

International Journal of Biosciences | IJB | ISSN: 2220-6655 (Print), 2222-5234 (Online) http://www.innspub.net Vol. 8, No. 4, p. 103-110, 2016

**RESEARCH PAPER** 

**OPEN ACCESS** 

# Effect of mycotoxins on rheological characteristics of stored wheat

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Key words: Amylograph, Farinograph, Wheat, Mycotoxins.

http://dx.doi.org/10.12692/ijb/8.4.103-110

Article published on April 23, 2016

# Abstract

Wheat and their products are the chief source of food and energy globally. Wheat grain quality are lessen due to mycotoxin contamination and becomes a major cause of illness or death in human and animals. Flour superiority determined by eminence of protein gluten presence into various products, also some chemical, biochemical and physical characteristics principally examined for the products prepared from wheat flour for this purpose some experiential rheological tests are used to assess quality of flour and its usage in specific baked products. During storage wheat contaminated with various mycotoxin due to poor storage conditions, improper moisture and temperature control. For to analyze the mycotoxins effects on quality of wheat, the wheat samples were taken from the eleven storage houses of Hyderabad division and the results revealed that samples contaminated with mycotoxin found with damaged gluten fractions because protein and starch start to break due to proteolytic action of amylolytic enzymes. From the study it was concluded that wheat quality reduced by fungus availability which deteriorate and break the glutinious complexes.

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#### Introduction

Wheat as food consumed by 36% of population around the globe and providing 20% of the total food calories consumed which delivering 55% of carbohydrates. Wheat also a rich source of vitamins, minerals, proteins and essential amino acids except lysine (Beriman & Graur, 1995 Khan & Zeb, 2007). In Pakistan wheat is the basic food and bulk quantity of wheat milled into flour which used to make chapaties, various naans, pastas and bakery goods (Anonymous, 2010; Anjum et al., 1991). Quality of wheat is measured to be the key factor for specific product preparation which is rely on several rheological dough properties, milling, chemical, baking and storage conditions (Pasha, 2006). Physical, chemical and biochemical characteristics along with the gluten protein are examined to produce different products from wheat flour (Weegels et al., 1996) and Wheat flour Product quality can be enhanced if the rheological properties of wheat are known because rheological tests are used to test quality of flour and its usage in specific baked products (Khatkar & Schofi, 2002).

Many methods has applied for improvement and quality control of wheat that muddle productivity (Raja et al., 2010) beside this 20% of wheat has lost every year (Fakir, 1999). Wheat can be contaminated by various microscopic fungi during its development that can affect grain quality, chemical properties as well as its rheological characteristics and make wheat unsuitable for consumption (Embaby et al., 2012). The contamination caused by the fungal secondary metabolite called 'Mycotoxin' that can grow well due to environmental and ecological factors, during growth at field, after harvest, during transportation or during storage. Several seed-borne pathogens are known to be associated with wheat seed which are responsible for deteriorating seed quality during storage (Doohan et al., 2003). These causal agents engendering severe agricultural issues and are known as potent carcinogenic (Pittet, 1998; Wagacha & Muthomi, 2008). During storage uncontrolled moisture and temperature changes colonize these toxins in wheat and Pakistan's hot and humid environment is most promising for the growth of these toxins (Styriak *et al.*, 1998; Paterson & Lima, 2011; Alam *et al.*, 2012).

Compositional changes, rheological and technological properties of stored wheat grains due to infection with fungi were studied by several investigators. Fusarium leads to destroy wheat kernels and many characteristics changes in endosperm and many region were found in Infected kernels, such as partial or complete lack of the protein damage to large and small starch granules caused by fungal amylolytic enzyme, and starch granules decreased with increase in colonization of fungi that produce mycotoxins, Fusarium also found to involved in extracellular hydrolytic enzymes and phytotoxic compounds production (Jackowiak et al., 2005; Meyer et al., 1986). Fusarium germanium also produce this type of destruction in grains (Nightingale et al., 1999; Schwarz, 2003), different types of damages after mycotoxin production was detected in oat endosperm cells composed of starch granules (Packa, 2005). Dexter, et al., (1996) reported that gluten originating from mycotoxin infected wheat have reduced content of glutenin fraction. Barabara, et al., 2004 and Chandra, et. al., 2011 assessed that the damage caused by fungi adversely affects the quality of wheat and reduces its nutritional composition. Marija et al., (2004) detected a higher contribution of major mycotoxigenic molds in flour, making the product more susceptible to the accumulation of mycotoxins. Therefore it is the need of time to detect deterioration

caused by fungal toxins this will help to Improve the quality and safety of food produce from wheat and as well as to limit the risk of food borne illnesses and their intoxications. Having collected extensive experimental materials, we launched a study to investigate and document damage to the wheat kernels and its effects on quality of wheat during storage in the common storage house (godowns) at Pakistan.

