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Notes - Research

\*EVALUATION OF SOME CASSAVA VARIETIES TO SUPPORT GROWTH

OF TRIBOLIUM LARVAE

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Bioassays based on the growth response of larvae of Tribolium castaneum have been used in the nutritional evaluation of starches, rice flours and carbohydrates (Vohra et al., 1979). We have studied the growth response of the Tribolium larvae fed some varieties of cassava meals. The cassava varieties were grown in Philippines. The harvested tubers were washed with water, gated, dried and ground to a fine powder to pass

A 100 mesh sieve. As received, the samples were in this powder form. The samples were analyzed for moisture, crude protein (CP), and ether extract (EE) following procedures described in AOAC (1979). The method of Hellendoorn et al. (1975) was used for determination of indigestible residue (IDR), a measure of unavailable carbohydrates. Available carbohydrates (ACHO) were measured by the method of Southgate (1969).

A part of the sample was autoclaved at 121 degree C for 20 minutes. Another was washed with water using 100 ml water per 20 g sample. After allowing the cassava to settle, supernatant was discarded and acetone was added to the sediment to precipitate the powder. After filtering the slurry on a suction filter, the residue was air dried till free from acetone odor. This was the leached material. A part of this was heated at 105 degree for 5 minutes, and another leached sample was heated for 2 hours. A part of the leached sample was autoclaved at 121 degree C for 20 minutes and another for

40 minutes.

The bioassay has been described by Vohra et al. (1979). Tribolium stock was maintained on a diet containing 90% unbleached wheat flour and 10% brewer’s yeast. The eggs from a 24 hour collection were kept in the stock diet and the larvae were transferred to the test diets for acclimatization on day 4. At day 6, three replicates of 10 larvae were fed the test diets. They were sieved out of the diets on day 14, and weighed. The test diets contained 80% cassava powder, 11.25% brewer’s yeast and 8.75% isolated soybean protein to supply 12.5% CP. The average larval weight on a test diet reflects the relative nutritive value of cassava powder. These weights were compared against the larval weights on the stock diet.

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Table 1 - Chemical composition of some varieties of cassava meals as obtained from Philippines

Cassava Moisture CP EE Carbohydrates HCN

Varieties % % % IDR ACHO mg/kg

% %

1 Branco 9.80 2.62 4.98 7.69 64.00 2.00

2 CMC – 76 6.90 2.01 5.17 13.12 60.00 0.72

3 Hawaiian-4 10.95 1.87 5.16 9.77 60.33 0.79

4 Java Brown 9.63 1.95 5.62 8.83 61.73 1.48

5 N-Mex-59 10.93 1.69 5.71 9.68 63.30 0.97

6 M-Ven 218 10.58 1.43 5.59 8.55 63.13 1.00

7 SMI-150 10.58 1.50 5.46 8.27 58.57 0.78

8. Vaseaininha 10.33 1,74 4.63 9.38 62.53 0.74

9 W-78 10.32 1,74 5.96 9.82 64.20 0.77

SEM 0.58 0.12 0.34 0.42 1.56 0.10

CP = crude protein, EE = ether extract, IDR = indigestible carbohydrate,

ACHO = available carbohydrate, HCN = hydrogen cyanide.

Table 2 – Average body weights of larvae of Tribolium castaneum fed diets

Containing various cassava meal samples as obtained and after

Various treatments

Average larval weight (mg) on diet containing cassava meal

Cassava variety Untreated Untreated Leached Leached Leached Leached

Autocl. Heated heated autocl. autocl.

121 deg. C 5 min. 2 hr, 121deg.C 121 deg.C

20 min 105 deg.C 105 deg.C 20 min. 40 min.

None (control) 2.95e 3.03g 3.13f 3.08f 3.03e 3.03e

1 Branca 0.90bc 1.55ef 1.06b 1.68cd 1.96cde 2.02ab

2 CMC-76 0.94bc 1.36d 1.28c 1.36ab 1.77cd 2.01ab

3 Hawaiian-4 0.71a 1.08ab 0.87a 1.21a 1.92cde 2.12b

4 Java brown 1.04c 1.56ef 1.08b 1.48bc 1.84cd 2.38c

5 M-Mex-59 1.02c 1.29cd 2.07e 1.64cd 1.91cde 2.55d

6 M-ven 218 0.80ab 1.52e 1.23c 1.69cd 1.97cde 2.83d

7 SMI – 150 .0.96bc 1.20bc 1.08b 2.18e 1.32a 2.41c

8 Vaseaininha 1.46d 1.65f 1.74d 1.79d 2.01ed 2.58d

9. W-78 0.72a 1.00a 1.05b 1.32ab 1.57b 1.97a

SEM 0.03 0.02 0.02 0.04 0.03 0.02

F-ratio 28.58 41.87 59.74 11.46 13.71 15.52

Treatment means 1.1a 1.5b 1.5b 1.7bc 1.9c 2.3d

Varietal means 1.5ab 1.5ab 1.3a 1.5ab 1.9c 1.3a

a to g. Values with different letters in a column are significantly different (P 0.01).

The results of the chemical composition of the tested varieties of cassava meals are given in Table 1. There were significant (P 0.01) differences in the determined parameters.

The data on the ability of cassava varieties to support larval growth when fed at a level of 80% in diets containing about 12.5% protein are given in Table 2. No significant (P 0.05) correlation could be established between any of the compositional parameters and the growth of Tribolium larvae. However, some varieties were significantly (P 0.01) better than the other in supporting larval growth. The difference was not due to their HCN content. Cyanide levels present in the cassava meals did not influence larval growth.

The processing procedures significantly influenced larval growth. The meals were improved in their ability to support optimal larval growth after leaching, and after heating in an oven at 105 degree C for 5 minutes. Autoclaving of leached meals at 121 degree C for 40 minutes was better than for 20 minutes, or dry heating even for 2 hours for larval growth.

In summary, significant differences were observed in the ability of different varieties of cassava meals, which was unrelated to their HCN content, to support the growth of Tribolium larvae.

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\*Egg size of Tribolium anathe Hint. (Coleoptera: Tenebrionidae) reared on some

Cereal flours and their mixtures.

Tribolium anaphe Hint. Is an Ethiopean species, originating in Africa and infesting groundnuts there. Hinton (1948) considers it to be a potential pest in the tropical countries. This beetle attacks a great variety of stored commodities. Barley (Hordeum vulgare) and rice (Oryzu sativa) are two important cereals and are consumed in various forms.

The eggs of T. anaphe, when washed free from flour particles, are small, oval, clearwhie and sticky. The eggs vary in size, which. According to Stanley 1965), is inherent in the egg and is not the result of environmental factors after oviposition. Steinhaus (1965) and Sokoloff (1974) reviewed the various types of abnormalities found in eggs. Egg size is an important taxonomic character of insects.

The present investigation aims at determining the effect of wholemeal, barley ad rice flours, and their combinations in equal proportions on the egg dimensions of T.anaphe.

Beetles (T. anaphe) wee collected from a laboratory culture on wholemeal flour in the Department of Zoology, Rajshahi University, Rajshahi, and were put on a thin layer of whole-meal flour in a Petri dish for oviposition. Eggs were sieved on the following day and were incubated at 30 degree C. Newly hatched larvae, 300 for each food, were transferred to individual glass jars (20 x 8 cms), each containing 250 g of wholemeal, barley, rice, wholemeal-barley, wholemeal-rice and barley-rice flours respectively, with a fine brush. After pupation insects were segregated according to sexes (Halstead, 1963) in separate Petri dishes for adult eclosion. Pairs of freshly emerged adults of opposite sexes were introduced into separate glass vials (3.5 x 1.8 cms) containing the experimental foods for oviposition.

