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RELATIONSHIP BETWEEN TIME OF EMBRYONIC AND POSTEMBRYONIC DEVELOPMENT AND WEIGHT IN TWO SPECIES OF *TRIBOLIUM* BEETLES

ABSTRACT: Time of embryonic (T_e), larval (T_l), and whole postembryonic (T_p) development as well as body weight at pupal stage have been examined in *Tribolium castaneum* Hbst and *T. confusum* Duval. T_e is strongly correlated with T_l as well as with T_p. The 6- and 7-instar individuals differing in respect of T_l and weight, show however no evident difference in T_e so that the correlation seems to be independent of the instar-diversity in populations. No dependence between T_e and weight of pupae has been observed. There is a tendency towards positive correlation between T_l and weight in whole populations, however both 6-instar and 7-instar groups of individuals, considered separately, show a tendency towards negative correlation (R).

KEY WORDS: *Tribolium castaneum, Tribolium confusum*, time of development, body weight, intrapopulation differentiation, trade-off.

1. INTRODUCTION

In studies on bioenergetics and population biology of *Tribolium* beetles (P r u s 1976 and B i j o k 1986), and other store-product insects (H o w e 1961), it was found that individuals differ strongly in respect of the number of larval instars, development time and body weight they attain. Two clearly different groups can be distinguished within populations: the so called 6-instar individuals, which develop faster but have lower weight, and 7-instar ones which develop longer but their bodies become heavier.

Further investigations concerned with analysing differences between 6- and 7-instar-individuals in such features as length of the period of subsequent stages, fecundity, weight and hatchability of eggs (T. P r u s and M. P r u s 1987, P r u s et al. 1989), the reproductive effort (P r u s et al. 1988). Heterogeneity can be explained as the way of adapting the species to a changeable environment and can be described in terms of life-history theory (I m u r a 1990, S i b l y and C a l o w 1989, M ø l l e r et al. 1989a)

When determining the 6-instar to 7-instar individuals ratio in *Tribolium* populations, it was found that this value varies considerably in time (T. P r u s and M. P r u s 1990). It might suggest a methodical error which could be made during preparation of the material for development assays and for distinguishing between 6- and 7-instar individuals (B i j o k 1989). We could make an unintended preselection of animals choosing larvae hatching earlier or later from the batch of eggs. If the instar-group-diversity affects the time of embryonic development such preselection can distort the real proportions between the two groups in population.

The aim of present paper is to find out if the time of embryonic development of an individual relates with its further growth and development and the fact it belongs to the 6- or 7-instar group.

2. MATERIAL AND METHODS

Two strains of two species: cI – *T. castaneum* and bIV – *T. confusum*, both from the group of genetic strains, reared at the Chicago University (P a r k et al. 1961), were used in these investigations.

All experiments were carried out in standard conditions at 29°C, 75% relative humidity, in culture medium consisting of 95% of wheat flour and 5% of baker's yeast by weight.

Groups of 200–300 adult individuals were placed in glass jars containing about 100–150 g of culture medium previously kept in incubator to reach adequate temperature and humidity. After 3 hours of egg laying the animals were replaced by sieving jar content through a coarse mesh. Eggs were separated from the medium by sifting through fine mesh and incubated in glass containers till young larvae started to hatch. Newly hatched larvae were collected every 3 hours and placed into numerated glass vials, each containing 1 g of culture medium. For each individual the time of hatching was recorded and the period of embryonic development (T_e) was calculated. The larvae were collected over the night until the last larva was hatched. Vials were placed in the incubator and left for 15 days.

The following procedure was an observation of final period of development in synchronised individual cultures (B i j o k 1989): Starting from the 15-th day of larval development the content of each vial were sifted through fine mesh, individuals were found and the exuviae were counted. When a pupation took place its time was

recorded and the time of larval development (T_l) was calculated. Newly emerged pupa was weighed on CAHN electrobalance and its sex was determined according to methods described by S o k o l o f f (1972). Then the animal and medium were put back into the vial. Such observations were continued until adult individual emerged (T_p).

