



## Effect of liquid urea ammonium nitrate application at varying depths on root and shoot growth in wheat (*Triticum aestivum*)

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### ABSTRACT

An experiment was conducted to optimise the desired concentration of urea ammonium nitrate (UAN), a liquid fertilizer, and its suitable depth of application in wheat (*Triticum aestivum* L.) during 2016-17 in a completely randomized factorial design. The wheat seed germination was affected both on blotting paper and in soil mixed with different UAN:water dilutions. Viability test showed that when soil was mixed with UAN:water dilution ratio of less than 1:20, all the embryos were destroyed. The cultivar HD 2967 was grown in cylindrical poly vinyl chloride tubes where two different dilution ratio of UAN:water, viz. 1:5 ( $U_1$ ) and 1:10 ( $U_2$ ) were used as fertilizer @ 120 N kg/ha. The fertilizer was placed at three depths below the seed, i.e. at 1 ( $D_1$ ), 2 ( $D_2$ ) and 3 cm ( $D_3$ ). The shoot and root growth parameters up to 34 days after sowing were recorded. Both fertilizer concentration and depth of its placement significantly affected plant height, root length, root surface area and root tips at 8, 20 and 34 DAS. At higher concentration of UAN ( $U_1$ ), the root growth significantly reduced by about 43% as that of  $U_2$  after 8 DAS. However, after 34 DAS, it was reduced to 16% which shows that the plants were able to overcome the initial shock given by high concentration of UAN. Root surface area was maximum at  $U_2D_3$  (7.04, 18.26 and 40.89 cm<sup>2</sup>) and minimum at  $U_1D_1$  (0.91, 7.46 and 15.24 cm<sup>2</sup>) after 8, 20 and 34 DAS, respectively. Root tip numbers were also affected at 8 DAS. However, it was non-significantly affected by concentration and depths at 34 DAS. Deep placement of fertilizer at 3 cm showed significantly better growth of both root and shoot in plants. Higher fresh root weight at all the growth stages was observed in  $D_3$ , which was at par to  $D_2$  at 8 DAS. Similarly, highest root and shoot growth were observed at  $U_2D_2$  which was statistically at par to  $U_1D_3$ . The UAN may be applied in 1:10 dilution ratio at 2 cm depth or 1:5 dilution ratio at 3 cm depth below the seed in order to get better growth of seedlings. Hence, sowing machines for wheat shall be developed keeping in mind these application depths of liquid UAN.

**Key words:** Placement depth, Root and shoot growth, Urea ammonium nitrate, Wheat

Nitrogen management is a crucial component for sustainable crop production. Granular fertilizers are widely used in crop production. However, Urea has a major disadvantage in terms of losses of N through volatilization, if it is not incorporated into soil soon after application (Chen *et al.* 2008).

Rochette *et al.* (2009) reported that broadcast Urea lost the greatest proportion of applied nitrogen (64%) followed by banded (2 cm) urea (31%). Liquid fertilizers

have relatively more fertiliser effectiveness as compared with granular ones (Holloway *et al.* 2001). Urea ammonium nitrate (UAN) is a solution of urea [ $CO(NH_2)_2$ ] and ammonium nitrate [ $NH_4NO_3$ ] containing about 28-32% N. Nitrogen uptake in point injection method of fertilizer placement was found better than the broadcast method (Blackshaw *et al.* 2002). UAN can be injected into the soil beneath the surface, sprayed onto plant leaves as a source of foliar nutrition and can be added to irrigation water for fertigation. Liquid UAN solution is becoming popular because of its versatility, viz. convenience to mix with other nutrients and agriculture chemicals, safe to handle and can be evenly applied in field. UAN is the most efficient N source as compared with calcium ammonium nitrate (CAN) and anhydrous ammonium (AA), as it provides the greatest crop response and greatest inorganic soil availability (Gagnon and Ziadi 2010). Fluid fertilizers can be blended to precisely meet the specific needs of a soil or crop (IPNI 2017).

