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Mineral Fertilizer Demand for Optimum Biological Nitrogen Fixation and Yield Potentials of Legumes in Northern Ethiopia

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Abstract: Farmers in Northern Ethiopia integrate legumes in their cropping systems to improve soil fertility. However, biological nitrogen fixation (BNF) potentials of different legumes and their mineral nitrogen (N) and phosphorus (P) demands for optimum BNF and yields are less studied. This study aimed to generate the necessary knowledge to enable development of informed nutrient management recommendations, guide governmental public policy and assist farmer decision making. The experiment was conducted at farmers' fields with four N levels, three P levels, and three replications. Nodule number and dry biomass per plant were assessed. Nitrogen difference method was used to estimate the amount of fixed N by assuming legume BNF was responsible for differences in plant N and soil mineral N measured between legume treatments and wheat. The result revealed that the highest grain yields of faba bean (2531 kg ha^{-1}), field pea (2493 kg ha^{-1}) and dekeko (1694 kg ha^{-1}) were recorded with the combined application of 20 kg N ha^{-1} and 20 kg P ha^{-1}. Faba bean, field pea and dekeko also fixed 97, 38 and 49 kg N ha^{-1}, respectively, with the combined application of 20 kg N ha^{-1} and 20 kg P ha^{-1}; however, lentil fixed 20 kg ha^{-1} with the combined application of 10 kg N ha^{-1} and 10 kg P ha^{-1}. The average BNF of legumes in the average of all N and P interaction rates were 67, 23, 32 and 16 kg N ha^{-1} for faba bean, field pea, dekeko and lentil, respectively. Moreover, faba bean, field pea, dekeko and lentil accumulated a surplus soil N of 37, 21, 26 and 13 kg ha^{-1}, respectively, over the wheat plot. The application of 20 kg N ha^{-1} and 20 kg P ha^{-1} levels alone and combined significantly ($p < 0.05$) increased the nodulation, BNF and yield of legumes; however, 46 kg N ha^{-1} significantly decreased BNF. This indicated that the combination of 20 kg N ha^{-1} and 20 kg P ha^{-1} levels is what mineral fertilizer demands to optimize the BNF and yield of legumes. The results of this study can lead to the development of policy and farmer guidelines, as intensification of the use of legumes supplied with starter N and P fertilizers in Northern Ethiopian cropping systems has the multiple benefits of enhancing inputs of fixed N, improving the soil N status for following crops, and becoming a sustainable option for sustainable soil fertility management practice.

Keywords: legumes; nodule characteristics; soil fertility; sustainable crop production

1. Introduction

The global concern regarding food demand for the increasing human population has been strengthening the importance of sustainable agricultural production [1]. Smallholder farmers practicing rain-fed agriculture in sub-Saharan Africa (SSA) including Ethiopia, face food insecurity due to poor soil fertility and climate change [2]. As a result, soil fertility management is critical to improve sustainable agricultural production and food security in Ethiopia [3–7]. Moreover, soils in northern Ethiopia are deficient in nitrogen and phosphorus, which negatively affects nodulation and biological nitrogen fixation (BNF) potentials of legume crops [7,8].

The government of Ethiopia has made tremendous effort to improve soil fertility and increase agricultural productivity [9]. However, most small holder farmers cannot afford the cost of mineral fertilizer. On the other hand, farmers use manure to enhance soil fertility; however, its nutrient content is less, which requires bulk application to satisfy plant nutrient demand. Previously, farmers were fallowing their land to restore soil fertility; however, today fallowing is abandoned because of the limited availability of arable lands. Due to high cost of the inorganic fertilizers, it is rare for poor farmers to use mineral fertilizers; as a result, crop yield is declining. Owing to rising costs of inorganic fertilizers and growing environmental concerns, there is an ever increasing interest in the role of BNF [8]. Therefore, soil fertility management practices, which are easily accessible for resource poor farmers like BNF and sustainably improve agricultural productivity, are essential.

Many studies conducted in Ethiopia and elsewhere in Africa have suggested that BNF in different legume crops supplies sufficient N for optimum and sustainable crop production [10–15]. On the other hand, farmers in the study area grow diverse legume crops with low or no fertilization because they thought that growing legume crops in their field would improve the fertility of degraded soils. However, since legumes cannot develop nodules at an early stage, legumes require starter N fertilizer for nodule initiation and P fertilizer for optimum nodulation and BNF [16,17]. Though farmers in the study region incorporate legumes into their cropping system, the cultivated soils are still deficient in N and P nutrients [4,18,19]. This deficiency in N and P affects nodulation and BNF in the study area [8]. Literatures also stated that though a high rate of N fertilizer suppressed nodule formation and BNF, only starter N fertilizer is important for nodule initiation and BNF in Northern Ethiopia [13–15]. Symbiotic N fixation also requires P fertilizer as a source of energy generating metabolism [15,20]. Moreover, many studies also confirmed that different legumes have different nodulation and BNF potentials [8,9,12,14,15]. However, since these studies were conducted in different areas, it was difficult to compare the BNF capacities of these different legumes. While it has been hypothesized that the additional starter N and P fertilizer could improve nodulation and BNF, the actual rates required to optimize BNF, and which legume species might provide the highest inputs of fixed N to support productivity and improve soil N, is currently unknown for Northern Ethiopia. This study aimed to generate the necessary knowledge to enable the development of informed nutrient management recommendations, guide governmental public policy, and assist farmer decision making.

