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Full-text article

SALIVARY GLAND ALPHA-AMYLASES RETAIN ACTIVITY IN WHEAT GRAINS DAMAGED BY *EURYGASTER* (HETEROPTERA: SCUTELLERIDAE) SUNN BUGS

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The Sunn pest *Eurygaster integriceps* and related wheat bugs (Sunn bugs) cause great damage to the wheat crop by injecting hydrolytic enzymes into the grain that liquefy the endosperm during extraintestinal digestion. The main harmful factor of bugs is considered to be their proteases, which are retained in the endosperm of damaged grain and flour and hydrolyze gluten proteins during dough kneading. Thus, the quality of bread deteriorates sharply. Salivary gland proteases provide protein nutrition for the wheat bugs. However, the main source of energy that allows them to accumulate fat necessary for wintering is the endosperm starch, hydrolyzed by alpha-amylases. Alpha-amylases of wheat bug salivary glands have not been sufficiently studied. There is still no consensus on the role of these enzymes in the nutrition of bugs, on their presence in damaged mature grain, and on their influence on the technological qualities of flour, which may be partly due to the lack of methods for detecting the actual bug alpha-amylases in grain. Using a new simple method, including inactivation of plant beta-amylases interfering with the analysis by sodium p-chloromercuribenzoate, we obtained alpha-amylase banding patterns similar to the patterns of wheat bug salivary gland alpha-amylases as a result of protein isoelectric focusing of some samples of damaged grain. The developed approach can be recommended for use in the technological assessment of damaged grain, in studying the mechanisms of plant immunity to pests as well as for diagnostics of grain damage by insects.

Keywords: Sunn pest, digestive enzymes, grain quality, isoelectric focusing, damage diagnostics

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Introduction

The Sunn pest *Eurygaster integriceps* Put. and related wheat bugs (or Sunn bugs) of the genus *Eurygaster* Lap. cause great damage to the wheat crop by injecting hydrolytic enzymes into the grain that liquefy the endosperm during extraintestinal digestion (Mehrabadi et al., 2014; Pavlyushin et al. 2015; Dizlek, 2018; Konarev, 2020; Kapustkina, Frolov, 2022; Ouaarous et al., 2025). The main factor causing harmfulness of wheat bugs is considered to be proteases remaining in the endosperm after incomplete ingestion of hydrolysis products by insects. Proteases remaining in damaged grains and flour prepared from them hydrolyze gluten proteins during dough kneading, which is why the technological qualities of flour and bread baked from it deteriorate sharply (Sivri et al., 2004; Kamenchenko et al. 2010; Salis et al., 2010; Dizlek, Özer, 2016; Konarev et al., 2019). Salivary gland proteases provide protein nutrition for bugs by converting insoluble under normal conditions grain proteins into a liquid state available for ingestion. However, the main source of energy that makes it possible to accumulate the fat necessary for bugs to overwinter is endosperm starch (Vilkova, 1980; Ghanbari et al 2022).

The main enzyme that hydrolyzes starch to maltose and dextrins, including in insects, is α-amylase (Da Lage, 2018). The important role of α-amylases for insect vital activity makes them a promising target for modern molecular approaches to plant protection (Wang et al., 2023). On the other hand, amylolytic activity is an essential element of the dough maturation process during kneading. The level of this activity can be affected by various factors, including premature germination of grains or insect damage, which can alter the level of monosaccharides involved in fermentation processes (Dizlek and Özer, 2024).

