

Nutrient deficiencies and soil fertility constraints
for common bean (*Phaseolus vulgaris* L.) production
in the Usambara Mountains, northern Tanzania

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1. Summary

Common bean (*Phaseolus vulgaris* L.) is the most important food grain legume crop in Tanzania. It is widely grown as a subsistence crop by smallholder farmers to provide an important source of dietary protein. Despite the relatively high importance and demand, the yields of common bean have remained low, especially under resource-poor farmer conditions and low soil fertility. This thesis research was conducted within the N2Africa framework in Tanzania, by studying the current nutrient deficiency problems for common bean production in the West-Usambara Mountains (Lushoto region, northern Tanzania). On-farm fertilizer and inoculation trials were carried out at nine farmer field sites in the short rainy season (vuli) from November 2013 until February 2014. The field trials were designed as factorial experiments with two different fertilizers and rhizobia inoculation as the main factors; phosphorus (26 kg P ha⁻¹ as triple superphosphate), potassium (25 kg K ha⁻¹ as muriate of potash) and rhizobia inoculant mixture. Nitrogen (25 kg N ha⁻¹ as calcium ammonium nitrate) was applied in an additional treatment together with P and K to analyse the effect of N fertilizer without inoculation. Nutrient limitations were identified with the use of soil and leaf sample analysis. Bean leaf samples were analysed for macro- and micronutrients, and the Diagnosis and Recommendation Integrated System (DRIS) was applied to rank nutrients according to their degree of limitation to bean production. Furthermore this study looked to the possible interaction between nutrient deficiencies and field management.

The combination of different analyses indicated K and P deficiency as a major constraint for bean production in the Usambara Mountains. Where soil analysis indicated deficiencies of the nutrients K and P and partly N, Ca and Mg. Growth and yield results revealed positive responses to fertilisers P and K. Analysis of bean leaf tissue indicated deficient nutrient concentrations levels for P, K, N and Zn when compared with critical nutrient concentration ranges. Application of P and K (and partially N) fertilisers increased leaf concentrations of the respective elements but depressed the concentrations of Ca, Mg, Cu and Zn. Ranking the obtained leaf nutrient concentrations with the DRIS approach, showed consistent results for nutrient deficiencies of P and K. Severe K deficiency became also visible at some fields through chlorotic and necrotic leaf symptoms. *Rhizobium* inoculation used in the experimental trials, gave poor or mixed results.

The results indicated that, besides the use of high value inputs, other factors played a major role in the determination of final bean yield. A lack of rainfall decreased nutrient uptake from the soil and plant growth at some fields. The farmers involved in the experimental trials in the Usambara region were not used to apply any type of manure and or rhizobia inoculant to beans. Declining soil fertility together with an increase in human population throughout the Usambara region, highlight the need to support farmers with high quality inputs and inform them on how to manage their (bean) production systems in an adequate way.

2. Introduction

2.1 Background

2.1.1 Research framework

Approximately 84% of the population of Tanzania is employed in the agricultural sector and crop production is important for both national food supply and foreign exchange through export commodities (Ndakidemi and Semoka, 2006), (Table 1, FAOSTAT, 2013). Common bean (*Phaseolus vulgaris* L.) is the most important food grain legume crop in Tanzania and the country ranks among the top 20 in global bean production (Hillocks *et al.* 2006; Ndakidemi *et al.* 2006). However, most of Tanzanian bean production is carried out by smallholder farmers for their own consumption, with an average 20% surplus being marketed (Hillocks *et al.*, 2006). Bean is widely grown in the Northern highlands of Tanzania (mid to high altitude areas with more reliable rainfall and cooler temperatures) as a subsistence crop, often intercropped with maize by local farmers to provide an important source for both dietary protein and carbohydrates in human diets (Amijee and Giller, 1998; Hillocks *et al.* 2006). In these highlands each year consists of two main growing seasons when rainfall is adequate; the long rains (*masika*) from March to June and the short rains (*vuli*) from November to January, where common bean is grown in both seasons (Smithson *et al.* 1993). In addition to the high dietary value, legumes have the ability to develop root nodules in symbiosis with rhizobia to fix 'freely' available atmospheric dinitrogen gas (N₂) and converting it to a biologically useful, combined form of N - ammonia, for use by the host plant or by associated or subsequent crops (Graham and Vance, 2003; Giller, 2001). Despite the relatively high importance and demand for grain legumes, the yields of common bean have remained low, especially under resource-poor farmer conditions, on average ranging from 700-900 kg ha⁻¹ in Tanzania over the last five years (Table 1, FAOSTAT, 2013) compared with a potential yield of about 1500-3000 kg ha⁻¹ (Hillocks *et al.* 2006). The most important constraints for bean production and N₂ fixation in Tanzania are: use of low quality seed and local bean cultivars prone to pest and diseases, nutrient limitations and low soil fertility, drought, soil acidity and poor crop management (Graham and Vance, 2003; Hillocks *et al.* 2006; Ndakidemi *et al.* 2006).

Table 2.1 Characteristics of dry bean production in Tanzania from 2008 to 2012 (FAOSTAT, 2013)

	2008	2009	2010	2011	2012
Area harvested (ha)	749 540	868 310	1 208 690	737 661	1 330 000*
Production (t)	570 750	773 720	867 530	675 948	1 199 267
Average yield (kg/ha)	762	891	725	916	902
Import quantity (t)	698	4 097*	468	832	n.a.
Export quantity (t)	*2 844	11 235	16 064	11 944	n.a.

* Unofficial figure (FAO estimate)

The Lushoto district is an important bean growing area in the West Usambara Mountains in northeastern Tanzania due to its favourable climatic conditions (Mbagal-Semgalawe and Folmer, 2000). However, In this highly populated area most farms are managed by relatively poor farmers, with few resources to purchase (in)organic fertilizer inputs and many of the production areas are located on slopes which are intensively cultivated and highly degraded (Ndakidemi and Semoka, 2006). Nutrient limitations in bean production in the Usambara Mountains have been studied for a long time already (Giller *et al.* 1989; Smithson *et al.* 1993; Amijee and Giller, 1998; Giller *et al.* 1998;

Ndakidemi and Semoka, 2006). Smithson et al. (1993) observed leaf chlorosis symptoms, referred to as 'Usambara mottle', as an expression of K deficiency and they pointed at the substantial benefits of phosphorus (P) and potassium (K) fertilizers in yield of common bean and the use of nitrogen (N) fertilizer as a relatively small starter dose to stimulate initial growth. The results confirmed the need for on-farm experiments in the area to specify the nutrient limitations for common bean in the Usambara region to be able to support sustainable soil fertility management in future.

2.1.2 Approach and aim of the study

The overall aim of this thesis research was to study the current nutrient deficiency problems for common bean production in the West-Usambara Mountains, to support soil fertility management in future. Fertilizer (N, P and K) and inoculation trials were carried out at nine locations with the use of Lymangu 90 in the short-rainy season in 2013. Chemical and physical soil analysis was conducted prior to planting and plant leaf nutrient concentrations were determined during growth. Plant leaf nutrient concentration results were compared to critical nutrient levels available from literature and the Diagnosis and Recommendation Integrated System (DRIS) methodology was applied to rank the nutrients according to their degree of limitation to bean production. Similar measurements were conducted at ten additional farmer fields in the region, where farmers managed bean production themselves. Furthermore this study looked to the possible interaction between nutrient deficiencies and field management (land use and input use in the past) and field type (soil properties, microclimate, slope and height) with the use of additional data analyses and interview sessions with the farmers involved in the fertilizer field trials.

2.1.3 N2Africa in (North) Tanzania

The N2Africa project runs since 2009 in several African countries as a large scale, science-based project with the overall objective of "Putting nitrogen fixation to work for smallholder farmers in Africa". With the start of the second phase on the first of January 2014, the project will run at least for five more years funded by the Bill & Melinda Gates Foundation and led by Wageningen University together with the International Institute of Tropical Agriculture (IITA) and the International Livestock Research Institute (ILRI). Tanzania is, next to Ghana, Nigeria, Ethiopia and Uganda, one out of five focus countries. At the official project launch in Dar es Salaam on 19-20 February 2014 the director for Research and Development in the Ministry of Agriculture, Food Security and Cooperatives from Tanzania, Dr Fidelis Myaka said: "Despite the obvious benefit of legumes to Tanzanian food security, employment, and even contribution to GDP, the productivity is low and legume yields are far below their potential" (Baijukya, 2014). Main legumes N2Africa will work on in Tanzania are common bean, soybean and groundnut with the focus niches for common bean in the Northern Highlands (Lushoto, Hai, Kilimanjaro and Rural). One of the potentials for change in growing bean in those regions, indicated by N2Africa, are soil fertility problems, next to pest and diseases, improved agronomy and varieties, with an overall impact on production of about 30%. N2Africa builds on the ($G_L \times G_R$) \times E \times M framework to improve legume technologies. In which: G_L = Legume genotype, G_R = Rhizobium strain, E = Environment and M = Management. Research on limiting nutrients and effective fertilizer recommendations is one of the activities to improve agricultural management. Field trials including inoculants, P, K and other limiting nutrients as treatments are conducted to be tested as relevant technologies (N2Africa, 2013).

2.2 Problem analysis

2.2.1 Soil fertility constraints

Geography and climate

To be able to unravel soil fertility we first need to take a closer look to the so called soil-forming factors, like; (palaeo-)climate, parental materials and vegetation (Stoorvogel *et al.* 1993). The West-Usambara mountains are part of the Eastern Arc Mountains (EAM) stretching from the Taita Hills in south-eastern Kenya to the Udzungwa Mountains in south-central Tanzania. The chain of mountains was uplifted by faulting during different periods, at least 30 million years ago and consists of Precambrian basement rocks (Mumbi *et al.*, 2008). The climate in the EAM is mainly influenced by the Indian Ocean and the passage of the Intertropical Convergence Zone (ITCZ). The mountains contain a very diverse and unique mixture of tropical habitats as they have probably been ecologically isolated since the Miocene (Hamilton, 1982). The (south-)eastern slopes historically supported a continuous forest cover due to the somewhat wetter climate (facing the Indian Ocean), while the (north-)western slopes were prone to a relatively drier climate and thereby supported deciduous woodland at lower elevations and evergreen coniferous forest at higher elevations (Burgess *et al.* 2007). On the top plateaus of the mountains chain, tall evergreen forest could be found, created by the continuous fog over the highlands during the night (Burgess *et al.* 2007; Mumbi *et al.* 2008). Soil under natural vegetation can be seen as being in a virtual steady state and from the moment of a land use change onwards this state can no longer be maintained (Stoorvogel *et al.* 1993). As a consequence, soil fertility can become prone to degradation at a rate dependent on cropping intensity and land management, with declining soil organic matter levels, leaching of nutrients and erosion (Stoorvogel *et al.* 1993).

Human impact

Up to the beginning of the 18th century most of the north-eastern mountains in Tanzania were covered with natural vegetation and agriculture only took place at small areas. Farmers made use of shifting cultivation and fallow practices to maintain soil fertility (Mbagal-Semgalawe and Folmer, 2000). A variety of crops was grown including: bananas, beans, sweet potatoes, taro, pumpkin, maize, rice, millet and yam (Huijzendveld, 2008). From the 20th century onwards the human population started increasing rapidly, with a major part of the area suitable for agriculture brought into cultivation by 1936 as a result. Land scarcity became the major problem in the mountainous areas and the attention for soil conservation practices faded away (Mbagal-Semgalawe and Folmer, 2000). In those years Tanzania was led by the German and British colonial governments which also contributed to ongoing land pressure as considerable areas in the Usambara Mountains were used to establish coffee and tea plantations. As a response and to sustain their livelihood, farmers diverted to the valley bottoms, lowlands, steep slopes and even forests and wetlands for crop cultivation, livestock grazing and settlement. Those practices together with socio-economic factors (e.g. traditional values and economic policies) led to increasing soil degradation (Figure 2.1). The combination of efforts initiated by farmers (mulching, crop rotations, intercropping, increase of manure use efficiency and irrigation furrows) together with activities directed by the colonial government (research programmes, advisory services, physical measures and demonstration plots) made it possible to improve soil productivity again from 1930 onwards. The British introduced the Land Usage Schemes in 1947 in both the Usambara and Pare mountains, with the focus on controlling land degradation and ensuring sustainable cultivation measures. The regulations were

strengthened by the implementation of laws and controlling institutions, which led to heavy resistance by the local people until the Land Usage Schemes collapsed in 1955 (Mbagal-Semgalawe and Folmer, 2000). History repeated itself and after independence in 1961 there was minimum attention to soil conservation measures and much more emphasis was put on new technologies introduced from developed countries; breeding for new crop varieties (cash crops), use of chemical fertilizers and pesticides. To 'solve' the land scarcity problem, considerable amounts of natural forests were allocated to farmers and cleared for agricultural production. During the late seventies of the 20th century, the integrated Soil Erosion Control and Agroforestry Programme (SECAP) was initiated to prevent further soil erosion by applying a combination of improved soil and water conservation measures and the implementation of agroforestry systems. Furthermore in 1989 the Dutch Volunteer Service (SNV) started an irrigation programme (TIP) to improve irrigation structures and access to irrigation water. Both projects also focused on enhancing awareness and cooperation of local people by using extension services, farmer training, providing inputs and establishing tree nurseries. Still, it turned out to be difficult for farmers to adopt new strategies and change their behaviour, mainly due to limited access to information, risk avoidance and lack of investment capacity (Mbagal-Semgalawe and Folmer, 2000).

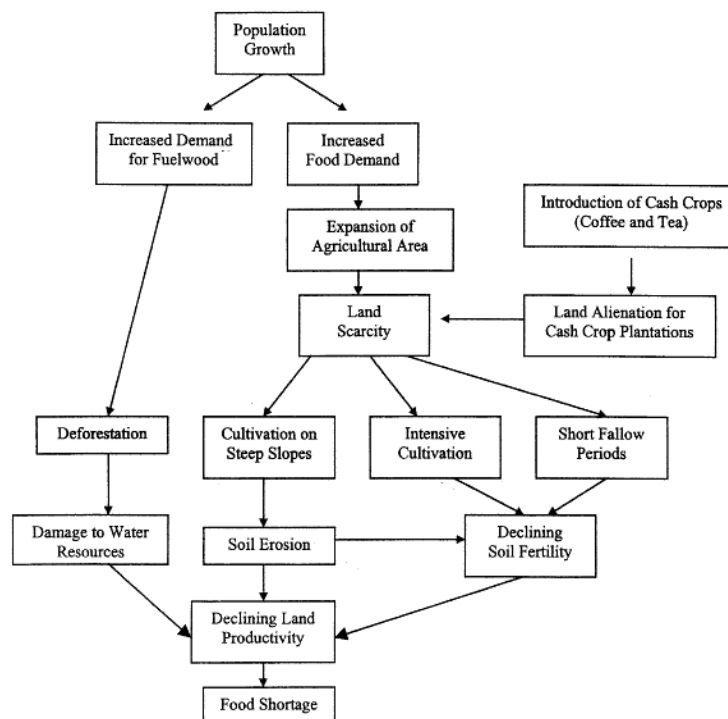


Figure 2.1 Land degradation scheme in the north-eastern mountains in Tanzania (Mbagal-Semgalawe and Folmer, 2000).

Current situation and problems for bean production in the West-Usambara mountains

Poor soil fertility is one of the most important constraints to crop production in most African countries. Land use intensification without adequate nutrient inputs led to nutrient removal, resulting in nutrient deficiencies and decrease of crop yields (Nziguheba *et al.*, 2009). In many parts of Tanzania factors such as population growth, deforestation and poor farming techniques also caused severe soil erosion (Mbagal-Semgalawe and Folmer, 2000). Eroded topsoil is richer in nutrients as fine particles are dislodged first and erosion, together with nutrient removal through the harvest of crops, have a strong negative impact on the nutrient balance (Stoorvogel *et al.* 1993). Furthermore Mowo *et al.* (2006) found out that soils at the top of the slopes were more degraded.

The pH, soil organic matter (SOM), available K and P all increased down the slope, most likely as a consequence of accumulated erosion material. They also indicated that there is very little use of inorganic fertilizers in the Lushoto district, especially in the case of smallholder farmers producing grain legumes for home consumption. Which can mainly be explained by the high prices of fertilizers, the absence of suppliers and information on how to use fertilizers. The application of animal manure also varies a lot in the Lushoto district, and is usually restricted to fields with cash crops and/or maize close to the farmers' homestead (Amijee and Giller, 1998). Due to the small amounts of nutrient inputs, mineral fertilizers and manure application are not capable to completely compensate nutrient outputs (Stoorvogel *et al.* 1993). Considering bean production, low fertility conditions are also likely to decrease the ability of bean to fix atmospheric nitrogen in symbiosis with *Rhizobium*, which decreases final yields even further (Amijee and Giller, 1998). Smithson *et al.* (1993) sampled bean leaf tissue already in 1989 to compare the samples from poorly growing bean plants (with the so called 'Usambara mottle' (UM) symptoms) with leaves from healthy plants in the Lushoto district. The leaf samples were analysed for the nutrient concentrations of P, K, Ca, Mg, Na, Fe, Al, Zn, Cu, Mn and B with the use of inductively coupled plasma emission spectrometry (ICPES). The results are shown in Table 2. Especially the concentration of K seems to be deficient in leaves with UM symptoms but also the P and Zn concentrations were at concentrations considered deficient for optimum bean growth (Smithson *et al.*, 1993). Phosphorus is an important growth factor as it plays a major role in the formation of ATP, improves flowering and root nodulation by *Rhizobium* bacteria (Smithson *et al.*, 1993; Giller *et al.*, 1998). Furthermore, the responses of nutrients were much greater when they were applied together instead of separately, especially for the nutrients P and K (Smithson *et al.*, 1993). More recently, Ndakidemi and Semoka (2006) indicated P deficiency as the major production constraint in the Usambara Mountains, followed by inadequate N and limiting availability of Mg and K in the soil.

Table 2.2 Dry weight element concentrations ($\mu\text{g/g}$) of trifoliolate leaves of *Phaseolus vulgaris* L. with and without Usambara mottle (UM) leaf symptoms from two locations in the Usambara Mountains in 1989 (Smithson *et al.* 1993).