The total of 81 *T. castaneum* individuals and 66 *T. confusum* individuals was examined. Distinction between 6- and 7-instar group individuals was made on the basis of weight, time of pupae emergence and the number of found exuviae (B i j o k 1989).

The relationship between measured values in examined species and groups was checked by an analysis of correlation and regression in linear model ($Y = a + bX$). Significance of differences between mean values was analysed by student t-test.

3. RESULTS AND DISCUSSION

A strong correlation between embryonic (T_e) and larval (T_l) development times in both *Tribolium* species was found (Fig. 1). This correlation is a bit stronger in *T. confusum* (R = 0.55) than in *T. castaneum* (R = 0.43) (Table 1). In both cases R-coefficient is significantly different from zero at the confidence level p <0.01. The situation is almost identical when analysing the dependence of total postembryonic (T_p) on embryonic (T_e) development time. This is quite obvious because T_p and T_l

Table 1. Comparison of correlation coeficient of larval development time (T_l) on embryonic development time (T_e) in two *Tribolium* species (numbers of replications, significance of R-difference from zero, and confidence level marked)

Compared elements	*Tribolium castaneum*			*Tribolium confusum*		
	whole populat.	6-instar indiv.	7-instar indiv.	whole populat.	6-instar indiv.	7-instar indiv.
Average	R = 0.43	R = 0.69	R = 0.23	R = 0.55	R = 0.68	R = 0.46
	n = 81	n = 27	n = 45	n = 66	n = 32	n = 31
	p<0.001	p<0.001	*N.S.*	p<0.001	p<0.001	p<0.01
Males	R = 0.28	R = 0.21	R = 0.20	R = 0.50	R = 0.60	R = 0.23
	n = 44	n = 17	n = 25	n = 31	n = 22	n = 8
	p<0.1	p<0.1	*N.S.*	p<0.005	p<0.005	*N.S.*
Females	R = 0.54	R = 0.88	R = 0.28	R = 0.68	R = 0.67	R = 0.50
	n = 37	n = 10	n = 20	n = 35	n = 10	n = 23
	p<0.001	p<0.001	*N.S.*	p<0.001	p<0.05	p<0.02

