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Farmyard Manure as a Green Solution for Mitigating Arsenic Induced Phytotoxicity in *Pisum sativum*

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Keywords

Arsenic toxicity;
Farmyard manure;
Soil contamination;
Sustainable agriculture;
Plant growth;
Environmental remediation

Abstract

Arsenic contamination in soil is a major environmental hazard, reducing plant development and agricultural productivity. This study investigates how farmyard manure (FYM) can help reduce arsenic-induced toxicity in *Pisum sativum*. The major goal was to determine how FYM improved seed germination, growth characteristics, and metabolic reactions in *Pisum sativum* subjected to various arsenic concentrations (control, 5, 10 and 20 ppm). Our findings show that arsenic exposure dramatically reduces seed germination, root and shoot growth, and chlorophyll content in *Pisum sativum.* However, the addition of FYM considerably reduced the harmful effects, increasing germination rates, root and shoot growth, and chlorophyll content when compared to without arsenic-treated soils. FYM amendment also improved soil properties, potentially reducing arsenic bioavailability and facilitating better plant growth under contaminated conditions. These findings suggest that FYM is a promising and sustainable approach for mitigating arsenic toxicity in agricultural soils, offering an environmentally friendly solution to improve crop yield and food safety in arsenic-affected regions.

1.Introduction

The contamination of soil and water by arsenic (As) has become a critical global environmental and agricultural challenge, particularly affecting South and Southeast Asia. In these areas, the extensive use of arsenic-contaminated groundwater for irrigation significantly jeopardizes crop yields and food safety (Singh *et al.,* 2023). In India, regions such as West Bengal, Bihar, Uttar Pradesh, Assam and Jharkhand often report groundwater arsenic levels that surpass the World Health Organization's recommended threshold of 10 µg/L (WHO, 2020). Furthermore, recent research highlights rising arsenic concentrations in irrigation water sourced from the Indus River basin in Punjab and Haryana, exacerbating worries about arsenic buildup in agricultural soils (Shaji *et al*., 2021). Arsenic is a harmful metalloid that disrupts various physiological and biochemical processes in plants. Its presence hinders photosynthesis, affects nutrient absorption, and causes oxidative stress, which can result in stunted growth and lower yields (Zemanova *et al*., 2021). Among leguminous crops, *Pisum sativum* (pea) is crucial for Indian agriculture due to its nutritional benefits and ability to enrich soil. However, it is notably vulnerable to arsenic toxicity, making it an ideal candidate for researching arsenic-related stress in plants. Traditional methods for reducing arsenic toxicity in crops typically include the application of organic soil amendments like FYM (Brunet *et al*., 2020). Research indicates that FYM can improve soil structure, boost microbial activity, and enhance organic matter levels, which aids in the immobilization of arsenic and decreases its availability to plants (Mukhopadhyay *et al.,* 2011). However, there is a lack of extensive research on the combined effects of FYM and arsenic stress in *Pisum sativum*. This study seeks to explore the potential of FYM to alleviate arsenic toxicity in *Pisum sativum*. The research will specifically assess variations in plant growth metrics, significant metabolic responses, and arsenic levels in various plant tissues under conditions of combined FYM and arsenic treatment. The results are estimated to support sustainable agricultural practices by providing valuable insights into environmentally friendly approaches for managing arsenic stress in vulnerable crop species.

2. Material and Methods

2.1. Plant material and treatment details

Certified seeds of *Pisum sativum* PB89 were obtained from the Directorate of Agriculture, Boileuganj, Shimla, and the ICAR-National Bureau of Plant Genetic Resources, Regional Station, Shimla, India. Arsenic-induced metal stress was administered using Sodium Arsenate. A stock solution of arsenic (1000 ppm) was prepared and subsequently diluted to achieve concentrations of 5, 10 and 20 ppm with a control treatment (without arsenic), using distilled water.

2.2. Preparation of Pots and Experimental design

The pot culture experiment used soil as a growing medium. Soil was gathered from the field, sieved and equalized in the laboratory. The physicochemical characteristics of the soil used in the experiment are given in Table 1. For the pot culture experiment, sixteen 5*5-inch pots were used. Eight pots were prepared and filled with 250 grams of field soil, followed by eight pots filled with 250 grams of soil mixed with FYM in a 1:1 ratio. The pots were labelled according to the experimental design.

