.10. Conclusion

The incorporation of the green manure legumes led to substantial increases in soil organic carbon content, highlighting their role in promoting soil organic matter accumulation and long-term fertility. Notable increases were observed in available nitrogen, phosphorus, and potassium following the incorporation of green manure legumes. The observed enhancements suggest that studied legume weeds can serve as effective sources of plant-available nutrients, reducing the reliance on external inputs such as chemical fertilizers. Significant increases in the soil biological properties were observed, indicating improvements in soil microbial activity and nutrient cycling. The incorporation of green manure significantly impacted tomato plant growth parameters and yield over two consecutive cropping years. Overall, the incorporation of *C. micans* consistently resulted in superior plant growth and higher fruit production, to other treatments and the control. The findings indicate that the use of green manure practices using the local legume weeds can be effective in enhancing soil health and enhancing the tomato plant growth, yield, and economic returns, thereby contributing to sustainable agricultural practices and improved crop production.

CHAPTER 8

GENERAL DISCUSSION

Organic manures have been viewed as an economical and primary means of providing plant nutrients and improving soil quality. The exclusive reliance on chemical fertilizers is causing the soil to become infertile and less productive in the absence of organic matter. Green manuring is the most significant form of organic manure. Green manuring is the act of ploughing or incorporating un decomposed green plants or their residue into the soil to enhance its physical structure and fertility. The implementation of green manure offers numerous benefits in terms of chemical, physical, and biological attributes. These include enhancements in organic matter levels, microbial activity, nutrient cycling, alleviation of compacted layers, reduction in erosion, incidence of pests and diseases, and suppression of weed growth. These improvements contribute to the establishment of a sustainable fruit production system.

The fertility of the soil determines its ability to give nutrients to crops during their active development stage. The presence of organic matter in soil creates a favourable environment for plant growth. As a result, green manuring is a vital technique in the soil. Mineralization of organic matter in soil releases plant nutrients such as nitrogen, phosphorus, potassium, and micronutrients. Aside from its own stock, the organic acid created during the decomposition of green manure crops converts a number of additional elements found in the soil's mineral components into accessible forms. Reducing soil pH temporarily accelerates nutrient release for plants. Green manures are a potential way to maintain sustainable nutrients for crop growth.

Mizoram is rich in biodiversity, with many plant species that would ordinarily be classified as weeds yet have the potential to be used as green manure crops. In the present study an attempt has been made to assessed the green manuring potential of three leguminous weeds of Mizoram. The overall findings of the study have been discussed herewith in connection to the various objectives mentioned in Chapter 1.

8.1. Biomass and nutrient accumulation

Several factors hinder crop growth and development. Ideal environmental conditions and soil fertility help crops thrive and produce more. The leguminous species' genetic makeup during the investigation undoubtedly affected their development characteristics. Soil moisture, temperature, and physical, chemical, and biological properties affect root growth (Fageria, 2009).

According to the present findings *C. micans* produced higher shoot and root biomass. Crop biomass yield depends on genotype, sowing date, management, soil and meteorological conditions, growing season, and plant population (de Silva *et al.*, 2020). Similar observation was recorded by Naidu *et al.* (2017), where the crop growth and yield varied significantly among genotypes. The species' biomass differences may also be due to temperature, climate, and light (Rahman *et al.*, 2020).

The N content in the shoot biomass of *C. micans* (28.25 g kg⁻¹), *C. mucunoides* (14.40 g kg⁻¹), and *A. indica* (21.28 g kg⁻¹) was lower than the N content of the green manure crops reported by Duarte *et al.* (2013). Similarly, Pereira *et al.* (2007) found substantial differences (p < 0.05) in N, P, K, Ca, and Mg levels among cover crops. The legume under examination had more phosphorus than Duarte *et al.* (2013) and Cazatta *et al.* (2005). Our species had a substantially greater total P and K content (shoot and root) than Mauad *et al.* (2019). The micronutrient content was in the order Fe > Na > Zn > Cu. The nutrient accumulation in the shoot and root biomass, except for Ca in the root biomass. Genetic variations in leguminous species may explain the differences in shoot and root biomass nutrient accumulation in this and other studies.

Green manure	Harvesting days	Biomass accumulation (t ha ⁻¹)		References	
	uuys	Shoot	Root		
Indigofera tinctoria	30	0.10	0.01		
	45	0.34	0.06		
	60	2.20	0.41		
Sesbania aculeata	30	0.63	0.13	Miah <i>et al.</i> (2015)	
	45	3.78	0.42		
	60	11.30	1.52		
Sesbania rostrata	30	0.49	0.27		
	45	2.57	0.46		
	60	9.72	1.59		
Sesbanea aculeata		4.51			
Sesbanea rostrata		5.12			
Crotolaria juncea		5.25		Irin and Biswas	
Vigna radiata	45	2.86		(2021)	
Vigna unguiculata		3.1	-		
Leucaena leucocephala		3.19			
Vigna mungo		2.6			
Mimosa pudica		2.68			
Crotolaria juncea		1.10			
Crotolaria spectabilis		1.29		Dos Santos	
Mucuna pruriens	60	1.16	_	Nascimento et al.	
Dolichos lablab		1.04		(2021)	
Canavalia ensiformis		1.65			
Crotolaria junceae		4.7			
Sesbania aculeata	60	5.2	_	Rani et al. (2022)	
Vigna radiata		4.2			
Crotolaria micans	90	15.6	1.65		
Aeschynomene indica		2.1	0.36	Present study	
Calopogonium mucunoides		2.36	0.28		

Table 8.1. Dry biomass accumulation of green manure species.

8.2. Decomposition and nutrient dynamics

C. Micans observed 45 % reduction in dry mass in the first 30 days, and about 90 % in 90 days. 30 % of the initial dry mass for *C. mucunoides* and *A. indica* was loss in the first month. *C. mucunoides* experiences a reduction of around 90% of its dry mass by the 5th month, but *A. indica* exhibits a gradual decomposition pattern after the second month. Decomposition across species was faster initially, then slowed. This phenomenon has been described by several writers (Bohara, 2019; Hou, 2021). Each species exhibited distinct decomposition patterns over the 180 days of incubation period. *C. micans* decomposed most rapidly, followed by *C. mucunoides* and *A. indica*. Decay rate constants ranged from 5.5 to 8.21 *k* year⁻¹, with corresponding half-life values for mass loss ranging from 32.99 to 46 days. The decay rate constant of various green manure legume species is represented in Table 8.2.

C. micans and A. indica released K > P > N during decomposition, but C. mucunoides released K > N > P. Hasanuzzam and Hossain (2014) found that agroforest tree litter released K > N > P. Lalremsang *et al.* (2022), Das and Mondal (2016), had comparable release patterns, with K having the highest rate. Due to its high mobility and leaching of water-soluble minerals, K concentration released during decomposition is substantial (Jeong et al., 2015; Patricio, 2012). Strong microbial activity increases nitrogen demand, causing the rapid drop in N content during breakdown. The N mineralization process of green manures correlated positively with the N content and negatively with cellulose, hemicellulose, C/N, and C/P ratios (Table 5.5). The findings match with Halde and Entz (2016), Watthier et al. (2020). Incubation showed that C. micans and C. mucunoides plant residue decomposed and released nitrogen faster than A. indica. Palm et al. (2001) recommended that high nitrogen (> 2.5 %) and phosphorus (> 0.25 %) during decomposition enhance P mineralization, and in this study, C. micans had a higher phosphorus content than A. indica and C. mucunoides (Table 5.1), which may explain why it released phosphorus faster. Fast phosphorus (P) mineralization is connected to the depletion of soluble P in plant tissue vacuoles (Watthier et al., 2020). The residues' inorganic and soluble phosphorus concentration and microbial activity in the organic fractions affect phosphorus release (Giocomini *et al.*, 2003). The nutrient mineralization of various green manure legume species is represented in Table 8.3.

