Biblioteca Digital
FCEN-UBA

BIBLIOTECA CENTRAL LUIS F LELOIR
BIBLIOTECA CENTRAL LELOIR
FACULTAD DE CIENCIAS EXACTAS Y NATURALES UBA

Tesis Doctoral

Influencia de los rasgos de historia de vida y del uso de información en la adquisición de recursos y dispersión en el parasitoide Ibalia leucospoides Hochenwarth (Hymenoptera: Ibaliidae)

Fischbein, Deborah

2011

Este documento forma parte de la colección de tesis doctorales y de maestría de la Biblioteca Central Dr. Luis Federico Leloir, disponible en digital.bl.fcen.uba.ar. Su utilización debe ser acompañada por la cita bibliográfica con reconocimiento de la fuente.

This document is part of the doctoral theses collection of the Central Library Dr. Luis Federico Leloir, available in digital.bl.fcen.uba.ar. It should be used accompanied by the corresponding citation acknowledging the source.

Cita tipo APA:
Fischbein, Deborah. (2011). Influencia de los rasgos de historia de vida y del uso de información en la adquisición de recursos y dispersión en el parasitoide Ibalia leucospoides Hochenwarth (Hymenoptera: Ibaliidae). Facultad de Ciencias Exactas y Naturales. Universidad de Buenos Aires.

Cita tipo Chicago:
Fischbein, Deborah. "Influencia de los rasgos de historia de vida y del uso de información en la adquisición de recursos y dispersión en el parasitoide Ibalia leucospoides Hochenwarth (Hymenoptera: Ibaliidae)". Facultad de Ciencias Exactas y Naturales. Universidad de Buenos Aires. 2011.

EXACTAS UBA
Facultad de Ciencias Exactas y Naturales

UBA
Universidad de Buenos Aires

Dirección: Biblioteca Central Dr. Luis F. Leloir, Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires.
Intendente Güiraldes 2160 - C1428EGA - Tel. (++54 +11) 4789-9293
Contacto: digital@bl.fcen.uba.ar

UNIVERSIDAD DE BUENOS AIRES

Facultad de Ciencias Exactas y Naturales

Departamento de (Ecología, Genética y Evolución)

Influencia de los rasgos de historia de vida y del uso de información en la adquisición de recursos y dispersión en el parasitoide *Ibalia leucospoides* Hochenwarth (Hymenoptera: Ibaliidae)

Tesis presentada para optar al título de Doctor de la Universidad de Buenos Aires
en el área de Ciencias Biológicas

Deborah Fischbein

Directores de tesis: Juan Carlos Corley y Carlos Bernstein
Consejero de Estudios: Juan Carlos Reboreda

EEA INTA Bariloche, Grupo de Ecología de Insectos. Modesta Victoria, 4450 (8400), S.C. de Bariloche, Río Negro, Argentina.

Buenos Aires, 2011

N° d'ordre 105-2011 Année 2011

THÈSE PRESENTÉE DEVANT L´UNIVERSITE DE BUENOS AIRES
Et préparée en cotutelle avec
L´UNIVERSITE CLAUDE BERNARD LYON 1
Pour l´obtention du
DIPLOME DE DOCTORAT
(arrêté du 7 août 2006)

ECOLE DOCTORALE

Évolution Écosystèmes Microbiologie Modélisation
Soutenue publiquement le 15 Juin 2011

par

Deborah FISCHBEIN

Influence des traits d'histoire de vie et de l'utilisation des informations pour l'acquisition des ressources et la dispersion chez le parasitoïde *Ibalia leucospoides* Hochenwarth (Hymenoptera: Ibaliidae)

Directeurs de thèse: CORLEY Juan Carlos /BERNSTEIN Carlos

Jury :

M FREDERIC MENU	Président du Jury et Examinateur
M Diego SEGURA	Examinateur
M Esteban HASSON	Examinateur et Rapporteur
M Xavier FAUVERGUE	Examinateur et Rapporteur

Influencia de los rasgos de historia de vida y del uso de información en la adquisición de recursos y dispersión en el parasitoide *Ibalia leucospoides* Hochenwarth (Hymenoptera: Ibaliidae)

RESUMEN

El objetivo central de esta tesis es estudiar como la selección natural moldea los rasgos de historia de vida y el comportamiento de los animales de acuerdo al entorno ecológico en el que viven. Este marco teórico nos conduce a las hipótesis especificas respecto a los procesos de reproducción, dispersión y forrajeo de recursos en el parasitoide *Ibalia leucospoides*. Los resultados principales de la tesis son (1) que la proporción de huevos maduros que posee la hembra al momento de la emergencia respecto de la fecundidad potencial es alta; es decir que *I. leucospoides* tiende a la pro-ovigenia como estrategia de maduración de huevos; (2) las hembras adultas no dependen de la alimentación para sostener la maduración de huevos, ni tampoco para el mantenimiento somático y la capacidad de vuelo; (3) bajo nuestras condiciones experimentales la capacidad de vuelo obedece a las características morfológicas del parasitoide (tamaño y carga alar) y finalmente; (4) respecto a la búsqueda de hospedadores, las hembras son capaces de discriminar diferencias en la calidad de los parches de hospedadores a la distancia, sin la necesidad de llevar a cabo un proceso de muestreo de los mismos. Además, para la explotación de los parches utilizan información de los parches de hospedadores vecinos. El éxito reproductivo de *I. leucospoides*, parasitoide pro-ovigenico, no está limitado por la ausencia de alimento durante la vida adulto. Ambas estrategias, la de asignación de recursos hacia diferentes funciones biológicas y la comportamental de forrajeo, adoptadas por esta especie, probablemente sean en respuesta a características del ambiente tales como la distribución fuertemente agregada de *Sirex noctilio*, el hospedador, y la disponibilidad de recurso (i.e. hospedador y/o alimento).

Palabras claves

Estrategias de historia de vida, compromisos (*trade-offs*), asignación de recursos, inversión reproductiva, tamaño corporal, índice de ovigenia, dispersión, comportamiento de forrajeo, reglas de decisión.

Influence of life history traits and information use on resource acquisition and dispersal in the parasitoid *Ibalia leucospoides* Hochenwarth (Hymenoptera: Ibaliidae)

ABSTRACT

The central aim of this thesis is to explore how natural selection shapes life history traits and behaviuors according to the ecological environment in which an animal exists. This overall framework leads to specific hypotheses concerning the reproductive, dispersal and foraging processes in the parasitoid *Ibalia leucospoides.* The experiments in behavioural ecology and eco-physiology in female parasitoids of *Ibalia leucospoides* show as main results, that (1) the proportion of the potential lifetime eggs complement that is mature at female emergence is high; (2) low dependency on adult female feeding, not only for egg maturation but also for survival and flight; (3) under our experimental conditions flight capacity depends on the morphological characteristics of the parasitoid (size and wing loading); and (4) regarding host foraging, females accurately assess differences in host patch quality from a distance without the need of displaying a sampling process; in addition that patch exploitation times depend on the information obtained from the surrounding patches. The reproductive success of *I. leucospoides,* a proovigenic parasitoid, is not constrained by adult food deprivation. Both the resource allocation strategy toward the different biological functions and the foraging behaviour adopted by this parasitoid species may well be in respond to habitat characteristics, such as a strongly aggregated distribution of *Sirex noctilio*, the host, as well as to the resources availability (i.e. host and food).

KEY WORD:

Life history strategy, trade-off, resource allocation, reproductive investment, body size, ovigeny index, dispersal, foraging behaviour, patch decision rules.

Influence des traits d'histoire de vie et de l'utilisation des informations pour l'acquisition des ressources et la dispersion chez le parasitoïde *Ibalia leucospoides* Hochenwarth (Hymenoptera: Ibaliidae)

RÉSUMÉ

L'objectif principal de cette thèse est d'étudier comment la sélection naturelle façonne les traits d'histoire de vie et le comportement des animaux en fonction de l'environnement écologique dans lequel ils évoluent. Ce cadre théorique soulève des hypothèses spécifiques concernant les processus de reproduction, de dispersion et d'approvisionnement chez le parasitoïde *Ibalia leucospoides.* Les expériences d'écologie comportementale et d'éco-physiologie menées sur des parasitoïdes femelles d' *Ibalia leucospoides* démontrent principalement que (1) la proportion d'oeufs matures que possède la femelle à l'émergence est élevée par rapport à sa fécondité potentielle : *I. Leucospoides* adopte une stratégie proovigénique ; (2) l'alimentation a un effet négligeable sur les femelles adultes : elle n'influence ni la maturation des oeufs, ni la survie, ni les capacités de vol ; (3) dans nos conditions expérimentales, la capacité de vol dépend des caractéristiques morphologiques du parasitoïde (taille et charge alaire) ; et (4) concernant la recherche d'hôtes, les femelles sont capables de discerner des différences dans la qualité des patchs d'hôtes à distance, sans les échantillonner. D'autre part, le temps d'exploitation d'un patch dépend des informations obtenues des patchs voisins. Le succès reproductif du parasitoïde proovigénique *Ibalia leucospoides,* n'est pas limité par le manque de nourriture chez l'adulte. La stratégie d'allocation des ressources entre les différentes fonctions biologiques, ainsi que le comportement d'approvisionnement adopté par cette espèce de parasitoïde pourraient vraisemblablement être une réponse aux caractéristiques de l'habitat, telle que la distribution fortement agrégée des *Sirex noctilio*, l'hôte, ainsi que la disponibilité des ressources (i.e. hôtes et nourriture).

MOTS CLEFS :

Stratégies d'histoire de vie, compromis évolutifs, allocation des ressources, investissement reproductif, taille corporelle, indice ovigénique, dispersion, comportement d'approvisionnement, règles de décision.

INTITULE ET ADRESSE DE L'U.F.R. OU DU LABORATOIRE :

Laboratoire de Biométrie et Biologie Évolutive, CNRS UMR 5558, Université Claude Bernard Lyon 1, 43 bd du 11 novembre 1918, 69622 Villeurbanne Cedex.

EEA INTA Bariloche, Grupo de Ecología de Insectos. Modesta Victoria, 4450 (8400), S.C. de Bariloche, Río Negro, Argentina.

Agradecimientos

Quiero agradecer especialmente a Juan Corley y Carlos Bernstein por haberme guiado durante todos estos años y por su total confianza, paciencia y libertad en el trabajo. Quiero agradecerles también el haberme involucrado en sus proyectos de investigación que sin duda han enriquecido enormemente mi formación como investigadora. Además, valoro sus consejos, su continua motivación y sentido del humor en el trabajo. Gracias a Carlos y a Juan por transmitirme sus conocimientos y modos de pensar y trabajar en ciencia. Le quiero dar las gracias a Carlos por haberme abierto las puertas de su laboratorio en Lyon, cada estadía en Francia fue una experiencia de vida insuperable a nivel laboral como humano.

Gracias a Nélida, Maite, Ana, José y Octavio por su ayuda en el trabajo y compañerismo. Gracias José, Nelly y Mai por las charlas materas y los buenos momentos que pasamos juntos.

Quiero agradecer al Laboratorio de Suelos, en especial a Javier y Silvana, y a todos los chicos del Laboratorio de Genética Forestal por haberme prestado lugar y materiales para realizar mis ensayos.

Je tiens à remercier ici Louise, Perrine et Thibaut pour votre amitié et pour ces moments passés ensemble. Je vous souhaite tout le meilleur pour la suite. Je remercie Perrine Pelosse pour ses conseils sur l’analyse statistique des données et nos discussions sur les parasitoïdes.

Je remercie Emmanuel Desouhant, nos discussions et vos conseils ont grandement participé à l’orientation de ma thèse.

Quiero agradecer especialmente a Louise por haber sido tan buena y soportarme en su casa durante tantos días, semanas. Por los paseos en bici, las cervezas y largas charlas. Gracias por tu ayuda y buena onda en todos mis viajes.

Gracias a todos mis amigos. Yu, Guille, Roxy, Lula, Dami, Flor, Maco y Euge por estar desde el primer momento que pisé Bariloche, por hacerme sentir en casa, son una gran familia para mí. Gracias a Laurita y a Monchi por estar tan cerca aun a la distancia. A Richard por su amor, planes sorpresa, vinos y charlas. A Totoro por su siempre buena onda y por ser mi topo amigo.

Mi más grande agradecimiento a mi mamá y papá por ayudarme y apoyarme en todo y en todos los momentos. Gracias por sus consejos y amor. Dahlu, Franka, Richard, Gus, Marce, Ivi y Axel, gracias por estar siempre presentes.

TABLE OF CONTENTS

IBALIA LEUCOSPOIDES LIFE HISTORY TRAITS II

IBALIA LEUCOSPOIDES FLIGHT PERFORMANCE

IBALIA LEUCOSPOIDES FORAGING BEHAVIOUR

Preface

Research on how variations in life-history traits lead to variations in fitness among individuals strongly contributes to our understanding of life-history evolution. Variations in life-history traits (i.e. fecundity, longevity, size at birth, age and size at maturity, or number of offspring) can be approached through studies at different levels; for example, focusing on genetic, ecological, physiological and/or behavioural aspects of individuals.

It is widely known that individuals cannot simultaneously maximize all life-history traits to maximize fitness, partly because many traits are not independent. Generally, the existence of genetic, physiological or behavioural constraints on the evolution of life history traits, are expressed in the manner of *trade-offs.* That is, the fitness gain derived from changes in one trait may only be achieved at the cost of another trait. *Trade-offs* thus emerge mainly from the need for allocating limited energy resources to different competing traits.

For parasitoids, those insects that lay eggs in or on other arthropods (their hosts), foraging success is in direct connection with offspring production and consequently with individual fitness. This fact has made parasitoids ideal subjects to test evolutionary and behavioural hypothesis as well as excellent models to study state-dependent behaviour (Godfray, 1994). In parasitoids, the most commonly studied *trade-off* is that between individual survival and egg production. This is mainly because maintenance and reproduction are key factors limiting reproductive success. However, other life history traits such as mate, food and host finding, in addition to dispersal may also represent significant constraints on progeny production.

Parasitoids, like other holometabolous insects, have to experience contrastingly different habitats during their development. Immature stages (the larvae) may live and develop in some specific environment (eg: inside wood or under the soil) while adults may feed and reproduce in totally different habitats (eg: woodlands, prairies, etc). The habitat characteristics, including resource availability, experimented by each stage may well play an important role in shaping their life history and resource allocation strategies.

The central aim of this thesis is to explore how natural selection shapes life history traits and behaviuors according to the ecological environment in which an animal exists. This overall framework leads to specific hypotheses concerning the reproductive, dispersal and foraging processes in the parasitoid *Ibalia leucospoides*. An important effort is directed to study the mechanisms involved in the expression of behaviour and life history traits. In addition, particular attention is given throughout this study to the role that resource availability and the habitat context might have in shaping observed resource allocation patterns and life-history strategies in the female parasitoid. *Ibalia leucospoides* is a hymenopteran parasitoid of the wood wasp *Sirex noctilio*. *S. noctilio* is a primitive invasive forest insect which has become a significant pest of softwood forestation in Argentina.

In the first section of this manuscript, an introduction to parasitoid energy resource allocation strategies, foraging decisions and dispersal is presented. In a second section, the outcome of specific experiments is described. Chapter I examines the relationship between size and several life history traits and the consequences of the environmental characteristics on the observed larval resource allocation strategies. Chapter II deals with the study of adult resource allocation to several life-history traits. Research into the "food effects" on fecundity and survival is carried out, and food acquisition by *I. leucospoides* females in the wild explored. Chapter III, in turn, examines the effects of food provisioning on flight performance of *I. leucospoides*, under laboratory conditions. Finally, through Chapter IV, I address host foraging strategies and patch time allocation by the parasitoid. A third section of this dissertation discusses the end results, the significance of this study, as well as the perspectives for future work it opens.

SECTION 1
INTRODUCTION

1. General overview

1.1 Insect parasitoids: Definitions and life history

Parasitoids are insects whose adult females lay their eggs in or on other insects, and immature larvae develop by feeding on the host, this resulting inevitably in the death of the host (Godfray, 1994). Only larval stages are parasitic whereas adults are free-living. Hence, parasitoids are seen as intermediate between predators and parasites: like parasites, they can not survive without attacking hosts; like predators, they always kill the host they attack. Parasitic insects are found in many orders but, the greater part of species are members of the order Hymenoptera (50,000 species) and the order Diptera (15,000 species). Parasitoids constitute about 8.5% of all insects known (Godfray 1994).

Different criteria may be used to classiffy parasitoids. One way to do so is according to the host stage they attack. There are *egg parasitoids*, *larval parasitoids*, *pupal parasitoids* or *adult parasitoids* (the latter case being rarer). The feeding behaviour of the larvae is another criterion used. Some parasitoids develop within the body of their host, so- called *endoparasitoids*, and others develop externally, so-called *ectoparasitoids.* In addition, parasitoids which a single adult emerges from the host (regardless of the number of eggs that were laid) are known as *solitary* and, when several individuals can emerge from a single host, they are known as *gregarious.* Moreover, there are *specialist* parasitoids, those that attack single host species or *generalist* parasitoids*,* those who attacks different host species. Parasitoids can also be divided into *idiobiont* and *koinobionts.* While the former kill or paralyze their host at oviposition, the latter allow hosts to continue to feed and grow refraining from feeding on vital organs of the host. When deposition of an extra conspecific egg in an already parasitized host occurs, it is said that there is *superparasitism.* If a second female of a different species lays her eggs on an already parasitized host *multiparasitism* or *hyperparasitism* can occur. In the case of multiparasitism the larvae (the resident and the second species) compete for host resources. However, in the case of hyperparasitism the larvae of the second species feed on the parasitoid larvae already present, not on the host.

Besides, parasitoids differ in their egg maturation strategies. While some species emerge with their entire or nearly entire, complement of mature eggs, the so-called *pro-ovigenic parasitoids*; some others continue to mature eggs throughout their reproductive life, the so-called *synovigenic parasitoids* (Flanders, 1950; Jervis *et al.*, 2001). Adult parasitoids can feed from the hosts themselves (in *host-feeding* species) or from non-host sources, such as extra-floral and floral nectar or honeydew (Jervis & Kidd, 1986; Jervis *et al.*, 1992; Rivero & Casas, 1999).

All characteristics mentioned above have significant consequences on female parasitoid life history. Hence, those of our interest in this thesis will be more thoroughly reintroduced along this section.

1.2 Life-history trade-offs

Trade-offs represent the costs paid in fitness terms when a beneficial change in one trait is linked to a detrimental change in another (Stearns 1989; Roff & Fairbairn, 2007). The term '*trade-off*' can be used to describe the functional relationship between two traits or the statistical correlation between them. *Trade-offs* have a fundamental place in the theory of life-history evolution because they constrain the direction and rate of evolution, and therefore limit the range of possible life-history combinations. For example, the evolutionary biologists argue that organisms are not able to attain perfection through natural selection due to the antagonistic pleiotropy. Antagonistic pleiotropy is when one gene controls for more than one trait, and at least one of these traits is beneficial and another is detrimental to the organism's fitness. That is, antagonistic pleiotropic genes can be one of the reason of the existence of the *fitness trade-offs.* Nevertheless, *trade-offs* are often described as the result of the allocation of limited resources to different competing traits. Resources invested in one life history trait cannot be invested in another (Zera & Harshman, 2001). The most studied *trade-offs* are those between reproduction and survival, reproduction and growth, current and future reproduction, and between size and number of offspring (Stearns 1989).

To maximize their number of offspring, organisms have to allocate their energy in an efficient way to several components of their reproductive success. Survival and reproduction are major components of lifetime reproductive success. However, not less important are the contributions of other activities such as locomotion, dispersal, host finding, quiescence, that are also energy-demanding. Hence, differences in energy allocation toward these traits result in the observed inter and intraspecifics differences in life histories.

Therefore, life-history *trade-offs* can have genetic, phenotypic or physiological basis. Studies dealing with these three levels would make contribution to our understanding about how a trade-off works Stearns (1989). At the phenotypic level, researches commonly look for negative relationships among observations of populations' phenotypic variability whereas at the genetic level, *trade-offs* are sought through the measurement of genetic correlations among traits, either with quantitative genetics methods or selection experiments. Finally, research in the between level of these extremes involve the study of the physiological and developmental mechanisms that result in the allocation of resources among reproduction, growth, maintenance, storage and survival functions.

The habitat in which organisms exist is, actually, of great importance in determining the optimal resource allocation strategy. It is expected so, that natural selection favours that combination of trait values that result in maximum fitness gain under a given environment circumstance.

1.3 Egg load evolution

Since the reproductive success of parasitoids mainly depends on the time and energy required in producing eggs and locating hosts to finally oviposit on; the allocation of resources to survival and reproduction is under strong selective pressure. This trade-off between reproduction and somatic maintenance has been widely studied in parasitoids and evidence that these two life-history traits use the same common resources has been provided (Rosenheim 1996; Sevenster *et al.*, 1998; Ellers *et al.*, 2000; Ellers, 1996).

Parasitoids that invest too much in eggs will not live enough to search for hosts on which to deposit their eggs, whereas parasitoids that invest too much in life span possibly find more hosts than eggs they have to lay. However, parasitoids seem to have evolved resource allocation strategies to reduce either the risk of becoming time-limited (when females have not sufficient time to oviposit all their mature eggs) or egg-limited (when females experience a temporary or permanent exhaustion of their egg supply). That is, strategies that allow females to balance the energy resource allocation to survival and reproduction in order to maximize their lifetime reproductive success are to be selected.

Despite the fact that natural selection is expected to result in reproductive strategies that approach a quantitative match between egg supply and oviposition opportunities (Rosenheim 1996; Sevenster *et al.*, 1998), several factors create stochastic variation in reproductive opportunities making a precise matching of egg load with host availability not possible. That is, for example, variations in weather conditions, host density, food availability and predation on adult parasitoids. Rosenheim (1996), Sevenster *et al.* (1998) and Ellers *et al.* (2000) through theoretical works formally postulated various sources of stochasticity which can potentially have an impact on reproductive opportunities and in this way preclude the aforementioned matching. For instance, stochastic variation in individual life span (namely, time available to search for hosts), stochastic variation in the distribution of host patches in space (uniform, random or clustered) and stochastic variation in the number of hosts per patch (uniform vs. aggregated distribution of hosts across patches). In these papers, models that evaluate the effects of habitat quality and stochasticity on egg load evolution in proovigenic and synovigenic parasitoid were developed as well as the conditions under which egg limitation and time limitation are likely to occur were identified.

Summarizing the findings of Ellers *et al.* (2000), one model assumes that female parasitoids with a *proovigenic* reproductive mode have a fixed amount of resources that have to be allocated to eggs and life span before the start of their adult life. The model also takes into account that the optimal allocation at emergence is determined by the initial egg load associated with the highest expected reproductive success. The predictions are that the initial egg load and the number of hosts found during lifetime can match exactly in absence of stochasticity but, in contrast, in presence of stochasticity the allocation of resources to survival and reproduction is affected and the perfect match is then no longer possible. Hence, when conditions of overall stochasticity present, the optimal egg load exceeds the average number of host encountered in poor habitat whereas in rich ones is lesser than the expected number of host. Besides, the model predicts a low incidence of egg limitation at optimal strategy, increasing only when habitat quality is high and stochasticity is low. It is expected, from the model that parasitoids, especially those that experience low habitat qualities, maximize the number of offspring gain per unit time by becoming eventually time-limited (Rosenheim, 1996; Sevenster *et al.*, 1998; Ellers *et al.*, 2000).

For parasitoids with *synovigenic* reproductive mode, the model assumes that a fixed amount of resources has to be allocated to the initial egg load and the remaining energy reserves. In contrast to the proovigenic model, in which these remaining energy reserves are completely invested in life span (used to search for host), in the synovigenic model the remaining resources can be used either to cover travel costs or to mature more eggs during the adult life. Therefore, even though synovigenic parasitoids are able to adjust their reproductive output to variation in host numbers, the optimal strategy adopted in stochastic environments still being an excess of eggs compared to the expected number of hosts encountered. Furthermore, as they can continue to mature eggs throughout the adult life, the incidence of egg limitation in synovigenic individuals is even lower than in proovigenic parasitoids.

Although there are few studies concerning the incidence of time and egg limitation under field conditions, those that are reported suggest that most individuals end their lives being time limited (Driessen & Hemerik, 1992; Weisser *et al.*, 1997; Ellers *et al.*, 1998).

1.4 Ovigeny index

The ‘ovigeny index’ (OI) is the ratio of the initial egg load to the lifetime potential fecundity. Since this index measures the proportion of the potential lifetime eggs complement mature upon female emergence (Jervis *et al.*, 2001), it can be used not only as a measure of the concentration of egg production early in life, but also as a quantitative measure of the allocation of capital resources (larval resources carried-over to the adult) to reproduction at the start of adult life (Jervis & Ferns, 2004).

An index of 1 (strict pro-ovigeny) denotes that all the oocytes are mature upon emergence, whereas an index of 0 (extreme synovigeny) denotes emergence with no mature oocytes. A continuum between these extremes of OI has been shown to exist within the orders Hymenoptera, Lepidoptera and Diptera (Jervis *et al.*, 2001; Jervis & Ferns, 2004; Jervis *et al.*; 2005). The variety in the OI implies different levels of reproductive plasticity. In parasitic wasps, the emergence with a high proportion of mature eggs leads to a maximization of lifetime potential eggs complement ready to be laid at the start of the adult life, however, at the expense of a reduction in reproductive plasticity. Namely, females with high ovigeny index have a reduced capacity to adjust their egg supply to variations in host availability. On the contrary, the emergence with low proportion of mature eggs implies a gain in reproductive plasticity eventually at the cost of missing oviposition opportunities (Rosenheim *et al.*, 2000; Ellers & Jervis, 2003).

Several hypotheses, that relate the ovigeny to a variety of life-history traits involved in resource acquisition and allocation, have been proposed (Jervis *et al.* 2001; Jervis *et al.* 2003; Jervis & Ferns, 2004). For example, correlations between the degree of egg maturation at emergence and egg type (Jervis *et al.* 2001), egg resorption capability (Jervis *et al.*, 2001), body size (Ellers & Jervis 2003; Jervis *et al.*, 2003), life-span (Jervis *et al.*, 2001, 2003) and feeding habit (Flanders 1950; Boggs 1997; Jervis & Kidd 1986; Jervis *et al.*, 2001).

It is worthwhile mentioning here that the OI provides a more precise measure to test correlations between ovigeny and other life-history traits than the dichotomy between pro- and synovigeny, given that it is a continuous variable ranging between 0 and 1.

1.4.1 *Ovigeny index and trade-offs based on teneral reserves allocation patterns*

As the ovigeny index is a measure of the degree of initial investment in reproduction, it is expected to reflect trade-offs between other initial resource allocation patterns. For example, two hypothesized capital resource trade-offs are between soma and non-soma, and within non-soma, between initial eggs and initial reserves (Figure 1). Capital resources comprise the nutrients obtained through larval feeding. Soma mainly involves the adult's exoskeleton and musculature, and non-soma comprises the reproductive tissues and oocytes, together with initial nutrient reserves (i.e. larval quantities of blood sugars and lipids, glycogen, and protein in the fat body).

In holometabolous insects most of the investment in soma occurs during the pupal stage, therefore it is at this instance that soma and reproduction (non-soma) compete for initial resource allocation. This trade-off was first postulated by Boggs (1981) who argued that species whose females are longer-lived and which have higher resource intake prospects should invest more in building a "sturdy body" (soma) at expense of non-soma than shorter-lived species (resulting thus in an OI<1 , and *vice versa*).

The other hypothesized capital resources trade-off occurs within non-soma with regard to the relative allocation to initial eggs versus initial reserves (that can be used, during adult life, as a source of nutrients for producing additional eggs and for fueling somatic functions). Therefore, this trade-off implies a reproductive cost. An investment in eggs early in life entails a cost upon the adult lifespan of females. This occurs both at intraspecifics and interspecific level (Ellers & van Alphen, 1997; Jervis *et al.*, 2001)

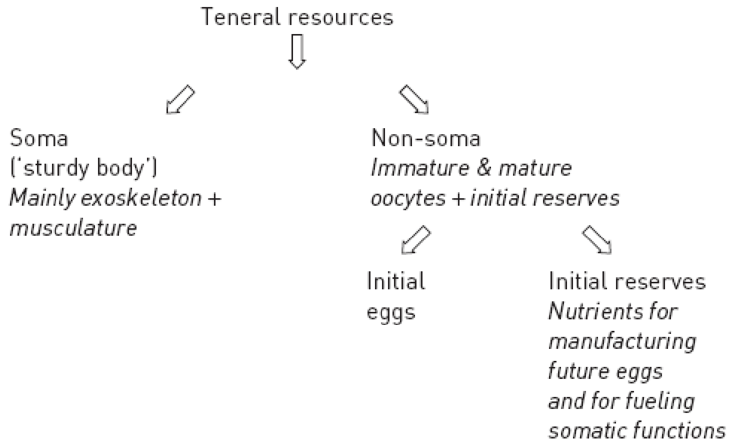


Figure 1. Allocation of carried-over (capital) resources to competing physiological functions in holometabolous insects. Modified from Jervis *et al.* (2005) and taken from Bernstein & Jervis (2008).