Table 1. Physico-chemical characteristics of the soil used for pot culture experiment

Parameters with units	Soil	Soil +FYM
pH	7.5	8.1
EC(dS/m)	0.109	0.181
N (kg/ha)	175.6	238.3
P(kg/ha)	0.341	0.386
K(kg/ha)	1456	2553
TOC(%)	2.7	2.7
Zn(ppm)	3.7	3.2
Fe(ppm)	2.0	1.55
Mn(ppm)	1.61	1.67
Cu(ppm)	2.52	2.56

The design of pot culture experiment is as follows:

A: 2 Replicate * 4 treatments with As (0, 5, 10, 20ppm) = 8 Pots.

B: 2 Replicate * 4 treatments with As (0, 5, 10, 20ppm) +FYM = 8 Pots.

The experiment was conducted in duplicates. 8 seeds were then sown in each pots having soil As and As + FYM. These were kept for germination at 25°C. Different concentrations of As 0, 5, 10, 20ppm was given three times weekly in respective pots.

2.3. Analysis of growth parameters

2.3.1 Germination percentage and percent inhibition

Germination percentage of seed was calculated using equation:

$$GP = (n / N) \times 100 \quad \text{---------------------------------(1)}$$

where ‘n’ is the number of grown seeds and N is the complete number of tested seeds.
Percent inhibition of germination was calculated with following formula given by Baruah. (Baruah *et al.*2019)

Percent inhibition of germination =100-GI of treatment/ GI of control*100 ---------(2)

2.3.2. Length of root and shoot

Ruler was used to measure the length of root and shoot. The roots and shoots were separated by cutting down them with the help of sharp blade. They were lay down on white sheet of paper and ruler was placed parallel to them to measure the length.

2.3.3. Fresh and dry weight of root, shoot and cotyledon

Fresh weight of whole plant was done by keeping it at weighing balance. The roots, shoots and cotyledons were separated by cutting them with the help of blades. Dry weight was measured by oven drying plant material at 50^{o}C overnight.

2.3.4. Germination index

Germination index (GI) was determined by the formula given by (AOSA 1983):

$$GI = \frac{No.of\ germinated\ seeds}{Days\ of\ first\ count} + \cdots + \frac{No.of\ germinated\ seeds}{Days\ of\ final\ count} \quad \text{------------(3)}$$

2.3.5. Chlorophyll content

The chlorophyll content of leaf tissue of *Pisum sativum* was estimated following (Harborne, 1973) method.100 mg leaf tissue was homogenized with 80% acetone in pestle mortar. The homogenate was transferred, and the procedure was repeated till the residue becomes colourless. The final volume of homogenate was made 2 ml with 80% acetone. This homogenate was centrifuged at 5000-10000 rpm for 5 minutes and a supernatant was obtained. This supernatant was transferred to cuvette and measured absorbance with the help of spectrophotometer at 645 nm - 663nm against the solvent (acetone) blank. The total chlorophyll was calculated by the formula given by Arnon (1949):

$$\text{Total chlorophyll (mg/g) FW} = 20.2\ (A_{645}) + 8.02\ (A_{663}) \times V/1000 \times W \quad \text{---------------(4)}$$

Where, A= Absorbance at specific wavelengths i.e. 663 and 645; V= Final volume of chlorophyll extract in 80% acetone and FW= Weight of the fresh tissues extracted.

2.3.6. Vigor index

Seed vigor index was calculated as under following (Maisuria and Patel, 2009):

$$Vigor\ index = Root\ length + Shoot\ length \times Seed\ germination\ \%$$ ---------------(5)

2.3.7. Tolerance index

The tolerance index (T.I.) was calculated using the formula given by (Iqbal and Rahmati 1992).

$$T.I. = \frac{Mean\ root\ length\ in\ the\ metal\ solution}{Mean\ root\ length\ in\ control} \times 100$$ -----------------------(6)

2.3.8. Relative germination rate: The relative germination rate (RGR) and the root and shoot length was measured, as well as the ratio between them.

$$RGR = \frac{Germination\ percentage\ in\ the\ metal\ concentration}{Germination\ Percentage\ in\ the\ control}$$ ---------------------(7)

2.3.9. Percent Phytotoxicity

Percent phytotoxicity was calculated on 8th day after germination using the formulae of Choui *et al*., 1976.

Percent Phytotoxicity = Radical length in control – Radical length in treatment /Radical length of control *100 ---------------------------(8)

2.4. Statistical Analysis

The seed germination and seedling growth data were analysed by using student's t test done in XLSTAT and two-way Analysis of Variance (ANOVA) done by using SPSS software followed by Tukey-Kramer Significance Difference (HSD) test between the means of treatment to determine the significant difference. All values in this experiment are mean of three in petriplates.

