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EFFECTS OF WATER STRIDER (HETEROPTERA: GERRIDAE) EGG QUALITY ON DEVELOPMENT AND FITNESS OF *TIPHODYTES GERRIPHAGUS* MARCHAL (HYMENOPTERA: SCELIONIDAE)

by

JOÃO MANUEL SOUSA ©

A thesis submitted to the Faculty of Graduate Studies and Research in partial fulfillment of the requirements for the degree of Master of Science

DEPARTMENT OF BIOLOGICAL SCIENCES

EDMONTON, ALBERTA

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The undersigned certify that they have read, and recommend to the Faculty of Graduate Studies and Redearch for acceptance, a thesis entitled **Effects of water strider (Heteroptera: Gerridae) egg quality on development and fitness of *Tiphodytes gerriphagus* Marchal (Hymneoptera: Scelionidae)** submitted by João Manuel Sousa in partial fulfillment of the requirements for the degree of Master of Science.

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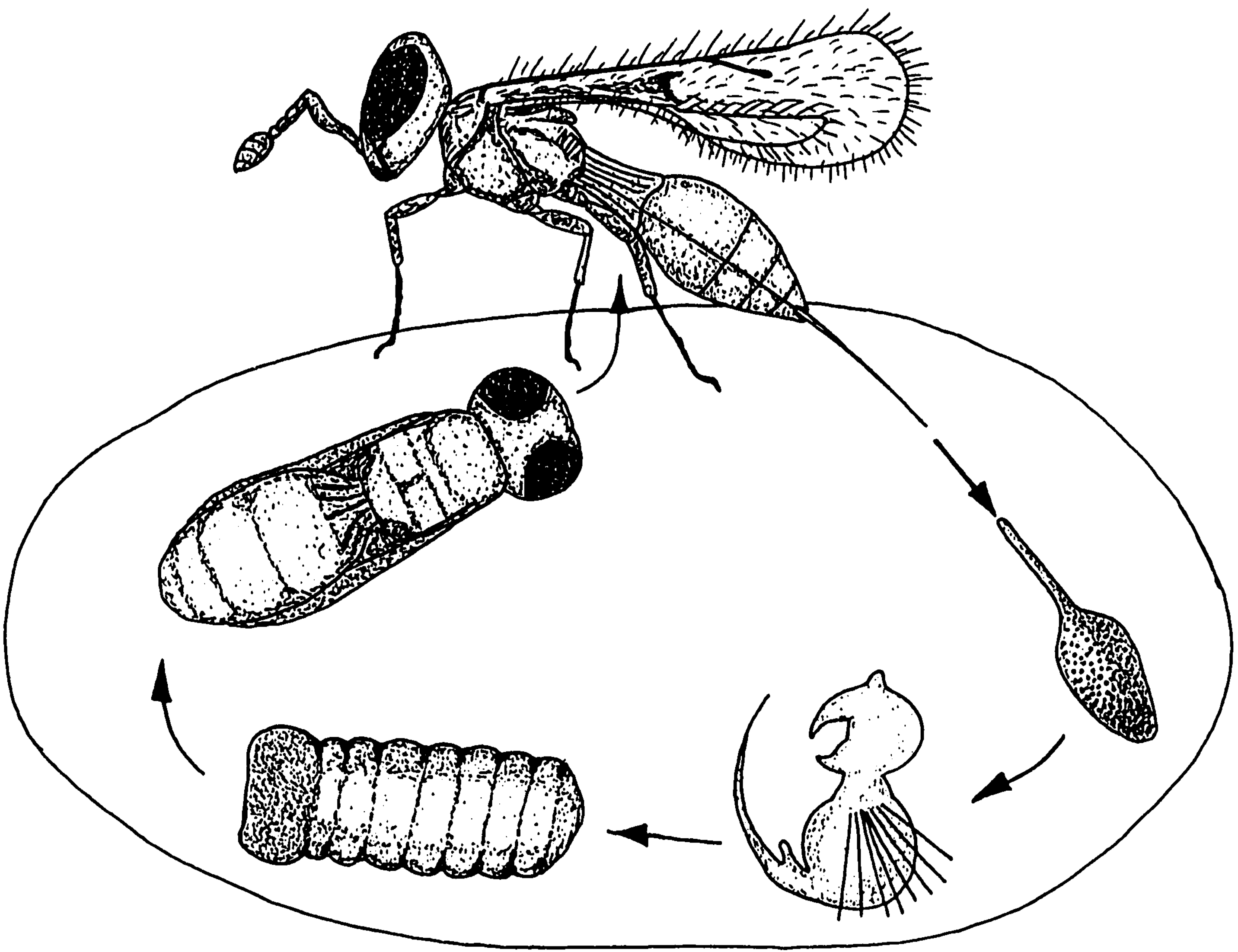
ABSTRACT

The effects of water strider (Heteroptera: Gerridae) egg quality on development and fitness of *Tiphodytes gerriphagus* Marchal (Hymentoptera: Scelionidae) were investigated.

Following preimaginal development of *T. gerriphagus* within *Limnoporus dissortis* Drake & Harris eggs, revealed two larval instars and a pupal stage. Confirmation of number of instars was based on counting exuviae and measuring mandible length. These findings are at odds with previous descriptions of *T. gerriphagus*.

Tiphodytes gerriphagus is able to develop within *L. dissortis* eggs of various ages with varying success rates. The greatest wasp emergence occurred with intermediate aged eggs because infertile eggs are less suitable for parasitoid survival. *L. dissortis* eggs which are larger than *Gerris buenoi* Kirkaldy eggs may sometimes be too large for proper wasp development. Increasing foundress number lowered parasitoid survival and body size. Sex ratio of progeny was independent of egg age, host species, and foundress number.

As wasp density increases, the number of probes per egg and the number of larvae per egg increases. Superparasitism occurred most frequently from groups of mated wasps than groups of unmated wasps. Development time of wasps in superparasitised eggs was longer than from singly parasitised eggs. Longevity was not affected in males emerging from superparasitised eggs, but female longevity was significantly decreased. Mated and unmated females were equally fecund regardless of whether they emerged from superparasitised or singly parasitised eggs.



Frontispiece: Lifecycle of *Tiphodytes gerriphagus* Marchal

Poucas pessoas irão ler parte da minha tese e ainda menos lerão o trabalho inteiro. Entristece-me saber que o meu pai não será uma destas pessoas. Em memória de José Adelino Sousa.

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1. INTRODUCTION

1.1 Aquatic Hymenoptera

The order Hymenoptera contains over 108 000 known species and include the insects perhaps most beneficial to humankind since their adults pollinate most insect-pollinated plants and are important in regulating populations of pest insects (Borror *et al.*, 1989). Prior to Lubbock's (1862) discovery of two wasps swimming beneath a pond's surface, it was thought that hymenopterans were excluded from aquatic habitats. Since then, 51 North American and 74 European species of aquatic Hymenoptera have been described from ten families (Hagen, 1978; Ward, 1992).

All aquatic hymenopterans are parasitoids; their immature stages must feed on tissues of another arthropod to develop into an adult. Parasitoids differ from parasites in that their development results in host death. They differ from predators in that only one prey item is consumed per immature parasitoid, and only adult females search for prey (Godfray, 1994).

Aquatic parasitoids may be broadly defined as those parasitising any stage in the life cycle of an aquatic invertebrate, even if the attack occurs on land (Hedquist, 1967), or may be narrowly defined as those entering the water to parasitise a host (Hagen, 1956). A more biologically meaningful definition of an aquatic parasitoid is one that parasitises an aquatic stage (Burghele, 1959), for this definition includes parasitoids attacking aquatic insects beneath the water surface and semi-aquatic insects on the surface film.

Generally, aquatic hymenopterans lack special morphological adaptations for underwater respiration because the microhabitats of water and host tissues are similar with respect to oxygen availability (Hagen, 1956). A large surface/volume ratio, due to the small size of most aquatic hymenopterans, allows for sufficient underwater respiration (Ward, 1992).

Understanding host-parasitoid interactions in the field will be incomplete without consideration of behaviour and biology of individuals in the population (Salt, 1934). Eggs of two genera of water strider (Heteroptera: Gerridae), *Gerris* and *Limnoporus*, are regularly parasitised by *Tiphodytes gerriphagus* Marchal (Hymenoptera: Scelionidae) in western Canada (Spence, 1986a). Despite knowledge of this parasitism since the beginning of this century (Marchal, 1900; Bradley, 1902; Matheson and Crosby, 1912; Martin, 1927; Hoffmann, 1932; Hagen, 1956; Masner, 1972), its significance for gerrid populations and parasitoid behaviour have been mostly ignored (Spence, 1986a). This thesis presents new information about the development of *T. gerriphagus* within gerrid eggs and variation in fitness of successfully emerging wasps.

1.2 *Limnoporus dissortis* Drake & Harris and *Gerris buenoi* Kirkaldy adults

Populations of all pond skater species can be partially bivoltine in western Canada (Spence and Scudder, 1980; Spence, 1989). Overwintered adults breed only after a reproductive diapause spent away from the water (Spence, 1989). After ice melt, mating begins in late May and continues to mid August depending upon whether summer generation reproductives are produced (Wilcox and Spence, 1986).

Limnoporus dissortis Drake & Harris prefers temporary ponds, and dispersal between ponds is a conspicuous feature of its life history (Spence and Scudder, 1980).

Temporary habitats seem to provide a refuge from egg parasitoids (Spence, 1986a). Eggs of *L. dissortis* are heavily parasitised by *T. gerriphagus*, and this and instability of their preferred habitats probably contribute to observed rates of local extinction (Spence, 1986a). Eggs of *L. dissortis* are easily recognized by their linear arrangement and conspicuous jelly coat (Spence and Wilcox, 1986), and it has been suggested that the latter may offer some protection against egg parasitoids (Martin, 1927; Andersen, 1982; Spence, 1986a).

Gerris buenoi Kirkaldy is a common, often abundant habitat generalist (Spence, 1981), and is the smallest of the water strider species found in western Canada (Spence, 1986b). Eggs are deposited in groups of one to four at the water surface on floating vegetation or below the surface on submerged or decaying vegetation, but most eggs are laid on floating vegetation (Spence, 1986a). *G. buenoi* eggs are not enveloped by a jelly coat and any protection against egg parasitoids is probably achieved by oviposition in sites inaccessible to wasps (Spence, 1986a).

Overwintered *L. dissortis* females are two to three times as fecund as those of any *Gerris* spp., but fecundity of direct breeders is more similar to that of other western Canadian species (Spence, 1989). Overall, *L. dissortis* is more fecund and has a longer reproductive life than *Gerris* spp. (Spence, 1989).

1.3 Hosts: gerrid eggs (Heteroptera)

Gerrid eggs are deposited on the underside edge of floating vegetation, or on submerged vegetation if floating vegetation cannot be found (Andersen, 1982). Eggs are glued lengthwise to the substrate and are surrounded by a gelatinous mass considered to be ancestral for Gerromorpha (Heteroptera) (Andersen, 1982). Eclosion of first instars is aided by an egg burster (ruptor ovi) which ruptures the egg shell in a median slit from the anterior pole to two-thirds posteriorly (Andersen, 1982).

1.4 Parasitoid: *Tiphodytes gerriphagus* Marchal

While collecting gerrid eggs from Trivaux pond, near Paris, France, Marchal (1900) discovered a solitary wasp developing within several eggs. It was a previously unknown scelionid, and Marchal (1900) published the first description of adults under the species name *Limnodytes gerriphagus* Marchal. Bradley (1902) pointed out that the genus name *Limnodytes* was preoccupied by a salamander described by Dumeril and Bibron (1841) and proposed the genus name *Tiphodytes* in its place.

Tiphodytes gerriphagus was first discovered to parasitise gerrid eggs in North America by Matheson and Crosby (1912), but no new information on adult behaviour or larval morphology was provided. Martin (1927) described two different oviposition methods used by *T. gerriphagus* involving either complete or partial submergence of the parasitoid. He also provided the first description of all immature stages of *T. gerriphagus* developing within eggs of *Trepobates* spp.

Tiphodytes gerriphagus appears to specialise on gerrid eggs. Hoffmann (1932) introduced various belostomatid, hydrometrid, gyrinid, corixid, and notonectid eggs to females, but no parasitism was observed. *Tiphodytes gerriphagus* successfully parasitises eggs of five *Gerris* and two *Limnoporus* species in western Canada (Spence, 1986a), and has been found to emerge from three different *Gerris* and one other *Limnoporus* species in Finland (Nummelin *et al.*, 1988). At present, *T. gerriphagus* is known to parasitise eggs of 13 species in three different genera (Table 1.1).

1.5 Thesis objectives

Scelionid development has been studied by several authors (McColloch and Yuasa, 1915; Chopard, 1923; Martin, 1927; Bakkendorf, 1934; Schell, 1943; Pickford, 1964; Safavi, 1968; Rothschild, 1970; Gerling, 1972; Ticehurst and Allen, 1973; Strand *et al.*, 1986; Navasero and Oatman, 1989; Volkoff and Colazza, 1992), and most studies involve parasitoids of economically important pests. In contrast, there is little information about development of scelionids that do not attack pest species.

Tiphodytes gerriphagus life stages have been described within *Trepobates* sp. eggs (Martin, 1927), but development time was not provided. Differences between first instars of *T. gerriphagus* as described by Marchal (1900) and Martin (1927) have been noted by Clausen (1940), Hagen (1956), and Masner (1972), but an explanation for the apparent contradictions has not been provided. The actual form of Martin's (1927) second instar has also been questioned (Clausen, 1940). Therefore, in chapter two I consider development and larval behaviour of *T. gerriphagus* within *L. dissortis* eggs, and present the first data about sex ratio allocation for this species.

Successful development of a parasitoid within a host is affected by host age, species, and whether the host has been previously parasitised. These conditions may alter host quality and can affect the fitness of an emerging parasitoid (Pak, 1986). In chapter three, I consider possible effects of egg quality on survival, body length and sex ratio allocation of *T. gerriphagus*.

In solitary parasitoids, only one individual can emerge successfully. Therefore, when two solitary parasitoids develop within a host (superparasitism), they must compete for resources and one or both will die (Salt, 1961). Supernumerary larvae of *T. gerriphagus* have been found within field collected gerrid eggs (Martin, 1927), but the implications of superparasitism for surviving wasps has not been examined. Thus, in chapter four I examine the effects of mating status and wasp density on frequency of superparasitism, parasitoid development, and the longevity and fecundity of the resulting wasp progeny.

In chapter five, I summarise conclusions of my studies, and discuss possible future research involving parasitoid development and fitness.

Table 1.1. Review of literature on successful parasitism of different gerrid eggs by *Tiphodytes gerriphagus*.

Gerrid species	Reference
Gerris	
G. buenoi	Spence 1986
G. comatus	Spence 1986
G. incognitus	Spence 1986
G. lacustris	Nummelin *et al.* 1988
G. odontogaster	Nummelin *et al.* 1988
G. paludum	Nummelin *et al.* 1988
G. pingreensis	Spence 1986
G. remigis	Matheson and Crosby 1912; Spence 1986
G. spp.	Marchal 1900
Limnoporus	
L. dissortis	Spence 1986
L. notabilis	Spence 1986
L. rufoscutellatus	Nummelin *et al.* 1988
Trepobates	
T. spp.	Martin 1927

1.6 LITERATURE CITED

Andersen, N. M. 1982. *The semiaquatic bugs (Hemiptera, Gerromorpha), phylogeny, adaptations, biogeography and classification.* Scandinavian Science Press, Klampenborg.

Bakkendorf, O. 1934. Biological investigations on some Danish Hymenopterous egg-parasites, especially in Homopterous and Heteropterous eggs, with taxonomic remarks and descriptions of new species. *Entomologiske Meddelelser* **19**: 1-134.

Borror, D. J., C. A. Triplehorn, and N. F. Johnson. 1989. *An introduction to the study of insects.* Harcourt Brace College Publishers, Florida.

Bradley, J. C. 1902. A recently discovered genus and species of aquatic hymenoptera. *The Canadian Entomologist* **34**: 179-180.

Burghele, A. 1959. New Rumanian species of Dachnusni (Hym.: Braconidae) and some ecological observations upon them. *Entomology Monthly Magazine* **95**: 121-126.

Chopard, L. 1923. Les parasites de la Mante Religieuse. *Annales de la Société Entomologique de France* **91**: 249-274.

Clausen, C. P. 1940. *Entomophagous insects.* McGraw-Hill, New York.

Dumeril, A. H. and G. Bibron. 1841. *Erpétologie générale: Ou histoire naturelle complète des reptiles* **8**. Roret, Paris.

Gerling, D. 1972. The developmental biology of *Telenomus remus* Nixon (Hymenoptera: Scelionidae). *Bulletin of Entomological Research* **61**: 385-388.

Godfray, H. C. J. 1994. *Parasitoids: behavioral and evolutionary ecology.* Princeton University Press, Princeton.

Hagen, K. S. 1956. Aquatic Hymenoptera. In R. L. Usinger, ed., *Aquatic insects of California.* pp. 289-292. University California Press, Berkeley.

Hagen, K. S. 1978. Aquatic Hymenoptera. In R. W. Merritt and K. W. Cummins, eds., *An introduction to aquatic insects of North America.* pp. 233-241. Kendall/Hunt, Dubuque.

Hedquist, K. J. 1967. Hymenoptera. In Joachim Illies, ed., *Limnofauna Europaea.* pp. 242-244. Verlag Gustav Fischer, Stuttgart.

Hoffmann, C. H. 1932. Hymenopterous parasites from the eggs of aquatic and semi-aquatic insects. *Journal of Kansas Entomological Society* **2**: 33-37.

Lubbock, J. 1862. On two aquatic Hymenoptera, one of which uses its wings in swimming. *Transactions of the Linneaen Society, London* **24**: 135-142.

Marchal, P. 1900. Sur un nouvel hyménoptère aquatique, le *Limnodytes gerriphagus*, n. gen., n. sp. *Annales de la Société Entomologique de France* **69**: 171-176.

Martin, C. H. 1927. Biological studies of two hymenopterous parasites of aquatic insect eggs. *Entomologica Americana, New Series* **7**: 105-156.

Masner, L. 1972. The classification and interrelationships of Thoronini (Hymenoptera: Proctotrupoidea, Scelionidae). *The Canadian Entomologist* **104**: 833-849.

Matheson, R. and Crosby, C. R. 1912. Aquatic Hymenoptera in America. *Annals of the Entomological Society of America* **5**: 65-71.

McColloch, J. W. and H. Yuasa. 1915. Further data on the life economy of the chinch bug parasite. *Journal of Economic Entomology* **8**: 248-261.

Navasero, R. C. and E. R. Oatman. 1989. Life history, immature morphology and adult behavior of *Telenomus solitus* (Hymenoptera: Scelionidae). *Entomophaga* **34**: 165-177.

Nummelin, M., J. R. Spence, and K. Vepsäläinen. 1988. Infection of gerrid eggs (Heteroptera: Gerridae) by the parasitoid *Tiphodytes gerriphagus* Marchal (Hymenoptera: Scelionidae) in Finland. *Annales Zoologici Fennici* **25**: 299-302.

Pak, G. A. 1986. Behavioural variations among strains of *Trichogramma* spp. A review of the literature on host age selection. *Journal of Applied Entomology* **101**: 55-64.

Pickford, R. 1964. Life history and behaviour of *Scelio calopteni* Riley (Hymenoptera: Scelionidae), a parasite of grasshopper eggs. *The Canadian Entomologist* **96**: 1167-1172.