1.4.2 *Ovigeny index and life history traits*

The ovigeny index can be related to different life history traits both intra and interspecifically amongst parasitoids. The following paragraphs will deal with those relationships of particular interest in this thesis.

Intraspecifically, body size maintains a positive relationship with various life history traits such as total amount of resources, ovariole number, egg load and longevity (Ellers *et al.*, 1998; Visser, 1994; Jervis *et al.,* 2003); however, ovigeny index declines with body size. That is, smaller wasps have higher proportion of mature eggs at emergence than larger ones. Note that, the ovigeny index numerator (the initial egg load) represents the initial energy investments in reproduction and the denominator (lifetime potential fecundity) represents the initial reserves (fat body storage) which would contribute to maintenance and lifetime fecundity. Taking this into account, the negative relationship between OI and body size within species can be explained as a differential allocation of capital resources to initial egg load and initial reserves (Ellers & Jervis, 2003). As body size increases, the total amount of capital resources increases and allocation to initial egg and initial reserves also increases. Nevertheless, the increase in allocation to initial eggs load is proportionately smaller than the increase in allocation to initial reserves, leading this to a decrease in the ovigeny index (Ellers & Jervis, 2003; Figure 2, **a**).

Indeed, among species, OI is also inversely correlated with body size but the explanation for this relationship is still unclear. Jervis *et al.* (2003) suggested that this negative correlation between OI and body size can be explained in the same way as within species, due to a differential investments of capital resources to reproduction and survival. However, no knowledge about size-related changes in resource allocation, among species, is currently available. Alternatively, they proposed other candidate forces that explain the evolution of the negative correlation. These potential factors should lead to size-related shifts in the egg maturation schedule and they could be variations in habitat quality (host and food) or size-related extrinsic mortality in adult females. For example, small species suffering higher mortality risk than larger ones either by predation, desiccation or starvation and so the tiny species are selected for a shift towards early egg production (Jervis *et al.*, 2003). Anyway, experimental results in Jervis *et al.* (2003) show that as initial egg load declines with increasing body size and potential fecundity is uncorrelated with body size, across species, the decrease in ovigeny index with size is largely explained only by its numerator, the initial egg load (Figure 2, **b**).

All together, the effects of body size on the relative allocation of resources to the initial egg load and the potential lifetime fecundity and, ultimately on variation in the ovigeny index, should be different for the two types of egg maturation strategies. For example, in strict pro-ovigenic parasitoid species, initial egg load always equals the maximum potential lifetime fecundity. Therefore, even though larger individuals may allocate more of the carried-over larval resources to reproduction and emerge with a larger mature egg load, ovigeny index should not be altered with body size (Ellers & Jervis, 2003; Figure 2, **c**). Synovigenic wasps, on the contrary, show variation in ovigeny index across species and within species (Jervis *et al.,* 2001), again mainly, because of body size changes in resource allocation.

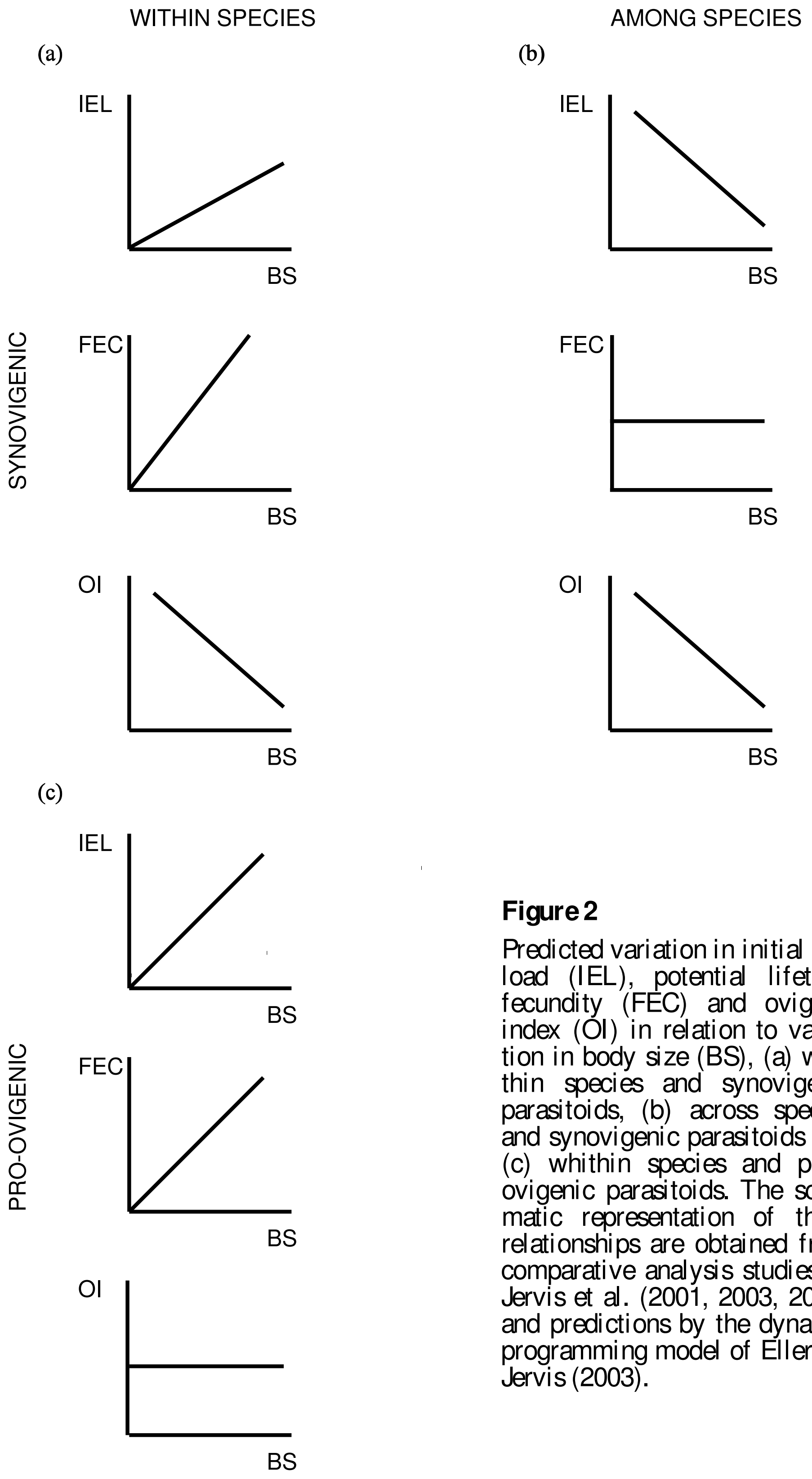


Figure 2

Predicted variation in initial egg load (IEL), potential lifetime fecundity (FEC) and ovigeny index (OI) in relation to variation in body size (BS), (a) whithin species and synovigenic parasitoids, (b) across species and synovigenic parasitoids and (c) whithin species and proo-ovigenic parasitoids. The schematic representation of these relationships are obtained from comparative analysis studies of Jervis et al. (2001, 2003, 2004) and predictions by the dynamic programming model of Ellers & Jervis (2003).

As life history theory based on resource allocation predicts, lifespan and reproductive effort are inversely correlated. Among parasitoid wasps, a negative relationship between OI and life span within and between species is observed. Within species, this correlation could be explained once more as a differential allocation of capital resources to initial egg load and fat body reserves (capital resources trade-off within non-soma; Jervis *et al.*, 2001; Ellers & Jervis, 2003; Jervis & Ferns, 2004). A pre-emergence energy investment in reproduction brings on a post-emergence life-span cost, and as would expected, an increase in the ovigeny index. Under this argument it is hypothesized that pro-ovigenic species should be shorter-lived than synovigenic ones, due to the fact that the former have a greater proportion of mature eggs at birth.

Ovigeny index can also be related to the reliance on adult female feeding. Boggs (1986, 1997) and Jervis *et al*. (2001, 2005) studies suggest, through comparative analysis among species of Lepidoptera and Hymenoptera orders, that the lower the ovigeny index is, the more dependent females are on external nutrient acquisition for fueling post-emergence egg maturation. An OI=1 or nearly one indicates that nutrients used for maturation of the lifetime egg complement is carried over mostly or entirely from larval resources. Conversely, an OI lower than one, indicates that the nutrients used for the egg manufacture derive only partly from larval resources and the remaining of resources needed are obtained by income nutrient at adult stage. Therefore, they suggest interactions between patterns of capital resource allocation to reproduction and survival (i.e., represented by the ovigeny index) and adult feeding habits (i.e., host feeders, nectar only feeders, pollen feeders and non feeding adult).

1.5 Nutritional biochemical resources and utilization

Carbohydrates, proteins, and lipids are the primary nutrient classes and the fat body is the main site of nutrient storage and metabolism for all insects (Chapman, 1998). Dietary sugars (i.e. exogenous) can be used for maintenance immediately, or transformed in trehalose or glycogen for future use. Trehalose (a glucose-glucose dimer) is mainly stored in blood and glycogen is stored in fat body. Lipids are also stored as tryglycerides in the fat body and many key molecules including the lipoprotein vitellogenin, the primary constituent of yolk, and trehalose, can be synthesized there. Insect hemolymph can be considered as another site of nutrient storage as it usually contains high levels of free amino acids, storage proteins, and sugars (trehalose) required for maintenance, metamorphosis, and reproduction (Chapman, 1998; Strandan & Casas, 2008).

Teneral resources comprise the resources obtained through larval feeding. The allocation of these teneral resources to different biological functions can occur both during larval development and as adult individuals. “Capital resources” or “carried over resources” are also terms generally used to refer to those nutrients that are acquired during larval stages and then used during adult life. In this way, insects can use both capital resources and additional nutrients acquired through adult feeding (although not all species feed as adult) to fuel metabolic processes such as reproduction, survival and locomotion (Boggs & Freeman, 2005; Figure 3).

Sugars and glycogen mainly fuel somatic maintenance and locomotion. Carbohydrates appear to be the main nutrient used for flight by Hymenoptera. To some extent, carbohydrates also contribute to egg manufacture over the reproductive life of synovigenic parasitoids. For example, Giron *et al.* (2004) have shown that eggs of the host-feeding, yolk-rich egg-producer wasp *E. vuilleti*, contain an amount of carbohydrates equivalent to that of either protein or lipids. Proteins fuel egg production and proteins obtained presumably through host feeding, fuel both egg production and somatic maintenance. Lipid reserves fuel both egg manufacture and maintenance. Hymenoptera parasitoids are not able to synthesize new lipids from carbohydrates as adults, making lipids acquired during larval development a non-renewable resource that eventually constrains egg production (Ellers, 1996; Rivero & Casas, 1999; Olson *et al.*, 2000; Rivero *et al.*, 2001).

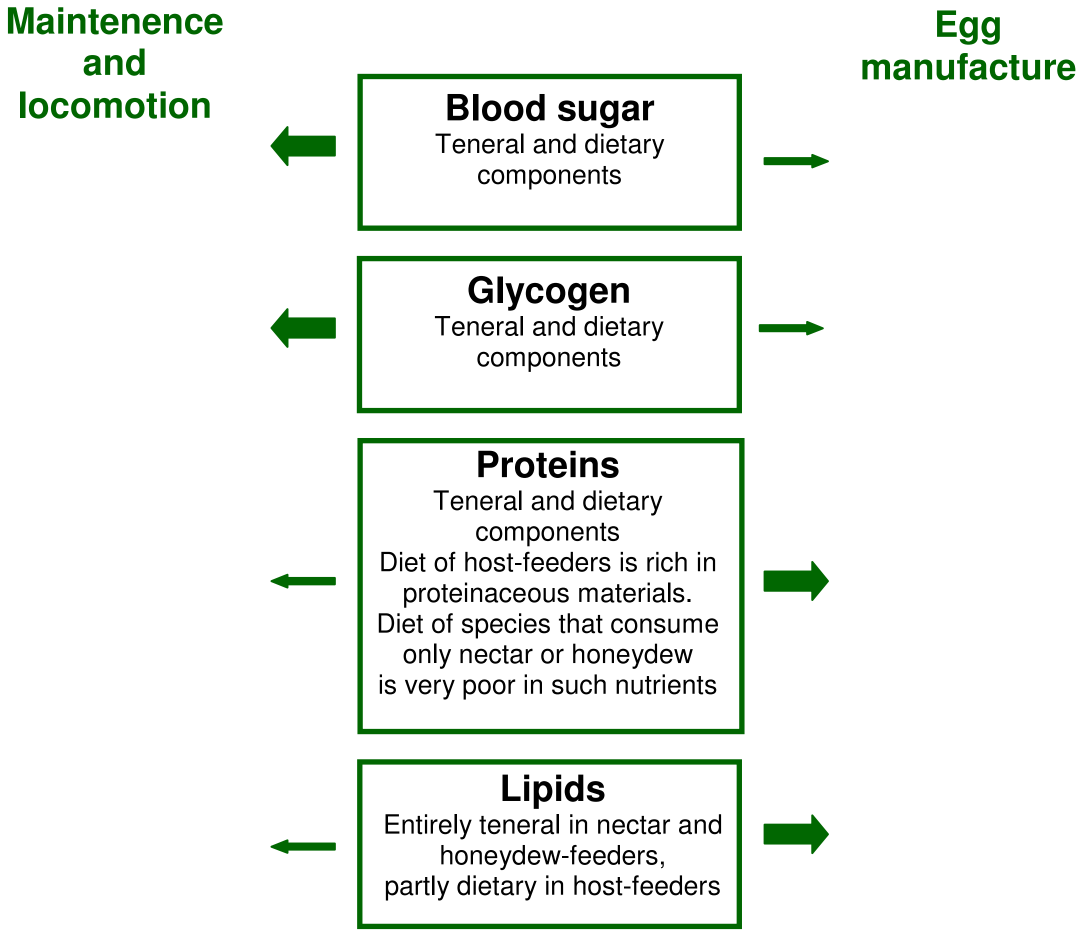


Figure 3. Utilization of nutrient resources by parasitoid wasps. Nutrients can be acquired both by larval and adult feeding. The width of the arrows indicates the presumed importance of a particular resource to a particular physiological function. Non host-feeding parasitoids use blood sugar and glycogen mainly for fueling somatic maintenance and locomotion (flight, walking) but they use proteins and lipids mainly for fueling egg manufacture (modified from Bernstein & Jervis, 2008).

1.6 Nutrient acquisition and allocation strategies of larval and adult parasitoids

1.6.1 *Teneral resources*

Nutrient acquisition and allocation strategies of parasitoids are strongly linked to two components of life-history: (i) the egg maturation strategy (pro-ovigeny *vs.* synovigeny) and (ii) the larval development mode (idiobiosis *vs.* koinobiosis) (Strand & Casas, 2008;

Jervis *et al.*, 2001). *Teneral reserves* obtaining by larval feeding can be mainly allocated to the ovaries or the fat body (Rivero & Casas, 1999; Jervis *et al.*, 2001). Through larval stages, pro-ovigenic parasitoids would invest most of the nutrient resources in egg production and little resources would be allocated to the fat body storage. Then, during the adult stage capital reserves are allocated to body maintenance and locomotion. In contrast, synovigenic parasitoids result because a major part of the teneral resources is allocated to storage in fat body and less to initial eggs (mature eggs available at emergence); as higher the degree of resource allocation to fat body is, the stronger would be the degree of synovigeny. In this way, synovigenic parasitoids as adults, have to deal with the problem of how allocate teneral reserves not only to body maintenance and locomotion but also to egg production (Jervis & Kidd, 1986; Rivero & Casas, 1999; Strand & Casas, 2008).

The egg maturation strategy and the larval development mode are also tightly linked factors. Generally, the extreme synovigenic parasitoids are idiobionts and produce yolk-rich (anhydropic) eggs, whereas the weak synovigenic or pro-ovigenic parasitoids are koinobionts and tend to produce yolk deficient (hydropic) eggs (Strand & Casas, 2008). Host feeding behavior (see below) by the adult females appears to be more prevalent in idiobiont than in koinobionts. Given that koinobionts are unable to host feed, many species have evolved compensatory strategies for enhancing the acquisition of nutrients during larval development that are lacking in the adult diet. Several koinobiont species manipulate host nutritional physiology in a manner that increases nutrient availability in the host hemolymph and disrupts nutrient uptake by host tissues (redirecting energetic resources away from the host towards the larval parasitoid) and so parasitoids enhance their capital reserves (Kaeslin *et al.*, 2005; Pennacchio & Strand, 2006).

1.6.2 *Income resources: origin and composition*

There are two major kinds of foods for adult parasitoids allowing them to be classified accordingly. Parasitoids that feed upon their hosts are called *host-feeders* and those that do not feed on host are termed *non-host feeders.* Whereas the former feed on their hosts may also do so on sugar- rich sources (Jervis & Kidd, 1986; Heimpel & Collier, 1996; Heimpel *et al.*, 1997; Casas *et al.*, 2005), the latter species rely mainly on sugary sources (Bernstein & Jervis, 2008).

The main non-host foods for parasitoids comprise floral and extra floral nectar and the honeydew excreted by some Homoptera (Jervis & Kidd, 1986; Jervis *et al.,* 1992; Rivero & Casas, 1999). Consumption of pollen is practiced only by a few parasitoids (mutillids, scoliids, and some bombyliids) and feeding on other materials, such as fungi, is likewise generally rare (Jervis *et al.*, 2008). Nectar and honeydew are rich in carbohydrates but poor in amino acids and lipids (Baker & Baker, 1983).That is, nectar composition mainly consists in glucose, fructose, and sucrose, however other sugars (maltose, melibiose, erlose and melezitose) or substance (organic acids, terpenes, alkaloids, flavonoids, vitamins, phenolics and oils) are reported but are present in small quantities (Carter *et al.*, 2006; Nicolson & Thornburg, 2007). Sugar proportions vary among different plant species (Baker & Baker, 1983). Hence, nectar from different flower species can have different effects on life history traits (i.e.: flight, longevity, fecundity) and thus parasitoids can have specific floral preferences (Wäckers, 2004; Wanner *et al.*, 2006). Honey is widely employed in the laboratory and under mass-rearing conditions as a substitute for nectar or honeydew because of its similar biochemical composition.

Host-feeding is widespread among parasitic wasps. Host feeding, consists in the consumption of the host's body fluids or tissues, such as host's hemolymph and fat body (Rivero & West 2005; Jervis *et al.*, 2008). Host blood is rich both in nitrogen and carbohydrates, with a lesser amount of lipid components (Giron *et al.*, 2002, 2004). That is, the host hemolymph provides amino acids and proteins, and the main carbohydrates are comprised by sugar trehalose.

Apparently, parasitoid wasps can synthesize storage proteins *de novo* from dietary amino acids. However, only host-feeders have a dietary income which allows this to occur. On the contrary, as it was previously mentioned, there is no evidence of lipogenesis in adult parasitoid wasps (Rivero & Casas, 1999). Therefore in these organisms larval lipid reserves are non-replaceable resources and thus potentially limiting.

1.7 Effects of adult feeding on life history traits and behaviours

Ingested sugars can be converted in carbohydrate reserve (trehalose or glycogen) or used for fueling mainly somatic maintenance, locomotion and in lesser extent for egg production. It is widely known, from an array of laboratory studies, that adult feeding can increase longevity and fecundity in many parasitoid species (Heimpel *et al.*, 1997; Olson *et al.*, 2000; Schmale *et al.*, 2001; Siekmann *et al.*, 2001; Tylianakis *et al.*, 2004; Bezemer *et al.*, 2005; Desouhant *et al.*, 2005), and can also increase several components of locomotory activity such as flight initiation and capacity or walking activity (Wanner *et al.*, 2006; Jervis *et al.*, 2008). Despite the important fitness consequences of food evidenced from the numerous laboratory measurements, knowledge of food foraging in field conditions is limited (Sisteron & Averill 2002; Lee & Heimpel, 2008; Desouhant *et al.*, 2010).

An increase in the rate of ovigenesis in sugar fed females compared to unfed females as observed by Bezemer *et al.* (2005) could be the result of an indirect effect of this sort of food. In other words, the effect of food on egg production could be attributable to the nutrients in the food lessening the catabolic drain that somatic maintenance exerts upon carried-over resources that fuel ovigenesis (Bernstein & Jervis, 2008). It is likely, that parasitoid wasps whose diet is confined to both nitrogen- and lipid-poor non host foods, such as nectar and honeydew, can gain little in terms of egg manufacture by feeding (Bernstein & Jervis, 2008). Host feeding, on the other hand, can increase egg production as well as life expectancy (Heimpel *et al.*, 1997; Rivero & West, 2005). Whether both or one of these fitness components is enhanced depends on the parasitoid species (Jervis *et al.,* 2008). To actually know whether a direct involvement of exogenous nutrient exists in egg production nutrient tracking techniques (i.e. radiotracer or isotopes) should be used.

1.7.1 *Parasitoid foraging decisions: implication on fitness of food or host searching*

Taken account that females of the vast majority of parasitoid wasp species consume some kind of food during their adult live and the potential positive effects of the dietary nutrients have on key life-history traits such as longevity and fecundity, several modeling works have been developed in order to investigate fitness consequences of food foraging.

In many parasitoids, oviposition and feeding do not occur simultaneously and host and food patches are usually concurrently available but may be spatially separate. Visiting food patches is likely to increase the risk of mortality mainly because of extrinsic factors such as weather, predation and desiccation (Heimpel *et al.*, 1997; Rosenheim, 1998; Jervis *et al.*, 2003). Under this scenario, females have to make decisions about which of the two resources (host or food) they should forage to balance the *trade-off* between immediate and future reproduction. On the one hand, while foraging for hosts will increase 'immediate' fitness gains, life expectancy will decrease. On the other, by foraging for food, females will postpone egg-laying to a later stage in adult life (which translates into 'immediate' reproductive opportunities skipped) but gain future oviposition opportunities, due to the potentially positive effects of food on life expectancy and egg production. In addition**,** parasitoid foraging decisions are likely to be state-dependent, that is, individual behaviour relies on the nutritional state, age, egg load etc (Bernstein & Jervis, 2008).

Sirot & Bernstein (1996) and Tenhumberg *et al.* (2006) formulated state-dependent models, posing the question of how females should allocate their time and energy in food and host foraging in order to maximize their fitness gains. These food foraging models assume that parasitoids are pro-ovigenic and time limited and also that host and food patches are concurrently but spatially separate. However, these models differ in some assumptions with respect to parasitism and food consumption rates, number of resource patches and which factor more strongly determines the strategy to adopt: the availability of food (environmental factor) or energy reserves (internal factor).

Sirot & Bernstein (1996) model, giving more relevance to the environmental factor, predicts that under conditions of high food availability, parasitoids should delay searching for food until reserves become low. However, when food availability is moderate, parasitoids should not delay food searching, and when food availability is low, parasitoids should never search for food. In contrast, the Tenhumberg *et al.* (2006) model, giving more relevance to the internal factors, predicts that parasitoids should always search for food if energy reserves drop to low levels, even if there is a low probability of finding food and also if the average food reward is low.

The weaknesses of these models are that none of them takes into account egg dynamics (egg depletion). In addition, these models are unsuitable for synovigenic parasitoids.

Bernstein & Jervis (2008) proposed two new models that deal with the shortcomings of the Sirot & Bernstein (1996) and Tenhumberg *et al.* (2006). Basically, they take account two egg maturation strategies (synovigeny and pro-ovigeny specifically non host feeders), egg dynamics and the compartmentalization of biochemical resources in the synovigenic model, i.e. sugars and glycogen into metabolic resources, and protein and lipids into egg production resources. This latter compartmentalization of resources becomes to solve the unrealistic assumption that egg manufacture and somatic functions are fueled from a "single resource pool". Remember that parasitoids can utilize different resources to fuel different functions (Figure 3). Moreover, patterns of resource utilization can be altered during lifetime due to either fuel depletion and/or limitations as result of low food availability and low food quality. Both models(synovigenic and pro-ovigenic models), assume that optimal behaviour is influenced by two *environmental factors*: (1) inter-patch distance and, (2) the probability of finding food as well as by three *physiological factors*: (1) energy reserves (metabolic resources), (2) egg load in the case of pro-ovigeny or egg production resources in the case of synovigenic and (3) the ovigeny index.

Although the detailed outcome of these models escapes the interest of this dissertation, a few results are worth mentioning. For some combinations of parameters values, the models of Bernstein & Jervis (2008), yield similar results to those of Sirot & Bernstein (1996) and Tenhumberg *et al.* (2006). In addition, the main differences between pro-ovigeny and synovigeny are firstly, that for synovigenic, food-searching is less likely not only at low egg loads but also at high egg loads, while for pro-ovigenic, when egg load is low, animals should not feed to satiation as feeding entails a mortality risk and moderate resources would suffice to lay the remaining eggs. Secondly, in terms of the expected frequency of food-searching, pro-ovigenic parasitoids are more likely to feed compared with synovigenic species. The latter finding can be explained given that in the synovigenic model, females eventually draw upon egg production resources to fuel maintenance, which thus reduces the need to obtain exogenous nutrients. Finally, Bernstein & Jervis, 2008 suggest that differences in behavior between the Sirot and Bernstein (1996) and Tenhumberg *et al.* (2006) models stem more from their parameter values used rather than from the biological scenarios depicted.

While the role of female foraging behavior in parasitoid-host population dynamics has been well studied (Hassell & Waage 1984; Hassell, 2000), surveys on the dynamical consequences of food-foraging are still limited (Krivan & Sirot, 1997)

1.8 Dispersal behaviour

Most animals move but the degree of movement varies greatly between individuals and species. For example, insects spend a large part of their lives moving between locations that offer different resources (i.e. food, mates and oviposition sites). Such movements can take place at very different scales, from moving between patches to long-distance displacements between remote populations (Johnson, 1969; Desouhant *et al.*, 2003). It is generally assumed that dispersal implies displacements at larger scales.

Dispersal plays an important role in several ecological and evolutionary processes. For example, when individuals disperse between local populations they may well affect the densities and dynamics of the metapopulation system (Hassell *et al.*, 1991; Murdoch *et al.*, 1992). Likewise, dispersal promotes colonization of previously unoccupied habitats, altering the distribution and abundance of species (Roland & Taylor, 1997; Brigg & Latto, 2000).

Dispersal is either strongly influenced by environmental factors (i.e. habitat fragmentation, light, wind, vegetation density) or internal factors (reproductive status, nutritional reserves, age, health state) (Desouhant *et al.*, 2003; Villacide & Corley, 2008; Elliott & Evenden, 2009). In addition, dispersal can involve different mechanisms that are broadly categorized as active (locomotory) or passive (i.e. wind, water currents, animal vectors), and the relative contributions of both components of dispersal are highly variable among species**.**

Flight is energetically very costly in insects and relies for fuelling on different biochemical nutrients such as carbohydrates, lipids and proteins (Chapman, 1998). The kind of fuel used in flight will depend basically on the species, individual nutritional state and foraging habits (O'Brien, 1999). In some species of Lepidoptera (butterflies or moths) and Orthoptera (locust) lipids are the main sources of energy for sustained flight while in Diptera (mosquitoes) and Hymenoptera (wasps) flight is primarily fueled by carbohydrates. However, there are species that vary the fuel use temporally presumably due to specific nutrient depletion or ecological factors such as variation in food availability (O'Brien, 1999; Rivero & Casas, 1999). Hence, nutritional condition and metabolic reserves may be critical factors determining the success of dispersal events (Dudley & Srygley, 2008).

