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FORTIFICATION OF IRON IN FINGER MILLET (*ELUSINE CORACANA* L.): IMPACT OF CONVENTIONAL AND NANO NPK FERTILIZERS COMBINED WITH INORGANIC AND ORGANIC IRON SOURCES IN AN ACIDIC SANDY LOAM SOIL

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Abstract

The purpose of the present work was to assess the effect of combined application of conventional and nano NPK fertilizer and different iron (Fe) sources on yield and Fe uptake towards iron fortification of finger millet (*Elusine coracana* L.). The experiment was carried out in a net house using factorial completely randomized design (FCRD). The factors consist of different Fe sources (Iron enriched biochar, iron enriched farmyard manure & ferrous sulphate), and different levels (75%, 100% &125 %) of conventional and nano NPK fertilizers application and replicated thrice. Results of the net house experiment revealed that the application of iron enriched biochar (FeEBC) with 125% of Nano NPK recorded higher grain (14.5 g pot^{-1}) and straw (36.7 g pot^{-1}) yield, Fe content (45.6 & 58.1 mg kg^{-1}) and uptake (1.78 & 2.30 mg pot^{-1}) in grain and straw, respectively. However, statistically similar results obtained by the application of Fe enriched biochar with 100% of Nano NPK on grain (13.6 g pot^{-1}) and straw (35.6 g pot^{-1}) yield, Fe content in grain (45.5 mg kg^{-1}) and straw (58.6 mg kg^{-1}) and Fe uptake in grain (1.73 mg pot^{-1}) and straw (2.28 mg pot^{-1}),respectively. So, considering the input reduction and higher net yield, nutrient content and uptake; the application of 100% Nano NPK with Fe enriched biochar (FeEBC) can be a significant and suitable form and proportion of nutrient source for Fe fortification of finger millet grown in acidic soil conditions. From this study we found that application of nano NPK fertilizer plus FeEBC can significantly fortify the iron content in finger millet grain up to~3.92 % over combined application of recommended dose of NPK conventional fertilizers and $FeSO_4$ to finger millet.

Keywords: Ferrous sulphate, finger millet, iron enriched biochar, iron fortification, nano NPK, yield

Article Highlights

- Iron enriched biochar (FeEBC) and Nano NPK supply the nutrient constantly from soil to crops.
- FeEBC changed the soil environment and slowly supply the nutrients to the crop.
- FeEBC reduce Fe losses, increase availability to crop through change the soil properties.
- Iron enriched biochar and Nano NPK application influence grain and straw yield.
- FeEBC and Nano NPK addition significantly enrich Fe into the finger millet grains.

Introduction

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Current farming practices have ignored the appropriate fertilizer application, and we have been dumping a lot of inorganic fertilizers since we implemented the green revolution strategy (Lal, 2018; Ren *et al,* 2021). And, in order to increase yields, we abandoned the use of organic fertilizers in favor of just adding inorganic fertilizers to the soil for quick and maximum returns and neglect the soil health. As a result, without knowing the soil's fertility state, addition of higher or lesser quantity of inorganic fertilizers to the soil consequently leads to lower the soil productivity (Ren *et al,* 2021). The practice of blending inorganic and organic fertilizers has mostly been discontinued due to lower yield and scarcity of sources (Rajakumar, 2015). As a consequence, soil fertility and productive potential have declined, causing environmental degradation and food insecurity (Liu, 2016). Such fertilization procedures deprive the important nutrients to crops and impede the crops from absorbing micronutrients from the soil (Semida *et al*. 2019). Therefore, assessing fertilizer requirements and blending inorganic with organic fertilizers is need of the hour to save soil health and productivity. Similarly, detecting micronutrient deficiency and applying the necessary quantity might contribute to a sustainable future. This will enhance the soil's physicochemical and biological qualities (Jin *et al.*, 2006).

Supplementing organic fertilizers to soils not only supply nutrient and energy for microorganisms but also serve as a habitat. As a result, microbes thrive and aid in the availability of nutrients to plants (Yuan *et al.,* 2022). Organic source application that binds or chelates micronutrients in the soil to reduce micronutrient loss. This prevents nutrient loss and makes them accessible to plants. Therefore, quantity of fertilizers is needed to be lower of and thus less impact on soil contamination (Li *et al.,* 2020). Naturally existing poor soils and land barriers are the key agricultural limiting concerns owing to different inappropriate physical and chemical components or less suited for cultivation, leading in crops that are unable to grow and provide as low as normal yields. Soils like alkaline, acid, acid sulphate, sandy, organic and shallow soils are all naturally occurring soils (Bonnefoy and Holmes. 2012). Acid soils are the most problematic soils all around the world, including in India (Mandal *et al*., 2019).

Acidic soils are defined as having a pH less than 7.0. Soils with pH less than 5.5 are problem because silicate clay contains more hydrogen, aluminum, and iron ions at exchangeable sites. Acid soils cover 3950 million hectares of the world's ice-free land (Maji, 2012). Almost half of the total soil orders of USDA classification that have acid soils i.e. they are being members of the soil orders Spodosols, Alfisols, Inceptisols, Histosols, Ultisols, and Oxisols, and have udic or ustic moisture regimes Around 179 M ha of these lands are utilized for arable crops, with the rest 33 M ha used for perennial tropical crops(Maji 2015 and ISSS, 2015).Parent materials, temperature, vegetation, and topography all have an impact on the development of acid soils, which are created by base leaching from heavy precipitation and acidic parent materials. The formation of laterite and podzol process regulates the acidity of the soil (Bonnefoy and Holmes. 2012).

National Bureau of oil Survey and Land Use Planning (NBSS&LUP), Indian Council of Agricultural Research (ICAR), India and Maji *et al*. (2012) reported that acid soils occupy around 90 million hectares, and more than one-fourth of the country's total land area. Over half of the territory is used for agriculture, while the other half is used for forestry and other purposes. A just over 59 million hectares (65%) of cultivated land are moderately to slightly acidic, with a pH of 5.5 to 6.5, and about 31 million hectares (35%) are strongly acidic with a pH value less than 5.5 and are severely degraded with extremely poor physical, chemical, and biological characteristics. The use of lime as a soil amendment for acidic soil management, one of the best methods is to cultivate acid-tolerant plants. Rice, minor millets, finger millets are acid-tolerant and respond poorly to liming (Bonnefoy and Holmes, 2012). Acid red soils need continual application of organic manures and amendments owing to their lighter texture and high mineralization rate, both of which increase soil's productivity (ISSS, 2015).

