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Comparative Effects of Hydropriming and Iron Priming on Germination and Seedling Morphophysiological Attributes of Stay-Green Wheat

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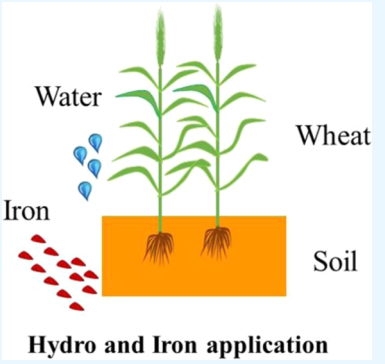
ABSTRACT: Seed priming is considered to play an essential role in the overall improvement of agricultural crops. The current research work was carried out in order to investigate the comparative effects of hydropriming and iron priming on the germination behavior and morphophysiological attributes of wheat seedlings. The experimental materials consisted of three wheat genotypes including a synthetically derived wheat line (SD-194), stay-green wheat genotype (Chirya-7), and conventional wheat variety (Chakwal-50). The treatments included hydro (distilled and tap water)- and iron priming (10 and 50 mM) of wheat seeds for 12 h duration. Results indicated that both priming treatment and wheat genotypes exhibited highly different germination and seedling characteristics. These included germination percentage, root volume, root surface, root length, relative water content, chlorophyll content, membrane stability index, and chlorophyll fluorescence attributes. Furthermore, the synthetically derived line (SD-194) was the most promising in majority of the studied attributes by exhibiting a high germination index (2.21%), root fresh weight (7.76%), shoot dry weight (3.36%), relative water content (19.9%), chlorophyll content (7.58%), and photochemical quenching coefficient (2.58%) when compared with stay-green wheat (Chirya-7). The study also found that hydropriming with tap water and priming wheat seeds with low concentrations of iron yielded better results when a comparison was made with wheat seeds primed at high concentrations of iron. Therefore, wheat seed priming with tap water and iron solution for 12 h is recommended for optimum wheat improvement. Furthermore, current findings suggest that seed priming may have the prospect of an innovative and user-friendly approach for wheat biofortification with the aim of enhanced iron acquisition and accumulation in grains.



INTRODUCTION

Wheat (*Triticum aestivum* L.) is the most important crop in the world after rice and maize with a total production of 765 million tons, covering 216 million hectares.[1−3] The wheat grain contains 20% food calories and 55% carbohydrates and is a chief source of food across the globe.[4−8] However, besides the risks of climate change and increased population, wheat producers and breeders are facing bigger challenges of malnutrition and food insecurity.[9−11] Furthermore, it is believed that a substantial increase in the production of agricultural crops after green revolution has led to a gradual decline in the nutritional quality of elite high-yielding cereal crops, particularly wheat.[12−15] An exceptional rise in atmospheric CO_2 up to 420 ppm has directly affected the carbon assimilation, the result being decreased accumulation of both grain micro- and macronutrients.[16−19]

At least 18 mineral nutrients are vital and required for the proper growth, development, and reproduction of plants as well as humans.[20,21] Iron (Fe) and zinc (Zn) with atomic numbers 56 and 30, respectively, are among the important transition metals on earth, which have been classified as essential micronutrients needed in small quantities and play regulatory, catalytic, signaling, and structural roles.[22] Fe has a vital role in plant physiological and biochemical processes, and many electron transport reactions and plant hormone synthesizing machinery need iron as cofactor for their accomplishment.[23,24] Leaf chlorosis is a condition during which a young leaf is turned yellow, which results mainly from the deficiency of iron. However, if it is present in high concentrations, it can lead to different types of oxidative stresses including OH^- (hydroxyl radicals), O^{-2} (superoxide radicals), and H_2O_2, i.e., hydrogen peroxide.[6,25,26] Deficiency of Fe and Zn is prevalent in both humans and crops, for instance, anemia is among the major challenges in developing

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Table 1. List and Pedigree of Wheat Genotypes Studied during Current Research Work[a]

acc. no.	genotype	pedigree
AS-01	Chirya-7	CHINESE-SPRING/AG.CU//GLENNSON-81/3/ALONDRA/ PAVON-76/4/NINGMAI-4/OLESEN//ALONDRA/YANGMAI-4[1281]
AS-02	Chakwal-50	ATTILA/3/HUI/CARC//CHEN/CHTO/4/ATTILA
AS-03	SD-194	CHAPIO/INQALAB 91/4/PICUS/3/KAUZ*2/BOW//KAUZ

[a]Source: Bioresources Conservation Institute (BCI), National Agriculture Research Centre, Islamabad, Pakistan.

countries, which is due to the consumption of iron-deficient diet; hence, there is an urgent need to develop an appropriate strategy to provide ample iron in the daily diet for addressing the problem.[27−30] In cereal crops, an increase in the concentrations of iron and zinc may result in better crops and it may also improve human health globally, especially micronutrients, which are beneficial for enhanced wheat grain quality, biomass, harvest index, and plant height.[31−35]

