

Effect of drought stress on grain quality of wheat and its mitigation through arbuscular mycorrhiza fungi

B. Rani^{1*}, M. Jatttan², Pooja³, N. Kumari⁴, J. Prashad⁵, A. Kumari¹, K.D. Sharma⁶ and S. Madan⁴

¹Biochemistry, Chaudhary Charan Singh Haryana Agricultural University, Hisar-125 004, India

²Plant Breeding, Chaudhary Charan Singh Haryana Agricultural University, Hisar-125 004, India

³Plant Physiology, ICAR – Sugarcane Breeding Institute, Regional Centre, Karnal-132 001, India

⁴Biochemistry, Chaudhary Charan Singh Haryana Agricultural University, Hisar-125 004, India

⁵Microbiology, Chaudhary Charan Singh Haryana Agricultural University, Hisar, 125 004, India

⁶Plant Physiology, Chaudhary Charan Singh Haryana Agricultural University, Hisar-125 004, India

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*Corresponding Author Email : babitachahalkharb@gmail.com

*ORCID: <https://orcid.org/0000-0002-0723-2168>

Abstract

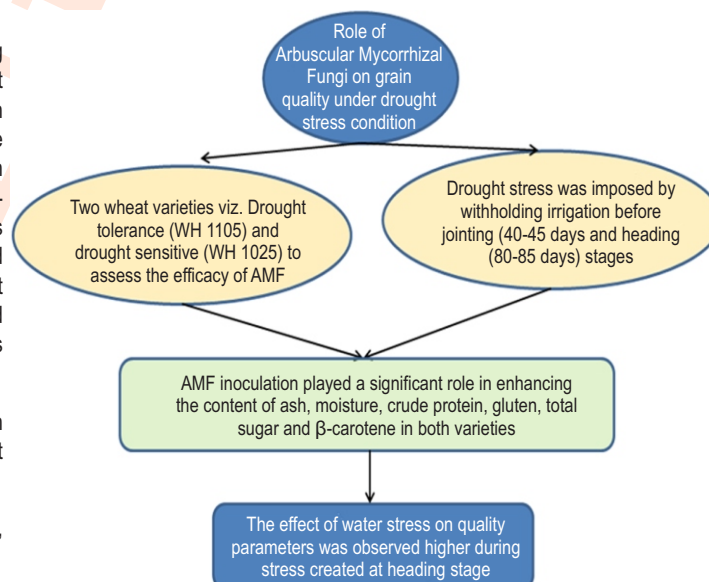
Aim: Grain quality is an important aspect to ensure food and nutritional security of the ever-increasing population of the world, and maintaining wheat grain quality under stressful conditions is decisive for end-use functional properties. Therefore, this study was carried out to evaluate the potential of arbuscular mycorrhizal fungi in improving grain quality under stress condition.

Methodology: The present study was conducted on two wheat varieties viz. Drought tolerance (WH 1105) and drought sensitive (WH 1025) under drought stress condition. This experiment was conducted in CRD factorial design and drought stress was imposed by withholding irrigation before jointing (40-45 days) and heading (80-85 days) stages.

Results: Drought stress imposed at either jointing or heading stage deteriorated all quality traits except for crude protein content and gluten content. Arbuscular mycorrhizal fungi inoculation played a significant role in enhancing the ash, moisture, crude protein, gluten, total sugar and β -carotene contents in both varieties, however, the interactive effects were found non-significant. Crude protein, which defines nutritional properties increased by 14.6 and 15.9% in WH 1025 grains and 13.9 and 14.9% in WH 1105 grains of AMF untreated and treated plants at heading stage. In both wheat varieties, sedimentation value and grain hardness were observed higher at heading stage as compared to the jointing stage, irrespective of AMF inoculation.

Interpretation: AMF inoculation played a significant role in improving quality traits by enhancing water relations and nutrient uptake under drought conditions.

