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AGROBIOCHEMICAL STUDIES OF TRITICALE SEED USING FOLIAR APPLICATION OF IRON AND ZINC AT VARIOUS POTASSIUM LEVELS

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Abstract

Triticale (X Triticosecale Wittmack) has higher yield potential and is tolerant to several environmental stresses, however, its grain quality is not as good as wheat. If the quality nutrient ie potassium (K) and the micronutrients zinc (Zn) and iron (Fe), are applied; its grain quality might be improved. The current study was initiated to evaluate quantitative and qualitative improvement in grains of triticale through the application of Zn and Fe with K fertilization. The experiment was laid out in RCBD replicated four times during rabi 2020-2021 and 2021-2022 at The University of Agriculture, Peshawar-Pakistan. Triticale cultivar QASWA-18 was sown using recommended seed rate of 120 kg/ha. The seed was planted in a 2.4 x 3m^2 size plot. Eight rows were sown in each plot using 3m row length. Recommended nutrients i.e. NP were used @ 120:90. Zn and Fe were applied as $ZnSO_4.7H_2O$ & FeO respectively. Three levels each of K ie. 60, 90 & 120 kg/ha, Fe ie. 3, 6 & 9 kg/ha and Zn ie. 2, 4 & 6 kg/ha were used. One control plot was used in each replication. K was used as soil applied at sowing and Fe and Zn were foliar applied at stem elongation stage. Results revealed significant variation in biochemical and yielding attributes of triticale grain during both the years. Averaged over the two years data, plots where K was applied @ 120 kg/ha recorded higher grain yield (GY) (5385 kg/ha), harvest index (HI) (39.6%), grain Fe content (32.7 mg/kg) & Zn content (24.6 mg/kg), grain protein content (GPC) (13.66%) and gluten content (GC) (26.37%). HI both at 90 and 120 kg K/ha were not significantly different. Plots where Fe was applied @ 9 kg/ha produced higher GY (4866 kg/ha), grain Fe content (32 mg/kg), GPC

(13.48%) and GC (26.02%). Plots where Zn was applied @ 6 kg/ha produced higher GY (4722 kg/ha), grain Zn content (24.2 mg/kg), GPC (13.42%) and GC (25.84%). GY and GC at 6 and 4 kg Zn/ha were not significantly different. In control plots where no K, Fe or Zn were applied, lower GY (2702 kg/ha), and HI (29.2%), lower grain Fe content (28.3) and Zn content (19.6 mg/kg), lower GPC (12.33%) and GC (23.9%) were observed. The interaction of K × Zn was significant for grain Zn content only. It was concluded that GY, GPC and GC of triticale were increased with increase in the levels of K, Fe and Zn. The K × Zn interaction significantly affected grain Zn content, with the highest value recorded under 120 kg K ha⁻¹ combined with 6 kg Zn ha⁻¹. A significant K × Fe interaction was also observed for GY, HI, GPC and GC, with maximum values obtained at 120 kg K ha⁻¹ and 9 kg Fe ha⁻¹. It is recommended that K @ 120 kg/ha and Fe @ 9 kg/ha should be used combine for higher GY and grain quality. The combined use of K @ 120 kg/ha & Zn @ 6 kg/ha is recommended to enhance grain Zn content of triticale.

INTRODUCTION

Triticale (X *Triticosecale* Wittmack) is recognized as the first artificially developed cereal crop, produced through the hybridization of wheat (*Triticum aestivum* L.) and rye (*Secale cereale* L.) by plant breeders from Germany and Scotland (Igne *et al.,* 2007). Main credit goes to Scottish botanist S. A. Wilson who developed it in 1875 in Scotland (Wilson, 1875). While German researchers first bred triticale in 1888, it remained agriculturally underutilized for nearly a century; its commercial viability as a crop only emerged in 1969 through the creation of Hungary's Triticale No. 57 and No. 64 varieties (Martinez-Moreno et al. 2025). Triticale was developed by combining desirable traits from both parental species, including the food-processing qualities of wheat and the greater adaptability of rye to drought and cold, along with disease resistance and suitability for low-input conditions. It was grown on 3.8 million ha area around the world half of which is produced in Poland, Germany and Belarus combined (Martinez-Moreno et al. 2025). Presently, it has more than 200 cultivars in around 30 countries (FAO, 2014).

Triticale produces higher grain yield as compared to wheat both under optimal and suboptimal conditions. It is more competitive with weeds and other pests and survives better under drought conditions (Beres et al., 2010). Its grain contains more protein (19.71%) than wheat, maize, rye, oat, and rice (Hale et al., 1985). However, its flour shows brownish color and has less stickiness as compared with wheat, which makes it less attractive for the consumers. This less stickiness is due to comparatively lower amount of gluten which makes its quality poorer as compared to wheat in terms of bread making. The ratio of globulin and albumin in triticale grain is higher having minimum amount of prolamin thus enhancing the digestibility of triticale products. Triticale grain contains approximately 20 % higher lysine amino acid than wheat grain (Doxastakis et al 2002).

Potassium (K) application contributes significantly to triticale yield and quality by supporting plant growth, development, assimilate translocation, and protein formation (Marschner, 1995). Crop growth significantly improved with the application of K (Marschner et al., 1996). Potassium fertilization could positively promote metabolic activities like enzyme activation, neutralization

of organic acids and also enabling the plant more efficient to drought, insect pests and disease conditions. Furthermore, K being a macro nutrient can significantly promote yield of triticale crop. It is macro nutrient but not properly considered in Pakistan especially for cereals (MNFSR, 2020). The critical limit for soil K deficiency is 150 mg extractable K per kg of soil (Tahir et al., 2008; DAFWA, 2015),

Zinc (Zn) is an essential micronutrient that supports plant growth, contributes to nitrogen metabolism, and facilitates enzyme activation (Inamullah and Khan, 2022; Mengel et al., 2001). Due to deficiency of Zn, decline occurs in root development system of crop (Fageria, 2005). Unavailability of Zn may adversely affect absorption of water and nutrients as a result plant growth and yield decrease. Biochemical processes in plants depend upon Zn because it is the most active and essential nutrient (Mousavi et al., 2013). Zn also helps in increasing the protein concentration in grain due to which seed quality is improved (Broadley et al., 2007).

