The influence of wheat cultivar on bug (*Nysius huttoni*) damage to wheat

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Abstract

New Zealand wheat cultivars were compared for their resistance to attack by the New Zealand wheat bug, *Nysius huttoni*, which deposits a proteinase into grain while feeding. None of the cultivars tested showed distinct resistance to bug attack in the field. A flour containing bug proteinase was blended with sound flour from different cultivars and the effects on bread baking and the level of proteolysis of gluten by bug enzyme were measured. Cultivar Oroua had gluten of most resistance to the effects of proteolysis by bug enzyme. The poorest bread-baking cultivar, Karamu, had gluten of least resistance to the effects of proteolysis. The soft white wheat, Arawa, was more resistant to the effects of bug proteinase on baking than all the semi-hard red wheat cultivars that were tested except Otane. Of the red wheat cultivars, Otane showed greatest resistance to the effects of bug proteinase on baking, and Kotare and Rongotea showed least resistance. Within a cultivar group, samples of the highest baking quality showed the greatest resistance to the effects of bug proteinase on baking.

Additional key words: *susceptibility, resistance, bug infestation, bread baking, gluten proteolysis, bug proteinase, loaf volume, crumb texture, caged insects.*

Introduction

Wheat in various parts of the world can be infested by certain Heteropterous insects (wheat bugs) which seriously affect the end use of wheat, particularly for bread-baking (Paulian and Popov, 1980; Swallow and Cressey, 1987). The wheat bugs deposit proteolytic enzymes into immature grain while feeding. The proteinases remain in the grain during its subsequent maturation and milling into flour. When bread is made from such flour, the proteinase can destroy gluten structure and thereby produce slack, sticky dough and poor quality bread.

There is evidence that wheat cultivars vary in their susceptibility to bug attack. When wheat was caged with controlled numbers of the New Zealand wheat bug, *Nysius huttoni* White (Heteroptera : Lygaeidae), cv Karamu suffered far greater visible bug damage than cv Rongotea (Every *et al.*, 1989). In Europe, *Eurygaster integriceps* Put. (Pentatomidae) attacks hard wheats more severely than soft wheats (Paulian and Popov, 1980). In America, certain wheat cultivars are relatively resistant to attack by *Blissus leucopterus* Say which belongs to the same family as *N. huttoni* (Bonnemaison, 1980).

There is also evidence that cultivars vary in their resistance to the effects of bug proteinase on gluten structure and dough properties. Kretovich (1944) showed that wheats with strong dough properties were more resistant to the effects of bug proteinase (as measured by dough extensibility after incubation) than wheat with weak dough properties. Cressey (1987) found that the gluten of certain New Zealand wheat cultivars had a degree of resistance to the effects of bug proteinase (as measured by a sodium dodecyl sulphate (SDS)-sedimentation test) but this was not related to dough strength properties. Over many years of bake testing in New Zealand, cv Karamu has shown more effects and the cv Arawa fewer effects of bug damage than other cultivars (Simmons, 1983; Swallow and Cressey, 1987). It is not known whether these cultivars vary in their susceptibility to insect attack or their ability to resist the effects of bug proteinase on baking.

It would be useful to the wheat industry to find cultivars that either resist bug infestation in the field or resist the effects of bug proteinase on baking. This paper investigates the susceptibility of various New Zealand wheat cultivars to attack by *N. huttoni* in controlled cage experiments. It also examines different cultivars for their resistance to gluten proteolysis by bug enzyme and resistance to loss in baking quality caused by proteinase action. Finally, the effect of baking quality of wheat on resistance to bug damage is analysed.

Materials and Methods

Caging insects on wheat

For this pilot study on cultivar resistance to insect attack, four common New Zealand wheat cultivars in 1987, Karamu, Rongotea, Otane, and Oroua, were sown on 17 November 1987 at the Lincoln DSIR Research farm. The wheat was sown late in order to ensure availability of *N. huttoni* adults, which are locally most abundant in February, at the time of grain-filling. On 9 February 1988, groups of three ears at the watery ripe growth stage were enclosed in nylon gauze bags and colonised with 50 field-collected adult *N. huttoni* per bag. Ten replicates were set up in each cultivar. In addition, three groups of three ears of Karamu were bagged but not infested as a control treatment. Cages were removed and grain was harvested on 15 March. The number of kernels with distinctive feeding marks was visually determined. Feeding damage consisted of an opaque yellow patch, usually with a dark puncture mark at the centre.