For the seedling establishment of wheat, a sowing depth of 2-6 cm is recommended (Hadjichristodoulou *et*

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al. 1977). It is necessary to place liquid fertilizer at such a depth which will give better seed germination without damaging the seeds as well as easy availability of nutrients to the primary and secondary roots. During designing of liquid fertilizer seed drill, it is very important to maintain a vertical gap between seed and liquid fertilizer so that toxicity have no adverse effect on seed germination, seedling growth and early plant growth. Bremner and Krogmeier (1989) reported the adverse effect of urea fertilizers on seed germination, seedling growth, and early plant growth in soil. Also, Ammonium at a high concentration inhibits primary root development (Liu *et al.* 2013). However, no such study has been conducted on the effect of UAN solution on germination and early root growth. Hence, an experiment was designed to assess the effect of UAN fertilizer on seed germination and early seedling growth of wheat (*Triticum aestivum* L.) seed.

#### MATERIALS AND METHODS

Two different experiments were conducted to evaluate the effect of UAN solution mixed with water in different ratios on the germination as well as early seedling growth of wheat seed. UAN (28%, w/w) solutions were provided by National Fertilizer Ltd, Nangal Unit, Punjab. UAN 28 is a clear transparent liquid with a specific gravity of 1.25 (Bijarniya 2016). HD 2967 wheat variety was used in these studies. The soil used in both the experiments was sandy loam having pH 7.5 obtained from the experimental farm of Division of Agricultural Engineering, IARI, New Delhi.

The seed germination percentage was determined using between the papers in which four replications of 100 seeds were placed in petriplate method (ISTA 2015). The petriplates were placed in germinator at  $20 \pm 1^\circ\text{C}$ . The number of normal, abnormal seedlings and dead seeds were calculated after 12 days. Seed germination percentage of wheat variety was calculated on the basis of normal seedling by following formula:

$$\text{Per cent germination} = \frac{\text{No. of germinated seed}}{\text{Total no. of seed}} \times 100$$

Seed was germinated with different dilution ratios of UAN by two methods:

*Germination on blotting paper:* Three layers of blotting paper were placed in a Petri-dish of 100 mm diameter. Four replications of 25 seeds were used for each lot, and placed on the paper; the Petri dish was covered with a lid and kept in the dark in an incubator maintained at  $20 \pm 1^\circ\text{C}$ . Different UAN water dilution ratios (1:0, 1:1, 1:5, 1:10, 1:15, 1:20, 1:25 and 1:30) were used for moistening of the blotting paper.

*Germination with soil:* Bremner and Krogmeier (1989) advised a method for finding out seed germination using soil. Air-dried soil (40 g) was placed in a 15 mm  $\times$  100 mm Petri dish and moistened with 10 ml of water containing the UAN solutions. Different UAN water ratio used were 1:0, 1:1, 1:5, 1:10, 1:15, 1:20, 1:25 and 1:30. Four replications of 25 seeds were used for each lot, and placed on the soil;

the Petri dish was covered with a lid and kept in the dark for 7 days in an incubator maintained at  $20 \pm 1^\circ\text{C}$ .

Tetrazolium (TZ) test is a biochemical test to rapidly estimate seed viability and alternative quick method for seed's germination potential (ISTA 2015). It is based on the change in colour of living tissues in the presence of a tetrazolium salt solution. This change in colour reflects the activity of specific enzyme systems, closely related to seed viability (Carvalho *et al.* 2013). Four replicates of 25 seeds each per lot were taken from the petri dishes after the above germination test was over. Seeds were bisected longitudinally through the midsection of the embryonic axis and then placed in beakers covered (15 ml) with a 0.075% 2, 3, 5 tri-phenyl tetrazolium chloride solution and incubated at  $40^\circ\text{C}$  for 3 h (Santos *et al.* 2007).

