PHENOTYPIC PLASTICITY IN SOYBEANS (*Glycine max (L.*) Merrill) ASSOCIATED WITH PLANT DENSITY AND PHENOLOGICAL STAGE AT THINNING

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A Dissertation Submitted to the School of Agricultural Sciences in Partial Fulfilment of the Requirements for the Award of the Degree of Master of Science in Agronomy-Plant Science

SCHOOL OF AGRICULTURAL SCIENCES, DEPARTMENT OF PLANT SCIENCES

THE UNIVERSITY OF ZAMBIA LUSAKA

July 2020

DECLARATION

I, Noah Manda, hereby declare that the work presented in this dissertation was my own and has never been submitted for a degree at this or any other university.

Signature:

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APPROVAL

This dissertation of Noah Manda has been approved as partial fulfilment of the requirements for the award of Master of Science in Agronomy by the University of Zambia.

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DEDICATION

To my wife- Maiwase and our lovely children- Chisomo and Nkwela Manda, for enduring my long absence from home during my study and for their unwavering support. To my late father-Mr. Gilbert Manda for recognising my potential for academics and encouraging me to pursue further.

ACKNOWLEDGMENTS

I am sincerely grateful to my academic supervisor, Dr Mebelo Mataa. I further thank my supervisors at work, Dr Martin Chiona (ZARI) and Dr Pheneas Ntawuruhunga (IITA) for their mentorship and all-season support in my academic and profession endeavours.

Special thanks go to Trevor Chola Mulenga and Debra Kapemba for their assistance and commitment during field data collection at different times of the experiment. Mr. Justin Njobvu and Mr. Kabaja Banda whose technical and moral contribution to my work cannot go unrecognised.

Further gratitude goes to my classmates, Inonge 'Noni' Siziya and Shadreck Chola Mubanga, for their encouragements, reminders of important deadlines and for being a dependable academic support system during my study period.

I wish to thank ZARI management for granting me the fellowship through the APPSA project and particularly Mr. Ivor Mukuka.

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ABSTRACT

Soybean (Glycine max L.) is reported to demonstrate plasticity responses when exposed to either supra or sub optimal plant density stress. However, it is not known whether determinate and indeterminate varieties respond the same and at what phenological stage a soybean plant is able to exhibit adaptive plasticity or elasticity response after thinning. The relationship between plant density stress and plant performance was studied in soybean varieties. The objective of the study was to determine phenotypic plasticity and its consequence on grain yield associated to plant population density-stress recovery capacity on different soybean varieties. The trials were conducted at Mansa Research Station of the Zambia Agricultural Research Institute (Agroecological Region III). A split-split plot design was used replicated four times, with variety occupying the main plot, plant density in sub plot and thinning time in sub-subplot. The Zambian soybean varieties used were two determinate types (Lukanga and SC Semeki) and one indeterminate type (Mwembeshi). Planting density stress was imposed by planting at supra optimal densities (700 K, 600 K, and 500 K plants ha⁻¹, where K represented 1000) and stress was removed by thinning to the recommended density (400 K plants ha⁻¹) at different crop phenological stages $(V_0, V_4, R_1 and R_8)$. V₀ was time of planting, where seeds were sown at the recommended density level of 400K plants ha⁻¹. V₄ was the vegetative stage where the fourth trifoliate leaf had completely unrolled and not touching, R1 was the onset of the reproductive growth phasebeginning bloom, where the plant had developed at least one open flower at any node and R_8 was a stage when the plant had reached full maturing with at least 95 % of the pods had attained their full maturity colour. In practice V₀ and R₈ treatments were maintained with no thinning. The results showed that variety had significant effects on plant height, biomass weight, number of grains per pod, number of pods per plant, grain yield and harvest index (HI). Plant density only affected biomass, grain weight and HI. Thinning time influenced root to shoot ratio, number of grains per pod, grain yield and HI. All the interaction levels exerted significant effects on most of the observed parameters. Grain yield and biomass weight were highly influenced by variety. Lukanga had the highest grain yield (2.43 tons.ha⁻¹), followed by Mwembeshi (1.95 tons.ha⁻¹) the least was recorded in SC Semeki (1.17 tons.ha⁻¹). The significantly high yield observed in Lukanga was associated to the significant number of grains per pod while for Mwembeshi was owing its significant yield value to its high number of pods per plant. There was an observed inverse relationship between biomass weight and plant density stress duration, particularly for SC Semeki.

Chapter 1

1. INTRODUCTION

Soybean (*Glycine max* (L.) Merrill) is an oil seed crop produced worldwide in varying environments (FAO, 2016). In 2017, soybeans accounted for almost 60% of the world oilseed crops production and contributed 70 % to the world protein- meal consumption (Soystats, 2018). Despite having its origin in Asia (Hymowitz, T. and Shurtleff, 2005), the Americas (i.e. USA, Brazil and Argentina) have dominated the world soybeans production in recent years, accounting for over 80 % of the total production (Zhang *et al.*, 2017; FAO 2016. Soystats 2018).

Soybean is reported to be efficient at accumulating oils and proteins in its seed grain, hence producing more protein and oil per unit land compared other cultivated crop plants (Franklin and Martin, 1988; Hartman *et al.*, 2011; Mingjue, 2014). Soybeans' ability to have high oil and protein content places the crop among the most important cultivated crops in the world and its importance has been increasing, hence in 2017, it ranked third, (only after wheat and maize respectively) as the most traded crop commodity in the world (Soystats, 2018). The crop is versatile regarding utilisation, ranging from human and animal consumption to primary feed stock in many industrial and pharmaceutical products (Manavalan *et al.*, 2009; Hartman *et al.*, 2011; Mingjue, 2014).

According to McWilliams *et al.*, (2004) soybeans productivity is highly influenced by the plant's ability to tolerate environmental stress and to efficiently partition photoassimilates into economically important components (genetic potential). Responses to abiotic stress factors are manifested in the morpho-physiological character embodiments (Tilman *et al.*, 2014; Rahmawati *et al.*, 2019). Plant competition, whether intraspecific or interspecific results in resource limitation; to which, plants respond by modifying their morphological traits and physiological functions (Mataa and Sichilima, 2019). This is so, to maximise the plant's access to limiting resources by reallocating its produced biomass (here referred to as photoassimilates) to plant parts which are responsible for acquisition of the resource in limitation (Aroca, 2013; Tilman, Isbell and Cowles, 2014; Rondanini *et al.*, 2017).

Field crops are assumed to manifest a general plant growth characteristic of having phenotypic adaptive plasticity, which enables plants to counter abiotic stress factors. Soybean response capacity to counter abiotic stresses imposed by high population density may vary among genotypes. An understanding of phenological development provides the basis for enhanced crop management. Specifically, a good understanding of the growth habits of soybean plants and its function can help growers and plant development experts refine their management practices to achieve better yield and maximise profits (Counce *et al.*, 2000). During the process of plant development- the sum total of plant growth and differentiation- there are many physiological processes that occur and are controlled or influenced by plant hormones available only at specific growth stages (Maggio *et al.*, 2018). Accurate identification of the different phasic growth stages is essential for growth environment manipulation to influence plant development, accurately apply pesticides and determine potential yield loss from environmental stresses (Wright and Lenssen, 2013; Agalave, 2017).

Increasing soybeans plant population per unit area generally results in higher grain yields (Rahman and Hossain, 2011; Ibrahim, 2012; Mondal *et al.*, 2014). However, the phenological stage at which a soybean plant is able to exhibit adaptive plasticity or elasticity after thinning when exposed to high plant population density is not yet known.

1.1. Objectives

The main objective of the study was to determine the extent to which plant density and thinning time influences crop development and yield in soybeans, thus the study evaluated vegetative plasticity in soybeans. Specifically, the study sought to determine;

- i. The stage at which plant density stress is removed affects plant developmental responses
- ii. Whether these plasticity responses are genotype dependent.

1.2. Research Hypothesis

- i. Plant density stress and stage at which this stress is removed has an effect on vegetative plasticity responses.
- ii. These plasticity responses are variety dependant and soybeans may also show yield and morphological elasticity by being able to recover if stress is removed at a suitable time.

Findings of the study may contribute to understanding how crops respond to stress and assist in optimising cultural practices for new crop varieties, thereby contribute to better resource use and improve yield efficiency.

Chapter 2:

2. LITERATURE REVIEW

2.1. Taxonomy and Origin of Soybean Plant

Soybean (*Glycine max* (L.) Merrill) is an oil seed crop that belongs to the *Leguminosae* family, with the subfamily *Papilionoideae* and in the genus *Glycine*, L (Mingjue, 2014). Soybean is thought to have originated from China some 4000 to 5000 years ago (Barbalho and Farinazzi-Machado, 2011; Mingjue, 2014). However, Hymowitz and Shurtleff, (2005) suggest that production in China only goes back to the 11th century B.C.E or slightly earlier, therefore the crop is only about 3100 years old under domestication. Soybean was introduced to North America and Europe around 1765 by Samuel Bowen (Hymowitz, T. and Shurtleff, 2005). Initially, soybeans were mainly used as a forage crop in North America. By the early 1950s, USA surpassed China in production and this was mainly due to the expanded utilization portfolio to include human consumption, livestock feed, the pharmaceuticals and other industrial processes (Sonderegger, 2013). Shurtleff and Aoyagi, (2019) suggest that soybean was brought to Africa through North Africa in 1857. The same authors also dated earliest soybean production in Zambia to be 1910 and India to be the source of the seed.

2.2. Soybean Plant Development and Phenology

Growth, development and yields of a plant are a result of a variety's genetic potential interacting with the environment (Wright and Lenssen, 2013) and management practices. The Plant Ontology Consortium (POC) recently developed the common ontology that describes the anatomy, morphology, and growth stages of all angiosperms. This computer assisted plant development staging technology was developed to counter the challenge of many volumes of literature available that detail growth stages for individual plant species or closely related groups of species (Pujar et al., 2006). However, for easy application in field crops research, Fehr and Caviness (1971) staging system of soybean plant development was found to be most ideal, because of its use of the plant's morphological markers. The system uses the cumulative leaf number (CLN) to describe growth stages in the vegetative phase and discrete morphological criteria for staging growth in the reproductive phase (Counce et al., 2000). The CLN system results into further division of vegetative morphogenesis into specific V(n) stages where (n) represents a numeric value for the last node. The specific (n) stage is determined by counting the number of nodes on the main stem starting with the unifoliate node which has a completely unrolled leaf. There is an exception for the first two stages though where (n) is represented by letters (i.e. VE and VC). It has been observed that

maximum V(n) is varietal specific and is also influenced by the environment (Wright and Lenssen, 2013).

A leaf is considered completely unrolled when the leaf at the node immediately above it has unrolled sufficiently so the two edges of each leaflet are no longer touching. At the terminal node on the main stem, the leaf is considered completely unrolled when the leaflets are flat and similar in appearance to older leaves on the plant (Fehr and Caviness, 1971; Counce, Keisling and Mitchell, 2000; Pujar *et al.*, 2006; Wright and Lenssen, 2013).

The discrete morphological features in the reproductive stages start with the beginning bloom (R1) where the plants have at least one open flower at any node and ends at R8 where soybean is considered to have reached physiological maturity or cessation of dry matter accumulation (Wright and Lenssen, 2013).

2.3. Plant stress

Environmental constraints occur in form of abiotic or biotic stresses on the plant (Chang and Turner, 2019). Environmental constraints imposed on plants due to abiotic factors are as a result of the plant's need for different essential and trace nutrients from the soil/growth media, water and light (Craine and Dybzinski, 2013; Sonderegger, 2013). Being sessile organisms, plants must make do with environmental stresses such as soil salinity, extreme temperatures, flooding and drought as well as ultra violet radiation for adaptation and survival (De Bruin and Pedersen, 2008; Wani *et al.*, 2016; Zhu, 2016; Takahashi and Shinozaki, 2019). The biotic factors acting as environmental constraints on the plant's growth include the plant's susceptibility to pests and diseases, competition from neighbouring crops and weed plant species (De Bruin and Pedersen, 2008). Plants respond to adverse conditions from the environment by invoking stress signals that induce appropriate physiological and molecular responses to maintain their development and productivity (Ramegowda and Senthil-kumar, 2015; Zhu, 2016; Mataa *et al.*, 2019).

2.3.1. Biotic stresses

Soybean is susceptible to pests and diseases that cause stress in plants and hence negatively impact seed production and productivity. The most common diseases and pests of economic importance for soybeans growers in Zambia are bacterial pustule (*Xanthomonas axanapodis pv. Glycines*), soybean rust (*Phakopsora pachyr- hizi*), soybean mosaic virus, frogeye leaf spot (*Cercospora sojina* Hara), common soybean fly (*Melanagromyza sojae*), Soybean aphids (*Aphis glycines*), pod sucking bugs (*Nezara viridula*) and pod borers (*Helicoverpa*

armigera). (Mwase and Kapooria, 2001; Hartman *et al.*, 2011; Hartman and Murithi, 2014; Alfy, 2017; Gaur and Mogalapu, 2018; Chigeza *et al.*, 2019).

2.3.2. Stress signalling

In order to acquire stress tolerance and resistance, plants have developed unique stress signalling mechanisms that involve rapid short-term responses to adverse changes in the environment such as high temperatures and excessively low soil moisture content among others, to prevent severe/permanent damage and long-term adaptation responses to acquire tolerance (Kollist *et al.*, 2019;Ramegowda and Senthil-Kumar, 2015; Takahashi and Shinozaki, 2019). A number of genes that regulate specific signal transduction have been identified (Takahashi and Shinozaki, 2019) and the stress signalling pathways elucidated.

Among the best-known signalling pathways are the reactive oxygen species (ROS), once considered by-products of metabolic pathways at sub-cellular level (Mhamdi and Van Breusegem, 2018), but now understood and appreciated for being responsible for critical signalling in plants for stress response and plant development. By interacting with other phytohormones (such as salicylic acid-SA, jasmonic acid-JA, ethylene-ET, abscisic acid-ABA and gibberellic acid-GA), ROS effect physiological and molecular responses of plants to both biotic and abiotic stresses by varying their concentration levels through the redox perturbations, thus influencing the formation of responsive molecules (Mabuchi *et al.*, 2018; Mhamdi and Van Breusegem, 2018; Takahashi and Shinozaki, 2019).

Changes in the hydrostatic pressure is one form of stress signalling that triggers a chain of complex reactions at subcellular level to mediate the low water potential in the root zone of the growth media (Takahashi and Shinozaki, 2019).

2.3.3. Plant performance due to competition as a stress factor

It is an established theory that minimizing abiotic and biotic stresses will maximize soybean yield (Wright and Lenssen, 2013). An understanding of how plants grow and responds to intraspecific competition provides an insight into how stress can be positively manipulated to enhance plant performance and maximise yields.

