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Protein and uric acid contents of cereal grains as affected by insect infestation

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A significant increase was recorded in the levels of total nitrogen, total protein, non-protein nitrogen and uric acid contents of wheat, maize and sorghum grains having 25, 50 and 75% infestation caused by *Trogoderma granarium* Everts and *Rhizopertha dominica* Fabricius separately and in mixed population. On the other hand, there was a significant reduction in protein nitrogen and true protein contents of three cereal grains at 75% infestation level but the decrease was non-significant at the 25 and 50% levels of grain infestation as compared to control (uninfested grains). Both insect species produced uric acid contents above acceptable limits (10 mg/100 g) even at 50% infestation levels in three cereal grains. Variations in contents of nitrogenous compounds during storage (1, 2 and 4 months) were marginal.

INTRODUCTION

Cereal grains are an important dietary component for the majority of populations in Asia and Africa. In addition to being an inexpensive source of carbohydrates, B-vitamins and some minerals, cereal grains also meet much of the protein requirement. Cereals are attacked by several insects during storage (Atwal, 1976). Of these *Rhizopertha dominica* Fabricius and *Trogoderma granarium* Everts are the most serious in tropical and sub-tropical climates (Salunkhe *et al.*, 1985). Insect infestation has been reported to impair protein quality of cereals by increasing uric acid (White & Sinha, 1980; Wehling *et al.*, 1984), creating unhygienic conditions through addition of insect fragments (Swaminathan, 1977) and reducing protein digestibility (Hira *et al.*, 1988; Jood & Kapoor, 1992). The effect of insect infestation has been an increase in total nitrogen content of grains (Pingale *et al.*, 1954; Sudhakar & Pandey, 1987) but a few studies (Pushpamma & Reddy, 1979; Sharma *et al.*, 1979; Nirmala & Kokilavani, 1980) have noted a decrease in protein content in insect-infested grains. However, such information with respect to *T. granarium* and *R. dominica* infestations is scanty. This paper reports the effects of three levels of grain infestation (25, 50 and 75%) caused by two insect species and also different storage periods (1, 2 and 4 months), on total protein, true protein, non-protein nitrogen and uric acid contents of wheat, maize and sorghum grains.

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MATERIALS AND METHODS

Mass culture of two insect species, viz. *Trogoderma granarium* Everts and *Rhizopertha dominica* Fabricius, was maintained in the laboratory at ambient temperature (28–39°C) and relative humidity (60–90%) conditions. The grains of commonly consumed cultivars of three cereals, WH-147 (wheat), Composite Vijay (maize) and JS-20 (sorghum), apparently free from insect infestations, were procured and further subjected to aluminium phosphide fumigation to eliminate hidden insect populations. After fumigation such grains were put in 108 glass jars (20 cm × 15 cm), each containing 1·5 kg of grains. Each cereal grain formed a group of 36 glass jars. The jars were covered with muslin cloth and kept in the laboratory for 10 days. On the 10th day, the moisture level of the grains ranged from 10 to 11% which is congenial for multiplication of both insects (Pingale & Girish, 1967). The jars of each grain were subdivided into three sets.

In the first set of each grain (9 jars), 60 larvae of *T. granarium* per jar were released to obtain three levels of infestation (25, 50 and 75% in three replicates). In the second set, 60 adults of *R. dominica* were released while in the third set mixed populations of both species (30 larvae of *T. granarium* + 30 adults of *R. dominica*) were released to achieve three infestation levels in three replications. In addition to this, in each set, controls (jars without insects) were also kept. It took 1, 2 and 4 months to obtain 25, 50 and 75% levels of infestation, respectively. To obtain the desired infestation levels, grain samples (500 grains/jar) were inspected twice a week after the release of insects and the grains which

showed signs of insect damage were considered as infested and the same were used to calculate infestation percentage. The observation frequency was increased at later stages to ensure 25, 50 and 75% grain infestation, respectively, at ambient laboratory conditions. On the day that the desired levels of infestation were actually achieved, the grains were immediately fumigated with aluminium phosphide. At the end of each experimental storage period, control grains were not subjected to fumigation as they were free from insect infestations due to initial exposure to aluminium phosphide. After fumigation at each infestation level, grains were cleaned by passing over a 4 mesh sieve to separate insects, frass (dust) and insect remains from infested grains. Control grains were free from frass and insect fragments during sieving. Grains, after cleaning, were powdered in a cyclotec mill to pass a 60 mesh sieve and then stored in air-tight polyethylene bottles for further chemical analysis.

True protein was estimated by the method of Osborne and Voogt (1978). Total nitrogen was determined by the micro-Kjeldahl method (AOAC, 1980). Non-protein nitrogen was calculated by subtracting true protein nitrogen from total protein nitrogen. Total protein was calculated by multiplying the N content by 6·25 for maize and sorghum and 5·70 for wheat. Extraction of uric acid was carried out by the AOAC method (1980) and it was estimated by the method of Oser (1971). The data were analysed by analysis of variance (ANOVA) to determine significant differences among treatments (Snedecor & Cochran, 1968).