Wheat cultivar (HD 2967) were grown in cylindrical poly vinyl chloride (PVC) pots (150 mm high, 54 mm diameter). Each pot contained 420 g of dry soil, which had been passed through a 5 mm sieve and packed in 20 mm thick layers to achieve a uniform bulk density of 1.3 g/cc. The packing was done with a small piston of diameter 50 mm. Two different solutions of UAN:water in the ratio 1:5 ( $U_1$ ) and 1:10 ( $U_2$ ) were used as fertilizer doses (120 N kg/ha), which were placed at three different depths from seed i.e. at a depth of 1 ( $D_1$ ), 2 ( $D_2$ ) and 3 cm ( $D_3$ ). The ratios of 1:5 and 1:10 were chosen, as it would be easy for a farmer to mix water with UAN in the fertilizer tank of seed drill during sowing operation. Two seeds were planted at a depth of 3 cm. The initial moisture content of the soil was  $11 \pm 0.55\%$  (db).

The length of the shoot was measured daily from the time of emergence. Plants were used for destructive measurements from each replication on days 8, 20 and 34 after emergence. The shoots were cut from the roots after removing and washing the loose soil. The fresh weights of the two fractions, roots and shoots were immediately measured. The dry weight of roots was also measured after oven-drying for 48 hr at  $40^\circ\text{C}$ . The root length, root surface area, no. of root tips of each plant were measured with an image-analysis system (Win-RHIZO); at Water Technology Centre, IARI, New Delhi. RHIZO system measured the root length by scanning the length of the root skeleton. Measurements of root morphological characteristics were based on Regent's non-statistical method with overlap compensation (Arsenault *et al.* 1995, Aggarwal *et al.* 2006). Plants with a high SRL value have more root length for a given dry-mass and are considered to have higher rates of nutrient and water uptake (per dry mass). Most interactions between plant and soil take place at root-soil interface (Jozefaciuk and Lukowska 2013), meaning more interaction with more surface area of roots.

Specific root length (SRL) and specific root surface area (SRA) are indicators of efficiency of plant nutrient uptake. Roots with a small radius have a higher specific root length (Eissenstat 1992). Plants with a high specific root length are more competitive for below ground nutrients than plants with a low specific root length (Eissenstat and Caldwell

1989). Specific root length and specific root surface area were calculated as follows (Saidi 2010)

$$\text{SRL}=\text{RL}/\text{RW}$$

$$\text{SRA}=\text{RA}/\text{RW}$$

where, SRL, specific root length; SRA, specific root surface area; RL, root length; RA, root surface area and RW, dry weight of roots.

The data thus obtained on various shoot and root parameters of wheat were analysed to decipher the treatments main and interaction effects using SAS 9.3 software in factorial completely randomized design using general linear model. The Tukey's Honest Significant test was used to decipher the treatment effects with probability levels.

## RESULTS AND DISCUSSION

The initial germination percentage of wheat var. HD 2967 was 99.67%. The effect of urea ammonium nitrate on seed germination and different parameters of plant growth are discussed below.

### *Effect of UAN on seed germination and its viability*

Seed germination was affected both on blotting paper and in soil. There was no germination of seed even when the water level was increased upto 30 times (Table 1). However, coleoptiles emerged in soil in 1:30 (UAN:water), but it became brown, scorched and there was no growth thereafter. This may be due to toxicity of ammonia ion released from UAN which is harmful for germinating seeds both due to the increased osmotic potential of the solution and through direct ammonia toxicity. The germination of wheat seed is affected if they are in contact with UAN solution. Cooke (1962) got similar scorched root tips while studying the toxic effect of urea on maize plant roots. Hence proper care must be taken so that both seed and fertilizer doesn't come in contact with each other while designing the sowing equipment.