Eggs were washed free from adhered flour particles in 0.9% normal saline solution. They were measured for their length and width using a micrometer (40X).

The effect of cereal flours and their mixtures was to decrease the egg dimensions of T. anaphe (Table 1). The highest and lowest egg lengths were observed for the eggs laid by the females reared on wholemeal and barley-rice flours respectively and the highest and lowest egg widths were noted for the eggs laid by the females grown on wholemeal and wholemeal-rice, and barley-rice flours respectively (P 0.001). Thus the results imply that the nature of the rearing media must be taken into consideration for taxonomic studies, and the mention of the rearing media should be made in any biometric analysis.

The authors would like to extend grateful thanks to the Pest Infestation Control Laboratory, Slough, England, for providing the experimental beetles. They extend sincere thanks to Dr. M. Sayedur Rahman, Chairman, Department of Zoology, Rajshahi University, Rajshahi, Bangladesh, for furnishing the necessary laboratory facilities.

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\*Dietary efficiency of corn flour for Tribolium anaphe Hint. (Coleoptera: Tenebrionidae).

Tribolium anaphe Hint.is included into the confusum species group of the nenus Tribolium Macleay (Hinton, 1948) and is a serious pest of several stored commodities, especially in the tropics. Many researchers recorded deleterious effects of corn flour on beetles (Chapman, 1924; Khalifa and Badawy, 1955 a, b; Inouye and Lerner, 1965); Sokoloff et al. 1966; Khare and Pant, 1978). Corn (Zea mays) is an important cereal and is used both as food and fodder.

Howe (1968) pointed out that one of the criteria used in nutritional studies as a guide to the adequacy of a diet is the weight of larvae. However, according to Sokoloff (1974), this is a less satisfactory criterion than the developmental period, because its increase is episodic, with intervals in which it decreases.

The present work is concerned with the determination of the effect of corn flour and a mixture of wholemeal and corn flours in equal proportions on the growth and development of T. anaphe, which seemed important from the point of nutritional regulation of the pest.

A large number of adult T. anaphe were sieved from a culture maintained on wholemeal flour in the Department of Zoology, Rajshahi University, and were placed on a thin layer of wholemeal flour in a Petri dish for egglaying. Eggs, collected on the next day, were incubated in a Petri dish at 30 degree C for hatching. Neonate T.anaphe larvae, 300 for each food, were transferred to individual glass jars (25.4 x 11.4 cms) containing 250 g wholemeal (control), corn, and wholemeal + corn flour mixture in equal weights. The jars were closed at their mouths with a piece of fine-netted cloth. The growth of the larvae was assessed at two stages : after 10 days and at maturity. They were individually weighed on an electric balance, and their length was measured with a scale and their headcapsule width with a micrometer (40X). The insects were carefully observed for pupation. Pupae were sieved, cleaned from adhered flour particles, and were sexed by the microscopic examinations for the exogenital processes of the females (Halstead, 1963). They were weighed and measured according to their sexes similarly. Larval periods and pupation (%) on foods were carefully noted. Pupae were segregated according to sexes in separate Petri dishes for adult eclosion. Freshly eclosed adults were then weighed and measured according to sexes. Pupal periods and adult eclosion (%) on various foods of the beetle were recorded.

\*For correspondence.

The coefficient of variance (C.V.) was calculated using the formula : V = (100Xv)/M, where of the V = coefficient of variance, v = standard deviation of the mean of the sample, and M = mean of the sample (Simpson and Roe, 1939). The growth indices (GI) of T.anaphe on different foods were determined according to the formula : GI = Adult eclosion (%) / sum of larval and pupal periods. All the experiments were conducted at 30 degree C.

Corn flour significantly reduced larval, pupal and adult growths of T.anaphe (P (0.001) (Tables 1 and 2). It also decreased pupation and adult emergence of the beetle significantly (P (0.001), the order was wholemeal ) wholemeal + corn) corn flours

(Table 3). In addition, corn flour significantly lengthened the larval and pupal periods of T. anaphe (P (0.001) followed by wholemeal + corn flour mixture (Table 4). The values for the growth indices indicate that corn flour proved to be the least and wholemeal flour to be the best rearing media for T.anaphe respectively.

Hasan and Khan (unpublished data) observed significantly reduced fecundity and fertility of T. andphe when grown on corn flour and a mixture of wholemeal and corn flours. Insect, like other animals, develop nutritional diseases when subjected to faulty nutrition. The familiar symptoms of most nutritional defects are : slow or arrested growth and development, diminutive size, and high or complete mortality of the immature stages and little or no reproduction in the adult. According to Geier (1966), one way to manage insect pests is to modify intrinsically favourable habitats in such

a way that they no longer furnish adequate environments for the pest populations involved. The results obtained in the present investigation are promising as regards the control of T. anaphe through nutritional regulations.

The authors are grateful to the Slough Laboratory, MAFF, England, for supplying the experimental insects and to Dr. M. Sayedur Rahman, Chairman, Dept. of Zoology, Rajshahi University, for providing laboratory facilities and also to Mr. Shamimul Haque, Computer center, Rajshahi University, for typing this manuscript.

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\*Life Table for Tribolium freeman Hinton.

Mortality and natality are important factors which affect population numbers. A life and fertility table is a convenient format for describing the mortality and reproduction schedules of a population (Krebs, 1972). Several life table studies have been made for Tribolium species of economic importance, i.e., T. castaneum (Mertz, 1969; Young, 1970), T. confusum (Pearl et al., 1941; Young, 1970) and T. madens (Park, 1945). The present note aims at determining the life table of Tribolium freeman Hinton at an optimal condition of 32.5 degree C and 70% r.h. (Imura & Nakakita, 1984) with an ample supply of food. The data derived from a demographic study of T.freemani (Imura, 1987).

The insects used were the offspring of T. freeman found in 1978 in Japan (Nakakita et al., 1981). Two hundred newly hatched larvae were introduced into a plastic container (13 cm diam. By 7 cm deep) with 300 g of a medium consisting of fine wheat shorts and dried Brewer’s yeast (19:1 by weight), which had been tempered in a dark experimental room at 32.5 + 0.5 degree C ad 70 + 7% r.h. Pupae were separated from the medium with a No.14 mesh sieve as the larvae pupated and transferred to a glass bottle (5.5 cm diam. By 9 cm deep) containing 100 g of a mixture of finely sifted wheat flour and dried Brewer’s yeast (19:1 by weight). The bottle, which was covered with a cotton gauze, was kept in the experimental room. A census of the adults and eggs in the bottle was taken at 3-days intervals by sifting the flour medium with sieves of No.14 and 40 mesh and the medium was replenished at 9 days intervals. Living adults were returned to the bottle after counting and dead adults were kept in 10% KOH solution for latter examination of sex. Hatchability of eggs was also determined on an adhesive tape (Imura \* Nakakita, 1984).

