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Evaluation of nitrogen fixation using ^{15}N dilution methods and economy of a maize-tepary bean intercrop farming system in semi-arid SE-Kenya

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Abstract

Tepary bean has become popular among poor small-scale farmers in semi-arid Kenya, where it is intercropped with maize. This study aimed at i) evaluating the N-economy of maize/tepary bean intercrop versus sole crop using natural abundance and ^{15}N enriched fertilizer methods, and ii) assessing the contribution of fixed N_2 by tepary bean to the total N balance in the intercrops and sole cropping systems assessed from harvested seed and residues. Experiments were carried out during the short rains of 2001/2002 and long rains of 2003 at Kenya Agriculture Research Institute (KARI) Kiboko, Kenya. Randomised block design was used with one block devoted to the ^{15}N natural abundance (–N), the other ^{15}N labelled fertilizer (+N), replicated 4 times. Above ground biomass and total N were determined in sole crops or intercrops (–N or +N). Tepary bean received 53–69% of its N supply from N_2-fixation with N_2-fixation slightly affected by intercropping or N fertilizer application. N_2-fixation of tepary in greenhouse experiment was lower (36–66%) than in the field study and more affected by N supply. Budgets for N were estimated for field intercrops based on above-ground seed yields, return of crop residues, input of fixed N and fertilizer N. N_2-fixation was 59 kg N ha^{-1} in plots receiving no N fertilizer, and 73 kg N ha^{-1} in plots receiving N as urea. Corresponding fixation by sole tepary was high (87 and 82 kg N ha^{-1}, respectively), but this advantage was outweighed by greater land use efficiency in intercrop than sole crop

Key words: ^{15}N dilution methods; Kenya; intercropping; maize; Tepary bean

Introduction

The Government of Kenya has underscored the important role played by the arid and semi-arid lands (ASALs) of Kenya in food production (Republic of Kenya, 1993). Currently, food production has been declining in these ASALs as population growth increases (Rao and Mathuva, 2000; Maingi et al., 2001; Shisanya, 2002), and also because long periods of fallow are no longer practiced and the land is cropped continuously after clearing. Fertilizer use is low because of socio-economic constraints, its unavailability at the right time, its high cost, and risks from erratic rainfall. As a result, yields of cereals do not exceed 1 t ha^{-1}, and legumes 0.5 t ha^{-1} per crop season (Tiffen et al., 1994). It is therefore a major challenge to sustain crop yields and economic returns in such low input agricultural systems, predominantly by small-scale farm.

Various researchers have emphasized the importance of research on drought tolerant crop species of short cycle as a priority in addressing the food deficit problem in the ASALs (Hornetz et al., 2000; Shisanya, 2002, 2004). Unfortunately, this has not received adequate attention (Shisanya, 1999). Most smallholder farmers in the ASALs cannot afford the required external inputs in the form of chemical N fertilizer to improve their food production. Researchers in Kenya have exploited

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the legume *Rhizobium* symbiosis as a substitute for the expensive N fertilizers in these ASALs (Gitonga et al., 1999; Hornetz et al., 2000; Maingi et al., 2001; Shisanya, 2002, 2004). Nitrogen (N) contribution by legumes to other crops in the system depends on the species, biological N_2 fixation and growth of legumes as determined by climate and soil, and management of residues.

In semi-arid Kenya, maize (*Zea mays* L.) is commonly intercropped or rotated with bean (*Phaseolus vulgaris* L.), pigeon pea (*Cajanus cajan* L. Millsp.) or cowpea (*Vigna unguiculata* L. Walp), although the relative proportion of the legume in these mixed systems is small. Tepary bean (*Phaseolus acutifolius* A. Gray var. latifolius), a drought tolerant legume (Hornetz, 1990) has recently assumed importance in the intercrop farming systems of semi-arid Kenya (Shisanya, 2002). The N removed by maize in this region is estimated to be as much as 25–40 kg ha^{-1} per season, which means that a matching amount of N needs to be supplied for long term sustainability of production (Rao and Mathuva, 2000). Nitrogen fixation by bean is notoriously inconsistent, with or without inoculation (Maingi et al., 2001), but cowpea nodulates well by the ubiquitous *Bradyrhizobia* sp. and fixes up to 20 kg N ha^{-1} (Pilbeam et al., 1995). Recently, Shisanya (2004) found that tepary bean (TB) nodulates very well with *Rhizobium* sp. strain R3254 and fixes up to 260 kg N ha^{-1}. However, this study by Shisanya (2004) did not investigate the effect of intercropping maize and TB on nitrogen fixation and the actual amount fixed by the latter under the semi-arid conditions. Further, the study did not make an assessment of the contribution of fixed N_2 by tepary bean to the total balance of the intercrop farming system. Earlier studies (Gitonga et al., 1999; Maingi et al., 2001) investigated the effect of intercropping on nitrogen fixation by common bean and green gram (*Vigna radiata* L. Wilczek) under semi-arid conditions.

In view of the above, the main objectives of this study were, therefore to: (1) evaluate the relative efficiency of maize/tepary bean intercrop versus sole cropping situation under the semi-arid conditions of southeast Kenya, (2) evaluate the N-economy of maize/tepary bean intercrop versus sole crop using the natural abundance and ^{15}N enriched and (3) assess the contribution of fixed N_2 by tepary bean to the total balance of N in the intercrops and sole cropping farming systems as assessed in terms of harvested seed and crop residues, under the semi-arid conditions of southeast Kenya.

Materials and methods

Experimental site

The experiments were carried out at Kenya Agriculture Research Institute (KARI) Kiboko sub-centre (latitude 02° 12' S, longitude 37° 43' E, altitude 975 m a.s.l.), located at about 160 km southeast of Nairobi, the capital town of Kenya. The climates of the experimental site is described as hot and dry (Hornetz et al., 2000). The soils are well drained Fluvisols, Ferralsols and Luvisols (Eichinger, 1999). Rainfall is bimodally distributed, with median monthly maximum in April (126 mm) and November (138 mm). The medial annual rainfall is about 582 mm $year^{-1}$. The short rains (SR) (October–January) generally have more rainfall and are more reliable than the long rains (LR) (March–June) (Hornetz et al., 2000). The lengths of the agrohumid periods for drought-adapted crops are 50–55 days (LR) and 65–70 days (SR) (Jaetzold and Schmidt, 1983). Average monthly temperatures are highest in February (24.3°C) and October (23.4°C) (Shisanya, 1996) prior to the onset of the rains in March and November, respectively. Preliminary soil analysis indicated that the N and P in 0–60 cm soil depth were 0.7 mg N kg^{-1} and 3.0 mg P kg^{-1} soil respectively. The C/N ratio and CEC are 11.7 and 7.8 ML^{-1}, respectively. The soil pH of the experimental field was acid (5.3) (measured in 0.01 ML^{-1} $CaCl_2$).