The most copious and rich element on the earth crust is iron (Fe). Generally, soil contains 1-5% of Fe. It helps in formation of enzymes. Fe plays essential role in metabolism of nucleic acid and in the processes of oxidation (Havlin et al., 2104). Among the micronutrients, Fe and Zn are more important for humans (Bouis and Welch, 2010). Improvement of Zn and Fe concentration in cereal crops will enhance the quality and yield of crops. According to Velu (2014), due to deficiency of essential micronutrients such as Zn and Fe, more than two billion people are affected in world which especially include children having less than five years age and pregnant women.

The strategic application of Zn and K fertilizers significantly enhances nutrient availability, which directly translates into better growth, higher yields, and superior crop quality for wheat (Bashir et al. 2023). Beyond macro-nutrient benefits, targeted K nutrition can also trigger enhanced Fe absorption across both monocot and dicot plant varieties, making it an essential tool for Fe bio-fortification strategies (Awad-Allah and Elsokkary, 2020). Enhancing these specific elements in food crops is a major public health priority, as Zn and Fe deficiencies trigger “hidden hunger” posing severe health risks to newborn infants and breastfeeding mothers (Hafeez et al. 2021).

Zn and Fe deficiencies are among the most widespread micronutrient-related public health concerns worldwide. Inadequate Zn intake may contribute to gastrointestinal disorders, impaired physical growth, elevated blood pressure, and hormonal imbalances (Prasad et al., 2004; Levenson et al., 2011). Similarly, Fe deficiency is associated with anemia, pregnancy-related complications, weakened immunity, fatigue, reduced reproductive performance, and impaired cognitive and work capacity (Bouis, 2002). Although nutrient-rich foods such as apples provide essential micronutrients, their high cost may limit accessibility among low-income populations. Agronomic bio-fortification through Zn and Fe fertilizer application can improve the nutritional quality of triticale flour, offering an affordable dietary approach to addressing micronutrient deficiencies (Kotecha, 2008)

Triticale has higher yield potential and is tolerant to several environmental stresses like heat and drought, therefore, it can be a suitable substitute of wheat in the present and forthcoming scenario of global warming and climate change. However, its bread making quality is not considered as good as wheat. Its flour shows brownish color and has less stickiness as compared with wheat, which makes it less attractive for consumers. If the quality nutrient ie K and the micronutrients Zn and Fe, the deficiency of which may cause hidden hunger, are applied; the grain quality of Triticale may be improved.

The current study was therefore initiated to evaluate quantitative and qualitative improvement, if any, in triticale through the foliar application of Zn and Fe levels with soil applied K fertilization.

Materials and Methods

A two years field experiment was carried out to study the effects of various Fe and Zn levels on grain yield and quality of triticale with various K levels during the winter growing seasons 2020-21 and 2021-22 at the Agronomy Research Farm, The University of Agriculture Peshawar, Pakistan. The research was carried out within a subtropical, semi-arid zone positioned at coordinates 34.022°N and 71.471°E, at an elevation of 359m. Long-term meteorological data spanning three decades indicates that the region receives a mean yearly precipitation of approximately 400 mm. The thermal profile of the site includes hot summers, where average maximum temperatures reach 40°C and minimums settle around 25°C. Winters are mild to cool, with average temperatures ranging from a low of 4°C to a peak of 18.35°C (Inamullah and Khan, 2022). The experimental field was irrigated using water of Kabul river through Warsak canal. The textural class of the soil was silty clay loam, pH of the soil was 8.2, EC 0.18 dSm^{-1}, OM 0.84%, K 89 mg kg^{-1}, Zn 0.7 ppm and Fe 2.3 ppm of the soil of the experimental field.

A Randomized Complete Block design with four replications was utilized to evaluate triticale (cv. Qaswa-18) performance. Seeds obtained from CCRI, Nowshera, were planted at a standard rate of 120 kg ha^{-1} using a tractor-driven seed drill into a well-prepared, moisture-sufficient seedbed initially tilled via disc harrow and rotavator. Each 2.4 × 3 m^2 experimental plot contained eight rows, with a row length of 3 m and an inter-row spacing of 30 cm. Recommended NP fertilizers were added at a baseline rate of 120:90 kg ha^{-1}, whereas potassium was applied to the soil at sowing using three distinct concentrations (60, 90, and 120 kg ha^{-1}). Foliar interventions of Zn (2, 4, and 6 kg ha^{-1} as $ZnSO_4 \cdot 7H_2O$) and Fe (3, 6, and 9 kg ha^{-1} as FeO) were executed between the stem elongation and booting stages. These treatments were delivered as surfactant-free 5% solutions sprayed during the afternoon, with one control plot assigned per replication. Chemical weed control was enforced using standard narrow- and broad-leaf herbicides, and the recommended irrigation routine was adapted based on prevailing weather patterns. Meteorological indicators for the experimental timeline (Fig. 1) were sourced from the Peshawar regional station of the Pakistan Meteorological Department.

Data were taken on following parameters.

