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Nano Iron Sulphide (FeS_2) Application and its Influence on Growth and Yield of Oilseed *Brassica* Species

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ABSTRACT: Nano fertilizers are a topic of greater interest for efficient nutrient supply and has drawn attention of the scientific community of soil and plant scientists. Currently various forms of plant fertilizers are experimented for their applicability in nano form and its influence on crops. With this background, a field experiment was conducted in the agricultural research farm of Banaras Hindu University during *rabi* of 2017-18 and 2018-19 to evaluate the effect of nano FeS_2 on the growth and yield of *Brassica* sp. From the two-year experiment, it was found that nano-FeS_2 significantly influenced the growth and yield of *Brassica sp.*and foliar application at 8 ppm at 35 DAS resulted in highest plant height, dry matter accumulation, relative growth rate, as well as seed and stover yield. Moreover, among *Brassica* sp., *B. juncea* showed superiority in terms of growth and yield over *B. napus* and *B. carinata.*

Keywords: Nano-fertilizer, Oilseed *Brassica*, Iron sulphide**,** Relative growth rate, Seed yield.

INTRODUCTION

Rapeseed-mustard called as oilseed *Brassicas*, is the second most important group of oilseed crops globally after soybean. India is the fourth largest rapeseed-mustard growing country in the world, occupying the fourth position in area and production after Canada, China and European Union. The estimated area, production and productivity of rapeseed-mustard in the world is 36.59 million hectares (mha), 72.37 million tonnes (mt) and 1980 kg/ha, respectively, during 2018-19 (DRMR, 2023). Although India accounts for 19.8% of the total acreage, it is responsible for only 9.8% of the total production (DRMR, 2023). In order to realise the full yield and productivity potential, nanotechnology holds a promising solution, at the same time ensuring sustainable soil health and crop production (Prasad *et al*., 2017; Kumari *et al*., 2021). Nano-fertilizers are synthesized in order to regulate the release of nutrients depending on the requirements of the crops and are more efficient than ordinary fertilizers (Madzokere *et al*., 2021). According to Siddiqui *et al*. (2015) engineered nanoparticles are able to enter into plants cells and leaves, and also can transport DNA and chemicals into plant cells. Thus, the high performance of iron sulphide (F_eS_2) nanoparticles is expected to enhance the growth and yield of oilseed *Brassica* crops. In view of this, a field experiment was undertaken to evaluate and understand the effect of nano iron sulphide (F_eS_2) on growth, and yield parameters of oilseed *Brassica* species.

MATERIALS AND METHODS

The experiment was conducted at the Agricultural Research Farm, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi during two consecutive years of 2017-2018 and 2018-19 in the winter (rabi) season. The site is located 83°03′E longitude, 25°18′N latitude with an altitude of 75.7 meters above average sea level. The total amount of rainfall received during 1st year and 2nd year was 27.20 mm and 59 mm, respectively during the crop growth period. The mean weekly maximum temperature recorded during crop seasons of 2017-18 ranged between 16.1°C to 41°C while during 2018-19 it ranged between 19.8°C to 43.8°C. The sunshine hours recorded in the range of 4.3 to 10.1hrs during entire cropping season of 2017-18 while during next year, it was ranged between 4.7 to 10.1 hrs. Initial soil analysis reported low in the available N (195.56 kg/ha), moderate P_2O_5 (19.03 kg/ha) and K_2O (216.25 kg/ha) and neutral pH (7.32). The experiment was conducted in a factorial randomised block design (RBD) with three factors *viz.,* three levels of genotype (G1: *Brassica carinata*, G2: *Brassica napus* and G3: *Brassica juncea*), two levels of time of application

(T1: 35 DAS and T2: 65 DAS) and four levels of nano F_eS_2 (N0: control or 0 ppm, N1: 4 ppm, N2: 8 ppm and N3: 12 ppm), replicated thrice with a total of 72 treatments combined. Nano iron sulphide powder having particle size of about 43 nm collected from NANO SHELL- Intelligent Materials Pvt. Ltd. Punjab was used for the experiment. Ten plants from each plot were tagged and observation were recorded using standard procedures and scale. Analysis of variance was done as suggested by Gomez and Gomez (1984).

