Hungarian University of Agriculture and Life Sciences

Doctoral PhD Dissertation

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Gödöllő, Hungary

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INFLUENCE OF *FUSARIUM* HEAD BLIGHT ON TECHNOLOGICAL QUALITY OF WHEAT

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Influence of *Fusarium* Head Blight on Technological Quality of Wheat

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LIST OF ABBREVIATIONS

%	percent
ANOVA	analysis of variance
BP	before present
df	degrees of freedom
EFSA	European Food Safety Authority
F	F statistic
FHB	*Fusarium* head blight
ICC	International Association for Cereals and Technology
ISO	International Organization for Standardization
Mv	Martonvásár
NIR	near infrared reflectance
P	significance value
R	coefficient of correlation
R^2	coefficient of determination
SPSS	Statistical Program for Social Sciences
Std	standard

1. INTRODUCTION

Cereals have been a basic agricultural product since ancient times due to their nutritional properties, moderate cost and ability to achieve immediate satiation. Wheat is a plant grown on more land area than any other commercial crop. Wheat provides carbohydrate staple foods that form the basis of most diets, both ancient and modern, around the world (Jones et al., 2015). Wheat flour is used in a diverse range of end use products including bread, cake, noodle, cracker, cookie, and pasta (Khan, 2019). Wheat kernel is composed of endosperm (81–84%), bran (14–16%) and germ (2–3%). The endosperm is rich in carbohydrate, lipid and protein (Hung, 2016). The bran is the outer layer protecting the kernel. The germ is the kernel's embryo that will grow into a new plant (Shewry, 2004).

Wheat is a cereal of special importance in the world cereal production. During crop production, both abiotic and biotic stresses occur, often acting in combination under field conditions (Mittler, 2002) and potentially increase sensitivity to pathogens. *Fusarium* head blight (FHB) is one of the most devastating fungal diseases of wheat and other small grain cereals and has caused serious epidemics worldwide (Bai et al., 2003). The fungal pathogen associated with this disease in wheat is *Fusarium* spp. (Kikot et al., 2011). During the wheat's flowering stage, *Fusarium* infection occurs when weather conditions become favorable. The infection begins in the middle of the wheat spike and then spreads throughout the rest of it, eventually causing the entire ear spike to turn white and the kernels to become light-weight and shriveled (Kelly et al., 2015). Occurrence of FHB poses a serious problem because of considerable economic losses caused by lowered yield and deteriorated grain quality (Bottalico and Perrone, 2002; Argyris et al., 2003; Prange et al., 2005), and possible contamination of infested grain with mycotoxins that are known to be harmful for both consumer and livestock health (Dexter and Nowicki, 2003).

Crop yield losses due to FHB represent a significant problem worldwide (Mesterházy et al., 2020). In many regions, severe intensity of FHB occurs in cultivated wheat approximately two to three times per decade (Shaner, 2003; Stack, 2003; Champeil et al., 2004). Severe yield losses can occur during the epidemic year which are largely determined by the weather (Mesterházy et al., 2020). Thus, growers use multiple control measures to protect crops against FHB infections and prevent yield loss.

FHB poses a toxicological risk due to the mycotoxin contamination of wheat. In addition, it may reduce the quality of grains, as it is manifested in their weight loss, carbohydrate and protein composition changes and the presence of fungal toxins (Magliano and Kikot, 2013). It can destroy starch granules, storage proteins, and grain cell wall and subsequently affect the quality of wheat flour (Dexter et al., 1997). Nightingale et al. (1999) showed that the consistency and the extensibility of the dough was decreased when *Fusarium* damaged grains were included in the flour constituents. This resulted in substantial reductions in loaf volume and was attributed to the presence of fungal proteases. Fungal proteases have been shown to lead to weak dough properties and unsatisfactory bread quality (Wang et al., 2005b). Additionally, *Fusarium* infection reduces gluten strength in wheat due to lower proportions of glutenin (Dexter et al., 1997). Those biochemical changes in grain composition and subsequent changes in wheat quality traits are caused by the incomplete accumulation of the kernel constituents through the mechanical blocking of vascular bundles by fungal mycelium (Goswami and Kistler, 2004; Kang and Buchenauer, 2000; Ribichich et al., 2000) and the secretion of fungal enzymes (Eriksen and Pettersson, 2004).

Moreover, during the invasion of the kernel, *Fusarium* spp. secretes enzymes such as carbohydrases and proteases that degrade the cell wall and the kernel components (Eggert et al., 2011; Dexter and Nowicki, 2003; Pekkarinen and Jones, 2000). As a result, FHB infection leads to poor end use quality (Dexter and Nowicki, 2003). The most important method for FHB control and the reduction of mycotoxin concentration is the use of FHB resistant wheat varieties, appropriate cultural practices, fungicides, biological control and crop rotation (Mesterházy et al., 2015).

Previous studies reported the effect of fertilization (Horváth, 2015; Kismányoky and Ragasits, 2005; Pepó, 2007; Salgó and Gergely, 2012; Matić et al., 2022), storage time (Móré et al., 2014) and transgenic plant (Rakszegi et al., 2005) on wheat quality. Research on the influence of *Fusarium* infection on technological quality of wheat seems relatively scarce compared to literature dealing with *Fusarium* mycotoxins, prevention strategies and epidemiology of FHB (Spanic et al. 2021). Although it is understandable, that the management of FHB has in the past primarily focused on food safety and therefore on the avoidance of mycotoxin contamination in grain, the effects of FHB on grain quality are not to be underestimated. Researchers have just recently become aware that quality assurance in FHB affected wheat is essential for wheat marketing. Because the wheat price is directly determined by its processing attributes, influence of FHB on wheat milling performance, flour properties and endproduct quality have just recently confirmed its partially adverse effects (Dexter and Nowicki, 2003).

1.1 Objectives

The aim of the research lies on studying the influence of *Fusarium* head blight on technological quality of wheat, while taking the following questions into account

- Is there a relationship between wheat quality parameters and *Fusarium* infection?
- In what way does *Fusarium* infection have an impact on wheat quality?
- Does the effect of *Fusarium* infection on wheat quality vary between the different wheat varieties used?

2. LITERATURE REVIEW

2.1 Wheat morphology and taxonomy

Wheat is used to produce bread, bulgur, coucous, pastas, semolina, biscuits, and baked goods. Wheat belongs to the kingdom *Plantae*, order *Poales* (*Glumiflorae*), family *Poaceae* (*Gramineae*), tribe *Triticeae*, genus *Triticum* (Shewry, 2009). *Triticum aestivum* is a cereal of temperate climates. The northern limit of wheat cultivation in Europe lies in southern Scotland (60° latitude) and occasionally beyond (central Scandinavia up to 64°). In North America wheat is grown to about 55° latitude (Williams et al., 2008). Wheat occurrence follows a similar pattern in the southern hemisphere. In the Alps, it is grown to an altitude of 1 500 metres above sea level (Hömmö and Pulli, 1993; Diallo et al., 2012). The minimum temperature for germination of *T. aestivum* seeds is between 3 and 4°C. Flowering begins above 14°C. The vegetative period is 120 to 145 days for spring wheat and 280 to 350 days for winter wheat. Some varieties of *T. aestivum* need long photoperiods; some, especially those cultivated in southern Europe, are insensitive to day length (Diallo et al., 2012; Distelfeld et al., 2009). Plants of the genus *Triticum* are annuals with spring or winter forms. They show the following morphological features: short ligule and spikelets that are sometimes hairy and a smooth, bald, usually hollow culm, 0.7-1.6 metre in height. Generally, the spike or ear is four sided and have a brittle or tough rachis (Zohary, 2012). The spikelets have two to five florets, each floret can produce one grain (caryopsis). The glumes are long, keeled with serrated lemmas and are either bearded or unbearded. Grains are loosely enclosed and easily threshed (Figure 1). The rachilla has thin walls and does not disarticulate on maturity. In case of *T. aestivum* spp. *spelta* (spelt wheat) the grains are hulled. For this reason, they cannot be dropped during the process of threshing (Zohary, 2012).

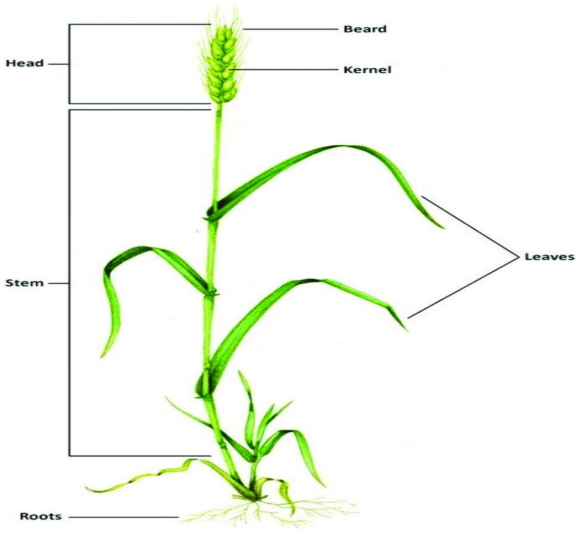

Figure 1. Wheat plant main parts (Mohammed et al., 2021)

2.2 Evolution of wheat

The first cultivation of wheat occurred about 10 000 years ago, as part of the 'Neolithic Revolution', which saw a transition from hunting and gathering of food to settled agriculture. The spread of wheat from its site of origin across the world has been described by Feldman (2001). The main route into Europe was via Anatolia to Greece (8000 BP) and then both northwards through the Balkans to the Danube (7000 BP) and across to Italy, France and Spain (7000 BP), finally reaching the UK and Scandanavia by about 5000 BP. Similarly, wheat spread via Iran into central Asia reaching China by about 3000 BP and to Africa, initially via Egypt. It was introduced by the Spaniards to Mexico in 1529 and to Australia in 1788 (Feldman, 2001).

These earliest cultivated forms were diploid (genome AA) (einkorn) and tetraploid (genome AABB) (emmer) wheats and their genetic relationships indicate that they originated from the south-eastern part of Turkey (Heun et al., 1997; Nesbitt, 1998; Dubcovsky and Dvorak, 2007). Hexaploidy bread wheat has only existed in cultivation by hybridization of cultivated emmer with the unrelated wild grass *Triticum tauschii* (also called Aegilops tauschii and Ae. Squarosa).

2.3 Cultivated wheat today

Currently, about 95% of the wheat grown worldwide is hexaploid bread wheat, with most of the remaining 5% being tetraploid durum wheat. The latter is more adapted to the dry Mediterranean climate than bread wheat and is often called pasta wheat to reflect its major end use (Nalam et al., 2006). However, it may also be used to bake bread and is used to make regional foods such as couscous and bulgar in North Africa (Jantasuriyarat et al., 2004). Small amounts of other wheat species (einkorn, emmer, spelt) are still grown in some regions including Spain, Turkey, the Balkans, and the Indian subcontinent. In Italy, these hulled wheats are together called faro (Szabó and Hammer, 1996) while spelt continues to be grown in Europe, particularly in Alpine areas (Fossati and Ingold, 2001).

The recent interest in spelt and other ancient wheats (including kamut, a tetraploid wheat of uncertain taxonomy, related to durum wheat) as healthy alternatives to bread wheat (Abdel- Aal et al., 1998) may also lead to wider growth for high value niche markets in the future.

2.4 Wheat success

Despite its relatively recent origin, bread wheat shows sufficient genetic diversity to allow the development of over 25 000 types (Feldman et al., 1995) which are adapted to a wide range of temperate environments. Provided sufficient water and mineral nutrients are available and effective control of pests and pathogens is ensured, yields can exceed 10 tonnes ha^{-1}, comparing

well with other temperate crops. However, deficiencies in water and nutrients and the effects of pests and pathogens cause the global average yield to be low, at about 2.8 tonnes ha^{-1}. Wheat is also readily harvested using mechanical combine harvesters or traditional methods and can be stored effectively indefinitely before consumption, provided the water content is below about 15% dry weight and pests are controlled (Feldman et al., 1995).

There is no doubt that the adaptability and high yields of wheat have contributed to its success, but these alone are not sufficient to account for its current dominance over much of the temperate world (Williams et al., 2008). The key characteristic which has given it an advantage over other temperate crops is the unique properties of doughs formed from wheat flours, which allow it to be processed into a range of breads and other baked products (including cakes and biscuits), pasta and noodles, and other processed foods (Barneix, 2007). These properties depend on the structures and interactions of the grain storage proteins, which together form the 'gluten' protein fraction (Mckevith, 2004).

2.5 Wheat production and trade

World wheat production is almost entirely based on just two wheat species; common wheat or bread wheat that accounts for about 95% of the world production and durum wheat that accounts for the remaining 5% of production (Shewry, 2009). As a result of extensive efforts made through national and international breeding programs wheat production per hectare has almost tripled and the area sown to wheat has doubled over the last 50 years (Curtis, 2002).

China ranks first in world wheat production having the largest cultivated land area for wheat (29.4 million hectares). United States, India, Russian Federation, Kazakhstan and Canada are the other top wheat producers in the world having 25, 24.9, 23.6, 12.6 and 11.5 million hectares of wheat cultivated wheat land, respectively (Shewry, 2009).

Wheat is also the world's most widely traded food grain with about 150 million tons or about 18% of world production traded each year. China being the world's largest wheat producer is also the world's largest wheat importer. Russian Federation, Egypt, Japan and Brazil rank among the other top wheat importers in the world. The United States, Canada, France and Australia are the largest wheat exporters in the world (Cisse et al., 2013).

2.6 Agronomic practices

In the Northern Hemisphere, depending on the location and the preceding crop, winter wheat can be sown from late August to late December. Sowing usually occurs between mid September and late October. Seeds of winter wheat need 40 to 70 days of vernalization with a temperature between

-1°C and +8°C. Hömmö and Pulli (1993) reported a maximum cold tolerance for winter wheat of about -25°C. Seeds of spring wheat need only 3 to 5 days or 0 to 14 days of vernalization. The commencement of growth of shoots is decisively influenced by the photoperiod in the case of spring wheat. The cold tolerance for seedlings of spring wheat is about -5°C (Hömmö and Pulli, 1993). The sowing season for spring wheat is from January to May.

In normal agricultural practice *T. aestivum* is used in a crop rotation schedule. Sugar beet, grain legumes and corn (*Zea mays*) or fodder maize make good preceding crops. Oilseed rape and winter barley occupy large areas and are part of many crop rotation systems that include winter wheat. Wheat/fallow rotations are commonly used in the western Great Plains region of the United States. Problems with plant diseases may arise from the frequent use of wheat as part of the crop rotation system (Denčić et al., 2012).

A great number of dicotyledonous and fewer monocotyledonous weeds have been reported to occur in fields used for wheat production. Seeds of some of these, when harvested and mixed with the wheat grain, can reduce flour quality (Barclay, 1975). As with all crops cultivated and harvested at the field scale, some seeds may escape and remain in the soil until the following season when they germinate either before or following seeding of the succeeding crop. In some instances, these “volunteers” may give considerable competition to the seeded crop and warrant chemical and/or mechanical control. The problem of volunteer plants in succeeding crops is common to most field crop species. Much depends on the management practices used in the production of the crop, e.g. the speed of the harvesting operation which will determine whether more or less seed is lost by the harvester. A suitable soil treatment after the harvest can considerably reduce the volunteer problem (Cisse et al., 2013).

2.7 Growth and development of wheat plant

The growth cycle of wheat consists of five phases: 1) Germination, seedling establishment and leaf production 2) Tillering and spike differentiation 3) Stem and spike growth 4) Spike emergence and flowering 5) Grain filling and maturity.

Germination starts when a kernel is sown in the presence of water. The radical and seminal roots initiate from the level of the seed and at the crown of the seedling. Consequently, the growth of the coleoptile ceases and the first leaf emerges through the tip as soon as it has grown out of the soil.

After seedling establishment, the plant starts to produce leaves at a rate of one per 4 or 5 days. Usually there will be a total of 8 or 9 leaves produced. Emergence of the flag leaf is an important

stage in the growth of the plant and this helps for timing the application of plant growth regulators (Simmons et al., 1995).

Tillering is one of the most important stages in the growth of the plant; it allows plants to compensate for low plant populations. Lower leaves on the main shoot and the coleoptile are the points of attachment from which tillers are formed. Variety and the growing conditions are the decisive factors influencing the number of tillers formed in a plant. Even though all tillers do not produce grain, other than the main shoot, there may be a total of three tillers per plant under usual growing conditions. The tillers which appear fourth to sixth from the main shoot have the higher probability of producing grain (Evers et al., 1999).

Parallel to the tillering, formation of spikes on the tillers and main shoot can be observed. Parts which will possibly become kernels and floral structures are being formed in this microscopic spike. The "boot" stage is just prior to spike emergence, when the flag leaf sheath encloses the growing spike. Booting is defined as the stage, when the spike can be felt within the whorl of leaf sheaths, but this is not visible, booting stage ceases when heading of the plant begins (McMaster, 2009).

The ‘heading’ of the plant occurs when stem continues to elongate in such a way that the spikes pushed out of the flag sheath. Flowering, also known as pollination, initiates in the spike. It starts with the florets in the central spikelets. Flowering can be observed by when anthers are thrust out from the floret. But this depends on the conditions such as weather. When pollination starts, anthers within a floret turn to yellow or gray. Pollination within a single spike takes around four days. The young kernels within a spike vary considerably in size at pollination and maintain this size variation throughout grain filling to maturity (Figure 2).

Usually, the growth of the kernel takes around three weeks. The kernel grows in three phases. Under the first phase, numerous cells in the endosperm are formed. Endosperm is the main protein and starch storage section of the kernel. Secondly, a couple of weeks after pollination, the kernel starts producing protein and starch. It causes the dry weight of the wheat grain to increase. Unfavourable environmental conditions during the above mentioned growth periods of the kernel might decrease the yield as a result of a low rate of dry matter accumulation. Necessarily, the longer the harmful weather conditions exist during grain filling, the greater the effect on yield. Products of photosynthesis that are produced during the grain filling stage contribute 70 to 90% of the final grain yield. The flag leaf and the spike also give a significant contribution (about 50%) of the photosynthate to grain filling, but the amount depends on the environmental conditions;

therefore maintenance of green and functional upper leaf blades, sheaths and spikes during grain filling results in better grain yields (Kirby and Appleyard, 1987).