Key words: Arbuscular Mycorrhizal Fungi, Drought, Grain quality, Wheat



Introduction

Among the abiotic stresses, drought is most severe and frequent stress that hinders morphological, physiological, biochemical and molecular processes and limits crop productivity as well as alters quality traits in many arid and semi arid areas (Pooja et al., 2019, 2021). Such aberrations arising due to changing climate are likely to reduce the quality of grains and yield production. Wheat is a major global crop, cultivated on more than 200 million ha land with more than 772 million tons of annual production in 2020-2021. Wheat grains are enriched with various nutritional and health beneficial compounds like iron, thiamin, niacin, riboflavin and, more recently, folic acid and calcium (Slavin et al., 2000). In addition to yield production, wheat quality is central to the well being. Historically, wheat breeding programs have focused on improving yield production, disease resistance, earlier maturity and wide adaptability (Zhang et al., 2005), but due to increasing market demand and food diversity, nowadays more priority is given to improve the grain quality (Wang et al., 2005c; Zhang et al., 2004b). Arbuscular mycorrhizal fungi (AMF) are mycorrhizal symbionts that have potential to improve plant performance, change the plant-water relationship, and increase plant productivity under drought stress (Augé, 2001; Amer et al., 2023). Arbuscular mycorrhizal (AM) fungi have many roles in various developmental processes in plants, especially in cereals. AM fungi have an effect on nutrient availability and uptake, increase the photosynthetic rate, improve antioxidant activities, and increase tolerance against environmental stress (Khan et al., 2022., Rani et al., 2018a). AMF, being a natural root symbiont, provides essential plant inorganic nutrients to host plants, thereby improving growth and yield under unstressed and stressed regimes (Li et al., 2019). The role of AMF as a bio-fertilizer can potentially strengthen plants' adaptability to changing environment.

The well-known beneficial effects of AM fungus on plant roots include: improved absorption and translocation of micro and macronutrients; more robust root system and resistance against many biotic and abiotic stresses (Rani et al., 2017; 2018b). Different environmental factors directly affect the wheat quality, but how these factors affect the grain quality, is not fully understood. Wheat flour technological and rheological properties are governed by most important component of the grains proteins content which is closely linked with end use quality (Zhao et al., 2010). Various experiments have been conducted to study the effect of different environmental factors on wheat grain quality, but we are unable to achieve a reciprocal correlation between wheat varieties and environment because of the less availability of good quality genotypes and insufficiency of related research (Zhao et al., 2010). Improvement of plant fitness by AMF under drought may possibly be due to the increased surface area for water absorption provided by AMF hyphae, increased access to small soil pores, or improved apoplastic water flow. Though, numerous studies have witnessed the beneficial role of AMF in drought mitigation (Jerbi et al., 2022; Rani et al., 2023, Khan et al., 2022), however, studies on the improvement in grain quality is meagre. Therefore, the present work was conducted to study the effect of drought stress on wheat

grain quality and its mitigation by the use of Arbuscular Mycorrhiza Fungi.

Materials and Methods

The present investigation was carried out on two varieties of wheat viz. WH 1025 (drought tolerant) and WH 1105 (drought susceptible) in pots. Seeds were procured from Wheat and Barley Section, Department of Genetics and Plant Breeding, College of Agriculture, CCS HAU, Hisar. Cultures of AMF (*Glomus mosseae*) were procured from Indian Agriculture Research Institute, New Delhi. In half number of pots, seeds were inoculated (20g kg⁻¹ seeds) with AMF (*Glomus mosseae*) and in another half pots, seeds were not inoculated with AMF. Six seeds were sown in each pot at uniform depth. Thinning was done one week after germination to leave two plants of uniform growth in each pot. Drought stress was created by withholding irrigation at jointing (40-45 days) and heading (80-85 days) stage and grain samples were retained from AMF inoculated and uninoculated plants under normal and stressed conditions. The experiment was laid out in a complete randomized design in net house.

Moisture, ash, fat, crude fiber, crude protein and β -carotene contents were analyzed by the standard methods (AOAC, 2000). Gluten content was measured on weight basis. Total soluble sugars (TSS) was estimated following the method of Yemm and Willis (1954) using anthrone reagent and glucose (20-100 μ g) standard curve. Starch content was estimated by the method of Clegg (1956) using glucose (20-100 μ g) standard curve. Grain hardness (kg/seed) was estimated by grain hardness tester (Kiya Seisakusho Ltd., Japan) by using ten average sized well fitted grains. Sedimentation value was measured by the method of Axford et al. (1979). All data were statistically analyzed by analysis of variance using SAS software (Version 9.3, SAS Institute Inc., Cary, NC, USA) on the three replicated data and each replicate was estimated in duplicate. TUKEY's Honest Significant Test was put at 5% probability level to compare the mean differences.