RESULT AND DISCUSSION

Plant height: Data on plant height is presented in Table 1. The data indicated that *Brassica juncea* consistently exhibited the highest plant height (116.78 cm and 146.04 cm) at 60 and 90 DAS, respectively, outperforming *Brassica napus* (91.72 cm and 122.27 cm) and *Brassica carinata* (98.09 cm and 141.23 cm) respectively. Application of iron sulphide nanoparticle had significant effect on the plant height at 60 DAS. Treatment with 8 ppm iron sulphide nanoparticle spray recorded the highest mean plant height (108.03 cm) at 60 DAS. Data recorded from 90 DAS also indicated that highest plant height (142.94 cm) was observed with treatment involving 8 ppm iron sulphide nanoparticle spray. This was followed by 12 ppm (104.84 and 141.08 cm), 4ppm (104.34 and 134.51 cm) and 0 ppm (91.61 and 127.27 cm), respectively at 60 and 90 DAS with application of iron sulphide nanoparticle spray. Time of application had no significant effect on plant height at 60 DAS, however at 90 DAS, application at 35 DAS was found superior to application at 65 DAS. Application of nano Fe had magnetic and chemical properties influencing the enzymes involved in photosynthesis resulting in higher accumulation of photosynthates leading to higher plant height (Mahto *et al*., 2022). In a similar vein, Seif *et al*. (2011) reported an increase in plant height in Borago plants due to the influence of Ag-NP, which led to alterations in GA content.

Dry matter accumulation: The data pertaining to dry matter accumulation of *Brassica* species as affected by time of application and concentration of iron sulphide nanoparticle is presented in Table 2. *Brassica* species displayed varied response to dry matter accumulation. At 60 DAS, *B. juncea* recorded the highest dry matter of 13.58 g, followed by *B.carinata* (12.02 g) and lastly, *B. napus* (9.99 g). Similarly, at 90 DAS, *B.juncea* continued to outperform the other two genotypes and recorded the highest dry matter (30.17 g), followed by *B.carinata* (19.53 g) and *B.napus* (16.05 g). Under influence of nano F_eS_2, at 60 and 90 DAS, dry matter accumulation was in the decreasing order of 8 ppm, 4 ppm, 12 ppm and 0 ppm. Indicating that, increase in iron sulphide nanoparticle beyond 8 ppm will have a declining effect on the dry matter. Time of application had no significant effect on dry matter at 60 DAS. At 90 DAS, application of nano FeS2 at 35 DAS resulted in higher dry matter (23.59 g) than at 65 DAS (20.24 g). The above observations could be attributed to the pivotal role of iron in chlorophyll, a vital component for photosynthesis, underscores its significance in promoting overall plant growth (Hosikian *et al*., 2010). The application of 8 ppm iron sulphide nanoparticles is postulated to uphold the integrity of cell membranes, enabling the plant to allocate its resources towards growth, rather than expending energy on the detoxification of reactive oxygen species (ROS) generated during metabolic processes. This is further supported by the research of Yuan *et al*. (2021), who reported an increase in the dry matter of oilseed rape with the application of nano sulphur.

Relative growth rate (RGR): At different stages, there was a significant variation in the relative growth rate of *Brassica* Species, which was significantly influenced by iron sulphide nanoparticles and time of application and is presented in Table 3. *Brassica juncea* had the highest RGR (0.118 g/g/day) at 30-60 DAS, followed by *Brassica carinata* (0.107 g/g/day), and *Brassica napus* (0.100 g/g/day). However, at 60-90 DAS, a change in trend was observed, where *Brassica napus* outperformed *Brassica carinata* by 6.06%, whereas *B. juncea* remained the highest performing species (0.071 g/g/day). No significant effect of nano F_eS_2 and time of application was observed on the relative growth rate of the crop. This finding is supported by the work of Mahto *et al*. (2022), where the highest RGR was observed in *Brassica juncea*.