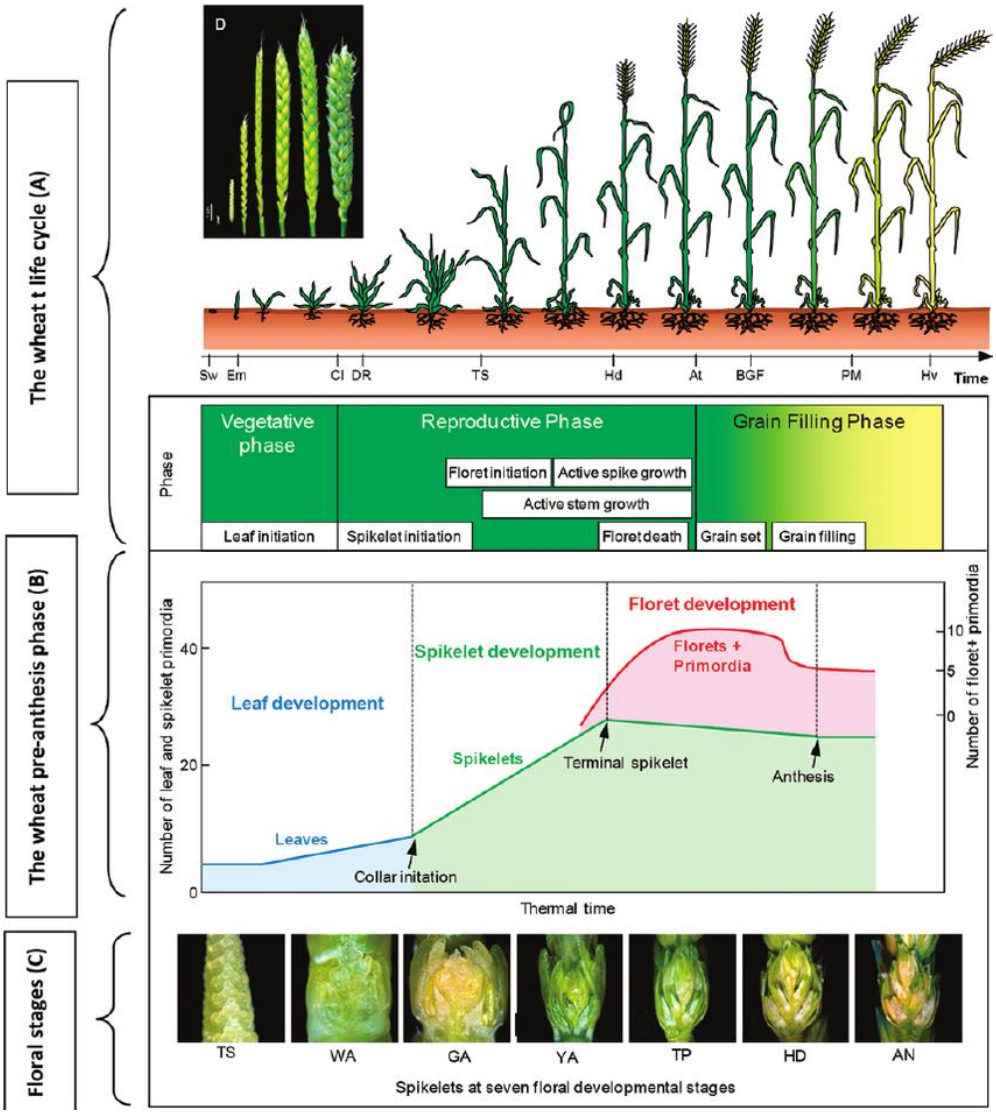

Figure 2. Different developmental stages in the wheat life cycle. (A) The wheat life cycle, Sw (sowing); Em (emergence); DR (double ridge appearance); TS (terminal spikelet initiation); Hd (heading); At (anthesis); BGF (beginning of grainfilling period); Pm (physiological maturity); Hv (harvesting). (B) The wheat pre-anthesis phase, including leaf, spikelet, and floret initiation. (C) The spikelets at the seven floral developmental stages, TS (terminal spikelet stage), WA (white anther stage), GA (green anther stage), YA (yellow anther stage), TP (tipping stage), HD (heading stage), and AN (anthesis stage) (from left to right). (D) The spikes at the seven floral developmental stages: TS, WA, GA, YA, TP, HD, and AN (from left to right) during floral development (floret initiation) (Kirby and Appleyard, 1987)

2.8 Characterization of the grain

2.8.1 Grain anatomy

Wheat grains is a single seeded fruit, called a caryopsis, in which that the pericarp is tightly fused with the seed. The seed itself consists of the testa, the endosperm and the embryo (Figure 3).

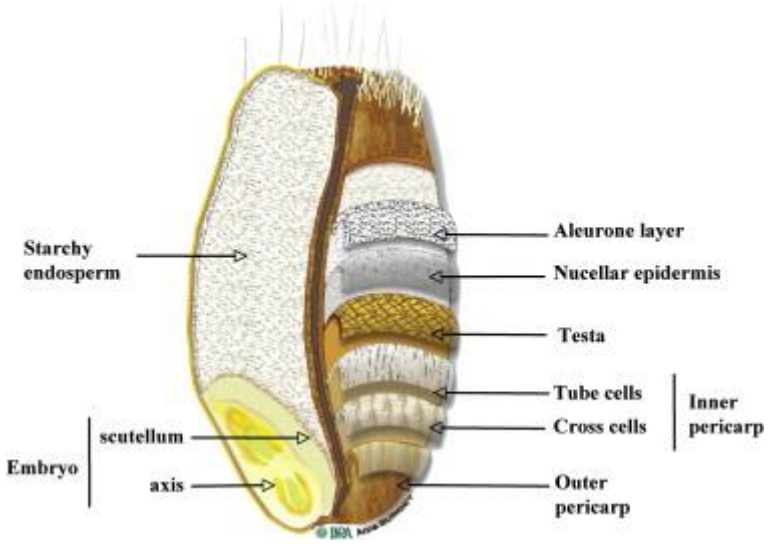

Figure 3. Anatomy of wheat grain (Surget and Barron, 2005)

2.8.1.1 Pericarp and testa

Pericarp and testa are the external layers of the grains, which provide support and protect the interior parts of grains during development from the external environment (Bewley et al., 2013). They comprise more than 80 % fibres, mainly water insoluble fibres, which serves as semi permeable barrier (Šramková et al., 2009). In addition, they have a cuticle layer, consisting fatty and waxy substances, which play a role in regulating water and gaseous exchange in growing grains, but become leaky on drying (Bewley et al., 2013).

Wheat grain has a unique morphological structure on the ventral side i.e., the opposite side to the embryo called “crease”, which is parallel to grain long axis. The role of crease is to facilitate the translocation of nutrients from the vascular strand to the developing grain. Furthermore, hairs, also named trichomes, are present on the ventral side of grain (Evers and Millar, 2002). In addition, the chloroplasts are present in wheat’s pericarp in which gives the grain its green color in early stage of development (Evers and Millar, 2002).

2.8.1.2 Endosperm

Endosperm has the largest morphological proportion in cereal grains, it makes up 80 - 84 % in wheat grain (Stevenson et al., 2012). It consists of the outer aleurone layer and the inner starchy endosperm. Most of the aleurone layer is removed as part of the bran during milling (Stevenson et al., 2012). The aleurone layer is a uniform single layer of cells (Šramková et al., 2009; Balconi et al., 2007), it surrounds the starchy endosperm and the embryo (Evers and Millar, 2002). It plays a vital role in both grain development and germination. In developing grain, the aleurone layer has the ability to divide starchy endosperm cells (Evers et al., 1999). A further function of some aleurone cells, which are called "transfer aleurone layer", is to facilitate the uptake of nutrients from the maternal tissues into starchy endosperm and embryo during the grain maturation (Balconi et al., 2007). The inner endosperm is referred to as mealy or starchy endosperm (Šramková et al., 2009). It consists of cells packed with nutrients, starch, protein and lipid storage, which can mobilize to support the growth of embryo on germination process (Stevenson et al., 2012). At maturity, endosperm becomes non-living, but aleurone cells remain alive for long period (Fox and Manley, 2009).

2.8.1.3 Embryo

The embryo is the reproductive organ, in which contains the scutellum and an embryonic axis. The scutellum acts as a secretory and absorptive organ, leading to de novo synthesis and secretion of hormones and enzymes as well as absorption of solubilized nutrients during germination. The embryonic axis is the plant of the next generation. It is composed of the radical and plumule (Evers and Millar, 2002).

2.8.2 Grain composition

Starch is the most abundant storage carbohydrate in all cereal grains, it provides the energy for seedling during germination (Beckles and Thitisaksakul, 2014). It accounts for 65 % of the dry matter of the wheat grain (Evers et al., 1999). Starch synthesis and storage take place mostly in the amyloplast in the growing endosperm, while some temporary starch reserves appear during development of the embryo and testa (Tetlow, 2011; Keeling and Myers, 2010). Protein content is in a relatively narrow range in cereal grains with 10 - 15 % of the grain dry weight (She et al., 2011). Proteins of wheat and maize have primary deficiency in lysine and methionine, whereas the secondary deficiency is threonine in wheat and tryptophan in maize. Sugars occur in low content in mature cereals grains 1 - 2 % of the grain dry weight, while relatively high amount of them is accumulated in immature grains (Evers et al., 1999; Watson and Ramstad, 1994). They are

composed of monosaccharides, glucose and fructose, and disaccharides, sucrose and maltose (Halford et al., 2011). Enzymes present in cereals are relatively low in mature grain, but they are necessary for grain development and germination (Koehler and Wieser, 2013). The majority of enzymes activity during developing period is concerned with synthesis, particularly the synthesis of storage components mainly starch and protein. However, some hydrolytic enzymes are found in the pericarp of the developing grain and may persist (Evers et al., 1999). Upon germination, the hydrolytic enzymes involved in the breakdown of starch and protein are predominant in order to provide the embryo with nutrients and energy (Koehler and Wieser, 2013; Minic, 2008). Furthermore, cereal grains contain various enzyme inhibitors such as α-amylase inhibitors (Juge and Svensson, 2006) and protease inhibitors (Sharma and Gupta, 2001). Maize grain is rich in lipids with 5 % of dry matter (Watson and Ramstad, 1994), whereas wheat grain contains 0.8 - 1.5 % of its dry matter (Black et al., 2006). Cereal grain contains fibers, minerals and vitamins from E and B-group (Koehler and Wieser, 2013).

2.9 *Fusarium* head blight in wheat

Fusarium head blight (FHB) is one of the most important diseases of wheat jeopardizing food and feed safety in many regions of the world. FHB was first described in 1884 in England and since then there have been increased FHB incidences across the world (Goswami and Kistler, 2004). *Fusarium* species belong to the Kingdom: *Fungi*, Order: *Hypocreales*, Phylum: *Ascomycota*, Family: *Hypocreaceae*, Class: *Euascomycetes*, Genus: *Fusarium* (Liddell, 2003). FHB pathogens are ubiquitous in nature (Miraglia et al. 2009) and cause disease sporadically, being of high incidence in some seasons yet low in others (Jennings and Turner, 1996). There were reports of a large rise in epidemic FHB outbreaks in America during the 1990s, and in areas of North America during 1993 it caused one of the greatest economic losses of any plant disease in a single year (McMullen et al., 1997). This epidemic was attributed to severely wet weather followed by humid periods and confirms how variable this disease can be between years. FHB can cause severe losses in grain yield (Saur, 1991), reductions in grain quality (Nightingale et al., 1999) and can cause contamination with mycotoxins (Prandini et al., 2009; Edwards, 2004).

2.9.1 Causal organisms and geographical distribution

A range of *Fusarium* spp. have been associated with the development of FHB in small grain cereals in Europe. These predominantly include *F. graminearum*, *F. avenaceum* and *F. culmorum*, but other less frequently encountered species can also be present including, *F. poae, F. cerealis, F. equiseti, F. sporotrichioides, F. tricinctum* and some even more sporadically identified species include *F. acuminatum, F. subglutinans, F. solani, F. oxysporum, F. verticillioides, F. semitectum*

and *F. proliferatum* (Bottalico and Perrone, 2002). Bottalico and Perrone (2002) describe how it is common to isolate from a single fragment of infected tissue, up to nine differing *Fusarium* species, and up to 17 in freshly harvested grain. The species profile of FHB pathogens in different geographic areas depends on multiple factors, but primarily upon climatic conditions, especially temperature (Miraglia et al., 2009). *F. graminearum* is associated with warm and humid conditions, *F. avenaceum* and *F. culmorum* with cooler, damper and humid conditions, *F. poae* with drier and warmer climates (Xu et al., 2008). The incidence of *Fusarium* species throughout Europe is split between the cooler north and central regions, and the warmer south. Isolates of *F. graminearum*, originating from both America and China, have also been shown to vary in their cultural characteristics and ability to cause FHB (Bai and Shaner, 1996) showing that that there could be significant differences in an isolates ability to cause disease may vary by geographical location. Significant quantitative variation in the aggressiveness of *F. graminearum* isolates has been observed within populations, this variation however did not differ between the Asian or European populations studied (Gagkaeva and Yli-Mattila, 2004).

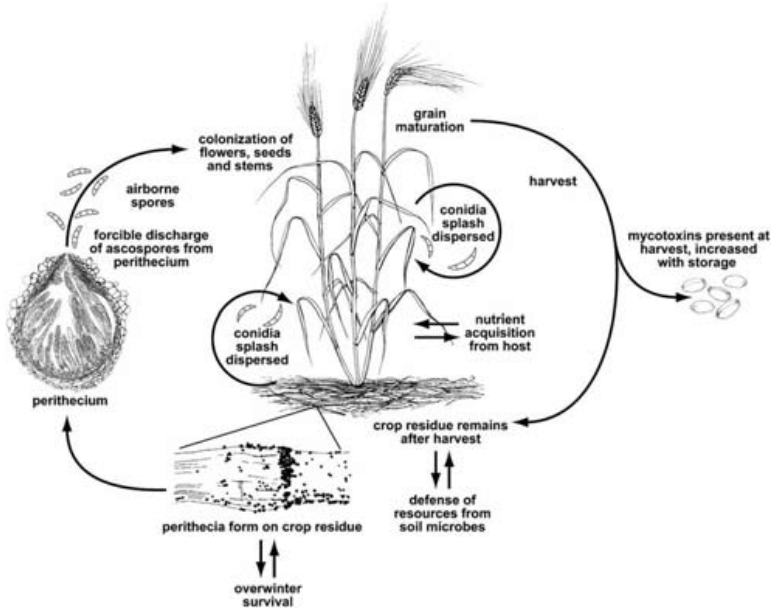

Figure 4. *Fusarium* spp. life cycle (Trail, 2009)

2.9.2 Dispersal and spread

Fusarium species are both saprophytes and facultative parasites that can colonize living hosts before establishing themselves in crop stubble through their presence in senescent tissue (Liddell, 2003). FHB inoculum can come from several sources, including soil and infected seed, as well as crop debris (Edwards, 2004; Nielsen et al., 2014). The plant debris is a primary source of FHB inoculum as it allows reserves of *Fusarium* pathogen to survive between seasons. This ensures FHB inoculum to be abundant within the environment (Osborne and Stein, 2007; Maiorano et al.,

2008). Both conidia and ascospores are able to cause FHB in cereals (Champeil et al., 2004; Snijders, 1990). Areas with an abundance of infected residues, such as regions producing cereal crops, contribute to the presence of airborne spores within the local area (Osborne and Stein, 2007). Such airborne spores can travel long distances (Maldonado-Ramirez et al., 2005) and should be considered an important dispersal method of FHB inoculum. Sutton (1982) found that disease severity and incidence of FHB had close relations to the quantity of primary inoculum present in the crop, and that since the disease is polycyclic it does not spread between ears.

2.9.3 Infection

The successful colonisation of a wheat ear by *Fusarium* pathogens causing FHB can be split into three distinct stages. These stages are: spore dispersal onto the wheat ear, entry into floral tissues and colonisation of the floret and spike (Figure 4).

The principal source of FHB inoculum being deposited onto the wheat ear is thought to be through the splashing and wind dispersal of ascospores and conidia, both of which are considered to be forms of aerial contamination (Champeil et al., 2004; Clement and Parry, 1998). Splashing is the main route by which macroconidia are spread, since they are too heavy to be transported by wind, whereas ascospores rely more on wind dispersal. (Champeil et al., 2004; Snijders, 1990). The spread of inoculum via leaf to leaf contact has been suggested as a route for FHB dispersal to the ear (Champeil et al., 2004; Atanassov, 1920) and this possibility has shown how, under field conditions, spores attached to the upper sides of leaves can be further spread to higher leaves towards the wheat ear by continuous splash events (Horberg, 2002). Furthermore to the role that wind dispersal plays in deposition of spores on ears, *Fusarium* spores have been found in the atmosphere at a height of 60 m, and consideration must therefore be given to the role that long distance transport of *F. graminearum* inoculum can play in contribution to regional epidemics, as atmospheric transport may move spores in the order of tens to hundreds of kilometres (Maldonado-Ramirez et al., 2005). Significantly more spores were found in the atmosphere during cloudy conditions than during clear conditions (Maldonado-Ramirez et al., 2005).

Once *Fusarium* spores have been deposited onto the ears of wheat, spores can exploit multiple pathways to gain entry into wheat floral tissues. Several pathways of entry have been shown, including via stomatal openings, direct penetration, and hyphal extension into the floret mouth (Lewandowski et al., 2006). There are large differences in the susceptibility of floral cells to infection between the internal and external surfaces. The external surface cells of the palea, lemma and glumes that are in contact with the environment, have extremely thick epidermal and hypodermal cell walls, which prevent direct penetration by the fungus (Shaner, 2003). The surface

cells within florets however have thin walls and lack resistance to *Fusarium* invasion (Lewandowski et al., 2006) making the internal surfaces of floral parts much more easily invaded than their external surfaces.

Primarily, *Fusarium* spores produce short infection hyphae that can penetrate the epidermal cuticle of the internal surfaces of the palea, lemma and glumes via the use of hydrolyzing enzymes e.g. cutinases and lipases (Walter et al., 2010). Intercellular fungal growth then occurs. Pectinases are released which begin to degrade the middle lamella and cell walls, and this, in turn, exposes other polysaccharides in the cell wall to degradation from hemicellulases and cellulase.

2.9.4 Disease development and symptoms

Once the *Fusarium* pathogen has gained initial entry into the floral tissues, the pathogen will continue to develop by colonising the floret and spreading within the ear (Brown et al., 2010; Walter et al., 2010). When the colonisation of the floral tissues has been achieved, *F. graminearum* spread down the rachis node to the central rachis, before eventually spreading up and down (Brown et al., 2010; Kang and Buchenauer, 2002). This spread leads to the invasion of further spikelets and the appearance of bleached ears. A distinctive symptom of FHB is premature bleaching of one or more spikelets or the entire immature wheat head. The bleaching can start anywhere on the head and spread until the entire head is bleached. Bleached spikelets are sterile or contain shriveled and/or discolored seed (Bushnell et al., 2010; Proctor et al., 1995). During humid conditions, white or pink fungal growth with orange spore masses may be seen on bleached spikelets. During warm temperatures (77^{o}F to 86^{o}F), blight symptoms on heads appear within three days following infection (Brown et al., 2010). Often, the infected kernels have a rough, shriveled appearance, ranging in color from pink, soft-gray, to light-brown. If this grain is used as seed for the following year's crop, severe seedling blight can result (Xu et al., 2008). Several factors influence the incidence of FHB epidemics each year, including previous cropping and tillage, varietal resistance and climatic conditions during flowering (Schaafsma et al., 2005).