In wheat bugs, salivary gland α-amylases have been studied to a much lesser extent than proteases, and the information about them is quite contradictory. There is still no consensus in the scientific literature on the role of salivary gland α-amylases in the nutrition of bugs, as well as on their influence on the baking and other technological qualities of flour. A number of authors believe that the salivary glands of wheat bugs produce α-amylases, which convert starch granules into a state suitable for ingestion by the insect. The process of starch digestion is completed in the intestine under the action of another set of α-amylases and carbohydrases (Vilkova 1980; Mehrabadi et al., 2009, 2014; Ravan et al., 2009; Konarev, 1996, 2020). Meanwhile some researchers find no direct relationship between wheat bug damage and the level of amylolytic activity in grain, although they confirm the destruction of starch granules in the endosperm (Lorenz and Meredith, 1988; Every et al., 1990), others confirm such a relationship (Dizlek, Özer, 2024), and still others generally deny the involvement of α-amylases of the salivary glands in the nutrition of these insects (Rosell et al., 2002). Many authors do not find α-amylases at all in the salivary glands of various representatives of the suborder Heteroptera that feed on plant seeds (unlike predatory bugs), and see the role of the secretion

of these glands only in moistening food before ingestion. They associate the assimilation of seed starch by bugs only with intestinal enzymes (Silva, Terra, 1994; Woodring et al., 2007; Rocha et al., 2014; Da Lage, 2018). Such different opinions may be related to varietal peculiarities of protective mechanisms of neutralization of foreign enzyme activity in the grain, to the phase of grain development upon damage, as to the specifics of the assay methods used.

Typically, total amylase activity in grain is determined by standard methods of technological assessment or biochemical methods (Cornaggia et al., 2016; Dizlek and Özer, 2024), which, even when using substrates specific for, for example, α-amylase, do not provide information on the origin of the enzyme being analyzed. The total amylase activity in mature intact grain is usually determined by the presence of β-amylases (Ziegler, 1999), but it can be increased by endogenous or exogenous α-amylases. As a result, it may remain unknown whether changes in amylase activity are related to insect damage or, for example, to the appearance of endogenous α-amylase during premature grain germination.

A simple approach to identifying (attributing) the enzyme would be be to compare the electrophoretic or isoelectric spectra of α-amylase components (banding patterns) from salivary glands and bug-damaged grains, as was previously done for these insects' proteases (Konarev et al., 2017). Amylolytic enzymes are usually easily detected by electrophoresis or isoelectric focusing (IEF) of proteins, followed by incubation of the gel with starch and iodine staining (Thomson et al., 1990; Ravan et al., 2009; Konarev, Lovegrove, 2012; Netsvetaev et al., 2014). Detecting insect α-amylases in grain in this way is complicated by the presence of highly active endogenous β-amylases in the extract. In the case of endogenous α-amylases of cereal grain, the problem is solved by inactivating less heat-stable β-amylases by heating at 70–71 °C (Konarev, 1982; Doehlert, Duke, 1983; Evans et al., 2003), however, insect α-amylases, including those of the *Eurygaster* bugs, are also inactivated under these conditions (Ravan et al., 2009; Mehrabadi et al., 2014). It is also known that grain β-amylases are inactivated by mercury salts, e.g., $HgCl_2$, whereas the effect of these salts on α-amylases depends strongly on their origin and remains poorly studied (Doehlert, Duke, 1983). We found no information in the scientific literature on the identification of α-amylases of wheat bugs in the electrophoretic spectra of proteins of damaged wheat grains.

To determine whether active bug α-amylases are present in damaged grain, as is the case for proteases, we developed a simple method involving the selective inactivation of grain β-amylases that interfere with analysis using sodium p-chloromercuribenzoate (pCMB).

Materials and Methods

The study material consisted of grain samples from several soft and durum wheat cultivars (cv.) that were damaged by the *E. integriceps* and other *Eurygaster* bug species. The samples were obtained from different regions of Russia in 2016–2022: Krasnodar Krai (*Triticum aestivum* L. cv. Grom, Bezostaya 100, Svarog, Elanchik), Saratov Oblast (T. aestivum cv. Dzhangal) and Altai Krai (*T. aestivum* cv. Altayskaya 70, Altayskaya 75, Tobolskaya stepnaya, Altayskaya zhnitsa, Union; *T. durum* Desf. cv. Salyut Altaya, Oasis, Pamyati Yanchenko), as well as from Turkey in 2006 (*T. aestivum* cv. Ikizce and Bayraktar; *T. durum* cv. Ege-88). Grain samples from Turkey were kindly provided by Professors D. Sivri Ozay and H. Koksel (Hacettepe University, Ankara)