Different approaches that are currently in practice for improving the grain quality include reducing time from seed sowing to seedling emergence; minimizing the deleterious effects of different biotic and abiotic stresses at the germination stage, and different seed treatments such as pelleting, coating of seeds, or seed priming.[11,36−40] Seed priming treatment has been proven to have several benefits such as seed dormancy breakdown, rapid and uniform emergence, deeper roots, enhanced root growth, stand establishment, better competition with weeds, formation of sexual organs, early flowering, resistance to different climatic conditions, and better grain production in wheat.[41−45] It is considered an innovative and user-friendly approach for wheat biofortification, which increases iron acquisition and accumulation in grains. In the seed priming technique, seeds are soaked in solutions of low water potential to enhance pregerminative metabolic activities.[46−48] It may be categorized as hydro-, osmo-, thermo-, halo-, and matrix priming.[49] Every priming technique responds differently in different crops and has mostly been successfully applied for their beneficial effects in many agronomic crops including soybean, wheat, sunflower, and rice. Among these, osmopriming can strengthen the antioxidant system to increase seed germination potential, resulting in an enhanced stress tolerance capacity in germinated seedlings.[32,50] It is generally believed that changes in wheat seeds, due to priming agents including water and iron, may have positive prospects in seedling establishment and subsequent physiological performances. Furthermore, seed priming with different media has regularly been employed for the general improvement of seed crops including wheat. However, the comparison of hydropriming and iron priming needs to be investigated, as very little work in this perspective has been done so far. The present study was therefore aimed at studying the germination characteristics and morphophysiological and germination performance of wheat seedlings through hydropriming and iron priming.

MATERIALS AND METHODS

Plant Materials. The present study was conducted at the Centre for Plant Sciences and Biodiversity, University of Swat, in order to investigate the comparative effects of hydropriming and iron priming on the germination behavior and morphophysiological characteristics of wheat. The research was carried out on three wheat genotypes including a synthetically derived wheat advanced line (SD 194), stay-green drought-tolerant wheat cultivar (Chirya-7), and conventional wheat variety (Chakwal-50). The selection was based on two years of detailed characterizations of a core collection of 325 diverse bread wheat for drought stress effects on stay green and chlorophyll fluorescence with focus on it yield characteristics. The experimental genotypes originated through Wheat Wide Crosses and Cytogenetics Program at National Agriculture Research Centre, Islamabad, Pakistan (pedigree is given in Table 1).

Seed Priming Experiment. Clean and healthy seeds of all wheat genotypes were primed with iron (Fe) as iron sulfate and water (distilled and tap). Briefly, seeds of each genotype were separately soaked in a 100 mL solution of $FeSO_4$ (10 and 50 mM) and distilled and tap water in a beaker for 12 h. Ten seeds from each medium were then transferred to Petri dishes containing filter paper and were kept completely in the dark for 72 h. All the primed seeds were observed on a daily basis, and germination was considered to have taken place when the radical reached 2 mm length.[51] The germination percentage (GP; which is the percentage of number of germinated seeds to total seeds sown multiplied by 100) of seeds was obtained when the experiment was completed. Mean germination time (MGT) was measured when the experiment was terminated. Seed germination index (SGI) was measured as described in the Association of Official Seeds Analyst (1983)

$$GI = \frac{\sum TiNi}{S}$$

where Ti represents the number of days counted after planting, Ni is the number of germinated seeds on day 1, and S is the total number of seeds sown.

The experiment was continued further by transferring three uniform-sized plants from each replicated treatment to pots for further morphological and physiological investigation. The growing medium was a mixture of mineral soil collected from a nearby field and commercial organic soil, which comprised deep, well-drained and moderately fine-textured particles. It was slightly calcareous and nonsaline, with pH 8.1 and an electrical conductivity (EC) of 0.24 dS/m. The soil was amended with a commercial organic soil mix with a ratio of 70:30 (v/v) and placed into 2.5 L (24 × 30 cm^2) pots, ensuring a bulk density of 1.2 $g{\cdot}cm^{-3}$. Pots were arranged in a randomized complete block design (RCBD). In pot soil, $FeSO_4$ (10 and 50 mM) and H_2O (control) were employed for watering purposes on a need basis. All the pots were kept under light at room temperature. The experiment was continued for up to 45 d until each plant attained maximum shoot length. Images were taken with a digital camera (Meiji infinity DK-5000, Japan). The root surface (RS), root volume (RV), root diameter (RDia), and root length (RL) were measured using SmarRoot software,[52−54] while shoot length (SL) was determined with the help of a scale. Sampling was then accomplished for further physiological analysis.

Leaf Relative Water Content (RWC). For the RWC, the flag leaf samples were taken from all the treatment plants and then cut immediately into small discs (0.8 cm diameter). These discs were weighed (fresh weight, FW). The leaves were then