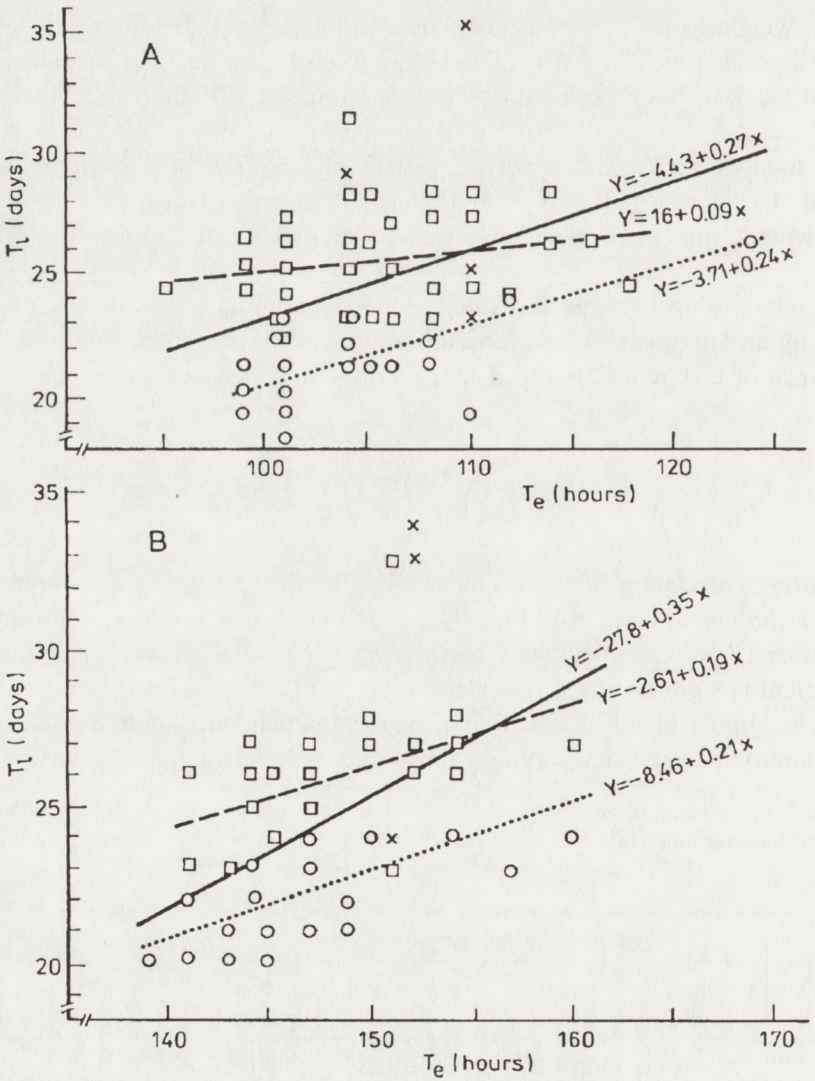

Fig. 1. Dependence of larval development time (T_l) on embryonic development time (T_e) in *Tribolium castaneum* – (A), and *T. confusum* – (B)
Circles – 6-instar-individuals, squares – 7-instar-individuals, x-marks – undetermined individuals, continuous line – regression line for whole population, dotted line – regression line for 6-instar individuals, dash line – regression line for 7-instar individuals

are values differing from one another by the duration of pupal stage which is ca 6 days and differs only slightly for each species. Therefore, further in the paper only larval development time (T_l) will be discussed as a value characterising more precisely the period of intensive growth of these animals.

One could expect that such correlation between T_e and T_l values is due to existing in populations 6- and 7-instar groups which differ from one another in the

time of larval and embryonic development. These two groups of individuals would be shown on the T_e/T_l scale (Fig. 1) by two clouds of points which can partly overlap but their centres would be pushed one from another along the regression line. One could expect then, that the 6-instar larvae would hatch earlier than the 7-instar ones. Empirical data, however, do not support this supposition. As in *T. confusum*, we may observe a tendency in the 6-instar group towards shortened embryonic development (Fig. 2) and mean T_e-values show a small difference which is still significant at a confidence level $p<0.05$. In *T. castaneum* there is no difference between mean T_e-values, and frequency histograms for 6- and 7-instar individuals are almost identical. Clouds are pushed along a vertical rather than a regression line which can not produce any correlation.

When analysing the dependence of T_l on T_e within two instar-groups of both species (Fig. 1) it has been found that these values are also correlated. So the correlation between these values seems to be independent of instar-group existence in populations of these species. The regression coefficient is higher in 6-instar groups: $R = 0.69$ and $R = 0.68$ than in 7-instar groups: $R = 0.23$ and $R = 0.46$ for *T. castaneum* and *T. confusum*, respectively. Females show stronger correlation between these values: $R = 0.50$ and $R = 0.68$ than males: $R = 0.28$ and $R = 0.50$ for *T. castaneum* and *T. confusum*, respectively.

The relationship between time of embryonic development and biomass of body built up till the end of larval growth (means weight of pupae) should be analysed separately for males and females because of considerable difference in the weight of pupae of different sexes. When analysing the weight/T_e dependence (Table 2) a weak correlation was found, in most cases insignificant. It is, however, interesting that generally R-coefficient is smaller than zero which suggests a tendency towards inversely proportional relation of these values. It means: the longer the egg develops the smaller is the weight of pupa which, eventually originates from this egg. This tendency is always observed when analysing both sexes and when the material is divided into both instar-groups.