Cohort life tables for females and males (Table 1) were constructed according to Deevey (1947). Survivorship curves of females and males are also illustrated in Fig. 1. Males lived longer than females. The mean duration of life was 329. 1 days for females and 346.7 days for males and the maximum life-span recorded was 639 days for females and 750 days for males. The shorter longevity of females resulted in the lower life expectancies (e x) in females (Fig.1). Imura (1987) reasoned that the continuous reproductive effort of female (Table 1) decreased her survival rate. The life expectancy of both sexes was maximum in the age group of 30-60 days (Fig. 1); in average a male of this age group is expected to live ca. 400 days further while a female ca. 350 days. The survivorship curves were a convex type, i.e., type I (Deevey, 1947), indicating that juvenile mortality was small and most of the deaths occurred in old age (see also d x and q x columns in Table 1). The life and fertility table for females.suggested that the type of life-history of T. freeman was a continuous interoparity (Begon et al., 1986), which is thought to have evolved under an environment with uncertain pre-reproductive survival and relatively stable survival of reproductive ages (Murphy, 1968). Therefore, T.freemani might exhibit different survivorships from those observed in this study in their natural habitats. The gross reproduction rate (G.R.R. = m x) and the net reproduction rate (R o = 1 x m x ) (Leslie & Park, 1949) was 529 and 351, respectively. The estimated mean generation time (T) was 81 days and the intrinsic rate of natural increase (r m) was 2.182 per 30 days (Imura, 1987), indicating that T. freeman had a relatively low population growth rate.

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\*Combined action of pirimiphos-methyl and tobacco (Nicotiana tabacum) leaf powder on the larval mortality of the onfused flour beetle, Tribolium confusum Duval.

Flour and other cereal products are subjected to attached by a large number of beetles belonging to the Tenebrionidae. Of these, the most abundant and attractive is the conused flour beetle, Tribolium confusum Duval occurring in flour mills, granaries and other places where grains are stored (Park, 1934; Khalifa and Badaway, 1955). Most workers on this insect studied its biology and ecology, and a few studies have been done on their behavior and control. Khan (1981) used pirimiphos-methyl and microsporidian, Nosema whitei on T. castaneum and Mondal (1984) methyl-quinone, aggregation pheromone and Pirimiphos methyl on the same insect. The present work aims at finding out the combined effect of pirimiphos-methyl on the same insect. The present work aims at finding out the combined effect of pirimiphos-methyl and tobacco

(Nicotiana tabacum) leaf powder on the mortality of the confused flour beetle larvae.

The beetles were collected from the stock culture of the Entomology Laboratory of the Department of Zoology, Rajshahi University. About 1000 beetles were collected from the culture and placed in an 1 lb Kilner jar containing whole wheat flour previously passed through a 60 mesh sieve. On the next day the contents were passed through a 18 mesh sieve to separate the adults and then through a 60 mesh sieve to collect the eggs. Neonate larvae from the eggs, 100 for each treated food in a petridishes with five replications were reared. The doses were made with 2 ppm and 3 ppm pirimiphos-methyl (dust) and each of them mixed with 5 X 10 3 ppm and 10 X 10 3 ppm of tobacco leaf powder with whole wheat flour. A control group was also maintained on fresh whole wheat flour to record control mortality. Experiments were conducted at 30 degree C without light and relative humidity control. The mortality of the larvae was assessed after 1, 2, 3, 10 and 20 days after treatment. The results were tested by using Chi-square (x 2) based on an expected mortality for the sum of the two individual effects (Mather, 1940). Significant Chi-square result indicate observed mortality of combined effect is greater than expected and synergism occurring. The combined effect on larval mortality were classified on the criteria for synergism (Howlett, 1960) as described by Benz (1971).

The mortality percentage of T. confusum larvae in two constant doses of pirimiphos- methyl and tobacco leaf powder has been presented in Table – 1. It was found that in some cases the combination effect offered synergism. The synergistic action of tobacco leaf dust is, to some extent similar to the results of Dyte and Rowlands (1970), who reported higher mortality of T. castaneum adults in combined doses of insecticides and synergists in comparison with the mortality due to individual action of chemicals.

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The combined action of compounds with insecticides may vary from synergism to antagonism according to both the test insect (Hadaway et al., 1962) and the toxicant used (Sun and Johnson, 1960, 1965; O’Brien, 1961; Sun et al., 1967). The synergistic action of various pyrethrum synergistic with different insecticides on several species of insects and mites were studied by Hewlett (1960, 1968) and Sun and Johnson (1960). The action of pyrethrum synergists, sesame, SKF 525A, and PAPB-1 (Dyte and Rowlands, 1970); piperonyl butoxide (Ishaaya et. Al. 1983); and low frequency sound (Mullen and Upson, 1974) in combination with several insecticides were studied on Tribolium. The synergistic action of microsporidian N. whitei with sublethal doses of pirimiphos-methyl on T. castaneum was studied by Khan (1981), Mondal (1984) studied the effect of synthetic methyl-quinone in combination with low doses of pirimiphos-methyl on the same insect in their larval stage.

The present results suggest that synergism could be achieved by applying either constant or variable doses of tobacco leaf powder in combination with variable doses of pirimiphos-methyl.

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\*VARIATION OF FECUNDITY AND HATCHABILITY IN STRAINS:

TRIBOLIUM CASTANEUM HBST. cI AND T. CONFUSUM DUVAL Biv.

INTRODUCTION

In the course of bioenergetic studies on T. castaneum Hbst differentiation within strain has been observed in : duration of developmental cycle, the maximum weight attained by larvae, time of reaching it and weight of adult beetles (Prus 1976). Similar differences were found in T. confusum bIV strain (Bijok 1986). These observations enhanced us to examine such features as fecundity, mean individual egg weight, and hatchability within the diserened dgroups in both species. Information on these autecological features is thought to be useful for further interpretation of the populational phenomena in terms of life strategy theory.

Phenotypic differentiation for T. castaneum cI strain was described in details in an earlier study (Prus and Prus 1987). Significant differences were found in development duration, maximum larval weight, fecundity and hatchability of eggs in the two substrains (6- and 7- instar groups).

The period of more than three months of censusing of individual pairs of beetles was considered to be long enough to grasp time changes in the characteristics studied. It was based on the fact that similar period of censusing of Tribolium populations for primary characteristics had been adopted by Park (Park at el. 1961).

The present paper aims at detecting significance of differences in these features mentioned between the substrains. Differences observed between species will be useful for evaluation of position of these two strains on r - K continuum suggested by Stearns (1980).

MATERIAL AND METHODS

All experiments were carried out in standard conditions of 29 degree C, 75% relative humidity, in medium consisting of 95% of wheat flour and 5% of baker’s yeast.

Material used for experiments consisted of beetles representing 6- and 7- instar substrains. They were separated on the basis of differences in attaining pupal stage, similarly as in earlier study (Prus, Prus 1987). The design of experiment consisted of four series (2 species x 2 substrains) with 10 replications in each, except for 7-instar T.confusum, where there were only 4 replications due to the shortage of material

Fecundity of individual pairs kept in 8 g of standard medium was measured as number of eggs laid by a female in 3 day interval. At each census the culture medium was renewed. Fecundity was expressed as number of eggs laid by female during 24 hours, or during 72 hours.

The collected batch of eggs from each pair was weighed on Cahn model G electrobalance in order to assess mean fresh weight of eggs. Thus collected eggs were left over in the incubator to hatch. Number of hatched larvae was counted after six days elapsed from the moment of their deposition. This duration exceeds much the time needed for all eggs to hatch under the experimental conditions.

The results were presented for 3-day intervals in the sub-strains examined (6- and

7-instar groups). However, in order to make the interpretation of interspecies differences more compact the results were grouped in 9-day intervals (3 censuses together).

The significances of difference between substrains were tested using t-test method for combined presentation of results. Probability of p=0.05 was accepted as the significance level.