3. Results and Discussion

3.1. Germination Percentage

The experiment described investigates the germination percentage of *Pisum sativum* (pea) seeds under different concentrations of arsenic (0, 5, 10, 20 ppm) with and without FYM in soil. The daily germination percentage of *Pisum sativum* at different concentrations of arsenic (0, 5, 10, 20 ppm) and As + FYM sown in soil is described in Fig. 1. Exposure to arsenic (As) markedly diminished the germination rates of *P. sativum* seeds in a dose-dependent manner. The germination percentage decreased from 34.09% in the control group to 22.72%, 13.06% and 6.81% at concentrations of 5, 10 and 20 ppm As, respectively. This demonstrates the detrimental impact of arsenic on initial seed development, due to impaired respiration and water absorption. Conversely, the addition of FYM enhanced germination rates across all experimental groups. In the control + FYM group, germination increased to 44.31%, while at 5, 10, and 20 ppm As + FYM, the rates improved to 31.8%, 26.7%, and 21.5%, respectively. By adding FYM as amendment to the metal contaminated soil at each concentration germination percentage showed statistically significant increase ($p = 0.00037$; $p \leq 0.05$) according to student's t test.

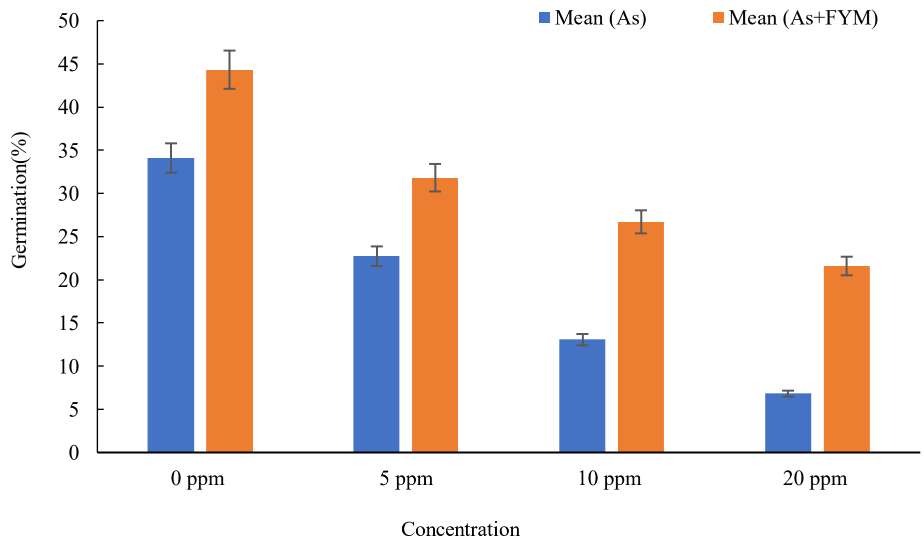

Figure 1. Variation in Mean Germination Percentage of *Pisum sativum* at various concentrations in Soil with As and As+FYM (Day 11).

The mean values at different concentrations of As and As+FYM are statistically significant according to Multivariate Tukey (HSD) at ($P \leq 0.05$) and student's 't' test between two groups.

It is likely that FYM lessens the impact of arsenic toxicity by improving soil quality and decreasing arsenic bioavailability, as indicated by (Mukhopadhyay *et al.* 2011). These results are consistent with earlier studies (Tripathi *et al.,* 2012; Adeloju *et al*., 2021) and imply that FYM can effectively mitigate arsenic stress, thereby enhancing seed germination and providing a viable strategy for cultivation in contaminated soils.