Siliqua number on the main shoot: Data on siliqua number on the main shoot as influenced by *Brassica* species, iron sulphide nanoparticles and time of application is presented in Table 4. Among the *Brassica* species, *Brassica napus* recorded the highest siliqua number on the main shoot (62.8), which was significantly higher than that of the other two species under trial. The lowest value (11.2) was recorded for *Brassica carinata*. This increase observed in *Brassica napus* may be attributed to an increase in the number of inflorescences owing to its indeterminate growth habit (Setia *et al*., 1996). All iron sulphide nanoparticles treatment were significantly superior over control (0 ppm). The highest number of siliquae on the main shoot (41.4) was observed under the 8 ppm application, which was significantly higher than that of all other treatments. The lowest value (30.7 was recorded for the 0 ppm treatment. Time of application had a significant effect on the number of siliquae on the main shoot, where application at 35 DAS recorded that higher value of 37.2 as against 34.0 under application at 65 DAS. The photosynthetic machinery of the plant may have functioned more efficiently by providing iron at 35 DAS. This increase in photosynthesis could have contributed to the greater energy and carbon availability for siliqua development (Yeasmin *et al*., 2014).

Test weight: As per the data presented in Table 4, *Brassica juncea* recorded a 48.08% higher test weight than *Brassica napus* and 20.0% higher than *Brassica carinata*. This indicates the superiority of *Brassica juncea* in terms of test weight over the other species under trial. Application of iron sulphide nanoparticles had a substantial influence on the test weight, where the highest value of 3.96 g was observed with 8 ppm application. The order of deceasing test weight was 8 ppm> 12 ppm> 4 ppm> 0 ppm. The application of 8 ppm resulted in an increase of 27.53% in the test weight compared to 0 ppm. Time of application also significantly influenced the test weight during both years, where application at 35 DAS displayed 14.92% higher test weight over application at 65 DAS. The positive impact on siliqua development naturally translates into higher seed yield and test weight. With more seeds per siliqua and potentially improved nutrient translocation to the developing seeds, the result was larger and heavier seeds.

Seed yield: The data on seed yield (Table 5) revealed that *Brassica juncea* displayed impressive superiority over *Brassica carinata* and *Brassica napus* with a seed yield of 2054.5 kg/ha, which was 14.04% and 24.53% higher that than of *Brassica carinata* (1766.0 kg/ha) and *Brassica napus* (1550.5 kg/ha), respectively. Iron sulphide nanoparticles had a positive effect on the seed yield, as an increase in the concentration of iron sulphide nanoparticles led to an increase in seed yield until it reached its peak and further increase led to a decline in the yield. A significant increase in seed yield above 0 ppm was observed for all nanoparticle treatments. The highest seed yield (1946.8 kg/ha) was recorded with the application of 8 ppm, followed by 12 ppm (1819.4 kg/ha), 4 ppm (1783.9 kg/ha), and the lowest seed yield (1609.4 kg/ha) was observed at 0 ppm. The time of application also significantly influenced the seed yield, where application at 35 DAS displayed superiority over application at 65 DAS, accounting for 6.33% higher seed yield. The observed increase in yield can be attributed to the improvement in various growth and yield parameters of *Brassica* species, which is likely facilitated by enhanced nutrient availability. The role of iron as a cofactor and enzyme activator in essential physiological processes, such as photosynthesis, nitrogen fixation, chlorophyll synthesis, respiration, and DNA synthesis, has been well documented (Rout and Sahoo 2015). In this regard, the optimal availability of iron likely resulted in increased cell proliferation, heightened chlorophyll production, and subsequently, elevated dry matter accumulation with efficient nutrient translocation from the source to sink.

The Stover yield: Data on stover yield as influenced by *Brassica* species, iron sulphide nanoparticles, and time of application are presented in Table 5. *Brassica* species displayed variation in stover yield, which was significantly different from one another. The highest stover yield (4838.7 kg/ha) was recorded under *Brassica carinata*, followed by *Brassica juncea* (4658.0 kg/ha) and the lowest (3327.0 kg/ha) under *Brassica napus*. The stover yield as influenced by iron sulphide nanoparticles ranged from 4754.6 to 3839.4 kg/ha. The highest and lowest stover yields were recorded at 8 ppm and 0 ppm, respectively. The time of application also significantly influenced the stover yield during both years, where application at 35 DAS was superior to application at 65 DAS, which accounted for a 6.25% higher stover yield.