Results and Discussion

Mutualistic root microorganisms have high potential to improve plant nutrient uptake and improve plant growth most efficiently under stress conditions (Mäder et al., 2011). Mycorrhiza helper bacteria have been reported to stimulate AMF spore germination, hyphal growth and AMF root colonization directly and conversely, AMF influence the chemical composition of root exudates, which in turn are a major nutrient source for rhizosphere bacteria (Pellegrino et al., 2020). In recent years, numerous studies have focused on the qualitative aspects of wheat production to face the widespread malnutrition due to lack of nutrients, which leads to the development of several biofortification strategies for increasing the concentration of minerals in cereals and improvement of grain quality traits (Liu et al., 2020; Parashuram et al., 2021). Mycorrhizal treated plants

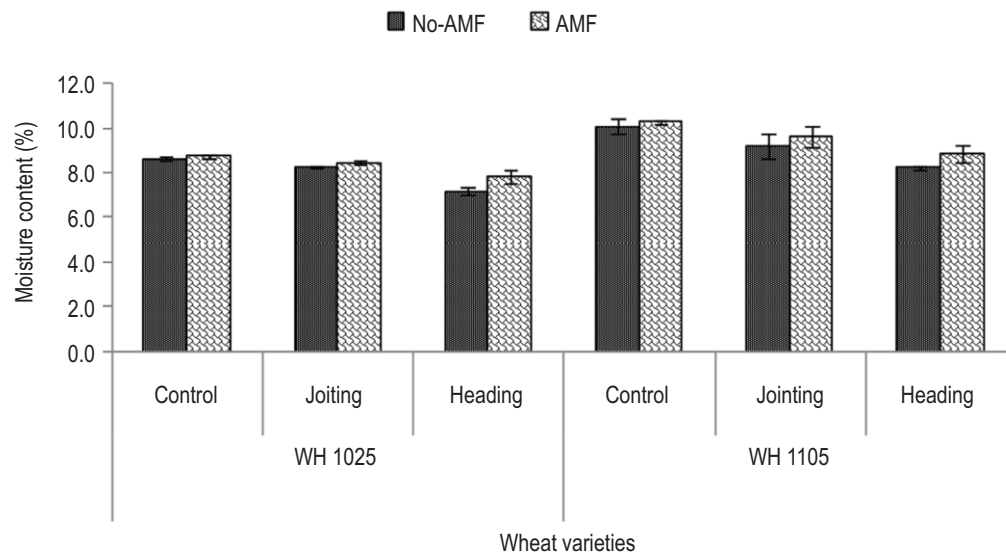


Fig. 1: Effect of arbuscular mycorrhizal fungi on moisture content in wheat plants under control and drought stress condition at different growth stages.