3.2. Root and Shoot Length

The study examined the impact of arsenic stress on *Pisum sativum* seedlings, with and without the addition of farmyard manure (FYM). The root and shoot lengths were measured at 0 ppm, with 100% representing the control values as shown in Fig. 2. At 5 ppm arsenic, root length decreased to 3.7 cm, while shoot length increased to 17.9 cm. FYM mitigated the negative effects of arsenic and promoted shoot growth at this concentration. At 10 ppm arsenic, root length declined to 2.8 cm, but with amendment of FYM it showed improvement to 3.8 cm. Shoot length also decreased under arsenic stress, with the As treatment exhibiting a length of 12.1 cm. At the highest arsenic concentration (20 ppm), both root and shoot growth were significantly affected. FYM helped alleviate these negative effects, especially at lower arsenic levels (5 ppm), with a more noticeable recovery in shoot length than root length. However, at higher arsenic concentrations (10 and 20 ppm), the ameliorative effects of FYM were less pronounced, suggesting its limited capacity to counteract severe arsenic toxicity. Shoot length was also affected by arsenic. Without FYM, shoot lengths decreased from 14.3 cm at 5 ppm to 12.1 cm at 10 ppm, and down to 10.5 cm at 20 ppm. However, the presence of FYM consistently enhanced shoot growth under arsenic stress. According to Student's t-test, adding farmyard manure (FYM) as an amendment to metal-contaminated soil caused a statistically significant increase in root length at each concentration (p = 0.00082; p ≤ 0.05). The mean shoot length of *Pisum sativum* also increased statistically significantly (p = 0.00089; p ≤ 0.05), suggesting that FYM has a beneficial impact on plant growth in the presence of metal stress. This demonstrates the role of FYM in supporting shoot elongation under heavy metal stress. The results are supported by the work of other researchers (Castillo-Michel *et al.,*2007) on *P. sativum*, which showed decrease in root and shoot length by increasing arsenic concentrations but increase in shoot and root length by adding amendments (Srivastva *et al.,* 2013) on black gram.

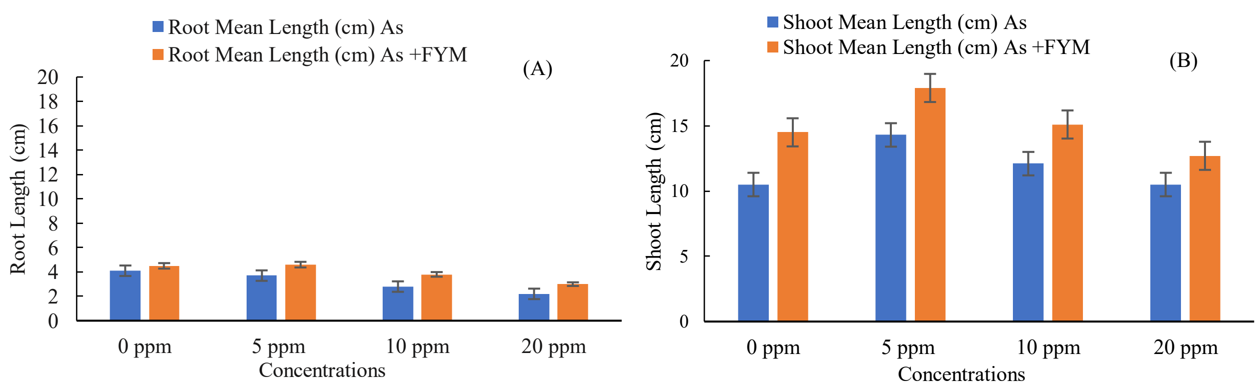

Figure 2. Effect of arsenic applied either singly or in combination with FYM on Root and Shoot Length of *Pisum sativum* (A) Root Mean Length; (B) b) Shoot Mean Length

The mean values at different concentrations of As and As+FYM are statistically significant according to Multivariate Tukey (HSD) at (P≤0.05) and student's 't' test between two groups.

3.3. Fresh Weight and Dry weight (Root, Shoot)

Table 2 shows how different concentrations of arsenic (As), applied either alone or with FYM, affect the fresh and dry weight of roots and shoots in *P. sativum*. At the control (0 ppm), plants grown in soil with arsenic had a fresh root weight of 0.49 g and a fresh shoot weight of 0.52 g, while plants grown in FYM-treated soil had slightly higher weights (0.55 g for roots and 0.58 g for shoots). With addition of arsenic to the soil at 5 ppm, the fresh root weight dropped to 0.34 g but stayed almost the same (0.37 g) with FYM-treated soil. Whereas the fresh shoot weight increased in both As and As+FYM treatments compared to the control, with values of 0.81 g and 0.85 g, respectively. The dry weights followed a similar pattern, with arsenic treatment causing a decrease in root and shoot dry weights, but FYM helped reduce the loss, especially in shoot dry weight. At 10 ppm of arsenic, the fresh root weight decreased further to 0.32 g in As-treated soil, while it was 0.34 g in As+FYM soil. The fresh shoot weight also decreased but was still higher in FYM-treated soil (0.68 g) than in As soil (0.59 g). The dry weight of both roots and shoots continued to decline with arsenic exposure, although FYM treatment helped to some extent. At the highest concentration of 20 ppm, the fresh root weight in As-treated soil decreased to 0.26 g, while it remained higher (0.32 g) in FYM-treated soil. Similarly, fresh shoot weight was 0.51 g in As soil and 0.64 g in As+FYM soil.

