



Article Integrated Fertilizers Synergistically Bolster Temperate Soybean Growth, Yield, and Oil Content

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Abstract: For ensuring food security and imparting sustainability to modern commercial-oriented and highly intensive temperate farming systems, organic wastes from poultry and dairy industries constitute biologically viable strategy to improve crops productivity under changing climate. A field trial was undertaken to appraise the impact of broiler litter (BL = 5 tons ha⁻¹), farm yard slurry (FYS = 10 tons ha⁻¹), and chemical fertilizers including di-ammonium phosphate (DAP = 60 kg ha^{-1}) and single super phosphate (SSP = 60 kg ha^{-1}) applied solely and in conjunction with each other, along with a control treatment (NM). The synergistic fertilization regime encompassing BL+DAP triggered the vegetative growth of soybean as indicated by taller plants having thicker stems and higher leaf area per plant compared to NM. In addition, this fertilization management system improved reproductive yield attributes including pods number and 100-seeds weight which maximized the seed yield, harvest index, seed oil content, and biological yield by 66%, 5%, 31%, and 23% respectively than NM. Moreover, this fertilizers combination was followed by SSP + BL, while BL performed better than FYS and DAP remained superior to SSP. Furthermore, the correlation analyses indicated moderately stronger direct association of seed yield with vegetative growth traits and highly stronger linear relationship with reproductive yield attributes. Thus, co-application of broiler litter (5 tons ha^{-1}) with reduced doses of DAP (60 kg ha^{-1}) might be recommended to temperate soybean growers having access to poultry wastes.

Keywords: sustainable farming; chicken manure; farm yard slurry; single super phosphate; harvest index

1. Introduction

Globally, leguminous crops are of vital pertinence for ensuring the food and nutritional security of rapidly increasing population especially in temperate regions of developing countries in South Asia (Pakistan, India, and Bangladesh). Among leguminous crops, soybean (*Glycine max* (L.) Merrill) constitutes one of the largest cultivated crops of Fabaceae family in the world [1]. It finds multi-dimensional uses in recent times and has been declared as the meat of plant origin and king of beans due to its high quality protein [2].



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Copyright: © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). During 2020–2021, global soybean production remained over 361 million metric tons harvested from an area of around 121 million hectares [3]. Soybean has been classified among oilseed crops instead of pulse crops as over 85% of global soybean is processed into vegetable oil that fulfills half of the edible oil demand worldwide. In south Asia, soybean production has remained suboptimal owing to low soil fertility and inadequate fertilization in terms of quantity, quality, and time of application [4–6]. Despite rapid increase in food requirement, declining soil fertility has seriously slowed the growth of food production in temperate regions of Asia [7]. In addition, sole reliance on mineral fertilizers in legumes production systems has been the prime cause of significant slicing in food production on per capita basis.

The temperate Himalayan regions of Pakistan and India have witnessed rapidly decreasing soil fertility over time leading to decline in crop productivity [6,8]. It needs emphasis that for ensuring food security, healthy soils form the foundation of food production, conservation of water and biodiversity especially bacterial community in the rhizosphere and nutrient cycling in agricultural eco-systems [9]. Recently, soil health (soil potential as a living system for sustaining plant and animal productivity along with maintaining water and air quality) and environmental pollution caused by excessive use of chemical fertilizers have received due attention [10,11]. The changing climate scenario has further aggravated the need to sustainably supply nutrients to soybean crop for maximizing grain yield and economically sustain the health of the agricultural soils. The organic manures applied solely or in conjunction with reduced doses of mineral fertilizers hold potential to improve soil health and soybean productivity [12]. These can effectively fulfill crop nutritional needs along with supporting associated biological, chemical, and physical processes to produce soybean yield as per varietal potential and support the rural-based farm economy [13,14]. Among organic manures, wastes of poultry sheds and dairy farms yard slurry have recently been recognized as rich and concentrated source of macro and micro nutrients. It is interesting to mention that world poultry meat production has recently surpassed 100 million tons, which generates over 75 million tonnes of litter (BL) annually that have contributed to environmental pollution owing to inappropriate disposal [15]. The BL generated by broiler chicken (Gallus gallus domesticus) industry is a mixture of chicken excreta and bedding material. Typically, every broiler chicken produces 1.1 to 1.5 kg litter which may supplement chemical fertilizers provided optimization of their dose and application time are done based on field studies under specific pedo-climatic conditions [16]. Similarly, dairy industry also generates huge tonnage of farm yard slurry that might serve as rich source of plant nutrients. The composted BL and cattle farm yard slurry (FYS) integration with mineral fertilizers hold bright perspectives to decrease environmental challenges including eutrophication [17]. In addition, phosphorous (P) recycling from BL and FYS may constitute as vital strategy for extending the life of mineral P reserves along with attenuating the environmental contamination. The FYS application tend to improve soil organic matter (OM) content, microbial biomass in the rhizosphere, cation exchange capacity (CEC), metals complexation, and steady nutrient release for a longer period of time during critical crop growth stages [18]. Likewise, BL applied solely and in conjunction with reduced doses of chemical fertilizers resulted in lesser N losses through negligible leaching and volatilization compared to solo mineral fertilizers (MF) especially urea and nitrophos [19]. Furthermore, slow-release characteristic of BL and FYS significantly reduced P losses by adsorption to soil minerals and this property may assist considerably in improving P fertilization in temperate weathered soils having low P recovery from conventional water soluble P fertilizers [20]. Typically, the soils of Pakistan's temperate regions (especially autonomous region of Azad Jammu and Kashmir) are low in OM and water holding capacities along with being extremely eroded owing to water erosion, while BL and FYS application to such soils may warrant significant increase in OM and overall soil health. However, Deeks et al. [21], and Morais and Gatiboni [22] concluded that there was no significant difference among different doses of BL, FYS, and P mineral fertilizer in terms of P availability in the soil solution and P absorption estimated in plant

