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Effect of guar-gum grafted clay/pomace-based composites loaded with iron and zinc on Thompson seedless grapes (*Vitis vinifera* L.)

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Abstract
Use of clay-polymer composites is the new avenue of research in the field of agriculture due to their role in controlled release of nutrients synchronizing with crop demand. A 28 days laboratory incubation experiment was conducted to study the release pattern of Fe and Zn from clay/pomace-based composites. The DTPA-Fe was 16.18 ppm at 0 day of incubation and decreased to 11.65 ppm after 28 days of incubation in case of Fe-EDTA fertilizer whereas the DTPA-Fe was 15.05 ppm at the start of incubation and 16.12 ppm after 28 days of incubation in case of pomace-based composite loaded with Fe. Similarly, the DTPA-Zn was 8.49 ppm at 0 day of incubation and decreased to 5.06 ppm after 28 days of incubation in case of Zn-EDTA fertilizer whereas the DTPA-Zn in case of composite loaded with Zn was 5.75 ppm at the start of incubation and 9.65 ppm after 28 days of incubation. Application of polymer composites loaded with Fe and Zn resulted in an increase in Fe and Zn content in grapes. Iron content in grapes increased by 10.24% after application of Guar-gum grafted Pomace-Polymer composite as compared to conventional iron fertilizer. However, the zinc content in grapes increased by 8.41% after application of Guar-gum grafted Clay + Pomace-Polymer composite as compared to conventional zincatic fertilizer. A significant increase in Fe and Zn uptake was also recorded in case of polymer composite treated soil. Hence, the use of Fe and Zn loaded clay/pomace-based composites emerged as a promising option for increasing Fe and Zn content in grapes.

Keywords: Composites, iron, zinc, guar-gum, grapes

Introduction
Among the various constraints in Indian Viticulture, calcareous nature of soil is one of the most important factors which adversely affect the production of quality grapes. The calcareous nature of soil due to high pH often accentuates micronutrients deficiency. Micronutrients are crucial for enhancing crop yields as well as improving the nutritional value of food since they are vital components of proteins and enzymes. Among the micronutrients, iron (Fe) and zinc (Zn) are an essential component of many enzymes involved in the metabolism of grapes (Brataševec *et al*., 2013) [3]. The only metal found in all six enzyme classes and the second-most prevalent transition metal in organisms is zinc (Zn) (Drenjanˇcevi´c, 2012) [7]. In addition to its structural and metabolic significance at the cellular level, Zn is a fundamental trace element and one of the most prevalent in the human body (Li *et al*., 2017; Cataldo *et al*., 2020) [11, 5]. Iron increases photosynthesis and carbohydrate synthesis in grapes whereas, Zn is essential for normal leaf development, shoot elongation, pollen development, and the set of fully developed berries**.**
The majority of micronutrients used nowadays are chelated salts that are water soluble, namely sulphates or their analogues (EDTA, DTPA, etc.). Soil application of Fe and Zn fertilizers (dominantly $ZnSO_4.7H_2O$ and $FeSO_4.7H_2O$), due to their high solubility leads to fixation by soil constituents, runoff and leaching, high dosage requirements, which results in severe wastage and heavy economic loss of these nutrients. As a result, the agronomic efficiency of applied fertilizer rarely exceeds 1-5%. Iron and Zn deficiency is most widespread deficiency not only in India but also in world. One third of the arable land on this planet has Fe deficiency (Mori, 1999; Li *et al*., 2017) [14, 11]. The e-atlas on micronutrients shows deficiency of Zn in Maharashtra (major grape growing area) - Nasik 12.7%, Sangli -32.9%, Solapur - 60.7%, Pune - 0.9% and Fe - Nasik 27.4%, Sangli -27.6%, Solapur - 63.5%, Pune - 6.8%. Lime induced chlorosis, is a common term used to describe Fe deficiency in calcium carbonate containing soils. It is a significant issue in viticulture that has an immediate impact on both grape output and quality. Younger leaves with intervascular chlorosis, in which the leaf ribature stays green, are a sign of Fe deficiency in grapevines, which is followed by marginal necrosis and leaf fall (Drenjanˇcevi´c, 2012) [7].