Finger millet (*Elusine coracana* L.) is a dynamic minor millet crop grown in India (Encyclopaedia of food science and nutrition, 2016). India is the world's greatest producer of different millets and among these finger millets contributes for about 85% of total minor crop (Sakamma *et al*., 2018). Finger millet's adaptability, ease of growing, lack of significant pests and diseases, and drought tolerance have made it an obvious option in dry agricultural systems (AICSMIP, 2013). Finger millet is grown on 1.27 million hectares with a total yield of 2.61 million tonnes, with an average productivity of 1489 kg ha^{-1} (Agricultural statistics in a nutshell. 2017). Millions of people in India and Africa eat finger millet as staple food. It also contributes to the country's nutritional security and is seen as a climate-resilient crop for the future (Timmaiah *et al*., 2016). It has a greater nutritive value in terms of macronutrients (Ca - 344 mg $100g^{-1}$), potassium - 408 mg $100g^{-1}$, micronutrients (Fe - 4.62 mg $100g^{-1}$ (Dayakar Rao et al. 2017), Zn - 3.5 - 3.87 mg $100g^{-1}$ (Wafula *et al*., 2018), protein - 7.16 g $100g^{-1}$, sulphur-containing amino acids, and fewer vitamins (Shobana *et al*., 2013).

Iron is an important nutrient and its deficiency affects two billion individuals globally and it causes anemia in pregnant women and children (WHO, 2015). It may be treated with dietary diversity, micronutrient supplements, medications, and surgery, depending on severity. Geographic and economical barriers may prevent everyone from accessing such treatments. Iron is the most challenging mineral to utilize in food fortification since the most soluble and absorbable compounds (e.g., $FeSO_4$) affect the flavor or colour of fortified food, giving it discomforting (Singh *et al*., 2017).

Cereal crops literally cannot afford most micronutrients, while non-staple foods employed for a micronutrient-rich diet are at risk for deficiencies (Bouis and Saltzman., 2016). Most millet crops contain more micronutrients than cereal crops. Due to millets' high micronutrient concentration (Kumar et al., 2016), if scientists enrich such nutrients in these crops would pave a path to fighting human nutrient deficiency. Finger millet includes calcium, potassium, iron, and zinc, thereby iron is used for biofortification in the edible part and nutrient enrichment depends on crop species and variety (Prasad et al., 2014). The process of enhancing the micronutrient content of food grains or edible plant parts is referred to as biofortification. This might be accomplished by breeding and agronomic methods (Andrew De *et al.,* 2018).

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Fe-biofortification may be accomplished using agronomic measures like as fertilizer or foliar feeding (Phattaraku *et al*., 2012). Because not all nutrients are transported, agronomic approaches must account for iron bioavailability at various stages (Cakmak, 2008, Velu *et al*., 2014 and de Valença, 2017). Several important factors, such as bioavailability of nutrient uptake from the soil, nutrient distribution in different parts of the plants, milling or dehusking, and human ability to absorb and utilize nutrients, may all contribute to nutrient loss at various stages (Valenca *et al*., 2017). These aspects must be addressed to ensure iron biofortification via agronomic methods. Mainly, Iron availability and absorption in plants depends on soil factors such as pH, composition, aeration, and moisture. Higher plants may release protons into the soil to augment iron solubility and pH of the rhizosphere, enhancing iron availability and uptake (Singh and Chaturvedi. 2016).

Biofortification is a viable technique for a long-term, sustainable approach to alleviating micronutrient deficient, however biofortification success at the expense of environmental degradation is unacceptable (Singh *et al*., 2016). Leaching is one of the primary issues in the application of fertilizer in agronomic practices because it degrades the environment. Nevertheless, most micronutrients are not vulnerable to leaching since they could develop a strong association with the soil.

Biochar is the porous, carbon-rich byproduct of the thermal decomposition of agricultural leftovers in an oxygen-depleted environment by slow pyrolysis. Biochar as a soil amendment has been discovered as a unique climate change mitigation method that improves soil carbon storage and crop production with using non-feed crop wastes effectively (Peiris *et al*., 2019). The addition of biochar may improve the soil organic carbon content, fertility in Alfisols and it does not equal fertilizer or organic manure (Li *et al.,* 2020) and Yan *et al.,* 2021) Incorporating biochar with the required quantity of inorganic fertilizer may increase crop performance. Biochar is stable compared to manure, compost, and other soil additions; consequently, it is not applied with each crop. The beneficial impacts of biochar may increase over the course of many growing seasons (Venkatesh *et al*., 2018). Liming has been the standard method for modifying acidic soils. Notwithstanding, biochar application as a soil amendment has received a great deal of attention for a number of reasons, including neutralizing soil acidity, making a carbon (C) sink to alleviate global warming, increasing soil water holding capacity, lowering greenhouse gas emissions, and sustaining mobile heavy metals, pesticides, and other organic pollutants in soil (Berek *et al*., 2018).

Biochar is a carbon-rich material formed by pyrolysis of biomass in the absence of oxygen. Normally, pyrolysis of plant biomass yields extremely alkaline biochar (Lehmann *et al*. 2009). Nonetheless, alkalinity changes depending on the feedstock properties employed in biochar production. The higher the alkalinity of biochar, the reduced the acidity (Yuan *et al,*. 2011). It has been found that adding biochar to nutrient-deficient soil improves nitrogen availability and increases plant biomass (Alburquerque *et al*., 2014). The addition of peanut shell biochar with very acidic red soil has been shown to improve cabbage growth by lowering Al toxicity due to higher soil pH and nutrient availability. Together with the liming effect, high surface charge density, vast surface area and internal porosity, and the existence of both polar and non-polar sites on the surface of biochar all play important roles in metal adsorption (Lehmann e*t al.,* 2015). Here, we found certain research gap on use of materials that should be act both as amendment as well as nutrient source. With these backgrounds, we synthesis biochar using tapioca stem wastes and Fe enriched biochar; the prelude work of this doctoral research work was accepted in Nano Impact (Elsevier). And the key purpose of this study was to evaluate the impact of different iron enriched sources and various levels of NPK through conventional and nano fertilizers for iron fortification in finger millet crop under acid soil condition.

