# STUDIES ON THE BIOLOGY OF RHYSSA PERSUASORIA (L.) (HYMENOPTERA: ICHNEUMONIDAE) INCORPORATING AN X-RAY TECHNIQUE

# By Helen Hocking\*

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#### Abstract

Development of the immature stages of *Rhyssa persuasoria* (L.) on the larvae, prepupae and pupae of *Sirex noctilio* F. was followed *in situ* in *Pinus radiata* D. Don. by an X-ray technique.

*R. persuasoria* females were allowed to oviposit in wood in which the location of immature stages of *S. noctilio* had been determined radiographically. By this means it was found that they could not locate their hosts with certainty. When a host was located the female parasite paralysed it by stinging and laid an egg upon it. The host was consumed within five weeks and the parasite passed through four larval instars before pupation.

#### INTRODUCTION

Chrystal and Myers (1928) observed feeding of larvae of *Rhyssa persuasoria* (L.) reared from an early stage on *Sirex cyaneus* F. larvae in gelatin capsules. Such a technique, however, introduces a variety of artificial conditions and leaves doubts as to the true course of events in the host trees. The use of soft X-rays to study insects in grain, wood and other media (for example Berryman and Stark 1962) provides a useful method for the study of the biology of *Rhyssa* spp., and in fact it was found that soft X-rays would penetrate through radial and tangential faces of suitable specimens of *Pinus radiata* D. Don. timber, permitting satisfactory study of the parasitisation of *Sirex noctilio* F. by *R. persuasoria*.

#### METHODS

Logs from trees naturally infested with Sirex noctilio were exposed to oviposition by *Rhyssa persuasoria*. On the following day 12 sample blocks approximately  $1\frac{1}{2}$  in.  $\times 1\frac{1}{2}$  in.  $\times 7$  in. in size were cut from these logs. The surface of each sample block was flamed to sterilize it against fungal infection. To reduce excessive drying the blocks were then coated with several layers of microcrystalline wax or a mixture of equal parts of petroleum jelly and paraffin wax. The samples were stored individually in numbered polythene bags in an open insectary cubicle.

The X-ray apparatus used was a converted Watson machine, catalogue number S3307, with a stationary copper anode, modified to produce 24kV and passing approximately 10 milliamps of current across the tube. The specimen block was placed 1 metre from the tube and centred with a plumbline. "Kodirex" envelope packed medical X-ray films were used for the exposures. The sample was placed on one half of a film and the other half was screened by a lead block. After one exposure for the appropriate time, the specimen was turned through 90° and another exposure made from the other surface on the unused half of the plate. By this method the precise locus of the larva could be determined. Exposure times were established by experiment for radial and tangential aspects of each sample and were found to range from 15-25 seconds depending on the thickness and nature of the wood in the sample. The films were developed in "Kodak" X-ray developer type 2, and then fixed for 10 minutes in "Kodak" X-ray fixative.

Contact radiographs of sample blocks containing R. persuasoria larvae were taken on the day after oviposition and then at weekly intervals until feeding was completed.

As each weekly exposure of the sample blocks was taken, other wood containing R. persuasoria larvae of the same age was dissected. These larvae and their exuviae were examined and the appearance and behaviour of the stages were noted.

<sup>\*</sup>Division of Entomology, C.S.I.R.O., Canberra, A.C.T. Present address: 3 Ascot Avenue, Sandy Bay, Hobart, Tasmania.

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The parasite eggs were too small to be positively identified on the radiographs at the time of parasitisation, but at a later stage the larvae could be seen and then it was possible to identify from the series of radiographs the stage of the host when it was parasitised. If further tunnelling by the host larva had occurred this could have been traced.

Using the same X-ray technique, a set of contact radiographs was taken of a slab of wood (2 ft long  $\times$  2 in. thick  $\times$  7 in. wide), with the bark surfaces intact on the two narrow sides. Five adjacent plates were exposed in turn for 20 seconds along the length of the slab, giving a complete X-ray picture and showing the areas in which the host larvae were located. The slab was then placed in a cage containing *R. persuasoria* females to see whether there was any relationship between their probing and the whereabouts of host larvae.

# RESULTS

# Searching and oviposition by R. persuasoria females

When the wood slab in which the locations of *S. noctilio* larvae had been established by radiographs was exposed to parasite females they were found to palpate the exposed bark surfaces vigorously with their antennae and to insert their ovipositors at numerous points on the surface. This probing, however, was unrelated to the positions of the larvae or their tunnels, even where several occurred close together. It was apparent therefore that there was no specific ability to locate individual host larvae.

#### Stage of host attacked

Assessment from the radiographs of parasitism in the 12 sample blocks showed that *R. persuasoria* deposited eggs randomly on larvae, prepupae and pupae of *S. noctilio*. Thus no more preference was shown for the more active larval stages than for the more or less immobile prepupal and pupal stages (Table 1). One example was encountered of parasitisation of a female *S. noctilio* on the point of emergence.