For the bake test and SDS-sedimentation test experiments, cv Karamu was sown on 25 May 1987. On 15 December 1987 five 1 m^2 areas of wheat at the watery ripe growth stage were caged in nylon gauze and colonized with adult *N. huttoni* at a rate of $5000/m^2$. Five caged control plots of cv Karamu were free of *Nysius*. The cages were removed and grain was harvested on 10 February 1988.

Bake test comparisons of cultivars

Grain samples of the most common New Zealand cultivars in 1988 (Otane, Kotare, Oroua, Arawa, and Rongotea) were selected from the 1988 Grain Processing Laboratory harvest file to represent bake scores of 20, 24, and 28. Another common cultivar, Karamu, was not included because no samples could be found to fit the above bake score groups. These samples and the bulk caged samples of control and bug damaged Karamu were conditioned to 15% moisture content. White flour was prepared on a Brabender Junior Quadrumat at an extraction rate of 63% for bug damaged Karamu and about 70% for all other samples. Flours were blended as described below and bread was made by the Wheat Research Institute (WRI) 50g Mechanical Dough Development process (Swallow and Baruch, 1986). Doughs were produced from 50g white flour using average water absorption and work input values as determined on a 125g Mitchell Mixer (Mitchell, 1984) for the variety of sound wheat being tested. The dough was proved for 8 min at 32°C, moulded, proved again for 45 min at 40°C then baked for 25 min at 210°C. Loaves were left overnight at room temperature (20°C) before assessment of volume, by rape seed displacement, and crumb texture. Loaf volume (ml) was converted to volume score (arbitrary units) and combined with a crumb texture score to give a bake score as described by Swallow and Baruch (1986).

Flour samples from three initial bake scores (20, 24, and 28) of five cultivars (Otane, Oroua, Kotare, Rongotea, and Arawa) were baked with the addition of 15% bug-damaged Karamu flour (bug group) or with addition of 15% sound Karamu flour (control group). Loaf volume, crumb texture, and bake score results for bug and control groups were determined for each combination of bake score group and cultivar. The data analysed were the differences in loaf volume, crumb texture and bake score between bug and control group. These differences were analysed as a completely randomized 3x5 factorial design (3 bake score groups by 5 cultivars) with 4 replicates. Each replicate was a separate loaf of bread. The bake scores of control samples were reduced from their initial bake scores, possibly due to the addition of 15% Karamu flour, and consequently bake score groups were categorized as high (>22), medium (20-22) or low (<20). No samples were categorized as high for Rongotea or Arawa and results from one Otane bug group were lost because of baking problems. This resulted in an unbalanced design with three missing treatments. Means for bake score groups and cultivars as main effects were separated by Waller-Duncans K-ratio. All results were analysed by SAS statistical package on a Sperry XT personal computer.

Cultivars and proteolysis of gluten by bug enzyme

Grain samples of the New Zealand cultivars Advantage, Arawa, Karamu, Kotare, Oroua, Otane, and Rongotea were randomly selected from the 1988 Grain Processing Laboratory harvest evaluation file. Arawa is a soft white wheat cultivar, the others are semi-hard red wheats. White flour was prepared on a Brabender Junior Quadrumat at an extraction rate of about 70%. Each flour sample was blended with 13% sound Karamu flour (control samples) or with 13% bug damaged Karamu flour (bug samples) from the large cage experiment. Pilot experiments showed that addition of 13% bug-damaged flour gave the best discrimination between proteolysis values of different cultivars. The blended flour samples were tested for proteolysis of gluten by a modification of the SDS-sedimentation test described by Cressey and McStay (1987). To 0.5 g flour in a 10 ml measuring cylinder was added 5 ml of 0.05 M glycine-NaOH buffer, pH 9.0. The mixture was shaken vigorously for 15 sec, placed in a 37°C water bath and shaken again at 2, 5, 10, 20, 25 and 30 min. Immediately after the final shake SDS-lactic acid reagent (5 ml) was added and mixed in by inverting the cylinder five times. Inversion was repeated after a further 1, 2, 3, 4, and 5 min. The contents of the cylinder were then allowed to settle for 15 min before sedimentation volume was read to the nearest 0.1 ml. Ten samples could be tested simultaneously. The sedimentation volume for a sample showing proteolysis was lower than control sample. The amount of proteolysis (proteolysis value) was defined as the difference between the sedimentation volumes of control and bug-damaged samples. Each sample was tested in duplicate. Duplicates were all within 0.2 ml of one another.