In an attempt to describe mechanistic approach to resource competition among plants, Pacala and Tilmant (1994) stated that relative growth rate and weight per unit plant demonstrates a negative relationship, whereby an increase in plant density results in decreased individual total plant biomass. These findings were authenticated by Li *et al.*, (2019) who observed that as competition between plants increases-i.e. increased plant population density, there are decreases in individual plant dry matter accumulation. The plant organs most negatively

affected by increases in population density are the yield components as observed by Ibrahim, (2012) who associated the decrease to biomass allocation plasticity. However, when observed on a unit area basis, total dry biomass increased with an increase in plant population (Rahman and Hossain, 2011; Ibrahim, 2012) due to the individual plant influences as well as the increased Photosynthetic Active Radiation (PAR) interception owing to the large canopy (Park and Runkle, 2018; Li et al., 2019). The above observations were valid in both resource poor and rich nutrient environments. Pacala and Tilman's (1994) hypothesis was later shared by Arenas and Fernández, (2000) who observed that there was a negative biomass-density relationship, in their efforts to understand size structure and dynamics in plant populations. Reduced competition for resources from the environment can be attributed to the observed association of the reduction in plant density which theoretically implies increased individual plant biological yield (Ibrahim, 2012; Li et al., 2019). High plant density introduces abiotic stress due to onset of competition for resources from the environment i.e. light, nutrients and moisture among others (Rondanini et al., 2017). Higher plants elicit their light quality and quantity perception mechanisms using three main photoreceptor families; R:FR ratio phytochrome, the Blue/UV-A light absorbing cryptochromes and the phytotropins to respond to light stress in their environment (Park and Runkle, 2018).

When competition is asymmetric, it results in weaker plants to be out competed and the vigorous plants are induced into positive morpho-physiological responses (Mellendorf, 2011; Rondanini et al., 2017). As defined by Park, Benjamin and Watkinson, (2003) asymmetrical competition results when smaller plants get a disproportionately lower share of the available resources to their detriment, because larger plants have had excessively higher share of the resource in question to the point of limiting growth and productivity of the surrounding plants. The induced positive morpho-physiological response raises the plants resource acquisition competitiveness per unit photoassimilate produced, thereby enhancing the plants development capacity despite the resource limitation (Craine and Dybzinski, 2013). This concept postulates that the photoassimilate partitioning plasticity is functional at some critical phenological phase of a plant (Mataa and Sichilima, 2019). Therefore, early removal (in the developmental cycle) of the induced abiotic stress such as the one that results from high plant density before the full physical manifestation of the morpho-physiological responses, theoretically builds the plants yield capacity (Rondanini et al., 2017). However, workers on plant population studies (Sultan, 2003, Zamir et al., 1999, Kamil, 1983, Mondal, 2014) hold a consensus that grain yield does not holdfast the above established principle of negative relationship between biomass yield and plant density.

The contrast that, reduced competition increases individual plant productivity (Rondanini *et al.*, 2017; Mellendorf, 2011; Mataa and Sichilima, 2019) and findings that grain yield increases with an increase in plant density per unit area (Sultan, 2003; Ahmad and Latif, 2011; Mondal *et al.*, 2014) and hence an increase in competition, sets the basis for evaluating the morpho-physiological response of soybean plants to population density and the associated photoassimilate partitioning patterns into yield components.

2.4. Phenotypic plasticity

Plants being sessile, organisms maintain stability under varying environmental conditions by adjusting their morphophysiological characteristics. Bradshaw, (1965) described such responses as plasticity. This is achieved when a plant trades off its available resources (photoassimilates) by investing more of the photoassimilates into the enhancement of the plant structure which will help maximize the plant's access to the resource in limitation (Murren *et al.*, 2015). The preceding is one form of plant plasticity (i.e. morphological) as discussed by (Grime and Mackey, 2002), who postulated that plant plasticity involves change of the meristematic tissue parts of the plant with different characteristics-a resource costly option for changes in the environment (Bradshaw, 1965). For instance, an individual plant under high population growth conditions with a closed canopy will undergo physiological changes which signal apical growth enhancement, thereby influencing the plants phenotypic expression (i.e. modified morphology) to allow it to positively compete in low photon flux density, (Bradshaw, 1965; Park, Benjamin and Watkinson, 2003; Park and Runkle, 2016; Maggio *et al.*, 2018).

The second form is one which does not involve visible changes to the eye but occurs at subcellular level in already differentiated tissues where cues prompt rapid reversible responses of the plant-a resource cheap option to changes in the environment (Bradshaw, 1965; Grime and Mackey, 2002). Shade avoidance syndrome has been cited in literature as the manifestation of physiological plasticity in response to cues for presence of neighbouring plants, where the plant responds to the reduced quality of white light (i.e. R:FR ratio-a parameter that describes the natural light environment), a reversible process that occurs rapidly (Franklin and Whitelam, 2005; Park and Runkle, 2016; Maggio *et al.*, 2018). Sultan (2003) described this capacity for specific functionally appropriate response to environmental conditions as adaptive plasticity. Soybeans, like other field crops are exposed to various levels of intraspecific competition (interference), the negative influence of neighbouring plants. However, there is little evidence which demonstrates if there are differential responses among genotypes.

Intraspecific competition reduces the rate of development of some plant parts. For example, leaf size changes in response to light quality or quantity or when plants are grown on sub optimal soils (Agalave, 2017; Maggio *et al.*, 2018; Rahmawati *et al.*, 2019). In response to reduced light, Sultan, (2003) observed that genetically identical plants increased photoassimilate partitioning to leaves, thus maximising leaf surface area. This in turn increased the leaf's capacity to capture more light under low photon flux density condition. The stress factor of low light induced a positive morpho-physiological response in the plant to raise its photosynthetic effectiveness per unit photoassimilate produced, hence enhancing the plants development capacity despite the resource limitation. The above notion implies that the photoassimilate partitioning plasticity is functional in all phenological phases of a plant (Pacala and Tilmant, 1994; Sultan, 2003; Rondanini *et al.*, 2017).

Plasticity responses to adverse environmental cues that are restricted to vegetative phenotypic traits such as described by Rondanini *et al.*, (2017) [i.e. plant height, petiole length, leaf size, rosette diameter] is referred to as vegetative plasticity (Rondanini *et al.*, 2017). Increasing root biomass for instance in the early phenotypic stages of the plant in order to increase the relative root surface area and therefore their absorptive capacity of the soil resources constitute vegetative plasticity and root biomass being the trait thereof (Bradshaw, 1965; Sultan, 2003). On the other hand, reproductive plasticity was describe by Sultan, (2003) in light of any developmental adjustments to phenotypic traits that directly affect reproductive success of an individual plant in response to various environmental stress signals. This response involves disproportionate photoassimilates partitioning to reproductive structures, total reproductive output, adjustment in reproductive timing and the size and quality of their offspring. Rondanini *et al.*, (2017) associated branching dynamics observed in rapeseed to reproductive plasticity, where branching was the trait of attribution. Agudamu *et al.*, (2016) noted that soybean varieties with determinate growth type exhibited more reproductive plasticity than their indeterminate counterparts.

2.5. Plant competition and yield development

Plant competition is generally categorised as either intraspecific; when common resources are inadequate for all the plants of a similar specie in a given land unit, such as between the established crop plants at a given crop canopy (Mellendorf, 2011; Craine and Dybzinski, 2013). On the other hand, plant competition or interspecific- between multiple species and is usually demonstrated as competition between a cultivated crop and weed/volunteer plant species (Mellendorf, 2011). When faced with either kind of competition, plants may respond by modifying their morphological characteristics and physiological functions. This is so as

to maximise the plant's access to limiting resources by reallocating its produced biomassphotoassimilates-to plant parts/organs which are responsible for the acquisition of the resource in limitation (Mellendorf, 2011; Al-Suhaibani, El-Hendawy and Schmidhalter, 2013; Craine and Dybzinski, 2013; Tilman, 1988; Schulze and Mooney, 1993).

2.5.1. Water and soil stresses

Soybean manifests an allorhizic root system, typical of the dicot plants, the root system has a tap root with branching out lateral roots which provide the plant with anchorage and adsorption of mineral nutrients and moisture from the soil (Atkinson *et al.*, 2014). Poorly developed root system renders the plant ineffective at competing for soil-based resources (i.e. nutrients, soil water). A plant which has the capacity to respond to the dehydration stress signalling due to water deficit in the developing root system so that it can acquire more of the said limiting resource eventually out performs its competitors (Takahashi and Shinozaki, 2019). The plant shifts its photoassimilates by partitioning them in favour of the growth points in the root system (McWilliams *et.al.*, 2004). For the below ground growth points, this is to allow the plant to reach more water and nutrients by covering a wider root zone and deep lying soil moisture (Souza *et al.*, 2013). Ku *et al.*, (2013) reported such morphological and growth adjustments by measuring the root system parameters such as root density, root length, root-shoot biomass ratio and root distribution in both low and high moisture environments. It was further observed that there were differential growth adjustments among varieties of different soybean plants (Ku *et al.*, 2013).

In their studies of the developmental windows on soybeans phenology, Kron *et al.* (2008) observed that subjecting soybean plants to water stress in the early growth stages (up to V_4) of the plants, the crop showed increased tolerance to water shortages in later growth stages. A similar conclusion was drawn by Manavalan *et al.*, (2009); who stated that if a plant develops a large root system during its early vegetative growth, it would be in an exceptional and better position to sequester the resource in limited supply later on in its growth phases. This finding was authenticated by Ku *et al.*, (2013) that the plant partitions more of its photosynthates in favour of the root system to enable the plant to acquire more of the limiting resource-moisture.

The interactive effects of phosphorous and water stress revealed that phosphorous (P) addition to soils with low P content improves plant performance in moisture stressed environments, by increasing grain yields; according to findings of Mataa *et al.*, (2019). Phosphorous is involved in the maintenance of hydrolytic conductance, energy metabolic systems and enhancement of root development in plants and by extension improved water

use efficiency through enhancement of stomata closures in moisture deficit environments (Jin *et al.*, 2015; Mataa *et al.*, 2019).

2.5.2. Light stress effects

Light is one of the environmental resources that are competed for in crop stands (Chang and Turner, 2019). This competition is strongly influenced by the number of plants in a unit areaplant density (Al-Suhaibani *et al.*, 2013). Plant density is reported to exert strong influence on general plant performance, yields and economic profitability (Al-Suhaibani *et al.*, 2013; Sonderegger, 2013; Mataa and Sichilima, 2019; Ciampitti and Vyn, 2011). Arenas and Fernández (2000) who reported that there was a negative biomass-plant density relation among crop stand agree with the above notion. Although the influence of plant density on light interception maybe negligible in the early stages of plant growth, plant density exerts strong influence on the amount of light intercepted by an individual plant as the crop canopy closes (Mataa and Sichilima, 2019).

Plants growing under a closed canopy of a plant stand are subjected to reduced amounts of irradiance and the quality of light received is also poor (Zhang *et al.*, 2017). This is because red (R) light is preferentially absorbed by chlorophyll in plant leaves and the far-red (FR) light is weakly absorbed or largely reflected (Park and Runkle, 2016). Consequently, the R:FR ratio reduces as sunlight progresses through the crop canopy (Franklin and Whitelam, 2005; Park and Runkle, 2016). The reduced R:FR ratio at lower level canopy structure signals a plant's morpho-physiological response (Franklin and Whitelam, 2005; Lin *et al.*, 2008) that is transduced into increased apical dominance, decreased branching, stem extension and internode elongation (Li *et al.*, 2014; Franklin and Whitelam, 2005; Mellendorf, 2011). In other plant species, the response is by varying their photosynthetic capacity (Li, *et al.* 2014).

In a related work, Mataa and Sichilima, (2019) observed that soybeans can adjust its plant architecture to available space. This enables it to achieve optimum yields over a broad range of plant arrangements and densities. Mellendorf, (2011) suggested that varying plant density at a critical growth stage is a viable alternative of positively manipulating plant productivity of soybeans under different environmental conditions through their changes in morphophysiological responses. However, the mechanism responsible for this yield compensation is not yet fully understood.

2.6. Soybean Production Management Practices

Production management practices are best optimised when growers of the crop utilize the understanding and knowledge of physiology of the crop, climate conditions, soil types, pest concerns, crop nutrient requirements, and geographical location of the production site (Kansas State University, 2016). Knowledge of the aforesaid production parameters is a tool which growers can use for planning or react to conditions they have for many of these factors (Mellendorf, 2011). However, there are limits to production management factors that producers can easily control. This include time of sowing the soybean seed, seed rates per unit area, seed sowing arrangements-row spacing, soil nutrient/moisture management and control measures for biotic stresses- weeds, insects and diseases (Sonderegger, 2013; Kansas State University, 2016).

Soybean is a short-day plant that responds to change in the duration of light (about 8 to 10 hours) and dark (about 14 to 16 hours) periods of the 24-hours day by transitioning from vegetative to reproductive growth phase (Rattunde *et al.*, 2016; Yang *et al.*, 2019). Plants have back and forth physiological processes that occur during the light and dark periods where primary and secondary metabolites are formed (Seaton *et al.*, 2018). Changes in the quantity of the metabolites formed during the light period signals a growth phase change (Franklin and Martin, 1988).

Day length varies during a calendar year for different geographic locations. Understanding the climatic calendar in relation to photoperiod of a specific geographical site is important as it helps growers to prescribe the appropriate planting time of the crop (Zhang *et al.* 2019). Delaying planting may result in shorter vegetative growth phase, thereby rendering the plant to develop lower yield component capabilities (Sonderegger, 2013). Soybean requires about 6 weeks of vegetative growth to develop sufficiently in size (61 cm to 91 cm) for optimum yield by the time reproductive growth phase sets in (Rattunde *et al.*, 2016). In most parts of Zambia, planting is scheduled for the mid-month of December through to the first week of January (MACO, 2002; Miti, 1995).

Recent plant breeding efforts have contributed to genetic advances in soybean yield potential (IITA, 2017; Chigeza *et al.*, 2019). Between 2003 and 2014, Zambia recorded an annual soybean yield increase of 85.2 Kg/year, one of the highest genetic gains in the world (Chiona, *et al.*, 2017). The genetic gains include reduced yield loss to lodging, and improved N₂-fixation ability and better stress tolerance (Kron *et al.*, 2008; Ku *et al.*, 2013). A rise in atmospheric CO₂ and advances in agricultural production practices technology have also contributed to increased soybean yields (Sonderegger, 2013). Soybeans is efficient at

utilising residual nutrients from previous crops; hence it forms an excellent rotation plant in a legume-cereal system (Sonderegger, 2013; Lamptey *et al.*, 2015).

Given the increasing human population, declining land availability and soil productivity, increasing yield is becoming an important goal of agriculture. As the crop's importance increases, growers seek for options to increase yields. Various alternative practices to maximise soybeans yields such as higher than recommended seeding rates, reduced row spacing, use of N fertilizers, seed treatment to enhance stand establishment, induced branching through breaking of apical dominance and soybeans diseases management through application of fungicides, establishing symbiotic relationships of soybean plants with mycorrhiza to help deal with salinity stress in suboptimal soils among others have been tried with varying success stories (De Bruin and Pedersen, 2008; Sonderegger, 2013; Rahmawati *et al.*, 2019)

Due to the increasing economic importance of soybean in Zambia and challenges of climatic variabilities, new varieties are being developed (SCCI, 2013; IITA, 2017) and released but production recommendations have not changed to take advantage of emerging varieties' variable phenotypic characteristics (Chigeza *et al.*, 2019). The high cost of soybean seed necessitates the re- evaluation and optimization of planting densities recommendations. Soybean yield is considered a function of four basic factors, commonly called 'yield components', which include seed mass, number of seeds/pod, number of pods/plant, and number of plants per given area (Hall, 1999). Detailed studies on the influence of planting densities may present more options to maximize soybeans plant productivity and thus increase yields.

Chapter 3:

3. MATERIALS and METHODS

3.1. Description of the Study Site

3.1.1. Site Location

The experiment was conducted at Mansa Research Station ((latitude 11° 14' 27.0" S and longitude 28° 57' 23.2" E) which is located in Agro-ecological region III. This region is characterised by high rainfall of about 1200 mm; with mean minimum and maximum temperature of 11.1 °C and 30.5 °C (figure 1). The site elevation was 1231 meters above sea level. The site had fallowed for at least two years.