tissues. Thus, changing climate scenario and conflicting findings regarding BL efficacy as a supplement and/or substitute of chemical fertilizers have further aggravated the need to conduct field studies in order to bridge the research and knowledge gaps.

Being a leguminous crop, soybean plants primarily meet their nitrogen (N) requirement from symbiotic fixation (BNF) process, however, a number of field studies have reported contradictory findings regarding the impact of organic manures on soybean yield [7,9,23]. Schmidt et al. [24] inferred that soybean yield was enhanced by 1.4 kg per kg of applied N in the form of well-composted organic manures especially chicken manure. Likewise, Varvel and Peterson [25] noted that soybean crop yield of 2.5–3.4 Mg on per hectare basis needed at least 200 kg N that could not be met from BNF process and deficient nutrients were supplied in the form of farm yard manure. It was inferred that organic manures especially BL remained effective in supplying various micronutrients that ensured optimal growth and grain yield of soybean [26]. However, Gates and Muller [27] found that co-application of organic manures with mineral fertilizers of N, P, and S in soybean crop resulted in a stronger symbiotic association and greater fixation of N. These findings are further supported by Adeli et al. [28] who inferred that soybean yield was increased by 9% owing to BL application which supplied abundant quantities of secondary and micro nutrients. Likewise, Garcia and Blancaver [29] reported that organic manures improved the grain yield of soybean by 62%. In complete disagreement, Quinn and Steinke [26] reported no significant impact of organic manures from plant and animal origin on soybean yield in traditional and intensive farming systems. Similarly, Slaton et al. [30] also inferred unresponsiveness to BL application in eleven fertilization trails in comparison to P and K conventional fertilizers applied at equivalent rates. Thus, it becomes clear that previous findings pertaining to effectiveness of organic wastes in crop production are contradictory and to the best of our study, reports are scant on their performance under temperate climate.

Therefore, it becomes pertinent to bridge research and knowledge gaps regarding source and dose optimization of organo-mineral fertilization regimes which may constitute a potent approach for mitigating the adverse effects of environmental pollution along with boosting soybean yield and seed oil content. To this end, it was hypothesized that organo-mineral synergistic fertilization regimes could exert a growth boosting influence on soybean plants by improving different micro and macro nutrients availability slowly over a longer period of time during crop growth stages. In contrast, temperate rainfed conditions might also potentially cause growth-restricting effects on soybean plants that are supplied organic fertilizers owing to slow release of nutrients compared to chemical fertilizers. Therefore, this multi-year field study was undertaken to appraise the influence of the organo-mineral fertilization regimes entailing broiler litter, farm yard slurry, single super phosphate, and di-ammonium phosphate, on growth, yield attributes, grain yield, and oil content of soybean sown under rainfed conditions in temperate climate.

2. Materials and Methods

2.1. Description of Meteorological and Physico-Chemical Characteristics of Experimental Site

The Research Farms of the University of Poonch Rawalakot, Azad Jammu and Kashmir, Pakistan (33.8584° N, 73.7654° E, altitude of 1638 m) [31], was the location of the experiments for two consecutive years (2018 and 2019). The locality of the experiment is illustrated in Figure 1. The sowing of the experimental crop (soybean) was performed after winter wheat harvesting on 23 June and 28 June during 2018 and 2019 respectively. The mean meteorological data regarding temperature and precipitation of our experimental location are presented in Figure 2. The locality of trials is characterized by rainfed farming systems that usually receives sufficient precipitation for growing crops like soybean, sorghum, maize, wheat, and various vegetables.



Figure 1. The map of the trial's location (Rawalakot, Azad Jammu, and Kashmir, Pakistan) prepared using QGIST software whereby black doted place indicates the proximate location of the trial and half-arrow depicts North direction.



Figure 2. The trial location's (Rawalakot, Azad Jammu and Kashmir, Pakistan) meteorological characteristics (temperature and precipitation) over crop growing seasons, (data with exponential trend lines).