Table 2. Effect of arsenic applied either singly or in combination with FYM on fresh weight of root and shoot of *Pisum sativum*

Concentration (ppm)	Fresh Root Mean Weight (g)		Fresh Shoot Mean Weight (g)		Root Mean Dry Weight (g)		Shoot Mean Dry Weight (g)	
	As	As+FYM	As	As	As + FYM	As + FYM	As	As+ FYM
0 ppm	0.49±0.05 (100)	0.55±0.05 (100)	0.055±0.008 (100)	0.364±0.003 (100)	0.52±0.09 (100)	0.071±0.02 (100)	0.58±0.09 (100)	0.72±0.1 (100)
5 ppm	0.34±0.07 (69.3)	0.37±0.05 (67.2)	0.043±0.004 (78.1)	0.29±0.003 (80)	0.31±0.05 (60)	0.06±0.01 (85.7)	0.81±0.05 (139)	0.85±0.02 (118)
10 ppm	0.32±0.08 (65.3)	0.34±0.07 (61)	0.038±0.006 (69)	0.212±0.001 (59)	0.23±0.02 (44)	0.04±0.008 (57)	0.59±0.09 (101)	0.68±0.02 (94.4)
20 ppm	0.26±0.03 (53)	0.32±0.04 (58)	0.025±0.002 (45.4)	0.19±0.001 (52)	0.21±0.01 (40)	0.022±0.002 (30)	0.51±0.08 (87.9)	0.64±0.07 (88.8)

The mean values at different concentrations of As and As+FYM are statistically significant according to Multivariate Tukey (HSD) at ($P \leq 0.05$) student's 't' test between two groups.
Values in parenthesis represent relative values, respective control taken as 100.

Both root and shoot dry weights were significantly lower in As-only soil compared to the FYM-treated soil. This shows that organic amendments like FYM can partially reduce the harmful impact of arsenic on plant growth. Adding FYM as an amendment to metal-contaminated soil at each concentration significantly increased shoot dry weight ($p = 0.031$, $p \leq 0.05$) based on student's t-test. The improvement in growth parameters with FYM suggests that organic amendments may help mitigate arsenic toxicity by improving soil health and nutrient availability, as noted in other studies on heavy metal stress (Alam *et al.*, 2020)

3.4. Chlorophyll content

The data presented Fig. 3 illustrates the impact of varying arsenic concentrations on total chlorophyll content in soils with and without farmyard manure. Chlorophyll content serves as a crucial indicator of photosynthetic activity and overall plant health. The total chlorophyll content of *Pisum sativum* was significantly affected by arsenic concentrations, with lower values observed in As-treated soils compared to those amended with FYM. At 0 ppm (control), chlorophyll content was 0.061 mg/g in As-treated soil and 0.118 mg/g in FYM-treated soil. As the concentration of arsenic increased, chlorophyll content decreased sharply in As-treated soil, with values of 0.026 mg/g at 5 ppm, 0.054 mg/g at 10 ppm, and 0.059 mg/g at 20 ppm. In contrast, FYM treatment improved chlorophyll content, especially at higher arsenic concentrations, showing 0.209 mg/g at 5 ppm, 0.084 mg/g at 10 ppm, and 0.080 mg/g at 20 ppm (Fig. 4.). For instance, (Khan *et al.* 2023) demonstrated that compost and FYM enhanced biomass and chlorophyll content in maize grown in arsenic-contaminated soil (Alam *et al.* 2020) found that the addition of organic matter improved photosynthetic pigments and growth in rice exposed to arsenic. Collectively, these studies highlight the importance of organic amendments in enhancing plant resilience to heavy metal stress through improved nutrient availability, microbial activity and detoxification processes.