To determine grain yield, the threshed grains from each experimental unit were weighed. The raw plot weights were subsequently converted into kilograms per hectare (kgha^{-1}). Similarly, the harvest index (HI) was computed as the ratio of economic yield to total biological yield and Fe content in triticale grain (mg/kg or ppm of seed) was determined by using the spectrometric Fe determination method according to AOAC (2000) and Jha (2023) methods. For the determination of zinc in grains, DTZ staining method was used (Ozturk et al., 2006, Duarte et al. 2016).

Zn concentration was assessed using 1 g of whole-grain flour placed in borosilicate glass test tubes. Milling equipment was thoroughly cleaned between samples to minimize cross-contamination. Each sample was mixed with 5 mL of 300 mg L^{-1} diphenyl thiocarbazone (DTZ)-based solution prepared in analytical-grade methanol and homogenized for 2 minutes using a Vortex mixer and 15 minutes to facilitate colour development. Zn levels were initially estimated through a visual scoring scale ranging from 1 to 3, corresponding to light pink/red, moderate red, and deep red coloration, respectively. The reliability of this screening method was assessed by comparing the observed colour intensity with quantitative Zn conc. obtained through Atomic Absorption Spectroscopy (AAS) following acid digestion of the tissue samples. Although the DTZ-based colour reaction remained stable for approx. 120 minutes, noticeable fading occurred after 3 hours.

Grain N content was determined using the Kjeldahl method. For digestion, 0.2 g of grain sample was mixed with 1.3 g of digestion mixture (10 g $CuSO_4$ + 100 g K_2SO_4 and 1 g selenium powder)

and 3 mL of diluted H_2SO_4 in a digestion tube. The mixture was heated continuously until a clear greenish solution was obtained, followed by filtration and volume adjustment to 100 ml. A 20 ml aliquot of the prepared solution was transferred to the Kjeldahl apparatus, mixed with 4 ml of NaOH, and heated for distillation. The liberated ammonia was collected in a conical flask containing 5 ml of boric acid solution and an indicator, followed by titration with 0.005 N HCL. Blank and sample readings were recorded to calculate grain N content. Grain protein content was estimated by multiplying the N concentration by 6.25 (Bremner, 1982). Wet gluten (% of grain total protein) was determined using the hand-washing method (AACC, 2000). 25 g of triticale flour from each treatment was mixed with 15 mL of water to prepare a uniform dough. The dough was rested at room temperature for one hour and subsequently washed by hand to remove starch and soluble components, leaving a cohesive gluten mass. The resulting gluten ball was placed in a previously tared container, and its weight was recorded to determine wet gluten content. The experimental data were analysed according to the procedures of the Randomized Complete Block Design (RCBD). Treatment means were compared using the Least Significant Difference (LSD) test at the 0.1%, 1%, and 5% significance levels, as appropriate (Steel et al., 1997).

Results & Discussion

Grain yield

Data on triticale grain yield in response to foliar Fe and Zn application under different K levels during the Rabi seasons of 2020–21 and 2021–22 are presented in Table 1. Statistical analysis revealed that K levels and foliar Fe and Zn applications significantly influenced grain yield, whereas the year of experimentation had no significant effect. Across both years, the highest grain yield (5385 kg ha^{-1}) was recorded with K application at 120 kg ha^{-1}, followed by 90 kg ha^{-1} K (4795 kg ha^{-1}), while the lowest yield (3613 kg ha^{-1}) was obtained at 60 kg ha^{-1} K. Renata et al. (2012) similarly reported improved biological and grain yields in winter triticale with increasing K application rates. In wheat, Hussain et al. (2004) and Khan et al. (2014 & 2015) also documented positive effects of K on biological yield, grain yield, and harvest index. Foliar Fe application also significantly affected grain yield. The application of 9 kg ha^{-1} Fe produced the highest grain yield (4866 kg ha^{-1}), which was significantly greater than the yield obtained with 6 kg ha^{-1} Fe (4550 kg ha^{-1}). The lowest grain yield (4377 kg ha^{-1}) was recorded at 3 kg ha^{-1} Fe. These findings are consistent with the reports of Gulser et al. (2019) and Hafeez (2021), who observed improvements in biological and grain yields with increasing Fe application rates. Similarly, Zn application at 6 kg ha^{-1} resulted in the highest grain yield (4722 kg ha^{-1}), although it remained statistically comparable to the yield recorded with 4 kg ha^{-1} Zn (4577 kg ha^{-1}). The lowest grain yield (4493 kg ha^{-1}) was observed at 2 kg ha^{-1} Zn. Mousavi et al. (2013) and Hafeez et al. (2021) reported enhanced biological and grain yields, along with improvements in other yield-related traits, with increasing Zn application in wheat. The untreated control plots, where K, Fe, and Zn were not applied, produced a significantly lower grain yield (2702 kg ha^{-1}) than the nutrient-treated plots, which recorded an average yield of 4598 kg ha^{-1}. The K×Fe interaction was significant at the 5% probability level, as illustrated in Fig. 2. Grain yield increased more markedly with K application from 60 to 120 kg ha^{-1} when Fe was applied at 9 kg ha^{-1}. At Fe application rates of 3 and 6 kg ha^{-1}, a sharper yield increase was observed as K increased from 60 to 90 kg ha^{-1}; however, a comparatively smaller improvement occurred with a further increase to 120 kg ha^{-1}. The maximum grain yield was obtained from the combined application of 120 kg ha^{-1} K and 9 kg ha^{-1} Fe. In contrast, the K×Zn, Fe×Zn, and K×Fe×Zn interactions did not significantly affect triticale grain yield.

