Boll. Ist. Ent. "G. Grandi" Univ. Bologna, 50: 143-153, 1996.

EGIDIO MELLINI, GUIDO CAMPADELLI
Istituto di Entomologia "G.Grandi" dell'Università di Bologna

Latest results in the rearing of the parasitoid *Exorista larvarum* (L.) on oligidic diets. (*) (1)

(Research supported by MURST 40%)

Introduction

The present study concerns the *in vitro* rearing of *Exorista larvarum* (L.) with special attention to the most appropriate techniques for reducing the costs of rearing this parasitoid on artificial oligidic diets. The best diet developed by us in a previous study (Mellini and Campadelli, 1995), employing skim milk as diluent and yeast and egg yolk as main nutrients, was adopted as the basic medium.

In particular, the study was aimed at determining the effects of changing egg yolk rates for the same amount of pabulum, the ideal thickness of the pabulum in the Petri dishes employed for the collective rearing of the parasitoid depending on the behaviour of the larvae, and the possibility of using inexpensive cotton wool as a substrate for the liquid diet in place of costly agar.

Moreover, given that many newly hatched larvae from dehiscent eggs placed on the surface of the pabulum were observed not to sink immediately into it (as, vice versa, is normally the case on the host) but to travel often large distances, even reaching the walls and the lids of the containers where after a few days they starve to death, an attempt was made to ascertain whether the introduction into the diet of homogenized host larvae could significantly reduce this behaviour. Finally, the addition to the diet of small amounts of soya meal, a very cheap and highly protein ingredient, as a partial substitute for the more expensive fresh chicken egg yolk, was tested. This was done not only in an attempt to reduce costs even if only to a limited extent but also because this ingredient permits to reduce the doses of agar required for appropriately gelling the diet.

In the present experiments, emergence rates of between 85% and 90% for *in vivo* reared puparia samples were obtained, while the viability of the eggs

(*) Accepted for publication: March 4, 1996.
(1) Studies on Diptera Tachinidae. LXIX contribution.

employed confirmed the results of previous studies, with hatching rates of around 90%. It is important to note that in *in vivo* rearing the eggs and puparia do not undergo those manipulations typical of *in vitro* rearing which can to some extent adversely affect their viability.

It should also be noted that the mean weight of *in vitro* reared puparia was generally found in these experiments, as in previous ones, to be greater, and even considerably so, than that of the puparia developing on the host. This finding depends not only on the quality of the diet but also on the amount available for each larva, which is far greater than that available in the larvae of the factitious host *Galleria mellonella* L. In fact, the latter are parasitized within the first few days of their last instar, that is when they are not yet fully developed, being then left unfed and having to bear a considerable number of antagonist eggs.

Materials and methods

Almost all the experiments described below were conducted using glass Petri dishes with a diameter of 5 cm which were, in turn, enclosed in dishes with a diameter of 17 cm wrapped up in aluminium foil so as to always keep their contents in the dark. Unless otherwise specified, 13 cc of diet and 30 non-incubated eggs of *Exorista larvarum* were placed in each dish. Throughout the experiments, the temperature ranged between 24 and 28 °C.

In addition to the ingredients shown in the various tables, the diets were also added with the usual doses of ascorbic acid (0.01%) and gentamycin sulphate (0.03%).

As a considerable difference was always observed between the number of eggs introduced and the number of puparia formed, the viability of the eggs was tested for each experiment by rearing the *Galleria* larvae which supported the eggs remaining after the experiment until the formation of the antagonist puparia.

The following tests were performed with 4 to 6 replications as the case required.

