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**Full Length Article**

# Performance and Nitrogen Use of Wheat Cultivars in Response to Application of Allelopathic Crop Residues and 3, 4-dimethylpyrazole Phosphate

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## Abstract

This study investigated the impact of allelopathic crops residue amendments and a nitrification inhibitor DMPP (3, 4-dimethylpyrazole Phosphate) on germination, growth, yield and nitrogen (N) use of wheat. In the first study, wheat cultivars [Lasani 2008 (LS-08) and Faisalabad 2008 (FSD-08)] were grown in soil filled pots (18 kg soil) amended with sorghum and sunflower residues (0, 8 and 12 Mg ha$^{-1}$) in presence and absence of nitrogen fertilizer sources [urea and ammonium sulphate (N at 110 kg ha$^{-1}$)]. Plants were raised up to maturity; sorghum residue amendments improved the performance of both wheat cultivars; however sunflower residues initially inhibited wheat germination and stand establishment but at later growth stages, the inhibitory effects were diminished. Both N sources improved grain yield of wheat cultivars with greater by ammonium sulphate in LS-08. Sorghum residue at 12 Mg ha$^{-1}$ in addition with ammonium sulphate had significant influence on grain yield of *cv.* LS-08 than other treatments. In second experiment, *cv.* LS-08 was grown in soil filled pots amended with sorghum and sunflower residue (16 and 32 Mg ha$^{-1}$) in presence or absence of DMPP (0.36 µg g$^{-1}$) in greenhouse. DMPP in combination with N in the absence of crop residues improved dry matter production and reduced the C:N ratio; however any residue addition at any rate immobilized N and decreased the dry matter accumulation. In crux, residue amendments in combination with inorganic fertilizers could improve wheat grain yield, while combined use of DMMP and N can improve plant N availability and dry biomass. 

**Keywords:** Allelopathy; Cultivars; DMPP; Nitrogen; Organic residues; Wheat

## Introduction

Returning agricultural wastes/crop residues to soil is one of the best management practices to improve soil physical properties, biological activities, soil organic matter (SOM) and nutrients availability (Malhi *et al*., 2011; Abbasi and Khizar, 2012; Abalos *et al*., 2013). The SOM has a critical role in sustainability of soil fertility, carbon sequestration, nitrogen (N) cycling and soil structure formation (Herrick and Wander, 1997). Soils in the semi-arid climates are inherently deficient in SOM due to significant drought spells (Alvaro-Fuentes *et al*., 2008) and high temperatures, which decreases the chance to attain high levels of SOM to enhance crop productivity (Albaladejo *et al*., 2012). The long term deficiency of SOM has multiple adverse effects on soil fertility, structure and surrounding environment (Capriel, 2013). Therefore, these challenges necessitate the development of sustainable countermeasures for managing organic residues to improve SOM and productivity of crops.

Regarding the potential benefits of SOM, the residues of sorghum and sunflower offers some additional benefits, for example; upon their decomposition or water washing they release some alleopathic compounds inhibitory to specific surrounding plants, germinating weeds or microbes. These allelopathic compounds have been found to possess promising potential of weed control and nutrient cycling (Cheema and Ahmad, 1992; Ahmad *et al*., 1995, 2000; Cheema and Khaliq, 2000; Macias *et al*., 2002; Alsaadawi, 2013). Moreover, sorghum and sunflower residues/root exudates/water extracts have been shown to improve plant nitrogen uptake and fertilizer nitrogen use efficiency due to their significant roles in nitrification (the biological oxidation of $NH_4^+$ to $NO_3^-$ via $NO_2^-$) inhibition (Alsaadawi, 1986a, b; 1988). The suppression of soil nitrification through the release of organic compounds from plant roots is a function of plants and is termed as BNI ‘biological nitrification inhibition’ (Subbarao *et al*., 2012). These compounds from root exudates of potentially allelopathic plant species inhibit the activity of AMO (ammonia mono-oxygenase) and HAO (hydroxylamine oxidoreductase), the two bacterial enzymes responsible for catalysis of nitrification reactions in soil (Subbarao *et al*., 2008). These compounds include, isothiocyanates mostly found in crucifer tissues (Bending and Lincoln, 2000),

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brachialactone, isolated from root exudates of *B. humidicola* (Subbarao *et al*., 2009), sorgoleone released from sorghum roots and methyl 3, (4-hydroxyphenyl) propionate, MHPP from sorghum roots (Zakir *et al*., 2008). The potential of BNI-activity have also been found in sunflower due to the presence of linoleic and linolenic acids (the potential compounds to block AMO and HAO) in its essential oils (Subbarao *et al*., 2008). The crops which could not release these BNI compounds from their roots e.g., crucifers (Bending and Lincoln, 2000), their residues could be incorporated into soil for nitrification inhibition to improve crops N use (Subbarao *et al*., 2012).

Synthetic nitrification inhibitors could be a useful agricultural practice to investigate the comparative nitrification inhibition potential of allelopathic crop residue amendments in soil. Several synthetic compounds like dicyandiamide (DCD), nitrapyrin and 3, 4-dimethyl pyrazole phosphate (DMPP) have shown promising nitrification inhibition results in laboratory and agronomic field studies (Calderon *et al*., 2005; Di and Cameron, 2008). Kleineidam *et al*. (2011) reported that application of DMPP can reduce N losses to environment and improve N supply to cultivated crops.

Although several studies has been conducted to evaluate the impact of various nitrogen sources and residue amendments on wheat yield but data on combined influence of various rates of allelopathic residues and various inorganic fertilizer sources is missing. Similarly, role of DMMP accomplished with inorganic nitrogen and various allelopathic residues in improving the N use and dry matter accumulation has rarely been explored. This study was, therefore, conducted to evaluate the impact of residue amendments (stimulating or adverse) on germination, growth and yield of two wheat cultivars applied at varying rate. Evaluating the role of combined use of residues and N sources in improving efficiency of N fertilizers for improving wheat yield, monitoring the combined influence of DMPP, residue sources and N to improve wheat performance and N utilization efficiency was also an important aspect of this study.