Grain material from Turkey was previously analyzed with respect to Sunn pest proteases (Konarev et al., 2013). According to the methodology adopted by the VIZR (Kapustkina and Frolov, 2022; Kapustkina, 2024), severely damaged individual grains with damage level – 3–4 points out of 5 were selected. At least ten individual damaged grains or flour from ten grains were analyzed for each sample. Grains without visible signs of damage were used as a controls. Grains were ground in a mortar. Then, proteins were extracted from the flour using a 2-fold (v/w) volume of 0.05 M Tris-HCl buffer at pH 8.5 containing 0.1 % Triton X-100 for 30 minutes. After centrifugation, the extracts were analyzed immediately or placed in a freezer after an equal volume of 80 % glycerol and 1 % CHAPS detergent was added. Detergents and glycerol preserved enzyme activity in the extracts at -24 °C for an extended period. α-amylases of the Sunn pest *E. integriceps* (Krasnodar Krai, 2015) were extracted from salivary glands stored at -80 °C by homogenization them with 0.01 % Triton X-100 with 0.5 % CHAPS (Konarev et al., 2019) followed by centrifugation.

To investigate the potential for selective inhibition of β-amylase activity and to determine the optimal conditions for analysis, 0.1 M pCMB dissolved in 0.1 M NaOH and diluted 10 times with water was added to the extract, resulting in a final concentration of 1–10 mM. Samples with high amylase activity were diluted 20-fold with the extractant; however, to simplify the comparison of results the volumes applied to the gel in terms of undiluted extract are indicated in the experiment results description.

The proteins were fractionated by IEF using IEF PhastGel gels with a pH range of 3–9 and 4–6.5 on a PhastSystem device (Pharmacia, Sweden). Instead of using the branded applicator untreated and pCMB-treated protein extracts were applied to 5x1 or 10x1 mm filter paper strips (LKB, Sweden), placed on the gel near the anode (20 pieces per gel) in a volume of 0.2–3 µl. IEF was performed at voltage 1000 V, a power 2.5 W and temperature 15 °C for 180 Vh (for pH 3–9 gels) or 230 Vh (for pH 4–6.5 gels). Protein separation was monitored using colored protein markers of isoelectric points (pI) cytochrome c (10.6) and horse myoglobin (7.3).

Amylases were detected in protein spectra using a new version of the substrate replica. To prepare the replica, 45 mg of soluble potato starch was boiled in 30 ml of water, 200 mg of agarose (SERVA, Germany, gel point 40 °C) was added to the solution which was boiled again. Then, the hot agarose solution was poured onto a GelBond film for agarose (SERVA, 250 x 125 mm), which was placed on the horizontal thermostated plate of the Multiphor II IEF device (LKB, Sweden), heated to 50 °C. The plate temperature was lowered to 5 °C for the gel to form. Then, the temperature was increased again to 50 °C and the gel was dried with a fan. Before use a fragment of the replica film was cut to the size of the area of interest in the separating gel (30 x 42 mm). It was immersed in 0.1 M Na acetate buffer (pH 5.4) containing

0.1 M NaCl for 20 seconds, overlaid on the separating gel, and covered with plastic film to protect it from drying. A weight was placed on top to ensure tight contact between the substrate replica and the gel. The entire structure was transferred to a thermostated plate and kept at 37 °C for an hour. Then, the replica was immersed in an iodine solution containing 30% alcohol. Light bands on a blue background corresponded to the components of the protein spectra with amylase activity. Dried replicas preserved the visible banding patterns for a long time. An alternative analysis method that does not require separate treatment of the samples with pCMB consisted of preliminary soaking the replica in a 0.1 M solution of this reagent diluted 20-fold in a Na-acetate buffer at pH 5.4.