RESULTS

In T. castaneum cI strain females of 6-instar group lay more eggs than those of

7-instar group (Fig. 1). Maximum fecundity occurs during the first month of adult life. In this period the differences between substrains are statistically significant (Table IA). Later on, at diminishing fecundity these differences became obscured. In T. confusum bIV strain the differences of fecundity between substrains are rather significant throughout the whole experiment and the decreasing tendency in egg laying with time is less conspicuous (Fig. 2, Table IA).

From comparison of the smoothed curves of fecundity in both species it is evident that cI strain is much more fecund than bIV strain. There is more than 50% difference in this feature in favour of T. castaneum This tendency is maintained during the whole period of the experiment.

The mean individual weight of eggs laid by 6- and 7-instar females show practically no significant difference in the two species. The only tendency observed was slightly increasing individual weight of an egg with time, i.e. age of female, in both species. In T. castaneum 6-instar substrain, such increase was from 42.82 ug fresh weight during first 9-day period to 46.06 ug in the end of experiment, and in T. confusum in 6- instar substrain the corresponding values are 54.93 ug fresh weight and 66.08 ug fresh weight. In 7-instar substrains of both species the corresponding values are: 39.71 – 49.45 in T. castaneum and 54.56 – 68.69 ug fresh weight in T.confusum (Table IB).

The analysis of mean weights of eggs reveals that T. confusum lays heavier eggs than those of T. castaneum. There is about 20% difference in favour of the former species. However, there is a similar tendency in both species which depends on increased individual weight of eggs with time, i.e. age of females.

The data on fecundity and mean individual weight of eggs indicate that younger females lay more eggs but the eggs are smaller, and older ones lay less eggs but of a higher individual weight.

Hatchability of eggs laid by T. castaneum females seems to show no difference between substrains, maintaining rather constant level during the experimental period. It oscillates around 60% of total number of eggs laid within 72 h-period (Fig. 3, Table Ic). In T. confusum there is no difference in hatchability between egg laid by females representing two substrains but there is a constant decreasing tendency in hatchability with time from about 60% in the beginning of egg laying to about 40% in the end of experiment (Fig. 3, Table Ic).

Higher hatchability was observed in T. castaneum cI strain tan in T. confusum bIV strain. Due to the decreasing tendency of hatchability in bIV strain with time, the almost equal values of percentage of eggs hatched at the beginning of the experiment in both species were becoming more and more different in the course of the experiment.

DISCUSSION

Strains used in the experiment were derived from the University of Chicago where they have been created and cultured for many years in the laboratory run by prof. Thomas Park. Primary characteristics of these strains are given in paper by Park et al. (1961). If we compare fecundity of cI strain in that time with fecundity of the two substrains of this strain obtained in our laboratory we can state that fecundity of 7-instar group resembles most the fecundity of the initial cI strain. The average number of eggs laid by female per day within first month in cI strain was 16.3 (Park et al. 1961, modified), and that of 7-instar female during first 27 days was 15.9. The 6-instar group shows higher fecundity than that described in the paper cited. It’s fecundity amounts for 27-day period 18.8 eggs per female per day. However, in our earlier experiments (Prus and Prus 1987) the reversed situation was observed, i.e. fecundity of 6-instar group during first month of adult life was significantly higher than that of 7-instar group. So, the whole array of possibilities was observed in mutual relation of fecundities in this two groups (Table II). In T. confusum bIV fecundity of 6-instar group (11.3) resembles most that of bIV strain examined by Park et al. (1961) (11.7 eggs per female per day). Fecundity of 7-instar group is somewhat lower (9.9). Unfortunately, due to inadequate number of experiments it is not possible to infer about long term changes in this strain.

For comparison of fecundity we have chosen first month of adult life since the early performances of individuals seem to be most important in survival success of

a population. In later period the situation is different in the two compared strains, since in T. castaneum cI strain differences become nonsignificant, whereas in T.confusum bIV strain differences ae statistically significant in spite of a low number of replicates.

The diminishing of fecundity with time observed in the present paper (Fig. 1) finds its corroboration in earlier studies performed by Park et al. (1961) for both species and by Takahashi and Yamamoto (1972) for T.confusum wild strain.

It seems that fecundity of examined substrains is not a well established trait yet. Both the experimental work and selection procedure should be continued in order to achieve univocal results.

Considering the characteristic of mean individual egg weight no significant differences were observed between 6- and 7-instar groups in both species (Fig. 2). However, T. confusum bIV strain laid heavier eggs than T. castaneum cI strain, the difference being about 20%. So. T.castaneum cI strain lay more smaller eggs than T. confusum bIV (Fig. 1 and 2). A tendency of larger eggs being laid with increasing age of female is well known in poultry studies (Hutt 1949). It was observed here, too.

Hatchability of eggs in both species shows much higher variation than other traits examined in this paper (cf. standard deviation in Table I). Hatchability of eggs when referred to its first assessment (Park et al. 1961) for cI and bIV strains showed substantial differences. The observed percentages of eggs that hatch in both strains were much lower than these reported by Park et al. (1961) and the interspecies difference became evidently smaller.

When characterizing both strains in terms of life strategy performance it can be said that T. castaneum cI strain is characterized by shorter developmental cycle, earlier maturity, lower energetic value of larval and adult body, higher fecundity, lower individual weight of eggs and higher hatchability. All this predisposes this strain to be r-regulated strategist as compared to T. confusum bIV strain. Our earlier studies (Prus 1976, Bijok 1986, Prus and Prus 1987) showed differentiation of the examined strais into 6- and 7-instar groups. Wen these substrains were tested for autecological traits considered to be substantial for life history strategy, it turned out that 6-instar strains in both species have strategy more of “r” type as compared with 7-instar substrains.

The constant existence of phenotypic groups in populations can have an important significance for their survival. In colonizing new habitats 6-instar groups seem to be more important whereas 7-instar groups are necessary for further maintaince of these populations in already occupied habitats.

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Table II – Changes in fecundity of 6- and 7-instar females of T. castaneum cI and

T. confusum bIV strains in subsequent experiments

(No. eggs x Female – 1 x 72 h)

No. Date 6-instar 7-instar Reference

T. c a s t a n e u m cI

I Jun 1982 50.8 57.5 Prus and Prus 1987  
II Dec 1982/

Jan 1983 55.9 57.9 “  
III Mar/Jun 1986 56.5 48.0 Present paper

T. c o n f u s u m b I V

1 Jan/Feb 1983 38.7 36.3 Bijok 1985  
II Mar/Jun 1986 35.1 33.3 Present paper

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\*COMPARATIVE STUDY OF REPRODUCTIVE EFFORT IN TWO SPECIES OF

TRIBOLIUM

INTRODUCTION

The ratio between lipid content or energetic value of offspring to the body weight of a female or its energetic value has been often used to measure the reproductive effort in animal (Tinkle, Hardley 1975, Clarke 1977, 1979, Grahame 1977). I depicts investment of an organism in its progeny. Other measure were also used to describe the reproductive effort such as ratio of clutch size and clutch weight to the body weight. In the case of Tribolium, a continuous egg laying brings about that daily egg production is the most convenient quantity for calculation of the proportion mentioned. According to Tinkle and Hardley (1975), the best way to estimate relative reproductive effort is finding proportion between the total energy consumed by an organism to that allocated to reproduction. The latter approach will be developed in a more extensive paper compiling data of the present study and energy budgets of T. castaneum cI strain (Klekowsk 1967), and of T. confusum bIV strain (Bijok in press).

The aim of this study is to test differences in reproductive effort between the 6- and 7-instar groups discerned earlier Prus 1976, Bijok 1984) in the above mentioned strains.