Species	k year ⁻¹	Reference
Crotalaria juncea,	0.84	
Cajanus cajan,	0.91	Pereira et al. (2016)
Crotalaria spectabilis,	1.94	
Mucuna deeringiana	1.09	
Canavalia ensiformis	4.16	
Cajanas Cajan	2.56	Mangaravite et al. (2023)
Mucuna deeringiana	3.58	
Crotolaria juncea	1.93	
Calopogonium mucunoides	0.95	
Crotalaria spectabilis	0.89	Mendonca et al. (2017)
Dolichus lablab	0.84	
Crotolaria micans	8.21	
Calopogonium mucunoides	7.15	Present study
Aeschynomene indica	5.5	

 Table 8.2. Decay constants of different leguminous green manure species.

Species	<i>k</i> day ⁻¹	Reference
Crotalaria juncea,	$k_{\rm N} = 0.008$	
	$k_{\rm P} = 0.012$	
	$k_{\rm K} = 0.01$	
Cajanus cajan,	$k_{\rm N} = 0.008$	
	$k_{\rm P} = 0.009$	
	$k_{\rm K} = 0.01$	
Crotalaria spectabilis,	$k_{\rm N} = 0.011$	Pereira et al. (2016)
1	$k_{\rm P} = 0.013$	× ,
	$k_{\rm K} = 0.013$	
Mucuna deeringiana	$k_{\rm N} = 0.003$	
0	$k_{\rm P} = 0.005$	
	$k_{\rm K} = 0.007$	
Canavalia ensiformis	$k_{\rm N} = 0.012$	
0	$k_{\rm P} = 0.020$	
	$k_{\rm K} = 0.027$	
	$k_{\rm Ca} = 0.007$	
	$k_{\rm Mg} = 0.011$	
Mucuna deeringiana	$k_{\rm N} = 0.01$	
5	$k_{\rm P} = 0.022$	
	$k_{\rm K} = 0.023$	
	$k_{\rm Ca} = 0.006$	
	$k_{ m Mg}=0.01$	Mangaravite et al. (2023)
Cajanas Cajan	$k_{\rm N} = 0.01$	
	$k_{\rm P} = 0.023$	
	$k_{\rm K} = 0.02$	
	$k_{Ca} = 0.006$	
	$k_{\rm Mg} = 0.008$	
Crotolaria juncea	$k_{\rm N} = 0.004$	
	$k_{\rm P} = 0.024$	
	$k_{\rm K} = 0.019$	
	$k_{\rm Ca} = 0.006$	
	$k_{\rm Mg} = 0.008$	
Crotolaria micans	$k_{\rm N} = 0.006$	
	$k_{\rm P} = 0.007$	
	$k_{\rm K} = 0.011$	
Calopogonium mucunoides	$k_{\rm N} = 0.006$	Present study
	$k_{\rm P} = 0.003$	
	$k_{\rm K} = 0.008$	
Aeschynomene indica	$k_{\rm N} = 0.005$	
	$k_{\rm P} = 0.005$	
	$k_{\rm K} = 0.01$	

 Table 8.3. Nutrient release rate of different green manure species.

8.3. Nodulation and Nitrogen Fixation

In the present study significant variation in the number of nodules were observed among the three species. *A. indica* had the most nodules, regardless of rootlet nodulation, of the three species whereas in the case of *C. mucunoides*, no nodulation was observed in the first month. Root nodulation is affected by species genetics, environmental conditions, growth conditions, sample site, and plant species (Gehlot *et al.*, 2012). In all three species, nodules originated in the primary root and grew in the secondary and tertiary roots later in plant growth. Reduced surface area, infection site, and enzymatic activity may explain the decrease in nodulation per rootlet compared to its parent root. The total nodule produced (irrespective of rootlets) in the present study was within the range that was higher than the study reported by Ranjan *et al.* (2007) who studied on the nodulation behaviour of *Acacia mangium*, *Albizia lebbek*, *Gliricidia sepium*, *Leucaena diversifolia* and *Leucaena leucocephala*. The nitrogen content in plant sections differed significantly between the three species in this investigation. Both *C. micans* and *C. mucunoides* had increased shoot and root nitrogen levels in this study.

In accordance with the data presented in Table 6.8, there was significant variation (p < 0.05) in the nitrogen uptake where, highest nitrogen uptake was recorded by *C. micans*, which was 550.97 mg plant⁻¹. The rate at which plants grow, the active-passive mechanism of the root surface, the availability of nitrogen, the physiological qualities of the soil, and the prevailing climatic conditions are some of the elements that have an impact on the amount of nitrogen that plants are able to take in. This difference can be related to the varying efficiency of nutrient uptake by the species, which is impacted by various soil conditions, as well as the efficiency of nutrient usage. Both of these factors are responsible for the observed difference. In addition, different plant components have different ways of utilizing nutrients, which may also be influenced by the microclimate of the area. The nitrogen fixation of various green manure legumes is represented in Table 8.4.

Species	Method	Nitrogen fixed	Reference
	used		
Crotolaria juncea	N-	58 kg ha ⁻¹	Nezomba <i>et al.</i> (2008)
Crotolaria pallida	difference method	173 kg ha ⁻¹	
Acacia catechu	Difference	14.10 kg ha ⁻¹ yr ⁻¹	
Albizia chinensis	method	8.84 kg ha ⁻¹ yr ⁻¹	Kashyap <i>et al</i> . (2012)
Alnus nitida		7.41 kg ha ⁻¹ yr ⁻¹	
Lupinus albus	-	167.7-196.0 kg ha ⁻¹	Sulas <i>et al.</i> (2016)
Crotolaria juncea	-	174 kg ha ⁻¹	Perin <i>et al</i> . (2006)
Crotolaria micans	Difference	243.36 kg ha ⁻¹ yr ⁻¹	
Calopogonium mucunoides	method	72.46 kg ha ⁻¹ yr ⁻¹	Present study
Aeschynomene indica		152.07 kg ha ⁻¹ yr ⁻¹	

 Table 8.4. Nitrogen fixation by different species

8.4. Soil properties

8.4.1. Soil physico-chemical properties

Significant differences (p < 0.05) in the soil pH, available N, P and K and the soil organic carbon were identified among the different treatments before and after incorporating the green manure legumes into the soil and also the interaction between the two components (Table 7.3, 7.4, 7.5, 7.6 and 7.7). However, the soil moisture in the surface and sub-surface did not show any variation among the treatments before and after incorporating the green manure legumes (Table 7.2).

The significant decrease in the soil pH of the plots treated with the leguminous green manure crops as compared to the control plot could be attributed to its acidic character due to the CO₂ and organic acid generation during the decomposition process (Agbede *et al.*, 2018b).

The decomposition of green manure crops recycles nitrogen, phosphorus, and potassium in the plant nutrients cycle (Carvalho *et al.*, 2015). Green manure can offer nutrients by legume nitrogen fixation or quick decomposition of high-nutrient crops (Chimouriya *et al.*, 2018). Green manure crops, particularly legumes, can boost soil N supply for following crops by biological N fixation. Not only can green manure crops produce nitrogen, but they also add organic matter to the soil.

In inorganically-managed or integrated soil systems, soil organic phosphorus mineralization has been demonstrated for significantly contributing to plant phosphorus requirements (Oehl *et al.*, 2001). Additionally, the incorporation of green manure crops enhances biological phosphorus cycling in soil and improves the dissolution and bioavailability of sparingly-soluble phosphates (Cavigelli and Thien, 2003). Mineralization occurs in the soil through the recycling of microbial P following microbial mortality and predation which helps in generating the organic P (Rani *et al.*, 2022). The presence of K in the soil may be attributed to the input of potassium to the soil, as well as the decrease in potassium fixation and the release of potassium owing to the interaction of degraded green manure biomass with clay (Das *et al.*, 2004).