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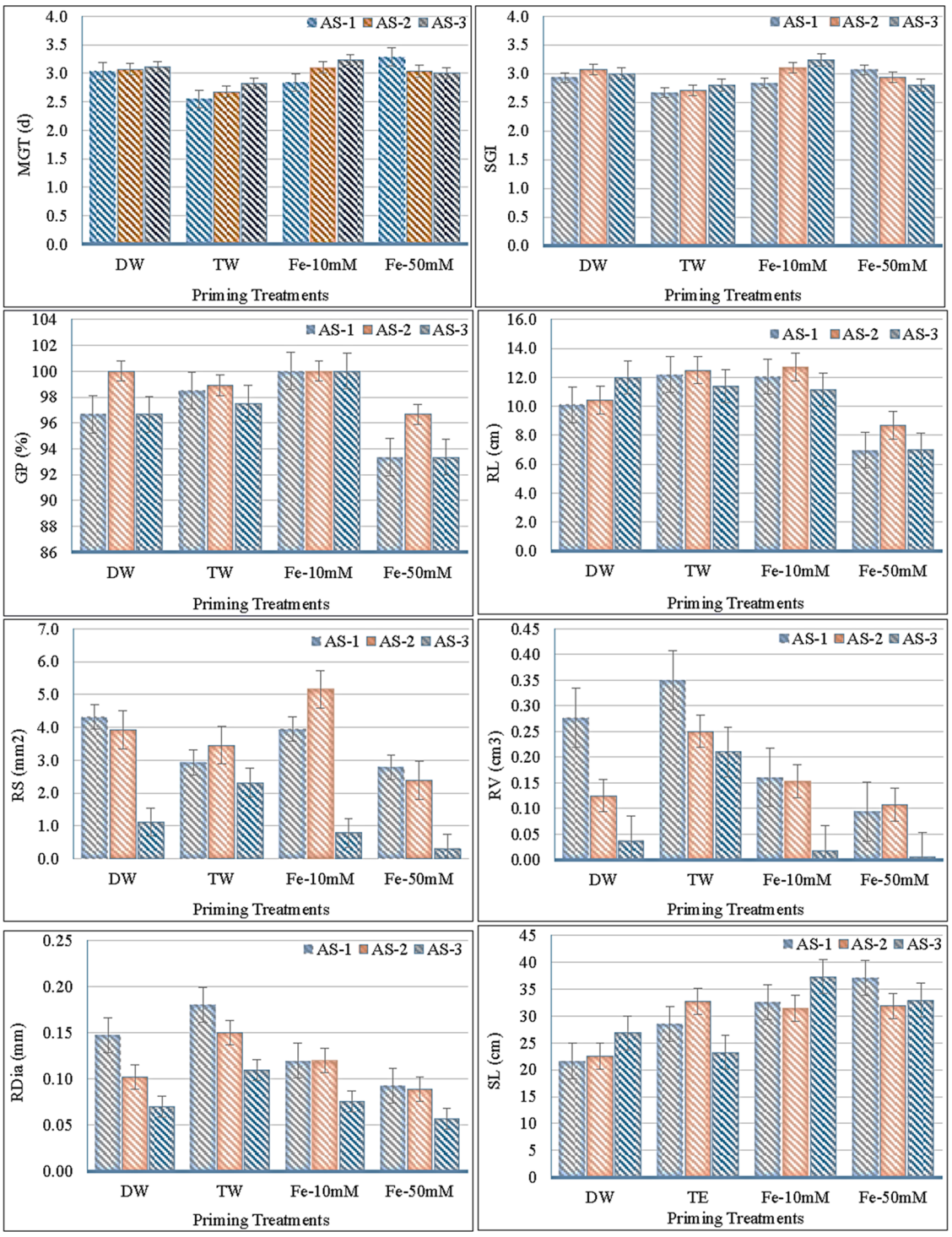

Figure 1. Mean germination time (MGT), seed germination index (SGI), germination percentage (GP, %), root length (RL, cm), root surface (RS, mm^2), root volume (RV, cm^3), root diameter (RDia, mm), and shoot length (SL, cm) of the studied wheat lines under different priming media. Error bars represent standard deviation (n = 3).

kept in test tubes and floated in distilled water in the dark at 4 °C for 24 h and weighed again (turgid weight, TW). Finally, the leaves were dried in an oven at 80 °C for 48 h, and the weight was determined again (dry weight, DW). Measurement of the relative water content was accomplished as previously reported.[55]

Chlorophyll Analysis. To measure the content of photosynthetic pigments (chlorophyll a, chlorophyll b, and total chlorophyll), fresh leaves were collected from each pot, weighed, wrapped in aluminum foil, and then kept in a freezer for 72 h. The method of Hiscox and Israelstam[56] was employed for chlorophyll determination with minor modifications. For this purpose, all the samples were kept in test tubes containing 5 mL of dimethyl sulfoxide (DMSO) and heated in a water bath at 65 °C for 35 min. Absorbance of all the sample extracts was then taken using a spectrophotometer (BMS-6702) at 645 and 663 nm wavelengths. The contents of