Element	Irente		Miegeo	
	+ symptom	- symptom	+ symptom	- symptom
Phosphorus (P)	2835	3307	1695	2002
Potassium (K)	4958	16138	2210	6190
Calcium (Ca)	28572	23137	41169	19247
Magnesium (Mg)	8346	5679	10303	5775
Sodium (Na)	56	31	81	37
Iron (Fe)	163	120	190	82
Aluminium (Al)	113	57	107	30
Zinc (Zn)	26	29	18	24
Copper (Cu)	9	7	6	6
Manganese (Mn)	181	65	139	82
Boron (B)	36	30	32	22

2.2.2 Indicating nutrient deficiencies

To be able to efficiently apply any given fertilizer it is important to know which specific nutrients are limiting and in which order they are constraining growth. By using nutrient diagnosis based upon (leaf) tissue analysis and concurrent soil tests, fertilization methods can highly be improved on an 'as needed' basis. In general, nutrient contents of foliar tissue are useful indicators to determine the

nutritional status of a given plant or crop (Beverly, 1992). But due to its dynamic nature, leaf tissue composition is affected by both physiological and environmental factors and analysis and interpretation of the results can become quite complicated. The most widely used systems are based on a comparison of analytical results to standard values. The Critical Nutrient Level (CNL) method, in which each nutrient is interpreted individually, nutrient relationships are more difficult to study (Beverly, 1992). To be able to cope with those difficulties the Diagnosis and Recommendation Integrated System (DRIS) was developed and introduced by Beaufils (1973). In DRIS, the use of an integrated index of elemental concentration ratios was suggested, to reduce the influence of environmental terms and to study nutrient availability (Wortmann *et al.* 1992). DRIS evaluates the adequacy of each nutrient in relation to all other nutrients by rating each pair of nutrients. In this way DRIS largely eliminates the common source of error in CNL method, caused by the effect of increase or decrease of nutrient concentrations occurring during plant growth. First, standard values or norms need to be established with the use of a survey approach based on a crop response model. Data on high-yielding populations of a specific crop are collected and averaged to obtain estimates of tissue parameter optima. In addition the coefficients of variation (CVs) of the same data are used as a measure of the relative spread of the yield response curve. Standard values are then derived for all nutrient ratios to be used in index calculations (Walworth and Summer, 1987) by considering the hypothetical nutrients A through N:

$$A \text{ index} = \frac{f(A/B) + f(A/C) + f(A/D) + \dots + f(A/N)}{z}$$

$$B \text{ index} = \frac{-f(A/B) + f(B/C) + f(B/D) + \dots + f(B/N)}{z}$$

$$N \text{ index} = \frac{-f(A/N) - f(B/N) - f(D/N) - \dots - f(M/N)}{z}$$

Where,

$$f(A/B) = \left(\frac{A/B}{a/b} - 1 \right) \frac{1000}{CV} \quad \text{if } A/B \geq a/b$$

or,

$$f(A/B) = \left(1 - \frac{a/b}{A/B} \right) \frac{1000}{CV} \quad \text{if } A/B < a/b$$

In which A/B is the value of the ratio of the two elements in the tissue of the bean plant being diagnosed, a/b is the optimum value or norm for that ratio, CV is the coefficient of variation associated with the norm, and z is the number of functions comprising the nutrient index (Walworth and Summer, 1987). In the end the most important step still lies in the interpretation of the index values in order to identify the nutrient needs of the crop and to come up with appropriate fertilizer recommendations. Single DRIS indices in itself have no meaning as they need to be compared to data available for other nutrients to identify the order of limitation (Beverly, 1992). DRIS indices can range from negative to positive depending on whether a nutrient is deficient, sufficient or excessive relative to other nutrients considered. DRIS was first applied to beans by Wortmann *et al.* (1992), they determined and validated DRIS norms from bean leaf sample data collected from several

tropical countries. They concluded that DRIS gave the better results for foliar tissue analysis than CNL in the tested environments (Wortmann *et al.* 1992).

Applying DRIS in practice

DRIS diagnosis can be applied in practice with the use of factorial fertilizer trials. In that case the following procedure is shown as an example (Wallworth and Summer, 1987):

1. Using data from an experiment in which yield responses have been obtained to the nutrients e.g. being studied, plants from control or lowest level treatment are diagnosed. And the most needed nutrient(s) are determined and used as treatments in a trial.
2. The treatment with additions prescribed by the initial diagnosis is located and the yields are compared. If yield increases when the appropriate treatment is applied, then the diagnosis is considered a success; if not, it is considered a failure.
3. Then proceed with an evaluation of the nutritional status of the second nutrient indicated deficient and so on, until all indices equal zero or, more commonly, until the prescribed treatment cannot be found as part of the experimental layout.

2.3 Research questions and approach

2.3.1 Research questions and hypotheses

Based on the available information indicated above and corresponding to the N2Africa-Tanzania phase II research objectives, three main research questions were developed. Hypotheses were formulated for each (sub)question.

1. Can nutrient limitation in bean production in the West-Usambara Mountains (northern Tanzania) be reliably assessed with leaf nutrient analysis?

It is hypothesised that nutrient deficiencies in bean production in the West-Usambara Mountains (northern Tanzania) can be assessed reliably with the use of leaf nutrient analysis in comparison to critical nutrient levels and existing DRIS data norms for bean.

2. Which nutrients are most limiting bean production in the West-Usambara Mountains (northern Tanzania)?

It is expected that phosphorus (P) and potassium (K) will show up as the main nutrient limitations in growing common bean in the West-Usambara mountains. The responses of nutrients in the fertilizer field trials are therefore likely to increase when nutrients (especially P and K) are applied together.

3. Is there any relationship between farm management, field type and nutrient deficiencies in bean production in the West-Usambara Mountains (northern Tanzania)?

a) Can land use and farmer input management for common bean practices in the past, be related to the observed nutrient deficiencies?

It is hypothesised that nutrient deficiencies are mainly due to low (in)organic fertilizer use by smallholder farmers in the area.

b) Is there any relation between field type characteristics like; soil properties, slope and altitude, and the observed nutrient deficiencies in common bean production in the West-Usambara Mountains (northern Tanzania)?

A direct link between relatively low quality soil properties (assessed by chemical soil analyses) and

high nutrient deficiencies is expected to be found. Furthermore it is hypothesized that soil nutrient deficiencies increase with increasing slope and altitude, as a result of erosion problems causing relocating of nutrients down the slope and a decrease in soil organic matter and water holding capacity up the slope.

3. Material & Methods

3.1 Experimental set-up

3.1.1 Experimental field selection

An experimental field trial was conducted in the short rainy season (*vuli*) from November 2013 until February 2014, on 10 different farmer fields in the West-Usambara Mountains (Tanga region, northern Tanzania), (Figure 1). To include the major field types in the region, fields with a range of soil types, altitudes (between 1200-1700 m above sea level) and slopes were selected (Table 1). Furthermore, the following conditions were set for field site selection; (1) the field was been used for (common) bean production until recently; (2) no presence of non-removable rock, termite, shrubs and or trees within the field; (3) the slope of the field is constant. The fields were selected within a radius of 20 km of the district capital Lushoto in the villages Mabughai, Jaegertal, Lushoto, Kikurunge, Mshizii, Kwemsanga, Ngulwi and Mbuzii (I and II), in agreement with the local agricultural extension officer and owners of the fields (Figure 2). Each field was considered as one experimental site on which two or even three replicate blocks could be implemented, depending on the field size available, ranging from 150 m² to 225 m².

Next to the experimental farmer fields, ten neighbouring fields, cultivated with bean were included in the research (Table 3.2). At those fields no experimental plots were laid out and only soil samples and leaf samples were taken. To be able to get an indication of local farm management and bean production and to compare the results with data obtained with the experimental field trials.



Figure 3.1 General map of Tanzania (East Africa), the research area in the West-Usambara mountains is indicated with a red circle.

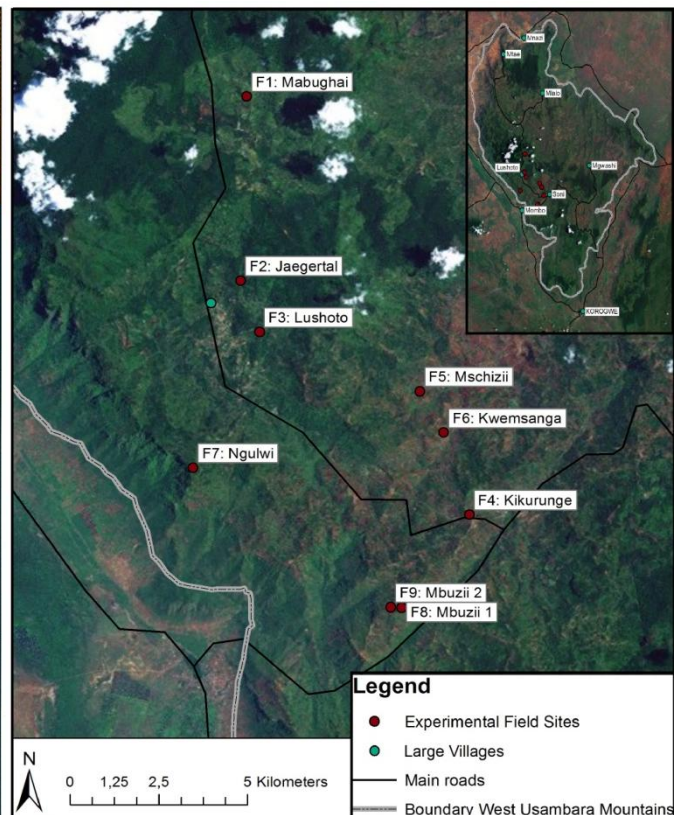


Figure 3.2 Map with the experimental farmer field locations within the research area centred around the main village Lushoto in the West-Usambara mountains.

Table 3.1 Field characteristics of ten selected experimental sites in Lushoto region, West-Usambara Mountains, Tanzania.

Location	Coordinates (DMS)		Altitude (m)	Slope (%)	Position on the hill
	Latitude, S	Longitude, E			
Mabughai	04° 44.215'	038° 17.904'	1667	0-5	Foot slope
Jaegertal	04° 47.111'	038° 17.808'	1415	0	Foot slope - valley bottom
Lushoto	04° 47.920'	038° 18.094'	1444	0	Uphill slope
Kikurunge	04° 50.795'	038° 21.290'	1340	25-30	Uphill slope
Mshizii	04° 48.854'	038° 20.538'	1256	10-15	Foot slope - valley bottom
Kwemsanga	04° 49.509'	038° 20.902'	1253	20-25	Slope
Ngulwi	04° 50.053'	038° 17.082'	1423	10-15	Slope
Mbuzii I	04° 52.256'	038° 20.256'	1218	5-10	Foot slope
Mbuzii II	04° 52.252'	038° 20.090'	1286	10-15	Uphill slope
Mbuzii III	04° 52.409'	038° 19.890'	1221	15-20	Uphill slope

Table 3.2 Field characteristics and leaf sampling dates of ten additionally selected farmer fields, planted with common bean by the farmer involved in the experimental trial or other local smallholder farmers.

Location	Farmer field	GPS coordinates		Altitude (m)	Slope (%)	Position on slope	Date leaf sampling
		Latitude, S	Longitude, E				
Kwemsanga	1	04° 49.509'	038° 20.902'	1253	20-25	Slope	24-12-'13
	2	04° 49.509'	038° 20.902'	1247	10-15	Slope	24-12-'13
Ngulwi	3	04° 50.053'	038° 17.082'	1420	10-15	Slope	25-12-'13
	4	04° 49.986'	038° 17.050'	1440	20-25	Uphill slope	25-12-'13
Kikurunge	5	04° 50.797'	038° 21.294'	1324	20-25	Slope	14-01-'14
Mshizii	6	04° 48.854'	038° 20.538'	1258	10-15	Foot slope - valley bottom	4-01-'14
	7	04° 48.854'	038° 20.538'	1254	10-15	Foot slope - valley bottom	4-01-'14
Mbuzii I	8	04° 52.248'	038° 20.247'	1220	10-15	Foot slope	5-01-'14
Mbuzii II	9	04° 52.251'	038° 20.110'	1284	10-15	Uphill slope	5-01-'14
	10	04° 52.248'	038° 20.076'	1301	5-10	Uphill slope	5-01-'14

3.1.2 Experimental design

The field trials were designed as factorial experiments consisting of two or three replicate blocks (depending on the available area) and with P and K fertilizers and rhizobia inoculation as the main factors (2³). Phosphorus (26 kg P ha⁻¹ as triple superphosphate), Potassium (25 kg K ha⁻¹ as muriate of potash) and rhizobia inoculant mixture containing *Rhizobium* strain CIAT-899 (Legume Technology, UK), containing at least 8-10⁹ cells g⁻¹ of *Rhizobium* bacteria on a peat carrier. Nitrogen (25 kg N ha⁻¹ as calcium ammonium nitrate) was used in one additional treatment together with P and K to analyse the effect of N fertilizer without inoculation. Furthermore one extra control treatment was taken up in the design to increase the comparability with the other treatments, giving 10 treatments in total (Appendix I). The treatment inputs were prepared at the Nelson Mandela Institute for Science and Technology (NM-AIST) in Arusha (TZ) (Appendix II). Taking the size, position and possible fertility gradient of each experimental site into account, two or three replicate blocks, each containing ten treatment plots, were implemented. At the locations Mabughai, Jaegertal, Kikurunge, Kwemsanga, Ngulwi and Mbuzii I all three blocks and for Lushoto, Mshizii, Mbuzii II and Mbuzii III all two blocks.

3.1.3 Field lay-out and cultural practices

Individual plots were 2.5 m x 3.0 m and consisted of five rows, each of 3 m in length with 50 cm in between rows (Appendix I). After clearing and ploughing the soil, the plots were laid out. Fertilizers

for each plot were mixed before being applied in furrows prior to seeding at a depth of approximately 20 cm. Thereafter fertilizers were covered with a 10 cm layer of soil. To keep conditions equal at plots without any fertilizer treatment, furrows were also created at a depth of 20 cm and filled with soil to a depth of 10 cm. Ten fields were sown halfway November 2013 at the beginning of the vuli season (Table 3.3). Three seeds of the early maturing bush bean cultivar Lyamungu 90 (*Phaseolus vulgaris* L.), obtained from Selian Agricultural Research Institute (SARI) in Arusha (TZ), were sown at 20 cm distance within rows at a depth of 5 cm. Plots without the rhizobia inoculant treatment were sown first. Then, the remaining seeds were mixed with approximately 10g of the inoculant mixture and immediately sown at plots with inoculation treatment. One to two weeks after emergence plants were thinned to two plants per stand to give a plant density of approximately 2×10^6 plants ha^{-1} (Table 3.3). Thinning was done by removing the weakest seedling. The seedlings were cut just aboveground to prevent any damage to the other seedlings in the rooting zone. Weeding was carried out once by the farmers 3-4 weeks after emergence (Table 3.3). Due to severe drought in the beginning and at the end of the growing season, the farmer fields at Mabughai and Jaegertal received irrigation water on the initiative of the farmers, once and three times respectively. The plants in Mshizii, Mbuzii I and Mbuzii II showed some symptoms of root rot problems 20-30 days after emergence. To promote additional root development, an extra layer of soil was added to the direct surrounding of each plant (Table 3.3). No further crop protection measures were taken.

Table 3. 3 Dates of cultural practices and field measurements at the experimental sites.

Location	Soil sampling and sowing	Thinning (d.a.s.)	Weeding (d.a.s.)	Levelling up of soil ¹ (d.a.s.)	Irrigation ² (d.a.s.)	Leaf sampling (d.a.s.)	Nodulation scoring (d.a.s.)	Harvest (d.a.s.)
Mabughai	8-11-'13	20	29	-	18	48	48	96
Jaegertal	7-11-'13	21	32	-	9, 25, 30	46	46	96
Lushoto	12-11-'13	18	37	-	-	53	53	91
Kikurunge	11-11-'13	24	64	-	-	64	67	91
Mshizii	15-11-'13	18	35	35	-	50	50	86
Kwemsanga	9-11-'13	23	26	-	-	76	76	79 ⁴ ,92 ⁵
Ngulwi	13-11-'13	16	-	-	-	42	42	86
Mbuzii I	19-11-'13	13	20	20	-	47	59	81
Mbuzii II	19-11-'13	13	20	20	-	47	59	81
Mbuzii III	19-11-'13	18	- ³	- ³	- ³	- ³	- ³	- ³

¹ After discovering root rot problems, soil was levelled up around the plant to promote new root development

² Irrigation measures were taken by the farmers themselves due to drought problems at the beginning (Mabughai and Jaegertal) and halfway the growing season (Jaegertal, twice).

³ Due to severe drought at the beginning of December, the plants at Mbuzii III were not able to survive after germination

⁴ Replicate 1

⁵ Replicate 2 and 3

3.1.4 Interviews

To establish cropping history, inputs used in the past and general management practices, farmers responsible for the field sites used in this study were interviewed after completion of the experiments. Questions were divided into general information about the location, general information about the household, management practices used on the field in the past and perception on agricultural characteristics by the farmer (Appendix III). There were two females and seven men involved in the experimental trials. Seven out of nine, were farmer as a main occupation. They were between 32 and 60 years old and took care of a household with two or three adults and

four children on average. The location in Lushoto served as a research station and was managed by different employees, both male and female. The location in Kikurunge belonged to a primary school and was managed by the agricultural teacher (female) (Table 4.7).

Table 3.4 Farmer and family information for each site being used in the experimental trials in Lushoto region (Tanzania).

Location	Gender farmer	Main occupation	Age farmer	Education (level)	Household	
					Adults	Children
Mabughai	male	Head teacher	43	diploma	2	2
Jaegertal	male	Farmer	45	secondary st. 7	2	6
Lushoto	male	Farmer (research station)	-	-	-	-
Kikurunge	female	Agricultural teacher primary school	-	-	-	-
Mschizii	male	Farmer	53	secondary st. 7	2	8
Kwemsanga	male	Farmer	32	primary	2	4
Ngulwi	female	Farmer	38	secondary st. 7	2	4
Mbuzii I	male	Farmer	60	secondary st. 6	2	3
Mbuzii II	male	Farmer	49	primary	3	3

3.1.5 Weather conditions

To get an indication of rainfall differences in the area, additional rainfall measurement were carried out with the use of simple rain gauges, produced from 1.5 litre water bottles (top turned upside down to prevent evaporation) and put at each field. Rain gauges were emptied once every week or two weeks and the diameter of each bottle was noted down to be able to determine the surface area and calculate the amount of rainfall in mm (Figure 3.3). Due to severe drought at the end of December until the beginning of January, the plants at Mbuzii III were not able to survive after germination and therefore excluded from the research (Table 3.3). Heavy rainfall occurred at almost all fields at the beginning of the growing season.

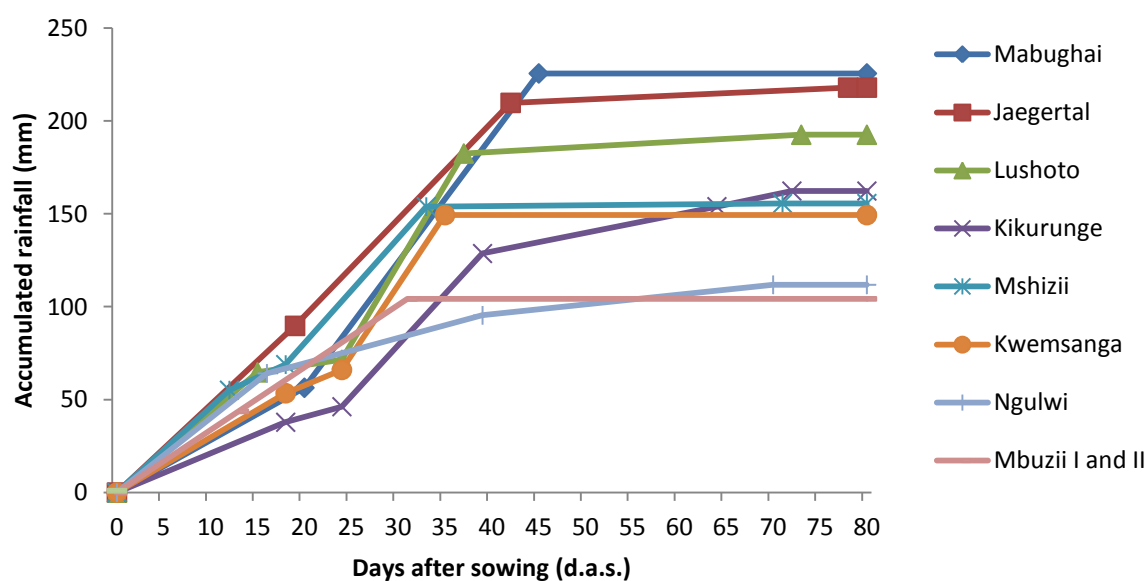


Figure 3.3 Accumulated rainfall data (mm) as a function of days after sowing (d.a.s.) during the vuli season (2013/2014). Measured at the different research sites used in the fertilizer field trials in the Lushoto region (West-Usambara Mountains, Tanzania).

