Boll. Ist. Ent. "G. Grandi" Univ. Bologna, 49: 15-19, 1994.

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Comparison of different commercial veal homogenates in artificial diets for *Brachymeria intermedia* (Nees) (Hymenoptera Chalcididae) (*)

(Research supported by MURST 40%)

Introduction

Complete *in vitro* rearing has so far been obtained for several idiobiotic hymenopterous parasitoids (see Bratti, 1990 for a review). Dindo *et al.* (1994) cultured *Brachymeria intermedia* (Nees), a solitary polyphagous pupal endoparasitoid of Lepidoptera, from the egg to adult stage on oligidic diets. These media contained 20% extract of *Galleria mellonella* L. (Lep. Galleriidae) pupae (Bratti, 1989) and 80% commercial meat homogenate (Plasmon®), a food especially designed for human babies. Moreover, Dindo *et al.* (submitted) demonstrated that diets based on meat homogenates for babies at the beginning of weaning (a) are more effective for *B. intermedia* than are those containing homogenates for babies well on in weaning (b). This difference is probably related to the higher protein, carbohydrate, lipid and calorie level of the a- than of the b-homogenates.

The two homogenate types, however, are different also in texture and added sodium chloride content. In fact the texture of the a-homogenates is smoother than that of the b-homogenates. Moreover, the former are no salt added, whereas the latter contain 0.25% added sodium chloride. We decided, therefore, to compare diets based on commercial veal homogenates with different texture and added sodium chloride content, to investigate the possible influence of these factors on the development of *B. intermedia*. Veal was selected among the other meat types, because diet veal-a produced the highest adult yield (27.4%) (Dindo *et al.*, submitted).

The results are reported in the present paper.

Materials and Methods

For the preparation of the diets, commercial veal homogenates (Plasmon®) for babies at the beginning of weaning (a), well on in weaning (b) and starting to chew (c) were used. Their composition and calorie content are reported in Table 1. The texture of the c-homogenate is much coarser than that of the b-homogenate.

(*) Accepted for publication on May 3, 1994.

Table 1 Nutrient content (g/100 g), % sodium chloride added and calories (/100 g) of the Plasmon® veal homogenates for babies at the beginning of weaning (a), well on in weaning (b) and starting to chew (c) used for the preparation of the artificial diets for *Brachymeria intermedia*. The values are drawn from those reported on the labels.

	dry residue	proteins (NX6.25)	lipids	carbohydrates	minerals	%sodium chloride	calories
a	≥19.5	9.5	≤6	6.4	0.5	0	118
b	16.8	7.5	≤5.2	3.5	0.6	0.25	91
c	20	10	5.2	5.6	0.83	0.25	107
d	16.8	7.5	≤5.2	3.5	0.6	0.25	91

Four diets were compared. The first three (a, b and c) were respectively based on a- b- and c- homogenates, whereas the fourth (d) was based on b-homogenate that had been further homogenized in the laboratory at 20,000 revolutions per minute for 1' 30", to make its texture as smooth as that of the a-homogenate.

All diets contained 80% homogenate and 20% extract of *G. mellonella* pupae prepared by the method described by Bratti (1989). They were supplemented with gentamycin sulphate (0.006%) to inhibit bacterial contamination (Bratti & Monti, 1988) and set in 1.2% agar.

B. intermedia eggs were obtained and sterilized by the method described by Dindo & Campadelli (1992), than placed singly on 0.4 cc of diet that had previously been pipetted into wells of 24-well plates (Nunclon, Denmark). The plates were sealed and kept in darkness at 26±1°C .

Instruments and glassware were sterilized by autoclaving for 20 minutes at 120°C and 1 bar. All operations, including daily visual examinations, were performed in a laminar flow hood.

Four repilicates were carried out, each comprising 23 eggs per diet.

Results were evaluated in terms of percentages of hatched eggs, mature larvae, which eliminated faeces, pupae and adults. Percentages of pupae developing to adults (= % adult emergence) were also calculated. Development times from egg to elimination of faeces by mature larvae and to adult emergence were recorded and newly emerged adults were weighed.

The data were analysed by one-way analysis of variance and means were compared using the least significance difference (LSD) multiple range test. Percentages were transformed for the analysis using the values tabulated by Mosteller & Youtz (1961) for small samples according to Snedecor & Cochran (1980). Means and standard errors in the tables are based on untrasformed values.