1.9 Host foraging, information acquisition and patch time allocation

Hosts usually occur in discrete patches bearing a variable number of them. In addition, host patches can be separated by important distances so that parasitoids must distribute their time between them to maximize the number of offspring that they can produce. Female parasitoids start searching for hosts shortly after emergence. During host searching, parasitoids respond to a variety of chemical, visual or physical cues that provide information which enables them to restrict their foraging behaviour to that part of the environment in which hosts are most likely to be encountered (Godfray, 1994; van Alphen *et al.*, 2003). For example, chemical cues are generally volatiles produced by the plant in response to herbivory (synomones), volatiles derived from the host (so-called kairomones; e.g. host aggregation or sex pheromone) and/or emitted by host associated organisms (i.e. microbes and fungus); and can be used as stimuli in host habitat location from a distance. The non-volatiles cues, such as visual (i.e. damage pattern and host movement) and physical cues (i.e., substrate vibrations) or non-volatile host products (i.e. feces and secretions of mandibular and accessory glands) are perceived and used by parasitoids in short distance host location (Vet and Dicke, 1992; Turlings & Wäckers, 2004). After arrival in the patch, the presence and concentration of kairomones can inform the parasitoid about host density.

To maximize their reproductive success, females should not only optimize host exploitation strategies, but also optimize host patch exploitation rules. Therefore, parasitoids should be able to adjust times allocated to each patch encountered. In this respect, The Marginal Value Theorem (MVT, Charnov, 1976) is the first model that predicts the optimal moment at which an animal, that forages its own, should leave a patch and search for another one. An optimal forager should leave a resource patch when the depletion of that patch is such that the instantaneous rate of fitness gain falls below the average rate of gain expected for the habitat. The main predictions from this theorem are that (1) patch time should increase with host density in the patch; (2) patch times should increase with increasing average travel time in the habitat and should decrease with increasing average host density in the patches; and (3) patches of different quality should be reduced to the same level of profitability when leaving (Figure 4). These predictions were repeatedly tested on several parasitoid species (see Wajnberg, 2006 for a review; Cook & Hubbard 1977; Hubbard & Cook, 1978; Waage, 1979; Corley, 1999, Driessen & Bernstein, 1999; Thiel & Hoffmeister, 2004). It is important to note that the model assumes that the foragers are omniscient and thus possess a perfect knowledge about the environment (Stephens & Krebs, 1986). This assumption is highly unlikely to occur in nature, especially as most habitats are highly variable. It is likely therefore, that animals estimate the actual habitat quality by means of gathering and updating information on host availability while searching for the host itself (Pierre *et al.*, 2003; Tentelier *et al.*, 2006; 2007).

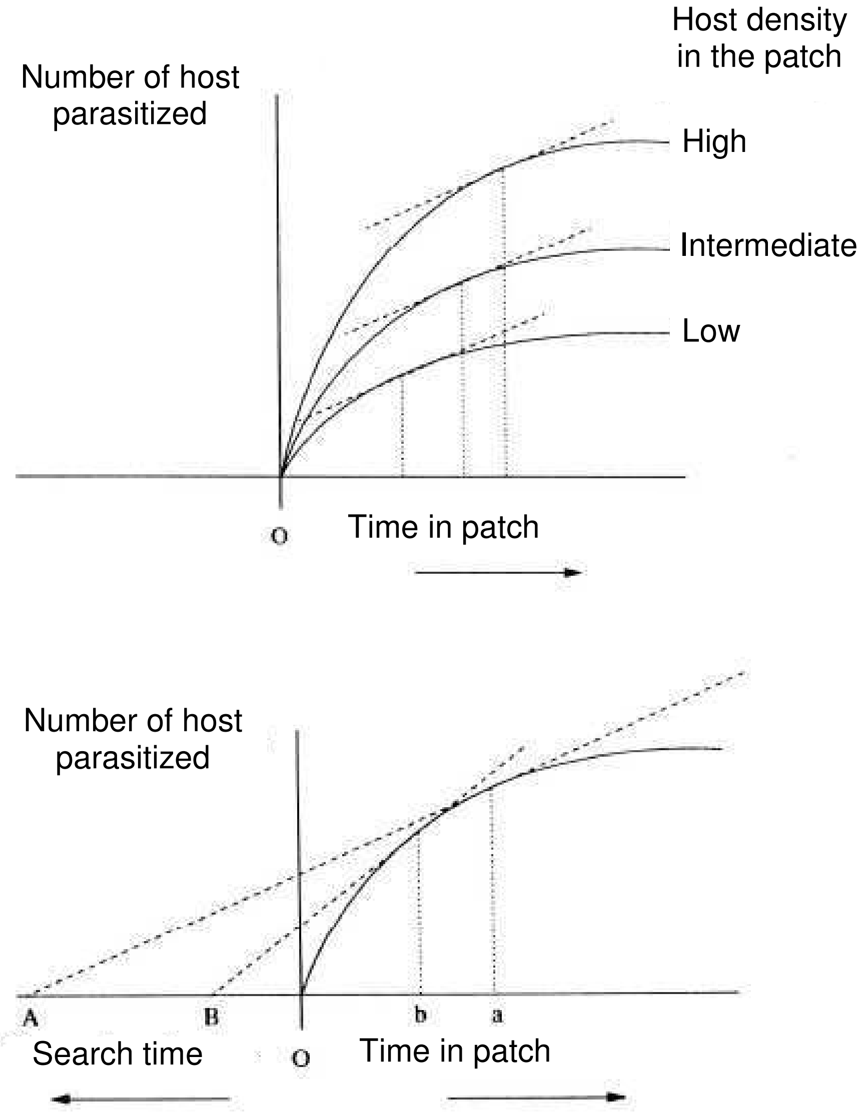


Figure 4. The Marginal Value Theorem (Charnov, 1976). The curve represents the cumulative number of hosts attacked by parasitoids. **(a)** The parasitoid should leave a patch when its marginal rate of gain of fitness (the slope of the cumulative parasitism curve) falls to the maximum average gain rate that can be achieved in the environment (represented by the slope of the three dashed lines). As higher is the quality a patch longer time the parasitoid should stay there. **(b)** If the travel time between patches is *A*, the optimal patch residence time *a* is the point where a line rooted at *A* is just tangent to the cumulative parasitism curve. Then, shorter travel time (*B*) leads to shorter optimal patch residence times (taken from Godfray, 1994).

Others models have been proposed to better understand the mechanism that foragers should employ to achieve the optimal residence time, the so-called rules of thumb. The best known rules for patch leaving decisions predict that a parasitoid should leave a patch, (1) after not having found an unparasitized host during a fixed period of time; (2) after a fixed period of time or (3) after a fixed number of hosts has been found (Iwasa *et al.*, 1981; McNair, 1982; Green, 1984; Stephens and Krebs, 1986). The rule used by a forager depends largely on the characteristics of the environment. In addition, more complex and dynamic models, based on motivational processes, have been proposed. They are the incremental and decremental mechanisms for deciding when to leave a patch (see Figure 5 for mechanisms description; Waage, 1979; Driessen *et al.*, 1995; van Alphen *et al.,* 2003). Which of these mechanisms parasitoids are more likely to use depends on the distribution of the hosts and on the reliability of the information about host availability. Parasitoids foraging for hosts with an aggregated distribution (high variability in the number of host per patch) should use incremental mechanisms, whereas those foraging for uniformly distributed hosts (low variability in the number of hosts per patch) should use decremental mechanisms. At the same time, when the number of hosts in a patch is known, as when the kairomone concentration is a reliable indicator of the number of hosts in the patch, each oviposition implies a subsequent decrease in host availability. The patch becomes progressively less valuable compared with the rest of the environment and animals should use a decremental mechanism. On the other hand, when information on host availability is limited due to parasitoids can detect the presence of hosts in a patch but not their number (e.g. when host numbers are high) the incremental mechanisms would be more adequate. In this case, the incremental mechanism will lead to a longer patch residence time and to a more thorough exploitation of the richest patches (van Alphen *et al.,* 2003).

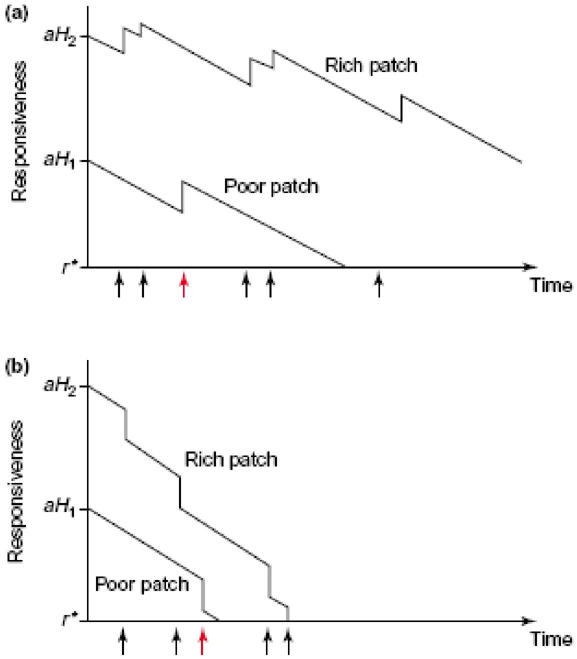


Figure 5. Patch leaving rules adopted by parasitoid females to manage their foraging time on patches of hosts. (a) Incremental mechanism (Waage [1979] model). Parasitoids are assumed to have a certain level of responsiveness (tendency to stay in the patch), and the initial responsiveness (aHi) is determined by the kairomone concentration, which is proportional to host number (Hi). Responsiveness decreases linearly with the time spent on the patch. As a result of ovipositions, responsiveness increases and the increment being dependent on the time elapsed since the last oviposition. When the level of responsiveness has dropped below the critical threshold, *r**, the parasitoid leaves the patch. (b) A decremental patch-leaving decision rule. As a result of ovipositions, the initial responsiveness to kairomones decreases, which means that the parasitoid is more likely to leave a particular patch. Black arrows denote ovipositions in a relatively rich patch; red arrow denotes an oviposition in a relatively poor patch (Reproduced from van Alphen *et al.,* 2003).

There are many studies that examine the different factors which can influence patch exploitation strategies by parasitoid females (for a review see Wajnberg, 2006). Either *environmental factors* such as host patch availability (Thiel *et al.,* 2006), number of host in the patch (Boivin *et al*., 2004; Amat *et al*., 2006), host quality (in term of host species, size, instar, healthy or parasitized host); or *internal factors* such as parasitoid physiological state (egg load, nutritional reserves, age) and parasitoid previous experience, show strong influence on patch leaving decision and therefore on patch residence time (Li *et al*., 1997; Frechette *et al.,* 2004; Goubault *et al*., 2005; Thiel & Hoffmeister, 2004; Thiel *et al*., 2006).

1.10 State dependency

Many behavioural decisions parasitoids make are dependent on the state of the animal (i.e. age, energy reserves, egg load). This is because behaviuors not only rely on external environmental conditions but also on the internal state of the organism. For example, choices can be contingent upon physiological variables such as nutritional state and egg load. Females having low nutritional levels are expected to food-search rather than host-search (Wäckers, 1994; Sisterton & Averill 2002; Desouhant *et al*., 2005). Likewise, when parasitoids are host-feeders the amount of mature eggs in the ovaries can determine whether female should feed or oviposit on hosts that they encounter (Heimpel & Collier, 1996). In these cases, food may provide the nutrients required to fuel egg production but, this might not be the case for the non-host feeding species, where whatever their egg load, the reproductive gain derived from the act of feeding is still unclear (Bernstein & Jervis, 2008). Stochastic dynamic models have been used on several occasions to find optimal patch time allocation of female parasitoids (Nonacs, 2001; Wajnberg *et al.,* 2006). On the contrary to the static MVT, their aim is to find optimal behavioral strategies, taking into account the state of the foraging females (McNamara & Houston 1990; Houston *et al.,* 1988; Clark & Mangel, 2000). While *foraging models* usually define the optimal strategy as the one that maximizes the net rate of host encounters, *dynamic modeling* defines a set of optimal strategies according to time-dependent factors such as the level of energy reserves and the number of eggs carried.

1.11 Behavioral ecology of insect parasitoids and biological control: is there a link?

Biological control is defined as the reduction of the pest population by natural enemies. Natural enemies of insect pests, the biological control agents, include predators, parasitoids, and pathogens. Insect parasitoids are the most important agent used in the context of pest bio-control.

There are three broad kinds of bio-control that describe different ways in which natural enemies can be used: *importation, augmentation, and conservation*. *Importation or classical biological control* makes use of host-specific parasitoids imported from the region of origin of invasive pests. Success in this category of biological control requires a long-term regional suppression of pest population. An *augmentative biological control*, involving the periodic release of insectary-produced parasitoids, can be effective, when parasitoids of invasive or indigenous pests are unable to persist or to increase in numbers quickly enough to suppress pest damage. Augmentative biological control includes *inoculation or inundation* approaches. *Inoculation* consists on the release of small numbers of parasitoids to improve colonization and provide season-long control. *Inundation* consists on the release of large numbers of parasitoids for immediate suppression, without a lasting impact, at very local scale. Finally, *conservation biological control* focused on the enhancement of the environment suitability for parasitoid populations through provisioning of limiting resources. For each of these categories, success in the suppression of an insect pest is dependent upon the behavioral decisions made by parasitoids in both searching for and parasitizing hosts (Mills & Wajnberg, 2008). Thus, the behavioral ecology of insect parasitoids should be central to the successful implementation of biological control programs.

For example, an introduction of an exotic parasitoid for the implementation of classical biological control implies two phases: *establishment and impact*. On the one hand, *the establishment* of an exotic species is determined by the ability of a small founder population to survive and reproduce in a novel environment (Liebhold & Tobin, 2008). Therefore, the establishment phase of importation biological control agent seems likely to be influenced by general population processes (i.e. Allee effects, genetic bottlenecks, demographic stochasticity, etc). However, success in establishment could be enhanced through manipulation of parasitoids holding conditions in order to maximize the fitness of individuals destined for field release. That is, rearing conditions might have influence on reproductive capacity and/or behavior characteristics of parasitoids, such as patch and host finding and dispersal rates, improving in this way performance (Hougardy *et al.*, 2005; Hougardy & Mills, 2006; 2007) and establishment.

On the other hand, *the impact* of established parasitoid is determined by the pest suppression achieved. In this way, the impact could be supported or constrained by, parasitoid host patch choice decision and exploitation rules (Mills & Wajnberg, 2008). For example, parasitoids can assess variation in host densities among patches from a distance using volatile infochemical signals (Geervliet *et al.,* 1998, Corley, 1999; Martinez *et al.*, 2006; Fischbein *et al.* in prep. Section 2, Chapter IV). Then, patch choice decisions determine the distribution of parasitoid foraging effort among host resulting in more o less host-parasitoid match. That is, the greater the ability of parasitoids to match their foraging effort to the heterogeneity in host densities among patches, the bigger the potential impact of parasitoids on the host population.

In addition, considerable attention has been paid to the influence of food on lifetime reproductive success in parasitoids (Schmale *et al.*, 2001; Siekmann *et al.*, 2001). The availability and quality of food sources are important for sustain biological functions. It is a widely held view that under some conditions food provisioning, in the context of conservation biological control, may improve parasitoid success as biological control agents (Jervis & Kidd, 1996; Lewis *et al.*, 1998; Cortesero *et al.*, 2000). Note however that the provisioning of nectar subsidies raises questions of optimal patch choice such as whether females should search for nectar subsidies to support future reproduction or for hosts to maximize current reproduction.

1.12 Study system: the host-parasitoid interaction

The woodwasp *Sirex noctilio* Fabricius (Hymenoptera: Siricidae) is a phytophagous insect that naturally infest conifers in Eurasia and North Africa. In the last century, *S. noctilio* has invaded several other regions of the world such as Australasia (Madden, 1988), South Africa (Tribe, 1995), South America (Iede *et al.*, 1988; Corley *et al.,* 2007; Villacide & Corley, 2008) and North America (Hoebeke *et al.*, 2005). *S. noctilio,* as an invasive forest insect displaying pulse-like eruptive outbreaks dynamics, has become one of the most economically significant pests of softwood forestation in most invaded areas (Madden, 1988; Hurley *et al.*, 2007).

This wood-boring wasp lay their eggs in the sapwood usually of stressed trees during the summer months and larvae grow and develop inside the wood throughout the rest of the year. *S. noctilio* larvae need of a prior alteration of wood to feed on it and thus attain their development. The decomposition of wood is carried out by a symbiont organism, the basidiomycetous fungus *Amylostereum areolatum*, whose spores (stored in mycangial sacs of *S. noctilio* females) are inoculated into trees during oviposition. Together with fungal spores phytotoxic mucus is also inoculated, that creates the favorable conditions for fungal growth (Coutts, 1969).

Figure 6. *Sirex noctilio* female wasp during oviposition.

The overall spatial pattern of attacked trees within plantations is strongly aggregated. Corley *et al.* (2007) have showed that this spatial pattern increases its aggregation with an increase in the number of attacked trees, through time. This can be the consequence of limited female woodwasp spatial redistribution; namely, newly individuals redistribute within the area of the original aggregation attacking healthy pine trees. It has been observed that through the repeated inoculations of fungal spores and mucus on healthy trees, these may become more attractive and susceptible to *S. noctilio* attacks (Coutts & Dolezal, 1969; Madden, 1974; Spradbery, 1977).

The strong spatial aggregation of attacks by *S. noctilio* and intermittent drought levels occurring during the woodwasp emergence season may well contribute to the observed population outbreaks dynamics (see Madden, 1988 and Corley *et al.*, 2007 for an extensive explanation). The epidemic levels reached by this insect can cause significant economic damage producing extensive trees mortality in pine plantations; therefore, much effort has been focused on successfully managing populations. Integrated pest management programmes have included timely silvicultural treatments of forests and the introduction of biological control agents such as the parasitoids *Ibalia leucospoides* (Hymenoptera: Ibaliidae), *Rhyssa persuasoria* and *Megarhyssa nortoni* (Hymenoptea: Ichneumonidae), and the parasitic nematode *Beddingia* (=*Deladenus*) *siricidicola* (Tylenchida: Neotylenchidae) (Haugen *et al.*, 1990; Haugen & Underdown, 1993; Villacide & Corley, 2006; Hurley *et al.*, 2007; Corley & Bruzzone, 2009).

Ibalia leucospoides Hochenwarth (Hymenoptera, Ibaliidae) is a parasitoid of the woodwaps *Sirex noctilio* (Spradbery, 1977; Madden, 1981). This wasp species is native to Europe but has been introduced into Australasia in the early sixties to control damaging woodwaps populations. Since then, it has established together with its host throughout the invasion range of *S. noctilio* (Madden, 1988). In Patagonia (South of Argentina) pine plantations, where the host and its parasitoid inhabit, are mainly located in the steppe. There, low scrub like vegetation and short grasses predominates, so pine plantations appear as distinct woodland patches (Figure 7).

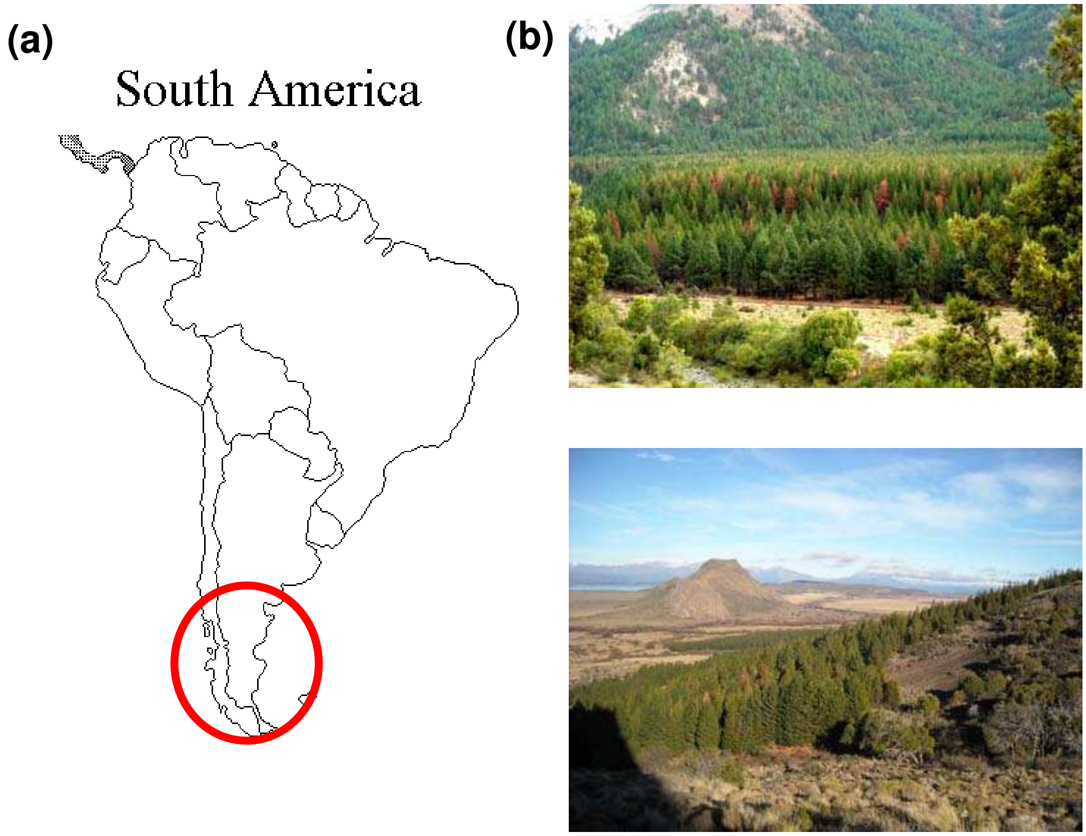


Figure 7. (a) Map of South America, the red circle indicates the location of Patagonia; (b) a view of typical pine tree plantations in Patagonia (Photo by J. Villacide).

I. leucospoides is a solitary, koinobiont parasitoid that attacks eggs and first instar larva of *S. noctilio*. Female parasitoids behave as endoparasitoids until the third instar, a stage at which it becomes an ectoparasitoid. Parasitoid life cycle is closely related with the development of the woodwasp larvae, usually lasting one year (but see Corley *et al.*, 2004).

Parasitoids are attracted to stressed, host-attacked trees by using volatile cues emitted by trees (α- and β- terpenes) and a host symbiotic fungus (Madden, 1968, 1988; Spradbery, 1970a,b; Martinez *et al.*, 2006). Once on tree, *I. leucospoides* searches for hosts exploring tree stems with theirs antennae (Figure 8).

Figure 8. *Ibalia leucospoides* female.

1.13 Aim of this thesis

The central aim of this thesis is to explore how natural selection shapes life history traits and behaviuors according to the ecological surroundings experienced by the animals. This overall framework leads to specific hypotheses of how natural selection might have shaped the reproductive, dispersal and foraging processes in the parasitoid *Ibalia leucospoides.* Understanding the mechanisms involved in the expression of behaviour and life history traits is also a central issue and an important part of my efforts were directed to study them. In addition, particular attention is given throughout this study to the role that resource availability and the habitat context might have in shaping observed resource allocation patterns and life-history strategies in the female parasitoid.

I. leucospoides is a parasitoid whose attractiveness relies on the fact that is a key biological control agent of what is probably the most important pest of softwood forestations in the Southern hemisphere. In this sense, providing information on those central and fundamental aspects of their biology may improve substantially our understanding of the role of parasitoids in bio-control of forest pests in general.

SECTION 2

RESULTS

Chapter I

Life history traits and body size

1. INTRODUCTION

Trade-offs arise as a consequence of the differential allocation of limited resources to different competing biological traits, such as reproduction, body maintenance, growth or locomotion (Roff & Fairbairn, 2007; Stearns, 1989). Since survival and reproduction are often the main factors limiting the reproductive success of parasitoids, the *trade-off* between them is one of the best studied (Ellers, 1996; Ellers & van Alphen, 1997; Hougardy *et al.*, 2005).

Even though adult parasitoids are free-living, their larvae feed on and develop on a host species. Therefore, adult size and reproductive success of parasitoid depend on both: i- the size and nutritional quality of the individual hosts attacked and, ii- the nutrient uptake and host resource allocation carried out by parasitoid as larvae. In fact, adult body size varies intra-specifically and is strongly correlated with larval feeding history and with several other life history traits (Godfray, 1994). For example within species, body size covaries with initial mature egg load, ovariole number, potential and realized fecundity and longevity, where larger wasps have larger resources to allocate to different functions (Honek, 1993; Visser, 1994; Ellers *et al.*, 1998a; Ellers *et al.*, 2001; Jervis *et al.*, 2003; Ellers & Jervis, 2003).

The ovigeny index - defined as the ratio between the initial egg load and the potential lifetime fecundity - has been suggested by Jervis *et al.* (2004) as a good measure of the relative distribution of parasitoid larval resources to reproduction and other functions (Jervis *et al.*, 2001). In contrast to the positive relationships between size and life-history traits, the ovigeny index countervaries with body size. It has been suggested that the reason is that while the two fitness components involved in the ovigeny index positively correlate with body size within species, the proportionate increase of initial egg load with body size is smaller than the proportionate increase in potential fecundity, resulting in a decline of the ovigeny index with body size (Olson & Andow 1998; Ellers & Jervis, 2003; Jervis & Ferns, 2004). Consequently, this negative relationship would mirror a higher degree of allocation of larval resources to fat body (and lifespan fecundity) at the expense of reproduction at the start of adult life. For more details refer to the general introduction (*Ovigeny index and life history traits*).

The differential energy allocation towards life history traits shape, therefore, intra and also interspecific differences among individuals. As noted previously, the size of female wasps can play an important role in the resulting resource allocation strategy. In synovigenic parasitoids, the observed variation in the ovigeny index within and across species could be a direct outcome of body size, affecting the relative allocation of resources to the initial egg load and potential lifetime complement of eggs. However, in strict pro-ovigenic parasitoids, initial egg load always equals the maximum potential lifetime fecundity. Hence, even though larger individuals may allocate more larval resources to reproduction and emerge with a larger number of mature eggs, the ovigeny index should not be affected by body size (Ellers & Jervis, 2003; Figure 2. in the introduction).

Here, we examine how fitness-related variables are size dependent and ultimately affect the ovigeny index in parasitoid wasps. Generally, life history traits increase with increasing body size. Thus I expect that larger females of *I. leucospoides* have higher egg loads upon emergence (and potential lifetime fecundity) and be alive for longer periods than smaller individuals. I assume that the shape of the relationship between the studied traits and body size, will allow speculations on the larval resources allocation strategy.

2. MATERIAL AND METHODS

Parasitoids were reared from pine (*Pinus contorta*) logs collected from several heavily attacked plantations located in Patagonia, Argentina. Once felled, trees were cut into 1 m-long logs and kept individually in locker-type cages under ambient indoor conditions until insect emergence occurred. Each morning, newly emerged insects were collected from the cages and immediately placed in individual vials in order to be randomly assigned to each treatment.

2.1 Estimation of the initial egg load, lifetime fecundity and ovigeny index

Emerging female parasitoids were stored in diluted ethanol (60% V/V) until the day of dissection when the number of initial matures eggs was tallied (initial egg load). As Ellers & Jervis (2003) noted, parasitoids have a limited number of oogonia, which constrains the total number of eggs they can produce, and so maximum potential fecundity can be estimated as the mean total egg production by females under the most favourable conditions. Under this assumption, I counted all eggs of females 24 day old, fed *ad libitum* and maintained in the absence of hosts. Next, these data were utilized to obtain a mean total egg production which was considered an estimate of lifetime fecundity. Finally, the ovigeny index was estimated as the mean initial egg load divided by the mean total egg production. Hind tibia length and weight at emergence were measured for each female as estimates of adult body size. These two variables are positively correlated (r^2=0.89, df = 110, p-value < 0.0001) and from now on they will be used interchangeably as indicators of body size.

2.2 Quantification of initial energy reserves

Quantification of the levels of carbohydrates of newly emerging female wasps was carried out as a proxy of initial energy reserves, using a modification of a colorimetric technique developed for mosquitoes (van Handel 1985a, b; van Handel & Day 1988). For each newly emerged female, the hind tibia was severed and the initial weight measured. Then, small metal balls and 200µl of 2% sodium sulphate were added into an Eppendorf tube containing the wasp. In a mixer, the sample was crushed for 1 minute at 30 beats/s and then the crushing balls removed. One ml of methanol was added to the sample and the tube was then vortexed. After centrifugation for 2 minutes at 12000 rpm, sugars remain dissolved in the supernatant. Seven hundred µl of the supernatant were transferred into another Eppendorf tube and the sample was evaporated to nearly 200µl at ambient temperature. This aliquot was collected and transferred into a volumetric flask in which anthrone reagent was added up to 25 ml were completed. Finally, samples were placed at 90ºC for 15 min, then cooled and read in a spectrophotometer at 625 nm to estimate the *total sugar content* for each female. Sugar concentrations were assessed from absorbance curve of known concentrations of glucose.

2.3 Longevity

Newly emerged females were placed in individual vials with access to water only, and maintained in a rearing chamber under semi-controlled temperature of 21 ± 0.1ºC and a LD 16:8h photoperiod until death.