Harvest index

Analysis of the data shows that K had significant while and Fe and Zn had no-significant effect on harvest index of triticale. Years of sowing of the experiments also showed no significant on harvest index. Averaged over the two years' data, plots where K was applied @ 120 kg ha^{-1}, recorded maximum harvest index of 39.6 % followed by plots supplied with 90 kg ha^{-1} K (38.8 %) while minimum harvest index of 34.3 % was noted in plots where K was applied @ 60 kg ha^{-1}. Renata et al (2012) reported increase in biological & grain yields in winter triticale due to increase in K levels. Hussain et al (2004) and Khan et al (2014 & 2015) reported that K increased biological & grain yields and harvest index of wheat. In control plots where K, Fe or Zn were not applied, significantly lower harvest index of 29.2 % was observed as compared with higher harvest index of 37.6 % in rest of the plots. Gulser et al. (2019) & Hafeez (2021) reported increase in biological and grain yields with increase in level of Fe. Mousavi et al (2013) and Hafeez et al (2021) reported increase in biological and grain yields along with other yield parameters of wheat with increase in the level of Zn. The interaction of K x Fe was found significant at 0.01% probability level for harvest index of triticale. As shown in Fig. 3, harvest index increased markedly with an increase in K application from 60 to 90 kg ha^{-1} across all three Fe levels, with the most pronounced response observed at 3 kg ha^{-1} Fe. However, a comparatively smaller increase in harvest index was recorded when K application was further increased from 90 to 120 kg ha^{-1} at Fe levels of 3, 6, and 9 kg ha^{-1}. The highest harvest index was observed with the combined use of K level of 120 kg ha^{-1} and any of the Fe levels ie. 3, 6 and 9 kg ha^{-1}. The rest of the interactions of the nutrients i.e. K x Zn, and Fe x Zn were observed to have no significant effect on harvest index.

Grain iron (Fe) content

K and foliar Fe significantly affected grain Fe content, while the year of experimentation also showed a significant effect. Averaged over the two years' data, plots where K was applied @ 120 kg ha^{-1}, recorded comparatively higher grain Fe content of 32.7 mg kg^{-1} followed by plots supplied with 90 kg ha^{-1} K (32.2 mg kg^{-1} while minimum grain Fe content of 30.6 mg kg^{-1} was noted under reduced K application. Awad-Allah and Elsokkary (2020) reported increase in the uptake of Fe in monocot and dicot plants due to increasing K levels. Similarly, plots where Fe was applied @ 9 kg ha^{-1} recorded higher grain Fe content of 32 mg kg^{-1} which was significantly higher from plots supplied with 6 kg ha^{-1} Fe (31.8 mg kg^{-1}). Significantly lower grain Fe content of 31.7 mg kg^{-1} was noted in plots where Fe was applied @ 3 kg ha^{-1}. Hafeez (2021) and Velu et al (2014) reported that grain Fe content increased with increase in sole application of Fe in wheat cultivars. In control plots where K, Fe or Zn were not applied, significantly lower grain Fe content of 28.3 mg kg^{-1} was observed as compared with higher grain Fe content of 31.8 mg kg^{-1} in rest of the plots where these nutrients were applied. The interactions of the nutrients i.e. K x Fe, K x Zn and Fe x Zn were observed to have no significant effect on grain Fe content of triticale. Hafeez et al (2021) reported sole application of Zn improved grain Zn content in wheat cultivars and had no effect on grain Fe content.

Grain zinc (Zn) content

Statistical evaluation confirmed that both Soil-applied K fertilization and foliar Zn treatment levels, both independently improved the Zn content within triticale grains. Foliar Fe application had no impact on grain Zn content. Years of sowing of the experiments were also significant for grain Zn content of triticale. Averaged over the two years' data, plots where K was applied @ 120 kg ha^{-1}, recorded maximum grain Zn content of 24.6 mg kg^{-1} followed by plots supplied with 90 kg ha^{-1} K (24.1 mg kg^{-1}) while lower grain Zn content of 22.9 mg kg^{-1} was noted under reduced K

incorporated. Bashir et al (2023) reported that uptake of Zn increased with increase in K level in wheat genotypes. Similarly, plots where Zn was applied @ 6 kg ha^{-1} recorded higher grain Zn content, considerably higher from plots supplied with 4 kg ha^{-1} Zn (23.8 mg kg^{-1}). Significantly lower grain Zn content of 23.6 mg kg^{-1} was noted in plots where Zn was applied @ 2 kg ha^{-1}. Hafeez et al (2021) reported sole application of Zn improved grain Zn content in wheat cultivars and had no effect on grain Fe content. In control plots where K, Fe or Zn were not applied, significantly lower grain Zn content of 19.6 mg kg^{-1} was observed as compared with higher grain Zn content of 23.9 mg kg^{-1} in rest of the plots where these nutrients were applied. A significant K×Zn interaction was observed for grain Zn content. As shown in Fig. 4, grain Zn increased markedly with K application from 60 to 90 kg ha^{-1} across all Zn levels, particularly at 2 kg ha^{-1} Zn. A comparatively smaller increase was observed with a further increase in K from 90 to 120 kg ha^{-1}. The highest grain Zn content was recorded with the combined application of 120 kg ha^{-1} K and 6 kg ha^{-1} Zn. The K×Fe, Fe×Zn, and K×Fe×Zn interactions were non-significant.