## Materials and Methods

### Experiment 1

**Experimental setup and growth conditions:** This experiment was conducted at Field Research Laboratory at Agronomic Research Farm, University of Agriculture Faisalabad (31°26´ N, 73°06´ E, at 184 m above sea level), Pakistan during 2009-2010. Experimental soil was collected from the plough layer of the experimental area. It was sandy clay loam in nature and belongs to Lyallpur soil series (Aridisol-fine-silty, mixed, hyperthermic Ustalfic, Haplargid in USDA classification and Haplic Yermosols in FAO classification scheme. The detailed soil properties are given in Table 1. For pot (45 × 30 cm) filling, before use, the soil was air-dried, thoroughly mixed, and sieved (5 mm) to remove stones and organic stubbles. Organic residues were produced from the sorghum (*Sorghum bicolor*) and sunflower (*Helianthus annuus*). At maturity, both crops were harvested and their stalks were shade dried for one growing season and then chopped into 1.25 cm pieces. Chemical composition and moisture content of the residues was determined before use (Table 2).

Experiment was conducted in completely randomized design in factorial arrangement. The two factors included crop residue sources, their application rates and N fertilizer sources. Pots were filled with 18 kg soil, soil residues mixture according to following treatments: (1) no residue + no fertilizer, (2) no residue + urea, (3) no residue + ammonium sulphate, (4) sorghum residue 8 Mg ha$^{-1}$ + no fertilizer, (5) sorghum residue 8 Mg ha$^{-1}$ + Urea, (6) sorghum residue 8 Mg ha$^{-1}$ + ammonium sulphate, (7) sorghum residue 12 Mg ha$^{-1}$ + no fertilizer, (8) sorghum residue 12 Mg ha$^{-1}$ + urea, (9) sorghum residue 12 Mg ha$^{-1}$ + ammonium sulphate, (10) sunflower residue 8 Mg ha$^{-1}$ + no fertilizer, (11) sunflower residue 8 Mg ha$^{-1}$ + urea, (12) sunflower residue 8 Mg ha$^{-1}$ + ammonium sulphate, (13) sunflower residue 12 Mg ha$^{-1}$ + no fertilizer, (14) sunflower residue 12 Mg ha$^{-1}$ + urea and (15) sunflower residue 12 Mg ha$^{-1}$ + ammonium sulphate. In all combinations urea and ammonium sulphate were applied on the basis of N at 110 kg ha$^{-1}$. Urea in granular and ammonium sulphate in powdered form was thoroughly mixed in soil along with residues at the time of pot filling. Soil of each pot, except no fertilizer treatments was fertilized with 100 and 50 kg ha$^{-1}$ of phosphorous as $Ca(H_2PO_4)_2.2H_2O$ and potassium sulphate as $K_2SO_4$, respectively. Twenty seeds of ~98% germination were sown in each pot during first week of November 2009 and then pots were supplied with water to bring the required WHC (above 60%). Subsequent watering was done when needed. Plants were thinned 35 days after sowing and five plants were maintained per pot until maturity.

**Measurements and calculations:** Daily observation of seed emergence was made to measure following indices of stand establishment according to standard procedures: (i) final emergence percentage (FEP) according to Handbook of Association of Official Seed Analysts (1990), (ii) energy of emergence (EE) according to Farooq *et al*. (2008), (iii) mean emergence time (MET) (Ellis and Robert, 1981) and (iv) time taken to 50% emergence ($E_{50}$) according to the formulae of Coolbear *et al*. (1984), modified by Farooq *et al*. (2005). Plant height, spike length, spikelet per spike, grains per spike, 100-grain weight, total biomass and grain yield per pot was recorded at harvest. Harvest index was calculated as ratio of grain yield to total biomass yield. Straw and grains were then oven dried and analysed for N concentration according to micro-Kjeldhal method (Bremner and Mulvaney, 1982).

Nitrogen use efficiency indices were calculated on the basis of above ground biomass as dry matter produced per unit of N applied or total N uptake in above ground biomass by following equations according to the terminology of N efficiency parameters by Delogu *et al.* (1998) and Lopez-Bellido and Lopez-Bellido (2001).

Nitrogen use efficiency (NUE, kg $kg^{-1}$) = Gy/N supply (eq. 1)
Nitrogen harvest index (NHI, %) = (Ng/Nt) ×100 (eq. 2)
Nitrogen utilization efficiency (NUtE, kg $kg^{-1}$) = Gy/Nt (eq. 3)

Where Gy is the grain yield, Ng is total grain N uptake determined by multiplying dry weight of grain by its N concentration. Nt is total plant N uptake, which was determined by dry weight of plant parts by N concentration and summing over parts for total plant uptake.

### Experiment 2

**Experimental setup and growth conditions:** This pot study was conducted in the wire house of University of Agriculture, Faisalabad (31°26´ N, 73°06´ E, at 184 m above sea level), Pakistan. Soil used in greenhouse experiment was a medium sandy loam by texture with pH (0.01 M $CaCl_2$) of 6.4, total carbon 169 mg $kg^{-1}$ and total nitrogen 31 mg $kg^{-1}$. Organic residues of sorghum and sunflower were chemically composed of (carbon 45.49%, nitrogen 0.79% with C:N 58.58) and (carbon 40.41%, nitrogen 0.90% with C:N 44.68%), respectively. In this completely randomized two factor experiment, plastic pots (8 × 6.5 cm) were filled with 250 g of soil or soil + residue (sorghum and sunflower each in separate set of experiment) according to following treatments. (1) no N + no residue, (2) no N + residue 16 Mg $ha^{-1}$, (3) no N + residue 32 Mg $ha^{-1}$, (4) N + no residue, (5) N + residue 16 Mg $ha^{-1}$, (6) N + residue 32 Mg $ha^{-1}$, (7) N + DMPP + no residue, (8) N + DMPP + residue 16 Mg $ha^{-1}$, and (9) N + DMPP + residue 32 Mg $ha^{-1}$. Nitrogen as ammonium sulphate was applied at 171.44 mg $kg^{-1}$ of dry soil in respective treatments. DMPP was applied at recommended rate of 0.36 µg $g^{-1}$ of dry soil. Both N, DMPP in solution and residues in powdered form were thoroughly mixed before pot filling. Ten seeds of wheat *cv.* LS-08 with 98% germination were sown in each pot and were thinned out to five per pot after complete emergence. Experiment was extended up to 45 days after sowing.