2. Methodology

2.1. The Study Area

This study was conducted in the Alaje district, Northern Ethiopia, located at 39°25′52′′ to 39°44′50′′ E and 12°15′28′′ to 12°59′21′′ N (Figure 1). The average elevation of the area is 2824 m a.s.l.

The total amounts of rainfall received during the 2017 and 2018 cropping seasons were 417 and 479 mm, respectively (Figure 2). The mean minimum and maximum temperatures were 12.6 and 23.3 °C for the 2017 cropping season and 11.6 and 22.3 °C for the 2018 cropping season, respectively (Figure 2). Alaje has cold sub-moist highland agro-ecology [21]. The main farming system in Alaje is mixed, meaning that farmers integrate crop and animal husbandry to enhance agricultural production and increase productivity. The dominant cereal food crops in the study area are wheat (*Triticum* spp.), barley (*Hordeum* spp.) and pulses such as faba-bean (*Vicia faba* L.), field pea (*Pisum sativum*), dekeko (*Pisum sativum* var. *abyssinicum*) and lentil (*Lens culinaris*).

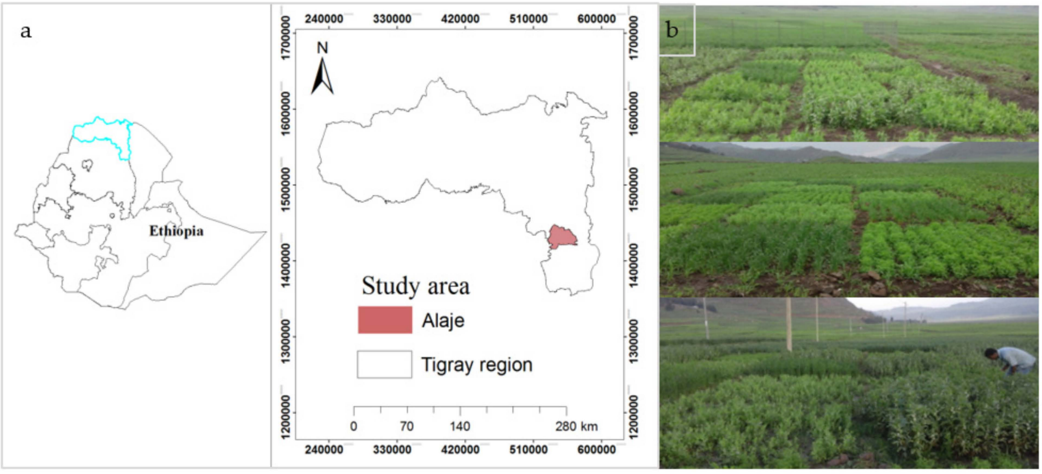

Figure 1. (**a**) Map of study area, and (**b**) four legume crops and the wheat field experiment layout in the Alaje district, Northern Ethiopia.

