

Impact of lectin (phytohemagglutinin) isolated from beans on grain aphid *Sitobion avenae* (F.) bionomy

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Introduction

The presence of certain groups of chemical compounds in plant tissues may influence the increase of their resistance to insects. One of those are lectins (phytohemagglutinin), which are considered to belong to the so called protein markers of immune system activation. The expression of these genes may result in a breeding of plants with the genotype, resistant to insects which are their pests (SAUVION *et al.*, 2004). Toxic interaction of particular lectins in relation to many phytophagous was confirmed both in the *in vitro* research as well as in experiments on plants that are characteristic for the expression of genes which code these proteins (SAHA *et al.*, 2006; LEITE *et al.*, 2005; CARLINI & GROSSI-DE-SÁ, 2002). There is only fragmentary data concerning the impact of lectins on physiology of aphids, including cereal aphids (VASCONCELOS & OLIVEIRA, 2004). The results of research that has been carried out so far point out to the fact that particular plant agglutinins did not show the same level of toxicity in relation to different species of these insects. A number of lectins which have a high level effect of toxicity in relation to particular aphid species may be tolerated by other species (BANDYOPADHYAY *et al.*, 2001). Such a diversified level of toxicity of individual lectins mainly results from their structural differences which condition different biochemical properties of these proteins and in particular their ability to bind specific carbohydrate radicals (SHAH *et al.*, 2005).

The aim of this paper is to study the reaction of lectin isolated from beans (*Phaseolus vulgaris* L.) (PHA) on grain aphid *Sitobion avenae* (F.) bionomy.

Material and method

The laboratory experiments were carried out under control: in temperature of 25°C and 18°C (day-night), relative humidity of 65 5% ± 5% and photoperiod of 16:8 hours. The experiment was concerned with wingless females of the grain aphid which were offered artificial diets of optimal composition (check), containing essential nutrients (KIECKHEFER & DERR, 1967). Moreover, the examined diets that were prepared contained additionally soluble lectin PHA in eight concentrations: 10, 50, 250, 500, 750, 1000, 1250, 1500 µg · cm⁻³. The prepared diets were introduced between layers of Parafilm M[®], which were placed on plastic rings (diameter 35 mm, height 15 mm). Inside the rings prepared in this way, 5 specimens of *S. avenae* aphids were placed, securing their basis with bolting-cloth.

The phagostimulating test was about determining the number of feeding specimens on controlled diets and those which contained specific concentration of lectin (phytohemagglutinin) PHA for 1 and 24 hours since the beginning of the test. Moreover, for the following 8 days the number of larvae of the tested diets was recorded in order to determine the average daily fertility of *S. avenae*. Weight tests were also carried out to determine the body mass of the wingless females of grain aphid before the onset of the test and after 24 and 48 hours of feeding. Observations on the impact of the analysed density of the lectin PHA on *S. avenae* survival were carried out daily (8 days), registering the number of living wingless grain aphid.

All the tests were carried out in 10 replications for each concentration of lectin and controlled diet and their significance underwent variant analysis (ANOVA). Differences in the reactions of the studied concentrations of lectin PHA on *S. avenae* fertility were determined by Duncan's test where $p \leq 0.01$.

Results

The phagostimulating test did not show clear interdependence between the concentration of lectin in the tested diets and the number of wingless females grain aphids both after the first and the 24th hour of feeding (Fig. 1.). The highest fertility, next to specimens on checked diets was found in wingless *S. avenae* females which fed on diets containing lowest concentration of lectin PHA (Tab. 1.). The increase of concentration of lectin in a diet lowered the daily fertility of the insect in relation to insects feeding on the checked diet. The

highest reduction of fertility, about 30%, was recorded for the wingless females of grain aphid which fed on diets containing the highest tested concentration of lectin PHA (Fig. 2.).

The weight tests that were carried out showed that the increase in concentration of lectin PHA in a diet as well as lengthening of feeding time induced the lowering of body mass of wingless females. With respect to the analysed concentration of lectin PHA a 20% decrease of body weight of the studied wingless individuals was registered (Fig. 3.).