Results

Loading 2 µl of protein extracts from bug-damaged (d) and intact (u) grains from different wheat samples onto a separating gel, followed by fractionation and of amylase detection, resulted in heavily overloaded lanes with barely distinguishable individual components. As an example, the Figure 1 (lanes 1 and 2) shows the results for a durum wheat sample (cv. Ege-88) from Turkey.

A slightly higher degree of "brightening" can be noted in lane 1d, corresponding to the extract from the damaged grain, compared to lane 2u in the zone of isopoints (pI) near those of the main *E. integriceps* salivary gland α-amylase components (lanes 5, 10 and 15). Loading significantly smaller volumes of the damaged grain extract (lanes 8 and 9, 0.1 and 0.02 µl) revealed several more distinct components. Some individual components with amylase activity in lane 8 were close to the salivary gland α-amylase components (lane 10) although this banding pattern was not clear. When even smaller amounts of the extract were applied (lane 9), distinct components with relatively low pI became discernible, apparently corresponding to the main components of β-amylases, while the bands with higher pI disappeared. A similar pattern was observed when small amounts of extract were applied to analyze other grain samples. For example, damaged and undamaged cv. Grom grains from the Krasnodar region exhibited this pattern (lanes 3d and 4u).

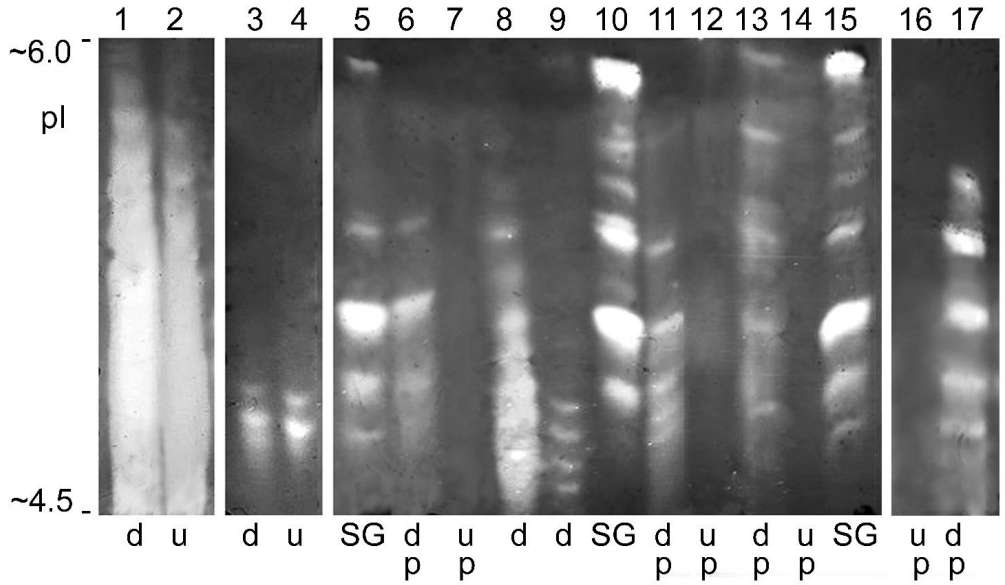

Figure 1. Detection of α-amylases from *Eurygaster* Lap. Sunn bugs in wheat grains damaged by them**.** Proteins were separated by isoelectric focusing (IEF) on a PhastGel with a pH range of 4–6.5. Amylases were detected using an agarose replica containing starch. d and u – extracts from damaged and undamaged grains, respectively; SG – salivary glands of *E. integriceps* Put.; p – extracts with added sodium p-chloromercuribenzoate (pCMB). Given isoelectric point (pI) values are approximate.
Lanes 1 and 2, 8 and 9, 11 and 12: *Triticum durum* Desf. culyivar (cv.) Ege-88. Lanes 1 and 2: 1 µL extract each; lanes 8 and 9: 0.1 µL and 0.03 µL; lanes 11 and 12: 2 µL each. Lanes 3, 4, 6, 7, 13, 14: *T. aestivum* L. Lanes 3 and 4: cv. Grom, 0.05 µL each; lanes 6 and 7: cv. Grom, 2.5 µL each; lanes 13 and 14: cv. Bayraktar, 2 µL each. Lanes 16 and 17: *T. durum* cv. Salyut Altaya, 1 µL each. Proteins were extracted from flour from 10 damaged and 10 undamaged grains (Ege-88, cv. Grom, and Bayraktar samples) or from individual grains (cv. Salyut Altaya)