Grape is quite sensitive to Zn deficiency also [3]. Chlorosis, necrotic patches, plant contraction, and small leaves are all obvious signs of Zn deficiency in grapevines [3, 6].

For improved sustainability of grapes, vineyard soil management is crucial (Cataldo *et al*., 2020) [5]. The basic role of soil is to provide enough water and nutrients to the plants during the growing season in addition to providing the space for root growth and development. The pH of the soil is the main determinant of the availability of Fe, Zn and other microelements in the soil (Ivezi´c *et al*., 2013) [9]. The main cause of the low Fe supply is the intensive synthesis of poorly soluble iron compounds in carbonate soils, such as Fe $(OH)_2^+$, Fe $(OH)_3$ and Fe $(OH)_4$. The pH induced Fe and Zn deficiencies in calcareous soil occurs due to precipitation of Fe and Zn as insoluble amorphous soil Fe and soil Zn and/or $FeSiO_4$ and $ZnSiO_4$ and, which reduces available Fe and Zn in the soil. In addition to the pH reaction of the soil (Tagliavini and Rombolà, 2001) [17]. the presence of carbonates ($CaCO_3$) in the soil, low soil aeration, soil compaction, coarse texture, leaching low temperature in the root zone, poor biological properties and inadequate soil nitrogen status are also important factors controlling the supply of microelements and often increases the Fe and Zn deficiency. Iron and Zn availability to plant is interfered by different factors like high soil pH, adsorption by negatively charged clay-colloids, presence of other competitive cations like Cu^{2+}, anions like PO_4^{3-}, SO_4^{2-} etc., low pCO_2 and adsorption on the surface of precipitated $CaCO_3$.

The use of agronomic practices to raise the amount of a nutrient in the edible part of food crops, via application of fertilizers through soil or foliar spraying, is gaining attention in order to combat nutrient deficiencies in the human diet. (Cakmak, 2008; Hussain *et al*., 2022) [4, 8]. Iron and Zn fertilisation is one of effective strategies that is efficient in the short term (Cakmak, 2008) [4], but its uptake is highly dependent on the soil's pH, organic matter content, antagonistic cations (particularly in calcareous soils), and the type of Fe and Zn complex. Therefore, a supply system which can minimize their loss and can supply Fe and Zn to the crop throughout the growth period synchronizing its demand maintaining higher crop yield can be a promising option in reducing Fe and Zn losses. (Bandopadhyay *et al*., 2008) [1]. Consequently, it is believed that low water-soluble slow-release fertilizers are the solution. Clay-polymer composites (CPCs) are a reaction product of clay and polymer and exhibit promising properties to be used as superabsorbent and slow-release fertilizer systems (Cinnamuthu and Boopathi, 2009) [6]. Clay-polymer composites make the loaded fertilizer nutrients available for a long duration as compared to the conventional fertilizers with high solubility, which release their nutrients rapidly. To enrich grapes with iron and zinc, slow-release fertilizers were developed using guar-gum and grape pomace and bentonite clay as a filler. The efficacy of these products was examined in laboratory incubation study and pot culture experiment.

Materials and Methods

Clay/ pomace-based composites were synthesized using guar-gum, bentonite clay, and pomace as a filler. The method of Liang and Liu (2007) [10] was followed to synthesize composites. The synthesized products were subsequently loaded with Fe-EDTA and Zn-EDTA. A 28 days laboratory incubation experiment was conducted to study the release pattern of Fe and Zn from clay/ pomace based composites. Fe-EDTA and Zn-EDTA were added @ 0.8mg/50g soil, polymer composite loaded with Fe-EDTA @ 2.16mg/50g soil and clay-polymer composite loaded with Zn was added @ 2.2mg/50g soil. The soil available Fe and Zn were analysed through the method of Lindsay and Norvell (1978). Analysis of variance test in the completely randomized design (CRD) was applied (Snedecor and Cochran, 1980) [16].