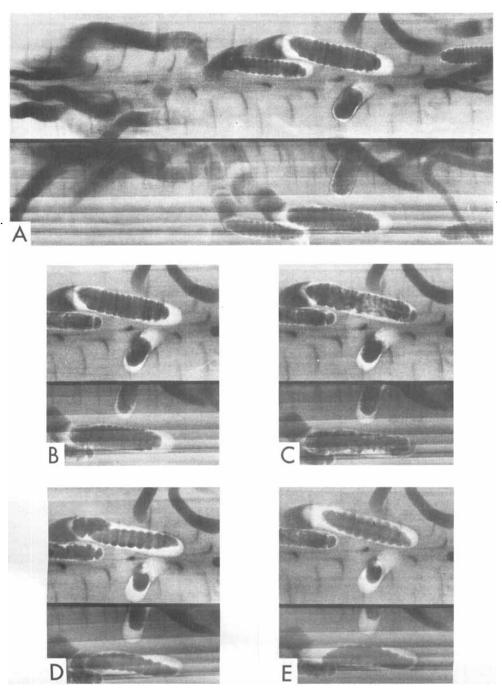
Stage	Total Insects	Per cent. of Each Stage	Number Parasitized	Per cent. Parasitism	Per cent. of Total Parasitized
Larvae	53	80	18	34	78
Prepupae	9	14	3	33	13
Pupae	4	6	2	50	9

TABLE 1

# Development of R. persuasoria larvae

Radiographs of one of the samples taken at weekly intervals (Plate 1, A-E) show clearly the manner and rate of feeding of R. persuasoria in situ. Examination of parasite larvae and their exuviae in samples exposed to the parasites at the same time, and cut out at different times during the experiment, showed that there were four larval instars. In addition, two R. persuasoria reared on host larvae in tubes, from the egg stage to the final instar, passed through four instars.

The parasite larva visible in Plate 1B was probably in the first instar. The first instar is quite active and wriggles vigorously if disturbed on the host. It has a prognathous head capsule with prominent curved mandibles (Fig. 1) which open widely when seeking a grip on the host. The second and third instars (Plate 1C) are very similar in structure, becoming progressively more hypognathous and resembling the final instar, though lacking the characteristic pointed extension of the labial sclerite (Fig. 2). The final instar larva has been figured and described by Beirne (1941) and Short (1959).





A-E.—Contact radiographs of a wood sample after oviposition by *Rhyssa persuasoria* on 29.x.63, exposure—radial face 15 secs, tangential face 20 secs, approximately natural size: (A) 30.x.63, one day after oviposition, a prepupa of *S. noctilio* is visible; (B) 6.xi.63, eight days after oviposition, *R. persuasoria* larva, probably 1st instar, can be seen on the left side of the host prepupa near the posterior end; (C) 13.xi.63, fifteen days after oviposition, parasite larva is now about 1 cm long, lying near head of host which has begun to shrivel up and become flaccid; (D) 22.xi.63, twenty-four days after oviposition, most of the host has been consumed and the parasite larva is probably in its final instar; (E) 28.xi.63, thirty days after oviposition and feeding is complete. Later dissection of this sample block, one year after oviposition, showed a female R. persuasoria ready to emerge, at the locus shown in the radiographs.

All four instars of the larvae were ectoparasitic. Feeding was completed within five weeks. The first three instars appeared to feed by piercing the host's integument with the mandibles and sucking out the fluid contents. During the final instar the remains of the host were consumed, and some shredding of the integument with the mandibles occurred.

The fully-fed larva constructed a cocoon, which was a very thin loose covering of silk and wood fibres adhering to the walls of the chamber. It remained in the cocoon in a state of diapause until the following spring. Preliminary tests using manometric techniques showed that the rate of respiration of R. persuasoria larvae during the resting stage was about one-tenth of the rate during spinning or immediately prior to pupation.

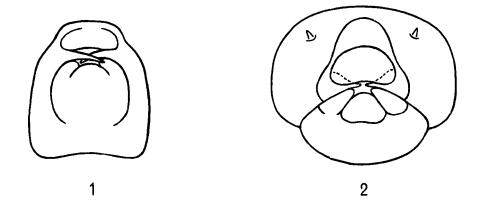
It was found that there was a distinct prepupal stage, lasting about one week, during which the integument became wrinkled, and eye-spots appeared.

### DISCUSSION

The duration of the larval stages determined in the present study (35 days) agrees well with that of 36 days reported by Chrystal and Myers (1928) under insectary conditions in England. Chrystal and Myers suggested that the host larva may continue to burrow whilst being consumed by the parasite, but the radiographs of parasitised larvae reported here showed that this did not occur. It appeared that the Sirex larva was paralysed by the *R. persuasoria* female at the time of oviposition and so was unable to burrow further.

Mandibular movements, which were elicited by tactile stimulation of healthy larvae, did not occur in parasitised larvae, or in larvae which, although not parasitised, appeared to have been stung. The latter, characterised by the presence of one or more dark brown or black spots on the integument, were only found in logs which had been exposed to attack by *R. persuasoria*. Myers (1928) observed similar larvae with "blackish" spots, and suggested that these spots were "the seats of punctures". Therefore it appears that penetration of the host integument by the ovipositor is almost certainly accompanied by the injection of venom, resulting in paralysis. From a general knowledge of the rate of action of venoms, it is likely that this would take effect rapidly, and prevent further boring by the larva. Chrystal and Myers (1928) observed that the larva of *R. persuasoria* fed

Chrystal and Myers (1928) observed that the larva of R. persuasoria fed externally from the nineteenth day onwards. Those observed in the course of the present study, either *in situ* in the wood, or cut from the blocks, were ectoparasitic for the whole of their feeding period.



FIGS. 1, 2.—Head capsules of exuviae of early instars of *Rhyssa persuasoria*: (1) first instar; (2) third instar.

Most oviposition by R. persuasoria takes place in the spring, prior to the pupation of the host larvae. However, during the present investigation it was found that a variable proportion of R. persuasoria individuals did not undergo a prolonged larval diapause but completed their development in three or four months, thus emerging during the summer. The females were then able to parasitise Sirex stadia which had been deep in the wood in spring and hence unavailable to the R. persuasoria females emerging in spring. In summer they were accessible because of the tendency of siricid larvae to tunnel towards the bark before pupation.

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