Rothschild, G. H. L. 1970. Parasites of rice stemborers in Sarawak (Malaysian Borneo). *Entomophaga* **15**: 21-51.

Safavi, M. 1968. Étude biologique et écologique des hyménoptères parasites des oeufs des punaises des céréales. *Entomophaga* **13**: 381-495.

Salt, G. 1934. Experimental studies in insect parasitism. 1. Introduction and technique. *Royal Society of London Proceedings Series B* **114**: 450-476.

______. 1961. Competition among insect parasitoids. *Mechanisms in Biological Competition, Symposium of the Society for Experimental Biology* **15**: 96-119.

Schell, S. C. 1943. The biology of *Hadronotus ajax* Girault (Hymenoptera: Scelionidae), a parasite in the eggs of Squash-bug (*Anasa tristis* DeGeer). *Annals of the Entomological Society of America* **36**: 625-635.

Spence, J. R. 1981. Experimental analysis of habitat selection in waterstriders (Heteroptera: Gerridae). *Ecology* **62**: 1505-1514.

______. 1986a. Interactions between the scelionid egg parasitoid *Tiphodytes gerriphagus* (Hymenoptera) and its gerrid hosts (Heteroptera). *Canadian Journal of Zoology* **64**: 2728-2738.

______. 1986b. Relative impacts of mortality factors in field populations of the water strider *Gerris buenoi* Kirkaldy (Heteroptera: Gerridae). *Oecologia* **70**: 68-76.

______. 1989. The habitat template and life history strategies of pond skaters (Heteroptera: Gerridae): reproductive potential, phenology, and wing dimorphism. *Canadian Journal of Zoology* **67**: 2432-2447.

Spence, J. R. and G. G. E. Scudder. 1980. Habitats, life cycles and guild structure of water-striders (Heteroptera: Gerridae) on the Fraser Plateau of central British Columbia. *The Canadian Entomologist* **112**: 779-792.

Spence, J. R. and R. S. Wilcox. 1986. The mating system of two hybridizing species of water striders (Gerridae). II. Alternative tactics of males and females. *Behavioral Ecology and Sociobiology* **19**: 87-95.

Strand, M. R., S. M. Meola, and S. B. Vinson. 1986. Correlating pathological symptoms in *Heliothis virescens* eggs with development of the parasitoid *Telenomus heliothidis*. *Journal of Insect Physiology* **32**: 389-402.

Ticehurst, M. and D. C. Allen. 1973. Notes on the biology of *Telenomus coelodasidis* (Hymenoptera: Scelionidae) and its relationship to the saddled prominent, *Heterocampa guttivitta* (Lepidoptera: Notodontidae). *The Canadian Entomologist* **105**: 1133-1143.

Volkoff, N. and S. Colazza. 1992. Growth patterns of teratocytes in the immature stages of *Trissolcus basalis* (Woll.) (Hymenoptera: Scelionidae), an egg parasitoid of *Nezara viridula* (L.) (Heteroptera: Pentatomidae). *International Journal of Insect Morphology and Embryology* **21**: 323-336.

Ward, J. V. 1992. *Aquatic insect ecology: Biology and habitat.* 1. John Wiley and Sons, New York.

Wilcox, R. S. and J. R. Spence. 1986. The mating system of two hybridizing species of water striders (Gerridae). I. Ripple signal functions. *Behavioral Ecology and Sociobiology* **19**: 79-85.

2. DEVELOPMENT OF *Tiphodytes gerriphagus* Marchal (HYMENOPTERA: SCELIONIDAE) IN *Limnoporus dissortis* Drake & Harris EGGS (HETEROPTERA: GERRIDAE)

2.1 SYNOPSIS

Investigations of preimaginal development in *Tiphodytes gerriphagus* Marchal reveals two larval instars, based on counting exuviae and comparing mandible length during development within *Limnoporus dissortis* Drake & Harris eggs. *Tiphodytes gerriphagus* eggs are stalked, as is typical of scelionids, and are 282.6 ± 3.48 µm (mean ± *SE*) total length. Chorion rupture at 8-9 h post-oviposition (PO) releases a non-feeding embryo into the gerrid egg. Feeding begins at 18-20 h PO, after embryonic cuticle is shed and a fully differentiated and active larva is released. The first larval stage is teleaform which lasted from 18-120 h PO, and grew from 183.6 ± 3.35 µm to 517.0 ± 14.67 µm total length. The second larval stage is hymenopteriform which lasted from 114-312 h PO, and grew from 920.2 ± 24.65 µm to 1352.4 ± 11.89 µm total length before pupating. The pupal period lasted about 11 days with male pupae being shorter and thinner than female pupae. These findings are at odds with previous descriptions of *T. gerriphagus*, and reasons for this are discussed. The sex ratio under laboratory conditions was female-biased (22% males), and males were smaller but did not emerge significantly before females as anticipated from previous work.

2.2 INTRODUCTION

Scelionid wasps are egg parasitoids of Lepidoptera and Heteroptera (Waage, 1982), and there are over 2700 described species (Johnson, 1992). The genus *Tiphodytes* Bradley (1902) contains five described species worldwide, all of which are suspected to parasitise eggs of aquatic and semi-aquatic Heteroptera (Masner, 1972). *Tiphodytes gerriphagus* Marchal is a solitary egg parasitoid of water striders (Heteroptera: Gerridae), and was first accidentally discovered during field collections of *Gerris* Fabricius (1794) eggs for embryological studies (Marchal, 1900).

Marchal described adults of this newly discovered species, and also provided a drawing of the first instar larva. Martin (1927) more fully described the immature stages of *T. gerriphagus* to include egg, three larval instars and pupa, based on preserved specimens. However, data about development time, examination of exuviae, and observations of larval behaviour are not available. Published data about number of larval instars of scelionids varies between two (McColloch and Yuasa, 1915; Chopard, 1923; Bakkendorf, 1934; Pickford, 1964; Safavi, 1968; Gerling, 1972; Navasero and Oatman, 1989), and three (Martin, 1927; Schell, 1943; Rothschild, 1970; Ticehurst and Allen, 1973; Strand *et al.*, 1986; Volkoff and Colazza, 1992).

Sex ratio output (proportion of males) of many Hymenoptera is generally female-biased when hosts are abundant and competing foundresses are absent from the patch (Hamilton, 1967; Waage, 1982). Although *T. gerriphagus* has been collected and studied several times (Marchal, 1900; Matheson and Crosby, 1912; Martin, 1927; Hoffmann, 1932; Masner, 1972; Spence, 1986; Nummelin *et al.*, 1988; Henriquez and Spence, 1993), sex ratio output has not been reported.

In this work, I describe egg and larval development in *T. gerriphagus* within eggs of the natural host *Limnoporus dissortis* Drake & Harris (Heteroptera: Gerridae) and observations of larval behaviour and sex ratio output. Field observations suggest that superparasitism by *T. gerriphagus* is not uncommon (Martin, 1927), and developmental data of singly parasitised eggs is required to study potential interactions and effects within superparasitised eggs.

2.3 MATERIALS AND METHODS

2.3.1 Host and parasitoid cultures

Tiphodytes gerriphagus in *L. dissortis* eggs were collected from various ponds at George Lake Field Site of the University of Alberta near Dunstable, Alberta (114°06'W, 53°57'N) and used to start a culture. Emerging wasps were collected and the culture was maintained on *L. dissortis* eggs from a laboratory culture reared at 20 °C and 19 L: 5 D photoperiod. Styrofoam strips served as oviposition sites for gerrids and were collected daily to supplement host and parasitoid cultures (Henriquez and Spence, 1993).

2.3.2 *Tiphodytes gerriphagus* egg size

On day of emergence, individual female and male parasitoids were paired for mating inside 12 dram vials with 15 mL of water. After pairing for 24 h, the reproductive system of mated females was dissected in Ringer's solution (Barbosa, 1974). One parasitoid egg, from the base of a randomly selected ovariole, was removed, placed in a well-depression slide, and a coverslip was placed on top. Egg length and width were measured at × 25 and × 40 using a light microscope fitted with an ocular micrometer. The above procedure was repeated for 20 new females.

2.3.3 *Tiphodytes gerriphagus* development

Females were paired and mated as above and then groups of ten mated females were placed into 500 mL widemouth jars with 100 mL of water and 100 *L. dissortis* eggs (0-24 h old, at 20 °C and 19 L: 5 D). After one hour, all parasitoids were removed and date and time of the oviposition trial were recorded to serve as a reference point to determine *T. gerriphagus* age. Eggs were selected at random from within a widemouth jar every 6 h for the first 24 h, and then every 24 h thereafter. This was repeated 15 times for each time interval. Each egg was placed at the center of a well-depression slide and dissected with tungsten wire pins (Norton and Sanders, 1985) in Ringer's solution. Dissections were covered with a coverslip before observation and measurement. Only eggs found to contain a single viable immature wasp during dissection were used for data on development time, body length, mandible length, and body width. Wasp larvae were measured at × 25 and × 40 using a light microscope fitted with an ocular micrometer. Feeding activity and locomotion were video recorded using a Hitachi camera (VK-C350) mounted on a Polyvar light microscope (Reichert-Jung).

2.3.4 Development time, length and sex ratio of adult *Tiphodytes gerriphagus*

Ten mated females were placed individually into widemouth jars with 20 *L. dissortis* eggs (0-24 h old, at 20 °C and 19 L: 5 D) for 24 h. Parasitised eggs were checked every 24 h, starting at 15 days post-oviposition (PO), and adult wasps were preserved in 70 % ethanol upon emergence for subsequent measurement of maximum body length (frons to tip of last abdominal tergum) at × 50 with a dissecting microscope. Total sex ratio of progeny produced was recorded.

2.4 RESULTS

2.4.1 *Tiphodytes gerriphagus* egg size

Parasitoid eggs dissected from ovarioles are stalked (Plate 2.1A) and found floating freely within eggs. Total length is 282.6 ± 3.48 µm (mean ± *SE*; $n = 20$) with the stalk comprising 146.1 ± 2.21 µm ($n = 20$) of that length. Egg and stalk width are 74.9 ± 2.05 µm ($n = 20$) and 7.2 ± 0.15 µm ($n = 20$), respectively.

2.4.2 *Tiphodytes gerriphagus* development

Four to 5 h PO, a black, circular puncture wound, approximately 18 µm in diameter, can be found on the surface of parasitised eggs. The parasitoid egg appears to increase in size within the host until the chorion ruptures at 8-9 h PO. A spherical group of cells (blastosphere) about 110 µm in diameter passes out from the shriveled remains of the parasitoid egg. Surrounded by host contents, the embryo continues to differentiate and develop into an early first instar (Plate 2.1B). The hatching membrane (serosa) ruptures at 18-20 h PO, releasing an active, feeding larva.

First instars are teleaform, characterized by large mandibles, circumabdominal setae, a caudal horn with a tooth, and lack of body segmentation (Clausen 1940; Plate 2.1C, 2.1D, and 2.1E). Inspection of early eclosed first instars shows a constriction between head and thorax and thorax and abdomen. However, within a brief period of time, the constriction between thorax and abdomen disappeared, apparently due to expansion of cuticle with larval growth.

First instars move slowly within the host by combining body bending with movement of caudal horn and circumabdominal setae. The caudal horn, about 137 µm long, appears to tear host tissue and to direct food into the larva's mouth. Total body length and maximum abdominal width were 183.6 ± 3.35 µm (n = 15) and 73.4 ± 1.57 µm (n = 15), respectively. Mandibles of 24 h old larvae were 48.0 ± 0.45 µm (n = 15) in length (Figure 2.1).

First instars continue to feed for 3 days, increasing in length and width approximately 2.8 and 3.5 fold, respectively (Plate 2.1E; Table 2.1), but mandible length remains constant (Figure 2.1). Most size increase is in the abdominal region. External movements decrease in both range and duration as first instars age, and 4 day old first instars no longer raise their caudal horn toward their mandibles during feeding (Plate 2.1E). In contrast, internal movements increase as first instars age. For example, peristaltic contractions of the digestive tract occur quite frequently, mixing ingested host material and apparently aiding digestion. Contractions may begin at either end or in the center of the midgut, and sometimes two contractive movements may travel in opposite directions.

At 114-120 h PO, molting behaviour begins and the remains of the first exoskeleton are shed. Mandibles, circumabdominal setae, and caudal horn are cast off during this first larval molt, but tracheae were not found with this exuviae (Plate 2.1F). Second instars are hymenopteriform, and characterized by spherical or spindle-shaped body with visible segmentation (Clausen 1940). Larvae have 11 visible body segments (Plate 2.1F) and total body length and maximum abdominal width was 920.2 ± 24.65 μm ($n = 15$) and 311.2 ± 4.82 μm ($n = 15$), respectively.

Mandibles are not easily seen soon after the first larval molt (114-120 h PO), but can be observed and measured after they have sclerotised (*ca.* 144 h; Figure 2.1). They are directed cephalad, instead of caudad as in first instars, and are significantly smaller than in first instars (Figure 2.1; *ANOVA*: $F = 1004$; df = 5, 84; $P < 0.0001$).

Second instars align their antero-posterior axes with that of the egg, but their dorso-ventral orientation is the reverse. In contrast, highly mobile first instars have a random orientation as they consume the host. Second instars snugly fit the end of their abdomen into the posterior end of the host egg, and as they are nearly the size of the egg (Table 2.1), have limited movement to locate and ingest any remaining host material. As they continue to grow and extend in an anterior direction, the larvae wiggle back and forth to move any remaining food in front of themselves. Mandibles are 24.4 ± 0.34 μm ($n = 15$) in length and about 2 μm wide at the base (Figure 2.1), and show limited movement so food appears to be sucked into the oral opening rather than chewed.

Second instars attain full egg size between 156-162 h PO after the entire host has been consumed. This coincides with the first external sign of parasitism in *L. dissortis* eggs that can be seen without the aid of a microscope. At this point, alternating dark and light rings (*ca.* nine each) can be seen encircling the width of the egg. These rings are caused by the segmented, second instar cuticle pressing against the chorion. Before the appearance of these rings, parasitised gerrid eggs cannot be distinguished from infertile eggs unless an oviposition mark is discovered with the aid of a microscope.

Second instars remain the size of the host egg for about four more days and as the prepupal transition begins (10-11 d PO), the larvae begin to decrease in length and the meconium is voided. Molting behaviour begins at 12-13 d PO with shedding of the remains of the second larval exoskeleton (Plate 2.1G). Mandibles and tracheae can be seen with the exuviae (Plate 2.1H). Second instars require a functional tracheal system because larvae are no longer surrounded by host fluids and must obtain oxygen directly from air inside the host.

Unlike the two larval instars, pupae and adults are sexually dimorphic. Males have moniliform antennae (mn) while female antennae are clavate (cl) (Plate 2.1I), and male pupae are significantly smaller in total body length and maximum abdominal width than females (Table 2.2). Pupal cuticle is colourless and transparent, and a pharate adult within the pupal cuticle begins as entirely white and gradually becomes yellow with red ocelli and compound eyes. Approximately 17 days PO, pharate adults become black and ocelli and compound eyes become brown and then black.

After adult eclosion, they rotate so that they no longer face the substrate that their host is attached to. Adults chew a roughly circular emergence hole 372.7 ± 10.62 µm (*n* = 20) in diameter on the ventro-anterior surface of the egg. In contrast, unparasitised *L. dissortis* eggs have a slit along the ventral surface, about half of egg length, after a gerrid emerges. Adult parasitoids crawl through the emergence hole into water and swim with their wings to the surface. When they break through the surface film, they either walk on top of the water or fly away. *Tiphodytes gerriphagus* development is summarized in Figure 2.2.

2.4.3 Development time, length and sex ratio of adult *Tiphodytes gerriphagus*

Mated females produced progeny with a sex ratio of 0.22 males (*n* = 121). Males emerged at 22.7 ± 0.40 days (*n* = 27) and females at 23.6 ± 0.23 days (*n* = 94), but these differences are not statistically significant (Wilcoxon Rank Sum test: $\chi^2 = 1.90$; $P = 0.17$). Adult males are significantly smaller than females (Table 2.2; *t-test*: $t = 2.88$; df = 119; $P = 0.005$).

2.5 DISCUSSION

2.5.1 *Tiphodytes gerriphagus* description comparisons

My work with *T. gerriphagus* revealed two larval instars rather than three as Martin (1927) suggested. Martin (1927) described second instars as having mandibles larger than first instars, clumped circumabdominal setae, and a reduced caudal horn without a tooth. Using ecdysis to define larval instars (Wigglesworth, 1973; Fink, 1983), I found neither setae nor caudal horn on second instar larvae (Plate 2.1F), and mandibles of second instars were reduced (Figure 2.1). In studies of other scelionids providing data about development time, second instars also lack setae and caudal horn and have reduced mandibles (McColloch and Yuasa, 1915; Bakkendorf, 1934; Schell, 1943; Pickford, 1964; Safavi, 1968; Gerling, 1972; Strand *et al.*, 1986; Navasero and Oatman, 1989; Volkoff and Colazza, 1992).

Range of total body length of first and second instar larvae reported by Martin (1927) overlap with my first instar measurements of *T. gerriphagus*, and Martin's third instar measurements coincide with my measurements of the second and final larval instar (Figure 2.3a). I suggest that Martin's (1927) first and second instars are, in fact, both first instars measured at different ages, and his third instar describes the second instar of *T. gerriphagus*. First instars of other scelionids greatly increase in size before molting (McColloch and Yuasa, 1915; Bakkendorf, 1934; Schell, 1943, Pickford, 1964; Safavi, 1968; Gerling, 1972; Navasero and Oatman, 1989). Furthermore, lack of distinct segmentation in Martin's (1927) illustration of third (actually second) instars is probably due to measuring larvae while in the prepupal transition period.

The range of Martin's (1927) mandibular measurements for first and second instars is smaller than my measurements for first instar mandibles, but his measurements for the last instar correspond well with my second instar (Figure 2.3b). Martin (1927) measured mandibles from tip to base but this is imprecise because the base is arbitrary; in contrast, I measured first instar mandibles from their tip to the anterior bifurcation caused by insertion of the dorsal mandibular fibres (Figure 2.3c). This reference point is easily located in first instars of *T. gerriphagus* and other scelionids (Bakkendorf, 1934; Schell, 1943). Mandibular length appeared to increase as first instars of *Trichogramma australicum* Girault aged; however, injecting fluorescent dye and repeating measurements revealed that mandibular length was constant (Dahlan and Gordh, 1996). As time passes, mandibles become more sclerotised and easier to measure in larvae of *T. gerriphagus* and other egg parasitoids (Volkoff *et al.*, 1995; Dahlan and Gordh, 1996), and may appear larger if clear reference points are not used.

Immersing parasitised gerrid eggs in hot alcohol may have complicated Martin's (1927) study. The delicate exoskeletons of parasitoid larvae probably shriveled under this treatment leading to clumped setae and perhaps to the broken caudal horn observed by Martin (1927). Since live specimens were not studied by Martin (1927), he could not determine the cause of the appearing and disappearing motile rings (peristaltic gut contractions) inside the egg, nor observe how older larvae manage to manipulate food. Clausen (1940) questioned the true form of Martin's (1927) second instar larvae since some specimens had dorsal, plate-like thickenings and others not. These thickenings are not external, but are an impression of the brain outlined from a highly stretched exoskeleton of well-fed first instars (Plate 2.1E; Schell, 1943; Safavi, 1968).