Table 2. Comparison of correlation coeficient of pupal weight on embryonic development time (T_e) in two *Tribolium* species (significance of R-difference from zero, and confidence level marked)

Compared elements	*Tribolium castaneum*			*Tribolium confusum*		
	all indiv.	6-instar indiv.	7-instar indiv.	all indiv.	6-instar indiv.	7-instar indiv.
Males	$R = +0.01$	$R = -0.57$	$R = -0.13$	$R = -0.02$	$R = -0.25$	$R = -0.36$
	N.S.	$p<0.02$	*N.S.*	*N.S.*	N.S.	N.S.
Females	$R = -0.38$	$R = -0.20$	$R = -0.60$	$R = -0.16$	$R = -0.19$	$R = -0.48$
	$p<0.02$	*N.S.*	$p<0.005$	*N.S.*	N.S.	$p<0.05$

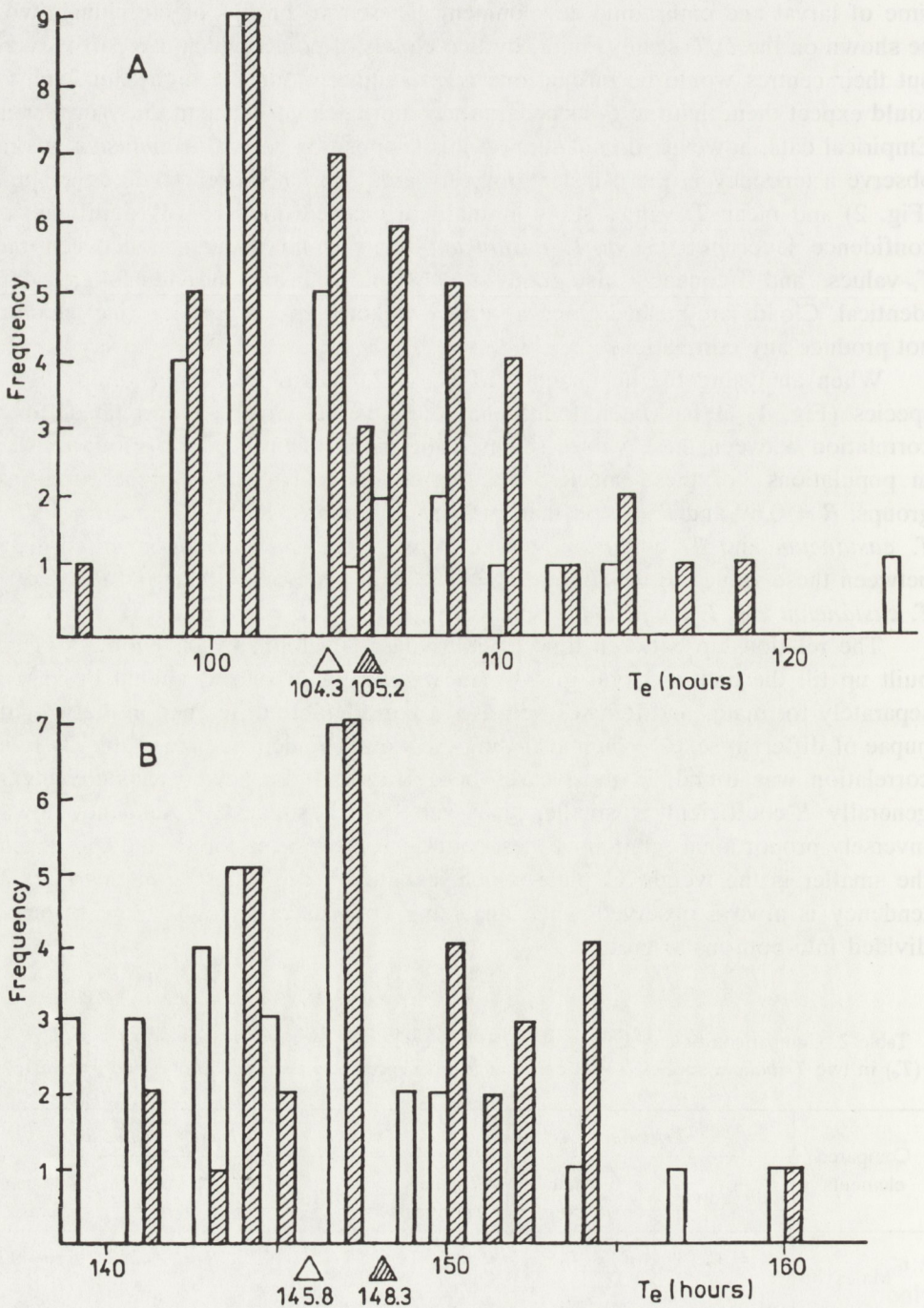

Fig. 2. Frequency histograms – numbers of eggs hatched after T_e (hours) in *Tribolium castaneum* – (A) and *T. confusum* – (B)
Dotted bars – 6-instar-individuals, striped bars – 7-instar-individuals, triangles – mean time of embryonic development

This phenomenon stimulates the analysis of relationship between the time of larval development (T_l) and weight of pupa. A separate approach to two sexes of both species shows a strong positive correlation – except for *T. castaneum* females – (Table 3). However, the situation was quite different, when the material was arranged into 6- and 7-instar groups. In that case there was a general tendency towards a negative correlation ($R < 0$).

Table 3. Comparison of correlation coeficient of pupal weight on larval development time (T_l) in two *Tribolium* species (significance of *R*-difference from zero, and confidence level marked)

Compared elements	*Tribolium castaneum*			*Tribolium confusum*		
	all indiv.	6-instar indiv.	7-instar indiv.	all indiv.	6-instar indiv.	7-instar indiv.
Males	$R = +0.30$	$R = -0.45$	$R = -0.80$	$R = +0.69$	$R = -0.01$	$R = +0.38$
	$p<0.05$	$p<0.1$	$p<0.001$	$p<0.001$	N.S.	N.S.