Field experiments

Field experiments were conducted over two seasons, i.e. short rains (SR) 2001/2002 (October–January) and long rains (LR) 2002 (March–June). A basal dose of triple superphosphate (TSP) fertilizer was applied at the rate of 40 kg ha^{-1} on all the plots to alleviate phosphorus deficiency. The N treatment plots (+N) received calcium ammonium nitrate fertilizer (CAN) (26% N) at the same rate as TSP. The experimental layout comprised two randomised block designs (Figure 1). One block was devoted to the ^{15}N-natural-abundance (–N) study, the other to the ^{15}N-labelled fertilizer (+N) experiment. The (–N) and (+N) blocks each consisted of four replicate plots of sole maize (M), sole tepary bean (TB), or intercropped maize/tepary bean (MTB) (Figure 1). The plots were sown on 3rd November and 4th April for the short rains and long rains experiments, respectively. In the (+N) study, all treatments were given a basal dose of 40 kg ha^{-1}

of N as urea, except for the unconfined microplots (3 m × 0.75 m), which were carefully watered (to avoid contamination) with ^{15}N-urea solution (1.37% atom excess ^{15}N) after post emergence thinning, at an equivalent rate of 40 kg N ha^{-1}. Unlabelled urea was added to the remainder of the block at the same rate. Each experimental plot measured 8 m × 6 m and consisted of 6 raised seedbeds separated by an irrigation furrow. The sole crops M, TB and TB comprised a two-row layout on each seedbed, with rows 75 cm apart with a spacing of 30 cm between plants in the row for (M), yielding a plant density of 44,000 plants/ha. The TB and SB spacing were 50 cm between rows and 20 cm within rows, giving a density of 100,000 plants/ha. The intercrop plots consisted of central plots of maize with flanking rows of TB. The above planting densities are those recommended for semi-arid southeast Kenya (Hornetz et al., 2000; Shisanya, 1998). TB was inoculated with a peat culture in gum *arabic* incorporating the effective Rhizobium strain R3254 according to the method described by Kibunja (1984).

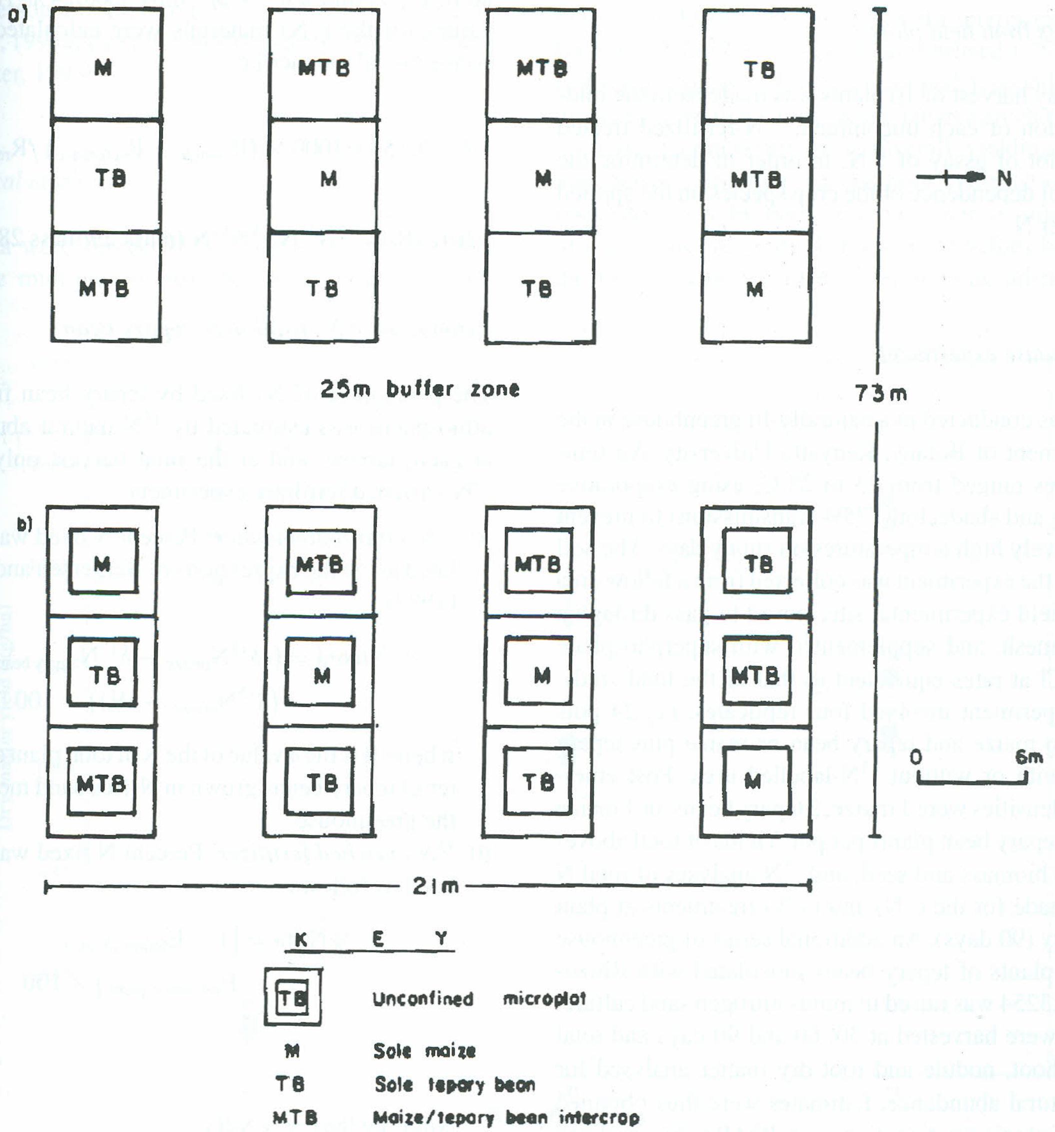

Figure 1. Plan of field site, (a) No added fertilizer N (–N experiment) (b) Fertilizer added (40 kg ha^{-1}) (+N experiment).