Results and discussion:

Initially the Fe as well as Zn content was more in case of conventional fertilizers but polymer-composites loaded with Fe and Zn resulted in higher DTPA-Fe and DTPA-Zn over the period of soil incubation due to their slow-release property (Figure 1a and 1b). High solubility of Fe and Zn fertilizers in soil solution resulted high concentration and rapid fixation in soil. The DTPA-Fe was 16.18ppm at 0 day of incubation and decreased to 11.65 ppm after 28 days of incubation in case of Fe-EDTA fertilizer whereas the DTPA-Fe was 15.05ppm at the start of incubation and 16.12 ppm after 28 days of incubation in case of composite loaded with Fe (Figure 1a). Similarly, the DTPA-Zn was 8.49 ppm at 0 day of incubation and decreased to 5.06 ppm after 28 days of incubation in case of Zn-EDTA fertilizer whereas the DTPA-Zn in case of composite loaded with Zn was 5.75 ppm at the start of incubation and 9.65 ppm after 28 days of incubation (Figure 1b).

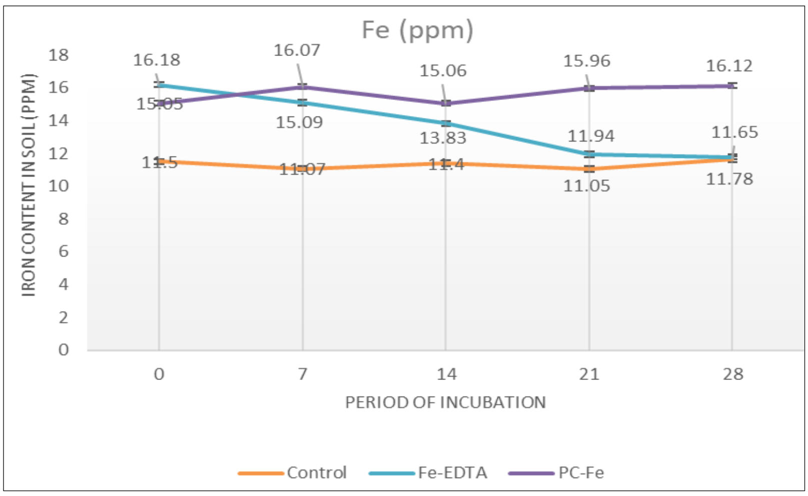

Fig 1a: Release pattern of Fe from polymer-composite loaded with Fe-EDTA and Fe-EDTA fertilizer

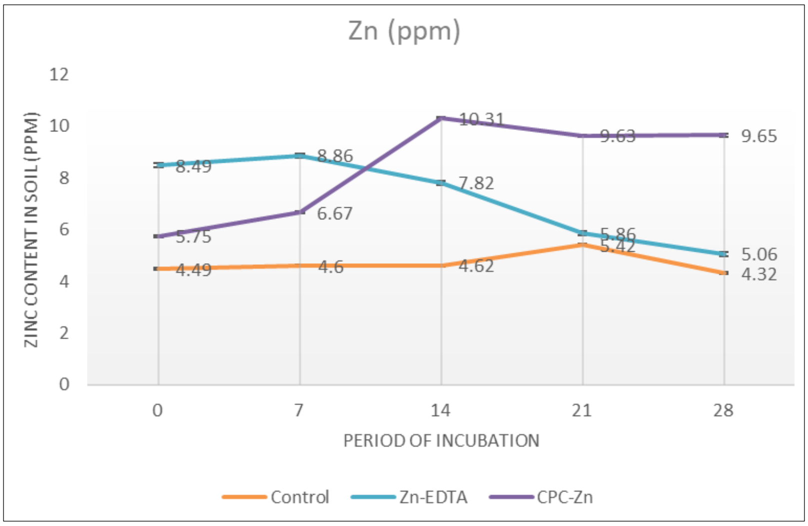

Fig 1b: Release pattern of Zn from clay-polymer composite loaded with Zn-EDTA and Zn-EDTA fertilizer

A 6 months pot culture experiment was conducted to study the effect of these slow-release fertilizers on Fe and Zn utilization by grapes. For conducting the pot experiment, Fe and Zn deficient soil was collected from MRDBS block MS1. (GPS location: N18°29.566'E073°59.355'). The pH of the soil was 9.29. The soil available Fe and Zn were 6.49 and 0.5ppm respectively. The soil was deficient in Zn. The EC of soil was 0.38 dSm^{-1}. The organic carbon was 0.26%, available N=72.8pp, available P=11ppm, available Na=4443.6ppm, available potassium=69.66ppm, available Ca=5457.8ppm, available Cu=1.42ppm and available Mn=4.24ppm.