Females	$R = -0.20$	$R = -0.41$	$R = -0.51$	$R = +0.50$	$R = -0.53$	$R = -0.38$
	N.S.	N.S.	$p<0.02$	$p<0.005$	N.S.	$p<0.1$

The results could be explained as follows: the 6- and 7- instar groups, with individuals which differ in body weight and time of development are shown on the graph (Fig. 3 and 4) as two separate clouds of points. (This is applied when determining these groups – B i j o k 1989). A distance between these clouds produces a positive correlation. We have in this case a trade-off between components of the fitness: a development rate which affects the population growth rate and body weight which may affect such features as fecundity longevity (M ø l l e r et al. 1989a). An example of such trade-off is described in *Callosobruchus maculatus* (F.) by M ø l l e r et al. (1989b). The existence of two such groups improves the fitness of population in a changeable environment.

Each of the instar groups, considered separately, generally shows a reverse correlation. Probably this is because of the genotype variability – existence in these groups of stronger and weaker genotypes. These which develop fast and also attain a high body weight are placed at the top left hand side of particular cloud of points. These that are not successful in this respect are placed at the bottom right hand side and are probably the successively born individuals of the population. T. Prus (unpublished data), when investigating the development of many *Tribolium* strains and mutants, found in most cases the not numerous individuals with a prolonged development and relatively low body weight (points producing a tail to the cloud of points on T_l/weight scale). These poorly developing individuals are supposed to be eliminated from the population and the reason for their appearance requires further investigations.

