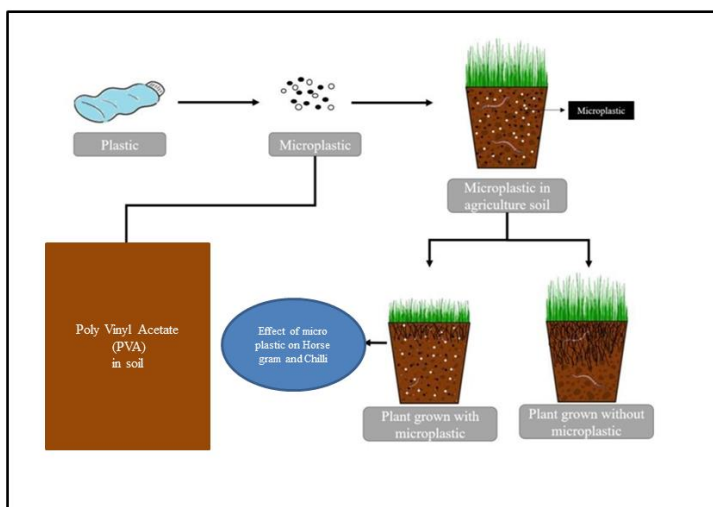


INVESTIGATING THE IMPACTS OF MICROPLASTICS ON THE PLANTS GROWTH IN AGRICULTURE SOIL

ABSTRACT

MP in the environment thrive for a long time and enter the ecosystem in vast amounts every day, microplastics pose a serious threat to the entire planet at the moment. The purpose of this study was to ascertain whether MP contamination affects plant development, the physical and chemical qualities of the soil, and soil metabolism. Several research databases about the identification and consequences of MP pollution of soil are available to evaluate its sources, impacts, mitigation, and global perspective. The effects of artificially spiked Poly Vinyl Acetate (PVA) micro plastics soil at different concentrations (0%, 0.5%, 1 %, 1.5%, 2%) on the growth parameters and nutrient contents of horse gram, green gram and chili grown soil. The germination percentage, seedlings length and vigor index was also reduced due to the presence of PVA particles which may be suspected to hinder the growth of the seeds, also the Boron and the Sulphur concentrations showed drastic reduction at higher concentrations of PVA micro plastics in green gram, horse gram and chili grown soil. Along with he reduced pH and Chlorophyll a due to the presence of PVA particles. Hence this study can help to focus on future need of remediation of micro plastics contaminated soil.

Keywords: Micro plastics, Poly Vinyl Acetate, Green gram, Horse gram and Chilli.



Graphical abstract

INTRODUCTION:

Plastic is ambidextrous by nature, which makes it extremely advantageous to manufacture goods with economic value. The world's output of plastics has increased dramatically over the past 50 years, especially since the first large-scale manufacture of synthetic plastic (Bakelite) in the 1940s and 1950s [1, 2]. In actuality, 367 million metric tonnes (Mt) of plastics were created globally in 2020 [3]. Additionally, it has been calculated that between 1950 and 2020, more than 8000 Mt of plastic garbage were produced [1,4], and that 4900 Mt of the total amount of plastic ever manufactured will end up in landfills or the environment. Due to decades of plastic manufacture and use in almost every industry, micro plastics smaller than 5 mm, are becoming more and more common in the environment. These micro plastics can worsen human health when exposed to them, as they are capable of adsorbing hazardous substances. They are so light that they may be readily flung by themselves, and many aquatic creatures eat them because they mimic food [17]. Microplastics (MP) are divided into two categories based on their source of production i.e. primary micro plastics (made for industrial uses), and secondary micro plastics (produced by degradation of larger plastics overtime).