Table 1: Plant height (cm) of Oilseed *Brassica species* as influenced by iron sulphide nanoparticles and time of application at 60 and 90 days after sowing (DAS) (Average data of two years)

Iron sulphide nanoparticles (ppm)	60 DAS						Mean	90 DAS						Mean
	Oilseed *Brassica species*							Oilseed *Brassica species*						
	B. carinata		*B. napus*		*B. juncea*			*B. carinata*		*B. napus*		*B. juncea*		
	Time of application		Time of application		Time of application			Time of application		Time of application		Time of application		
	35 DAS	65 DAS	35 DAS	65 DAS	35 DAS	65 DAS		35 DAS	65 DAS	35 DAS	65 DAS	35 DAS	65 DAS	
0	91.95	88.80	85.48	82.88	100.55	99.98	91.61	133.46	135.75	113.12	110.32	137.52	133.43	127.27
4	99.33	95.17	94.98	93.38	117.23	116.90	104.34	138.00	137.10	124.77	119.50	149.74	137.99	134.51
8	103.25	101.48	95.91	92.12	128.45	123.17	108.03	150.36	142.06	133.25	123.35	158.27	150.37	142.94
12	101.25	100.28	95.65	93.38	124.90	123.17	104.84	148.08	145.10	132.47	119.80	152.99	148.06	141.08
Mean	99.45	96.73	93.00	90.44	117.78	115.81		142.47	140.00	125.90	118.24	149.63	142.46	
	SEm±			**C.D.at 5%**				**SEm±**			**C.D.at 5%**			
Oilseed *Brassica* species (A)	**1.26**			**3.59**				**1.81**			**5.16**			
Time of application (B)	**1.03**			**NS***				**1.48**			**4.21**			
A × B	**1.775**			**NS***				**2.56**			**NS***			
Iron sulphide nanoparticle (C)	**1.45**			**4.23**				**2.09**			**5.96**			
A × C	**2.52**			**7.17**				**3.62**			**NS***			
B × C	**2.06**			**NS***				**2.95**			**NS***			
A × B × C	**3.56**			**NS***				**5.12**			**NS***			

*NS- non-significant

Table 2: Dry matter accumulation (g/plant) of Oilseed *Brassica species* as influenced by iron sulphide nanoparticles andtime of application at 60 and 90 days after sowing (DAS) (Average data of two years).

Iron sulphide nanoparticles (ppm)	60 DAS						Mean	90 DAS						Mean
	Oilseed *Brassica species*							Oilseed *Brassica species*						
	B. carinata		*B. napus*		*B. juncea*			*B. carinata*		*B. napus*		*B. juncea*		
	Time of application		Time of application		Time of application			Time of application		Time of application		Time of application		
	35 DAS	65 DAS	35 DAS	65 DAS	35 DAS	65 DAS		35 DAS	65 DAS	35 DAS	65 DAS	35 DAS	65 DAS	
0	10.38	9.11	9.03	7.01	11.73	10.64	9.65	16.24	13.71	13.79	11.32	25.78	22.64	17.24
4	12.92	11.28	11.23	11.25	14.31	11.19	12.03	21.07	18.75	18.74	19.38	29.68	26.15	22.29
8	15.23	12.00	12.00	10.32	19.54	14.61	13.95	25.38	19.47	19.47	16.28	39.35	36.05	26.00
12	14.45	10.81	10.81	8.24	14.21	12.46	11.83	24.19	17.44	17.25	12.22	32.14	29.55	22.13
Mean	13.24	10.80	10.77	9.21	14.94	12.23		21.72	17.34	17.31	14.80	31.74	28.60	