For performing the physico-chemical analyses, pre-sowing soil samples (0–15 cm and 15–30 cm depths) were collected from the center and four corners of the experimental block. Subsequently, the soil samples (belonging to both soil depths) were homogenized thoroughly by hand mixing and thereafter, the samples were shade dried, grounded, and sieved (using sieve having 2 mm pore size). For estimating the pH of soil, pastes of soil using 1:2.5 ratios of soil:water were prepared which were then subjected to the glass electrode (HI-98107, Hanna Instruments, Szeged, Hungry) for recording the pH [32]. The conductivity meter (Hi-8033, Hanna Instruments, Szeged, Hungry) was used for estimation of electrical conductivity (EC) of the soil samples [33]. In addition, for estimating organic carbon (OC) content of the soil, wet oxidation method was put into practice. Mean-

Rawalakot, Poonch, Azad Kashmir, Pakistan

while, organic matter (OM) content was also determined by following the methodology of Walkley–Black [34]. Among primary plant nutrients, total nitrogen (N) content of soil samples was estimated using Kjeldahl apparatus (ATN-300, Drawell, Shanghai, China) for distillation and subsequent titration using H_2SO_4 (concentrated acid) (Sigma-Aldrich, St. Louis, MI, USA) [35]. Phosphorous (P) content was measured by following Olsen's method involving 0.5 N NaHNO₃ (Sigma-Aldrich, St. Louis, MI, USA) at 8.5 pH from paste of soil: extractant in 1:10 ratio and thereafter spectrophotometer (MTAM-0743, Hanna Instruments, Hungary) having wavelength set at 882 nm in a system containing sulfuric acid was used [36]. Finally, potassium (K) content in the soil samples was estimated by following the standard protocols which involved ammonium acetate extraction (shaking soil samples with ammonium acetate solution of 0.5 M for 30 min) that resulted in positively charged K ions displacement and their detection was done using a flame photometer (FP-910, PG Instruments, Lutterworth, UK). Regarding micronutrients estimation in soil samples, extraction method involving ammonium acetate solution (CH_3COONH_4) (Sigma-Aldrich, St. Louis, MI, USA) by maintaining 3.0 pH of the soil paste was used for measuring available iron (Fe). Thereafter, colorimetric method and spectrophotometer (MTAM-0743, Hanna Instruments, Szeged, Hungary) (510 nm wavelength) were used for determining the Fe content. Moreover, other micronutrients such as boron (B), zinc (Zn), copper (Cu), and manganese (Mn), were determined using the extraction method involving diethylenetriaminepentaacetic acid [37,38]. The soil had a loam texture, with a pH 7.8, while OM content was 1.05%, indicating sub-optimal fertilization management of the soil. In addition, bulk density and EC of the experimental block remained 1.24 cm⁻³ and 0.45 dS m⁻¹, respectively indicating normal soil with respect to salinity level. Among macro-nutrients, NPK contents were 83, 5.2, and 171 mg kg⁻¹, respectively. The micronutrients including B (1.03 mg kg⁻¹), Mn (23.2 mg kg⁻¹), Fe (12.6 mg kg⁻¹), Cu (1.93 mg kg⁻¹), and Zn (1.34 mg kg⁻¹) were also determined.

2.2. Experimentation Details

The trial was consisted of different organo-mineral fertilization regimes involving single super phosphate (SSP) (60 kg P ha⁻¹), di-ammonium phosphate (DAP) (60 kg P ha⁻¹), farmyard slurry (FYS) (10 tons ha⁻¹), broiler litter (BL) (5 t/ha), SSP + BL (60 kg P ha⁻¹ + 5 tons ha⁻¹), SSP + FYS (60 kg P ha⁻¹ + 10 tons ha⁻¹), DAP + FYS (60 kg P ha⁻¹ + 10 tons ha⁻¹), DAP + BL (60 kg P ha⁻¹ + 5 tons ha⁻¹), and a control treatment (NM) in each replication was maintained for comparison purpose. After calculation, doses of manures were optimized in order to maintain the parity of applied plant nutrients in all experimental treatments. The organic manures (OM) were incorporated into soil (20 cm depth) as a basal dose before sowing as the trial was conducted under rainfed conditions so no splitting of fertilizers was done. The OM were applied as per treatment and incorporated in the soil 23 days before sowing in order to allow sufficient time for decomposing and initiation of nutrients release process. Additionally, considering the rapid release of nutrients from chemical fertilizers, SSP and DAP were applied at the time of sowing in accordance with treatments. The field trial was executed in the regular arrangement of a randomized complete block design (RCBD) having three replications of every experimental treatment. The net plot size of experimental units (after deducting the area under water channel, plot bunds and walking paths) was $4 \text{ m} \times 3 \text{ m}$ (1.5 feet wide earthen bunds surrounded the experimental plots that were separated by fellow area of 3 feet, while replications were separated by maintaining fellow areas of 5 m). In order to obtain fine tilth of the seed-bed, three ploughings with common cultivator (tractor-driven) followed by plankings were performed which thoroughly pulverized the soil of the experimental block. Soybean seeds (cv. Rawal-1 developed by National Agricultural Research Center, Islamabad, Pakistan in 1993, is an early maturing, determinate, erect-type, and high yielding variety being grown in rainfed regions of Punjab, Pakistan) were subjected to hydro-priming (seed soaking in sterilized water) for 30 h for obtaining higher germination rate and attain vigorous growth of the young seedling. Thereafter, shade drying of soaked seeds was performed and seed

storage was done at 10 °C. Soybean crop using 100 kg seed rate ha⁻¹ was sown using single row hand drill in 50-cm apart rows by maintaining plant to plant spacing of 25 cm. Soybean crop was kept weed free by manually uprooting the weeds four to five times during both the crop growing seasons. The nutritional profiles of broiler litter (N = 31.6, P = 13.2, K = 8.3, Ca = 9.2, Mg = 3.7 mg kg⁻¹ of the soil) and farm yard slurry (N = 16.1, P = 7.9, K = 10.1, Ca = 6.8, Mg = 3.9 mg kg⁻¹ of the soil) used as treatments are illustrated in Figure 3.