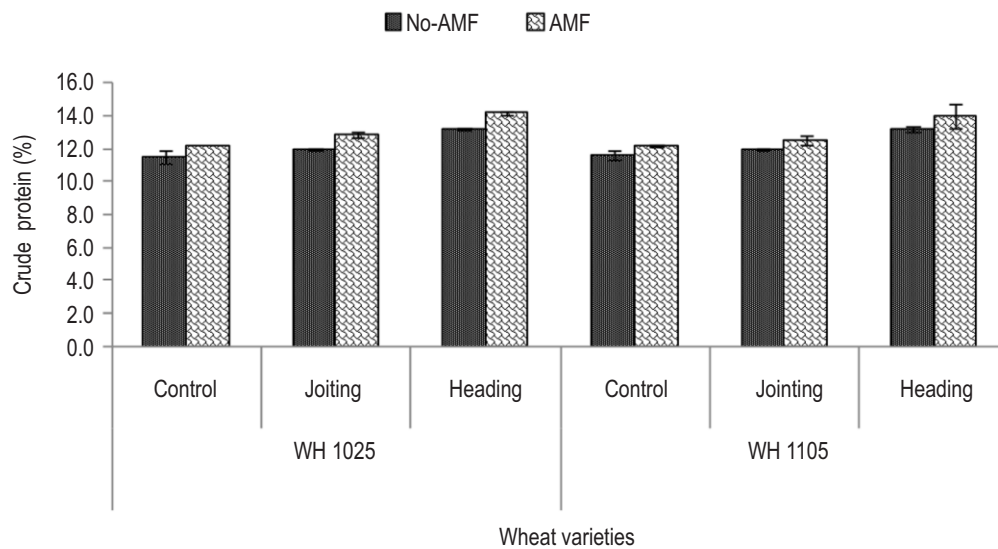


Fig. 2: Effect of arbuscular mycorrhizal fungi on crude protein content in wheat plants under control and drought stress condition at different growth stages.

often have greater tolerance to drought than untreated plants as AM fungi strengthen the plants' ability to increase their tolerance to biotic and abiotic stress conditions (Gao *et al.*, 2020).

The present work described the mutualistic root microorganisms on the quantitative changes in quality traits of wheat under unstressed and stressed conditions. AMF inoculation and drought stress significantly affected the moisture

stress but the interactive effect of AMF × TRT was found non-significant on both wheat varieties (Fig. 1). Under drought stress at jointing and heading growth stage, moisture content decreased in both tolerant and sensitive wheat varieties. In WH 1025, it decreased by 11.1% and in WH 1105 decreased by 15.8% under drought stress at heading stage. AMF inoculation played statistically no significant role in moisture content in both varieties, but the difference between moisture content of

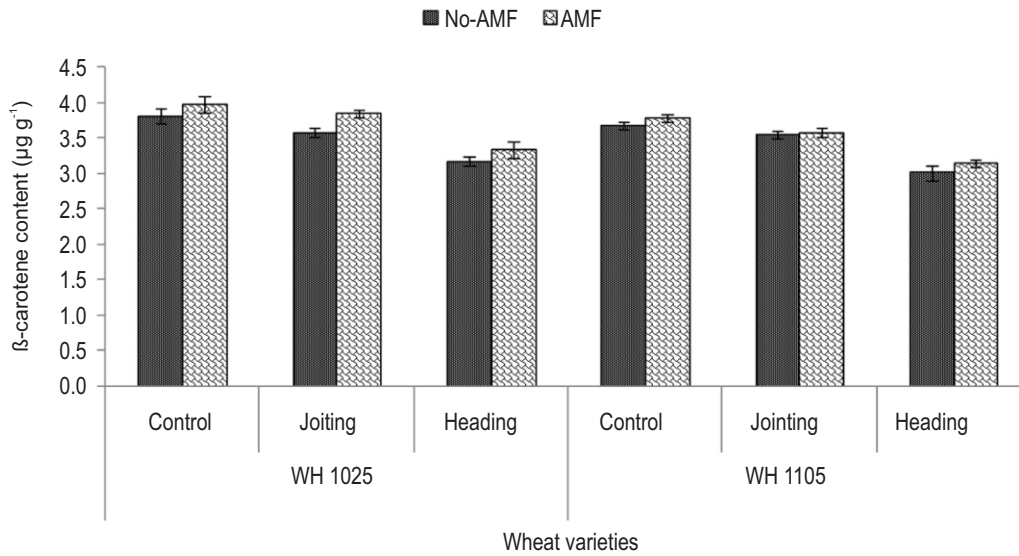


Fig. 3: Effect of arbuscular mycorrhizal fungi on β -carotene content in wheat plants under control and drought stress condition at different growth stages.