MPs have been found to enter agricultural soils by major agricultural methods such irrigation and organic fertilisation using compost made from municipal solid wastes, manure, or sewage sludge [6,13,14]. In contrast to sewage sludge, which can contain MPs from laundry, personal care products, and urban runoff, home waste compost can contain both plastic debris from packaging materials and food waste that has been contaminated with MPs [8-10]. Films from coated seeds and plastic coatings from slow-release fertilisers have also been found to be possible sources of MPs in agricultural soils [11,12]. Plants have the ability to alter the distribution of MPs in the soil profile at the root level. MPs can change soil structure when they are incorporated into the soil matrix [25]. MP's harm plants in a number of ways, including i) blocking cell wall pores or cell connections, which prevent plant nutrients from being absorbed and transported; (ii) reducing or delaying seed germination by preventing water absorption; (iii) altering root and shoot growth [15,16]; and (iv) disrupting the balance of plant chlorophyll a/b ratios [17]. MP's are more likely to encourage the phytotoxicity of other soil pollutants when they are present near plant roots. The shape, size, type, and concentration of MP's [15,18-20], as well as the kind of soil [14,21], all affect how much of an impact they have on the soil-plant system.

The agricultural systems are high receivers of MPs is majorly owing to soil management practices, and also due to atmospheric deposition [7]. Ironically, a variety of agricultural techniques used to boost crop yields and preserve soil quality, moisture, and nutritional status may have a negative impact on soil health due to MP build up [5,7]. MPs have been found to enter agricultural soils by major agricultural methods such irrigation and organic fertilisation using compost made from municipal solid wastes, manure, or sewage sludge [6,13,14]. These MP particles increases the soil porosity and hence allowing less dense MPs to float when the pores are filled with water, plants with mostly primary and secondary roots push MPs closer to the soil surface, whereas plants with mostly tertiary roots keep MPs deeper in the soil layers [14,15]. There are a multitude of different toxins in plastics, according to regulatory authorities like the European Commission. They can ultimately accumulate in body, disrupt cellular signalling and metabolic activities, induce oxidative stress in cells and are also neuro toxic in nature.

The ecosystem of the soil is altered by MP contamination, which significantly affects plant life. Thus, the purpose of this review was to investigate how MP affects plants in terrestrial environments. Plant root systems, germination ability, photosynthesis, and the quality of plant products are all significantly impacted by MP contamination. Plants are severely harmed by cytotoxicity and oxidative damage, both of which are made easier by MP. The sources and buildup of MP in agro ecosystems and the natural world are also covered in this study. This work will eventually contribute to a better understanding of terrestrial ecosystems MP contamination. There has been discussion of terrestrial MP by certain studies [10]. Thus, our following study focuses on examining the effects of micro plastics on the growth of plants and its effects on the nutrient content of the soil. To understand the impact of MP on plant performance, all pertinent data was gathered for this article. It appears that more research is required to fully understand the impact of MP on terrestrial plants. This paper's goal was to examine plant behaviour in the presence of MP contamination and offer insights that should enhance agricultural output. Policymakers implementing measures to reduce MP contamination will benefit from this study. Micro plastics also tend to accumulate in food chain leading to the process of bio magnification.

2. MATERIALS AND METHODS

2.1 Study Area and characterization of experimental soil

The experimental soil sample was collected from an agricultural land in Iledu village Chengalpettu district, India (12°.04'63.05"N, 79°.84'84.95"E). Soil samples were collected and dried for 2–3 days, ground, sieved (2 mm sieve), packed in polyethylene cover, and were used for further analysis. The soil was shade dried, sieved to get rid of plant residues, large rocks and gravel and digested using 30 % Hydrogen peroxide to remove the organic matter. pH was measured with a pH meter [26], Electrical Conductivity (EC) with a conductivity meter [26], organic carbon with a wet digestion method [27]. NPK analysis was performed by using Diacid extract (5:2- H₂SO₄ : HClO₄)- semi automatic Kjeldahl distillation method and (Piper 1966) Triacid extract (9:2:1- HNO₃: H₂SO₄ : HClO₄) Vanadomolybdate yellow colour method [26]. The estimation of Available Sulphur and Exchangable Calcium and Magnesium was performed using the methods: 0.15 per cent CaCl₂-Turbidimetry method [28], Neutral normal ammonium extract- Versenate titration method respectively.