Figure 3. Chemical composition regarding primary and secondary plant nutrients (mg kg⁻¹) in organic manures (broiler litter BL, farm yard slurry FYS) (data with exponential trend lines).

2.3. Data Recordings of Response Variables

Data pertaining to growth attributes of soybean including plant height (measured using tailor's tap from base of plant up till leaf tip), leaf number, leaf area (using leaf area meter), stem girth (with the help of vernier caliper), number of branches and pods per plant, number of seeds per pod, 100 grains weight (using digital balance) were recorded by randomly selecting 10 plants from interior rows of experimental plots, and subsequently their averages were computed. The grain yield of soybean was estimated by harvesting all plants in every experimental unit and bundled separately for subsequent threshing. Thereafter, seed yield was converted into per hectare basis using Equation (1). For estimation of biological yield, plants harvested from an area of 1 m² in each experimental plot were weighed separately using spring balance in the field and thereafter converted into per hectare by using formula 2. Additionally, harvest index (a measure of reproductive efficiency depicting grain ratio to total biomass) was also estimated using Equation (3). Finally, oil content of soybean sown under synergistic fertilization regimes was determined by using Soxhlet apparatus having n-hexane (60 °C) as an organic solvent by following the methodology as prescribed by [38].

Grain yield of soybean =
$$\frac{\text{Yield per plot}}{\text{Plot area } (m^2)} \times 10,000 \text{ m}^2$$
 (1)

Biological yield of soybean =
$$\frac{\text{Whole plants biomass per plot}}{\text{Plot area } (m^2)} \times 10,000 \text{ m}^2$$
 (2)

Harvest index of soybean =
$$\frac{\text{Total seed yield}}{\text{Total biological yield}} \times 100$$
 (3)

2.4. Statistical Analyses

The recorded data pertaining to response variables were arranged and subjected to statistical analysis by employing Bartlett's test, that indicated non-significant effect of the year. Thus, year-wise data were transformed into the mean values and subsequently used for determining statistical significance among treatment means. Thereafter, Fisher's ANOVA (analysis of variance) technique was employed for estimation of overall significance, while treatment means were compared with the help of Tukey's honest significant difference (HSD) test employed at 5% level of probability using SAS statistical package (Version 9.2, SAS Institute, Cary, NC, USA) [39,40].

3. Results

3.1. Vegetative Growth Attributes

The findings revealed that fertilization regimes had significant influence on vegetative growth of temperate soybean in comparison to control treatment (NM = no mineral or organic soil amendment) (Table 1). The synergistic organo-mineral fertilization management system involving combined application of DAP and BL remained superior by recording the tallest plants of soybean (11%, 14%, and 13% taller than NM, sole BL and sole DAP respectively) and it remained statistically at par to BL applied in conjunction with SSP that exhibited the plant height of 80.85 cm. The treatment combination entailing co-application of mineral fertilizers with FYS followed it. In addition, sole BL and FYS recorded 11% and 10% respectively taller plants than NM. For stem girth, BL applied in conjunction with DAP and SSP outperformed rest of the fertilization regimes by recording the maximum stem diameter of 10.47 cm, while NM recorded the most thin stemmed (8.29 cm) plants. Interestingly, sole BL remained statistically equivalent to integrated application of DAP and FYS, while combined application of FYS and SSP followed them as far as stem diameter was concerned. In contradiction, the combined application of BL + DAP, BL + SSP, and FYS + DAP was performed in parity with each other by giving the maximum number of branches per plant. On the other hand, sole BL remained non-significant to combined application of FYS + SSP, while these treatment combinations were trailed by solo FYS regarding branch numbers per plant of temperate soybean. Pertaining to number of leaves of per plant, treatment combinations of BL + DAP and FYS + DAP remained statistically at par by exhibiting 106% greater leaf number than NM. However, BL + FYS remained superior to sole BL and FYS + SSP which in turn outmatched solo application of both DAP and FYS. Similar to previous trend, DAP + BL and DAP + FYS performed statistically at par to each other by recording leaf areas of 1786 cm² and 1775 cm² respectively that were 149%, 6%, and 9% higher compared to NM, sole BL and, sole DAP respectively. Among rest of the fertilizers management systems, SSP + FYS showed 140% higher leaf area per plant in comparison to NM.

Table 1. Vegetative growth attributes of soybean as influenced by organo-mineral synergistic fertilization regimes under temperate conditions. (mean values).