**Measurements and calculations:** Relative leaf chlorophyll contents were measured were measured with the SPAD-502 device (Minolta, USA) on the first fully developed leaf. Plant height was measured and number of leaves was counted at the time of harvesting. Plant's fresh and oven dry biomass was weighed. Plants dry matter was milled using a Retsch mill type SM300 (Hahn, Germany) with a 0.5 mm sieve. An aliquot of the plant material (~200 mg) was combusted in a CN analyzer (Vario MAX, Elementar Analysensysteme Gmbh, Hanau, Germany) for the determination of N concentration. The total amount of N removed from the pot by harvesting the aboveground biomass was calculated by multiplying N concentration in the aboveground biomass with the total amount of dry biomass (leaves + stems) per pot. Soil mineral nitrogen ($NO_3^-$ and $NH_4^+$) was quantified using the methods of Keeney and Nelson (1982). Twenty grams of soil was weighed in 80 mL 2 M KCl, shaken for one hour at 100 rpm and filtered (Round filter ø 70 mm S&S type 595). Concentration of $NH_4^+$-N and $NO_3^-$-N was determined colorimetrically using an auto-analyzer (Seal, Germany).

### Statistical analysis

The data were analysed statistically by using fisher analysis of variance technique and the difference between treatments was calculated by using least significant difference (LSD) test at $p \leq 0.05$ level using statistical software Sigma Plot 11.0 (Systat, Inc., Richmond, USA).

## Results

### Experiment 1

**Stand establishment, yield and N use of LS-08:** The FEP, EE, MET and $E_{50}$ of LS-08 was significantly affected by residue amendments (Table 3). Sunflower residues at either rate (8 and 12 Mg $ha^{-1}$) significantly ($p \leq 0.05$) decreased EE (44.7 to 66.1%) over control; whereas sorghum residues showed no significant adverse effect on FEP, EE, MET and $E_{50}$ compared to control (Table 3). Sunflower residues delayed emergence with increasing rate of application as indicated by increased MET and $E_{50}$ (Table 3); however the FEP was not decreased significantly over control, except at higher (12 Mg $ha^{-1}$) rate of application (Table 3).

Nitrogen and residue sources in tested combinations significantly influenced grains per spike, 100-grains weight, biological and grain yield over control (Table 4). Sorghum residue at 12 Mg $ha^{-1}$ with either N source generally or in combination with ammonium sulphate increased the yield components leading to increased grain yield compared to control (Table 4). Sunflower residues also enhanced 100-grains weight and grain yield at either rate of application in combination with N sources (Table 4); however, sole application of sunflower had no stimulating effect on yield compared to control treatment. Nitrogen sources significantly improved yield components and grain yield, with leading effect by ammonium sulphate (Table 4).

Nitrogen sources significantly stimulated plant N uptake as indicated by response of grain protein contents, NUE, NUtE and NHI (Table 5). Nitrogen and residue sources at any rate significantly influenced grain protein contents and NUtE (Fig. 1a, d; Table 5). Urea in absence of residues showed maximum NUtE, while sorghum residue at higher and sunflower residue at lower rate of application showed maximum increase in NUtE (Fig. 1d; Table 3). The NUE or NHI was not improved significantly by using residues in combination with N sources (Fig. 1b c; Table 5).

**Stand establishment, yield and N use of cv. FSD-08:** The stand establishment of *cv.* FSD-08 was also influenced significantly by residue amendments or by their combined use (Table 3). Sunflower residue at higher rate of application

**Table 1:** Soil physico-chemical properties and chemical composition of crop residues

| Soil physico-chemical properties | | | Chemical composition of crop residues | | |
|---|---|---|---|---|---|
| Characteristics | Units | Values | chemical composition | Sorghum | Sunflower |
| Sand | % | 48.31 | Carbon, % | 41,85 | 38,80 |
| Silt | % | 27.87 | Nitrogen, % | 0,65 | 0,52 |
| Clay | % | 22.29 | C:N | 64.4 | 74.6 |
| pH | | 7.98 | Sulphur, mg $g^{-1}$ | 0,346 | 0,423 |
| EC | dS $m^{-1}$ | 1.05 | Zinc, mg $kg^{-1}$ | 28.95 | 31.55 |
| Organic matter | % | 0.68 | Copper, Cu mg $kg^{-1}$ | 2.95 | 7.6 |
| Total nitrogen | % | 0.062 | Iron, Fe mg $kg^{-1}$ | 783.85 | 229.35 |
| [1]CAL-P | mg $kg^{-1}$ | 9.91 | Manganese, Mn mg $kg^{-1}$ | 29.6 | 13.1 |
| [1]CAL-K | mg $kg^{-1}$ | 187.97 | Phosphorous, P mg $kg^{-1}$ | 1960.98 | 771.17 |

[1]Available P and K in soil samples extracted by adopting CAL method (Schueller, 1969)

**Table 2:** Weather data during the experimental period

| Months | Temperature (°C) | | Relative Humidity (%) | Rainfall (mm) |
|---|---|---|---|---|
| | Maximum | Minimum | | |
| November | 25.7 | 18.2 | 64.7 | 0.7 |
| December | 22.1 | 14.5 | 64.4 | 0 |
| January | 16.2 | 11.1 | 82.3 | 0.8 |
| February | 22.0 | 15.7 | 62.7 | 11.9 |
| March | 30.4 | 23.5 | 57.5 | 8.8 |
| April | 38.4 | 29.9 | 36.8 | 1.3 |