3.5. Growth indices

Table 3 presents the effects of arsenic (As) and FYM on various growth indices of *P. sativum*, including the Germination Index, Vigour Index, Relative Germination Rate, Tolerance Index and Percent Phytotoxicity. When arsenic was applied at 5 ppm, the Germination Index increased to 9.69 in As-treated soil but remained higher at 13.55 in FYM-treated soil, showing that

FYM continues to support better germination even under low arsenic stress. At 10 ppm, the Germination Index decreased in both treatments, with values of 7.31 for As-treated soil and 10.95 for FYM-treated soil, reflecting the negative impact of arsenic on germination, but still demonstrating the mitigating effect of FYM. At the highest concentration of 20 ppm, the Germination Index dropped drastically to 3.6 in As-treated soil, compared to 8.7 in FYM-amended soil, further emphasizing the protective role of FYM in reducing arsenic-induced damage to seed germination. Overall, the results show that FYM significantly improved the Germination Index at all arsenic concentrations, indicating that it helps mitigate the toxic effects of arsenic on seedling growth. FYM helps mitigate arsenic toxicity by immobilizing arsenic through organic matter complexation and promoting microbial transformations. It improves soil structure, moisture retention, and nutrient availability, promoting germination. FYM also enhances antioxidant defense systems in plants by increasing enzyme activity, reducing oxidative damage caused by arsenic-induced reactive oxygen species (Nazir *et al*., 2024; Nahar *et al.,* 2022).

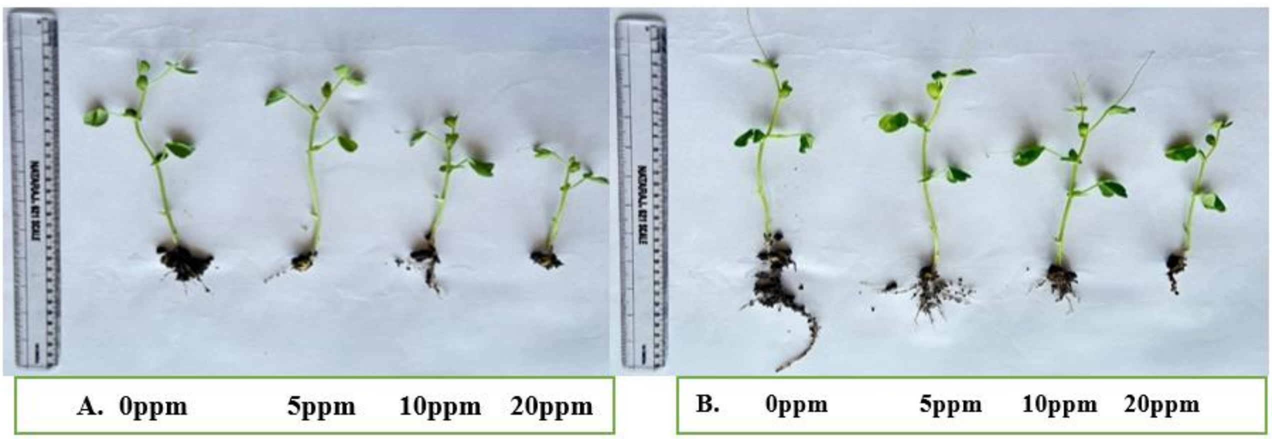

Figure 3. Photograph showing growth of P. *sativum* at different concentration of As (a) and As+FYM (b)

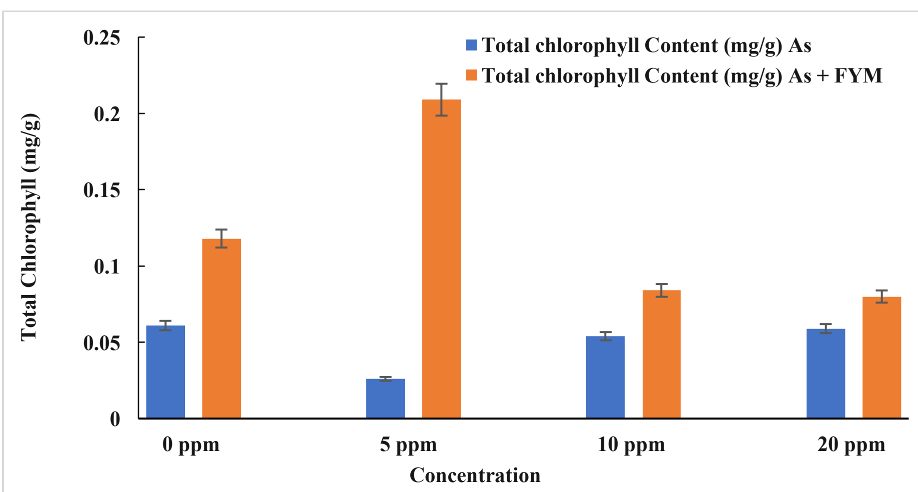

Figure 4. Effect of arsenic applied either singly or in combination with FYM on total chlorophyll content of *Pisum sativum*

The mean values at different concentrations of As and As+FYM are statistically significant according to Multivariate Tukey (HSD) at ($P \leq 0.05$) and student's 't' test between two groups.