1. Variations in the number of eggs for the same amount of pabulum.

The tests comprised four treatments, the amount of pabulum invariably being 6.5 cc for each dish, while the number of eggs introduced was gradually reduced from a maximum of 40 to a minimum of 10. In other words, the food ration per egg varied from between 162.5 mg (which corresponds more or less to the mean weight of *Galleria* larvae subject to parasitization) for the highest population density to 650 mg for the lowest one. In actual fact, however, given that the number of puparia which develop is much less than that of the eggs introduced (generally a bit less than half), the amount of food available to the developing larvae was twice as much as the nominal doses mentioned above, and therefore more than enough even for the treatments with higher population densities. It should however be noted that, given that the pabulum is not very thick, it tends to dry out and this has the effect of slightly extending larval development times and of slightly reducing puparia weight.

Four replications were made.

Diet formulation:

Skim milk %	Yeast %	Egg Yolk %	Saccharose %
74	8	16	2

2. A comparison of diets containing *Galleria mellonella* larval homogenate with diets devoid of this ingredient.

This issue has already been dealt with in previous studies published by us, the purpose being that of ascertaining whether host material in the diet is indispensable for the development of the parasitoid (Mellini and Campadelli, 1994, 1995). As it has been demonstrated that the presence in the diet of such material is in fact not essential, new tests were carried out in order to verify whether the larval homogenate of the factitious host attracts the newborn larvae of *Exorista*. As has already been mentioned, in fact, once hatched on the artificial diet, the larvae tend to disperse (with often over 40% not sinking into the pabulum), reaching the lid where after a few days they starve to death. Vice versa, on the *Galleria* host generally the newborn larvae immediately pierce the integument in the area adjacent to the cephalic pole of the dehiscent egg.

So as not to significantly increase parasitoid production costs in the case of mass rearing, only small amounts of the homogenate were added to the diet. The amount of saccharose was doubled in treatment D as many authors hold that this sugar possesses phagostimulating properties for insects in general. In treatments C and D the lack of homogenate was compensated for with a corresponding greater amount of skim milk.

In view of the surprising and unexpected results of the first tests, two additional replications were made in addition to the usual four.

Diet formulation:

Diet	Homogenate (%)	Skim milk (%)	Yeast (%)	Egg yolk (%)	Saccharose (%)
A	10	64	8	16	2
B	5	69	8	16	2
C	–	74	8	16	2
D	–	72	8	16	4

3. The importance of pabulum thickness.

Given the subvertical position in which the larvae sink into the diet, a position they generally maintain up to the advanced stages of the third instar, it was decided to compare the effects of two pabulum thicknesses amongst the ones most commonly adopted throughout our experiments, namely 6.6 and 3.3mm. The latter value was obtained by simply halving the usual amount of pabulum employed in 5cm diameter Petri dishes (13 cc). Thinner pabulum layers are not recommendable since, as other trials have shown, the diet tends to dry up, while thicker layers means uselessly employing greater amounts of pabulum, although it is worth mentioning that in case of diet contamination by bacteria or moulds, thicker layers favour the survival of the larvae which manage to continue feeding at deeper levels.

As the number of eggs (30) introduced was the same for all treatments, the amount of diet available to each egg varied from 433 to 216 mg in the two experimental set-ups. Moreover, as already mentioned, these nominal doses are always considerably less than the real ones since during their development the number of larvae usually drops to less than half with respect to the number of eggs introduced.

The tests were carried out using two types of very simple diets which had proven to be ideal in previous studies. One of these diets contained small amounts of *Galleria* larval homogenate while the other was devoid of this ingredient, as shown in the table below.

Diet formulation:

Diet	Homogenate (%)	Skim milk (%)	Yeast (%)	Egg yolk (%)	Saccharose (%)	Pabulum thickness (mm)	Pabulum quantity/egg (mg)
A	10	64	8	16	2	6.6	433
B	10	64	8	16	2	3.3	216
C	–	74	8	16	2	6.6	433
D	–	74	8	16	2	3.3	216

It should finally be mentioned that as far as the amount of ingredients in the diet is concerned, the present tests were similar to those described in section 1 above, except for the amount of eggs which was unvaried for all diets.