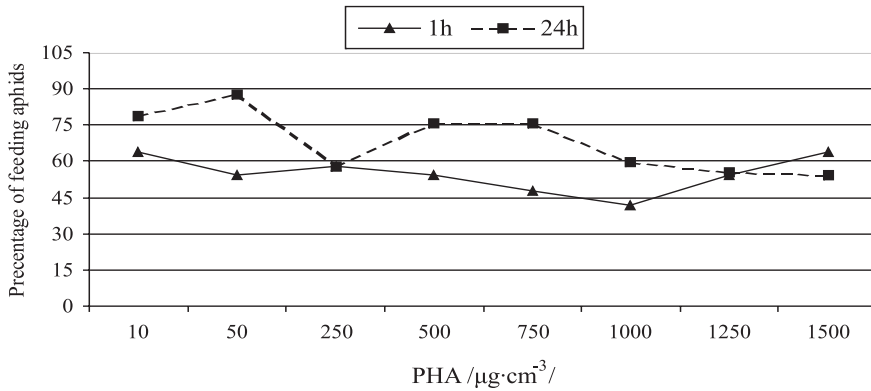


Fig. 1. Effect of PHA on the feeding of wingless females of grain aphid

Table. 1. Daily fecundity of wingless females of grain aphid feeding on diet containing different concentrations of PHA

PHA /µg · cm-3/	average daily fecundity (number of larvae / female /day)
None (check)	1.57 a
10	1.56 a
50	1.56 a
250	1.31 b
500	1.29 c
750	1.25 d
1000	1.21 e
1250	1.18 f
1500	1.10 g

Values in the column followed by various letters are significantly different at $p \leq 0.01$ (Duncan's test)

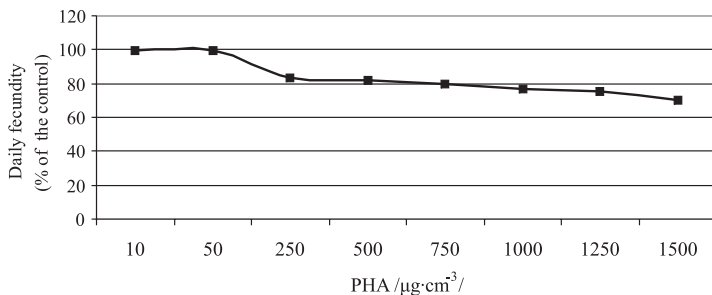


Fig. 2. Effect of PHA on daily fecundity of wingless females of grain aphid

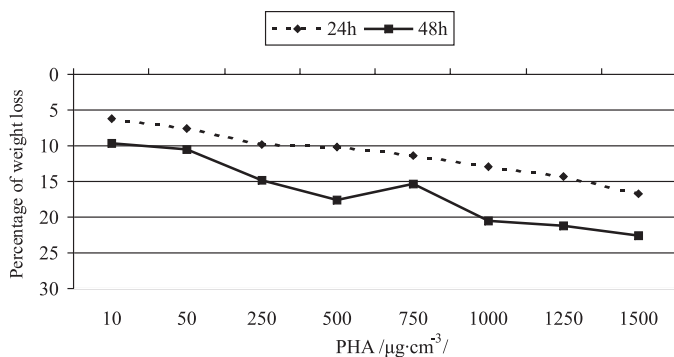


Fig. 3. Changes in weight of wingless females of grain aphid induced by tested concentrations of PHA

The laboratory tests that were carried out showed that the increase in lectin concentration induced the decrease of *S. avenae* survival after 24 hours of the test in operation. Such a tendency was observed also after 96 hours of insect feeding (Fig. 4.).