2.10 Factors affecting fungal growth

Climate factors play a key role in determining fungal occurrence (Smith et al., 2016) so the activity of the fungi and their level of colonization are much determined by predominant environmental conditions most importantly humidity and temperature specially on the field (Magan and Olsen, 2004). Those factors, according to Doohan et al. (2003) influence the development, survival, distribution, and frequency of mycotoxigenic fungi and their subsequent toxin accumulation (Doohan et al., 2003). Temperature and humidity also affect plant growth, strength, and health and influence the competitiveness of mycotoxigenic fungi (Richard et al., 2003). When humidity and

temperature conditions are favorable, fungal invasion can take place and occur at different stages on the field (Perdoncini et al., 2019; Joubrane et al., 2020).

As for temperature requirements, most fungal species grow within a temperature range of 5 to 35 ℃ with optimum growth taking place at a range between 25 to 30 ℃ (Dix and Webster, 1995). Temperature conditions that promote growth are related to chemical reactions necessary for development that happens inside the fungi most efficiently at the optimal temperature ranges allowing for an accelerated growth pace. However, when the temperature shifts from the optimal range the reaction rate declines or may even stop leading to a growth halt (Kamil et al., 2011). Conditions that promote fungal growth may not always lead to mycotoxin production. However, generally, a temperature range between 25 and 30 ℃ and relative humidity between 88% and 95% are considered as favorable for fungal growth and subsequent mycotoxin production (Thanushree et al., 2019).

2.11 Losses due to *Fusarium* head blight

FHB causes crop losses in a multitude of ways, including reductions in grain yield, reductions in grain quality and contamination with harmful mycotoxins.

2.11.1 Grain yield

Growers use multiple control measures to protect crops against FHB infections and prevent yield loss. Severe yield losses can occur during epidemic years, which are largely determined by the weather (Parry et al., 1995). Yield reductions of between 6-39% have been reported in experiments containing over 500 wheat lines, inoculated with *F. graminearum* and *F. culmorum* spores (Saur, 1991). Parry et al. (1995) revised studies that quantified the loss of yield in wheat in the field under natural occurrence of the disease from 15-70 % in different countries of the world. Controlled inoculation trials with different *Fusarium* spp. caused losses from 3-60 %. In China, wheat yield losses ranged from 20-40 % in severe epidemical years, in Canada, losses from 30- 70 % were mentioned for spring wheat (Bai and Shaner, 1994). McMullen et al. (1997) reported average losses to FHB of 45% in commercial crops under high disease pressure years. FHB epidemics lead to the increased production of shrivelled grains with a white or pink discolouration. Additionally, FHB epidemics can lead to seed-borne contamination with *Fusarium* spores. Infected grain, when used to establish new crops, can lead to poor establishment and reduced grain yield (Jones, 1999).

Snijders (1990) showed a significant positive relationship between yield reduction and FHB in wheat as well as that yield reduction mainly resulted from the loss of kernel weight and reduction

of kernel amount that was caused by FHB. Several studies have demonstrated that FHB damaged grain obtains a lower test weight (Shotwell et al., 1985; Tuite et al., 1990; Jones and Mirocha, 1999) and generally shows a reduced thousand kernel weight compared to healthy grain (Seitz and Bechtel, 1985; Meyer et al., 1986; Dexter et al., 1996; Jones and Mirocha, 1999). Schade-schütze et al. (2000) demonstrated that thousand kernel weight of wheat was reduced in a range from 14-61% after artificial inoculation with *F. culmorum*.

2.11.2 Grain quality

Fungal growth leads to reduced nutritional and technical quality of cereal grains. Hence, diseased grain will have quantitative and qualitative losses (Turnbull and Rahman,2002; Wall,1979; Wicklow, 1995). Quantitative losses include the loss in vitamins, protein (albumins, globulins, gliadins, and glutenins), lipid, and carbohydrate (starch) content of the kernel. The quantity and quality of endosperm proteins are the major factors responsible for baking quality, and nutritional value of wheat (Finney and Barmore, 1948; Pomeranz, 2009; Blandino et al., 2015). Qualitative losses include discoloration, caking, and abnormal odors. With increasing fungal invasion, grain loses its natural luster and becomes rather dull and lifeless in appearance. General appearance alone is considered a quality factor in the routine inspection and grading of cereals. According to Christensen (1955), Papavizas and Christensen (1957), and Ranalli et al. (2003), it is highly probable that invasion of the germ or embryo of seeds by fungi is a major cause of discoloration. Damaged kernels can be identified by the brown to black color of the germ. Wheat damaged in this manner is frequently referred to as "sick wheat" (Pomeranz, 2009).

FHB epidemics have detrimental effects on the quality of wheat crops making them less suitable for their respective end uses. Epidemics reduce the hectolitre weight of grain (McMullen et al., 1997), reducing quality premiums from the milling sector or leading to the end use of grain being downgraded. Wheat grains, damaged by *F. graminearum* and *F. avenaceum*, have been shown to suffer from significant degradation of endosperm proteins (Nightingale et al., 1999) and starch granules (Snijders, 2004). Nightingale et al. (1999) showed that the consistency and resistance to extension of dough was decreased when *Fusarium* damaged grains were included in the flour constituents. This resulted in substantial reductions in loaf volume and was attributed to the presence of fungal proteases. Fungal proteases have been shown to lead to weak dough properties and unsatisfactory bread quality (Wang et al., 2005a). Additionally, reductions in gluten strength in durum wheat, through the inclusion of *Fusarium* damaged grains is likely caused by lower proportions of glutenin (Dexter et al., 1997; Bouachra et al., 2017; Aghagholizadeh et al., 2017) of which there was a linear relationship between *Fusarium* damaged grains and dough quality. The

quality of grain, for use as seed to establish new crops, is reduced through FHB epidemics as it can lead to poor establishment of crops (Haigh and Hare, 2012).

Fusarium head blight poses a toxicological risk due to the mycotoxin contamination of wheat. In addition, it may influence grain components such as starch and proteins (Siuda et al., 2010) and impair wheat quality essential for baking performance (Lancova et al., 2008). Those biochemical changes in grain composition and subsequent changes in wheat quality traits are caused by the incomplete accumulation of the kernel constituents through the mechanical blocking of vascular bundles by fungal mycelium (Goswami and Kistler, 2004; Kang and Buchenauer, 2000; Ribichich et al., 2000) and the secretion of fungal enzymes (Eriksen and Pettersson, 2004; Alconada and Kikot,2013). During the invasion of the kernel, *Fusarium* spp. secretes enzymes such as carbohydrases and proteases that degrade the cell wall and the kernel components (Eggert et al., 2011; Dexter and Nowicki, 2003; Pekkarinen et al., 2000). As a result, FHB infection leads to poor end use quality (Alconada et al., 2019).

2.12 *Fusarium* mycotoxins and their occurrence in grains

The word “mycotoxin” is derived from “myco” and “toxin”, greek words “mykes” and “toxikon”, meaning mold and a poison produced by a living organism. The term “mycotoxins” defines secondary fungal metabolites (metabolites not essential to the normal growth and reproduction of the fungus) that cause biochemical, physiologic, and/or pathologic changes in other species, including vertebrates, other animal groups, plants, and other microbes. Mycotoxins have low molecular weight molecules (M_w < 700) and are toxic in low concentrations (Soriano, 2007; Haschek and Voss, 2013). Even if hundreds of compounds have been classified as mycotoxins, and have been isolated and chemically characterized, only approximately 50 have been studied in detail (Cast, 2003).

The major mycotoxins occurring in wheat, at levels of potential concern for human and animal health, are *Fusarium* mycotoxins (Binder et al., 2007; Rodrigues and Naehrer, 2012). Results from worldwide mycotoxin occurrence studies indicate that deoxynivalenol (DON), zearalenone (ZEN) and fumonisins (FBs), are the most common mycotoxin contaminant of wheat and wheat-based products. Moreover, results highlighted the presence of considerable differences regarding the type and prevalence of mycotoxin contamination in different regions of the world, confirming that contamination is strongly dependent on regional climatic conditions (Streit et al., 2012; Schatzmayr and Streit, 2013). Differences in mycotoxin occurrence and concentration between distant geographical areas are uncontroversial. Within each geographical area, seasonal and local weather conditions during critical crop growing stages are of great importance to explain the

variation in mycotoxin occurrence. In general, environmental conditions, such as excessive moisture, temperature extremes, humidity, drought conditions, insect damage, crop systems, and some agronomic practices, can cause stress and predispose wheat in the field to mold and determine the severity of mycotoxin contamination (Hussein and Brasel, 2001; Cotty and Jaime-Garcia, 2007). Moreover, the high variability in the occurrence and level of mycotoxins may be the results of several factors, such as the years of the surveys, the annual weather fluctuations, and the storage conditions (Visconti and Pascale, 2010; Yan et al.,2022).

Another important point highlighted from studies on the worldwide mycotoxin occurrence in wheat and cereals is that the levels of detected mycotoxins are extremely variable. Average levels of mycotoxin contamination may be low and rarely exceed risk threshold levels, but as the content range is very wide, several samples may exceed the maximum or recommended levels for mycotoxin contamination (Streit et al., 2012; Schatzmayr and Streit, 2013).

Another important point highlighted from mycotoxin research is that mycotoxin co-contamination is more the rule than the exception. Several studies reported a high incidence of multi-mycotoxin contamination in cereals and agricultural commodities (Streit et al., 2012; Schatzmayr and Streit, 2013). A recent survey showed that in 2015, 46% of wheat samples were co-contaminated by two to six mycotoxins (Alkadri et al., 2014). A study carried out in Italy showed that at least 80% of wheat samples were contaminated with one mycotoxin, while two mycotoxins were found in 27% of contaminated samples; 38% of the analyzed samples were contaminated with three or more mycotoxins (Alkadri et al., 2014). Multi-mycotoxin contamination is a topic of great concern, as co-contaminated samples, although at lower levels than those indicated by EU regulations, might still exert adverse effects on animals due to additive/synergistic interactions of the mycotoxins.

A further scenario is represented by the climate changes. Estimates suggest that climate change will reduce wheat production globally by 29–34% by 2050 in developing countries (Hellin et al., 2012). This will have a great impact on food security. In terms of food safety and mycotoxin contamination, although aflatoxin is the mycotoxin that is most likely to increase under near future climate scenario, problems concerning also *Fusarium* toxins may represent a challenge if the temperature increases in cool or temperate climate countries (Marroquín et al., 2014).

2.13 Mycotoxin exposure and health impact

Mycotoxins commonly enter the food chain through contaminated food and feed crops, mainly cereals. FAO estimated that approximately 25% of the cereals produced in the world are contaminated by mycotoxins, but perhaps this value is closer to 50%, if one takes into account

emerging mycotoxins of which so far have limited data. The accumulation of mycotoxins in foods and feeds represents a major threat to human and animal health as they are responsible for many different toxicities, and it has a big economic significance (Oancea and Stoia, 2008; Coppock and Dziwenka, 2014). Acute exposure to high levels of mycotoxins is not very common, but the adverse effects in a chronic exposure continue to attract worldwide attention because of their impact on human health (Ferrante et al., 2012; Haschek and Voss, 2013; Marroquín et al., 2014).

Mycotoxins cause diseases in human and animals called mycotoxicosis and its severity depends on the toxicity rate of the mycotoxin (Peraica et al., 1999), the exposure route, the extent of exposure (duration and intensity), the age and nutritional status of the individual, and the potential synergistic effects with other chemicals, including other mycotoxins, to which the individual has been exposed (Peraica et al., 1999). Mycotoxicosis may be manifested as acute to chronic, and ranges from rapid death to tumor formation. Mycotoxicosis is the consequence of ingestion of grains or forage containing toxic metabolites produced by certain fungi. Fungi that produce toxins often do so only under specific conditions of warmth, moisture and humidity.

According to EFSA and FAO there are three mycotoxins, that occur quite often in food deooxynivalenol, zearalenone and fumonisins.

3.13.1 Deoxynivalenol

Deoxynivalenol is commonly present in cereal grains, animal feeds and forages. DON is often used as a marker for *Fusarium* infection (Sypecka et al., 2004). DON causes economic losses in livestock production and may pose a health risk to humans consuming contaminated cereal products (Völkl et al., 2004; Rotter et al., 1996).

Reduction in weight gain and impaired resistance to infection are the clinical manifestation observed in animals and poultry exposed to DON. Clinical signs and pathology observed during acute DON exposure include nausea, vomiting, feed refusal, skin irritation, and gastrointestinal lesions (Sypecka et al., 2004). Ingestion of DON at moderate to low concentrations by livestock is associated with reduced performance and immune function. The overall effect of ingestion of a low concentration of DON appears to be reduced feed consumption but higher concentrations are associated with vomiting (Rotter et al., 1996). DON can induce rapid diminution of lymphoid tissues and lymphopenia in chickens and mammals (Moon et al., 2007).

3.13.2 Zearalenone

Zearalenone (ZEN) is a natural toxin produced by *Fusarium graminearum* and *Fusarium culmorum* (Cast, 2003). ZEN may occur in infected wheat and is often found together with DON.

A common feature of many *Fusarium* species is their ability to synthesise ZEN and DON leading to toxicological interactions between these mycotoxins which can be additive or synergistic (Placinta et al., 1999). ZEN, a widely distributed oestrogenic fusariotoxin, constitutes a potential risk for human and animal health. ZEN is known to cause oestrogenic effects in animals including reproductive disorders and decreased fertility in variety of species with varying sensitivity (Kolf-Clauw et al., 2008).

Zearalenone is widely distributed in different commodities and its production is also favoured by environmental conditions such as high humidity and low temperatures (10-15 °C) (Domijan et al., 2005; Almeida et al., 2011; Grajewski et al., 2012; Pleadin et al., 2012; Pleadin et al., 2013; Krnjaja et al., 2013). Maize is the cereal at the highest risk of frequent and high level ZEN contamination, while wheat, oat and soybean products have been found to be contaminated only occasionally (Zinedine et al., 2007; Placinta et al., 1999).

3.13.3 Fumonisins

They are metabolites of *Fusarium* (*F. Verticillioides, F. proliferatum*). Fumonisins B1, B2 and B3 are the major fumonisins produced. The most prevalent is fumonisin B1 (FB1), which is believed to be the most toxic, i.e. nephrotoxic and hepatotoxic (Voss et al., 2007). High concentrations of fumonisins are associated with hot and dry weather, followed by periods of high humidity (Domijan et al. 2005). FB1 is toxic and carcinogenic to rodents and there are data suggesting that fumonisins or *F. verticillioides* cause esophageal cancer or other human health problems. Contamination of feed with FB1 resulted in a diverse range of damage to animal tissues, including lesions to the esophagus, gastrointestinal tract, liver, lungs, and brain. In the animals hepatotoxic, nephrotoxic, neurotoxic and carcinogenic effects were observed. It is considered highly toxic to horses and pigs, while in poultry significantly higher levels generally not cause a change in the production characteristics (Leeson et al., 1995).

2.14 *Fusarium* head blight control strategies

2.14.1 Cultural control

Cultural control is an environmentally friendly approach that can be used to reduce the risk of FHB epidemics. Numerous studies have been done to evaluate the effect of crop rotation on FHB development. Depending on the previous crop, the severity of FHB can be affected. Rotation of wheat with non host crops reduces the amount of inoculum in the crop residues (Sutton, 1982; Parry et al., 1995, Dill-Macky and Jones, 2000). Research showed that when wheat was grown following maize, FHB infection increased by 15% compared to only 4% infection when wheat was sown following alfalfa or oats (Pirgozliev et al., 2003). In another study, it has been found

that cultural practices such as tillage do not have significant effects on the disease severity and kernel infection (Miller et al., 1998). Miller et al, (1998) examined the effect of tillage on FHB disease incidence and suggested that the use of FHB resistant cultivars is more important in controlling FHB epidemics than tillage practices. Dill-Macky and Jones, (2000) evaluated the effect of crop rotation and tillage on FHB of wheat and reported that, FHB severity and incidence was less when wheat was grown after soybean than after wheat or corn irrespective of the tillage practice. Dill-Macky and Jones, (2000) also reported that conventional tillage and no till systems contributed to FHB epidemics in the Upper Midwest. Schaafsma et al. (2005) reported that previous crop, field size and tillage affect the FHB index, DON accumulation and *Fusarium* damaged grains in infected fields. Studies done by Schaafsma et al. (2001) also reported that tillage had no effect on DON levels in infected wheat grains. Guo et al. (2010) quantified the effects of cropping practices on *F. graminearum* inoculum levels and developed a cropping practice index (CPI) model to express the relationship. Applications of nitrogen fertilizers, can, however, increase the incidence of *Fusarium* damaged grains in wheat, barley and triticale (Martin et al., 1991). But Teich and Hamilton, (1985) reported that application of nitrogen fertilizers had no significant effect on the FHB disease incidence. According to Yi et al. (2001) application of nitrolime reduced the incidence of FHB by 59% but no significant reduction of DON accumulation was observed. Weed control is another cultural practice that can be adopted to reduce the FHB. Instead weed can act as an alternative source of FHB inoculum, control of weeds can reduce the availability of alternative FHB inoculum (Pirgozliev et al., 2003). Fields with higher weed densities had higher numbers of infected heads than the weed-free fields (Teich and Nelson, 1984).