3.1.2. Rainfall and Temperature

Annual rainfall is documented to be above 1130 mm. The wettest months are usually December, January and February, registering above 200 mm of rain (Chileshe and Chirwa, 1990). The mean temperatures during the growing season for the months of December through to March were above 15 °C. The highest minimum temperatures at Mansa Research Station coincided with the wettest months (figure 1). Soil temperature regimes for the area as described by Spaargaren (1987) fall within the isohyperthermic regime where soil temperatures at depths of up to 50 cm ranges between 22 to 27.9 °C.

The 2015/2016 planting season did not vary much from the rainfall and temperature distribution for the last 10 years. However, it was noted that the recorded mean rainfall for the months of January and February were less than the mean for the last 10 years. Though the total rainfall was above 100 mm for each month through the growing season, this particular year, distribution was very poor. The number of rain days were few during the critical growth stages of plant establishment and grain filling. The site experienced a partial drought of about 14 days in the 2nd and 3rd week of January 2016. This was at 1 week after seed germination.

3.1.3. Soil classification

According to Chileshe and Chirwa, (1990), the area is mostly covered by granite as a parent material, which is an acidic rock hence, the prevalence of acidic soils (pH water 5.1-5.5) (ZEMA, 2013). The soils at Mansa Research Station are *Acrisols* according to the world reference base (WRB) classification system (FAO, 2001; Sichinga, 2014). These soils are generally very deep to moderately deep, strong brown to yellowish red (5-7.5 YR), highly weathered fine loamy (18-34 % clay and more than 15 % fine or coarse sand) to clay sub soils which are also strongly leached (Chileshe and Chirwa, 1990; JAICAF, 2012). In most

cases these soils are overlain by sandy loam soils with the cation exchange capacity (CEC) of less than 24 meq. 100^{-1} g clay and the base saturation is below 50 % and thus of low fertility (FAO, 2001; JAICAF, 2012).

3.2. Soil chemical characteristics

The composite baseline soil samples were analysed using the Bray-1 method for determination of Phosphorus content in soils as described by Murphy and Riley, (1962). The exchangeable bases were assessed using the method described by Soltanpour and Schwab, (1977) to determine the presence and concentrations of Potassium (K), Calcium (Ca), Magnesium (Mg), and Sodium (Na). Organic carbon was determined using the Walkley and Black, (1934) method. Table 1 shows the soil chemical characteristics of the test site. The soil was acidic, as the mean pH (water) was low (4.4). Soil Phosphorous, organic Carbon and Magnesium were also observed to be below the critical values (i.e. 6.4 ppm and 0.77 % respectively). However, Calcium (Ca), Sodium (Na), and Potassium (K) were noted to be within range as they did not exceed the maximum critical level to support plant growth.

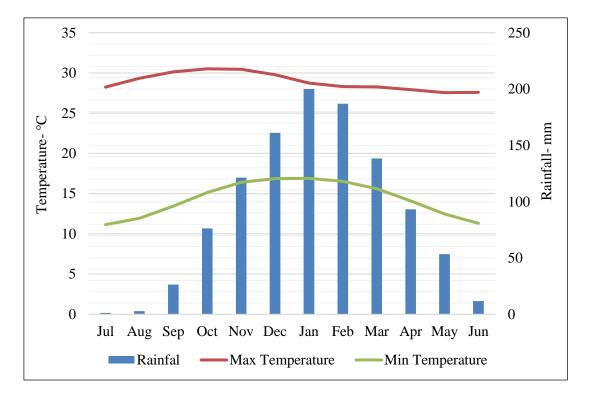


Figure 1: Mansa Research Station mean annual temperature and rainfall distribution 2015-2016.

Table 1: Mansa Research Station Soil Chemical Properties							
Parameter	Unit	Mean	Critical Levels				
рН		4.4	<4.5				
Р	ppm	6.4	<15				
Corganic	%	0.77	<1.58				
Ca	ppm	101.64	>200				
Mg	ppm	25.34	<50				
Κ	ppm	91.54	<40				
Na	ppm	2.71	>200				

(Analysis was done at ZARI, Mount Makuru Soils Lab-2016)

3.3. Plant Materials

3.3.1. Soybean genotypes

Three soybean varieties were used in the study. They were; Mwembeshi, Lukanga and SC Semeki.

Mwembeshi is an indeterminate, self nodulating and an early maturing type developed by IITA. It is reported to be tolerant to drought and resistant to bacterial pustule (*Xanthomonas axanapodis* pv. *Glycines*), soybean rust (*Phakopsora pachyr- hizi*), soybean mosaic virus, frogeye leaf spot (*Cercospora sojina* Hara), common soybean fly (*Melanagromyza sojae*), Soybean aphids (*Aphis glycines*), pod sucking bug (*Nezara viridula*) and pod borers (*Helicoverpa armigera*). (Mwase and Kapooria, 2001; Hartman, West and Herman, 2011; Alfy, 2017; Chigeza *et al.*, 2019)

Lukanga; developed by ZARI and obtained from Zamseed, is a determinate non-self nodulating variety. In terms of maturity it is a medium maturing variety. It is strong to shattering resistance and to most common soybeans' diseases and pests such as bacterial pustule (*Xanthomonas axanapodis* pv. *Glycines*), soybean mosaic virus, frogeye leaf spot, common soybean fly (*Melanagromyza sojae*) and Soybean aphids (*Aphis glycines*). The variety is reported to be drought tolerant (SEEDCO, 2015; Afriseed Stewards, 2018; Chigeza *et al.*, 2019).

SC Semeki is a determinate type of variety developed by SeedCo. SC Semeki is a non-self nodulating variety with a medium maturity. It has a long pod shatter free period, high resistance to Frogeye Leaf Spot (*Cercospora sojina* Hara), Wildfire (*Pseudomonas syrin-gae*) and Downy Mildew (*Peronospora manshurica*). It also demonstrates acceptable levels of tolerance to Red Leaf blotch (*Phoma glycinicola*) and Bacterial Blight (*Pseudomonas savastanoi pv. glycinea*). However, it is susceptible to Soybean rust (*Phakopsora pachyrhizi*). The variety is also reported to be drought tolerant (SEEDCO, 2015; Murithi *et al.*, 2016).

3.4. Cultural practices

Land preparation was done by ploughing and harrowing and later formed into seed-beds. Prior to seeding, each plot was applied with a stimulative dose (33kg ha⁻¹) of D-Compound (NPK 10:20:10+6S) fertilizer as recommended (Miti, 1995). In the early stages of crop development, the nitrogen fixing systems is inoperative, hence the need to apply an external N to meet the plants demand (Oyatokun and Oluwasemire, 2014). The fertilizer was broadcast and worked into the topsoil. Planting was done on 9th January 2016. Seeds were drilled by hand and thus the spacing ranged from 2.5 cm to 7 cm within rows and a 30 cm distance between rows was maintained.

The seeds were inoculated with rhizobia (*Rhizobium leguminosarum*) at the time of seeding. Hand weeding, pests and disease control were done as the need arose. Supplemental irrigation was done only when the rains were inadequate and soil moisture was deemed to be below field capacity. Harvesting was done by hand approximately 120 days after sowing when the crop had reached physiological maturity (8th to 10th May 2016).

3.5. Treatment factors and Experimental Design

The experiment had three factors; variety, planting density and thinning time and it was set as a split- split plot design. Variety was assigned to the main plots, plant density levels - the subplots and time of thinning or phenological stage was the sub- subplot. The upper limit of soybeans recommended planting density for Zambia of 350,000 to 400,000 plants ha⁻¹ was used as a benchmark or reference point (Miti, 1995). Three planting densities- 500,000; 600,000; and 700,000 plants ha⁻¹ densities were used. For thinning time, plants planted under the various densities were thinned down progressively to the recommended (control) density level of 400,000 plants ha⁻¹ at V₄, R₁ and R₈ of the plant phenological phases (Miti, 1995). V_0 was planted at the recommended density but for the purpose of the design used was treated as thinning at planting. Using Fehr and Caviness, (1971) soybean development staging system of cumulative leaf number, V_0 ; was assumed to be the planting stage, where "thinning" (i.e. seeding at the control density level of 400,000 plants ha⁻¹) was done at the time of seeding. V₄ is the vegetative growth stage where the fourth trifoliate leaf had completely unrolled and not touching (Wright and Lenssen, 2013). R₁; is the onset of the reproductive growth phase, where the plant had developed at least one open flower at any node and R₈ is when the plant had reached full maturing where at least ninety five percent of the pods have attained their full maturity colour (Fehr and Caviness, 1971; Wright and Lenssen, 2013). In practice V₀ and R₈ treatments were maintained with no thinning. Each of the main plots had an absolute area of 9.5 m by 5.5 m by three varieties and replicated four times. The smallest unit plot, a Sub-subplot measured 1.5 m by 1.5 m.

Equation 1: Split split plot model

The linear additive model for split split plot design was:

$$y_{ijkh} = \mu + \tau_i + \beta_j + (\tau\beta)_{ij} + \gamma_k + (\tau\gamma)_{ik} + (\beta\gamma)_{jk} + (\tau\beta\gamma)_{ijk} + \delta_h + (\tau\delta)_{ih} + (\beta\delta)_{jh} + (\tau\beta\delta)_{ijh} + (\gamma\delta)_{kh} + (\tau\gamma\delta)_{ikh} + (\beta\gamma\delta)_{jkh} + (\tau\beta\gamma\delta)_{ijkh}$$

Where: y_{ijkh} = the selected vegetative or reproductive parameter of the ith soybeans variety at the kth plant population density when thinned at the hth thinning time at each jth replicate.

$\mu =$	Overall mean of the population
$ au_i = \mathrm{i}^{\mathrm{th}}$	soybeans variety (main plot)
$\beta_j =$	j th block (replication) effect
$(\tau\beta)_{ij} =$	main plot (soybeans variety) error
$\gamma_k =$	k th plant population density (subplot)
$(\tau \gamma)_{ik} =$	Interaction between soybean variety and the plant population density
$(\beta \gamma)_{jk} =$	Error specific to plant population density
$(\tau\beta\gamma)_{ijk} =$	Subplot error
$\delta_h =$	h th thinning time (sub-subplot)
$(\tau\delta)_{ih} =$	Interaction between soybeans variety and thinning time
$(\beta\delta)_{jh} =$	error specific to processing method
$(\tau\beta\delta)_{ijh} =$	error specific to the interaction between soybeans variety and thinning
	time
$(\gamma \delta)_{kh} =$	Interaction between plant population density and thinning time
$(\tau\gamma\delta)_{ikh} =$	Interaction between soybeans variety, plant population density and
	thinning time
$(\beta\gamma\delta)_{jkh} =$	error specific to interaction between plant population density and
	thinning time
$(\tau\beta\gamma\delta)_{ijkh} =$	sub-subplot error.

(Gomez and Gomez, 1984).

3.6. Data Collection

Data was collected on plant height, biomass weight, and root-shoot weight ratio for vegetative parameters. Reproductive parameters that were monitored included number of grains per pod, number of pods per plant, 100 grain weight, grain yield and harvest index.

3.6.1. Vegetative Parameters

Plant height

Plant height was collected using a hand rule in centimetres. Five plants were randomly selected in a sub- subplot and their height was taken from the plant base to the upper most shot tip. Measurement were taken at R1 and R8 and expressed as a mean of the five individual plants.

Biomass

Biomass weight was determined using destructive sampling method where five plants were carefully uprooted from the border rows of the sub subplot. After prior moistening of the soil, the plants were dug up carefully to retain as much root mass as possible using a hand hoe. Soil debris were carefully washed off using tap water. Biomass was determined at V3-V4 and R8 phenological stages. Fresh weights were taken and thereafter the samples were air- dried under shade (for about 2 weeks) to constant mass and then reweighed to determine dry weights (Mataa and Sichilima, 2019).

Root-shoot weight ratio

The fresh and dry mass of the below ground and above ground dry biomass from five randomly selected plants were taken at V3-V4 phenological stages and means based on the five plants were used to compute the root and shoot ratios.

3.6.2. Reproductive Parameters

Grains per pod

The parameter was determined at harvest by counting number of grains from five randomly selected plants and expressed as a mean value.

Pods per plant

Number of pods per plant is one of the components of yield in soybeans and as such it is an important factor in the calculation of crop economic yield. Pods per plant were determined by taking counts of number of pods at maturity of the five randomly selected plants.

100- grain weight

The weight of the 100 grain seeds is one of the yield components for soybeans, hence it is an important factor in the determination of crop yield. It is generally positively correlated with yield (Zhang *et al.*, 2015). It was measured by taking a triplicate sample of 100- grains per subplot of soybeans in grams to come up with a representative average weight.

Grain yield

Yield is a function of planting density per unit area, number of pods per plant and number of grains per pod (Mataa *et al.*, 2019). A net plot (i.e. sub-sub plot excluding border rows) was harvested when the crop had attained at least 95 % senescence and dried under shade for two weeks to constant moisture content. Grain yield was determined from each plot by weighing the total grains harvested in the net plot and expressed as tons per hectare.

Harvest index

Harvest index (HI) is a genotype fixed trait which has been used by crop plant breeders as a parameter in the selection criteria for high yielding genotypes due to its positive correlation to yield in general. HI expressed as a ratio of the crop's economic yield over biological yield (Amanullah, 2016) was determined by dividing grain yield with the total dry biomass yield.

3.7. Statistical analysis

Data was analysed using *GenSTAT* (version 18) and graphical illustrations were generated using Microsoft Excel. Data was subjected to analysis of variance (ANOVA), correlation, multiple regression and mean separation was done using least significant differences (LSD).

Chapter 4

4. **RESULTS**

4.1. Summary of factor significance

Table 2 presents a summary of factor significances for the measured parameters at both vegetative and reproductive growth stages. Variety (G) affected most parameters, except for root to shoot weight ratio and the 100 grains weight. Yield was highly influenced by variety ($p \le 0.001$). Biomass at R1 was very significantly influenced by variety ($p \le 0.01$). Plant height, biomass at R8, grains per pod, number of pods per plant and harvest index were significantly influenced by variety ($p \le$ 0.05). Plant density (D) (subplot) had no significant effect on most parameters (p > 0.05) except for biomass at R8, 100 grain weight and harvest index ($p \le 0.05$). Developmental stage at which thinning was done (T) influenced significantly root to shoot weight, grains per pod and harvest index ($p \le 0.05$). Thinning time affected yield only among all parameters very significantly ($p \le 0.01$).

Significant interactions between G and D were observed for all parameters ($p \le 0.05$) but for 100 grains weight (p > 0.05) and harvest index ($p \le 0.01$) which were non- significant and very significant respectively.

Significant interactions between G and T were observed in all parameters except 100 seed weight (p > 0.05). G and T interaction for biomass at V4 and grain yield were very significant ($p \le 0.01$) and highly significant (≤ 0.001) respectively.

D and T significant interactions ($p \le 0.05$) were observed for plant height at R8, biomass at R8, root to shoot weight, number of grains per pod, number of pods per plant and harvest index. D and T interaction for 100 seed weight were very significantly influenced (≤ 0.01). The interactions of D and T for biomass at R1 and grain yield were highly significant (≤ 0.001).