Maize, tepary seeds, and Rhizobium culture

Seeds of maize (cv. Makueni DLC1) and tepary bean (cv. latifolius) were obtained from local farmers. Undamaged seeds were carefully selected to ensure uniformity in size. Commercial *Rhizobium leguminosarum* biov. *phaseoli* strain R3254 used to inoculate TB was obtained from the Microbiological Resource Centre (MIRCEN), University of Nairobi. The infectivity and effectivity of this strain had been established in an earlier study (Shisanya, 2002).

Harvests from field plots

A 90-day harvest of 10 plants was made from the middle region of each unconfined, ^{15}N-fertilized treated microplot of assay of ^{15}N, in order to determine the extent of dependence of the crop species on the applied fertilizer N.

Greenhouse experiment

This was conducted in a naturally-lit greenhouse in the Department of Botany, Kenyatta University. Air temperatures ranged from 23 to 28°C, using evaporative cooling and shadecloth (75% transmission) to prevent excessively high temperatures on sunny days. The soil used in the experiment was collected from a fallow area at the field experimental site, sieved to pass through a 2-mm mesh, and supplemented with superphosphate and KCl at rates equivalent to that of the field study. The experiment involved four replicates, i.e. 24 pots sown to maize and tepary bean or maize plus tepary bean, with or without ^{15}N-labelled urea. Post emergence densities were 1 maize, 2 tepary beans, or 1 maize plus 2 tepary bean plants per pot. Yields of total aboveground biomass and seed, and ^{15}N analyses of total N were made for the (–N) and (+N) treatments at plant maturity (90 days). An additional series of greenhouse grown plants of tepary beans inoculated with Rhizobium R3254 was raised in minus nitrogen sand culture. Plants were harvested at 30, 60 and 90 days and total N of shoot, nodule and root dry matter analysed for ^{15}N natural abundance. Estimates were thus obtained of the isotopic fractionation (so called B value) realized by nodulated tepary bean subsisting on atmospheric N_2 (Amarger et al., 1979).

^{15}N analysis of soil and plant dry matter

Samples of soil or plant dry matter were prepared and analysed according to the methods described by (Boddey et al., 2000; Bergersen, 1980). ^{15}N analyses were done at the UFZ Centre for Environmental Research in Leipzig-Halle, Germany, using a triple ion collector, dual inlet mass spectrometer (SIRA 9, VG Isogas, Middlewich, Cheshire, UK), incorporating a direct inlet line from hypobromite oxidation system similar to that described by Porter and O'Deen (1977). The standard reference source for ^{15}N analysis was a sample of $(NH_4)SO_4$ showing 0.36598 ± 0.00001 atom% ^{15}N and a $\delta^{15}N$ of $-0.87 \pm 0.024\%$. All $\delta^{15}N$ values for the (–N) materials were calculated in the conventional manner i.e.:

$$\delta\ ^{15}N\,(5) = 1000 \times \left(R_{sample} - R_{reference}\right) / R_{reference} \quad (1)$$

where: $R = {}^{15}N^{14}N/{}^{14}N^{14}N$ (mass 29/mass 28)

Estimation of N_2 fixation by tepary bean

The percentage of N_2 fixed by tepary bean from the atmosphere was estimated by ^{15}N natural abundance at each harvest, and at the final harvest only in the ^{15}N-enriched fertilizer experiment.

a) *^{15}N natural abundance*: Percent N fixed was calculated using the expressions of Bergersen and Turner (1983):

$$\%Nfixed = \left(\left(\delta^{15}N_{maize} - \delta^{15}N_{tepary\ bean}\right) / \left(\delta^{15}N_{maize} - \delta B\right)\right) \times 100 \quad (2)$$

where: B is the δ value of the N of total plant dry matter of tepary beans grown in N-free sand medium in the greenhouse.

b) *^{15}N-enriched fertilizer*: Percent N fixed was calculated as follows:

$$\%Ndfa = \left[1 - E_{fixing\ plant} / E_{reference\ plant}\right] \times 100 \quad (3)$$

$$Ndfa\ (kg/ha) = \%Ndfa \times \text{Total N in fixing crop}\,(kg/ha)/100 \quad (4)$$

where: %Ndfa = Percent N derived from the atmosphere, and
E = atom% excess of either N_2-fixing plant or the reference plant

Soil analysis

Soil samples were collected to a depth of 60 cm using a soil auger before planting. Five sub-samples were collected from each plot, mixed thoroughly in polythene bags and transported to the laboratory. Soil organic C was analysed according to the method described by Anderson and Ingram (1993), total N according to (Forster, 1995a; Bremner and Mulvaney, 1982) and soil P (Forster, 1995b).

Statistical analysis

The data were subjected to analysis of variance and pairwise multiple comparisons were based on least significance difference (LSD) among treatment means (Steel and Torrie, 1981).

Results

Dry matter yields, N and land equivalent ratio for field experiments

Dry matter gains by sole crops and intercrops of maize in the presence or absence of fertilizer N were as plotted in Figures 2a and 2b. Analysis of variance for data at final harvest showed significant ($p < 0.05$) reductions in yield of either species with intercropping in both the N fertilized (+N) or unfertilised (–N) plots. Both crop species, sole or intercropped, responded positively to N fertilizer application, but to a greater degree in maize than in tepary bean. Overall, yielding capacities between the two seasons bore evidence of yield responses to N by both crops whether as sole crops or intercrops, and remarkably similar values between the two seasons for LER. Intercropping advantages,