2.14.2 Biological control

Biological control is an important part of an integrated FHB management system. Biological control is an environmentally friendly, durable method that is compatible with other control strategies (Schisler et al., 2002). The strategies for biological control of FHB include the control of the pathogen by disrupting the fungal life cycle using nonpathogenic microorganisms. Spikelet infection, colonization, ascospore production and dispersal are considered to be potential points for this biological intervention (Luz et al., 2003). Biological control of FHB mainly includes treatment of crop residue with antagonists to reduce the pathogen inoculum or application of antagonists to wheat heads during anthesis to reduce fungal infection (Schisler et al., 2002). The biological control of the pathogen may be achieved by aborting, curtailing or delaying the germination of the spores in the infection court of the head (Fernando et al, 2002). Antibiosis, competition, mycoparasitism, induced resistance and inhibition of mycotoxin synthesis are considered to be the major modes of action of biocontrol agents (Schisler et al., 2002; Luz et al.,

2003). Various research groups have examined the use of a wide range of microorganisms against the development of FHB (Stockwell et al., 1997; Luz et al., 2003). The Brazilian isolates of Bacillus and Paenibacillus are found to be the most effective biocontrol agents that can reduce the FHB disease severity in field by 50-67% (Luz et al., 2003). Schisler et al. (2002) isolated microbial strains from wheat anthers during anthesis and examined the feasibility of using those organisms in biological control of FHB. They could identify four strains that utilize tartaric acid and three that did not utilize tartaric acid as potential biocontrol agents from wheat anthers. These strains reduced the FHB disease severity up to 95% under greenhouse conditions and 56% under field conditions (Schisler et al., 2002). In another study, Schisler et al. (2006) identified 31 choline metabolizing strains from wheat flower tissue; all of them reduced FHB disease severity by 25% in a greenhouse trial where 17 of them reduced the disease severity up to 50% on wheat. Khan and Doohan (2009) reported that, Pseudomonas fluorescens strains MKB 158 and MKB 249 significantly reduced both the FHB severity and mycotoxin contamination caused by *F. culmorum* on wheat and barley. Another Pseudomonas strain, Pseudomonas frederikbergensis also significantly reduced the FHB severity under both greenhouse and field conditions (Khan and Doohan, 2009). Shi et al. (2014) reported on use of Bacillus subtilis strains 53 and 71, and Pseudomonas fluorescens biov1 strain 32 and Streptomyces spp. strain 3 as potential biological agents for control of FHB. Also Fernando et al. (2002) examined three bacterial strains of Bacillus substilis (Ehrenberg) Cohn strains H-08, S-01, and L-01 and were found to reduce FHB disease severity. Ramarathnam et al. (2007) reported that Bacillus substilis strain DFH08 significantly inhibited the radial mycelial growth of *F. graminearum* by 60% compared to the control and reduced the disease severity in a green-house study. Khan et al. (2001) isolated seven novel antagonists from wheat anthers and examined the efficacy of those antagonists against three isolates of G. zeae on the wheat cultivar Norm. Xu et al. (2008) found that, strain ACM941 of Clonostachys rosea significantly reduced the FHB index, *Fusarium* damaged grains and DON content but less effective than the fungicide tebuconazole. Schisler et al. (2002) demonstrated the feasibility of using biocontrol agents to control FHB on durum wheat and found that yeast antagonists were more successful in reducing the FHB symptoms than bacterial antagonists on durum wheat. There is a lack of consistency between the performance of biocontol agents under controlled environmental conditions and natural field conditions. Biocontrol agents that are proven to be effective under controlled conditions do not perform in the same way under field conditions. This is one of the major issues in commercial biocontrol production. Therefore, future research should focus on identifying biocontrol agents and effective application technologies to restrict the FHB colonization and mycotoxin accumulation both under large scale field conditions and glass-house conditions (Luz et al., 2003). As biological control agents are also living organisms, they

may require specific or optimum conditions for their functioning (Fernando, 2003). Therefore presence of a favourable environment for a particular biocontrol agent ensures an effective control against *F. graminearum*.

2.14.3 Chemical control

Chemical control is one of the main parts of an integrated FHB management approach. Fungicides are currently used at both flowering stage and before flowering stage to reduce quantitative yield loss and mycotoxin contamination (Mullenborn et al., 2008). Effective fungicides should be safe products with short pre-harvest interval and have high efficacy in reducing FHB and DON. Traditionally should have optimum application rates and techniques and a reasonable price. To date, many fungicides with different active ingredients are being used to manage FHB (Vanova et al.,2001).

Little studies have been done regarding control of FHB with fungicides (Mesterházy, 2003). Several factors such as level of inoculum, cultivar resistance, climatic conditions, crop sensitivity and yield potential affect the success of fungicide application in controlling FHB (Mesterházy, 2003). Fungicides based on the triazole chemistry (tebuconazole, metconazole or prothioconazole) are considered to be the most effective among all available registered fungicides (Mesterházy et al., 2003; Edwards et al., 2001; Pirgozliev et al., 2008; Simpson et al., 2001). Triazole based fungicides inhibit the 14α demethylase, an enzyme that is essential for ergosterol biosynthesis (Klix et al., 2007). The efficacy of use of fungicides and the effects on FHB and mycotoxin contamination in the field are often conflicting. In some studies, it has been found that triazole fungicides such as metaconazole, tebuconazole, prothioconazole and tebuconazole were effective, resulting in reductions of head blight severity and mycotoxin contamination by 50-80% and 5-90% respectively (Matthies and Buchenauer, 2000). On the contrary, in another study, application of fungicides has resulted in an increased trichothecene accumulation (Gareis and Ceynowa, 1994; Simpson et al., 2001). Gareis and Ceynowa, (1994) observed that application of the fungicide, Matador to *F. culmorum* infected winter wheat, increased the NIV content in infected seeds. Application of the fungicide, Azoxystrobin also increased the DON content in infected grains (Simpson et al., 2001). Therefore, presence of conflicting evidence in the use of fungicides to control the development of FHB needs to be clarified. Fungicides may affect the severity of FHB symptoms and the amount of DON in harvested grain by either altering the proportion of trichothecene producing *Fusarium* spp. or altering the rate of DON synthesis (Hasan, 1993; Edwards et al., 2001). Hasan et al. (1993) studied the effect of fungicides on diacetoxyscirpenol and zearalenone produced by *F. graminearum* and observed that fungicides significantly reduced

the toxin content and fungal growth. It has been suggested that fungicides are more effective at the early stages of the *Fusarium* infection process such as during spore germination and growth of the germ tube. An in vitro study done by Klix et al. (2007) found triazole based fungicides inhibit ascospore germination.

The two main critical factors in use of fungicides to control FHB are the timing and rate of application. The best time to apply fungicides is the period after the emergence of the head. Because the systemic triazole fungicides do not move from leaves to head from the point of contact, early application can protect only the leaves not the heads (Mesterházy, 1995). Application of fungicides several weeks before wheat anthesis may be more harmful for non toxigenic microorganisms and can promote subsequent spread of toxigenic *Fusarium* species in the field (Henriksen and Elen, 2005). Fungicides should be applied from both sides of the plots as partial coverage reduces the control of FHB. The rate of application may vary with the type of fungicides applied and comes with the fungicide label. It has been found that the concentration of fungicides was highest in the glumes and gradually decreases when moving to lemma and the embryo (Mesterházy, 1995).

It is always recommended that fungicides be used with other management strategies such as tillage, crop rotation and resistant cultivars. The combined effect of several strategies would provide a better control with higher yield and less infection (Birr et al., 2022).

3. MATERIALS AND METHODS

3.1 Experimental design and statistical analysis

The experiment was conducted during two growing seasons 2020 and 2021 at the experimental field and laboratories of the Hungarian University of Agriculture and Life Sciences (MATE), Agronomy Institute, Gödöllő, Hungary. The experimental site is in a hilly area with a close to average climatic zone of the country (47° 35′ 40.8″ N 19° 22′ 08.4″ E, 210 m above sea level). The soil type of the experimental field is sand-based brown forest soil (Chromic Luvisol). The textural classification of the soil was sandy loam with parameters shown in table 1. A crop rotation of soybean, wheat and maize was implemented in the field. Prior to sowing, the field was cleared, ploughed, rotor-tilled and the seedbed was prepared. The plots were sown in october and harvested in middle of July with plot machines. The sowing depth was 5 cm. The rate of sowing was 450- 500 seeds per square meter. The wheat varieties used in the experiment were Alföld, Mv Kolompos and Mv Karéj. Each variety had a total plot area of 75 m^2. Each plot was then divided into 15 sub-plots of 5 m^2 each to create replications. At the end of the growing season, wheat grain samples were collected from each sub-plot, stored under laboratory conditions and measured for *Fusarium* infection level, protein content, gluten content, test weight, thousand kernel weight, falling number and Zeleny sedimentation index. *Fusarium* infection level percentage was calculated by counting the number of colonies that formed on wheat kernels disinfected with a solution of PCNB and chloramphenicol (100 kernels from each sample) incubated for 7 days under laboratory conditions on Nash and Snider *Fusarium* selective medium (Distilled water 1 l, Peptone 15 g, KH_2PO_4 1 g, $MgSO_47H_2O$ 0.5 g, Agar 20 g, PCNB 1 g, Chloramphenicol 100 ppm). Protein content, gluten content and Zeleny sedimentation index were measured with Mininfra Scan-T Plus 2.02 version. Falling number was measured with Perten 1400 system (ICC method No. 107/1 1995). Test weight was measured with chondrometer hectoliter grain tester (ISO 7971-3:2019). Thousand kernel weight and test weight were measured with the KERN EMS and the Sartorius MA-30 precision scales. IBM SPSS V.21 software was used for the statistical evaluation of the results, the linear regression module at 5% significance level was performed to determine the effect of *Fusarium* infection level on wheat quality parameters, and the analysis of variance (ANOVA) module at 5% significance level was performed to determine the effect of the growing season on *Fusarium* infection and wheat quality parameters.

Table 1. Soil type of the experimental field

	Humus %	pH (H_2O)	K_A	Sand %	Silt %	Clay %	$CaCO_3$
Medium	1.32	7.08	40	49	25	26	0

3.2 Meteorological properties of the experimental field

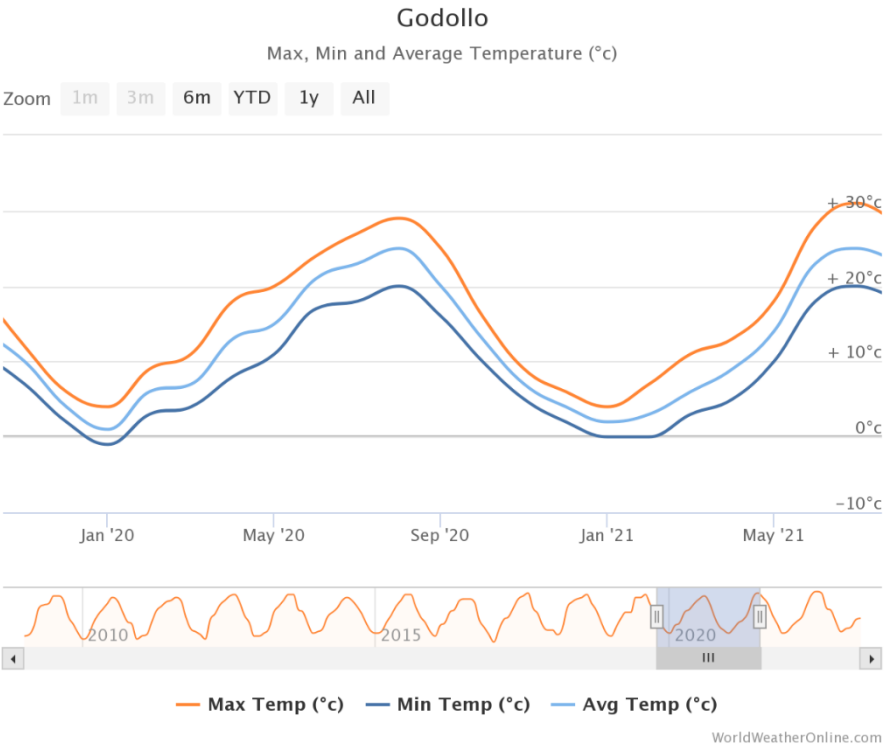

Figure 5.Temperature in 2020 and 2021 (worldweatheronline.com)

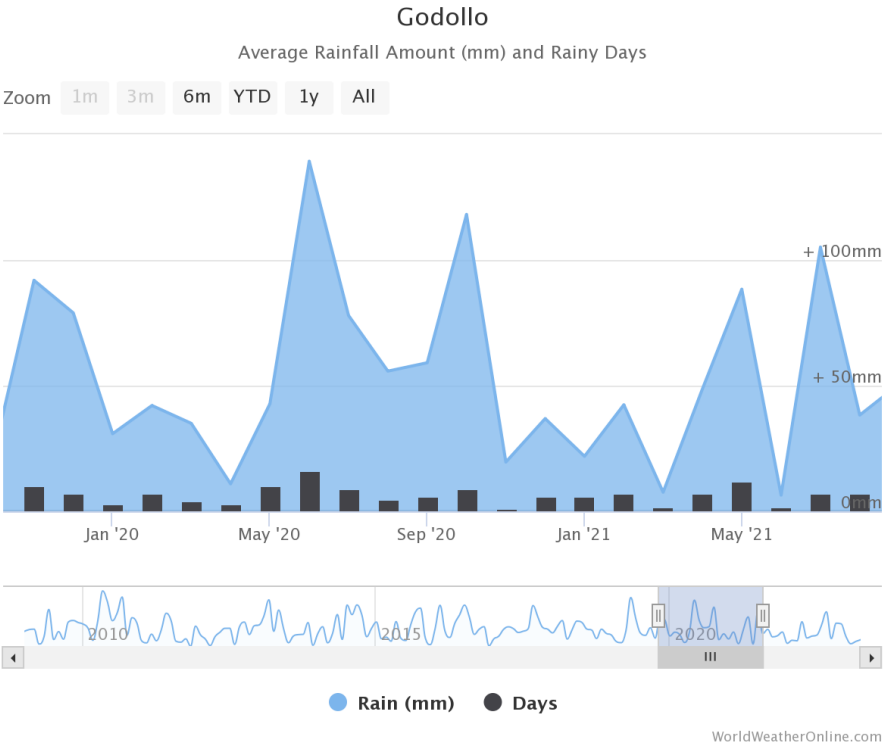

Figure 6. Rainfall in 2020 and 2021 (worldweatheronline.com)

Figure 5 provides information about the temperature, and figure 6 shows the rainfall amount and rainy days of Godollo experimental field in 2020 and 2021.

3.3 Studied winter wheat varieties

The present study examined the effect of *Fusarium* infection on wheat quality parameters of three winter wheat varieties:

- Alföld
- Mv Kolompos
- Mv Karéj

The characteristics of the three wheat varieties used are presented in table 2.

Table 2. Characteristics of the three wheat varieties used (Martonvásár 2020)

Quality values specific to the variety	Alföld	Mv Kolompos	Mv Karéj
Thousand Kernel Weight (g)	39-44	50-55	45-50
Hectolitre Weight (kg/hl)	78-82	75-78	82-85
Falling Number (seconds)	350-400	350-400	350-400
Protein Content (%)	14-17	12-14	11-13
Gluten Content (%)	34-40	30-35	29-32
Zeleny Sedimentation Index (ml)	31-55	36-46	31-55
FHB Disease Resistance (1-9)	6	6	6

3.4 Determination of falling number

3.4.1 General description

The Falling number (FN) method is an internationally standardized method for the determination of the level of alpha-amylase in grain, flour and other starch containing products, in particular wheat. It determines alpha-amylase activity using the starch in the sample as substrate. The method is based upon the rapid gelatinisation of a suspension of flour or meal using a boiling water bath and the subsequent measurement of the liquefaction, by alpha-amylaze, of the starch contained in the sample. FN values bear a complex inverse relationship with the quality of alpha-amylase in the sample. This relationship is known as the Perten Liquefaction Equation.(Figure 7).

3.4.2 Application of falling number

Falling number results are used to segregate grain into good quality for bread making and poorer grades suitable only for feedstuffs, or controlled mixing. FN product, it may also be used to calculate optimum milling blends or flour blends for better product control. FN results are used by the grain trader to establish the quality of the grain, for export or the local trade. FN results can be used at the bakery to determine the quality of the flour supplied, and to optimise flour blends to

suit individual products. FN results can be used to monitor the ripening process of grain to determine the optimum harvesting date, especially in areas that are subject to rain during the harvest period.

3.4.3 Equipment

• Falling number apparatus

• Digital balance

• Perten Type 1400 system

• Moisture balance meter

Figure 7. Falling number apparatus

3.4.4 Procedure

- A moisture test is done on a wheat sample that has been selected and ground
- The sample weight depends on the moisture content of the ground sample
- Distilled water (25 ml) is added to the ground sample in a falling number test tube
- The ground wheat and water mixture is thoroughly shaken, forming a slurry
- A stirrer is placed in each falling number tube
- Tubes containing the slurry are immersed in the boiling water bath of the falling number apparatus. The slurry is stirred with the stirrer for 60 seconds then the stirrer is allowed to drop by its own weight through the ground wheat and water slurry
- The total time in seconds it takes the stirrer to reach the bottom including the 60 seconds stirring time is the falling number result

3.4.5 Interpretation of the results

Below 200: High amylase activity, sprout damaged wheat. Bread crumb is likely to be sticky.

200 – 300: Optimal amylase activity. Bread crumb is likely to be good.

Above 300: Low amylase activity, sound wheat. Bread crumb is likely to be dry, and loaf volume reduced.

3.5 Determination of hectoliter weight

Hectolitre weight (or weight per unit volume) is the weight of 100 litres of wheat and is the simplest criteria of wheat quality. It gives us a rough index of flour yield. In USA and Canada, hectoliter weight is expressed in terms of lb/bushel (pounds per bushel) whereas in India and Europe it is expressed as kg/hectolitre. Higher the hectoliter weight, the better is the flour yield. The factors affecting the hectoliter weight are kernel shape and uniformity of kernel size, orientation of kernels in container when it is filled, density of the grain influenced by structure of grain and its chemical composition.

3.5.1 Equipment

- Digital balance
- Plastic container
- Chondrometer
- Plunger weight
- Cut-off slide

3.5.2 Procedure

- Insert the cut-off slide into the designated opening of the chondrometer
- Drop the plunger weight into the chondrometer, ensuring that it is resting flat on the cut-off slide
- Place the chondrometer with the cut-off slide and the plunger weight on the weighing platform
- Press on the tare/on button
- Remove the chondrometer from the balance
- Pour the grain sample into the plastic container
- Slowly fill the chondrometer to the top with the grain sample
- Remove the cut-off slide carefully, allowing that the plunger weight together with the grain to descent into the lower part of the chondrometer

- Re-insert the cut-off slide through the slot of the chondrometer and push it through the grain in one go; intervening grains are cut through
- Tip out the surplus grain
- Place the chondrometer on the balance
- Read off the mass and take the value for the hectoliter weight (kg/hl) from the calibration chart

3.6 Determination of thousand kernel weight

Thousand kernel weight (TKW) measures the mass of the wheat kernel and is an essential parameter for the selection of cultivars with the best physical and physiological seed quality. Generally, higher TKW values are positively related to potential flour extraction or yield, because this property is closely related to grain size and proportion of endosperm to germ and pericarp tissues. Wheat breeders and flour millers employ this method as a complement to test weight to better describe wheat kernel composition and potential flour extraction. TKW could be used as an index of wheat milling value and is a good parameter for evaluation of kernels as seed material. When the grain is undamaged may be expected high test weight, due to a greater endosperm to bran ratio. The values range from 30 to 50 g. Higher the value, better is the wheat for milling in terms of flour yield.