Three-way interactions between G, D and T were observed in plant height at R1 ($p \le 0.01$), biomass at V4 ($p \le 0.001$), biomass at R8, root to shoot weight, number of grains per pod, number of pods per plant 100 grains weight and harvest index ($p \le 0.05$); and grain yield ($p \le 0.001$).

Source of Variation	d. f.	Plant	Plant	Biomass	Biomass	Root:	Grains	Pods	100-	Grain	Harvest
		height	height.	(V4) ^z	(R8) ^x	shoot ^w	per pod	per	Grain	Yield	index
		(R1) ^y	(R8) ^x					plant	weight		
Replication Stratum	3										
Variety	2	*	*	**	*	ns	*	*	ns	***	*
Residual	6										
Density	2	ns	ns	ns	ns	ns	ns	ns	*	ns	*
Variety x Density	4	*	*	*	*	*	*	*	ns	*	**
Residual	18										
Thinning	3	ns	ns	ns	ns	*	*	ns	ns	**	*
Variety x Thinning	6	*	*	**	*	*	*	*	ns	***	*
Density x Thinning	6	ns	*	***	*	*	*	*	**	***	*
Variety x Density x Thinning	12	**	ns	***	*	*	*	*	*	***	*
Residual	81										
Total	143										

Table 2: Summary ANOVA table showing significance of different sources of variation.

Factor significance; *** highly significant ($p \le 0.001$); **very significant ($p \le 0.01$); * significant ($p \le 0.05$) and ns- nonsignificant.

^z Vegetative stage V4, ^y Reproductive stage R1, ^x Reproductive stage R8, ^w Root: shoot ratio- determined at vegetative stage V4.

4.2. Vegetative parameters

4.2.1. Plant height at R1

The main effect of variety showed significant effects (p < 0.05) on plant height (R1) as shown in Table 3. Lukanga had the tallest plants followed by Mwembeshi and SC Semeki was the shortest at 29.30 cm, 24.63 cm and 24.42 cm respectively.

Table 4 presents the two-way factor interactive effects of plant height during the vegetative growth phase. There were significant variety and planting density effect (p < 0.05) observed in G₁ x D₁ (29.32 cm), G₁ x D₂ (30.13 cm) and G₁ x D₃ (28.47 cm) interactions for the plant height parameter at R1.

The interaction of variety and thinning (G x T) exhibited significant differences (p < 0.05), specifically for G₁ x T ₁, G₁ x T ₂ and G₁ x T₃ interactions with values of 29.61 cm, 30.76 cm and 29.48 cm respectively.

There were no significant differences (p>0.05) among means of plant height at R1 when it was subjected to different levels of planting density and thinned at different growth stages (D x T).

The three-way factor interactive effects as presented in Figure 2 show that variety by density by thinning time interaction was very significant (p<0.01) for plant height taken at the end of the vegetative growth phase. The tallest plants were observed in the Lukanga and Mwembeshi at 600 K plants ha⁻¹ density when thinned at V4 growth stage (32.61cm). The shortest plants where observed in Mwembeshi at 500 K plants ha⁻¹ population density when thinned at R1 (17.52 cm).

4.2.2. Plant height at R8

The single effects of treatments on plant height at R8 are presented in Table 3. They show that there were significant differences (p<0.05) between varieties in plant height at harvest (R8). At 34.32 cm, 33.14 cm and 31.48 cm respectively, Lukanga had the tallest plants followed by Mwembeshi and SC Semeki recorded the shortest plants. The other main effects of planting density and thinning time did not have significant effect on plant height at R8.

There were significant two-way interactions (p < 0.05) between factors on plant height at R8 as shown in Table 4. Variety and planting density effect observed in $G_1 \times D_1$ (33.90 cm), $G_1 \times D_2$ (34.62 cm), $G_1 \times D_3$ (34.45 cm) and $G_2 \times D_2$ (34.71 cm) interaction for the plant height parameter at R8 revealed significant differences at lsd (5 %) of 3.73. The interaction of variety and thinning (G x T) exhibited significant differences (p < 0.05), specifically for G_1

x T ₃, G₁ x T₄ and G₁ x T₂ interactions with values of 34.76 cm, 34.23 cm and 34.18 cm respectively at lsd (5 %) value of 3.12. Planting density and thinning time interaction exhibited significant differences (p < 0.05) specifically at D₂ x T₃ (35.84 cm), D₁ x T₄ (34.32 cm), D₂ x T₂ (33.65 cm), D₁ x T₁ (33.49 cm), D₃ x T₄ (33.13 cm) and D₁ x T₂ (33.04 cm) in their respective descending order.

Figure 3 shows the three- way interactions of variety, planting density and thinning time (G x D x T) on plant height at R8. Generally, for Lukanga at 500 and 600 K density there was a trend where plant height increased with an advance of the phenological phase at thinning. The tallest plants occurred at 600 K density. For Mwembeshi the 600 K density had the tallest plants and at this density plant height increased with phase at thinning.

	Parameter						
		Plant h	-	Biomass (ton.		Deets Ol	
Factor		(cm	1)	ha ⁻¹)		Root: Shoot Ratio	
		R1	R8	V4	R8	Ratio	
Variety (G)							
Lukanga	(G ₁)	29.30	34.32	1.99	5.81	10.87	
Mwembeshi	(G ₂)	24.63	33.14	1.72	5.44	9.00	
SC Semeki	(G ₃)	24.42	31.48	2.76	2.90	11.24	
Lsd (G)		4.13	2.12	0.44	1.85	2.7	
Planting Density (D) ^z							
500	(D ₁)	25.29	33.01	2.20	4.24	10.5	
600	(D ₂)	27.35	33.56	2.30	5.70	11.6	
700	(D ₃)	25.72	32.37	1.97	4.21	8.8	
Lsd (D)		2.98	2.35	0.40	1.42	3.7	
Thinning Stage (T)							
$\mathbf{V}_0^{\mathbf{w}}$	(T ₁)	25.38	32.74	2.08	5.25	9.50	
$V_4{}^v$	(T ₂)	27.02	33.08	2.18	4.73	9.2	
R_1^y	(T ₃)	25.81	32.96	2.33	4.43	9.7	
R_8^x	(T ₄)	26.26	33.14	2.03	4.45	12.9	
Lsd (T)		2.04	1.71	0.51	1.06	3.1	

Table 3: Single effects of variety, planting density and thinning stage on vegetative parameters of soybeans (*Glycine max*).

^z Planting density in thousands of plants per hectare.

^w No thinning, planted at control density of 400 K plants ha⁻¹.

^v Plants at stage where four nodes on main stem beginning with the unifoliate node are fully developed.

^y Reproductive stage where there is at least one flower at any node.

^x 95 % of pods are brown- harvest maturity.

4.2.3. Biomass (V4)

Variety had exhibited very significant effects (p < 0.01) on biomass at V4. Plant density and phenological stage at thinning did not have a significant effect on biomass (Table 3). There was very significant G x T (p < 0.01) and highly significant D x T and G x D x T interactions (p < 0.001).

In terms of varietal effect at V4, SC Semeki (2.76 tons Ha⁻¹) had significantly more biomass than the other two varieties (1.8 tons ha⁻¹) (Table 2 and Table 3). There was no difference between Mwembeshi and Lukanga.

The two-way interactive effect as presented in Table 4 revealed some significant differences (p < 0.05) on biomass weight for variety at different levels of planting densities. The biomass significant differences were recorded in the G₃ x D₃ (2.92 tons Ha⁻¹), G₃ x D₂ (2.76 tons Ha⁻¹) and G₃ x D₁ (2.60 tons ha⁻¹) interactions.

The interaction of variety and thinning time treatments on biomass weight was highly significant (P<0.01). The highest biomass (3.58 tons ha⁻¹) was observed in SC Semeki variety for treatment that were thinned early at V0 (G₃ x T₁). Biomass were accumulated least (2.03 ton. ha⁻¹) in treatments that were not thinned at all (R8) in SC Semeki (G₃ x T₄). The case was however different for Lukanga and Mwembeshi whose highest dry biomass weights (2.46 and 2.32 ton. ha⁻¹) were observed in thinning times R1 and R8 respectively and their respective least dry biomass weights (1.64 and 1.10 ton. ha⁻¹) were observed in V4 and R1 thinning time treatments.

There were highly significant differences (p<0.001) in biomass at V4 when planting density was subjected to different levels of thinning time (D x T). The treatment that accumulated the highest dry biomass weight (2.88 ton. ha⁻¹) were those that were planted at 600 K plants ha⁻¹ and thinned at V4 (D₂ x T₂). The overall least accumulated BM (1.35 ton. ha⁻¹) was observed in treatment that were planted at the density of 500 K plants ha⁻¹ and thinned at V4. Treatment plots that were subjected to higher planting density (700 K plants ha⁻¹) recorded their highest biomass weight (2.29 ton. ha⁻¹) when they were thinned at V0 growth stage. The least weight (1.62 ton. ha⁻¹) at the preceding density was recorded in plots that were thinned at R1 growth stage (D₃ x T₃).

The three-way interaction effect as presented in Figure 4 revealed that there were highly significant differences (p<0.001) between treatments. With the variety Lukanga at 500 K plants ha⁻¹, BM increased with delay in thinning. At 700 the BM reduced with delayed

thinning. Mwembeshi at 600 and 700 K, BM tended to decline with delayed thinning stage. For SC Semeki at 500 and 600 K BM declined with delay in thinning stage. At 700 it increased with delay in thinning time.

Biomass (R8)

The main effect of variety on biomass weight at R8 was significant (p<0.05). As can be noted from Table 3, SC Semeki variety had significantly lower mean dry biomass weight (2.90 tons. ha⁻¹) than Lukanga and Mwembeshi which had 5.81 and 5.44 tons. ha⁻¹ respectively at lsd (5 %.) value of 1.85. Planting density exerted significant effect (p<0.05) on biomass at R8. Density 600 K plants yielded the highest BM (5.70 tons ha⁻¹) and the least was observed in density 700 K plants ha⁻¹. Thinning time however did not exert any significant difference (p>0.05) on BM.

Table 4 show the two- way interactions on biomass. In terms of G x D interactions, Lukanga at the three planting densities had the highest BM. For G x T interactions Lukanga had higher BM across thinning times. The highest BM occurred at $G_2 \times D_2$ and $D_2 \times T_3$.

At R8, significant three- way interactions (p<0.05) were observed (Fig. 5). For Lukanga, thinning at V0 caused the lowest BM. At 700 K the highest BM was observed at V0 thinning and the lowest at V4. At 600 K there were no significant differences. For Lukanga the highest BM was at 600 K with no differences due to thinning stage.

BM increased with delay in thinning in Mwembeshi and the highest BM was at 600 K thinned at R1. Generally, Lukanga had the highest BM and SC Semeki had lowest BM at all planting densities compared with the other two varieties and at 500 K and 600 K the BM declined with delayed thinning time.

4.2.4. Root-Shoot Ratio

There were no significant differences on the main effects for variety and planting density (P>0.05). Thinning time however showed significant differences (p<0.05) together with all the factors' interactions (Table 2 and Table 3).

The ratio of root to shoot was higher and significantly different when thinning was withheld until R8 growth stage.

Table 4 presents two- way interactions. SC Semeki had the highest ratio when planted at 600 K plants ha⁻¹ (G₃ x D₂) and Lukanga exhibited a significantly (p<0.05) higher ratio when planted at 500 K plants ha⁻¹ (G₁ x D₁). Variety was significantly influenced by thinning at

V4 for both Lukanga and SC Semeki ($G_1 \times T_2$ and $G_3 \times T_2$). When density interacted with thinning time, it resulted in significant effects, specifically for density 600 K plants ha⁻¹ when thinned at V4 and R1, and for density 500 K plants ha⁻¹ when thinned at V4.

The three- way interaction showed significant effects (p<0.05) for root to shoot ratio. For Lukanga, thinning at V4 generally resulted in higher root to shoot ratio across planting densities. The highest ratio for Lukanga was observed at density 600 K when thinned at V4. Mwembeshi had its highest ratio recorded in density 600 K plants ha⁻¹ when thinned at R1. Mwembeshi showed tendencies to increase the ratio for every subsequent thinning stage across planting densities. However, the opposite was true for SC Semeki which demonstrated the propensity to reduce the root to shoot ratio for every successive thinning stage across all planting densities. The highest ratio across all varieties and densities was observed in SC Semeki at density 600 K and thinning at V4 while the least was recorded in the same variety (SC Semeki) but at the density of 700 K and thinning at R1.

	Parameter					
Factor	Plant he	ight (cm)	Biomass (ton. ha ⁻¹)	Root: Shoot	
	R 1	R 8	V4	R 8	Ratio	
G ₁ x D ₁	29.32	33.90	2.27	5.24	13.07	
$G_1 \ge D_2$	30.13	34.62	2.25	6.49	10.15	
G ₁ x D ₃	28.47	34.45	1.45	5.68	9.40	
$G_2 \ge D_1$	22.68	32.23	1.72	3.87	7.90	
$G_2 \ge D_2$	27.54	34.71	1.89	7.47	10.34	
$G_2 \ge D_3$	23.67	32.47	1.53	4.98	8.74	
$G_3 \ge D_1$	23.86	32.91	2.60	3.60	10.69	
$G_3 \ge D_2$	24.38	31.33	2.76	3.13	14.52	
G ₃ x D ₃	25.01	30.18	2.92	1.98	8.52	
Lsd(G x D)	5.46	3.73	0.68	2.53	5.65	
$G_1 \ge T_1$	29.61	34.13	1.67	5.70	8.88	
$G_1 \ge T_2$	30.76	34.18	1.64	5.35	14.85	
G ₁ x T ₃	29.48	34.76	2.46	6.19	9.02	
$G_1 \ge T_4$	27.37	34.23	2.19	5.99	10.74	
$G_2 \ge T_1$	23.33	32.84	1.73	4.52	8.46	
$G_2 \ge T_2$	25.36	33.31	1.71	5.19	9.74	
$G_2 x T_3$	23.30	33.02	1.10	6.74	9.44	
G ₂ x T ₄	26.54	33.38	2.32	5.30	8.34	
$G_3 \ge T_1$	23.20	31.90	3.58	3.07	12.00	
$G_3 \ge T_2$	24.93	31.93	2.76	2.81	14.26	
G ₃ x T ₃	24.66	30.44	2.67	2.83	10.04	
G3 x T4	24.87	31.64	2.03	2.90	8.68	
Lsd(G x T)	4. 78	3.12	0.84	2.27	5.22	
$D_1 \ge T_1$	24.69	33.49	2.03	4.04	10.95	
$D_1 \ge T_2$	25.65	33.04	1.35	4.03	13.67	
$D_1 \ge T_3$	24.02	31.20	2.58	4.03	8.58	
$D_1 \ge T_4$	26.79	34.32	2.83	4.85	9.02	
$D_2 \ge T_1$	26.47	32.92	2.66	5.03	9.58	
$D_2 \ge T_2$	28.85	33.65	2.88	5.61	14.70	
$D_2 \ge T_3$	28.06	35.84	2.03	7.56	13.35	
$D_2 \ge T_4$	26.01	31.80	1.63	4.60	9.06	
$D_3 \ge T_1$	24.97	32.45	2.29	4.23	8.81	
$D_3 \ge T_2$	26.55	32.72	1.87	3.72	10.49	
D ₃ x T ₃	25.36	31.17	1.62	4.16	6.57	
$D_3 \ge T_4$	25.98	33.13	2.08	4.73	9.68	
Lsd(D x T)	4.18	3.40	0.85	2.09	5.88	

 Table 4: Two-way interactive effects of variety, planting density and phenological stage at thinning on vegetative parameters of soybeans (*Glycine max*).