Table 1: Effect of polymer-composite loaded with Fe-EDTA and Zn-EDTA fertilizer on growth parameters of grapes

Treatments	Plant height (cm)	No. of leaves	No. of nodes
T_1(Control)	80.85± 7.14 ^{a}	23.50 ± 3.09^a	16.33±1.11^a
T_2(Zn-EDTA)	88.33±8.03^a	27.00±2.46^a	21.00±1.37^{ab}
T_3 (CPC-Zn)	100.50±5.09^a	30.33±4.75^{ca}	23.16±3.07^b
T4 (Fe-EDTA)	97.00±10.64^a	26.14±2.53^a	22.85±1.92^b
T5 (PC-Fe)	104.00±5.10^a	30.33±3.71^a	23.66±1.89^b

Application of polymer composites loaded with Fe and Zn resulted in an increase in plant growth parameters (Table 1). Biomass yield when recorded was found to be lowest in case of control and highest in case of soils receiving polymer-composite loaded with Fe and Zn. It was due to lesser nutrient availability in control at critical growth stages where no Fe and Zn was applied while highest value in polymer- composites was attributed to higher nutrient availability by these slow-release fertilizers at critical growth stages of crop (Table 2 a and 2 b).

Table 2a: Effect of clay-polymer loaded with Zn-EDTA and Zn-EDTA fertilizer on dry matter yield of grapes

Treatments	Dry wt. (shoot)	Dry wt. (root)
T_1(Control)	22.83±0.36 ^{a}	20.93 ± 0.72^a
T_2 (Fe-EDTA)	25.19±0.41^b	24.36±0.71^b
T_3 (PC-Fe)	33.35±0.36^c	33.26±1.07^c

*PC-Fe= Polymer composite loaded with Fe-EDTA

Table 2b: Effect of polymer composite loaded with Fe-EDTA and Zn-EDTA fertilizer on dry matter yield of grapes

Treatments	Dry wt. (shoot)	Dry wt. (root)
T_1 (Control)	22.83±0.36 ^{a}	20.93 ± 0.72^a
T_2 (Zn-EDTA)	28.85±0.21^b	27.58±0.50^b
T_3 (CPC-Zn)	38.36±0.48^c	36.34±0.62^c

*CPC-Zn=Clay polymer composite loaded with Zn-EDTA

Application of polymer composites loaded with Fe and Zn resulted in more grapes Fe and Zn content. Iron content in grapes increased by 10.24% after application of Guar-gum grafted Pomace-Polymer composite as compared to conventional iron fertilizer. However, the zinc content in grapes increased by 8.41% after application of Guar-gum grafted Clay+Pomace-Polymer composite as compared to conventional zincatic fertilizer (Table 3 a and 3b).

Table 3a: Effect of polymer composite loaded with Fe-EDTA fertilizer on Fe content of grapes

Treatments	Fe content in leaves (ppm)	Fe content in stem (ppm)	Fe content in root (ppm)
T_1 (Control)	353.40±1.21^a	296.59±0.59^a	687.68±2.44^a
T_2 (Fe-EDTA)	558.29±0.61^b	320.58±0.92^b	965.33±0.99^b
T_3 (PC-Fe)	623.15±0.52^c	357.75±1.72^c	1074.47±1.83^c

Table 3b: Effect of clay-polymer composite loaded with Zn-EDTA fertilizer on Zn content of grapes

Treatments	Zn content in leaves (ppm)	Zn content in stem (ppm)	Zn content in root (ppm)
T_1(Control)	74.87±0.57 ^{a}	91.91±0.13^a	61.55±1.07^a
T_2(Zn-EDTA)	118.62±0.45^b	126.77±0.51^b	100.51±1.74^b
T_3 (CPC-Zn)	129.62±0.65^c	138.55±0.73^c	109.50±1.28^c