Experimental set up

The polyvinyl acetate microplastics (PVA-MPs) powder used in the study was obtained from online E commerce market. To reduce microbiological contamination, the MP's were microwaved at 0.8 KW for 2 minutes [29]. MP's were added to the farm soil after 3 days of processing the soil at five different concentrations (T₁- 0, T₂ - 0.5, T₃- 1.00, T₄- 1.50 and T₅- 2.00% on w/w basis). The concentrations were fixed based on the previous studies that quantified MP's in different soils [30]. Mixing of MP's with soil was done by stirring with ethanol sterilized glass rod in a glass beaker. The Green gram, Horse gram and Chilli seeds were bought from the local market.

Treatment details

The test soils (200g) with different MP's concentrations were transferred to clean containers. The seedlings were maintained for duration of 15 days after sowing. To assess the influence of soil types in the presence of PVA-MPs in Green gram, Horse gram and Chilli seedling growth, the critical parameters like germination rate, germination time, root length, shoot length, vigour index, and chlorophyll content were recorded after 15 days.

Evaluation of chlorophyll content

Leaves were collected separately from seedlings grown in control and micro plastics added soil. 0.1g leaf tissue was cleaned. The leaf tissues were placed in a glass beaker with 8ml of acetone and 2ml of ethanol for 18 hours in darkness. After 18 hours the chlorophyll a and chlorophyll b was analysed at 665nm and 649nm respectively using UV spectrophotometer and absorbance values were noted as mentioned in [31] and the chlorophyll content was estimated as per the equation:

$$C_a = 13.95A_{665} - 6.88A_{649}$$

$$C_b = 24.96A_{649} - 7.32A_{665}$$

3. RESULTS AND DISCUSSION

3.1 Characteristics of the Initial soil

The physico-chemical characterization was carried out as per the standard procedures and the results obtained were given in Table 1. The water holding capacity of the soil was found to be 51%. The pH and EC of the experimental soil was found to be 7.28, 0.42 mS/cm. The value of organic carbon was found to be 1.29 in the initial soil. The NPK concentrations were found to be 61.89 mg/kg, 28.9 mg/kg and 389 mg/kg in the initial soil. The other parameters such as Boron, Calcium and Zinc were found to be 0.4, 2201, 1.21. MP contamination of soil organic carbon (SOC), a key indicator of soil quality and the global carbon cycle. The effects of MPs on heavy metals have the potential to alter the chemical speciation of heavy metals as well as the physicochemical characteristics of soil, including pH, organic matter (OM), bulk density, water holding capacity, and enzyme content. By altering the cycle and concentration of accessible C, N, and P in the soil, they can even have an impact on plant growth [42].

3.2 Characteristics of growth parameters of the seeds

The presence of PVA-MPs influenced the germination of the Green gram seedlings. The germination percentage of Green gram reduced from 90% (Control T₁ – No presence of PVA-MPs) to 30% (T₅ – 2% PVA-MPs). The total seedling length decreased from 31cm (Control T₁ - 0% PVA-MPs) to 26.6cm (T₅ – 2% PVA-MPs). The germination percentage of Horse gram reduced from 100% (Control T₁ - 0% PVA-MPs) to 40% (T₅ – 2% PVA-MPs). The total seedling length decreased from 18.2 cm (Control T₁ - 0% PVA-MPs) to 10.4 cm (T₅ – 2% PVA-MPs). The reduction of germination percentage of chilli was seen from 90% (Control T₁ - 0% PVA-MPs) to 10% (T₅ – 2% PVA-MPs). The total seedling length

decreased from 11.9 cm (Control T₁ - 0% PVA-MPs) to 8.2 cm (T₅ - 2% PVA-MPs). MP's may physically or chemically interfere with seed imbibition or water uptake, which would reduce germination rates [32]. Many plants, including rape, cucumbers, rice, and others, are negatively impacted by microplastics. Polystyrene microplastics inhibited metabolism and degraded the quality of rice [42]. As the amount of MP's in the soil increased, it was seen that the vigour index gradually declined. The vigour index of Green gram, Horse gram and Chilli were found to be the lowest with the values of 798, 416, 82 (T₅ - 2% PVA-MPs). Previous studies have stated that the MP's lead to lower biomass in maize [38], duckweed [39], plantain [40], broad bean [39], and cress [33]. Moreover, MPs significantly inhibit the growth and procreation of soil fauna, including earthworms, springtails, and nematodes. MPs may be harmful to the soil biota if contamination changes the ecology of the soil [42].