4. Introduction into the diet of small amounts of soya meal.

This ingredient has already been tested by us in a previous paper (Mellini and Campadelli, 1995) without, however, any satisfactory results owing to the large doses employed. In fact, amongst other effects, this ingredient was found to overly harden the diet, thus making it difficult to prepare and for the larvae to efficiently exploit it.

In view, however, of the low cost of soya meal and of the fact that, thanks to its outstanding protein content, it is widely employed in artificial diets for insects in general, including parasitoids, a new experiment was set up adopting the following changes. In the first place, the amount of soya meal was reduced, varying therefore from between 2 and 8% instead of from between 6 and 12% and the amount of egg yolk was increased by 2% for all treatments while that of yeast, which is one of the most expensive ingredients, was correspondingly reduced by 2%.

In the second place, considerable changes were made with regards the amounts of agar employed. Given that soya meal, and to a lesser extent egg yolk, tend to harden the pabulum, the usual amount of agar was reduced from 1.5 to 1.0% in diets A and B and to as low as 0.5% in diets C and D, which contained greater doses of soya meal. In this manner, an ideal consistency was attained for the various diets. Larval growth was thus seen to be favoured both in terms of weight and development times as compared to the first treatments conducted, where the diets had been added with the usual amount of 1.5% of agar. Moreover, this also

reduced costs, albeit only slightly, seeing as this gelling agent is one of the most expensive components in the diet.

Diet formulation:

Diet	Skim milk (%)	Larval homo-homogenate (%)	Yeast (%)	Egg yolk (%)	Soya meal (%)	Saccharose (%)
A	75	5	4	12	2	2
B	75	5	4	10	4	2
C	75	5	4	8	6	2
D	75	5	4	6	8	2

5. The use of cotton wool as a support for liquid diets.

Wads of cotton wool were used as an agar substitute for supporting the liquid diets. The purpose was to use a material which, while avoiding the introduction of an indigestible substance in the pabulum, permitted considerable savings both in terms of initial purchase cost and ease of application.

This issue has already been dealt with in previous papers by us (Mellini *et al.*, 1993). In the present study the use of cotton wool was further investigated applying new techniques and in connection with recently developed diets richer in nutrients, the considerable validity of which has been proven.

The pabulum employed was made up as follows

Skim milk (%)	Larval homogenate (%)	Yeast (%)	Egg yolk (%)	Saccharose (%)
75	5	6	12	2

Two treatments were compared. In treatment A the diet was added with agar to the amount of 1%, while in treatment B the diet was made to soak into in 0.5 g of cotton wool. In both cases standard 5cm diameter Petri dishes were employed, each containing 6.5 cc of pabulum and 30 *Exorista* eggs. The cotton wool was sterilized by autoclaving at 135 °C for 20 minutes. Given the considerable practical importance of this test, six replications were made.

Results

1. Variations in the number of eggs for the same amount of pabulum.

The table below shows the findings of the four replications.

As can be seen, the mean weight of the puparia is the same in all diets, which means that this factor remained unaffected by high food rations. No significant differences in emergence rates (always rather high) can be observed either. Obviously, the number of puparia is less as the number of eggs introduced is less, albeit not proportionally. In fact, the ratio between puparia and eggs increases as the latter decrease, going from 33% for the 40 eggs/dish trials to over 57% for the 10 eggs/dish ones. This means that lower population density conditions tend

Diet	No. eggs	Pabulum/ egg (mg)	Pabulum/ puparium (mg)	No. puparia	Mean weight (mg)	Emergence (%)	No. pup./ No. eggs	Biomass pup./ pab. weight
A	40	162.5	490	53	34.31	83.01	33.12	7.0
B	30	216.6	500	52	35.48	88.46	43.33	7.1
C	20	325.0	702	37	35.18	81.08	46.25	5.0
D	10	650.0	1130	23	38.41	82.64	57.50	3.4

to favour the development of a higher percentage of larvae. This finding is rather surprising when one considers that *Exorista* is a gregarious parasitoid and, moreover, that the space and amount of food available are more than required even in the trials with the greater number of eggs.