#### Material and methods

Wheat samples was collected from all government godowns of Hyderabad division namely Bolhari, Hali

road, Fatah chowk, Hala city, Matiyaari, Thatta, Tandpallhyaar, Dadu, K.N shah, Sehwan and Aarazi. Sampling was done form upper, central and bottom sacks of one selected stack after seven months of wheat storage at godowns. During sampling temperature was recorded 37-52°C and moisture of all godowns were in the range of 40-70%.

#### Mycological Study

Mycoflora was determined by the method of agar plate described by Mathur *et al.* (2003). Whereas relative isolation frequency (Fq.) of fungi (*Aspergillus parasiticus and Aspargillus flavus*) that produce mycotoxins was calculated (Fatma Bensassi *et al.*, 2011).

#### Sample Extraction and analysis

Wheat sample was extracted by the method available in commercially available immunoassay kit the Neogen ELISA Kit (Veratox , Product no. 8030) and analysis was based on competitive direct enzyme linked immunosorbent assay format. Concentration was calibrated and calculated by ELISA 'state fax 2100' (Awareness technology).

#### Sample Preparation for Rheological analysis

After quantitative analysis through ELISA wheat samples were used for rheological properties to observe the effect of mycotoxin in wheat, the grains samples were taken free from dockage and foreign matter and were subjected to tempering at a moisture content of 14-15 percent in plastic containers at room temperature for 24 hours and amount of water required for tempering was calculated by following the expression given in AACC (2000) than were milled by using Brabender quadrametic junior mill to yield flour.

#### Rheological analysis of wheat flour

# Measurement of alpha-Amylase Activity with the Amylograph

Amylase activity and gelatinization temperature of wheat flour were obtained by placing the samples in Brabender amylograph-E (GmbH & Co. Germeny). This method uses the amylograph to estimate alpha-

#### Rheological Behaviour of Flour by Farinograph

The Farinograph measures and records the resistance of dough to mixing and quantitatively analyze the flour protein through Brabender Farinograph-E (GmbH & Co. Germeny).

Methods used to assess rheological properties were meets standard recommendations of method by AACC 2000.

#### Data analysis

All the experiments had three replicates. Data was analyzed for one-way analysis of variance followed by Student-Newman-Keuls multiple test at 0.05 level using compare means procedure of SPSS 16.

#### **Results and discussion**

# Measurement of alpha-Amylase Activity with the Amylograph

The gelatinization properties of starch in wheat depends on alpha amylase activity and can be used to make important predictions about the baking quality of flour, Amylograph enables continuous measurement of changes of viscosity in flour and water suspension during heating.

The amylographic results revealed that the beginning of gelatinization, gelatinization temperature and gelatinization maxima of upper and central portion were found within the standard limits except results of bottom portioned wheat that shows higher temperature to begin the gelatinization in flour. Higher temperature for beginning of gelatinization was noted in bottom wheat samples of Dadu (83°C), Aarazi (87°C) and Sehwan (84°C) along with higher temperature for gelatinization was found in Dadu, K.N Shah, Sehwan and Aarazi. Lower values for gelatinization maxima were prominent in wheat of bottom portion where the mycotoxin contamination ratio was elevated and results shows that Dadu with 100 GM, Sehwan and Aarazi wheat were recorded with 90 and 95 gelatinization maxima respectively.

This alteration of results was noted because of breakdown of starch granules due to proteolytic action of amylolytic enzymes. Wheat found at bottom portion was more contaminated with mycotoxin due to which starch garnules broken and gel formation could not form properly which in turn reduces the quality of wheat. The results are in consistent with Jackowiak, *et. al.* (2005) that reported that decomposition of Fusarious wheat kernel by Fungal amylolytic enzyme leads to degradation of small starch granules.

Table 1. Amy	lographic	characteristics	of godown wheat.