Table 3. Effect of arsenic applied either singly or in combination with FYM on Germination Index, Vigor Index, Relative Germination Rate, Tolerance Index, Percent Phytotoxicity% of *Pisum sativum* sown in Soil

Concentration (ppm)	Germination Index		Vigour Index		Relative Germination Rate		Tolerance Index		Percent Phytotoxicity	
	As	As+FYM	As	As +FYM	As	As+ FYM	As	As + FYM	As	As+ FYM
0 ppm	7.25	16.82	766.5	1157.1	1	1	100	100	0	0
5 ppm	9.69	13.55	680.5	870.9	0.72	0.75	90.2	102.2	9.75	2.22
10 ppm	7.31	10.95	451.47	825.9	0.57	0.71	68.2	84.4	31.7	15.55
20 ppm	3.6	8.7	194.3	555.7	0.29	0.48	53.6	66.6	46.3	55.55

The study examined the effect of arsenic (As) concentrations on the vigor index of plants grown in soil, with and without FYM. The results showed that as arsenic concentration increased, the vigor index decreased significantly. At 0 ppm As, the vigor index was 766.5, but decreased to 194.3 at 20 ppm As. This suggests that higher levels of As negatively affect plant vigor when organic matter is not present. In soil supplemented with As + FYM, initial vigor indices were higher across like 1157.1, 870.9, 825.9 and 555.7 all As+FYM concentrations compared to soil with As. However, as As concentration increased with FYM, the vigor index also declined, less drastically compared to soil containing As. Similar findings were reported by (Yadav *et al.* 2020), who discovered that FYM increased growth and biomass in arsenic-contaminated soils by increasing microbial activity while decreasing arsenic bioavailability. FYM delivers organic ligands that bind to arsenic and convert it to less harmful forms, all while boosting soil nutrient content and aeration.

The Relative Germination Rate of *P. sativum* was significantly lower in arsenic-treated soil compared to FYM-amended soil. At the control (0 ppm), both treatments exhibited an RGR of one, indicating no difference in germination. However, when arsenic concentrations grew, the RGR in when-treated soil reduced dramatically. At 5 ppm, the RGR fell to 0.72, whereas in FYM-amended soil, it stayed slightly higher at 0.75. At 10 ppm, the RGR dropped to 0.57 in As-treated soil, versus 0.71 in FYM-treated soil. The greatest dramatic decrease came at 20 ppm, when the RGR was just 0.29 in As soil and 0.48 in FYM-amended soil. These results are consistent with earlier studies. Arsenic drastically lowered seed germination rates by decreasing water intake and enzymatic activity, which are required for early seed development (Shri *et al.* 2009). However, organic amendments like as FYM have been demonstrated to reduce this effect by enhancing soil structure, moisture retention, and microbial activity, resulting in improved germination (Gunes *et al.,* 2009).

The Tolerance Index data illustrate in Table 3 shows how FYM improves the stress tolerance of *P. sativum* to As. Under no As condition (0 ppm As), both As-only and FYM soils showed a Tolerance Index of 100 which indicates normal growth (control). However, with increasing As concentration, the Tolerance Index in As-only treatments decreased significantly—90.2 at 5 ppm, 68.2 at 10 ppm, and 53.6 at 20 ppm. On the other hand, FYM-treated soils showed better tolerance with 102.2, 84.4, and 66.6 at the same As concentrations, demonstrating that FYM not only reduces arsenic toxicity but also increases the ability of plants to withstand stress. These findings are consistent with those of (Chahal *et al.*, 2020), who found that FYM enhances plant tolerance to heavy metal stress by increasing soil fertility and microbial activity, which together assist preserve plant physiological systems under adverse conditions. FYM aids in arsenic immobilization by enhancing adsorption onto organic matter and stimulating microbial reactions that convert arsenic into less accessible forms. Furthermore, (Brunet *et al.* 2020) discovered that organic supplements like FYM can improve plant tolerance by improving antioxidant defenses and nutrient uptake efficiency, which are frequently affected by metal stress.