2.4 Statistical analysis

The relationships between initial egg load and total egg production with body size were analyzed using linear models whereas the relationships between initial energy reserves and longevity with body size was analyzed using a generalized linear model assuming a Gamma distribution of residuals and an inverse link function. Finally, residuals were examined to confirm that the final models accurately fitted the data. All data analyses were done using the R statistical environment (R Development Core Team, 2008).

3. RESULTS

3.1 Body size and life history traits

Initial egg load is significantly and positively correlated with female body size, measured as hind tibia length ($r^2 = 0.470$; $F_{(1,\ 19)} = 5.415$; $P = 0.0311$; **Figure 1**). Likewise, the total egg production increased with tibia length ($r^2 = 0.731$; $F_{(1,9)} = 10.341$; $P = 0.0105$; **Figure 2**). However, the slope of the size–initial egg load relationship did not significantly differ from the slope of size-total egg production relationship (test for different slopes: $t_{(2,30)} = 0.36$; $P = 0.722$). The mean ovigeny index for *Ibalia leucospoides* was estimated as 0.77 and as expected for parasitoids with a high ovigeny index, it appears to remain constant with body size (**Figure 3).**

The initial energy reserves (total sugar content at emergence) significantly increased with body size, both when size is measured as tibia length ($F_{(1,14)} = 11.371$; $P = 0.0045$) and as initial weight ($F_{(1,14)} = 4.696$; $P = 0.0479$) (**Figure 4**). In addition, lifespan (age at death) of females without access to food was size-dependent. It increased slightly significantly with tibia length ($F_{(1,\ 16)}$= 4.185; P=0.0575) (**Figure 5**) but significantly with initial weight ($F_{(1,17)}$= 6.448; P=0.0211). On the contrary, the longevity of females kept with *ad libitum* access to diluted honey did not significantly change with size (data not shown).

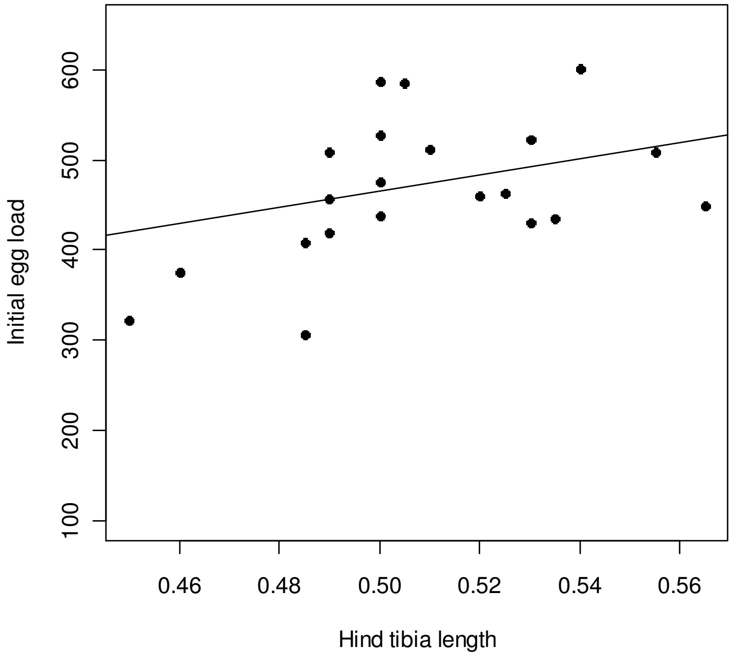


Figure 1. Relationship between initial egg load and body size (hind tibia length, cm) of *Ibalia leucospoides* females. The line in the plot is the regression line corresponding to: number of initial eggs = 1284.4 (size) -185.6.

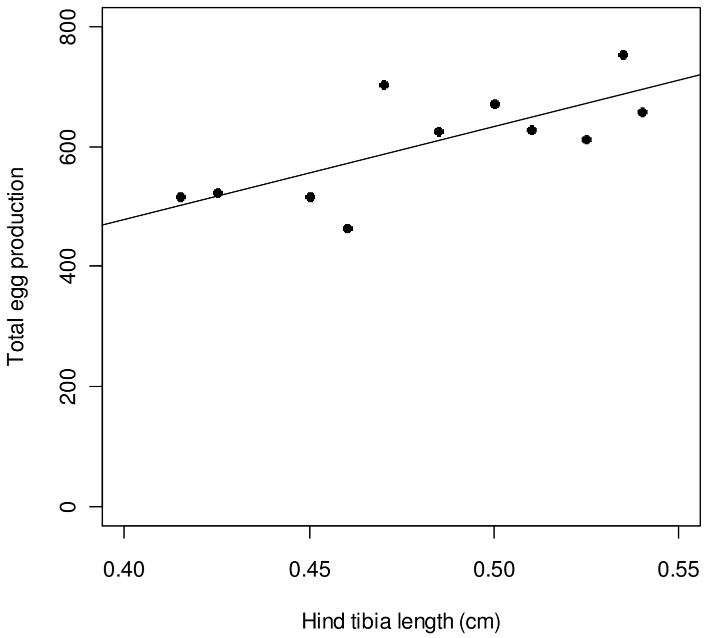


Figure 2. Total egg production in relation to body size (hind tibia length, cm) in *Ibalia leucospoides* females. The line in the plot is the regression line corresponding to: number of egg produced= 1547.7(size) – 139.9.

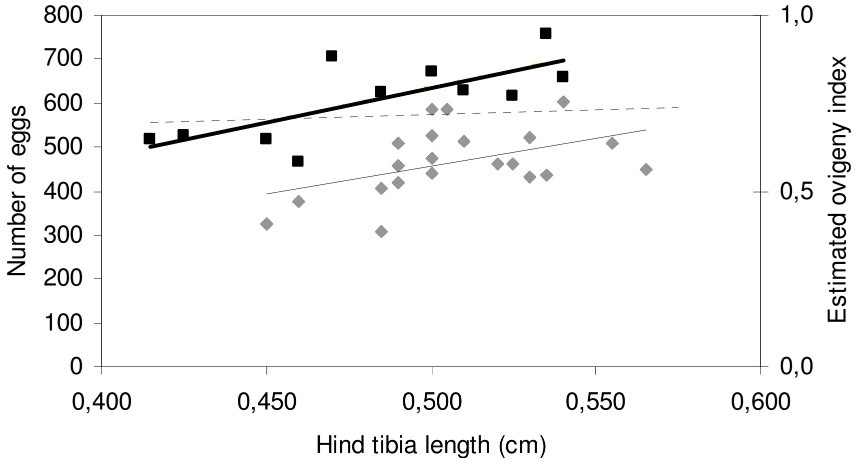


Figure 3. Regression for total egg production (black squares) and regression for initial egg load (grey circles) as a function of adult size (1284.4 x -185.6 and 1545.7 x -130.9, respectively). Estimated ovigeny index as a function of adult size, obtained from the ratio of initial egg load and total egg production predicted by the regressions (dotted line).

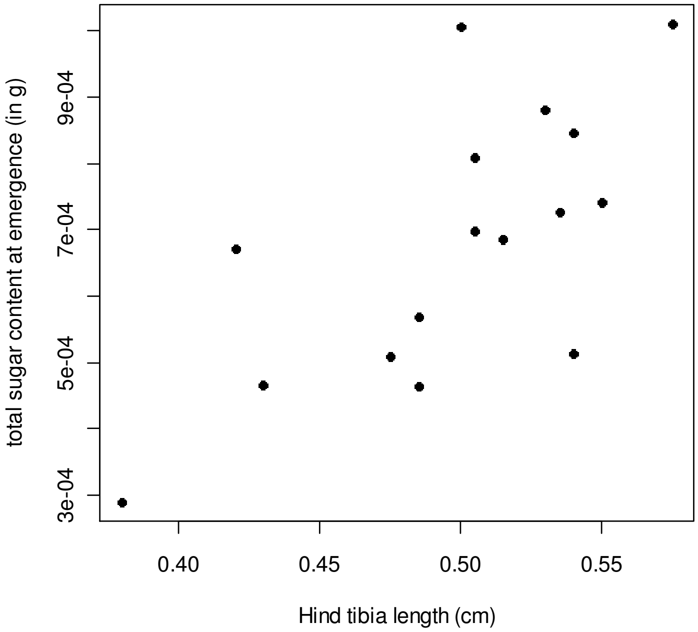


Figure 4. Total sugar content at emergence (in g) according to body size measured as hind tibia length (cm) in *Ibalia leucospoides* females.

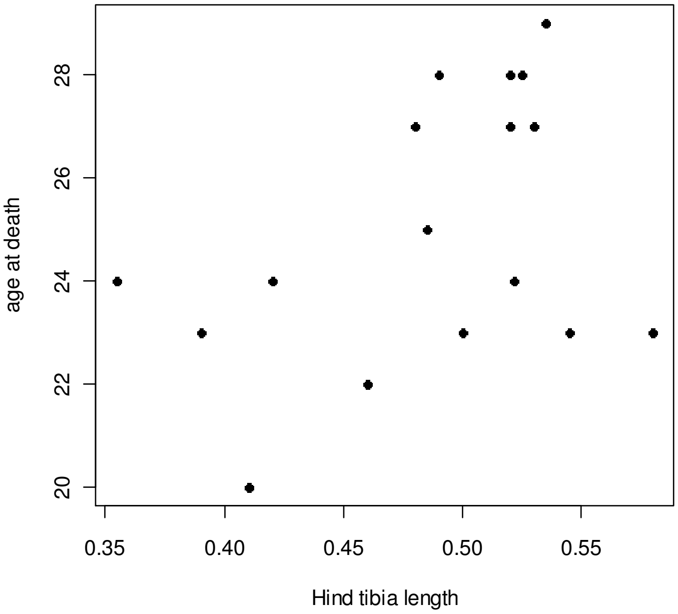


Figure 5. Lifespan (in days) in relation to body size measured as hind tibia length (in cm) in *Ibalia leucospoides* females.

4. DISCUSSION

The results of this study reveal that *I. leucospoides*, like other parasitoids, exhibits positive relationships between body size and life history traits (Visser, 1994; Ellers *et al.*, 1998a; Ellers *et al.*, 2001; Rivero & West, 2002). With increasing body size, initial egg load and total egg production increase in the same proportion. This suggests that there would be not a differential resources allocation to the initial egg load and the total egg production as has been proposed by Ellers & Jervis (2003) for synovigenic parasitoids. Precisely, in line to this, *I. leucospoides* females show a mean ovigeny index of 0.77 which enable us to place this species between weak synovigenic and nearly pro-ovigenic parasitoids.

The initial energy reserves, measured in this case as the total sugar content, co-vary with body size parameters. Nevertheless, when initial weight was used as indicator of size, this correlation was marginally significant. This may be due to the fact that weight at emergence may be explained largely by lipid reserves rather than by sugars. For example, in *N. vitripennis*, the largest females emerge with twice as much lipid as the smallest females (Rivero & West, 2002). Additionally, large unfed females live longer than smaller ones. At this point, initial weight rather than tibia length seems to reflect better that larger females possess greater total amount of resources than small individuals and in consequence live longer. Lipids and glycogen (in fat body) are the main sources of stored energy that fuel survival without food (Ellers, 1996; Rivero & Casas, 1999). Alternatively, the eggs themselves could be a nutrient source but while egg resorption is believed to provide nutrients for metabolic maintenance in parasitoids, studies quantifying its role in extending female longevity are still lacking (Strand & Casas, 2008).

In holometabolous insects, energy allocation towards competing needs occurs during both juvenile and adult stages. This is probably why the nutrient acquisition and allocation strategies adopted during the larval stages may have also impact on adult reproductive success. It is expected, that ontogeny patterns of resource allocation adjust to environmental conditions encountered by the adult. In the remainder of the discussion, based on theoretical works, I examine the environment factors (i.e. resource availability and habitat characteristics) that could influence the larval pattern of resource allocation in parasitoids in general, and particularly in *I. leucospoides*. I attempt to understand what factors may lead to the observed nearly proovigenic egg maturation strategy.

Ellers *et al.* (2000) have showed through a theoretical model, what environmental characteristics may determine the initial resource allocation strategy to initial egg load and survival (represented in the ovigeny index by the denominator). Namely, the variation in the optimal initial egg load can be almost completely explained by habitat quality (measured as the number of patches encountered multiplied by the mean number of hosts per patch) and by overall stochasticity in oviposition opportunities (host distribution among patches and host patch distribution). In a following work, Ellers & Jervis (2003) have attempted to identify size-related changes in resource allocation. They suggested that the allocation to initial egg load would not be strongly subject to parasitoid size but rather to habitat characteristics. Still, large individuals would have a greater total amount of resources to be allocated than smaller parasitoids.

According to the Ellers *et al.* (2000) model, as overall stochasticity in reproductive opportunities increases, the optimal initial egg load exceeds the expected number of hosts found. Consequently, the selection pressure of stochasticity could determine size related intra-specific variation in initial egg load and ultimately in ovigeny index. Following, Ellers & Jervis (2003) suggest that when hosts are aggregated across patches, the ovigeny index is always high, irrespective of habitat quality; but when hosts display a random distribution across patches (low stochasticity) then an increase in habitat richness causes an increase in the ovigeny index. Here, the higher allocation to initial eggs in host-rich habitats is due to the greater oviposition opportunities that such habitats offer. Hence, the ovigeny index, in theory, would depend on body size, mean habitat quality and stochasticity in the number of host per patch.

I. leucospoides attacks a host with outbreak dynamics. Thus host density can vary with time, across many orders of magnitude. In addition, as Corley *et al.* (2007) have shown, the overall spatial pattern of attacked trees by *S. noctilio* within plantations is strongly aggregated and over the time this aggregation becomes more intense together with an increase in the number of attacked trees. These observations could be translated into a high stochasticity in oviposition opportunities and surely, this host abundance and distribution variability could be key selective forces in the evolution of egg load in this parasitoid, by leading to higher egg production toward the start of adult life. A high ovigeny index despite of being disadvantageous compared with low ovigeny index, because it implies low reproductive plasticity to deal with variation in host availability; may prove an optimal strategy in the stochastic habitat where *I. leucospoides* inhabits.

Two strategies could be proposed to deal with habitat heterogeneity (Ellers, 1998b). On the one hand, animals may develop a plastic strategy (i.e. an adaptive phenotypic plasticity to face with habitat variability, which leads to a higher fitness) that can be adapted to any circumstance encountered. On the other hand, a fixed strategy that maximizes fitness given the probability of finding different scenarios may be also selected. The development of plastic strategy requires the presence of reliable information that indicates the future quality of the habitat. In the case that this information is lacking, no accurate predictions can be made about the future habitat quality and so the plastic strategy may prove inappropriate. Therefore, if the animal inhabits a highly unpredictable habitat, a fixed life history strategy could be a good option. Yet, the selection on a fixed trait could lead to adopt two strategies with different modalities. One form could be a risk-prone strategy, that is, with the trait value adapted to above the average quality of the habitat. The other form, on the contrary, could be a more risk-averse strategy with a trait value adapted to below the average habitat quality. Which of these strategies is more likely to evolve depend on habitat characteristics and the shape of the fitness function (Kacelnik & Bateson, 1993; Ellers, 1998b). A resource allocation strategy which likely leads to time limitation can be interpreted as a risk prone-strategy, with high benefits when host availability is high but associated with a high probability of not finding available hosts. On the contrary, a resource allocation strategy that leads to egg limitation can be understood as a risk-averse strategy with a probability of finding hosts, but decreasing the number of oviposition opportunities because eggs may not be ready to be laid fast enough. *I. leucospoides* dealing with an unpredictable pulse- like outbreak host population dynamics should be able to use all opportunities when hosts are available. Thus, female individuals presumably developed a tending time limitation strategy due to fitness gains from deviations above the average host availability over producing eggs are greater than the fitness loss from equal deviations below average.

Here, aspects to take into account are the frequency and duration of host outbreaks. Consider four potential scenarios: (1) frequent host outbreaks of a short duration; (2) frequent host outbreaks of a long duration; (3) infrequent host outbreaks of short duration and; (4) infrequent host outbreaks of a long duration. This means that, under these settings there may well be different degrees of stochasticity (host distribution), as well as different habitat quality (host density). One would expect so, based on model predictions of Ellers *et al.* (2000, 2003), that the optimal initial egg load should increase with increasing habitat quality as well as with increasing stochasticity, in addition parasitoids would always produce more eggs than they expect to be able to lay. Therefore, a higher ovigeny index (as a prone-risk strategy) would be selected as the best allocation strategy for the first two proposed scenarios while, on the contrary, a low ovigeny index may fit well with the last two scenarios, by resulting in a more accurate match between egg load and oviposition opportunities. Nevertheless, field data on frequency and duration of host outbreaks are required to accurately test these predictions.

To sum up, the differential allocation of teneral resources has important consequences for parasitoid foraging behavior, by determining egg load, the amount of reserves that are available for fueling egg manufacture, maintenance and locomotion and the role that income resources will play in nutrient dynamics.

5. REFERENCES

Bernstein, C. and Jervis M.A. (2008) Food-searching in parasitoids: the dilemma of choosing between ‘immediate’ or future fitness gains. *Behavioral Ecology of Insect Parasitoids: from theoretical approaches to field applications* (ed. by E. Wajnberg, C. Bernstein & J.J.M. Van Alphen), pp. 129-171. Blackwell Publishing, Oxford, U.K.

Ellers, J. and van Alphen, J. J. M. (1997). Life history evolution in *Asobara tabida*: plasticity in allocation of fat reserves to survival and reproduction. *J. Evol. Biol.* 10: 771-785.

Ellers, J. (1996) Fat and eggs: an alternative method to measure the trade-off between survival and reproduction in insect parasitoids. *Netherlands Journal of Zoology* 46: 227–35.

Ellers, J., van Alphen, J. J. M. and Sevenster, J. G. (1998a). A field study of size-fitness relationships in the parasitoid *Asobara tabida. J. Anim. Ecol.* 67: 318–324.

Ellers, J. (1998b) Life-history evolution in the parasitoid *Asobara tabida*: on the trade-off between reproduction and survival. Ph.D.-thesis, Leiden University, The Netherlands.

Ellers, J., Sevenster, J. G. and Driessen, G. (2000). Egg load evolution in parasitoids. *Am. Nat.* 156: 650-665.

Ellers, J., Bax, M. and van Alphen, J. J. M. (2001). Seasonal changes in female size and its relation to reproduction in the parasitoid *Asobara tabida*. *Oikos* 92: 309–314.

Ellers, J. and Jervis, M. A. (2003). Body size and the timing of egg production in parasitoid wasps. *Oikos* 102: 164-172.

Godfray, H.C.J. (1994) *Parasitoids. Behaviour and Evolutionary Ecology*. Princeton University Press, Princeton.

Honek, A. (1993). Intraspecific variation in body size and fecundity in insects: a general relationship. *Oikos* 66: 483–492.

Hougardy, E., Bezemer, T.M. and Mills, N.J. (2005) Effects of host deprivation and egg expenditure on the reproductive capacity of *Mastrus ridibundus*, an introduced parasitoid for the biological control of codling moth in California. *Biol. Cont.* 33: 96–106.

Jervis, M.A., Heimpel, G.E., Ferns, P.N., Harvey, J.A. and Kidd, N.A.C. (2001) Life-history strategies in parasitoid wasps: a comparative analysis of ‘ovigeny’. *J. Anim. Ecol.* 70: 442-458.

Jervis, M.A., Ferns, P.N. and Heimpel, G.E. (2003). Body size and the timing of egg production in parasitoid wasps: a comparative analysis. *Funct. Ecol.* 17:375–83

Jervis, M.A. and Ferns, P.N. (2004) The timing of egg maturation in insects: ovigeny index and initial egg load as measures of fitness and of resource allocation. *Oikos* 107: 449-460.

Kacelnik, A. and Bateson, M. (1996) Risky theories – the effects of variance on foraging decisions. *Am. Zool.* 36: 402–434.

Olson, D. M. and Andow, D. A. (1998). Larval crowding and adult nutrition effects on longevity and fecundity of female *Trichogramma nubilale* Ertle & Davis (Hymenoptera: Trichogrammatidae). *Environ. Entomol.* 27: 508–514.

R Development Core Team (2008) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL http://www.R-project.org.

Rivero, A. and Casas, J. (1999). Incorporating physiology into parasitoid behavioral ecology: the allocation of nutritional resources. *Res. Popul. Ecol.*, 41: 39–45.

Rivero, A. and West, S.A. (2002) The physiological costs of being small in a parasitic wasp. *Evol. Ecol. Res*. 4: 407–420.

Roff, D.A. and Fairbairn, D.J. (2007). The evolution of trade-offs: where are we? *J. Evol. Biol.* 20: 433-447.

Stearns, S.C. (1989). Trade-offs in life-history evolution. *Funct. Ecol.* 3: 259-268.

Strand, M.R. and Casas, J. (2008) Parasitoid and host nutritional physiology in behavioral ecology. *Behavioral Ecology of Insect Parasitoids: from theoretical approaches to field applications* (ed. by E. Wajnberg, C. Bernstein & J.J.M. Van Alphen), pp. 113-128. Blackwell Publishing, Oxford, U.K.

van Handel E (1985) Rapid determination of glycogen and sugars in mosquitoes. *J. Amer. Mosquito Control Assoc*. 1:299–301

van Handel E and Day JF (1988) Assay of lipids glycogen and sugars in individual mosquitoes: correlations with length in field-collected *Aedes vexans*. *J. Amer. Mosquito Control Assoc.* 4:549–550

Visser, M. E. (1994). The importance of being large: the relationship between size and fitness in females of the parasitoid Aphaereta minuta (Hymenoptera: Braconidae). *J. Anim. Ecol.* 63: 963-978.

Chapter II

This manuscript is in preparation

The influence of adult nutrition on resource allocation to fitness-related physiological functions

AUTHORS

Deborah Fischbein (1,2), Carlos Bernstein (2), Juan C. Corley (1).

(1) Grupo de Ecología de Insectos, INTA EEA Bariloche CC 277, San Carlos de Bariloche 8400, Río Negro, Argentina.

(2) Laboratoire de Biométrie et Biologie Evolutive UMR 5558. Université de Lyon Université Claude Bernard Lyon 1, 43 Bd du 11 Novembre 1918, 69622 Villeurbanne cedex.

1. INTRODUCTION

Egg production and life span are crucial life-history parameters in parasitoids because they directly limit the lifetime reproductive success (Rosenheim, 1996; Heimpel *et al.*, 1998; Sevenster *et al.*, 1998; Rosenheim *et al.*, 2000). Parasitoids can be *egg-limited* (when females have insufficient eggs to lay in all available hosts) or *time- limited* (when females risk of dying without discharging all available mature eggs). The incidence of these two aspects of animal life histories strongly relies on the evolutionary optimization of resource allocation. Hence, parasitoid reproductive success is mainly dependent on: (1) the available physiological resources for egg maturation; (2) the available time for host searching and oviposition. Natural selection is expected to lead to reproductive strategies that approach a quantitative match between egg load and hosts availability (Rosenheim, 1996; Sevenster *et al.*, 1998) and thus reduce the risk of becoming time or egg limited. Actually, most species are at an intermediate position along the egg-/time-limitation continuum, with a bias towards time-limitation (Jervis *et al.*, 2004).

For parasitoids, adult metabolic resources originate from nutrient reserves accumulated during larval development (the 'teneral resources' or capital reserves) and by adult feeding. The combination of incoming adult nutrients and stored larval nutrients is then used in specific proportions to support adult functions such as reproduction, survival and dispersal (Boggs & Freeman, 2005). Given that both exogenous (food intake) and endogenous (fat body) resources could be limiting, *trade-offs* between competing fitness functions may arise.

Floral nectar is the most important plant-derived food source for adult parasitoids. Nectar is rich in carbohydrates but generally poor in proteins and lipids (Petanidiou, 2007). While it is often the main food source for *non-host feeding* parasitoids, *host feeders* can consume nectar or honeydew in addition to feeding on hosts hemolymph and/or other tissues (Rivero & Casas, 1999; Heimpel *et al.*, 1997; Casas *et al.*, 2005). Several species of parasitoids greatly enhance their longevity, fecundity and flight through feeding (Heimpel, 1997; Rivero & Casas, 1999; Olson *et al.,* 2000; Siekmann *et al*., 2001; Giron *et al.,* 2002; Mitsunaga *et al.*, 2004; Wanner *et al*., 2006; Bernstein & Jervis, 2008). Fecundity can increase by feeding through diverse mechanisms. For example, egg load may change as a direct result of feeding because of a reduction of the egg resorption rate (Heimpel *et al.*, 1997) or an increase of egg maturation (Tylianakis *et al.*, 2004; Bezemer *et al.*, 2005). At the same time, access to adult food sources may also increase the number of progeny by increasing host attack rates (Schmale *et al.*, 2001; Bezemer *et al.*, 2005). Likewise, feeding can indirectly affect realized fecundity through an increase in longevity, allowing for more time to mature and lay eggs.

For *non-host feeders*, hosts and food patches in natural conditions are generally spatially separated; consequently the acts of oviposition and feeding do not occur simultaneously. Under these circumstances, foraging females must make important foraging decisions regarding whether to allocate their time and energy to food-searching or host-searching. Taking into account the *trade-off* between current and future reproduction, feeding constitutes a missed opportunity in terms of 'immediate' fitness gain. However, by choosing to feed, females gain in future oviposition opportunities, given the potentially positive effects that nutrients have on life expectancy and may have on egg maturation rates (in the case of synovigenic wasps) (Heimpel & Collier 1996; Sirot & Bernstein 1996; Desouhant *et al.*, 2005; Tenhumberg *et al.*, 2006, Bernstein & Jervis, 2008). Likewise, the availability of food sources is usually highly variable in space and time in the field or even, not available where hosts are found. In this way, ecological conditions such as variations in food foraging opportunities can occur and may influence both female nutritional conditions and their foraging behaviour.

Since sugar-rich foods consumption is beneficial for parasitoid reproduction, through an increase in longevity and egg load (Schmale *et al.*, 2001; Siekmann *et al.*, 2001) as well as by changes in insect behaviors (Wanner *et al*., 2006; Takasu and Lewis 1995; Staple *et al.*, 1997) considerable attention has been paid to the influence of food on adult fitness in the context of biological control (Jervis *et al.*, 1996; Wäckers, 2004; Heimpel & Jervis, 2005; Lavandero *et al.*, 2006). In cultivated pine forests in Patagonia, where *Ibalia leucospoides* is established, food sources are rare, may not occur concurrently with the host or else are unavailable where hosts are found, namely, within the pine plantations and their immediate surroundings. In addition, there is no available information on whether food sources are present in the natural environments where *I. leucospoides* evolved. Should feeding behaviour be a significant aspect in this species, food scarcity may be a handicap for parasitoid performance in the cultivated/ artificial environment in which the species is used for the control for damaging woodwasp populations. Therefore, given the empirical evidence on fitness gains of sugar feeding, we predict that *I. leucospoides,* a *non-host feeding* parasitoid, can be beneficed extending adult life and enhancing egg maturation through feeding.

In this chapter, I first test whether adult food provisioning affects longevity in *I. leucospoides*. As in Patagonia cultivated pine forest, temporal starvation could be a likely scenario, I also examine: (a) whether variations in the timing (duration) during which female wasps are exposed to food (e.g. one day or *ad libitum*) affects longevity and (b) whether the time when wasps are provisioned with food affects adult lifespan (at emergence, middle age or later in their life). Secondly I test the influence of adult feeding on egg production. To address this, female wasps are allowed to feed *ad libitum* on a honey diluted solution and their egg load compared with unfed wasps at different ages. Finally, despite the important fitness consequences of food acquisition, knowledge of parasitoids food foraging remains limited especially in the wild (Lewis *et al.* 1998; Sisteron & Averill; 2002; Lee & Heimpel, 2008). Therefore, a third aim is to gain an insight into food use under field conditions. For this purpose, levels of body sugars of female wasps collected in the field were quantified using colorimetric techniques (Van Handel 1985; Van Handel & Day, 1988). Laboratory individuals of known nutritional state were used as reference values to compare with field captured individuals. The results of this investigation are discussed with respect to adult resources allocations strategies to lifespan and reproduction.

2. MATERIAL AND METHODS

Ibalia leucospoides parasitoids were reared from pine (Pinus contorta) logs collected from several, heavily attacked plantations located in NW Patagonia, Argentina. Once felled, trees were cut into 1 m-long logs and kept individually in locker-type cages under ambient indoor conditions until insect emergence occurred. Each morning newly emerged insects were collected from the cages and immediately placed in individual vials (251 cm^3) in order to be randomly assigned to the different experiences.