Materials and methods

The pot experiment was carried in the farmer's field (doctoral researcher's own field due to COVID-19) in Narippalli village, Harur Firka, Dharmapuri District, is located at 12° 06' N latitude and 78 ° 68' E longitude, and at an altitude of +297 m above mean sea level, during March – June, 2020. In this experiment to assess the fortification of iron, yield and Fe uptake of finger millet (*Elucine coracana* L.) variety PAIYUR 2 was used in red sandy loam soil (Alfisol).

Experimental details

The bulk of an unfertilized surface soil samples (0-15 cm) were collected randomly at own farm (where the field experiments were planned to carry out), processed and used for pot experiment. The findings of soil properties are presented in table 1. The study was conducted in Factorial Complete Randomized Design (FCRD) with three replications. There are two factors

Table 1. Experimental soil properties

Particulars	Observations
Mechanical Composition	
Fine Sand (%)	36.07
Coarse Sand (%)	28.0
Silt (%)	16.43
Clay (%)	19.08
Soil textural class	Sandy loam
Taxonomic class	*Fluventic Haplustalf*
Physical properties	
Bulk density (Mg m^{-3})	1.53
Particle density (Mg m^{-3})	2.22
Total porosity (%)	31.08
Available moisture content (%)	1.37

Physico-Chemical properties	
pH (1:2:5)	4.86
EC (dS m^{-1})	0.08
CEC (cmol (+) kg-1)	3.55
Organic Carbon (g kg^{-1})	2.5
Available N (kg ha^{-1})	182
Available P (kg ha^{-1})	36.50
Available K (kg ha^{-1})	120.7
Available Ca (mg kg^{-1})	288.6
Available Mg (mg kg^{-1})	49.8
Available S (mg kg^{-1})	7.49
Available Fe (mg kg^{-1})	3.37

viz., Fe sources and NPK fertilizers (conventional and nano NPK) i.e. the first factor has 3 levels and the second factors has 6 levels. For each iron sources applied in 6 pots. In the six pots 3 pots were supplied conventional fertilizer 75%, 100% and 125% and another 3 pots were supplied nano fertilizer 75%, 100% and 125% as per the treatment schedule.

Treatments used are as follows: Factor 1 containing 3 levels of Fe sources were Fe enriched biochar (FeEBC) (F_1), Fe enriched farm yard manure (FeEFYM) (F_2) and ferrous sulphate (FS) (F_3). The factor 2 containing 6 levels of NPK fertilizers were 75% RDF through Conventional NPK (75% - CF) (T_1), 100% RDF through Conventional NPK (100% - CF) (T_2), 125% RDF through conventional NPK (125% - CF) (T_3), 75% RDF through Nano NPK (75% - NF) (T_4), 100% RDF through Nano NPK (100% - NF) (T_5) and 125 % RDF through Nano NPK (125% - NF) (T_6). Totally18 pots per replication. The study was conducted in green shade net house.

Fertilizer application and assessment of yield, nutrient content and uptake

For this study the plastic pot was taken with the capacity of 10 kg which was 18-inch (1.5 ft) height 12-inch (1 ft) diameter. 10 Kg of processed soil was filled into the plastic pot. As stated in the treatment schedule calculated amount of Fe sources applied based on Fe content in Biochar, FYM and Ferrous sulphate i.e., depicted in table 2, conventional fertilizers viz., (60:30:30 N, P_2O_5, K_2O kg ha^{-1} in the form of urea, DAP and MOP) and Nano fertilizers (61.75 Kg ha^{-1}in the form of granular Nano NPK) were applied on dry weight basis at 75%. 100 % and 125% respectively and mixed well uniformly with soil.

Table 2. Details of iron sources used in the experiment

Sources	Fe content (%)	Sources application based on iron content	
		kg ha^{-1}	g pot^{-1} (10 kg of soil)
Ferrous sulphate ($FeSO_4$)	19	25	0.125
Iron enriched biochar (FeEBC)	0.87	548.49	2.7
Iron enriched farm yard manure (FeEFYM)	0.91	520.83	2.6

After application of Fe sources and fertilizers as per treatment schedule and watered to maintain the required soil moisture in pots, then 25 days old finger millet seedlings 4 numbers were planted for each pot. At harvest stage, grain yield and straw yield was recorded and expressed in g pot^{-1} at 15 - 20 % moisture level. Dry matter production of the plant sample was assessed after shade drying after that oven dried at 60 - 70 ºC to attain constant weight, and the weight was recorded. The average dry weight was taken as dry matter production per plant and is expressed in g pot^{-1}. The plant grain and straw samples were collected at harvest stage from various treatments in each replication. The grain and straw samples were shade dried and then oven dried at 70 ºC for 24 hours to attain constant weight. They were ground into fine powder using Willey mill and used for chemical analysis to estimate Fe content in grain and straw. The grind powder sample 1 gram weighed and transferred to digester tube. Then 15 ml of Nitric acid and perchloric acid mixture was put into the digester tube and digest the content for 45 minutes to clear solution. Then the digested solution was made up to 100 ml and then Fe content was analyzed by using Atomic Absorption Spectrophotometer (Miller, 1998 and Jackson, 1973). The uptake of Fe was worked out by the formula of nutrient content (%) multiplied with DMP (grain and straw) and computed to g pot^{-1}. The percentage increases of yield, Fe content and uptake were calculated by the difference of highest value and lowest value is divided by lowest value and calculated value is multiplied by 100.