3.3 . Electrical conductivity and pH characteristics of the soil

The EC values of the Green gram, Horse gram and Chilli grown in PVA micro plastics spiked soil were not much affected by the addition of PVA micro plastics. The pH values significantly reduced from 6.85 to 6.10 in green gram grown soil. The pH values significantly reduced from 6.83 to 6.52 in horse gram grown soil. The pH values drastically reduced from 6.87 to 5.72 in Chilli. These results collectively indicated that the micro plastics decrease the pH values of soil up to a greater extent but do not have significant impact on the electric conductivity of the soil. The impact of PE on soil pH from a study suggested that HDPE may also cause an increase in soil pH. As an example, low density polyethylene (LDPE) films may increase soil pH whereas, high density polyethylene (HDPE), may have the opposite pattern [30]. MP sorption in soils was influenced by pH, and as pH increased, the sorption capacity reduced. The physicochemical properties of the soil have a considerable impact on the sorption of polystyrene microplastic particles (PSMPs) [43].

3.2 Chlorophyll content of the seedlings

The chlorophyll content in the leaves of green gram didn't vary much significantly and more over less similar values were obtained as mentioned in table 4. The chlorophyll a and b values decreased at the concentration of 2% of PVA micro plastics where the concentrations were found to be 3.9121 and 1.6815 in Horse gram leaves as mentioned in table 4. In Chilli leaves, the concentration of chlorophyll b decreased with value of 1.998. Similar study, on lettuce (*Lactuca sativa*) revealed that plants treated with MP had lower levels of chlorophyll and some photosynthetic indices [43]. These results indicate that PVA did not have any impact on the chlorophyll content of green gram leaves and the chlorophyll content got reduced at higher concentration that is 2% of PVA in horse gram and the chlorophyll b value of Chilli got

reduced at 2% PVA concentration. However, 50 nm red fluorescent nanoplastics at a concentration of 10^4 particles/mL did not statistically significantly affect the amount of chlorophyll b in duckweed (*Spirodela polyrhiza*) in an experiment [43].

3.3. Sulphur content present in soil

The concentration of sulphur decreased drastically from 22.8 mg/L to 9.4 mg/L in Green gram grown soil especially from 1% concentration of micro plastics as mentioned in table 5 (a) and was significantly reduced from 29.2 mg/L to 20.0 mg/L in horse gram grown soil as mentioned in table 5 (a). The concentration of sulphur didn't vary at lower concentrations of PVA but reduced drastically at 1.5%, 2% of micro plastics. The concentration of sulphur didn't significantly vary in Chilli grown soil as mentioned in table 5 and hence it can be concluded that micro plastics affected the concentration of sulphur in green and horse gram grown soil and it can be assumed that sulphur had got accumulated along with micro plastics making it unavailable to the seedlings as well as the soil. Similar results were reported in previous studies were 1% unplastitized PVC MP's significantly reduced available Phosphorus content from 38.4 ± 2.5 mg/kg to 26.9 ± 1.3 mg/kg ($p < 0.05$) (Chen et al., 2021). MPs can also affect the soil nutrient cycle by changing organic matter and available nutrients. MPs age as a result of long-term enrichment and emission migration, changing in size, shape, and surface characteristics. Additionally, MPs have the ability to adsorb or desorb additional contaminants during these processes, such as organic pollutants and toxic chemicals, which hurt the soil organisms [44].