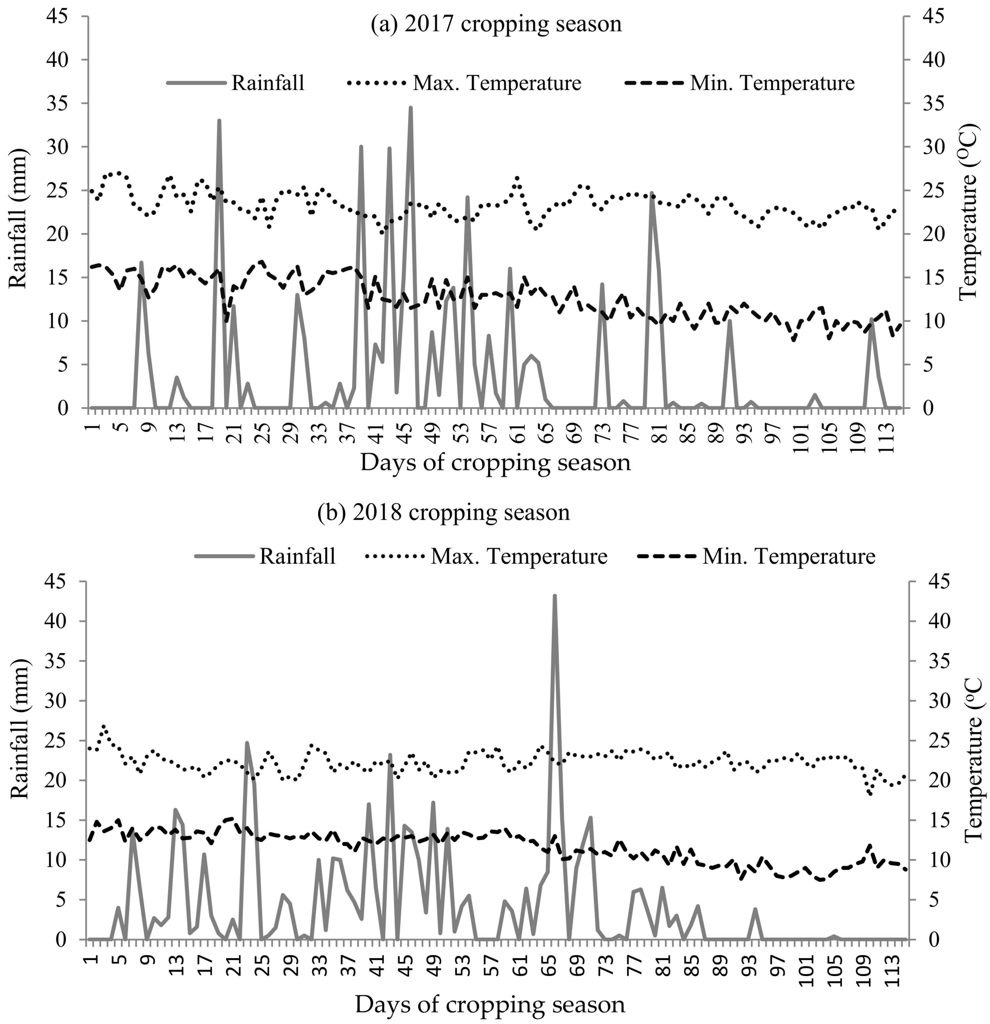

Figure 2. Daily rainfall and temperature in Alaje during July to mid-October of (**a**) 2017 cropping season and (**b**) 2018 cropping season in Northern Ethiopia.

2.2. Experimental Set Up and Management

Soil samples were collected by auger at the depth of 0–20 cm prior to sowing and after harvesting legumes and wheat from each experimental plot during the two experiment seasons. These samples were air-dried, crushed, and sieved by a 2 mm sieve and preserved for analysis. The dominant soil types of the study site are Cambisols, Regosols, Leptosols and Vertisols [21]. To assess the soil characteristics of the experimental plots before sowing and investigate soil N change due to legumes after harvest, soil properties were analyzed using different methods: soil organic carbon (SOC) using Walkley and Black method [22], total nitrogen (TN) using Kjeldahl method [23], available P (Av.P) using Olsen method [24], exchangeable potassium (Exc. K) using ammonium acetate method [25], electrical conductivity (EC) using EC meter, and soil pH using pH meter in 1:2.5 soil-water ratio [26]. The important soil chemical properties of the study area before legume sowing were acidic (pH 6.0) to moderately alkaline (pH 8.0) with average pH of 6.6. The plots show an average EC of 0.3 (0.0–0.9), soil organic carbon (g kg^{-1} soil) of 14.5 (9.4–24.0), soil TN (g $kg^{-1\ soil)}$ of 1.2 (0.5–2.4), available P (mg kg^{-1} soil) of 15.7 (1.6–51.5), and exchangeable potassium (g kg^{-1} soil) of 0.63 (0.05–2.8).

The experiment was conducted on farmers' fields in Alaje. The experiment consisted of four legumes and wheat: faba bean, field pea, dekeko and lentil, and wheat as control. The experiment was conducted for two consecutive cropping seasons (2017 and 2018) and arranged in a randomized complete block design in factorial combinations of four legumes and one cereal crop, four levels of N (0, 10, 20, 46 kg N ha^{-1}) and three levels of P (0, 10, 20 kg P ha^{-1}) with three replications. The experiment was also replicated in six farmers' fields during both cropping seasons. Experimental plots with plot size of 3 m by 3 m were prepared manually with a 20 cm space between rows, 0.5 m between experimental plots, and 1 m between blocks. The sources of fertilizers were urea for N and triple superphosphate for P. The triple superphosphate was applied during planting whereas urea was applied through top dressing after crop emergence. Both fertilizers were applied in a band along the crop lines. Faba bean and field pea were sown early July whereas dekeko, lentil and wheat were sown mid-July. During the growing period, all plots received enough rainfall, and weeds were removed manually. All legumes were harvested early October and wheat early November in both cropping seasons.

2.3. Nodule Assessment and Agronomic Parameters

In both cropping seasons, nodulation was assessed at the mid flowering stage for faba bean, field pea, dekeko and lentil. Five randomly selected plants from each plot were uprooted by excavating the soil with a spade around the plant to a depth of 30 cm. All nodules including detached ones were collected and soil adhered to the root was removed by washing with tap water over a metal sieve. The nodules from each plant root were removed and separately spread on a sieve for some minutes until the water completely drained from the surface of the nodules. The number of nodules per plant was determined by counting all the nodules on each of the five plants. The number of effective nodules was determined by cutting nodules and observing the color inside the nodule. Effective or active nodules were identified by a pink to reddish internal color [27]. The percentage of effective nodules was then calculated through the ratio of effective nodules to total nodules per plant for each plot. All nodules per plant were kept in labeled envelops and oven dried at 70 °C for 24 h, then weighed to determine nodule dry mass. A quadrant (1 by 1 m) sample from the middle of each treatment plot was harvested to determine dry biomass (shoot and grain) yields. Crops were harvested manually, dried, and weighed, while grains were weighed after threshed and winnowed.

2.4. Biological Nitrogen Fixation (BNF)

Sample grain and straw were taken from each plot to analyze their N content. The samples were oven dried at 70 °C for 24 h. The nitrogen content of all legumes and wheat from both seasons were analyzed using the Kjeldahl method for an estimation of BNF. The nitrogen uptakes of each test crop were estimated by multiplying N concentrations with their respective grain and straw yields. Finally, the nitrogen difference method was used to estimate amount of fixed N [28–30] by assuming legume BNF was responsible for the differences in plant N and soil mineral N measured between legume treatments and wheat (Equations (1) and (2)).