3.2 Analyses

3.2.1 Chemical soil analysis

Prior to seeding, the soils of the experimental sites (Table 3.1) and the selected neighbouring fields managed by the farmer (Table 3.2) were sampled and chemically analysed. Per site, 20 soil samples were randomly taken at a depth of 0-20 cm. Samples were bulked and air-dried and a subsample of about 500 g was taken for chemical analysis. Which was then mixed, sieved (2 mm) and analysed at at Cropnuts, Crop Nutrition laboratory services Ltd (Nairobi, Kenya). The following soil properties with corresponding methods between brackets were measured; pH (H₂O), available P (Olsen), cation exchange capacity (CEC) (extraction with ammonium acetate), cations (K, Ca, Na and Mg content) (atomic absorption spectrophotometry), electrical conductivity (EC) and soil particle size (Bouyoucos). The results were compared with critical levels obtained from literature.

3.2.3 Plant growth

At emergence the number of plants in the plots were counted (percentage emergence). Further on during the growing season the following parameters were recorded; days to flowering (number of days to flowering of 50% of the plants), crop vigour (visual overview and scoring from 1 (poor growth) to 5 (healthy and good growth)), plant height in cm (mean of ten plants per plot at maturity) and nodulation scoring. To record nodulation 6 plants per plot were carefully uprooted within the area assigned for nodulation scoring (Appendix I) at the moment up to flowering. The following parameters were assessed: number (number of nodules on each plant), crown root nodulation, and colour (active: red, pink or brown; inactive: green, grey or white) to obtain a nodulation score ranging from 0 to 5 in which: 0=root nodules were absent, 1=rare (<5 active nodules), 2=few (5-10 active nodules), 3=moderate (11-20 active nodules), 4=abundant (20-50 active nodules) and 5=super nodulated (>50 active nodules).

3.2.2 Leaf sampling and nutrient concentration analysis

Bean leaf samples were taken from the uppermost, fully expanded trifoliolate leaf on the main stem, harvested at the time of 50% flowering (Wortmann *et al.*, 1992), approximately 6-8 weeks after seeding. Recently matured leaves were picked as they best reflect the general nutrient status of the whole plant during the period of most intensive growth. Considering the experimental trial, 20 leaves were sampled from the harvest area (Appendix I) in each treatment plot, for all replications (Table 2). Furthermore, 20 bean leaves were sampled from ten additionally selected farmer fields, planted with common bean by the farmer: Mshizii (2), Kwemsanga (2), Ngulwi (3), Mbuzii I (1) and Mbuzii II (2) (Table 3.2). Leaves from diseased, damaged or dead plants were not included in the sampling procedure. Samples were carefully washed with distilled water prior to sun drying. The dried samples were subsequently grinded with a stone mortar and pestle and repacked in plastic sealed bags, each bag containing 5-10 g of dried and grinded leaf sample.

To analyse nitrogen content of the leaves, a Carbon-Hydrogen-Nitrogen (CHN) elemental analysis was used. About 1 mg of the collected leaf samples was transferred to tin cups and analysed with the use of a CHN EA-1110 analyser at the KU Leuven (Belgium). Each subsample was burned in an excess of oxygen to trap the combustion products carbon dioxide, water and nitric oxide (Jimenez and Ladha, 1993). The obtained masses were then used to calculate the N percentages of the samples. For the determination of the nutrient concentrations of P, K, Ca, Fe, Mg, Mn, Cu and Zn inductively coupled plasma optical emission spectrometry (ICP-OES) analysis was used (Nölte, 2003). First 1 ml of nitric acid (HNO₃) was added to approximately 50 mg of the dry leaf (exact weights were noted) in 10

ml glass tubes. After a minimum of 12 hours, the tubes were digested on a hot plate to 180°C and mixed occasionally to promote the digestion process. This procedure continued up to the moment at which approximately 0.5 cm of liquid was left in each glass tube. After which the digest was cooled to room temperature and MQ water was added to dilute to 10 ml (Appendix IV). Two internal maize laboratory standards and two blanks (1 ml HNO₃) were included in each sample batch (Appendix IV). The nutrient content in the digest was determined with the use of ICP-OES analysis (Varian 720 ES, KU Leuven, Belgium). After the analysis the blank concentration was subtracted from the sample concentration (at equal dilution) and the solution concentration (mg l⁻¹) was converted to a dry weight basis using the following equation:

$$\frac{mg/l \times volume\ liquid\ (l)}{sample\ weight\ (kg)}$$

The obtained nutrient concentration data from the leaf samples was used to apply the Diagnosis and Recommendation Integrated System (DRIS; Beaufils, 1973) to generate nutrient indices. DRIS indices are calculated based on ratios of each nutrient relative to all other nutrients using the equations below provided by Walworth and Summer (1987). Consider the hypothetical nutrients A through N:

$$\begin{aligned} A\ index &= \frac{f(A/B) + f(A/C) + f(A/D) + \dots + f(A/N)}{z} ; \\ B\ index &= \frac{-f(A/B) + f(B/C) + f(B/D) + \dots + f(B/N)}{z} ; \\ N\ index &= \frac{-f(A/N) - f(B/N) - f(D/N) - \dots - f(M/N)}{z} ; \end{aligned}$$

where,

$$f(A/B) = \left| 1 - \frac{a/b}{A/B} \right| \frac{1000}{CV}$$

In which A/B is the value of the ratio of the two elements in the tissue of the bean plant being diagnosed, *a/b* is the optimum value or norm for that ratio, CV is the coefficient of variation associated with the norm, and *z* is the number of functions comprising the nutrient index (Walworth and Summer, 1987). In this study the indices for N, P, K, Ca, Mg, Mn, and Zn were calculated and compared to the norms for dry bean, determined by Wortmann *et al.* (1992) (Table 3.5).

Table 3.5 DRIS norms and the coefficient of variation (CV) associated with the norms for beans generated from a broad-based database (Wortmann et al., 1992).

	Number of samples	Norms	CV (%)		Number of samples	Norms	CV(%)
N/P	306	13.588	25.8	K*Ca	227	5.469	34.9
N/K	306	2.098	37.7	K*Mg	227	1.567	41.8
N*Ca	227	10.767	59.2	K*Mn	227	683.959	77.6
N*Mg	227	2.764	31.7	K/Zn	227	0.058	70.7
N*Mn	227	1044.700	62.0	Ca/Mg	227	3.564	24.7
N/Zn	227	0.116	56.9	Ca/Mn	227	0.013	46.1
P/K	306	0.157	26.7	Ca*Zn	227	116.304	60.2
P*Ca	227	0.816	36.0	Mg/Mn	227	0.003	100.0
P*Mg	227	0.218	51.4	Mg*Zn	227	34.176	61.4
P*Mn	227	82.939	78.2	Mn*Zn	227	12912.000	80.9
P/Zn	227	0.008	50.0				

3.2.4 Harvest

At harvest (Table 3.3) the number of plants in the harvest area was recorded. The harvest area consisted of the inner three rows horizontally, excluding three border rows at the vertical side at which the nodulation scoring was carried out and one border row at the other vertical side, giving a harvest area of 1.5 m x 2.2 m (Appendix I). In the case of a relatively low yielding field (Kikurunge, Kwemsanga, Ngulwi, Mbuzii I and Mbuzii II) all above ground biomass was harvested from the harvest area. From higher yielding fields (Mabughai, Jaegertal and Lushoto), 20 plants were randomly harvested from the harvest area. At Mshizii the farmer already started harvesting some plants and pods, so no harvest results could be obtained at this site. The number of plants harvested per plot were counted once again. The pods were separated from the stems and the remaining leaf material and soil particles were removed. Thereafter both pods and stems were weighted separately to determine the fresh weight. The number of pods was counted before creating a subsample of 20 pods (if possible), the FW of this subsample was determined. In the lab at NM-AIST the pods were threshed to separate the husks from the seeds and the number of seeds was counted. Stems, husks and seeds were oven dried at 65°C for 24 hours and weighed to determine the dry matter. From these measurements the final yield components were derived: percentage of plants which reached maturity, average number of seeds per pod, 100-seed weight (g), dry stem yield (kg ha⁻¹), dry grain yield per plant (g plant⁻¹) and dry grain yield ha⁻¹ (kg ha⁻¹).

3.3 Statistical Analysis

The research was set up as a randomized complete block design at each field, with plots randomized within replicate blocks. Treatment effects on agronomic indicators were analysed through an analysis of variance (ANOVA) using the F-test. Treatment effects were analysed while accounting for Block and Field effects. In which Block and Field were used as blocking factors and subsequent LSD tests were performed ($\alpha = 0.05$). Treatment 1 to 9 were analysed for possible main effects and interactions. The addition of nitrogen fertiliser (treatment 10) was analysed separately by comparing to treatment 6 and 9. To look for possible site effects, treatment effects were also analysed for each location separately, with Block as a blocking factor. All analyses were carried out with the use of the statistical software R[®] version 3.0.3.

4. Results

4.1 Soil analysis

4.1.1 Fertilizer and inoculation field trial sites

Soil analysis results are shown in Table 4.1. At the locations Mabughai, Jaegertal and Lushoto, $\text{pH} \leq 5.8$ was measured, which is below the optimal pH range for bean production of 5.8 to 6.5, indicated by Lunze (2012). Acidic soils (pH below 5.2) did not occur. Available P (P-Olsen) ranged from 10.9- indicating deficiencies in soil available phosphorus at all nine field sites being tested, when being compared to the critical value of 15.0 mg kg^{-1} (Ndakidemi and Semoka, 2006). Where Mabughai and Lushoto gave relatively higher values in comparison to the other fields (Table 4.1). The exchangeable bases Ca, Mg, K and Na were measured. Exchangeable K in the soil ranged from 0.11-0.25 $\text{cmol}(+) \text{ kg}^{-1}$, with six out of nine locations scoring lower than the recommended value of $0.20 \text{ cmol}(+) \text{ kg}^{-1}$ for adequate crop growth in East Africa (Anderson, 1973). Three locations showed relatively low levels for Ca, when being compared with the critical level of $5.0 \text{ cmol}(+) \text{ kg}^{-1}$ (Lunze, 2012). For exchangeable magnesium, both analyses showed Mg levels below the critical level of $2.0 \text{ cmol}(+) \text{ kg}^{-1}$ (Ndakidemi and Semoka, 2006) at seven out of nine field sites (Table 4.1).

Different textural classes were found between the sites; (1) sandy clay loam at Mabughai, Lushoto and Mbuzii I; (2) sandy clay at Jaegertal and Kikurunge; (3) clay at Mschizii, Kwemsanga, Ngulwi and Mbuzii II (Table 4.1).

4.1.2 Farmer Fields

Chemical soil analysis was also carried out for nine additional farmer fields used in the research (Table 4.2). At all those field sites measured pH was relatively high, with an average pH of 6.5. Available P (P-Olsen) revealed extremely low values (below 3.0 mg kg^{-1}) for six out of nine locations, when being compared to a critical value of 15.0 mg kg^{-1} (Ndakidemi and Semoka, 2006). Exchangeable K values were below the critical deficiency level for seven out of nine locations. The values for Mg and Ca fluctuated around the critical deficiency level. Remarkably high values were obtained for soil available P and CEC.

Table 4.1 Chemical soil properties from the selected experimental farmer field locations in the West-Usambara Mountains, Lushoto district, Tanga region, Tanzania (vuli season 2013-2014). Analysed at Cropnuts, Crop Nutrition laboratory services Ltd (Nairobi, Kenya).

Location	pH (H ₂ O)	P-Olsen Avail. P (mg kg ⁻¹)	CEC (cmol(+) kg ⁻¹)	Exchangeable bases (cmol(+) kg ⁻¹)				EC (mS cm ⁻¹)	Particle size analysis			Texture	
				K	Ca	Mg	Na		Clay (%)	Silt (%)	Sand (%)	Abbreviation	Classname
Mabughai	5.4	6.1	17.6	0.11	3.92	0.84	0.24	1.13	24	18	58	SCL	Sandy clay loam
Jaegertal	5.5	1.5	21.7	0.18	5.56	1.42	0.22	2.69	36	23	40	CL	Clay loam
Lushoto	5.3	10.9	12.5	0.20	2.40	0.70	0.19	1.21	28	20	52	SCL	Sandy clay loam
Kikurunge	6.3	2.6	24.2	0.13	8.00	3.07	0.29	0.67	44	18	42	C	Clay
Mshizi	6.3	5.7	15.7	0.18	4.96	1.79	0.33	0.67	46	16	38	SC	Sandy clay
Kwemsanga	6.3	2.3	16.8	0.17	5.36	1.58	0.23	0.70	42	12	46	SC	Sandy clay
Ngulwi	6.2	2.5	16.7	0.20	5.16	1.30	0.16	0.69	40	18	42	CL/C	Clay (loam)
Mbuzii I	6.0	2.7	21.5	0.19	6.26	2.23	0.30	0.78	32	18	50	SCL	Sandy clay loam
Mbuzii II	6.1	1.6	18.7	0.25	5.36	1.99	0.32	1.24	50	12	38	C	Clay

Table 4.2 Chemical soil properties from additional farmer fields (FF) in the West Usambara Mountains, Lushoto district, Tanga region, Tanzania (vuli season 2013-2014). Analysed at Cropnuts, Crop Nutrition laboratory services Ltd (Nairobi, Kenya).

Location	pH (H ₂ O)	P-Olsen Avail. P (mg kg ⁻¹)	CEC (cmol(+) kg ⁻¹)	Exchangeable bases (cmol(+) kg ⁻¹)				EC (mS cm ⁻¹)
				K	Ca	Mg	Na	
FF1	6.5	2.1	16.2	0.14	5.31	1.52	0.27	0.28
FF2	6.5	2.7	14.6	0.14	4.69	1.46	0.19	0.26
FF4	5.0	1.4	9.16	0.10	1.49	0.51	0.20	0.48
FF5	6.6	2.8	26.2	0.14	8.33	3.15	0.31	1.06
FF6	6.7	1.9	14.8	0.13	4.66	1.82	0.29	0.53
FF7	6.7	2.1	12.3	0.19	3.72	1.59	0.34	0.44
FF8	6.7	7.0	18.2	0.23	5.71	2.18	0.40	0.56
FF9	6.1	1.8	18.3	0.12	5.46	1.78	0.32	0.96
FF10	6.7	17.6	25.0	2.24	7.61	2.38	0.31	2.43

4.2 Growth parameters (experimental trial)

4.2.1 Crop vigour

At approximately 50% flowering, crop vigour at each plot was evaluated and scored with the use of a crop vigour scale ranging from 1 (poorest growth) to 5 (best growth) (Figure 4.1). In the factorial experiment both the application of P and K fertiliser ($p < 0.001$) gave a significant increase in crop vigour in comparison with the control and inoculation treatment. Furthermore a significant and positive interaction effect of K, P and I was found ($p < 0.05$). When comparing inoculation and nitrogen as an addition to the original 'K+P' treatment in a separate statistical analysis of variance, the addition of nitrogen fertiliser had a positive significant main effect on crop vigour ($p < 0.001$) (Figure 4.1). The addition of inoculant to P and K fertiliser however, had no significant effect. During the growing season, leaf tissue appearance between different treatments and fields was surveyed. Interveinal leaf chlorosis symptoms, referred to as 'Usambara mottle' by Smithson et al. (1993), were clearly visible at Mabughai and Ngulwi in treatments where only P fertilizer was added (Figure 4.3). Furthermore, visual overviews were created at the moment of 50% flowering for each field site, to represent the treatment effects on crop vigour (Appendix V).

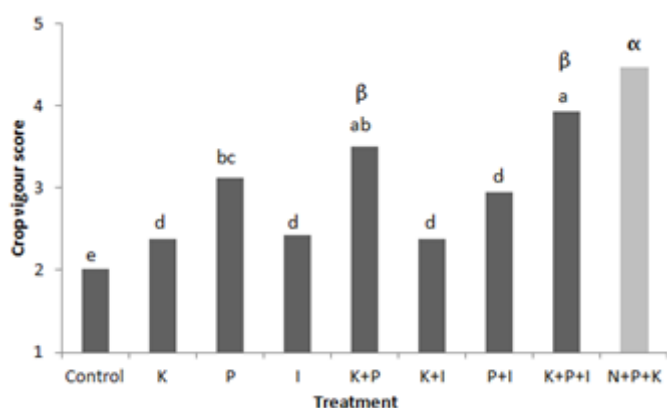


Figure 4.1 Crop vigour ranging from 1 (poorest growth) to 5 (best growth) plotted against treatment. The 'N+P+K' treatment is shown as an additional treatment to the factorial fertiliser (P and K) and inoculation (I) treatments. Significant differences (LSD=0.483, $\alpha=0.05$) are indicated with alphabetic letters for the treatments within the factorial design. 'N+P+K' was statistically compared with the 'K+P' and the 'K+P+I' treatment to look for possible effects of addition of nitrogen fertilizer in comparison to inoculation, significant differences are indicated with Greek letters (LSD=0.418, $\alpha=0.05$).

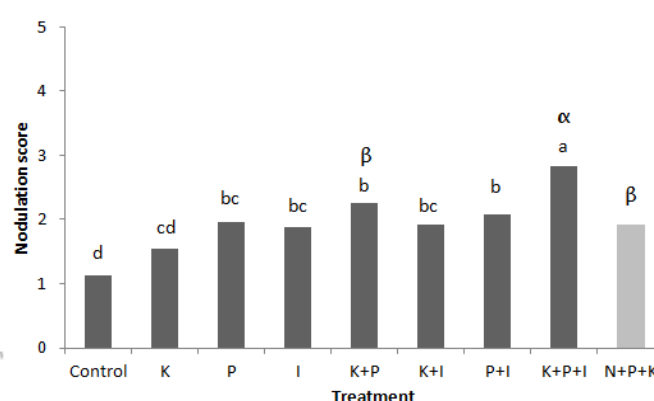


Figure 4.2 Average nodulation score per plant ranging from 0 (no nodules present) to 5 (50 or more active nodules present) plotted against treatment. The 'N+P+K' treatment is shown as an additional treatment to the factorial fertiliser (P and K) and inoculation (I) treatments. Significant differences (LSD=0.426, $\alpha=0.05$) are indicated with alphabetic letters for the treatments within the factorial design. 'N+P+K' was statistically compared with the 'K+P' and the 'K+P+I' treatment to look for possible effects of addition of nitrogen fertilizer in comparison to inoculation, significant differences are indicated with Greek letters (LSD=0.353, $\alpha=0.05$).

4.2.2 Nodulation

All three treatments of P, K or I gave a significant increase in nodulation score in comparison with the control ($p < 0.001$) (Figure 4.2). No significant interactions were found for P or K fertilizer in combination with inoculation. However, a significant positive interaction between the application of inoculant in addition to P and K fertilizer was found ($p < 0.05$). The addition of inoculant or nitrogen fertilizer gave a nodulation score significantly different from the 'P+K' treatment. Inoculation had a significant positive effect on the nodulation score ($p < 0.005$) and nitrogen fertilizer had a significant negative effect on the nodulation score ($p < 0.001$).