Treatments	Plant Height (cm)	Stem Girth (cm)	Number of Branches per Plant	Number of Leaves per Plant	Leaf Area per Plant (cm ²)
NM	64.01 d	8.29 f	10.10 e	32.23 f	717.7 g
$SSP = 60 \text{ kg P ha}^{-1}$	71.63 c	9.31 e	13.30 de	38.16 e	1311.0 f
$DAP = 60 \text{ kg } P \text{ ha}^{-1}$	71.94 c	9.36 de	13.66 d	43.83 d	1628.3 e
$FYS = 10 \text{ tons } ha^{-1}$	70.83 cd	9.40 d	14.53 c	42.60 d	1626.7 e
$BL = 5 t ha^{-1}$	71.29 с	10.05 b	15.31 b	48.61 c	1690.0 d
$SSP + BL = 60 \text{ kg P ha}^{-1} + 5 \text{ tons ha}^{-1}$	80.85 ab	10.46 a	16.60 a	52.41 b	1701.8 c
$SSP + FYS = 60 \text{ kg } P \text{ ha}^{-1} + 10 \text{ tons } \text{ha}^{-1}$	74.80 b	9.62 c	15.43 b	44.82 c	1715.6 b
$DAP + FYS = 60 \text{ kg } P \text{ ha}^{-1} + 10 \text{ tons } \text{ ha}^{-1}$	74.24 b	10.04 b	16.33 a	65.47 a	1775.3 ab
$DAP + BL = 60 \text{ kg} P ha^{-1} + 5 \text{ tons } ha^{-1}$	81.3 a	10.47 a	16.73a	66.12 a	1786.4 a

SSP = single super phosphate, DAP = di-ammonium phosphate, FYS = farm yard slurry, BL = broiler litter, NM = no amendment as control treatment. Letters in columns indicate difference among values having statistically significant difference ($p \le 0.05$) from each other while values having same letter within column depict statistical parity among those values.

3.2. Reproductive Yield Attributes, Seed Yield, and Biological Yield

The research findings showed quite significant impact of OM including BL and FYS applied solely and in conjunction with chemical fertilizers (DAP and SSP) pertaining to reproductive yield attributes of soybean (Table 2). Regarding the number of pods per plant of temperate soybean, it varied from 25 to 37 whereby NM (no soil amendment) gave the lowest value while DAP + BL recorded the highest value (48%, 6%, and 14% higher than NM, BL + SSP and DAP + FYS respectively). It was also observed that BL + SSP remained superior to DAP + FYS, that in turn performed better than SSP + FYS, while sole BL was statistically equivalent to sole DAP as far as pods number per plant of soybean were concerned. However, all fertilization regimes remained insignificant for number of seeds per plant of soybean. In contrast, 100-seeds weight exhibited by integrated fertilization of DAP + BL was 53% higher in comparison to NM. It was followed by SSP + BL which showed 9% lesser seed weight than DAP + BL, while sole BL outperformed FYS and sole DAP application surpassed sole SSP treatment. Additionally, integrated fertilization encompassing DAP + BL recorded the highest values of seed yield (66% greater than NM) and biological yield (23% higher compared to NM), while corresponding values for following treatment combination of SSP + BL were 3% and 1.2% lower in comparison to DAP + BL (Table 2). Interestingly, FYS performed better in conjunction with DAP compared to FYS + SSP. Similarly, sole BL outperformed sole FYS in terms of seed yield and biological yield by recording significantly higher corresponding values of 4% and 1% respectively. On similar fashion, DAP surpassed SSP by exhibiting 8% and 1.4% greater values of seed and biological yields respectively. Overall, all fertilization regimes outperformed control treatment, however integrated fertilization involving chemical fertilizers especially DAP and organic manures especially BL remained superior regarding reproductive yield attributes and seed yield of soybean.

Table 2. Reproductive yield attributes, seed yield and biological yield of soybean as influenced by synergistic fertilization regimes under temperate conditions. (mean values).

Treatments	Number of Pods per Plant	Number of Seeds per Pod	100 Seed Weight (g)	Seed Yield (t ha ⁻¹)	Biological Yield (t ha ⁻¹)
NM	25.53 f	2.39	13.36 f	0.93 h	3.25 f
$SSP = 60 \text{ kg P ha}^{-1}$	27.33 f	2.38	14.06 ef	1.16 g	3.52 e
$DAP = 60 \text{ kg P ha}^{-1}$	31.80 d	2.41	15.36 de	1.25 f	3.57 d
$FYS = 10 \text{ tons } ha^{-1}$	29.96 e	2.39	14.20 e	1.35 e	3.55 de
$BL = 5 \text{ tons } ha^{-1}$	31.70 d	2.45	15.10 de	1.40 d	3.58 d
$SSP + BL = 60 \text{ kg } P \text{ ha}^{-1} + 5 \text{ tons } \text{ha}^{-1}$)	35.80 b	2.42	19.43 b	1.51 b	3.96 b
$SSP + FYS = 60 \text{ kg P ha}^{-1} + 10 \text{ tons ha}^{-1}$	31.33 d	2.45	18.06 c	1.41 d	3.78 d
$DAP + FYS = 60 \text{ kg } P \text{ ha}^{-1} + 10 \text{ tons } \text{ha}^{-1}$	32.90 c	2.43	15.53 d	1.48 c	3.88 c
$DAP + BL = 60 \text{ kg} P ha^{-1} + 5 \text{ tons } ha^{-1}$	37.70 a	2.46	20.40 a	1.56 a	4.01 a

SSP = single super phosphate, DAP = di-ammonium phosphate, FYS = farm yard slurry, BL = broiler litter, NM = no amendment as control treatment. Letters in columns indicate difference among values having statistically significant difference ($p \le 0.05$) from each other while values having same letter within column depict statistical parity among those values.