The Percent Phytotoxicity values point towards the protective role of FYM against arsenic-induced toxicity in *P. sativum*. At the control (0 ppm), there was no phytotoxicity in either treatment. But with increased arsenic concentration, phytotoxicity rose in both treatments. At 5 ppm, Percent Phytotoxicity was 9.75% in As-treated soil, but significantly lower at 2.22% in FYM-treated soil, which shows that FYM alleviated some of the arsenic toxic effects. At 10 ppm, phytotoxicity increased to 31.7% in As soil, whereas in FYM-amended soil, it was 15.55%. At the maximum dose (20 ppm), Percent Phytotoxicity rose to 46.3% in As-treated soil, whereas it increased sharply to 55.55% in FYM-amended soil. These outcomes indicate that although FYM lowers arsenic-induced phytotoxicity, the effect lessens at greater levels of arsenic, where phytotoxicity rises steeply in both the treated and amended soils. Overall, the use of FYM alleviated the adverse effects of arsenic on seedling development, enhancing germination, vigor, and resistance to arsenic stress. This indicates that organic amendments such as FYM can be a valuable approach to diminishing the toxicity of heavy metals in soils, as also indicated by other similar studies (Prasad *et al*., 2015; Hussain *et al*., 2021; Kwiatkowska *et al.,* 2018).

Co-relation matrix between different Growth Indices of *Pisum sativum* with different concentrations of As (0,5,10,20 ppm) is shown in Table 4. The germination index and vigour index exhibits a strong positive correlation (0.817, $P \leq 0.05$); germination index and relative germination rate shows a moderate positive correlation (0.660, $P \leq 0.05$); vigour index and relative germination rate are strongly positively correlated (0.967, $P \leq 0.05$); tolerance index and percent phytotoxicity: They show a perfect negative correlation (-1.00, $P \leq 0.05$)at different concentrations of As 0, 5, 10, 20ppm respectively.

Table 4. Co-relation matrix between different Growth Indices of *Pisum sativum* with different concentrations of As (0,5,10,20 ppm)

Germination Indices	Germination Index	Vigour Index	Relative Germination Rate	Tolerance Index	Percent Phytotoxicity
Germination Index	1				
Vigour Index	0.817**	1			
Relative Germination Rate	0.660**	0.967**	1		
Tolerance Index	0.746**	0.990**	0.971**	1	
Percent Phytotoxicity	-0.747**	-0.990**	-0.970**	-1.000	1

**Correlation is significant at the 0.05 level

Co-relation matrix between different Growth Indices of *Pisum sativum* with different concentrations of As +FYM (0,5,10,20 ppm) is shown in Table 5. The germination index shows high positive correlations with both vigour index (0.970, $P \leq 0.05$) and relative germination rate (0.971 $P \leq 0.05$);vigour index and relative germination rate demonstrate a perfect positive correlation (1.0, $P \leq 0.05$). the tolerance index shows strong positive correlations with vigour index (0.851, $P \leq 0.05$); relative germination rate (0.848, $P \leq 0.05$), and a slightly weaker correlation with germination index (0.889, $P \leq 0.05$);percent phytotoxicity exhibits strong negative correlations with tolerance index (-0.980, $P \leq 0.01$), vigour index (-0.857, $P \leq 0.01$), and relative germination rate (-0.853, $P \leq 0.01$).

Table 5. Co-relation matrix between different Growth Indices of *Pisum sativum* with different concentrations of As +FYM (0,5,10,20 ppm)

Germination Indices	Germination Index	Vigour Index	Relative Germination Rate	Tolerance Index	Percent Phytotoxicity
Germination Index	1				
Vigour Index	0.970**	1			
Relative Germination Rate	0.971**	1.000	1		
Tolerance Index	0.889**	0.851**	0.848**	1	
Percent Phytotoxicity	-0.849**	-0.857**	-0.853**	-0.980**	1

**Correlation is significant at the 0.05 level

4. Conclusion

The present study demonstrates that farmyard manure plays a significant role in mitigating the adverse effects of arsenic toxicity on *Pisum sativum.* Arsenic exposure, particularly at higher concentrations markedly reduced seed germination, root and shoot growth, chlorophyll content and various growth indices, highlighting its detrimental impact on plant development. However, the incorporation of FYM substantially improved these parameters across all arsenic treatments. FYM not only enhanced soil fertility and reduced arsenic bioavailability but also boosted the plants physiological tolerance and metabolic responses to arsenic stress. These findings underscore the potential of FYM as a sustainable and environmentally friendly soil amendment for promoting healthy crop growth in arsenic contaminated soils. Thus, the application of FYM could be a practical strategy for improving food security and agricultural productivity in arsenic affected regions.

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Conflict Interest: The author declare that they have no conflict of interest.

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