Seeds picked from blotting paper of each treatment were non viable during Tetrazolium test, meaning that the direct contact with UAN solution has killed the embryo of

Table 1 Germination behaviour of wheat seed with different UAN:water solution

Treatment (UAN:water ratio)	Germination % after seven days	
	Blotting paper	Soil
1:0	No germination	No germination
1:1	No germination	No germination
1:5	No germination	No germination
1:10	No germination	No germination
1:15	No germination	No germination
1:20	No germination	No germination
1:25	No germination	No germination
1:30	No germination	Scorched Coleoptile and thereafter no growth was noticed
Control (only water)	98%	92%

Table 2 Viability (%) determined by the tetrazolium test performed on four lots of wheat seeds on soil

Seed lot	Seed viability (%)							
	UAN:water							
	1:0	1:1	1:5	1:10	1:15	1:20	1:25	1:30
1	0	0	0	0	0	28	76	88
2	0	0	0	0	0	36	80	80
3	0	0	0	0	0	24	76	92
4	0	0	0	0	0	28	80	92

the seeds. However, the seed lots taken from soil showed some viable seeds which are presented in Table 2.

To facilitate proper germination of wheat seeds, the UAN:water dilution ratio should not be less than 1:30. Here the seed was in contact with the soil and UAN:water mixture. However, during sowing in field with the help of fertilizer applicator, space between seed and liquid fertilizer need to be maintained so that UAN does not come in direct contact of seeds. While designing a fertilizer applicator, water should be mixed as little as possible so that the applicator acreage is maximum in a single filling of tank. Application of UAN:water in ratio of 1:30 will require frequent refilling of fertilizer tank in field. Hence a lesser ratio should be preferred. Dilution ratio of 1:5 and 1:10 (UAN:water) was selected for further studies as its pumping is easy than pure UAN.

### *Effect of UAN concentration and application depth on root growth parameters*

Seedlings emerged uniformly across all treatments within 5 days, except in pots containing UAN:water (1:5) and placed at a depth of 1 cm; where it was delayed by 1-2 days.

Root length of wheat seedlings showed significant difference of UAN concentration at all the three (8, 20 and 34 DAS) stages. At lower concentration of UAN, i.e.  $U_1$ (1:5), the root growth significantly reduced by about 43% as compared to  $U_2$  (1:10) after 8 days (Table 3). The higher concentration of UAN had a detrimental effect on the root growth. However, after 34 DAS, the root length of wheat seedling in  $U_1$  (1:5) reduced only by 16% as compared to  $U_2$  (1:10). This suggests that wheat roots were able to overcome the initial metabolic shock due to higher concentration of urea ammonium nitrate solution. This is in accordance with Liu *et al.* (2013), Britto and Kronzucker (2002) who concluded that ammonium at a high concentration usually inhibits primary root development. Deep placement ( $D_3$ ) of UAN resulted in significantly higher root length at all the growth stages over  $D_1$  and  $D_2$ . The greater depth placement might have caused lesser shock to the roots of wheat seedling due to lateral contact of roots with placed fertilizer. Xiaobin *et al.* (1995) concluded that excess concentrations of ammonium ion ( $\text{NH}_4^+$ ) from Urea can produce seedling damage, due to osmotic stress and direct ammonia toxicity. However, no seedling damage was observed during these experiments, but the root growth of

wheat was hampered significantly. There was a significant interaction between UAN concentration and its application at different depth (Table 3). Root growth was maximum in U<sub>2</sub>D<sub>3</sub> and was minimum in U<sub>1</sub>D<sub>1</sub> at all the three growth stages. Similar trends were also seen with root fresh weight and root dry weight.

Placement depths significantly affected the fresh root weight as well as root dry weight at all the growth stages. However, at initial stage (8 DAS), they were not affected by placement depth of 2 and 3 cm. Deep placement (D<sub>3</sub>) of UAN resulted in significantly higher fresh root weight as well as root dry weight at all the growth stages over D<sub>1</sub> (Table 3). Interaction effect of depth with UAN concentration was also found significant where U<sub>1</sub>D<sub>3</sub> at 8 and 34 DAS; and U<sub>2</sub>D<sub>3</sub> gave maximum root dry weight and lowest was observed in U<sub>1</sub>D<sub>1</sub> at all the growth stages (Table 3).