CONCLUSION

Growing concerns about environmental pollution in general and MPs contamination in particular call for a continuous measurement and redefining of plastic pollution, scientific understanding has advanced dramatically in the last several years. We have covered several kinds of MP - plant interactions in this essay. Therefore, it is important to research how each polymer behaves in various soil-plant systems. The study conducted by growing Green gram, Horse gram, Chilli in PVA micro plastics spiked soil concludes that the nutrient concentration of the soil especially sulphur, varies significantly. The sulphur concentration along with Chlorophyll a showed drastic reduction at higher concentrations of PVA micro plastics in Green gram, Horse gram, Chilli. Hence, the study given an insight on the effects of MP's on the growth on plants. In this instance, an attempt has been made to highlight the MP contamination, its destiny during transportation, and the actions done to eliminate it from the standpoint of solid waste. Furthermore, standardising and combining the analysis techniques is essential for further research. Improved coordination of research and understanding

regarding the possible hazards of MP their detrimental impacts, and their origins in contemporary agriculture can lead to potential remedies. Therefore, more research is required to continuously assess the impact of MP contamination on a variety of previously unexplored soil health indices.

Disclaimer (Artificial intelligence)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of manuscripts.

UNDER PEER REVIEW

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Table 1: Initial characteristics of the soil

Parameters	Unit	Value
pH	-	7.28
EC	mS/cm	0.42
Organic Carbon	%	1.29
Boron	mg/kg	0.4
Available phosphorus	mg/kg	28.9
Available sulphur	mg/kg	38.01
Exchangeable potassium	mg/kg	389
Exchangeable calcium	mg/kg	2201
Exchangeable magnesium	mg/kg	621
Available Iron	mg/kg	3.89
Available copper	mg/kg	3.12
Nitrate	mg/kg	61.89
Available Zinc	mg/kg	1.21

Table 2: Growth parameters of seedlings

	Spiked Concentration Of MPs	Total seedling Length (In cm)	Germination Percentage (In %)	Vigour index
Green gram	Control	31	90	2790
	0.5%	30.5	70	2135
	1%	29.7	60	1782
	1.5%	26.8	50	1340
	2%	26.6	30	798
Horse gram	Control	18.2	100	1820
	0.5%	18.0	80	1440
	1%	17.2	80	1376
	1.5%	15.5	50	775
	2%	10.4	40	416
Chilli	Control	11.9	90	1071
	0.5%	10.6	50	530
	1%	11.7	30	351
	1.5%	7.9	20	158
	2%	8.2	10	82

Table 3: electrical conductivity and pH of soil

	Spiked Concentration Of MPs	EC	pH
Green gram	Control	0.108	6.85
	0.5%	0.103	6.72
	1%	0.097	6.65
	1.5%	0.090	6.64
	2%	0.083	6.10
Horse gram	Control	0.068	6.83
	0.5%	0.057	6.72
	1%	0.057	6.62
	1.5%	0.053	6.58
	2%	0.051	6.52
Chilli	Control	0.072	6.87
	0.5%	0.061	6.72
	1%	0.057	6.48
	1.5%	0.051	6.40
	2%	0.050	5.72

Table 4 Effect of PVA-MPs at concentrations 1.5% and 2 % on chlorophyll content of leaves

Concentration of PVA micro plastics						
	Control		1.5%		2%	
	Chlorophyll a	Chlorophyll b	Chlorophyll a	Chlorophyll b	Chlorophyll a	Chlorophyll b
Green gram	4.2321	2.2678	4.2034	2.1976	4.1983	2.1956
Horse gram	4.1863	2.2798	4.1862	2.1743	3.9121	1.6815
Chilli	4.2567	2.3765	4.2489	2.264	4.2287	1.9987

Table 5. Sulphur content present in soil after the addition of microplastics

	Green gram	Horse gram	Chilli
Control	22.8	29.2	29.2
0.5%	18.5	26.8	29.0
1%	14.6	22.8	28.1
1.5%	14.5	20.9	28.0
2%	9.4	20.0	28.0