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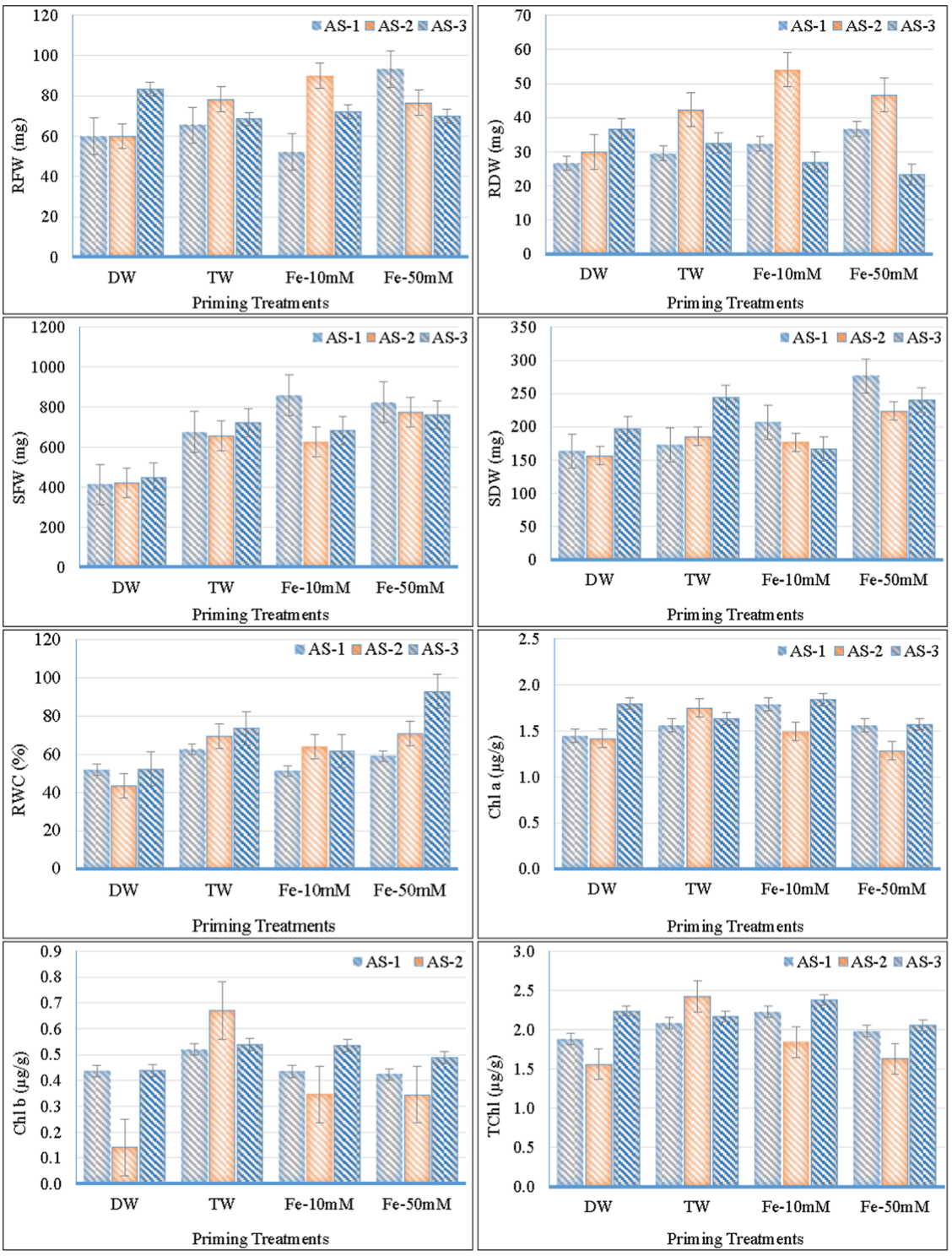

Figure 2. Root fresh (RFW, g) and dry (RDW) weight and shoot fresh (SFW, g) and dry (SDW) weight, relative water content (RWC, %), chlorophyll contents (Chla, Chlb, and TChl, μg/g) of the studied wheat genotypes under different priming media. Error bars represent standard deviation (n = 3).

these photosynthetic pigments were then calculated using the following equations of Arnon's.[57]

$$\text{Chla} = 0.0127\, A_{663} - 0.00269\, A_{645}$$

$$\text{Chlb} = 0.0029\, A_{663} - 0.00468\, A_{645}$$

$$\text{total Chla} = 0.0202\, A_{663} - 0.00802\, A_{645}$$

Membrane Stability Index (MSI). The study of Sairam[58] was followed for the determination of the leaf MSI. Briefly, two sets of about 100 mg of leaf tissue were taken in test tubes and dipped in 10 mL of distilled water. One set was heated in a water bath for 30 min at 40 °C, and the other was boiled for 10 min at 100 °C. Electrical conductivity of both solutions was recorded on a conductivity bridge, i.e., C1 and C2 (Elico,