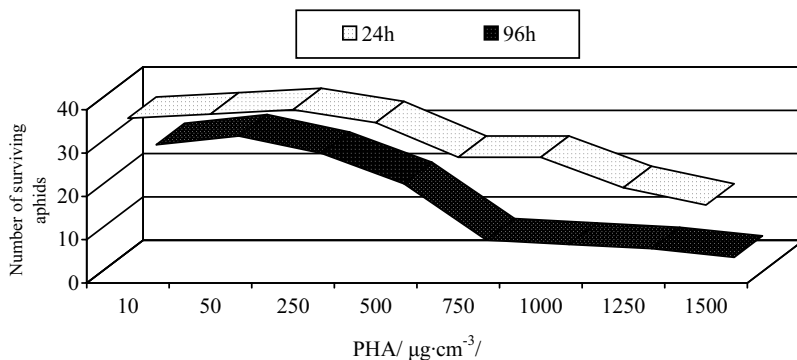


Fig. 4. The influence of PHA on the survival of wingless females of grain aphid

Discussion

The laboratory tests did not reveal any phagostimulating properties of lectin PHA in relation to wingless females grain aphids. Similar results were obtained by RAHBE & FEBVAY (1993), who showed that none of the 25 proteins which they tested contained the properties of feeding stimulator in relation to *Acyrtosiphon pisum* (H.). According to YAMASHITA *et al.* (1990) many proteins, including lectin are usually deprived of phagostimulating properties excluding some specific compounds such as polypeptide sweets or taste modifiers.

There is little data directly concerned with the effect of lectin PHA on aphid bionomy. It is limited to *A. pisum* only. RAHBE *et al.* (1995) showed that lectin PHA negatively influences the survival and mass increase of *A. pisum* in the tested concentration $/10-250\mu\text{g} \cdot \text{cm}^{-3}/$. HABIBI *et al.* (1993), who studied the impact of 15 plant lectins on the survival of *Empoasca fabae* (H.) (*Homoptera*) females found out that the lectin isolated from beans is one of the most toxic of all in relation to this insect species. It has been pointed out that it significantly increased the mortality of this pest with concentration of $200\mu\text{g} \cdot \text{cm}^{-3}/$, but on the second day of the experiment total mortality of *E. fabae* females was observed. Those females were feeding on a diet containing the highest tested concentration of lectin PHA ($1000\mu\text{g} \cdot \text{cm}^{-3}/$ and $1500\mu\text{g} \cdot \text{cm}^{-3}/$). The presented results confirm this tendency in case of *S. avenae*, because its mortality was much correlated with the concentration of lectin PHA in the diet and with the time of feeding. The toxic effect of lectin PHA in relation to *Callosobruchus maculatus* (F.) was confirmed also by SAGEDHI *et al.* (2006). It was shown that agglutinin significantly reduced female fertility (78.1%) and was one of the most toxic of all. Similar results were obtained by MACHUKA *et al.* (1999), who observed a negative impact of lectin PHA on the mortality of larvae *Maruca vitrata* (F.).

There is also little data on the impact of other lectins on grain aphid bionomy. In a research on transgenic wheat which was characteristic for the expression of lectin GNA isolated from *Galanthus nivalis* (STROGER *et al.*, 1999; SHAH *et al.*, 2005) no significant impact on grain aphid and rose-grass aphid (*Metopolophium dirhodum* Walk.) survival was registered. However, the reduction of fertility (65%) of the feeding females was recorded. A different level of toxicity of lectin PHA and GNA in relation to *S. avenae* may have been conditioned by the ability of these specific proteins to bind specific carbohydrate radicals. Lectin PHA is highly specific towards galactose and N-acetylglucosamine, whereas lectin GNA is capable of binding mannose. SHAH *et al.* 2005 claim that the toxicity of plant lectins has a strong connection with their substrate specificity towards saccharine which directs interactions with given receptors placed in the insects' digestive tract.

Conclusions

The inclusion of protein markers of immune system activation, including the plant lectins to transgenic resistance programs was preceded by detailed research which let one assume the toxicity of particular proteins in relation to a given insect species. The results of this research imply that lectin PHA, due to its negative impact on *S. avenae* bionomy, may be an important factor of resistance which inhibits the development of this insect species. Nevertheless, the fact that lectins have different mechanisms and diversified level of toxicity, their reaction in relation to particular insect species should be researched individually.