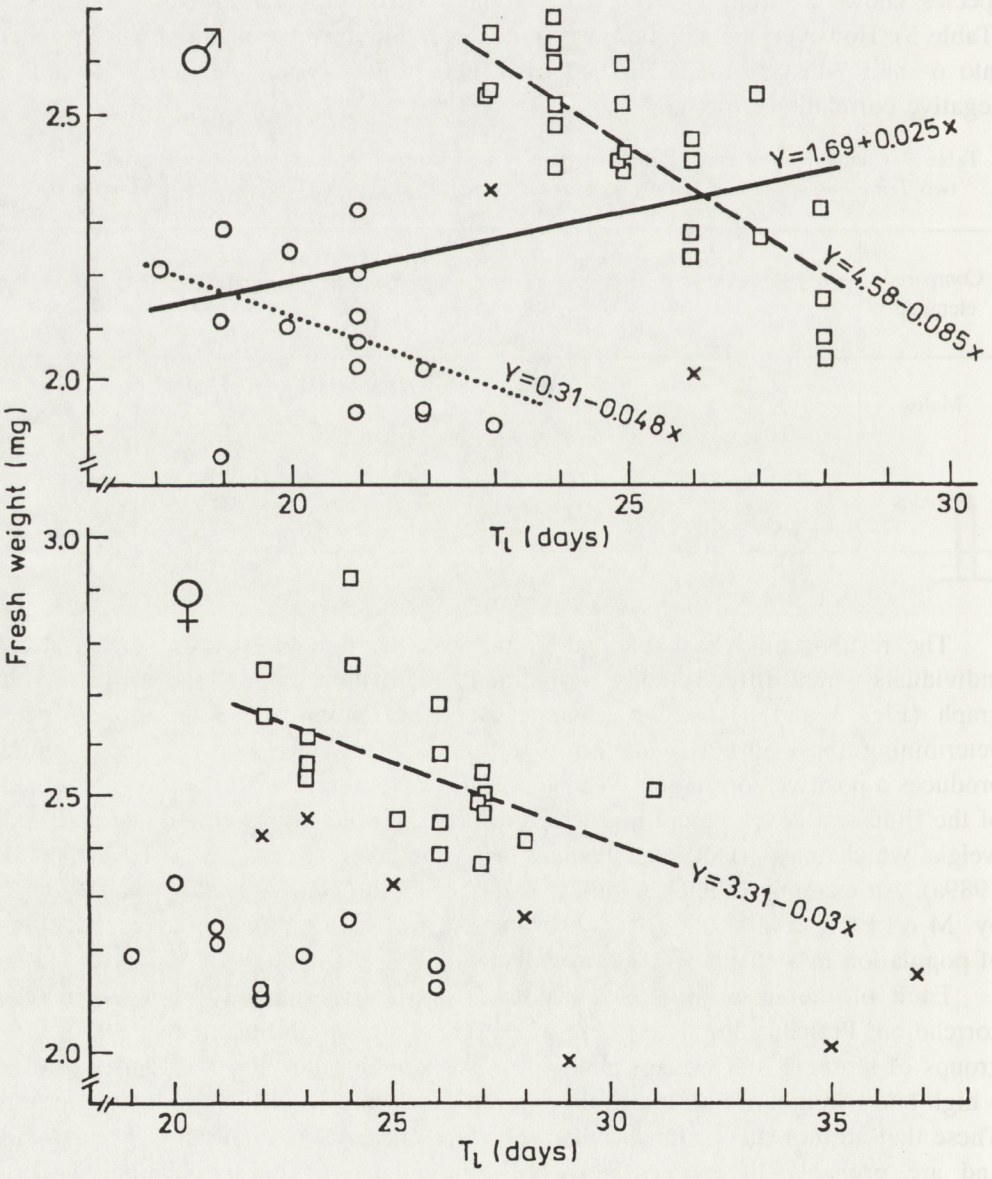

Fig. 3. Dependence of pupal fresh weight on time of larval development (*Tl*) in *Tribolium castaneum* Circles – 6-instar-individuals, squares – 7-instar-individuals, x-marks – undetermined individuals, continuous line – regression line for all individuals, dotted line – regression line for 6-instar individuals, dash line – regression line for 7-instar individuals