3.6.1 Equipment

• Digital balance

• Clean, dry wheat samples

• Plastic bowls

3.6.2 Procedure

• Clean the wheat samples

• Count 1000 wheat grains

• Record the weight of 1000 kernels

3.7 Determination of protein content, gluten content and zeleny sedimentation value

Wheat protein content is an important consideration for all end products from bread baking to noodles, paste, cakes, and biscuits. Wheat protein content varies widely depending on wheat class, growing region, type and quality of soil, the health of the grain and of course fertilizers input, nitrogen in particular. All other factors being equal, flour from higher protein wheat has greater

water absorbing capacity and thus greater bread volume potential, depending somewhat on the baking process used. Wheat protein content is strongly correlated to gluten content and both are used as quality indicators when wheat price is being negotiated between sellers and buyers.

Gluten, the protein component of flour which gives the dough elasticity and strength, can be defined as the rubbery mass that remains when wheat dough is washed to remove starch granules and water soluble constituents. Gluten ranges from 0 to 100. Gluten is thus classified as weak (0- 25), sufficient (26-45), medium (46-65), strong (66-85) or very strong (> 85). Gluten plays a key role in determining the unique baking quality of wheat by conferring water absorption capacity, cohesiveness, viscosity, and elasticity on dough. Generally, the higher a flour's protein content, the higher the gluten formation. During dough mixing, wheat flour is hydrated and the gluten proteins are transformed into a continuous cohesive viscoelastic gluten protein network.

Knowing the protein and gluten content is also not enough to fully characterize wheat flour. Therefore, it is useful to measure sedimentation value. The sedimentation value according to Zeleny (Zeleny value) describes the degree of sedimentation of flour suspended in a lactic acid solution during a standard time interval and this is taken as a measure of the baking quality. It is determined on the ground kernel or on the flour and it ranges from 0 to 80. Wheat having a Zeleny index below 20 is generally regarded as unsuitable for baking. Swelling of the gluten fraction of flour in lactic acid solution affects the rate of sedimentation of a flour suspension. Both a higher gluten content and a better gluten quality give rise to slower sedimentation and higher Zeleny test values. The sedimentation value of flour depends on the wheat protein composition and is mostly correlated to the protein content, the wheat hardness, and the volume of pan and hearth loaves.

The baking industry requires flours to have defined quality characteristics including protein content, gluten, and Zeleny sedimentation. This supposes that the flour milling industry must complete many physical and chemical analyses, with consequent economic and time-related costs. Therefore, NIR spectroscopy (NIRS), which is one of the various technologies for the evaluation of wheat flour quality, has become more popular and is attracting more attention from food researchers, recently.

The protein content, gluten content and Zeleny sedimentation index were determined by using NIR technique by running the grain samples through Mininfra Scan-T Plus 2.02 version. NIR spectroscopy is a method that makes use of the near-infrared region of the electromagnetic spectrum (from about 700 to 2500 nanometers). By measuring light scattered off of and through a sample, NIR reflectance spectra can be used to quickly determine a material's properties without altering the

sample. The instrument requires no sample preparation other than cleaning or removal of large impurities (Figure 8).

Figure 8. NIR apparatus

Advantages of near infrared spectroscopy

- Requires little or no sample preparation
- Highly flexible form of analysis
- Cost-effective
- Capable of examining irregular surfaces
- Can be carried out on the production line since the analysis is very simple and very fast
- Non destructive, it preserves the sample after the measurement for further analysis

3.8 Determination of *Fusarium* infection level

Kernels from each sample were surface disinfected with PCNB and chloramphenicol (100 kernels from each sample) and then cultured on Nash and Snyder *Fusarium* selective medium (Distilled water 1 l, Peptone 15 g, KH_2PO_4 1 g, $MgSO_47H_2O$ 0.5 g, Agar 20 g, PCNB 1 g). The culture media was supplemented with 100 ppm of chloramphenicol. Plates were incubated at 25^0C, and differential counting of fungal growth was made after 7 days of incubation. Colonies that were developed on kernels were not identified to species level but were classified as either *Fusarium* spp. or other fungal species. The percentage of kernels infected with *Fusarium* spp. or other fungal species was recorded (infection level (%) = total number of infected kernels × 100⁄total number of tested kernels).

4. RESULTS AND DISCUSSION

4.1 *Fusarium* infection level

Fusarium infection in 2021 (91.47%, 94% and 95.20%) was higher than in 2020 (44.33%, 48.4% and 40.27%), the difference was statistically significant [F = 135.813, P = 0.000], [F = 62.869, P = 0.000] and [F = 100.952, P = 0.000] in the three wheat varieties Alföld, Mv Kolompos and Mv Karéj used respectively (Table 3). Rainfall measurements were collected from World Weather Online Meteorological Service. Wheat heads are most susceptible to FHB infection during flowering period (May). Rainfall during the flowering period in 2021 (88.39 mm) was higher than in 2020 (42.8 mm), this increase in rainfall could explain the increase in *Fusarium* infection level. Simple linear regression is used to test the effect of *Fusarium* infection on the following wheat quality parameters: protein content, test weight, thousand kernel weight, falling number, gluten content and Zeleny sedimentation index.

Table 3. Descriptive statistics and ANOVA for the influence of growing season on *Fusarium* infection in Alföld, Mv Kolompos and Mv Karéj wheat varieties

Descriptive statistics		Mean	Std. Deviation	Std. Error	Minimum	Maximum
Alföld	2020	44.33	14.60	3.77	20	66
	2021	91.47	5.68	1.47	80	100
	Total	67.90	26.32	4.81	20	100
Mv Kolompos	2020	48.40	21.37	5.52	12	82
	2021	94.00	6.28	1.62	84	100
	Total	71.20	27.88	5.09	12	100
Mv Karéj	2020	40.27	20.71	5.35	12	88
	2021	95.20	4.39	1.13	88	100
	Total	67.73	31.57	5.76	12	100
ANOVA		Sum of Squares	*df*	Mean Square	*F*	Sig.
Alföld	Between Groups	16661.633	1	16661.633	135.813	0.000
	Within Groups	3435.067	28	122.681		
	Total	20096.700	29			
Mv Kolompos	Between Groups	15595.200	1	15595.200	62.869	0.000
	Within Groups	6945.600	28	248.057		
	Total	22540.800	29			
Mv Karéj	Between Groups	22632.533	1	22632.533	100.952	0.000
	Within Groups	6277.333	28	224.190		
	Total	28909.867	29			

4.2 Influence of *Fusarium* infection on protein content

In Alföld, protein content was lower in 2021 (13.41 %) compared to 2020 (14.75 %) the difference was statistically significant [$F = 20.862$, $P = 0.000$] (Table 4). *Fusarium* infection had a strong negative effect on protein content [$R = - 0.682$], protein content decreased when the infection increased. The fitted regression model between *Fusarium* infection and protein content is y = - 0.027x + 15.917. The regression is statistically significant [$R^2 = 0.465$, $F = 24.309$, $P = 0.000$] (Figure 9, Table 5).

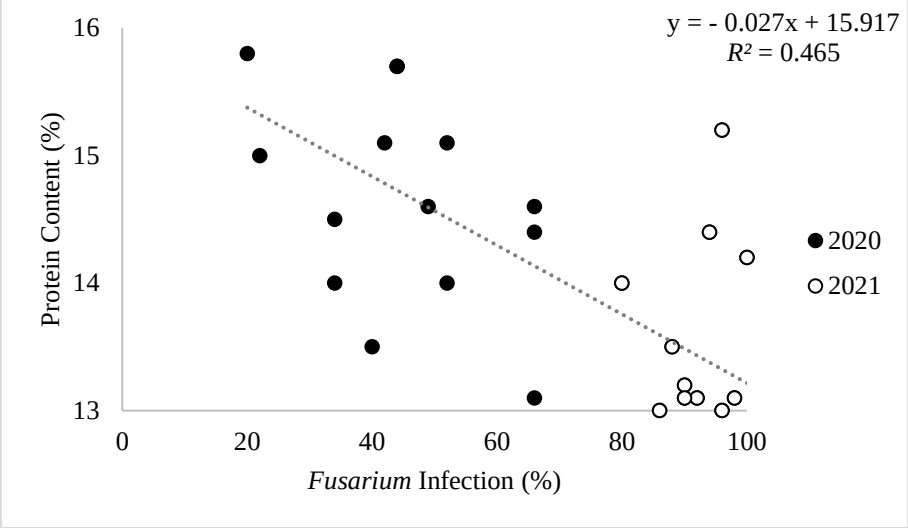

Figure 9. Influence of *Fusarium* infection (%) on protein content (%) in 2020 and 2021 growing seasons in Alföld wheat variety

In Mv Kolompos, protein content was lower in 2021 (14.49 %) compared to 2020 (15.2 %) the difference was statistically significant [$F = 10.559$, $P = 0.003$] (Table 4). *Fusarium* infection had a moderate negative effect on protein content [$R = - 0.426$], protein content decreased when the infection increased. The fitted regression model between *Fusarium* infection and protein content is y = - 0.011x + 15.667. The regression is statistically significant [$R^2 = 0.182$, $F = 6.218$, $P = 0.019$] (Figure 10, Table 5).

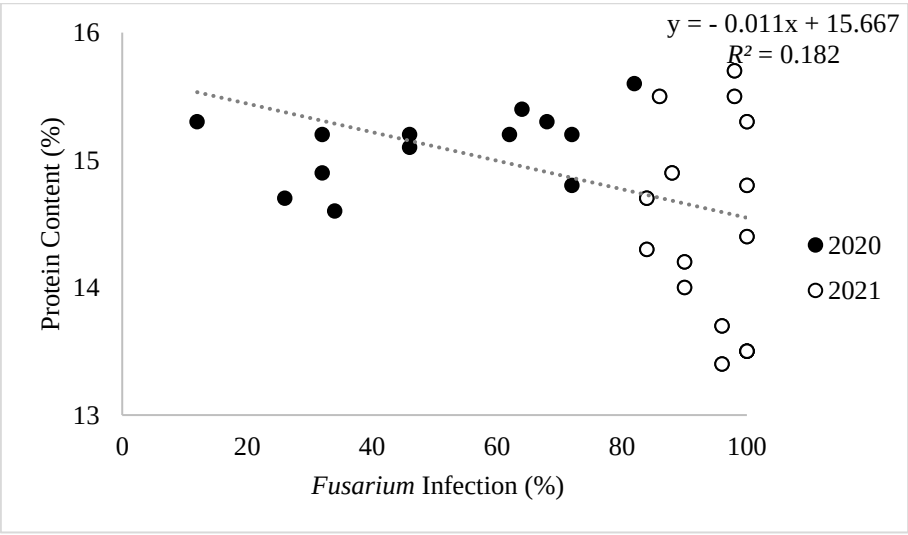

Figure 10. Influence of *Fusarium* infection (%) on protein content (%) in 2020 and 2021 growing seasons in Mv Kolompos wheat variety

In Mv Karéj, there was no statistically significant difference in protein content in 2020 and 2021 [F = 3.443, P = 0.074] (Table 4). *Fusarium* infection had no effect on protein content [R = - 0.310]. The fitted regression model between *Fusarium* infection and protein content is y = - 0.007x + 15.047. The regression is not statistically significant [R^2 = 0.096, F = 2.974, P = 0.096] (Figure 11, Table 5).

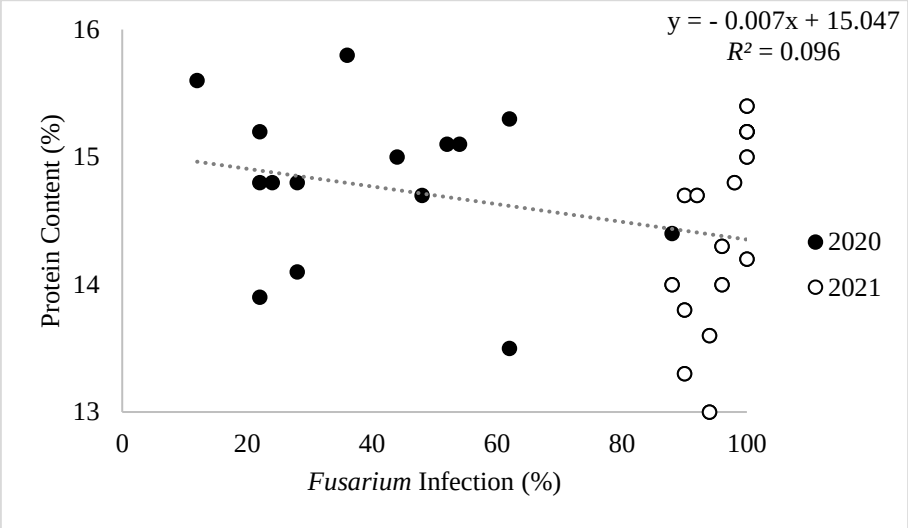

Figure 11. Influence of *Fusarium* infection (%) on protein content (%) in 2020 and 2021 growing seasons in Mv Karéj wheat variety

Table 4. Descriptive statistics and ANOVA for the influence of growing season on protein content in Alföld, Mv Kolompos and Mv Karéj wheat varieties

Descriptive statistics		Mean	Std. Deviation	Std. Error	Minimum	Maximum
Alföld	2020	14.75	0.88	0.23	13.10	16.20
	2021	13.41	0.71	0.18	12.70	15.20
	Total	14.08	1.04	0.19	12.70	16.20
Mv Kolompos	2020	15.25	0.44	0.11	14.60	16.10
	2021	14.49	0.78	0.20	13.40	15.70
	Total	14.87	0.73	0.13	13.40	16.10
Mv Karéj	2020	14.81	0.62	0.16	13.50	15.80
	2021	14.35	0.73	0.19	13	15.40
	Total	14.58	0.71	0.13	13	15.80
ANOVA		Sum of Squares	*df*	Mean Square	*F*	Sig.
Alföld	Between Groups	13.467	1	13.467	20.862	0.000
	Within Groups	18.075	28	0.646		
	Total	31.542	29			
Mv Kolompos	Between Groups	4.256	1	4.256	10.559	0.003
	Within Groups	11.287	28	0.403		
	Total	15.543	29			
Mv Karéj	Between Groups	1.587	1	1.587	3.443	0.074
	Within Groups	12.907	28	0.461		
	Total	14.494	29			

Table 5. Model summary, ANOVA and coefficients for the influence of *Fusarium* infection on protein content in Alföld, Mv Kolompos and Mv Karéj wheat varieties

Model summary		*R*	*R*^{2}	Adjusted *R*^{2}	Std. Error of the Estimate	
Alföld		0.682	0.465	0.446	0.777	
Mv Kolompos		0.426	0.182	0.152	0.674	
Mv Karéj		0.310	0.096	0.064	0.684	
ANOVA		Sum of Squares	*df*	Mean Square	*F*	Sig.
Alföld	Regression	14.658	1	14.658	24.309	0.000
	Residual	16.884	28	0.603		
	Total	31.542	29			
Mv Kolompos	Regression	2.824	1	2.824	6.218	0.019
	Residual	12.719	28	0.454		
	Total	15.543	29			
Mv Karéj	Regression	1.392	1	1.392	2.974	0.096
	Residual	13.102	28	0.468		
	Total	14.494	29			
Coefficients		Unstandardized Coefficients		Standardized Coefficients	*t*	Sig.
		B	Std. Error	Beta		
Alföld	*Fusarium*	-0.027	0.005	-0.682	-4.930	0.000
	(Constant)	15.917	0.398		39.989	0.000
Mv Kolompos	*Fusarium*	-0.011	0.004	-0.426	-2.494	0.019
	(Constant)	15.667	0.342		45.745	0.000
Mv Karéj	*Fusarium*	-0.007	0.004	-0.310	-1.725	0.096
	(Constant)	15.047	0.300		50.196	0.000

4.3 Influence of *Fusarium* infection on test weight

In Alföld, test weight was lower in 2021 (72.76 kg/hl) compared to 2020 (75.10 kg/hl) the difference was statistically significant [F = 25.338, P = 0.000] (Table 6). *Fusarium* infection had a strong negative effect on test weight [R = - 0.626], test weight decreased when the infection increased. The fitted regression model between *Fusarium* infection and test weight is y = - 0.041x + 76.714. The regression is statistically significant [R^2 = 0.391, F = 18.005, P = 0.000] (Figure 12, Table 7).

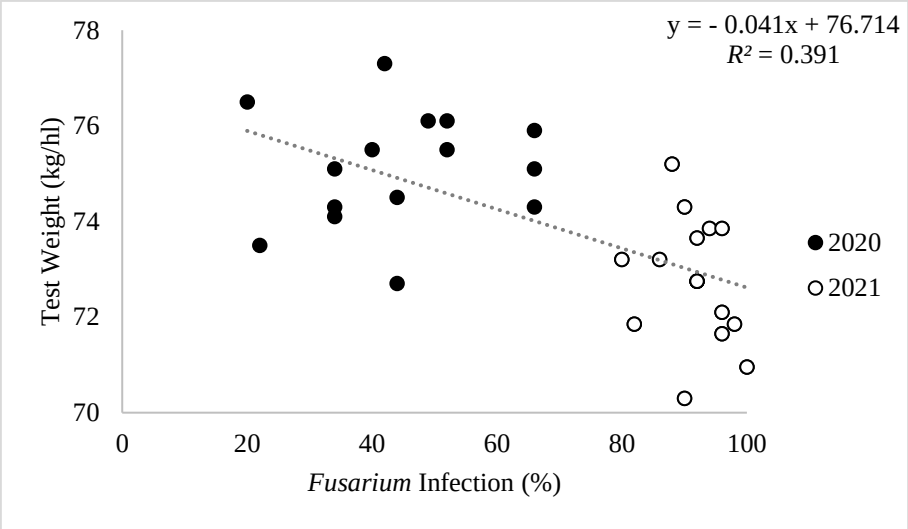

Figure 12. Influence of *Fusarium* infection (%) on test weight (kg/hl) in 2020 and 2021 growing seasons in Alföld wheat variety

In Mv Kolompos, test weight was lower in 2021 (79.44 kg/hl) compared to 2020 (81.15 kg/hl) the difference was statistically significant [F = 71.975, P = 0.000] (Table 6). *Fusarium* infection had a strong negative effect on test weight [R = - 0.770], test weight decreased when the infection increased. The fitted regression model between *Fusarium* infection and test weight is y= - 0.028x + 82.304. The regression is statistically significant [R^2 = 0.592, F = 40.701, P = 0.000] (Figure 13, Table 7).