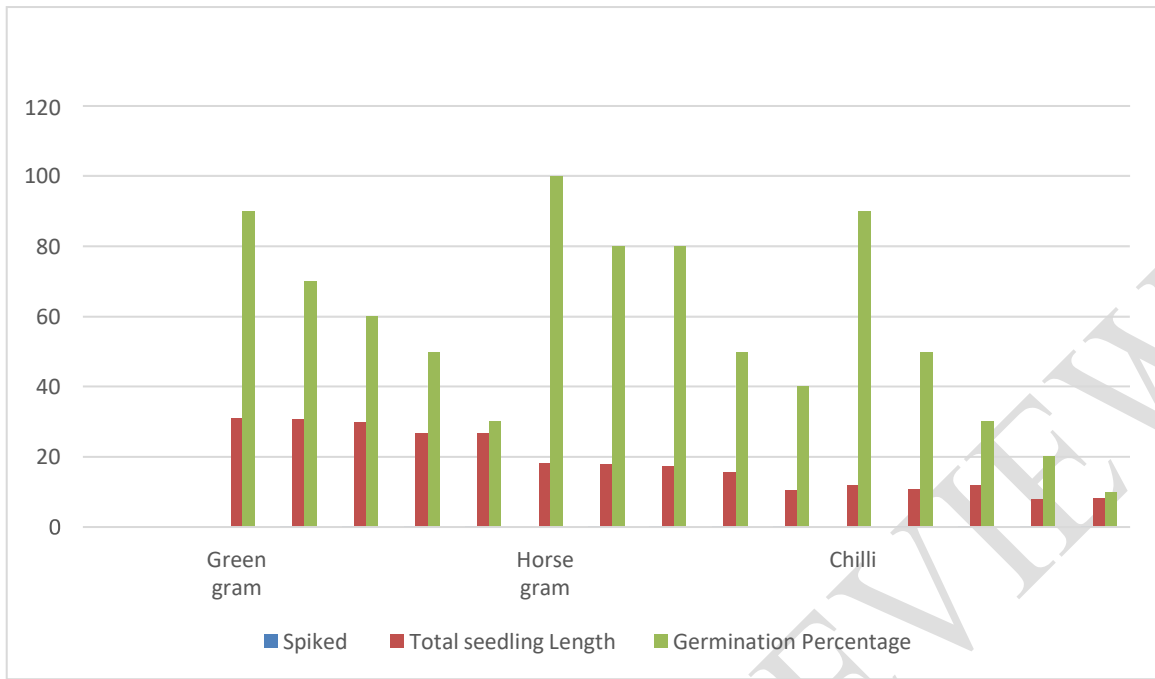


Figure 1: Effects of microplastics on growth of seedling

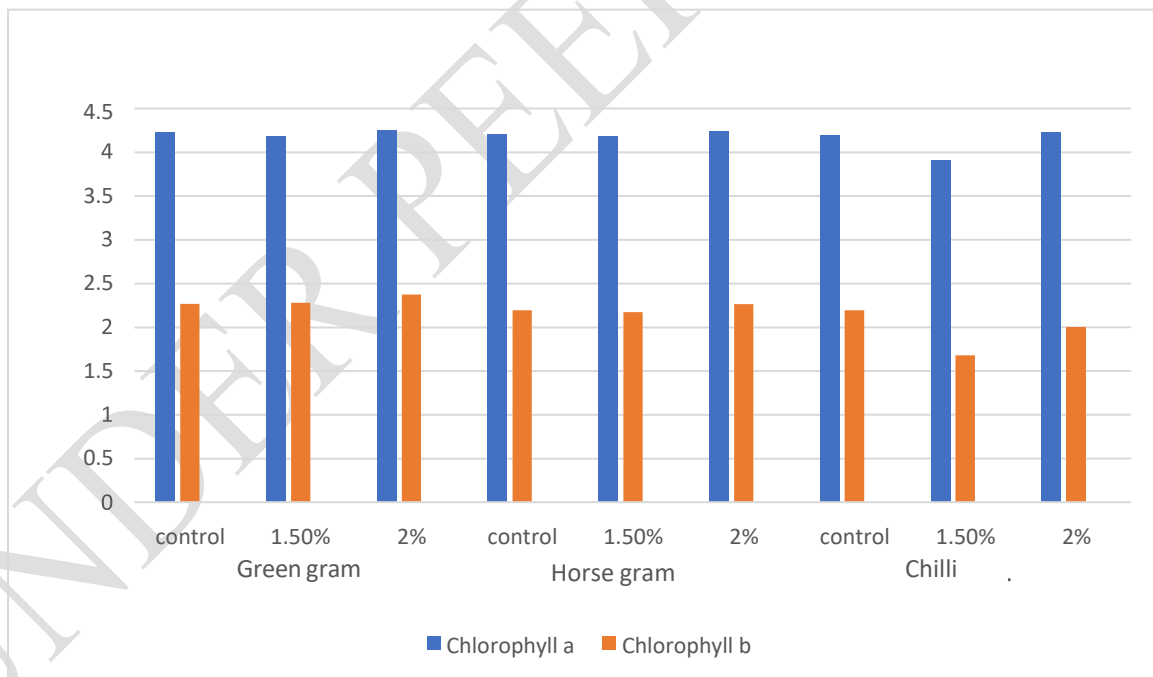


Figure 2: Graphical representation of PVA MPs affecting the chlorophyll content in leaves