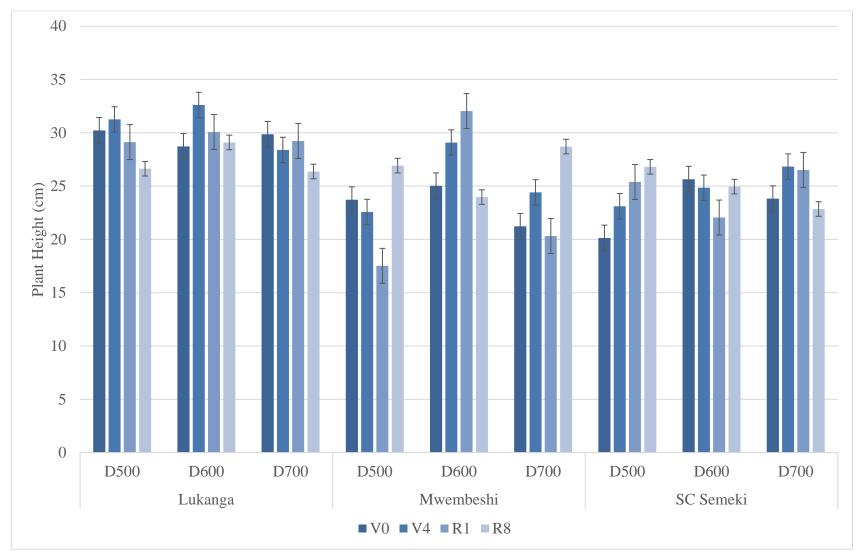


Figure 2: Plant height as Measured at R1. Vertical bars are standard errors.

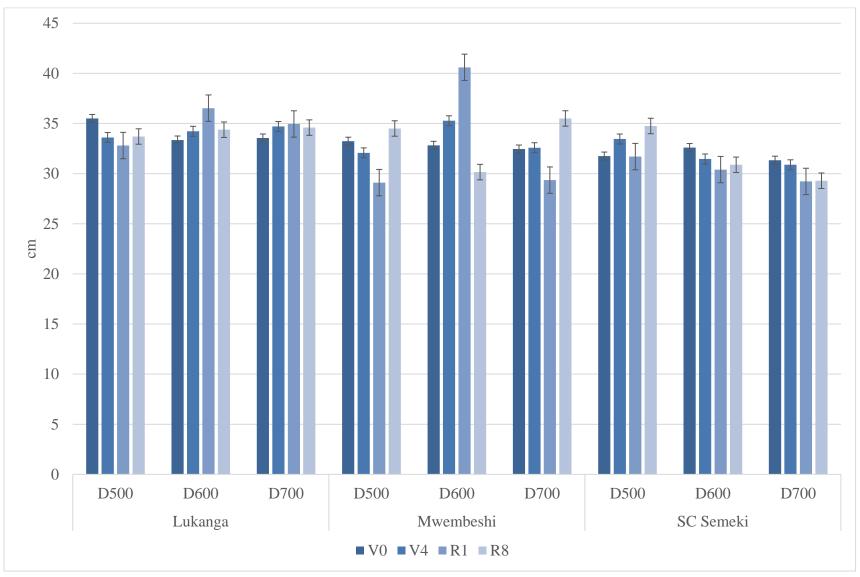


Figure 3: Plant height measured at R8. Vertical bars are standard errors.

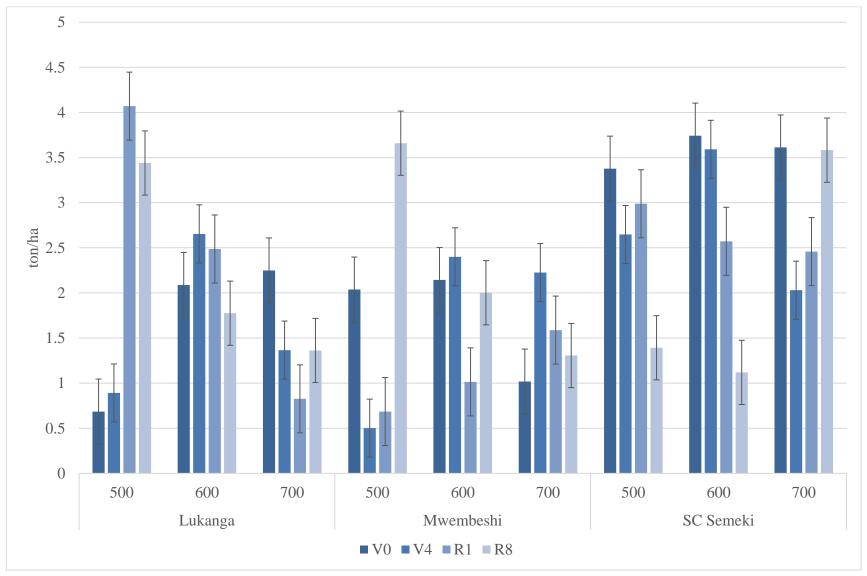


Figure 4: Biomass measured at V4. Vertical bars are standard errors.

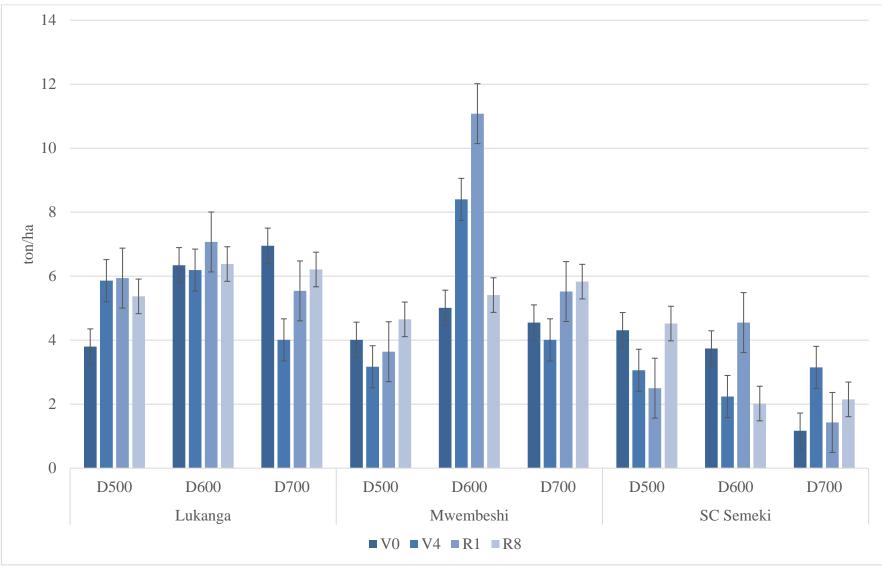


Figure 5: Biomass measured at R8. Vertical bars are standard errors.

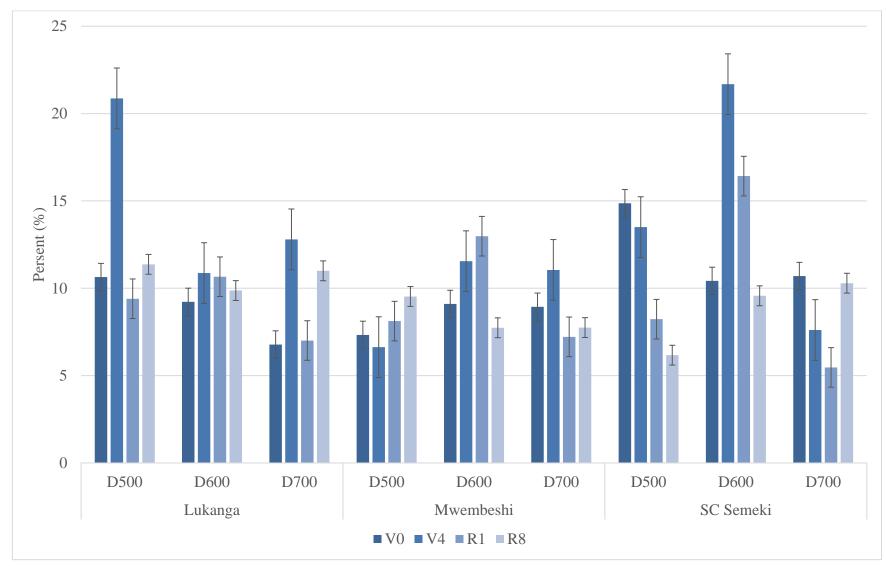


Figure 6: Root: Shoot weight ratio. Vertical bars are standard errors.

4.3. Reproductive parameters

4.3.1. Number of grains per pod

There were significant differences (p<0.05) in the number of grains per pod among varieties. Lukanga variety had the highest and only significantly different mean number of grains per pod (2.41) compared to SC Semeki. There was no significant difference between Mwembeshi and Lukanga.

Planting density did not exert any significant effect on grains per pod (p>0.05).

Thinning stage however exerted significant difference (p < 0.05). This was noted at T₃ thinning stage.

The two- way interactions were observed to be significant (p < 0.05) at all factor levels i.e. G x D, G x T and D x T. For Lukanga, at 700 K density had significantly more grains per pod compared to SC Semeki at the same density. The significant interactions of variety (G) with thinning time (T) was observed at all levels of thinning for Lukanga, at two levels of thinning for Mwembeshi and SC Semeki (T₁ and T₃ respectively). The interaction of planting density with thinning time (D x T) showed significant effect for density D₁ and D₂ which was thinned at T₂, T₃ and T₃ respectively.

The three-way interaction effects (Figure 7) of variety, planting density and time of thinning (V x D x T) on number of grains per pod showed some significant effects (p < 0.05) despite lack of perceivable trends.

Parameter						
	Grains per pod	Pods per plant	100-Grains weight (g)	Yield (ton. ha ⁻¹)	HI (%)	
(G ₁)	2.41	12.24	13.27	2.43	37.99	
(G ₂)	2.32	16.35	13.56	1.95	35.24	
(G ₃)	2.24	13.73	13.42	1.17	34.56	
	0.10	3.87	0.46	0.24	3.28	
(D ₁)	2.34	14.38	13.29	1.83	36.47	
(D ₂)	2.34	13.72	13.41	1.89	36.13	
(D ₃)	2.30	14.21	13.55	1.83	35.20	
	0.07	2.19	0.26	0.21	1.07	
(T ₁)	2.34	14.45	13.27	2.02	36.95	
(T ₂)	2.31	13.64	13.44	1.86	34.53	
(T ₃)	2.38	14.81	13.38	1.67	36.80	
(T ₄)	2.26	13.53	13.57	1.85	35.45	
	0.11	2.42	0.37	0.19	1.93	
	(G1) (G2) (G3) (D1) (D2) (D3) (T1) (T2) (T3)	Grains per pod (G1) 2.41 (G2) 2.32 (G3) 2.24 (G3) 2.24 (D1) 2.34 (D2) 2.34 (D3) 2.30 (T1) 2.34 (T2) 2.31 (T3) 2.38 (T4) 2.26	Grains per podPods per plant(G1)2.4112.24(G2)2.3216.35(G3)2.2413.73(G1)2.3414.38(D2)2.3413.72(D3)2.3014.21(D3)2.3014.21(T1)2.3413.72(T1)2.3413.72(T1)2.3414.21(T1)2.3014.21(T1)2.3414.45(T2)2.3113.64(T3)2.3814.81(T4)2.2613.53	Grains per podPods per plant100-Grains weight (g)(G1)2.4112.2413.27(G2)2.3216.3513.56(G3)2.2413.7313.42(G1)2.2413.730.46(D1)2.3414.3813.29(D2)2.3413.7213.41(D3)2.3014.2113.55(D4)10.072.190.26(T1)2.3414.4513.27(T2)2.3113.6413.44(T3)2.3814.8113.38(T4)2.2613.5313.57	Brains Pods per plant Hon-Grains weight (g) Yield (no. har') (G1) 2.41 12.24 13.27 2.43 (G2) 2.32 16.35 13.56 1.95 (G3) 2.24 13.73 13.42 1.17 (G3) 2.24 13.73 13.42 1.17 (G4) 2.34 13.73 0.46 0.24 (D1) 3.87 0.46 1.83 (D2) 2.34 13.72 1.83 (D2) 2.34 13.72 1.83 (D2) 2.34 13.72 1.83 (D3) 2.30 14.21 13.55 1.83 (T1) 2.34 14.45 13.27 2.02 (T1) 2.34 14.45 13.27 2.02 (T1) 2.34 14.45 13.27 2.02 (T2) 2.31 13.64 13.41 1.86 (T3) 2.38 14.81 13.38 1.67 (T4) 2.26 13.53 13.57 1.85	

Table 5: Treatment Effects on selected parameters during the reproductive growth phase

Parameter

^z Planting density in thousands of plants per hectare.

^w No thinning; planted at control density of 400 K plants ha⁻¹.

^v Plants at stage where four nodes on main stem beginning with the unifoliate node are fully developed.

^y Reproductive stage where there is at least one flower at any node.

^x 95 % of pods are brown- harvest maturity.

4.3.2. Number of Pods Per Plant

Table 5 shows the effect of treatments on reproductive parameters. Mwembeshi had significantly more pods per plant compared to Lukanga but there was no difference between Lukanga and Semeki.

Planting density and thinning stage did not exert any significant effect on number of pods per plant.

The two- way interaction revealed significant differences (p < 0.05). Mwembeshi had significantly more pods per plant when it was planted at densities i.e. 600 K and 700 K plants per ha⁻¹ compared to Lukanga and SC Semeki. The interaction of variety and thinning stage revealed that Mwembeshi had significantly more pods per plant when it was thinned at T_1 and T_3 . Density had no influence on the number of pods except when density 600 was thinned at R1 (T_3).

The three way- interactive effect showed significant effects (Figure 8). Mwembeshi revealed significant differences at density 600 K at all levels of thinning except R8. SC Semeki only showed some significant differences at density 500 K and then thinning was done at T_2 and T_4 . For Mwembeshi, it was observed that the number of pods increased when thinning was delayed at low density. However, at higher density, delayed thinning reduced the number of pods per plant.

4.3.3. Grain weight

The single factor effects were non-significant except for density (Table 2 and Table 5). The highest grain weight was observed in density D_3 and the least was recorded in density D_1 .

There were no significant differences (p > 0.05) among means of grain weight when variety was observed at different levels of planting density (G x D) as presented in Table 6. Observing variety at different thinning stages (G x T) did not show any significant differences, except for Mwembeshi when it was thinned at R1.

Treatment combination of planting density and thinning time (D x T) done at different stages of plant growth however demonstrated very significant differences ($p \le 0.01$). The highest grain weight (14.18 grams) was recorded in the treatment combination of density 700 K plants ha⁻¹ and thinning done at R8 (D₃ x T₄). Density 500 K plants ha⁻¹ and thinning done at R1 (D₁ x T₃) yielded the least grain weight (12.99 grams).

Significant three-way interactive effects of variety, planting density and time of thinning were observed (p<0.05) with respect to grain weight. The variety with the highest 100 grains

weight was Mwembeshi, which occurred at the population density of 600 K plants ha⁻¹ when thinned at R1 (15.07 grams). The lowest 100 grains weight was observed in the interaction of SC Semeki at density 500 K plants ha⁻¹ when it was thinned at R1 (12.28 grams). Thinning time did not have any effect on Mwembeshi at the density level of 500 K plants ha⁻¹ thus all the resulting means were statistically the same. Density 700 K plants ha⁻¹ for Mwembeshi revealed a significantly higher grain weight (14.68 grams) when thinning was at R8. Thinning Lukanga at R8 from density 700 K plants ha⁻¹ resulted in higher grain weight (14.22 grams). When Lukanga was thinned from its original density of 600 K plants ha⁻¹ at R1 stage, the lowest 100 grains weight across all thinning and density levels was recorded (12.93 grams).