Following the methodology proposed by Jackson (1973) the soil pH and EC were determined by using a modified dilution of 1:2.5 (soil: deionized water). Cation exchange capacity was determined by replacement of exchangeable cations by ammonium acetate (pH 7) by Piper (1966). Soil organic carbon was determined by chromic acid wet digestion method (Walkley and Black. 1934). To assess the availability of Fe in the present study by the DTPA extractant procedure, which was described by Lindsay and Norvell. (1978). Available N was determined by alkaline permanganate method (Subbiah and Asija., 1956), available P was determined by Bray 1 extractant method by using Spectro photometer (Bray and Kurtz, 1945), available K was determined by 1N NH_4OAc extractant method using with Flame photometer and available Ca and Mg were determined by Versenate method using 0.02 N EDTA (Jackson, 1973) and available sulphur was determined by 0.15 per cent $CaCl_2$ by using turbidimetry (Piper, 1966).

Statistical analysis

The data on yield, Fe content, and uptake parameters of this study were statistically scrutinized as suggested by Gomez and Gomez (1976), using AGRES package. The treatment differences were found significant (LSD test), critical differences were worked out at five percent (p=0.05) probability level and the value were furnished. Correlation and regression analysis were carried out using OPSTAT package to determine the strength and relationship among the yield and nutrient characters.

Results and discussion

Grain yield

The grain and straw yields of finger millet were significantly impacted by the various sources of iron, the varying rates of NPK fertilizers, and the complex interplay between sources and levels significantly (Table 3). The grain yield from 8.60 g pot^{-1} to 16. 3 g pot^{-1}. Therefore, the incorporation of Fe sources resulted in an increase in the grain yield. Application of Fe enriched biochar (FeEBC) recorded the mean greatest grain yield of 13.2 g pot^{-1}, followed by Fe enriched FYM (FeEFYM) with grain yield of 11.8 g pot^{-1}. Whilst the lowest grain yield of 11.3 g pot^{-1} was recorded with ferrous sulphate incorporated pots. When compared to FeEFYM and FeSO4, the application of FeEBC proved with a grain yield 11 % and 16% higher grain yield, respectively. The average grain yield varied from 14.5 g pot^{-1} to 9.3 g pot^{-1} depending on the form of macronutrient fertilizers used i.e. conventional NPK sources like urea, di-ammonium phosphate and muriate of potash or Nano NPK (granular protein gluconated). The grain yield that was recorded with 125% nano NPK (NF) was recorded the highest grain yield of 14.53 g pot^{-1}, and it was on par with 100% nano NPK (13.62 g pot^{-1}), and the lowest grain yield was registered with 75% conventional NPK (CF) of 9.29 g pot^{-1}. The addition of nano NPK at 125%, 100%, and conventional NPK at 100% indicated substantial differences, although the level of CF to NF at 100% and 75% was comparable.

There was a consistent trend observed among NPK levels with respect to grain yield at different Fe sources. Regardless of Fe sources, the NPK fertilizer application 125% NF registered highest grain yield while 75% CF recorded the lowest grain yield. The percentage of increase that was achieved as a direct result of the application of Fe and NPK fertilizers varied from 20.6 to 56.4. This might be due to interaction effect of iron enriched biochar with nano NPK fertilizer sources significantly increased the nutrient supply by moderating the physicochemical properties of the acid soil. Besides, ensuring the sustainable release of Fe and primary nutrients and it might have supported the uptake of nutrient elements in consequence with improved productivity was realized by the application of Fe enriched biochar and Nano fertilizer. Crop yields were similar to those achieved by addition of biochar and chemical fertilizers as reported by Li *et al.* (2020) and Blackwell *et al*. (2010). Biochar can be attributed to increased CEC of soil, pH, nutrient retention and increase plant available water. Ultimately, it might have increased the grain yield of finger millet (Yan *et al.,* 2021 and Lehmann e*t al.* 2015). And it could be attributed to better uptake of essential nutrients and translocation to the economic parts as well as improving yield parameters. These kinds of responses were reported by Bair *et al*. 2020; Van Zwieten *et al*. (2010) and Major *et al*. (2010).

Table 3. Effect of different Fe sources and NPK fertilizers on Grain yield of finger millet (g pot^{-1})

NPK levels	Iron sources			
	FeEBC	**FeEFYM**	**$FeSO_4$**	**Mean**
CF 75%	10.3	9.0	8.60	**9.29**
CF 100%	12.0	10.9	10.8	**11.2**
CF 125%	13.0	12.7	11.6	**12.4**
NF 75%	12.2	10.8	11.4	**11.5**
NF 100%	15.4	13.1	12.4	**13.6**
NF 125%	16.3	14.6	12.8	**14.5**
Mean	**13.2**	**11.8**	**11.3**	
	F	**T**	**FxT**	
SEd	0.19	0.27	0.47	
CD (p=0.05)	0.39	0.55	0.95	

[CF – Conventional fertilizer, NF-Nano fertilizer, $FeSO_4$ - Ferrous sulphate, FeEBC –Iron enriched biochar, FeFYM– Iron enriched Farmyard manure]

Results of the present investigation also describes the encouraging effect of biochar on soil CEC following which the ability to hold or bind the plant nutrient cations increases, thereby increasing the retention and reducing leaching losses. The alkaline nature of biochar that lowered the exchangeable acidity thus raising the soil pH has provided a wide range of benefits in terms of soil quality especially by chemically improving nutrient availability (Amoah *et al.,* 2020) Application of biochar alone or together with fertilizers would have resulted in higher nutrient uptake and yield as reported from many investigations with biochar on crop yield (Elangovan, 2014 and Dainy, 2015).