MATERIAL AND METHODS

The collected material for pupae and eggs in 6- and 7-instar groups of both the species was dried at a constant temperature of 60 degree C. Then it was homogenized in mortar. A part of material that was devoted for lipids determination was weighted into aliquot; from which lipids were extracted by a mixture of chloroform methanol and naphta ether. After extracting lipids were weighted and its percentage content in dry matter content was calculated.

Another part of the material was used for assessment of energetic value by forming pellets and combusting them in the Phillipson microbomb-calorimeter (Prus 1975). The obtained results were expressed in calories per mg dry weight. The lipids content and energetic value were also determined in the cultured medium consisting of 95% of wheat flour and 5% of powdered baker’s yeast. The cultures for collecting eggs and pupae were run at 20 degree C, 75% RH in a dark incubator.

The compared strains were cI of T. castaneum and bIV of T. confusum, both originating from Thomas Park’s laboratory, from where they were brought to Poland many years ago. Within each of these strains, 6- and 7-instar groups have been distinguished in our laboratory and kept as separate substrains. When collecting the material precaution was made to select both eggs and pupae that were at the same age. In addition, the pupal material was divided into males and females. So, the eggs taken for analyses were 0 to 24 hr old and pupae were taken on the third day of the pupal stage duration. This was meant to make the material as comparable as possible since the energetic values of both the eggs and pupae diminish alsong the development process.

RESULTS AND DISCUSSION

The results on lipid contents in eggs and in pupae of Tribolium revealed a very high differentiation of this trait. In general, the lipid content in eggs was much lower, around 7 – 10% of dry weight, whereas that in pupal stage ranged from about 37 to 51% (Table I). Percentage of lipids in T. castaneum eggs was slightly but consistently lower than that in T. confusum. It was also lower in both the 6-instar groups than in 7-instar groups. Thus we know that T. confusum lays fatter eggs than T. castaneum and fatter are also eggs laid by females representing 7-instar groups than 6-instar groups.

The resources of lipids accumulated in pupae are rather of a crucial importance for the further life of adults. In Tribolium there is a rather long period of non-feeding stages (prepupae, pupae) amounting one fifth or one sixth of the whole developmental cycle, before the callow beetles start feeding again. High content of lipids in pupae is a convenient situation for seeking differences between the discerned groups. That is why we consider this stage as the most appropriate for comparison. Puapal of T. confusum show generally higher lipid content than those of T. castaneum (39-51% versus

37-39%, respectively). In the former species males show higher lipid percentage content than do females, whereas in T. castaneum the difference is irregular. The lipid content in the food is rather low and it amounts to 4.05% of dry weight (Table I).

Energy values (cal/mg dry wt) measured point to a very uniform energy value of eggs in two groups of both species. These values are rather low, lower by about 2 cal/mg dry wt than that of pupae. Energetic value of female and male pupae of T. confusum is higher than that of T.castaneum and the sex differences in the four compared pairs are rather small (Table II). The energy equivalent of the food is 4.12 + - 0.11 cal/mg dry wt.

On the basis of results listed above the reproductive effort was estimated using three different measures. Namely, the progeny-to-female ratios were calculated on the basis of (a) lipid contents in eggs laid daily by a female to live weight of reproducing female, (b) lipid content in daily egg production to lipid content in a pupal female, and (c) energy equivalent of both eggs produced and pupal female body (Table III).

All ratios are higher in T. castaneum than in T.confusum, pointing to a higher reproductive effort in the former species. If we compare 6- and 7-instar group is characterized by higher reproductive effort.

Ratios of lipid contents in eggs produced daily by a female to its fresh weight depict real situation since the weights of females were assessed on 72nd day of life and about 42nd day after mating, when reproduction took place. The other two indices involved both lipid content and energetic equivalents of females calculated from data of Table I and II, referring to pupal stage. Thus they did not reflect the real proportion of lipids and energy amount transferred to progeny by a female because these values in a reproducing female are much lower than that of pupal stage (Klekowski et al. 1967). Nevertheless, they are valid for estimation of trends observed between the groups and strains in order to characterize the differences between the compared pairs.

The results presented in this paper are in accordance with suggestions that were made elsewhere (Prus at al. 1988), concerning the life history strategy in Tribolium,

i.e. T.castaneum is more r-strategist than is T. confusum.

There is an opinion (e.g. Tinkle, Hadley 1975) that estimation of reproductive effort, as based on the total energy budget, is the best mean of characterizing the compared species or strains. However, the energy budget assessments differ so much one from another, depending on developmental cycle, type of feeding, reproduction and behavior, that even estimates based on total energy budgets can be illusive and not valid for comparison of more distant taxonomic units. For example in Tribolium, data on total reproduction during the whole life span are still missing or it is difficult to measure them. Such are also budget parameters. It seems, however, that for future comparison we should choose only instantaneous daily energy budgets of a reproducing female in the case of Tribolium.

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Table I -- Lipid contents in dry matter (%) of Tribolium

Stage T. castaneum cI T. confusum bIV

6-instar 7-instar 6-instar 7-instar

Egg 7.50 9.90 9.44 10.39

Pupae (males) 38.64 37.27 42.53 51.21

Pupae (females) 37.93 39.46 39.02 46.71

Lipid content in food (wheat flour + yeast) – 4.05%.

Table II - Energetic value (cal/mg dry wt.) in Tribolium

Stage T. castaneum cI T. confusum bIV

6-instar 7-instar 6-instar 7-instar

Egg 4.57 4.63 4.45 4.40

+- 0.25 +- 0.05 +- 0.25 +- 0.15

Pupae (males 6.54 6.47 6.68 6.65

+- 0.64 +- 0.15 +- 0.25 +- 0.23

Pupae (females) 6.34 6.50 6.57 6.61

+- 0.15 +- 0.17 +- 0.29 +- 0.12

Energetic value of food – 4.12 +- 0.11 cal/mg dry wt.

Table III - Calculation of reproductive effort indices in Tribolium

Stage T. castaneum cI T.confusum bIV

6-instar 7-instar 6-instar 7-instar

Lipids (ug) in:

Pupal female (1) 346.3 440.2 485.2 637.8

Egg (2) 1.61 1.97 3.28 3.59

Daily egg production by

A female (3) 32.20 33.40 38.11 34.95

Weight of female

(ug) (4) 1950 1300 2580 2830

Reproductive effort

(3) : (4) % 1.65 1.46 1.48 1.23

Reproductive effort

(3) : (1) % 9.30 7.59 7.85 5.48

Energetic equivalent

(cal) in:

Pupal female (5) 5.789 7.251 8.169 9.025

Egg 0.098 0.092 0.155 0.152

Eggs laid daily (6) 1.956 1.562 1.796 1.480

Reproductive effort

(6) : (5) % 33.79 21.54 21.99 16.40

\*EVALUATION OF BREAKFAST CEREALS TO SUPPORT THE GROWTH OF

RED FLOUR BEETLE (Tribolium castaneum)

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ABSTRACT

Sixteen proprietary breakfast cereals were tested for their ability to support the growth of red flour beetle (Tribolium castaneum) larvae. They were tested without or with the addition of either fat-free dried milk powder, or brewer’s yeast, or both at levels of 40% and 10% respectively. Without supplementation, only one of the 16 cereals supported as good larval growth as on unbleached white wheat flour control. The addition of yeast improved larval growth on all but a high bran cereal. Milk powder depressed larval growth on the control diet. Only 5 cereals were improved by the addition of milk powder. In a number of cases, milk powder interfered with the growth enhancing effects of yeast when both supplements were present. The detrimental effects of milk powder were not caused by lactose. Galactose but not Sucrose, glucose or fructose has any detrimental effect on larval growth. No significant correlation could be found between larval growth, and vitamin and mineral supplementation by the manufacturers, or protein content of the various processed cereals. The results suggest that the breakfast cereals are nutritionally deficient to support an optimal growth of T. castaneum larvae.