Thus,

$$N_2 \text{ fixed} = N_{\text{fixed}=} (\text{plant } N_{\text{legume}} - \text{plant } N_{\text{ wheat}}) \pm (\text{soil } N_{\text{legume}} - \text{soil } N_{\text{wheat}}) \quad (1)$$

where

$$\text{N in plants} = \frac{\left(\text{Dry matter weight kg ha}^{-1} \times \text{ \% N in plants}\right)}{100} \quad (2)$$

All collected data (dry biomass and grain yield, nodule and BNF) were subjected to analysis of variance for factorial experiment using the Gen-stat statistical package. Data normal distribution and homogeneity tests were checked before variable tests. Data with abnormal distribution and unequal variables were transformed using logarithm to the base 10 and checked again for normal distribution and homogeneity tests. Analysis of variance (ANOVA) was used to detect differences. In this experiment, N and P rates were the main factors, and a separate analysis was performed for each legume. The effect of N and P factors and their interactions were compared by computing the standard deviation (SD). Treatment means were compared using the least significant differences (LSD) at $p \leq 0.05$. Since agronomic parameters, nodule characteristics and BNF data had no significant differences between seasons, mean data of the two cropping seasons were presented. The coefficients of variation for all response variables were below 20%.

3. Results

3.1. Dry Biomass and Grain Yield

The result revealed that different N fertilizer rates significantly ($p < 0.05$) affected the dry biomass and grain yield of faba bean, field pea and dekeko (Table 1). Sole application of 20 and 46 kg N ha^{-1} significantly ($p < 0.05$) increased the dry biomass and grain yield of faba bean, field pea and dekeko (Table 1). The sole application of 20 kg N ha^{-1} has increased the yield of faba bean, field pea, dekeko and lentil by 304, 222, 231 and 100 kg ha^{-1}, respectively, over the plot with nil nutrient supply. However, the dry biomass and grain yield of lentil were not significantly affected by N fertilizer rates (Table 1). Like N rates, application of 20 kg P ha^{-1} significantly ($p < 0.05$) increased the grain yield of faba bean, field pea and dekeko (Table 1). However, the dry biomass of faba bean, field pea and dekeko, as well as both the dry biomass and grain yield of lentil, were not significantly affected by P rates (Table 1). The sole application of 20 kg P ha^{-1} has also increased the yield of faba bean, field pea, dekeko and lentil by 316, 302, 302 and 142 kg ha^{-1}, respectively, over the plot with nil nutrient supply. The interaction of different N and P rates also significantly ($p < 0.05$) affected the dry biomass and grain yield of faba bean, field pea and dekeko, which were highest in the interaction of 20 kg N ha^{-1} and 20 kg P ha^{-1} (Table 1). However, the dry biomass and grain yield of lentil were not significantly different among the different interaction of N and P rates. The highest yield increments of faba bean (796 kg ha^{-1}), field pea (507 kg ha^{-1}), dekeko (408 kg ha^{-1}) and lentil (196 kg ha^{-1}) were recorded by the combined application of 20 kg N ha^{-1} and 20 kg P ha^{-1} over the plot with nil nutrient supply (Table 1).

3.2. Effects of N and P Fertilizers on Nodulation and BNF

With the exception of lentil, highest nodule number, nodule dry weight per plant, and BNF were recorded by the application of 20 kg N ha^{-1} (Table 2).

Table 1. Effects of nitrogen (N) and phosphorus (P) on the dry biomass and grain yields of faba bean, field pea, dekeko and lentil. Values representing the mean of the two cropping seasons ± SD are presented.

N and P Rates (kg ha^{-1})	Grain Yield (kg ha^{-1})	Increased Yield (kg ha^{-1}) Over Nutrient Nil Plots	Price of Increased Yield in Birr (US$)	Biomass (kg ha^{-1})	Grain Yield (kg ha^{-1})	Increased Yield (kg ha^{-1}) Over Nutrient Nil Plots	Price of Increased Yield in Birr (US$)	Biomass (kg ha^{-1})
	Faba Bean				**Field Pea**			
0N or 0P	1735 ± 223d	0	0(0)	4104 ± 345d	1986 ± 203c	0	0 (0)	5280 ± 235c
10N	1773 ± 370d	38	950 (29)	4159 ± 432cd	1993 ± 178c	7	245 (8)	5357 ± 657c
20N	2039 ± 237bc	304	7600 (234)	4925 ± 543b	2208 ± 109b	222	7770 (239)	5769 ± 219b
46N	2024 ± 169b	289	7225 (222)	4736 ± 765b	2201 ± 245b	215	7525 (232)	5703 ± 290b
10P	1815 ± 201cd	80	2000 (62)	4382 ± 629c	2004 ± 92c	18	630 (19)	5287 ± 225c
20P	2051 ± 452b	316	7900 (243)	4251 ± 321c	2288 ± 334b	302	10570 (325)	5531 ± 269bc
20N × 20P	2531 ± 87a	796	19900 (612)	5209 ± 154a	2493 ± 192a	507	17745 (546)	6411 ± 285a
lsd	211			271	201			343
cv	19.9			20.0	18.7			19.7
p value	0.01			0.03	0.03			0.01
	'Dekeko'				*Lentil*			
0N or 0P	1286 ± 192c	0	0 (0)	2280 ± 435cd	1561 ± 262a	0	0 (0)	3842 ± 350a
10N	1307 ± 314c	21	1050 (32)	2317 ± 676c	1616 ± 155a	55	1925 (59)	3912 ± 450a
20N	1517 ± 109b	231	11550 (355)	2439 ± 245b	1661 ± 250a	100	3500 (108)	4108 ± 289a
46N	1511 ± 229b	225	11250 (346)	2503 ± 385b	1712 ± 271a	151	5285 (163)	4248 ± 520a
10P	1311 ± 306c	25	1250 (38)	2189 ± 289d	1743 ± 317a	182	6370 (196)	3823 ± 251a
20P	1588 ± 288ab	302	15100 (465)	2231 ± 209cd	1703 ± 215a	142	4970 (153)	3840 ± 371a
20N × 20P	1694 ± 307a	408	20400 (628)	2794 ± 412a	1957 ± 354a	196	6860 (211)	3890 ± 602a
lsd	118			121	662			758
cv	18.8			17.7	18.0			15.8
p value	0.03			0.02	0.23			0.67