3.3. Harvest Index and Seed Oil Content

The results of our field trial depicted that organo-mineral fertilization regimes significantly influenced the harvest indices (HI) and seed oil content (SOC) of temperate soybean (Figure 4). The HIs ranged from 30–39%, whereby co-application of BL and DAP superpassed rest of treatments by recording the maximum HI of 39.2% (9% higher than control treatment) while sole application of BL performed statistically at par to it (Figure 4A). In addition, BL + SSP performed statistically equivalent to DAP + FYS by recording 5% greater HI compared to NM. Overall, BL remained superior to FYS while DAP performed better than SSP, however all integrated fertilization management systems statistically outperformed control treatment (NM). Pertaining to SOC, DAP + BL recorded significant superiority (31% higher than NM), while it was followed by combined application of DAP + FYS which recorded 11% lower and 17% higher SOC than DAP + BL and NM respectively (Figure 4B).



To sum up, BL performed better with DAP and was trailed by SSP + BL and sole BL. Interestingly among solo manures, BL resulted in 3% higher SOC than DAP which in turn recorded 4% higher corresponding value in comparison to sole SSP.

Figure 4. (A) Harvest index (%) and (B) seed oil content (%) of temperate soybean under synergistic fertilization regimes involving no manure (NM), DAP (di-ammonium phosphate), SSP (single super phosphate), FYS (farm yard slurry), and BL (broiler litter) applied solely and in conjunction with each other. (mean values having different letter indicating statistical difference at $p \le 0.05$)).

3.4. Correlation among Yield Attributes, Seed Yield, and Biological Yield

The correlation analyses among seed yield and yield attributes (both vegetative and reproductive) of temperate soybean under organo-mineral synergistic fertilization regimes are of vital pertinence owing to varying requirement of spreading and towering type cultivars. It indicated moderate degree of positive relationships ($R^2 = 0.76$ *) between seed yield and plant height as the minimum plant height of 64 cm resulted in 0.93 tons ha⁻¹ of seed yield, while 26% increment in plant height maximized the seed yield to the extent of 64% (Figure 5a). In addition, linear relationship of seed yield with stem girth ($R^2 = 0.97$ **) was recorded whereby 23% enhancement in stem diameter yielded 48% higher productivity (Figure 5b). Likewise, leaf area also exhibited stronger direct association with seed yield of soybean ($R^2 = 0.85$ **) whereby 1070 cm² increment in leaf area boosted seed yield by 0.63 tons ha⁻¹ that corresponds to over 65% increment in seed yield (Figure 5c). On similar fashion, reproductive traits of soybean including number of pods per plant recorded direct association ($R^2 = 0.89$ **) with seed yield by improving it to the degree of 65% as the number

of pods was increased from 25 to 37 (Figure 5d). Importantly, 100-seeds weight was strongly correlated ($R^2 = 0.95$ **) with seed yield as the addition of 7 g weight in 100-seeds weight resulted in bolstering of seed yield by 0.63 tons ha⁻¹ (Figure 5e). Interestingly, reproductive yield attributes (number of pods and 100-seeds weight) were more strongly associated with seed yield of soybean compared to vegetative growth traits like plant height.



Figure 5. Cont.



Figure 5. Correlation analyses depicting relationship between seed yield and yield attributes such as plant height (**a**), stem girth (**b**), leaf area per plant (**c**), number of pods per plant (**d**), and (**e**) 100-seeds weight of soybean. * = significant at $p \le 0.05$, ** = highly significant significant at $p \le 0.05$.

4. Discussion

The findings of this study pertaining to unmatched impact of broiler litter and DAP on vegetative growth traits especially plant height, stem girth, and number of branches per plant corroborate with those of Watts and Torbert [41] who concluded that achieving potential growth of crop plants remains challenging in most soils having inadequate fertility to appropriately fulfill plant nutrient requirements, while reduced doses of chemical fertilizers applied with OM increased vegetative growth traits especially plant height, stem diameter, and leaf area per plant. In contrast to our findings, Slaton et al. [30] observed no significant influence of BL on plant height and stem diameter of plants and they attributed it to equivalent N, P, and K levels in the soil for all fertilization systems. However, we interpret that non-significant effect of BL in contrast to our findings could be owing to application of BL for a short-term (i.e., one year), while longer term application (i.e., two years) remained biologically viable in triggering the growth of soybean plants which manifested as taller plants having significantly higher stem thickness and greater leaf area per plant. Our findings also exhibited BL superiority over FYS and DAP efficiency over SSP in terms of higher vegetative growth (plant height, stem girth, number of branches and leaves per plant along with leaf area) probably owing to better chemical composition of BL. However, it has been reported that based on soil tests for estimation of temporal change in nutrient dynamics, BL (over 5 Mg ha⁻¹ yr⁻¹) incorporated at the depth of 15 cm in the soil remained inadequate for boosting the concentrations of extractable nutrients except K, while increased dose $(11.2 \text{ Mg ha}^{-1} \text{ yr}^{-1})$ of BL remained instrumental in increasing P, K, Zn, and Cu concentrations which increased plant height and stem girth of crop plants [42,43]. Thus, we interpret our findings as not only BL dose but duration in years of applying BL and FYS cumulatively determine their efficacy in boosting soil fertility status and their subsequent growth promoting effect on soybean growth and development. Our findings are also consistent with those of Adeli et al. [28] who opined that significant increment in soil macro-nutrients (especially N and P), micronutrients (B, Zn, and Cu), and vegetative growth was recorded in experimental plots receiving BL (5 Mg ha⁻¹) in comparison to farm yard manure and other composted materials. However in contrast to our findings, Werner [44] and Drinkwater et al. [45] inferred that nutrient loading from cattle manure was a slow and steady process that required more time for releasing nutrients