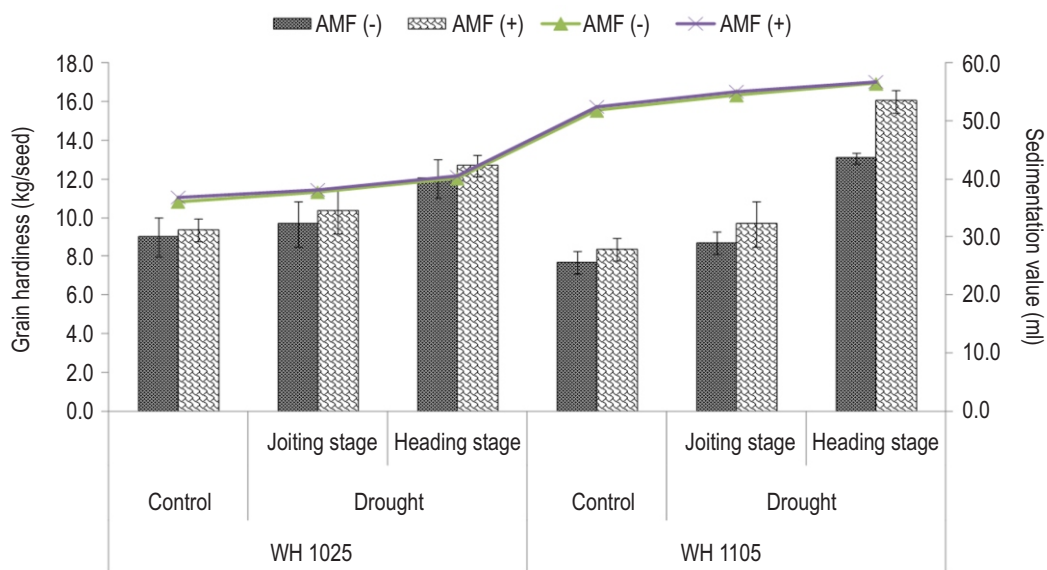


Fig. 4: Effect of arbuscular mycorrhizal fungi on grain hardiness (kg per seed) and sedimentation value (ml) of wheat varieties under control and drought stress at different growth stages.

untreated and treated plants was found significant (Fig. 1). Under drought stress, ash content decreased in both varieties, however, higher per cent (Table 1) reduction was observed when stress was created at heading stage as compared to jointing stage. In drought tolerant variety, ash content decreased by 3.1% and in drought sensitive variety 2.4% reduction was observed under drought condition created at heading stage as compared to AMF

uninoculated control plants. No significant effect was observed in both the varieties after AMF inoculation. Under stress condition, crude fiber also decreased in AMF inoculated and uninoculated plants of WH 1105 and WH 1025 wheat varieties (Table 1). Under control and stressed conditions, more crude fiber content was reported in drought sensitive variety as compared to tolerant wheat variety. No significant effect of AMF inoculation was

Table 1: Effect of arbuscular mycorrhizal fungi on ash content and crude fibre in wheat plants under control and drought stress condition at different growth stages

Treatments/Traits		Ash content (%)		Crude fibre (%)	
		No-AMF	AMF	No-AMF	AMF
WH 1025	Control	1.54	1.56	1.90	1.94
	Drought at Jointing stage	1.53	1.54	1.87	1.91
	Drought at Heading stage	1.51	1.52	1.79	1.80
WH 1105	Control	1.56	1.57	1.98	2.00
	Drought at Jointing stage	1.55	1.57	1.95	1.96
	Drought at Heading stage	1.52	1.53	1.86	1.87
CD @ 5%		AMF – 0.01; Stress – NS; AMF × Stress - NS	AMF – 0.009; Stress – 0.013; AMF × Stress - NS	AMF – 0.178; Stress – NS; AMF × Stress - NS	AMF – 0.212; Stress – 0.212; AMF × Stress - NS

Table 2: Effect of arbuscular mycorrhizal fungi on fat content and gluten fibre in wheat plants under control and drought stress condition at different growth stages

Treatments/Traits		Fat content (%)		Gluten content (%)	
		No-AMF	AMF	No-AMF	AMF
WH 1025	Control	0.96	0.96	30.67	31.33
	Drought at Jointing stage	0.96	0.96	32.33	33.0
	Drought at Heading stage	0.91	0.92	33.33	34.33
WH 1105	Control	0.95	0.95	28.0	29.33
	Drought at Jointing stage	0.94	0.95	29.67	31.67
	Drought at Heading stage	0.89	0.90	29.67	32.33
CD @ 5%		AMF – 0.01; Stress – NS; AMF × Stress - NS	AMF – 0.009; Stress – 0.013; AMF × Stress - NS	AMF – 0.530; Stress – 0.530; AMF × Stress - NS	AMF – 0.467; Stress – 0.467; AMF × Stress – 0.661