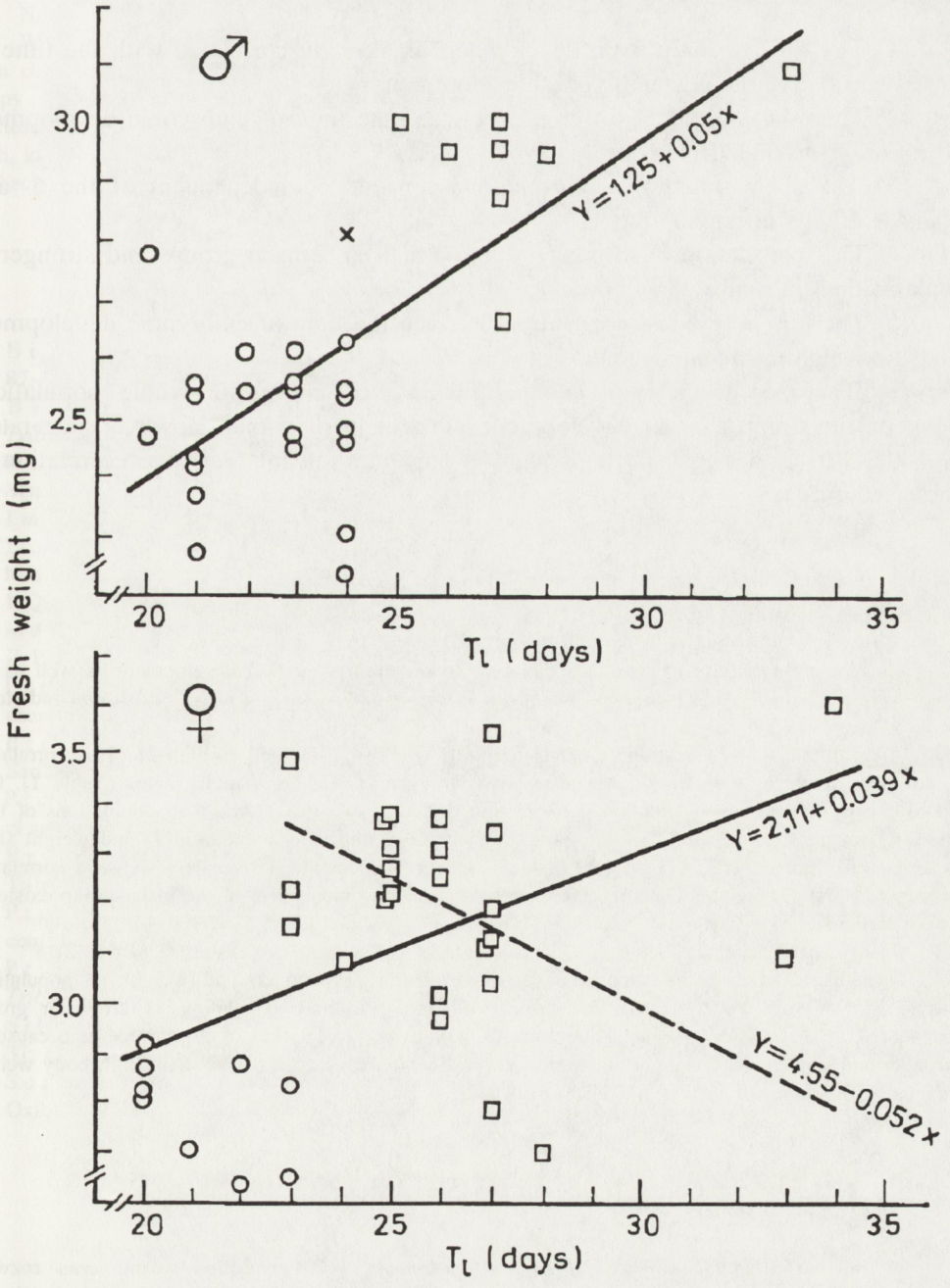

Fig. 4. Dependence of pupal fresh weight on time of larval development (T_l) in *Tribolium confusum*
Circles – 6-instar-individuals, squares – 7-instar-individuals, x-marks – undetermined individuals, continuous line – regression line for all individuals, dash line – regression line for 7-instar individuals

4. CONCLUSIONS

(1) The time of embryonic development is strongly correlated with the time of larval and the whole postembryonic development.