4.3.4. Grain yield

As presented in Table 5, varietal effect on grain yield was highly significant (p < 0.001). Means of variety on grain yield varied from the lower limit of 1. 17 tons ha⁻¹ exhibited by SC Semeki to upper limit of 2.43 tons ha⁻¹ exhibited by Lukanga. Mwembeshi yielded 1.95 tons ha⁻¹, which was statistically different from the yield values of the other two varieties (Lukanga and SC Semeki).

Planting density showed no significant (p > 0.05) effect on grain yield.

Thinning time showed very significant effect (p < 0.01) on grain yield. Plots that were thinned at V0 (T₁) had the highest mean grain yield (2.02 tons ha⁻¹). Treatments that were thinned at V4 (1.85 tons. ha⁻¹) growth stage were statistically the same as those that were maintained without thinning until harvest time at R8 (1.86 tons ha⁻¹). Plots that were thinned at R1 yielded the least amount of grain (1.67 tons ha⁻¹).

Table 6 presents the two-way interactive effects. Varietal effect on grain yield when observed at different levels of planting density was not significant (p < 0.05). Lukanga variety performed significantly better than Mwembeshi and SC Semeki at every tested density. However, within Lukanga, there were no differences in grain yield at different densities. SC Semeki was the least performing regarding grain yield.

Phenological stage of thinning (V0, V4, R1 and R8) had a significant effect on yield and therefore G x T and D x T were highly significant (p < 0.001).

In terms of G x T, the highest yield was observed in Lukanga (2.68 ton. ha⁻¹), when thinning was done at R1 (T₃). The minimum value of 0.74 tons ha⁻¹ was observed in SC Semeki variety when thinning was done at V0 (T₁). Lukanga and Mwembeshi had their respective

best (2.68 and 2.14 ton. ha⁻¹) yields occurring when thinning was done at the onset of the reproductive growth phase-R1.

Interaction effect of planting density and time of thinning (D x T) at different phenological stages on grain yield was very highly significant (p<0.001). Planting density of 600 K plants ha⁻¹ exhibited the highest yield value of 2.51 tons ha⁻¹, when thinning was done at R1. The lowest yield (1.42 ton. ha⁻¹) was observed at density 700 K plants ha⁻¹, when thinning was done at V0. Density 500 K and 700 K plants ha⁻¹ had their respective best yields (2.08 and 2.09 ton. ha⁻¹) happen in plots that were maintained without thinning-R8 respectively. The lowest yields for density 500 K plants ha⁻¹ level occurred when thinning was done at V4 (1.45 ton. ha⁻¹).

The three-way interaction of variety, planting density and time of thinning had highly significant effects (p<0.001) on grain yield (Fig. 10). The overall highest yield (2.92 ton. ha⁻ ¹) was recorded at treatment plots with variety Mwembeshi, which was planted at density 600 K plants ha⁻¹ and thinned at R1 growth stage (G₂ x D₂ x T₃). Lukanga's density of 700 K plants ha⁻¹ recorded its highest yield value of 2.88 tons ha⁻¹ at the thinning time stage of V0 ($G_1 \times D_3 \times T_2$). The highest yield value (2.92 ton. ha⁻¹) for Mwembeshi however was observed at the planting density of 600 K plants ha⁻¹, when thinning was done at R1 (G₂ x $D_2 \ge T_3$). Crops left without thinning (T₄) resulted in the least performance of grain yield values of 1.76- and 1.54- tons ha⁻¹ for Lukanga (600 K plants ha⁻¹) and Mwembeshi (500K plants ha⁻¹) respectively. Variety SC Semeki had its best performance on grain yield (2.52) ton. ha⁻¹) achieved at the treatment combination of density 500K plants ha⁻¹ and thinning time at R8 (G₃ x D₁ x T₄). The least yield (0.62 ton. ha⁻¹) was recorded in the combination of SC Semeki variety at density 500 K plants ha⁻¹ when thinned at R1 stage of growth. However, the highest yield for SC Semeki (2.06 ton. ha⁻¹) at density 600 K plants ha⁻¹ occurred when thinning was done at R1. At density 700 K plants ha⁻¹ for SC Semeki, thinning at R8 resulted in the best yield $(1.31 \text{ ton. ha}^{-1})$.

4.3.5. Harvest Index

The effects of treatments on harvest index are presented on Table 2 and Figure 11. Lukanga variety had the highest harvest index (0. 379) and it was significantly different from SC Semeki which had the ratios of 0. 345 but not from Mwembeshi which had the ratio of 0. 352. Hence, there was no significant difference between the means of HI ratios for Lukanga and Mwembeshi.

The treatment of planting density (D) revealed significant effect on HI (p < 0.05). Density 500 K plants ha⁻¹ had the highest HI (0.365) from density 700 K plants ha⁻¹ (0.352) but was not significantly different from density 600 K plants ha⁻¹ (0.361).

Thinning time (T) had a significant effect on HI (p < 0.05). Treatments that were thinned at R1 had the highest mean HI ratios (0.37). The least HI was recorded in treatments thinned at R8 (0.35).

The two-way interaction G x D was very significant (p<0.01). The highest HI ratio was recorded on Lukanga when it was planted at the density of 600 K plants ha⁻¹ (0.395). The least HI ratio (0.328) was observed in SC Semeki variety when it was planted at the density of 700 K plants ha⁻¹ (Table 6).

At an interaction of G x T, Lukanga had recorded significant values of HI at all stages of thinning, although the highest was recorded at T_1 (0.396). Mwembeshi only revealed a significant HI value when it was thinned at R1.

Significant effects were detected at the D x T interaction. Density 500 K plants ha⁻¹ yielded significant values of HI at all the thinning stages except for T_2 interaction. The highest ratio was achieved when density 500 K plants ha⁻¹ was thinned at V0 (D₁ x T₁). Density 600 K plants ha⁻¹ only yielded significant interactive effects when thinning was conducted at T₃. Significant differences were noted at T₁ and T₃ for density 700 K plants ha⁻¹.

The three- way interaction revealed some significant effects (Figure 11). Lukanga recorded its highest HI in density 600 K plants ha⁻¹ and thinning at R1. Mwembeshi and SC Semeki had their highest ratios achieved in density 500 K plants ha⁻¹, thinned at R1. There were observed tendencies for lower planting densities to increase the HI when thinning was done early. Delayed thinning resulted in reduction in the HI particularly for Lukanga and SC Semeki varieties.

		Parameters				
Factor	Grains	Pods per	100-Grains	Yield	HI (%)	
	per pod	plant	weight (g)	$(ton. ha^{-1})$	ПІ (%)	
$G_1 \ge D_1$	2.36	12.49	13.19	2.42	37.25	
$G_1 \ge D_2$	2.49	12.41	13.11	2.30	39.51	
$G_1 \ge D_3$	2.39	11.81	13.5	2.56	37.22	
$G_2 \ge D_1$	2.38	15.55	13.5	1.83	36.27	
$G_2 \ge D_2$	2.24	16.96	13.6	2.09	33.90	
$G_2 \ge D_3$	2.35	16.55	13.57	1.93	35.57	
$G_3 \ge D_1$	2.29	15.11	13.18	1.25	35.88	
G ₃ x D ₂	2.29	11.80	13.51	1.27	34.98	
G ₃ x D ₃	2.15	14.27	13.58	1.00	32.80	
Lsd(G x D)	0.14	4.57	0.55	0.36	3.40	
$G_1 \ge T_1$	2.38	12.62	13.10	2.68	39.62	
$G_1 \ge T_2$	2.38	11.88	13.40	2.26	37.50	
$G_1 \ge T_3$	2.48	13.43	13.12	2.32	38.05	
$G_1 \ge T_4$	2.40	11.02	13.45	2.44	36.80	
$G_2 \ge T_1$	2.42	17.23	13.46	2.14	35.07	
$G_2 \ge T_2$	2.27	15.32	13.19	1.75	33.69	
G ₂ x T ₃	2.35	17.05	13.83	1.94	36.91	
$G_2 \ge T_4$	2.25	15.82	13.74	1.98	35.30	
$G_3 \ge T_1$	2.23	13.50	13.24	1.25	36.17	
G3 x T2	2.28	13.72	13.73	1.58	32.39	
G ₃ x T ₃	2.32	13.93	13.19	0.74	35.43	
G3 x T4	2.13	13.77	13.53	1.12	34.23	
Lsd(G x T)	0.18	4.94	0.68	0.35	4.07	
$D_1 \ge T_1$	2.33	14.43	13.36	1.90	38.33	
$D_1 \ge T_2$	2.40	14.77	13.45	2.08	34.53	
$D_1 \ge T_3$	2.40	13.05	12.99	1.90	36.29	
$D_1 \ge T_4$	2.23	15.28	13.36	1.45	36.71	
$D_2 \ge T_1$	2.37	13.95	13.42	2.51	35.90	
$D_2 \ge T_2$	2.30	13.50	13.19	1.42	35.83	
$D_2 \ge T_3$	2.40	16.85	13.83	1.67	37.03	
$D_2 \ge T_4$	2.28	10.60	13.18	1.95	35.75	
$D_3 \ge T_1$	2.33	14.97	13.03	1.66	36.63	
$D_3 \ge T_2$	2.23	12.65	13.68	2.09	33.21	
D ₃ x T ₃	2.35	14.52	13.32	1.42	37.08	
D ₃ x T ₄	2.27	14.72	14.17	2.15	33.87	
Lsd(D x T)	0.17	4.17	0.61	0.35	3.06	

Table 6: Two-way interactive effects of genotype, planting density and phenological stage at thinning on reproductive parameters of soybeans (*Glycine max*).

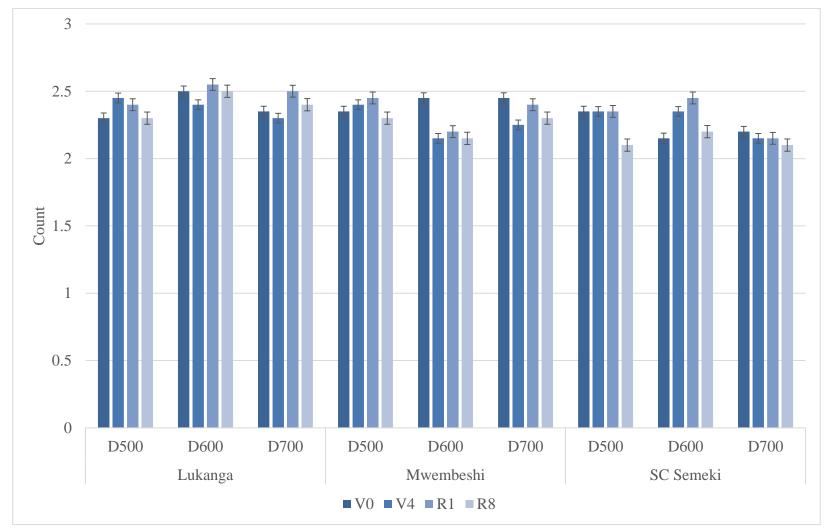


Figure 7: Number of grains per pod for different soy varieties grown under various plant densities and thinned at different phenological stages. Vertical bars are standard errors.

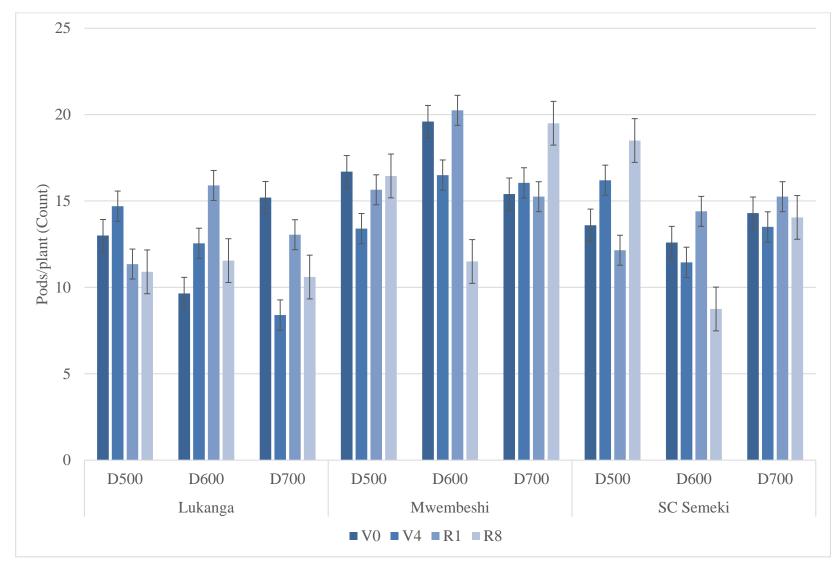


Figure 8: Number of pods per plant of different soy varieties grown under various plant densities and thinned at different phenological stages. Vertical bars are standard errors

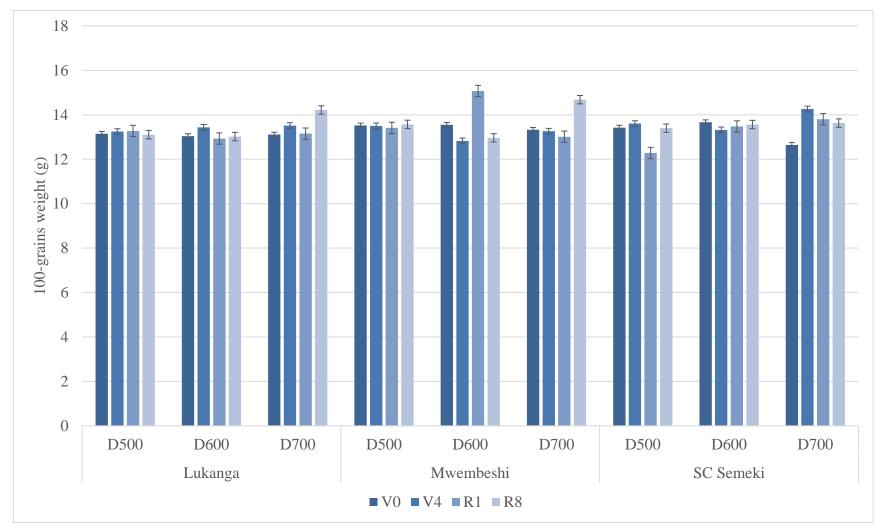


Figure 9: 100-Grains weight of different soy varieties grown under various plant densities and thinned at different phenological stages. Vertical bars are standard errors