2.1 Longevity estimations

To study the effects of food provisioning on longevity in females of *Ibalia leucospoides*, a laboratory experiment was conducted to answer the following questions: Does unlimited access to food increase the longevity of females? Can an increase in longevity be achieved by a single day access to food? Is the temporal occurrence of this one day of access to food critical for life expectancy?

Newly emerged female parasitoids were randomly assigned to each of the following five treatments: (1) access to water only (n=22; *unfed wasps*); (2) *ad libitum* access to 30% diluted honey solution (n= 22; *fed ad libitum*); (3) access to honey solution for one day at emergence (n=23; *fEd*); (4) access to honey solution for one day at day 7 (n=24; *f7d*) and; (5) access to honey solution for one day at day 14 (n=22; *f14d*). In treatment two, food was replenished every day. All tested wasps were kept individually in plastic vials (251 cm^3) under semi-controlled temperature, humidity conditions (temperature 21 ± 0.1ºC; 54% r.h.) and LD 16:8h photoperiod until death.

2.2 Egg load

To examine the effects of sugar feeding on the egg complement at different ages, newly emerged female parasitoids were randomly assigned to one of two feeding treatments: (1) Fed *ad libitum* with 30% diluted honey solution and, (2) access only to water. In both treatments, wasps were deprived from hosts and males, and females were dissected at each of the following ages: after emergence to determine the number of mature eggs present at birth (initial egg load), and at 3, 14 and 24 days since emergence. The experiment was terminated 24 days post emergence because this corresponds to the mean life span of food-deprived wasps, obtained in the longevity experiments (see below, results section). Since in the current experiment unfed wasps did not survive as long as in longevity experiments, egg load at death of unfed wasps was estimated at day 19, which was the mean life span in this assay. Field captured wasps were also included in this assay and their egg load recorded. In treatment one, food was replenished every day. All wasps were kept individually in plastic vials (251 cm^3) under semi-controlled temperature, humidity conditions (temperature 21 ± 0.1ºC; 54% r.h.) and LD 16:8h photoperiod to the age of interest. Then, females were kept in alcohol 60% until dissection. Only mature eggs were counted, because the degree of egg maturation may vary with age and food exposition whereas the number of immature eggs is considered to be fixed at emergence.

2.3 Feeding in the wild

To gain an insight into *I. leucospoides* food use under field conditions, quantification of carbohydrates levels of wasps collected in the field was carried out using a colorimetric technique (Van Handel 1985; Van Handel & Day 1988). This method has been used efficiently to compare variations in sugar content between different feeding treatments in other studies (Casas *et al.*, 2003; Pelosse *et al.*, 2007; Desouhant *et al.*, 2009). Therefore, since extrafloral and floral nectars sugars are predominantly glucose, fructose and sucrose as in honey meal; I measured levels of total sugar content (hot anthrone test) in field and laboratory reference females. Body sugars allows visualize changes in teneral reserves in starved wasps on the one side, and on the other, is a reliable indicator of feeding occurrence because increase with nectar or honey food ingestion. By comparing with reference values of sugar content obtained from laboratory-reared individuals whose nutritional state is known, I determined whether captured wasps actually fed on sugary sources in the field.

2.3.1 *Wasps captured in the field*

Female wasps were captured in pine tree plantations located approximately 10 km from the city of El Bolson (42º01' S - 71º33' O) from late January to late March 2010 (within the wasp flight season). Females for egg load counting (n=9) were captured with traps baited with α- and β-pinene placed in inside and in the vicinity of pine stands and revised every 15 days. Individuals for biochemical analyses (n=9) were captured, inside pine plantations while flying using an insect net. Captured wasps were immediately placed in cold (camping coolers) and carried within a few hours to the laboratory where they were then frozen at -20ºC.

2.4 Reference values

Newly laboratory emerged females were randomly divided to each of the following feeding treatments: emergence; one, seven and fourteen days, unfed (*unfed1, unfed7, unfed14*); one, seven and fourteen days, fed *ad libitum* since emergence (*fed1, fed7, fed14*); fourteen days fed and starved (seven days fed *ad libitum* and seven days unfed; *fed&starved*); twenty four days fed *ad libitum* (*fed24*) and; at death. These treatments allowed us to create reference values with which to compare the sugar levels obtained from females collected in the field. The sugar-rich meal consisted of a 30% honey/water diluted solution. Food deprived wasps allowed us to assess changes in teneral reserves through the time. Wasps were conserved at -20ºC up to the day of biochemical analysis**.** All measurements of sugar content were expressed relative to wasp weight at time of emergence.

2.5 Biochemical analysis

For the sugar analysis (hot anthrone test), small metal balls and 200µl of 2% sodium sulphate were added into an Eppendorf tube containing a single wasp (laboratory or field individuals). In a mixer, the sample was crushed for 1 minute at 30 beats/s and then all added balls removed. One ml of methanol was added to the sample and the tube was vortexed. After centrifugation for 2 minutes at 12000 rpm sugars remain dissolved in the supernatant. Seven hundred µl of the supernatant was transferred into another Eppendorf tube and the sample was evaporated to nearly 200µl at ambient temperature. This aliquot was collected and transferred into a volumetric flask in which anthrone reagent was added up to 25 ml were completed. Finally, a fraction of the sample was placed at 90ºC for 15 min, then cooled and read in a spectrophotometer at 625 nm to estimate the *total sugar content* for each female. Body sugars (total sugar content) are mainly constituted of glucose, however, other sugars such as, fructose, trehalose (storage source of energy) or melezitose (obtained by adult nutrition) are present. Yet, as non-glucose sugars concentrations are usually low in parasitoids (Desouhant *et al.*, 2009), we considered total sugar content as equivalent of glucose. A calibration curve with a standard solution of glucose (0.1 mg / ml in ethanol 25%) allowed us to transform absorbance into concentrations.

2.6 Statistical analysis

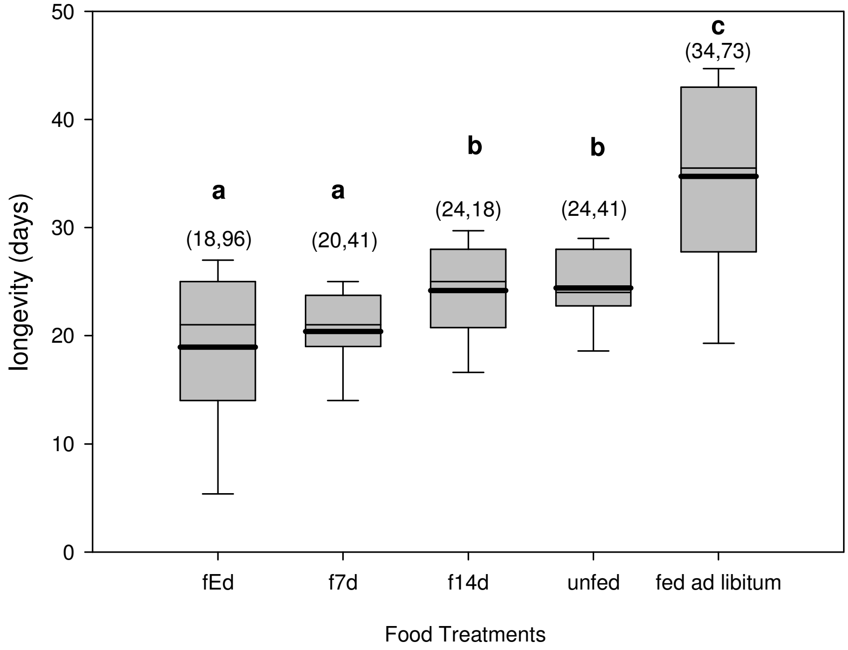
Longevity was analyzed using both survival analysis and a matrix of contrasts to perform *a posteriori* tests of coefficients (a way to specify specific contrasts between treatments). Egg load was analyzed using a linear model and a significance coefficients test. To study changes on total sugar content over life span for laboratory-reared individuals, linear models and generalized linear models assuming a Gamma distribution of residuals and an inverse link function were used. Feeding treatment and age were introduced in the models as explanatory variables. To compare the total sugar content of wild-caught female wasps with laboratory reference values, we used a generalized linear model assuming a Gamma distribution of residuals and an inverse link function. The data were organized such as, that as a consequence of the contrast matrix used, wild-caught animals constituted the focal group against which all other groups (the different treatments in the laboratory) were compared. The differences between the focal group and laboratory treatments are given by the regression coefficients and their significance levels. Finally, residuals were examined to confirm that the final models accurately fitted the data. All data analyses were done using the R statistical environment (R Development Core Team, 2008).

3. RESULTS

3.1 Longevity

Females with *ad libitum* access to sugar-rich food gained, in average, ten days of life compared with starved wasps. Hence, the increase in longevity was of 42% (**Figure 1; a, b**). Without access to food, females lived up to 30 days while in the presence of honey, females stayed alive for up to 46 days. There was no significant difference in longevity between females fed during a one day-long span, at emergence or at day 7 (z= -0.687; P=0.492). Likewise, females with one day honey solution access at day 14, had similar longevity than unfed wasps (z= -0.313; P=0.755). However, longevity significantly decreased (4.61 days, which correspond to a decrease of 19%) when female wasps were fed early in life (fEd and f7d) compared with unfed and fed wasps at day 14 (z= - 6.546; P< 0.0001).

(a)



(b)

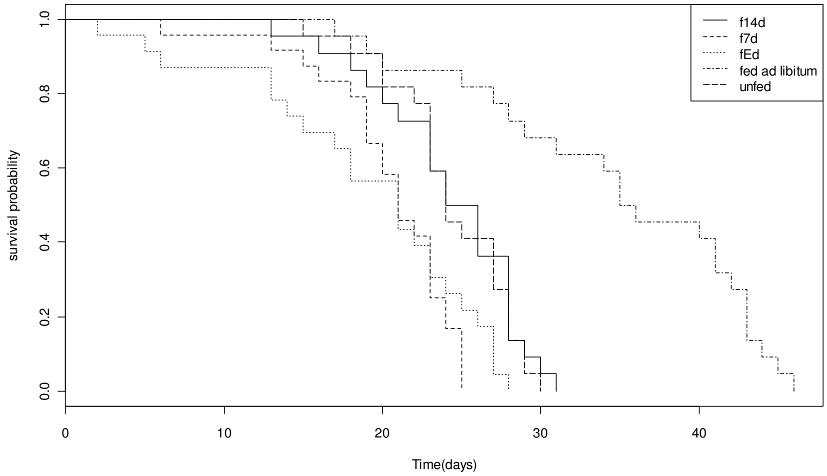


Figure 1. Effects of the frequency and temporal access to food on the longevity of *I. leucospoides* females. **(a)** The thin horizontal line in each box plot marks the median of the sample and the thick horizontal line marks the sample mean. The mean for each group of wasps is shown above each box. Significantly different treatments are shown with different letters (a, b, c). fEd: access to honey solution for one day at emergence; f7d: access to honey solution for one day at day 7; f14d: access to honey solution for one day at day 14; unfed: access to water only and fed *ad libitum*: continuous access to 30% diluted honey solution. **(b)** Kaplan-Meier Survival curves for *Ibalia leucospoides,* according to the above described five food treatments.

3.2 Egg load

The egg load of *I. leucospoides* ranged from 178 to 806 eggs per female (field-caught wasps not included), that is, a 4.5-fold difference between extremes. Sugar-rich food had no significant effect on egg load, at any of the studied ages (fed *vs* unfed wasps of 3 days old: t = 0.857, P = 0.394; fed *vs* unfed wasps of 14 days old: t = -0.234, P = 0.815; and 24 days fed *vs* death of unfed wasps: t = -0.715, P = 0.476). However, age had a significantly positive effect on the egg complement, (emergence *vs* 3 days old: t = -7.119, P<0.0001; 3 *vs* 14 days old: t = -5.280, P< 0.0001; **Figure 2**). Yet, there were no significant differences in the egg complement between wasps 14 days old and those 24 and 19 days old (t = 0.640, P= 0.5238; t= -0.089, P = 0.929, respectively). Finally, field captured females had a lower number of mature eggs than newly emerged females (t=-6.174, P<0.0001).

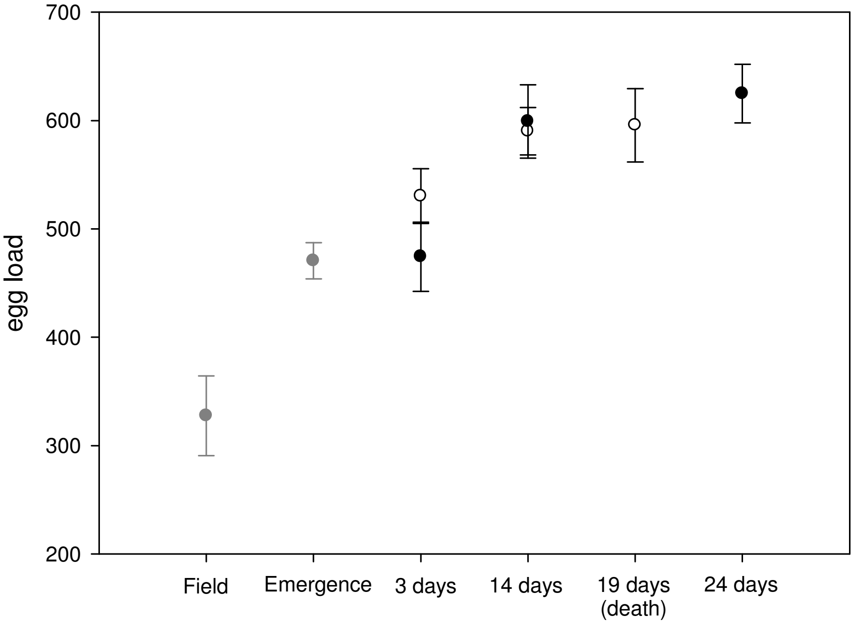


Figure 2. Egg load (mean ± SE) of *I. leucospoides* females as a function of age, in the absence or presence of food. Sample sizes are $n_{field} = 9$, $n_{emergence} = 22$, $n_{3\ days\ unfed} = 15$, $n_{3\ days\ fed} = 15$, $n_{14\ days\ unfed} = 15$, $n_{14\ days\ fed} = 15$, $n_{death} = 14$, $n_{24\ days\ fed} = 13$. *Grey circles*: field and newly emerged wasps; *white circles*: unfed wasps; *black circles*: fed wasps.

Realized ovipositions in the field until the moment of capture was estimated as the difference between the mean total egg production of reference females and the average egg load of females captured in the wild. Total egg production of reference females was obtained averaging the egg load of 14 to 24 day old females since egg load remained constant after 14 days in the absence of hosts. As a result, wild caught females should have discharged in average, 277.13 eggs.

3.3 Changes in reference total sugar content values for laboratory-reared individuals

The decline of total sugar content between birth and death was of 44.68 % ($F_{(1,34)}$ = 17.820; $P<0.001$; Table 1). Age had a significant effect on total sugar content for starved females ($F_{(3,58)}$ = 6.929 $P<0.001$). Fourteen days unfed females had a significantly lower total sugar content than recently emerged females and than one day and seven day old unfed females (t = 2.572, P = 0.0127; t = 3.544, $P<0.001$; t = 4.176, $P<0.001$; respectively). However, similar quantities of body sugars were observed in newly emerged, one day old and seven day old unfed females (emergence *vs* unfed1, t = 1.047, P = 0.299; emergence *vs* unfed7, t = 1.624, P = 0.109; unfed1 *vs* unfed7, t = -0.539, P = 0.591; see Table 1; **Figure 3**). The interaction between feeding treatment and age had a significant effect on total sugar content for females of one, seven and fourteen days of age ($F_{(2,85)}$ = 13.620; $P<0.001$). As expected**,** the access to food *ad libitum* produced an increase in total sugar content of 29% in one day old fed wasps compared to one day old unfed wasps. Likewise, the total sugar content of seven day old fed females was four times larger than the quantity recorded for seven day old unfed wasps. Finally, fed females fourteen days of age, had nine times more total sugar content than unfed ones of the same age.

3.4 Total sugar content of wild-caught female wasps

The total sugar content of females captured in the wild (0.0126 ± 0.0036 g body sugars/g wasps) did not significantly differ from the laboratory reference values of starved females, regardless of age (t = -0.548, $P_{emergence}$ = 0.5845; t = -0.982, P_{unfed1} = 0.3277; t = -1.202, P_{unfed7} = 0.2310; t = 0.824, $P_{unfed14}$ = 0.4114, **Figure 3**). Still, field wasps had significantly higher total sugar content than wasps at death (t = 2.472, P = 0.0145). Finally, field wasps showed a total sugar content significantly lower than that recorded for females fed under laboratory conditions, irrespective of age ($t = -2.374$, $P_{fed1} = 0.0188$; $t = -4.889$, $P_{fed7} < 0.001$; $t = -2.415$, $P_{fed\&starved} < 0.0169$; $t = -5.241$, $P_{fed14} < 0.0001$; $t = -5.175$, $P_{fed24} < 0.0001$).

Only one out of the nine field captured wasps showed a high sugar content (0.0393 g body sugars/g wasps). This was of 2 times that observed value for the individual with the highest sugar content at emergence (teneral level), indicating that this wasp had probably fed on sugars in the field. Therefore, the preceding results would be likely be affected by this wasp. By removing this individual from the analysis the results were different. On the one hand, in contrast to the previous results, the total sugar content of females captured in the wild *did* significantly differ from the laboratory reference values of young unfed females of one day old and seven day old ($t = -2.247$, $P_{unfed1} = 0.0260$; $t=-2.432$, $P_{unfed7} = 0.0161$) and, marginally differed from the recently emerged wasps ($t = -1.896$, $P_{emergence} = 0.0597$). Field caught females average sugars were 1.5, 1.69 and 1.77 lower than reference values at emergence, one day old and seven day old starved females, respectively (**Table 1**). On the other hand, the body sugars of wild females did not significantly differ from the laboratory reference values of fourteen day old unfed wasps and wasps at death ($t = -0.700$, $P_{unfed14} = 0.4850$; $t= 0.838$, $P_{death}= 0.4032$). Field wasps still show a total sugar content significantly lower than the content of females fed under laboratory conditions.

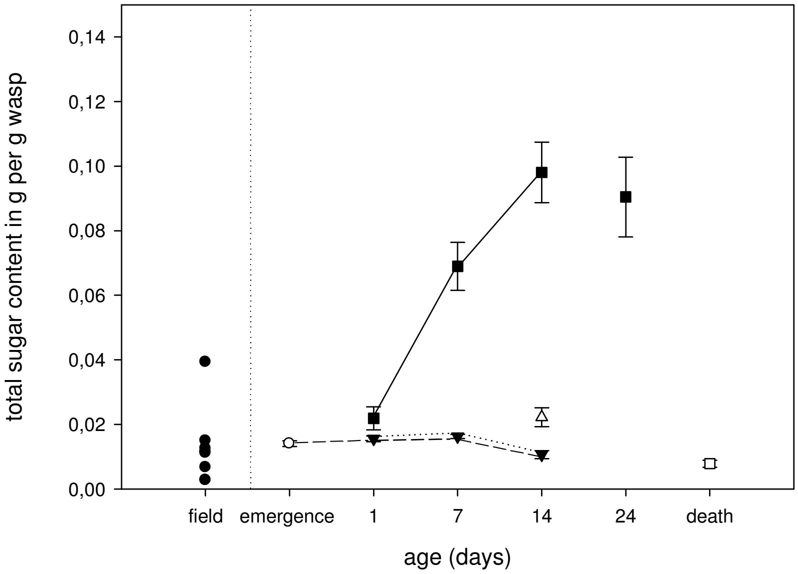


Figure 3. Total sugar content (mean ± SE) as a function of age. *Open circle*: emergence females*; filled triangles*: one, seven, fourteen day old unfed females; *Open triangle*: fourteen day old fed and starved females; *full squares*: one, seven, fourteen and twenty four day old wasps fed *ad libitum*; *open squares:* females at time of death. Field captured wasps are represented as individual data with *full circles*.

TABLE 1. Mean ± SE of laboratory reference values of total sugar content for *I. leucospoides* females.

Treatment	Mean ± SE (g body sugars/g wasps)	n
Field	0.0126 ± 0.0036	9
Field *without fed individual*	0.0092 ± 0.0016	8
emergence	0.0141 ± 0.0008	16
1 day unfed	0.0156 ± 0.0009	14
1 day fed	0.0219 ± 0.0035	16
7 days unfed	0.0163 ± 0.0006	15
7 days fed	0.0689 ± 0.0074	14
14 days unfed	0.0106 ± 0.0013	17
14 days fed	0.0981 ± 0.0093	15
14 days, fed and starved	0.0222 ± 0.0030	15
24 days fed	0.0904 ± 0.0123	17
at death	0.0078 ± 0.0011	20

4. DISCUSSION

In this chapter, I studied under laboratory conditions, the benefits of feeding on sugar for *I. leucospoides* females and I draw a first inference about the use of food in the wild by this parasitoid was made. While sugar-rich food provisioning *ad libitum* increases lifespan, this has no effect on egg load at any given age. Females emerge with a considerable amount of mature eggs and continue to mature several more over the course of adult life. The access to honey based food over a one day long interval, has no strong influence on longevity regardless at which moment in the lifespan this occurs. Currently, there is scant information on feeding behaviour of free foraging wasps of this species. However, the results presented here suggest that food intake in the wild although possible might not be a common event.

It is widely known that sugar consumption improves parasitoid longevity in laboratory conditions (Leius, 1961; Olson *et al.*, 2000; Lee *et al.*, 2004; Fadamiro *et al.*, 2005). For example, a single sugar meal can reduce the risk of death by starvation by up to 73% in the parasitoid *Cotesia rubecula* (Siekmann *et al.*, 2001) while *Aphidius ervi* females need at least two food intakes per day to attain maximum longevity (Azzouz *et al.*, 2004). In turn, *Meteorus pulchricornis* fed continuously with a 30% (w/v) 1:1:1 sucrose–glucose–fructose-mixture lives six times longer than unfed parasitoids (Heping *et al.*, 2008), and in the same way sugar feeding can increase longevity 15-fold in *Cotesia glomerata* (Wäckers, 2001; Lee & Heimpel, 2008) and 10-fold in *Mastrus ridibundus* (Bezemer *et al.*, 2005).

In this study, honey intake has a positive effect on longevity in *I. leucospoides* but only when it is provided *ad libitum*. In this species, females with full access to honey based foods, can increase by ten days their lifespan. In addition, while one day access to food much later in life (*f14d*) increases longevity by 4.61 days compared to individuals fed much earlier in adult life (*fEd* and *f7d*), this does not result in a significant difference in longevity with unfed wasps. Perhaps, *I. leucospoides* has physiological or morphological constraints and thus a single day of access to food is not sufficient to acquire enough nutrients to allow for differences with the nutritional state of the starved individuals. At the same time, why females fed at emergence and at day 7 significantly decrease, thought a few days, their lifespan with respect to starved wasps, could be explained by food triggering the development of some organ of the insect early in life. Once food is removed, the insect would have to draw resources from reserves to continue to fuel the organ development resulting in a reduction of longevity. Given that in field conditions food is scant and feeding seems to be a rare event, it is possible that the lack of influence of limited access to food (during a singe day) results from other adaptations to reduced food availability. Females thus may emerge with robust fat body reserves, enough to support maintenance function requirements without feeding (this will be discussed later on this section).

Furthermore, whether the augmentation of lifespan for females fed *ad libitum,* in perspective to improve rearing conditions of parasitoid destined to field release, translates in an increase of lifetime reproductive success would depend on some additional factors. Firstly, the extent in which this species is *time-limited*. That is, if *I. leucospoides* is able to deposit almost all mature eggs within a period of time shorter than their lifespan in the wild, 15 days approximately, then an increase in longevity does not imply an additional gain in fitness. Secondly, the benefits of feeding may be insignificant if a strong predation pressure exists, killing parasitoids before starvation becomes a limiting factor (Rosenheim, 1998; Lee & Heimpel, 2008). Likewise, other extrinsic mortality factors such as extreme temperatures or storms could have a strong impact on insect longevity (Dyer & Landis, 1996; Fadamiro *et al.*, 2005).

For parasitoids species whose adults do not host-feed, resources necessary for egg production are mainly obtained during larval development and then are carried over to the adult stage (fat body storage, Rivero & West, 2002). Adult parasitoids must then metabolize fat body reserves for egg production and/or somatic maintenance. In the presence of a carbohydrates source, the catabolic drain that somatic maintenance exerts upon these carried-over resources that fuel ovigenesis, could be attenuated. That is, feeding enables parasitoids to maintain high levels of glycogen and sugar reserves, on the one hand, and on the other, to keep fat body available, which may promote egg maturation. There is much empirical evidence on the positive role of sugars on egg maturation (Olson & Andow, 1998; Tylianakis *et al.*, 2004; Bezemer *et al.*, 2005) but there are also cases in which feeding does not influence this process (Sisterson &

Averill, 2002). In the case of *I. leucospoides*, feeding has no effect on egg load. Egg load analysis showed that the egg number increases with age; however it remains constant after 14 days. Furthermore, female wasps emerge with more than 75% of its total egg complement mature. Consequently, the lack of effect of feeding on egg maturation may be related to the fact that life-history traits and nutritional strategies of *I. leucospoides* would be closer to a pro-ovigenic parasitoid than synovigenic ones. Pro-ovigenic species emerge more or less with a fixed complement of eggs and do not need any income resource to be assigned to egg production or maturation, hence only somatic maintenance demands to be fuelled after emergence (Rivero *et al.*, 1999; Rivero *et al.*, 2001; Pelosse, 2008). In line with this, females of pro-ovigenic species should, in theory, have lower need for supplementary food (Jervis & Heimpel, 2007). In fact, it is possible to suggest that in this species, female carried-over resources would be sufficient to meet all their maintenance and reproduction needs.

When comparing egg load of females at death and 24 day-old fed individuals, eggs number is similar. In all cases female wasps were host deprived and it could be that egg resorption took place. However, this seems not to be the case, in agreement with the assumption that egg resorption is concentrated mostly among synovigenic species (Jervis *et al.,* 2001). This assumption arises from the facts that egg resorption implies a delay and that synovigenic parasitoids commonly have yolk-rich eggs (anhydropic) while pro-ovigenic bear yolk poor eggs (hydropic). *I. leucospoides* females would not reabsorb mature eggs during periods of host and food scarcity due to its condition of nearly pro-ovigenic parasitoid (with many likely hydropic eggs). Moreover, while it is commonly held that egg resorption is a strategy for recovering nutrients under conditions of resource limitation by reallocating them to maintenance, few studies with parasitoids have reported a positive correlation between the number of eggs reabsorbed and extended longevity.

Therefore, with respect to the limited impact of adult nutrition on egg production or survival, one would expect that parasitoids evolved a strategy to optimize the uptake and allocation of limited host resources to these competing fitness-related functions during larval development, independently of adult food acquisition.

Only one of the nine field captured wasps showed a high level of body sugars. Given that this field female had well above twice sugar level than the teneral levels, presumably this result could be the consequence of food intake. In field conditions, *I. leucospoides* females emerge within host patch aggregates inside pine forests, where the probability of finding food is low or else unavailable (personal observation). Despite this, wasps are able to forage for hosts within the plantation. Feeding surely incurs at a cost for the female parasitoid, both in terms of time and energy investment (especially when hosts and food are found in different parts of the environment). In the Sirot & Bernstein (1996) food foraging model for pro-ovigenic parasitoid it is pointed out that when food availability is low, parasitoids should avoid searching for food. *I. leucospoides* as a nearly pro-ovigenic parasitoid seems to be closer to this strategy. Furthermore, taken into account the Bernstein & Jervis (2008) model for pro-ovigenic parasitoids, *I. leucospoides* probably should not search for food. On the one hand, the model predicts that for short travel distances and a high egg load when food is scarce, parasitoids should avoid searching for food and should remain in the host patch. On the other hand, when egg load is low, animals should not feed to satiation due to the fact that continuing feeding entails a mortality risk, and moderate resources would suffice to lay the remaining eggs. In addition, when the travel time increases, females become more reluctant to leave their current patch so for even low egg loads, females avoid searching for food still in cases of high food availability.