2.5.2 Development time, length and sex ratio of adult *Tiphodytes gerriphagus*

Male scelionids generally emerge before females (Waage, 1982), but if observations are only done every 24 h, emergence may not be significantly different (Table 2.2). Regardless of equal food being available for progeny of both sexes, males of *T.*

gerriphagus are smaller than females (Spence, 1986). In my laboratory experiments, female-biased sex ratios (22% males) were produced when eggs were abundant and competing adults were absent (*i.e.* foundress number = 1). Female-biased sex ratios are theoretically predicted for many Hymenoptera with low foundress number per patch (Hamilton, 1967), and scelionids attacking egg batches of moderate size (*ca.* 20 to 50 eggs) produce female-biased sex ratios because one wasp can parasitise the entire batch (Waage, 1982).

2.5.3 Scelionid eggs

All scelionids studied to date have similar morphology and development of early stages. Stalked eggs occur in females of several superfamilies of Hymenoptera (Clausen, 1940) and has been the only egg type found in scelionids (McColloch and Yuasa, 1915; Martin, 1927; Schell, 1943; Pickford, 1964; Safavi, 1968; Rothschild, 1970; Gerling, 1972; Strand *et al.*, 1986; Navasero and Oatman, 1989; Volkoff and Colazza, 1992). Scelionid eggs increase in size during embryonic development (McColloch and Yuasa, 1915; Schell, 1943; Strand *et al.*, 1986; Volkoff and Colazza, 1992), probably from both differentiation and from diffusion of host material into the egg.

2.5.4 Scelionid development

Rupture of the parasitoid chorion does not immediately release an active larva but rather an early embryonic stage (blastosphere) still enveloped by serosal cuticle (Schell, 1943; Gerling, 1972; Strand *et al.*, 1986; Volkoff and Colazza, 1992). Such embryos can survive because they are surrounded by food and are protected from desiccation while within a healthy egg.

Rupture of the serosa releases an active feeding teleaform larva which is superficially divided into two distinct regions, usually a cephalothorax and abdomen (Clausen, 1940; Schell, 1943; Pickford, 1964; Strand *et al.*, 1986; Volkoff and Colazza, 1992) or head and fused thoracico-abdomen (Gerling, 1972). All first instar *T. gerriphagus* larvae, examined within one hour of eclosing from the serosa, had three distinct body regions: head, thorax, and abdomen. The 'neck' (= thorax) of first instars was not considered a distinct tagma in other scelionid studies, but a reason for this has not been provided. Description of only two body regions is probably due to both historical interpretation and dissection difficulties.

The term 'teleaform larva' comes from Ganin's (1869) description of a *Teleas* larva possessing only two body regions. Scelionid first instars are all defined as teleaform (Clausen, 1940), so a third body region was probably not looked for by investigators. Rapid change in body shape during development and long circumabdominal setae, which are about half the first instar's body length (Plate 2.1C), can hide a distinct third region. First instars do not have an active tracheal system (McColloch and Yuasa, 1915; Schell, 1943; Gerling, 1972; Strand *et al.*, 1986; Volkoff and Colazza, 1992) and respiration is believed to be cutaneous.

Accounts of second instar form and total number of larval instars differ among Scelionidae. Some authors have described two larval instars (McColloch and Yuasa, 1915; Chopard, 1923; Bakkendorf, 1934; Pickford, 1964; Safavi, 1968; Gerling, 1972;

Navasero and Oatman, 1989), and others three (Ganin, 1869; Martin, 1927; Schell, 1943; Rothschild, 1970; Ticehurst and Allen, 1973; Strand *et al.*, 1986; Volkoff and Colazza, 1992). Of those authors suggesting three larval instars, only Schell (1943), Strand *et al.* (1986), and Volkoff and Colazza (1992) successfully followed all life stages and provided development time. By not counting exuviae and observing obvious changes in larval morphology and behaviour, as documented with *T. gerriphagus*, instar number may be inflated.

Plate 2.1. Development of *Tiphodytes gerriphagus*. (A) Mature egg dissected from ovariole. (B) Newly released early embryo floating within *Limnoporus dissortis* egg. (C) Dorsal and (D) lateral view of 24 h old actively feeding first instar (se, setae). (E) 96 h first instar. (F) Dorsal view of second instar with shed exuviae of first instar (ex, exuviae; md, first instar mandibles). (G) Dorsal view of pupa. (H) Posterior view of pupa with shed exuviae of second instar (tr, tracheae). (I) Adult female and male (cl, clavate antennae; antennae; mn, moniliform antennae). Scale bar = 50 µm (A-G). Scale bar = 100 µm (H). Scale bar = 250 µm (I).

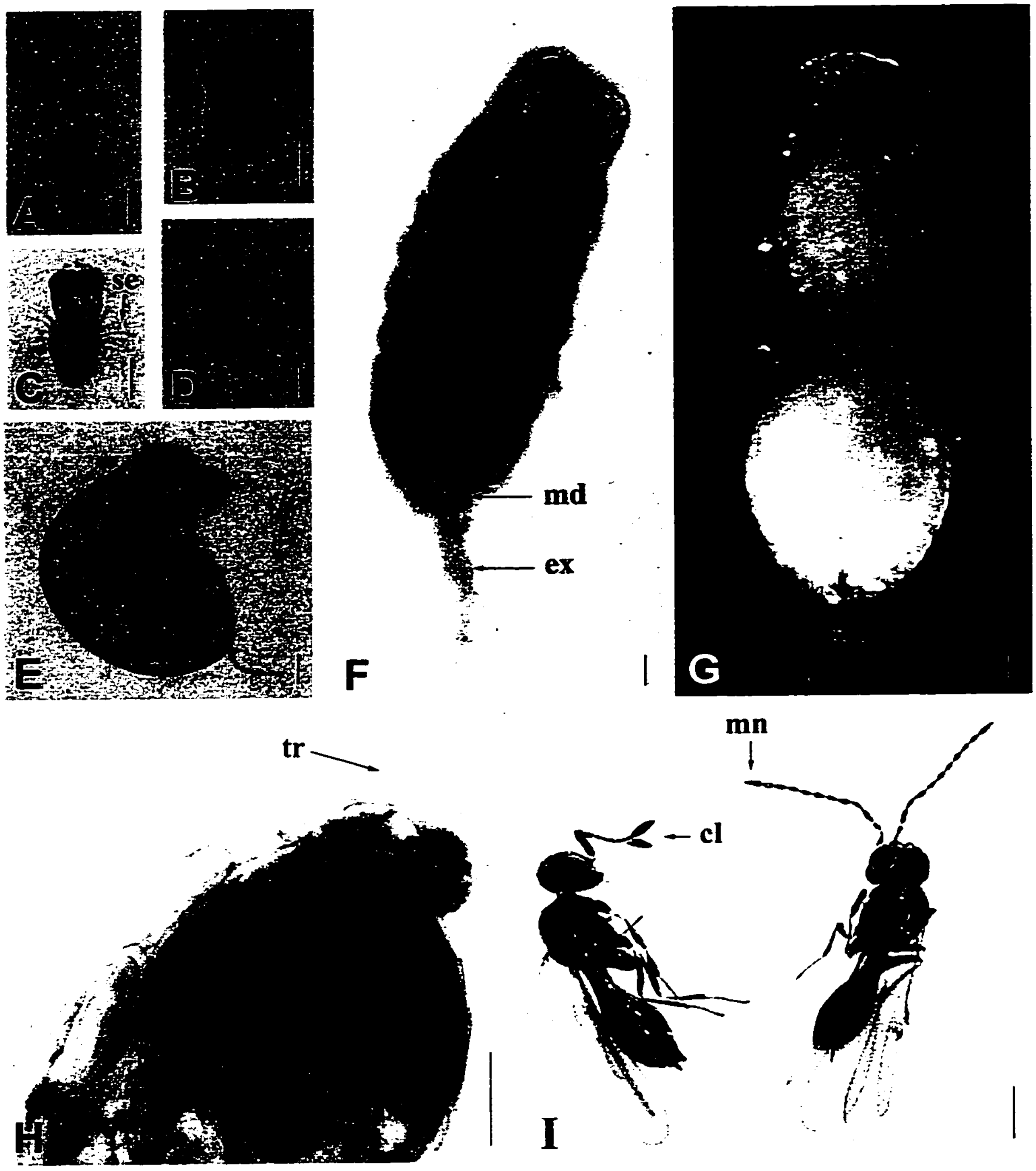
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Table 2.1. Mean body length and abdomen width of *Tiphodytes gerriphagus* larvae at daily intervals (*n* = 15/interval).

Hours after oviposition	Instar	Total body length (μm ± SE)	Abdomen width (μm ± SE)
24	first	183.6 ± 3.35	73.4 ± 1.57
48	first	227.4 ± 3.92	106.9 ± 2.09
72	first	357.2 ± 4.29	171.7 ± 3.01
96	first	517.0 ± 14.67	256.5 ± 5.07
120	second	920.2 ± 24.65	311.2 ± 4.82
144	second	1117.5 ± 16.57	411.9 ± 5.30
168*	second	1352.4 ± 11.89	486.7 ± 5.28

* *n* = 30.

Table 2.2. Mean body length and abdomen width of male and female *Tiphodytes gerriphagus* pupae and adults.

	Pupa			Adult	
Gender	*n*	Total body length (mm ± SE)	Abdomen width (mm ± SE)	*n*	Total body length (mm ± SE)
male	*15*	1.08 ± 0.019^{a}	0.36 ± 0.004^{a}	*27*	1.13 ± 0.005^{a}
female	*15*	1.14 ± 0.010^{b}	0.40 ± 0.005^{b}	*94*	1.16 ± 0.007^{b}

Values within a column followed by same letters are not significantly different (*t-test*; $P > 0.05$).

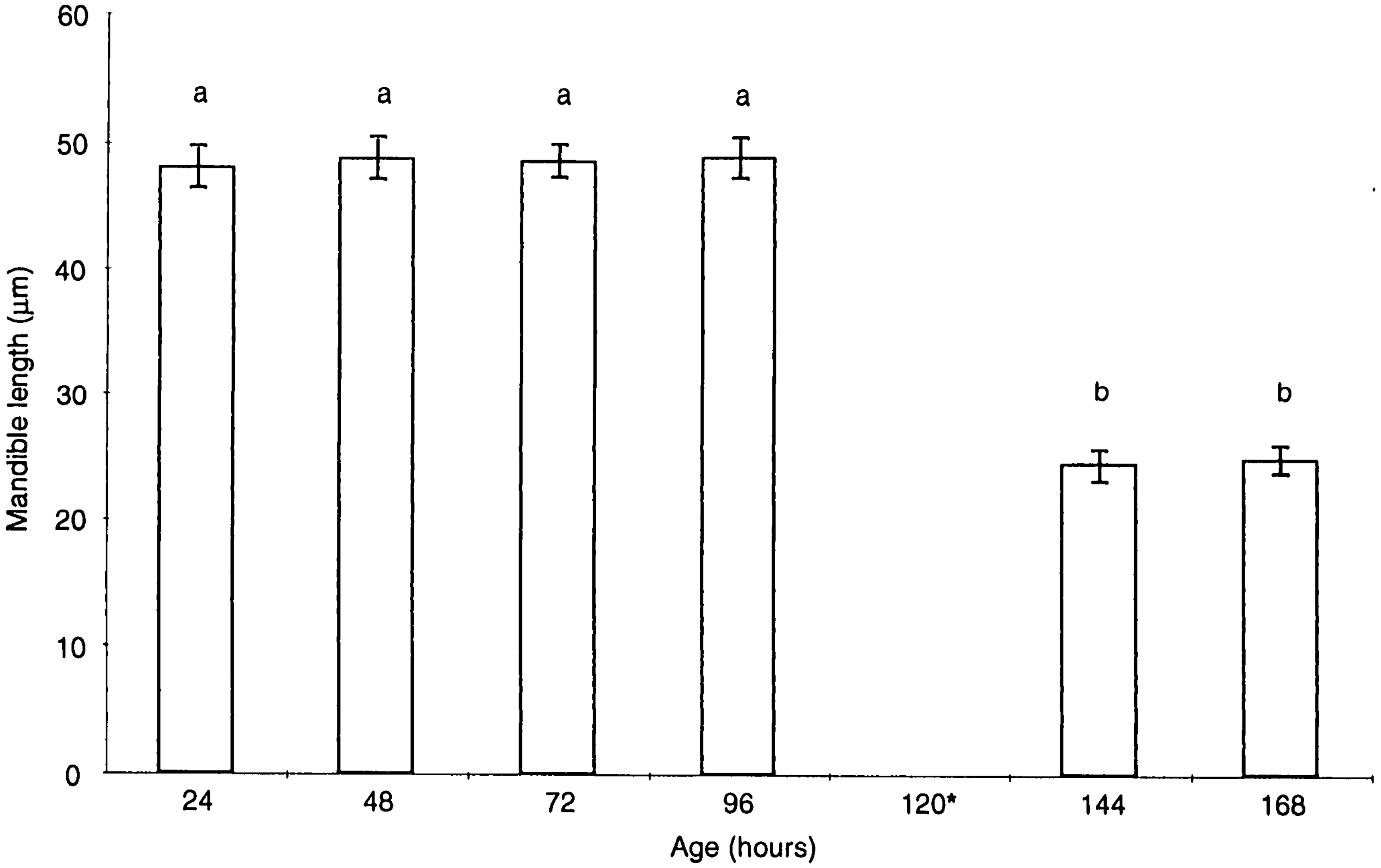


Figure 2.2. Mandibular length (mean ± *SE*) of *Tiphodytes gerriphagus* larvae at daily intervals (*n* = 15). * Mandibles are not sclerotized and difficult to measure. Means followed by same letters are not significantly different (Student-Newman-Keuls aposterior test).

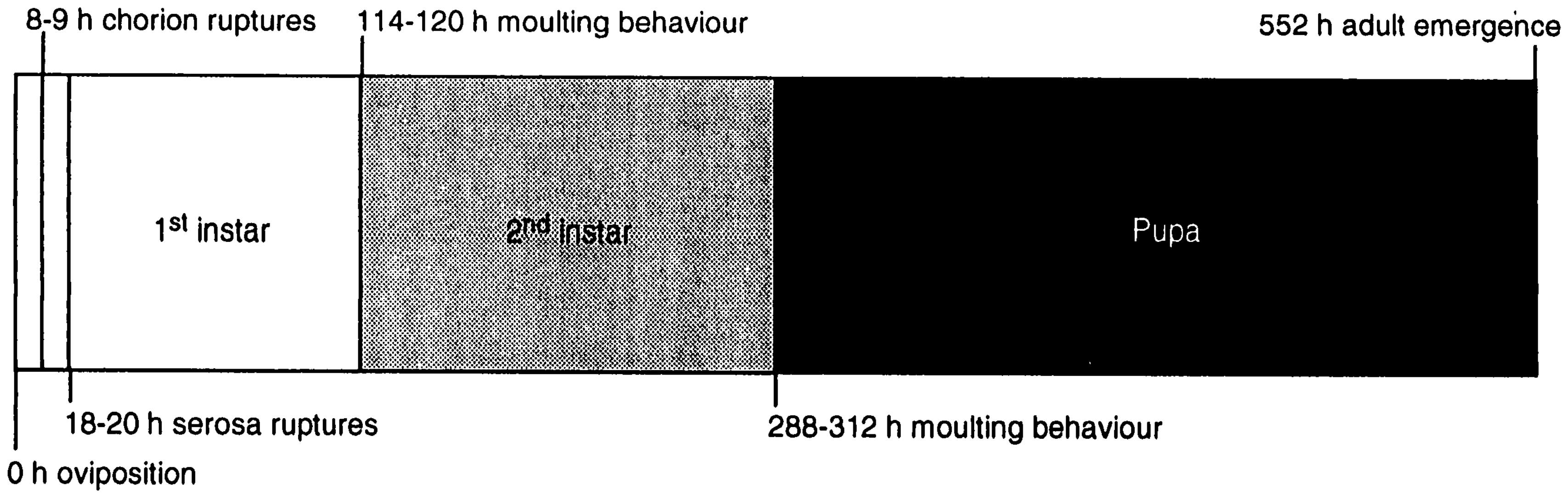


Figure 2.3. Temporal course of *Tiphodytes gerriphagus* development within *Limnoporus dissortis* eggs at 20 °C and 19 L : 5 D photoperiod. Major developmental features are demarcated.

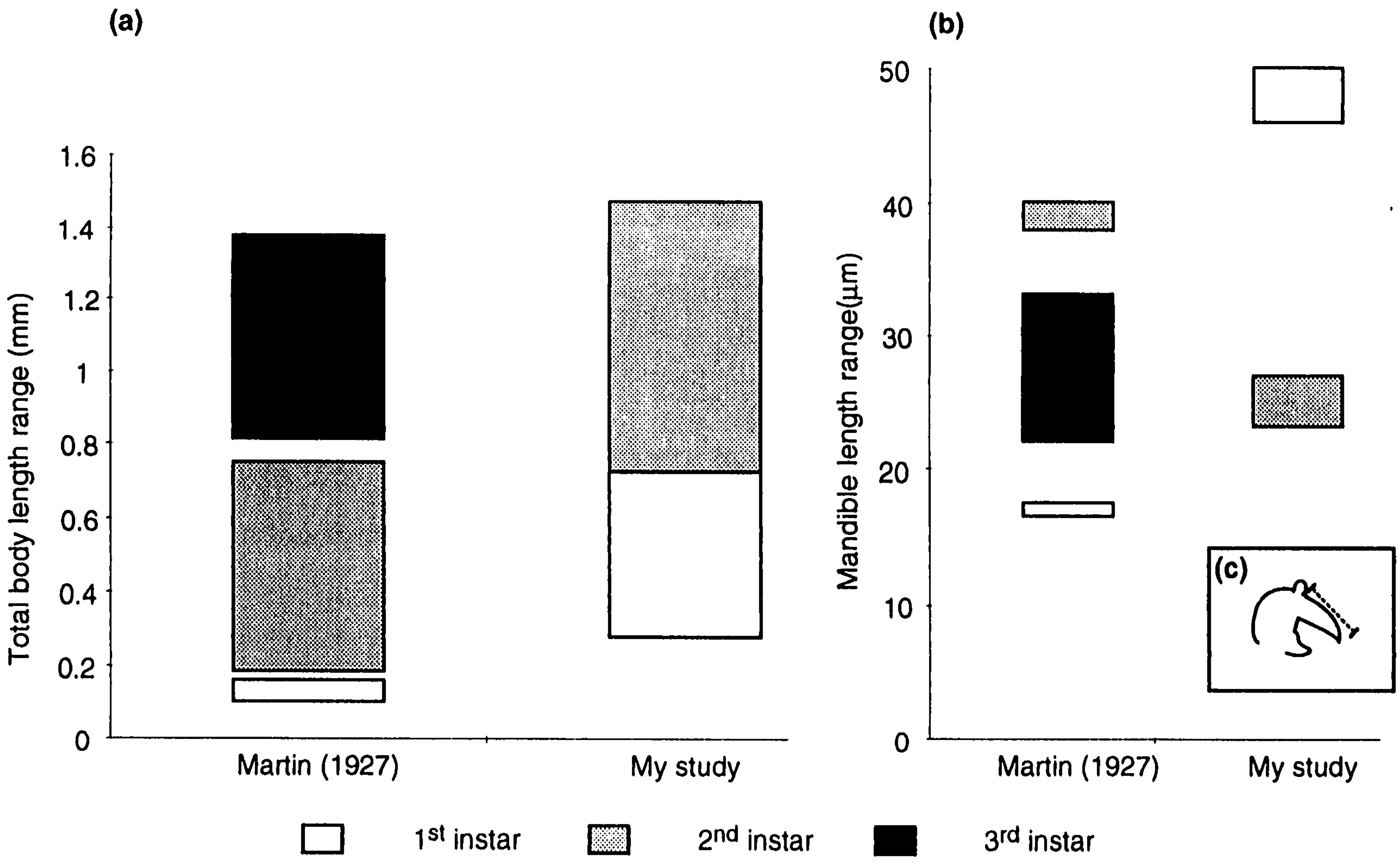


Figure 2.4. Comparison of *Tiphodytes gerriphagus* larval measurements. (a) Range of total body length, and (b) range of mandibular length according to Martin (1927) and my study. Inset (c) depicts measurement of first instar mandibles from dorsal mandibular fibres to tip.