Grain protein content

Analysis revealed that K levels, and foliar Fe and Zn application had significant effects on grain protein content of triticale. Years of sowing of the experiments were also significant for grain protein content. Averaged over the two years' data, plots where K was applied @ 120 kg ha^{-1}, recorded maximum grain protein content of 13.66 % followed by plots supplied with 90 kg ha^{-1} K (13.49 %) while minimum grain protein content of 13 % was noted in plots where K was applied @ 60 kg ha^{-1}. Grain protein and wet gluten contents of wheat were improved due to increasing K levels (Khan et al., 2015). Quinlan et al (2017) also reported that grain protein and wet gluten contents increased with increase in K levels. Tababtabael and Ranjbar (2012) reported increase in grain protein content of triticale with increase in K quantity. Similarly, plots where Fe was applied @ 9 kg ha^{-1} recorded higher grain protein content of 13.48 % which was significantly higher from plots supplied with 6 kg ha^{-1} Fe (13.38 %). Significantly lower grain protein content of 13.28 % was noted in plots where Fe was applied @ 3 kg ha^{-1}. Habib (2009), Velu et al (2014) and Hafeez (2021) reported that grain protein and wet gluten content increased with increase in application of Fe in wheat. Similarly, plots where Zn was applied @ 6 kg ha^{-1} recorded higher grain protein content of 13.42 % which was significantly higher from plots supplied with 4 kg ha^{-1} Zn (13.39 %). Significantly lower grain protein content of 13.33 % was noted in triticale experimental plots where Zn was applied @ 2 kg ha^{-1}. Habib (2009), Tahir et al (2013), Mousavi et al (2013), Hafeez et al (2021) and many others reported that application of Zn improved grain protein and wet gluten content in wheat and other grains. In control plots where K, Fe or Zn were not applied, significantly lower grain protein content of 12.33 % was observed as compared with higher grain protein content of 13.38 % in rest of the plots. The K×Fe interaction significantly affected grain protein content at the 1% probability level. As portrayed in Fig. 5, grain protein content increased sharply with K application from 60 to 90 kg ha^{-1} across all Fe levels, particularly at 3 kg ha^{-1} Fe. A smaller increase was observed with further K application. The highest grain protein content was recorded at 120 kg ha^{-1} K with all Fe levels (3, 6, and 9 kg ha^{-1}). Similar results were reported by Awad-Allah and Elsokkary (2020) in wheat. The K×Zn and Fe×Zn interactions were non-significant.

Grain wet gluten content

K levels, and foliar Fe and Zn application levels had significant effects on grain wet gluten content of triticale. Years of sowing of the experiments were also significant for grain wet gluten content. Averaged over the two years' data, plots where K was applied @ 120 kg ha^{-1}, recorded maximum grain wet gluten content of 26.37 % followed by plots supplied with 90 kg ha^{-1} K (25.98 %) while

minimum grain wet gluten content of 24.93 % was noted in plots where K was applied @ 60 kg ha^{-1}. Hussain et al (2020) and Khan et al (2015) reported increase in the grain protein and wet gluten contents of wheat due to increasing K levels. Quinlan et al (2017) reported that grain protein and wet gluten contents increased with increase in K levels. Tababtabael and Ranjbar (2012) reported increase in grain protein content of triticale with increase in K quantity. Similarly, plots where Fe was applied @ 9 kg ha^{-1} recorded higher grain wet gluten content of 26.02 % which was significantly higher from plots supplied with 6 kg ha^{-1} Fe (25.75 %). Significantly lower grain wet gluten content of 2552 % was noted in triticale plots where Fe was applied @ 3 kg ha^{-1}. Habib (2009), Velu et al (2014) and Hafeez (2021) reported that grain protein and wet gluten content increased with increase in application of Fe in wheat. Similarly, plots where Zn was applied @ 6 kg ha^{-1} recorded higher grain wet gluten content of 25.84 % which was not significantly different from plots supplied with 4 kg ha^{-1} Zn (25.83 %). Significantly lower grain wet gluten content of 25.61 % was noted in triticale experimental plots where Zn was applied @ 2 kg ha^{-1}. Habib (2009), Tahir et al (2013), Mousavi et al (2013), Hafeez et al (2021) and many others reported that application of Zn improved grain protein and wet gluten content in wheat and other grains. In control plots where K, Fe or Zn were not applied, significantly lower grain wet gluten content of 23.9 % was observed as compared with higher grain wet gluten content of 25.8 % in rest of the plots. The K×Fe interaction had a significant effect ($P \leq 0.01$) on the grain wet gluten content of triticale. As illustrated in Fig. 6, raising the K rate from 60 to 90 kg ha^{-1} produced a marked increase in wet gluten content at all three Fe levels, the response being most pronounced at 3 kg Fe ha^{-1}. A further increase in K from 90 to 120 kg ha^{-1} resulted in a comparatively modest gain across all Fe levels (3, 6 and 9 kg ha^{-1}). The maximum wet gluten content was recorded where 120 kg K ha^{-1} was applied in combination with 9 kg Fe ha^{-1}. Awad-Allah and Elsokkary (2020) reported similar increase in wheat grains. The rest of the interactions of the nutrients i.e. KxZn, and FexZn were observed to have no significant effect on grain wet gluten content of triticale.

Conclusions and Recommendations

The study concluded that increasing K, Fe, and Zn levels improved GY, grain protein, and gluten contents of triticale. A significant K × Zn interaction was observed for grain Zn content, with the highest value recorded at 120 kg K ha^{-1} and 6 kg Zn ha^{-1}. The K×Fe interaction significantly affected GY, HI, grain protein, and gluten contents, with maximum values obtained at 120 kg K ha^{-1} and 9 kg Fe ha^{-1}. The Fe × Zn interaction was non-significant for all studied parameters. Based on these findings, application of 120 kg K ha^{-1}, 9 kg Fe ha^{-1}, and 6 kg Zn ha^{-1} is recommended to improve grain yield and quality of triticale. For interaction effects, 120 kg K ha^{-1} + 9 kg Fe ha^{-1} is recommended for improving GY and quality traits, while 120 kg K ha^{-1} + 6 kg Zn ha^{-1} is recommended for enhancing grain Zn content.