8.4.2. Soil biological properties

Significant differences (p < 0.05) in the soil microbial biomass carbon, microbial biomass phosphorous, soil's bacterial and fungal population, were observed between the treatments both before and after incorporating the green manure legume weeds in both the cropping year. However, the soil's MBC, MBP and the bacterial and fungal population decreased from the surface to the sub-surface layer.

Any soil management approach that increases soil organic matter affects soil microbial biomass and biological characteristics. Therefore, the inclusion of green manure legumes weeds may be a suitable suggestion for these soils, particularly for enhancing soil biological health. According to Chen *et al.* (2005), a reduction in soil organic carbon leads to a decrease in the amount of microbial biomass in the soil. The rise in soil microbial biomass carbon and phosphorous may also be attributed to the addition of readily decomposable organic molecules, which enhances microbial activity (Tajeda *et al.*, 2008). The soil microbial biomass exhibited a notable decline as the soil depth increased. This could be attributed to a reduction in the amount of organic carbon in the soil as the soil depth increases. The soil microbial biomass is highly responsive to even little alterations in the organic matter composition of the soil, as this immediately functions as a source of energy (Wang *et al.*, 2007).

The results demonstrated that the incorporation of green manure legume weeds significantly increased the bacterial and fungal population in both the cropping years. The rise in microbial population in soil may be attributed to the increased amount of organic matter introduced by green manure legumes and the intensity of cropping (Stromberger *et al.*, 2007; Shah *et al.*, 2009). Other studies have also reported a comparable enhancement in the population of bacteria and fungi due to the use of green manure legumes (Ding *et al.*, 2021; Zhu *et al.*, 2022).

8.5. Growth and yield of tomato

The study found that tomato crop growth metrics such as plant height, number of primary branches, and collar diameter at all growth stages varied greatly across treatments (Figure 7.20, 7.21, and 7.22) over both years of trial. However, all the growth variables of the tomato plants were lower in the second year. The incorporation of *C. micans* (T₁) resulted in the maximum growth of the tomato plant followed by T₂ (*C. mucunoides*), T₃ (*A. indica*) and the minimum by T₀ (Control).

Green manuring is a sustainable agricultural practice that affects the physical features of plants. It effectively improves growth parameters by supplying adequate nutrient levels for crop development (Meng *et al.*, 2019). The decomposition of the green manure legumes is facilitated by soil microorganisms, which convert nutrients from inaccessible forms into available ones. This process has a positive impact on growth parameters and generates sustainable soil conditions, leading to enhanced crop production compared to non-green manure fields. Green manure releases organic matter, nitrogen, and other essential nutrients, that may explain why tomato plants in the green manure plots grew better than those in the control plot (Liu *et al.*, 2007, Tonfack 2009). Total biomass of the legumes integrated determines nitrogen availability (Liu *et al.*, 2007). *C. micans* biomass releases more nitrogen minerals than other legumes, helping tomato plants absorb N. This encourages vegetative growth, increasing plant height, collar diameter, and primary branches. Green manure releases nutrients slowly, improving nutrient use, according to Muazu and Yusuf (2021).

The fruit yield was significantly influenced by the incorporation of the green manure crops in the first and second year. The yield of tomato was significantly (p < 0.05) higher in the plot incorporated with *C. micans* (T₁) in both the cropping year. The treatments' favourable effect on tomato yield can be attributed to the rise in organic matter, which subsequently enhanced soil physical and chemical conditions. This assertion is consistent with the findings of Mandal *et al.* (2003), Adekiya *et al.* (2019a). The increase in tomato output resulting from the incorporation of green manure legumes is consistent with the findings reported by other studies (Togun *et al.*, 2004; Akanni and Ojeniyi, 2007). The study found a strong and favourable relationship

between the growth and yield characteristics of tomato plants and the amounts of soil organic carbon, nitrogen, phosphorus, and the soil's biological features. This connection is supported by the data presented in Table 7.20 and Table 7.21. The study revealed that these soil factors have a significant impact on the growth and productivity of tomato plants. Moreover, the beneficial influence of legume green manuring on soil organic matter and other soil characteristics, which results in improved nutrition for the growing crop, clearly increased the crop yield by several other researchers.

However, the reduction in crop yield during the second year (Figure 7.27) compared to the first year may be due to the slight decrease in pH following the addition of green manure. This decrease in pH could have hindered nutrient uptake due to increased acidity and subsequent damage to the roots (Agbede *et al.*, 2018b).

8.6. Economics

Significant variation (p < 0.05) was observed in the Gross return, Net return and the Benefit Cost Ratio among the treatments as well as different cropping years. However, their non-significant difference was observed in the interaction between the two components (Figure 7.28 and 7.29).

Among all the other treatments, the inclusion of *C. micans* (T_1) resulted in higher gross return, net income, and a superior B:C ratio, that is attributed to the higher tomato fruit productivity compared to the other treatments. In addition to increasing crop yields, organic farming with green manure can also lead to cost savings and higher profits (Madhukumar *et al.*, 2018).

Moreover, *in situ* green manuring is a viable and cost-effective approach that has the capacity to enhance both output and profitability (Sajjad *et al.*, 2019). Another study revealed that incorporation of *Crotolaria junceae* as green manure resulted in the highest net returns and B:C ratio of Rs 21581 ha⁻¹ and 2.25 respectively (Bhayal *et al.*, 2018).

Overall, the study revealed that that leguminous weed species exhibits significant biomass and nutrient buildup. Additionally, it was further demonstrated that the rate of breakdown and the capacity to fix nitrogen were both high. As the biomass of the legume species undergo decomposition, they enhance microbial activity, which in turn promotes the creation of humus and enhances soil fertility. As a result, these activities provide a more advantageous environment for the tomato plants, improving their development and productivity which ultimately increased the benefit cost ratio. However, the tomato fruit yield was lower in the second year, possibly due to the nutrients being trapped in the soil microbial biomass, resulting in reduced yield.

SUMMARY AND CONCLUSION

8.1. Summary

The study entitled "Assessment of green manuring potential of leguminous weeds of Mizoram" was carried out in Mizoram University, Tanhril, Aizawl, Mizoram, India (23°44'22"N, 92° 39' 54" E and 950 masl).

Agriculture has always endowed human civilization with crucial resources for existence and progress. As the global population grows, demand rises, putting unprecedented pressure on agricultural systems. Optimizing agricultural productivity requires appropriate soil fertility. This can be achieved with proper soil and crop management. To maintain soil fertility for long-term crop production, organic manures, particularly green manuring, are essential. Green manuring has been popular in India for decades, but food production pressure, competitive and profitable crops, and cheap and expensive chemical fertilizers have limited its appeal. Organic producers and low-input farms using conventional agronomic methods have rediscovered it because to soil access, fertility depletion, and public concerns about abuse and energy conservation. Humus and organic matter in green manure technology improve soil fertility, structure, water retention, erosion prevention, and microbiological growth.

Recent government interest in organic and natural farming has led to laws in most nations. Long-term agricultural system viability and eco-friendly agriculture awareness are rising. To address these issues, agricultural production with minimal inorganic and maximum organic inputs for soil health is suggested. Recent global climate change has led governments and scientists to adopt land management strategies, particularly agricultural systems, to minimize GHG emissions and boost carbon sequestration. The Indian government have approved the entire North East region as an organic hub.

Mizoram is diverse and has many weeds that can be used as green manure. Green manuring crops can increase soil fertility, in situ water conservation, weed control, and soil-borne pathogen management. These green manure crops help enrich organic soil. Farmers might choose new locally available plants as green manure crops using the data.