*Effect of UAN concentration and application depth on plant height and fresh shoot weight*

Significant differences was observed at 34 days due to UAN concentration where U<sub>2</sub> gave higher (9%) plant height compared to U<sub>1</sub> (Table 4), though it was statistically non-significant at 8 and 20 DAS. It showed that initial plant vigour can sustain injuries caused by higher concentration but at later stage plant growth was affected due to initial metabolic shock. Deep placement (D<sub>3</sub>) of UAN concentration resulted in significantly higher plant height at all the growth stages over D<sub>1</sub> and D<sub>2</sub>. However, it was

at par with D<sub>2</sub> after 20 DAS. The greater depth placement might have resulted in lesser shock to the wheat seedling due to lesser contact of roots with deep placed fertilizer.

*Effect of UAN concentration and application depth on root surface area, no. of tips, SRL and SRA*

Surface area of roots of wheat seedlings were found significantly affected by UAN concentration as well as its application at different depths at all the growth stages. Roots had more surface area at U<sub>2</sub> (1:10) than U<sub>1</sub> (1:5) concentration at all the growth stages (Table 5). Higher concentration of UAN significantly affected root surface area even after 34 DAS. Deep placement of the fertilizer facilitated more root length, thereby increasing its surface area. Root surface area increased significantly as the depth of placement was increased from D<sub>1</sub> to D<sub>3</sub> after 8, 20 and 34 DAS. Interaction effect of depth with UAN concentration was also found significant. Root surface area was maximum in U<sub>2</sub>D<sub>3</sub> (7.04, 18.26 and 40.89 cm<sup>2</sup>) and minimum in U<sub>1</sub>D<sub>1</sub> (0.91, 7.46 and 15.24 cm<sup>2</sup>) after 8, 20 and 34 DAS, respectively (Table 5). Number of root tips depicts the growth and efficiency of rooting system. Its number decreased significantly as the concentration of UAN increased from U<sub>2</sub> (1:10) (241.5) to U<sub>1</sub> (1:5) (197.2) after 8 days. However, after 34 DAS, the numbers of tips were at par at both the dilution level of UAN (Table 5). This may be due to reduced effect of UAN after a certain interval of time. Similarly, shallow placement of UAN mixture had a

Table 3 Root length and weight in wheat as affected by application of different UAN concentrations at varying depths