2.6 LITERATURE CITED

Bakkendorf, O. 1934. Biological investigations on some Danish Hymenopterous egg-parasites, especially in Homopterous and Heteropterous eggs, with taxonomic remarks and descriptions of new species. *Entomologiske Meddelelser* **19**: 1-134.

Barbosa, P. 1974. Manual of basic techniques in insect histology. Autumn, Amherst.

Chopard, L. 1923. Les parasites de la Mante Religieuse. *Annales de la Société Entomologique de France* **91**: 249-274.

Clausen, C. P. 1940. *Entomophagous insects*. McGraw-Hill, New York.

Dahlan, A. N. and G. Gordh. 1996. Development of *Trichogramma australicum* Girault (Hymenoptera: Trichogrammatidae) on *Helicoverpa armigera* (Hübner) eggs (Lepidoptera: Noctuidae). *Australian Journal of Entomology* **35**: 337-344.

Fink, T. J. 1983. A further note on the use of the terms instar, stadium, and stage. *Annals of the Entomological Society of America* **76**: 316-318.

Ganin, M. 1869. Beiträge zur Erkenntniss der Entwickelungsgeschichte bei den Insekten. *Zeitschrift fur wissenschaftliche Zoologie* **19**: 381-451.

Gerling, D. 1972. The developmental biology of *Telenomus remus* Nixon (Hymenoptera: Scelionidae). *Bulletin of Entomological Research* **61**: 385-388.

Hamilton, W. D. 1967. Extraordinary sex ratios. *Science* **156**: 477-488.

Henriquez, N. P. and J. R. Spence. 1993. Host location by the gerrid egg parasitoid *Tiphodytes gerriphagus* Marchal (Hymenoptera: Scelionidae). *Journal of Insect Behavior* **6**: 455-466.

Hoffmann, C. H. 1932. Hymenopterous parasites from the eggs of aquatic and semi-aquatic insects. *Journal of Kansas Entomological Society* **2**: 33-37.

Johnson, N. F. 1992. Catalog of world species of Proctotrupoidea, exclusive of Platygasteridae. *Memoirs of the American Entomological Institute* **51**: 1-825.

Marchal, P. 1900. Sur un nouvel Hyménoptère aquatique, le *Limnodytes gerriphagus* N. Gen., N. Sp. *Annales de la Société Entomologique de France* **69**: 171-176.

Martin, C. H. 1927. Biological studies of two hymenopterous parasites of aquatic insect eggs. *Entomologica Americana, New Series* **8**: 105-157.

Matheson, R. and Crosby, C. R. 1912. Aquatic Hymenoptera in America. *Annals of the Entomological Society of America* **5**: 65-71.

Masner, L. 1972. The classification and interrelationships of Thoronini (Hymenoptera: Proctotrupoidea, Scelionidae). *The Canadian Entomologist* **104**: 833-849.

McColloch, J. W. and H. Yuasa. 1915. Further data on the life economy of the chinch bug parasite. *Journal of Economic Entomology* **8**: 248-261.

Navasero, R. C. and E. R. Oatman. 1989. Life history, immature morphology and adult behavior of *Telenomus solitus* (Hymenoptera: Scelionidae). *Entomophaga* **34**: 165-177.

Norton, R. and F. Sanders. 1985. Superior micro-needles for manipulating and dissecting soil invertebrates. *Quaestiones Entomologicae* **21**: 673-674.

Nummelin, M., J. R. Spence, and K. Vepsäläinen. 1988. Infection of gerrid eggs (Heteroptera: Gerridae) by the parasitoid *Tiphodytes gerriphagus* Marchal (Hymenoptera: Scelionidae) in Finland. *Annales Zoologici Fennici* **25**: 299-302.

Pickford, R. 1964. Life history and behaviour of *Scelio calopteni* Riley (Hymenoptera: Scelionidae), a parasite of grasshopper eggs. *The Canadian Entomologist* **96**: 1167-1172.

Rothschild, G. H. L. 1970. Parasites of rice stemborers in Sarawak (Malaysian Borneo). *Entomophaga* **15**: 21-51.

Safavi, M. 1968. Étude biologique et écologique des hyménoptères parasites des oeufs des punaises des céréales. *Entomophaga* **13**: 381-495.

Schell, S. C. 1943. The biology of *Hadronotus ajax* Girault (Hymenoptera: Scelionidae), a parasite in the eggs of Squash-bug (*Anasa tristis* DeGeer). *Annuls of the Entomological Society of America* **36**: 625-635.

Spence, J. R. 1986. Interactions between the scelionid egg *parasitoid Tiphodytes gerriphagus* (Hymenoptera) and its gerrid hosts (Heteroptera). *Canadian Journal of Zoology* **64**: 2728-2738.

Strand, M. R., S. M. Meola, and S. B. Vinson. 1986. Correlating pathological symptoms in *Heliothis virescens* eggs with development of the parasitoid *Telenomus heliothidis*. *Journal of Insect Physiology* **32**: 389-402.

Ticehurst, M. and D. C. Allen. 1973. Notes on the biology of *Telenomus coelodasidis* (Hymenoptera: Scelionidae) and its relationship to the saddled prominent, *Heterocampa guttivitta* (Lepidoptera: Notodontidae). *The Canadian Entomologist* **105**: 1133-1143.

Volkoff, N. and S. Colazza. 1992. Growth patterns of teratocytes in the immature stages of *Trissolcus basalis* (Woll.) (Hymenoptera: Scelionidae), an egg parasitoid of *Nezara viridula* (L.) (Heteroptera: Pentatomidae). *International Journal of Insect Morphology and Embryology* **21**: 323-336.

Volkoff, A., J. Daumal, P. Barry, M. Francois, N. Hawlitzky, and M. M. Rossi,. 1995. Development of *Trichogramma cacoeciae* Marchal (Hymenoptera: Trichogrammatidae): time table and evidence for a single larval instar. *International Journal of Insect Morphology and Embryology* **24**: 459-466.

Waage, J. K. 1982. Sib-mating and sex ratio strategies in scelionid wasps. *Ecological Entomology* **7**: 103-112.

Wigglesworth, V. B. 1973. The significance of "apolysis" in the molting of insects. *Journal of Entomology* **47**: 141-149.

3. EFFECTS OF EGG QUALITY AND PARASITOID DENSITY ON REPRODUCTIVE OUTPUT OF *Tiphodytes gerriphagus* Marchal (HYMENOPTERA: SCELIONIDAE)

3.1 SYNOPSIS

Laboratory studies were conducted to investigate the effects of egg quality and wasp density on mortality, adult body size and sex ratio of *Tiphodytes gerriphagus* Marchal, a parasitoid of water strider eggs. *Tiphodytes gerriphagus* is able to develop within *Limnoporus dissortis* Drake & Harris eggs that have undergone greater than 40 % of their embryological development. Parasitoid mortality was significantly lower from eggs 48-72 h old at 20 °C (4.7 %) than from those less than 24 h old (10.6 %). However, wasp body size did not change significantly with increasing egg age. Larval mortality was significantly higher when parasitoids were reared from the larger eggs of *L. dissortis* (6.3 %) than from the smaller eggs of *Gerris buenoi* Kirkaldy (2.2 %). As expected, larger wasps emerged from eggs of the larger host species. As foundress number was increased, parasitoid survival and body size significantly decreased. Sex ratio of progeny was independent of egg age, host species, and foundress number.

3.2 INTRODUCTION

Parasitoids use a variety of physical and chemical stimuli to locate patches with potentially suitable hosts in a heterogeneous environment (van Alphen and Vet, 1986). After locating a patch with hosts, a parasitoid may still not oviposit in any of the hosts encountered. Host selection is influenced by the physiological state (Salt, 1935) and experience of the parasitoid (Strand and Vinson, 1982), and by the quality of the host itself. Host quality is determined by such factors as age, size, species, and parasitism state (*i.e.*, parasitised vs. unparasitised), and these factors can affect the fitness of the emerging parasitoid (Pak, 1986).

Tiphodytes gerriphagus Marchal (Hymenoptera: Scelionidae) is a solitary egg parasitoid of several species of water striders (Heteroptera: Gerridae) (Chapter 1, Table 1.1; Marchal, 1900; Martin, 1927; Hoffmann, 1932; Spence 1986; Nummelin *et al.*, 1988; Henriquez and Spence, 1993a), and is the dominant parasitoid of field collected *Limnoporus* eggs in western Canada (Spence, 1986).

During host searching under natural conditions, most *T. gerriphagus* females will encounter gerrid eggs of various ages and species and these may differ in quality for a developing parasitoid. Host quality may also be affected by the number of female parasitoids (foundresses) in a patch because superparasitism is more likely when parasitoid to host ratio is high (van Alphen and Vet, 1986). In solitary parasitoids, host quality is dramatically affected by superparasitism because only one parasitoid can emerge successfully.

Female survival and reproductive output (fitness) are usually more sensitive to host quality than are those of males (Charnov *et al.*, 1981). It has thus been hypothesized that parasitoids will produce more male-biased sex ratios from hosts of lower quality (Charnov *et al.*, 1981). *Tiphodytes gerriphagus* parasitises greater than 80 % of *L. dissortis* eggs during mid-summer in western Canada (Spence, 1986), and the availability and quality of eggs probably fluctuate during the season. It is not known how gerrid egg quality affects *T. gerriphagus*. In this work, I investigate the effects of egg age, host species, and foundress number on *T. gerriphagus* fitness and sex ratio allocation.

3.3 MATERIALS AND METHODS

Adult *T. gerriphagus*, obtained from the University of Alberta George Lake Field Site, near Dunstable, Alberta during 1997, were used to start a laboratory culture. Eggs from laboratory reared *L. dissortis* served as the culture host, and both parasitoid and host cultures were maintained at 20 °C and a photoperiod of 19 L: 5 D. Newly oviposited *L. dissortis* eggs were collected daily from styrofoam oviposition strips and used to maintain the culture.

On day of adult emergence, individual female and male parasitoids were paired for mating inside 12 dram vials provided with 15 mL of water. After 24 h, the females were placed into 500 mL widemouth jars provided with 100 mL of water and eggs of a specified treatment. Each trial lasted 24 h, then the eggs were placed inside 12 dram vials with 15 mL of water until wasp emergence.

Three experiments were established, as described below, and a common set of observations were made on each experiment. I observed and recorded percent probing and parasitoid mortality, maximum body length, and total sex ratio (proportion of males). The external surface of eggs was searched for puncture wounds (probes) using a dissecting microscope for each replicate, and the mean percent of eggs probed was calculated for each treatment. All probed eggs not producing a wasp were counted among the total number attacked in calculations of percent parasitoid mortality. Maximum body length of emerging wasps was measured at × 50 using a dissecting microscope fitted with an ocular micrometer. The sex-specific median of body size of progeny of an individual female were compared among treatments with analysis of variance (*ANOVA*). Any wasps failing to emerge were not measured, but all male and female wasps (dead and alive) were counted and used to calculate total sex ratio (proportion of males). Any replicate producing less than five wasps identifiable to gender was not included in calculating total sex ratio for that treatment. Sex ratio as considered in this study is the observable proportion of male progeny (2°) as opposed to the actual sex ratio at oviposition (1°) (Godfray, 1994). However, assuming differential mortality to be low or negligible, 2° sex ratio approximates actual sex ratio.

3.3.1 Effects of *Limnoporus dissortis* egg age

An experiment was designed to test parasitoid ability to develop within *L. dissortis* eggs of various age. Eggs were grouped into the following age classes: 0-24, 24-48, 48-72, and 72-96 h, and 20 eggs of each age class were presented to a parasitoid for 24 h. Each age class treatment was replicated 15 times.

3.3.2 Effects of host species

Possible differences in egg quality between gerrid species and their effects on parasitoid fitness were tested using *L. dissortis* and *G. buenoi* eggs. Host eggs were randomly selected from gerrid cultures and measured for length, width, and dry mass.

Measurements were made at × 50 using a dissecting microscope fitted with an ocular micrometer. Eggs were also oven-dried for 24 h at 60 °C and weighed on a Cahn model 6926 electrobalance. Female wasps were presented with either 20 *L. dissortis* or *G. buenoi* eggs for 24 h. The experiment consisted of ten replicates with *L. dissortis* eggs and 15 replicates with those of *G. buenoi.*

3.3.3 Foundress number

To asses how wasp density might affect wasp fitness, 20 *L. dissortis* eggs were presented to one, three, or five female parasitoids for 24 h. The experiment consisted of ten replicates with the one and five parasitoids and nine replicates with three parasitoids.

3.4 RESULTS

3.4.1 Effects of *Limnoporus dissortis* egg age

Hosts of all age classes were equally probed during the 24 h interval (Table 3.1; *Kruskal-Wallis test*: $\chi^2 = 2.87$; df = 3; $P = 0.41$). However, parasitoid mortality decreased significantly as egg age increased (Table 3.1; $\chi^2 = 12.2$; df = 3; $P < 0.01$). Sex ratio did not differ significantly among age treatments (Table 3.1; $\chi^2 = 0.29$; df = 3; $P > 0.95$).

Male wasps emerging from eggs of each age class were significantly smaller than females, but wasp length did not change significantly as egg age increased (Table 3.1). Any effects of egg age were small, as indicated by the non-significant interaction term in the analysis (Table 3.2).

3.4.2 Effects of host species

Limnoporus dissortis eggs were significantly longer, wider, and heavier than *G. buenoi* eggs (Table 3.3). Eggs of both species were probed equally in no-choice experiments (Table 3.4; Kruskal-Wallis test: $\chi^2 = 0.09$; df = 1; $P = 0.76$), but parasitoid mortality was significantly greater in *L. dissortis* eggs (Table 3.4; $\chi^2 = 4.01$; df = 1; $P < 0.05$). Sex ratio did not differ significantly between hosts (Table 3.4; $\chi^2 = 0.47$; df = 1; $P > 0.25$).

Male wasps were significantly smaller than females from eggs of both host species (Table 3.5), and larger wasps emerged from the larger *L. dissortis* eggs (Table 3.4).

3.4.3 Foundress number

The proportion of eggs probed did not differ significantly among parasitoid density classes (Table 3.6; *Kruskal-Wallis test*: $\chi^2 = 3.40$; df = 2; $P = 0.18$), but mean percent probing appeared to be higher for three and five wasps (Table 3.6). Parasitoid mortality increased significantly as foundress number increased (Table 3.6; $\chi^2 = 5.99$; df = 2; $P < 0.05$), but sex ratio did not differ between foundress treatments (Table 3.6; $\chi^2 = 0.75$; df = 2; $P > 0.25$).

Male wasps were significantly smaller than females from eggs exposed to densities of one or three wasps, but male and female length did not differ significantly in the five wasp density treatment (Table 3.6). Wasp length decreased significantly as wasp density increased (Table 3.6). Body size of parasitoids of both sexes decreased equally as parasitoid : host ratio increased (Table 3.7).

3.5 DISCUSSION

3.5.1 Effects of *Limnoporus dissortis* egg age

Development of *L. dissortis* from oviposition to first instar requires approximately nine days at 18 °C (Spence, 1986), and *T. gerriphagus* oviposits into and successfully develops from eggs that have progressed through greater than 40% of embryogenesis (Table 3.1; Spence, 1986). Unfortunately, it is not known which age precludes parasitoid development because egg dissections were not done in these experiments. As an insect egg develops, its contents change from a liquid yolk to a young insect with cellular tissue and a chitinous exoskeleton. This makes searching for immature parasitoids difficult in older eggs. However, scelionids are generally able to parasitise and successfully emerge from eggs of almost any stage of development (Jubb and Watson, 1970; Rabinovich, 1970; Fedde, 1977; Harris, 1977; Le Pelley, 1979; Strand and Vinson, 1983a; Austin, 1984; Spence, 1986; Strand, 1986; Strand *et al.*, 1986; Ohno, 1987; Navasero and Oatman, 1989; Nechols *et al.*, 1989). Avoidance of older eggs has been reported for certain phoretic scelionids (Clausen, 1976; Orr *et al.*, 1986) probably because eggs become inaccessible as time progresses due to hardening of protective egg coatings.

Percentage probing did not differ significantly among the age classes tested perhaps because of the limited egg batch size provided during experiments (20 gerrid eggs). A female of *T. gerriphagus* is able to parasitise greater than 30 gerrid eggs on its first day of oviposition (Chapter 4). A difference in percent probing might be seen if a wider range of egg age classes were provided with more than 20 eggs per replicate. Mean percent probing was highest for the oldest age class of eggs tested (72-96 h). However, this probably does not indicate a preference for older eggs, but rather results from breakdown of the protective jelly coat surrounding *L. dissortis* eggs and perhaps to increased visibility of older eggs. Jelly coats are produced by ovarian follicular cells of gerrids (Anderson, 1982) and are present on all eggs of *Limnoporus* spp. (Spence, 1986). Jelly coats may offer eggs some protection against desiccation, parasitoids, and predators for several days after oviposition (Martin, 1927; Anderson, 1982; Spence, 1986). However, degradation of the jelly coat in older eggs probably decreases handling time and allows *T. gerriphagus* to parasitise more eggs.

As *L. dissortis* eggs develop, they change in colour from white, to gray, to yellow, and finally to grayish-black before hatching. Apparently, *T. gerriphagus* locates egg patches by visually searching for vegetation and adult gerrids (Henriquez and Spence, 1993a), and it is possible that they may locate eggs within a patch by also using visual cues. Since gerrid eggs are oviposited underwater, on the underside edge of floating vegetation (Andersen, 1982), it is certain that wasps find eggs initially without referring to colour. Also egg colour has been found to be unimportant in host selection by the scelionid *Telenomus heliothidis* (Strand and Vinson, 1983a).

As in other scelionids (Rabinovich, 1970; Gerling and Schwartz, 1974; Strand and Vinson, 1983a; Navasero and Oatman, 1989), the highest egg-adult survival of *T. gerriphagus* was observed from eggs of intermediate age at the time of oviposition. This could result from the disproportionate action of different mortality factors on young and old eggs. Parasitoid eggs oviposited into young eggs (< 25 % development) are at risk of being placed into infertile eggs. Several scelionids do not develop normally from infertile insect eggs (Egwuatu and Taylor, 1977; Fedde, 1977; Strand and Vinson, 1983a) because they lack a serosal endocuticle which is apparently important in water regulation (Strand, 1986). Scelionids generally survive better in young infertile eggs compared to old infertile eggs because a parasitoid can develop into a pupa and resist desiccation if oviposited early enough into an egg (Strand, 1986). However, fertile eggs yield higher parasitoid survival than infertile eggs (Strand, 1986). Healthy *L. dissortis* eggs less than 48 h old are impossible to distinguish visually from infertile gerrid eggs because both are white. Since viable eggs were preferentially selected for experiments based on normal egg colour, it is reasonable to assume that a larger proportion of infertile eggs were included in the earlier age classes tested (0-24 h and 24-48 h) because such eggs are all white. I hypothesize that *T. gerriphagus* mortality is greater in infertile eggs resulting in greater death in earlier ages.