**Table 3:** Influence of allelopathic mulches and nitrogen sources on stand establishment of wheat

| TREATMENTS | LS-08 | | | | FSD-08 | | | |
|---|---|---|---|---|---|---|---|---|
| Crop residues (R) | FEP (%) | MET (days) | $E_{50}$ (days) | EE (%) | FEP (%) | MET (days) | $E_{50}$ (days) | EE (%) |
| $R_1$ No residue | 95.54 a | 10.82 bc | 7.50 c | 76.30 a | 94.88 a | 10.30 b | 7.58 | 66.00 a |
| $R_2$ Sorghum 8 Mg $ha^{-1}$ | 94.06 a | 10.17 c | 7.89 abc | 62.22 a | 87.88 b | 10.90 b | 7.74 | 51.00 b |
| $R_3$ Sorghum 12 Mg $ha^{-1}$ | 90.37 a | 10.31 c | 7.63 bc | 63.71 a | 94.66 a | 10.63 b | 7.86 | 47.22 bc |
| $R_4$ Sunflower 8 Mg $ha^{-1}$ | 88.90 a | 12.91 a | 8.07 ab | 42.23 b | 82.88 b | 11.12 b | 7.58 | 38.33 cd |
| $R_5$ Sunflower 12 Mg $ha^{-1}$ | 80.00 b | 12.28 ab | 8.19 a | 25.93 b | 74.88 c | 12.72 a | 7.90 | 33.33 d |
| LSD at $p \leq 0.05$ | 7.405 | 1.736 | 0.481 | 17.120 | 5.194 | 0.990 | ns | 8.982 |
| Nitrogen sources | | | | | | | | |
| C Control | 92.87 a | 11.66 | 7.88 | 54.23 | 84.20 b | 10.76 | 7.71 | 47.33 |
| U Urea | 85.78 b | 11.45 | 7.73 | 57.33 | 89.26 a | 11.29 | 7.68 | 49.40 |
| AS Ammonium sulphate | 90.67 ab | 10.79 | 7.96 | 50.68 | 87.66 ab | 11.34 | 7.80 | 44.80 |
| LSD at $p \leq 0.05$ | 5.736 | ns | ns | ns | 4.024 | ns | ns | ns |

Mean sharing the same letters within a column for a factor do not differ significantly at $P < 0.05$
FEP = Final emergence percentage, MET = Mean emergence time, $E_{50}$ = Time to 50% emergence, EE = Energy of emergence

had maximum adverse effect on FEP, MET and EE; however sorghum residue at higher (12 Mg $ha^{-1}$) rate of application significantly affected only EE (Table 3). Moreover, sorghum residue amendments improved yield components but not grain yield over control; however, lower application rate had more pronounced effect to stimulate number of grains per spike and biological yield (Table 4). Both nitrogen sources improved grain yield than control. Combined use of residues and N sources had no significant influence on grain yield; though different combinations increased grain yield over control (Table 4). Residue amendments at either rate of application in combination with ammonium sulphate improved N uptake resulting in higher grain protein contents over control (Fig. 2a; Table 5). Both nitrogen sources improved NUE over control but no significant effect was found due to combined use of N and residue sources (Fig. 2b; Table 5). Sunflower application at 12 Mg $ha^{-1}$ had shown highest NHI (Fig. 2c; Table 5), while NUtE significantly improved with control and at higher rate of sunflower residue application (Fig. 2d; Table 5).

**Experiment 2**

**Sorghum residue impact on plant soil N dynamics and biomass yield:** Nitrogen fertilization in combination with DMPP significantly increased plant N uptake as indicated by higher leaf chlorophyll contents, plant height, dry biomass and lowest values of C:N ratios and mineral $NH_4^+$-N (Table 6). The significant effect was further followed by sole N application and combined application of N + DMPP + sorghum 16 Mg $ha^{-1}$ (Table 6). Sorghum residue amendments without N or at higher rate 32 Mg $ha^{-1}$ with N did not improved plant N uptake or dry biomass yield than control (Table 6), which is indicated by the presence of significant amount of $NH_4^+$-N in soil at harvest (Table 6).

**Table 4:** Impact of various nitrogen sources and crops residues applied at various rates on yield-related traits, biological and grain yields, and harvest index of wheat