Figure 4.3 Interveinal leaf chlorosis symptoms at the P fertilizer treatment at Mabughai (at the moment of approximately 50% flowering).

4.3 Bean yield

At harvest, bean grain and stem yields at nine field locations were measured and several yield components were calculated. To be able to analyse overall effects and possible site effects, the results were statistically analysed for all locations together and for each location individually.

4.3.1 Yield - all locations combined

Bean grain yields were significantly increased by the application of both P and K fertilizer ($p < 0.001$ and $p < 0.01$ respectively). There was no significant main effect of inoculation and none of the interactions among treatments were significant. A separate analysis indicated that addition of nitrogen fertilizer nor inoculation significantly affected bean grain yield relative to the 'K+P' treatment. Largest bean grain yields were obtained within the 'K+P', 'K+P+I' and 'N+P+K' treatments, whereas smallest bean grain yields were found in the control and inoculation plots (Figure 4.4). Differences in bean grain yield reflected differences in pod number m^{-2} (Figure 4.5). The application of both P and K fertilizer led to a significant increase of pods m^{-2} ($p < 0.001$ and $p < 0.01$ respectively).

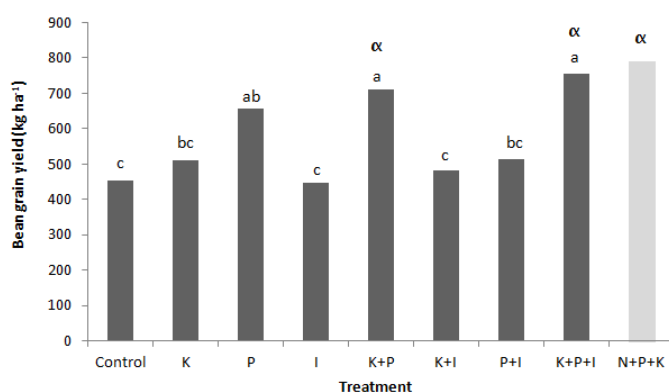


Figure 4.4 Bean grain yield ($kg\ ha^{-1}$) plotted against treatment. The 'N+P+K' treatment is shown as an additional treatment to the factorial fertiliser (P and K) and inoculation (I) treatments. Significant differences (LSD, $\alpha=0.05$) are indicated with alphabetic letters for the treatments within the factorial design and with Greek letters to compare the treatments 'K+P', 'K+P+I' and 'N+P+K' treatment.

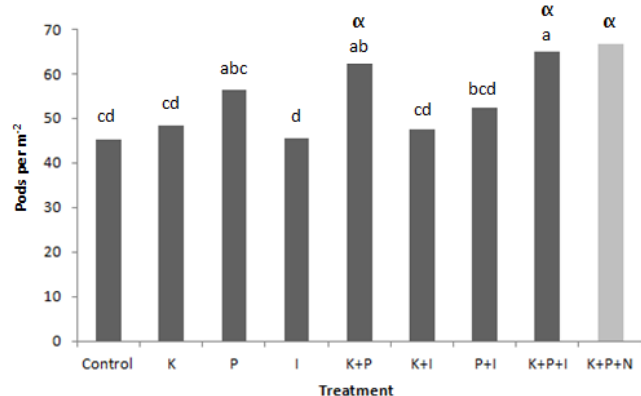


Figure 4.5 Pods per m^{-2} plotted against treatment. The 'N+P+K' treatment is shown as an additional treatment to the factorial fertiliser (P and K) and inoculation (I) treatments. Significant differences (LSD, $\alpha=0.05$) are indicated with alphabetic letters for the treatments within the factorial design and with Greek letters to compare the treatments; 'K+P', 'K+P+I' and 'N+P+K'.

4.3.2 Yield - at individual locations

The treatment effects on several yield components, were statistically analysed for each location separately, with replicate block as the blocking factor (Table 4.4). Highest bean grain and stem yields were obtained at Jaegertal, Mabughai and Lushoto respectively. At Jaegertal, the addition of P had a significant and positive effect on both bean grain and stem yield ($p < 0.001$). Here the highest average grain yield was obtained after addition of P fertilizer, followed by the 'K+P+I' treatment and 'N+P+K' treatment. Smallest grain and stem yields were obtained in the control and inoculation plots. At Mabughai, a significant increase of grain yield was obtained for all three single factor treatments; phosphorus ($p < 0.005$), potassium ($p < 0.001$) and inoculation ($p < 0.05$), where bean grain yields ranged from 522 kg ha⁻¹ for the control plots to 1477 kg ha⁻¹ for the 'K+P' treatment. K fertilizer increased different yield components, especially in comparison with P fertilizer. A significant and positive main effect of K fertilizer on grain yield was shown within the factorial fertilizer and inoculation experiment at Lushoto ($p < 0.05$). Largest bean grain and stem yields at Lushoto were measured when both P and K fertilizer, in combination with N fertilizer or inoculation were applied. Average grain yields obtained at Ngulwi, Kikurunge, Kwemsanga, Mbuzii I and Mbuzii II, were relatively low, with a maximum of 400 kg ha⁻¹. The same was true for dry stem yields. Still, some significant differences between treatments were found here. At Kikurunge grain yield was not significantly different between treatments and the average grain yield obtained in the control plots was remarkably high, explaining low responses to fertilizers and inoculation. However, dry stem yield was significantly increased through additional N fertilizer application on top of P fertilizer, within the 'N+P+K' treatment in comparison with the 'P+K' treatment ($p < 0.001$). At Kwemsanga no significant treatment effects were found for any of the yield components. Relatively low yields were found for the field experiments at Ngulwi and grain yield was significantly increased by P ($p < 0.001$) and the combination of both P and K fertilizer ($p < 0.05$). Largest grain yields were obtained when P and K fertilizers were applied in combination with inoculation. 100 seed weight was significantly increased by P fertilizer ($p < 0.05$). N fertilizer in addition to P and K gave no substantial increase of grain yield, however it did significantly increase stem yield ($p < 0.005$). Also P and K fertilizer had a significant positive effect on stem yield ($p < 0.0001$). Both Mbuzii I and II had to face major growth problems due to drought. No significant effects or interactions were found for any of the yield components obtained from Mbuzii I. At Mbuzii II no clear results were found for grain yield. Stem yield was significantly positively affected by P fertilizer and highest stem yields were obtained for the combination of P and K fertilizer together with inoculation or N fertilizer.

Table 4.4 Yield components at individual sites, in the fertilizer and inoculation trials in the West-Usambara Mountaints, Lushoto region (Tanga district, Tanzania).

Location	Trt ¹	Yield (kg ha ⁻¹)	Yield (g plant ⁻¹)	Stem yield (kg ha ⁻¹)	100 seed weight (g)	Location	Trt ¹	Yield (kg ha ⁻¹)	Yield (g plant ⁻¹)	Stem yield (kg ha ⁻¹)	100 seed weight (g)
Mabughai	Control	521.6	2.9	180.1	40.6	Kwem-sanga	Control	175.9	1.1	93.7	33.3
	K	938.3	5.0	291.6	46.1		K	293.6	1.6	161.0	33.8
	P	885.1	4.9	305.8	42.9		P	340.6	1.8	165.3	31.5
	I	606.4	3.4	191.2	48.8		I	348.8	2.2	155.1	35.7
	K+P	1476.6	7.8	557.5	47.8		K+P	245.3	1.3	146.5	34.1
	K+I	604.6	3.4	221.9	43.6		K+I	349.1	2.3	100.2	34.8
	P+I	598.2	3.3	236.0	41.3		P+I	276.5	1.5	163.8	34.1
	K+P+I	1107.2	6.2	337.0	46.4		K+P+I	167.7	1.0	101.3	32.5
LSD ²		357.6	2.01	118.06	8.47	LSD		274.7	1.33	115.82	5.39
	K+P+N	1450.9	8.2	540.9	43.4		K+P+N	287.9	1.8	168.6	36.7
	LSD	844.7	4.66	318.27	3.72		LSD	245.7	1.34	143.25	5.90
Jaegertal	Control	1630.0	8.8	365.2	42.3	Ngulwi	Control	226.9	1.3	73.2	25.2
	K	1575.2	8.1	346.0	42.1		K	180.8	0.9	95.3	19.8
	P	2737.6	15.0	883.3	47.0		P	281.5	1.5	112.9	27.5
	I	1348.7	7.6	299.1	40.5		I	184.3	1.0	79.3	21.6
	K+P	2256.0	11.9	731.2	44.2		K+P	370.9	2.0	163.7	25.1
	K+I	1843.9	10.0	381.9	40.0		K+I	162.5	0.8	96.5	20.8
	P+I	2131.8	11.0	697.9	42.4		P+I	285.4	1.5	119.2	26.7
	K+P+I	2684.2	14.5	848.0	46.4		K+P+I	404.3	2.1	168.2	25.6
LSD		696.3	3.88	250.33	6.01	LSD		146.3	0.74	24.36	6.69
	K+P+N	2529.6	14.3	837.9	44.2		K+P+N	365.3	1.8	213.4	27.2
LSD		313.3	1.32	290.49	5.07	LSD		368.1	1.87	25.14	9.40
Lushoto	Control	389.3	2.1	189.3	33.1	Mbuzii I	Control	205.0	1.2	146.4	27.4
	K	617.7	3.1	228.2	34.9		K	199.6	1.2	146.4	28.2
	P	551.0	2.8	229.1	38.1		P	130.8	0.7	194.0	32.1
	I	568.5	2.9	201.1	35.3		I	123.1	0.7	174.7	27.8
	K+P	718.3	3.8	294.1	37.6		K+P	141.1	1.2	111.2	27.0
	K+I	425.6	2.3	185.3	34.8		K+I	122.3	0.9	117.8	26.2
	P+I	252.1	1.3	150.1	31.4		P+I	192.1	1.3	142.6	26.8
	K+P+I	1091.4	5.6	627.4	44.8		K+P+I	148.1	1.1	125.1	27.8
LSD		551.0	2.88	381.55	12.22	LSD		<i>n.a.</i> ³	<i>n.a.</i> ³	<i>n.a.</i> ³	<i>n.a.</i> ³
	K+P+N	1084.2	5.8	560.3	46.1		K+P+N	126.1	0.8	203.4	27.9
LSD		1857.3	9.14	1326.59	46.19	LSD		61.9	1.06	50.39	4.98
Kikurunge	Control	229.6	0.8	85.0	25.2	Mbuzii II	Control	97.0	0.8	93.4	26.8
	K	143.8	0.9	103.4	22.2		K	4.4	0.1	54.7	18.1
	P	60.7	0.4	115.3	22.8		P	32.1	0.2	107.4	22.6
	I	174.2	1.1	99.0	26.0		I	10.6	0.1	70.5	13.2
	K+P	178.5	1.1	129.3	23.6		K+P	111.6	0.7	164.2	26.1
	K+I	92.7	0.7	73.5	20.9		K+I	97.3	0.5	91.5	26.8
	P+I	65.2	0.4	95.8	26.6		P+I	81.0	0.5	157.0	28.0
	K+P+I	225.3	1.3	133.3	25.6		K+P+I	106.3	0.6	169.0	26.5
LSD		131.9	0.87	44.69	4.09	LSD		165.6	1.02	57.94	13.21
	K+P+N	208.8	1.2	196.6	25.1		K+P+N	164.7	1.0	215.0	26.7
LSD		160.1	1.22	19.10	4.23	LSD		712.12	4.23	359.27	11.77

¹ Treatment

² Least significant differences were calculated based on the results of the analysis of variance with $\alpha=0.05$

³ LSD is not available due to one missing observation

4.4 Leaf nutrient analysis

4.4.1 Leaf nutrient concentrations

The leaf nutrient concentrations of the macronutrients N, P, K, Ca, Mg and micronutrients Cu, Zn and Mn were compared to the critical deficiency concentrations (CDCs) for bean, obtained from Reuter and Robinson (1997), (Table 4.5). Treatment effects were largest for leaf K concentration. 84.8% of all observations were below the adequate potassium concentration range of 1.5-3.5%. With the smallest value of 0.19% obtained at Kikurunge and the largest value of 2.09% at Kwemsanga. Application of K fertilizer significantly increased overall leaf K concentration ($p < 0.0001$). The obtained N concentrations ranged from a minimum value of 2.89% at Kikurunge to a maximum value of 7.83% at Jaegertal in which 89.2% of the total observations were below the adequate range of 5.2-5.5% (Reuter and Robinson, 1997). Both P and K fertilizer significantly increased leaf N concentration ($p < 0.05$). In the case of phosphorus 94.9% of the total observations were below the adequate phosphorus concentration range of 0.4-0.6% (Reuter and Robinson, 1997), with the smallest concentration of 0.10% measured at Mbuzii II and the largest concentration of 0.52% at Lushoto. P fertilizer had an overall significant effect on leaf P concentration ($p < 0.01$). The leaf nutrient concentrations for calcium, magnesium, copper and manganese fell within the adequate ranges obtained from literature for almost all plots (Table 4.5). Measured leaf zinc concentrations, however, were in 99.2% of the total measurements, less than the adequate range of 35-100 mg kg⁻¹ (Reuter and Robinson, 1997).

Leaf nutrient concentrations measured at ten additional farmer fields located in the region, which did not receive any treatment, indicated overall deficiencies for the nutrients K, P and Zn (partly N) relative to the CDCs. Ca, Mg, Cu and Mn scored well within the adequate nutrient ranges proposed for bean growth (Table 4.6).

4.4.2 Relationship between grain yield and leaf nutrient concentrations

Bean leaf nutrient concentrations (N, P, K, Ca, Mg, Zn and Mn) were compared with grain yield (Figure 4.6) to study the relationship between plant growth and nutrient concentration in shoots. Leaf N concentrations varied from 3.0-6.5% within the low grain yield section (below 750 kg ha⁻¹). However, a slight increase in leaf N concentration was observed in the relatively higher grain yield section, consisting of yields obtained at Mabughai, Jaegertal and Lushoto. The relationship between leaf P concentration and grain yield showed a similar pattern, with a broad nutrient range at the base, after which growth increases with only small changes in leaf P concentration. A more linear relationship was found for leaf K concentration, still a broad nutrient range was observed within the relatively low yield section. Clear C-shaped curves were found (Figure 4.6), when comparing grain yield (y-axis) with the leaf concentrations for Ca and Mg and partly for Cu and Zn (x-axis). Where the leaf nutrient concentrations decreased with a slight increase in grain yield, when grain yields were below approximately 750 kg ha⁻¹. After which nutrient-induced increases in grain yield were observed. No clear relationship between leaf Mn concentration and grain yield could be found.

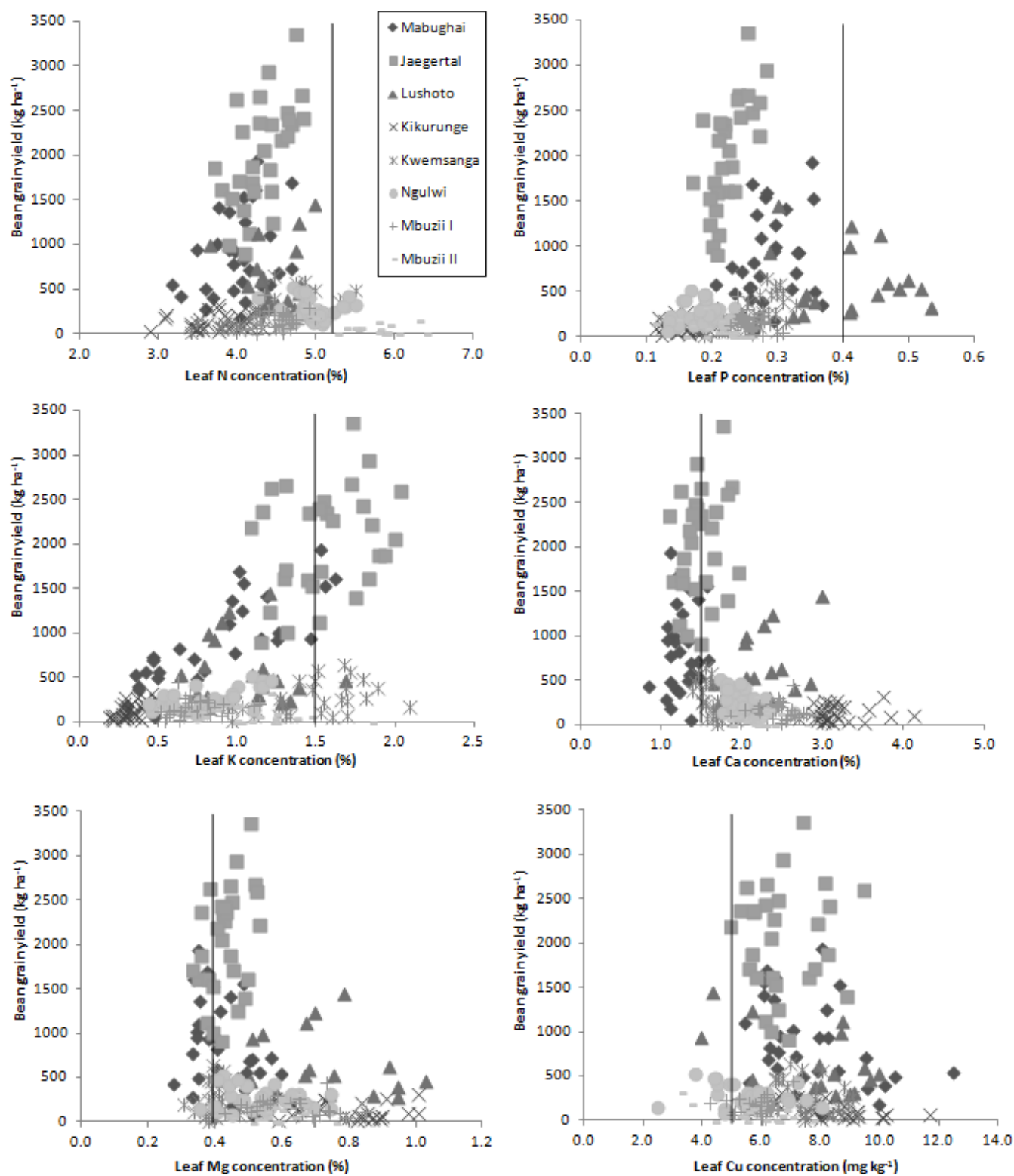
Relationships between grain yield and leaf nutrient concentrations differed between sites, where treatment effects became more clear for Mabughai and Jaegertal, which made up the major part of obtained yields above 750 kg ha⁻¹, and for Ngulwi (Table 4.5). Highly significant effects of K were found for leaf K concentrations when comparing among treatments at all three locations. Where K fertilizer application significantly increased leaf K concentration ($p < 0.001$). At both Mabughai and

Jaegertal, leaf N concentration was significantly increased by P fertilizer application ($p < 0.05$ and $p < 0.01$ respectively) and highest N concentrations were measured when P fertilizer was combined with K or I at Mabughai and with K+I and K+N at Jaegertal. However, leaf N concentration measured at Ngulwi, was significantly decreased by K ($p < 0.005$). P significantly increased leaf P concentration at Jaegertal ($p < 0.001$) and Ngulwi ($p < 0.01$). Whereas K gave a significant increase in leaf P concentration at Jaegertal ($p < 0.05$) and a significant decrease at Ngulwi ($p < 0.001$).