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Straw yield

With respect to straw yield, it ranged from 21.0 g pot^{-1} to 39.9 g pot^{-1}. Addition of Fe sources might have enhanced the straw yield. Application of Fe enriched biochar (FeEBC) recorded the mean highest straw yield (32.84 g pot^{-1}), the second highest (29.72g pot^{-1}) was recorded by Fe enriched FYM (FeEFYM) and Ferrous sulphate registered lowest straw yield (27.15 g pot^{-1}). And the per cent increases were 16 and 34 of straw yield over FeEFYM and $FeSO_4$, respectively. Among conventional and Nano NPK fertilizers, the mean straw yield ranged from 36.7 g pot^{-1} to 22.3 g pot^{-1}. The 125% nano NPK (NF) was recorded the highest straw yield (36.7 g pot^{-1}) which was comparable with 100% NF (35.6 g pot^{-1}) and 75% conventional NPK (CF) registered lower straw yield (22.3 g pot^{-1}). With respect to straw yield, the addition of nano NPK 125%, 100% and conventional NPK 100% showed significant difference and the 100% CF and 75% NF were on par.

Table 4. Effect of different Fe sources and NPK fertilizers on Straw yield of finger millet (g pot^{-1})

NPK levels	**Iron sources**			
	FeEBC	**FeEFYM**	**$FeSO_4$**	**Mean**
CF 75%	23.97	22.00	20.98	**22.32**
CF 100%	28.64	25.29	24.89	**26.27**
CF 125%	35.01	32.11	27.00	**31.37**
NF 75%	30.03	26.16	25.05	**27.08**
NF 100%	39.46	36.00	31.43	**35.63**
NF 125%	39.95	36.74	33.53	**36.74**
Mean	**32.84**	**29.72**	**27.15**	
	F	T	FxT	
SEd	0.4	0.6	1.05	
CD(p=0.05)	0.87	1.23	2.13	

[CF – Conventional fertilizer, NF-Nano fertilizer, $FeSO_4$ - Ferrous sulphate, FeEBC –Iron enriched biochar, FeFYM– Iron enriched Farmyard manure]

Similar to grain yield, there existed significant difference among NPK levels on straw yield in the absence of Fe sources application. Irrespective of Fe sources, the NPK fertilizer application 125% NF registered highest straw yield while 75% CF recorded the lowest straw yield. The per cent increase due to addition of Fe and NPK fertilizers ranged from 32 to 119.

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The application of Fe enriched biochar and nano fertilizer provide energy to soil microbes and helps in improving microbial activity and soil physical environment. The interaction effect of biochar and nano fertilizer helps to make long term availability of both micro and macro nutrient to crop, which aids in growth and development of plants (Agegnehu *et al*., 2016). The very high porosity and surface area of biochar enable it to retain more water (Martinsen *et al.,* 2015) and nutrients in addition to providing an ideal habitat for the soil microorganisms which may be the probable reason for crop yield improvement as reported by Lehmann *et al.* (2006). Due to its resistance to decomposition in soil, one-time application of biochar can provide beneficial effects on crop growth and productivity has also been narrated by Islami *et al*. (2013), Elangovan (2014), Widowati *et al*. (2017), Sikder and Joardar (2018) and Sara *et al*. (2018).

Grain and straw Fe content (Iron fortification)

There was a significant effect of different levels of NPK fertilizer and Fe sources applications on Fe content in grain and straw. Among the different Fe levels, the grain Fe content ranged from 44.4 mg kg^{-1} to 46.1 mg kg^{-1} and 56.4 mg kg^{-1} to 59.2 mg kg^{-1} in grain and straw, respectively. The addition of FeEBC significantly fortify the Fe content in finger millet grain and straw. The per cent increase in grain Fe content by 1.75 and 3.92 and in straw 2.96 and 5.03, respectively over FeEFYM and $FeSO_4$ applied plants.

While comparing the NPK levels implemented to finger millet based on the RDF, the maximum of Fe content was registered with 125 % Nano NPK (NF) which was on par with 100% NF and the minimum was registered in 75% CF. Fe content in grain ranged from 44.4 to 45.6 mg kg^{-1} and 56.4 to 58.7 mg kg^{-1} in straw, respectively. The per cent increase of Fe content in grain was ranged from 1.92 to 2.72 and 2.19 to 4.02 in straw. However, there is no variation among the different NPK fertilizer levels and interaction effect also was found to non-significant.

The results showed that maximum grain and straw Fe content with the combination of FeEBC and 125% NF (47.0 mg kg^{-1}) and (60.1 mg kg^{-1}) which was on par with FeEBC and 100% NF (46.9 mg kg^{-1}) and (60.1 mg kg^{-1}) application and the minimum values of iron content recorded with combined application of $FeSO_4$ and 75% CF (44.3 mg kg^{-1}) and (55.1 mg kg^{-1}) respectively.