	SEm ±	CD at 5%	SEm±	CD at 5%
Oilseed *Brassica* species (A)	0.31	0.86	0.71	2.02
Time of application (B)	0.25	0.71	0.58	1.65
A × B	0.42	NS*	1.00	NS*
Iron sulphide nanoparticle (C)	0.35	0.99	0.82	2.33
A × C	0.60	1.71	1.41	4.03
B × C	0.49	NS*	1.15	NS*
A × B × C	0.85	NS*	2.01	NS*

*NS- non-significant

Table 3: Relative growth rate (g/g/day) of Oilseed *Brassica species* as influenced by iron sulphide nanoparticles andtime of application between 30-60 and 60-90 days after sowing (DAS) (Average data of two years).

Iron sulphide nanoparticles (ppm)	30-60 DAS						Mean	60-90 DAS						Mean
	Oilseed *Brassica species*							Oilseed *Brassica species*						
	B. carinata		*B. napus*		*B. juncea*			*B. carinata*		*B. napus*		*B. juncea*		
	Time of application		Time of application		Time of application			Time of application		Time of application		Time of application		
	35 DAS	65 DAS	35 DAS	65 DAS	35 DAS	65 DAS		35 DAS	65 DAS	35 DAS	65 DAS	35 DAS	65 DAS	
0	0.107	0.099	0.099	0.087	0.113	0.107	0.102	0.062	0.062	0.064	0.062	0.068	0.072	0.065
4	0.110	0.102	0.102	0.100	0.117	0.110	0.107	0.065	0.058	0.068	0.065	0.075	0.075	0.068
8	0.110	0.114	0.114	0.100	0.127	0.123	0.115	0.065	0.065	0.065	0.065	0.068	0.065	0.066
12	0.107	0.109	0.109	0.090	0.123	0.127	0.111	0.067	0.055	0.068	0.067	0.075	0.068	0.067
Mean	0.109	0.106	0.106	0.094	0.120	0.117		0.065	0.060	0.067	0.065	0.072	0.070	

	SEm±	CD at 5%	SEm±	CD at 5%
Oilseed *Brassica* species (A)	0.001	0.004	0.001	0.003
Time of application (B)	0.001	0.003	0.001	0.003
A × B	0.002	NS*	0.002	NS*
Iron sulphide nanoparticle (C)	0.002	0.005	0.001	NS*
A × C	0.003	0.008	0.002	NS*
B × C	0.002	NS*	0.002	NS*
A × B × C	0.004	NS*	0.003	NS*

*Non-significant

Table 4: Siliqua number on the main shoot and Test weight (g) of Oilseed *Brassica species* as influenced by iron sulphide nanoparticles andtime of application (Average data of two years).

Iron sulphide nanoparticles (ppm)	Siliqua number on the main shoot							Test weight (g)						
	Oilseed *Brassica species*						Mean	Oilseed *Brassica species*						Mean
	B. carinata		*B. napus*		*B. juncea*			*B. carinata*		*B. napus*		*B. juncea*		
	Time of application		Time of application		Time of application			Time of application		Time of application		Time of application		
	35 DAS	65 DAS	35 DAS	65 DAS	35 DAS	65 DAS		35 DAS	65 DAS	35 DAS	65 DAS	35 DAS	65 DAS	
0	10.4	9.2	57.6	50.6	29.9	26.3	30.7	2.92	2.62	2.43	2.05	3.95	3.30	2.87
4	11.2	9.8	62.8	57.6	32.9	30.1	34.1	3.43	3.13	2.95	2.56	4.46	3.81	3.39
8	14.4	11.8	74.6	70.9	38.9	37.6	41.4	4.00	3.70	3.52	3.13	5.03	4.38	3.96
12	12.1	10.8	66.8	61.7	35.0	32.3	36.5	3.83	3.53	3.34	2.96	4.86	4.21	3.78
Mean	12.0	10.4	65.4	60.2	34.2	31.6		3.55	3.25	3.06	2.68	4.58	3.93	
	SEm±			**CD at 5%**				**SEm±**			**CD at 5%**			
Oilseed *Brassica* species (A)	**0.7**			**2.0**				**0.07**			**0.21**			
Time of application (B)	**0.6**			**1.6**				**0.06**			**0.17**			
A × B	**1.0**			**NS***				**0.10**			**NS***			
Iron sulphide nanoparticle (C)	**0.8**			**2.3**				**0.52**			**0.24**			
A × C	**1.4**			**4.0**				**0.14**			**0.42**			
B × C	**1.1**			**NS***				**0.12**			**NS***			
A × B × C	**2.0**			**NS***				**0.20**			**NS***			