	BG (°C)	GT (°C)	GM	
	Food Gra	ain godown Bolhari		
Upper Portion	62 a	83 a	935 °	
Central Portion	53 ª	<b>81</b> a	820 <sup>b</sup>	
Bottom Portion	71 <sup>b</sup>	95 <sup>b</sup>	<b>200</b> <sup>a</sup>	
	Food Grai	n Godown Hali Road		
Upper Portion	<b>60</b> <sup>a</sup>	80 <sup>a</sup>	871 °	
Central Portion	61 <sup>a</sup>	81 a	632 <sup>b</sup>	
Bottom Portion	78 <sup>b</sup>	95 <sup>b</sup>	451 <sup>a</sup>	
	Food Grain	Godown Fateh Chowk		
Upper Portion	58 ª	79 <sup>a</sup>	776 <sup>c</sup>	
Central Portion	60 <sup>a</sup>	81 <sup>a</sup>	710 <sup>b</sup>	
Bottom Portion	75 <sup>b</sup>	98 <sup>b</sup>	552 <sup>a</sup>	
	Food Grai	in Godown Hala City		
Upper Portion	55 <sup>a</sup>	<b>82</b> <sup>a</sup>	<b>821</b> <sup>c</sup>	
Central Portion	63 <sup>a</sup>	78 <sup>a</sup>	809 <sup>b</sup>	
Bottom Portion	73 <sup>b</sup>	98 b	<b>321</b> <sup>a</sup>	
	Food Gra	ain Godown Matiari		
Upper Portion	64 <sup>a</sup>	88 a	632 <sup>c</sup>	
Central Portion	60 <sup>a</sup>	84 <sup>a</sup>	610 <sup>b</sup>	
Bottom Portion	81 <sup>b</sup>	95 <sup>b</sup>	551 <sup>a</sup>	
	Food Grain (	Godown Thatta (Makli)		
Upper Portion	61 <sup>a</sup>	82 a	720 °	
Central Portion	63 ª	80 a	672 <sup>b</sup>	
Bottom Portion	79 <sup>b</sup>	92 <sup>b</sup>	<b>200</b> <sup>a</sup>	
	Food Grain	Godown Tando Alliyar		
Upper Portion	<b>60</b> <sup>a</sup>	81 a	810 c	
Central Portion	59 <sup>a</sup>	85 <sup>a</sup>	778 <sup>b</sup>	
Bottom Portion	85 <sup>b</sup>	98 b	415 <sup>a</sup>	
	Food Gr	ain Godown Dadu		
Upper Portion	60 <sup>a</sup>	87 <sup>a</sup>	620 <sup>c</sup>	
Central Portion	83 <sup>a</sup>	92 <sup>a</sup>	<b>232</b> <sup>b</sup>	
Bottom Portion	87 <sup>b</sup>	98 <sup>b</sup>	100 <sup>a</sup>	
	Food Grain Gode	own Khairpur Nathan sha	ah	
Upper Portion	61 <sup>a</sup>	<b>90</b> <sup>a</sup>	770 <sup>c</sup>	
Central Portion	65 <sup>a</sup>	86 a	643 <sup>b</sup>	
Bottom Portion	85 <sup>b</sup>	100 <sup>b</sup>	105 <sup>a</sup>	
	Food Gra	in Godown Sehwan		
Upper Portion	63 <sup>a</sup>	87 <sup>a</sup>	637 °	
Central Portion	78 a	<b>91</b> <sup>a</sup>	110 <sup>b</sup>	
Bottom Portion	84 <sup>b</sup>	94 <sup>b</sup>	90 <sup>a</sup>	
		ain Godown Aarazi		
Upper Portion	65 ª	88 a	552 °	
Central Portion	61 a	<b>83</b> <sup>a</sup>	131 <sup>b</sup>	
Bottom Portion	87 <sup>b</sup>	99 b	95 ª	

Values followed by the same letter are not significantly different at 0.05 level Student-Newman-Keuls test.

BG= Beginning of gelatinization (F= 31.73, P=0.00, df = 32);

GT= Gelatinization temperature (F= 42.73, P=0.00, df = 32);

GM= Gelatinization maxima (F= 14.72, P=0.00, df = 32)

Fusarious kernel progress more disintegration of starch due to fungal alpha amylase and reduction of total number of starch granules and higher damage of starch components of endosperm due to this the reduction in amylographic highest viscosity was observed. Oat endosperm cells composed of starch granules was also found destroyed after mycotoxin attack (Packa, 2005).

Table 2.	Farinog	raphic	charac	eteristics	of g	odown	wheat.