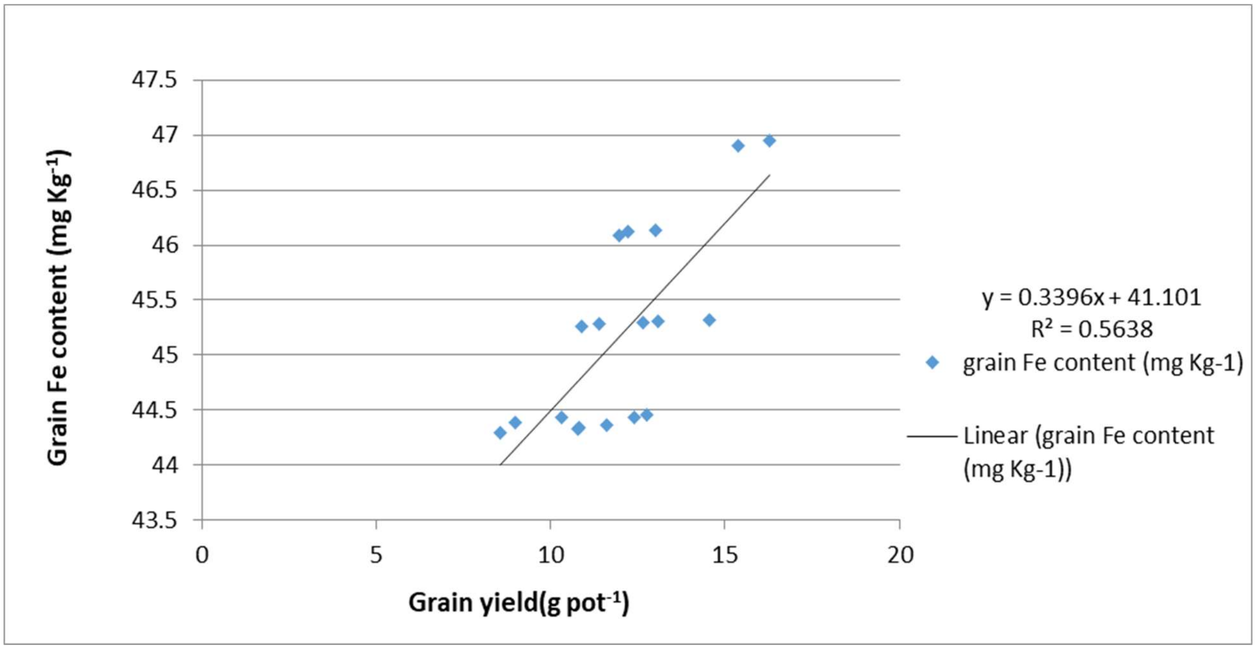

Fig.1. Linear relationship between grain yield and grain Fe content

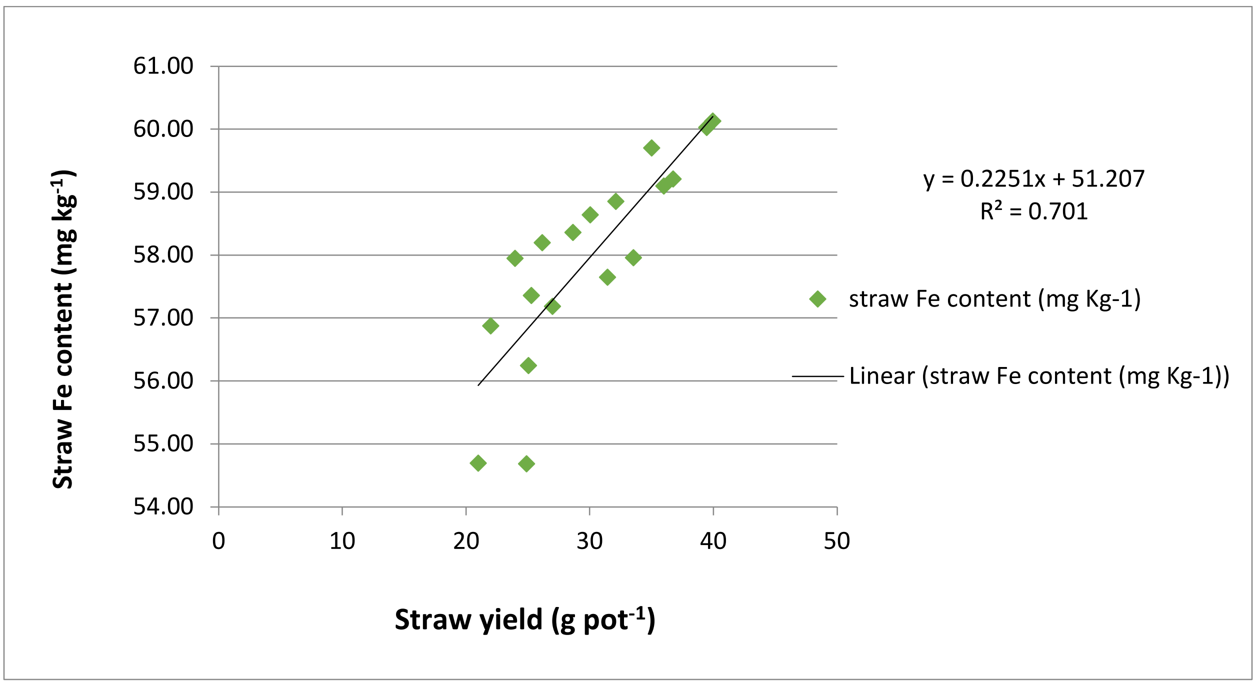

Fig.2. Linear relationship between straw yield and straw Fe content

This iron fortification may be attributed due to significant increase in the Fe availability in the rhizosphere for subsequent storage of Fe content by the added iron sources especially FeEBC and it might have focused on the active release of Fe mobilizing substance from roots. Further, the higher micronutrient content especially that of Fe due to Fe-enriched organics along with different

NPK fertilizer application could also be attributed to the priming effect causing higher crop growth, yield and nutrient content to biochar addition either alone or in combination with NPK (Abbas *et al*., 2017; Walter and Rao., 2015). In addition to that, in the present study found a significant and positive relationship between grain and straw yield with grain ($y=0.3396x+41.101$, $R^2 = 0.5638$) and straw ($y=0.2251x + 51.207$, $R^2 = 0.701$) Fe content (Fig. 1 and Fig.2) in finger millet by the addition of different Fe and NPK levels. Yet another reason for increased nutrient content and uptake suggested by many researchers is the favorable effect on soil pH, especially in an acidic soil following biochar addition which decreases Al activity, bettering root growth and in turn a higher nutrient uptake. Nigussie *et al*. (2012) highlighted the presence of essential plant nutrients, its high surface area, porous nature and the capacity of biochar to act as a medium for soil microorganisms as the prime reasons for the enhancement in soil properties, leading to increased nutrient content and uptake in plants whenever supplied with biochar. Increase in micronutrient content and uptake might be due to the presence of chelated micronutrients in the applied biochar, as opined by Nanda *et al*. (2016), and Major *et al.* (2010).

Grain and straw Fe uptake

There was a significant effect of different levels of NPK fertilizers and Fe sources application on uptake of Fe in grain and straw (Table 5). Fe sources effect on iron uptake was found to be similar trend as registered in Fe content. Among the Fe sources, the grain uptake ranged from 1.20 to 1.54 mg pot^{-1} and straw Fe uptake ranged from 1.54 to 19.8 mg pot^{-1}. The addition of FeEBC registered higher Fe uptake over other two iron sources used. Fe uptake increases with an average of 15. 6 % and 27.7 % in grain, and 16.6 % and 28.5 % in straw over FeEFYM and $FeSO_4$, respectively. And the lower Fe content was recorded by $FeSO_4$ which was par with FeEFYM.