NB. Different letters in the same column represent significant differences of mean at $p < 0.05$.

Table 2. Effects of N and P application on the nodulation and nitrogen fixation of legumes in Northern Ethiopia. Values represent the mean for the two cropping seasons ± SD (n = 1080).

N Rates (kg ha^{-1})	P Rates (kg ha^{-1})	TNP	ENP (%)	NDWP (mg)	BNF (kg ha^{-1})	TNP	ENP (%)	NDWP (mg)	BNF (kg ha^{-1})
		Faba Bean				Field Pea			
0	0	52 ± 5f	60.0	118 ± 14g	58 ± 12.9de	5 ± 2g	70	36 ± 11d	19 ± 7c
	10	61 ± 1e	68.6	218 ± 24e	63 ± 15cd	6 ± 2ef	85	67 ± 27bc	20 ± 7c
	20	77 ± 18c	76.3	245 ± 46d	83 ± 22b	7 ± 1d	81	49 ± 16 cd	32 ± 9ab
10	0	67 ± 5d	60.8	225 ± 35e	60 ± 21cd	5 ± 1fg	84	76 ± 29b	20 ± 10c
	10	77 ± 6c	65.8	317 ± 42c	76 ± 19bc	10 ± 2c	88	109 ± 40a	31 ± 12ab
	20	90 ± 4b	68.4	367 ± 62b	88 ± 17ab	11 ± 2b	90	116 ± 15a	32 ± 13ab
20	0	70 ± 4d	69.0	251 ± 68d	78 ± 28b	7 ± 2de	86	83 ± 27b	29 ± 10ab
	10	75 ± 17c	76.9	315 ± 83c	90 ± 17ab	10 ± 1c	88	109 ± 17a	29 ± 10abc
	20	119 ± 11a	80.8	619 ± 99a	97 ± 15a	14 ± 2a	92	223 ± 17a	38 ± 19a
46	0	34 ± 6h	86.4	98 ± 9g	38 ± 10f	3 ± 1i	72	31 ± 11d	9 ± 2d
	10	38 ± 12gh	84.6	154 ± 56f	41 ± 16f	4 ± 1h	82	42 ± 17d	7 ± 2d
	20	39 ± 10gh	75.2	161 ± 35f	45 ± 20ef	3 ± 1hi	76	41 ± 9d	11 ± 5cd
	lsd	5.2		25.0	17.7	0.96		15.5	8.5
	cv%	15.4		11.5	17.6	12.3		15.3	17.8
	p-value	<0.001		<0.001	0.02	<0.001		0.04	0.75
		'Dekeko'				Lentil			
0	0	9 ± 2e	63	66 ± 19f	24 ± 4de	3 ± 1gh	64	6 ± 3gh	9 ± 3bc
	10	9 ± 1e	77	122 ± 48ed	29 ± 6cde	4 ± 1efg	73	10 ± 2fg	14 ± 5abc
	20	13 ± 4cd	76	105 ± 17e	32 ± 7cd	5 ± 1cde	76	14 ± 4ef	15 ± 3abc
10	0	12 ± 4d	70	135 ± 38d	27 ± 6cde	5 ± 2def	75	20 ± 5cd	13 ± 7abc
	10	14 ± 3c	79	150 ± 56d	34 ± 10cd	6 ± 1bcd	78	17 ± 4de	20 ± 6a
	20	17 ± 4b	85	231 ± 56b	36 ± 9bc	7 ± 1b	83	28 ± 4b	19 ± 8ab
20	0	14 ± 4c	79	174 ± 56c	36 ± 12b	6 ± 1bc	79	25 ± 5c	9 ± 3bc
	10	18 ± 4b	83	223 ± 48b	44 ± 12ab	9 ± 1a	82	23 ± 5c	13 ± 5abc
	20	21 ± 5a	88	256 ± 48a	49 ± 18a	8 ± 1a	89	33 ± 2a	14 ± 5abc
46	0	4 ± 1g	63	39 ± 12g	21 ± 8e	2 ± 0h	65	4 ± 1h	5 ± 2c
	10	6 ± 1f	68	104 ± 24e	22 ± 5e	4 ± 1fg	70	12 ± 3f	6 ± 1c
	20	8 ± 2ef	68	73 ± 12f	27 ± 14cde	4 ± 1efg	75	10 ± 3fg	7 ± 3c
	lsd	1.7		22.0	8.5	1.2		4.4	6.0
	Cv%	13.2		11.1	18.6	17.6		13.2	19.7
	p-value	0.004		<0.001	0.01	0.04		<0.001	0.82

NB: Different letters in the same column represent significant differences of mean values at $p < 0.05$. TNP = total nodules per plant, ENP = effective nodules per plant, NDWP = nodule dry weight per plant, and BNF = biological nitrogen fixation.