| Treatments | LS-08 | | | | | FSD-08 | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Residues (R) | Grains per spike | 100-GW (g) | BY per pot (g) | GY per pot (g) | HI (%) | Grains per spike | 100-GW (g) | BY per pot (g) | GY per pot (g) | HI (%) |
| $R_1$ No residue | 33.58 b | 3.25 b | 57.07 b | 22.74 | 39.19 b | 38.71 b | 3.30 c | 58.32 b | 23.76 | 39.77 |
| $R_2$ Sorghum 8 Mg $ha^{-1}$ | 35.72 a | 3.32 ab | 64.74 a | 24.20 | 37.46 b | 46.14 a | 3.66 b | 65.57 a | 25.47 | 38.74 |
| $R_3$ Sorghum 12 Mg $ha^{-1}$ | 35.43 a | 3.32 ab | 58.49 b | 24.43 | 41.80 ab | 44.83 a | 3.91 a | 60.54 b | 23.88 | 40.30 |
| $R_4$ Sunflower 8 Mg $ha^{-1}$ | 35.81 a | 3.43 a | 51.48 c | 23.36 | 45.40 a | 43.66 a | 3.71 b | 50.88 c | 22.64 | 44.34 |
| $R_5$ Sunflower 12 Mg $ha^{-1}$ | 36.46 a | 3.16 b | 51.66 c | 22.96 | 45.09a | 43.22 a | 3.33 c | 49.28 c | 21.38 | 43.83 |
| LSD at $p≤0.05$ | 1.62 | 0.74 | 3.60 | ns | 4.96 | 3.47 | 0.17 | 3.58 | ns | ns |
| Nitrogen sources | | | | | | | | | | |
| C Control | 35.23 b | 3.21 b | 50.59 c | 20.93 c | 41.25 | 41.66 b | 3.58 | 45.70 b | 18.52 b | 40.46 |
| U Urea | 36.95 a | 3.28 ab | 55.54 b | 23.80 b | 43.37 | 43.1 ab | 3.57 | 62.14 a | 26.9 a | 43.78 |
| AS Ammonium sulphate | 34.01 b | 3.40 a | 63.93 a | 25.88 a | 40.73 | 45.21 a | 3.59 | 62.91 a | 24.78 a | 39.95 |
| LSD at $p≤0.05$ | 1.30 | 0.35 | 2.79 | 1.89 | ns | 2.69 | ns | 2.77 | 3.38 | ns |
| Interactions | | | | | | | | | | |
| $R_1 \times C$ | 31.53 g | 2.91 e | 45.17 f | 17.10 f | 43.90 | 33.83 | 3.04 h | 39.23 h | 12.84 | 32.66 |
| $R_1 \times U$ | 34.30 d-g | 3.41 abc | 54.53 de | 23.93 a-e | 38.04 | 41.38 | 3.37 fg | 64.10c | 31.48 | 49.23 |
| $R_1 \times AS$ | 34.90 c-f | 3.43 abc | 71.50 a | 27.20 ab | 37.99 | 40.88 | 3.50 d-g | 71.63ab | 26.92 | 37.46 |
| $R_2 \times C$ | 38.30 ab | 3.42 abc | 60.60 cd | 23.03 b-e | 37.78 | 43.88 | 4.04 b | 56.33 ef | 22.17 | 39.13 |
| $R_2 \times U$ | 36.30bcd | 3.31 bc | 67.63 ab | 25.50 abc | 36.59 | 45.63 | 3.49 d-g | 66.00 bc | 27.29 | 41.56 |
| $R_2 \times AS$ | 32.57 fg | 3.22 ab | 66.00 ab | 24.07 a-e | 41.35 | 48.88 | 3.44 efg | 74.37a | 26.88 | 35.53 |
| $R_3 \times C$ | 35.50 b-e | 3.16 cde | 56.73 de | 23.40 a-e | 40.71 | 43.10 | 4.41 a | 54.47f | 22.93 | 42.32 |
| $R_3 \times U$ | 37.53 bc | 3.28 bcd | 55.57 de | 22.50 cde | 43.35 | 47.50 | 3.74 d | 61.97cde | 26.02 | 43.60 |
| $R_3 \times AS$ | 33.57 efg | 3.53 ab | 63.17 bc | 27.40 a | 45.22 | 43.88 | 3.58 def | 65.20c | 22.67 | 34.98 |
| $R_4 \times C$ | 36.40bcd | 3.22 cd | 46.07 f | 20.83 def | 46.68 | 44.13 | 3.37 fg | 40.00h | 17.37 | 43.51 |
| $R_4 \times U$ | 36.13bcd | 3.43 abc | 53.33 e | 24.77 a-d | 44.34 | 39.25 | 4.03 bc | 55.97 ef | 23.46 | 41.93 |
| $R_4 \times AS$ | 34.90 c-f | 3.64 a | 55.03 de | 24.47 a-e | 46.07 | 47.50 | 3.75 cd | 56.67def | 27.03 | 47.57 |
| $R_5 \times C$ | 34.40 def | 3.34 abc | 44.40 f | 20.30 ef | 47.86 | 43.33 | 3.07 h | 38.47 h | 17.17 | 44.65 |
| $R_5 \times U$ | 40.50 a | 2.99 de | 46.63 f | 22.30 cde | 41.38 | 41.53 | 3.24 gh | 62.67cd | 26.57 | 42.56 |
| $R_5 \times AS$ | 34.40 def | 3.16 cde | 63.93 bc | 26.27 abc | 43.90 | 44.75 | 3.69 de | 46.70g | 20.37 | 44.25 |
| LSD at $p≤0.05$ | 2.84 | 0.30 | 6.23 | 4.24 | ns | ns | 0.29 | 6.20 | ns | ns |

Mean values within the columns sharing the same letter are not significantly different at $p≤0.05$

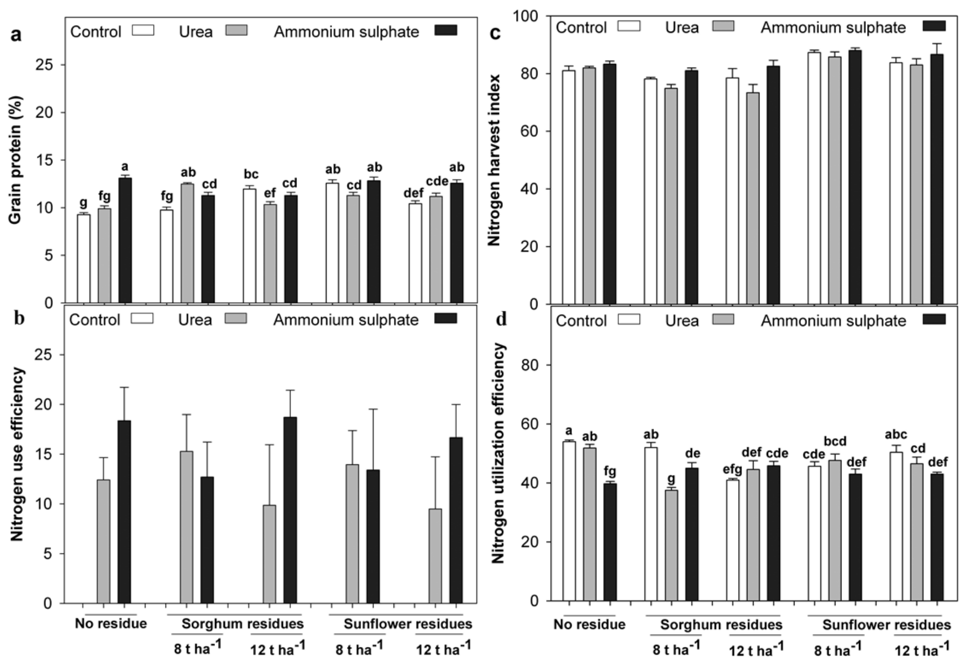


**Fig. 1:** (a) Grain protein concentration, (b) nitrogen use efficiency, (c) nitrogen harvest index and (d) nitrogen utilization efficiency of *cv.* LS-08 as influenced by different rates of sorghum and sunflower residues alone and in combination with urea and ammonium sulphate as nitrogen sources. Bars with similar letters or having no letters are not significantly different at p<0.05