Treatment	Root length (cm)			Root fresh weight (g)			Root dry weight (g)		
	8 DAS	20 DAS	34 DAS	8 DAS	20 DAS	34 DAS	8 DAS	20 DAS	34 DAS
<i>UAN:Water</i>									
U <sub>1</sub> =1:5	26.9 <sup>b</sup>	121.4 <sup>b</sup>	250.8 <sup>b</sup>	104.9 <sup>b</sup>	183.7 <sup>b</sup>	229.4 <sup>b</sup>	21.7 <sup>b</sup>	36.7 <sup>b</sup>	46.4 <sup>b</sup>
U <sub>2</sub> =1:10	38.1 <sup>a</sup>	135.7 <sup>a</sup>	291.1 <sup>a</sup>	122.9 <sup>a</sup>	201.7 <sup>a</sup>	249.9 <sup>a</sup>	24.9 <sup>a</sup>	40.7 <sup>a</sup>	49.6 <sup>a</sup>
P-value	<.0001	<.0001	<.0001	0.0046	0.0027	0.0040	0.0099	0.0038	0.0145
HSD	1.40	2.45	4.46	11.94	11.10	13.27	2.40	2.59	2.49
<i>Depth of application</i>									
D <sub>1</sub> =1cm	10.3 <sup>c</sup>	78.8 <sup>c</sup>	151.2 <sup>c</sup>	89.6 <sup>b</sup>	159.2 <sup>c</sup>	199.1 <sup>c</sup>	18.6 <sup>b</sup>	32.2 <sup>c</sup>	39.8 <sup>c</sup>
D <sub>2</sub> =2 cm	24.4 <sup>b</sup>	130.0 <sup>b</sup>	280.6 <sup>b</sup>	122.1 <sup>a</sup>	195.6 <sup>b</sup>	229.1 <sup>b</sup>	24.9 <sup>a</sup>	38.9 <sup>b</sup>	45.9 <sup>b</sup>
D <sub>3</sub> =3 cm	62.8 <sup>a</sup>	177.0 <sup>a</sup>	381.2 <sup>a</sup>	130.0 <sup>a</sup>	223.4 <sup>a</sup>	290.7 <sup>a</sup>	26.4 <sup>a</sup>	45.1 <sup>a</sup>	58.3 <sup>a</sup>
P-value	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
HSD	2.08	3.63	6.61	17.69	16.45	19.66	3.55	3.83	3.69
<i>UAN:Water × Depth</i>									
U <sub>1</sub> D <sub>1</sub>	5.99	74.68	141.21	61.40	147.80	180.40	13.44	29.76	36.32
U <sub>1</sub> D <sub>2</sub>	17.38	127.60	251.62	121.60	192.00	209.60	24.48	37.58	42.84
U <sub>1</sub> D <sub>3</sub>	57.32	162.06	359.66	131.60	211.40	298.20	27.08	42.84	60.02
U <sub>2</sub> D <sub>1</sub>	14.62	82.98	161.10	117.80	170.60	217.80	23.76	34.68	43.22
U <sub>2</sub> D <sub>2</sub>	31.43	132.30	309.52	122.60	199.18	248.60	25.34	40.20	48.98
U <sub>2</sub> D <sub>3</sub>	68.26	191.87	402.68	128.40	235.40	283.20	25.66	47.36	56.52
P-value	0.0121	<.0001	<.0001	0.0004	0.3776	0.0027	0.0009	0.7289	0.0026



Table 4 Plant height and shoot weight in wheat as affected by application of different UAN concentrations at varying depths

Treatment	Plant height (mm)			Fresh shoot weight (g)		
	8 DAS	20 DAS	34 DAS	8 DAS	20 DAS	34 DAS
<i>UAN:Water</i>						
U <sub>1</sub> =1:5	32.8 <sup>a</sup>	152.1 <sup>a</sup>	206.2 <sup>b</sup>	99.1 <sup>b</sup>	230.7 <sup>a</sup>	280.8 <sup>b</sup>
U <sub>2</sub> =1:10	34.3 <sup>a</sup>	157.7 <sup>a</sup>	229.1 <sup>a</sup>	113.4 <sup>a</sup>	220.3 <sup>a</sup>	293.4 <sup>a</sup>
P-value	0.5485	0.2671	<.0001	0.0003	0.2731	0.0174
HSD	5.20	10.29	8.49	6.92	19.12	10.18
<i>Depth of application</i>						
D <sub>1</sub> =1cm	23.60 <sup>c</sup>	126.10 <sup>b</sup>	199.60 <sup>c</sup>	84.20 <sup>b</sup>	214.82 <sup>a</sup>	256.10 <sup>b</sup>
D <sub>2</sub> =2 cm	33.20 <sup>b</sup>	166.10 <sup>a</sup>	218.80 <sup>b</sup>	114.10 <sup>a</sup>	229.01 <sup>a</sup>	295.80 <sup>a</sup>
D <sub>3</sub> =3 cm	43.90 <sup>a</sup>	172.50 <sup>a</sup>	234.50 <sup>a</sup>	120.40 <sup>a</sup>	232.78 <sup>a</sup>	309.40 <sup>a</sup>
P-value	<.0001	<.0001	<.0001	<.0001	0.2677	<.0001
HSD	7.71	15.25	12.57	10.25	28.34	15.09
<i>UAN:Water × Depth</i>						
U <sub>1</sub> D <sub>1</sub>	16.80	96.20	180.20	60.60	208.40	241.40
U <sub>1</sub> D <sub>2</sub>	33.80	173.40	213.20	113.40	239.60	294.60
U <sub>1</sub> D <sub>3</sub>	47.80	186.60	225.20	123.20	244.20	306.40
U <sub>2</sub> D <sub>1</sub>	30.40	156.00	219.00	107.80	221.24	270.80
U <sub>2</sub> D <sub>2</sub>	32.60	158.80	224.40	114.80	218.42	297.00
U <sub>2</sub> D <sub>3</sub>	40.00	158.40	243.80	117.60	221.36	312.40
P-value	0.0063	<.0001	0.0311	<.0001	0.2278	0.0719