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Novelty Statement

"Agro-biochemical studies of triticale seed using foliar application of iron and zinc at various potassium levels" has been presented in this paper which has not been previously studied. This

research evaluated the effects of foliar application of iron and zinc at various potassium levels to improve grain yield and grain biochemistry of triticale.

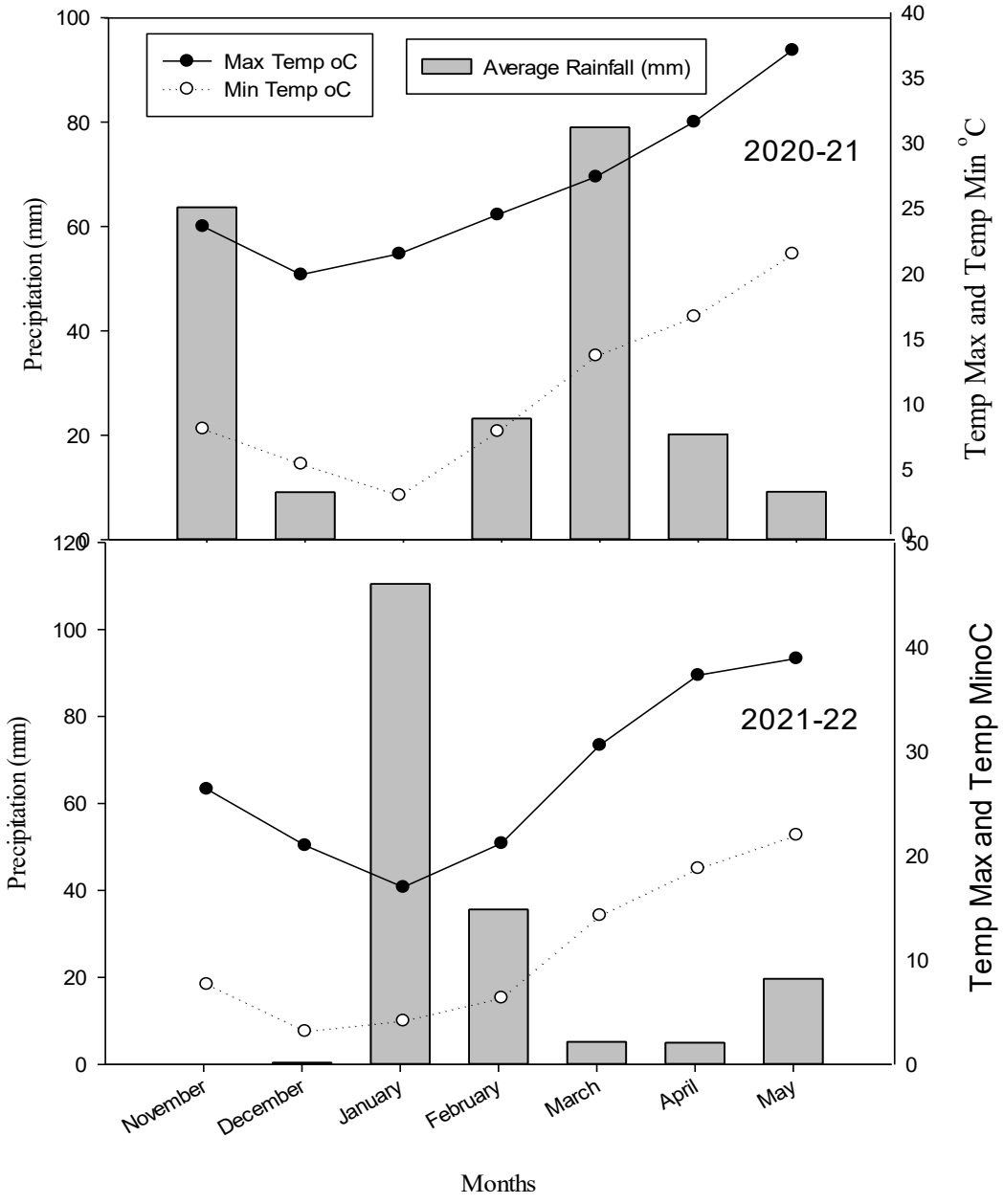


Fig. 1. Monthly average weather data (maximum and minimum temperature) of two year 2020-21 and 2021-2022 for the experimental location for the crop growth season (Pakistan Meteorological Department)

Table 1.Grain yield & harvest index of triticale as affected by foliar iron (Fe) and zinc (Zn) application with various potassium levels during the growing seasons of the year 2020-21 & 2021-22.

	Grain yield (kg/ha)			Harvest index (%)		
K levels (kg ha^{-1})	2020-21	2021-22	Mean	2020-21	2021-22	Mean
60	3628	3598	3613 c	34.2	34.4	34.3 b

90	4839	4751	4795 b	39.4	38.3	38.8 a
120	5404	5365	5385 a	39.7	39.6	39.6 a
$LSD_{(0.05)}$ for K			188			2.6
Fe levels (kg ha^{-1})						
3	4385	4368	4377 b	37.1	36.8	37.0
6	4586	4514	4550 b	37.9	37.5	37.7
9	4899	4833	4866 a	38.3	37.9	38.1
$LSD_{(0.05)}$ for Fe			188			NS
Zn levels (kg ha^{-1})						
2	4503	4483	4493 b	37.6	37.3	37.5
4	4608	4546	4577 ab	37.6	37.2	37.4
6	4759	4685	4722 a	38.1	37.7	37.9
$LSD_{(0.05)}$ for Zn			188			NS
Control (no K, Fe or Zn applied)	2739	2664	2702 b	29.0	29.3	29.2 b
Rest of the plots	4624	4572	4598 a	37.8	37.4	37.6 a
Significance			***			***
Interaction			Sig			Sig
K x Fe			*			***
K x Zn			NS			NS
Fe x Zn			NS			NS
K x Fe x Zn			NS			NS

Means in the same category of rows or columns followed by different letters are significantly different from each other. * & *** means significance at 5% & 0.1% level of probability, respectively.