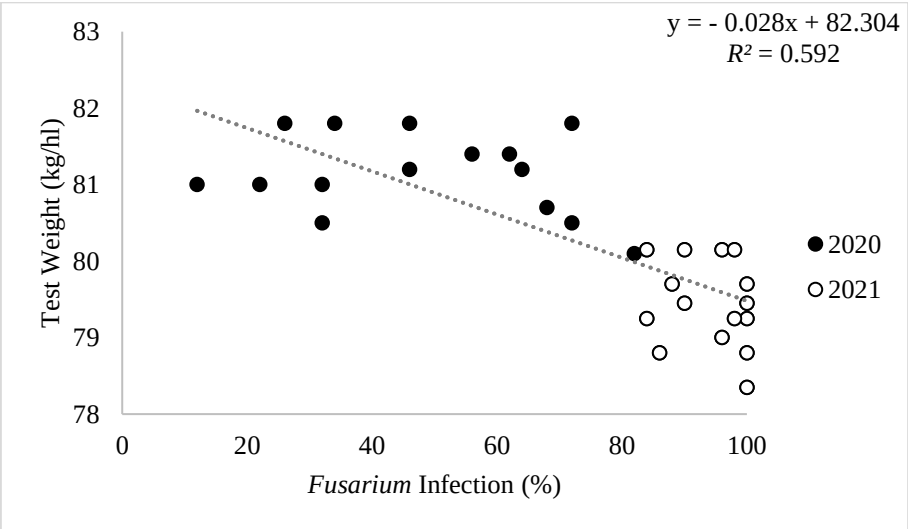

Figure 13. Influence of *Fusarium* infection (%) on test weight (kg/hl) in 2020 and 2021 growing seasons in Mv Kolompos wheat variety

In Mv Karéj, test weight was lower in 2021 (79.33 kg/hl) compared to 2020 (81.04 kg/hl) the difference was statistically significant [F = 48.936, P = 0.000] (Table 6). *Fusarium* infection had a strong negative effect on test weight [R = - 0.692], test weight decreased when the infection increased. The fitted regression model between *Fusarium* infection and test weight is y = - 0.024 + 81.802. The regression is statistically significant [R^2 = 0.479, F = 25.724, P = 0.000] (Figure 14, Table 7).

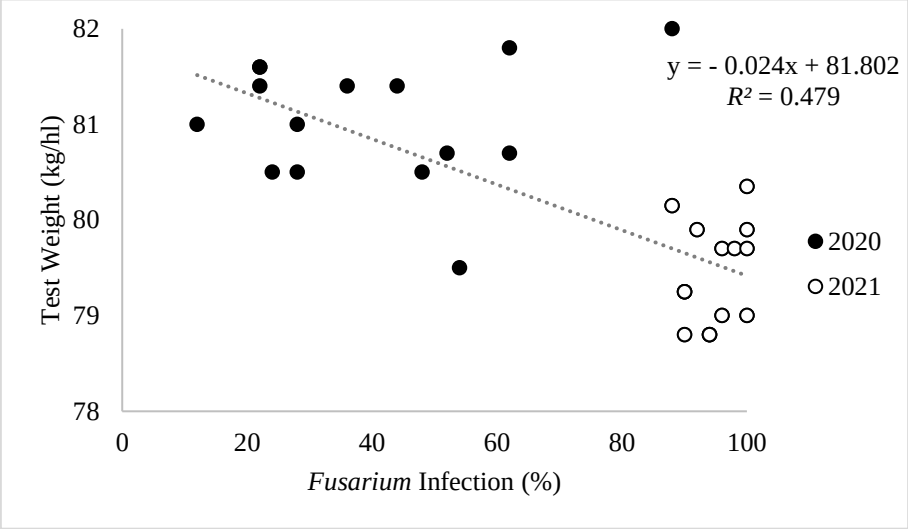

Figure 14. Influence of *Fusarium* infection (%) on test weight (kg/hl) in 2020 and 2021 growing seasons in Mv Karéj wheat variety

Table 6. Descriptive statistics and ANOVA for the influence of growing season on test weight in Alföld, Mv Kolompos and Mv Karéj wheat varieties

Descriptive statistics		Mean	Std. Deviation	Std. Error	Minimum	Maximum
Alföld	2020	75.10	1.22	0.31	72.70	77.30
	2021	72.76	1.32	0.34	70.30	75.20
	Total	73.93	1.72	0.31	70.30	77.30
Mv Kolompos	2020	81.15	0.54	0.14	80.10	81.80
	2021	79.44	0.56	0.15	78.35	80.15
	Total	80.29	1.02	0.19	78.35	81.80
Mv Karéj	2020	81.04	0.65	0.17	79.50	82.00
	2021	79.33	0.68	0.18	77.70	80.35
	Total	80.19	1.09	0.20	77.70	82.00
ANOVA		Sum of Squares	*df*	Mean Square	*F*	Sig.
Alföld	Between Groups	40.950	1	40.950	25.338	0.000
	Within Groups	45.252	28	1.616		
	Total	86.202	29			
Mv Kolompos	Between Groups	21.845	1	21.845	71.975	0.000
	Within Groups	8.498	28	0.304		
	Total	30.344	29			
Mv Karéj	Between Groups	21.845	1	21.845	48.936	0.000
	Within Groups	12.499	28	0.446		
	Total	34.345	29			

Table 7. Model summary, ANOVA and coefficients for the influence of *Fusarium* infection on test weight in Alföld, Mv Kolompos and Mv Karéj wheat varieties

Model summary		*R*	R^2	Adjusted R^2	Std. Error of the Estimate	
Alföld		0.626	0.391	0.370	1.369	
Mv Kolompos		0.770	0.592	0.578	0.665	
Mv Karéj		0.692	0.479	0.460	0.800	
ANOVA		Sum of Squares	*df*	Mean Square	*F*	Sig.
Alföld	Regression	33.737	1	33.737	18.005	0.000
	Residual	52.466	28	1.874		
	Total	86.202	29			
Mv Kolompos	Regression	17.977	1	17.977	40.701	0.000
	Residual	12.367	28	0.442		
	Total	30.344	29			
Mv Karéj	Regression	16.445	1	16.445	25.724	0.000
	Residual	17.900	28	0.639		
	Total	34.345	29			
Coefficients		Unstandardized Coefficients		Standardized Coefficients	*t*	Sig.
		B	Std. Error	Beta		
Alföld	*Fusarium*	-0.041	0.010	-0.626	-4.243	0.000
	(Constant)	76.714	0.702		109.332	0.000
Mv Kolompos	*Fusarium*	-0.028	0.004	-0.770	-6.380	0.000
	(Constant)	82.304	0.338		243.703	0.000
Mv Karéj	*Fusarium*	-0.024	0.005	-0.692	-5.072	0.000
	(Constant)	81.802	0.350		233.474	0.000

4.4 Influence of *Fusarium* infection on thousand kernel weight

In Alföld, thousand kernel weight was lower in 2021 (39.65 g) compared to 2020 (45.91 g) the difference was statistically significant [F = 96.249, P = 0.000] (Table 8). *Fusarium* infection had a strong negative effect on thousand kernel weight [R = - 0.765], thousand kernel weight decreased when the infection increased. The fitted regression model between *Fusarium* infection and thousand kernel weight is y = - 0.105x + 49.920. The regression is statistically significant [R^2 = 0.585, F = 39.441, P = 0.000] (Figure 15, Table 9).

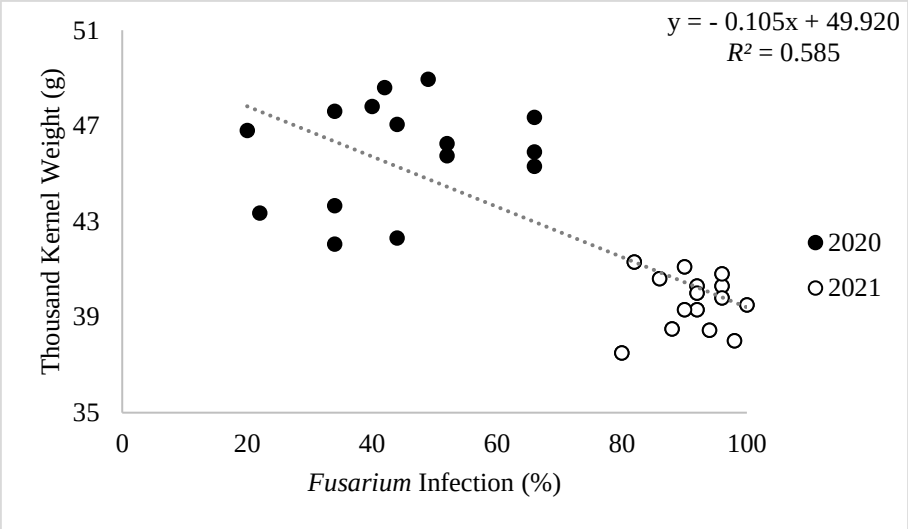

Figure 15. Influence of *Fusarium* infection (%) on thousand kernel weight (g) in 2020 and 2021 growing seasons in Alföld wheat variety

In Mv Kolompos, thousand kernel weight was lower in 2021 (38.68 g) compared to 2020 (42.48 g) the difference was statistically significant [F = 26.306, P = 0.000] (Table 8). *Fusarium* infection had a moderate negative effect on thousand kernel weight [R = - 0.516], thousand kernel weight decreased when the infection increased. The fitted regression model between *Fusarium* infection and thousand kernel weight is y = - 0.051x + 44.230. The regression is statistically significant [R^2 = 0.266, F = 10.155, P = 0.004] (Figure 16, Table 9).

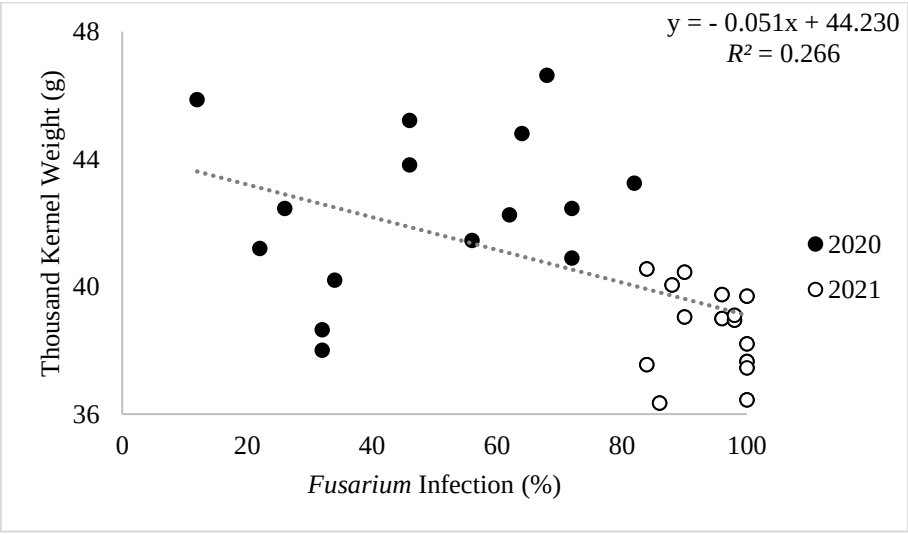

Figure 16. Influence of *Fusarium* infection (%) on thousand kernel weight (g) in 2020 and 2021 growing seasons in Mv Kolompos wheat variety

In Mv Karéj, there was no statistically significant difference in thousand kernel weight in 2020 and 2021 [F = 3.743, P = 0.063] (Table 8). *Fusarium* infection had a moderate negative effect on thousand kernel weight [R = - 0.454], thousand kernel weight decreased when the infection increased. The fitted regression model between *Fusarium* infection and thousand kernel weight is y = - 0.031x + 44.483. The regression is statistically significant [R^2 = 0.206, F = 7.264, P = 0.012] (Figure 17, Table 9).

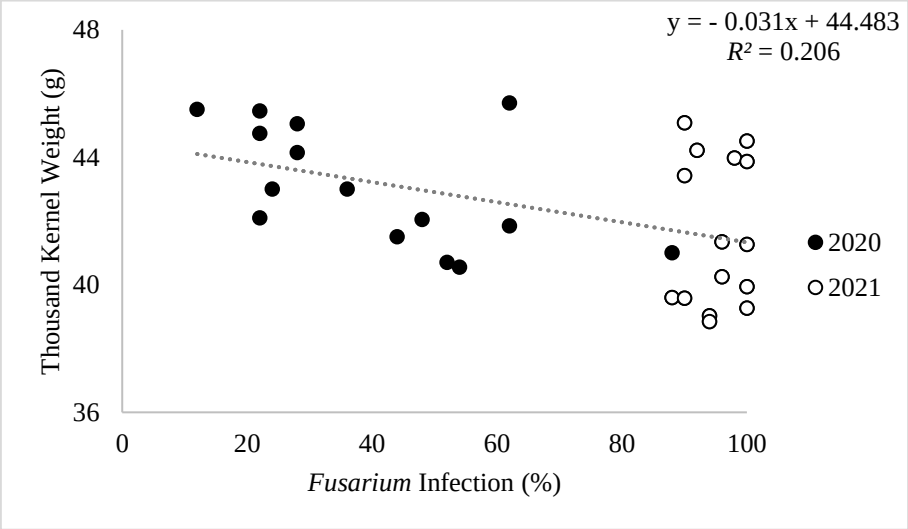

Figure 17. Influence of *Fusarium* infection (%) on thousand kernel weight (g) in 2020 and 2021 growing seasons in Mv Karéj wheat variety

Table 8. Descriptive statistics and ANOVA for the influence of growing season on thousand kernel weight in Alföld, Mv Kolompos and Mv Karéj wheat varieties

Descriptive statistics		Mean	Std. Deviation	Std. Error	Minimum	Maximum
Alföld	2020	45.91	2.19	0.57	42.05	48.95
	2021	39.65	1.15	0.30	37.50	41.30
	Total	42.78	3.62	0.66	37.50	48.95
Mv Kolompos	2020	42.48	2.52	0.65	38	46.63
	2021	38.68	1.35	0.35	36.35	40.55
	Total	40.58	2.77	0.51	36.35	46.63
Mv Karéj	2020	43.09	1.86	0.48	40.55	45.70
	2021	41.61	2.30	0.59	38.84	45.08
	Total	42.35	2.19	0.40	38.84	45.70
ANOVA		Sum of Squares	*df*	Mean Square	*F*	Sig.
Alföld	Between Groups	294.220	1	294.220	96.249	0.000
	Within Groups	85.592	28	3.057		
	Total	379.812	29			
Mv Kolompos	Between Groups	107.844	1	107.844	26.306	0.000
	Within Groups	114.790	28	4.100		
	Total	222.634	29			
Mv Karéj	Between Groups	16.398	1	16.398	3.743	0.063
	Within Groups	122.656	28	4.381		
	Total	139.054	29			

Table 9. Model summary, ANOVA and coefficients for the influence of *Fusarium* infection on thousand kernel weight in Alföld, Mv Kolompos and Mv Karéj wheat varieties

Model summary		*R*	R^2	Adjusted R^2	Std. Error of the Estimate	
Alföld		0.765	0.585	0.570	2.373	
Mv Kolompos		0.516	0.266	0.240	2.416	
Mv Karéj		0.454	0.206	0.178	1.986	
ANOVA		Sum of Squares	*df*	Mean Square	*F*	Sig.
Alföld	Regression	222.123	1	222.123	39.441	0.000
	Residual	157.689	28	5.632		
	Total	379.812	29			
Mv Kolompos	Regression	59.254	1	59.254	10.155	0.004
	Residual	163.380	28	5.835		
	Total	222.634	29			
Mv Karéj	Regression	28.644	1	28.644	7.264	0.012
	Residual	110.411	28	3.943		
	Total	139.054	29			
Coefficients		Unstandardized Coefficients		Standardized Coefficients	*t*	Sig.
		B	Std. Error	Beta		
Alföld	*Fusarium*	-0.105	0.017	-0.765	-6.280	0.000
	(Constant)	49.920	1.216		41.038	0.000
Mv Kolompos	*Fusarium*	-0.051	0.016	-0.516	-3.187	0.004
	(Constant)	44.230	1.228		36.032	0.000
Mv Karéj	*Fusarium*	-0.031	0.012	-0.454	-2.695	0.012
	(Constant)	44.483	0.870		51.119	0.000

4.5 Influence of *Fusarium* infection on falling number

In Alföld, there was no statistically significant difference in falling number in 2020 and 2021 [$F = 0.449$, $P = 0.508$] (Table 10). *Fusarium* infection had no effect on falling number [$R = -0.142$]. The fitted regression model between *Fusarium* infection and falling number is y = - 0.238x + 441.002. The regression is not statistically significant [$R^2 = 0.020$, $F = 0.580$, $P = 0.453$] (Figure 18, Table 11).

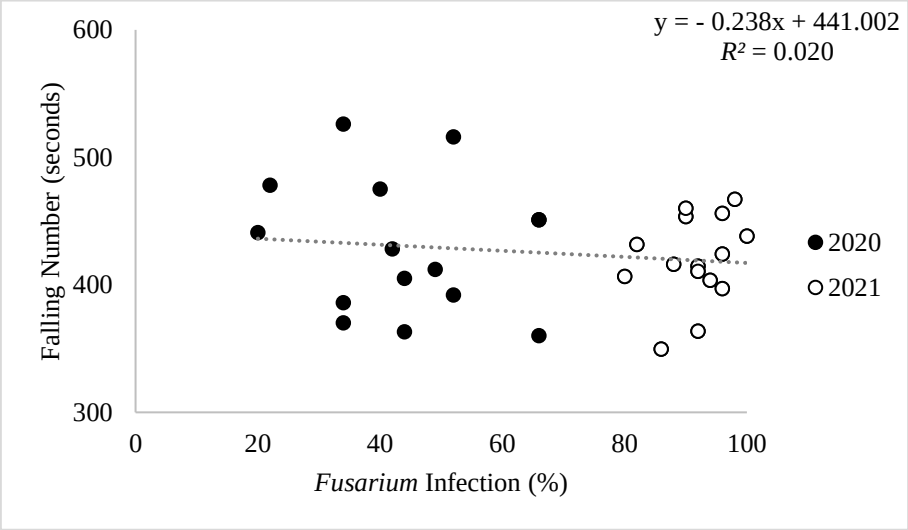

Figure 18. Influence of *Fusarium* infection (%) on falling number (seconds) in 2020 and 2021 growing seasons in Alföld wheat variety

In Mv Kolompos, falling number was lower in 2021 (358.87 seconds) compared to 2020 (544.93 seconds) the difference was statistically significant [$F = 101.038$, $P = 0.000$] (Table 10). *Fusarium* infection had a strong negative effect on falling number [$R = -0.758$], falling number decreased when the infection increased. The fitted regression model between *Fusarium* infection and falling number is y = - 2.908x + 658.924. The regression is statistically significant [$R^2 = 0.575$, $F = 37.831$, $P = 0.000$] (Figure 19, Table 11).