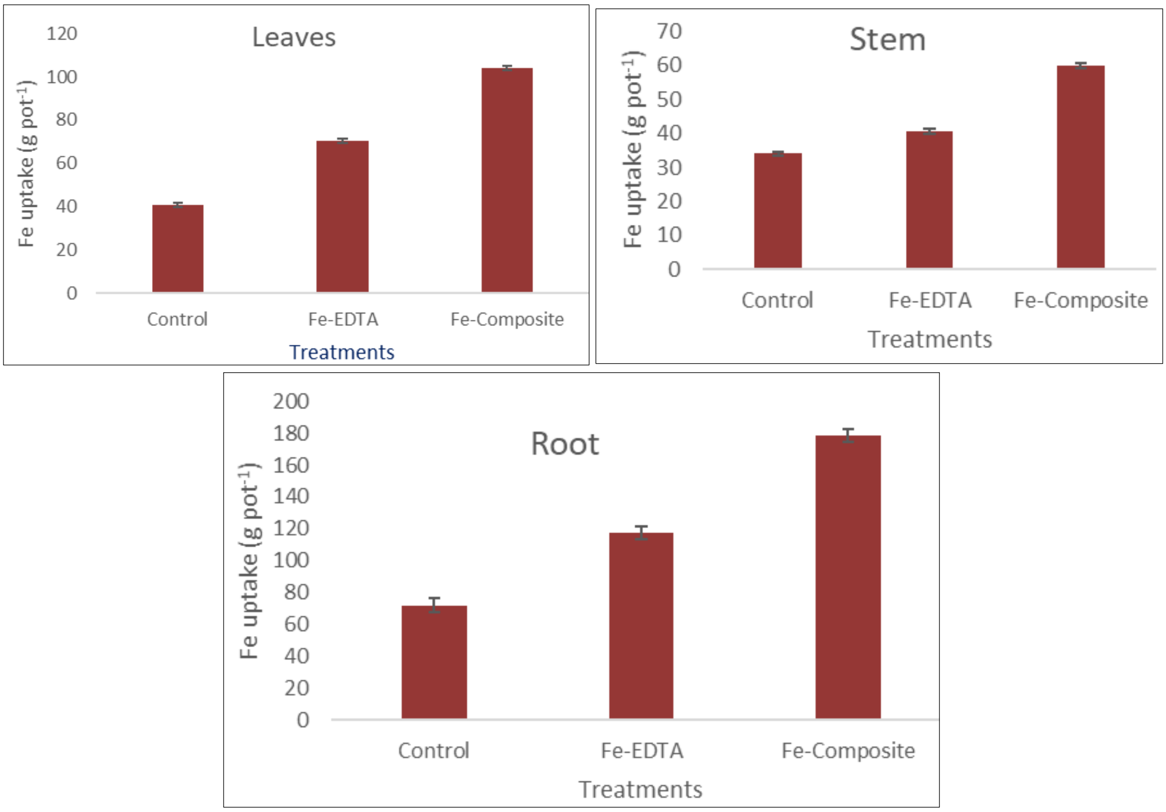

Fig 2: Effect of polymer composite loaded with Fe-EDTA fertilizer on Fe uptake by (a) leaves (b) stem, and (c) roots