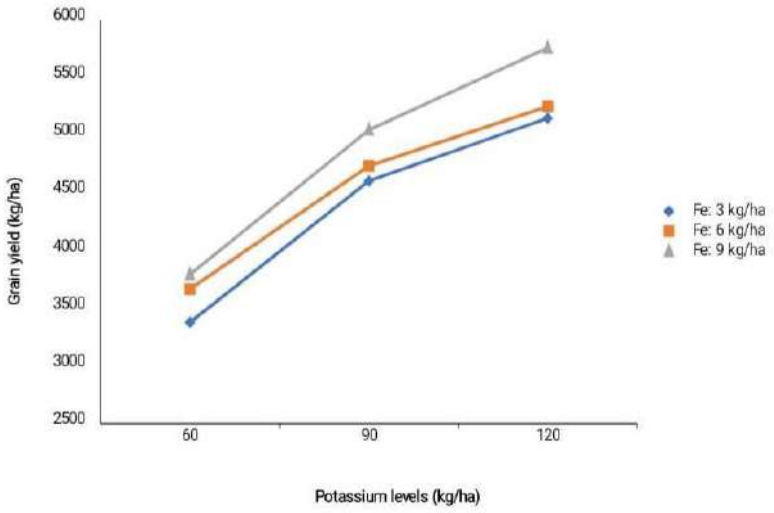


Fig.2. Interaction of potassium and iron (K x Fe) for grain yield (kg ha^{-1}) of triticale averaged over two years (rabi season 2020-21 & 2021-22).

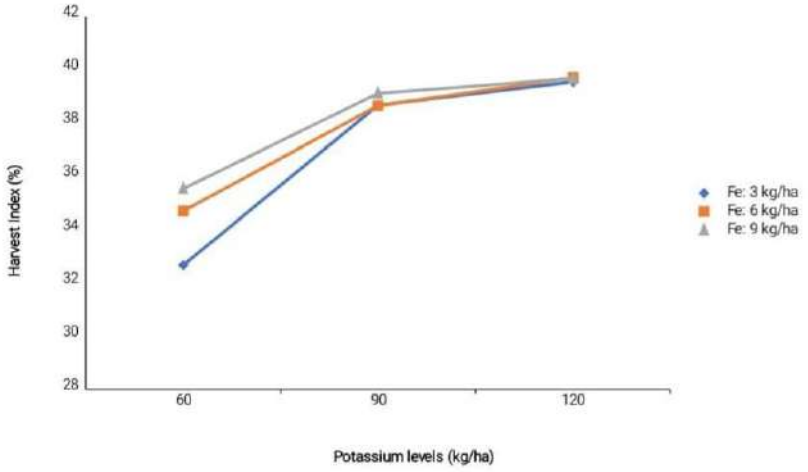


Fig.3. Interaction of potassium and iron (K x Fe) for harvest index (%) of triticale averaged over two years (rabi season 2020-21 & 2021-22).

Table 2.Grain Fe content & Grain Zn content of triticale as affected by foliar iron (Fe) and zinc (Zn) application with various potassium levels during the growing seasons of the year 2020-21 & 2021-22.

	Grain Fe content (mg kg^{-1})			Grain Zn content (mg kg^{-1})		
K levels (kg ha^{-1})	2020-21	2021-22	Mean	2020-21	2021-22	Mean
60	30.6	30.5	30.6 c	23.3	22.4	22.9 c
90	32.4	32.0	32.2 b	24.4	23.9	24.1 b
120	32.7	32.7	32.7 a	24.7	24.4	24.6 a
$LSD_{(0.05)}$ for K			0.1			0.1
Fe levels (kg ha^{-1})						
3	31.7	31.6	31.7 b	24.1	23.6	23.9
6	31.9	31.7	31.8 b	24.2	23.6	23.9
9	32.0	31.9	32.0 a	24.1	23.6	23.8
$LSD_{(0.05)}$ for Fe			0.1			NS
Zn levels (kg ha^{-1})						
2	31.9	31.7	31.8	23.8	23.3	23.6 c
4	31.9	31.8	31.8	24.2	23.5	23.8 b
6	32.0	31.7	31.8	24.5	23.9	24.2 a
$LSD_{(0.05)}$ for Zn			NS			0.1
Control (no K, Fe or Zn applied)	28.5	28.0	28.3 b	20.1	19.0	19.6 b
Rest of the plots	31.9	31.7	31.8 a	24.1	23.6	23.9 a
Significance			***			***
Interaction			Sig			Sig
K x Fe			NS			NS
K x Zn			NS			**
Fe x Zn			NS			NS
K x Fe x Zn			NS			NS

Means in the same category of rows or columns followed by different letters are significantly different from each other. * & *** means significance at 5% & 0.1% level of probability, respectively.