Most likely, in *I. leucospoides,* larval and adult resource allocation strategies are strongly influenced by environment characteristics such as host abundance and distribution, and reproductive success would not be constrained by adult food deprivation. Note that *I. leucospoides*, emerges with a high proportion of eggs mature ready to be laid which can be translated in a lower dependency on external nutrient acquisition for fueling post-emergence egg maturation. In addition, females seem to emerge with sufficient teneral reserves which allow meet maintenance needs. Thus females could use their life time completely to lay their mature eggs without feeding. The potential concentration of their effort towards host exploitation and do not on food consumption is likely to have been selected as an adaptive strategy to habitat characteristics (strongly aggregated host distribution) and resource availability (unpredictable of host and food abundance).

5. REFERENCES

Azzouz, H., Giordanengo, P., Wäckers F. L. and Kaiser L. (2004) Effects of feeding frequency and sugar concentration on behavior and longevity of the adult aphid parasitoid: *Aphidius ervi* (Haliday) (Hymenoptera: Braconidae). *Biol. Conl.* 31: 445-452.

Bernstein, C. and Jervis, M.A. (2008) Food-searching in parasitoids: the dilemma of choosing between 'immediate' or future fitness gains. *Behavioral Ecology of Insect Parasitoids: from theoretical approaches to field applications* (ed. by E. Wajnberg, C. Bernstein & J.J.M. Van Alphen), pp. 129-171. Blackwell Publishing, Oxford, U.K.

Bezemer, T. M., Harvey, J. A. and Mills, N.J. (2005) Influence of adult nutrition on the relationship between body size and reproductive parameters in a parasitoid wasp. *Ecol. Entomol.* 30: 571–580.

Boggs, C.L. and Freeman , K.D. (2005) Larval food limitation in butterflies: effects on adult resource allocation and fitness . *Oecologia* 144: 353 – 361 .

Casas, J., Driessen, G., Mandon, N., Wielaard, B., Desouhant, E., Van Alphen, J.J.M., Lapchin, L., Rivero, A., Christides, J.P. and Bernstein, C. (2003) Energy dynamics in a parasitoid foraging in the wild. *J. Anim. Ecol.* 72:691–697.

Candy, D.J., Becker, A. and Wegener, G. (1997) Coordination and integration of metabolism in insect flight. *Comp. Biochem. Physiol.* 117B: 497–512.

Casas, J., Pincebourde, S., Mandon, N., Vannier, F., Poujol, R. and Giron, D. (2005). Lifetime nutrient dynamics reveal simultaneous capital and income breeding in a parasitoid. *Ecology* 86:545–54

Desouhant, E., Driessen, G., Amat, I. and Bernstein, C. (2005). Host and food searching in a parasitic wasp *Venturia canescens*: a trade-off between current and future reproduction? *Anim. Behav.* 70:145–52.

Desouhant, E., Lucchetta, P., Giron, D., Bernstein, C. (2010) Feeding activity pattern in a parasitic wasp when foraging in the field. *Ecol. Res.* 25: 419-428.

Dyer, L.E. and Landis, D.A. (1996) Effects of habitat, temperature, and sugar availability on longevity of *Eriborus terebrans* (Hymenoptera: Ichneumonidae). *Environ. Entomol.* 25: 1192–1201.

Fadamiro, H.Y., Chen, L., Onagbola, E.O. and Graham, L.F. (2005) Lifespan and patterns of accumulation and mobilization of nutrients in the sugar-fed phorid fly, *Pseudacteon tricuspis*. *Physiol. Entomol.* 30: 212–224.

Giron, D., Rivero, A., Mandon, N., Darrouzet, E. and Casas, J. (2002) The physiology of host feeding in parasitic wasps: implications for survival. *Func. Ecol.* 16: 750–7.

Heimpel, G.E. and Collier, T.R. (1996) The evolution of host-feeding behavior in insect parasitoids. *Biol. Rev. Camb. Philos. Soc.* 71:373–400.

Heimpel, G.E., Rosenheim, J.A. and Kattari, D. (1997) Adult feeding and lifetime reproductive success in the parasitoid *Aphytis melinus*. *Entomol. Exp. Appl.* 83: 305–15.

Heimpel, G.E., Mangel, M. and Rosenheim, J.A. (1998) Effects of time limitation and egg limitation on lifetime reproductive success of a parasitoid in the field. *Am. Nat.* 152: 273–289.

Heimpel, G.E. and Jervis, M.A. (2005) Does floral nectar improve biological control by parasitoids? *Plant-provided Food and Plant – Carnivore Mutualism* (ed . by F . Waeckers , P . van Rijn and J . Bruin), pp . 267 – 304. Cambridge University Press, Cambridge.

Heping, W., Ling, M. and Baoping, L. (2008) Effects of feeding frequency and sugar concentrations on lifetime reproductive success of *Meteorus pulchricornis* (Hymenoptera: Braconidae). *Biol. Cont.* 45: 353-359.

Jervis, M.A., Kidd, N.A.C. and Heimpel, G.E., (1996) Parasitoid adult feeding behaviour and biocontrol—a review. Biocontrol News Inform. 17: 11–26.

Jervis, M.A. and Ferns, P.N. (2004) The timing of egg maturation in insects: Ovigeny index and initial egg load as measures of fitness and of resource allocation. *Oikos* 107: 449–60.

Jervis, M.A. and Heimpel, G.E. (2007) Phytophaghy. *Insects As Natural Enemies: A Practical Perspective* (ed. by M.A. Jervis), pp 543. Springer Publishing, The Netherlands.

Lavandero, I. B., Wratten, S.D., Didham, R. K. and Gurr, G. (2006) Increasing floral diversity for selective enhancement of biological control agents: A double-edged sward? *Basic Appl. Ecol.* 7: 236—243.

Leius, K. (1961) Influence of food on fecundity and longevity of adults of *Ictoplectis conquisitor* (Say) (Hymenoptera: Ichneumonidae). *Can. Entomol.* 93: 771–780.

Lee, J.C., Heimpel, G.E. and Leibee, G.L. (2004) Comparing floral nectar and aphid honeydew diets on the longevity and nutrient levels of a parasitoid wasp. *Entomol. Exp. Appl.* 111: 189–199.

Lee, J.C. and Heimpel, G.E. (2008) Effect of floral nectar, water, and feeding frequency on *Cotesia glomerata* longevity. *BioControl* 53:289–294.

Lee, J.C. and Heimpel, G.E. (2008) Floral resources impact longevity and oviposition rate of a parasitoid in the field. *J. Anim. Ecol.* 77:565–572.

Lewis, W.J., Stapel, J.O., Cortesero, A.M. and Takasu, K. (1998) Understanding How Parasitoids balance food and host needs: importance to biological control. *Biol. Cont.* 11: 175-183.

Mitsunaga, T., Shimoda, T. and Yano, E. (2004) Influence of food supply on longevity and parasitization ability of a larval parasitoid, *Cotesia plutellae* (Hymenoptera: Braconidae). *Appl. Entomol. Zool.* 39: 691–7.

Olson, D. M. and Andow, D. A. (1998) Larval crowding and adult nutrition effects on longevity and fecundity of female *Trichogramma nubilale* Ertle & Davis (Hymenoptera: Trichogrammatidae). *Environ. Entomol.* 27: 508–514.

Olson, D.M., Fadamiro, H., Lundgren, J.G. and Heimpel, G.E. (2000) Effects of sugar feeding on carbohydrate and lipid metabolism in a parasitoid wasp. *Physiol. Entomol.* 25:17–26.

Pelosse, P., Bernstein, C. and Desouhant, E. (2007) Differential energy allocation as an adaptation to different habitats in the parasitic wasp *Venturia canescens*. *Evol. Ecol.* 21:669–685.

Pelosse, P. (2008) Rôle des compromis évolutifs dans la spécialisation et la coexistence d'espèces compétitrices. Développements théoriques et expérimentaux chez les insectes parasitoïdes. PhD thesis, Lyon 1 University, France.

Petanidiou, T. (2007) Ecological and evolutionary aspects of floral nectars in Mediterranean habitats. In: Wilson SW, Nepi M, Pacini E (eds) Nectaries and nectar. Springer, Berlin Heidelberg New York.

Rivero, A. and Casas, J. (1999) Incorporating physiology into parasitoid behavioural ecology: the allocation of nutritional resources. *Res. Popul. Ecol.* 41: 39-45.

Rivero, A., Giron, D. and Casas, J. (2001) Lifetime allocation of juvenile and adult nutritional resources to egg production in a holometabolous insect. *Proc. R. Soc. Biological Sciences,* 268: 1231-1238.

Rivero, A. and West, S.A. (2002) The physiological costs of being small in a parasitic wasp. *Evol. Ecol. Res.* 4: 407–420.

Rosenheim, J.A. (1996) An evolutionary argument for egg limitation. *Evolution* 50: 2089–94.

Rosenheim, J.A. (1999) The relative contributions of time and eggs to the cost of reproduction. *Evolution* 53: 376–385.

Rosenheim, J.A., Heimpel, G.E. and Mangel, M. (2000) Egg maturation, egg resorption, and the costliness of transient egg limitation in insects. *Proceedings of the Royal Society of London B,* 267: 1565–1573.

Sevenster, J.G., Ellers, J. and Driessen, G. (1998) An evolutionary argument for time limitation. *Evolution* 52:1241–1244.

Schmale, I., Wäckers, F.L., Cardona, C. and Dorn, S. (2001) Control potential of three hymenopteran parasitoid species against the bean weevil in stored beans: the effect of adult parasitoid nutrition on longevity and progeny production. *Biol. Cont.* 21: 134-139.

Siekmann, G., Tenhumberg, B. and Keller, M.A. (2001) Feeding and survival in parasitic wasps: sugar concentration and timing matter. *Oikos* 95:425–430.

Sirot E. and Bernstein, C. (1996). Time sharing between host searching and food searching in parasitoids: state-dependent optimal strategies. *Behav. Ecol.* 7:189–94.

Sisterson, M. and Averill, A.L. (2002) Costs and benefits of food foraging for a braconid parasitoid. *J. Insect Behav.* 15: 571–588.

Stapel, J.O., Cortesero, A.M., De Moraes, C.M., Tumlinson, J.H. and Lewis, W.J. (1997) Extrafloral nectar, honeydew and sucrose effects on searching behavior and efficiency of *Microplitis croceipes* (Hymenoptera: Braconidae) in cotton. *Environ. Entomol.* 26: 617-623.

Takasu, K. and Lewis, W.J. (1995) Importance of adult food sources to host searching of the larval parasitoid *Microplitis croceipes*. *Biol. Cont.* 5: 25-30.

Tenhumberg, B., Siekmann, G. and Keller, M.A. (2006) Optimal time allocation in parasitic wasps searching for hosts and food. *Oikos* 113:121–31.

Tylianakis, J.M., Didham, R.K. and Wratten, S.D. (2004) Improved fitness of aphid parasitoids receiving resource subsidies. *Ecology* 85: 658–66.

van Handel, E. (1985) Rapid determination of glycogen and sugars in mosquitoes. *J Amer Mosquito Control Assoc* 1:299–301.

van Handel, E. and Day J.F. (1988) Assay of lipids glycogen and sugars in individual mosquitoes: correlations with length in field-collected *Aedes vexans*. *J Amer Mosquito Control Assoc* 4:549–550.

Wackers, F.L. (2001) A comparison of nectar– and honeydew sugars with respect to their utilization by the hymenopteran parasitoid *Cotesia glomerata*. *J. Insect Physiol.* 47:1077–1084.

Wäckers, F.L. (2004) Assessing the suitability of flowering herbs as parasitoid food sources: flower attractiveness and nectar accessibility. *Biol. Cont.* 29: 307 – 314 .

Wanner, H., Gu, H. and Dorn, S. (2006) Nutritional value of floral nectar sources for flight in the parasitoid wasp, *Cotesia glomerata*. *Physiol. Entomol.* 31: 127-133.

Chapter III

This manuscript has been accepted for publication in the *Journal of Insect Behavior*

The influence of food and con-specifics on the flight potential of the parasitoid *Ibalia leucospoides.*

AUTHORS

Deborah Fischbein (1,2)*, Juan C. Corley (1), José M. Villacide (1),Carlos Bernstein (2).

(1)Laboratorio de Ecología de Insectos, INTA EEA Bariloche CC 277, San Carlos de Bariloche 8400, Río Negro, Argentina.

(2) Laboratoire de Biométrie et Biologie Evolutive UMR 5558. Université de Lyon Université Claude Bernard Lyon 1, 43 Bd du 11 Novembre 1918, 69622 Villeurbanne cedex, France.

*Correspondence: E-mail: dfischbein@bariloche.inta.gov.ar
Telephone number: +54(0)2944-422731
FAX number: +54(0)2944-42 49 91

Running head: Flight potential of *Ibalia leucospoides*

1. ABSTRACT

For insect parasitoids, knowledge of their flight capability is essential for a general understanding of the relationship with their hosts. For instance, flight capacity might partly determine their efficacy as biological control agents. *Ibalia leucospoides* Hochenwarth (Hymenoptera: Ibaliidae) is a solitary, pro-ovigenic parasitoid of the woodwasp, *Sirex noctilio* Boidin (Hymenoptera, Siricidae), an economically important pest of softwood forestation. This study explores the flight capacities of *I. leucospoides* females and assesses the effects of a sugar-rich food supply and crowding on female flight performance, by using computer-linked flight mills. The present study shows: (1)

a high variability in flight potential of *I. leucospoides* females, (2) no effects of food supply and grouping on wasp flight (flight distance and speed), (3) a significant effect of body size and wing loading on flight performance and, (4) a significant body mass loss during the flight dependent on the total distance flown. The lack of effect of food on a highly energy-demanding activity as flight may be related to the life-history traits and nutritional strategies of this parasitoid. The relevance of these observations for the use of *I. leucospoides* as a biological control agent is discussed.

Key words: Hymenoptera, *Ibalidae*, insect flight, flight mills, parasitoids feeding, *Sirex noctilio.*

2. INTRODUCTION

Insects often engage in foraging flights in search for different resources, such as food, mating partners, nesting sites or hosts. Such movements can take place at very different scales, from moving between patches to long-distance displacements between distant populations (Johnson, 1969; Desouhant *et al*., 2003). For parasitoids, insects that lay eggs in or on other arthropods, long distance dispersal may contribute to the stability and persistence of species interactions (Hassell & May, 1973; Roland & Taylor, 1997). In turn, among several attributes, the dispersal capability of parasitoids could affect success of biological control programs. High flight capability may encourage fast expansion from the initial release point, as well as colonization of uninhabited sites (i.e. isolated host patches) and re-invasion of areas where the parasitoids become extinct (Kidd & Jervis, 2007).

For many insects, in metabolic terms, flight is the most costly of all activities (Chapman, 1998). The large amounts of energy consumed in flight can be obtained from different sources such as carbohydrates, fat and protein. Generally, in the Diptera and the Hymenoptera, carbohydrates constitute the main fuel burnt during flight (Chapman 1998; Roff, 1977; Vogt *et al*., 2000; Suarez *et al*., 2005), whereas to sustain flight Lepidoptera, Orthoptera and Hemiptera mainly burn lipids (Cockbain, 1961; Beenakkers, 1969; Sappington *et al*., 1995; Rivero & Casas, 1999). In parasitoids, adult metabolic resources originate from nutrients accumulated during larval development (the 'teneral resources') and from the food obtained as adults. Food can be obtained from the hosts themselves (in host-feeding species) or from non-host sources, such as extra-floral and floral nectar or honeydew (Jervis & Kidd, 1986; Jervis *et al*., 1992; Rivero & Casas, 1999).

In many agro-ecosystems where parasitoids are introduced as biological control agents, the availability of plant-derived food sources can be highly variable (Siekmann *et al*., 2001). In many cases food is unavailable where hosts are found (Jervis & Kidd, 1996; Sirot & Bernstein, 1996; Tenhumberg *et al*., 2006; Bernstein & Jervis, 2008). Thus, it is widely held that sugar-rich food supplementation in target areas and/or prior to parasitoid release should improve the success of biological control (Jervis & Kidd, 1996; Lewis *et al*., 1998; Bianchi *et al*., 2006). This is because adult feeding with sugar sources increases longevity and fecundity in many parasitoids species (Schmale *et al*., 2001; Siekmann *et al*., 2001). Furthermore, food provisioning may also increase flight capability (Wanner *et al*., 2006), allowing a faster spread of parasitoids and a higher host searching efficiency (Staple *et al*. 1997; Lewis *et al*. 1998; Kidd & Jervis, 2007). Consequently, the effects of sugar-rich food supply during rearing and in field conditions on life history traits should be carefully evaluated.

Ibalia leucospoides Hochenwarth (Hymenoptera, Ibaliidae) is a solitary, koinobiont and pro-ovigenic parasitoid of the woodwasp *Sirex noctilio* Boidin (Hymenoptera, Siricidae) (Spradbery, 1977; Madden, 1981). Woodwasps are primitive xylophages that attack pine trees. While *S. noctilio* is native to Mediterranean Europe, in the last century it has successfully invaded Australia, New Zealand, South Africa, South America and North America (Hoebeke *et al*., 2005). In most regions where *S. noctilio* has established, it has become an important pest of pine tree forestation, due to its reported outbreak population dynamics during which tree mortality can be severe (Corley *et al*., 2007). *I. leucospoides* is one of several bio-control agents adopted for pest management purposes of woodwasp populations (Hurley *et al*., 2007). The parasitoid, also native to Europe, was introduced into Australasia in the early 1960′s. *I. leucospoides* has established throughout the invasion range of *S. noctilio*, mostly through accidental introductions, together with its host (Madden, 1988). Under field conditions, *I. leucospoides* may parasitize up to 40% of its hosts.

Because of its applied importance there is a growing research effort on *I. leucospoides.* Most work has focused on its hosts foraging behaviour (Chrystal, 1930; Madden, 1968; Spradbery, 1970 a, b; Fernández-Arhex & Corley, 2005; Martinez *et al*., 2006; Fernández-Arhex & Corley, 2010), but to date, no attempts to evaluate its flight capacity and its relationship with rearing conditions (i.e.: food provisioning; housing in groups) have been made. In cultivated forest systems, trees attacked by *S. noctilio* are spatially aggregated, especially during long-lasting endemic population phases, during which overall *S. noctilio* densities are typically low (Corley *et al*., 2007). Although host foraging and feeding behaviour of *I. leucospoides* are not known in its natural environment, in cultivated forests *I. leucospoides* emerge within host aggregates, where floral nectar is rare or absent. Given the importance of this parasitoid in *S. noctilio* management programs, knowledge on the effects of housing conditions (i.e.: food provisioning; crowding) on flight performance can be helpful in the context of mass rearing and release protocols.

The present study investigates whether sugar supply and crowding influence flight capacity in *I. leucospoides.* The study focuses on the effects of sugar-rich food supply and housing with con-specifics, on female flight performance, as measured in flight mill systems (Schumacher *et al.*, 1997; Blackmer *et al.*, 2004; Wanner *et al.*, 2006; Senger *et al.*, 2007; Bruzzone *et al.*, 2009). Morphological traits such as size and wing loading which are likely contributors to flight abilities are also taken into account (Roff, 1977; Byrne *et al*., 1988; Dudley & Srygley, 1994; Shirai, 1995; Cronin, 2000).

3. MATERIALS AND METHODS

Ibalia leucospoides attacks eggs and first instar larvae of *Sirex noctilio* woodwasps. *I. leucospoides* life cycle is closely related to the development of woodwasp larvae, usually lasting one year (but see Corley *et al*., 2004). This species was first recorded in pine tree plantations in NW Patagonia in 1993, at the time woodwasps were first detected. Since, it has become established in most pine plantations in the region.

Parasitoids were reared from pine (*Pinus contorta*) logs collected in the field, from several heavily attacked plantations located in NW Patagonia (Argentina). Once felled, trees were cut into 1m-long logs and kept individually in locker-type cages under ambient indoor conditions, at 21 ºC, until insect emergence occurred. Each morning newly emerged insects were collected from the cages and immediately placed in individual plastic containers with food or water (see below in experimental design section).

3.1 ***Experimental design***

The influence of two factors, food access ("*feeding*") and crowding ("*conspecifics*") was tested according to a factorial design. Feeding had two conditions: fed animals provided with 30% diluted honey solution *ad libitum* and unfed wasps with access only to distilled water. In turn, conspecifics also had two levels: isolated females (housed in individual containers, immediately after emergence) and grouped females (housed in the same container, in groups of four). The storage containers were of 251 cm^3, a relatively small volume for four rather large individuals (13.41 mm, mean body length). Wasps to be tested of the grouped treatment were picked at random from the cage immediately before the experiment.

All tested animals were kept under the corresponding treatment for seven days within a rearing chamber under semi-controlled temperature and humidity conditions (temperature 21 ± 0.1ºC; 54% r.h.), and a LD 16:8h photoperiod. Flight trials began 4 h after to the onset of the photo-phase. Each female flew in the mills for 22 h and was weighed (Scientech SA210; Scientech, Boulder, Colorado, USA; to the nearest 0.0001g.) immediately before and after flight. The differences in weight before flight were used as a measure of any interference between individuals while feeding in groups. In addition, the weight before flight and the weight after flight were used to calculate loss of body mass (see below). Seven days of food deprivation ensured that fed and unfed wasps differed in their carbohydrate energy reserves (D. Fischbein, unpublished data).

Two morphological variables were measured: hind tibia length (TL, in cm) and wing loading (WL = body mass (g)/ wing area (cm^2). The left and right wings of each wasp were digitally scanned and with the aid of an image processing software (HOJA

Software, Verga A.R., INTA IFIVE Argentina, 2001) the forewing and hind wing areas measured.

3.2 *Flight mills*

To study the effects of food consumption and grouping on the flight performance of *I. leucospoides*, flight mills were used based on the model of Bruzzone *et al.* (2009). Four mills were run simultaneously on each day of experimentation. Each female was anesthetized with CO_2 for 20 s (Bruzzone *et al.*, 2009; Yu *et al.*, 2009) and the dorsal side of the thorax was glued to one end of an L shaped insect pin, using cyanoacrylate glue. The pin was shaped in order to create a suitable flight angle and greater surface area of contact between the pin and the insect thorax. Finally, the opposite free end of the pin was attached to one end of a wooden, horizontal rod. Wasps were left to recover for 15 min before flight tests.

The rotation of the mills was detected by infrared optical sensors connected through a circuit to the parallel port of a microcomputer. A Python script was designed to collect data from the interface board. Each spin made by the wasp was recorded when the beam of light generated by a LED was interrupted by the end of the wooden rod opposite to the one where the insect was held. The computer recorded each revolution (48.7 cm) as the wasp propelled the wooden rod (15.5 cm long). For each flight mill, the programme recorded every spin, speed rotation ($m.s^{-1}$) and accumulated flight distance (km). The data series, which consisted of a series of irregularly separated events (records of each time the wasp activated the sensor), were re-sampled to a series of speed data uniformly separated by 1 second. On this basis, the following flight parameters were estimated: total distance flown in kilometres, maximum 30 minutes-running mean speed throughout the whole flight (*V30min*, $m.s^{-1}$) and maximum 60 seconds-running mean speed, also through the duration of flight (*V60s*, $m.s^{-1}$).

The running mean speed is a mean obtained from a speed data set using a fixed time interval (i.e.: 60 s and 30 min). This running mean speed reduces the noise and reveals the speed tendencies. As new data were added to the series, the first data is eliminated, enabling the period to remain constant. As aforementioned, we calculated the running mean speed using two different periods (short and long time intervals) to ensure better quality information. The short periods (60 s) result in a closer and more sensitive tracking of speed changes while long intervals (30 min) enable us to track a long term tendency in speed variation. *I. leucospoides* females started to fly soon after they were introduced in the flight mills. Wasps flew for several hours and stops of variable periods were frequently observed (D. Fischbein, pers. obs). Again, the use of moving intervals allowed us to cope with inactivity periods, reducing the noise produced by them.

3.3 *Statistical analysis*

For the analyses of the influence of feeding, conspecifics and morphological variables on the different flight parameters, a linear model assuming a normal distribution of residuals was used. Flight parameters and morphological variables were log transformed to take into account allometric relationships. The data were first fitted to an initial model and then least significant variables were progressively removed from the model until a minimal appropriate model was obtained (i.e. a simplified model in which all terms are significant). We tested the effect of removing successive factors and variables by means of ANOVA. Single factors or variables incorporated into significant interactions were maintained in the minimum adequate model (Crawley, 2007). The initial model fitted to the data was $\log_e$ (*flight parameter*) = $\log_e$(WL)* feeding * conspecifics + $\log_e$(TL) * feeding * conspecifics, where * represents the sum of the influences of three-way interactions, two-way interactions and the four single variables. Inspection of residuals confirmed the fit to a normal distribution.

To assess whether the presence of conspecifics would cause interference while feeding, the effects of feeding, conspecifics and hind tibia length on the weight before flight were analyzed. A linear model assuming a normal distribution of residuals was used and the weight and hind tibia length were log transformed. A backward procedure was used to remove non-significant factors and interactions between factors, and to select the final models. Residuals were examined to confirm that the final models accurately fitted the data.

To further compare the effect of access to food on metabolic expenses in flight, the loss of body mass during flight as a function of log-transformed total distance flown and feeding was examined. The total body mass change was estimated as log (initial mass – final mass)/initial mass. All data analyses were done using the R statistical environment (R Development Core Team, 2008).

4. RESULTS

There was high variability in flight performance of females of *Ibalia leucospoides* (Fig.1). Flight parameters studied were neither affected by feeding (*total distance flown*: $F_{(1,61)} = 0.03$, $P = 0.856$; *V30min*: $F_{(1,62)} = 2.10$, $P = 0.152$; *V60s*: $F_{(1,60)} = 0.62$, $P = 0.432$) nor by mass housing (conspecifics; *total distance flown*: $F_{(1,60)} = 0.004$, $P = 0.946$; *V30min*: $F_{(1,60)} = 0.05$, $P = 0.823$; *V60s*: $F_{(1,61)} = 0.82$, $P = 0.367$).

The variability in total distance flown by female wasps was mostly accounted for by morphological variables (wing loading and tibia length) (Table.1). Larger females flew farther than smaller females (TL: $F_{(1,62)} = 6.11$, $P = 0.016$) and, females with high wing loading flew longer total distances than females with low wing loading (WL: $F_{(1,62)} = 7.32$, $P= 0.008$). Wing loading had a significant effect on the maximum running mean speeds, both *V30min* and *V60s* increasing with wing loading ($F_{(1,63)} = 7.73$, $P= 0.007$ and $F_{(1,63)} = 17.66$, $P< 0.0001$, respectively).

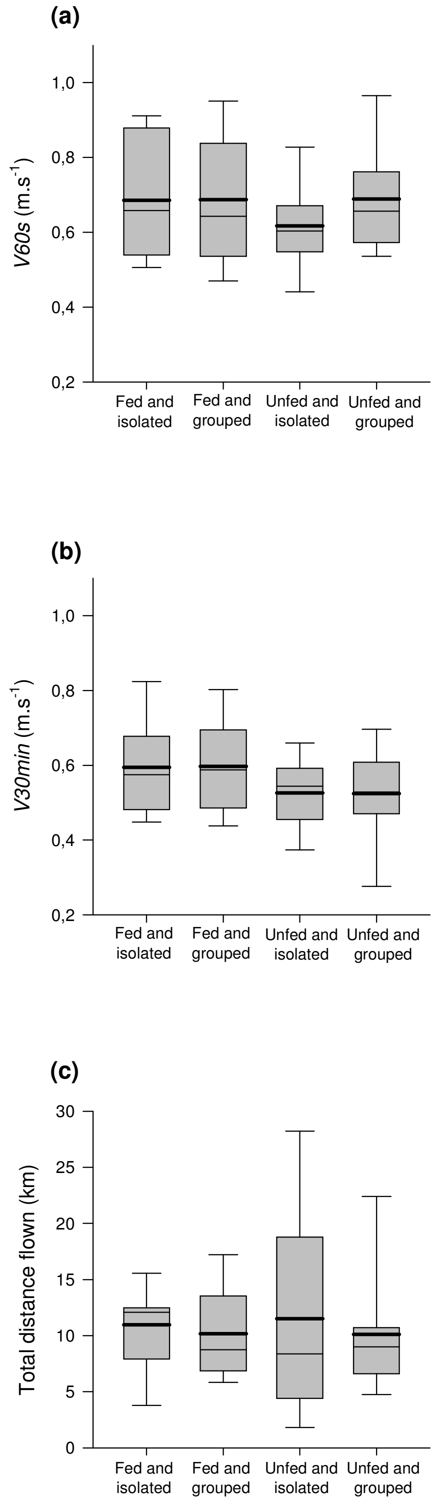


Figure 1. Flight parameters of *I. leucospoides* females according to the factorial design; (a) *V60s*, (b) *V30min* and (c) total distance flown. Flight parameters studied were neither affected by *feeding* nor by *conspecifics* (backward linear model: P<0.005). The thin horizontal line of each box plot marks the median of the sample and the thick horizontal line marks the mean of the sample.