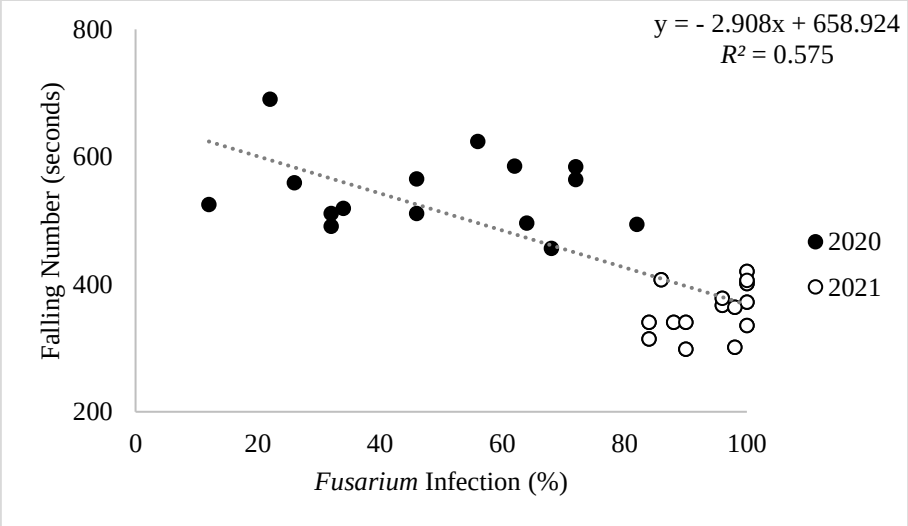

Figure 19. Influence of *Fusarium* infection (%) on falling number (seconds) in 2020 and 2021 growing seasons in Mv Kolompos wheat variety

In Mv Karéj, falling number was lower in 2021 (364.30 seconds) compared to 2020 (415.67 seconds) the difference was statistically significant [F = 16.984, P = 0.000] (Table 10). *Fusarium* infection had a moderate negative effect on falling number [R = - 0.428], falling number decreased when the infection increased. The fitted regression model between *Fusarium* infection and falling number is y = - 0.578x + 429.057. The regression is statistically significant [R^2 = 0.183, F = 6.285, P = 0.018] (Figure 20, Table 11).

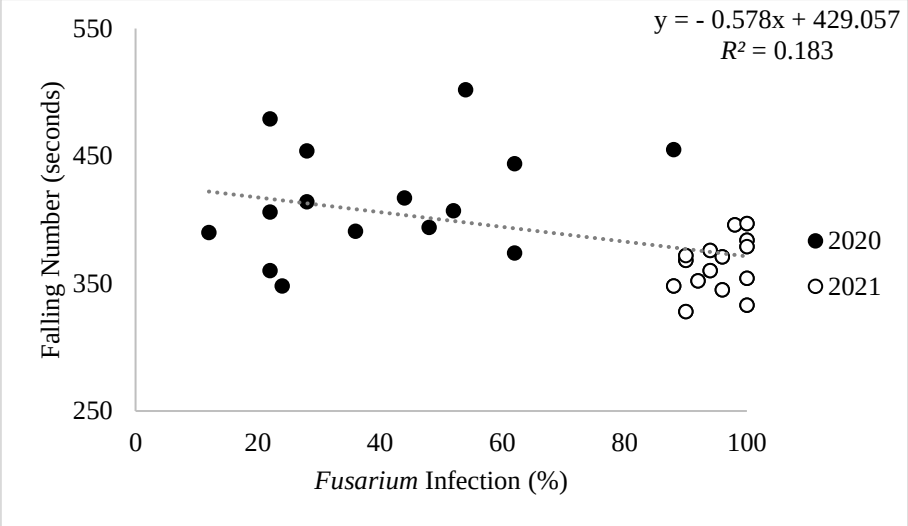

Figure 20. Influence of *Fusarium* infection (%) on falling number (seconds) in 2020 and 2021 growing seasons in Mv Karéj wheat variety

Table 10. Descriptive statistics and ANOVA for the influence of growing season on falling number in Alföld, Mv Kolompos and Mv Karéj wheat varieties

Descriptive statistics		Mean	Std. Deviation	Std. Error	Minimum	Maximum
Alföld	2020	430.27	52.96	13.67	360	526
	2021	419.40	33.78	8.72	349.50	467
	Total	424.83	43.99	8.03	349.50	526
Mv Kolompos	2020	544.93	60.10	15.52	456	690
	2021	358.87	39.08	10.09	298	420
	Total	451.90	106.93	19.52	298	690
Mv Karéj	2020	415.67	43.59	11.26	348	502
	2021	364.20	20.95	5.41	328	397
	Total	389.93	42.60	7.78	328	502
ANOVA		Sum of Squares	*df*	Mean Square	*F*	Sig.
Alföld	Between Groups	885.633	1	885.633	0.449	0.508
	Within Groups	55241.533	28	1972.912		
	Total	56127.167	29			
Mv Kolompos	Between Groups	259656.033	1	259656.033	101.038	0.000
	Within Groups	71956.667	28	2569.881		
	Total	331612.700	29			
Mv Karéj	Between Groups	19866.133	1	19866.133	16.984	0.000
	Within Groups	32751.733	28	1169.705		
	Total	52617.867	29			

Table 11. Model summary, ANOVA and coefficients for the influence of *Fusarium* infection on falling number in Alföld, Mv Kolompos and Mv Karéj wheat varieties

Model summary		*R*	R^2	Adjusted R^2	Std. Error of the Estimate	
Alföld		0.142	0.020	-0.015	44.315	
Mv Kolompos		0.758	0.575	0.559	70.974	
Mv Karéj		0.428	0.183	0.154	39.176	
ANOVA		Sum of Squares	*df*	Mean Square	*F*	Sig.
Alföld	Regression	1139.541	1	1139.541	0.580	0.453
	Residual	54987.626	28	1963.844		
	Total	56127.167	29			
Mv Kolompos	Regression	190567.506	1	190567.506	37.831	0.000
	Residual	141045.194	28	5037.328		
	Total	331612.700	29			
Mv Karéj	Regression	9645.185	1	9645.185	6.285	0.018
	Residual	42972.682	28	1534.739		
	Total	52617.867	29			
Coefficients		Unstandardized Coefficients		Standardized Coefficients	*t*	Sig.
		B	Std. Error	Beta		
Alföld	*Fusarium*	-0.238	0.313	-0.142	-0.762	0.453
	(Constant)	441.002	22.715		19.414	0.000
Mv Kolompos	*Fusarium*	-2.908	0.473	-0.758	-6.151	0.000
	(Constant)	658.924	36.067		18.270	0.000
Mv Karéj	*Fusarium*	-0.578	0.230	-0.428	-2.507	0.018
	(Constant)	429.057	17.167		24.993	0.000

4.6 Influence of *Fusarium* infection on gluten content

In Alföld, gluten content was lower in 2021 (24.79 %) compared to 2020 (30 %) the difference was statistically significant [F = 29.351, P = 0.000] (Table 12). *Fusarium* infection had a strong negative effect on gluten content [R = - 0.716], gluten content decreased when the infection increased. The fitted regression model between *Fusarium* infection and gluten content is y = - 0.101x + 34.234. The regression is statistically significant [R^2 = 0.512, F = 29.383, P = 0.000] (Figure 21, Table 13).

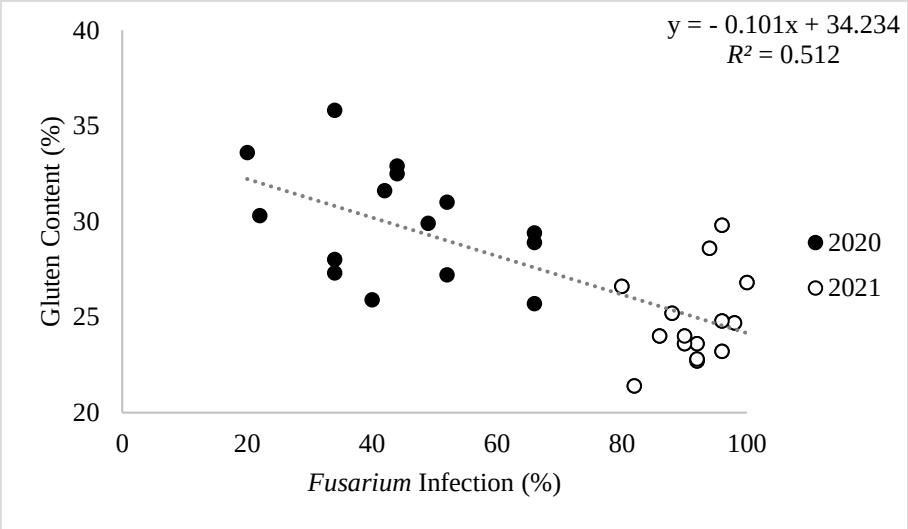

Figure 21. Influence of *Fusarium* infection (%) on gluten content (%) in 2020 and 2021 growing seasons in Alföld wheat variety

In Mv Kolompos, gluten content was lower in 2021 (29.07 %) compared 2020 (31.25 %) the difference was statistically significant [F = 8.235, P = 0.008] (Table 12). *Fusarium* infection had a moderate negative effect on gluten content [R = - 0.432], gluten content decreased when the infection increased.The fitted regression model between *Fusarium* infection and gluten content is y = - 0.036x + 32.727. The regression is statistically significant [R^2 = 0.186, F = 6.414, P = 0.017] (Figure 22, Table 13).

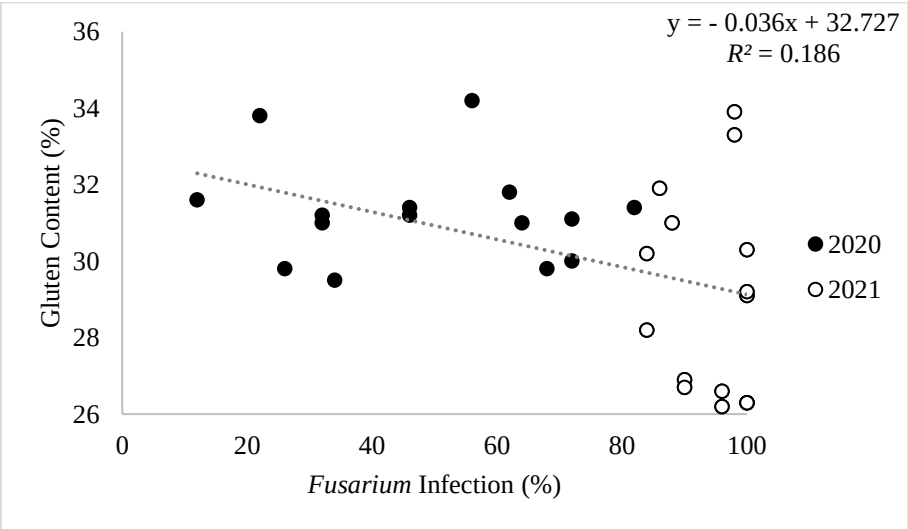

Figure 22. Influence of *Fusarium* infection (%) on gluten content (%) in 2020 and 2021 growing seasons in Mv Kolompos wheat variety

In Mv Karéj, there was no statistically significant difference in gluten content in 2020 and 2021 [F = 0.557, P = 0.462] (Table 12). *Fusarium* infection had no effect on gluten content [R = - 0.009] The fitted regression model between *Fusarium* infection and gluten content is y = - 0.001x + 28.944. The regression is not statistically significant [R^2 = 0.000, F = 0.002, P = 0.962] (Figure 23, Table 13).

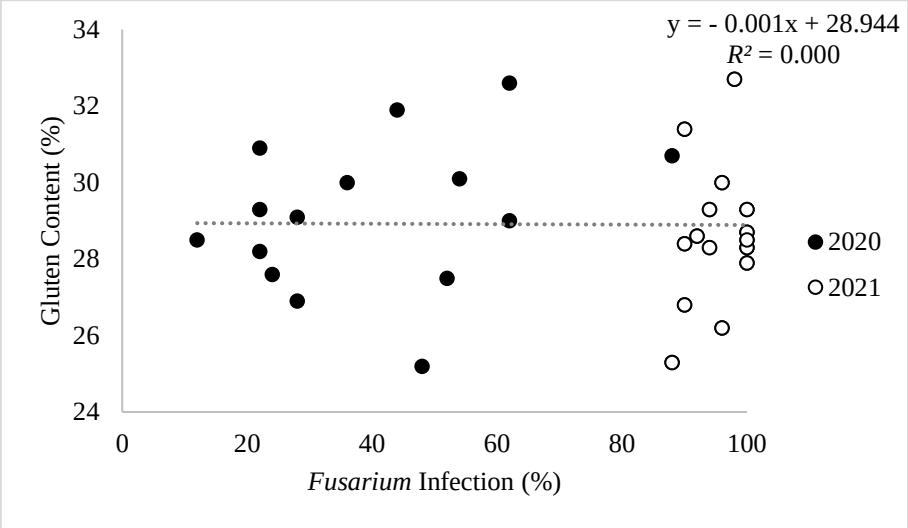

Figure 23. Influence of *Fusarium* infection (%) on gluten content (%) in 2020 and 2021 growing seasons in Mv Karéj wheat variety

Table 12. Descriptive statistics and ANOVA for the influence of growing season on gluten content in Alföld, Mv Kolompos and Mv Karéj wheat varieties

Descriptive statistics		Mean	Std. Deviation	Std. Error	Minimum	Maximum
Alföld	2020	30.00	2.94	0.76	25.70	35.80
	2021	24.79	2.29	0.59	21.40	29.80
	Total	27.39	3.71	0.68	21.40	35.80
Mv Kolompos	2020	31.25	1.33	0.34	29.50	34.20
	2021	29.07	2.63	0.68	26.20	33.90
	Total	30.16	2.33	0.42	26.20	34.20
Mv Karéj	2020	29.17	1.96	0.51	25.20	32.60
	2021	28.65	1.86	0.48	25.30	32.70
	Total	28.91	1.89	0.35	25.20	32.70
ANOVA		Sum of Squares	*df*	Mean Square	*F*	Sig.
Alföld	Between Groups	203.841	1	203.841	29.351	0.000
	Within Groups	194.457	28	6.945		
	Total	398.299	29			
Mv Kolompos	Between Groups	35.643	1	35.643	8.235	0.008
	Within Groups	121.187	28	4.328		
	Total	156.830	29			
Mv Karéj	Between Groups	2.028	1	2.028	0.557	0.462
	Within Groups	101.891	28	3.639		
	Total	103.919	29			

Table 13. Model summary, ANOVA and coefficients for the influence of *Fusarium* infection on gluten content in Alföld, Mv Kolompos and Mv Karéj wheat varieties

Model summary		*R*	R^2	Adjusted R^2	Std. Error of the Estimate	
Alföld		0.716	0.512	0.495	2.635	
Mv Kolompos		0.432	0.186	0.157	2.135	
Mv Karéj		0.009	0.000	-0.036	1.926	
ANOVA		Sum of Squares	*df*	Mean Square	*F*	Sig.
Alföld	Regression	203.948	1	203.948	29.383	0.000
	Residual	194.351	28	6.941		
	Total	398.299	29			
Mv Kolompos	Regression	29.228	1	29.228	6.414	0.017
	Residual	127.602	28	4.557		
	Total	156.830	29			
Mv Karéj	Regression	0.009	1	0.009	0.002	0.962
	Residual	103.910	28	3.711		
	Total	103.919	29			
Coefficients		Unstandardized Coefficients B	Standardized Coefficients Std. Error	*t* Beta	Sig.	
Alföld	*Fusarium*	-0.101	0.019	-0.716	-5.421	0.000
	(Constant)	34.234	1.350		25.350	0.000
Mv Kolompos	*Fusarium*	-0.036	0.014	-0.432	-2.533	0.017
	(Constant)	32.727	1.085		30.168	0.000
Mv Karéj	*Fusarium*	-0.001	0.011	-0.009	-0.049	0.962
	(Constant)	28.944	0.844		34.287	0.000

4.7 Influence of *Fusarium* infection on Zeleny sedimentation index

In Alföld, Zeleny sedimentation index was lower in 2021 (38.40 ml) compared to 2020 (53.5 ml) the difference was statistically significant [F = 52.412, P = 0.000] (Table 14). *Fusarium* infection had a strong negative effect on Zeleny sedimentation index [R = - 0.747], Zeleny sedimentation index decreased when the infection increased. The fitted regression model between *Fusarium* infection and Zeleny sedimentation index is y = - 0.270x + 64.266. The regression is statistically significant [R^2 = 0.557, F = 35.257, P = 0.000] (Figure 24, Table 15).

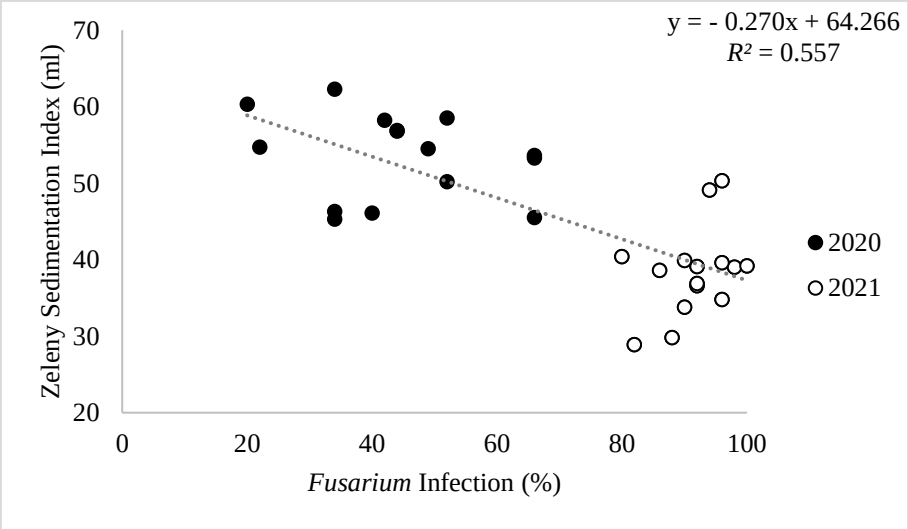

Figure 24. Influence of *Fusarium* infection (%) on Zeleny sedimentation index (ml) in 2020 and 2021 growing seasons in Alföld wheat variety

In Mv Kolompos, Zeleny sedimentation index was lower in 2021 (42.37 ml) compared to 2020 (63.09 ml) the difference was statistically significant [F = 67.705, P = 0.000] (Table 14). *Fusarium* infection had a strong negative effect on Zeleny sedimentation index [R = - 0.678], Zeleny sedimentation index decreased when the infection increased. The fitted regression model between *Fusarium* infection and Zeleny sedimentation index is y = - 0.305x + 74.443. The regression is statistically significant [R^2 = 0.460, F = 23.879, P = 0.000] (Figure 25, Table 15).