	WA	DDT	DST	MIT
Food Grain Godown Bolhari				
Upper Portion a	51.2	5.0	4.9	80
Central Portion <sup>a</sup>	51.0	5.2	5.0	72
Bottom Portion <sup>a</sup>	42.1	3.4	4.1	52
Food Grain Godown Hali Road				
Upper Portion <sup>a</sup>	52.1	5.0	5.5	71
Central Portion <sup>a</sup>	50	4.2	5.0	65
Bottom Portion <sup>a</sup>	39	3.4	4.5	50
Food Grain Godown Fateh Chowk				
Upper Portion <sup>a</sup>	50	4.9	5.0	68
Central Portion a	52	5.1	5.0	65
Bottom Portion <sup>a</sup>	49	3.6	3.9	46
Food Grain Godown Hala city				
Upper Portion a	53	5.1	6.1	72
Central Portion a	51	4.6	5.4	65
Bottom Portion <sup>a</sup>	48	3.8	4.1	50
Food Grain Godown Matiari				
Upper Portion a	55	5.0	5.2	68
Central Portion <sup>a</sup>	52	5.0	5.2	65
Bottom Portion a	45	4.8	4.0	55
Food Grain Godown Thatta (Makli)		•	•	
Upper Portion a	45	5.0	4.2	61
Central Portion <sup>a</sup>	43	5.0	4.2	54
Bottom Portion a	40	4.2	4.0	48
Food Grain Godown Tando Allahya	r	•	•	•
Upper Portion a	48	4.2	3.9	52
Central Portion a	42	4.0	4.0	48
Bottom Portion a	39	3.3	3.1	41
Upper Portion <sup>a</sup>	43	2.2	3.1	33
Central Portion a	40	2.4	3.1	30
Bottom Portion a	32	1.4	2.8	25
Food Grain Godown Khairpur Nath	an Shah			
Upper Portion <sup>a</sup>	41	3.2	3.8	32
Central Portion a	41	3.0	3.6	36
Bottom Portion <sup>a</sup>	35	2.1	2.5	21
Food Grain Godown Sehwan	30	<b>_</b> +±		
Upper Portion a	35	3.1	2.9	31
Central Portion a	<u> </u>	2.7	2.9	26
Bottom Portion a	21	2./		-
	21	1	1.7	100
Food Grain Godown Aarazi		<i>.</i> .		
Upper Portion a	27	2.1	1.4	24
Central Portion <sup>a</sup>	22	1.9	1.6	20

Values followed by the same letter are not significantly different at 0.05

21

1.4

1

92

level Student-Newman-Keuls test.

Bottom Portion a

WA= water absorption (F= 2.27, P=0.121, df = 32);

DDT= dough development time (F= 1.489, P= 0.240, df = 32);

DST= dough stability mixing index tolerance (F= 2.07, P=0.143, df = 32);

MIT= Mixing index tolerance (F= 0.228, P= 0.797, df = 32).

#### Rheological Behaviour of Flour by Farinograph

Farinograph measures dough properties including a reliable check for mixing phase and water absorption capacity of flour, mixing time and dough stability as indicator parameters of dough quality. The farinographic characteristics such as absorption of water, time of dough development, stability, mixing easiness of dough differed significantly. The more contaminated wheat of bottom portion of Dadu, Sehwan and Aarazi showed the lower water absorption, dough development time and dough stability time due to higher deterioration of protein by the mycotoxins present and due to reduced protein molecules availability water can'not form colloid with protein to withstand dough to stable for longer time as well to develop the dough quickly. The results are in steady agreement with Embaby et al. (2012) that the control sample had higher water absorption (68.5%), high dough development time (5.0 min), and high time for dough stability (6.0 min) as compared with the wheat samples contaminated with mycotoxins. The results are also in agreement with Alian et al., 1997.

Impacts of in Invasion of wheat grains by the some fungi are showed in farinographic parameters indicates that dough texture was not shaped properly in the samples showing high contamination i-e wheat of Bolhari, Sehwan, Aarazi and K.N Shah godown bottom wheat and these samples didn't give appropriate farinograms. Breakdown of dough protein leaves undesirable effect on dough properties because total number of protein or gluten particles per unit of dough and efficiency protein particles to swell by hydration are responsible factor for dough strength. Fungus availability decline quality of dough because the glutinious complex breakage. Elizabet, et al. (2011), specified that increase of invasion of Fusarium into kernels is directly proportional to decrease in Farinographic quality number, stability, and resistance of dough, and trends of rise in dough softening degree. It was suggested from many research studies that moulds deterioration during storage will enormously reduce baking quality of wheat flour. Baking performance was found abridged in the stored wheat contaminated with Aspergillus species and Penicillium species (Wang *et al.*, 2005). Gluten content into red spring wheat was found lower compared with healthy wheat which causes the weak dough properties and substandard the bread superiority (Dexter *et al.*, 1996). Boyacioglu & Hettiarchchy (1995) reported that *F. graminearum* was the causal agent of reduced albumin content and glutenine content by 33% and 88% respectively, Meyer *et al.* (1986) further confirmed the baking properties was adversely effected after contamination caused by *F. culmorum*.

#### Conclusion

It was concluded from present research that due to presence of fungi that produce mycotoxin abridged the wheat quality and also the deterioration caused by moulds during storage of wheat grain will enormously reduce baking quality of wheat flour due to breakdown of dough protein and starch granules, accordingly these samples didn't stretch appropriate farinograms and amylograms.

#### Acknowledgement

The authors are thankful to Department of Food, Government of Sindh, permitting for wheat sampling from all Government public godowns of Hyderabad division. This paper is the part of the Ph.D thesis of 1<sup>st</sup> author Mahvish Jabeen Channa

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