Рисунок 1. Выявление α-амилаз хлебных клопов рода *Eurygaster* Lap. в поврежденных ими зернах пшеницы. Белки разделены методом ИЭФ в геле PhastGel с интервалом pH 4–6.5. Амилазы выявлены с помощью агарозной реплики, содержащей крахмал. d и u – экстракты из поврежденных и неповрежденных зерен соответственно, SG – слюнные железы *E. integriceps* Put., p – экстракты с добавлением пара-хлормеркурибензоата Na (ПХМБ). Величины изоточек (pI) указаны ориентировочно.
Дорожки 1 и 2, 8 и 9, 11 и 12 – *Triticum durum* Desf., сорт Ege-88, 1 и 2 по 1 мкл экстракта, 8 и 9 – 0.1 и 0.03 мкл, 11 и 12 – по 2 мкл. 3, 4, 6, 7, 13, 14, 16 и 17 – *T. aestivum* L., 3, 4, 6 и 7 сорт Гром, 3 и 4 – по 0.05 мкл, 6 и 7 – по 2.5 мкл; 13 и 14 – сорт Bayraktar – по 2 мкл; 16 и 17 – сорт Салют Алтая – по 1 мкл.
Белки экстрагировали из муки из 10 поврежденных и 10 неповрежденных зерновок (сорта Ege-88, Гром, и Bayraktar) или из отдельных зерновок (Салют Алтая)

Excess grain β-amylases obviously prevent the acquisition of clear IEF spectra of bug α-amylases. The addition of a pCMB solution to proteins extracted from bug-damaged grains taken from various wheat samples including the cv. Grom (lane 6dp), Ege-88 (11dp), and Bayraktar (13dp) resulted in the appearance of distinct components with amylase activity that corresponded to the pI of the *E. integriceps* salivary gland α-amylases. Conversely, no amylase-active components were detected in preparations from intact pCMB-treated grains (lanes 7up, 12up, 14up and 16up).

Apparently, the effect of "cleaning" the spectra of α-amylases of insects was due to the inactivation of β-amylases of grain under the action of pCMB. Components of α-amylases corresponding or clozse in pI to the components of Sunn pest salivary gland α-amylases were found in some bug-damaged grains of samples from Krasnodar Krai (cv. Grom), Saratov Oblast (cv. Dzhangal), Altai Krai (cv. Altai 70, Altai 75, Salyut Altaya and Tobolskaya Stepnaya), as well as from Turkey (cv. Ege-88 and Bayraktar). Undamaged grains did not contain such amylase components.

It is noteworthy that not all damaged grains from the listed samples yielded discernible α-amylase spectra. When analyzing by 10 damaged grains of the listed above samples, bug amylases were detected in one or more of grains or in a sample of flour from 10 grains. Furthermore, in a number of samples, all analyzed grains exhibiting clear signs of severe bug damage contained no α-amylases in quantities sufficient for detection under the experimental conditions employed. This applies to the samples of cv. Bezostaya 100, Svarog, Elanchik (Krasnodar Krai), Oazis, Pamyati Yanchenko, YunIon (Altai Krai), and the sample of the cv. Ikizce from Turkey.

Comparison of the efficiency of different pCMB doses showed that the optimal pCMB concentration in the sample is 2 mM. Lower concentrations do not ensure complete inhibition of grain β-amylases, while concentrations above 6 mM may adversely affect the activity of insect α-amylases.

The alternative version of the method for detecting bug α-amylases, which involves treating the starch replica itself with a pCMB solution, showed somewhat lower effectiveness in inactivating β-amylases.