The N rates significantly ($p < 0.5$) affected nodule characteristics (Table 2). The application of 20 kg N ha^{-1} significantly ($p < 0.05$) increased nodule characteristics, except the nodule number of faba bean and dekeko and the nodule dry weight of field pea. On the other hand, 46 kg N ha^{-1} significantly ($p < 0.05$) decreased nodulation and BNF potentials of legumes (Table 2). Similarly, significantly higher mean nodule characteristics and BNF were observed in the treatment of 20 kg P ha^{-1}, except the nodule dry weight of field pea and the nodule dry weight and BNF of dekeko (Table 2). However, the nodule dry weight of dekeko and the nodule number of lentil did not significantly different between the application of 10 and 20 kg P ha^{-1} rates (Table 2).

The highest BNF (97, 38 and 49 kg ha^{-1}) fixed in faba bean, field pea and dekeko were recorded in plots that received a combined rate of 20 kg N ha^{-1} and 20 kg P ha^{-1} (Table 2) whereas the highest BNF (20 kg ha^{-1}) fixed in lentil was recorded with combined application of 10 kg N ha^{-1} and 10 kg P ha^{-1} (Table 2). Except for lentil the combined application of 20 kg N ha^{-1} and 20 kg P ha^{-} significantly ($p < 0.05$) improved the nodulation and BNF of the other legumes.

3.3. Effect of Legumes on Soil N and Biological Nitrogen Fixation

After the legume harvest, the average N content of soil significantly increased by 37, 21, 26 and 13 kg N ha^{-1} in the faba bean, field pea, dekeko and lentil plots, respectively, compared with the wheat plots and with no differences between the two cropping seasons (Figure 3). The soil N increases were significantly ($p < 0.05$) different among the legume plots which ranked as follows: faba bean (FB) > field pea (FP) = dekeko (D) > lentil (L) (Figure 3).

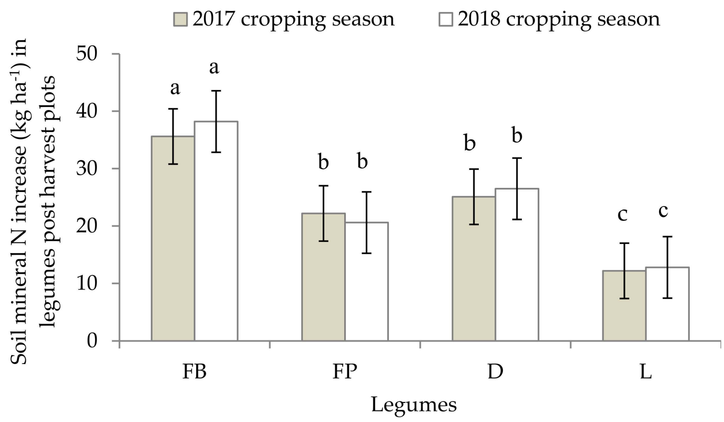

Figure 3. Increase in post-harvest soil mineral N (0–20 cm) following legume treatments relative to wheat. Different letters in the bar graph represent significant differences in mean values at $p < 0.05$. FB = faba bean, FP = field pea, D = dekeko and L = lentil. The soil mineral N in the wheat plot after harvest was 64 kg N ha^{-1} in 2017 cropping season and 58 kg N ha^{-1} in 2018 cropping season.

After harvest, the soil N in the legume and wheat plots were significantly ($p < 0.001$) different. Soil N in the faba bean plot was higher than all other legume and wheat plots (Table 3). The mean soil N accumulated in the faba bean, field pea, dekeko and lentil plots after legume harvest was increased by 37, 21, 26 and 13 kg N ha^{-1} over the wheat plot (61 kg N ha^{-1}), respectively (Table 3).

Faba bean fixed a higher (60%) proportion of N derived from the atmosphere (%Ndfa), whereas for field pea, dekeko and lentil it was 29, 45 and 20, respectively (Table 3). The %Ndfa, BNF and accumulated soil N were significantly ($p < 0.001$) different among the legumes. However, no significant differences in accumulated soil N were observed between field pea and dekeko plots (Table 3). The highest mean nodule characteristics and BNF of the different treatments were obtained by faba bean, which had the highest number of nodules (64) and dry weight of nodules (254 mg) per plant and BNF (69 kg ha^{-1}) (Table 3). The nodule characteristics, such as number of nodules and dry weight of nodules per plant, were significantly ($p < 0.05$) different among the four legume crops (Table 3). The BNF mean results of the four legumes revealed that faba bean, field pea, dekeko and lentil fixed 69, 23, 32 and 16 kg N ha^{-1}, respectively (Table 3).