reported in both varieties (Table 1). In AMF inoculated plants, crude fiber declined in WH 1025 by 7.6% and in WH 1105 by 6.7% as compared to control plants during stress created at heading stage. Optimum requirement of compositional and physical properties are set according to grain quality. The physical characteristics of whole grains, such as size and shape, affect milling yield and screening losses for staple food grains like wheat, which determine the processing capacity and value of the grain. Drought stress created for wheat plants by withholding irrigation at jointing and heading growth stages of plants showed significant diminishing effect on grain quality traits, however, the mycorrhizal association helped the plants in mitigating the effect by improving the traits (Perez *et al.*, 2016). Varieties displayed non-significant effect for crude protein content. In both the varieties, crude protein was significantly enhanced when stress was created at jointing and heading growth stages (Fig. 2).

A significant effect of AMF application was reported on crude protein content in WH 1105 and WH 1025 wheat varieties. In WH 1025, crude protein content enhanced by 14.6% and 15.9% while in WH 1105 it enhanced by 13.9% and 14.9% at heading stage under drought conditions in grains of AMF uninoculated and inoculated plants, respectively. Table 2 shows that fat content of both the varieties was non-significant. Although,

it was reported that fat content declined under drought stress but after AMF inoculation there was no significant change observed in drought tolerant and drought sensitive varieties. In WH 1025, fat content reduced from 0.96% to 0.92% after AMF inoculation when stress was created at heading stage. Significant differences were also noted for gluten content amongst varieties, AMF treatment and stress treatment (Table 2). Variety WH 1025 had higher gluten content (32.5%) than WH 1105 (30.11%). AMF treatment significantly enhanced gluten content by 4.35% in treated plants in comparison to untreated ones (Table 2). When drought stress was imposed at both growth stages, gluten content in WH 1025 and WH 1105 wheat varieties after AMF inoculation increased by 33.0 and 34.3% and 31.7 and 32.3%, respectively. As reported in this study, moisture content, ash, fat, crude fiber, total sugar and starch reduced however, sedimentation value, gluten content, protein content and grain hardness enhanced when stress was imposed at both the growth stages of drought tolerant and drought sensitive varieties. More effect of stress on quality parameters was reported at heading stage of both varieties. Different types of quality parameters observed in this study seemed to change significantly with environment. AMF inoculation played a significant role in improving the quality traits, except for crude fiber, fat and starch content. Similar reports have been observed earlier also (Noorka *et al.*, 2009;

Table 3: Effect of arbuscular mycorrhizal fungi on total sugar content content and starch content fibre in wheat plants under control and drought stress condition at different growth stages

Treatments/Traits		Total sugar content (%)		Starch content (%)	
		No-AMF	AMF	No-AMF	AMF
	Control	1.65	1.70	59.46	60.60
WH	Drought at Jointing stage	1.62	1.68	57.61	59.0
1025	Drought at Heading stage	1.51	1.63	54.58	55.8
	Control	1.65	1.66	59.36	60.84
WH	Drought at Jointing stage	1.53	1.61	58.39	58.92
1105	Drought at Heading stage	1.43	1.51	55.68	55.8
		AMF – 0.846;	AMF – 0.888;	AMF – 0.015;	AMF – 0.021;
		Stress – 0.846;	Stress – 0.888;	Stress – 0.015;	Stress – 0.021;
		AMF × Stress - NS	AMF × Stress - NS	AMF × Stress – 0.021	AMF × Stress – 0.029

Ozturk and Aydin 2004; Gudeir *et al.*, 2002; Mary *et al.*, 2001). Drought stress decreased the moisture and fat content, however, enhanced the crude protein, gluten content and sedimentation value (Hamid *et al.*, 2012; Zeleny *et al.*, 1960).