*Non-significant

Table 5: Stover yield (kg/ha) and Seed yield (kg/ha) of Oilseed *Brassica species* as influenced by iron sulphide nanoparticles andtime of application (Average data of two years).

Iron sulphide nanoparticles (ppm)	Stover yield (kg/ha)							Seed yield (kg/ha)						
	Oilseed *Brassica species*						Mean	Oilseed *Brassica species*						Mean
	B. carinata		*B. napus*		*B. juncea*			*B. carinata*		*B. napus*		*B. juncea*		
	Time of application		Time of application		Time of application			Time of application		Time of application		Time of application		
	35 DAS	65 DAS	35 DAS	65 DAS	35 DAS	65 DAS		35 DAS	65 DAS	35 DAS	65 DAS	35 DAS	65 DAS	
0	4349.5	4204.5	3343.0	2916.5	4204.5	4018.5	3839.4	1627.5	1547.5	1461.5	1320.5	1896.0	1803.5	1609.4
4	4514.5	4533.5	3763.5	3384.5	4533.5	4145.5	4145.8	1764.5	1667.5	1731.0	1559.5	2037.5	1943.5	1783.9
8	5727.5	5427.0	3614.5	3173.5	5427.5	5157.5	4754.6	1968.0	1943.5	1679.5	1523.0	2303.5	2263.5	1946.8
12	4882.5	5071.0	3433.5	2987.5	5071.0	4707.5	4358.8	1881.5	1726.5	1616.5	1510.0	2171.5	2010.5	1819.4
Mean	4868.5	4809.0	3538.5	3115.5	4809.0	4507.0		1810.5	1721.5	1622.5	1478.5	2102.5	2005.5	
	SEm±			**CD at 5%**				**SEm±**			**CD at 5%**			
Oilseed *Brassica* species (A)	**51.0**			**145.2**				**20.8**			**59.2**			
Time of application (B)	**41.6**			**118.5**				**17.0**			**48.4**			
A × B	**72.1**			**172.2**				**29.4**			**NS***			
Iron sulphide nanoparticle (C)	**58.9**			**167.6**				**24.0**			**68.4**			
A × C	**102.0**			**290.3**				**41.6**			**118.4**			
B × C	**83.3**			**NS***				**34.0**			**NS***			
A × B × C	**144.2**			**NS***				**58.8**			**NS***			

*Non-significant

CONCLUSION AND FUTURE SCOPE

From the results obtained based on a two-year experiment, it can be concluded that *Brassica juncea* showed superiority over *B. napus* and *B. carinata* in terms of plant height and accumulation of dry matter as well as test weight and seed yield, at the same time, this genotype was most responsive to the application of nano F_eS_2. The optimal level of nano F_eS_2 for foliar application was 8 ppm, regardless of the genotype of *Brassica* species. Additionally, the application of nano F_eS_2 was the most effective when sprayed at 35 DAS. This suggests that the timing and dosage are critical factors in maximizing the benefits of nano F_eS_2 application. However, further research is warranted to fully understand its implications and optimize its utilization in sustainable agricultural practices.

Understanding the genetic and physiological factors contributing to the genetic superiority observed among *Brassica* sp. could facilitate the development of cultivars with enhanced resilience and productivity. Long-term studies focusing on the sustainability and ecosystem effects of nano F_eS_2 supplementation would provide valuable insights into its role in promoting agricultural sustainability. Exploring the potential synergies between nano F_eS_2 and other agronomic practices, such as integrated pest and soil health management, could offer an opportunity to enhance crop productivity and resilience in diverse agro-ecosystems.

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Conflict of Interest. None.

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