Discussion

The study results showed that wheat grains damaged by wheat bugs in various regions of Russia (Krasnodar Krai, Saratov Oblast, Altai Krai) and in Turkey can contain active α-amylases with isozyme patterns similar to the α-amylases from the salivary glands of *E. integriceps* originating from Krasnodar Krai. It should be noted that in the European part of Russia, wheat is damaged mainly by *E. integriceps*, in the Altai Krai – by *E. maura* L. (Neymorovets, 2019; Kapustkina, 2024), while in Turkey it is believed that both of the mentioned species are harmful (Konarev et al., 2013; Mutlu et al., 2014), as well as *E. austriaca* Schrk. (Kınacı, Kınacı, 2004). Since the species of bugs that damaged the grain samples from Turkey analyzed in this work, unlike the samples from Russia, were not clearly identified, we limited ourselves to designating only their belonging to the genus *Eurygaster*. Researchers from Turkey often refer to these wheat bug species under the common name Sunn pests (Kınacı, Kınacı, 2004). This approach to species designation is supported by the results of our studies on the proteases of wheat grain damaged by wheat bugs. These results indicate some differences in the component composition of these enzymes between samples of Russian and Turkish origin, which may reflect both intra- and interspecific differences in the pests (Konarev et al. 2013, Konarev, Kapustkina, 2024).

As in the case of proteases (Konarev et al. 2013), even within a single sample, not all grains with distinct signs of severe damage by wheat bugs contained detectable amounts of salivary gland α-amylases. In a number of samples, α-amylases were absent in all damaged grains analyzed. It can be assumed that the level of activity of salivary gland hydrolytic enzymes remaining in the damaged grain may depend on the developmental stage of the grain at the time of damage, grain maturation conditions, the completeness of the insect's ingestion of the liquefied endosperm contents, and cultivar-specific features of the mechanisms for neutralizing the action of foreign enzymes in the grain. The seeds of various plants, including cereals, are known to contain protein inhibitors of insect α-amylases (Franco et al., 2002; Basso et al., 2025). It is possible that the absence of traces of α-amylases from the salivary glands of bugs in damaged grains of some samples may be somehow related to such inhibitors. α-amylase inhibitors from plant seeds are represented by a variety of forms, differing in their specificity to the α-amylases of various insect species (Konarev. 1996, 1999; Konarev and Lovegrove, 2012). Some inhibitors from the endosperm of common wheat *T. aestivum* inhibit the activity of intestinal α-amylases of the Sunn pest, but the sensitivity of these enzymes to these inhibitors is hundreds of times lower than the sensitivity of the α-amylase of the beetle *Tenebrio molitor* L. The α-amylases of the salivary glands of the bug did not show any sensitivity to them (Konarev, 1992, 1996). It was suggested that low sensitivity to wheat inhibitors or its absence is a result of the coevolution of species of the genus *Eurygaster* with the host plant (Konarev, 1996). *E. integriceps* is a highly specialized pest of wheat, and *T. molitor* is omnivorous. In turn, Mehrabadi et al. (2010, 2012) found that a protein inhibitor from the grain of a wheat-rye hybrid, *Triticale* (TAI), inhibits the activity of both intestinal α-amylases and salivary glands. Interestingly, rye is a much less preferred plant by *Eurygaster* bugs than wheat (Amiri et al., 2010), which may be partly due to its inhibitors. It has been shown that the salivary glands of the *Eurygaster* bug most intensively synthesize digestive enzymes, in particular proteases, which are capable of hydrolyzing the most readily available food component (Konarev et al., 2011), and this synthesis is highly dependent on the host plant (Amiri et al., 2016). These data suggest that the absence of bug α-amylases in some samples of damaged grain may be due to their reduced synthesis by the salivary glands in response to the presence of certain factors in the grain that impede starch digestion, such as inhibitors that make such synthesis energetically unjustifiable. At the same time, bug proteases may be present in such samples of damaged grain. In different samples with the same level of damage according to visual assessment, the component composition and activity of bug proteases may differ significantly (Konarev et al., 2013).