Parasitoid larvae developing from older eggs (> 50 % development) may face quantitative and, possibly, qualitative shortages in nutrition. As an egg ages, less digestable material is available for the parasitoid larva because nutrients such as free amino acids are incorporated into host tissues (Barrett and Schmidt, 1991) and host cuticle is not readily digested (Austin, 1984; Strand *et al.*, 1986).

Tiphodytes gerriphagus can successfully emerge from eggs greater than 120 h old (Spence, 1986), and the range of ages tested here (0 - 96h) may not have differed enough in quality and quantity of resources to affect development and size of parasitoids, nor to stimulate a change in sex ratio allocation (*i.e.*, more males in older hosts). However, sex ratio is constant among different aged eggs with other scelionids (Rabinovich, 1970; Navasero and Oatman, 1989; Nechols *et al.*, 1989; Noda and Hirose, 1989).

3.5.2 Effects of host species

Even though the jelly coat of *L. dissortis* eggs appears to interfere with *T. gerriphagus* oviposition (Martin, 1927; Spence, 1986), I found no difference in number off eggs probed between *L. dissortis* or *G. buenoi.* I used only 20 eggs for each treatment, and if a large number (> 50) of *L. dissortis* or *G. buenoi* eggs were provided to a wasp, more *G. buenoi* may be parasitised because they lack a jelly coat. Sticky exudates from tobacco leaves have been shown to decrease tobacco hornworm egg parasitism by the scelionid *Telenomus sphingis* (Rabb and Bradley, 1968).

Successful pupation and emergence of egg parasitoids requires a nearly dry host interior (Strand and Vinson, 1985). The scelionid *Telenomus lobatus* had greater mortality in large chrysopid eggs (Ruberson *et al.*, 1989) and the trichogrammatid *Lathromeroidea* sp. nova had poorer emergence from *L. dissortis* eggs as compared to *G. buenoi* eggs (Henriquez and Spence, 1993b). It is suspected that excess moisture or host material may have caused greater mortality of *T. gerriphagus* developing within *L. dissortis* eggs.

As shown here and in Spence (1986), larger wasps emerged from larger eggs. However, contrary to expectations, there was no concomitant change in sex ratio. Theory suggests that a more female-biased sex ratio should emerge from larger hosts (Charnov *et al.*, 1981), but this has not been demonstrated with solitary scelionids (Ruberson *et al.*, 1989). Scelionids appear to have evolved a fixed sex ratio allocation strategy in which the number of eggs parasitised determines sex ratio (Waage, 1982; van Welzen and Waage, 1987; Braman and Yeargan, 1989; Noda and Hirose, 1989; Weber *et al.*, 1996). As long as parasitism and emergence success is high, it appears that sex ratio allocation is independent of specific egg attributes such as size.

3.5.3 Foundress number

Several scelionids guard small egg masses to limit oviposition by other foundresses (Hokyo and Kiritani, 1966; Waage, 1982), but *T. gerriphagus* females have not been observed to guard egg masses. In the field, many *L. dissortis* oviposit on floating vegetation during the summer breeding season, and egg mass size per leaf fluctuates widely. Field collected *L. dissortis* egg batches greatly exceed the potential daily oviposition rate of *T. gerriphagus* early in the season (Spence, 1986). Lack of aggressive behaviour is expected for a parasitoid that commonly encounters large egg masses because it would be disadvantageous to waste time chasing away other foundresses without a corresponding increase in fitness (Hokyo and Kiritani, 1966; Rothschild, 1970). After mid-July, *L. dissortis* egg availability declines rapidly (Spence, 1986). This may increase the probability of interaction between female *T. gerriphagus* at oviposition sites because the parasitoid population would be larger and alternate patch choice would be limited. However, it is unlikely that aggressive behaviour would increase as egg batch size decreases because of the cost of defending a patch from large numbers of wasps. Aggressive behaviour is most likely to involve interactions between larvae within a superparasitised egg.

Parasitoid mortality appeared to increase with increasing foundress number, but since eggs were not dissected, actual oviposition could not be determined; therefore, some probed eggs may not have contained a parasitoid because of host rejection or parasitoid interference. If a scelionid is interrupted during oviposition, the host may not hatch even though a parasitoid egg is not deposited (Strand *et al.*, 1986). Though aggressive behaviour was not observed, accidental interruptions during oviposition may have occurred which would result in some eggs being probed but not parasitised.

When two solitary parasitoids are placed into a host (superparasitism), they must compete for resources and one or both parasitoids will die (Salt, 1961). Before larvae find each other within a superparasitised host, some of the host may be consumed. This may lead to less food being available for the surviving parasitoid and result in smaller wasps emerging (Table 3.6; Strand *et* al., 1988).

Sex ratio (proportion of males) is predicted to increase as foundress number increases within a patch (Hamilton, 1967). However, Waage (1982) argued that small egg masses (< 50 eggs) could be monopolised by one foundress so that sex ratio would not increase. Though sex ratio did not increase with increasing foundress number in this study, it is not because the egg mass was being monopolised by one foundress. Small egg masses may be attacked by more than one foundress without any noticeable change in behaviour nor sex allocation (Strand and Vinson, 1983b; van Welzen and Waage, 1987; Braman and Yeargan, 1989). The sex ratio of superparasitising scelionids is not significantly different from wasps ovipositing into unparasitised eggs as long as they are provided equal egg batch sizes (van Welzen and Waage, 1987). The fixed sex ratio allocation strategy of scelionids (Waage, 1982; van Welzen and Waage, 1987; Braman and Yeargan, 1989; Noda and Hirose, 1989; Weber *et al.*, 1996) may prevent superparasitising wasps from increasing the percentage of males they oviposit.

Tiphodytes gerriphagus has been known to parasitise gerrid eggs since the beginning of this century (Marchal 1900), but only recently have studies focused on parasitoid behaviours and life history (Spence, 1986). General trends of parasitoid fitness and sex allocation in relation to egg quality are quickly and easily attained by indirect methods of observation as outlined above. However, to obtain a better knowledge of host preference, host selection and primary sex ratio allocation, direct observations of parasitoid behaviour are still necessary (van Dijken *et al.*, 1986).

Table 3.1. Effect of egg age on parasitism, parasitoid mortality, maximum body length of emerging wasps, and sex ratio based on 15 replicates, each consisting of 20 *Limnoporus dissortis* eggs exposed to a mated female of *Tiphodytes gerriphagus* for 24 h.

Host age (h)	% eggs probed ± SE*	% parasitoid mortality†	Maximum length (mm ± *SE*)‡		Sex ratio (% male)†
			male	female	
0-24	75.7 ± 6.17^{a}	10.6^{ab}	1.13 ± 0.008^{ab}	1.17 ± 0.010^{a}	22.8^{a}
24-48	78.4 ± 6.44^{a}	13.0^{a}	1.14 ± 0.010^{a}	1.16 ± 0.008^{a}	22.7^{a}
48-72	77.3 ± 7.38^{a}	4.7^{c}	1.12 ± 0.007^{ab}	1.15 ± 0.007^{a}	23.4^{a}
72-96	90.7 ± 1.88^{a}	6.9^{bc}	1.11 ± 0.007^{b}	1.15 ± 0.008^{a}	21.5^{a}

Values within a column followed by same letters are not significantly different, $P > 0.05$.

* *Kruskal-Wallis test*

† χ^2 *test*

‡ *Fisher's least significant difference test*

Table 3.2. Results of two-way analysis of variance on measures of *Limnoporus dissortis* egg age and body length of emerging male and female *Tiphodytes gerriphagus*.

Source	df	Mean square	*F*	*P*
Host age	3	3.54×10^{-3}	3.45	< 0.05
Wasp gender	1	2.94×10^{-2}	28.57	< 0.001
Age x gender	3	4.99×10^{-4}	0.48	0.693
Residual	111	1.03×10^{-3}		

Table 3.3. Average egg length, width, and dry mass of host species.

Host species	Egg Length (mm ± *SE*)	Egg Width (mm ± *SE*)	Egg Dry mass (μg ± *SE*)
Gerris buenoi	1.11 ± 0.007^a (30)*	0.40 ± 0.003^a (30)	56.74 ± 0.797^a (43)
Limnoporus dissortis	1.35 ± 0.012^b (30)	0.49 ± 0.005^b (30)	69.30 ± 1.274^b (40)

Values within a column followed by same letters are not significantly different (*t-test*; $P > 0.05$).

* Numbers in parentheses represent sample sizes.

Table 3.4. Effect of host species on percent parasitism, parasitoid mortality, maximum body length of emerging wasps, and sex ratio based on replicates, each consisting of 20 eggs exposed to a mated female of *Tiphodytes gerriphagus* for 24 h.

Host species	*n*	% eggs probed ± SE*	% parasitoid mortality†	Maximal length (mm ± *SE*)‡ male	Maximal length (mm ± *SE*)‡ female	Sex ratio (% male)†
Gerris buenoi	*15*	75.3 ± 6.57^a	2.2^a	1.03 ± 0.011^a	1.07 ± 0.008^a	23.5^a
Limnoporus dissortis	*10*	72.0 ± 9.20^a	6.3^b	1.17 ± 0.012^b	1.22 ± 0.008^b	26.7^a

Values within a column followed by same letters are not significantly different, $P > 0.05$.

* *Kruskal-Wallis test*

† χ^2 *test*

‡ *Fisher's least significant difference test*

Table 3.5. Results of two-way analysis of variance on measures of gerrid egg species and body length of emerging male and female *Tiphodytes gerriphagus*.

Source	df	Mean square	*F*	*P*
Host species	1	2.35×10^{-1}	199.40	< 0.0001
Wasp gender	1	1.67×10^{-2}	14.15	< 0.001
Species x gender	1	1.64×10^{-4}	0.14	0.711
Residual	42	1.18×10^{-3}		

Table 3.6. Effect of wasp density on parasitism, parasitoid mortality, maximum body length of emerging wasps, and sex ratio based on replicates, each consisting of 20 *Limnoporus dissortis* eggs exposed to a group of mated female of *Tiphodytes gerriphagus* for 24 h.

Wasp density	*n*	% eggs probed ± SE*	% parasitoid mortality†	Maximum length (mm ± SE)‡		Sex ratio† (% male)
				male	female	
1	*10*	72.0 ± 9.20^{a}	6.3^{a}	1.17 ± 0.012^{a}	1.22 ± 0.008^{a}	26.7^{a}
3	*9*	88.3 ± 7.07^{a}	10.7^{ab}	1.16 ± 0.022^{a}	1.20 ± 0.011^{a}	31.0^{a}
5	*10*	83.6 ± 5.38^{a}	14.9^{b}	1.13 ± 0.006^{b}	1.15 ± 0.008^{b}	30.5^{a}

Values within a column followed by same letters are not significantly different, $P > 0.05$.

* *Kruskal-Wallis test*

† χ^2 *test*

‡ *Fisher's least significant difference test*

Table 3.7. Results of two-way analysis of variance on measures of wasp density and body length of emerging male and female *Tiphodytes gerriphagus*.

Source	df	Mean square	F	P
Wasp density	2	1.65×10^{-2}	12.36	< 0.0001
Wasp gender	1	1.85×10^{-2}	13.88	< 0.001
Density x gender	2	4.76×10^{-4}	0.36	0.701
Residual	51	1.33×10^{-3}		

3.6 LITERATURE CITED

van Alphen, J. J. M. and L. E. M. Vet. 1986. An evolutionary approach to host finding and selection. In J. K. Waage and D. Greathead, eds., *Insect parasitoids*. pp. 23-61. Academic Press, London.

Andersen, N. M. 1982. *The semiaquatic bugs (Hemiptera, Gerromorpha), phylogeny, adaptations, biogeography and classification.* Scandinavian Science Press, Klampenborg.

Austin, A. D. 1984. The fecundity, development and host relationships of *Ceratobaeus* spp. (Hymenoptera: Scelionidae), parasites of spider eggs. *Ecological Entomology* **9**: 125-138.

Barrett, M. and J. M. Schmidt. 1991. A comparison between the amino acid composition of an egg parasitoid wasp and some of its hosts. *Entomologia Experimentalis et Applicata* **59**: 29-41.

Braman, S. K. and K. V. Yeargan. 1989. Reproductive strategy of *Trissolcus euschisti* (Hymenoptera: Scelionidae) under conditions of partially used host resources. *Annals of the Entomological Society of America* **82**:172-176.

Charnov, E. L., R. L. Los-den Hartogh, W. T. Jones, and J. van den Assem. 1981. Sex ratio evolution in a variable environment. *Nature* **289**: 27-33.

Clausen, C. P. 1976. Phoresy among entomophagous insects. *Annual Review of Entomology* **21**: 343-368.

van Dijken, M. J., M. Kole, J. C. van Lenteren, and A. M. Brand. 1986. Host-preference studies with *Trichogramma evanescens* Westwood (Hym., Trichogrammatidae) for *Mamestra brassicae*, *Pieris brassicae* and *Pieris rapae*. *Journal of Applied Entomology* **101**: 64-85.

Egwuatu, R. I. and T. A. Taylor. 1977. Development of *Gryon gnidus* Nixon (Hymenoptera: Scelionidae) in eggs of *Acanthomia tomentosicollis* Stål (Hemiptera: Coreidae) killed by either gamma irradiation or by freezing. *Bulletin of Entomological Research* **67**: 31-33.

Fedde, G. F. 1977. A laboratory study of egg parasitization capabilities of *Telenomus alsophilae*. *Environmental Entomology* **6**: 773-776.

Gerling, D. and A. Schwartz. 1974. Host selection by *Telenomus remus*, a parasite of *Spodoptera littoralis* eggs. *Entomologia Experimentalis et Applicata* **31**: 391-396.

Godfray, H. C. J. 1994. *Parasitoids: behavioral and evolutionary ecology*. Princeton University Press, Princeton.

Hamilton, W. D. 1967. Extraordinary sex ratios. *Science* **156**: 477-488.

Harris, M. K. 1977. Egg parasites of the walnut caterpillar in Texas. *The Southwestern Entomologist* **2**: 170-178.

Henriquez, N. P. and J. R. Spence. 1993a. Host location by the gerrid egg parasitoid *Tiphodytes gerriphagus* (Marchal) (Hymenoptera: Scelionidae*)*. *Journal of Insect Behavior* **6**: 455-466.

______. 1993b. Studies of *Lathromeroidea* sp. nov. (Hymenoptera: Trichogrammatidae), a parasitoid of gerrid eggs. *The Canadian Entomologist* **125**: 693-702.

Hoffmann, C. H. 1932. Hymenopterous parasites from the eggs of aquatic and semi-aquatic insects. *Journal of Kansas Entomological Society* **2**: 33-37.

Hokyo, N. and K. Kiritani. 1966. Oviposition behaviour of two egg parasites, *Asolcus mitsukurii* Ashmead and *Telenomus nakagawai* Watanabe (Hymenoptera: Scelionidae). *Entomophaga* **11**: 191-201.

Jubb, G. L. and T. F. Watson. 1970. Parasitization capabilities of the pentatomid egg parasite *Telenomus utahensis* (Hymenoptera: Scelionidae). *Annals of the Entomological Society of America* **64**: 452-456.

Le Pelley, R. H. 1979. Some scelionid egg-parasites reared from coffee bugs (*Antestiopsis*) and from some unusual pentatomid hosts. *Entomophaga* **24**: 255-258.

Marchal, P. 1900. Sur un nouvel Hyménoptère aquatique, le *Limnodytes gerriphagus* N. Gen., N. Sp. *Annales de la Société Entomologique de France* **69**: 171-176.

Martin, C. H. 1927. Biological studies of two hymenopterous parasites of aquatic insect eggs. *Entomologica Americana, New Series* **8**: 105-157.

Navasero, R. C. and E. R. Oatman. 1989. Life history, immature morphology and adult behavior of *Telenomus solitus* (Hymenoptera: Scelionidae). *Entomophaga* **34**: 165-177.

Nechols, J. R., J. L. Tracy, and E. A. Vogt. 1989. Comparative ecological studies of indigenous egg parasitoids (Hymenoptera: Scelionidae; Encyrtidae) of the Squash Bug, *Anasa tristis* (Hemiptera: Coreidae). *Journal of the Kansas Entomological Society* **62**: 177-188.

Noda, T. and Y. Hirose. 1989. 'Males second' strategy in the allocation of sexes by the parasitic wasp, *Gryon japonicum*. *Oecologia* **81**: 145-148.

Nummelin, M., J. R. Spence, and K. Vepsäläinen. 1988. Infection of gerrid eggs (Heteroptera: Gerridae) by the parasitoid *Tiphodytes gerriphagus* Marchal (Hymenoptera: Scelionidae) in Finland. *Annales Zoologici Fennici* **25**: 299-302.

Ohno, K. 1987. Effect of host age on parasitism by *Trissolcus plautiae* Watanabe (Hymenoptera: Scelionidae), an egg parasitoid of *Plautia stali* Scott (Heteroptera: Pentatomidae). *Applied Entomology and Zoology* **22**: 646-648.

Orr, D. B., J. S. Russin, and D. J. Boethel. 1986. Reproductive biology and behaviour of *Telenomus calvus* (Hymenoptera: Scelionidae), a phoretic parasitoid of *Podisus maculiventris* (Hymenoptera: Pentatomidae). *The Canadian Entomologist* **118**: 1063-1072.

Pak, G. A. 1986. Behavioural variations among strains of *Trichogramma* spp. A review of the literature on host age selection. *Journal of Applied Entomology* **101**: 55-64.

Rabb, R. L. and J. R. Bradley. 1968. The influence of host plants on parasitism of eggs of the Tobacco Hornworm. *Journal of Economic Entomology* **61**: 1249-1252.

Rabinovich, J. E. 1970. Population dynamics of *Telenomus fariai* (Hymenoptera: Scelionidae), a parasite of Chaga's disease vectors. II. Effect of host-egg age. *Journal of Medical Entomology* **7**: 477-481.

Rothschild, G. H. L. 1970. Parasites of rice stemborers in Sarawak (Malaysian Borneo). *Entomophaga* **15**: 21-51.

Ruberson, J. R., C. A. Tauber, and M. J. Tauber. 1989. Development and survival of *Telenomus lobatus*, a parasitoid of chrysopid eggs: effect of host species. *Entomologia Experimentalis et Applicata* **51**: 101-106.

Salt, G. 1935. Experimental studies in insect parasitism III - Host selection. *Royal Society of London Proceedings Series B* **117**: 413-435.

______. 1961. Competition among insect parasitoids. *Mechanisms in biological competition, Symposium of the Society for Experimental Biology* **15**: 96-119.

Spence, J. R. 1986. Interactions between the scelionid egg parasitoid *Tiphodytes gerriphagus* (Hymenoptera) and its gerrid hosts (Heteroptera). *Canadian Journal of Zoology* **64**: 2728-2738.

Strand, M. R. 1986. The physiological interactions of parasitoids with their hosts and their influence on reproductive strategies. In J. K. Waage and D. Greathead, eds., *Insect parasitoids*. pp. 97-136. Academic Press, London.

Strand, M. R. and S. B. Vinson. 1982. Behavioral response of the parasitoid *Cardiochiles nigriceps* to a kairomone. *Entomologia Experimentalis et Applicata* **31**: 308-315.