**Table 5:** Analysis of variance for grain protein contents, nitrogen use efficiency (NUE), nitrogen harvest index (NHI) and nitrogen utilization efficiency (NUtE) of two wheat cultivars

| Source of variation | LS-08 | | | | FSD-08 | | | |
|---|---|---|---|---|---|---|---|---|
| | Mean sum of square | | | | | | | |
| | Grain protein | NUE | NHI | NUtE | Grain protein | NUE | NHI | NUtE |
| Crop residues (R) | 2.63** | 2.94 ns | 139.77** | 13.36 ns | 1.33** | 69.53 ns | 35.08 ns | 38.69* |
| Nitrogen sources (N) | 8.58** | 1044.0** | 75.92** | 9915.55** | 0.15 ns | 2867.33** | 70.37* | 11.31 ns |
| R × N | 4.33** | 19.68 ns | 8.54 ns | 42.99** | 1.06** | 54.20 ns | 50.25* | 46.35** |

* = Significant at $p \leq 0.05$; ** = Significant at $p \leq 0.01$; ns = non-significant

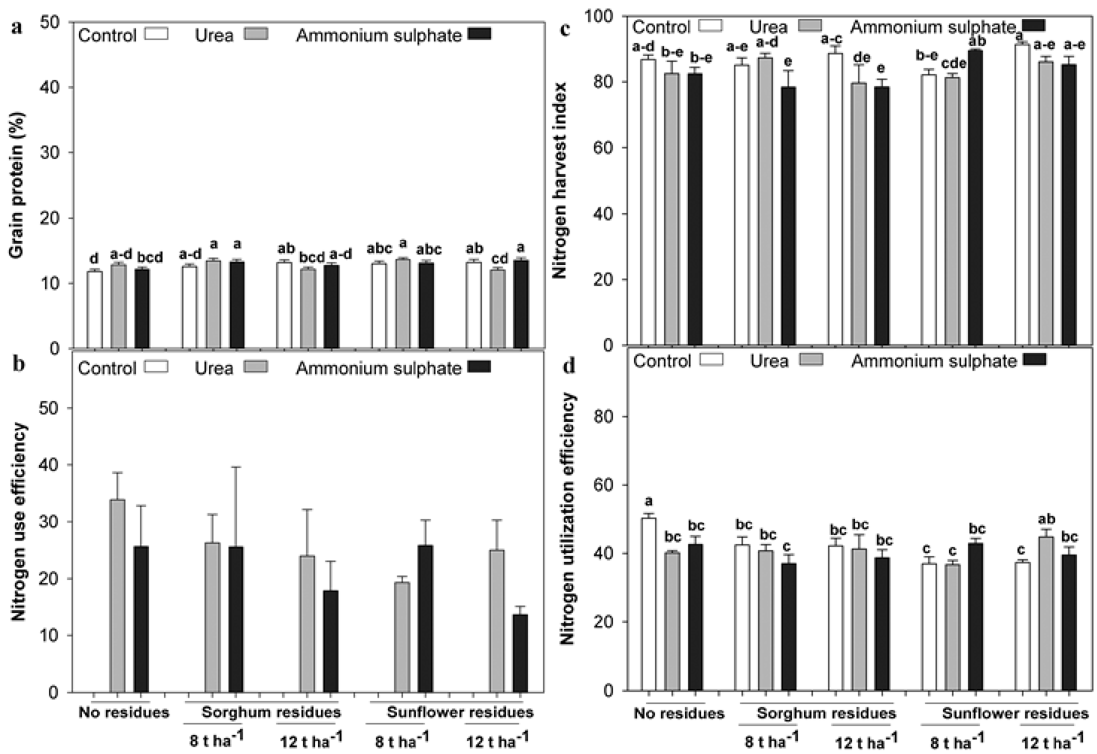


**Fig. 2:** (a) Grain protein concentration (%), (b) nitrogen use efficiency, (c) nitrogen harvest index and (d) nitrogen utilization efficiency of *cv.* FSD-08 as influenced by different rates of sorghum and sunflower residues alone and in combination with urea and ammonium sulphate as nitrogen sources. Bars with similar letters or having no letters are not significantly different at $p \leq 0.05$

**Sunflower residue impact on plant soil N dynamics and biomass yield:** Sunflower residue amendment in combination with N or N+DMPP significantly improved leaf chlorophyll content, plant height and dry biomass over control (Table 7). Combine application of N+DMPP enhanced N uptake which resulted into higher leaf chlorophyll content, plant height and ultimately greater dry biomass yield (Table 7). Sunflower residues at 32 Mg ha$^{-1}$ without N showed maximum leftover of $NH_4^+$-N in the soil analysed just after harvest (Table 7).

## Discussion

Sorghum and sunflower residue amendments significantly affected the stand establishment, grain yield and nitrogen use of the crop. However, sorghum residue amendments led to more pronounced effects on the improvement of plants growth, yield and N use than that of sunflower residues and the positive influence of sorghum residue was linearly increasing in terms of morphological traits in both tested cultivars. This improvement in morphological and yield related traits may be attributed to the allelopathic activity of these residues, which may have stimulated the production and release of phenolic acids and sorgoleone compounds upon their decomposition (Einhellig *et al.*, 1993; Ben-Hammouda *et al.*, 1995). In many earlier studies, the allelopathic activity of sunflower and sorghum residue/mulch/water extracts is well documented (Macias *et al.*, 1999; Cheema and Khaliq, 2000; Gawronski *et al.*, 2002, 2003; Alsaadawi, 2007), which may be a possible reason for the crop promotion or inhibition. We observed that sunflower residues inhibited the germination of wheat.