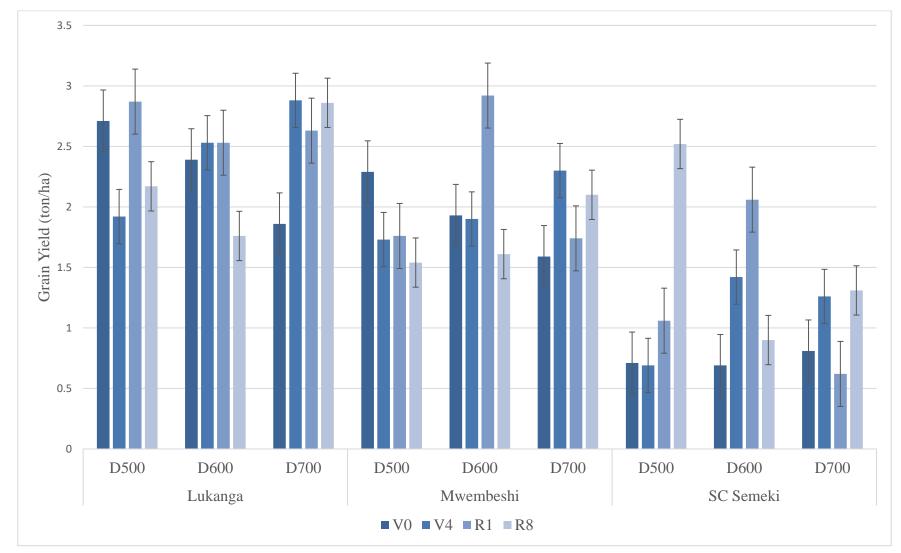


Figure 10: Grain yield of different soybean varieties grown under various plant densities and thinned at different phenological stages. Vertical bars are standard errors

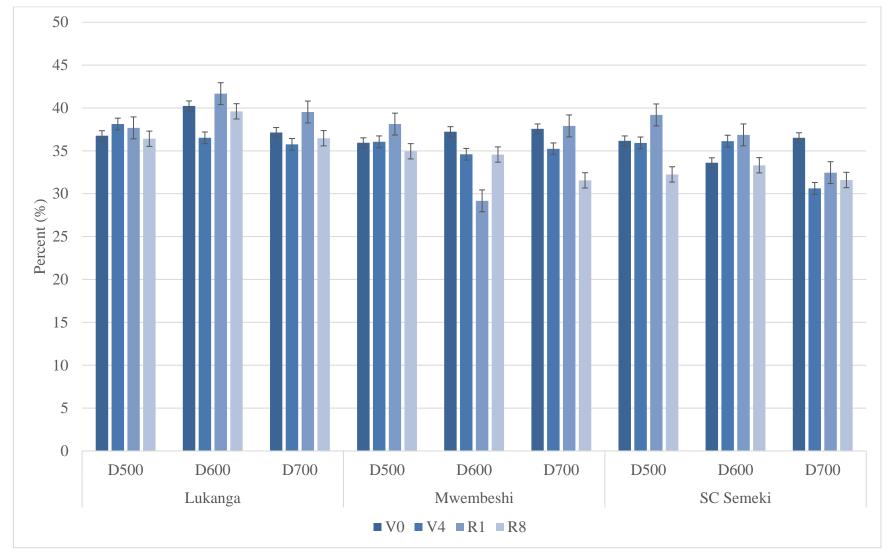


Figure 11: Harvest Index (HI) ratio of different soybean varieties grown under various plant densities and thinned at different phenological stages. Vertical bars are standard errors

4.4. Correlation

Table 7 shows that plant height at R8 (cm) was moderately positively correlated (r = 0.59) to plant height at R1 (cm) parameters. Biomass weight (ton. ha⁻¹) at R8 and grain yield were weakly correlated to plant height at R1 with correlation values of 0.44 and 0.38 respectively. Plant height at R8 had moderately positive correlation with biomass weight (ton. ha⁻¹) at R8 (r = 0.53) and with the number of pods per plant (r = 0.55). Grain yield was weakly positively correlated to biomass weight (ton. ha⁻¹) at R8 (r = 0.43). There was a moderately positive correlation (r = 0.53) between the number of grains per pod and harvest index. Harvest index was strongly negatively correlated (r = -0.88) to grain weight.

4.5. Multiple regression

The set dependent variable against morphophysiological parameters was grain yield (Table 8). Slight and significant contributions to the total variation was accounted for from the observed independent variables in our investigation. Parameter estimates revealed that biomass at R8, plant height at R8 and the number of pods per plant were highly significant and significant influence on grain yield. Additional parameters and their respective interactions to the model did not result in further significant effects on grain yield. Hence the generated grain yield prediction model was:

$$Yield = \sum ((-1.76 + 0.0194 PHR1 + 0.0286 PHR8 - 0.0897 BMV4 + 0.0755 BMR8 - 0.00377 R: S - (0.213 G/P) - (0.0238 P/P) + 0.12 G.wt + 0.0351 HI))$$

Where;

PH R1	:	Plant Height at R1 (cm)
PH R8	:	Plant Height at R8 (cm)
BM V4	:	Biomass Weight (ton. ha ⁻¹) at R1
BM R8	:	Biomass Weight (ton. ha ⁻¹) at R8
R:S	:	Root to Shoot weight Ratio
G/P	:	Number of Grains per Pod
P/P	:	Number of Pods per Plant
G. wt	:	100 Grain Weight (g)
HI	:	Harvest Index

The percentage variance accounted for was 26.1 and standard error of observations was estimated to be 0.681.

Parameter	Plant Height at R1 (cm)	Plant Height at R8 (cm)	Biomass wt. (ton. ha-1) at R1	Biomass wt. (ton. ha ⁻¹) R8	Root - Shoot wt.	Grains per Pod	Pods per Plant	100 Grain Weight (g)	Grain Yield (ton ha ⁻¹)	Harve st Index
Plant Height at R1 (cm)	-	, <i>, , , , , , , , , , , , , , , , , , </i>							, , , , , , , , , , , , , , , , , , ,	
Plant Height at R8 (cm)	0.5946*	-								
Biomass Weight (ton. ha-1) at R1	-0.0558	-0.1513	-							
Biomass Weight (ton. ha ⁻¹) at R8	0.4402*	0.5275*	-0.1522	-						
Root to Shoot weight Ratio	0.0146	0.1065	0.0107	0.0256	-					
Grains per Pod	0.2855	0.3015	-0.0586	0.2235	-0.0811	-				
Pods per Plant	0.3078	0.5568*	-0.067	0.4207*	0.0621	0.1701	-			
100 Grain Weight (g)	0.0001	0.1648	-0.129	0.1729	-0.0198	-0.089	0.1176	-		
Grain Yield (ton. ha ⁻¹)	0.3819	0.3853	-0.2379	0.4372*	-0.0294	0.2487	0.1224	0.0874	-	
Harvest Index	0.1203	0.0004	0.0671	-0.0393	-0.0287	0.5305*	-0.0305	-0.8808***	0.0508	-

Table 7: Correlation of different Morpho-physiological and yield parameters.

Response variate:	Grain Yield						
Fitted terms:	PH R1, PH R8, BM V4, BM R8, S:H, G/P, P/P, G.wt, HI						
Summary of analysis							
Source	d.f.	S.S.	m.s.	v.r.	F pr.		
Regression	9	27.62	3.068	6.61	<.001		
Residual	134	62.17	0.464				
Total	143	89.79	0.628				
Estimates of parameters							
Parameter	estimate	s.e.	t (134)	t pr.			
Constant	-1.76	2.41	-0.73	0.466	Wald statistic		
PH R1	0.0194	0.0106	1.82	0.071	4.683		
PH R8	0.0286	0.0183	1.56	0.121	0.694		
BM V4	-0.0897	0.0427	-2.1	0.038*	1.925		
BM R8	0.0755	0.0228	3.31	0.001***	3.339		
S:H	-0.00377	0.00827	-0.46	0.649	0.544		
G/P	-0.213	0.754	-0.28	0.778	4.939		
P/P	-0.0238	0.0122	-1.95	0.053*	0.505		
G.wt	0.12	0.165	0.73	0.468	5.254		
HI	0.0351	0.0497	0.71	0.482	4.896		

Table 8: Multiple regression of grain yield on morphophysiological traits in soybeans subjected to different plant density levels and thinned at varying phenological stages

Factor significance; *** highly significant ($p \le 0.001$); **very significant ($p \le 0.01$); * significant ($p \le 0.05$) and ns- nonsignificant.

<u>Key</u>

PH R1 : PH R8 :	Plant Height at R1 (cm) Plant Height at R8 (cm)
BM V4:	Biomass Weight (ton. ha ⁻¹) at R1
BM R8:	Biomass Weight (ton. ha ⁻¹) at R8
R:S :	Root to Shoot weight Ratio
G/P :	Number of Grains per Pod
P/P :	Number of Pods per Plant
G. wt :	100 Grain Weight (g)
HI :	Harvest Index

Chapter 5

5. DISCUSION

The environment under which organisms develop has influence on resulting morphological and physiological characteristics. Due to their sessile nature, plants contend with limitations of their environment which can be a source of stress depending on their degree of application (either as supra or sub optimal). Thus, to survive and remain productive, appropriate functional plasticity responses are inevitable (Bradshaw, 1965; Takahashi and Shinozaki, 2019). Using various signalling pathways such as Reactive Oxygen Species-ROS, phytochromes and an interaction of physiological metabolites with phytohormones at subcellular level, plants invoke morphophysiological plasticity that builds their competitive effectiveness at resource mobilization (Bradshaw, 1965; Wani et al., 2016; Mhamdi and Van Breusegem, 2018; Takahashi and Shinozaki, 2019). Plant density is one of the most pliable factors that can be altered in the row crop growth environment to improve crop production (Wright and Lenssen, 2013). This study investigated phenotypic plasticity in soybeans resulting from altered environment in terms of planting density. Different soybean varieties were subjected to various plant density levels and thinned progressively at different phenological stages to alter the plant density stress duration. The test varieties included indeterminate (Mwembeshi) and determinate (Lukanga and SC Semeki) varieties to evaluate their sensitivity to stress and ability to recover thereof. It is assumed that findings of this study would contribute to the understanding of the effects of inducing stress in row crops through use of supra optimal planting densities and removal thereof at specific phenological stages by crop thinning.

In Zambia, average yield of soybeans under farm conditions is about 2 ton/ha (Chiona *et al.*, 2017) which is comparatively low compared to yield potentials reported by researchers. For instance yield potentials in excess of 3 ton/ha have been reported (SEEDCO, 2015; Afriseed Stewards, 2018; Chigeza *et al.*, 2019). The low yield observed in this study can be attributed in part to poor soil fertility where pH, phosphorous, percentage organic carbon and magnesium were below the critical levels to support good plant growth. In addition, due to initial failure of the crop resulting from drought, the trial had to be replanted, which resulted in late planting.

The results showed that variety had a significant effect on all observed vegetative parameters except for root to shoot weight ratio. Lukanga had the highest plant height (at R1 and R8), biomass (R8) and these were significantly different with that of SC Semeki but not with Mwembeshi variety. The different performance of Lukanga and SC Semeki despite having the same growth characteristics (determinate type) is in contrast with the findings of Mataa and Sichilima, (2019) who found that there were no significant differences in growth characteristics among varieties that had contrasting growth habits (determinate and indeterminate types). These findings indicate that the responses are genotype specific (Tekola *et al.*, 2018). SC Semeki accumulated higher biomass at V4 growth stage than when the same parameter was measured at harvest time (R8). The comparative reduction in biomass accumulation for variety SC Semeki (high at V4-vegetative growth to low at R8-reproductive phase) to the other varieties could be attributed to vegetative allocational plasticity, a functional feature in environmental resource competition (Sultan, 2003).

Biomass and root- shoot ratio is an indicator of the plant's vigour in an environment where interplant competition exist (Yang *et al.*, 2019). Plants with higher biomass accumulation and comparative root mass have advantages in the competition for nutrients and moisture (Craine and Dybzinski, 2013). Plant density did not exert any significant effect on plant height, biomass at V4 and on root to shoot ratio. Only density 600 K plants ha⁻¹ however showed more assimilates being partitioned to biomass (5.70 tons. ha⁻¹) than the other two densities (4.22 tons. ha⁻¹). Our findings on plant height were similar to the observation made by Mellendorf, (2011), who associated the slight decrements/increments between densities to general plant growth due to row spacing and to the decreased/increased access to photosynthetically active radian-PAR (Park and Runkle, 2018). A similar study by Sichilima *et al.*, (2018) showed corresponding findings, where density had no significant effect on plant height.

Thinning time did not exert significant effects on vegetative parameters except for rootshoot ratio thinned at R8. Maintaining the crop without thinning increased the root to shoot ratio. The increased root biomass in relation to above ground biomass can be attributed to the sustained stress. Under stress, it has been suggested that plants invoke photoassimilate allocational plasticity (Murren *et al.*, 2015) which increases photoassimilate partitioning to roots to assist the plant to forage and compete for resources in the root zone (Rondanini *et al.*, 2017). The low yielding variety (SC Semeki) allocated most of the assimilates to vegetative tissues and therefore during the vegetative phase it had comparatively higher biomass. During the reproductive phase SC Semeki had lower biomass whereas the higher yielding varieties accumulated more biomass during the reproductive phase and most of this was directed to the grain. It can therefore be deduced that SC Semeki can better be used as a vegetative or forage genotype (Rondanini *et al.*, 2017)..

Significant variety and thinning interactions were observed with Lukanga in plant height. Treatments that were thinned early showed a slight reduction in plant height compared to late thinning and unthinned treatments. Mataa and Sichilima, (2019) made similar observations in their attempt to determine phenotypic response in soybeans- early density stress relief resulted in lower plant height. This low plant height (or reduced etiolation) may be due to decreased competition for light due to the available space left behind by the thinned-out plants. These findings are in support of the theory that an increase in plant density results in the change of the sink: source relationship to allocate more photoassimilates to support structures of the plant which allows it to be competitive at accessing solar radian (Park *et al.*, 2003; Mellendorf, 2011; Park and Runkle, 2016). Plants have developed mechanisms to sense neighbouring plants using phytochromes and shade avoidance cues to signal for change in resource partitioning to appropriate vegetative structures (Park and Runkle, 2018). Mwembeshi and SC Semeki were unresponsive to the thinning interaction. Hence phenotypic plasticity was operative only in Lukanga variety (Rondanini *et al.*, 2017).

The three- way interaction showed significant effects on some vegetative parameters especially on biomass (at V4). There was an inverse relationship between thinning time and biomass accumulation. Therefore, relieving density stress in the early stages of plant growth resulted in an increase in biomass especially for SC Semeki. This response was in conformity with the finding of Sekimura *et al.*, (2000) who observed that plants adjusted their morphological features in response to the closeness of their neighbours. The same response was observed by Sichilima *et al.*, (2018).

The number of grains per pod, number of pods per plant and grain weight are an important determinant of the final grain yield (Pereira-Flores and Justino, 2019). This parameter is among the five critical yield components described soybeans (Pereira-Flores and Justino, 2019). However, it is important to note that pod length and number of seeds per pod show little variation in soybeans and what is more critical is how many of these pods have filled

seeds (empty pods) and seed mass. Lukanga had a significantly higher number of grains per pod while Mwembeshi had a higher number of pods per plant. This led the two varieties to record significantly higher grain yields. The number of grains per pod contributed more to grain yield than the number of pods per plant because Lukanga which scored better at number of grains per pod also recorded the highest grain yield and the only significant value at harvest index.

A possible explanation why Mwembeshi did not have significantly high harvest index value could be that although it more pods most of these pods did not have seeds-a typical occurrence in indeterminate varieties. However, under optimal conditions these varieties are able to form more pods and ultimately translate into higher yield. This response is in conformity with the established theory that modular organisms have compensatory capabilities for any adverse shocks faced during plant growth (Murren *et al.*, 2015). Agudamu *et al.*, (2016) concluded that modular plants have a longer period over which they have to adapt to the environment through branch, leaf area or pod number development. Hence indeterminate varieties continue to grow post their juvenile stage, to which fact they owe their elasticity (Murren *et al.*, 2015).