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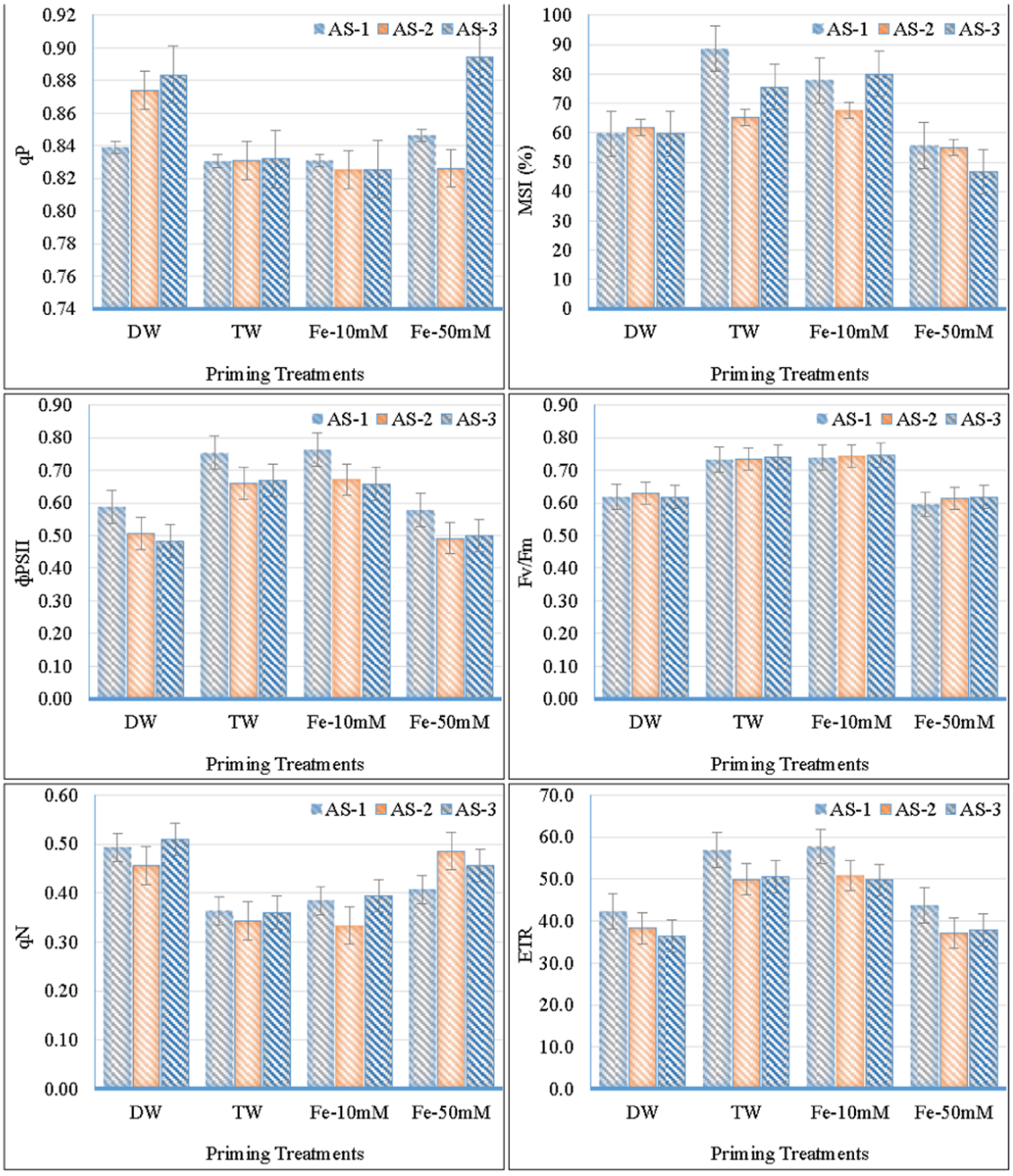

Figure 3. Membrane stability index fresh (MSI, %), photochemical quenching coefficient (qP), actual quantum yield of PSII electron transport (ϕPSII), maximal quantum yield of PSII photochemistry (FV/Fm), non-photochemical quenching coefficient (qN), and electron transport rate (ETR) of the studied wheat genotypes under different priming media. Error bars represent standard deviation (n = 3).

CM183EC-TDS analyzer, India) and MSI was calculated as MSI = [1 − (C1/C2)] × 100.

Measurement of Chlorophyll Fluorescence. The study of Genty et al.[59] was followed for the measurement of chlorophyll fluorescence attributes through a photosynthetic yield analyzer (Mini-PAM, Walz, Effeltrich, Germany), while fluorescence nomenclature was according to Maxwell and Johnson.[60] A 0.8 s saturating pulse of 8000 μmol m^{-2} s^{-1} in dark-adapted leaves was used to measure the maximum fluorescence (Fm) with all PSII reaction centers closed, while in light-adapted leaves, a second saturating pulse of 8000 μmol m^{-2} s^{-1} was applied again.

Statistical Analysis. The experimental design employed was a two-factor factorial RCBD with three replications, comprising two treatment factors, which included three wheat genotypes (Chirya-1, SD-194, Chakwal-50) and four priming media (tap and distilled water, Fe-10 mM and Fe-50 mM). The investigated germination, morphological, and physiological attributes were then subjected to ANOVA using STATISTIX 8.1 (Analytical Software, Tallahassee FL).[61] Comparison of means among experimental wheats for the studied physiological traits was accomplished using standard deviation and Fisher's least significant difference (p < 0.01 and 0.05).

RESULTS

Priming Effects on Germination and Seedling Attributes. Responses regarding germination percentage