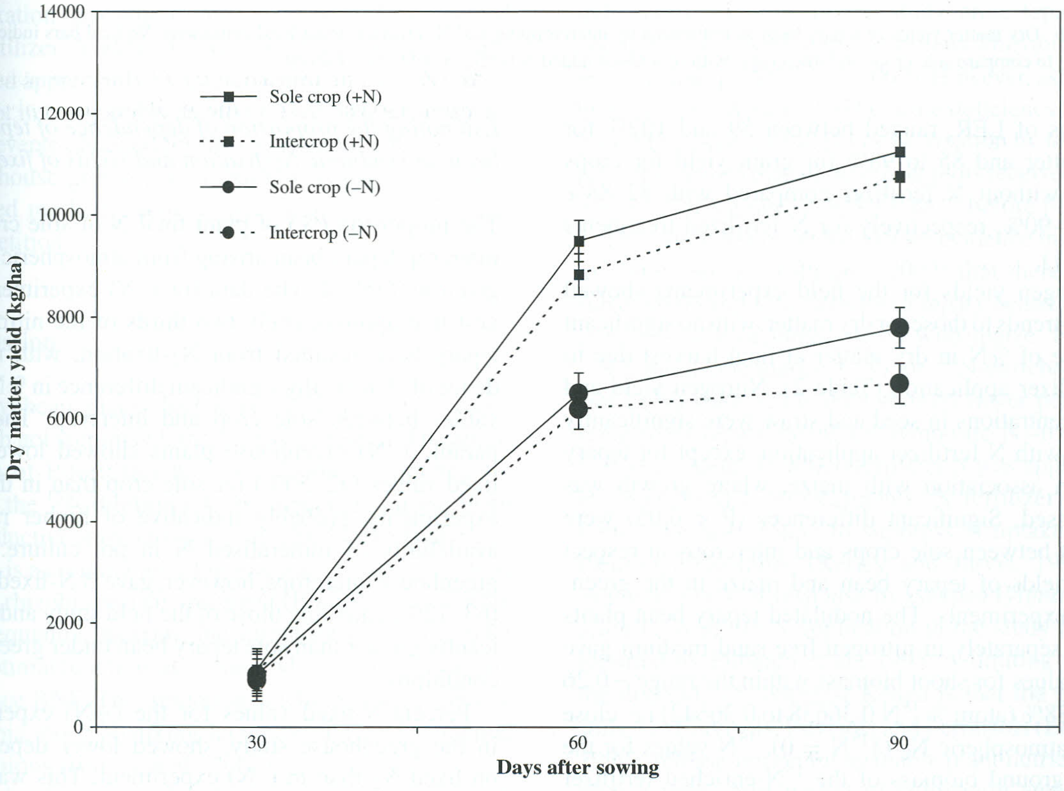

Figure 2a. Dry matter yields of maize as influenced by intercropping and N fertilizer under field conditions. Vertical bars indicate LSD ($p < 0.05$) to compare sole crops and intercrops, with or without added N fertilizer at 90 days harvest.

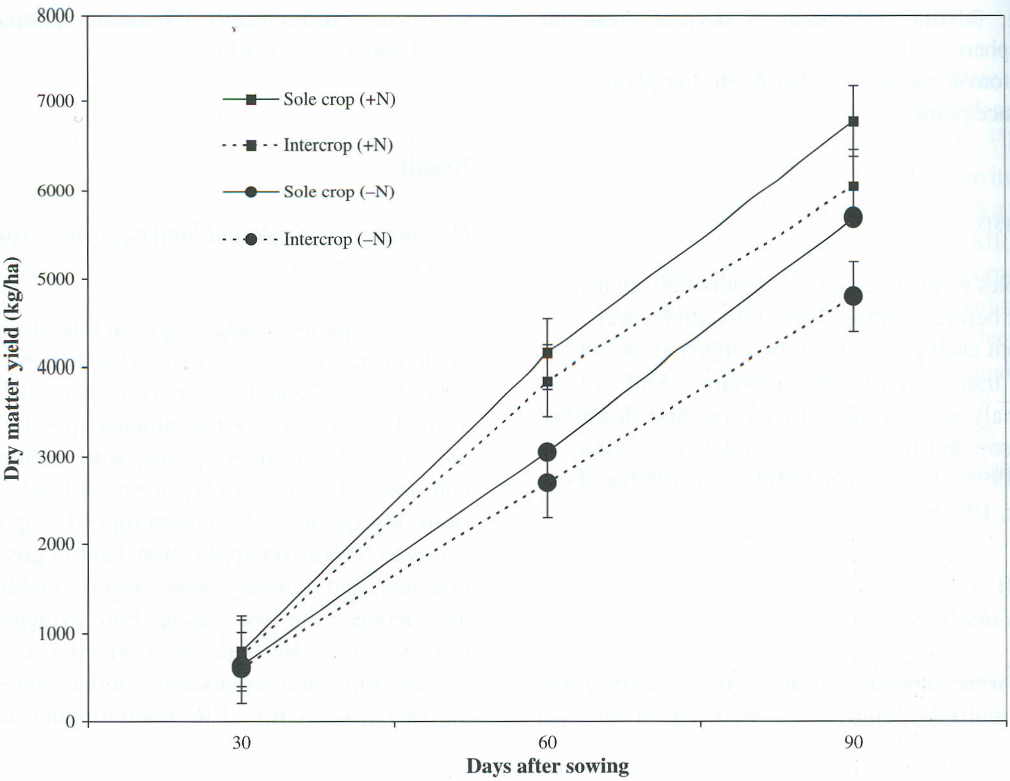

Figure 2b. Dry matter yields of tepary bean as influenced by intercropping and N fertilizer under field conditions. Vertical bars indicate LSD ($p < 0.05$) to compare sole crops and intercrops, with or without added N fertilizer at 90 days harvest.

in terms of LER, ranged between 59 and 102% for dry matter and 85 to 98% for grain yield for crops grown without N fertilizer compared with 82–88% and 82–90%, respectively for N fertilized treatments (Table 1).

Nitrogen yields for the field experiments showed similar trends to those for dry matter, with no significant increase of %N in dry matter at final harvest due to N-fertilizer application (Table 2). Nitrogen yield and N concentrations in seed and straw were significantly higher with N fertilizer application, except for tepary bean in association with maize, where growth was suppressed. Significant differences ($P < 0.05$) were evident between sole crops and intercrops in respect of N yields of tepary bean and maize in the greenhouse experiments. The nodulated tepary bean plants grown separately in nitrogen free sand medium gave $\delta^{15}N$ values for shoot biomass within the range +0.26 to +0.38% (atom % ^{15}N 0.36638 to 0.36642) i.e. close to the atmospheric N_2 ($\delta^{15}N = 0$). ^{15}N values for the above ground biomass of the ^{15}N-enriched fertilizer plots showed proportionality greater dilution of ^{15}N in tepary bean than in maize (Table 3).

Estimating the proportion of dependence of tepary bean on symbiotic N_2-fixation and yields of fixed N

The proportion (%) of plant final N of sole crop and intercrop tepary bean arising from atmospheric N_2 are given in Table 4. The data for (–N) experiment suggest that approximately two thirds of the nitrogen of tepary bean resulted from N_2-fixation, with no evidence of statistically significant difference in %N-fixed values between sole crop and intercrop. The comparable (–N) greenhouse plants showed lower %N-fixed values (42–50%) for sole crop than in the field experiments, probably indicative of higher rates of availability of mineralised N in pot culture. These greenhouse intercrops, however, gave %N-fixed values (63–72%) equalling those of the field study and significantly greater than sole tepary bean under greenhouse conditions.