To carry out the study, three abundantly found leguminous weed species viz: Crotolaria micans Link., Aeschynomene indica L. and Calopogonium mucunoides Desv. were selected to evaluate their green manuring potential after being crosschecked with their accession number by the Botanical survey of India, Shillong. The present study was carried out with the following objectives: to estimate the biomass production and nutrient content of the selected species as green manure plants, to estimate the decomposition and nutrient release pattern, to estimate the nitrogen fixation of the selected species and to estimate the effect of the selected species on the soil properties and the growth and yield of the tomato crops. The biomass productivity (both aboveground and belowground) of the selected legumes was estimated by harvest method. For this purpose, seeds of the selected species were collected from the wild and grown in experimental plots to monitor their growth and similarly harvested after 90 DAS, to estimate their biomass productivity and nutrient accumulation. The decomposition and nutrient release pattern of the weed species was determined by placing the air-dried samples in a litterbag and monitoring for 180 days. A poly pot experiment was conducted to determine the nitrogen and the amount of N fixed by each species was calculated by the difference method and expressed in terms of kg ha⁻ ¹yr⁻¹. In order to determine the impact of the legume weeds on the soil properties and the growth and yield of tomato plants, the experiment comprised of 4 treatments viz: T₁ (Crotolaria micans), T₂ (Calopogonium mucunoides), T₃ (Aeschynomene indica and T₀ (Control). Soil samples were collected before sowing the legume seeds and 60 days after incorporating the green manure into the soil. Analysis on various soil physical, chemical and biological properties were carried out. 60 days incorporating the legume weed species, tomato seedlings (Arkha Rakhshak variety) were transplanted to the plots and thereafter the growth parameters at successive stages were observed and recorded. The tomato yield and various yield parameters was calculated using standard methods.

The major findings of the study have been summarized as below:

1. Biomass production and nutrient accumulation of the selected species

- i. The shoot length and number of branches varied significantly (p < 0.05) among the species but showed no significant changes in the first and second year. While the collar diameter was found to vary significantly among the species and the cropping year. However, for the root length there was no significant difference among the species as well as the cropping year.
- ii. *C. micans* recorded for the maximum collar diameter (7.1 mm), shoot and root length (91.29 cm and 29.04 cm respectively) and number of branches (27.00).
- The fresh and dry biomass of the shoot and root of the species significantly vary among the species but showed no variation in the cropping year. However, the biomass decreased from 1st year to 2nd year.
- iv. Biomass production assessments underscored *C. micans* exceptional capacity to accumulate substantial amount of fresh and dry biomass of shoot (15.16 and 2.90 t ha⁻¹ respectively) root (1.65 and 0.54 t ha⁻¹ respectively).
- v. The shoot and root biomass of the three leguminous weed species exhibited significant differences in the nutrient content (p < 0.05) in both the cropping year.
- Vi. *C. micans* demonstrated statistical superiority (p < 0.05) in the accumulation of N (81.86 kg ha⁻¹), P (15.03 kg ha⁻¹), K (63.31 kg ha⁻¹), Ca (18.37 kg ha⁻¹), Mg (0.37 kg ha⁻¹), Na (357.03 g ha⁻¹), Fe (1499.89 g ha⁻¹), Zn (72.49 g ha⁻¹) and Cu (44.92 g ha⁻¹) in the shoot biomass.
- vii. C. micans had a greater capacity for N (12.42 kg ha⁻¹), P (2.56 kg ha⁻¹), K (6.84 kg ha⁻¹), Mg (1.04 kg ha⁻¹), Na (474.88 g ha⁻¹), Fe (952.94 g ha⁻¹), Zn (33.99 g ha⁻¹) and Cu (20.25 g ha⁻¹) accumulation in the root biomass. However, the C. mucunoides exhibited the highest Ca accumulation (0.30 kg ha⁻¹).

viii. The nutrient accumulation in the shoot and root biomass differed significantly in both the cropping years, with the exception of potassium and Cu accumulation in the root biomass, which did not show significant variation in cropping years.

2. Decomposition and nutrient release pattern of the selected legume weed species

- i. The initial chemical composition exhibited significant variation among the legume species. *C. micans* recorded the highest P (0.27 %), K (1.55 %) and C (38.12 %) content. *A. indica* observed highest Lignin (16.98 %), N (3.11 %), Total phenol (1. 80 %), crude fiber (21.74 %) and L/N ratio (5.45). *C. mucunoides* recorded the highest cellulose (32.19 %), C/N ratio (17.22) and C/P ratio (200.23).
- ii. Each species exhibited distinct decomposition patterns over the 180 days incubation period. *C. micans* decomposed most rapidly with decay rate 8.21 k year⁻¹ followed by *C. mucunoides* (7.15 k year⁻¹) and *A. indica* (5.5 k year⁻¹).
- iii. After 180 days, the decomposition time, there were 0.18 g (1.8 %), 0.25 g (2.85 %), and 0.66 g (6.64 %) of undecomposed dry material left from the original mass of *C. micans*, *C. mucunoides*, and *A. indica*, respectively.
- iv. The maximum half-life (t₅₀) and t₉₉ was recorded by *A. indica* with 46 and 331.89 days respectively.
- v. *C. micans* and *A. indica* released nutrients in the order of K > P > N, while *C. mucunoides* released them in the order of K > N > P.
- Vi. The N, P and K mineralization per year was higher in *C. micans* with value of k_N (2.31), k_P (2.45) and k_K (3.86) respectively. At the end of the incubation period N mass remaining (%) was 0.69 %, 1.50 % and 3.33 % for *C. micans*, *C. mucunoides* and *A. indica* respectively. The P mass remaining (%) were 0.72 %, 2.72 % and 3.41 % for *C. micans*, *C. mucunoides* and *A. indica* respectively. The percentage of K mass remaining in the plant residues for *C. micans*, *C. mucunoides*, and *A. indica* were found to be 0.32 %, 1 %, and 1.31 % correspondingly

- vii. Lignin, total phenol, Crude fiber, L/N and N/P ratio was negatively correlated with the decay rates of which the phenolic content ($R^{2}= 0.976$) of the plant litter is considered to have a strong influence on the decomposition rate.
- viii. While nitrogen release showed strong positive correlation with lignin and N content but negative correlation with the C/N ratio. Strong negative correlations were observed between hemicellulose and cellulose with phosphorus and potassium release rates, respectively.

3. Nodulation behavior and nitrogen fixation of the selected legume weed species

- Nodulation began on the primary roots for both *A. indica* and *C. micans* with 6.57 and 2.71 respectively. No nodulation was observed in the one-month-old seedlings of *C. mucunoides*.
- ii. In the second month, *A. indica* obtained the maximum number of nodules from the primary root with a value of 20.57. While *C. mucunoides* produced the highest number of nodules in the tertiary roots.
- iii. With respect to the root categories, the maximum number of nodules was obtained from the secondary roots for *A. indica* (306.74) and tertiary roots for *C. micans* (18.14) and *C. mucunoides* (12.43) in the third month.
- iv. With the increase in time the nodules in the primary root decreased for all the species.
- v. The fresh biomass of the primary root nodules of *A. indica* was higher comparing to the other species in all the study period (1st, 2nd, 3rd and 6th month). However, the secondary root nodules of *C. micans* in the 1st and 2nd month and the tertiary root nodules in the 1st, 2nd, 3rd and 6th month was higher comparing to the other legume species.
- vi. The dry biomass of the nodules was directly proportional to the fresh biomass of the nodules.
- vii. The nitrogen content exhibited variation across the shoots, roots, and nodules of different species, with *C. micans* demonstrating the maximum nitrogen level

in its shoot (2.07 %) and root nodules (4.78 %). However, the maximum nitrogen level in the root was recorded in *C. mucunoides* (1.96 %).

viii. C. micans (243.36 kg ha⁻¹ year⁻¹) was the most efficient nitrogen fixer, followed by A. indica (152.07 kg ha⁻¹ year⁻¹) and C. mucunoides (22.55 kg ha⁻¹ year⁻¹) with significant differences in nitrogen fixation rates.