**Table 6:** Impact of combine application of sorghum mulch and nitrogen applied alone or in combination with DMPP on chlorophyll concentration, plant height, dry biomass, C:N ratio and $NH_4^+$-N

| Factors | Chl. Concentration (SPAD values) | Plant height (cm) | Dry biomass (g) | C:N | $NH_4^+$-N (mg kg$^{-1}$) |
|---|---|---|---|---|---|
| Nitrogen treatments | | | | | |
| Control (C) | 23.05 c | 24.54 c | 0.40 c | 55.73 a | 3.60 b |
| Nitrogen (N) | 30.62 b | 33.72 b | 0.93 b | 42.25 b | 4.46 a |
| N+DMPP | 33.35 a | 37.17 a | 1.03 a | 38.06 b | 3.70 b |
| LSD at $p$≤0.05 | 1.583 | 1.633 | 0.080 | 5.528 | 0.433 |
| Crop residue rates | | | | | |
| Control (C) | 32.80 a | 35.19 a | 1.08 a | 37.12 b | 2.83 c |
| SR 16 | 28.92 b | 27.20 c | 0.55 c | 48.37 a | 4.76 a |
| SR 32 | 25.30 c | 33.03 b | 0.73 b | 50.56 a | 4.17 b |
| LSD at $p$≤0.05 | 1.583 | 1.633 | 0.080 | 5.528 | 0.433 |
| Interaction | | | | | |
| C × C | 22.31 d | 23.58 e | 0.39 d | 50.95 | 2.83 c |
| C × SR 16 | 23.62 d | 21.93 e | 0.36 d | 59.41 | 4.24 b |
| C × SR 32 | 23.24 d | 28.11 e | 0.45 d | 56.83 | 3.72 b |
| N × C | 36.85 a | 40.24 ab | 1.39 a | 31.23 | 2.90 c |
| N × SR 16 | 30.25 bc | 22.97 f | 0.37 d | 45.05 | 3.88 b |
| N × SR 32 | 24.76 d | 37.94 bc | 1.04 b | 50.46 | 6.59 a |
| N+DMPP × C | 39.26 a | 41.77 a | 1.47 a | 29.17 | 2.76 c |
| N+DMPP × SR 16 | 32.90 b | 36.71 c | 0.91 b | 40.64 | 4.38 b |
| N+DMPP × SR 32 | 27.91 c | 33.04 d | 0.70 c | 44.39 | 3.96 b |
| LSD at $p$≤0.05 | 2.742 | 2.828 | 0.138 | ns | 0.750 |

**Table 7:** Impact of combine application of sunflower mulch and nitrogen applied alone or in combination with DMPP on chlorophyll concentration, plant height, dry biomass, C:N ratio and $NH_4^+$-N

| Factors | Chl. Concentration (SPAD values) | Plant height (cm) | Dry biomass (g) | C:N ratio | $NH_4^+$-N (mg kg$^{-1}$) |
|---|---|---|---|---|---|
| Nitrogen treatments | | | | | |
| Control (C) | 22.08 c | 25.68 c | 0.44 c | 53.78 | 3.60 b |
| Nitrogen (N) | 32.96 b | 34.84 b | 0.96 b | 35.32 | 4.55 a |
| N+DMPP | 35.49 a | 38.92 a | 1.15 a | 32.79 | 3.70 b |
| LSD at $p$≤0.05 | 1.569 | 1.899 | 0.088 | 3.675 | .4963 |
| Crop residue rates | | | | | |
| Control (C) | 32.80 a | 35.19 b | 1.08 a | 37.12 | 2.83 c |
| SF 16 | 29.70 b | 27.01 c | 0.55 c | 41.28 | 3.58 b |
| SF 32 | 28.04 c | 37.23 a | 0.91 b | 43.49 | 5.05 a |
| LSD at $p$≤0.05 | 1.569 | 1.899 | 0.088 | 3.675 | 0.496 |
| Interactions | | | | | |
| C × C | 22.31 d | 23.58 ef | 0.390 e | 51.0 | 2.83 d |
| C × SF 16 | 22.16 d | 20.42 f | 0.295 e | 54.1 | 3.28 cd |
| C × SF 32 | 21.77 d | 33.06 d | 0.625 d | 56.3 | 6.46 a |
| N × C | 36.85 a | 40.24 ab | 1.390 a | 31.2 | 2.90 d |
| N × SF 16 | 33.40 b | 23.75 e | 0.345 e | 35.9 | 4.12 bc |
| N × SF 32 | 28.65 c | 40.52 ab | 1.135 b | 38.8 | 4.66 b |
| N+DMPP × C | 39.26 a | 41.77 a | 1.472 a | 29.2 | 2.76 d |
| N+DMPP × SF 16 | 33.54 b | 36.87 c | 1.007 bc | 33.9 | 3.34 cd |
| N+DMPP × SF 32 | 33.69 b | 38.13 bc | 0.972 c | 35.3 | 4.04 bc |
| LSD at $p$≤0.05 | 2.718 | 3.290 | 0.152 | ns | 0.86 |

Mean values within the columns sharing the same letter are not significantly different at $p$≤0.05 (SF 16 = Sorghum residue addition at 16 Mg ha$^{-1}$. SF 32 = Sorghum residue addition at 32 Mg ha$^{-1}$)

The sensitivity of cereals (in terms of germination, growth and total biomass production) to sunflower residues has also been reported due to production and release of toxic allelochemicals upon decomposition (Cheema and Khaliq, 2000; Ashrafi *et al*., 2008). Inhibitory effect was also observed when sunflower stubbles were left in the field prior to wheat sowing (Purvis and Jones, 1990). In wheat, Ciarka *et al*. (2002b) found a substantial reduction in seedling establishment in field conditions due to sunflower residues. Kumar and Goh (2002) found improved grain yield due to residue amendments in the following order white clover > pea > ryegrass > wheat due to improved N uptake. In this study, we found that, the effect of crop residues amendment was initially inhibiting for stand establishment which later on diminished and finally resulted in improved grain yield of wheat. Alsaadawi *et al*. (2011) found similar results of stimulatory effect due to residue incorporation of different cultivars of sunflower in wheat on morphological, yield related traits and grain yield of various wheat cultivars.