______. 1983a. Factors affecting host recognition and acceptance in the egg parasitoid *Telenomus heliothidis* (Hymenoptera: Scelionidae). *Environmental Entomology* **12**: 1114-1119.

______. 1983b. Host acceptance behavior of *Telenomus heliothidis* (Hymenoptera: Scelionidae) toward *Heliothis virescens* (Lepidoptera: Noctuidae). *Annals of the Entomological Society of America* **76**: 781-785.

______. 1985. *In vitro* culture of *Trichogramma pretiosum* on an artificial medium. *Entomologia Experimentalis et Applicata* **39**: 203-209.

Strand, M. R., S. M. Meola, and S. B. Vinson. 1986. Correlating pathological symptoms in *Heliothis virescens* eggs with development of the parasitoid *Telenomus heliothidis*. *Journal of Insect Physiology* **32**: 389-402.

Strand, M. R., S. B. Vinson, W. C. Nettles Jr., and Z. N. Xie. 1988. In *vitro* culture of the egg parasitoid *Telenomus heliothidis*: the role of teratocytes and medium consumption in development. *Entomologia Experimentalis et Applicata* **46**: 71-78.

Waage, J. K. 1982. Sib-mating and sex ratio strategies in scelionid wasps. *Ecological Entomology* **7**: 103-112.

Weber, C. A., J. M. Smilanick, L. E. Ehler, and F. G. Zalom. 1996. Ovipositional behavior and host discrimination in three scelionid egg parasitoids of stink bugs. *Biological Control* **6**: 245-252.

van Welzen, C. R. L. and J. K. Waage. 1987. Adaptive responses to local mate competition by the parasitoid, *Telenomus remus*. *Behavioral Ecology and Sociobiology* **21**: 359-365.

4. EFFECTS OF MATING STATUS AND WASP DENSITY ON REPRODUCTIVE BEHAVIOUR OF *Tiphodytes gerriphagus* Marchal (HYMENOPTERA : SCELIONIDAE)

4.1 SYNOPSIS

Effects of mating status and wasp density on frequency of superparasitism, development time, longevity, and fecundity of the egg parasitoid, *Tiphodytes gerriphagus* Marchal, were investigated. The number of probes per egg was significantly higher from single mated wasps (3.03 ± 0.212) (mean ± *SE*) than from single unmated wasps (2.41 ± 0.183), but there was no significant difference in the number of larvae per egg at (1.00 ± 0.000) and (1.03 ± 0.029), respectively. As wasp density increased, the number of probes per egg and the number of larvae per egg increased. Superparasitism occurred most frequently when more than one parasitoid was in a patch, and groups of mated wasps superparasitised more frequently than groups of unmated wasps at 78.1 % and 62.9 %, respectively. Development time of wasps from superparasitised eggs was significantly longer than of wasps from singly parasitised eggs. Longevity was not affected in males emerging from superparasitised eggs, but female longevity was significantly decreased. Mated and unmated females were equally fecund regardless of whether they emerged from superparasitised or singly parasitised eggs.

4.2 INTRODUCTION

Sex determination in haplodiploid Hymenoptera is proximally controlled by females through decisions about fertilization of eggs (Godfray, 1994). Males develop from unfertilized eggs and females from fertilized eggs. Thus, mated female wasps have some behavioural control over which sex will be oviposited into a host. In contrast, unmated wasps can produce only male progeny. Successful host location and production of offspring are interconnected for parasitoids and should be strongly influenced by natural selection (van Alphen and Vet, 1986). Because of the clear implications for sex ratio, mating status of parasitoids is potentially an important influence on current and future host-searching behaviours (Michaud, 1994).

Tiphodytes gerriphagus Marchal (Hymenoptera: Scelionidae) is a solitary egg parasitoid of several species of water striders (Heteroptera: Gerridae) (Marchal, 1900; Martin, 1927; Hoffmann, 1932; Spence, 1986; Nummelin *et al.*, 1988; Henriquez and Spence, 1993). Field collections of *Limnoporus dissortis* Drake & Harris eggs are parasitised at rates greater than 80 % during mid-summer in western Canada (Spence, 1986), and field sex ratios are generally less female-biased than laboratory sex ratios of the parasitoid (unpubl. data). Foundress number has been shown to have little effect on sex ratio output of *T. gerriphagus* (Chapter 3) and some other scelionids (van Welzen and Waage, 1987; Braman and Yeargan, 1989). Therefore, less female-biased sex ratios of field collected *L. dissortis* eggs may be due to searching behaviour of unmated wasps.

In this study, I investigate searching behaviour of mated and unmated *T. gerriphagus* in groups and alone that may affect the fitness of their progeny. The number of probes per egg and the number of larvae actually oviposited by females was observed. Also the frequency of superparasitism, development time, longevity, and fecundity of progeny were investigated.

4.3 MATERIALS AND METHODS

A *T. gerriphagus* culture was initiated and maintained as described in Chapter 3. Parasitised *L. dissortis* eggs were randomly selected and individually placed into 1.5 mL microcentrifuge tubes provided with 0.5 mL of water. On day of adult emergence, individual female parasitoids were placed inside 12 dram vials provided with 15 mL of water, and were either paired with males for mating or held singly and unmated. After 24 h, females were randomly allocated into one of four treatment groups: single unmated parasitoid, single mated parasitoid, five unmated parasitoids, or five mated parasitoids. Groups were placed into 500 mL widemouth jars provided with 100 mL of water and 15 *L. dissortis* eggs aged 48-72 h. Each trial lasted 24 h, then the eggs were placed inside 12 dram vials provided with 15 mL of water until wasp emergence. This experiment was repeated three times to determine superparasitism frequency, development and longevity, and fecundity of resultant progeny. Each treatment within an experiment varied in number of replicates depending on egg and wasp availability.

4.3.1 Superparasitism frequency

An experiment was done to test the probability of superparasitism due to mating status and wasp density. Forty-eight hours from first exposure to a wasp group, five eggs were randomly selected and searched for ovipositor wounds (Chapter 3). The total number of probes was counted on each egg which was then dissected. The number of parasitoid larvae per egg was counted, and frequency of superparasitism was calculated as the percent of eggs containing more than one parasitoid from total eggs probed. Median values of probes per egg and larvae per egg from each replicate were compared with *Kruskal-Wallis* analysis of variance.

4.3.2 Development and longevity

Possible differences in parasitoid fitness due to parent mating status and wasp density were tested by repeating the above experiment without dissections so that development time, longevity, and maximum length from each treatment could be recorded. Wasp emergence was checked every 8 h to record development time. Upon emergence, parasitoids were individually placed into a 1.5 mL microcentrifuge tube provided with a drop of water and no access to food so as to measure host derived resources. Wasps were checked every 8 h to assess longevity. After death, parasitoid body length was measured at × 50 using a dissecting microscope fitted with an ocular micrometer. Treatment specific median values of development time, longevity, and parasitoid body length for the progeny of each female were used in statistical analyses.

4.3.3 Fecundity

Actual lifetime fecundity was compared between female progeny emerging from eggs that had been exposed to either a single mated female or a group of five mated females. After 24 h exposure to parasitoids, parasitised *L. dissortis* eggs were individually placed into 1.5 mL microcentrifuge tubes provided with 1 mL of water. Two females were randomly selected from each replicate and either paired with a male from the same treatment for 24 h within a 12 dram vial, or held unpaired. Every 24 h until death, females were placed into a new vial with 50 *L. dissortis* eggs between 48-72 h old. Hosts that produce neither a gerrid nor wasp were dissected to determine if parasitoid oviposition had occurred. The total number of eggs parasitised was compared by *Kruskal-Wallis* analysis. Mating was confirmed *post facto* by observing if resultant progeny included females since unmated wasps produce only males. Wasps that did not parasitise any eggs during their lifetime were excluded from analysis.

4.4 RESULTS

4.4.1 Superparasitism frequency

Mated wasps probed each egg significantly more than did unmated wasps, and mean number of probes per egg increased as foundress number increased (Table 4.1; *Kruskal-Wallis* test: $\chi^2 = 36.6$; df = 3 $P < 0.0001$). Although there was no significant difference in the number of larvae found within an egg between mated and unmated parasitoids in the same foundress number group, the data suggest a trend toward more larvae from the mated five female group. Clearly, groups of five wasps produced significantly more larvae per egg than single wasps (Table 4.1; *Kruskal-Wallis*: $\chi^2 = 38.5$; df = 3; $P < 0.0001$).

Superparasitism occurred in all treatments with varying degree. Although the frequency of self-superparasitism did not differ significantly between single wasps of different mating status (Figure 4.1; $\chi^2 = 2.45$; df = 3; $P > 0.1$), it appears that unmated single wasps tend to superparasitise their own progeny less often than single mated wasps. Groups of five parasitoids superparasitised eggs significantly more often than did single females (Figure 4.1; $\chi^2 = 128.42$; df = 3; $P < 0.0001$), and unmated groups superparasitised significantly less than mated groups (Figure 4.1; $\chi^2 = 3.99$; df = 3; $P < 0.05$).

4.4.2 Development and longevity

Progeny emerging from eggs attacked by a single parasitoid developed significantly faster than wasps emerging from eggs subjected to attack by a group of five parasitoids (Figure 4.2; *Kruskal-Wallis test*: $\chi^2 = 20.4$; df = 5; $P < 0.001$). Also, only female wasps emerging from eggs exposed to five parasitoids generally had a shorter lifespan than wasps emerging from eggs exposed to one parasitoid (Figure 4.2; *Kruskal-Wallis* test: $\chi^2 = 11.3$; df = 5; $P < 0.05$). However, a clear pattern was not evident because male progeny from single unmated wasps had significantly lower longevity than males and females from single mated wasps (Figure 4.2).

The percentage of eggs probed out of 15 *L. dissortis* eggs did not differ significantly among different mating status and parasitoid density treatments (Table 4.2; *Kruskal-Wallis*: $\chi^2 = 5.93$; df = 3; $P = 0.12$). Probed eggs that produced neither a wasp nor gerrid were dissected to determine if parasitoid oviposition occurred, and any probed eggs that did not contain a parasitoid were considered unparasitsed. Based on parasitoid emergence and dissections, the actual percent parasitism did not differ significantly between treatments (Table 4.2; *Kruskal-Wallis*: $\chi^2 = 4.10$; df = 3; $P = 0.25$). However, it appears that unmated wasps tended to parasitise fewer eggs. Percent parasitoid mortality was calculated by counting the number of eggs that contained a dead wasp. Despite differences in larval densities within eggs of the different treatments (Table 4.1), percent wasp mortality was not significantly different between treatments (Table 4.2; $\chi^2 = 2.49$; df = 3; $P > 0.25$). Wasp size did not significantly differ between treatments (Table 4.2).

4.4.3 Fecundity

Fecundity did not differ significantly between female parasitoids emerging from singly parasitised eggs and superparasitised eggs, nor did it differ significantly between mated and unmated wasps (Table 4.3; two-way *ANOVA*: $F = 0.25$; df = 28; $P = 0.62$). However, there was a trend of wasps emerging from superparasitised eggs to parasitise fewer eggs in a lifetime.

4.5 DISCUSSION

4.5.1 Superparasitism frequency

Scelionids may probe eggs more than once because of accidental probes into adjacent eggs (Wilson, 1961; Austin, 1984), unsuccessful piercing (Bosque and Rabinovich, 1979), and difficulty in determining internal egg quality (Ables *et al.*, 1981). Female *T. gerriphagus* also make several attempts to oviposit into single gerrid eggs

(Martin, 1927; Spence, 1986), but this behaviour has not been previously quantified in relation to mating status for this or any other scelionid species. Multiple probing appears to be common in members of this family, and so it is not primarily due to the small egg batch size used in these experiments or to the jelly coating covering *L. dissortis* eggs. However, it remains to be tested how these factors affect the extent of multiple probes per egg.

External marking of eggs by scelionids is common when eggs are easily accessible to antennal examination by the parasitoid (Bosque and Rabinovich, 1979). Gerrid eggs are oviposited underwater, on the lower margin of floating vegetation, and *Limnoporus* eggs are covered by a jelly coat (Andersen, 1982; Spence, 1986). External examination of a *Limnoporus* egg is impossible and so it is probable that host discrimination is solely by internal examination with the ovipositor. This probably exacerbates multiple probing behaviour in female *T. gerriphagus*.

Mating status of *T. gerriphagus* appears to affect the number of probes per egg, with mated females tending to probe significantly more often than unmated females (Table 4.1). However, the average number of larvae per egg did not differ significantly between mated and unmated females (Table 4.1). Extra probing may be necessary for mated parasitoids because female progeny are usually more affected by egg quality than male progeny (Charnov *et al.*, 1981). As long as the egg does not rupture from multiple probing, extra probing may allow the parasitoid to assess if egg quality is suitable for female development. Unmated *T. gerriphagus* only produce male progeny, and males wasps can develop in eggs with less food because they pupate at smaller sizes compared to females (Chapter 2, Table 2.2.; Strand 1986).

Superparasitism frequently occurs when hosts are limited or parasitoid density in a patch is high (McColloch and Yuasa, 1915; Schell, 1943; Wilson, 1961; Rothschild 1970; Conde and Rabinovich, 1979; Waage 1986). It has been suggested that self-superparasitism may improve progeny survival by overwhelming host resistance (Michaud, 1994). However, insect eggs do not have a cellular defense response against parasitism (Strand, 1986), and so it is unlikely that self-superparasitism is a strategy to lower host defenses. This is further supported by the fact that self-superparasitism occurred at a low frequency (< 11 %) and did not significantly differ between single mated and unmated *T. gerriphagus* (Figure 4.1). The frequency of superparasitism was significantly greater with the five mated grouping than the five unmated grouping (Figure 4.1), but since behaviour was not observed directly, it cannot be determined if supernumerary larvae within an egg came from the same parasitoid (self-superparasitism) or different parasitoids (conspecific superparasitism).

Superparasitism has a selective advantage when there is a reasonable chance that offspring deposited later will survive (Bakker *et al.*, 1985), and it may increase the probability that one of a female's progeny will win against another competitor's offspring (Michaud, 1994). Superparasitism may be more common with groups of mated parasitoids than with groups of unmated parasitoids because mated females may be attempting to ensure that some of their female progeny survive. Mated parasitoids produce both males and females, and so there is a chance of different sexes being oviposited into a superparasitised egg. Male scelionids generally hatch a few hours before females (Waage, 1982; Field *et al.*, 1997), and the first parasitoid to eclose has the highest likelihood of survival (Visser *et al.*, 1992, Weber *et al.*, 1996, Field *et al.*, 1997). The fixed sex ratio allocation strategy of scelionids (Waage, 1982; van Welzen and Waage, 1987; Braman and Yeargan, 1989; Noda and Hirose, 1989; Weber *et al.*, 1996) may prevent mated females from selectively ovipositing more unfertilised eggs (males) when other foundresses are present. Revisiting a patch to superparasitise would increase the chance of a male being deposited into an egg containing a another wasp's female offspring.

On the other hand, unmated female *T. gerriphagus* only deposit males, and these tend to develop faster (Waage, 1982) and to pupate at smaller sizes than females (Strand, 1986). After oviposition, the progeny of an unmated parasitoid (all males) have a higher probability of surviving superparasitism than those of a mated parasitoid (female-biased). Thus, there is no selective advantage for an unmated parasitoid to revisit a patch and continue to parasitise eggs when other foundresses are present.

4.5.2 Development and longevity

Superparasitism involving solitary wasps tends to increase the total development time of the surviving wasp (McColloch and Yuasa, 1915; Gerling, 1972; Vinson and Sroka, 1978; Conde and Rabinovich, 1979; Harvey *et al.*,1993; Visser *et al.*, 1992). It has been suggested that larvae do not feed until competitors are eliminated and this is why wasps emerging from superparasitised eggs take longer to grow (Conde and Rabinovich, 1979). Observations of larval behaviour of *T. gerriphagus* during dissections (Figure 4.1) do not agree with this observation since they clearly feed in the presence of other larvae. Longer development time may result from an increase in concentration of maternal factors from repeated ovipositions (Strand *et al.*, 1986) and an increase in concentration of teratocytes from hatching larvae (Strand *et al.*, 1986; Strand *et al.*, 1988). A combination of these two factors may stimulate the larvae to feed for a longer period of time to offset the decrease of available food per individual and to ensure that no other living larvae remain in the egg when they are ready to pupate.

Longer development time may decrease the fitness of both sexes by limiting opportunities to mate or find hosts. Males of some scelionid species have been observed to await emergence of females from a parasitised egg batch and chase away late arriving males so that the guarding male obtains most of the matings (Wilson, 1961; Hokyo and Kiritani, 1966; Safavi, 1968; Rothschild, 1970; Harris, 1977, Waage, 1982). Late emerging female wasps may have difficulty finding unparasitised eggs because normal developing females have already exploited a patch (Godfray, 1994), or a territorial female may prevent access to eggs (Wilson, 1961; Hokyo and Kiritani, 1966; Safavi, 1968; Rothschild, 1970; Harris, 1977; Waage, 1982). Territorial behaviour has not been observed with *T. gerriphagus*, but it is expected that late emerging wasps (Figure 4.2) will have more difficulty in finding mates or hosts.

Studies of adult longevity of solitary parasitoids have generally involved comparisons of different sugar and/or water combinations (McColloch and Yuasa, 1915; Ticehurst and Allen, 1973; Harris, 1977; Austin, 1984; Cronin and Strong, 1990;

Navasero and Oatman, 1989), but the effect of superparasitism on longevity has not been studied. *Tiphodytes gerriphagus* emerging from superparasitised eggs tended to have shorter lifespans than wasps emerging from singly parasitised eggs (Figure 4.2). As expected, females appeared to be most affected by superparasitism (Figure 4.2). Females pupate at larger sizes compared to males (Chapter 2, Table 2.2.; Strand 1986), and overcrowding an egg may affect food quality.

It is unclear why males from single unmated wasps had lower longevity than males from mated wasps (Figure 4.2). Though not statistically significant, it appears that single unmated wasps rejected eggs more often than single mated wasps after they probed an egg (Table 4.2). The *L. dissortis* eggs used in this experiment may have been too large for proper male development, and so eggs were rejected more often by single unmated wasps. A single mated female would reject large eggs less often because she can deposit females into eggs that are too large to support male development. In contrast, there was no significant difference between actual oviposition of single mated and unmated wasps in the first experiment. Evidence for the eggs being too large include maximum length not being significantly different between wasps emerging from superparasitised and singly parasitised eggs (Table 4.2; *c.f.* Chapter 3, Table 3.6), male and female size not being significantly different in the single mated treatment (*Fisher's LSD*; $P = 0.12$; df = 43; *c.f.* Chapter 2 and Chapter 3), and two fully developed adults were found within one egg on four occasions. Surprisingly, from one of these eggs, a small male successfully emerged while a dead female remained in the egg. This is the first reporting of successful development of a solitary scelionid from an egg containing more than one adult (*c.f.* Schell, 1943; Gerling, 1972).

Hosts were randomly allocated to treatments and so it is unlikely that single unmated females received more eggs of poorer quality compared to the other treatments. It is possible that males tend to overeat and suffer greater mortality when oviposited into large eggs, but percent parasitoid mortality did not differ between treatments (Table 4.2). More experiments are needed to determine if males from unmated wasps are less fit than males from mated wasps.