KEY WORDS: breakfast cereals, Tribolium larvae, nutritional evaluation

INTRODUCTION

Processed grains are the main constituents of the proprietary ready to eat breakfast cereals. They are usually supplemented with vitamins and minerals. The level of added sugar is highly variable. The nutritive value of the cereals may be altered during their processing. The use of larvae of the red flour beetle, species Tribolium, a serious pest of cereals and their by-products, has been suggested for nutritional evaluation of different varieties of cereals and their by-products (1-6). We have evaluated some proprietary breakfast cereals to support the growth of larvae of Tribolium castaneum.

MATERIALS AND METHODS

The breakfast cereals were purchased from a market and assigned a number from

1 to 16. They were ground to a fine powder in an electric coffee been grinder and sieved though a 100 mesh screen. The process has to be repeated a few times till the bulk of the material could be sieved. The residual material on the sieve which could not be ground any further was added back to the finely ground material. Four feeding Trials were carried out with these ground cereals. The experimental diets were as follows: Trial 1, 100% ground cereal; Trial 2, 60% ground cereal plus 40% instant non-fat dry milk powder (Carnation Brand); Trial 3, 90% ground cereal plus 10% dried brewer’s yeast (Sigma Chem. Co., St. Louis, MO); and Trial 4, 50% ground cereal plus 40% dried milk powder plus 10% yeast. The amount of milk powder corresponded to the suggested level on the package of 0.5 cup (122 ml) liquid milk added to one ounce (28.4 g) dry cereal. A control diet for each Trial contained unbleached, white wheat flour in place of ground cereal. Crude protein (CP) in the samples was determined by multiplying the Kjeldahl nitrogen with 5.7 for wheat products and 6.25 for other cereals, milk powder, and brewer’s yeast. On this basis, wheat flour, milk powder, and yeast contained 10.2%, 35.75%, and 50.8% crude protein, respectively.

T. castaneum larvae were hatched from a 24 hour collection of eggs from a primary stock maintained on a control diet consisting of 90% unbleached white wheat flour plus 10% brewer’s yeast. Brewer’s yeast provides protein, vitamins, and minerals, and a sterol required for the optimal growth of larvae (7). The larvae were kept in an incubator at 33 + 1 degree C and 70 + 5% relative humidity (4). After rearing the larvae for 6 days on the control diet, they were transferred to the test diets. Three replicates of ten larvae each were placed in loosely capped plastic vials (2.5 cm diameter x 5 cm height) each containing 2 to 3 g test diet. Larvae were incubated until the fourteenth day after hatching when each replicate was weighed after sieving out from the diets. An average larval weight was determined and the data were subjected to statistical evaluation using a two-way analysis of variance using each Trial as a block. Tukey’s multiple comparison test was used for pair-wise comparison of treatment means at P 0.05 (8). A squared regression coefficient between growth and protein level in the diet was calculated for each Trial.

The effect of lactose, galactose, sucrose, glucose and fructose on larval growth was also evaluated. Lactose and galactose were tested to determine whether

T. castaneum larvae are lactose intolerant, and galactose is produced in the gut after hydrolysis of lactose by the enzyme lactase. Sucrose and its component monosaccharides were tested because high levels of sucrose are often added to processed cereals. Diets consisted of lactose, galactose, or sucrose at levels of 0.0, 1.5, 5.0, 10.0, 20.0, 50.0, or 75.0%; brewer’s yeast, 10%; and the balance of unbleached white, white wheat flour. As glucose and fructose are hygroscopic, they were evaluated separately with 0.5% Celite (Sigma Chem. Co., St. Louis, MO) added at the expense of flour to prevent stickiness of the diet in the humid environment of the incubator. The levels used to evaluate these two sugars were 0.0, 1.0, 2.0, 5.0, 10.0, and 30.0%. The feeding procedure was the same as that described for the cereal Trials. Fourteen-day larval weights were tested for statistical significance using analysis of variance and Tukey’s test (P 0.05).

RESULTS AND DISCUSSION

The sugar content of 15 of the cereals as given by Li and Schumann (9), the determined CP content, and the average weight of larvae fed cereals without any supplement are given in Table 1. The crude protein content of breakfast cereals without any supplementations varied from 3.8 to 17.3%. The average weights of larvae fed cereals 1 though 14 were not significantly different, but wee significantly less than of those fed cereals 15 and 16. The average weight of the larvae fed cereal 16 was not significantly different from that on the control diet based on wheat flour. It was the only cereal that provided the necessary nutrients for optimal larval growth.

Several investigators have reported that dietary protein concentrations influence growth and viability of Tribolium larvae (10-11). Medrano and Bressani (11) found that a minimum level of 8% protein from wheat flour and torula yeast was needed for optimum larval growth. In the present study, cereals No.12, 13, 14 and 15 with protein contents of higher than 8% did not support optimal growth of larvae. Cereal 13 had the highest protein content (17.3%), but failed to support the optimal growth. Poor correlation (r 2 = 0.26) was found between protein level of the cereals and the larval growth in this Trial, or in any of the other three (Table 1). This may be due to poor digestibility of the protein from the processed cereals.

Tribolium larvae appear to utilize dry fat-free milk powder poorly. The growth of larvae on the control diet containing 60% wheat flour plus 40% milk powder (Trial 2) was less than that on wheat flour alone in Trial 1 (Table 1). The addition of milk to cereals 3, 5, 10, 11, and 13, however, did improve larval growth (P 0,05). The depression in growth of larvae fed cereals 15 and 16 supplemented with milk powder was not statistically significant. The CP content of these diets varied from 16.5 to 24.5%.

The addition of yeast, a provider of essential growth factors, to test cereals was generally effective in enhacing larval growth in Trial 3 (Table 1). The maximum improvement with yeast occurred for cereal 1. The larval growth on test cereals 1, 4, 8, 9, 11, 14, 15 and 16 supplemented with yeast was not significantly different from that on the white flour plus yeast control. Calculated protein level of yeast supplemented diets varied from 8.4% to 20.5%, a level satisfying the recommendation by Medrano and Bressani (11). The addition of yeast improved larval growth for all the cereals, but growth was not correlated to protein level

(r 2 = 0.05). Cereal 16 based on oats supported the maximum growth of the larvae with or without yeast. Without yeast, all wheat-based cereals supported the larval growth poorly, but after supplementation with yeast, the larval growth o wheat-based cereals 8, 10, 11 and 14 was not significantly different from that on the control diet. The lack of response on wheat-based cereal 12 may be related to its high bran content.

T. castaneum larvae require essential amino acids, vitamins of the B-complex for growth and pupation, but requirements for other vitamins and minerals have not been studied thoroughly (8, 12, 13, 14). Riboflavin, niacin, and pantothenic acid are vital for growth. An absence of folic acid, pyridoxine and biotin from the diet inhibits growth and extends the larval period. Of less importance are inositol, choline and thiamin. Our present study shows that breakfast cereals supplemented with vitamins and minerals by the manufacturers did not seem to support optimal larval growth. Cereal 15 was the only one with no listed nutrient supplements on the label. Only niacin, thiamin, riboflavin and iron were added to the cereal 16. These two cereals, when fed without milk or yeast, were far better than other cereals in supporting larval growth, but only the larval growth on cereal 16 was not significantly different from that on the control diet. All the other test cereals had supplements of niacin, pyridoxine, thiamin, riboflavin, and some or all of the following: vitamin A, vitamin D, vitamin C, folic acid, vitamin B 12, pantothenic acid, zinc and iron. Our data indicate that besides these nutrients, some other factors are responsible for the growth-enhancing properties of brewer’s yeast for Tribolium larvae. The addition of milk powder to cereals 1, 2, 6, 8, 9, 10 and 16 supplemented with yeast depressed larval growth significantly (Trial 4, Table 1). This suggests an interaction between yeast and dried milk for larvae. The level and source of protein in the supplemented cereal diets did not appear to influence the pattern of larval growth.