Table 5 Surface area, tip and relative parameters of root in wheat as affected by application of different UAN concentrations at varying depths

Treatment	Root surface area (cm <sup>2</sup> )			No. of root tips			Specific root length (mg)			Specific root surface area (cm <sup>2</sup> /g)		
	8 DAS	20 DAS	34 DAS	8 DAS	20 DAS	34 DAS	8 DAS	20 DAS	34 DAS	8 DAS	20 DAS	34 DAS
<i>UAN:Water</i>												
U <sub>1</sub> =1:5	3.1 <sup>b</sup>	13.1 <sup>b</sup>	25.3 <sup>b</sup>	197.2 <sup>b</sup>	1175.8 <sup>b</sup>	1482.7 <sup>a</sup>	10.9 <sup>b</sup>	32.5 <sup>a</sup>	52.6 <sup>b</sup>	128.3 <sup>b</sup>	347.6 <sup>a</sup>	533.5 <sup>b</sup>
U <sub>2</sub> =1:10	4.2 <sup>a</sup>	15.1 <sup>a</sup>	29.9 <sup>a</sup>	241.5 <sup>a</sup>	1267.2 <sup>a</sup>	1509.3 <sup>a</sup>	15.4 <sup>a</sup>	32.7 <sup>a</sup>	57.5 <sup>a</sup>	170.0 <sup>a</sup>	372.4 <sup>a</sup>	592.1 <sup>a</sup>
P-value	<.0001	<.0001	<.0001	0.0064	0.0416	0.7691	<.0001	0.8552	0.0014	0.0005	0.0799	0.0011
HSD	0.28	0.44	0.65	30.61	87.62	185.37	1.44	2.24	2.82	21.39	27.91	32.57
<i>Depth of application</i>												
D <sub>1</sub> =1cm	1.37 <sup>c</sup>	9.57 <sup>c</sup>	15.75 <sup>c</sup>	181.30 <sup>b</sup>	592.80 <sup>c</sup>	813.10 <sup>c</sup>	5.78 <sup>c</sup>	24.66 <sup>c</sup>	38.32 <sup>c</sup>	79.19 <sup>c</sup>	296.56 <sup>b</sup>	400.49 <sup>b</sup>
D <sub>2</sub> =2 cm	3.24 <sup>b</sup>	15.06 <sup>b</sup>	28.91 <sup>b</sup>	216.30 <sup>ab</sup>	1260.40 <sup>b</sup>	1563.90 <sup>b</sup>	9.76 <sup>b</sup>	33.55 <sup>b</sup>	61.19 <sup>b</sup>	130.21 <sup>b</sup>	388.00 <sup>a</sup>	629.34 <sup>a</sup>
D <sub>3</sub> =3 cm	6.24 <sup>a</sup>	17.67 <sup>a</sup>	38.18 <sup>a</sup>	260.40 <sup>a</sup>	1811.30 <sup>a</sup>	2111.00 <sup>a</sup>	23.93 <sup>a</sup>	39.48 <sup>a</sup>	65.70 <sup>a</sup>	238.01 <sup>a</sup>	395.43 <sup>a</sup>	658.52 <sup>a</sup>
P-value	<.0001	<.0001	<.0001	0.0009	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
HSD	0.41	0.64	0.96	45.36	129.84	274.70	2.14	3.32	4.18	31.69	41.36	48.26
<i>UAN:Water × Depth</i>												
U <sub>1</sub> D <sub>1</sub>	0.91	7.46	15.24	150.40	459.40	685.20	4.47	25.22	38.95	67.41	252.24	420.89
U <sub>1</sub> D <sub>2</sub>	2.83	14.62	25.15	201.00	1237.60	1619.40	7.11	34.08	58.85	116.15	389.53	587.91
U <sub>1</sub> D <sub>3</sub>	5.44	17.08	35.47	240.20	1830.40	2143.40	21.19	38.08	60.00	201.29	401.11	591.73
U <sub>2</sub> D <sub>1</sub>	1.83	11.69	16.26	212.20	726.20	941.00	7.10	24.10	37.68	90.97	340.87	380.09
U <sub>2</sub> D <sub>2</sub>	3.65	15.49	32.66	231.60	1283.20	1508.40	12.41	33.01	63.52	144.27	386.46	670.78
U <sub>2</sub> D <sub>3</sub>	7.04	18.26	40.89	280.60	1792.20	2078.60	26.67	40.87	71.39	274.74	389.74	725.32
P-value	0.0557	<.0001	<.0001	0.684	0.0205	0.213	0.194	0.259	0.003	0.116	0.010	0.0005