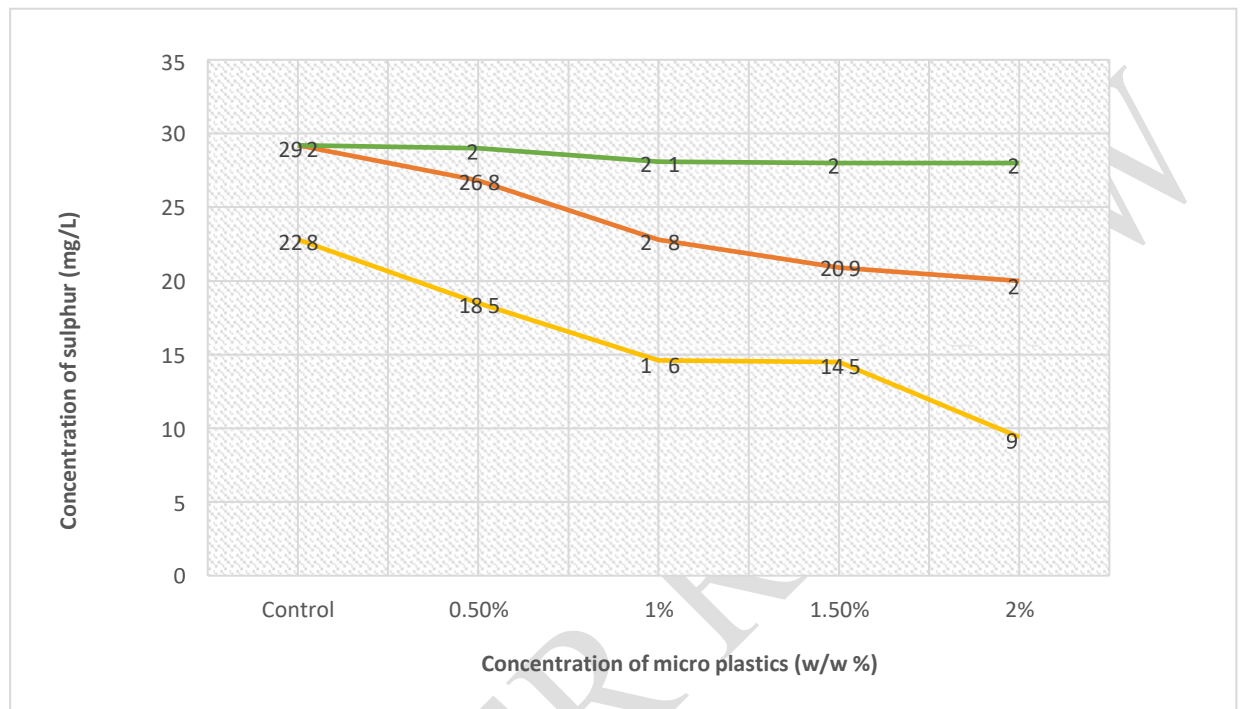


Figure 3: Graphical representation of PVA MPs affecting the sulphur content in soil (yellow-green gram, orange-horse gram, green- chilli)