Some earlier studies (15, 16, and 17) indicated that growth of Tribolium larvae can be affected by the carbonhydrate source in the diet. Wheat, rice and corn (maize) starch all promote growth, but starches with a high amylopectin content were better than those with a high amylose content.. The exact nature of the carbohydrate complex in the cereals used in the present Trials is not known, but oats, which generally have an amylopectin content similar to that of wheat or corn (18), were the major grain in cereals 15 and 16 which supported the greatest larval growth. These two cereals also have several other non-cereal ingredients other than sugars. Cereal 15 contains oats, wheat, coconuts and almonds, while cereal 16 is based in oats, soy, corn and casein. With only these exceptions, the other cereals had one or two main constituents, most were dereived from wheat, corn or rice.

The data on the role of various levels of dietary lactose (a major component of dried milk powder), galactose and of sucrose in supporting growth of Tribolium larvae are given in Table 2. A dietary lactose level of 75% was required to depress larval growth significantly, but the protein content of this diet was less than 8%. Since such a high level of lactose was required to depress larval growth, they could not be lactose intolerant. Galactose appeared to be highly toxic when it comprised more than 20% of the diet. If the enzyme lactase split dietary lactose to galactose and glucose in the gut of Tribolium larvae, a depression in growth would have been expected at a lactose level of 50%. The growth depression of larvae could not be due to the lactose content of the milk powder, but might be caused by some other toxicant component produced by heat during preparation of milk powder, possibly a protein carbohydrate complex.

Table 2 – Mean weights of Tribolium larvae fed increasing amounts of

Lactose, galactose, or sucrose

Larval weight (mg) on diet Containing

Sugar (%) Dietary protein (%) Lactose Galactose Sucrose

0 Control 14.3 2.94 2.99 2.70

1.5 14.1 2.78 2.91 3.11

5.0 13.8 2.47 2.84 2.78

10.0 13.2 2.73 2.88 2.62

20.0 12.2 2.67 1.61 2.78

50.0 9.2 2.45 0.63 2.88

75.0 6.6 2.05 0.73 2.25

Analysis of Variance

Source Sum of squares df Mean squares F Significance

P 0.05

Sugar level 12.48 6 2.08 35.68 \*

Sugar type 4.80 2 2.40 41.15 \*

Level x type 10.90 12 0.91 15.59 \*

Error 2.45 42 0.058

Previous studies have shown that high dietary levels of sucrose did not promote optimal growth of T. castaneum larvae (19) although a closely related species, Tribolium confusum, could utilize this sugar more efficiently (20). We could not show any significant growth depression by dietary sucrose until the level reached 75% and of dietary protein decreased to 6.6% (Table 2). Sucrose is not deleterious to Tribolium larvae and our data does not support the findings of Pant and Gabrani (19).

Fructose and glucose, in presence of Celite to prevent them from becoming sticky, are utilized equally well at the levels tested up to 30% of the diet, and support growth of larvae comparable to that on the control diet (Table 3). Some highly sweetened cereals such as cereal 7 tested in Trials 1 to 4 were hygroscopic. In absence of Celite, their hygroscopic nature may have contributed to poor growth by preventing larvae from tunneling through the diet easily. But this hygroscopic nature after opening the package does increase the shelf life of the proprietary breakfast cereals because it inhibits growth of larvae.

Table 3 - Mean weights of Tribolium larvae fed increasing levels of Fructose or glucose

Larval weight (mg) on diet

Sugar level (%)

Fructose Glucose

0 control 2.71 2.71

1 2.89 2.99

2 2.76 2.81

5 3.01 2.78

10 2.79 2.84

20 2.67 2.76

30 2.81 2.93

No significant difference at P 0.05.

Are the data derived from larval growth of any significance for the evaluation of cereals for humans? Each species of animals is unique and the data based from one species is not strictly applicable to another species even though the information developed by human nutritionists is based on the use of rats as experimental animals. Kies and Fox (21) measured nitrogen-balance in human adult males fed cereals which provided essentially all of the protein and 66% of the calories in the diet. They observed highest nitrogen retention on the cereals made from oats, especially those supplemented with other protein sources such as soy and casein (cereal 16). Differences in protein level and in digestibility of protein were responsible for the observed differences in nitrogen-balance. Cereals which supported the optimal growth of Tribolium larvae in our study had also induced positive N-balance in men and vice versa in studies of Kies and Fox (21).

Hackler (22) also observed that oat-based cereals, particularly those with added soy and milk products, gave higher protein efficiency ratios (PER) than wheat-based cereals when rats were fed 9.7% CP from cereals. He also found that PER improved in most cereals when case in was added. Womak et al. (23) determined PER in rats fed diets at fixed protein levels with cereal and dry milk mixtures containing 28.4 g cereal and 153.6 ml fluid milk equivalent. They found that milk protein complemented the cereal proteins in some, but not all instances.

It can be concluded that all but two of the breakfast cereals were nutritionally inadequate to support the optimal growth of Tribolium larvae.

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\*Reduced gena (rg), an interesting mutation in Tribolium brevicornis Lec. (Coleoptera: Tenebrionidae).

Tenebrionidae, the fifth largest family of beetles are a varied group, but they are identifiable by having a 5-5-4 tarsal formula, the front coxal cavities closed behind, the eyes usually notched, the antennae nearly always 11- segmented and either filiform or moniliform, and the abdomen consisting of five visible sterna (Borror, DeLong and Triplehorn, 1981).

Tribolium conforms to this set of characteristics. Hinton (1948) was able to work out a key for classifying the species with the genus by a combination of antennal, eye, leg and other morphological characteristics. With the similarities and differences in these traits, he was able to group the 30-plus known species into species-groups.

It is not the author’s intention to reproduce Hinton’s key (which has been reproduced in Sokoloff, 1972), but it is worthwhile to point out to what extent eye characteristics play

a role in placing a beetle of unknown species in placing it in one or another species group, and to emphasize that these characteristics are of great evolutionary importance.

The compound eye in the adult Tribolium may differ in the extent to which the dorsal portion or lobe of the eye extends over the total length of the head; the number of ommatidia or number of rows of facets forming a bridge between the dorsal and ventral lobes of the compound eye; and the distance involved in separating the ventral lobe of the left eye from the lobe of the right eye (usually expressed in terms of so many eye widths).

Species in the brevicornis species group of Tribolium are set aside from other species-groups by having eyes which are completely divided into a ventral and a dorsal lobe. There are no ommatidia to serve as a bridge between the ventral and the dorsal lobes. The separation of the ommatidia into a dorsal and ventral lobe is brought about by

A shelf-like structure which Hinton calls the “side of the front” and Sokoloff (1972) calls the gena. For the sake of brevity I will use gena to denote this structure.