Table 3. Nodulation and BNF potentials of different legumes in Tigray, Northern Ethiopia. Values represent the mean of the two cropping seasons ± SD.

Legumes and Wheat	Biomass N Accumulation	Ndfa (%)	BNF (kg ha^{-1})	Soil N After Harvest (kg ha^{-1})	TNP	ENP (%)	DWNP (mg)
Faba bean	82 ± 24a	60 ± 23a	69 ± 27 a	98 ± 31a	64 ± 26a	86	254 ± 147a
Field pea	44 ± 13c	29 ± 17c	23 ± 16 c	82 ± 28b	7 ± 4c	86	74 ± 43c
Dekeko	55 ± 18b	45 ± 19b	32 ± 14 b	87 ± 29b	12 ± 6b	75	142 ± 77b
Lentil	38 ± 11d	20 ± 7d	16 ± 9 d	74 ± 23 c	5 ± 2c	80	13 ± 7d
Wheat	31 ± 9e	-	-	61 ± 19d	-		-
lsd	3.7	5.34	3.7	9.9	2.8		17.8
cv	15.9	17.3	12.3	13.4	12.2		11.0
p-value	<0.001	<0.001	<0.001	<0.001	<0.001		<0.001

NB: Different letters in the same column represent significant differences of mean values at $p < 0.05$. Abbreviations for nodulation and BNF are described in Table 2. N= nitrogen, Ndfa = proportion of N derived from the atmosphere (%Ndfa) in the legumes. The values represent the mean for each legume species across all N and P treatment rates.

4. Discussion

4.1. Agronomic Responses to Nutritional Amendments

Application of N and/or P containing fertilizers had comparable effects on legumes' dry biomass and grain yield. The highest dry biomass and grain yields of faba bean, field pea and dekeko were obtained by the combined application of 20 kg N ha^{-1} and 20 kg P ha^{-1}. This is because starter N (20 kg ha^{-1}) together with P (20 kg ha^{-1}) improved early root formation and legume productivity through increased pod loading, dry biomass and grain yield. This could be attributed to an increase in growth parameters and plant photosynthesis, which contributed to higher dry biomass and grain production. The application of 20 kg N ha^{-1} together with 20 kg P ha^{-1} is favorable nutrition for greater grain production and dry biomass yields of the legumes. The interaction of N and P rates are also suitable for legumes to fix N from the atmosphere and contribute more N to legume production. The N derived from both fertilizer and improved BNF lead to greater dry biomass and grain yield of legumes than in the sole application of N and P rates and other interactions. Similar findings by [31] suggested that plant growth parameters of soybean (*Glycine max L. Merill*) increased due to the combination of starter N and P application. Other studies [8,16,32–36] suggested that starter N and P fertilizers significantly improved legume yields through the fulfilment of their N demand by fixing atmospheric N. This indicated that smallholder farmers can achieve dual benefits, such as better legume grain and dry biomass yields with lower N rate (20 kg N ha^{-1}) together with P, and improve their soil fertility for subsequent crops.

The grain yield of faba bean, field pea, dekeko and lentil has increased by 304, 222, 231 and 100 kg ha^{-1}, respectively, due to the sole application of 20 kg N ha^{-1} over the plots with nil nutrient supply. This is equivalent to a benefit of US$ 234, 239, 355 and 108, respectively (Table 1). Similarly, yield increments of 316, 302, 302 and 142 kg ha^{-1}, respectively, were recorded by sole application of 20 kg P ha^{-1} over the plots with nil nutrient supply. This yield increase is similar to US$ of 243, 325, 465 and 153, respectively (Table 1). The combined application of 20N and 20P also increased grain yield of faba bean, field pea, dekeko and lentil by 796, 507, 408 and 196 kg ha^{-1} over the plots with nil nutrient supply. This yield increase is equivalent to US$ 612, 546, 628 and 211, respectively. This is much higher than the price of 20 kg N (17 US$) and 20 kg P (46 US$). The responses to the treatment varied, with very low yields in the unfertilized plots and much higher yields in the treatments, mainly (20N*20P). This wide variation was mainly because of the variation in applied N and P rates. This is a result of the larger biomass N accumulation in the plots fertilized with (20 kg N ha^{-1} and 20 kg P ha^{-1}) as this treatment greatly boosts the BNF and biomass N accumulation of legumes [37]. Farmers in the study area often grew legumes in soils with low soil fertility because they thought that legumes resisted nutrient deficiency. However, the results of this study indicate that legumes require N and P supply to increase their yield and dry biomass. Some studies [8] suggested that although farmers in Africa preferentially allocate cereal crops to more fertile fields on their farms, legumes also demand mineral fertilizer for increased yield.

4.2. Nodulation and Biological Nitrogen Fixation

Highest nodulation and BNF of faba bean, field pea and dekeko were achieved by the combined application of 20 kg N ha^{-1} and 20 kg P ha^{-1}, whereas BNF in lentil was obtained with the combined application of 10 kg N ha^{-1} and 10 kg P ha^{-1}. The lowest nodulation and BNF, on the other hand, were observed where 46 kg N ha^{-1} was applied regardless of the rate of P. These results are consistent with previous studies demonstrating the benefits of low levels of starter N in conjunction with P for enhanced nodulation and BNF by a range of legume crops [8,9,12,14–16,31,32] and the well documented inhibitory effects of high soil nitrate on the BNF process [8,33]. However, it should also be noted that they are at odds with some studies in N-deficient soils where higher rates of N have been reported to be required to boost nodulation [34–36]. Therefore, the integration of legumes into the cropping system is an option to improve soil fertility in the study area and other semi-arid lands.