severe effect on number of tips. After 8 DAS, its number were significantly affected at depth  $D_1$  and  $D_3$ , but was at par at  $D_1$  and  $D_2$  as well as  $D_2$  and  $D_3$ . Number of root tips significantly increased as fertilizer application depth increased after 20 and 34 DAS.

Specific root length was significantly more at  $U_2$  concentration than  $U_1$  after 8 and 34 DAS. However, it was at par after 20 DAS (Table 5). Fertilizer placed at  $D_3$  (3 cm) gave significantly better SRL values than placed at  $D_2$  and  $D_1$  after 8, 20 and 34 DAS. There was almost 145% increase in value of SRL after 8 DAS at depth of 3 cm as compared to 2 cm. This may be due to less effect of UAN to the secondary roots of the wheat seedlings. Since, UAN mixture may be affecting the primary root of the seed at shallow depth, thereby could also affect the growth of secondary root systems. Interaction effect of depth with UAN concentration was also found significant on SRL values where  $U_2D_3$  at 8, 20 and 34 days gave maximum value and lowest was observed in  $U_1D_1$  at 8 days and  $U_2D_1$  at 20 and 34 days.

Specific root surface area is severely affected by high concentration of UAN  $U_1$  (1:5) than  $U_2$  (1:10) at initial growth period (Table 5). However, there was non-significant effect after 20 DAS, but was found significant after 34 DAS. After 8 DAS, the deep placement ( $D_3$ ) of fertilizer mixture significantly increased the SRA values to 238.01  $\text{cm}^2/\text{g}$  from 79.19  $\text{cm}^2/\text{g}$  ( $D_1$ ) and 130.21  $\text{cm}^2/\text{g}$  ( $D_2$ ). However, SRA was at par at a depth of 2 and 3 cm after 20 and 34 DAS. The total root surface of a plant is important for capturing water and nutrients. More value of SRA means more surface is available per unit dry weight of root, i.e. more nutrient and water inflow. Hence it is an indicator of healthy root system.

From the results, it can be concluded that seed became unviable through coming in contact with UAN (28% N). The viability of seed was affected at a dilution ratio of 1:20 (UAN:Water) due to toxicity of ammonium ion, hence, the seed became dead and could not germinate. On the other hand, the seed was viable at a dilution ratio of 1:30 (UAN:Water) and germination was observed but seedling establishment was affected. The pot study showed that space should be maintained while applying UAN along with seed in the field. It may be recommended to keep a vertical gap of at least 2 and 3 cm between seed and UAN with dilution ratio of 1:5 and 1:10 respectively while sowing. These results may form the basis of machinery development for placement of liquid UAN fertilizer with respect to seed.

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