Soybean has been noted to exhibit classical vegetative plasticity and therefore the plants make adjustments in morphological parametres to ensure stability of yield under a range of plant densities. Therefore, plant density was seen to have little or no effect on reproductive and vegetative parametres that were measured.

Thinning affected root: shoot ratios, number of grains per pod and the overall grain yield. Thinning early showed significant effects on grain yield and harvest index. Treatments that were thinned at planting recorded higher grain yields and harvest index. Relieving the imposed density stress after the plants have developed the competitive capacities for environmental resources in the early phenological stages allowed the soybean plants to change the sinks into reproductive structures such as number of pods per plant (Mellendorf, 2011).

Varietal effect exerted stronger influences on the significant differences observed in the grain yield and. In terms of yield components, the number of grains per pod had a larger contribution to grain yield and HI for Lukanga. While for Mwembeshi, the most contributions to grain yield was attributed to its high number of pods per plant. The source of reproductive plasticity can be from any of the yield components. Rondanini *et al.*, (2017) observed the reproductive plasticity in spring rapeseed to be determined by floral branching.

Based on the results obtained above, we postulate that Lukanga, a determinate variety exhibited reproductive plasticity mainly due to the changes in the number of grains per pod. Mwembeshi on the other hand had its reproductive plasticity caused by the number of pods per plant.

For Lukanga, thinning early generally resulted in higher grain yield and HI than late thinning. hence the degree and duration of the density stress affected varieties differently. This phenomenon was also observed by (Sichilima, Mataa and Mweetwa, 2018). The trend, however, was not discernible in Mwembeshi and SC Semeki. Hence it was supposed that the observed trend was a genotype specific effect and could not be associated to stress duration.

There were observed tendencies for lower planting densities to increase the HI when thinning was done early. Similar results were observed by Shamsi and Kobraee, (2011) that HI reduced with increase in plant density. Delayed thinning resulted in reduction in the HI particularly for Lukanga and SC Semeki varieties. This may be due to their determinate growth type nature. Despite not detecting differences due to competition relief in small cohorts Mellendorf, (2011) observed that large differences were apparent in the high plant density, where cohorts relieved of competition at V3 increased HI by 7.8 % compared to competition relief at R4 where HI was reduced by 10 %. It was therefore generalised that reproductive plasticity was at play when varieties interacted with density and thinning time.

In terms of grain yield, Mwembeshi- an indeterminate variety recorded the highest grain yield (2.92 tons ha⁻¹) when it was thinned at R1. This higher yield recorded for Mwembeshi could be due to its indeterminate growth type which have longer reproductive plasticity adjustment period compared to determinate varieties (Lukanga and SC Semeki). The main contributor to grain yield for Mwembeshi was number of pods per plant. Carpenter and Board, (1997) showed that the increase in yield was due to the increase in the number of pods per plant, an outcome that is in conformity with the present finding of our study. Pods emerge at branch nodes in plants (Carpenter and Board, 1997; Agudamu, Yoshihira and Shiraiwa, 2016). We did not determine the number of branches per plant but based on the high number of pods per plant, the shorter stature of the plants and significant values on biomass at R8, it can be speculated that Mwembeshi branched more than the other varieties. In growth environments where environmental perturbations are likely to impose longer stress duration, it would be safer to plant an indeterminate variety due to their mode of response to stress duration as demonstrated in this investigation.

Crop yield is a product of several crop and non-crop characters that exert their influence on the crop simultaneously. The degree of association between crop characters or morphological traits provides useful information on the performance of the crop (Gomez and Gomez, 1984). The positive inter component correlation observed in our investigation assert that phenotypic plasticity was at play in the tested varieties. For instance, a positive, though weak correlation between grain yield and biomass weight (r = 0.43) may suggest that larger plants tend to have higher yield. Mellendorf, (2011) noted that treatments that had high biomass were also reported to have a high number of branches, high number of pods per plant and higher grain yield. Biomass was found to be highly correlated to the plants' ability to forage for soil resources particularly N uptake, an important element in formation of assimilates that can be partitioning into grain yield and associated components (Ciampitti and Vyn, 2011). Plant height was also noted to be positively correlated to biomass ($r = 0.53^*$), signifying the density effect on biomass accumulation and subsequent grain yield.

A close relationship between grain yield and biomass was observed particularly at R8. Other parameters that showed significant contributions to variations in grain yield were plant height and number of pods per plant. The yield prediction model developed in this study however is restricted to the tested varieties and environmental conditions similar to the study site. It can be deduced therefore that one of the main contributors to grain yield is biomass. Similar findings were arrived at by Duncan, (1986) who associated higher biomass to higher grain yields and Sichilima *et al.*, (2018) also concluded that biomass had the strongest effect on yield. In summary of the three parametres variety and thinning time exerted more effects on plant developmental processes and yield compared to density which exerted very little influence.

Chapter 6

6. CONCLUSION AND RECOMMENDATIONS

Crop development and yield in soybeans was influenced by variety and thinning time. Variations in plant density and thinning at different phenological stages exhibited differences in their effects on morphophysiological parameters for the tested soybean varieties. Grain yield and biomass weight was highly influenced by variety. Plant density did not have exert significant effects on most observed vegetative and reproductive parameters. However, when density interacted with thinning time, it resulted significant effects on biomass and grain yield. Thinning time influenced grain yield significantly. Lukanga and SC Semeki are both determinate varieties but seem to follow different pathways in their response to density stress relief at different stages of growth. Lukanga exhibited reproductive plasticity while SC Semeki demonstrated vegetative plasticity. As for Mwembeshi, an indeterminate type of variety showed phenotypic elasticity to the effects of density stress relief. Vegetative plasticity however did not result in increased grain yield for SC Semeki. Early crop thinning induced reproductive plasticity and had an influence on grain yield in Lukanga. Thinning up to R1 stage resulted in the adjustment of the morphological features of the plant, particularly number of pods per plant which can be extrapolated to the number of branches per plant in Mwembeshi.

With the advent of climate change where rainfall and other climatic factors are increasingly becoming unpredictable, planting soybean varieties that exhibit phenotypic elasticity may be advantageous. Owing to their capacity to adjust their yield components over longer pheno phase. In developing new soybean varieties therefore, plasticity or elasticity characteristics should be considered. Varieties, that exhibit vegetative plasticity such as observed in SC Semeki can be recommended as a fodder and or a bioenergy crop due to their early high biomass accumulation capacity and less grain yield.

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Appendix



Figure 12: Location Map of Mansa Research Station

Annex 1: ANOVA Tables

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Replication stratum	3	2331.34	777.11	11.34	
Replication.Variety stratum					
Variety	2	732.58	366.29	5.35	0.046
Residual	6	411.05	68.51	1.42	
Replication.Variety.Density stratum					
Density	2	113.36	56.68	1.17	0.333
Variety.Density	4	129.61	32.4	0.67	0.622
Residual	18	871.26	48.4	2.56	
Replication.Variety.Density.Thin_Time stratum					
Thin_Time	3	52.79	17.6	0.93	0.429
Variety.Thin_Time	6	134.61	22.43	1.19	0.321
Density.Thin_Time	6	80.61	13.43	0.71	0.642
Variety.Density.Thin_Time	12	545	45.42	2.4	0.01
Residual	81	1530.16	18.89		
Total	143	6932.37			

Table 9: ANOVA Table for Plant Height in cm as measured at the onset of the R1 growth

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Replication stratum	3	703.31	234.44	13.05	
Replication.Variety stratum					
Variety	2	196.32	98.16	5.46	0.045
Residual	6	107.79	17.96	0.6	
Replication.Variety.Density stratum					
Density	2	33.97	16.99	0.56	0.579
Variety.Density	4	90.64	22.66	0.75	0.57
Residual	18	542.34	30.13	2.28	
Replication.Variety.Density.Thin_Time stratum					
Thin_Time	3	3.39	1.13	0.09	0.968
Variety.Thin_Time	6	19.84	3.31	0.25	0.958
Density.Thin_Time	6	189.41	31.57	2.39	0.036
Variety.Density.Thin_Time	12	257.53	21.46	1.62	0.101
Residual	81	1071.02	13.22		
Total	143	3215.57			

Table 10: ANOVA Table for Plant Height in cm as measured at R8 growth

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Replication stratum	3	6.348	2.116	2.7	
Replication.Variety stratum					
Variety	2	28.111	14.056	17.91	0.003
Residual	6	4.709	0.785	0.88	
Replication.Variety.Density stratum					
Density	2	2.755	1.377	1.55	0.24
Variety.Density	4	6.089	1.522	1.71	0.192
Residual	18	16.029	0.89	0.76	
Replication.Variety.Density.	Thin_Time st	ratum			
Thin_Time	3	1.852	0.617	0.53	0.665
Variety.Thin_Time	6	27.52	4.587	3.91	0.002
Density.Thin_Time	6	28.558	4.76	4.06	0.001
Variety.Density.Thin_Time	12	51.603	4.3	3.67	<.001
Residual	81	95.018	1.173		
Total	143	268.592			

 Table 11: ANOVA Table for Dry Biomass Weight in tonnes per hectare as measured at the onset of the R1 growth stage

Source of variation	d.f. s.s.		m.s.	v.r.	F pr.
Replication stratum	3	108.084	36.028	2.64	
Replication.Variety stratum					
Variety	2	239.977	119.99	8.79	0.016
Residual	6	81.912	13.652	1.24	
Replication.Variety.Density stratum					
Density	2	69.872	34.936	3.17	0.066
Variety.Density	4	74.559	18.64	1.69	0.196
Residual	18	198.366	11.02	2.14	
Replication.Variety.Density.Thin_Time stratum					
Thin_Time	3	15.752	5.251	1.02	0.388
Variety.Thin_Time	6	20.974	3.496	0.68	0.666
Density.Thin_Time	6	58.002	9.667	1.88	0.094
Variety.Density.Thin_Time	12	86.053	7.171	1.39	0.186
Residual	81	416.443	5.141		
Total	143	1369.99			

Table 12: ANOVA Table for Dry Biomass Weight in tons per hectare as measured at R8
growth stage

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Replication stratum	3	32.1	10.7	0.36	
Replication.Variety stratum					
Variety	2	139.6	69.8	2.37	0.174
Residual	6	176.45	29.41	0.39	
Replication.Variety.Density stratum					
Density	2	189.03	94.51	1.26	0.307
Variety.Density	4	276.52	69.13	0.92	0.472
Residual	18	1348.28	74.9	1.66	
Replication.Variety.Density.Thin_Time stratum					
Thin_Time	3	324.33	108.11	2.4	0.074
Variety.Thin_Time	6	184.13	30.69	0.68	0.665
Density.Thin_Time	6	249.74	41.62	0.92	0.482
Variety.Density.Thin_Time	12	472.14	39.35	0.87	0.576
Residual	81	3647.44	45.03		
Total	143	7039.76			

Table 13: ANOVA Table for Root to Shoot dry weight Ratio as measured at the onset of the R1 growth stage

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Replication stratum	3	3.39	1.13	26.25	
Replication.Variety stratum					
Variety	2	0.70167	0.35083	8.15	0.019
Residual	6	0.25833	0.04306	1.47	
Replication.Variety.Density stratum					
Density	2	0.06167	0.03083	1.05	0.369
Variety.Density	4	0.45167	0.11292	3.86	0.02
Residual	18	0.52667	0.02926	0.56	
Replication.Variety.Density.Thin_Time stratu	m				
Thin_Time	3	0.29	0.09667	1.86	0.143
Variety.Thin_Time	6	0.23833	0.03972	0.77	0.599
Density.Thin_Time	6	0.15167	0.02528	0.49	0.816
Variety.Density.Thin_Time	12	0.355	0.02958	0.57	0.86
Residual	81	4.205	0.05191		
Total	143	10.63			

Table 14: ANOVA Table for Number of Grains per Pod as measured at harvest-R8 growth stage

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Replication stratum	3	600.53	200.18	3.33	
Replication.Variety stratum					
Variety	2	417	208.5	3.47	0.1
Residual	6	360.38	60.06	2.3	
Replication.Variety.Density stratum					
Density	2	11.2	5.6	0.21	0.809
Variety.Density	4	104.99	26.25	1	0.431
Residual	18	470.58	26.14	0.98	
Replication.Variety.Density.Thin_Time stratum					
Thin_Time	3	41.54	13.85	0.52	0.669
Variety.Thin_Time	6	29.36	4.89	0.18	0.98
Density.Thin_Time	6	267.18	44.53	1.68	0.137
Variety.Density.Thin_Time	12	319.31	26.61	1	0.454
Residual	81	2150.08	26.54		
Total	143	4772.15			

 Table 15: ANOVA Table for Number of Pods per Plant as measured at harvest-R8 growth stage

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Replication stratum	3	618.1762	206.0587	237.92	
Replication.Variety stratum					
Variety	2	2.0309	1.0155	1.17	0.372
Residual	6	5.1965	0.8661	2.28	
Replication.Variety.Density stratum					
Density	2	1.6528	0.8264	2.18	0.142
Variety.Density	4	1.2997	0.3249	0.86	0.509
Residual	18	6.83	0.3794	0.61	
Replication.Variety.Density.Thin_Time	stratum				
Thin_Time	3	1.7121	0.5707	0.91	0.44
Variety.Thin_Time	6	4.7788	0.7965	1.27	0.28
Density.Thin_Time	6	11.8754	1.9792	3.16	0.008
Variety.Density.Thin_Time	12	15.2306	1.2692	2.03	0.032
Residual	81	50.7573	0.6266		
Total	143	719.5402			

Table 16: ANOVA Table for 100 Grain Weight in grams as measured at harvest-R8 growth stage

Source of variation	d.f.	S.S.	m.s.	v.r.	F pr.
Replication stratum	3	2.517	0.839	3.67	
Replication.Variety stratum					
Variety	2	38.5279	19.264	84.28	<.001
Residual	6	1.3715	0.2286	0.93	
Replication.Variety.Density stratum					
Density	2	0.0996	0.0498	0.2	0.819
Variety.Density	4	1.6463	0.4116	1.67	0.201
Residual	18	4.4443	0.2469	1.54	
Replication.Variety.Density.Thin_Time stratum					
Thin_Time	3	2.3029	0.7676	4.78	0.004
Variety.Thin_Time	6	4.1932	0.6989	4.35	<.001
Density.Thin_Time	6	12.4202	2.07	12.88	<.001
Variety.Density.Thin_Time	12	9.2434	0.7703	4.79	<.001
Residual	81	13.0201	0.1607		
Total	143	89.7864			

Table 17: ANOVA Table for Grain Yield as measured at harvest-R8 growth stage

Source of variation	d.f. s.s.		m.s.	v.r.	F pr.
Replication stratum	3	7942.21	2647.4	61.47	
Replication.Variety stratum					
Variety	2	317.65	158.83	3.69	0.09
Residual	6	258.42	43.07	6.95	
Replication.Variety.Density stratum					
Density	2	41.36	20.68	3.33	0.059
Variety.Density	4	141.43	35.36	5.7	0.004
Residual	18	111.62	6.2	0.37	
Replication.Variety.Density.Thin_Time stratum					
Thin_Time	3	144.28	48.09	2.84	0.043
Variety.Thin_Time	6	68.25	11.38	0.67	0.673
Density.Thin_Time	6	91.85	15.31	0.9	0.497
Variety.Density.Thin_Time	12	269.06	22.42	1.32	0.222
Residual	81	1373.44	16.96		
Total	143	10759.6			

Table 18: ANOVA Table for Harvest Index Ratio as measured at harvest-R8 growth stage