Besides the fertilizer rates, different legumes grown in the study area have different nodulation and BNF potentials (Table 3; Figure 4a–d). For instance, faba bean fixed higher N (69 kg N ha^{-1}) than dekeko (32 kg N ha^{-1}), field pea (23 kg N ha^{-1}) and lentil (16 kg N ha^{-1}). This is similar to research findings by [37,38] that suggested faba bean has high potential for nodulation and BNF. The nodules in all four legumes with a pink color (Figure 4e) have shown an active N-fixation because nodules with pink colors are effective nodules and actively fix N due to presence of leghaemoglobin [39]. The effects of legumes, N and P rates, and their interaction effects on nodulation and BNF, showed that the highest nodulation and BNF were obtained by faba bean, with a combination of 20 kg N ha^{-1} and 20 kg P ha^{-1} (Figure 4a–d). This indicated that faba bean has the highest N requirement, and as a result, it developed highest effective nodules per plant. The BNF difference among legumes is due to the ability of legumes to fix atmospheric N [8,9,14,15]. This helps smallholder farmers to select legume crops during crop rotation for their soil fertility management. The increased accumulated soil N from 13 kg N ha^{-1} in the lentil plot to 37 kg N ha^{-1} in the faba bean plot (Figure 2, Table 3) can be seen as evidence for the selection of legumes during crop rotation. It was also suggested [40] that an average of 25–35 kg N ha^{-1} change in soil N was observed in the legume plot over the wheat plots. This clearly showed that BNF is one of the most important sources of soil N in the agricultural system [41].

Figure 4. Nodules: (**a**) faba bean, (**b**) field pea, (**c**) dekeko, (**d**) lentil, and (**e**) pink color of effective nodules.

The legumes also accumulated considerable amounts of N in their biomass from the atmosphere (Ndfa) and stored N in the soil for the benefit of succeeding cereal crops. This indicated that legumes fix N from the atmosphere for their productivity and contribute more to improve soil N for sustainable cropping systems than wheat [42]. This is of paramount importance for farmers who are not able to apply expensive inorganic fertilizers. Based on the current urea price (12.85 ETB kg^{-1}) for the above mentioned amount of BNF for each legume, the estimated benefits of the fixed N were 1917.72, 881.05, 653.10 and 440.80 ETB ha^{-1}, respectively. This is equivalent to US$ ha^{-1} of 58.38, 26.82, 19.88 and 13.42, respectively. From this it can be concluded that farmers can save 1917.72 ETB ha^{-1} (58.38 US$ ha^{-1})

when they use faba bean for crop rotation. Therefore, as the study area has potential for legume crops, findings from this study have important policy implications. The policy implications are that the different N and P rates have increased the BNF potential of legumes, which enhances subsequent crop yields that can contribute to food security. Therefore, the BNF process offers an economically feasible and ecologically sound means of reducing external N input through improving soil nutrient content.

5. Conclusions

This study showed that 20 kg N ha^{-1} and 20 kg P ha^{-1} rates have increased the nodulation, BNF, dry biomass and grain yields of faba bean, field pea and dekeko. The application of 20 kg N ha^{-1} increased the grain yield of faba bean, field pea, dekeko and lentil by 304, 222, 231 and 100 kg ha^{-1}, respectively, whereas the application of 20 kg P ha^{-1} increased it by 316, 302, 302 and 142 kg ha^{-1}, respectively, compared with the plot with nil nutrient supply. On the other hand, the interaction of 20N and 20P increased the grain yield benefits of faba bean, field pea, dekeko and lentil by 796, 507, 408 and 196 kg ha^{-1}. This yield benefit is equivalent to US$ of 612, 546, 628 and 211, respectively, compared with the plot with nil nutrient supply. Moreover, the four legumes have accumulated atmospheric N in their biomass and store considerable amount of soil N. Faba bean showed a surplus accumulation of 37 kg ha^{-1} soil N over the wheat plot, indicating the potential of supplying soil mineral N for subsequent cereals. Starter fertilizer level (20 kg N ha^{-1}) in combination with 20 kg P ha^{-1} are required for optimum dry biomass and grain yields, nodulation and BNF of faba bean, field pea and dekeko in the study area, whereas 46 kg N ha^{-1} of fertilization inhibited the nodulation and N fixation potentials of faba bean, field pea, dekeko and lentil. Hence, it can be concluded that the studied legumes were poor at fixing adequate atmospheric N unless supplied with starter N together with P. The same can be said when higher N (46 kg N ha^{-1}) was applied. This study could be important input for soil fertility management strategies describing the N fixation potentials of legume crops. Faba bean has higher potential for nodulation and BNF than field pea, dekeko and lentil, which could be a major pathway to increasing subsequent crop yield and reducing environmental footprint. The policy implication of this study is that since mineral N fertilizer is expensive and sensitive to losses, maximizing BNF through supplying starter N and P appears to be a suitable way of improving legume yield. The N that is accumulated in the soil due to BNF can be used by subsequent crops to increase productivity.

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