The weight ratio between the puparia biomass and the amount of diet available is always very low. This finding does not depend on the low nutritional value of the pabulum but rather on the fact that the food is actually not exploited sufficiently as there is too much of it. In fact, considerable parts of the pabulum were found to be still perfectly intact after the puparia had formed. Obviously, this ratio progressively diminishes as the number of eggs introduced is less, reaching very low levels (from 7% for diet A to as low as 3.4% for diet D).

All in all, of the four experimental conditions examined, diet B gave the most satisfactory results, the number of puparia produced in relation to the number of eggs employed being good and the pabulum utilization index being amongst the highest recorded.

An additional test in which the diet was modified by substituting 10% skim milk with *Galleria* larval homogenate yielded similar results except for a slight drop in the number of puparia produced.

It can once again be noted that generally speaking there is a considerable gap between the number of inoculated eggs and that of the puparia formed. To a limited extent, this depends on the failure of some eggs to hatch (about 10%) and to a larger extent on the marked tendency of the newborn larvae to escape from the pabulum, moving up the walls of the Petri dish even up to the lid. As already mentioned, this tendency is all the more marked the higher the population density level even when there is ample space and food available. It may therefore be concluded that in order to increase the diet utilization index, reducing the quantity of food is a much more convenient strategy than increasing the number of eggs introduced.

The results of the present tests are fairly much in line with those of other tests already conducted by us in which, however, a different diet and different rearing techniques were employed (Mellini *et al.*, 1993).

2. A comparison of diets containing *Galleria mellonella* larval homogenate with diets devoid of this ingredient.

The results of the six replications are shown in the table below.

Puparia yield appeared to be considerably higher with diets devoid of host material. In fact, the puparia to egg ratio (180 eggs being used for each type of diet) increased from about 37 and 34% for diets A and B, respectively, to 50 and

Diet	No. of puparia	Mean weight (mg)	Emergence (%)	No. of puparia/No. of eggs
A	68	50.72	83.82	37.77
B	62	50.93	79.03	34.44
C	90	50.44	78.88	50.00
D	94	45.51	79.78	52.22

52% for diets C and D, respectively. This truly unexpected result was recorded for all replications. This means that, at least in the quantities employed and having been autoclaved, the homogenate not only does not exert any specific effect of attraction on the newborn larvae but that it actually seems to be tendentially repulsive to them.

Mean puparia weight, on the other hand, was found to be more or less the same in all diets and in any case sensibly higher than in the *Galleria* factitious host. Emergence rates were also found to be rather high, being practically the same for all experimental conditions.

Dissection of the unhatched puparia revealed the presence of either dried up eopupae or, less frequently, of dead adults which had not been capable of opening the cephalic cap of the puparium. Such failures were also frequently reported in the other tests as well as in *in vivo* rearing. A peculiar situation, encountered by us almost only in *in vitro* rearing, was represented by the presence of collapsed puparia, which preferably tended to involve specimens of considerable weight.

In conclusion, the problem of how to enhance the power of attraction of the diets for the newborn larvae so as to increase puparia yield, still remains to be solved. As far as other aspects are concerned, the diets employed in this test appear to be valid as mean puparia weight and adult emergence rates were found to be good. As regards the possibility of enhancing the diet's power of attraction, a further study will be conducted in which extracts of *Galleria* sterilized by means of microfiltering will be introduced in the diet in place of the homogenates sterilised at 135 °C, as this technique probably alters several components of the extract.