Leaf nutrient concentrations of Ca, Mg, Cu and Zn (which were not part of the fertilizer treatments) were generally lower after the application of P, K and/or N fertilizer, most likely due to dilution. At Mabughai, K significantly decreased leaf Mg ($p < 0.0001$), Ca ($p < 0.01$), Cu and Zn concentrations ($p < 0.05$). Where P also had a significant main effect on leaf Cu and Zn concentrations ($p < 0.05$).

However, inoculation significantly increased leaf Mg concentration ($p < 0.05$). When looking for significant treatment effects on those nutrients at Jaegertal, only Zn was significantly decreased by K ($p < 0.05$). At Ngulwi leaf Ca, Mg, Cu and Zn concentrations were significantly lower when K fertilizer was applied ($p < 0.05$, $p < 0.001$, $p < 0.001$ and $p < 0.01$ respectively). The same was true after addition of P fertilizer for leaf nutrient concentrations of Cu ($p < 0.05$) and Zn ($p < 0.001$).

The indicated relationships were also compared with CDCs for bean production obtained from literature (Reuter and Robinson, 1997) (Figure 4.6). Leaf N, P and Zn concentrations were all below the proposed CDC. However, this did not agree with the obtained relationships between grain yield and leaf nutrient concentration (Figure 4.6). Leaf nutrient concentrations of Ca, Mg and partly K, gave a clear increase in grain yield above the proposed CDC.



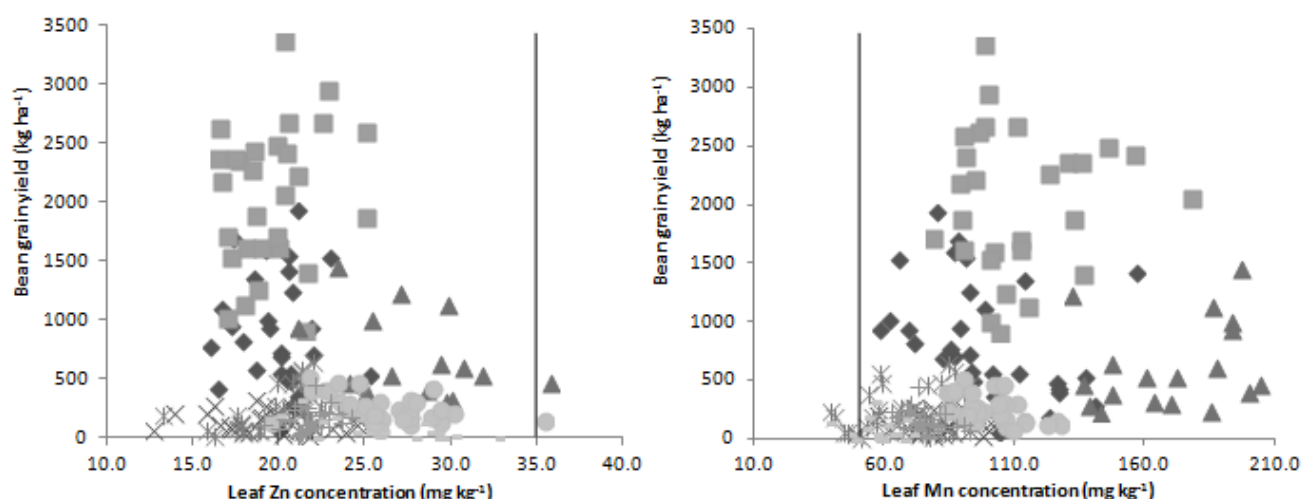


Figure 4.6 Bean grain yields as a function of determined leaf nutrient concentrations, for the nutrients N, P, K, Ca, Mg, Zn and Mn. Based on nutrient concentrations measured in the last developed mature bean leaf in the experimental fertilizer and inoculation trials. Different symbols are indicating the different locations where experiments were carried out. Vertical lines represent the critical nutrient concentration level for bean production, below which deficiencies can be assumed according to Reuter and Robinson (1997).

4.4.3 DRIS indices

DRIS indices were calculated for all plots used in the field trial (Table 4.5) as well as for the ten additional farmer fields (Table 4.6). After ranking the nutrients, it became clear that K, followed by P and N respectively, had the lowest DRIS indices overall, indicating relatively high importance of those nutrients in limiting yield. Some differences were observed between different locations, but the K-index ranked lowest for almost all locations. Where clearest results were obtained at Mabughai, Lushoto, Kikurunge, Mschizii, Ngulwi and Mbuzii I (Table 4.5) and for eight out of 10 farmer fields (Table 4.6). Whereas Zn and Mn had highest average DRIS index values, suggesting relatively low importance in limiting yield.

When comparing the DRIS indices with obtained grain yields, a clear and positive relationship was found between K-index and grain yield, but indices remained below zero, even for the highest yields obtained (Figure 4.7). A similar but weaker relationship was found for the P-index, where positive values were obtained at Lushoto and Mabughai. A negative response was found for Ca, Mg and Mn indices, where grain yield decreased with increasing index values. Those nutrients were not applied as fertilisers within the experiments. The N-index showed intermediate results and for the Zn-index no clear relationship with grain yield could be found, with all observations scoring index values well above zero (Figure 4.7).

Table 4.5 Leaf nutrient concentrations and DRIS indices for treatments at the individual field sites used in the experimental fertilizer and inoculation trials in the West-Usambara Mountains, Lushoto region (Tanga district, Tanzania).

Location	Treatment	Leaf nutrient concentration								DRIS indices						
		(%)					mg kg ⁻¹			N	P	K	Ca	Mg	Zn	Mn
		N	P	K	Ca	Mg	Cu	Zn	Mn							
Mabughai	Control	3.87	0.30	0.72	1.24	0.44	8.91	21.34	96.61	0.39	7.38	-118.71	35.50	24.49	22.59	28.36
	K	4.00	0.27	1.09	1.19	0.36	7.50	19.88	92.34	-6.96	-12.50	-54.29	17.90	13.83	10.27	31.76
	P	4.21	0.27	0.69	1.27	0.50	7.42	19.58	91.75	3.73	-1.79	-96.46	26.86	18.73	19.96	28.97
	I	3.89	0.30	0.52	1.43	0.53	10.16	22.64	108.56	4.85	12.04	-115.65	31.37	21.84	23.05	22.51
	K+P	4.46	0.27	1.19	1.16	0.35	5.99	17.76	91.28	-2.52	-11.84	-46.27	12.34	8.88	6.64	32.76
	K+I	3.88	0.26	1.00	1.20	0.37	7.25	19.62	97.80	-7.20	-13.35	-54.51	17.86	14.30	11.79	31.09
	P+I	4.45	0.29	0.43	1.39	0.52	7.60	20.46	92.36	13.60	12.12	-143.70	37.11	24.97	29.95	25.96
	K+P+I	3.81	0.30	1.26	1.01	0.32	6.73	18.30	98.92	-13.40	-9.82	-43.21	12.98	10.90	6.93	35.63
	LSD ¹	0.63	0.08	0.44	0.20	0.09	2.38	3.39	28.41							
LSD ¹	K+P+N	3.96	0.28	1.05	1.40	0.43	6.16	19.88	120.71	-3.96	-4.24	-42.38	11.26	9.16	7.40	22.75
	LSD ¹	0.51	0.08	0.68	0.31	0.10	1.54	4.15	32.17							
Jaegertal	Control	4.21	0.20	1.41	1.53	0.41	6.90	18.73	102.37	-0.60	-24.25	-23.15	9.85	6.50	5.99	25.66
	K	4.03	0.21	1.70	1.40	0.39	6.93	18.86	113.11	-4.98	-23.01	-15.78	7.77	5.41	3.83	26.75
	P	4.55	0.24	1.48	1.51	0.46	6.40	19.17	126.53	2.09	-13.32	-20.87	5.66	3.64	2.82	19.98
	I	4.09	0.21	1.31	1.47	0.44	6.98	19.56	105.70	-1.77	-19.33	-26.82	10.12	7.35	6.44	24.00
	K+P	4.38	0.25	1.68	1.45	0.44	6.58	20.05	108.38	-2.13	-14.98	-17.39	5.61	4.02	3.20	21.66
	K+I	4.16	0.21	1.79	1.41	0.41	6.77	21.39	123.26	-3.57	-21.90	-13.76	7.38	5.40	5.17	21.28
	P+I	4.56	0.21	1.27	1.41	0.41	5.34	17.14	171.72	5.08	-18.75	-29.18	7.25	5.04	5.90	24.65
	K+P+I	4.69	0.26	1.78	1.57	0.46	6.99	21.27	121.98	0.72	-12.15	-12.67	3.29	1.85	1.95	16.99
	LSD ¹	0.46	n.a. ²	n.a. ²	n.a. ²	n.a. ²	n.a. ²	n.a. ²	n.a. ²							
LSD ¹	K+P+N	5.42	0.24	1.61	1.38	0.42	6.88	19.36	105.68	3.97	-18.78	-23.28	6.54	3.70	2.59	25.26
	LSD ¹	2.54	0.041	0.60	0.29	0.064	1.60	1.60	14.67							
Critical nutrient range		5.2-5.4	0.4-0.6	1.5-3.5	1.5-2.5	0.4-0.8	5-15	35-100	50-400							

¹ Least significant differences were calculated based on the results of the analysis of variance with $\alpha=0.05$

² LSD not available due to one missing observation

Location	Treatment	Leaf nutrient concentration								DRIS indices						
		%					mg kg ⁻¹			N	P	K	Ca	Mg	Zn	Mn
		N	P	K	Ca	Mg	Cu	Zn	Mn							
Lushoto	Control	4.33	0.44	0.92	2.27	0.81	8.60	28.69	173.76	4.65	23.03	-45.80	4.46	3.48	5.00	5.20
	K	4.20	0.37	1.07	2.17	0.58	7.24	26.49	189.70	1.32	12.28	-33.89	5.01	4.18	4.04	7.08
	P	4.55	0.42	1.23	2.07	0.71	6.79	26.73	142.04	2.77	15.72	-33.92	2.52	1.55	3.48	7.88
	I	4.55	0.49	1.12	2.25	0.72	9.69	31.24	179.95	2.01	21.24	-31.40	1.21	0.86	2.67	3.42
	K+P	4.35	0.46	0.89	2.26	0.67	8.70	29.76	186.41	2.48	22.50	-43.18	4.62	3.75	4.67	5.15
	K+I	4.50	0.40	1.31	2.32	0.80	8.70	30.35	176.09	2.73	11.54	-20.51	-0.15	-0.39	1.60	5.18
	P+I	4.81	0.36	1.04	1.98	0.76	7.05	24.28	140.82	7.57	10.54	-37.03	3.66	2.86	3.96	8.44
	K+P+I	4.61	0.30	1.20	3.00	0.79	4.33	23.33	196.80	11.48	3.87	-17.60	0.24	-2.03	0.26	3.78
	LSD ¹	0.80	n.a. ³	n.a. ³	n.a. ³	n.a. ³	n.a. ³	n.a. ³	n.a. ³							
LSD ¹	K+P+N	4.77	0.30	1.20	3.00	0.79	4.33	23.33	196.80	8.18	9.65	-41.88	6.17	4.28	4.65	8.95
		1.50	n.a. ³	n.a. ³	n.a. ³	n.a. ³	n.a. ³	n.a. ³	n.a. ³							
Kikurunge	Control	3.59	0.16	0.29	3.21	0.86	8.42	21.93	79.55	22.03	-5.27	-165.09	34.68	27.94	60.08	25.64
	K	3.67	0.16	0.36	3.40	0.79	8.07	20.39	85.98	18.08	-8.45	-122.30	26.88	21.37	41.78	22.64
	P	3.88	0.17	0.28	3.00	0.87	8.8	19.91	69.37	25.11	-3.60	-191.41	37.00	26.95	76.28	29.65
	I	3.73	0.16	0.32	2.97	0.84	7.65	20.08	81.23	21.39	-7.09	-141.28	30.74	22.43	49.00	24.82
	K+P	3.83	0.15	0.4	2.99	0.64	6.73	18.98	82.65	16.78	-17.49	-111.44	26.97	22.35	37.52	25.31
	K+I	3.84	0.14	0.31	3.67	0.79	8.56	18.84	74.79	25.07	-20.66	-141.26	31.16	23.85	54.57	27.26
	P+I	3.45	0.16	0.22	3.03	0.84	10.31	17.97	83.10	29.26	0.25	-211.09	43.55	33.63	73.36	31.04
	K+P+I	3.72	0.15	0.34	3.19	0.68	7.71	18.19	80.16	19.06	-16.38	-139.32	30.86	25.22	51.16	29.41
	LSD ¹	0.52	0.036	0.11	0.44	0.14	1.94	4.8	18							
LSD ¹	K+P+N	3.96	0.17	0.41	3.02	0.62	6.93	17.56	90.41	17.90	-10.57	-107.05	24.16	19.15	31.57	24.84
		0.6	0.053	0.12	0.29	0.089	2.34	4.34	26.3							
Critical nutrient range		5.2-5.4	0.4-0.6	1.5-3.5	1.5-2.5	0.4-0.8	5-15	35-100	50-400							

¹ Least significant differences were calculated based on the results of the analysis of variance with $\alpha=0.05$

³ LSD not available due to two missing observation

Location	Treatment	Leaf nutrient concentration								DRIS indices						
		%					mg kg ⁻¹			N	P	K	Ca	Mg	Zn	Mn
		N	P	K	Ca	Mg	Cu	Zn	Mn							
Mschizii	Control	4.8	0.26	0.71	2.47	0.63	8.51	24.34	78.87	11.34	-1.38	-64.35	13.66	8.84	15.30	16.60
	K	4.6	0.23	0.79	2.33	0.60	7.90	21.31	80.34	8.39	-7.55	-55.34	13.31	8.52	13.12	19.55
	P	4.28	0.28	0.57	2.40	0.77	8.28	21.69	76.1	9.39	6.53	-86.08	17.00	12.22	21.17	19.77
	I	4.76	0.26	0.74	2.31	0.70	8.38	22.86	80.89	10.71	-2.45	-61.94	13.32	8.62	14.19	17.55
	K+P	4.82	0.26	0.77	2.16	0.61	7.85	23.35	87.67	9.46	-2.31	-58.48	12.83	8.75	12.93	16.83
	K+I	4.61	0.28	0.96	2.19	0.60	8.34	24.98	82.98	4.14	-3.08	-46.09	10.80	7.13	10.89	16.21
	P+I	4.36	0.35	0.61	2.62	0.81	9.02	23.46	103.3	9.99	18.69	-72.83	11.19	7.20	12.96	12.81
	K+P+I	4.16	0.3	0.71	2.35	0.66	8.47	23.45	113.71	5.41	7.29	-57.60	11.24	8.25	11.05	14.37
LSD ¹		0.88	0.1	0.28	0.49	0.25	2.17	4.68	27.46							
	K+P+N	4.74	0.25	0.95	2.33	0.61	6.66	21.36	89.9	7.95	-6.59	-40.26	9.44	5.38	7.52	16.58
LSD ¹		2.57	0.18	0.38	0.47	0.28	5.32	16.29	26.49							
Kwemsanga	Control	3.99	0.23	1.12	1.82	0.42	7.49	18.65	60.20	-7.60	-18.90	-44.19	14.28	9.12	11.95	35.35
	K	4.32	0.27	1.79	1.63	0.40	8.03	21.45	76.11	-6.79	-14.16	-17.69	6.90	3.64	3.54	24.55
	P	5.24	0.30	1.21	1.65	0.44	7.80	22.01	80.44	4.31	-6.62	-39.18	8.75	5.45	6.30	20.99
	I	4.45	0.28	1.63	1.66	0.40	7.84	21.33	62.18	-8.00	-13.54	-25.21	8.88	4.52	4.57	28.77
	K+P	4.68	0.27	1.67	1.50	0.39	7.40	19.94	78.90	-3.26	-14.65	-21.50	6.38	3.42	2.84	26.77
	K+I	4.54	0.27	1.56	1.65	0.39	8.08	20.99	57.54	-7.51	-15.76	-29.10	10.23	5.18	5.23	31.72
	P+I	4.65	0.28	1.52	1.61	0.41	7.48	20.04	60.95	-4.38	-13.62	-29.64	8.62	4.64	4.47	29.91
	K+P+I	4.62	0.25	1.36	1.69	0.37	7.49	17.16	64.28	-3.16	-19.35	-34.95	10.06	3.92	2.43	41.04
LSD ¹		0.76	0.05	0.30	0.16	0.06	0.88	4.06	23.37							
	K+P+N	4.41	0.28	1.43	1.66	0.38	7.22	20.80	70.57	-6.30	-11.94	-30.39	9.53	5.50	6.11	27.49
LSD ¹		0.86	0.08	0.30	0.21	0.07	1.12	7.15	30.94							
Critical nutrient range		5.2-5.4	0.4-0.6	1.5-3.5	1.5-2.5	0.4-0.8	5-15	35-100	50-400							

¹ Least significant differences were calculated based on the results of the analysis of variance with $\alpha=0.05$

Location	Treatment	Leaf nutrient concentration								DRIS indices						
		%					mg kg ⁻¹			N	P	K	Ca	Mg	Zn	Mn
		N	P	K	Ca	Mg	Cu	Zn	Mn							
Ngulwi	Control	5.07	0.18	0.58	2.17	0.59	6.86	29.70	51.01	20.85	-19.82	-81.88	22.69	17.29	24.07	16.80
	K	4.89	0.15	0.84	1.92	0.43	5.69	27.97	97.78	13.17	-36.56	-52.11	20.91	17.36	18.81	18.41
	P	5.23	0.20	0.58	2.12	0.64	5.76	26.21	105.72	21.12	-11.03	-77.02	19.00	13.50	19.02	15.39
	I	4.92	0.19	0.68	2.05	0.56	5.87	26.88	114.22	15.85	-16.02	-63.25	18.10	13.64	16.44	15.25
	K+P	4.68	0.17	1.03	1.84	0.48	4.80	22.80	87.26	7.76	-29.00	-38.28	15.07	10.51	13.20	20.76
	K+I	4.70	0.15	0.79	2.20	0.50	5.19	25.71	104.84	14.63	-32.69	-50.01	18.77	14.66	17.27	17.38
	P+I	5.14	0.21	0.64	2.00	0.62	6.09	25.54	100.73	14.63	-32.69	-50.01	18.77	14.66	17.27	17.38
	K+P+I	4.73	0.17	1.03	1.85	0.47	4.61	23.16	92.80	8.54	-28.65	-38.37	15.05	10.65	12.74	20.04
LSD ¹		0.40	0.03	0.23	0.31	0.13	1.26	3.15	19.86							
LSD ¹	K+P+N	4.80	0.17	1.15	1.85	0.41	3.77	22.51	100.62	7.81	-30.28	-31.94	13.88	9.93	10.03	20.56
		0.41	0.03	0.26	0.28	0.12	1.47	2.84	6.63							
Mbuzii I	Control	4.55	0.25	0.71	2.31	0.60	6.15	22.28	74.14	7.90	-3.84	-69.24	15.90	10.81	17.81	20.67
	K	4.42	0.24	0.67	2.52	0.63	5.96	21.92	76.56	8.69	-3.22	-69.98	16.12	10.50	17.84	20.05
	P	4.49	0.28	0.75	2.33	0.64	6.04	22.65	74.54	6.38	2.50	-68.16	14.33	9.59	16.28	19.09
	I	4.66	0.25	0.75	2.25	0.61	6.03	22.64	77.27	8.43	-4.25	-62.86	14.38	9.66	15.50	19.14
	K+P	4.35	0.26	0.76	2.64	0.68	5.52	21.60	73.88	6.37	-1.59	-59.47	13.73	8.18	13.61	19.16
	K+I	4.41	0.25	0.69	2.70	0.71	5.99	21.19	73.24	8.80	-1.96	-66.61	14.91	9.01	15.87	19.98
	P+I	4.71	0.26	0.84	2.30	0.56	6.11	22.34	72.76	6.49	-4.57	-58.37	13.89	8.76	13.46	20.33
	K+P+I	4.51	0.26	0.75	2.49	0.63	5.60	21.00	76.05	7.57	-1.14	-63.15	13.78	8.54	14.30	20.10
LSD ¹		0.32	0.02	0.15	2.27	0.11	0.75	1.65	12.49							
LSD ¹	K+P+N	4.60	0.25	0.87	2.49	0.61	4.84	20.23	81.74	7.65	-4.59	-49.84	11.70	6.17	9.20	19.70
		0.39	0.02	0.27	0.41	0.08	1.22	2.84	21.38							
<i>Critical nutrient range</i>		5.2-5.4	0.4-0.6	1.5-3.5	1.5-2.5	0.4-0.8	5-15	35-100	50-400							