4. Impact of the leguminous weed species on the soil physico-chemical properties

- i. The soil moisture contents were at par among the treatments hence displayed a non-significant variation, however, the moisture content vary after the incorporation of the leguminous weeds as green manure when compared to the initial stages. T₃ recorded the highest with value of 23.35 % and 22.21 % at 0-10 and 10-20 cm respectively and the least was recorded by T₁ had the lowest at the surface (20.44 %) and sub-surface layer (18.88 %).
- ii. The results revealed significant change in soil pH, with a decreasing trend observed following the incorporation of the selected species as green manure compared to treatments without green manure (T₀).
- iii. The incorporation of green manure legumes led to substantial increases in soil organic carbon content. T₁ exhibited the highest organic carbon with value of 2.02 % (0-10 cm) and 1.39 % (10- 20 cm) and the least was observed from the plot without any green manure legumes (T₀) with value 1.30 % (0-10 cm) and 0.79 % (10-20 cm).
- iv. Notable increase was observed in the soil available nitrogen following the incorporation of the selected green manure legume weeds. T_1 recorded the highest nitrogen levels both before sowing the legume seeds (359.45 kg ha-1 and 239.69 kg ha⁻¹ at 0-10 and 10-20 cm respectively as well as after incorporation of the legume respectively, with values of (512.35 kg ha⁻¹ and 369.27 kg ha⁻¹ at 0-10 cm and 10-20 cm).
- v. The incorporation of green manure legumes leads to significant (p < 0.05) increase in the soil's available phosphorus compared to the initial phases (before sowing of legumes) in both the cropping year. The highest accessible

phosphorous after the incorporation of green manure legume weed species was obtained from T_1 followed by T_3 and T_2 and the least was observed from T_0 .

vi. The plot that was incorporated with *C. micans* i.e. T₁ [474.88 kg ha⁻¹ (0-10 cm) and 307.69 (10-20 cm)] significantly had the highest increased in the soil available potassium content, while the plot without any green manure i.e. T₀ [351.68 kg ha⁻¹ (0-10 cm) and 268.32 kg ha⁻¹ (10-20 cm)] had the lowest in both the cropping year. However, the soil potassium decreased from the first year to 2nd year.

5. Impact on soil biological properties

- i. The soil biomass carbon content exhibited significant variations (p < 0.05) across different treatments at 0-10 cm and also before and after the incorporation of the green manure in first and second cropping year. However, at 10-20 cm in the second year there was no significant variation (p < 0.05) among the treatments but showed variation before and after incorporation of the green manure. After the incorporation of the green manure legumes, control (T₀) [212.47 μ g g⁻¹ (0-10 cm) and 157.10 μ g g⁻¹ (10-20 cm)] recorded the minimum and T₁ [354.30 μ g g⁻¹ (0-10 cm) and 200.59 μ g g⁻¹ (10-20 cm)] recorded the highest MBC content. Furthermore, the levels of soil microbial biomass carbon (MBC) exhibited a declining pattern as the depth of the soil increased.
- ii. Soil MBP at surface and sub-surface layer was observed to vary significantly (p < 0.05) across the various treatments before as well as after the incorporation of the green manure in both the cropping year. After the incorporation of the green manure, T₁ [13.28 µg g⁻¹ (0-10 cm) and 8.42 µg g⁻¹ (10-20 cm)], recorded the highest and T₀ [7.22 µg g⁻¹ (0-10 cm) and 4.45 µg g⁻¹ (10-20 cm)], the lowest soil MBP. Furthermore, the soil MBP was higher in the first year but gradually decreases as the cropping year increased.
- The soil microbial population increased significantly after the incorporation of the green manure legumes at the surface and sub-surface layer except for all the treatments at 10-20 cm in the first year were at par, therefore no significant

difference was observed. T₁ [2.44 x 10^8 CFU's ml⁻¹ (0-10 cm) and 1.80 x 10^8 CFU's ml⁻¹ (10-20 cm) bacteria, 0.97 x 10^6 CFU's ml⁻¹ (0-10 cm) and 0.80 x 10^6 CFU's ml⁻¹ (10-20 cm)] had the highest bacterial and fungal population after the incorporation of the green manure.

6. Impact of leguminous weeds on the growth and yield of tomato

- i. In the present study, irrespective of the treatments, the plant height gradually increased until 90 days after planting (DAT), after which the growth rate stabilized until maturity. *C. micans* (T₁) produced the tallest plants at 30 DAT (47.51cm), 60 DAT (75.47) and 90 DAT (96.39 cm) while, T₀ had the lowest values at 40.15 cm (30 DAT), 63.85 cm (60 DAT), and 83.54 cm (90 DAT). Significant differences (p < 0.05) were found in the tomato plant across several treatments and cropping years at different growth stages. However, the plant height decreased significantly from the first year to the second year.
- ii. T_1 (5.18, 6.35, and 7.72 mm at 30, 60 and 90 DAT respectively) had the highest collar diameter followed by T_2 and *A. indica* (T₃) with value of 4.83, 5.88 and 7.07 mm; 4.35, 5.76, 6.78 mm respectively at 30, 60 and 90 DAT, while the control (T₀) had the lowest. Also, significant (p < 0.05), decrease in the collar diameter from the first year to the second year.
- iii. The incorporation of green manure considerably affected the number of branches of the tomato plants at all stages in both years of the study. T₁ (7.87, 12.20 and 17.52 at 30, 60 and 90 DAT) recorded the maximum number of branches.
- iv. The incorporation of *C. micans* (T₁) resulted in a statistically significant increase in the maximum number of clusters plant⁻¹ and number of fruits cluster⁻¹ with value of 6.56 and 5.15 respectively. However, significant decrease in the number of fruits per cluster and number of fruits per cluster was observed from the first year to the next year.

- v. The incorporation of green manure had a significant effect on the number of fruits plant⁻¹; a maximum of 16.13 fruits plant⁻¹ was recorded from T_1 followed by T_2 (12.20), T_3 (10.32), and the least by the control (8.68).
- vi. Fruit length and breadth varied significantly (p < 0.05) with treatments. Control (T₀) had the minimum fruit length (3.47 cm) and breadth (3.41 cm), T₁ with the highest fruit length (4.12 cm) and breadth (4.00 cm). However, fruit length and breadth decreased from the first to the second year.
- vii. The fruit yield was significantly influenced by the incorporation of the green manure crops in the first and second year with T_0 (54.10 q ha⁻¹) having the least and T_1 (105.44 q ha⁻¹) the highest fruit yield. However, the fruit yield was found to be significantly higher in the first year.
- viii. The cost of cultivation was maximum with the green manure treatments comparing to the control.
 - ix. The incorporation of *C. micans* (T₁) had the highest gross return, net return and the B:C ratio, with values of Rs 5,27,176.90 ha⁻¹, Rs 3,52,743.30 ha⁻¹ and 3.02 respectively.
 - x. Moisture content showed no significant correlation with all the plant parameters except for plant height, collar diameter and yield. However, in the case of soil chemical properties, all the parameters except for the soil pH exhibited a significant positive correlation with all the plant growth and yield parameters. This indicates that green manure crops improved soil chemical characteristics, which may have increased tomato growth and productivity.
 - xi. The plant growth and yield parameters showed a positive significant correlation with all the soil biological properties (Table 7.22) suggesting that the soil biological properties had a significant influence on the growth and productivity of the tomato plants in both the cropping year.

8.2. Conclusion

It can be concluded that the leguminous weed species exhibited a significant accumulation of biomass and nutrients of which Crotolaria micans showed excellent performance, highlighting their capacity for nutrient cycling within the agricultural system. Crotolaria micans and Calopogonium mucunoides litter decomposed faster than the litter of Aeschynomene indica. The initial litter composition of plant materials utilized as green manures plays a significant role in the processes of decomposition and release of nutrients. The legume weed species due to its rapid decomposition can be useful in regulating the soil nutrient pool. The species showed significant difference in the nodulation behaviour and the nitrogen fixation of which C. micans and A. indica showed higher nitrogen fixing ability, suggesting their potential for being used in nitrogen-deficient lands. The study also shows that the incorporation of green manure significantly influences the soil properties, growth and yield of the tomato crop and economic returns, thereby contributing to sustainable agricultural practices and improved crop production. Thus, it may be inferred that the inclusion of legume weeds as green manure enhances the soil's physical, chemical, and biological characteristics. These enhancements have a beneficial impact on the growth and development of crops, ultimately contributing to the long-term sustainability of agricultural production for farming communities. Overall, it can be concluded that incorporation of the locally available leguminous species as green manure can function as a substitute for inorganic fertilizers, providing soil organic matter and nutrients. Nontheless, the nutrient mobilization and immobilization of the three legume species in the soils can be readily used in modelling studies to evaluate the long-term effect of organic inputs on soil fertility in abandoned lands.