3. The importance of pabulum thickness.

The table below shows the findings for the four replications:

Diet	No. of puparia	Mean weight (mg)	Emergence (%)	Amount of diet/pupa. (mg)	Puparia biomass/ pabulum weight
A	42	49.12	78.57	1238	3.96
B	39	40.60	84.61	666	6.08
C	73	49.00	65.75	712	6.87
D	61	40.92	77.04	426	11.02

Puparia weight was found to be tendentially less in treatments B and D in which diet thickness as well as its quantity were reduced by half. Even in this case, however, the ration per capita was by far in excess of that required, with

666 mg of pabulum being available on average for each pupated larva in diet B and 426 mg in diet D. Given that the second and third instar larvae have no difficulty in taking up a horizontal rather than a subvertical position, it may therefore be assumed that this tendentially lower puparia weight does not depend on the smaller amount of pabulum available or on the lower thickness of the pabulum itself. The reduction in puparia weight accompanied by a markedly longer larval growth period more than likely depends on the greater tendency of the "thin" pabulum to dry up and crack, thus taking on a consistency which, as already seen in other experiments, constitutes an obstacle for larval development.

The number of puparia was found to be sensibly greater in treatments C and D for which diets devoid of *Galleria* homogenate were employed. We are therefore faced with the same surprising situation already noted for the previous test. As the larvae rarely die in the course of their development, it may be concluded that they are more easily induced to feed on a diet devoid of host material, at least in the quantity and quality provided by us.

Emergence rates were lower in diet C which yielded the highest number of puparia, a good number of which with outstandingly high weights ranging from 80 to 90 mg (more than double the mean weights of *in vivo* reared specimens), with a slight delay in their development.

From a practical point of view, the thickness of the pabulum adopted for most of our experiments (6.6 mm) can therefore be reduced by half. Even if the weight of the puparia thus obtained is somewhat lower, it is nevertheless always greater than that obtained with the factitious host. It is in any case advisable to prevent the tendency of the pabulum to dry up by bringing to saturation the relative humidity of the air in the containers where the Petri dishes containing the pabulum are placed.

Diet utilization index, understood as the ratio between the puparia biomass and the trophic mass provided, was almost double in these diets in which the thickness of the pabulum was halved. The index recorded, however, is still too low especially if one considers that the highest value reported for diet D is only 11.02.

4. Base diet enriched with small amounts of soya meal.

The following table shows the findings for four replications.

Diet	No. of puparia	Mean weight (mg)	Emergence (%)	No. of puparia/ No. of eggs	Pabulum utilization index (%)
A	62	59.95	91.93	51.66	7.14
B	45	52.39	91.11	37.50	4.53
C	34	53.20	94.11	28.33	3.47
D	18	49.74	88.88	15.00	1.72

As can be seen, puparia yield progressively diminishes as the amount of soya meal increases and that of egg yolk correspondingly decreases. The percent ratio of puparia to the number of eggs introduced thus drops from a maximum of 51.66% in diet A to a minimum of only 15% in diet D. Since, as usual, no corresponding mortality in the developing larvae was observed, the lower yield of

puparia especially in diets C and D may be ascribed to the lower power of attraction exerted by these diets towards the newborn larvae, which tend to disperse in considerable number outside the pabulum.

Similarly to yield, mean puparia weight also tends to diminish, albeit less dramatically. In diet A it is exceptionally high, remaining even in diet D greater by over 30% than that reported for the *Galleria* host. As the amount of soya meal in the diet was increased, a rather sensible and progressive extension in larval growth time was also observed.

Diet utilization index was rather low, going in fact from 7.14 in the best conditions (diet A) to 1.72 in the worst (diet D). Indeed, this index could have been higher if the amount of diet had simply been reduced by half. In fact, the highest puparia yield was obtained in diet A where the amount of pabulum/larva was as high as 742 mg. It can safely be said, even on the basis of the specific tests currently being conducted on this topic, that this amount is really too much. Such a ration was however adopted in order to prevent any possibility of the diet progressively drying up.

In conclusion, amongst those so far tested, diet A was found to be one of the best both in terms of yield and costs. Its application for the mass rearing of *Exorista* is therefore worthy of the utmost consideration.

5. The use of cotton wool as a support for liquid diets.

The table below shows the results of the four replications.