Nitrogen use in wheat was also affected by the different residue amendments. Several studies have reported the presence of allelopathic effects on biological nitrogen cycle, in terms of nitrification or N fixation (Rice, 1984; Weston and Putnam, 1985; Alsaadawi *et al*., 1986a, b; Alsaadawi, 1988; Zwain *et al*., 1998). Alsaadawi *et al*. (1986a, b), while working with incubation study on residues incorporation of different sorghum genotypes, reported potential nitrification inhibition response varying with genotypes. A significant nitrification inhibition response has been reported in studies on root and shoots residues of different sunflower cultivars (Alsaadawi, 1988). Presence of linoleic and linolenic acids in essential oil of sunflower along with other crops has been found to inhibit nitrifying activity of microbes (Subbarao *et al*., 2008). The ultimate reduction in N losses increases the probability of N availability to the crops. Moreover, in present study the grain protein contents which is main quality factor of wheat quality and marketing (Jenner *et al*., 1991) was significantly improved with combined application of residue and N fertilizer sources. The availability of urea N was probably increased due to combined application with residue sources. The NUE being major challenge of crop breeding and production is usually partitioned into two important components: efficiency in uptake and efficiency in translocation to grain (Ortiz-Monasterio *et al*., 1997; Van Ginkel *et al*., 2001). We observed an improvement in the translocation efficiency of N due to combined application of N with residue sources; however uptake efficiency was not stimulated with combined application of fertilizer sources with residues. Nitrogen harvest index (NHI; the proportion of seed N to shoot N) is also considered to measure how efficiently the plants/specific cultivar utilize acquired N for grain protein production (Noulas, 2002). The NHI of wheat mostly reported is 70-80% (Calderini *et al*., 1995; Brancourt-Hulmel *et al*., 2003) or in extreme cases ranged 51-91% (Van Sanford and MacKown, 1987). Sunflower residue incorporation improved maximum NHI at both rates of application in present study. Nitrogen utilization efficiency is the ability of plants to convert absorbed N into grain yield (Ortiz-Monasterio *et al*., 1997). Sorghum residue amendments at 12 t $ha^{-1}$ and sunflower at lower rate of application improved maximum NUtE; sole urea fertilization has also improved NUtE over control indication higher N uptake and then successful conversion to grain yield. Yuan *et al*. (2010) also found higher NUtE from organic N source (swine manure 30%) as compared to chemical N source (24%) in wheat.

Residue amendments in soils lead to improvement of physical, chemical and biological properties of soil; maintain soil productivity by nutrient replenishment and reduce excessive evaporation to conserve soil water holding capacity (Hejazi *et al*., 2002). However, alleopathic crop residues on decomposition may have multiple effects on soil properties and weeds/crops (Wallace and Bellinder, 1992). We observed here differential response of sunflower and sorghum residue amendments on FSD-08 germination indices (FEP, MET and EE). Sunflower produced inhibiting influence increasing with rate of residue addition, while sorghum residue at higher rate of addition in combination with ammonium sulphate fertilizer showed highest improvement in FEP than control. Generally, sorghum residue amendments had improved agronomic parameters leading to improvement of grain yield of FSD-08, however lower (8 t $ha^{-1}$) rate of sorghum residue amendment had more pronounced effect. This phenomenon tentatively attributes to allelopathic compounds concentration released into soil at decomposition, which may produce concentration dependent positive or inhibitory influence. This phenomenon was also observed by Dietz *et al*. (2013) in their study with *Plantago lanceolata* L. leaf material or extract incubation; they found significant suppression of N mineralization and they strongly hypothesised that it happens due allelochemical effects of *P lanceolata*. Generally sole N sources produced greater grain yield compared to combined use of N and residue sources, however grain yield was still higher than control treatment but with no significant effect. Both N immobilization (conversion of inorganic N into organic N) and mineralization (conversion of organic N it no ammonium N) occur simultaneously in soil with the relative magnitudes defining whether the overall effect was net N immobilization or net N mineralization (Alexander, 1977; Cabrera *et al*., 2005). Therefore it is difficult to argue that no significant grain yield improvement in FSD-08 was due to N immobilizations when we have significant grain yield improvement in LS-08. Similarly no significant improvement in NUE and NUtE with combined use of N fertilizer and residue sources suggests further field studies keeping in view the soil organic matter development and net crop performance.

The only residue sources with < 24 C:N ratio can increase soil mineral N (Trinsoutrot *et al*., 2000). While the addition of organic residues with higher C:N ratios (25:1, 30:1 or 42:1) varying with soil conditions results in microbial N immobilization, which reduce nitrous oxide ($N_2O$) emission (Gentile *et al*., 2008). If this microbial N immobilization did not harm much to crop productivity could be a strategy to decrease environmental pollution and to improve N use efficiency. Similarly, addition of synthetic nitrification inhibitor DMPP also meant to the decrease greenhouse gases emission (Vannelli and Hooper, 1992) by inhibiting first phase of nitrification i.e. inhibiting the activity of catalytic enzyme (ammonia monooxygenase) responsible for conversion of $NH_4^+$ to hydroxylamine. Reductions in the activity of ammonia monoxygenase stabilize $NH_4^+$ and strongly inhibit $NO_3^-$ production (Chaves *et al*., 2006; Diez *et al*., 2010). In this study, we tested the DMPP in comparison with sorghum and sunflower residue amendments to estimate their ultimate effect on N availability of plants either by nitrification inhibition or by N immobilization. Nitrogen in combination with DMPP significantly enhanced N uptake which resulted into higher leaf chlorophyll content, plant height and ultimately greater dry biomass yield (Table 6, 7). However, residue addition combined with N+DMPP resulted in decreased N uptake, which was due to N immobilization and high C:N ratio (Bird *et al*., 2001) and this immobilization was increasing with increasing rate of residue amendments. Highest rate of residue amendments resulted in lowest N uptake as indicated by low chlorophyll contents and low final dry matter production. Similarly the presence of highest amount of $NH_4^+$-N in potted soil amended with higher rate of residue also indicate greater N immobilization with residue amendments and these responses can reduce crop growth and development at least for short term (Francis, 1995; Mary *et al*., 1996) as we observed in this study. Thus, the residue amendments alone or in combination with DMMP can effectively immobilize N to avoid losses and may result into beneficial effects as indicated by results of first experiment.

## Conclusion

Higher rates of sunflower residue amendment initially decreased wheat germination but finally improved grain yield while sorghum residue at either rate improved morphology and grain yield of tested wheat cultivars. Combined use of allelopathic residues with N sources did not improve NUE. Among nitrogen sources ammonium sulphate performed better than urea in terms of yield and N use. Use of DMPP in combination with allelopathic residues showed strong N immobilization. In crux, combine use of N sources with allelopathic crops residues did not improve NUE; however soil residue incorporation replenished the carbon losses which may improve soil biological and chemical properties.

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