The use of green manure legumes in agricultural systems can be promoted through practical guidance and recommendations, supporting sustainable land management techniques. The outcome of the study can also help in developing effective weed management strategies by transforming legume weed biomass to valuable organic inputs in organic and natural farming system required for addressing the present-day issues pertaining to sustainable and climate resilient agriculture.



Photo plate 1. Land preparation



Photo plate 2. Sowing of seeds



Photo plate 3. Cultural operations



Photo plate 4. Biomass of C. micans



Photo plate 5. Biomass of A. indica



Photo plate 6. Biomass of C. mucunoides



Photo plate 7. Air drying of samples.



Photo plate 9. Placing of nylon mesh



Photo plate 8. Chopping of air-dried samples.



Photo plate 10. Samples prepared for nylon mesh bag



Photo plate 11. Monthly retrieval of samples



Photo plate 12. Washing of litter bag



Photo plate 13. Estimation of Lignin, Cellulose and hemicellulose



Photo plate 14. Digestion of plant samples for nutrient analysis



Photo plate 15. Estimation of Total ash content



Photo plate 16. Polypot experiment for the leguminous species, (a) *Calopogonium mucunoides*, (b) *Crotolaria micans* and (c) *Aeschynomene indica*



Photo plate 17. Root nodules of the leguminous species, (a) *Crotolaria micans*, (b) *Aeschynomene indica* and (c) *Calopogonium mucunoides*



Photo plate 18. Harvesting and incorporation of the green manure legume species.



Photo plate 19. Transplanting of seedlings



Photo plate 20. Irrigation



Photo plate 21. Monitoring plant growth





Photo plate 22. Harvesting of ripe tomato



Photo plate 23. Measuring the Fruit length and fruit breadth



Photo plate 24. Soil pH



Photo plate 26. Soil Available Nitrogen



Photo plate 25. Soil Organic Carbon



Photo plate 27. Soil Available Phosphorous



Photo plate 28. Soil Microbial Biomass

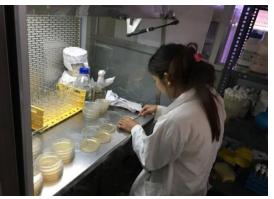


Photo plate 29. Soil Microbial population

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Name of Degree (Qualification)	Year	University/ Institution	Percentage	Subject
HSLC	2009	CBSE	69.8 %	English, Hindi, Science, Social Science, Mathematics
HSSLC	2011	CBSE	57 %	PCMB
Graduation	2016	Hemwati Nandan Bahuguna Garhwal University	77.77 %	Forestry
Post-Graduation	2018	Mizoram University	86.02 %	Forestry
NET	2018	ASRB-ICAR	-	Agroforestry

List of Publications

Research papers

- Jopir, J., Upadhyaya, K., Lalhmangaihzuali, B., and Rozar, K. P. (2023). Assessment of some leguminous weeds as potential green manure crops under Mizoram, North East India. *Legume Research*, 46(12), 1686-1691.
- 2. Jopir, J., and Upadhyaya, K. (2024). Nodulation Behavior of Three Leguminous Weeds of Mizoram with Potential Utility as Green Manure Crop. *International Journal of Ecology and Environmental Sciences*.
- Jopir, J., Upadhyaya, K., and Lalremsang, P. (2024). Decomposition and nutrient release pattern of three potential leguminous green manure crops of Mizoram. *Journal of Environmental Biology*, 45(1), 8-15.
- Lalthlamuanpuii, R., and Jopir, J. (2024). A Case Study on Farmers' Literacy in Agriculture Information in Lunglei District, Mizoram. *Indian Journal of Extension Education*, 60(2), 17-21.
- Rozar, K. P., Kumar, S., Sharma, R., Sharma, S. B., Nongrum, M. M., and Jopir, J. (2024). Comparison of phenolic content, flavonoid content and antioxidant activities of *Phyllanthus emblica* L. from North-East, India. *Indian Journal of Ecology*, 51(4), 732-737.

Book Chapters

 Jopir J and Upadhyay KK (2019). Ethno-medicinal Plants used by Adi tribes from Yingkiong circle of Upper Siang, Arunachal Pradesh. Medicinal Plants of India: Conservation and Sustainable Use: pp 2011-222, ISBN: 9788170196525. Today and Tomorrow's Printers and Publisher.

Papers presented in Workshop/Seminar

 Paper presentation on the topic "Biomass Production of three leguminous weeds and its Effect on the Yield of Tomato (*Lycopersicon esculentum* Mill)" in the 1st International Conference on "Global initiatives in Research, Innovation and Sustainable Development of Agriculture and Allied Sciences" organized by AEEFWS Society, Guru Kashi University, Talwandi Sabo on 6th-8th June 2022 at Guru Kashi University, Punjab.

- Paper presentation on the topic "Aboveground biomass production, nutrient content and accumulation of three leguminous weeds of Mizoram, with green manure potentiality" on the National Seminar on "Utilization and Conservation of Plant Resources for Sustainable Development organized by Biodiversity Research Centre, Department of Environmental Science, Mizoram University on 1st – 2nd June, 2023.
- 3. Paper presentation on the topic "Decomposition and nutrient release pattern of two potential leguminous green manure weeds of Mizoram" on the "International Conference on Biodiversity, Food Security, Sustainability & Climate Change" organized by College of Agriculture, Assam Agriculture University, Jorhat, Assam and Prof. H.S. Srivastava Foundation for Science, Lucknow, Uttar Pradesh on 26th – 28th April, 2023.

Seminars/Symposia/Course/ Workshop Attended

- Training program on "Climate Change for Natural Resource Management for the State of Mizoram" sponsored by DST during 11-15 March 2021 at Mizoram University.
- High-end workshop on Quality Control and Standardization of Herbal Raw Materials for Value Addition and Sustainable Development in North East organized by the Department of Horticulture, Aromatic and Medicinal Plants, Mizoram University from 16-21 September 2022.
- Participated in the "Nature Walk" event organized by 'Eco Club', Mizoram University and Mizoram Pollution Control Board on 14 October 2023
- Two-day training on Disease and Insect Pest Management in Forestry held by ICFRE- Bamboo and Rattan Centre, Aizawl on 23rd and 24th August 2023.
- 5-day workshop on "Sensitization of Students towards Science and Technology through demonstration and motivational talks at Kawkulh in Khawzawl district of Mizoram, Northeast India" on 30th May – 5th June 2023.

PARTICULARS OF THE CANDIDATE

NAME OF CANDIDATE	: Jyoti Jopir
DEGREE	: Ph.D
DEPARTMENT	: Forestry
TITLE OF THESIS	: Assessment of green manuring potential of
	Leguminous weeds of Mizoram

DATE OF ADMISSION : 29.08.2018

APPROVAL OF RESEARCH PROPOSAL

1.	DRC	: 22.03.2019
1.	DRC	: 22.03.2019

- 2. BOS : 18.04.2019
- 3. SCHOOL BOARD : 26.04.2019

MZU REGISTRATION NUMBER : 1600473

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& DATE	: MZU/Ph.D./1229 of 29.08.2018
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ABSTRACT

ASSESSMENT OF GREEN MANURING POTENTIAL OF LEGUMINOUS WEEDS OF MIZORAM

AN ABSTRACT SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF DOCTOR OF PHILOSOPHY

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DEPARTMENT OF FORESTRY SCHOOL OF EARTH SCIENCES AND NATURAL RESOURCES MANAGEMENT JULY, 2024

ASSESSMENT OF GREEN MANURING POTENTIAL OF LEGUMINOUS WEEDS OF MIZORAM

BY

JYOTI JOPIR Department of Forestry

Supervisor Prof. KALIDAS UPADHYAYA

Submitted

In partial fulfillment of the requirement of the Degree of Doctor of Philosophy in Forestry of Mizoram University, Aizawl.