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(GP), germination time, (GT), seed germination index (SGI), root surface (RS), volume (RV), diameter (RDia), length (RL), and shoot length (SL) are shown in Figure 1. A maximum GT of 3.04 was observed in SD-194, which was followed by Chakwal-50 (2.97) and Chirya-7 (2.93). Similarly, a SGI of 2.95 was observed in Chakwal-50, which was followed by SD-194 (2.94), while the lowest SGI of 2.88 was recorded in Chirya-7. A mean maximum RS of 3.73 mm^2 was noted in Chakwal-50, followed by 3.49 mm^2 in Chirya-7; however, the lowest (1.13 mm^2) was recorded in SD-194 (Figure 1). It is evident from the figure that a mean maximum GP of 98.9% was noted in Chakwal-50, while a GP of 97.3% was observed in Chirya-7 with 96.9% in SD-194. It is also evident from the figure that a mean maximum RV of 0.22 cm^3 was reported in Chirya-7, which was followed by 0.16 cm^3 in Chakwal-50, while the lowest RV of 0.07 cm^3 was noted in SD-194. Similarly, a mean maximum RDia of 0.13 mm was measured in Chirya-7, while the lowest RDia of 0.08 mm was recorded in SD-194. The result regarding root length showed that a mean maximum RL of 11.08 cm was noted in Chakwal-50, while the lowest RL of 10.33 cm was recorded in Chirya-7. Furthermore, a mean maximum SL of 30.05 cm was measured in SD-194 followed by Chirya-7, in which a SL of 29.93 cm was reported, while the least SL of 29.61 cm was observed in genotype Chakwal-50.

Results regarding root fresh (RFW) and dry (RDW) weight and shoot fresh (SFW) and dry (SDW) weight are shown in Figure 2. Results revealed that a maximum RFW of 76.3 mg was observed in Chakwal-50, followed by 73.5 mg in SD-194, while the least (67.8 mg) was recorded in Chirya-7. Furthermore, a mean maximum SFW of 692 mg was recorded in Chirya-7 followed by 654 mg in SD-194, while the lowest SFW of 618 mg was noted in Chakwal-50. Furthermore, a mean maximum MGT of 3.1 was recorded in Fe-50 mM-primed seeds followed by priming with distilled water (3.07), while the least (2.68) was noted in seeds primed with tap water. Similarly, a mean maximum GI of 3.06 was recorded in seeds primed in Fe-10 mM, while the lowest (2.70) was measured in seeds with tap water. Similarly, a mean maximum RS of 3.30 mm^2 was seen in seeds primed with Fe-10 mM, which was followed by distilled water (3.12 mm^2)- and tap water-primed seeds (2.90). However, the least mean RS of 1.83 mm^2 was measured in Fe-50 mM-primed seeds. Our results regarding GP revealed that the maximum GP of 100.0% was observed in Fe-10 mM-primed seed, while the lowest GP of 94.4% was seen in those plants treated with Fe-50 mM. Similarly, a mean maximum RWC of 74.2% was observed in Fe-50 mM followed by 68.5% in those treated with tap water, while the mean lowest RWC of 58.9% was measured in those primed in Fe-50 mM solution. Likewise, a mean maximum RL of 12.0 cm was observed in tap water-primed plants, which was followed by plants treated with Fe-10 mM (11.9 cm). A mean maximum SL of 33.9 cm was observed in Fe-50 mM followed by those treated with Fe-10 mM, in which a SL of 33.7 cm was observed, while the lowest mean SL of 23.6 cm was measured in those treated with distilled water. A mean maximum RV of 0.27 cm^3 was observed in tap water-primed plants, which was followed by those primed with distilled water, while the least (0.11) was noted in plants primed with Fe-10 mM. Moreover, the lowest RV of 0.07 cm^3 was observed in Fe-50 mM-primed plants. Likewise, a mean maximum RDia of 0.11 mm was observed in tap water-treated plants, while a minimum RDia of 0.08 mm was seen in those plants treated with Fe-50 mM.

Priming Effects on the Relative Water Content and Photosynthetic Pigments. Results regarding relative water content (RWC) and chlorophyll contents (Chla, Chlb, TChl) are shown in Figure 2. A mean maximum RWC of 70.1% was recorded in SD-194, which was followed by Chakwal-50 (61.9%), while the lowest RWC of 56.2% was observed in Chirya-7. It is evident from the figure that a mean maximum Chla of 1.71 μg/g was observed in SD-194, which was followed by Chirya-7, in which Chla was measured as 1.59 μg/g, while the lowest (1.49 μg/g) was noted in Chakwal-50. Similarly, a mean maximum Chlb of 0.50 μg/g was recorded in SD-194, which was followed by Chirya-7 and Chakwal-50 (0.45 μg/g and 0.37 μg/g, respectively). Furthermore, a mean maximum TChl of 2.21 μg/g was recorded in SD-194, which was followed by Chirya-7 (2.04 μg/g), while the lowest TChl of 1.86 μg/g was measured in Chakwal-50. The effect of the priming treatment on photosynthetic pigments including chlorophyll a (Chla), chlorophyll b (Chlb), and total chlorophyll (TChl) revealed that a mean maximum Chla of 1.71 μg/g was measured in Fe-10 mM-treated plants, followed by 1.65 μg/g in plants treated with tap water, while the lowest Chla of 1.47 μg/g was measured in Fe-50 mM. Likewise, the mean maximum TChl of 2.22 μg/g was observed in plants treated with tap water, which was followed by 2.14 μg/g in plants primed in Fe-10 mM, while the lowest TChl of 1.89 μg/g was observed in plants primed with Fe-50 mM.