¹ Least significant differences were calculated based on the results of the analysis of variance with $\alpha=0.05$

Location	Treatment	Leaf nutrient concentration								DRIS indices						
		%					mg kg ⁻¹			N	P	K	Ca	Mg	Zn	Mn
		N	P	K	Ca	Mg	Cu	Zn	Mn							
Mbuzii II	Control	6.09	0.15	0.98	2.21	0.57	6.20	24.67	48.42	24.09	-60.30	-58.49	22.44	13.03	31.27	27.97
	K	5.94	0.22	1.53	2.10	0.54	5.95	31.01	52.42	7.78	-28.02	-28.45	12.81	6.48	10.85	18.56
	P	5.62	0.23	1.05	2.09	0.56	6.42	27.07	60.95	9.83	-17.96	-45.78	13.41	7.86	13.47	19.17
	I	6.13	0.19	1.04	2.22	0.57	6.42	29.61	52.74	16.13	-34.63	-48.51	17.14	9.66	19.97	20.25
	K+P	5.24	0.22	1.16	2.17	0.55	5.31	24.49	61.82	7.03	-20.31	-37.66	12.55	6.87	10.72	20.82
	K+I	5.66	0.18	1.11	2.12	0.50	6.01	28.68	53.31	11.69	-34.68	-50.40	18.31	11.28	21.32	22.49
	P+I	5.70	0.21	1.00	2.21	0.64	6.02	24.84	61.14	14.40	-22.23	-48.54	14.14	8.11	13.97	20.16
	K+P+I	5.36	0.19	0.99	2.18	0.59	4.78	23.82	54.28	11.50	-31.50	-48.78	16.49	9.60	17.63	25.06
LSD ¹		0.86	0.10	0.52	0.40	0.09	1.97	6.25	17.23							
	K+P+N	5.12	0.20	1.21	2.14	0.70	3.76	21.53	61.06	8.24	-22.83	-31.58	10.24	6.00	7.43	22.51
LSD ¹		2.09	0.17	0.72	0.70	0.22	3.84	16.53	13.62							
Adequate range ⁴		5.2-5.4	0.4-0.6	1.5-3.5	1.5-2.5	0.4-0.8	5-15	35-100	50-400							

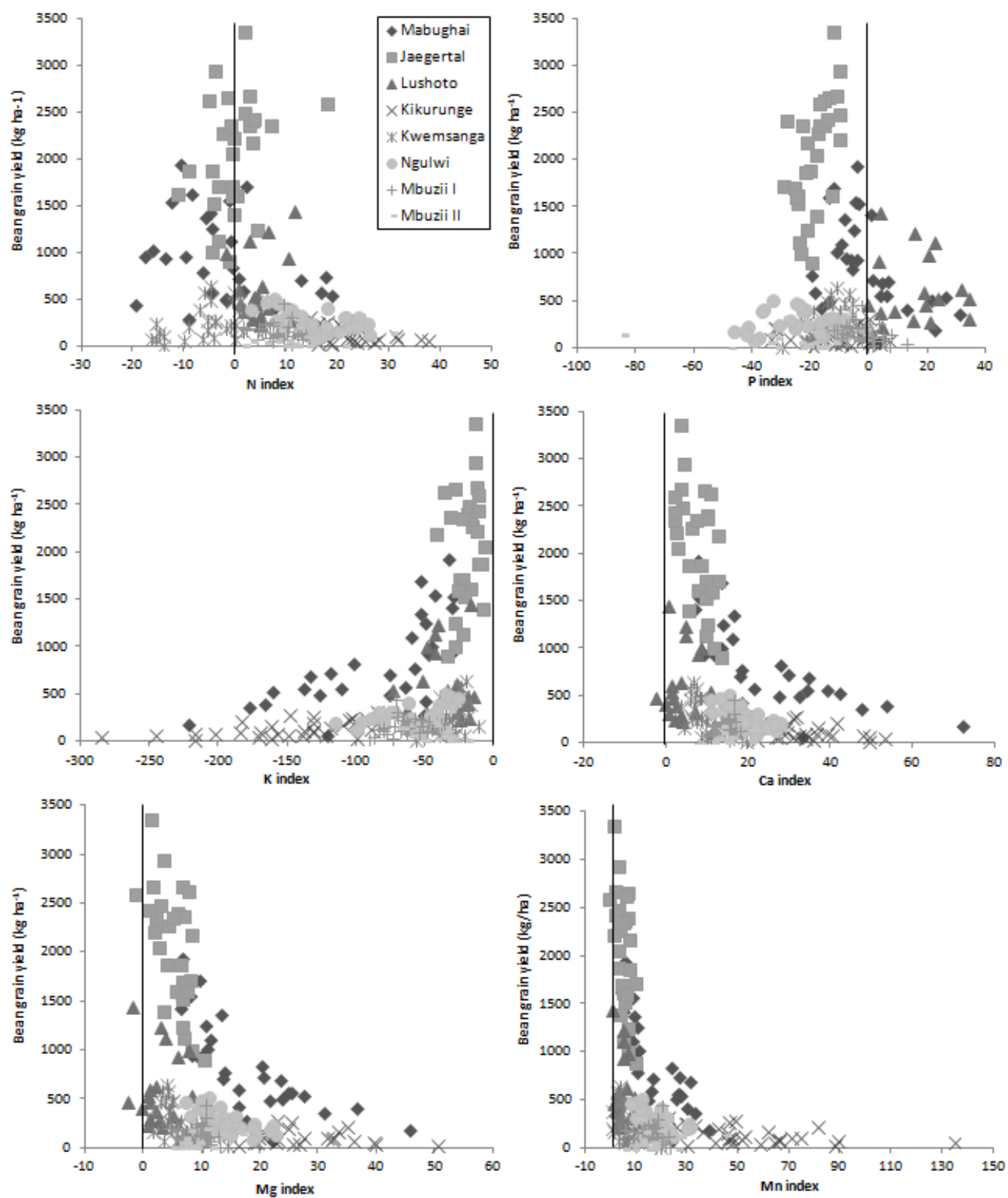
¹ Least significant differences were calculated based on the results of the analysis of variance with $\alpha=0.05$

⁴ Adequate ranges for bean leaf nutrient concentrations were obtained from literature (Reuter and Robinson, 1997).

Table 4.6 Bean leaf nutrient concentrations and DRIS indices measured at ten farmer fields in the West-Usambara Mountains, Lushoto region (Tanzania).

Farmer Field (FF)	Leaf nutrient concentrations								DRIS indices						
	%					mg kg ⁻¹			N	P	K	Ca	Mg	Zn	Mn
	N	P	K	Ca	Mg	Cu	Zn	Mn							
FF1	4.80	0.32	1.61	2.24	0.59	11.39	24.47	86.92	-0.73	-3.65	-15.61	2.23	-0.21	14.48	3.49
FF2	3.80	0.23	1.05	1.77	0.47	8.64	18.76	61.65	-9.66	-16.50	-42.52	11.12	7.80	33.41	16.35
FF3	5.49	0.17	1.01	2.13	0.53	7.95	27.46	97.46	5.58	-23.45	-32.22	12.48	8.40	15.42	13.79
FF4	5.25	0.13	0.92	2.33	0.57	6.30	21.87	403.37	10.78	-22.61	-19.82	8.02	6.56	9.86	7.21
FF5	4.09	0.17	0.64	1.99	0.50	5.49	25.23	116.08	0.69	-20.46	-57.31	21.40	17.91	18.26	19.51
FF6	4.17	0.16	1.00	1.70	0.47	4.93	17.24	45.83	-8.84	-40.26	-52.45	15.71	10.16	46.69	28.98
FF7	3.89	0.19	0.41	1.99	0.65	10.14	30.41	61.15	-4.82	-19.38	-119.81	32.42	22.88	25.26	63.46
FF8	5.09	0.31	0.64	2.44	0.82	8.65	27.43	66.81	2.03	-2.38	-62.73	10.88	6.86	17.35	27.99
FF9	5.55	0.21	0.63	2.46	0.77	6.68	30.51	61.57	4.43	-15.50	-67.25	14.19	8.91	18.55	36.67
FF10	5.58	0.31	3.07	2.36	0.49	4.18	32.76	39.51	-7.66	-19.36	-3.78	2.09	-1.87	20.61	9.96
Adequate range ¹	5.2-5.4	0.4-0.6	1.5-3.5	1.5-2.5	0.4-0.8	5.0-15.0	35-100	50-400							

¹ Adequate ranges for bean leaf nutrient concentrations were obtained from literature (Reuter and Robinson, 1997).



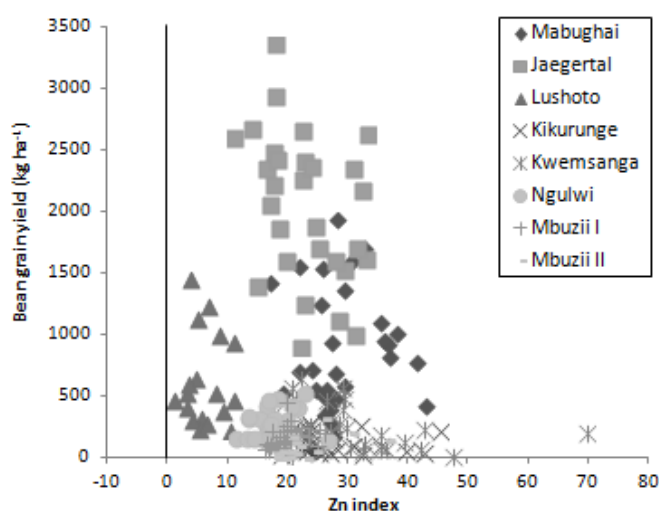


Figure 4.7 Bean grain yields as a function of calculated DRIS indices for the nutrients N, P, K, Ca, Mg, Mn and Zn. Based on nutrient concentrations measured in the lastly developed mature bean leaf in the experimental fertiliser and inoculation trials. The vertical line represents the zero index and different symbols are indicating different locations.

4.6 Interviews

Each farmer managed several plots ranging from three plots at Mabughai and Mbuzii II to 14 in co-op at Jaegertal (Table 4.7). The farmers with fields located on the slopes mainly cultivated maize, beans and some cassava. Farmers with additional fields in the valleys also cultivated cash crops like cabbage, carrot, tomato, potatoes, plantain and broccoli. Cows, sheep and goats served as the main livestock in the Lushoto region. The farmers at Jaegertal, Lushoto, Mschizii, Kwemsanga, Ngulwi, Mbuzii I and Mbuzii II took care of three cows on average. The farmers at Mschizii, Kwemsanga, Ngulwi and Mbuzii II also had two to seven goats or sheep. Livestock was fed with grasses and leftovers of the harvest. Animal manure was collected if possible but also purchased from outside the farm and mainly applied to maize and vegetables. Mineral fertilizers were used by some farmers, in the form of DAP, NPK or Urea and especially applied to high value crops but also to maize at Kikurunge and Mschizii. Farmers were not used to apply manure or mineral fertilizers to beans (Table 4.7).

Maize and bean were the main crops grown during the masika season in 2013. The farmers with fields at lower elevations indicated that the importance of the long rain masika season has increased over the years and that they often leave their land fallow during the vuli season, especially when irrigation is not possible (Table 4.8). The farmers involved in the experimental field trials normally planted local bean varieties (e.g. *Rosekoko* and *Soya*) and seeds often originated from their own stock or the local market. Farmers pointed at drought, root rot and aphids as the main causes of problems in the cultivation of beans. Problems with erosion were mainly indicated by farmers with fields on the steeper slopes. They often implemented erosion prevention techniques like creating terraces or planting grass strips at the field margins. The farmers judged the soil drainage of their fields as moderate or good (Table 4.8).

Table 4.7 General farm characteristics for each site used in the experimental trials in Lushoto region (Tanzania).

Location	number of plots	Crops produced	Livestock		Manure		Mineral fertilizer	
			Cows	Goats Sheep	Origin	Use	Type	Use
Mabughai	3	Maize, potatoes	0	0	purchased	maize, potatoes	DAP	Vegetables
Jaegertal	14 (co-op)	Maize, bean, potato, cabbage, carrot, tomato, brocolli, lettuce	4	0	livestock and purchased	maize, vegetables	DAP	Vegetables
Lushoto	-	Depended on research	7	0	livestock	Depended on research	DAP	Vegetables
Kikurunge	-	Maize, bean, cassava	0	0	purchased	Maize	DAP	mais
Mschizii	6	Maize, bean, tomato, cassava, cabbage, plantain	1	3	livestock and purchased	maize, vegetables	NPK	vegetables, mais
Kwemsanga	9	Maize, bean, carrot, tomato, cassava, cabbage, brocolli, lettuce, zuchini	4	3	livestock and purchased	maize, vegetables	Urea	vegetables
Ngulwi	4	Maize, bean, potato, plantain, cassava	2	7	livestock	Maize	-	-
Mbuzii I	5	Maize, bean, snappea, tomato	3	0	livestock	Maize	-	-
Mbuzii II	3	Maize, bean, cassava	3	2	livestock	maize	-	-

Table 4.8 Field crop history and characteristics of the specific sites used in the experimental trials in the Lushoto region (Tanga district, Tanzania).

Location	Cropping history			Problems in bean production	Perception of soil fertility by the farmer	Signs of soil erosion	Prevention techniques	Perception soil drainage
	Masika 2013	Vuli 2012	Bean variety					
Mabughai	maize, bean	maize, bean	-	Stunted growth	low	No	No	Good
Jaegertal	potatoes	Maize	Rosekoko	Aphids	good	No	No	Good
Lushoto	lettuce	zucchini, leek	-	-	moderate	No	Terraces, grass strips	Moderate ²
Kikurunge	maize, bean	Fallow	Jeska (a.o.)	Aphids	moderate	Yes	grass strips	Moderate
Mschizii	maize, bean	Bean	-	aphids, root rot	low	Yes	No	Good
Kwemsanga	maize, bean	Fallow	Rosekoko, Soya	Aphids, waterlogging	low	Yes	Grass strips	Good
Ngulwi	Fallow ¹	maize, bean	Soya, Injarna	Aphids, root rot	good	No	Grass strips	Good
Mbuzii I	Maize	fallow	Rosekoko, Soya	Drought	good	No	Terraces	Good
Mbuzii II	Bean	fallow	Soya	Root rot, diseases	poor	Yes	No	Moderate

¹ Land was left fallow as the farmer was not able to cultivate the land due to illness.² Problems with drought occurred more often close to trees at the borders of the field plot.

5. Discussion and Conclusion

5.1 Nutrient deficiencies

The effects of P, K, additional N fertilizer and inoculation were examined at field trials in the West-Usambara Mountains located in northern Tanzania during the Vuli season (2013), to diagnose nutrient limitations in bean production. Prior to seeding, soil analysis indicated deficiencies of the nutrients P, K and partly of Ca and Mg. Soil analyses carried out in the same area by Smithson et al. (1993) and Ndakidemi and Semoka (2006) showed soils mainly poor in P and K, limiting bean growth and productivity. Growth and yield results indicated differences in treatment responses, however effects were not that clear at all nine experimental fields. Sites where nutrients were clearly limiting revealed responses to fertilizers P and K, accompanied by more pods per m² and increased number of seeds per pod. Analysis of bean leaf tissue indicated deficient nutrient concentrations levels for P, K, N and Zn when compared with critical nutrient concentration ranges obtained from literature (Reuter and Robinson, 1997). At some sites application of P and K (and partially N) fertilizers increased leaf concentrations of the respective elements but depressed the concentrations of Ca, Mg, Cu and Zn (Table 4.5). Improved plant growth, initiated by the addition of K, P and/or N fertilizer(s), was likely to cause a dilution in the leaf tissue of the other major and minor nutrients not included in the experimental trials. Measured leaf Ca, Mg and partially N and Zn concentration in relation to bean grain yield showed a C-shaped response curve, which could be referred to as the 'Piper-Steenbjerg' effect (Figure 5.1, Bates, 1971). Where leaf nutrient concentration decreases with increase in bean grain yield at the bottom of the curve. A lack of remobilization from old leaves and stem is given as a possible explanation for this type of response by Reuter et al. (1981). Leaf nutrient analysis was also carried out for 10 neighbouring farmer fields next to the experimental field sites, planted with local bean varieties and management by smallholder farmers. Measured leaf nutrient concentrations pointed at major limitations of the elements K and P, followed by N and Zn, when being compared to the adequate nutrient range (Table 4.6).

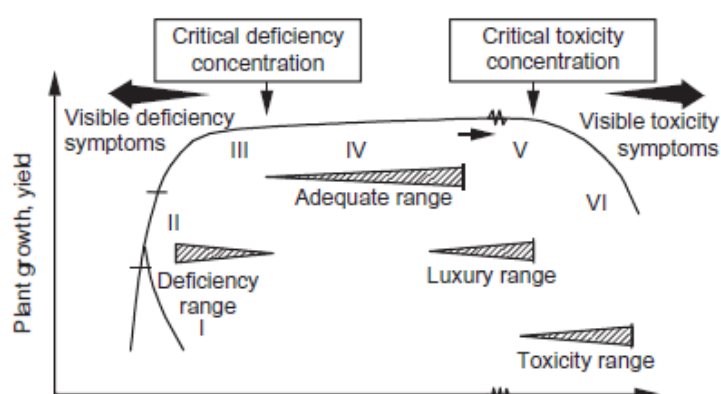


Figure 5.1 Relationship between nutrient concentrations and growth or yield (Marschner, 2011).

Ranking the obtained leaf nutrient concentrations with the DRIS approach, showed consistent results for nutrient deficiencies P and K (partly N). With K as the most limiting nutrient, followed by P and N respectively (Table 4.5 and 4.6). However, Zn was not indicated as one of the nutrients limiting bean production by the DRIS approach. Reuter and Robinson (1997) proposed a wide critical nutrient concentration range for Zn from 35-100 mg kg⁻¹. Only values lower than the minimum Zn level of 35

mg kg⁻¹ were obtained within this study, even at the relatively high yielding plots. Smithson et al. (1993) applied Zn as a trace element at Irete (West-Usambara Mountains), but recorded no or only slight effects on any of the characteristics recorded.