than plant growth duration. It might be inferred that organic amendments were left on the soil surface without appropriate incorporation in the soil that could result in loss of litter released NPK through surface water and ultimately no significant effect on plant growth was observed. In our trial, subsurface banding (at the depth of 20 cm) of organic amendments (both BL and FYS) hold potential in reducing the runoff of N, P, Zn, and Cu in comparison to surface application and thus, pronounced effects of BL and FYS applied solely or in conjunction with DAP and SSP were recorded in terms of vigorous vegetative growth of temperate soybean plants [46–48].

The research findings of this field trial evidenced that the organic fraction of nutrients from BL contributed significantly to improving the reproductive attributes including number of pods per plants and 100-seeds weight. The improved reproductive growth could be related to the enhanced supply of essential nutrients which triggered plant growth and resultantly more carbohydrates could have moved toward reproductive sinks (pod, seeds, etc.,) and ultimately 100-seeds weight was increased [49,50]. The seed yield of soybean is directly linked to yield attributes as evidenced from higher seed weight and pod numbers gave greater seed yield of soybean. Previously, it has been revealed that poultry litter served as rich source of slowly releasing N and P that resulted in vigorous reproductive growth of soybean. We may further interpret that BL performed better than SSP owing to contrasting chemical composition regarding P, where most P in BL exist as NH₄-P, whereas it is found as Ca-P in SSP. Since NH₄-P tends to have higher solubility in water in comparison to Ca-P, thus BL surpassed mineral fertilizers in terms of better reproductive traits and seed yield. Furthermore, decrease in soil pH by BL could also be declared a major contributing factor for increased seed yield of soybean which resulted in significant increment of organic P forms (phospho-monoesters and phospho-diesters) [51–53]. These organic P forms are released through hydrolysis of phosphatases produced by soybean roots and various microorganisms present in the rhizosphere of soybean [54]. Moreover, non-phytoavailable form of P might also be mobilized by anions (citrate, oxalate, and malate) released from FYS and Bl after decomposition which increased seed weight and ultimately higher seed yield was recorded for organic manures compared to sole mineral fertilizers. These findings are in concurrence with those of Carvalhais et al. [46] who noted significant P mobilization under low P soil conditions by organic anions and carbohydrates released from the organic manures that resulted in higher 100-seeds weight per plant. Similar underlying process has also been described for soybean crop by Wang et al. [49]. It also needs to be highlighted that slow release of nutrients from organic manures (broiler litter and farm yard slurry) in our trial might have reduced losses of nutrients especially P by adsorption on soil surface which could have increased its provision to soybean plants at all critical growth stages [55,56]. This slow and steady nutrient releasing property of BL might serve as potent strategy for improving P uptake and P-use efficiency in weathered soils similar to our study site, having high adsorption capacity and low recovery of P from conventional water-soluble mineral fertilizers [57,58]. However, in contrast to our findings, Deeks et al. [21], and Morais and Gatiboni [22] observed no pronounced difference in P availability to soybean plants by organic manures and conventional mineral fertilizers, while these insignificant findings could be due to inappropriate doses and time of application which must be adjusted keeping in view time needed (generally 3–4 weeks) for decomposition and subsequent release of absorbable nutrients. In accordance with our findings whereby BL applied in conjunction with DAP recorded the highest yield of soybean, Ronner et al. [48] also reported that chicken manure remained instrumental in boosting the nodulation capacity, roots growth, and the capacity of the nutrient sink which resulted in higher grain yield. The current results are also in agreement with those of [59–62] who opined that sufficient rainfall, co-application of organic manures, and reduced doses of chemical fertilizers significantly enhanced yield attributes, grain yield, and biological yield of crops, however these findings are limited in scope owing to varying nutrient release dynamics under different soil and climatic conditions. Additionally, it was inferred that optimum moisture and temperature might have contributed to speed-up the rate of decomposition of organic manures and