TABLE 1 Mean (± SE) morphological variables and weights of *I. leucospoides* females according to the factorial design.

	Wing loading (g/cm^2) Mean ± SE	Tibia length (cm) Mean ± SE	Weight before flight (g) Mean ± SE	Percentage body mass loss Mean ± SE	n
Fed and isolated	0.0869 ± 0.0015	0.4739 ± 0.0075	0.0419 ± 0.0016	9.48 ± 1.10	19
Fed and grouped	0.0833 ± 0.0026	0.4563 ± 0.0071	0.0381 ± 0.0022	11.86 ± 1.47	17
Unfed and isolated	0.0730 ± 0.0040	0.4492 ± 0.0125	0.0326 ± 0.0026	9.56 ± 1.43	13
Unfed and grouped	0.0792 ± 0.0030	0.4581 ± 0.0089	0.0377 ± 0.0022	8.14 ± 1.33	16

Variations in weight before flight were analyzed in order to test whether some interference between wasps might have occurred while they were fed under the grouped housing conditions. Weight was significantly affected by feeding; fed females were heavier than unfed ones (mean ± SE: 0.0401 ± 0.0014 g and 0.0354 ± 0.0017 g, respectively; $F_{(1, 62)} = 14.205$, $P< 0.001$), but this effect was independent of the presence of conspecifics. No interaction between feeding and conspecifics affected weight before flight ($F_{(1, 59)} = 1.52$, $P=0.22$). In addition, the variability in wasp' weight was also explained by tibia length ($F_{(1,62)} = 135.16$, $P< 0.0001$). Finally, the body mass loss during the flight trial was affected by total distance flown ($F_{(1,61)} = 5.89$, $P = 0.018$) (Fig. 2), but was neither affected by feeding ($F_{(1,60)} = 1.68$, $P = 0.198$) nor by interaction between the two explanatory factors ($F_{(1,59)} =0.91$, $P =0.342$).

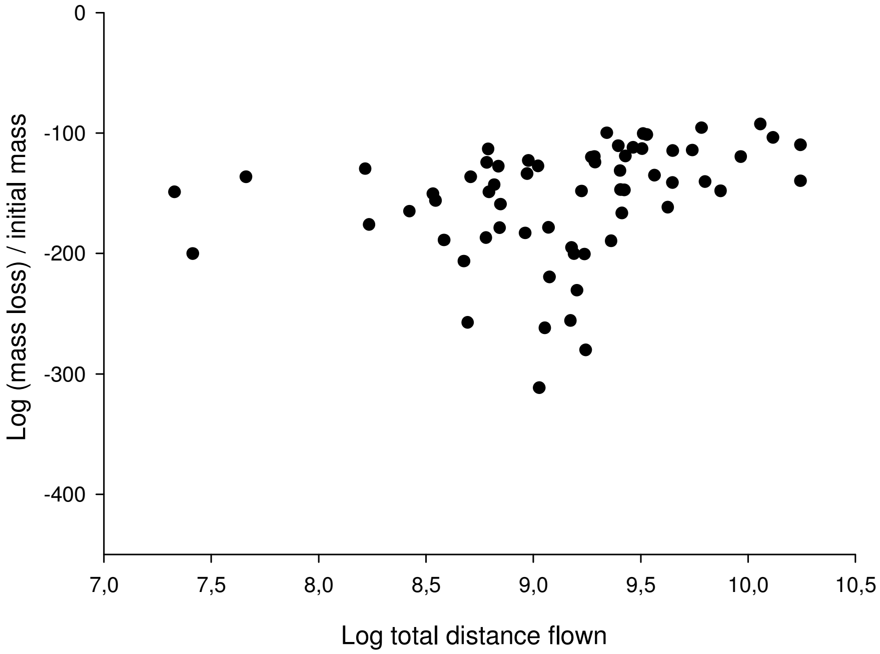


Figure 2. Female body mass loss during flight trials plotted against total distance flown (m, log-transformed). The total body mass change was estimated as log (initial mass – final mass)/initial mass.

5. DISCUSSION

This study is the first one to quantify the flight capacity of *Ibalia leucospoides*, a biocontrol agent of *Sirex noctilio,* which is one of the most economically significant pests of softwood forestation. The present study assessed whether flight performance of *I. leucospoides* was affected by unlimited access to sugar-rich food and the presence of conspecifics (data controlled for body size). Our findings show in general, a high variability in the flight performance of females of *I. leucospoides*. Two morphological variables (tibia length and wing loading), which are surrogate variables of body size, affected flight potential. The total distance that could be flown by wasps increased with tibia length and wing loading, and speed increased with wing loading. Conversely, feeding and presence of conspecifics did not affect flight parameters. According to the results obtained, access to food and body size (TL) increased female weight before flights, regardless of the presence of (and contact with) con-specifics. Finally, body mass loss of females during flight trials depended only on the total distance flown.

I. leucospoides females are capable of long-distance flights but exhibit about half of the flight potential of their host *S. noctilio* (the latter ranged between a minimum of 1 to a maximum of nearly 50 km per day in similar experimental conditions [Bruzzone *et al*., 2009]). However, recent field studies suggest that *S. noctilio* dispersal within a forest is very limited -less than 150 m- exhibiting a highly clumped spatial pattern of tree attacks during the endemic phase of host population growth (Corley *et al*., 2007). It is worth mentioning that even displaying half of host flight capacity, the flight potential of *I. leucospoides* may allow them to fly from tree to tree searching for hosts. Therefore, the flight potential displayed by *I. leucospoides* females could allow a thorough exploitation of host patches within and between aggregates of pest-attacked trees within pine plantations (see also Corley *et al*., 2010). However, whether *I. leucospoides* is able to follow its host during a regional spread process remains as an open question.

Flight capacity in insects is affected by their morphological traits. For example, Shirai (1995) showed that the larger moths of *Plutella xylostella* display greater flight abilities than smaller ones. Another example is *Sirex noctilio*; large females are capable of longer and faster flights than smaller ones (Bruzzone *et al*., 2009). Furthermore, Dudley *et al*. (1994) and Berwaerts *et al.* (2002) in their studies on the relationship between morphological variation and variation in flight performance have found a positive correlation between speed or acceleration capacity, and both size indicators and wing loading. According with this, in the present study, wasps with greater body sizes and wing loading ratios are able to fly longer distances than smaller ones. This fact may be due to a greater level of body teneral reserves. In addition, a high wing loading ratio increases flight speed. Therefore, the wing loading *per se* had an effect on flight performance in *I. leucospoides*, independently of adult feeding treatment. In this sense, this study provides additional evidence that morphological traits are crucial aspects of flight capability in insects.

Adult feeding with sugar is known to increase longevity and fecundity in many parasitoid species (Schmale *et al*., 2001; Siekmann *et al*., 2001), and to modify the propensity to search for hosts (Takasu & Lewis 1995; Staple *et al.,* 1997). Adult food consumption, namely by providing additional resources, may increase flight performance. For instance, Wanner *et al*. (2006) showed for the synovigenic parasitoid species *Cotesia glomerata,* that fed adult female wasps are able to increase flight capacity. They also showed that nectar with different nutritional values has different effects on flight activity in this species. In contrast to these findings, in this study flight parameters in *I. leucospoides* were not affected by prior access to food. It is possible that such an effect may manifest itself on subsequent days of flight. For instance, it is possible that carbohydrate energy reserves in unfed wasps are sufficient to fuel one day of flight at a similar level as fed wasps, but that flight on later days would be reduced or not feasible.

An alternative explanation for this lack of effect of food provisioning on flight capacity of *I. leucospoides* females may be related to the life-history traits and nutritional strategies of this species. Parasitoid wasps differ in their egg maturation strategies (Flanders, 1950; Jervis *et al.*, 2001). Some parasitoids emerge with their entire or near entire, complement of eggs (the so-called pro-ovigenic parasitoids) and some others continue to mature eggs throughout their reproductive life (the so-called synovigenic parasitoids). While the synovigenic parasitoids need to feed (from hosts or sugar sources) to fuel both somatic functions and reproduction, pro-ovigenic species may rely on sugar sources for maintenance and locomotion only (Rivero *et al.*, 1999; Jervis *et al.*, 2001; Rivero *et al.*, 2001; Pelosse, 2008). As a consequence, females of pro-ovigenic species should, in theory, have lower need for supplementary food (Jervis & Heimpel 2007). Given that *I. leucospoides* is a nearly pro-ovogenic parasitoid, it is possible that the effects of food obtained as an adult on flight performance is minimal. In this species, females could carry a nutrient storage (obtained during their larval development) sufficient to me*et al.*l their maintenance and locomotion needs.

Although the age-dependent variation in flight capacity can be a recurrent pattern attributed to species-specific temporal changes in physiological status (Johnson & Rowley, 1972; Schumacher *et al.,* 1997; Elliott & Evenden, 2009; Lukáš *et al.*, 2010) this is not a rule for all parasitoids. For example, flight duration of *Nasonovia vitripennis* does not differ between young and old individuals (King, 1993). In the present study, female wasps of a given age -seven days old- were used. This age lies in the middle range of the estimated female lifespan. Additional studies should be performed to discern if flight behavior at different ages retains the same characteristics.

No effects of housing in groups of 4 were noted on flight parameters. Females do not interfere with each other when feeding in groups and have a similar body mass increase compared with females housed alone with a food source. This observation, coupled with the results noted for the feeding treatment, can have important applied implications. The results indicate that mass rearing, even in the absence of food provisioning, is unlikely to negatively affect the subsequent flight performance of released parasitoids and so success in the biological control of *S. noctilio* populations in pine plantations. These results were obtained by analyzing tethered flight; the next step is to confirm these findings in free flight experiments. This study is a first attempt to evaluate conditions of food provisioning and grouping in the setting of laboratory mass rearing of this species. Additional efforts have to be made to understand how to manipulate the fitness of this parasitoid prior to release in order to maximize their efficiency in pest control.

The lack of effect of food intake on a highly energy-demanding activity as flight may reflect a particular property of pro-ovigenic parasitoids. However, further research is required to provide empirical evidence of the dynamics of nutrient utilization during flight and of the potential role that food could have on other life history traits (longevity and fecundity) of *I. leucospoides.*

ACKNOWLEDGMENTS

We acknowledge financial support from ANPCyT, Argentina (PICT 0-1200/06), CNRS, France (PICS 4144) and INTA, Argentina. We thank C. Lazzari, P. Pelosse and D. Giron for their valuable comments on an earlier version of the manuscript. We thank Enzo Sauro (Centro Atómico Bariloche) for helping us design the computer interface board.

6. REFERENCES

Beenakkers, A.M.T. (1969) Carbohydrate and fat as a fuel for insect flight. A comparative study. *J. Insect Physiol.* 15: 353-361.

Bernstein, C., and Jervis M.A. (2008) Food-searching in parasitoids: the dilemma of choosing between 'immediate' or future fitness gains. *Behavioral Ecology of Insect Parasitoids: from theoretical approaches to field applications* (ed. by E. Wajnberg, C. Bernstein & J.J.M. Van Alphen), Blackwell Publishing, Oxford, U.K. pp. 129-171.

Berwaerts K., Van Dyck H., and Aerts P. (2002) Does flight morphology relate to flight performance? An experimental test with the butterfly *Pararge aegeria. Func. Ecol.* 16: 484-491.

Bianchi, F.J.J.A., Booij, C.H.J., and Tscharnkte, T. (2006) Sustainable pest regulation in agricultural landscapes: a review on landscape composition, biodiversity and natural pest control. Proc. R. Soc. Lond. *B Biol. Sci*. 273: 1715-1727.

Blackmer, J.L., Naranjo, S.E., and Williams, L.H. (2004) Tethered and untethered flight by *lygus hesperus* and *lygus lineolaris* (Heteroptera: Miridae). *Environ. Entomol.* 33: 1389-1400.

Bruzzone, O.A., Villacide, J.M., Bernstein, C., and Corley, J.C. (2009) Flight variability in the woodwasp *Sirex noctilio* (Hymenoptera: Siricidae): an analysis of flight data using wavelets. *J. Exp. Biol.* 212: 731-737.

Byrne, D.N., Buchmann, S.L., and Spangler, H.G. (1988) Relationship between wing loading, wingbeat frequency and body mass in homeopterous insects. *J. Exp. Biol.* 135: 9-23.

Chapman RF. 1998. *The Insects: Structure and Function*. Cambridge, UK: Cambridge Univiversity Press

Chrystal, R.N. (1930) Studies of the *Sirex* parasites. Oxford Forestry Memories No 11. Oxford University Press, London, U.K.

Cockbain, A.J. (1961) Fuel utilization and duration of tethered flight in *Aphis fabae* Scop. *J. Exp. Biol.* 38: 163-174.

Corley, J.C., Capurro, A.F., and Bernstein, C. (2004) Prolonged diapause and the stability of host-parasitoid interaction. *Theor. Popul. Biol.* 65: 193-203.

Corley, J.C., Villacide, J.M., and Bruzzone, O.A. (2007) Spatial dynamics of a *Sirex noctilio* woodwasp population within a pine plantation in Patagonia. *Entomol. Exp. Appl.* 125: 231-236.

Corley, J.C., Villacide, J.M., and van Nouhuys, S. (2010) Patch time allocation by the parasitoid *Ibalia leucospoides* (Hymenoptera: Iballidea): the influence of con-specifics, host abundance and distance to the patch. *J. Insect Behav*. 23: 431-440.

Crawley, M.J. (2007) *The R Book*. John Wiley & Sons, Chichester, West Sussex, U.K.

Cronin, J.T., Reeve, J.D., Wilkens, R., and Turchin, P. (2000) The pattern and range of movement of a checkered beetle predator relative to its bark beetle prey. *Oikos,* 90: 127-138.

Desouhant, E., Driessen, G., Lapchin, L., Wielaard, S., and Bernstein, C. (2003) Dispersal between host populations in field conditions: navigation rules in the parasitoid *Venturia canescens. Ecol. Entomol.* 28: 257-267.

Dudley, R., and Srygley, R.B. (1994) Flight physiology of neotropical butterflies: allometry of airspeeds during natural free flight. *J. Exp. Biol.* 191: 125-139.

Elliott, C.G., and Evenden, M.L. (2009) Factors influencing flight potential of *Choristoneura conflictana. Physiol. Entomol.* 34: 71–78

Fernández-Arhex, V., and Corley, J.C. (2005) The functional response of *Ibalia leucospoides* (Hymenoptera: Ibaliidae), a parasitoid of *Sirex noctilio* (Hymenoptera: Siricidae). *Biocontrol Sci. Tech.* 15: 207-212.

Fernández-Arhex, V., and Corley, J.C. (2010) The effects of patch richness on con-specific interference in the parasitoid *Ibalia leucospoides* (Hymenoptera, Ibaliidae). *Insect Sci.* 17: 379-385.

Flanders, S.E. (1950) Regulation of ovulation and egg disposal in the parasitic Hymenoptera. *Can Entomol.* 82: 134-140.

Hassell, M.P., and May, R.M. (1973) Stability in insect host–parasite models. *J. Anim. Ecol.* 42: 693-736.

Hoebeke, E.R., Haugen, D.A., and Haack, R.A. (2005) *Sirex noctilio*: Discovery of a Palearctic siricid woodwasp in New York. *Newsletter of the Michigan Entomological Society.* 50: 24-25.

Hurley, B.P., Slippers, B., and Wingfield, M.J. (2007) A comparison of control results for the alien invasive woodwasp, *Sirex noctilio*, in the southern hemisphere. *Agric. Forest Entomol.* 9: 159-171.

Jervis, M.A., Kidd, N.A.C., and Walton, M. (1992) A review of methods for determining dietary range in adult parasitoids. *Entomophaga.* 37: 565-574.

Jervis, M.A., and Kidd, N.A.C. (1996) Parasitoid adult feeding behaviour and biocontrol: a review. *Biocontrol News and Information.* 17: 11-26.

Jervis, M.A., Heimpel, G.E., Ferns, P.N., Harvey, J.A., and Kidd, N.A.C. (2001) Life-history strategies in parasitoid wasps: a comparative analysis of 'ovigeny'. *J. Anim. Ecol.* 70: 442-458.

Jervis, M.A., and Heimpel, G.E. (2007) Phytophaghy. *Insects As Natural Enemies: A Practical Perspective* (ed. by M.A. Jervis). Springer Publishing. The Netherlands. pp 543.

Johnson, B.G.J, and Rowley, W.A. (1972) Age-related ultra-structural changes in the flight muscle of the mosquito, *Culex tarsalis*. *J. Insect Physiol.* 18: 2375–2389.

Kidd, N.A.C., and Jervis, M.A. (2007) Population Dynamics. *Insects As Natural Enemies: A Practical Perspective* (ed. by M.A. Jervis). Springer Publishing. The Netherlands. pp 515.

King, B. (1993) Flight activity in the parasitoid wasp *Nasonia vitripennis* (Hymenoptera: Pteromalidae). *J. Insect Behav.* 6: 313–321.

Lewis, W.J., Stapel, J.O., Cortesero, A.M., and Takasu, K. (1998) Understanding How Parasitoids balance food and host needs: importance to biological control. *Biol. Control.* 11: 175-183.

Lukáš, J., Bernstein,C., Gu, H., and Dorn, S. (2010) Could different environmental conditions select for different flight characteristics in sexual and asexual parasitoid *Venturia canescens*?. *Entomol. Exp. Appl.* DOI 10.1111/j.1570-7458.2010.01000.x

Madden, J.L. (1968) Behavioural responses of parasites to the symbiotic Fungus associated with *Sirex* noctilio F. *Nature.* 218: 189-190.

Madden, J.L. (1981) Egg and larval development in the woodwasp *Sirex noctilio*. *Australian J. Zool.* 29: 493-506.

Madden, J.L. (1988) *Sirex* in Australasia. Dynamics of forest insect populations: patterns, causes and implications (ed. By A.A. Berryman). Plenum Press. pp. 407-427.

Martínez, A., Fernández-Arhex, V., and Corley, J.C. (2006) Chemical information from the fungus *Amylostereum aerolatum* and host foraging behaviour in the parasitoid *Ibalia leucospoides. Physiol. Entomol.* 31: 1-5.

Pelosse, P. (2008) Rôle des compromis évolutifs dans la spécialisation et la coexistence d'espèces compétitrices. Développements théoriques et expérimentaux chez les insectes parasitoïdes. PhD thesis, Lyon 1 University, France.

Rivero, A., and Casas, J. (1999) Incorporating physiology into parasitoid behavioural ecology: the allocation of nutritional resources. *Res. Popul. Ecol.* 41: 39-45.

Rivero, A., Giron, D., and Casas, J. (2001) Lifetime allocation of juvenile and adult nutritional resources to egg production in a holometabolous insect. Proc. R. Soc. Lond. *Biol. Sci.* 268: 1231-1238.

R Development Core Team (2008) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL http://www.R-project.org.

Roff, D.A. (1977) Dispersal in dipterans: its costs and consequences. *J. Anim. Ecol.* 46: 443-456.

Roland, J., and Taylor, P.D. (1997) Insect parasitoide species respond to forest structure at different special scales. *Nature.* 386: 710-713.

Sappington, T.W., Fescemyer, H.W., and Showers, W.B. (1995) Lipid and carbohydrate utilization during flight of the migratory moth, *Agrotis ipsilon* (Lepidoptera: Noctuidae). *Arch. Insect Biochem. Physiol.* 29: 397- 414.

Schmale, I., Wäckers, F.L., Cardona, C., and Dorn, S. (2001) Control potential of three hymenopteran parasitoid species against the bean weevil in stored beans: the effect of adult parasitoid nutrition on longevity and progeny production. *Biol Control.* 21: 134-139.

Schumacher, P., Weyeneth, A., Weber, D.C., and Dorn, S. (1997) Long flights in *Cydia pomonella* L. (Lepidoptera: Tortricidae) measured by a flight mill: influence of sex, mated status and age. *Physiol. Entomol.* 22: 149-160.

Senger, S.E., Roitberg, B.D., and Thistlewood, H.M.A. (2007) Relative flight responses of *Rhagoletis indifferens* as influenced by crowding, sex, and resources. *Entomol. Exp. Appl.* 123: 91-100.

Shirai, Y. (1995) Longevity, flight ability and reproductive performance of the diamondback moth, *Plutella xylostella* (L.) (Lepidoptera: Yponomeutidae), related to adult body size. *Res. Popul. Ecol.* 37: 269-277.

Siekmann, G., Tenhumberg, B., and Keller, M.A. (2001) Feeding and survival in parasitic wasps: sugar concentration and timing matter. *Oikos*. 95: 425-430.

Sirot, E., and Bernstein, C. (1996) Time sharing between host searching and food searching in solitary parasitoids: State dependent optimal strategies. *Behav. Ecol.* 7: 189-94.

Spradbery, J.P. (1970a) Host finding by *Rhyssa persuasoria*, an ichneumonid parasite of siricid woodwasps. *Anim. Behav.* 18: 103-114.

Spradbery, J.P. (1970b) The biology of *Ibalia drewseni* Borries, a parasite of siricid woodwasps. *Proc. Entomol. Soc. London B.* 45: 104-113.

Spradbery, J.P. (1977) The oviposition biology of siricid woodwasps in Europe. *Ecol. Entomol.* 2: 225-230.

Stapel, J.O., Cortesero, A.M., De Moraes, C.M., Tumlinson, J.H., and Lewis, W.J. (1997) Extrafloral nectar, honeydew and sucrose effects on searching behavior and efficiency of *Microplitis croceipes* (Hymenoptera: Braconidae) in cotton. *Environ. Entomol.* 26: 617-623.

Suarez, R.K., Darveau, C.A., Welch, K.C., O'Brien, D.M., Roubik, D.W., and Hochachka, P.W. (2005) Energy metabolism in orchid bee flight muscles: carbohydrate fuels all. *J. Exp. Biol.* 208: 3573-3579.

Tenhumberg, B., Siekmann, G., and Keller, M.A. (2006) Optimal time allocation in parasitic wasps searching for hosts and food. *Oikos.* 113: 121-131.

Takasu, K., and Lewis, W.J. (1995) Importance of adult food sources to host searching of the larval parasitoid *Microplitis croceipes*. *Biol. Control.* 5: 25-30.

Vogt, J.T., Appel, A.G., and West, M.S. (2000) Flight energetics and dispersal capability of the fire ant, *Solenopsis invicta* Buren. *J. Insect Physiol.* 46: 697-707.

Wanner, H., Gu, H., and Dorn, S. (2006) Nutritional value of floral nectar sources for flight in the parasitoid wasp, *Cotesia glomerata*. *Physiol. Entomol.* 31: 127-133.

Yu, H., Zhang, Y., Wu, K., Wyckhuys, K.A.G., and Guo, Y. (2009) Flight potential of *Microplitis mediator*, a parasitoid of various lepidopteran pests. *BioControl*. 54: 183-193.

Chapter IV

This manuscript is in preparation

Patch choice from a distance and use of habitat information during foraging by the parasitoid *Ibalia leucospoides*

AUTHORS

Deborah Fischbein [1,3], Julieta Bettinelli [2], Carlos Bernstein [3], Juan C. Corley [1].

[1] Grupo de Ecología de Insectos, INTA EEA Bariloche CC 277, San Carlos de Bariloche 8400, Río Negro, Argentina.

[2] Julieta Bettinelli

[3] Laboratoire de Biométrie et Biologie Evolutive UMR 5558. Université de Lyon Université Claude Bernard Lyon 1, 43 Bd du 11 Novembre 1918, 69622 Villeurbanne cedex.

1. ABSTRACT

(1) In patchy environments where resources are heterogeneously distributed, patch choice and patch time-allocation by foragers are subject to strong selective pressures. This is particularly so for parasitoids, because in these animals foraging success translates directly into individual fitness.

(2) The aim of this study was to test whether females of *Ibalia leucospoides* (Hymenoptera: Ibaliidae), a parasitoid of the woodwasp *Sirex noctilio* Boidin (Hymenoptera, Siricidae), discriminate patches according to their host number and, once on patch, whether the time they spend on them is influenced by the number of hosts in the surrounding patches.

(3) *I. leucospoides* females were exposed to three different triple-patch environments which differed in host availability per patch, in the average environment host availability and in host distribution among patches. Each environment consisted of three host-containing pine logs (patches) which were placed in the corners of an imaginary triangle within a wire mesh cage ($1m^3$). The experimental wasp was released from the

exact centre of this arrangement and the first patch chosen and the time allocated to each patch visited were recorded.

(3) *I. leucospoides* is able to discriminate different levels in host availability among patches from a distance. The first patch chosen was always that bearing the highest number of hosts. In addition, the time allocated to the exploitation of the first patch visited depended not only on host availability in the patch but also on the quality of surrounding patches.

(4) These results indicate that *I. leucospoides* could make host searching decisions from the outset, by accurately assessing the initial density of host in patches at a distance without the need of a sampling process. Once on the patch, female exploitation rules were influenced by the information obtained from the surrounding patches, again without sampling.

(5) For parasitoid living in highly heterogeneous environments, such behavioural abilities may prove of high value since they may contribute to a better match between parasitoid foraging effort and host availability.

Correspondence: E-mail: dfischbein@bariloche.inta.gov.ar

Key words: *parasitic insects*, foraging behaviour, patch decision rules, Marginal Value Theorem.

2. INTRODUCTION

Animals usually live in patchy environments where resources are heterogeneously distributed. In these conditions, mechanisms for resource patch choice and patch time-allocation strategies are subject to strong selective pressures (Stephens & Krebs, 1986). This is particularly so for parasitoids, insects that lay their eggs in or on other arthropods, because in these animals foraging success translates directly into offspring production and so on individual fitness. This behavioural trait makes parasitoids ideal subjects to study foraging decisions. As a consequence, foraging strategies such as host patch choice and patch-time allocation, and other factors influencing parasitoids foraging behaviour, have been the subject of an important research effort (see Wajnberg, 2006 for a review).

Parasitoid females frequently search for their hosts in highly variable environments. In such settings, appropriate gathering and use information might help them make adaptive foraging decisions (Mc Namara & Houston, 1985; Haccou *et al.*, 1991; Hemerik *et al.*, 1993; Vos *et al.,* 1998; Thiel & Hoffmeister, 2006). Accordingly, parasitic wasps may sample patches when searching and, thus estimate the profitability of the current patch and of the surrounding habitat, by continuously updating information (Hubbard & Cook, 1978; Waage, 1979; Bernstein *et al.*, 1988; Pierre *et al.*, 2003; Tentelier *et al.*, 2006; 2007). This patch sampling process should help non-omniscient foragers to make better patch choice and usage decisions.

A great variety of environmental cues are used as sources of information for both locating and assessing host patches by parasitoids (Vet & Dicke, 1992). These cues can operate as indicators of host presence, host species (Thiel & Hoffmeister, 2006), host density (Waage, 1979; Li *et al.*, 1997; Shatiel & Ayal, 1998; Wang & Keller, 2004; Martinez *et al.*, 2006, Corley *et al.*, 2010) as well as of the presence of competitors in the current patch (Janssen *et al.,* 1995 a,b; Bernstein & Driessen, 1996; Castelo *et al.*, 2003). The use of such information is expected, therefore, to reduce uncertainty and so improve foraging success.

Taking patch choice decisions on the basis of an expected host availability (i.e. estimated through patch sampling or else through stimuli discrimination from a distance) might lead, in the long run, to an adaptive exploitation of resources. However, the quality of the estimate obtained depends on different factors. For instance, it depends on the number of patches already visited and the development of learning abilities. The initial inborn estimate can be highly inaccurate and lead at early stages of the learning process to inappropriate decisions. Alternatively, the quality of the estimate can rely on the ability to respond to host densities cues prior to patch exploitation (Geervliet *et al.*, 1998; Liu *et al.*, 2009).

In some circumstances much higher gains could be obtained if the animals are able to detect and exploit the best patch in a cluster, upon arrival. This strategy might be particularly appropriate at least under two circumstances: (i) when female that contain all or nearly all their oocytes mature upon emergence have the chance of finding patches in which the host availability is suffice for nearly match with female's egg load. This can so result in a reduction of time limitation incidence; (ii) whenever long visits to a cluster of patches might entail a risk, such as predation. This alternative strategy has been little explored both from theoretical and experimental points of view. In many host-parasitoid systems different cues such as secondary chemical compounds released by plants, visual indications of plant consumption by herbivores and volatile kairomones released by host can provide a rapid, if rough estimate of host availability and be the basis of an adaptive patch choice (Vet & Dicke, 1992; Turlings & Wäckers, 2004; Tentelier *et al.*, 2005; Tentelier & Fauvergue, 2007).