(2) There is no evident difference between the time of embryonic development of 6- and 7-instar-individuals.

(3) The above mentioned correlation seems to be independent of the 6- and 7-instar groups in populations.

(4) This correlation is stronger in the 6- than in 7-instar groups and stronger in females than in males.

(5) There is no evident correlation between the time of embryonic development and the weight at pupal stage.

(6) There is a tendency towards positive correlation in whole populations between the time of larval development and weight of pupae. However, separately considered 6- and 7-instar groups show a tendency towards negative correlation of these features.

5. SUMMARY

Time of embryonic (T_e), larval (T_l), and whole postembryonic (T_p) development as well as the body weight at pupal stage have been examined in *Tribolium castaneum* and *T. confusum* individual cultures.

It was found that T_e is strongly correlated with T_l (Fig. 1), as well as with T_p. This correlation is stronger in the 6- than in 7-instar group and stronger in females than in males (Table 1). One could expect that the above mentioned correlation is due to two instar groups in populations of both species. However, the 6- and 7-instar groups which differ one from another in T_l and weight show no evident difference in T_e (Fig. 2). Moreover, each group considered separately shows a correlation between T_e and T_l (Fig. 1). Thus, the correlation is probably independent of the instar-group existence in populations.

No evident dependence between T_e and weight of pupae has been observed (Table 2).

There is a tendency towards a positive correlation between T_l and weight in populations (Table 3), which is an effect of the existence of instar group in populations. Both instar groups considered separately show a tendency towards a negative correlation ($R<0$), probably because of stronger and weaker genotypes in these groups: these which develop fast and attain high body weight, and those that can not succeed in this respect (Fig. 3 and 4).

6. POLISH SUMMARY

W indywidualnych hodowlach *Tribolium castaneum* i *T. confusum* badano czas rozwoju embrionalnego (T_e), larwalnego (T_l) i postembrionalnego (T_p)oraz ważono masę ciała poczwarek.

Stwierdzono, że wartość T_e jest silnie skorelowana z T_l (rys. 1), a także z T_p. Korelacja ta jest silniejsza w grupie 6-stadialnej niż w 7-stadialnej, a także silniejsza u samic niż u samców (tab. 1). Można by przypuszczać, że korelacja ta jest spowodowana istnieniem grup 6- i 7-stadialnych w populacjach obu gatunków. Jednakże grupy te różniące się pod względem T_e i ciężaru ciała nie wykazują wyraźnych różnic w wartości T_e (rys. 2). Ponadto, obydwie grupy rozpatrywane osobno

wykazują również korelację pomiędzy wartościami T_e i T_l (rys. 1). Tak więc obserwowana korelacja jest prawdopodobnie niezależna od istnienia w populacjach grup stadialnych.

Nie stwierdzono wyraźnej zależności pomiędzy wartościa T_e a masą ciała poczwarek (tab. 2).

W populacjach obu gatunków stwierdzono tendencje do dodatniej korelacji pomiędzy wertością T_l a ciężarem poczwarek (tab. 3). Jest to efekt istnienia w populacjach grup stadialnych. Obydwie grupy stadialne rozpatrywane osobno wykazują tendencję do ujemnej korelacji pomiędzy tymi wartościami ($R<0$). Przypuszczalnie jest to efekt istnienia w tych grupach silnych i słabych genotypów: tych, które rozwijają się szybciej i osiągają większą masę ciała oraz tych, które im ustępują w tych cechach (rys. 3 i 4).

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