Treatment	No. of puparia	Mean weight (mg)	Emergence (%)
Agar	52	51.09	80.76
Cotton	53	48.04	100

As can be seen, no difference was observed in the production of *Exorista* when cotton was used in place of agar to support the diet, the number and mean weight of puparia being approximately the same in both cases. A slight difference was observed in larval growth times, which in the treatment employing cotton wool appears to be about half a day longer. Emergence rates, however, exhibited considerable differences, being 80.76% for the agar added diet and indeed as high as 100% for the cotton imbibed one.

From a practical point of view, therefore, the use of cotton wool in place of agar appears much more convenient not only because the latter is more expensive but also because it requires longer times for diet preparation.

The initial observations made by us in a previous study (Mellini *et al.*, 1993), in which however a diet of a very different composition and a different pabulum to cotton ratio were adopted, are so fully confirmed.

Neither Bratti and Nettles (1992) have observed any appreciable difference in the production of the *Eucelatoria bryani* Sab. tachinid by using agarized or cotton wool imbibed diets.

The above considerations are applicable to mass production. In laboratory applications, however, the use of an agarized diet is by far preferable for studying the larval behaviour of this parasitoid.

SUMMARY

This study, the sixth of the series, was aimed at investigating ways for improving artificial diets for the parasitoid *Exorista larvarum*. In particular, both technical and nutritional aspects were considered. A total of twenty-two tests were conducted during which over a thousand puparia were formed, from which about 850 adults emerged.

With regards the number of eggs to be introduced in the pabulum (which in the present study ranged from 40 to 10 in 13 cc of diet), puparia yield was seen to be percentagewise less in the diets with a high number of eggs, even though, it should be noted, the amount of diet available for each larva was in any case far greater than that actually required. To this it should also be added that the mean weight of the puparia was seen to be practically the same for all treatments. This means that, despite the parasitoid being gregarious, its development is not favoured by crowding which results in an increase in the number of newborn larvae escaping from the diet.

Having previous research shown that the presence of host material in the diet is not indispensable for the development of *Exorista* larvae, it was decided to test whether such an ingredient could limit the migration of newborn larvae away from the pabulum. Against all reasonable expectations, repeated experiments showed that puparia yield is about 50% greater for the diets containing no *Galleria mellonella* L. factitious host autoclaved larval homogenate which, when used, was introduced in doses of 10 and 5%.

Regarding pabulum thickness in the Petri dishes employed for collective rearing of the larvae, its reduction by half (that is, from 6.6 to 3.3 mm) with the number of eggs being kept the same, resulted in a slight decrease in puparia weight but not in their number. By adding to the diet a limited amount of host larval autoclaved homogenate (10%), a marked drop in puparia yield was observed as already reported in the previous experiment. In any case, the larvae proved to adapt themselves well to the thinner diet (on condition, however, that its tendency to dry up be prevented). The puparia biomass to pabulum quantity ratio, therefore, was found to be almost double in the treatments employing the thinner diet.

The progressive, albeit only partial, replacement of fresh chicken egg yolk with soya meal resulted in a sharp drop in puparia yield and in only a tendentially slight decrease in their weight. This means once again that, despite being valid from a nutritional point of view, this latter ingredient seems to favour the migration of the newborn larvae away from the pabulum. Notwithstanding, the addition of small doses of soya meal to the diet appears to be convenient as, amongst other things, it permits to diminish the dose of agar employed which is a very expensive gelling agent.

Bearing all this in mind and the fact that the use of agar makes diet preparation difficult from a technical point of view, a new attempt was made to replace this ingredient with cotton wool. As concerns puparia yield- and weight-wise, no differences were observed for the two treatments. Growth time, however, appeared to be tendentially longer and emergence rates markedly higher in the treatment with cotton wool. It may therefore be safely concluded that cotton wool represents a perfect substitute for agar, permitting considerable money saving. In a laboratory setting, however, the use of agarized diets appears to be by far more appropriate for the study of the larval behaviour of this parasitoid.