subsequent release of all essential nutrients over a longer period of time compared to sole DAP and SSP. It was also reported that being a leguminous crop, soybean could get requisite N through biological fixation process [63], however available P might not be quite enough and this is where BL might have fulfilled the deficiency in a better way than FYS in our trial. As per meteorological data of the experimental site presented in Figure 2, organic manures especially BL might have easily decomposed owing to sufficient soil moisture by virtue of good rains which boosted yield attributes, grain weight, grain yield, and biological yield of soybean. In addition, other soil factors including C:N ratio could contribute to enhancing the microbial populations that assist in decomposing the organic manures more rapidly [20,62]. In contrast to our findings, farm yard manure (FYM) (3 t ha^{-1}) applied to finger millet plantation following soybean crop produced comparable yield to those given by farmers practice (solo use of chemical fertilizers) under moisture-deficient conditions. Thus, it was suggested that FYM could replace chemical fertilizers and hence might be recommended to economically marginal farmers having access to sufficient FYM [43]. Our findings pertaining to superior performance of BL over FYS and sole use of chemical fertilizers are also in agreement with those of Wang et al. [49] who confirmed that poultry manure significantly reduced evaporation rate from soil which led to increment in soil moisture-retaining capacity especially during the period of the peak growth. Corroborating to our results, other findings indicated that organic manure remained effective in supplying nutrients to crop plants throughout the growth season of the crop, besides improving soil water holding capacity and, hence, increased grain yield and biological productivity of leguminous crops [20,62–67].

Regarding seed yield and harvest index findings, it might be interpreted that presence of numerous micronutrients especially Ca, Mg, S, etc., along with macronutrients in BL could be attributed to higher grain yield of soybean and HI [48,51]. These findings are in concurrence with those of Devi et al. [52] who opined that organic manures remained effective in improving the yield attributes which led to higher seed yield by 32% and resultantly harvest index was increased by 11%. In contrast to synthetic fertilizers, sulfur present in organic wastes, especially broiler litter, has been attributed to promote SOC of soybean [4,8]. In our study, SOC was improved owing to synergistic fertilization regimes involving DAP and BL that tend to supply numerous nutrients slowly over a longer period of time which improved seed weight and oil content compared to farm yard slurry or solo application of chemical fertilizers. However, our findings are in contradiction with those of Elicin et al. [50] who observed no significant difference in SOC of soybean under organic fertilization management systems, but it might be attributed to lower doses of organic manures, application of partially decomposed manures, inappropriate time of application, non-responsive soybean cultivars, sub-optimal agronomic management practices etc., that hampered the influence of organic manures on SOC of soybean. Previously, Bungau et al. [68] inferred that seed yield was impacted by a variety of factors including fertilization management, field management, irrigation practices, and more importantly climatic factors especially solar radiation which either promoted or restricted the microbial activity in the soil along with enzymatic action (caused by accumulated enzymes that were secreted by proliferating microorganisms). Likewise, Samuel et al. [69] reported that it was enzymatic activity that determines the quality of soil and might be used as a reliable indicator to gauge the soil fertility and productivity status, while fertilization systems involving organic manures boost microbial activity which holds potential to enhance enzymatic activity and ultimately economic yield might be increased in a biologically viable way.

The correlation results of this field study are in liaison with those of Iqbal et al.'s [6] who opined that plant height had linear association with seed yield for erect type cultivars of soybean, while antagonistic relationship might be observed for spreading type cultivars whereby more partitioning of assimilates toward vegetative sinks could reduce reproductive growth and ultimately lower seed weight tend to reduce seed yield of soybean. It has also been previously reported that stem girth and leaf area had stronger associations with grain yield of towering genotypes of soybean and also direct association of moderate

extent for spreading accessions as maximum leaf area recorded higher photosynthetic rate which synthesized maximum carbohydrates which boosted 100-seeds weight and seed yield [4,8,10]. In accordance with our findings, number of pods and 100-seeds weight were reported to have significantly stronger association with seed yield of soybean and it was opined that these were the most pertinent yield attributes among reproductive yield-contributing traits and might be used in genetic programs for developing soybean genotypes having potential to produce a greater number of productive pods per plants having the maximum number of heavier seeds. However, a moderate association was reported among pods and seed weight with seed yield of soybean [8,17,21] compared to stronger degree of association between these traits as recorded in our trial, might be owing to genetic divergence of soybean cultivars under investigation. Thus, these findings elucidate the effectiveness of broiler litter application with reduced doses of DAP for boosting growth, yield, and oil content of soybean under temperate conditions. However, research findings of this study might be limited in scope for testing organic amendments from animal origin only which necessitates further in-depth studies for evaluating the co-application of organic manures from plants and animal origins.

5. Conclusions

The findings of this field trial remained in line with the postulated hypothesis, as the organo-mineral synergistic fertilization regimes encompassing broiler litter and farm yard slurry applied solely and in conjunction with chemical fertilizers (DAP and SSP) varied significantly in terms of soybean growth, yield, and seed oil content. Among fertilization management systems, co-application of broiler litter (5 tons ha^{-1}) and DAP (60 kg ha^{-1}) remained unmatched for triggering the vegetative growth traits (plant height, stem girth, leaf are per plant, etc.,) and boosting the reproductive yield attributes (number of pods per plant, 100-seeds weight, etc.,) of soybean. The robust vegetative growth and vigorous reproductive traits maximized seed yield, biological yield, harvest index, and seed oil content. Overall, broiler litter remained superior to farm yard slurry and DAP outperformed SSP either as a sole source of plant nutrients or applied in combination with each other. Thus, these findings provide insights that broiler litter application with reduced doses of chemical fertilizers might be recommended to soybean growers having access to poultry wastes. However, there is need for conducting further in-depth studies entailing combination of organic manures from plant and animal origins, aimed at boosting nutrient availability and reducing environmental pollution caused by inappropriate disposal of wastes from dairy and poultry industries.

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