Other species-groups have a gap between the posterior portion of the gena and the parietal portion of the eye, and this gap is filled by several ommatidia or rows of ommatidia. Thus, species in the confusum species group have a compound eye in which the dorsal and ventral lobes are united by a lateral portion, the eye consisting of a single ommatidium (in T. confusum), or at most by two or three ommatidia or facets

(in T. destructor or T. anaphe). Species in the castaneum group have eyes which on the ventrum of the head, are separated by one eye-width. The narrowest part of the eye, in front of the gena, may have as few as two facets (in T. freeman) to as many as 6-7 facets (in T. castaneum).

Previously I have described several mutations in T. castaneum which affect the mosrphology of the eye and/or the shape of the head:

Microphthalmic (Mo) an autosomal dominant with recessive lethal effects and microcephalic (mc), and autosomal recessive, modify the size of the head behind the gena and result in a reduction of the compound eye. Mo reduces the head behind the gena more markedly tan mc, but mc can have a more drastic effect on the eye, sometimes eliminating the compound eyes completely on both sides of the head with the result that the beetles are blind. Neither Mo nor mc affect the gena or the anterior parts of the head.

Another autosomal dominant with recessive lethal effects is Bar eye (Be). It eliminates all of the ventral and lateral facets of the compound eye exposing a portion of the ocular diaphragm (an endoskeletal structure which serves to support the ommatidia). A few dorsal facets remain, giving the eye a bar-like appearance. A more severe effect is produced by squint (sq), an autosomal recessive. In sq, although the ocular diaphragm forms, all the ommatidia are eliminated and the beetles are blind. Neither Be nor sq affect the front parts of the head, nor do they seem to affect the size of the cranium (Sokoloff, 1966).

A more recently discovered autosomal recessive mutant in T. confusum, diminished eye (de), also appears to reduce the number of facets without affecting the size of the cranium: de has good viability but incomplete penetrance. The eye is mis-shapen to

a varying degree, ranging from a smooth faceted narrow area (similar to Be in T.castaneum) to an eye of normal appearance but slightly reduced in size (Blackman, 1982).

All of the above mutations fall in one of two categories: (i) The eye itself is affected, but the mutant gene has no effect on the head structures, or (ii) The eye is reduced or eleiminated completely, and there is a variable reduction of the cranium, but the head structures anterior to the compound eye are not affected.

The rg mutant was found accidentally. It occurred spontaneously in some crosses trying to elucidate the nature of a body size variation of descendants of beetles found in a natural population of T.brevicornis in Waterman Canyon, north of San Bernardino.

The rg mutant affects the morphology of the eye in a different way. As mentioned above, in normal Tribolium brevicornis the compound eye is separated into two discrete lobes by the gena. The gena extends posteriorly to the parietal portion of the cranium, eliminating the possibility of formation of the ommatidia wherever it forms (Fig.1, A, B, C). In the rg mutant, (Figs. 1. C,D,E), the gena is reduced to a variable degree even within the same individual. Sometimes the gena fails to reach the parietal portion of the cranium leaving a narrow gap between itself and the parietal portion of the cranium, a gap wide enough so that one ommatidium forms in that space. The net result is an eye similar to that in T. confusum, with the dorsal and ventral lobes connected by a single ommatidium. At the other extreme, the gap between the posterior portion of the gena and the parietal portion of the cranium may be sizable, so that as many as 6 or 7 rows of ommatidia form within the gap. In such a case, the compound eye may resemble the compound eye of T. castaneum. The full range of expression of the mutant is yet to be observed, but it is clear from preliminary observations that the extent to which the gena forms is the determining factor whether the compound eye in Tribolium brevicornis will consist of two discrete lobes, one dorsal and the other ventral, or a compound eye in which one or many ommatidia will interconnect the dorsal and ventral lobes.

The rg gene behaves as a phenodeviant: by selecting rg beetles each generation it is possible to increase the frequency of beetles that manifest the mutant phenotype. However, on outcrossing, the number of mutants in F2 may be variable, but small and significantly different from the 3:1 ratio expected for

an autosomal trait.

Nevertheless, the rg mutant is a useful one, because it points to the importance of the gena in determining the final morphology of the eye, not only in T.brevicornis, but other species as well.

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Fig.1. A,B,C, dorsal, lateral and fronto-ventral view of normal T.brevicornis. D, E, F, dorsal, lateral and ventral view of reduced gena (rg) beetles. The gena is located in front of the eye and is crosshatched.

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\*EFFECTS OF LOCALIZED REGIONS OF HIGH MOISTURE GRAIN ON

EFFICIENCY OF INSECT TRAPS CAPTURING ADULT TRIBOLIUM CASTANEUM

AND CRYPTOLESTES FERRUGINEUS IN STORED WHEAT

Introduction

Many species of stored-product insects are attractd to or aggregate in areas of high moisture grain (Watters 1969) especially the rusty grain beetle Cryptolestes ferrugineus (Loschiavo 1983) which exhibits a klinokinetic response to humidity (Surtees 1965) as opposed to a positive geotaxic response in grain of a uniform moisture content (Loschiavo 1975). The movement of C. ferrugineus is also affected by fungi in stored grain (Dolinski and Loschiavo 1973) and most stored product beetles breed and develop when feeding on fungi (Sinha 1971).

Insect detection traps have been developed for use in stored grain (Loschiavo 1973, 1974) but since stored-product insects tend to aggregate in areas of grain with high moisture content this behavior could affect the efficiency of the traps. There might be few insects in the traps but many in pockets of wet grain or, if the traps were placed in a area of wet grain, a disproportionately large number of insects in the traps.

Materials and Methods

The experimental apparatus described by White and Loschiavo (1986) was used. Nine small galvanized steel bins were each filled with wa8 kg of hard red spring wheat at 13% moisture content. Each bin had 1070 adult T. castaneum and 1070 adult C. ferrugineus added and a 6-section insect trap (15 cm long, 2.6 cm diam.) inerted vertically through the center of the grain bulk until the bottom extended 10 cm through a hole in the floor. Plastic mesh bags containin absorbent cotton soaked with water and 50 g of fresh wheat at 24% MC or 50 g of rotten wheat (moldy with bacterial deterioration) at 26% MC were singly placed in drums 20 cm below the grain surface near the central insect traps and bags were left at ambient conditions ranging from 17 to 31 degree C and were checked once a week for 6 weeks for insects in the bags and traps. All captured insects were returned to the grain bins and new grain was added to the bags. During each week 3 drums served as controls, 3 as replicates of one treatment and 3 as replicates of another. The treatments are outlined in Table 1 and consisted of bags of wet grain near the traps or at the bin wall, wet grain and moldy grain in the bags, and wet or moldy grain beside 500 caged adult insects of each species.

RESULTS AND DISCUSSION

Most T. castaneum adults were caught in traps at the top surface of the grain bulk while most C. ferrugineus adults were caught at the bottom. Negligible numbers of T. castaneum were attracted to the bags of wheat in any treatment regardless of temperature, although there were a few more in moldy grain.

C. ferrugineus numbers were appreciably higher than in the controls in all bags of wet or moldy grain but were largest in moldy grain beside the traps with caged insects at 21 degree C, wet grain beside the traps at 29 degree C, moldy grain beside the traps with caged insects at 31 degree C.

The bags of wet or moldy grain did not affect trap catches of T. castaneum, even at the same grain depth. However, wet and moldy pockets of wheat did result in larger numbers of C.ferrugineus being caught at the 12-24 cm depth in the traps. This effect was less evident at 31 degree C when caged insects were present.

The presence of localized areas of wet or rotten grain does not appear to affect the capture of T.castaneum in insect traps but C.ferrugineus do aggregate in these regions which has a moderate effect on insect capture.

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