This experiment compared proteolysis results for the seven cultivars in a completely randomized design. The proteolysis result for each cultivar was calculated as the mean for six randomly selected samples within each cultivar. The Waller-Duncan K-ratio was used to determine significant differences ($p < 0.05$) among cultivars.

Results

Cultivar resistance to bug infestation

None of the cultivars showed distinct resistance to attack by *N. huttoni* (Table 1). Comparison of the average percentage of damaged kernels suggested that cultivars Karamu, Otane, and Oroua were equally susceptible to bug damaged but less susceptible than Rongotea. This difference, however, was not significant because of the large variation in the percentage of damaged kernels in each caged sample. No bug-damaged kernels were found in the uninfested (control) ears of Karamu.

Table 1. Influence of wheat cultivar on *N. huttoni* damage.

Cultivar	Visibly damaged kernels (%) Mean[1]	Range
Rongotea	79	(1 - 100)
Otane	63	(34 - 81)
Oroua	61	(23 - 86)
Karamu	61	(15 - 79)

[1] Values are means of 10 samples.

Cultivar effect on baking of bug damaged wheat

The results are summarized in Tables 2 and 3. Analysis of variance indicated no significant interaction between bake score groups and cultivars for differences in loaf volume, crumb texture or bake score. However, Tables 2 and 3 show that volume, crumb texture, and bake scores for bug samples were always lower than those from control. For comparison of cultivars, data were combined for all bake score groups of each cultivar. For comparison of bake score groups, data were combined for all cultivars of each bake score group.

Loaf volume results showed that the greatest mean reduction was in Kotare (72 ml), but this was not significantly different from the lowest mean reduction in Rongotea (61 ml). Loaf volume results also showed no significant difference among bake score groups.

Crumb texture results showed that the mean reduction was significantly ($p < 0.05$) greater than in Rongotea (6.0) in all other cultivars. Mean reductions in Kotare (4.9) and Oroua (4.7) were significantly higher than in Otane (3.7) and Arawa (3.0). Crumb texture results also showed that the mean reduction in the high bake score group (3.4) was significantly smaller than for the medium (4.7) and low (5.3) groups.

Loaf bake score results showed the greatest mean reductions in Kotare (17.2) and Rongotea (16.9), and these were significantly greater than all other cultivars. Mean reduction in Oroua (15.2) was significantly greater than in Arawa (13.4) but neither of these were significantly different from Otane (14.4). Loaf bake score results also showed that the mean reduction in the high bake score (14.5) group was significantly lower than for the medium (16.0) and low (16.0) groups.

Cultivar effect on proteolysis of gluten by bug enzyme

The comparison of proteolysis results between cultivars is shown in Table 4. Statistical analysis of results showed the mean proteolysis value for Karamu was significantly higher than for all other cultivars, and the mean value for Oroua was significantly lower than for all other cultivars. There was no significant difference between Arawa, Advantage, Kotare, Rongotea, and Otane.

Discussion

All the cultivars tested appear to be equally susceptible to bug damage. The higher level of bug damage in Rongotea, although not significantly different from the other cultivars, was in contrast to an earlier cage experiment which indicated that Rongotea was less

Table 2. Influence of cultivar and wheat quality on the effect of bug-damaged wheat in baking.

Cultivar	Bake score group[1]	Loaf volume (ml)[2]			Crumb texture[b]		
		Control	Bug	Reduction	Control	Bug	Reduction
Otane	Low	306	242	64	6.5	2.3	4.2
	High	324	251	73	7.7	4.5	3.2
Oroua	Low	292	223	69	6.8	1.3	5.5
	Medium	305	232	73	8.0	2.8	5.2
	High	327	273	54	9.3	6.0	3.3
Kotare	Low	306	239	67	6.5	1.0	5.5
	Medium	314	241	73	6.8	1.5	5.3
	High	334	257	77	8.0	4.3	3.7
Rongotea	Low	287	234	53	7.3	1.0	6.3
	Medium	303	231	72	7.8	1.8	6.0
	Medium	317	259	58	8.3	2.8	5.5
Arawa	Medium	320	266	54	7.5	4.8	2.7
	Medium	330	259	71	6.5	3.3	3.2
SEM		7.2	7.2	6.1	0.6	0.6	0.5

[1] low <20; medium = 20-22; high >22
[2] Means of four replicates are presented; SEM calculated for the data as indicated.