Protein content and composition of grains are also important parameters which explains the nutritional and end use characteristics of dough mixing and rheological properties like dough strength, extensibility, breakdown, development time and loaf volume which influence the efficiency of bread making process and product quality. Similar results were also reported (Baric *et al.* (2004), who found that protein content of grains was influenced under different environmental conditions and genotypes. In both varieties, TSS content declined under drought stress conditions (Table 3) and the reduction was 3.01 % at jointing and 8.43 % at heading stage, respectively. When drought stress created at heading stage showed more effect as compared to jointing stage. AMF treated plants showed significant higher total sugar content than untreated plants (Table 3). In arbuscular mycorrhizal uninoculated plants of drought tolerant variety, TSS content reduced by 9.7% and in drought sensitive variety by 14.9% as compared to control plants during stress imposed at heading stage. Varieties didn't showed any significant effect on total soluble sugar content. Starch content also decreased in both the varieties under drought stress conditions (Table 3) at both the growth stages of drought tolerant and drought sensitive varieties. Varieties and AMF treatments showed non-significant effect on starch condition. In arbuscular mycorrhizal fungi inoculated plants of drought tolerant variety (WH 1025), starch content reduced by 7.9% and 8.3% in drought sensitive variety (WH 1105) under stress conditions imposed at heading stage (Table 3). In this study, a significant effect of AMF inoculation and drought condition was found on β -carotene (Fig. 3). In both the varieties, β carotene content increased after AMF inoculation, however, it decreased under drought stress conditions. Beta carotene content in drought tolerant variety reduced by 3.5% and 19.0%, and in drought sensitive variety it decreased by 5.6% and 20.2% under drought conditions created at jointing and heading stage, respectively (Fig. 3). Grain hardness enhanced with AMF

inoculation and under stress conditions (Fig. 4) in drought tolerant and drought sensitive wheat variety.

Higher effect of stress was reported at heading stage of both varieties. By imposing stress at heading stage, the grain hardness enhanced by 35.7% in drought tolerant and 40.0% in drought sensitive variety in AMF treated plants. Under stress conditions, sedimentation value increased in both the varieties (Fig. 4). The maximum value was noted in grains of AMF inoculated drought sensitive variety whereas minimum in drought tolerant variety under control conditions. Rharrbti *et al.* (2003) reported that different environment conditions affect different quality parameters of grains. . Noorka *et al.* (2009) also observed that different effects under stress and control conditions for grain yield production and ash, fat, gluten, moisture, protein levels.

Under water deficit condition, Hailu *et al.* (2007) observed that moisture content reduced, however other traits, especially protein content enhanced in while cross breeds. Zeki *et al.* (2010) observed a positive correlation of protein content with dry gluten content and sedimentation value, however, a negative correlation was found with grain ash under stress and control conditions. A negative correlation of protein and gluten content with grain yield and kernel weight was reported during stress (Yong *et al.*, 2004; Robert, 2002; Purchase *et al.*, 2000). Similar trends were reported by Hasanpour *et al.* (2012) that both AMF inoculation and Azotobacter leads to a significant enhanced percentage of grain protein during stress conditions. Accompanying with our finding differences in grain protein content due to drought and environmental conditions were observed by Beltrano *et al.* (2006). Whereas, Sumera and Asghari (2009) observed that changes in composition and percentage of wheat grain protein due to differences in genome rather than treatments indicated that composition of seed protein is mainly controlled by genetic makeup rather than environmental factors. The inoculation effects of AMF were highly dependent on planting density and soil moisture and The mechanism behind the inoculation effects of AMF was that, in low density and drought conditions, it enhanced rhizospheric interactions for better water and nutrient status (Duan *et al.*, 2024).

Biofertilizers, such as AMF inoculants, are an alternative source to meet the nutrient requirements of wheat and are of great importance in agriculture. The observation provides evidence to affirm that AMF application can be beneficial to wheat production under drought conditions, and the wheat growers looking to mitigate drought risk and increase grain yields in a sustainable manner would benefit from applying AMF to their soils. Drought stress decreased grain quality traits viz. moisture, ash, fat, crude fiber, total sugar and starch contents however, sedimentation value, gluten content, protein content and grain hardness enhanced in both the varieties. The effect of water stress on quality parameters was observed higher during stress created at heading stage. AMF inoculation played significant role in improving quality traits except crude fibre, fat content and starch content. These findings indicated that inoculation with AMF can enhance drought tolerance by alleviating the antioxidative system and nutrient uptake as discussed in our previous studies, which may be helpful in quality improvement of wheat grains.

One may claim that AMF inoculation has a beneficial influence on the development of wheat yield under drought stress, and this effect can be closely linked to enhanced rhizosphere soil water status and nutrient cycling. This could result in fresh approaches to the agricultural system's sustainable development in the face of global climate change.

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