RESULTS AND DISCUSSIONS

There was a progressive increase in total nitrogen, non-protein nitrogen and uric acid with increased levels of infestation in wheat (Table 1). Both species of insects caused almost equal increases in total nitrogen level but *R. dominica*, at the 75% level of infestation, caused a significantly ($P < 0·05$) higher increase in non-protein nitrogen than *T. granarium*. At the 50% level of infestation, *T. granarium* produced 15·5 mg/100 g uric acid and *R. dominica* added 19·0 mg/100 g uric acid. Infested wheat was reported to contain higher total nitrogen (Pingale *et al.*, 1954; Sudhakar & Pandey, 1987; Hira *et al.*, 1988) due to loss of endosperm and addition of insect fragments and uric acid. Increase in total nitrogen and total protein in this study was due to the increase of non-protein nitrogen. However, insect feeding has actually decreased the true protein content from 11·8% in uninfested grains to 9·0–11·0% in 75% infested grains. Similarly, there was a proportional decrease in protein nitrogen. Wheat grains contain higher proportions of true protein in the germ (26%) and bran (48%) components of seed (Aykroyd & Doughty, 1970) which are readily damaged by the germ feeder *T. granarium* (Girish *et al.*, 1975). Hence *T. granarium* decreased true protein more than *R. dominica* (endosperm feeder).

Results in maize were similar to those in wheat (Table 2). *T. granarium* decreased protein nitrogen and true protein more than *R. dominica*. Uric acid excretion, at the 75% level of infestation due to *T. granarium* (36·5 mg/100 g), was much higher than in

Table 1. Effect of insect infestation on nitrogen contents of wheat grains (percentage on a dry matter basis)

Insect species	Infestation level (%)	Total nitrogen	Non-protein nitrogen	Protein nitrogen	True protein	Total protein	Uric acid (mg/100 g)
T. granarium	25	2·28±0·02	0·33±0·00	1·95±0·02	11·1±0·04	13·0±0·15	5·20±0·08
	50	2·61±0·02	0·80±0·00	1·81±0·02	10·3±0·06	14·9±0·13	15·5±1·10
	75	3·02±0·01	1·44±0·01	1·58±0·01	9·0±0·03	17·2±0·08	40·2±1·20
	Mean	2·64	0·86	1·78	10·1	15·0	20·3
R. dominica	25	2·33±0·02	0·30±0·01	2·04±0·02	11·6±0·03	13·3±0·14	6·85±0·05
	50	2·67±0·03	0·71±0·02	1·96±0·03	11·2±0·02	15·2±0·15	19·0±0·18
	75	3·44±0·03	1·51±0·02	1·93±0·00	11·0±0·03	19·6±0·06	48·9±1·24
	Mean	2·81	0·84	1·98	11·2	16·0	24·9
T. granarium	25	2·29±0·01	0·29±0·00	2·00±0·02	11·4±0·04	13·1±0·12	6·30±0·20
+	50	2·62±0·03	0·69±0·00	1·93±0·01	11·0±0·05	14·9±0·09	18·0±2·25
R. dominica	75	3·34±0·02	1·51±0·01	1·83±0·02	10·5±0·05	19·0±0·15	44·8±3·20
	Mean	2·75	0·83	1·92	11·0	15·7	23·0
Control	0	2·11±0·01	0·04±0·00	2·07±0·02	11·8±0·02	12·0±0·09	0·04±0·00
Insect species	SE (m)	0·02	0·02	0·08	0·25	0·32	0·18
	CD ($P<0·05$)	NS	0·06	0·24	0·72	NS	0·52
Infestation level	SE (m)	0·04	0·03	0·08	0·26	0·32	0·20
	($P<0·05$)	0·12	0·09	0·22	0·75	0·97	0·60
Insect species	SE (m)	0·07	0·05	0·14	0·48	0·51	0·41
× infestation level	CD ($P<0·05$)	NS	0·15	NS	NS	1·52	1·20

Values are means ± SD of six independent determinations.

Table 2. Effect of insect infestation on nitrogen contents of maize grains (percentage on a dry matter basis)