Results related to membrane stability index and chlorophyll fluorescence attributes are shown in Figure 3. Membrane stability was noted to be the highest, i.e., 70.9% in Chirya-7, while it was the lowest in Chakwal-50 (62.4%). A mean maximum ϕPSII (actual quantum yield of PSII electron transport) of 0.670 was recorded in Chirya-7, which was followed by Chakwal-50 and SD-194 (0.58). Similarly, it is evident from the figure that a mean maximum FV/Fm (maximal quantum yield of PSII photochemistry) of 0.68 was observed in SD-194 and Chakwal-50, while the lowest was noted in Chirya-7. Also, a mean maximum qP (photochemical quenching coefficient) of 0.86 was recorded in SD-194, while the least qN (non-photochemical quenching coefficient) was recorded in Chirya-7. Regarding the effects of priming treatment on chlorophyll fluorescence attributes, it was revealed that a mean maximum ETR (electron transport rate) of 52.7 was measured in Fe-10 mM-treated plants, followed by 52.4 in plants treated with tap water, while the lowest (38.9) was measured in plants primed in distilled water. Likewise, the mean maximum ϕPSII of 0.70 was observed in plants treated with Fe-10 mM, while the lowest 0.52 was observed in plants primed with Fe-50 mM.

Analysis of variance of the studied germination, morphological, and physiological attributes using different priming media is given in Tables 2 and 3. The analysis indicated that the studied wheat genotypes as well as different priming media were highly different regarding GP, RS, RDia, RWC, Chlb, ϕPSII, and ETR. The wheat genotypes did not show significant differences for the rest of the studied attributes. Similarly, the employed priming treatment exhibited maximum differences with respect to GI, GT, RL, RS, RDia, SL, SFW, RWC, Chla, Chlb, TChl, MSI, ϕPSII, Fv/Fm, qN, and ETR. Furthermore, the interaction between wheat genotypes and priming treatment led to high differences regarding GT and MSI. However, this interaction yielded no significant differences for the rest of the studied characteristics.

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Table 2. Mean Squares of Germination and Morphological Traits among the Studied Wheat Genotypes[a]

SOV	DF	GP	GI	MGT	RDW	RFW	RL	RS	RV	RDia	SL	SDW	SFW
gen	2	14.63*	0.025^{NS}	0.067^{NS}	494.3^{NS}	135.2^{NS}	2.51^{NS}	19.59***	0.184^{NS}	0.008*	0.087^{NS}	2209.2^{NS}	4353.0^{NS}
treat	3	48.75^{NS}	0.186**	0.444**	119.2^{NS}	1207.4^{NS}	37.64***	3.03*	0.118^{NS}	0.009**	197.3***	9676.6^{NS}	221181***
gen × treat	6	3.07^{NS}	0.059^{NS}	0.087**	155.1^{NS}	548.2^{NS}	1.96^{NS}	1.30^{NS}	0.071^{NS}	0.001^{NS}	47.35^{NS}	2362.9^{NS}	8427.0^{NS}
error	22	21.21	0.030	0.021	257.4	1209.4	4.06	0.67	0.062	0.002	23.36	3535.7	16914.0

[a]DF, degree of freedom; GP, germination percentage; GI, germination index; MGT, mean germination time; RDW, root dry weight; RFW, root fresh weight; RL, root length; RS, root surface; RDia, root diameter; SL, shoot length; *, **, and ***, statistical significance by employing Fisher's LSD at 0.05, 0.01, and 0.001 α levels, respectively; and NS, nonsignificant.

DISCUSSION

Iron plays an important role in fundamental biochemical processes including chlorophyll synthesis, mitochondrial respiration, and nitrogen fixation. It is also a basic component of many important enzymes such as catalase, ferredoxin, superoxide dismutase, and peroxidase and hence may be a limiting factor for plant biomass production and quality.[62−68] Present research work focused on the potentiality of hydropriming and iron priming in terms of its effect on germination and morphophysiological responses of the studied wheats, which exhibited enhanced germination and early primary growth in young seedlings. Hydropriming specifically with tap water and priming with Fe-10 mM resulted in quicker MGT, high GP, better GI, ϕPSII, Fv/Fm, qP, and ETR along with other investigated morphological and physiological attributes. Regarding GP, the experimental wheats depicted maximum germination in seeds primed with Fe-10 mM. Similarly, minimum germination time was observed in the seeds of Chirya-7 (stay-green wheat cultivar) primed with Fe-10 mM, with the maximum being observed in those primed with Fe-50 mM. Regarding seedling RL, the maximum (12.7 cm) was observed in Chakwal-50 (rain-fed cultivar) primed with Fe-10 mM, while the minimum was recorded in the Chirya-7 wheat seedling primed with Fe-50 mM. Similar findings of wheat seed speedy germination after soaking for 12 h in water have been reported.[43] Previously, 85% germination of wheat seeds as a result of hydropriming for 12 h was demonstrated.[69] In the current study, 100% germination percentage, maximum mean germination time, high root length, and enhanced chlorophyll content were attained by some of the genotypes, which could be due to difference in the genetic background of the studied wheat genotypes and the optimal priming duration of 12 h as previously reported.[36] The same has previously been reported for other crops also, including maize, beans, finger millet, sorghum, pearl millet, rice, sorghum, maize, and cotton.[48] This effect could be credited to the leakage of germination inhibitors into the priming solution and seed reserve premobilization during the priming period.[70] Our results are generally in accordance with Giri and Schillinger,[71] who previously stated that soaking wheat seeds in water delivered better germination performance than that of the other used priming media. In another study by Dezfuli et al.,[72] it was reported that soaking maize seeds in water for still high duration could lead to a high germination percentage. Similarly, enhanced seed germination and root length were reported in wheat genotypes soaked in water.[36]