Percent-N-fixed values for the (+N) experiments in the greenhouse study, showed lower dependence on fixed N_2 than in (–N) experiment. This was to be expected from the well-known inhibitory effect of combined N on symbiotic activity. However, comparison

Table 1. Yields of total dry matter (TDM), grain and land equivalent ratios (LER) of maize and tepary beans planted with or without nitrogen (N) fertilizer in the field

Cropping system		SR 2001/2002				LR 2002			
		Yield (kg ha^{-1})							
		TDM	Grain	LER		TDM	Grain	LER	
Sole crop									
Maize	(–N)	8400	2800			7200	2500		
	(+N)	12200	4280			10300	4150		
Tepary bean	(–N)	6300	950			5100	1000		
	(+N)	7100	1280			6500	1320		
				TDM	Grain			TDM	Grain
Intercrop									
Maize	(–N)	6800	2650	0.81[a]	0.95[a]			1.08[a]	0.95[a]
	(+N)	11300	3960	0.93	0.93			0.99	1.01
Tepary bean	(–N)	5000	980	0.79[a]	1.03[a]			0.94[a]	0.90[a]
	(+N)	6300	1200	0.89	0.94			0.89	0.89
Total	(–N)	11800	3630	1.59	1.98			2.02	1.85
	(+N)	17600	5160	1.82	1.87			1.88	1.90

[a] Partial LER for single species of intercrop.

between percent-N-fixed values of field-grown (+N) and (–N) tepary beans showed reduced dependence on N_2-fixation only where sole tepary beans had received N fertilizer. Yields of fixed N (Table 4) were suppressed appreciably by intercropping in the (–N) plots, but not in comparably treated greenhouse experiment. The reverse obtained for the (+N) treatments in which greenhouse grown plants, but not field grown plants, showed markedly reduced yields of fixed N through competition.

Discussion

Tepary bean plants had lower ^{15}N atom % excess, though not significant, than maize (Table 3). The application of P-fertilizer has been found to significantly affect the ^{15}N dilution effects, especially during plant reproductive stage (Irungu et al., 2002). The BNF symbiosis is a culmination of interactions between the host plant, rhizobia and environment (Boddey et al., 2000). Consequently, tackling the limitations, which hinder the optimal functioning of the BNF symbiosis, can only enhance BNF. The limitations include lack of suitable rhizobia, poor N_2-fixing legumes, and environmental limitations (Irungu et al., 2002).

Lack of suitable rhizobia can be tackled through production and inoculation with efficient host-specific rhizobia or in the absence of an elaborate breeding programme, selection of a suitable legume that nodulates abundantly and forms effective symbiosis with indigenous rhizobia. In the study area, tepary bean nodulated very well with indigenous *Rhizobium leguminosarium* sp. (Shisanya, 2002). However, as reported by Giller and Wilson (1991), acute deficiency of P can prevent nodulation and hence N_2 fixation by legume. It is for this reason that all treatment plots received a basal dose of P at the rate of 40 kg P ha^{-1} before planting.

This study has provided further evidence in addition to a previous one (Shisanya,2002) that there exists a substantial symbiotic activity of tepary beans, whether grown as sole or intercropped with maize. From the ^{15}N-natural abundance (–N) or ^{15}N-enriched fertilizer (+N) studies, the data showed lower percentage reliance of tepary beans on symbiotic N_2-fixation in (+N) treatments in which fertilizer N was applied than in unfertilised (–N) treatments. N fertilizer applications have been found to suppress N uptake and N_2 fixation in legumes (Kerley and Jarvis, 1999). For example a lack of response by cowpea (*Vigna unguiculata* L. Walp) to N application in the study has been reported by Pilbeam et al., 1995. A limiting factor in the drylands of southeast Kenya is that the prevalent soils, i.e. Fluvisols, Luvisols and Ferralsols (Eichinger, 1999), release important available plant nutrients only after a very short period of cultivation and through leaching (Eichinger, 1999; Hornetz, 1997). In addition, N-uptake by crops may be constrained by low root

Table 2. Nitrogen yields and nitrogen concentrations in dry matter of maize and tepary beans as influenced by intercropping under field and greenhouse conditions

Harvest	Cropping System	Maize N yield matter	Maize %N in dry matter	Tepary bean N yield	Tepary bean %N in dry Matter
(–N) study.					
Field study		*kg ha^{-1}*		*kg ha^{-1}*	
30 days	Sole crop	17.5	0.55	29.9	1.62
(shoot)	Intercrop	15.3	0.52	15.6	1.49
60 days	Sole crop	32.8	1.01	49.9	2.68
(Shoot)	Intercrop	38.5	0.95	25.6	2.40
90 days	Sole crop	68.0	0.90	90.6[a]	1.95
(Shoot)	Intercrop	61.3	0.88	73.5	1.65
90 days	Sole crop	20.4	0.25	45.0	0.72
(Straw)	Intercrop	19.5	0.27	29.3	0.70
90 days	Sole crop	57.4	1.20	80.4[a]	4.50
(Seed)	Intercrop	46.4	1.10	60.2	4.30
90 days	Sole crop	77.8	0.72	125.4[a]	1.52
(Seed+Straw)	Intercrop	65.9	0.70	89.5	1.63
Greenhouse study		*mg N pot^{-1}*		*mg N pot^{-1}*	
90 days	Sole crop	520[a]	0.66	750a	0.94
(Straw)	Intercrop	330	0.56	500	0.98
90 days	Sole crop	1020[a]	1.20	1390[a]	4.30
(Seed)	Intercrop	700	1.18	860	4.20
90 days	Sole crop	1600[a]	0.82	2040[a]	1.86
(Seed+Straw)	Intercrop	1040	0.80	1370	1.89
(+N) experiment (40 kg ha^{-1})					
Field study		*kg ha^{-1}*		*kg ha^{-1}*	
90 days	Sole crop	31.5[a]	0.46	55.0[a]	1.06
(Straw)	Intercrop	27.8	0.41	39.9	1.05
90 days	Sole crop	73.1[a]	1.15	97.5	3.80
(Seed)	Intercrop	59.8	1.09	68.0	3.75
90 days	Sole crop	104.6[a]	0.80	152.5[a]	1.98
(Seed+Straw)	Intercrop	87.6	0.78	107.9	1.96
Greenhouse study		*mg N pot^{-1}*		*mg N pot^{-1}*	
90 days	Sole crop	705	0.60	785	1.05
(Straw)	Intercrop	680	0.66	460	1.00
90 days	Sole crop	1450[a]	1.40	1740[a]	3.90[a]
(Seed)	Intercrop	996	1.30	770	3.68
90 days	Sole crop	2100[a]	0.98	2510[a]	2.20
(Seed+Straw)	Intercrop	1620	0.96	1220	1.98

[a]Differences between sole crop and intercrop significant at $p < 0.05$.

density, which is typical of tepary bean and cowpea (Shisanya, 1998), and or inadequate soil moisture content by which mineral N can move to the plant root (Pilbeam et al., 1995). However, results from this study in +N treatments were contrary to the above observations as will be shown later on in the discussion.