It was found that the α-amylases of wheat bugs retain activity in damaged grains for many years – they were detected in samples from harvests 10–20 years old (e.g.,

samples of the cv. Grom and Ege-88). Bug proteases also remain active in damaged grains for equally long periods (Konarev et al., 2020). Other examples of the preservation of insect α-amylase activity in cereal grains and various biological materials of plant origin, including food products, are also known. Traces of active α-amylases from the granary weevil *Sitophilus granarius* (L.) were detected in barley grains fed on by adults of this pest (Piasecka-Kwiatkowska et al., 2014). α-Amylases are a component of the salivary gland secretion of *S. granarius* (Baker et al., 1984). Bee α-amylase was isolated from honey (Babacan and Rand, 2007). The long-term preservation of activity of non-ingested residues of insect α-amylases in cereal grains is possibly associated with the formation of their complexes with water-insoluble proteins and endosperm starch granules, followed by drying. This may provide stabilization of the enzyme molecules. The formation of such complexes has been demonstrated for endogenous α-amylases (Yu et al., 2018).

Conclusion

Using a novel method that enables the selective inactivation of interfering endogenous β-amylases, α-amylases similar to those found in the salivary glands of the *Eurygaster integriceps* bug (Sunn pest) were identified in wheat grains damaged by bugs of the genus *Eurygaster*. This finding demonstrates that not only proteases but also the α-amylases of these pests can persist for extended periods within damaged grain. The activity of these enzymes theoretically may manifest during the use of flour produced from damaged grain in dough preparation or other food technologies, potentially impacting the quality of the final product what requires more detailed study in the future. Some interpretations of results we obtained are still hypothetical in nature and also deserve separate experimental confirmation or refutation. The proposed approach for detecting bug α-amylases in insect-damaged cereal grains can be recommended for technological assessment of damaged grain, studying plant immunity mechanisms against pests, and diagnosing insect damage to grain. Refinement of the method, including its alternative version, will enhance its sensitivity, resolution and effectiveness thereby expanding opportunities for further research into insect-plant interactions.

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Полнотекстовая статья

АЛЬФА-АМИЛАЗЫ СЛЮННЫХ ЖЕЛЕЗ ХЛЕБНЫХ КЛОПОВ РОДА *EURYGASTER* СОХРАНЯЮТ АКТИВНОСТЬ В ПОВРЕЖДЕННЫХ ЗЕРНАХ ПШЕНИЦЫ

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Вредная черепашка *Eurygaster integriceps* Put. и другие виды клопов-черепашек наносят большой ущерб урожаю пшеницы за счет введения в зерно гидролитических ферментов, разжижающих эндосперм в процессе внекишечного пищеварения. Основным фактором вредоносности хлебных клопов являются протеазы, которые, сохраняясь в поврежденном зерне и муке из него, расщепляют белки клейковины при замесе теста, что приводит к резкому ухудшению качества хлеба. Протеазы слюнных желез обеспечивают белковое питание клопов, однако, основным источником энергии, позволяющим им накапливать необходимый для зимовки жир, является крахмал эндосперма, который гидролизуют альфа-амилазы. Несмотря на важную роль альфа-амилаз слюнных желез, их участие в процессе питании клопов-черепашек, присутствие в поврежденном зерне и воздействие на технологические качества муки остаются недостаточно изученными, что может быть отчасти связано с отсутствием методов выявления в зерне собственно альфа-амилаз клопов. Использование нового простого метода, включающего инактивацию эндогенных бета-амилаз п-хлормеркурибензоатом натрия, позволило при проведении изофокусировании белков некоторых образцов поврежденного зерна получить спектры альфа-амилаз, сходные со спектрами альфа-амилаз слюнных желез хлебных клопов. Разработанный подход может быть рекомендован для использования при технологической оценке поврежденного зерна, при изучении механизмов иммунитета растений к вредителям, а также для диагностики повреждения зерна насекомыми.

Ключевые слова: вредная черепашка, пищеварительные ферменты, качество зерна, изоэлектрическая фокусировка, диагностика повреждения

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