Table 3. Influence of cultivar, wheat quality and protein content on the effect of bug-damaged wheat in baking performance, as measured by bake score.

Cultivar	Bake score group[1]	Bake score[2]		
		Control	Bug	Reduction
Otane	Low	19	4	15
	High	23	9	14
Oroua	Low	18	1	17
	Medium	20	5	15
	High	25	12	13
Kotare	Low	19	2	17
	Medium	21	3	18
	High	24	8	16
Rongotea	Low	18	1	17
	Medium	20	3	17
	Medium	22	6	16
Arawa	Medium	22	10	12
	Medium	22	7	15
SEM		1.1	1.1	0.9

[1] low <20; medium = 20-22; high >22
[2] Means of four replicates are presented.

susceptible than Karamu (Every *et al.*, 1989). The large variation of results between different cages on the same cultivar we encountered in this experiment was also found by Morrison (1938) in cage experiments with *N. huttoni.* These large variations within an experiment and from year to year make it difficult to test for cultivar susceptibility. A new experiment is planned based on a randomized complete block design, lower insect densities, and including soft white wheat cultivars.

Table 4. Influence of cultivar on proteolysis of gluten by bug enzyme.

Cultivar	Proteolysis values (ml)	
	Mean[1]	Range
Oroua	1.8	(1.0 - 2.6)
Otane	2.6	(1.8 - 3.6)
Kotare	2.7	(1.8 - 3.1)
Rongotea	2.7	(1.8 - 3.5)
Arawa	2.9	(1.8 - 3.9)
Advantage	2.9	(2.4 - 3.5)
Karamu	3.8	(3.3 - 4.3)
SEM	0.6	

[1] Values are means of 6 samples.

None of the cultivars tested gave flour showing marked resistance to the effects of bug proteinase on baking. However, the effect of bug proteinase on loaf bake score results showed small but significant differences among cultivars, mainly because of the crumb texture results. Of the red-wheat cultivars, Otane showed greatest resistance to the effects of bug proteinase on baking. The only white wheat cultivar, Arawa, was more resistant than all red wheat cultivars except Otane. This result may partly explain why Arawa has been the cultivar with the lowest level of bug damage, as measured by test baking, for all the bad bug-damage seasons since 1962 (Swallow and Cressey, 1987; Every, D. and Lindley, T.N. unpublished). On the other hand, because Arawa is a soft wheat, it may have resisted insect attack in the field during the bad seasons. Soft wheats in Europe appear to be most resistant to bug attack (Paulian and Popov 1980). Arawa is the only soft wheat in the present experiment; the other cultivars are semi-hard, red wheats. Whether the softness characteristic of wheat is related to resistance to bug infestation or reduced effect of bug-proteinase on test baking requires further investigation.

The present results show that wheats with lower bake scores tend to be more susceptible to the effects of bug proteinase on baking. This is consistent with the results of Kretovich (1944) showing that wheat samples of poor bread baking quality are most susceptible to the effects of bug proteinase on dough extensibility. The results are also consistent with the observation that wheat cultivars of generally low baking quality, such as Karamu, show the effects of bug proteinase in routine bake testing more readily than cultivars of generally high baking quality (Every, 1991). In recent years, Otane has proved to be the cultivar of highest bread baking quality (Simmons, 1991) and, in relation to bug damage, there might be some advantage in growing Otane in regions prone to bug infestation.

The apparent resistance of Oroua gluten to proteolysis by bug enzyme confirms the results of Cressey (1987). This result does not appear to be related to the baking results where Oroua is not the most resistant cultivar to the effects of bug damage. Also, the resistance of Arawa to the effects of bug proteinase on baking does not appear to be related to the gluten proteolysis results where Arawa shows no special resistance. Compared to the other cultivars, Karamu appears to have a degree of susceptibility to proteolysis by bug enzyme. It is possible that the quality of the gluten proteins has an effect on the degree of protein degradation, as Karamu has the poorest protein quality of all the cultivars tested.

Overall, none of the cultivars tested showed marked resistance to bug infestation or the effects of bug proteinase on baking. However, because Arawa and Otane showed a degree of resistance in bake tests there might be some advantage in growing these cultivars in regions prone to *N. huttoni* infestation. The tendency for wheat samples of lower bread baking quality to be more susceptible to the effects of bug proteinase suggests that cultivars with generally poor bread baking quality should not be grown in regions prone to bug infestation. This does not apply to wheat that is destined for biscuit production in which case bug damage has negligible effect (Every, D. unpublished).

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