Nuovi dati per l'allevamento del parassitoide *Exorista larvarum* (L.) su diete oligidiche

RIASSUNTO

In questo sesto contributo al miglioramento delle diete artificiali per il parassitoide *Exorista larvarum* sono stati presi in considerazione più gli aspetti tecnici che quelli nutrizionali. Complessivamente sono state effettuate 22 prove nel corso delle quali si sono formati oltre un migliaio di pupari, da cui sono sfarfallati circa 850 adulti.

Con riferimento al numero delle uova da immettere sul pabulum (nelle presenti prove da 40 a 10 in 13 cc di dieta), si è notato che la resa in pupari è percentualmente minore nelle tesi con più elevato carico di uova, pur essendo, in ogni caso, il quantitativo di dieta disponibile per ciascuna larva di gran lunga superiore alle sue reali necessità. Ciò, unitamente al fatto che il peso medio dei pupari si mantiene in pratica costante in tutte le tesi, indica che le condizioni di affollamento sfavoriscono l'attecchimento di questo parassitoide, che pure è gregario, incrementando l'esodo delle larve neonate dal substrato trofico.

Stabilito, in precedenti ricerche, che la presenza nella dieta di materiali provenienti dall'ospite non è indispensabile per lo sviluppo delle larve di *Exorista*, si è saggiato se la sua introduzione potesse invece limitare l'allontanamento delle larve neonate. Ripetute prove sperimentali hanno però evidenziato, contro ogni ragionevole aspettativa, una resa in pupari di circa il 50 % superiore nelle tesi prive di omogeneizzato larvale dell'ospite di sostituzione *Galleria mellonella* L., impiegato nella misura del 10 e del 5 % previa sua sterilizzazione in autoclave.

Per quanto concerne lo spessore del pabulum nelle capsule Petri usate per l'allevamento collettivo delle larve, il suo dimezzamento (da 6,6 a 3,3 mm), mantenendo costante il carico di uova, ha portato ad una lieve flessione nel peso dei pupari ma non nel loro numero. Addizionando alla dieta limitati quantitativi di omogeneizzato larvale dell'ospite (10 %) si è assistito invece, come nell'esperienza precedente, ad un netto calo nella resa in pupari. In ogni caso le larve hanno dimostrato di adattarsi bene a spessori modesti della dieta (purché ne sia ostacolata la tendenziale disidratazione) così che il rapporto tra la biomassa dei pupari e il quantitativo di pabulum fornito è risultato quasi doppio nelle tesi a dieta dimezzata.

La progressiva, seppure parziale, sostituzione del tuorlo d'uovo fresco di gallina con farina di soia, ha portato ad una forte caduta nella resa in pupari e solo ad una tendenziale diminuzione nel loro peso. Ciò sembra indicare che tale prodotto, seppure valido anche in questo caso dal punto di vista nutrizionale, favorisca invece l'allontanamento delle larve neonate dal substrato trofico. Tuttavia, l'aggiunta di piccole dosi di soia nella dieta appare conveniente perché consente, tra l'altro, di diminuire la quota di agar che, oltretutto, è un gelificante molto costoso.

Tenuto conto di ciò e del fatto che l'agar comporta anche complicazioni di ordine tecnico nell'allestimento della dieta, si è provato a sostituirlo con cotone idrofilo. In pratica non sono emerse differenze tra le due condizioni sperimentali per quanto concerne la resa ed il peso dei pupari; invece la durata dell'accrescimento è apparsa tendenzialmente più lunga e le percentuali di sfarfallamento sensibilmente più elevate nelle tesi con cotone. Pertanto il cotone idrofilo può perfettamente sostituire l'agar con grande vantaggio economico. Con riferimento invece allo studio del comportamento larvale del parassita le diete agarizzate sono invece di gran lunga superiori.

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