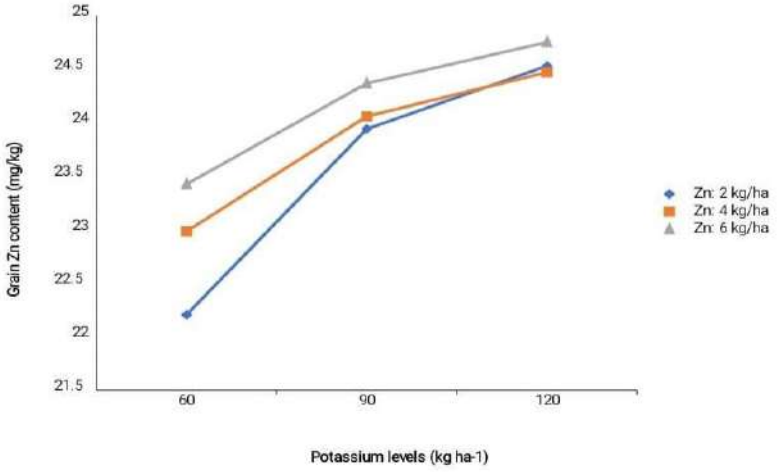


Fig.4. Interaction of potassium and zinc (K x Zn) for grain Zn content (mg kg^{-1}) of triticale averaged over two years (rabi season 2020-21 & 2021-22).

Table 3.Grain protein & Grain wet gluten content of triticale as affected by foliar iron (Fe) and zinc (Zn) application with various potassium levels during the growing seasons of the year 2020-21 & 2021-22.

	Grain protein content (%)			Grain wet gluten content (%)		
K levels (kg ha^{-1})	2020-21	2021-22	Mean	2020-21	2021-22	Mean
60	13.08	12.91	13.00 c	25.13	24.73	24.93 c
90	13.43	13.54	13.49 b	25.83	26.13	25.98 b
120	13.55	13.77	13.66 a	26.19	26.55	26.37 a
$LSD_{(0.05)}$ for K			0.05			0.11
Fe levels (kg ha^{-1})						
3	13.28	13.28	13.28 c	25.48	25.55	25.52 c
6	13.36	13.39	13.38 b	25.70	25.79	25.75 b
9	13.42	13.55	13.48 a	25.97	26.07	26.02 a
$LSD_{(0.05)}$ for Fe			0.05			0.11
Zn levels (kg ha^{-1})						
2	13.29	13.36	13.33 c	25.55	25.66	25.61 b
4	13.38	13.40	13.39 b	25.78	25.88	25.83 a
6	13.39	13.46	13.42 a	25.82	25.87	25.84 a
$LSD_{(0.05)}$ for Zn			0.05			0.11
Control (no K, Fe or Zn applied)	12.35	12.30	12.33 b	24.10	23.60	23.9 b
Rest of the plots	13.35	13.41	13.38 a	25.72	25.80	25.8 a
Significance			***			***
Interaction			Sig			Sig
K x Fe			**			**

K x Zn			NS			NS
Fe x Zn			NS			NS
K x Fe x Zn			NS			NS

Means in the same category of rows or columns followed by different letters are significantly different from each other. * & *** means significance at 5% & 0.1% level of probability, respectively.

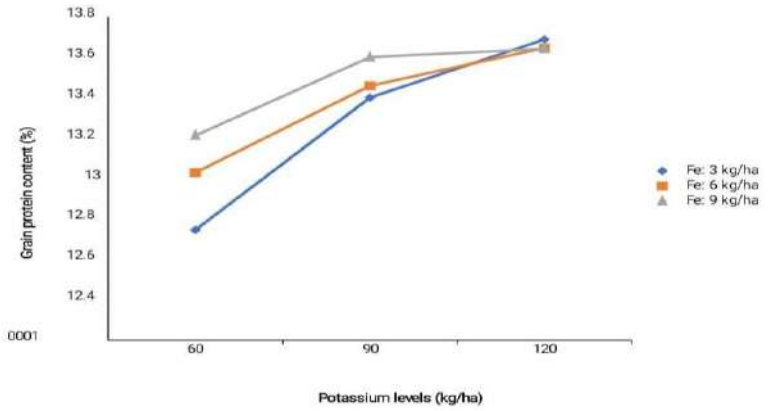


Fig.5. Interaction of potassium and iron (K x Fe) for grain protein content (%) of triticale averaged over two years (rabi season 2020-21 & 2021-22).

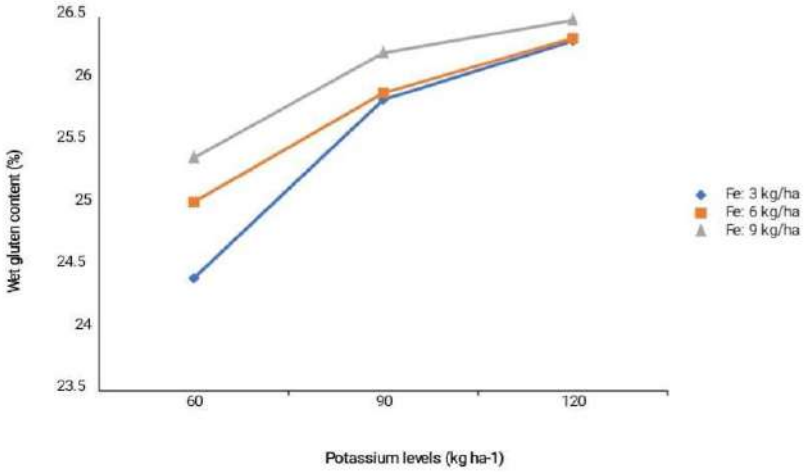


Fig.6. Interaction of potassium and iron (K x Fe) for grain wet gluten content (%) of triticale averaged over two years (rabi season 2020-21 & 2021-22).

Author's Contribution

Asifur Rehman: Principal author, conducted field trial, wrote the manuscript draft.

Inamullah: Supervised the trial, provided guidance for execution of this research experiment and proof reading.

Niamatullah: Provided lab facilities and technical guidance regarding analyzing the seed chemical parameters.

Conflict of interest

The authors have declared no conflict of interest.

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