Besides quantitative results, possible nutrient deficiencies were also visually analysed in the field during the experiments (Appendix V). Severe K deficiency became visible at some fields through chlorotic and necrotic leaf symptoms, earlier referred to as 'Usambara mottle' in beans by Smithson et al. (1993). K is the nutrient required in the largest amount by plants and when K is deficient, growth is retarded, enhancing net transport of K⁺ from mature leaves and stems (Marschner and Cakmak, 1989). Leaf senescence is induced by K deficiency (Armengaud et al., 2004) and in the form of leaf chlorosis of source leaves, it can readily be induced by high light intensity combined with K, Mg and/or Zn deficiency (Marschner and Cakmak, 1989). P requirements for optimal growth are highest during the vegetative growth phase as P functions as a structural element within plants. It is the main component of macromolecular structures like nucleic acids. Concentrations are therefore highest in expanding and newly formed leaves, where P is needed for rapid protein synthesis (Marschner, 2011). In the experimental field trials, lack of P resulted in reduced crop vigour, where both number of leaves as well as expansion of leaves was decreased. Furthermore the leaves had a darker green colour which can be explained as leaf expansion was probably more inhibited than chlorophyll formation (Hecht-Buchholz, 1967).

Addition of P (26 P kg ha⁻¹) and K (25 K kg ha⁻¹) fertilizers did not fully alleviate P and K deficiency, according to the leaf nutrient concentrations and related DRIS indices. However, increasing the application rate would hardly be economically feasible for smallholder farmers in the Usambara region. And it is likely that with an adequate and repeated supply of organic and/or inorganic fertilizers nutrient reserves will accumulate in the soil over the years, to give residual benefits.

5.1.3 Nutrient deficiencies in relation to N₂-fixation

Within the experiments, nodulation score was significantly increased when both P and K fertilizers were applied in combination with or sometimes even without *Rhizobium* inoculation. Deficiencies in essential plant nutrients are able to cause reductions in the number and size of root nodule formation and in the total amount of N₂ fixed (Giller, 2001). Essential mineral nutrients needed for symbiotic legume nitrogen fixation are those required for normal establishment and adequate functioning of the symbiosis. Many macro- and micronutrients are involved in the interactions process, either as a constituent of an essential element or required for enzyme activity (O'Hara et al., 1988). Phosphorus (together with sulphur) is mainly required for nodule metabolism, where capture and uptake of phosphorus depends on the root geometry and the interaction with mycorrhizas (which can assist in the uptake of phosphorus by increasing the volume of soil effectively explored by the plant) (O'Hara et al., 1988). Giller et al. (1998) also suggested that poor plant vigour and nodulation of *Phaseolus* in the bean growing areas of Tanzania, was related to the variation of extractable soil P concentration. Large indigenous populations of *Rhizobium* in those area were reported but nodulation remained poor. No direct role for potassium on N₂-fixation has been reported, but the element is of major importance for adequate plant growth and development, underlying a successful symbiosis.

5.2 Overriding yield limiting factors

The results indicated that besides the use of an improved seed variety, fertilizers and inoculant, other factors played a role in the determination of final bean yield, as major differences between locations were observed. Six out of nine sites were not clearly responding to treatment inputs and yields were very poor. This could partially be explained by differences in altitude and rainfall distribution, affecting the local climate within the experimental area (Table 3.1; Figure 3.3). Fields located in Ngulwi, Mbuzii I and II, Kikurunge, Mschizii and Kwemsanga were located at the somewhat lower altitudes and received relatively low amounts of precipitation, especially during the middle and second part of the growing season. The fields at Mabughai and Jaegertal, where the highest yields were obtained, plots were irrigated once and twice respectively during the growing season, when rain did not seem to be sufficient for bean growth. In general, nutrient availability in the topsoil in dry climates declines during the growing season because low soil water content becomes a limiting factor for nutrient delivery to the root surface and nutrient uptake will further decrease by impaired root growth in dry soil (Marschner, 2011). It could therefore be possible that N, P and K applied through fertilizers within this experiment became less available at drier locations where soil moisture content was relatively low. The uptake of K by the plant decreases with a decrease in soil water content due to low K mobility (Kuchenbuch et al., 1986). In addition, when K is limiting, plants become more susceptible to abiotic and biotic stresses and will also be more sensitive to drought due to several factors; oxidative stress avoidance, stomata regulation (major mechanisms controlling the water regime of higher plants) and high osmotic pressure in the vacuoles (Marschner, 2011). In the case of P, only about 10-20% of P applied is generally taken up by the first crop, and P applied through (TSP) fertilizer need to be provided in the soil solution to be available for plant uptake (Chien et al., 2011). So with inadequate water supply by rainfall and/or irrigation P uptake by the plant can even be lower than the proposed 10-20%.

High temperatures and relatively low soil water contents also affect nodulation. The numbers of rhizobia in soil decline as soil dries and drought stress drastically affects N_2 -fixation in legumes. Rates of N_2 -fixation are found to be more sensitive to reductions in soil water content than other plant physical and/or chemical processes (Giller, 2001).

Symptoms of root rot infestation were observed within plots at Mbuzii I and II and Mshizii after germination. Some plants were able to recover (by the development of new roots) but others were not even able to survive or produced poor yields (Appendix V). Bean root rots (*Pythium* spp., *Fusarium solani* subsp. *phaseoli*, *Rhizoctonia solani*) are triggered by particular climatic conditions and have a major impact on bean yields in Africa (Otsyula, 1994). The impact of the fungal disease varies through time with relatively low incidence in some years and entire crops wiped out in others (CIAT, 1992). Its distribution and severity throughout East Africa is related to the intensity of bean cultivation, human population density, soil properties and rainfall (Otsyula and Buruchara, 2001; Wortmann et al., 1998). Root rot infestation requires high soil moisture in the root-zone since the most important pathogen (*Pythium* spp.) is water-borne (Pieczarka and Abawi, 1978). The critical period for root rot infestation is immediately after germination, during the first weeks of plant development (Farrow et al., 2011). Around this time, major rainfall events took place at Mbuzii and Mshizii followed by a period of drought.

5.3 Leaf nutrient concentrations and the DRIS approach

Bean leaves were collected in the field at the moment of approximately 50% flowering. Differences in moment of flowering between treatments and/or locations have been observed. Translocation of nutrients to the storage organs (filling of the pods) sets in after flowering as the leaf undergoes a shift in which its function changes from a sink to a source of both nutrients and products from the photosynthesis process. Early induced flowering can be the result of stress factors, like drought, pest and diseases and/or nutrient deficiencies (Marschner, 2011). Nutrient concentration values were obtained through laboratory analysis of the leaf tissue. Values obtained in this way should never be regarded as absolute, but only as representative values of a possible range that is influenced by many uncontrolled and unknown factors. This also means that critical values obtained from literature are not without error and can only function as a rough guide when interpreting the data obtained during the experiments (Bergmann, 1992). The results in this study showed that relatively high yields could still be reached, even though the measured nutrient concentrations were lower than the associated critical nutrient value indicated by literature. This was especially true for the leaf nutrient concentrations of Zn (Figure 4.5).

The moment of leaf collection in the field can also be of major importance during the interpretation of the measured nutrient concentration. As extreme weather events and/or the developmental stage of the plant affect nutrient transport and allocation through the plant. Leaf potassium levels, for example, are expected to be low after prolonged periods of drought, but higher after regular rainfall events (Bergmann, 1992).

The DRIS approach was used to convert the obtained nutrient concentrations into DRIS indices. Alkoshab et al. (1988) indicated that DRIS can best be viewed as a supplement to sufficiency range diagnosis that provides additional information when severe imbalances are detected. DRIS results obtained in this study gave a clear overview of the nutrients expected to be most limiting; K and P respectively (Figure 4.6). For the nutrients Ca, Mg and Mn the DRIS indices decreased towards zero with increasing yields, as they were not included in the experimental treatments. The DRIS indices determined for Zn, showed no clear pattern and they were highly positive. To determine the possible limitation of Zn in bean growth and yield in the Usambara Mountains, further research needs to be done, taking Zn fertilization into account.

5.4 Socio-economic factors and agronomic performance

Farmers involved in the experimental trials in the Usambara region were not used to apply any type of manure and or *Rhizobium* inoculant to beans. As common bean is usually grown for household consumption and in some cases thought to play a role in maintaining or improving soil fertility (Mbaga-Semgalawe and Folmer, 2000). Animal manure and some chemical fertilizers (mainly NPK and DAP) are traditionally applied to cash crops like cabbage, tomatoes and potatoes but also to maize. Intercropping maize and bean is a common farmer practice in the Usambara region as they make up the main ingredients in household food provision. In this way bean can still profit from manure applied to maize. Farmers involved in the interviews also noticed the beneficial effects for bean when intercropped with maize. The heterogeneity among farmers involved in the experimental trials reflected the differences between farmers in the region. The farmers at Mabughai, Jaegertal and Lushoto were most wealthy in terms of knowledge, resources and investment capacity and managed several fields on the flat areas, less prone to erosion. The highest yields obtained in the experiment, were achieved at those sites. One of the most important differences with the other

farmers is that they had the possibility to irrigate during the growing season, when rainfall turns out to be inadequate. Furthermore they were more used to apply chemical fertilizers, as they grow a variety of crops, including cash crops with high input needs. The production and marketing of cash crops generates more income for the household, which can be used for general agricultural investments (livestock for animal manure, high quality seeds, chemical fertilizers, plant protection), but also for education of the children, machinery and extra labour for example. Mowo et al. (2006) studied the role of smallholder farmer community in soil fertility evaluation and management in northern Tanzania. They indicated that with an increase of income and knowledge, farmers were more likely to invest in soil fertility management. The distribution of high quality inputs throughout the Usambara region improved over the years, but there is still a need to inform farmers in which inputs they need the most and how they need to use them.

5.5 Implications for N2Africa

Results obtained within the experimental study in combination with the farmer interviews, indicated some implications for N2Africa research within this region in future:

- There is a need to increase the use of animal manure and/or fertilizers based on potassium and phosphorus deficiencies in the Usambara region. To be able to increase soil fertility and to maintain agricultural production on the long run. Fertilizers already used in the region are mostly based on N and P, so there need to be special attention for the application of K fertilizers.
- *Rhizobium* inoculation used in the experimental trials, gave poor or mixed results, probably due to low soil moisture contents and poor plant growth. The experiments in this study were carried out in the short rainy season (vuli) it would therefore be good to repeat similar experimental trials and/or dissemination trials in the long rainy season (masika) to look for possible differences, so concerning the availability of nutrients applied through fertilizers.
- The combination of soil analysis and leaf nutrient concentration analysis showed to be of value for the final results of this study. The use of the DRIS approach to rank nutrients according to their degree in limiting bean yield was useful in the visualization and interpretation of the leaf nutrient concentration results.

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Appendix I - Experimental plan of the field experiment

Design: Factorial experiment

Factors and Levels:

Phosphorus fertilizer (P):	P = 26 kg ha ⁻¹ as triple superphosphate (P)
Potassium fertilizer (K):	K = 25 kg ha ⁻¹ as muriate of potash (K)
Nitrogen fertilizer (N):	N = 25 kg ha ⁻¹ as calcium ammonium nitrate (N)
Rhizobia inoculation (Inoc):	Inoc = Rhizobia inoculant containing <i>Rhizobium</i> strain CIAT-899 inoculant contains at least 8-10 ⁹ cells/g of <i>Rhizobium</i>

Treatments (at least one replicate, two or even three when possible):

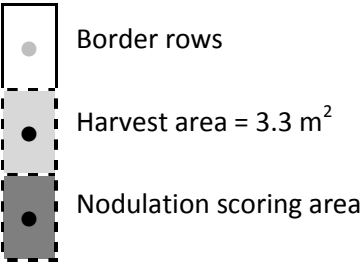
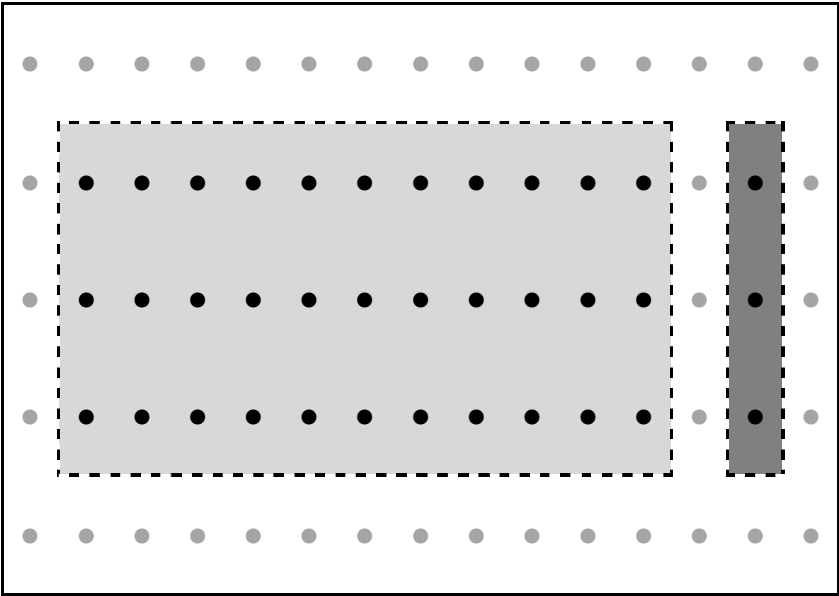
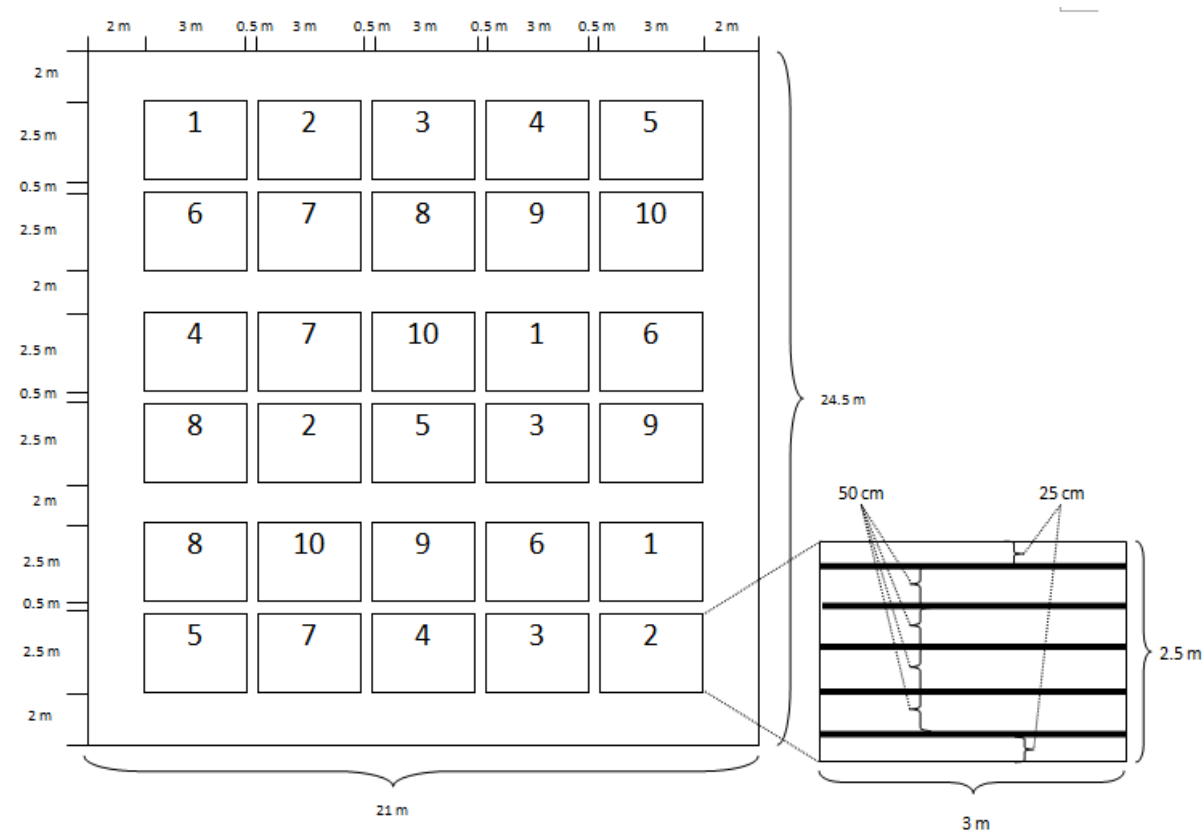
- 1: control 1 (no fertilizer or inoculant added)
- 2: control 2 (no fertilizer or inoculant added)
- 3: K
- 4: P
- 5: Inoc
- 6: K + P
- 7: K + Inoc
- 8: P + Inoc
- 9: K + P + Inoc
- 10: N + P + K

Further specifications:

Locations:	In agreement with local supervisor and farmers
Sowing date:	In between 7-19 th of November, 2013
Harvesting date:	In between 27 th of January and 12 th of February 2014
Row spacing:	50 cm, 5 rows per 2.5 m
Sowing density:	50 x 10 cm = 2 x 10 ⁵ plants ha ⁻¹
Sowing depth:	3-4 cm
Disease control:	-
Weed control:	Hand weeding
Fertilization:	According to the treatment
Dimension field:	parcel: 17 m x 21 (in the case of 2 replicates), net dimension experiment 10 m x 15 m exc. outer borders 24.5 m x 21 m (in the case of 3 replicates), net dimensions experiment: 15 m x 15 m exc. outer borders
Dimension gross plots:	150 m ² (in the case of 2 replicates) and 225 m ² (in the case of 3 replicates)
Dimension net plot:	7.5 m ²

Coordinates of the net field corners were recorded

Layout (not on scale)



Appendix II - Preparation experimental inputs

Calculations fertilizers

N as CAN (Calcium Ammonium Nitrate)

Treatment: 25 kg N/ha

CAN contains 27% N $> 25 * (100/27) = 92.59$ kg CAN/ha

- CAN applied per plot (7.5 m²)

$92.59 \text{ kg}/10000 \text{ m}^2 = 9.259 \text{ g/m}^2$

CAN per plot (7.5 m²) $= 9.259 * 7.5 = 69.44$ g CAN/plot

- CAN per replica block (1 plot) = 69.44 g CAN/replica

- CAN per field (3 rep.) $= 69.44 * 3 = 208.33$ g/field

- Total CAN (10 fields) = 2.08 kg CAN

P as TSP (Triple super phosphate)

Treatment: 26 kg P/ha

TSP contains: 46% P₂O₅

Molecular weight P₂O₅ : P = 30.973701 g/mol * 2 = O = 15.9994 g/mol * 5 = 141.94 g/mol P₂O₅

% P $(30.973701/141.94) * 100\% = 21.82\% > 46\% * 21.82\% = 10.04\%$ P in TSP

26 kg P/ha $> (100/10.04) * 26 = 258.96$ kg TSP/ha

- TSP/m² = 25.9 g/m²

- TSP/plot = 25.9 * 7.5 = 194.22 g/plot

- TSP/repl. = 194.22 * 5 = 971.1 g/repl.

- TSP/field = 971.1 * 3 = 2913.3 g/field

- Total TSP = 29.13 kg TSP

K as MOP (Muriate of potash)

Treatment: 25 kg K/ha

MOP contains 60% K₂O

Molecular weight K₂O: K = 39.0983 g/mol * 2 + O = 15.9994 g/mol = 94.196 g/mol K₂O

% K: $(39.0983/94.196) * 100\% = 41.51\% > 60\% * 41.51\% = 24.9\%$ K in MOP $> 25 \text{ kg K/ha} = (100/24.9)$

$* 25 = 100.4$ kg MOP/ha

- MOP/m² = 100400/10000 = 10.04 g/m²

- MOP/plot = 10.04 * 7.5 = 75.30 g/plot

- MOP/repl. = 75.30 * 5 = 376.51 g/repl.