One of the objectives of this study was to evaluate the economy of nitrogen in a maize/tepary bean intercrop. In the absence of fertilizer (–N experiment), the amounts of fixed N in above-ground biomass of tepary bean (59 kg N ha^{-1}), almost balanced with the amount of N removed as tepary bean seed (60 kg N ha^{-1}) (Table 2). The amount of N removed as maize seed was 46 kg N ha^{-1}. Thus, essentially, there was a deficit of 46 kg N ha^{-1} in the system (Figure 3a). In the presence of N fertilizer (+N experiments), the amounts of N in the above-ground biomass of tepary bean was 73 kg N ha^{-1}, while N removed as tepary seed was 68 kg N ha^{-1} (Table 2 and Figure 3b). The amount of N removed as maize seed was 60 kg N ha^{-1}. In this +N

Table 3. ^{15}N abundance values of maize and tepary beans as sole crops or intercrops under field and greenhouse conditions with and without ^{15}N-enriched fertilizer

Harvest	Cropping	Maize	Tepary bean
(–N) study.			
Field study		$\delta^{15}N$ %	$\delta^{15}N$ %
60 days	Sole crop	6.7 (0.73)[a]	6.3 (0.47)
(Shoot)	Intercrop	7.6 (1.07)	5.9 (0.41)
90 days	Sole crop	6.5 (0.97)	4.3 (0.90)
(Shoot)	Intercrop	6.2 (0.64)	3.1 (0.12)
90 days	Sole crop	6.8 (0.72)	2.1 (0.07)
(Straw)	Intercrop	5.0 (0.48)	2.1 (0.22)
90 days	Sole crop	7.0 (0.81)	2.4 (0.30)
(Seed)	Intercrop	6.6 (0.90)	2.3 (0.12)
Greenhouse study		$\delta^{15}N$ %	$\delta^{15}N$ %
90 days	Sole crop	2.4 (0.21)	1.5 (0.21)
(Straw)	Intercrop	3.6 (0.42)	1.2 (0.19)
90 days	Sole crop	4.1 (0.48)	2.2 (0.34)
(Seed)	Intercrop	3.9 (0.29)	1.6 (0.10)
(+N) experiment (40 kg ha^{-1})			
Field study		*Atom % ^{15}N*	*Atom % ^{15}N*
90 days	Sole crop	0.4151 (0.0061)	0.3919 (0.0049)
(Straw)	Intercrop	0.4233 (0.0031)	0.3864 (0.0059)
90 days	Sole crop	0.4149 (0.0064)	0.3872 (0.0037)
(Seed)	Intercrop	0.4150 (0.0050)	0.3813 (0.0037)
Greenhouse study		*Atom % ^{15}N*	*Atom % ^{15}N*
90 days	Sole crop	0.5554 (0.0216)	0.5069 (0.0081)
(Straw)	Intercrop	0.5389 (0.0179)	0.5047 (0.0119)
90 days	Sole crop	0.5227 (0.0168)	0.4586 (0.0087)
(Seed)	Intercrop	0.5384 (0.0160)	0.4591 (0.0084)

[a]Values in parentheses are standard errors of 2 replicate analyses 5 or 10 plant samples from each plot (see text for details).

experiment, there was a net loss to the soil of 21 kg N ha^{-1}. This comprised a relatively small deficit in comparison to that shown by the (–N) plots. Amounts of N in the total biomass, harvested seed and crop residues of maize and tepary bean components were higher in the fertilized than unfertilised, and somewhat surprisingly, N-fertilizer application appeared to have promoted a greater return of fixed N in tepary been shoots (73 kg N ha^{-1}) than in the unfertilised situation (42 kg N ha^{-1}). This may have been due to the greater competitiveness of maize than cowpea in acquiring the added fertilizer (Maingi et al., 2001).

Nitrogen derived from fertilizer by intercrop maize was 5.3 kg N ha^{-1} and fertilizer use efficiency 22%. In intercrop tepary bean, these values were 3.6 kg N ha^{-1} and 15%, respectively. Maize thus met 5.3% of its N requirement from the ^{15}N-labelled fertilizer compared with 1.7% for tepary bean. Overall, intercrop above ground biomass derived 59.5% of its N from the soil, 3.3% from the added fertilizer and 37.3% from atmospheric N_2 fixation, suggesting that proportional benefits from symbiotic fixation were not significantly depressed by N fertilizer application, as pointed out earlier in this discussion. However, a detailed cost benefit analysis would be required to determine whether N fertilizer application is warranted under conditions in semi-arid southeast Kenya, and, if so, how its effectiveness might be reduced were extensive leaching of applied fertilizer to occur during the crop cycle.

A major inadequacy of the present approach, and other similar studies (Amarger et al., 1979; Kohl et al., 1980; Bergersen et al., 1990; Hardarson et al., 1991), is that no measurements were made of N in root biomass, especially in relation to the amount of fixed N added by legume component through decay of roots and nodules at the end of the growing season. It is proposed that such an approach be considered in future in similar studies. The present data using the ^{15}N-natural-abundance technique were encouraging in

Table 4. Proportional dependence on N2-fixation and N_2 fixed by sole and intercrop estimated by ^{15}N-labelled fertilizer method under field and greenhouse conditions

Harvest	Cropping	% N fixed	N_2 fixed[a]
(–N) study.			
Field study			*kg N ha^{-1}*
90 days	Sole crop	73.1	32.9
(Straw)	Intercrop	62.5	18.3
90 days	Sole crop	69.3	55.7
(Seed)	Intercrop	68.9	41.5
90 days	Sole crop	70.0	87.8
(Straw + Seed)	Intercrop	63.5	42.4
Greenhouse study			*mg N pot^{-1}*
90 days	Sole crop	44.3b	330
(Straw)	Intercrop	74.2	370
90 days	Sole crop	50.9	657
(Seed)	Intercrop	62.7	537
90 days	Sole crop	46.5	947
(Straw + Seed)	Intercrop	66.1	896
(+N) experiment (40 kg ha^{-1})			
Field study			*kg N ha^{-1}*
90 days	Sole crop	47.6	26.2
(Straw)	Intercrop	64.9	25.9
90 days	Sole crop	57.1	55.7
(Seed)	Intercrop	69.3	47.1
90 days	Sole crop	53.4	81.9
(Straw + Seed)	Intercrop	67.7	73.0
Greenhouse study			*mg N pot^{-1}*
90 days	Sole crop	25.7	200[b]
(Straw)	Intercrop	19.8	90
90 days	Sole crop	41.0	706[b]
(Seed)	Intercrop	46.1	351
90 days	Sole crop	36.2	906[b]
(Straw + Seed)	Intercrop	36.2	440