Results

There were no significant differences among diets in percent of hatched eggs (d.f. 3,12; p = 0.3452) and mature larvae (d.f. 3, 12; p = 0.9710), as well as in larval development times up to elimination of faeces (d.f. 3, 12; p= 0.5774) (Table 2). On the opposite, the percentages of pupae (d.f. 3,12; p= 0.0223) and adults (d.f. 3,12; p= 0.0018) were significantly influenced by diet. The highest adult yields were produced on diet (a) and (c) (Table 2).

The % adult emergence were also significantly affected by diet (d.f. 3, 11; p= 0.0087). No differences were found in percent of emergence among diet a, c and d, but, on the latter, a few pupae formed in the first 3 replicates whereas no pupae were obtained in the fourth (Table 2).

The data on adult weights (W) and development times from egg to emergence (T) were pooled for the four replicates and not subjected to statistical analysis because of the few individuals obtained, especially on diet (b) and (d). Both males and females, however, were comparable in weight to those reared from *G. mellonella* pupae. Similarly, the development times (T) observed *in vitro* were comparable to those observed in the host, though the times recorded on diet (b) were considerably shorter than on other diets for unknown reasons (Table 3). Adults were fecund.

Table 2 *In vitro* rearing of *Brachymeria intermedia* on artificial diets based on Plasmon® veal homogenates (means ± S.E.). Means in the same column followed by the same letter are not significantly different (LSD test, = 0.05). us= number of hatched eggs; u= original number of eggs placed on the media(=23);ml= number of mature larvae which eliminated faeces; p= number of normal pupae; a= number of adults.

	% hatched eggs (us/u x 100)	% mature larvae (ml/us x 100)	larval development time (days)	% pupae (p/ml x 100)	% adult emergence (a/p x 100)	% yields of adults (a/us)
diet a	85.9±4.8a	84.4±8.3a	12.8±1.1a	41.5±10.2a	79.2±7.2a	27.4±7.2a
diet b	83.7±4.8a	91.2±0.9a	10.9±1.1a	18.7± 5.1bd	54.1±15.8b	8.1±1.9b
diet c	84.8±3.8a	88.4±3.8a	11.1±0.5a	39.5±3.3ab	89.6±6.3a	32.4±2.6a
diet d	74.0±6.5a	80.4±9.1a	11.3±0.6a	13.6±5d*	75±14.4ab*	7.2±3.1b

* On diet d, no pupae and no adults were obtained in the fourth replicate.

Table 3 Weights in mg (W) and development times from egg to adult in days (T) of *Brachymeria intermedia* parasitoids obtained on artificial diets based on Plasmon® veal homogenates and *in vivo* . Means ± S.E. The data for the 4 replicates were pooled.

	males			females		
	n	W	T	n	W	T
diet (a)	9	7.9±0.3	19.2±0.3	12	11.6±0.4	20.1±0.2
diet (b)	3	8.1±0.6	16.7± 0.8	4	10.7±0.6	16.5±0.3
diet (c)	12	7.5±0.4	19.7±0.6	11	10.8±0.2	20.4±0.3
diet (d)	1	7.7	20	4	12.5±0.7	19.5±0.3
in vivo	12	8.8±0.2	18±0.1	12	12.2±0.2	20.8±0.2

Conclusions

The results did not show any difference between diet (a), based on no-salt-added homogenate with very smooth texture, and diet (c), based on sodium chloride-added homogenate with coarse texture. Moreover, the homogenation to which homogenate (b) was subjected in the laboratory did not improve its efficiency as an ingredient in artificial diets for *B. intermedia*, as diet (d) produced very low yields of adults, not significantly different from those obtained on diet (b).

We can conclude that neither homogenate texture nor added sodium chloride (0.25%) affect the development of *B. intermedia*. Therefore the most plausible explanation for the higher efficiency of the a- than of the b-homogenates (Dindo *et al.*, submitted), is that the nutrient and calorie content is higher in the former than in the latter homogenates. A further demonstration is given by the fact that the level of proteins, carbohydrates and lipids of the veal c-homogenate is only slightly different from that in the veal a-homogenate (Table 1). For the preparation of the diets, however, the a-homogenate is more practical to use than the c-homogenate, because of its smooth texture.