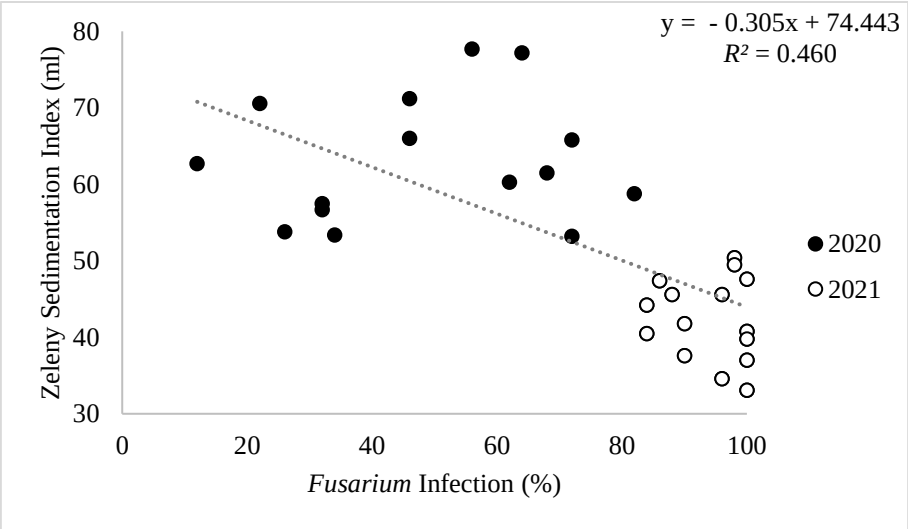

Figure 25. Influence of *Fusarium* infection (%) on Zeleny sedimentation index (ml) in 2020 and 2021 growing seasons in Mv Kolompos wheat variety

In Mv Karéj, Zeleny sedimentation index was lower in 2021 (49.47 ml) compared to 2020 (60.57 ml) the difference was statistically significant [F = 17.748, P = 0.000] (Table 14). *Fusarium* infection had a strong negative effect on Zeleny sedimentation index [R = - 0.613], Zeleny sedimentation index decreased when the infection increased. The fitted regression model between *Fusarium* infection and Zeleny sedimentation index is y = - 0.176x + 66.938. The regression is statistically significant [R^2 = 0.375, F = 16.823, P = 0.000] (Figure 26, Table 15).

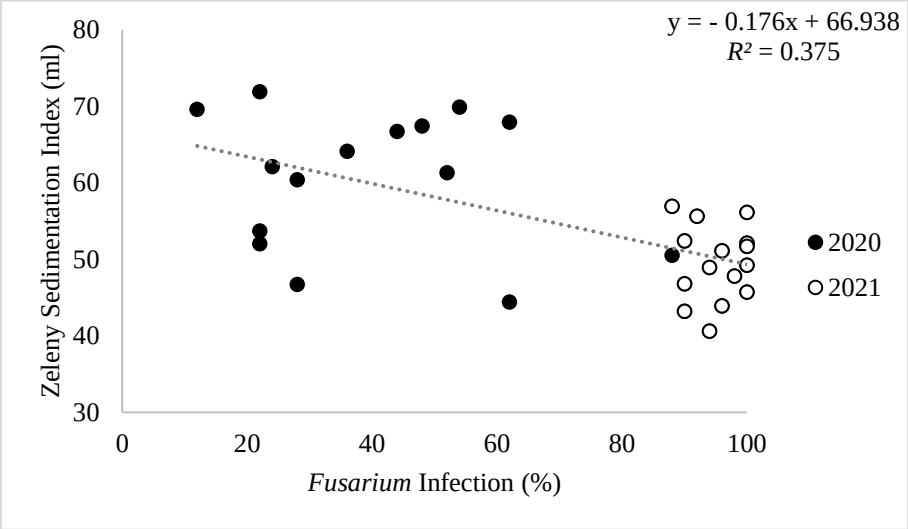

Figure 26. Influence of *Fusarium* infection (%) on Zeleny sedimentation index (ml) in 2020 and 2021 growing seasons in Mv Karéj wheat variety

Table 14. Descriptive statistics and ANOVA for the influence of growing season on Zeleny sedimentation index in Alföld, Mv Kolompos and Mv Karéj wheat varieties

Descriptive statistics		Mean	Std. Deviation	Std. Error	Minimum	Maximum
Alföld	2020	53.50	5.63	1.45	45.30	62.30
	2021	38.40	5.79	1.49	28.90	50.30
	Total	45.95	9.51	1.74	28.90	62.30
Mv Kolompos	2020	63.09	8.15	2.10	53.20	77.70
	2021	42.37	5.37	1.39	33.10	50.40
	Total	52.73	12.53	2.29	33.10	77.70
Mv Karéj	2020	60.57	8.97	2.32	44.40	71.90
	2021	49.47	4.88	1.26	40.60	56.90
	Total	55.02	9.07	1.66	40.60	71.90
ANOVA		Sum of Squares	*df*	Mean Square	*F*	Sig.
Alföld	Between Groups	1710.075	1	1710.075	52.412	0.000
	Within Groups	913.580	28	32.628		
	Total	2623.655	29			
Mv Kolompos	Between Groups	3221.960	1	3221.960	67.705	0.000
	Within Groups	1332.463	28	47.588		
	Total	4554.423	29			
Mv Karéj	Between Groups	925.185	1	925.185	17.748	0.000
	Within Groups	1459.583	28	52.128		
	Total	2384.768	29			

Table 15. Model summary, ANOVA and coefficients for the influence of *Fusarium* infection on Zeleny sedimentation index in Alföld, Mv Kolompos and Mv Karéj wheat varieties

Model summary		*R*	R^2	Adjusted R^2	Std. Error of the Estimate	
Alföld		0.747	0.557	0.542	6.440	
Mv Kolompos		0.678	0.460	0.441	9.370	
Mv Karéj		0.613	0.375	0.353	7.294	
ANOVA		Sum of Squares	*df*	Mean Square	*F*	Sig.
Alföld	Regression	1462.319	1	1462.319	35.257	0.000
	Residual	1161.336	28	41.476		
	Total	2623.655	29			
Mv Kolompos	Regression	2096.331	1	2096.331	23.879	0.000
	Residual	2458.092	28	87.789		
	Total	4554.423	29			
Mv Karéj	Regression	895.056	1	895.056	16.823	0.000
	Residual	1489.712	28	53.204		
	Total	2384.768	29			
Coefficients		Unstandardized Coefficients		Standardized Coefficients	*t*	Sig.
		B	Std. Error	Beta		
Alföld	*Fusarium*	-0.270	0.045	-0.747	-5.938	0.000
	(Constant)	64.266	3.301		19.468	0.000
Mv Kolompos	*Fusarium*	-0.305	0.062	-0.678	-4.887	0.000
	(Constant)	74.443	4.761		15.635	0.000
Mv Karéj	*Fusarium*	-0.176	0.043	-0.613	-4.102	0.000
	(Constant)	66.938	3.196		20.942	0.000

Two major reasons might be the cause for biochemical changes in grain composition and therefore subsequent changes in wheat quality traits: fungal enzymes and impaired synthesis of grain components.

Fungal enzymes, especially carbohydrases and proteases, are assumed to cause the major changes in flour composition and processing quality (Dexter and Nowicki, 2003). An increased activity of amylase, xylanase, cellulase, and glucanase in flours from *Fusarium* infected grain could partly explain weaker and stickier dough properties, lower water uptake and poorer baking quality of these flours in comparison to less severely infected flours (Dexter et al., 1996; Pawelzik et al., 1998; Nightingale et al., 1999; Matthaus et al., 2004; Wang et al., 2005a). It is assumed that the fungus secretes these enzymes during the invasion of the kernel thus degrading starch as well as cell wall components. Wang et al. (2005a) characterized alpha-amylase of *F. culmorum* and demonstrated that it was active in a wide pH range from 5.0 to 8.5 and at temperatures from 10 to 100 °C, enabling the enzymes to have an adverse effect on dough and baking properties, particularly in baking procedures involving longer proofing times. Even more important might be the activity of fungal proteases. According to Dexter and Nowicki (2003), proteases of fungal origin are the most reasonable explanation for poorer dough and baking qualities of *Fusarium* infected wheat in comparison to healthy wheat. Pekkarinen et al. (2000) showed that *F. graminearum*, *F. culmorum* and *F. poea* were able to produce acid, neutral or alkaline proteases

when grown on gluten media. Several studies showed that protease activity in severely *Fusarium* infected wheat samples was higher than in a control or less infected samples (Pawelzik et al., 1998; Matthaus et al., 2004; Wang et al., 2005b). The authors suggested that the digestion of storage proteins during processing could lead to change in dough properties such as loss of loaf shape during baking.

Beside enzymatic degradation of starch, storage proteins and other flour components, another reason for changed flour, dough and baking properties after *Fusarium* infection might be the incomplete accumulation of kernel constituents. One reason for this could be the mechanical blocking of vascular bundles by fungal mycelium.

Fungal mycelium growth within xylem and phloem may inhibit the nutrient supply for developing spikelets and leading to their premature death (Kang and Buchenauer, 2000; Ribichich et al,. 2000; Goswami and Kistler, 2004). The interruption of assimilate transport within the spike impairs the normal development of kernels and leads to the typically shrunken and shriveled kernels of *Fusarium* damaged grain (Meyer et al., 1986) and the reduction of thousand kernel weight and therefore also test weight (Simmonds, 1968; Bechtel et al.,1985).

The present study was carried out to determine the effect of *Fusarium* infection on wheat quality in 2020 and 2021 growing seasons. The different climatic conditions that prevailed during 2020 and 2021 growing seasons could be the reason for the increase in *Fusarium* infection which leads to the decrease in wheat quality. According to El Chami et al. (2022) climatic factors (especially during flowering period) play a key role in determining fungal occurrence. Thus, the activity of the fungi and their level of colonization are much determined by environmental conditions. The likelihood of *Fusarium* infection increases when favorable environmental conditions are present, ultimately resulting in a decrease in both yield and quality of wheat..In our study the increase in *Fusarium* infection showed a negative impact on wheat quality. Antes et al. (2001) and Prange et al. (2005) found that a strong *Fusarium* infection did not significantly influence wheat quality parameters. On the contrary, Gärtner et al. (2008) and Seitz et al. (1986) observed in their studies that *Fusarium* infection has negative effects on wheat quality parameters.

The results obtained in our study showed that *Fusarium* infection decreases protein content in Alföld and Mv Kolompos which is observed by Bechtel et al. (1985), Nightingale et al. (1999), Prange et al. (2005) and Gartner et al. (2008). However, in Mv Karéj *Fusarium* infection did not have an effect on protein content which is supported by the findings of other studies (Seitz et al,. 1986; Dexter et al.,1996; Prange et al., 2005; Wang et al., 2005b; Terzi et al., 2007). Other studies found an increase of protein content after severe *Fusarium* infection (Meyer et al., 1986; Boyacioglu and hettiarachchy, 1995; Pawelzik et al., 1998; Matthaus et al., 2004; Siuda et al., 2010).

The results obtained in our study showed that *Fusarium* infection decreases gluten content in Alföld and Mv Kolompos. Dexter et al. (1997) and Gärtner et al. (2008) agrees with the observations of other studies (Meyer et al., 1986; Boyacioglu and hettiarachchy, 1995; Pawelzik et al., 1998) who found a slight decrease in gluten content in wheat kernels after *Fusarium* infection. However, in Mv Karéj gluten content was not affected by *Fusarium* infection. Wang et al. (2005b) concluded that gluten content in the wheat grain was not affected by

Fusarium infection. However, Boyacioğlu and Hettiarachchy (1995) concluded that gluten content in wheat kernels increased following their contamination with *Fusarium* species.

The results obtained in our study revealed that *Fusarium* infection decreases falling number in Mv Kolompos and Mv Karéj . Fungal infection of ears increases degradation of starch due to the presence of enzymes, such as α-amylase, the activity of which is measured using falling number (Wang et al., 2008). A reduction of falling number after infection with *Fusarium* fungi could, therefore, be expected and has been confirmed (Dexter et al., 1996; Siuda et al., 2010). According to Hareland (2003), *Fusarium* infection increases the degradation of starch in wheat kernels due to the presence of enzymes such as α-amylase which in turn decreases the quality of wheat flour and results in lower falling number values. Howerver, in Alföld falling number was not affected by *Fusarium* infection which was observed by Gärtner et al. (2008), whereby falling number remained unchanged by the infection.

The results obtained in our study revealed that *Fusarium* infection, in the three wheat varieties used, decreases Zeleny sedimentation index. Papousková et al. (2011) observed that Zeleny sedimentation index showed distinctively decreased values in the infected samples. Meyer et al. (1986) and Gärtner et al. (2008) observed general reduction of Zeleny sedimentation index in wheat grains after *Fusarium* infection. On the other hand, Kreuzberger et al. (2015) did not observe a change in sedimentation index.

The results obtained indicated that test weight and thousand kernel weight, in the three wheat varieties used, were significantly decreased by *Fusarium* infection. Test weight is primarily a function of the size of the grains. The fact that *Fusarium* infection affects test weight has been well documented (McMullen et al., 2012; Wong et al., 1995). Spanic et al. (2017) findings suggest that *Fusarium* infection has a negative effect on test weight. Dexter et al. (1996), Wang et al. (2005b) and Dvojkovic et al. (2007) found that *Fusarium* infection resulted in a decrease of thousand kernel weight. *Fusarium* infected kernels are damaged, shriveled, shrunken and light weight showing a tendency towards a decrease in the endosperm to bran ratio due to fungal carbohydrate consumption. Results from the mentioned studies indicate that *Fusarium* infection may alter and lead to the deterioration of wheat quality parameters.

5. CONCLUSION AND RECOMMENDATIONS

- FHB has a substantial negative influence on the technological quality of wheat, impacting protein content, gluten content, Zeleny sedimentation index, falling number, test weight, and thousand kernel weight.
- FHB infection leads to a reduction in protein content. This decrease in protein levels can have implications for the overall quality of wheat-based products.
- Gluten content, a critical aspect of wheat quality, is negatively impacted by FHB. The reduction in gluten content can lead to poor dough strength, reduced volume, and undesirable texture in baked goods.
- Zeleny sedimentation index, is typically lower in FHB infected wheat. A lower index indicates poorer flour quality and potential baking challenges.
- FHB infected wheat grains often exhibit a decrease in falling number. A low falling number indicates starch degradation, affecting flour functionality and leading to poor bread quality.
- Test weight, a measure of grain weight per unit volume, is significantly reduced in FHB affected wheat due to the presence of lightweight and shriveled kernels. This reduction in test weight has implications for grain quality and market value.
- Thousand kernel weight, which reflects seed size and weight, is also negatively affected by FHB. Infected kernels tend to be smaller and lighter, resulting in a decrease in thousand kernel weight. This reduction can impact overall yield potential, grain quality and economic value.
- FHB can have significant economic impacts on wheat quality, as well as implications for the agricultural industry. FHB infected wheat kernels often exhibit quality issues, including discoloration, reduced test weight and damaged kernels. These factors can result in downgrading of the grain, limiting its marketability and potentially leading to lower prices for affected farmers. Grain quality issues can also disrupt export opportunities, affecting the overall competitiveness of the agricultural industry.
- Addressing the economic impact of FHB on wheat quality requires a comprehensive approach. This includes the development and adoption of resistant wheat varieties, improved disease management practices, effective monitoring and forecasting systems and educational programs to promote best management practices among farmers. Collaborative efforts between farmers, researchers, policymakers, and the industry are essential to minimize the economic losses in the face of FHB outbreaks.

6. NEW SCIENTIFIC RESULTS

- The effect of *Fusarium* infection on wheat quality varies between the three different wheat varieties used (Alföld, Mv Kolompos and Mv Karéj) as they show different response patterns against *Fusarium* head blight.

- In Alföld, *Fusarium* infection had a negative effect on protein content, gluten content, test weight, thousand kernel weight and Zeleny sedimentation index, whereas falling number was not affected.

- In Mv Kolompos, *Fusarium* infection had a negative effect on all wheat quality parameters used protein content, gluten content, falling number, test weight, thousand kernel weight and Zeleny sedimentation index.

- In Mv Karéj, *Fusarium* infection had a negative effect on test weight, thousand kernel weight, falling number and Zeleny sedimentation index, whereas protein content and gluten content were not affected.

- Although *Fusarium* infection reduced wheat quality, Mv Karéj showed a stable protein and gluten content whereas Alföld showed a stable falling number. Thus, Mv Karéj is the most tolerant to *Fusarium* infection, followed by Alföld and then Mv Kolompos being the least tolerant.

7. SUMMARY

Wheat is a cereal of special importance in the world cereal production. During crop production, both abiotic and biotic stresses occur, often acting in combination under field conditions and potentially increase sensitivity to pathogens. *Fusarium* head blight (FHB) is one of the most devastating fungal diseases of wheat and other small grain cereals and has caused serious epidemics worldwide. The fungal pathogen associated with this disease in wheat is *Fusarium* spp. During the wheat's flowering stage, *Fusarium* infection occurs when weather conditions become favorable. Occurrence of FHB poses a serious problem because of considerable economic losses caused by lowered yield and deteriorated grain quality. Moreover, during the invasion of the kernel, *Fusarium* spp. secretes enzymes such as carbohydrases and proteases that degrade the cell wall and the kernel components. As a result, FHB infection leads to poor end use quality. Beside enzymatic degradation of starch, storage proteins and other flour components, another reason for changed flour, dough and baking properties after *Fusarium* infection might be the incomplete accumulation of kernel constituents. One reason for this could be the mechanical blocking of vascular bundles by fungal mycelium. Results showed that *Fusarium* head blight has a substantial negative influence on the technological quality of wheat:

- FHB infection leads to a reduction in protein content. This decrease in protein levels can have implications for the overall quality of wheat-based products.
- Gluten content, a critical aspect of wheat quality, is negatively impacted by FHB. The reduction in gluten content can lead to poor dough strength, reduced volume, and undesirable texture in baked goods.
- Zeleny sedimentation index, is typically lower in FHB infected wheat. A lower index indicates poorer flour quality and potential baking challenges.
- FHB infected wheat grains often exhibit a decrease in falling number. A low falling number indicates starch degradation, affecting flour functionality and leading to poor bread quality.
- Test weight, a measure of grain weight per unit volume, is significantly reduced in FHB affected wheat due to the presence of lightweight and shriveled kernels. This reduction in test weight has implications for grain quality and market value.
- Thousand kernel weight, which reflects seed size and weight, is also negatively affected by FHB. Infected kernels tend to be smaller and lighter, resulting in a decrease in thousand kernel weight. This reduction can impact overall yield potential, grain quality and economic value.

8. SCIENTIFIC PUBLICATIONS

El Chami, J., El Chami, E., Tarnawa, Á., Kassai, K.M., Kende, Z., Jolánkai, M., (2023). Influence of *Fusarium* head blight on technological quality of wheat. *Acta Phytopathologica et Entomologica Hungarica*. https://doi.org/10.1556/038.2023.00179

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