[a]Computed by multiplying % N fixed by tepary bean N yield data (Table 2); [b]Difference between sole crop and intercrop significant at $P < 0.05$.

showing reproducible differences between legume and reference crop under field conditions, and sufficiently great differences between $\delta^{15}N$ values of tepary bean relying solely on atmospheric N_2 (B-values, Table 3) and $\delta^{15}N$ of soil and maize to suggest that the technique might be used in future occasions for monitoring N balance of legume/cereal intercrops. The values for percentage N_2 fixed by tepary beans using ^{15}N-natural abundance and ^{15}N-enriched fertilizer methods lay within the range 53–69%. Combining high symbiotic competence of tepary bean with considerable yield advantages of mixed culture with maize in comparison with growing either species separately, there would appear to be considerable potential advantages in practicing maize/tepary bean intercrop, especially where it is not feasible or economical to effect large inputs of fertilizer to the cropping system as is the case in semi-arid southeast Kenya.

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a) *No added fertilizer N (–N)(experiment)*

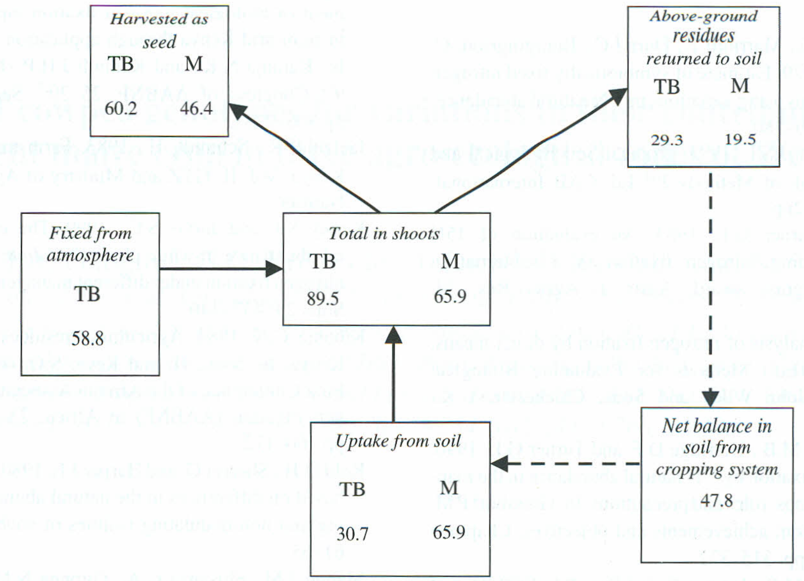

Figure 3a. Nitrogen budgets for maize tepary bean intercrops determined by ^{15}N dilution methods without N fertilizer.

b) *Fertilizer N added (40 kg N ha^{-1})(+N) experiment)*

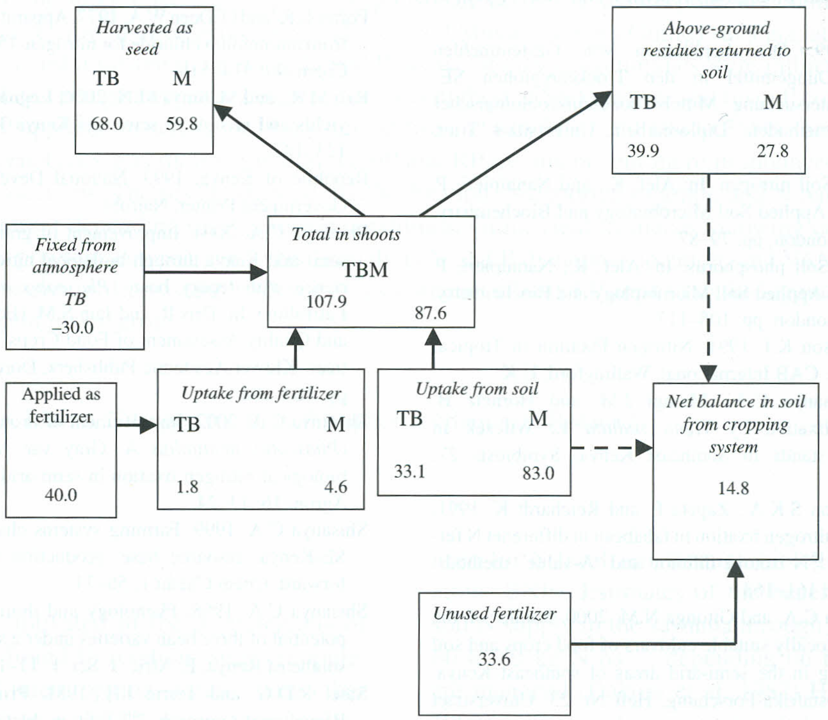

Figure 3b. Nitrogen budgets for maize tepary bean intercrops determined by ^{15}N dilution methods with added N fertilizer (40 kg N ha^{-1} urea).

References

Amarger N., Mariotti A., Marriotti F., Durr J.C., Bourguigrion, C. and Lagacherie B. 1979. Estimate of symbiotically fixed nitrogen in field grown soybeans using variations in ^{15}N natural abundance. Plant and Soil 52: 269–280.

Anderson, J.M., Ingram, J.S.I., 1993. Tropical Soil Biological and Fertility: A Handbook of Methods 2nd Ed. CAB International, Wallingford, U.K. 232pp.

Bergersen F.J. and Turner G.L. 1983. An evaluation of 15N methods for estimating nitrogen fixation in a subterranean clover-perennial ryegrass sward. Austr. J. Agric. Res. 34: 391–401.

Bergersen F.J. 1980. Analysis of nitrogen fixation by direct means. In: Bergersen F.J. (Ed.) Methods for Evaluating Biological Nitrogen Fixation. John Wiley and Sons, Chichester, U.K., pp. 65–110.

Bergersen F.J., Peoples M.B., Herridge D.F. and Turner G.L. 1990. Measurement of N_2 fixation by ^{15}N natural abundance in the management of legume crops: roles and precautions. In: Gresshoff P.M. (Ed.) Nitrogen Fixation: achievements and objectives. Chapman and Hall, New York, pp. 315–322.