ABSTRACT

Agriculture has always been the backbone of human civilization, providing the essential resources needed for survival and development. However, as the global population continues to grow, the demand increases, putting unprecedented pressure on the agricultural systems. The increased in demand comes at a cost: the degradation of soil health. Ensuring the soil fertility at a sufficient level is crucial for optimizing the crop yield. This goal can be accomplished by implementing suitable soil and crop management techniques. In this regard, the use of organic manures, including green manuring is a crucial tactic for preserving and enhancing soil fertility for long-term crop production. Green manuring has been widely carried out in India for decades, but growing food production pressure, the availability of competitive and profitable crops, and cheap and expensive chemical fertilizers have reduced its popularity. Due to soil access concerns, soil fertility loss, and public concern about abuse and energy conservation, organic growers and low-input farms utilizing conventional agronomic methods have rediscovered it. Green manure technology enhances soil fertility, structure, and water retention, prevents erosion, and promotes microbial growth by incorporating humus and organic matter

In recent years, governments have taken interest in organic farming, and most nations now have organic farming laws. Also, recent global climatic trends have forced policymakers and scientists worldwide to adopt appropriate land management practices, including agricultural systems, to reduce GHG emissions and increase carbon sequestration. The Indian government has designated the entire North Eastern region for organic farming. Therefore, maintaining organic farming in this location will require more organic amendment sources.

Mizoram is rich in biodiversity and has various weeds that could be used as green manure crops. Local plant resources like green manuring crops can improve soil fertility, in situ water conservation, weed control, and soil-borne pathogen management. Organic agriculture systems can use these green manure crops as soil additives. The information generated can help farmers in the region choose new locally available plant as green manure crops. The present investigation titled **"Assessment of green manuring potential of Leguminous weeds of Mizoram"** was conducted during 2020-2023 at Mizoram University, in Tanhril, Aizawl, Mizoram (23°44'22"N, 92° 39' 54" E and 950 meters above sea level) with the following objectives:

- 1. To estimate the biomass production and nutrient content of the selected species as green manure plants.
- 2. To assess the decomposition rate and nutrient release pattern of the selected species.
- 3. To assess the nitrogen-fixing ability of the selected species.
- 4. To determine the impact of the selected weeds on the soil properties and growth & yield of some agriculture crops.

The study aimed to evaluate the green manuring potential of three leguminous weed species abundantly found in Mizoram, India: Crotolaria micans Link., Aeschynomene indica L., and Calopogonium mucunoides Desv. The selected leguminous species were confirmed through the Botanical Survey of India in Shillong. The present study was carried out with the following objectives: to estimate the biomass production and nutrient content of the selected species as green manure plants, to estimate the decomposition and nutrient release pattern, to estimate the nitrogen fixation of the selected species and to estimate the effect of the selected species on the soil properties and the growth and yield of the tomato crops. Investigations on the biomass productivity (both aboveground and belowground) was estimated by harvest method. For this purpose, seeds of the selected species were collected from the wild and grown in experimental plots to monitor their growth and similarly harvested after 90 DAS, to estimate their biomass productivity and nutrient accumulation. A polypot experiment was conducted to determine the nitrogen and the amount of N fixed by each species was calculated by the difference method and expressed in terms of kg ha⁻¹yr⁻¹. The decomposition and nutrient release pattern of the weed species was determined by placing the air-dried samples in a litterbag and monitoring for 180 days. In order to determine the impact of the legume weeds on the soil properties and the growth and yield of tomato plants, the experiment comprised of 4 treaments viz: T1 (Crotolaria micans), T2 (Calopogonium

mucunoides), T_3 (*Aeschynomene indica* and T_0 (Control). Soil samples were collected before sowing the legume seeds and 60 days after incorporating the green manure into the soil. Analysis on various soil physical, chemical and biological properties were carried out. 60 days after incorporating the legume weed species, tomato seedlings (*Arkha Rakhshak* variety) were transplanted to the plots and thereafter the growth parameters at successive stages were observed and recorded. The tomato yield and various yield parameters was calculated using the standard methods.

Results

Biomass Production and Nutrient Accumulation

After 90 days of sowing the legume species, showed significant variation in the shoot length, but did not show any variation in the cropping year. The root length of all the three species were at par during both the cropping year. The collar diameter varied greatly by species and cropping year. Although there was no significant difference in branch count between cropping years, there was a significant variation (p < 0.05) among species. *C. micans* exhibited the greatest growth with the highest shoot length (91.29 cm), root length (29.04 cm), collar diameter (7.1 mm), and number of branches per plant (27.00).

C. micans also showed the highest fresh and dry biomass production in both shoot and root compared to the other species. The fresh biomass of the shoot and root was $15.16 \text{ t} \text{ ha}^{-1}$ and $1.65 \text{ t} \text{ ha}^{-1}$ respectively, and the dry biomass was $2.90 \text{ t} \text{ ha}^{-1}$ for the shoot and $0.54 \text{ t} \text{ ha}^{-1}$ for the root. The least fresh and dry biomass of shoot and root was observed by *A. indica* with value of $2.1 \text{ t} \text{ ha}^{-1}$ and $0.58 \text{ t} \text{ ha}^{-1}$ (fresh and dry shoot biomass) and $0.36 \text{ t} \text{ ha}^{-1}$ and $0.16 \text{ t} \text{ ha}^{-1}$ (dry shoot and root biomass).

Significant variations on the nutrient accumulation were observed among the three species, *C. micans* accumulated the highest amounts of N, P, K, Mg, Ca, Fe, Zn and Na in the shoot and root biomass, while Cu in the shoot biomass. However, *Calopogonium mucunoides* recorded the highest Cu accumulation in the root

biomass. It was observed that for all the species, the micronutrient content (Na, Fe, Zn and Cu) in the root biomass were higher comparing to the aboveground biomass.

Decomposition and Nutrient Release Patterns

The initial chemical composition exhibited significant variation among the legume species. *C. micans* recorded the highest P (0.27 %), K (1.55 %) and C (38.12 %) content. *A. indica* observed highest Lignin (16.98 %), N (3.11 %), Total phenol (1. 80 %), crude fiber (21.74 %) and L/N ratio (5.45). *C. mucunoides* recorded the highest cellulose (32.19 %), C/N ratio (17.22) and C/P ratio (200.23).

The decomposition of the species was monitored over 180 days. *C. micans* decomposed most rapidly with decay rate constants of 8.21 *k* year⁻¹ while *A. indica* (5.5 *k* year⁻¹) had the slowest decomposition rate. The maximum half-life (t_{50}) and t_{99} was recorded by *A. indica* with 46 and 331.89 days respectively. After 180 days, the decomposition time, there were 0.18 g (1.8 %), 0.25 g (2.85 %), and 0.66 g (6.64 %) of undecomposed dry material left from the original mass of *C. micans, C. mucunoides*, and *A. indica*, respectively.

C. micans and *A. indica* released nutrients in the order of K > P > N, while *C. mucunoides* released them in the order of K > N > P The N, P and K mineralization per year was higher in *C. micans* with value of k_N (2.31), k_P (2.45) and k_K (3.86) respectively.

The initial chemical composition of the plant materials plays significant role in the decomposition process of the species. Lignin, total phenol, Crude fiber, L/N and N/P ratio negatively correlated with the decay rates of which the phenolic content (R^2 = 0.976) of the plant litter strongly influence the decomposition rate.