4.5.3 Fecundity

Mating status does not appear to have a consistent effect on oviposition behaviour within the Hymenoptera. When compared to mated parasitoids, virgins may parasitise fewer eggs in a given period of time (McColloch and Yuasa, 1915; Donaldson and Walter, 1984; Tagawa, 1987; Antolin, 1989; Hooker and Barrows, 1989; Michaud, 1994), an equal number of hosts (Walter *et al.*, 1994), or more hosts (Cronin and Strong, 1990). Mating status of *T. gerriphagus* did not have a significant effect on total lifetime fecundity (Table 4.3). This means that host rejection by single unmated wasps in the development and longevity experiment (Table 4.2) was probably due to egg quality and not the effect of mating status on parasitoid behaviour. This also supports observations of less female-biased sex ratios in field collected *L. dissortis* eggs because unmated wasps can exploit egg patches as efficiently as mated females. As the summer season progresses and parasitism rates reach almost 100 % (Spence, 1986), it is likely that superparasitism frequency increases as well. This will result in slower developing wasps of both sexes (Figure 4.2) and increase the proportion of unmated females in the population. This may be an alternate explanation of why a survey of scelionid parasitised eggs collected from the field (Waage, 1982) had less female-biased sex ratios.

Though not significantly different, wasps emerging from parasitised eggs tended to have lower fecundity (Table 4.3). Scelionids usually oviposit most of their egg complement within a few days of emergence (McColloch and Yuasa, 1915; Austin, 1984). Unless longevity is greatly impaired by superparasitism, fecundity will not differ between females emerging from singly parasitised and superparasitised eggs.

Studies of superparasitism with solitary wasps have generally focused on host discrimination (Wilson, 1961; Hokyo and Kiritani, 1966; Gerling and Schwartz, 1974; Ables *et al.*, 1981; Strand and Vinson, 1983; van Alphen and Vet, 1986; Cave *et al.*, 1987; Okuda and Yeargan, 1988; Weber *et al.*, 1996), larval competition and fitness (McColloch and Yuasa, 1915; Schell, 1943; Gerling, 1972; Conde and Rabinovich, 1979; Strand, *et al.*, 1980, Austin, 1984; Strand, 1986; Visser *et al.*, 1992; Field *et al.*, 1997), or sex ratio (Waage, 1986; van Welzen and Waage, 1987; Braman and Yeargan, 1989; Dijken *et al.*, 1993). Few studies have focused on fitness effects on wasps surviving superparasitism (Bai and Mackauer, 1992; Harvey *et al.*, 1993; Ueno, 1997). In this study, fitness differences between wasps emerging from singly parasitised and superparasitised eggs were not as large as expected. A definite effect was seen with the increase of development time for superparasitised wasps (Figure 4.2), but maximum length, longevity, and fecundity were not significantly different. Other studies have also found few differences in fitness between solitary wasps that were singly parasitised and superparasitised (Bai and Mackauer, 1992; Harvey *et al.*, 1993; Ueno, 1997). It remains to be determined if any detrimental effects from superparasitism are increased in field tests as opposed to those conducted in a more benign laboratory setting.

Table 4.1. Effect of mating and foundress number on number of probes and parasitoid larvae based on replicates each consisting of 15 *Limnoporus dissortis* eggs exposed to *Tiphodytes gerriphagus* for 24 h. Five eggs were randomly selected from each replicate for observations.

Treatment						
Reproductive status	Parasitoid density	*n*	Probes per egg (# ± *SE*)*	Range (min.-max.)	Larvae per egg (# ± *SE*)*	Range (min.-max.)
Unmated	1	*17*	2.41 ± 0.183^{a}	1-8	1.00 ± 0.000^{a}	0-2
Mated	1	*17*	3.03 ± 0.212^{b}	1-13	1.03 ± 0.029^{a}	0-2
Unmated	5	*15*	5.83 ± 0.762^{c}	1-20	2.17 ± 0.291^{b}	0-8
Mated	5	*15*	8.60 ± 0.653^{d}	1-21	2.73 ± 0.153^{b}	0-6

Values within a column followed by same letters are not significantly different, $P > 0.05$.
* *Kruskal-Wallis* test.

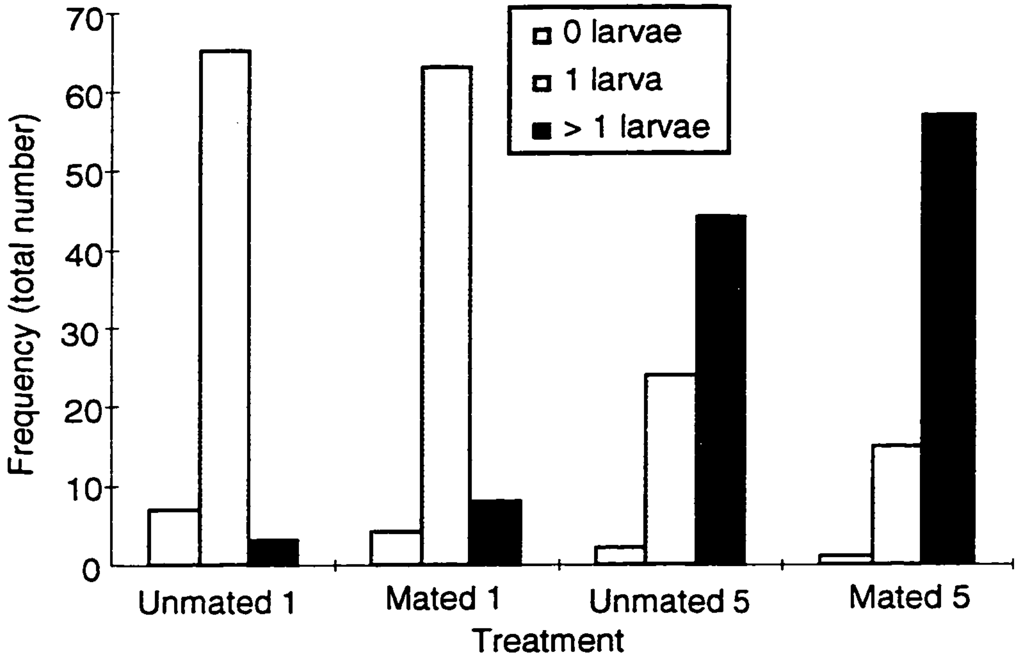


Figure 4.1. Effect of mating status and wasp density on frequency of superparasitism based on replicates, each consisting of 15 *Limnoporus dissortis* eggs exposed to *Tiphodytes gerriphagus* for 24 h. Five randomly chosen eggs from each replicate were selected for dissection.

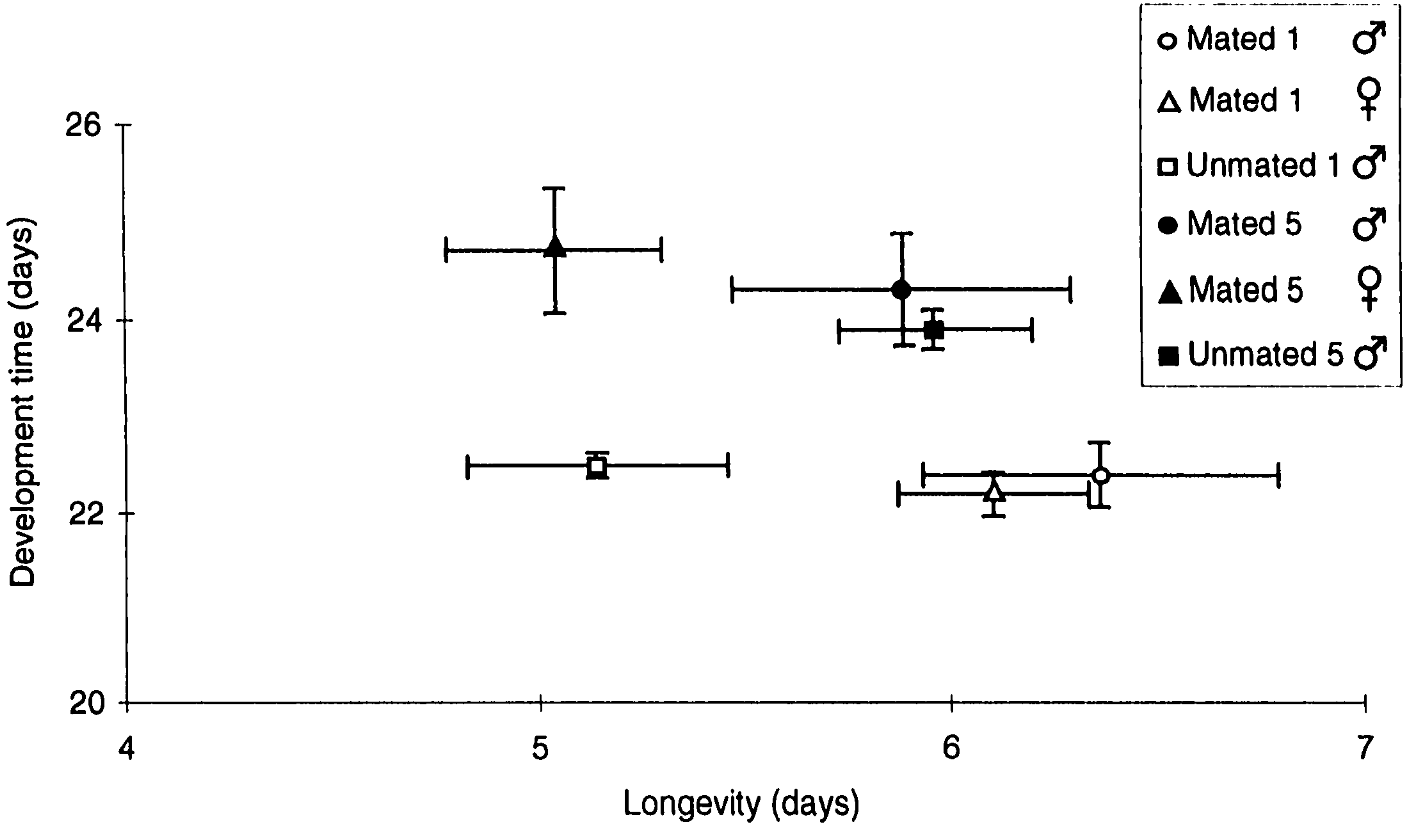


Figure 4.2. Effect of mating status and wasp density on development time and longevity of emerged wasps based on replicates, each consisting of 15 *Limnoporus dissortis* eggs exposed to *Tiphodytes gerriphagus* for 24 h.

Table 4.2. Effect of mating and foundress number on parasitism, parasitoid mortality, and maximum body length based on replicates, each consisting of 15 *Limnoporus dissortis* eggs exposed to *Tiphodytes gerriphagus* for 24 h.

Treatment			%	%	%	Maximum length (mm ± $SE^{‡}$)	
Reproductive status	Parasitoid density	*n*	eggs probed ± SE^{*}	actual parasitism ± SE^{*}	parasitoid mortality†	male	female
Unmated	1	*10*	90.7 ± 4.35^{a}	62.0 ± 12.29^{a}	5.4^{a}	1.21 ± 0.012^{a}	-
Mated	1	*6*	95.6 ± 3.30^{a}	86.7 ± 9.11^{a}	10.1^{a}	1.21 ± 0.012^{a}	1.24 ± 0.015^{a}
Unmated	5	*8*	100.0 ± 0.00^{a}	89.2 ± 4.53^{a}	9.6^{a}	1.20 ± 0.011^{a}	-
Mated	5	*8*	98.3 ± 1.09^{a}	93.3 ± 3.33^{a}	11.6^{a}	1.19 ± 0.009^{a}	1.24 ± 0.013^{a}

Values within a column followed by same letters are not significantly different, $P > 0.05$.

* *Kruskal-Wallis test.*

† χ^2 *test.*

‡ *Fisher's least significant difference test.*

Table 4.3. Effect of mating and larval competition on total fecundity based on replicates, each consisting of 50 *Limnoporus dissortis* eggs exposed to groups of mated *Tiphodytes gerriphagus* daily until death.

Parental exposure (parasitoid : host ratio)	Progeny		
	Treatment Reproductive status	*n*	Total lifetime fecundity ± SE^{*}
1:15	Unmated	*9*	61.2 ± 8.69^{a}
1:15	Mated	*5*	66.6 ± 5.64^{a}
5:15	Unmated	*9*	54.4 ± 3.87^{a}
5:15	Mated	*6*	53.2 ± 3.54^{a}

Values within a column followed by same letters are not significantly different, $P > 0.05$. * Two-way *ANOVA* test.

4.6 LITERATURE CITED

Ables, J. R., S. B. Vinson, and J. S. Ellis. 1981. Host discrimination by *Chelonus insulais* (Hym.: Braconidae), *Telenomus heliothidis* (Hym.: Scelionidae), and *Trichogramma pretiosum* (Hym.: Trichogrammatidae). *Entomophaga* **26**: 149-156.

van Alphen, J. J. M. and L. E. M. Vet. 1986. An evolutionary approach to host finding and selection. In J. K. Waage and D. Greathead, eds., *Insect parasitoids*. pp. 23-61. Academic Press, London.

Andersen, N. M. 1982. *The semiaquatic bugs (Hemiptera, Gerromorpha), phylogeny, adaptations, biogeography and classification.* Scandinavian Science Press, Klampenborg.

Antolin, M. F. 1989. Genetic considerations in the study of attack behavior of parasitoids, with reference to *Muscidifurax raptor* (Hymenoptera: Pteromalidae). *Florida Entomologist* **72**: 15-32.

Austin, A. D. 1984. The fecundity, development and host relationships of *Ceratobaeus* spp. (Hymenoptera: Scelionidae), parasites of spider eggs. *Ecological Entomology* **9**: 125-138.

Bai, B. and M. Mackauer. 1992. Influence of superparasitism on development rate and adult size in solitary parasitoid wasp, *Aphidius ervi*. *Functional Ecology* **6**: 302-307.

Bakker, K., J. J. M. van Alphen, F. H. D. van Batenburg, N. van der Hoeven, H. W. Nell, W. T. H. F. van Strien-van Liempt, and T. C. J. Turlings. 1985. The function of host discrimination and superparasitization in parasitoids. *Oecologia* **67**: 572-576.

Bosque, C. and J. E. Rabinovich. 1979. Population dynamics of *Telenomus fariai* (Hymenoptera: Scelionidae), a parasite of Chaga's disease vectors. VII. Oviposition behaviour and host discrimination. *The Canadian Entomologist* **111**: 171-180.

Braman, S. K. and K. V. Yeargan. 1989. Reproductive strategy of *Trissolcus euschisti* (Hymenoptera: Scelionidae) under conditions of partially used host resources. *Annals of the Entomological Society of America* **82**:172-176.

Cave, R. D., M. J. Gaylor, and J. T. Bradley. 1987. Host handling by *Telenomus reynoldsi* (Hymenoptera: Scelionidae), an egg parasitoid of *Geocoris* spp. (Heteroptera: Lygaeidae). *Annals of the Entomological Society of America* **80**: 217-223.

Charnov, E. L., R. L. Los-den Hartogh, W. T. Jones, and J. van den Assem. 1981. Sex ratio evolution in a variable environment. *Nature* **289**: 27-33.

Conde, J. E. and J. E. Rabinovich. 1979. Larval competition between *Telenomus costalimai* (Hymenoptera: Scelionidae) and *Ooencyrtus trinidadensis venatorius* (Hymenoptera: Encyrtidae) after simultaneous oviposition in *Rhodnius prolixus* eggs (Hemiptera: Reduviidae). *Journal of Medical Entomology* **16**: 428-431.

Cronin, J. T. and D. R. Strong. 1990. Biology of *Anagrus delicatus* (Hymenoptera: Mymaridae), an egg parasitoid of *Prokelisia marginata* (Homoptera: Delphacidae). *Annals of the Entomological Society of America* **83**: 846-854.

van Dijken, M. J., P. van Stratum, and J. J. M. van Alphen. 1993. Superparasitism and sex ratio in the solitary parasitoid *Epidinocarsis lopezi*. *Entomologia Experimentalis et Applicata* **68**: 51-58.

Donaldson, J. S. and G. H. Walter. 1984. Sex ratios of *Spalangia endius* (Hymenoptera: Pteromalidae), in relation to current theory. *Ecological Entomology* **9**: 395-402.

Field, S. A., M. A. Keller, and G. Calbert. 1997. The pay-off from superparasitism in the egg parasitoid *Trissolcus basalis*, in relation to patch defense. *Ecological Entomology* **22**: 142-149.

Gerling, D. 1972. The developmental biology of *Telenomus remus* Nixon (Hym.: Scelionidae). *Bulletin of Entomological Research* **61**: 385-388.

Gerling, D. and A. Schwartz. 1974. Host selection by *Telenomus remus*, a parasite of *Spodoptera littoralis* eggs. *Entomologia Experimentalis et Applicata* **17**: 391-396.

Godfray, H. C. J. 1994. *Parasitoids: behavioral and evolutionary ecology*. Princeton University Press, Princeton.

Harris, M. K. 1977. Egg parasites of the walnut caterpillar in Texas. *The Southwestern Entomologist* **2**: 170-178.

Harvey, J. A., I. F. Harvey, and D. J. Thompson. 1993. The effect of superparasitism on development of the solitary parasitoid wasp, *Venturia canescens* (Hymenoptera: Ichneumonidae). *Ecological Entomology* **18**: 203-208.

Henriquez, N. P. and J. R. Spence. 1993. Host location by the gerrid egg parasitoid *Tiphodytes gerriphagus* (Marchal) (Hymenoptera: Scelionidae*). Journal of Insect Behavior* **6**: 455-466.

Hoffmann, C. H. 1932. Hymenopterous parasites from the eggs of aquatic and semiaquatic insects. *Journal of Kansas Entomological Society* **2**: 33-37.

Hokyo, N. and K. Kiritani. 1966. Oviposition behaviour of two egg parasites, *Asolcus mitsukurii* Ashmead and *Telenomus nakagawai* Watanabe (Hymenoptera: Scelionidae). *Entomophaga* **11**: 191-201.

Hooker, M. E. and E. D. Barrows. 1989. Clutch size and sex ratios in *Pediobius foveolatus* (Hymenoptera: Eulophidae), primary parasites of *Epilachna varivestis* (Coleoptera: Coccinellidae). *Annals of the Entomological Society of America* **82**: 460-465.

Marchal, P. 1900. Sur un nouvel Hyménoptère aquatique, le *Limnodytes gerriphagus* N. Gen., N. Sp. *Annales de la Société Entomologique de France* **69**: 171-176.

Martin, C. H. 1927. Biological studies of two hymenopterous parasites of aquatic insect eggs. *Entomologica Americana, New Series* **8**: 105-157.

McColloch, J. W. and H. Yuasa. 1915. Further data on the life economy of the chinch bug egg parasite. *Journal of Economic Entomology* **8**: 248-261.

Michaud, J. P. 1994. Differences in foraging behaviour between virgin and mated aphid parasitoids (Hymenoptera: Aphidiidae). *Canadian Journal of Zoology* **72**: 1597-1602.

Navasero, R. C. and E. R. Oatman. 1989. Life history, immature morphology and adult behavior of *Telenomus solitus* (Hymenoptera: Scelionidae). *Entomophaga* **34**: 165-177.

Noda, T. and Y. Hirose. 1989. 'Males second' strategy in the allocation of sexes by the parasitic wasp, *Gryon japonicum*. *Oecologia* **81**: 145-148.

Nummelin, M., J. R. Spence, and K. Vepsäläinen. 1988. Infection of gerrid eggs (Heteroptera: Gerridae) by the parasitoid *Tiphodytes gerriphagus* Marchal (Hymenoptera: Scelionidae) in Finland. *Annales Zoologici Fennici* **25**: 299-302.