- MOP/field = 1129.52 g/field

- Total MOP = 1129.52 * 10 = 11.3 kg MOP

Rhizobia inoculation

Rhizobia inoculant mixture, containing *Rhizobium* strain CIAT-899 and a peat carrier, was obtained from Legume Technology (UK). To be able to use the inoculant at several fields the larger pack was repacked in smaller containers using the following method:

1. Sterile containers were transferred to a laminar flow cabinet (NM-AIST, Arusha), one container for each field and two additional containers
2. The large pack was opened in the laminar flow cabinet.
3. Approximately 10g of the inoculant mixture was transferred to each container in the laminar flow cabinet and the head space in the container was minimized, to prevent the

inoculant from drying out.

4. The small containers were kept cool in a fridge of about 4°C until further use.

Appendix III - Questionnaire

Part A: Location information

Village _____
GPS Coordinates Field N/S _____ E/W _____
Altitude _____ meter

Part B: General information

Gender of farmer M / F
Age of farmer _____
Is farmer head of the household Yes / No
If no, gender of the HH head is _____
Members of the HH, specify gender and ages _____
Highest education enjoyed in the household _____
Total area of arable land available for the household _____
Do you grow legumes, apart from the research plot No / Yes > Which? _____

Number of large livestock species owned or taken care of by the household, specify _____

If yes, what do you feed the livestock? _____
If yes, what do you do with the manure? _____
If applied to the field, to which crops or fields is it preferentially applied? _____

Part C: Field management (experimental fields)

Which crops did you grow (if intercropped, mention all crops and indicate relative
Shares: 1. Vuli 2012 _____
2. Masika 2013 _____
3. Vuli 2013 _____
Indicate origin and variety/ies _____
Do you leave land fallow during the cropping season Yes / No
if yes, how long is a field left fallow between crops (season) _____
Mineral fertiliser(s) applied Yes / No
If yes, specify type and amount _____
Organic input(s) applied Yes / No
If yes, specify type, origin and amount _____
Inoculant applied Yes / No
If yes, specify type and origin _____
Perception of soil fertility by farmer (very poor, poor moderate, fertile or very
fertile) _____

In relation to other fields of the farmer
1) Poorer than in most other fields of the farm
2) The as in most other fields of the farm
3) Better than in most other fields of the farm
Soil drainage in the plot (good, medium or poor) _____
Are there any signs of soil erosion in the plot Yes / No
Presence of soil conservation structures in or directly around the field Yes / No
If yes; tied ridges, bench terrace, ditches, grass strips, tree lines, contour ridges
and/or others? _____

Appendix IV - Protocol ICP-OES analyses - KU Leuven

Division Soil and Water Management, KU Leuven

Protocol: ICP plant destruction (10ml tubes)



14. DIGESTION OF PLANT MATERIAL FOR ICP-ANALYSIS (10ML TUBES)

1. Principle

Samples are digested on a hot plate by addition of concentrated nitric acid and measured by ICP (Inductively Coupled Plasma).

2. Apparatus

- Digestion tubes: disposable glass tubes of 10ml (10 ml is marked manually)
- dispenser 10 ml
- precision balance (0.0001g accuracy)
- digestion block (96 places)
- ICP

3. Reagents

- Nitric acid (HNO_3), min 69%, plasmaPURE

4. Protocol

4.1. Digestion

- Weigh approximately 50mg of the dried (75° C) plant material into clean, dry digestion tubes. Ensure that dried plant material settles well at the bottom. Record the weight of the sample.
- Add 1ml of nitric acid in such a way to rinse down any plant particles adhering to the tube edges.
- Swirl to mix and allow to stand overnight.
- Position the tubes in the digestion block.
- Turn the block up to 140°C (starting at 50°C and increase temperature gradually) and maintain that temperature for about two hours (until half of the liquid has evaporated), occasionally swirling each tube.
- Increase the temperature to 180°C and digest until "just NOT" dry, i.e. about 0.5 cm liquid left.
- Remove the digestion tubes from the digestion block and cool to room temperature.
- Another 1ml of nitric acid can be added if the digestion is not clear, again heat to 180°C and digest until "just NOT" dry.
- Dilute the digests to 10ml with MQ.
- Homogenize and allow to stand for a few hours.
- If particles are visible in the diluted digest, it is imperative to membrane filter the digest prior to dilution.
- Plant digests can be stored several days prior to analyses

Remarks:

- Digestion tubes need to be checked individually as some places in the unit heat quicker than others
- Always include 1 or 2 internal lab standards (Maize and/or another plant material) and 2 blanks (1ml HNO₃).

4.2. Calculations

- Subtract blank concentration from sample concentration (at equal dilution!)
- Solution concentration (mg/l) are converted to dry weight based concentration using:

$$\frac{mg/l \times volume\ liquid\ (l)}{sample\ weight\ (kg)}$$

5. Safety and chemical waste

Potential hazards: concentrated acid: wear protective gloves and safety glasses and work under a fumehood!

Disposal of chemical waste: collect the leftover digests into waste container Category 1.

Appendix V - Crop vigour overview

Field 1 - Mabughai 23-12-2013



R2T1 - Control



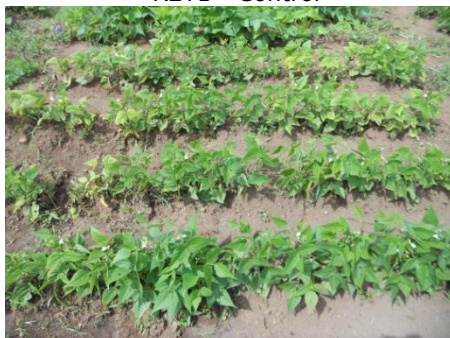
R2T2 - Control



R2T3 - K



R2T4 - P



R2T5 - Inoc.



R2T6 - K+P



R2T7 - K+Inoc.



R2T8 - P+Inoc.



R2T9 - K+P+Inoc.



R2T10 - P+K+N

Field 2 - Jeagertal 19-12-2013



R1T1 - Control



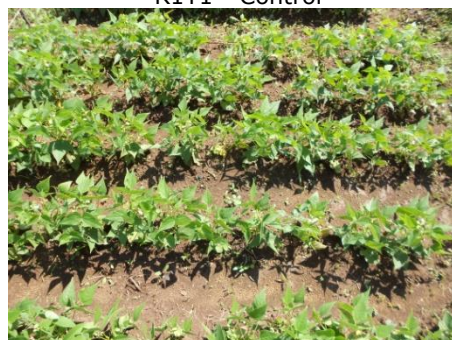
R1T2 - Control



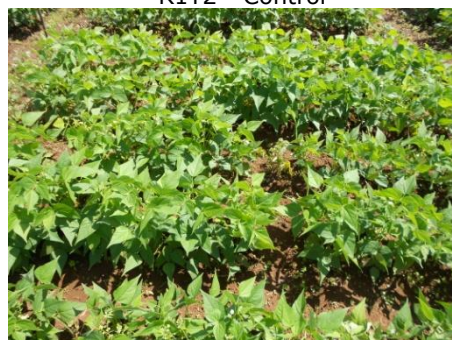
R1T3 - K



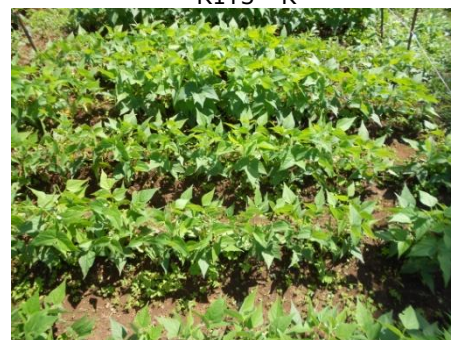
R1T4 - P



R1T5 - Inoc.



R1T6 - K+P



R1T7 - K+Inoc.



R1T8 - P+Inoc.



R1T9 - K+P+Inoc.



R1T10 - P+K+N

Field 3 - Lushoto 04-01-2014



R1T1 - Control



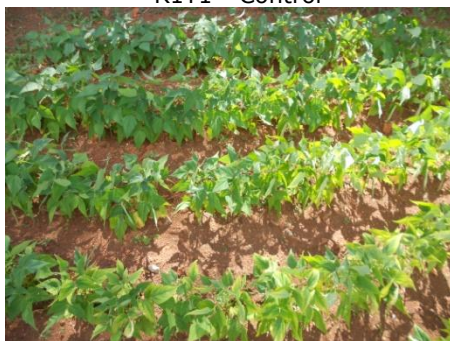
R1T2 - Control



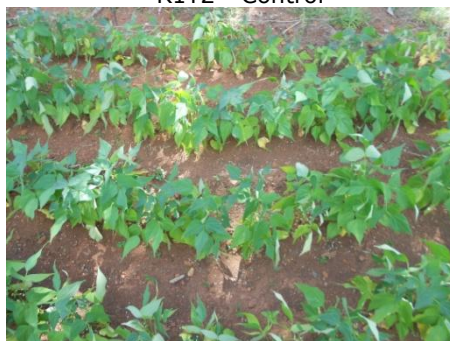
R1T3 - K



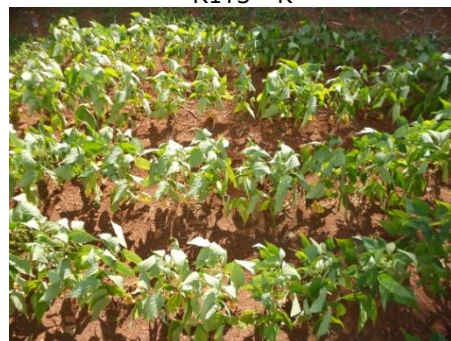
R1T4 - P



R1T5 - Inoc.



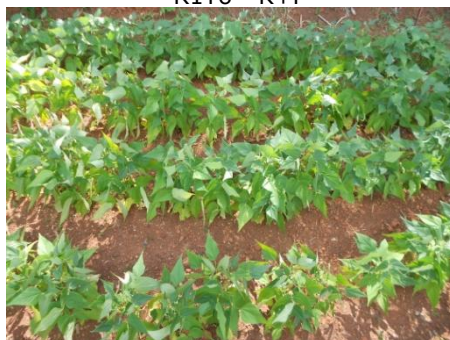
R1T6 - K+P



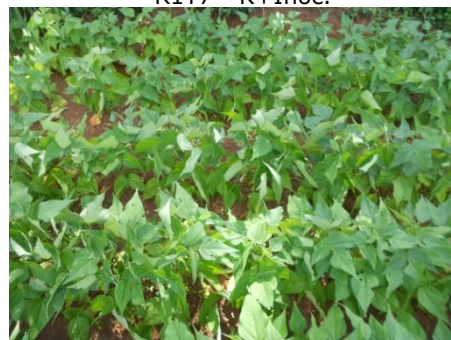
R1T7 - K+Inoc.



R1T8 - P+Inoc.



R1T9 - K+P+Inoc.



R1T10 - P+K+N

Field 4 - Kikurunge 14-01-2014



R2T1 - Control



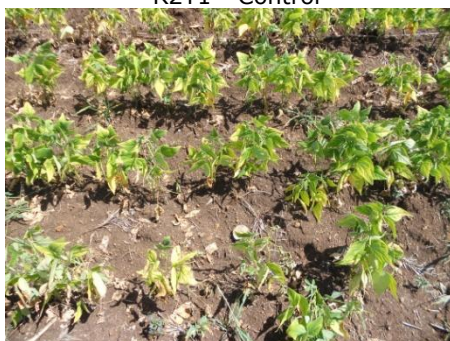
R2T2 - Control



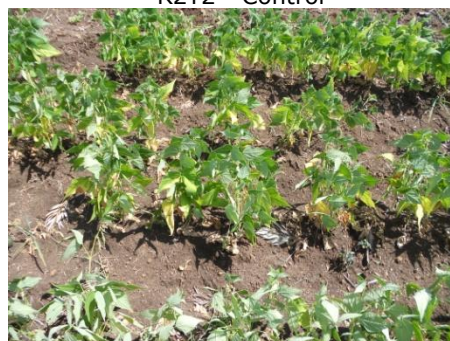
R2T3 - K



R2T4 - P



R2T5 - Inoc.



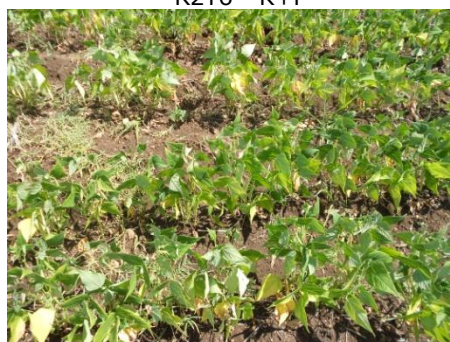
R2T6 - K+P



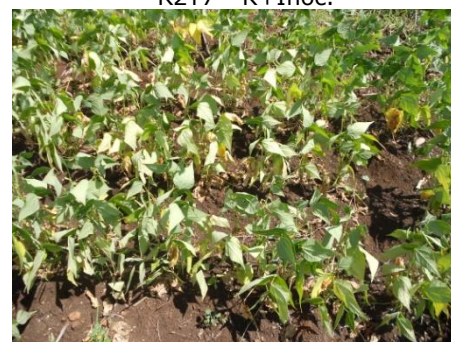
R2T7 - K+Inoc.



R2T8 - P+Inoc.



R2T9 - K+P+Inoc.



R2T10 - P+K+N

Field 5 - Mschizii 04-01-2014



R2T1 - Control



R2T2 - Control



R2T3 - K



R2T4 - P



R2T5 - Inoc.



R2T6 - K+P



R2T7 - K+Inoc.



R2T8 - P+Inoc.



R2T9 - K+P+Inoc.



R2T10 - P+K+N

Field 6 - Kwemsanga 24-12-2013



R2T1 - Control



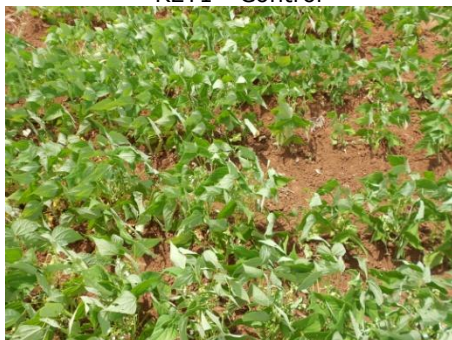
R2T2 - Control



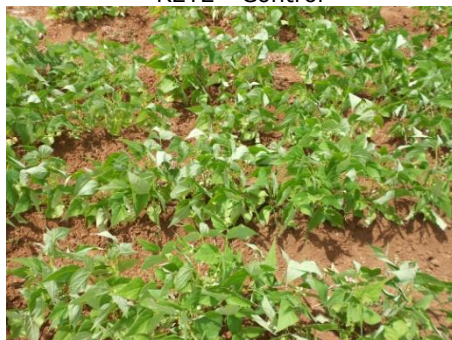
R2T3 - K



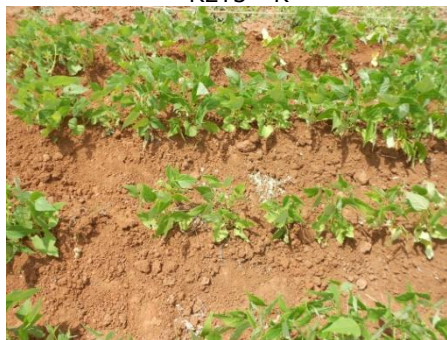
R2T4 - P



R2T5 - Inoc.



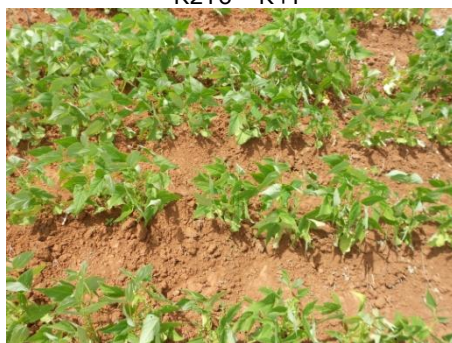
R2T6 - K+P



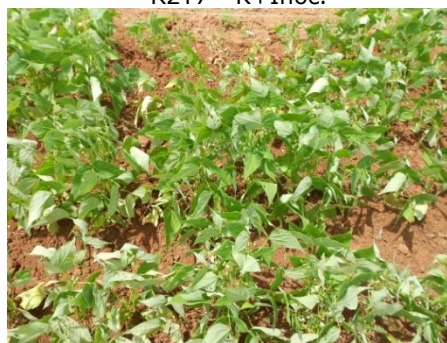
R2T7 - K+Inoc.



R2T8 - P+Inoc.



R2T9 - K+P+Inoc.



R2T10 - P+K+N

Field 7 - Ngulwi 25-12-2013



R1T1 - Control



R1T2 - Control



R1T3 - K



R1T4 - P



R1T5 - Inoc.



R1T6 - K+P



R1T7 - K+Inoc.



R1T8 - P+Inoc.



R1T9 - K+P+Inoc.



R1T10 - P+K+N

Field 8 - Mbuzii I 05-01-2014



R1T1 - Control



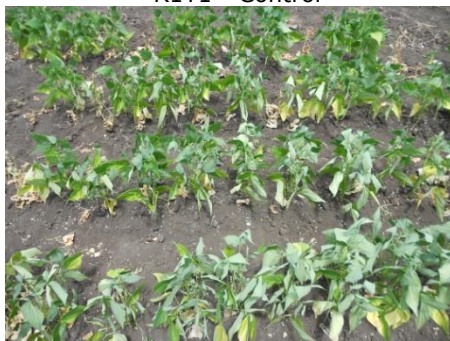
R1T2 - Control



R1T3 - K



R1T4 - P



R1T5 - Inoc.



R1T6 - K+P



R1T7 - K+Inoc.



R1T8 - P+Inoc.



R1T9 - K+P+Inoc.



R1T10 - P+K+N

Field 9 - Mbuzii II 05-01-2014



R1T1 - Control



R1T2 - Control



R1T3 - K



R1T4 - P



R1T5 - Inoc.



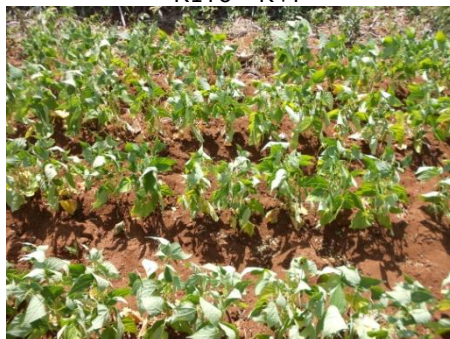
R1T6 - K+P



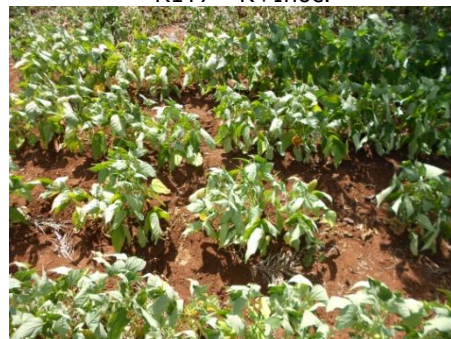
R1T7 - K+Inoc.



R1T8 - P+Inoc.



R1T9 - K+P+Inoc.



R1T10 - P+K+N