Insect species	Infestation level (%)	Total nitrogen	Non-protein nitrogen	Protein nitrogen	True protein	Total protein	Uric acid (mg/100 g)
T. granarium	25	1·95±0·02	0·27±0·00	1·68±0·01	10·5±0·06	12·2±0·15	4·20±0·05
	50	2·23±0·03	0·64±0·01	1·59±0·02	9·85±0·05	13·9±0·14	12·2±0·10
	75	2·70±0·00	1·31±0·00	1·39±0·01	8·70±0·06	16·9±0·25	36·5±1·02
	Mean	2·29	0·74	1·55	9·68	14·4	17·6
R. dominica	25	2·05±0·01	0·29±0·00	1·76±0·00	11·0±0·07	12·8±0·15	5·87±0·10
	50	2·38±0·02	0·64±0·00	1·74±0·01	10·9±0·05	14·9±0·20	16·2±0·23
	75	2·98±0·02	1·37±0·01	1·61±0·01	10·1±0·03	18·6±0·09	42·8±1·32
	Mean	2·47	0·77	1·70	10·7	15·4	21·6
T. granarium	25	1·96±0·00	0·22±0·02	1·74±0·01	10·9±0·06	12·3±0·15	5·50±0·10
+	50	2·24±0·01	0·57±0·01	1·67±0·00	10·5±0·04	14·0±0·09	14·1±0·22
R. dominica	75	2·80±0·02	1·21±0·02	1·59±0·00	9·91±0·04	17·5±0·17	40·0±1·39
	Mean	2·33	0·67	1·67	10·4	14·6	20·0
Control	0	1·83±0·01	0·02±0·00	1·81±0·02	11·3±0·05	11·5±0·15	0·06±0·00
Insect species	SE (m)	0·03	0·02	0·09	0·22	0·31	0·80
	CD ($P<0·05$)	NS	0·06	0·27	0·66	NS	2·20
Infestation level	SE (m)	0·04	0·03	0·10	0·25	0·32	0·90
	($P<0·05$)	0·12	0·09	0·30	0·75	0·96	2·62
Insect species	SE (m)	0·07	0·06	0·21	0·45	0·60	1·50
× infestation level	CD ($P<0·05$)	NS	0·18	0·63	NS	1·80	4·20

Values are means ± SD of six independent determinations.

the control (0·06 mg/100 g). Uric acid exceeded acceptable limits even at 50% grain infestation.

Insect infested sorghum grains also showed a significant ($P < 0·05$) increase in total nitrogen, non-protein nitrogen, total protein and uric acid (Table 3). The endosperm component of the sorghum grain contains 80·9% of the seed protein (Hubbard *et al.*, 1950) which is more readily damaged by the endosperm feeder *R. dominica*. Hence, unlike in wheat and maize, *R. dominica* caused a greater reduction in protein nitrogen than did *T. granarium*. Mixed populations of both insect species in three cereal grains produced intermediate results.

During storage (1, 2 and 4 months), there were

Table 3. Effect of insect infestation on nitrogen contents of sorghum grains (percentage on a dry matter basis)

Insect species	Infestation level (%)	Total nitrogen	Non-protein nitrogen	Protein nitrogen	True protein	Total protein	Uric acid (mg/100 g)
T. granarium	25	1·94±0·01	0·26±0·00	1·69±0·03	10·6±0·05	12·2±0·15	4·90±0·08
	50	2·21±0·02	0·57±0·00	1·64±0·03	10·3±0·06	13·8±0·14	13·5±0·10
	75	2·15±0·03	1·17±0·01	1·58±0·02	9·9±0·07	17·2±0·16	38·2±1·30
	Mean	2·30	0·67	1·64	10·2	14·4	18·9
R. dominica	25	1·97±0·01	0·29±0·01	1·68±0·01	10·5±0·05	12·3±0·09	6·00±0·08
	50	2·28±0·03	0·68±0·02	1·59±0·02	9·96±0·04	14·2±0·07	17·5±0·15
	75	2·86±0·02	1·42±0·02	1·44±0·03	9·00±0·03	17·9±0·07	44·8±2·35
	Mean	2·37	0·79	1·57	9·82	14·8	22·8
T. granarium	25	1·97±0·03	0·28±0·02	1·68±0·03	10·5±0·07	12·3±0·08	5·70±0·10
+	50	2·24±0·01	0·61±0·00	1·63±0·03	10·2±0·06	14·0±0·13	15·9±0·25
R. dominica	75	2·76±0·03	1·18±0·02	1·58±0·04	9·88±0·08	17·3±0·09	42·0±2·29
	Mean	2·32	0·69	1·63	10·2	14·5	21·2
Control	0	1·74±0·02	0·01±0·00	1·73±0·01	10·8±0·03	10·9±0·15	0·05±0·00
Insect species	SE (m)	0·02	0·02	0·08	0·23	0·32	0·85
	CD ($P<0·05$)	NS	0·06	NS	NS	NS	2·52
Infestation level	SE (m)	0·02	0·02	0·07	0·25	0·35	0·90
	($P<0·05$)	0·06	0·06	0·19	0·75	1·05	2·62
Insect species	SE (m)	0·04	0·03	0·14	0·45	0·70	1·15
× infestation level	CD ($P<0·05$)	NS	0·06	NS	NS	NS	3·45

Values are means ± SD of six independent determinations.

minor variations in the total nitrogen, protein nitrogen, non-protein nitrogen and uric acid contents of three cereal grains. Therefore, it may be inferred from these studies that insect infestation in cereal grains causes substantial reductions in protein nitrogen and true protein contents, and increases in total nitrogen, total protein, non-protein nitrogen and uric acid contents.

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