References

- BANDYOPADHYAY S., ROY A., Das S. 2001. Binding of garlic (*Allium sativum*) leaf lectin to the gut receptors of homopteran pests is correlated to its insecticidal activity. *Plant Sci.*, 161: 1025-1033.
- CARLINI C.R., GROSSI-DE-SÁ M.F. 2002. Plant toxic proteins with insecticidal properties. *Toxicon*, 40: 1515-1539.
- HABIBI J., BACKUS E.A., CZAPLA T.H. 1993. Plant lectins affect survival of the potato leafhopper (*Homoptera: Cicadellidae*). *J. Econ. Entomol.*, 86: 945-951.
- KIECKHEFER R.W., DERR R.F. 1976. Rearing three species of cereal aphids on artificial diets. *J. Econ. Entomol.*, 60: 663-665.
- LEITE Y., SILVA L., AMORIM R., FREIRE E., JORGE D., GRANGEIRO T., BENEVIDES N. 2005. Purification of the lectin from the marine red alga *Gracilaria ornata* and its effect on the development of the cowpea weevil *Callosobruchus maculatus* (*Coleoptera: Bruchidae*). *Bioch. Biophys. Acta*, 1724: 137-145.
- MACHUKA J., VAN DAMME E.J.M., PEUMANS W.J., JACKAI L.E.N. 1999. Effect on plant lectins on larval development of the legume pod borer *Maruca vitrata*. *Entomol. Exp. Appl.*, 93: 179-187.
- RAHBE Y., FEBVAY G. 1993. Protein toxicity to aphids-an in vitro test on *Acyrtosiphon pisum*. *Entomol. Exp. Appl.*, 67: 149-160.
- RAHBE Y., SAUVION N., FEBVAY G., PEUMANS W. J. 1995. Toxicity of lectins and processing of ingested proteins in the pea aphid *Acyrtosiphon pisum*. *Entomol. Exp. Appl.*, 76: 143-155.
- SADEGHI A., VAN DAMME E.J.M., PEUMANS W.J., SMAGGHE G. 2006. Deterrent activity of plant lectins on cowpea weevil *Callosobruchus masculatus* (F.) oviposition. *Phytochemistry*, 67: 2078-2084.
- SAHA P., MAJUMDER P., DUTTA I., RAY T., ROY S.C., DAS S. 2006. Transgenic rice expressing *Allium sativum* leaf lectin with enhanced resistance against sap-sucking insect pests. *Planta*, 223: 1329-1343.
- SAUVION N., NARDON CH., FEBVAY G., GATEHOUSE A.M.R., RAHBE Y. 2004. Binding of the insecticidal lectin Concanavalin A in the pea aphid *Acyrtosiphon pisum*

- (Harris) and induced effects on the structure of midgut epithelial cells. *J. Insect Physiol.*, 50: 1137-1150.
- SHAH P.A., GATEHOUSE A.M.R., CLARK S.J., PELL J.K. 2005. Wheat containing snowdrop lectin (GNA) does not affect infection of the cereal aphids *Metopolophium dirhodum* by the fungal enemy *Pandora neoaphidis*. *Transgenic Res.*, 14: 473-476.
- STROGER E., WILLIAMS S., CHRISTOU P., DOWN R.E., GATEHOUSE J.A. 1999. Expression of the insecticidal lectin from snowdrop (*Galanthus nivalis* agglutinin; GNA) in transgenic wheat plants: effect on predation by the grain aphid *Sitobion avenae*. *Mol. Breed.*, 5: 65-73.
- VASCONCELOS I.M., OLIVEIRA J.T.A. 2004. Antinutritional properties of plant lectins. *Toxicon*, 44: 385- 403.
- YAMASHITA H., THEERASILP S., AIUCHI T., NAKAYA K., NAKAMURA Y., KURIHARA Y. 1990. Purification and complete amino-acid sequence of a new type of sweet protein with taste-modifying activity, Curculin. *J. Biol. Chem.*, 265: 15770-15775.

Wpływ lektyny izolowanej z fasoli zwykłej na bionomię mszycy zbożowej *Sitobion avenae* (F.).

Streszczenie

Celem pracy była ocena wpływu lektyny izolowanej z fasoli zwykłej (PHA) na bionomię mszycy zbożowej. Badania prowadzono w warunkach laboratoryjnych, metodą „sztucznych diet” w czasie który testowano oddziaływanie różnych stężeń lektyny PHA na populację mszycy zbożowej. Przeprowadzone badania wykazały, że lektyna PHA nie wykazuje właściwości fagostymulujących w stosunku do bezskrzydłych samic mszycy zbożowej. Stwierdzono również, że testowana lektyna wykazuje toksyczny efekt w stosunku *Sitobion avenae* ograniczając przeżywalność i płodność bezskrzydłych samic tego gatunku mszycy oraz oddziałując negatywnie na przyrost masy ciała badanych *apterae*. Wyniki te wskazują, że lektyna PHA może być jednym z białkowych markerów odporności limitującym rozwój mszycy zbożowej.