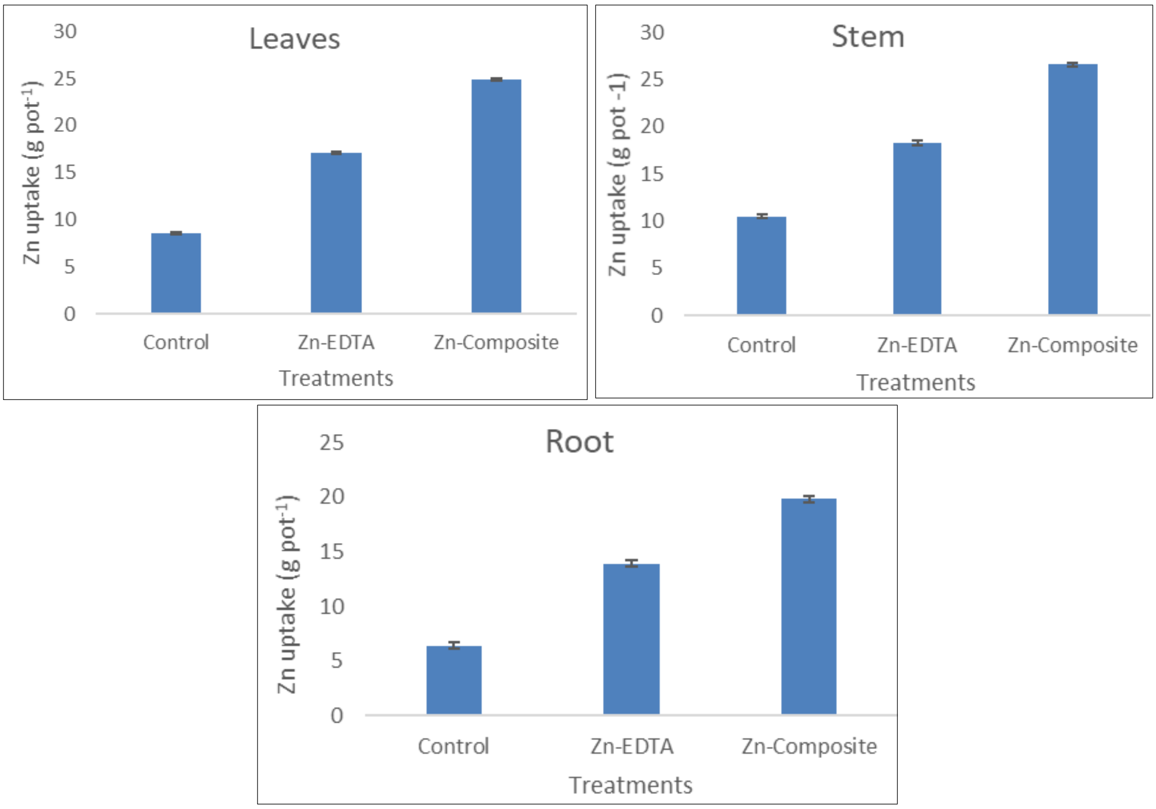

Fig 3: Effect of clay-polymer composite loaded with Zn-EDTA fertilizer on Zn uptake by (a) leaves (b) stem, and (c) root

A significant increase in Fe and Zn uptake was recorded in case of polymer-composites treated soil as compared to Fe-EDTA and Zn-EDTA. The increase was +32.39, 32.29 and 34.36% in leaves, stem and root when polymer-composite loaded with Fe-EDTA was applied to soil as compared to Fe-EDTA fertilizer. Similarly, the increase was +31.17, 31.23 and 29.85% in leaves, stem and root when clay-polymer composite loaded with Zn-EDTA was applied to soil as compared to Zn-EDTA fertilizer. It was attributed to more concentration of soil available Fe and Zn in case pf polymer-composites due to their low fixation in soil. The uptake rate of nutrient (Fe and Zn) is proportional to Fe and Zn concentration in soil solution near root surface (Figure 2 and 3). The results are in accordance with Sarkar *et al*. (2014) [15] and Mandal *et al*. (2014) [13].

Conclusion
The DTPA-Fe and DTPA-Zn was more in case of polymer composites as compared to Fe and Zn fertilizers due to the slow-release property of polymer-composites. Application of polymer-composites loaded with Fe and Zn resulted in more grapes Fe and Zn content. Iron content in grapes increased by 10.24% after application of guar-gum grafted pomace-polymer composite as compared to conventional Fe fertilizer whereas Zn content in grapes increased by 8.41% after application of guar-gum grafted clay + pomace-polymer composites as compared to conventional zincatic fertilizer. Higher biomass yield was recorded in polymer-composites due to higher nutrient availability by these slow-release fertilizers at critical growth stages of crop. An increase of +32.39, 32.29 and 34.36% in Fe (leaves, stem and root) and +31.17, 31.23 and 29.85% in Zn (leaves, stem and root) was recorded in case of polymer composite treated soil. Hence, the use of Fe and Zn loaded clay/pomace-based composites emerged as a promising option for increasing Fe and Zn content in grapes.

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