Changes in photosynthetic pigments and subsequently in chlorophyll fluorescence attributes may lead to an alteration in photosynthetic efficiency, including the capability to harvest light; hence, observing these traits is essential to determine the plant photosynthetic effectiveness. In the current research, a differential trend was observed in the chlorophyll content, which was positive in the case of plants primed with tap water and Fe-10 mM, which may be attributed to simultaneous changes in osmoprotectant and different response levels of the studied wheats.[73−77]

In addition to the risks of climate change and increased population, crops including wheat are also facing bigger challenges of malnutrition and food insecurity.[10] To ensure food security, exploration of genetic diversity and germplasm improvement are crucial to the reliable and sustainable production of crops.[78−81] In this context, hydropriming is

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Table 3. Mean Squares of Physiological Traits among the Studied Wheat Genotypes[a]

SOV	DF	RWC	Chla	Chlb	TChl	MSI	ϕPSII	Fv/Fm	qP	qN	ETR
gen	2	416.3*	0.218^{NS}	0.039^{NS}	0.323^{NS}	196.0^{NS}	0.033***	0.001^{NS}	0.002^{NS}	0.002^{NS}	160.1***
treat	3	599.8***	0.646*	0.131**	0.1267**	1232.0***	0.882***	0.046***	0.003^{NS}	0.035***	533.9***
gen × treat	6	125.6^{NS}	0.026^{NS}	0.034^{NS}	0.142^{NS}	140.5**	0.001^{NS}	0.0001^{NS}	0.001^{NS}	0.003^{NS}	2.274^{NS}
error	22	75.56	0.165	0.019	0.229	36.7	0.0009	0.0005	0.001	0.005	3.722

[a]DF, degree of freedom; NS, nonsignificant; *, **, and ***, statistical significances by employing Fisher's LSD at 0.05, 0.01, and 0.001 α levels, respectively; RWC, relative water content; Chla, chlorophyll a; Chl b, chlorophyll b; TChl, total chlorophyll; MSI, membrane stability index; ϕPSII, actual quantum yield of PSII electron transport; FV/Fm, maximal quantum yield of PSII photochemistry; qP, photochemical quenching coefficient; qN, non-photochemical quenching coefficient; and ETR, electron transport rate.

considered important for ensuring germination to be uniform and rapid; however, the underlying mechanisms involved have remained unclear and hence need further exploration. As a matter of fact, unprimed seeds may need extra time for germination and those that are hydroprimed may exhibit enhanced germination, probably through its action on embryo development and leakage of emergence inhibitors. It may also play its role through earlier initiation of metabolic processes accompanied by better synthesis of RNA, DNA, and proteins.[82] Accordingly, enhanced concentration of free amino acids including proline and soluble sugars has been reported in hydroprimed rice in an osmotic stress environment.[83] Furthermore, seeds of *Medicago truncatula* when hydroprimed have been reported to upregulate the genes involved in antioxidant activity and DNA damage repair. Similarly, hydropriming treatment for 4 h enhanced the activity of formamidopyrimidine DNA glycosylase involved in base excision repair.[84] The ANOVA result revealed that priming medium has affected the germination characteristics of the studied wheat genotypes. However, the effect was more prominent when priming was accomplished with water. The studied wheat genotypes, on the other hand, were highly different in response to different priming media used. The role of Fe-10 mM and priming with tap water was comparatively much more promising in terms of GP, RV, RS, RDia, RL, SL, RFW, RDW, SFW, chlorophyll content, ϕPSII, Fv/Fm, qP, and ETR. The wheat genotype interaction with priming treatment was also conducive in terms of germination percentage, chlorophyll a, chlorophyll b, and total chlorophyll content. Shahverdi et al.[85] have also reported previously that iron priming of *Stevia* seeds at low concentration has the potential of reducing the deleterious effect of salinity stress on the investigated attributes, which included mean germination time, germination percentage, shoot length, free proline, and total chlorophyll content as well as antioxidants activities. El Rasafi et al.[86] have reported similar findings of enhancing germination percentage with priming wheat seeds with iron, cadmium, and zinc. It has also been reported that iron when applied at low concentrations in the form of iron sulfate resulted in enhanced germination behavior and increased yield.[43] Nonetheless, iron sulfate when applied in high concentrations led to reduction in the number of leaves and nodules. Hence, care needs to be taken when selecting the proper concentration of iron for obtaining optimum beneficial output.

CONCLUSIONS

The research study has demonstrated that seed priming in general is a preliminary step to enhance the germination characteristics of wheat crop. Priming accomplished with distilled as well as tap water and low iron concentrations revealed better germination and seedling performances in the experimental wheats and was harmful or toxic in high concentrations. The optimum level of Fe must not exceed 10 mM for better performance of bread wheat in terms of germination behavior and other related seedling morphological and chlorophyll fluorescence attributes. Furthermore, current findings suggest that seed priming may have a prospect of an innovative and user-friendly approach for wheat biofortification with the aim of enhanced iron acquisition and accumulation in grains.

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Notes

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