Boddey R.M., Peoples M.B., Palmer P. and Dart P.J. 2000. Use of the ^{15}N natural abundance technique to quantify biological nitrogen fixation by woody perennials. Nutr. Cycl. Agroecosyst. 152: 25–52.

Bremner J.M. and Mulvaney C.S. 1982. Nitrogen-total. In: Page A.L., Miller R.H. and Keeney D.R. (Eds.) Methods of Soil Analysis, Part 2. Chemical and Biological Properties. Am. Soc. Agron. 9: 595–624.

Eichinger M. 1999. Die Applikation von Gesteinmehlen als Alternative Dungemittel in den Trockenregionen SE-Kenias: Eine Untersuchung Mittels bodenmikrobiologischer Feld- und Labormethoden. Diplomarbeit, Universitaet Trier, 140pp.

Forster J.C. 1995a. Soil nitrogen. In: Alef, K., and Nannipieri, P. (Eds.) Methods in Applied Soil Microbiology and Biochemistry. Academic Press, London, pp. 79–87.

Forster J.C. 1995b. Soil phosphorus. In: Alef, K., Nannipieri, P. (Eds.), Methods in Applied Soil Microbiology and Biochemistry. Academic Press, London, pp. 105–117.

Giller K.E. and Wilson K.J. 1991. Nitrogen Fixation in Tropical Cropping Systems. CAB International, Wallingford, U.K.

Gitonga N.M., Shisanya C.A., Maingi J.M. and Hornetz B. 1999. Nitrogen fixation by *Vigna radiata* L. Wilczek in pure and mixed stands in Southeast Kenya. Symbiosis 27: 239–250.

Hardarson G., Danson S.K.A., Zapata F. and Reichardt K. 1991. Measurements of nitrogen fixation in fababean in differenet N fertilizer rates using ^{15}N isotope dilution and "A-value" methods. Plant and Soil 131: 161–168.

Hornetz B., Shisanya C.A. and Gitonga N.M. 2000. Studies on the ecophysiology of locally suitable cultivars of food crops and soil fertility monitoring in the semi-arid areas of southeast Kenya. Materialien zur Ostafrika-Forschung, Heft Nr. 23. Universitaet Verlag, Trier.

Hornetz B. 1997. Resourcenschutz und Ernahrungssicherung in den semiariden Gebieten Kenyas. Reimer Verlag, Berlin.

Hornetz B. 1990. Vergleichende Stressphysiologie von Tepary Bohnen als 'Minor crop' und Mwezi Moja Bohnen als Hochleistungsleguminose im tropischen Landbau. J. Agron. Crop Sci. 164: 1–15.

Irungu J.W., Wood M., Okalebo J.R. and Warren G.P. 2002. Enhancement of biological nitrogen fixation input in smallholder farms in semi-arid Kenya through application of phosphorus fertilizer. In: Karanja N.K., and Kahindi J.H.P. (Eds.) Proceedings of the 9th Congress of AABNF, 25–29th September 2000, Nairobi, pp.306–323.

Jaetzold, R., Schmidt, H., 1983. Farm management handbook of Kenya. Vol. II. GTZ and Ministry of Agriculture, Eschborn and Nairobi.

Kerley S.J. and Jarvis S.C., 1999. The use of nitrogen-15 natural abundance in white clove (*Trifolium repens* L.) to determine nitrogen fixation under different management practices. Biol. Fert. Soils 29: 437–440.

Kibunja C.N. 1984. Agricultural residues as Rhizobia carriers in Kenya. In: Ssali, H. and Keya, S.O. (Eds.) Proceedings of the First Conference of the African Association for Biological Nitrogen Fixation (AABNF) in Africa, 23–28 September, Nairobi, pp 160–172.

Kohl D.H., Shearer G. and Harper J.E. 1980. Estimates of N_2 fixation based on differences in the natural abundance of 15N in nodulating and non-nodulating isolines of soybeans. Plant Physiol. 66: 61–65.

Maingi J.M., Shisanya C.A., Gitonga N.M. and Hornetz B. 2001. Nitrogen fixation by common bean (*Phaseolus vulgaris* L.) in pure and mixed stands in semi-arid south-east Kenya. European J. Agron. 14: 1–12.

Pilbeam C.A., Wood M., and Mugane P. G. 1995. Nitrogen use in maize-grain legume cropping systems in semi-arid Kenya. Biol. Fert. Soils 20: 57–62.

Porter L.K. and O'Deen W.A. 1977. Apparatus for preparing nitrogen from ammonium chloride for nitrogen-15 determinations. Analyt. Chem. 49: 514–516.

Rao M.R., and Mathuva M.N. 2000. Legumes for improving maize yields and income in semi-arid Kenya. Agric., Ecosys. Env. 78: 123–137.

Republic of Kenya, 1993. National Development Plan 1993–96. Government Printer, Nairobi.

Shisanya C.A. 2004. Improvement of grain legume production in semi-arid Kenya through biological nitrogen fixation: The experience with tepary bean (*Phaseolus acutifolius* A. Gray var. Latifolius). In: Dris R. and Jain S.M. (Eds.) Production Practices and Quality Assessment of Food Crops. Vol. 1: Preharvest Practices. Kluwer Academic Publishers, Dordrecht, The Netherlands, pp. 163–188.

Shisanya C.A. 2002. Improvement of drought adapted tepary bean (*Phaseolus acutifolius* A. Gray var. latifolius) yield through biological nitrogen fixation in semi-arid SE-Kenya. European J. Agron. 16: 13–24.

Shisanya C.A. 1999. Farming systems characteristics in semi-arid SE-Kenya: resource base, production dynamics and the way forward. Chem Chemi 1: 56–74.

Shisanya C.A. 1998. Phenology and diurnal course of leaf water potential of three bean varieties under a semi-arid environment in southeast Kenya. E. Afric. J. Sci. 1: 11–19.

Steel R.D.G. and Torrie J.H. 1981. Principles of Statistics: A Biometrical Approach. 2nd Edition. McGraw-Hill, London.

Shisanya C.A. 1996. Chances and risks of maize and bean growing in the semi-arid areas of SE-Kenya during expected deficient, normal and above normal rainfall of the short rainy seasons. Materialien zur Ostafrika-Forschung, Heft 14. 418pp.

Tiffen M., Mortimore M. and Gichuki F. 1994. More People, Less Erosion. ACTS Press, Nairobi.