Nodulation and Nitrogen Fixation

The initiation of nodulation began on the primary roots, however in the later stages of seedling growth, lateral roots developed a large number of nodules. The formation of nodules exhibited a substantial rise as the age of the seedling rose in all species. *A. indica* recorded the highest number of nodules from its secondary roots while in the case of *C. micans* and *C. mucunoides* highest number of nodules was observed in the tertiary roots.

In all months $(1^{st}, 2^{nd}, 3^{rd}, and 6^{th})$, *A. indica* primary root nodules had higher fresh biomass than the other species. However, *C. micans* had more secondary and tertiary root nodules in the 1^{st} and 2^{nd} months and 1^{st} , 2^{nd} , 3^{rd} and 6^{th} month respectively. *C. micans* had higher fresh biomass although the number of nodules were lesser comparing to *A. indica*, indicating more space which results in higher nitrogen fixing ability. The dry biomass of the nodules was directly proportional to the fresh biomass of the nodules.

The nitrogen content exhibited variation across the shoots, roots, and nodules of different species, with *C. micans* demonstrating the maximum nitrogen level in its shoot (2.07 %) and root nodules (4.78 %). However, the maximum nitrogen level in the root was recorded in *C. mucunoides* (1.96 %). *C. micans* was the most efficient nitrogen fixer, with a rate of 243.36 kg ha⁻¹ year⁻¹, followed by *A. indica* (152.07 kg ha⁻¹ year⁻¹) and *C. mucunoides* (22.55 kg ha⁻¹ year⁻¹).

Effect of the legume weed species as green manure on the soil properties

The soil moisture contents were at par among the treatments hence displayed a non-significant variation, however, the moisture content vary after the incorporation of the green manure when compared to the initial stages. T₃ recorded the highest with value of 23.35 % and 22.21 % at 0-10 and 10-20 cm respectively and the least was recorded by T₁ had the lowest at the surface (20.44 %) and sub-surface layer (18.88 %).

Significant change in soil pH, with a decreasing trend observed following the incorporation of green manure compared to treatments without green manure (T_0). The incorporation of green manure legumes led to substantial increases in soil organic carbon content. T₁ exhibited the highest organic carbon with value of 2.02 % (0-10 cm) and 1.39 % (10- 20 cm) and the least was observed from the plot without

any green manure legumes (T0) with value 1.30 % (0-10 cm) and 0.79 % (10-20 cm).

The soil available nitrogen, phosphorous and potassium following the incorporation of the green manure legumes significantly increased. T₁ recorded having the highest N levels after incorporation of the legume respectively, with values of 512.35 kg ha⁻¹ and 369.27 kg ha⁻¹ at 0-10 cm and 10-20 cm. The incorporation of *C. micans* recorded the highest accessible phosphorous followed by T₃ and T₂ and the least was observed from T₀. The plot that was incorporated with *C. micans* i.e. T₁ [474.88 kg ha⁻¹ (0-10 cm) and 307.69 (10-20 cm)] significantly had the highest increased in the soil available K content, while the plot without any green manure i.e. T₀ [351.68 kg ha⁻¹ (0-10 cm) and 268.32 kg ha⁻¹ (10-20 cm)] had the lowest in both the cropping year.

 T_0 had the lowest MBC content (212.47 µg g⁻¹ (0-10 cm) and 157.10 µg g⁻¹ (10-20 cm)) while T_1 had the highest (354.30 µg g⁻¹ and 200.59 µg g⁻¹). As soil depth grew, soil microbial biomass carbon (MBC) decreased. Green manure legumes boosted soil microbial population at the surface and subsurface layer, but all treatments at 10-20 cm in the first year were at par. After adding green manure, T_1 had the greatest bacterial and fungal population (2.44 x 10⁸ CFU's ml⁻¹ (0-10 cm) and 1.80 x 10⁸ CFU's ml⁻¹ (10-20 cm) and 0.97 x 10⁶ CFU's ml⁻¹ (0-10 cm) and 0.80 x 10⁶ CFU's ml⁻¹ (10-20 cm).

Effect of the legume weed species as green manure on the growth and yield of tomato

The plant height gradually increased until 90 days after planting (DAP), after which the growth rate stabilized until maturity. T_1 produced the tallest plants at 30 DAT (47.51 cm), 60 DAT (75.47), and 90 DAT (96.39 cm), while T_0 exhibited the lowest values at 40.15 cm, 63.85 cm, and 83.54 cm.

 T_1 (5.18, 6.35, and 7.72 mm at 30, 60 and 90 DAT respectively) followed by T_2 and *A. indica* (T_3) with value of 4.83, 5.88 and 7.07 mm; 4.35, 5.76, 6.78 mm

respectively at 30, 60 and 90 DAT, while the control (T_0) had the lowest. The incorporation of green manuring considerably affected the number of branches of the tomato plants at all stages in both years of the study. T_1 (7.87, 12.20 and 17.52 at 30, 60 and 90 DAT) recorded the highest.

The incorporation of *C. micans* (T₁) resulted in a statistically significant increase in the maximum number of clusters plant⁻¹ and number of fruits cluster⁻¹ with value of 6.56 and 5.10 respectively. However, significant decrease in the number of fruits per cluster and number of fruits per cluster was observed from the first year to the next year. The incorporation of green manure had a significant effect on the number of fruits plant⁻¹; a maximum of 16.13 fruits plant⁻¹ was recorded from T₁ followed by T₂ (12.20), T₃ (10.32), and the least by the control (8.68). Fruit length and breadth varied significantly (p < 0.05) with treatments. Control (T₀) had the minimum fruit length (3.47 cm) and breadth (3.41 cm), T₁ with the highest fruit length (4.12 cm) and breadth (4.00 cm). However, fruit length and breadth decreased from the first to the second year.

The fruit yield was significantly influenced by the incorporation of the green manure crops in the first and second year with T_0 (54.10 q ha⁻¹) having the least and T_1 (105.44 q ha⁻¹) the highest fruit yield. However, the fruit yield was found to be significantly higher in the first year. The incorporation of *C. micans* (T₁) had the highest gross return, net return and the B:C ratio, with values of Rs 5,27,176.90 ha⁻¹, Rs 3,52,743.30 ha⁻¹ and 3.02 respectively.

Conclusion

The present study concluded that the leguminous weed species exhibited a significant accumulation of biomass and nutrients of which *Crotolaria micans* showed excellent performance, highlighting their capacity for nutrient cycling within the agricultural system. *Crotolaria micans* and *Calopogonium mucunoides* litter decomposed faster than the litter of *Aeschynomene indica*. The initial litter composition of plant materials utilized as green manures plays a significant role in the processes of decomposition and release of nutrients. The legume weed species

due to its rapid decomposition can be useful in regulating the soil nutrient pool. The species showed significant difference in the nodulation behaviour and the nitrogen fixation of which *C. micans* and *A. indica* showed higher nitrogen fixing ability, suggesting their potential for being used in nitrogen-deficient lands. The study also shows that the incorporation of green manure significantly influences the soil properties, growth and yield of the tomato crop and economic returns, thereby contributing to sustainable agricultural practices and improved crop production. Overall, it can be concluded that incorporation of the locally available leguminous species as green manure can function as a substitute for inorganic fertilizers, providing soil organic matter and nutrients. Nonetheless, the nutrient mobilization and immobilization of the three legume species in the soils can be readily used in modelling studies to evaluate the long-term effect of organic inputs on soil fertility in abandoned lands.

The use of green manure legumes in agricultural systems can be promoted through practical guidance and recommendations, supporting sustainable land management techniques. The outcome of the study can also help in developing effective weed management strategies by transforming legume weed biomass to valuable organic inputs in organic and natural farming system required for addressing the present-day issues pertaining to sustainable and climate resilient agriculture.