Among the NPK fertilizer levels, the grain Fe uptake ranged from 0.85 to 1.78 mg pot^{-1}and straw Fe uptake ranged from 1.12 to 2.22 mg pot^{-1}. The per cent increase in grain Fe uptake ranged from 42.50 to 109.5 and straw Fe uptake ranged from 43.0 to 112.2. The maximum of iron uptake in grain and straw were recorded by 125% nano NPK of 1.78 mg pot^{-1} and 2.29 mg pot^{-1}, respectively. Which was on par with 100% nano NPK (173 mg pot^{-1} and 2.23 mg pot^{-1}) and the minimum uptake of 10.77 mg pot^{-1} and 17.75 mg pot^{-1} was recorded by 75% conventional NPK in grain and straw, respectively. Interaction of different levels of NPK fertilizer and Fe sources was found to be non-significant on grain and straw iron uptake. Further, there was significant positive linear relationship between yield and Fe uptake of finger millet grain ($y=0.1776x – 0.7638$, $R^2 = 0.9276$) (Fig.3) and ($y=0.8117x - 6.4974$, $R^2 = 0.9705$) (Fig.4) indicating that enhanced in grain and straw uptake caused increased in grain and straw yield.

Table 5. Effect of different Fe sources and NPK fertilizers on grain and straw Fe uptake of finger millet (mg pot^{-1})

	Grain Fe uptake				Straw Fe uptake			
NPK levels	Iron sources				Iron sources			
	FeEBC	FeEFYM	$FeSO_4$	Mean	FeEBC	FeEFYM	$FeSO_4$	Mean
CF 75%	0.89	0.84	0.81	**0.85**	1.20	1.11	1.05	**1.12**
CF 100%	1.36	1.20	1.00	**1.19**	1.77	1.55	1.27	**1.53**
CF 125%	1.67	1.59	1.31	**1.52**	2.15	2.05	1.67	**1.95**
NF 75%	1.42	1.23	1.06	**1.24**	1.83	1.58	1.35	**1.58**
NF 100%	1.94	1.74	1.51	**1.73**	2.45	2.21	1.88	**2.18**
NF 125%	1.98	1.77	1.58	**1.78**	2.46	2.25	1.96	**2.22**
Mean	**1.54**	**1.40**	**1.21**		**1.98**	**1.79**	**1.53**	
	F	T	FxT		F	T	FxT	
SEd	0.03	0.04	0.06		0.03	0.05	0.08	
CD(p=0.05)	0.05	0.07	0.13		0.07	0.09	0.16	

[CF – Conventional fertilizer, NF-Nano fertilizer, $FeSO_4$ - Ferrous sulphate, FeEBC – Iron enriched biochar, FeFYM– Iron enriched Farmyard manure]