SUMMARY

Brachymeria intermedia, a hymenopterous pupal endoparasitoid of Lepidoptera, was reared *in vitro* on 4 artificial diets based on commercial veal homogenates for human babies (Plasmon ®). The first 3 diets were respectively based on no-salt-added homogenate with very smooth texture (a), on sodium chloride-added (0.25%) homogenate with smooth texture (b) and on sodium chloride-added (0.25%) homogenate with coarse texture (c). The fourth diet (d) was based on b-homogenate that had been further homogenized in the laboratory at 20.000 revolutions per minute for 1' 30", to make its texture as smooth as that of the a-homogenate. All diets contained 20% pupal extract of *Galleria mellonella*. Diets (a) and (c) produced the highest adult yields (27.4% and 32.4% respectively), whereas on diets (b) and (d) only a few adults were obtained. Therefore, neither homogenate texture nor added sodium chloride (0.25%) affect the development of *B. intermedia*. The higher efficiency of diet (a) and (c) was seemingly due to the fact that the nutrient and calorie level of the a- and c-homogenate is considerably higher than that of the b-homogenate.

Confronto tra diversi omogeneizzati commerciali a base di carne di vitello in diete artificiali per *Brachymeria intermedia* (Nees) (Hymenoptera Chalcididae)

RIASSUNTO

L'imenottero *Brachymeria intermedia*, endoparassitoide di pupe Lepidotteri, è stato allevato *in vitro* su 4 diete artificiali, a base di omogeneizzati per bambini (Plasmon®) di carne di vitello. Le prime 3 diete contenevano, rispettivamente, omogeneizzato di struttura finissima e privo di sale aggiunto (a), omogeneizzato di struttura fine contenente cloruro di sodio aggiunto (0,25%) (b) e, infine, omogeneizzato di struttura grossolana pure contenente cloruro di sodio aggiunto (0,25%) (c). La quarta dieta (d) era invece a base di omogeneizzato (b), sottoposto in laboratorio ad una ulteriore omogeneizzazione a 20.000 giri/min per 1'30", allo scopo di rendere la sua struttura fine al pari di quella dell'omogeneizzato (a). Tutte e 4 le diete sono state integrate con estratto di crisalide di *Galleria mellonella* in misura del 20%. Sulle diete (a) e (c) sono state ottenute le più elevate rese in adulti, pari, rispettivamente, al 27,4% e al 32,4%. Viceversa, sulle diete (b) e (d) solo pochi individui hanno raggiunto lo stadio di immagine. Pertanto, nè la struttura dell'omogeneizzato, nè la presenza di cloruro di sodio aggiunto in misura del 25% influenzano lo sviluppo di *B. intermedia*. La maggiore validità delle diete (a) e (c) è verosimilmente da attribuirsi al fatto che il contenuto in

nutrienti e in calorie degli omogeneizzati (a) e (c) è considerevolmente maggiore rispetto a quello dell'omogeneizzato (b).

REFERENCES CITED

BRATTI A., 1989. - Allevamento *in vitro* di *Pseudogonia rufifrons* Wied. in estratti di omogeneizzato di crisalidi di *Galleria mellonella* L. e su diete meridiche. - *Boll. Ist. Ent. "G. Grandi" Univ. Bologna*, 44: 11-22.

BRATTI A., 1990. - Tecniche di allevamento *in vitro* per gli stadi larvali di insetti entomofagi parassiti. - *Boll. Ist. Ent. "G.Grandi" Univ. Bologna,* 44:169-220.

BRATTI A., MONTI M. , 1988.- Allevamento *in vitro* delle larve di *Pseudogonia rufifrons* Wied. (Dipt. Tachinidae) su omogeneizzato di crisalidi di *Galleria mellonella* L. (Lep. Galleriidae). - *Boll. Ist. Ent. "G. Grandi" Univ. Bologna*, 43: 115-126.

DINDO M.L., CAMPADELLI G., 1992. - Preliminary studies on the artificial culture of *Brachymeria intermedia* (Nees) (Hym. Chalcididae) on oligidic diets. - *Boll. Ist. Ent. "G. Grandi" Univ. Bologna,* 46: 93-99.

DINDO M.L., SAMA C., FARNETI R., 1994. - Allevamento *in vitro* di un endoparassitoide pupale, *Brachymeria intermedia* (Nees) (Hymenoptera: Chalcididae).-*Atti XVII Congresso Naz. Italiano Entomologia* - Udine, 13-18 giugno 1994 *(in press).*

DINDO M.L., SAMA C., FANTI P. - *In vitro* rearing of the pupal parasitoid *Brachymeria intermedia* (Nees) (Hym.: Chalcididae) on oligidic diets. - (submitted)

MOSTELLER, F.M. , YOUTZ C., 1961. -Tables of the Freeman-Tukey transformations for the binomial and Poisson distributions. - *Biometrika*, 48: 433-440.

SNEDECOR, G.W. & COCHRAN W.G. - 1980. Statistical methods. -*Iowa State University Press*, Ames.