Okuda, M. S. and K. V. Yeargan. 1988. Intra- and interspecific host discrimination in *Telenomus podisi* and *Trissolcus euschisti*. *Annals of the Entomological Society of America* **81**: 1017-1020.

Rothschild, G. H. L. 1970. *Gryon flavipes* (Ashmead) (Hymenoptera: Scelionidae), an egg-parasite of the Rice Earbug *Leptocorisa oratorius* (Fabricius) (Hem.: Alydidae). *Entomophaga* **15**: 15-20.

Safavi, M. 1968. Étude biologique et écologique des Hyménoptères parasites des oeufs des punaises des céréales. *Entomophaga* **13**: 381-495.

Schell, S. C. 1943. The biology of *Hadronotus ajax* Girault (Hymenoptera: Scelionidae), a parasite in the eggs of Squash-bug (*Anasa Tristis* DeGeer). *Annals of the Entomological Society of America* **36**: 625-635.

Spence, J. R. 1986. Interactions between the scelionid egg *parasitoid Tiphodytes gerriphagus* (Hymenoptera) and its gerrid hosts (Heteroptera). *Canadian Journal of Zoology* **64**: 2728-2738.

Strand, M. R. 1986. The physiological interactions of parasitoids with their hosts and their influence on reproductive strategies. In J. K.Waage and D. Greathead, eds., *Insect parasitoids*. pp. 97-136. Academic Press, London.

Strand, M. R. and S. B. Vinson. 1983. Host acceptance behavior of *Telenomus heliothidis* (Hymenoptera: Scelionidae) toward *Heliothis virescens* (Lepidoptera: Noctuidae). *Annals of the Entomological Society of America* **76**: 781-785.

Strand, M. R., S. M. Meola, and S. B. Vinson. 1986. Correlating pathological symptoms in *Heliothis virescens* eggs with development of the parasitoid *Telenomus heliothidis*. *Journal of Insect Physiology* **32**: 389-402.

Strand, M. R., S. Ratner, and S. B. Vinson. 1980. Maternally induced host regulation by the egg parasitoid *Telenomus heliothidis*. *Physiological Entomology* **8**: 469-475.

Strand, M. R., S. B. Vinson, W. C. Nettles Jr., and Z. N. Xie. 1988. In *vitro* culture of the egg parasitoid *Telenomus heliothidis*: the role of teratocytes and medium consumption in development. *Entomologia Experimentalis et Applicata* **46**: 71-78.

Tagawa, J. 1987. Post-mating changes in the oviposition tactics of the parasitic wasp, *Apanteles glomeratus* L. (Hymenoptera: Braconidae). *Applied Entomology and Zoology* **22**: 537-542.

Ticehurst, M. and D. C. Allen. 1973. Notes on the biology of *Telenomus coelodasidis* (Hymenoptera: Scelionidae) and its relationship to the Saddled Prominent, *Heterocampa guttivitta* (Lepidoptera: Notodontidae). *The Canadian Entomologist* **105**: 1133-1143.

Ueno, T. 1997. Effects of superparasitism, larval competition, and host feeding on offspring fitness in the parasitoid *Pimpla nipponica* (Hymenoptera: Ichneumonidae). *Annals of the Entomological Society of America* **90**: 682-688.

Vinson, S. B. and P. Sroka. 1978. Effects of superparasitism by a solitary endoparasitoid on the host, parasitoid, and field samplings. *The Southwestern Entomologist* **3**: 299-304.

Visser, M. E., B. Luyckx, H. W. Nell, and G. J. F. Boskamp. 1992. Adaptive superparasitism in solitary parasitoids: marking of parasitised hosts in relation to the pay-off from superparasitism. *Ecological Entomology* **17**: 76-82

Waage, J. K. 1982. Sib-mating and sex ratio strategies in scelionid wasps. *Ecological Entomology* **7**: 103-112.

______. 1986. Family planning in parasitoids: adaptive patterns of progeny and sex allocation. In J. K. Waage and D. Greathead, eds., *Insect parasitoids*. pp. 63-95. Academic Press, London.

Walter, G. H., K. Ruohomäki, E. Haukioja, and E. Vainio. 1994. Reproductive behaviour of mated and virgin females of a solitary sawfly *Dineura viridiorsata*. *Entomologia Experimentalis et Applicata* **70**: 83-90.

Weber, C. A., J. M. Smilanick, L. E. Ehler, and F. G. Zalom. 1996. Ovipositional behavior and host discrimination in three scelionid egg parasitoids of stink bugs. *Biological Control* **6**: 245-252.

van Welzen, C. R. L. and J. K. Waage. 1987. Adaptive responses to local mate competition by the parasitoid, *Telenomus remus*. *Behavioral Ecology and Sociobiology* **21**: 359-365.

Wilson, F. 1961. Adult reproductive behaviour in *Asolcus basalis* (Hymenoptera: Scelionidae). *Australian Journal of Zoology* **9**: 737-751.

5. CONCLUSION

5.1 Summary

There are three families within Hymenoptera that exclusively parasitise insect eggs, the fairy flies (Mymaridae), the scelionid wasps (Scelionidae), and the minute egg parasites (Trichogrammatidae) (Strand, 1986). Egg parasitoids are important in biological control programs for they eliminate insect pests before any feeding damage occurs. Effective development of biocontrol agents requires understanding parasitoid ecology and knowledge of the biology of individual parasitoids and hosts as well as their interactions (Salt, 1934). However, the study of egg parasitoid biology is difficult because of the small size of them and their hosts (Strand, 1986).

Tiphodytes gerriphagus Marchal (Hymenoptera: Scelionidae) has been known to parasitise water strider (Heteroptera: Gerridae) eggs since the beginning of this century (Marchal, 1900), but investigations of its biology and behaviour have only recently started (Spence, 1986; Henriquez and Spence, 1993a). This thesis has examined the development and fitness of successfully emerging *T. gerriphagus* in a laboratory setting in an attempt to better understand observations of field populations of hosts and wasps.

Successful parasitism involves the successful location of a host and development within. *Tiphodytes gerriphagus* has been shown to use visual cues such as vegetation and adult gerrids to locate eggs (Henriquez and Spence, 1993a), but little was known about development after oviposition. Development of immatures within *Trepobates* eggs was described by Martin (1927), but development time was not provided. Furthermore, controversies about the actual form of the immature stages surfaced (Clausen, 1940;

Hagen, 1956; Masner, 1972). In chapter two, I described the development of all immature stages of *T. gerriphagus* within *Limnoporus dissortis* Drake & Harris eggs. Using ecdysis to determine the number of instars after eclosion, *T. gerriphagus* was found to have two larval, a pupal, and an adult stage. The first instar dramatically increased in length and width as it fed on host material, and this stage lasted approximately four days after eclosion. The second instar used peristaltic gut contractions to mix ingested food, and its mandibles were greatly reduced compared to those of the first. It was determined that this second instar was the 'third instar' described by Martin (1927). Males were significantly smaller than females in both pupal and adult measurements.

After an egg has been selected for oviposition, successful development of the parasitoid is affected by the quality of the egg. Differences in egg quality can affect the fitness of an emerging parasitoid (Pak, 1986). In chapter three, I examined the potential effects of egg quality on *T. gerriphagus* fitness. I demonstrated that wasp mortality was significantly lower from intermediate aged eggs (48-72 h) than from eggs less than 24 h old. I have suggested that infertile eggs may contribute to increased wasp mortality in young eggs. Wasp size was not significantly affected within the egg age range used (0-96 h). Wasp mortality was significantly higher when reared on the larger eggs of *L. dissortis* than from the smaller eggs of *Gerris buenoi* Kirkaldy. Excessive food material may have contributed to wasp mortality (Strand *et al.*, 1988). Increasing the number of wasps searching on a patch (foundress number) significantly decreased wasp survival and size. Unexpectedly, sex ratio was not affected by egg age, host species, nor foundress number (Charnov *et al.*, 1981, Waage, 1982).

Mating status is potentially an important influence on hymenopteran foraging behaviour (Michaud, 1994). Though foundress number has been shown to have little effect on sex ratio output of scelionids (Chapter 3; van Welzen and Waage, 1987; Braman and Yeargan, 1989), field collections of parasitised *L. dissortis* eggs are less female-biased than laboratory cultures. Chapter four focussed on foraging behaviour of mated and unmated *T. gerriphagus* in groups and alone that may affect the fitness of their progeny. The number of probes and larvae per egg increased with increasing wasp density. Mated females tended to probe eggs more often than unmated females, and groups of five mated females significantly superparasitised eggs more often than groups of five unmated females. It is proposed that mated females probe and superparasitise more often than unmated females because they are trying to increase the chance of survival of their female progeny. Development time was significantly longer for wasps emerging from superparasitised eggs, but longevity and fecundity were not significantly different. Mating status had no significant affect on total lifetime fecundity, and it is suggested that unmated wasps foraging in the field contribute to the less female-biased sex ratios observed in the field.

5.2 Considerations for future research

Study of behaviour and biology of water striders, their egg parasitoids, and their interactions is relatively new. To progress in understanding field observations, further experiments need to resemble those established by biological control researchers.

Direct observation of oviposition behaviour needs to be carried out to obtain a better understanding of host preferences with respect to host egg age, species, and parasitism state (van Dijken *et al.*, 1986). Direct observation will let us know if *T. gerriphagus* can discriminate between parasitised and unparasitised eggs. Abdominal movements during oviposition may differ when fertilised eggs (female) are deposited compared to unfertilised eggs (male) (Suzuki *et al.*, 1984; van Dijken and Waage, 1987). This will make sex ratio allocation studies more accurate because successful emergence of adult wasps is not necessary to obtain data.

Wasp fitness is seldom explored in experiments such as those investigating the effects of superparasitism (Bai and Mackauer, 1992; Harvey *et al.*, 1993; Ueno, 1997). Data on development time, longevity, fecundity and perhaps male fertility should be collected for any experiments with *T. gerriphagus*. For superparasitsm, experiments should be conducted in which only two immature wasps are deposited into an egg instead of using experiments that have a range of larval densities. This will allow for possible effects of competition to be compared more accurately.

Future experiments should be repeated a minimum of three times involving:

1. a replicate that allows wasp adults to emerge,
2. a replicate in which all eggs are dissected, and
3. appropriate controls comparing gerrid emergence.

This will allow more accurate comparisons of % parasitism, % parasitoid mortality, and egg mortality. It is also suggested that eggs from which a wasp emerged be measured (length) to notice if any treatment receives larger eggs than other treatments.

The above suggestions and investigations covered by this thesis could also be used in exploring the biology and behaviour of *Lathromeroidea* sp. nova Girault (Hymenoptera: Trichogrammatidae) which is another aquatic egg parasitoid of water striders in western Canada (Spence, 1986; Henriquez and Spence, 1993b). Host range of *T. gerriphagus* and *L.* sp. nova overlaps (Spence, 1986; Henriquez and Spence, 1993b), and both species can be collected from the field within gerrid eggs on the same piece of floating vegetation. Understanding the biology of *L.* sp. nova will guide future experiments on possible multiparasitism effects on wasp fitness of both wasp species. In closing, field observations and experiments are needed to verify laboratory results and to understand parasitism rates in the field.

5.3 LITERATURE CITED

Bai, B. and M. Mackauer. 1992. Influence of superparasitism on development rate and adult size in solitary parasitoid wasp, *Aphidius ervi*. *Functional Ecology* **6**: 302-307.

Braman, S. K. and K. V. Yeargan. 1989. Reproductive strategy of *Trissolcus euschisti* (Hymenoptera: Scelionidae), under conditions of partially used host resources. *Annals of the Entomological Society of America* **82**: 172-176.

Charnov, E. L., R. L. Los-den Hartogh, W. T. Jones, and J. van den Assem. 1981. Sex ratio evolution in a variable environment. *Nature* **289**: 27-33.

Clausen, C. P. 1940. *Entomophagous insects*. McGraw-Hill, New York.

van Dijken, M. J. and J. K. Waage. 1987. Self and conspecific superparasitism by the egg parasitoid *Trichogramma evanescens*. *Entomologia Experimentalis et Applicata* **43**: 183-192.

van Dijken, M. J., M. Kole, J. C. van Lenteren, and A. M. Brand. 1986. Host preference studies with *Trichogramma evanescens* Westwood (Hym., Trichogrammatidae) for *Mamestra brassicae, Pieris brassicae, Pieris rapae*. *Journal of Applied Entomology* **101**: 64-85.

Hagen, K. S. 1956. Aquatic Hymenoptera. In. R. L. Usinger, ed., *Aquatic insects of California*. pp. 289-292. University California Press, Berkeley.

Harvey, J. A., I. F. Harvey, and D. J. Thompson. 1993. The effect of superparasitism on development of the solitary parasitoid wasp, *Venturia canescens* (Hymenoptera: Ichneumonidae). *Ecological Entomology* **18**: 203-208.

Henriquez, N. P. and J. R. Spence. 1993a. Host location by the gerrid egg parasitoid *Tiphodytes gerriphagus* (Marchal) (Hymenoptera: Scelionidae). *Journal of Insect Behavior* **6**: 455-466.

______. 1993b. Studies of *Lathromeroidea* sp. nov. (Hymenoptera: Trichogrammatidae), a parasitoid of gerrid eggs. *The Canadian Entomologist* **125**: 693-702.

Marchal, P. 1900. Sur un nouvel Hyménoptère aquatique, le *Limnodytes gerriphagus* N. Gen., N. Sp. *Annales de la Société Entomologique de France* **69**: 171-176.

Martin, C. H. 1927. Biological studies of two hymenopterous parasites of aquatic insect eggs. *Entomologica Americana, New Series* **8**: 105-157.

Masner, L. 1972. The classification and interrelationships of Thoronini (Hymenoptera: Proctotrupoidea, Scelionidae). *The Canadian Entomologist* **104**: 833-849.

Michaud, J. P. 1994. Differences in foraging behaviour between virgin and mated aphid parasitoids (Hymenoptera: Aphidiidae). *Canadian Journal of Zoology* **72**: 1597-1602.

Pak, G. A. 1986. Behavioural variations among strains of *Trichogramma* spp. A review of the literature on host age selection. *Journal of Applied Entomology* **101**: 55-64.

Salt, G. 1934. Experimental studies in insect parasitism. 1. Introduction and technique. *Royal Society of London Proceedings Series B* **114**: 450-476.

Spence, J. R. 1986. Interactions between the scelionid egg parasitoid *Tiphodytes gerriphagus* (Hymenoptera) and its gerrid hosts (Heteroptera). *Canadian Journal of Zoology* **64**: 2728-2738.

Strand, M. R. 1986. The physiological interactions of parasitoids with their hosts and their influence on reproductive strategies. In J. K. Waage and D. Greathead, eds., *Insect parasitoids*. pp. 97-136. Academic Press, London.

Strand, M. R., S. B. Vinson, W. C. Nettles Jr., and Z. N. Xie. 1988. In *vitro* culture of the egg parasitoid *Telenomus heliothidis*: the role of teratocytes and medium consumption in development. *Entomologia Experimentalis et Applicata* **46**: 71-78.

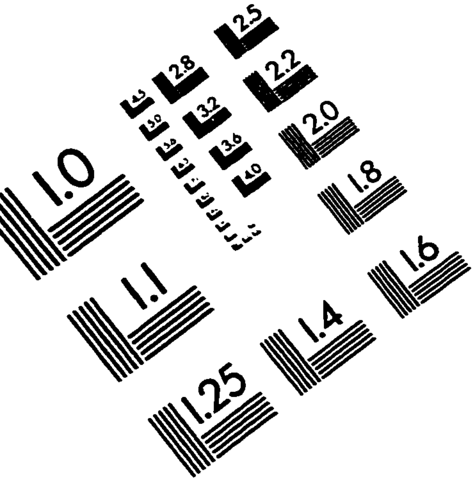
Suzuki, Y., H. Tsuji, and M. Sasakawa. 1984. Sex allocation and effects of superparasitism on secondary sex ratios in the gregarious parasitoid, *Trichogramma chilonis* (Hymenoptera: Trichogrammatidae). *Animal Behaviour* **32**: 478-484.

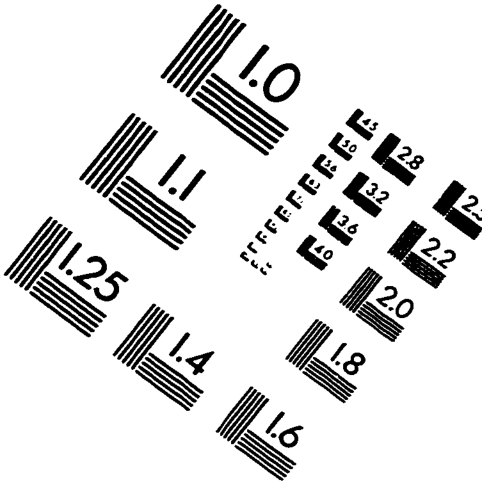
Ueno, T. 1997. Effects of superparasitism, larval competition, and host feeding on offspring fitness in the parasitoid *Pimpla nipponica* (Hymenoptera: Ichneumonidae). *Annals of the Entomological Society of America* **90**: 682-688.

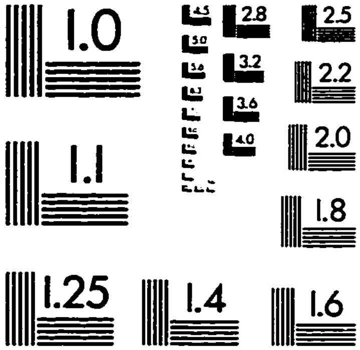
Waage, J. K. 1982. Sib-mating and sex ratio strategies in scelionid wasps. *Ecological Entomology* **7**: 103-112.

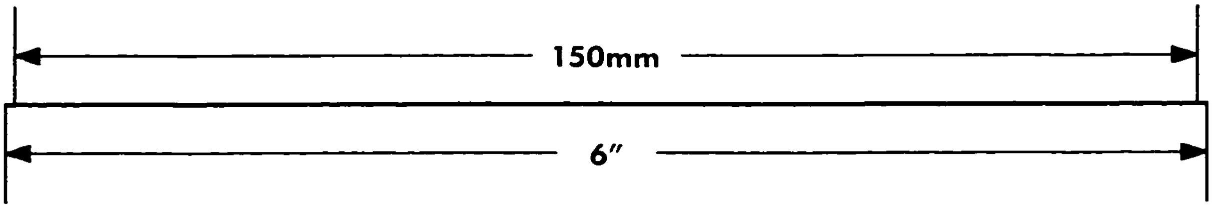
van Welzen, C. R. L. and J. K. Waage. 1987. Adaptive responses to local mate competition by the parasitoid, *Telenomus remus*. *Behavioral Ecology and Sociobiology* **21**: 359-365.

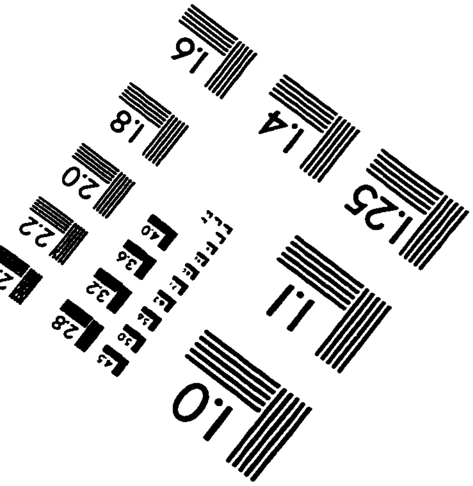
IMAGE EVALUATION TEST TARGET (QA-3)











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