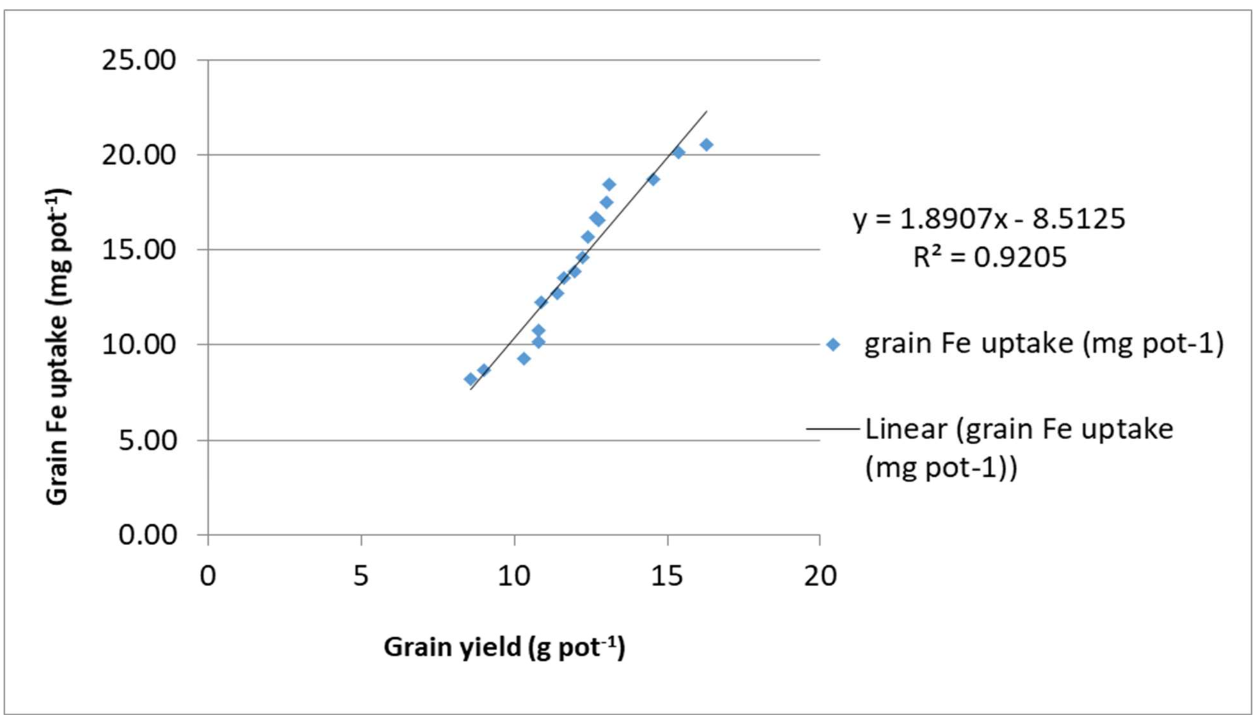

Fig.3. Linear relationship between grain yield and grain Fe uptake

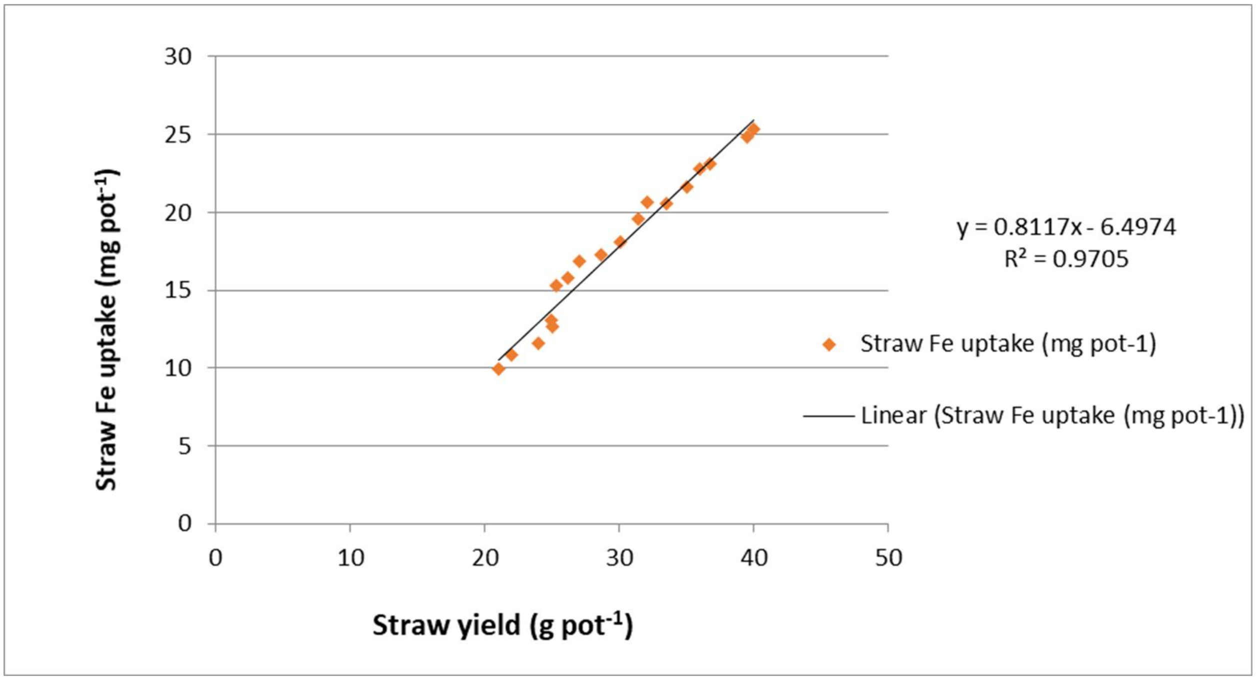

Fig.4. Linear relationship between straw yield and straw Fe uptake

The simple correlation studies among the yield and uptake had shown that Fe content was positively correlated with grain and straw yield, grain and straw uptake only with the application of different Fe sources during the green house study (Table 6). This could be attributed by the different Fe sources applied and they aptly increase Fe availability in the rhizosphere for subsequent uptake and realized the active release of Fe mobilizing substance from finger millet root. It might be due to the Fe enriched BC that caused higher utilization of Fe mainly due to its beneficial effects in mobilizing the native nutrients from soil. Further, the microbes near the root rhizosphere utilize Fe from both soil and enriched biochar successfully made the Fe in available form to uptake as naturally chelated form by crop and fortify the iron its seed. This might have provided better nutrition over longer time to cause better crop growth and higher yield. The higher removal of Fe by grain and straw also is attributed to the priming effect of externally added nutrients to improve crop growth (Yang et al., 2020). Hence higher content of Fe in grain and straw and also higher grain and straw yield under Fe sources application might have contributed towards higher uptake of Fe by grain and straw. The results are in accordance with those reported by Latha *et al*. (2001) and Patel *et al*. (2010). According to Abbas *et al*. (2017) the role of the change of oxidation state of Fe (popularly known as “redox wheel”) as a vital key for enhancing P, N and S availability and uptake in plants. Similar observations of higher nutrient content and uptake due to biochar addition either alone or in combination with NPK has also been reported by Hamdani *et al*. (2017).

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Table 6. Correlation analysis among the grain and straw yield, Fe content and uptake of Finger millet

	GY	GC	GU	SY	SC	SU
GY	1	0.280^{NS}	0.493**	0.600**	0.187^{NS}	1.000**
GC	0.280^{NS}	1	0.253^{NS}	0.440*	0.613**	0.280^{NS}
GU	0.493**	0.253^{NS}	1	0.173^{NS}	0.453**	0.493**
SY	0.600**	0.440*	0.173^{NS}	1	0.107^{NS}	0.600**
SC	0.187^{NS}	0.613**	0.453**	0.107^{NS}	1	0.187^{NS}
SU	1.000**	0.280^{NS}	0.493**	0.600**	0.187^{NS}	1

[GY-Grain yield, GC-grain Fe content, GU-Grain Fe uptake, SY-Grain yield, SC-grain Fe content, SU-Grain Fe uptake, NS-Non significant, ** -Significant at 0.01 level, * -Significant at 0.05 level, Kendall's tau correlation]

Conclusion

In conclusion, this study demonstrates that finger millet yield increased with nano NPK fertilizers but not with conventional NPK fertilizers. And the benefits of incorporation of iron enriched biochar (FeEBC) along with nano NPK fertilizers ensuring the higher yield and iron uptake of finger millet through fortifying the nutrient content in grains. The response of finger millet variety PAIYUR-2 to co-fertilization of iron enriched biochar with 125 % Nano NPK or 100 % nano NPK significantly enhanced the yield, Fe content and its uptake in under acidic stress condition. The additive effect of Iron enriched biochar proved its superiority over FeEFYM and $FeSO_4$ supplementations to acid soils is confirmed through its alkaline nature here in this study. Therefore, it can be concluded that the application 100% recommended dose of Nano NPK with Fe enriched biochar could be an effective and viable option for fortifying the Fe content in finger millet. However, the effect of FeEBC application should be further examined for confirmation by increasing the levels with various varieties of finger millet. In addition, the iron transport under acidic soils through iron enriched or doped biochar or nano biochar in finger millet deserve further investigation.

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