Once on a patch, parasitoids should be able to adjust the time allocated to it in order to maximize their lifetime reproductive success. The Marginal Value Theorem (MVT; Charnov, 1976), the most influential rate maximization model, predicts that an optimal forager should leave a resource patch when the instantaneous rate of fitness gain falls below the average rate of gain expected for the habitat. Therefore, optimal residence time on a patch would increase both with increasing resource availability in the patch or with decreasing availability in the habitat as a whole (i.e. habitat profitability). However, the MVT model does not suggest patch exploitation mechanisms that foragers should employ to achieve the optimal residence time. Simple behavioural rules such as the so-called rules of thumb (Iwasa *et al.*, 1981; McNair, 1982; Green, 1984; Stephens & Krebs, 1986) or more complex and dynamic incremental and decremental patch-leaving rules have been proposed as such mechanisms (Waage, 1979; Driessen *et al.*, 1995; van Alphen *et al.,* 2003).

The parasitoid *Ibalia leucospoides* Hochenwarth (Hymenoptera, Ibaliidae) is a solitary, koinobiont and nearly pro-ovigenic parasitoid of the woodwasp *Sirex noctilio* Boidin (Hymenoptera, Siricidae) (Spradbery, 1977; Madden, 1981). Woodwasps are primitive xylophagous insects that attack pine trees. Although *S. noctilio* is native to Mediterranean Europe, in the last century it has successfully invaded Australia, New Zealand, South Africa, South America, and more recently, North America (Hoebeke *et al.*, 2005). In most regions where *S. noctilio* has established, it has rapidly become a most important pest of pine tree forestation, due to its reported outbreak population dynamics, during which tree mortality can be severe (Corley *et al.*, 2007). *I. leucospoides* is one of several bio-control agents adopted for pest management purposes of woodwasp populations (Hurley *et al.*, 2007, Corley & Bruzzone 2009). The parasitoid, also native to Europe, was introduced into Australasia in the early sixties. Since then, it has established throughout the invasion range of *S. noctilio*, mostly through accidental introductions, together with its host (Madden, 1988). In field conditions, *I. leucospoides* may parasitize up to 40% of its hosts. Because of its applied importance, there is a growing research effort on this parasitoid. Most works have focused on its host location and patch use behaviour (Chrystal, 1930; Madden, 1968; Spradbery, 1970 a, b; Fernández-Arhex & Corley, 2005; 2010; Martinez *et al.*, 2006), but to date, no attempts have been made to evaluate its foraging behaviour in a multiple patch arrangement (but see Corley *et al.*, 2010).

In the field, trees attacked by *S. noctilio* are spatially aggregated and, during long-lasting endemic population phases, overall *S. noctilio* densities are typically low (Corley *et al.* 2007). However, during outbreak periods woodwasps can reach high densities and host distribution across the aggregate of attacked trees becomes more homogenous. *I. leucospoides* emerges within these clusters of attacked trees where patches (i.e. trees) can differ in host availability. In these contexts, *I. leucospoides* females should be able to choice patches and adjust their patch exploitation behaviour to the different levels of heterogeneity and availability of host.

I. leucospoides use chemical information derived from a fungal symbiont of *S. noctilio*, *Amylostereum areolatum,* as host locating cue (Madden, 1968; Spradberry, 1974). In addition, fungal volatiles may provide information on relative densities of hosts on the patch (Martinez *et al.*, 2006). The combination of fungal and host-derived chemical cues might be expected to provide a reliable and detectable source of information for host location, patch-quality classification and subsequent patch exploitation adaptation. Mainly, this odour combination could help to optimize patch choice decisions prior to patch exploitation.

All this led to the hypothesis that female parasitoids are able to discriminate patches with different host availability from a distance by using airborne cues. Once patch selection occurs, females are also capable to assess habitat profitability (e.g. in a multiple patch environment) and thus adjust their patch residence time on the current patch (maybe also using the volatiles cues). The aim of this study was to test: (i) whether *I. leucospoides* females are able to perceive differences in patch host availability from a distance by moving towards the best patch, as first choice (i.e.: no patch sampling process is involved); (ii) whether the surrounding context affects this choice as well as the patch time allocation; and finally, (iii) whether the perceived quality of a given patch is relative to the quality of the surrounding patches.

3. MATERIALS AND METHODS

3.1 *The study system*

Ibalia leucospoides Hochenwarth (Hymenoptera: Ibaliidae) is a solitary, pro-ovigenic parasitoid (females emerge with their entire or nearly entire egg complement), that attacks eggs and first-instar larvae of the pine woodwasp *Sirex noctilio* (Hymenoptera: Siricidae). The species was first obsreved in pine tree plantations in NW Patagonia in 1993, at the time woodwasps were first detected. Since, it has become established in most pine plantations in this region. The woodwasp *S. noctilio*, together with its eggs injects into the trunk tree, toxic mucus and spores of a symbiotic fungus, *Amylostereum areolatum.* The host larva develops inside trees feeding on wood decomposed by the fungus. It has been shown that the chemical information derived from the host fungal symbiont is used by parasitoids during host searching. In addition, these fungal volatiles may provide information on relative hosts availability and on the suitable development stage for host attacking by the parasitoids (Madden, 1968; Martinez *et al*., 2006; N. Jofré, unpub.data).

Female wasps used in this study were obtained from pine (*Pinus contorta*) logs collected in the field, at several heavily attacked plantations located in NW Patagonia (Argentina). Once felled, trees were cut into 1m-long logs and kept individually in locker-type cages under indoor conditions until insect emergence. Each morning, newly emerged insects were collected from the cages and immediately placed in individual plastic vials with food (see below in experimental design section).

3.2 *Experimental design*

In order to assess the capacity of *I. leucospoides* females to detect from a distance the best patch in an array and whether the patch residence time is proportional to host availability on it, as well as, to examine whether host spatial distribution and host availability (in the patch visited and in the surrounding patches) influence patch exploitation behaviour, female parasitoids were individually assigned at random to one of three different triple-patch environments (hereafter called habitats A, B, C). The settings differed in host numbers at the individual log level (i.e.: patch), in average environment host number (3 logs) as well as in the host distribution among patches (Table 1). Habitat A is characterized by low average host number for the setting as a whole and a heterogeneous host distribution among patches; habitat B by a high average host number and a homogeneous host distribution and habitat C, by high average environment host number and a heterogeneous host distribution among patches. The host patches chosen as first choice and the time allocated to each patch visited, were recorded as the response variables.

TABLE 1 Female parasitoid wasps were individually assigned to one of three different triple-patch environments (*habitats* A, B, C). The environments differ in individual patch hosts number, in average environment host number and host distribution among patches.

	n	Patch hosts number			Average environment host number	Host distribution among patches
		Low	Medium	High		
Habitat A	12	1	8	15	8	Heterogeneous
Habitat B	11	10	15	20	15	Homogeneous
Habitat C	13	2	15	28	15	Heterogeneous

Since the insects emergence season last only 2 months and have ready the experimental setup for each replica takes a prolonged time, the experiments were carried out in two different years during daytime hours (between 11:00 and 20:00 hours) and at 23 ± 1 ºC.

In order to create a symmetrical environment, three host patches were placed in the corners of an imaginary isosceles triangle (30cm on each side) within a wire mesh cage ($1m^3$). To obtain host patches we cut "clean" (i.e.: free from woodwasp attacks) pine logs, 0.8m in length, and left them to dry at ambient temperature for 10 days. This was done to ensure appropriate wood moisture conditions for woodwasp oviposition. Logs were then exposed to woodwasp females until the desired number of ovipositions was achieved and afterward were stored for another 10 days before the experiments. This ensured that host development and fungal growth were suitable and attractive to *I. leucospoides* females. The number of hosts per log is given in Table 1. The position of a given log within the array was set at random.

A female wasp was introduced at the exact centre of the arrangement, and after a 5 min-long adaptation period carried out inside a perforated plastic vial, it was gently released and the trial initiated. In each replicate of the experiment, both parasitoids and logs used were replaced. Wasps were observed continuously during an experiment. We recorded patch residence times (PRT) using a hand-held stopwatch. Although off-the-patch short excursions during a host patch visits are not common in *I. leucospoides*, we still arbitrarily defined the end of a patch visit when a wasp left the patch and remained off it for 20 min. The experiment was considered ended when, after having left a patch and landed elsewhere, wasps remained off any patch for 30 or more minutes.

Newly emerged parasitoid females were stored individually in plastic vials ($251cm^3$) for seven days in a common chamber free from host odours (temperature 23 ± 1ºC, 53% RH and 16:8 h light: dark cycle). All wasps had unlimited access to a diluted honey solution (30% honey-distilled water solution) since emergence and on the day prior to the experiment, food was removed and replaced with distilled water only.

3.3 *Data analysis*

The effect of patch host availability and the surrounding context on the first patch chosen were analyzed using three-way contingency tables. In these tables, frequencies were compared in a generalized linear model with a log link function and a Poisson distribution of errors. The initial model fitted to the data considered *patch host number* (high, medium and low; a three qualitative levels factor), the *habitats* (A, B, C; a three qualitative levels factor) and the *year* as explanatory variables. We were interested in testing for the presence of interactions starting with the most complex model (*n*= *patch host number* * *habitat* * *year*, where *n* refers to the number of wasps choosing a given hosts patch for the first time and the * symbol represents the sum of the influences of three-way interaction, two-way interactions and the three single variables). We were mainly interested in the term involving *patch host number* x *habitat* interaction. Non-significant interactions were excluded backwardly from the model starting with the highest-order interactions (Crawley, 2007; Sokal & Rohlf, 1995). We tested the effect of interaction removal by means of ANOVA.

The effects of host number within the patch and the habitat as a whole on patch time allocation to the first host patch chosen were analyzed using a generalized linear model assuming a Gamma distribution of residuals and an inverse link function. The *patch host number* (a quantitative variable, in this case, considering the number of hosts within the patches), the *habitats* (A, B, C; a three qualitative levels factor) and *year* were introduced in the model as explanatory variables. We used a backward procedure to remove non-significant factors and interactions between factors, and to select the final model. Residuals were examined to confirm that the final model accurately fitted the data. All data analyses were done using the R statistical environment (R Development Core Team, 2008).

4. RESULTS

4.1 Patch choice

In the analysis of the first patch chosen out of the three offered, we did not find a significant interaction between the three explanatory variables, *patch host number, habitat* and *year* ($G = 8.497$, df = 4, $p = 0.075$). The interaction between *patch host number* and *habitat* (A, B, C) was also non significant ($G = 6.842$, df = 4, $p = 0.144$). This shows that the choice by *I. leucospoides* females was independent of average environment host number and host distribution in the surrounding patches. However, *I. leucospoides* females were able to discriminate stimuli from patches with different host number from a distance, and showed a propensity to alight in and exploit the patch bearing the highest number of hosts ($G = 17.963$, df = 2, $p < 0.001$, Fig. 1).

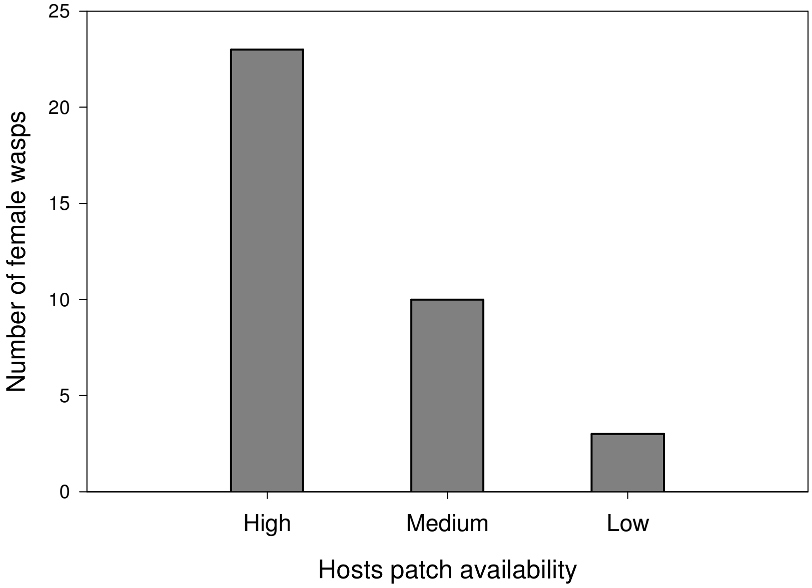


Figure 1. First host patch chosen by *Ibalia leucospoides* females among the three patches simultaneously offered as a function of patch host availability (high, intermediate, low).

4.2 The influence of patch host number and habitat as a whole on PRT

The time allocated to the exploitation of the first host patch visited was significantly affected by the interaction between host number in the patch and the habitat as a whole ($F_{(2,35)} = 9.45$; $p < 0.0001$). We observed a positive relationship between PRT and patch host number for habitat A and C (Fig. 2). However, for habitat B, where similar hosts number across patches is offered, the PRT- patch host number relationship was not as expected. This is because only one female wasp visited the patch containing ten hosts and she allocated a disproportionate amount of time to it compared with the time allocated by the other wasps to patches of comparable number of hosts. Anyway, in general terms, time spent on the first patch visited increased with host number in that patch ($F_{(1,35)} = 30.60$; $p < 0.0001$). After removing the aformentioned individual (who visited the patch of ten hosts) from the analysis, the interaction between host number in the patch and the habitat as a whole still being significant ($F_{(2,34)} = 3.89$; $p = 0.032$) and again, the time allocated to the first patch visited increased with host number in that patch ($F_{(1,34)} = 33.28$; $p < 0.0001$).

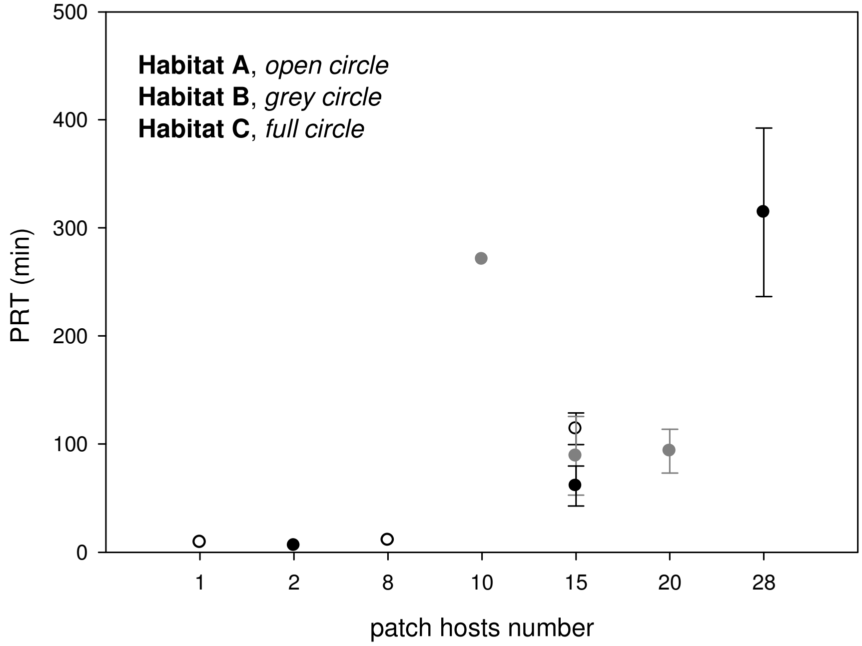


Figure 2. Patch residence times (PRT, mean ± SE) on the first host patch visited, as a function of patch host availability for *I. leucospoides* females experiencing different environmental conditions.

To further study the effects of host number in the environment and within the patch and the spatial host distribution across patches on patch time allocation, we analyzed differences in the time assigned to the patch holding fifteen hosts. This patch was included in the habitats A, B and C. We did not find any differences between habitats on the time spent on patches of fifteen hosts ($F_{(2,18)} = 0.983$; $p = 0.395$, Fig. 3). Only six out of thirty-six wasps visited more than one patch. The years in which the experiments were performed did not have a significant effect on any response variable analyzed.

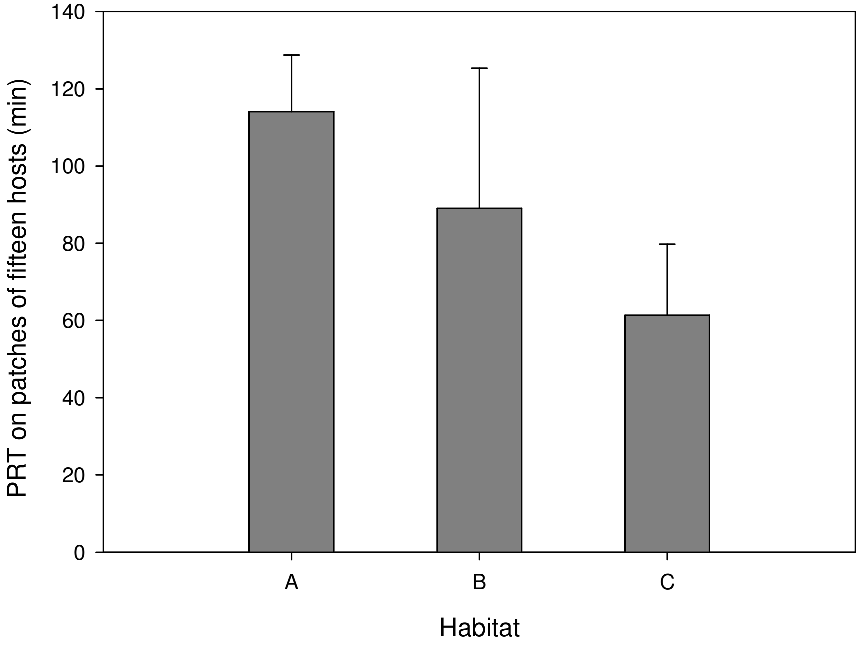


Figure 3. Patch residence times (PRT, mean ± SE) on patches of fifteen hosts when they were chosen as first option in systems A, B, or C.

5. DISCUSSION

Our results show that *I. leucospoides* females tend to alight in and exploit firstly, the logs holding the highest number of hosts. This is particularly remarkable considering that the first selection occurs from a distance. Once on the patch, the time allocated to the exploitation of the first host patch visited depends on host number in the patch and on the habitat setting. This finding suggests that the exploitation decisions are influenced by information obtained from the surrounding patches.

5.1 *Patch choice decisions*

The most striking result of our study is that *I. leucospoides* was capable of responding to differences in host patch quality from a distance. *I. leucospoides* seems to do this, using chemical cues derived from the host fungal symbiont (Madden, 1968; Martinez *et al*., 2006). While hosts could be avoiding information conveyance through their concealed development, fungus odours provides parasitoids with information on host.

Recall that feeding by immature stages of *S. noctilio* depend on *Amylostereum areolatum* decomposition of wood.

Choosing the best between the three nearby patches suggests, on the one hand, proportionality between odour concentration and host number. On the other hand, it requires at least a minimal capacity of appraising several patches in the local environment and the capability of detecting differences in volatile cues. It is generally assumed that animals need to sample first their environment (i.e.: make contact) in order to assess its profitability and make decisions (e.g.: Hubbard & Cook, 1978; Waage, 1979; Stephens & Krebs, 1986; Driessen *et al.,* 1995; Pierre *et al.*, 2003). However, our results suggest that *I. leucospoides* is able to take host searching decisions at a distance, by accurately assessing the initial density of host in patches, without the need of a sampling process. In addition, the assessment capacity in this parasitoid did not differ between habitats (i.e.: anywhere wasps be present they first select the best log), although we cannot rule out at this stage, that this is the consequence of a lack of power of our tests due to small sample sizes.

In *I. leucospoides*, the strategy of choosing and staying in the best patch might perform better than engaging in a process of sampling and continuous updating. In pine plantations *S. noctilio* densities and their spatial distribution among trees are highly variable, including large population outbreaks (Corley *et al.*, 2007). In these conditions, it is possible that a host infested tree, within the aggregate of attacked trees, bears several to hundred host larvae, hence surpassing the parasitoid egg complement. For parasitoids inhabiting highly variable environments, the ability to assess patch quality from a distance would be of value. Firstly, because it may result in a time saving strategy and secondly, because by an appropriate choice there is a chance to lay the entire egg load in a single high density hosts tree. In addition, by an accurate choice and by exploiting thoroughly first patch *I. leucospoides* may protect itself against a failure in finding another good hosts patch, mortality risk during travel and losses in term of their energy budget. This parasitoid species appears to be, hence, more tuned to the direct search than to the sampling process in the plantations environments.

5.2 *Patch residence time*

Under our experimental conditions, the first patch chosen is exploited according to the habitat. PRT increases with patch hosts number for heterogeneous habitats (A and C) but not for the homogeneous scenario (habitat B) where more similar host number across patches is offered to the foraging female parasitoid. In accordance with predictions of some theoretical studies (Mc Namara & Houston, 1985; 1987; Vos & Hemerik, 2003), the results of this survey suggest that wasps may use the information available from nearby patches to make patch leaving decisions. Additionally, the results qualitatively agreed with the general predictions of the Marginal Value Theorem in that patch residence times by parasitoids increases with increasing host number. The increased PRT could be a consequence of a density-dependence response of *I. leucospoides* to host chemical cues concentration.

While one of the study subjects was to examine whether *I. leucospoides* is capable to use habitat information to adjust their patch residence time on the current patch, we did not pretend to asses whether they behave optimally on the patch. Anyway, as aforementioned, our results show a general propensity to agree with foraging theory. The foraging theory predicts, that the time allocated to a patch of a given host density would depend upon the particular host environment. Thus, when habitat profitability is high, the residence times on a patch of particular host availability should decrease (Charnov, 1976). While we find that the number of hosts in the patch determines the time allocation to the first patch chosen in association with habitat characteristics, we do not observe a statistically significant habitat-dependent time allocation pattern to those patches bearing fifteen hosts. This apparent mismatch is probably a consequence of the low power of the statistical test (low sample size) and not of parasitoid behavioral rules. Note that parasitoids show a more variable response in patch of fifteen hosts when this patch is presented in the habitat B than when it is in habitats A and C. This could be the consequence of wasp exposure to homogeneous habitat where information on slight differences in the number of host per patch could be difficult to decode. As well, female wasps tend to spend more time on the patch with fifteen hosts, when this patch is within the poorer habitat (habitat A) than when it is in the richest one (habitat C).

In the experimental design used, female wasps experienced three different multiple patch environments in terms of hosts abundance and distribution. One environment represented a poor and heterogeneous habitat (habitat A), another one was rich and homogeneous (habitat B) and the last one mimicked a rich but heterogeneous scenario (habitat C). Including a *poor* environment with a *homogeneous* host distribution would have allowed a factorial design. However these characteristics are most difficult to achieve, given the fact that mean environment host number should be eight and host patches should bear different hosts numbers.

This study suggests the relevance of quantitative differences in volatiles cues in the process of distant patch selection. *I. leucospoides* showed a strong response to host abundance by choosing mainly the patches with the highest number of hosts. Wasps, once on patch, may well use or overlook the information about habitat characteristics. In this study individuals use habitat information but, whether this leads to an increase in reproductive success deserves further investigation. Anyway, it is worthwhile noting that female parasitoids emerge with almost all of their eggs completely mature and with the ability to respond to differences in host number on patches which likely allow this parasitoid to a rapid exploitation of the located host patches. This is of great importance due to might represent how well *I. leucospoides* match their foraging effort to host availability.

ACKNOWLEDGMENTS

We acknowledge financial support from ANPCyT, Argentina (PICT 0-1200/06), CNRS, France (PICS 4144) and INTA, Argentina.

6. REFERENCES

Bernstein, C., and Driessen, G. (1996). Patch-marking and optimal search patterns in the parasitoid *Venturia canescens*. *J. Anim. Ecol.* 65: 211–219.

Bernstein, C., Kacelnik, A. and Krebs, J.R. (1988) Individual decisions and the distribution of predators in a patchy environment. *J. Anim. Ecol.* 57, 1007–1026.

Castelo, M.K., Corley, J.C. and Desouhant, E. (2003) Conspecific avoidance during foraging in *Venturia canescens* (Hymenoptera: Ichneumonidae): the roles of host presence and conspecific densities. *J. Insect Behav*. 16: 307–318.

Charnov, E.L. (1976) Optimal foraging: the marginal value theorem. Theor Popul Biol 9:129–136.

Crawley, M.J. (2007) The R Book. John Wiley & Sons, Chichester, West Sussex, UK.

Chrystal, R.N. (1930) Studies of the *Sirex* parasites. Oxford Forestry Memories No 11. Oxford University Press, London,UK.

Corley, J.C., Villacide, J.M. and Bruzzone, O.A. (2007) Spatial dynamics of a *Sirex noctilio* woodwasp population within a pine plantation in Patagonia. *Ent. Exp. Appl.* 125: 231-236.

Corley, J.C., Villacide, J.M. and van Nouhuys, S. (2010) Patch time allocation by the parasitoid *Ibalia leucospoides* (Hymenoptera: Iballidea): the influence of con-specifics, host abundance and distance to the patch. *J. Insect Behav*. 23(6):431-440.

Corley, J.C. and Bruzzone, O.A. (2009) Delayed emergence and the success of parasitoids in biological control. *Biol. Control* 51: 471-474.

Driessen, G., Bernstein, C., van Alphen, J.J.M. and Kacelnik, A. (1995) A count-down mechanism for host search in the parasitoid Venturia canescens. *J. Anim. Ecol.* 64:117–125.

Fernández-Arhex, V. and Corley, J.C. (2005) The functional response of *Ibalia leucospoides* (Hymenoptera: Ibaliidae), a parasitoid of *Sirex noctilio* (Hymenoptera: Siricidae). *Biocont. Sc. Technol.* 15: 207-212.

Fernández-Arhex, V. and Corley, J.C. (2010) The effects of patch richness on con-specific interference in the parasitoid *Ibalia leucospoides* (Hymenoptera: Ibaliidae). *Insect Science* 17(4):379-385.

Geervliet, J.B.F., Ariens, S., Dicke, M. and Vet, L.E.M. (1998) Long-distance assessment of patch profitability through volatile infochemicals by the parasitoids *Cotesia glomerata* and *C. rubecula* (Hymenoptera: Braconidae). *Biol. Control* 11: 113–21.

Green, R.F (1984) Stopping rules for optimal foragers. *Am. Nat.* 123: 30-40.

Haccou, P., De Vlas, S.J., van Alphen, J.J.M., and Visser, M.E. (1991). Information processing by foragers: Effects of intra-patch experience on the leaving tendency of *Leptopilina heterotoma*. *J. Anim. Ecol.* 60: 93–106.

Hemerik, L., Driessen, G. and Haccou, P. (1993) Effects of intra-patch experiences on patch time, search time and searching efficiency of the parasitoid *Leptopilina clavipes*. *J. Anim. Ecol.* 62, 33-44.

Hoebeke, E.R., Haugen, D.A. and Haack, R.A. (2005) *Sirex noctilio*: Discovery of a Palearctic siricid woodwasp in New York. Newsletter of the Michigan Entomological Society 50: 24-25.

Hubbard, S.F. and Cook, R.M. (1978) Optimal foraging by parasitoid wasps. *J. Anim. Ecol.* 47, 593-604.

Hurley, B.P., Slippers, B. and Wingfield, M.J. (2007) A comparison of control results for the alien invasive woodwasp, *Sirex noctilio*, in the southern hemisphere. *Agr. Forest Entomol.* 9: 159–171.

Iwasa, Y., Higashi, M. and Yamamura, N. (1981) Prey distribution as a factor determining the choice of optimal foraging strategy. *Am. Nat.* 117:710–723.

Janssen, A., van Alphen, J.J.M., Sabelis, M.W. and Bakker, K. (1995a). Odour-mediated avoidance of competition in *Drosophila* parasitoids: The ghost of competition. *Oikos* 73: 356–366.

Janssen, A., van Alphen, J.J.M., Sabelis, M.W. and Bakker, K. (1995b). Specificity of odour mediated avoidance of competition in *Drosophila* parasitoids. *Behav. Ecol. Sociobiol.* 36: 229–235.

Li, C., Roitberg, B.D., Mackauer, M. (1997) Effect of contact kairomone and experience on initial giving-up time. *Entomol. Exp. Appl.* 84:101–104.

Liu, Q.Y., Thiel, A. and Hoffmeister, T.S. (2009) Odor-mediated patch choice in the parasitoid *Venturia canescens*: temporal decision dynamics. *Entomol. Exp. Appl.* 132: 110–117.

Madden, J.L. (1968) Behavioural responses of parasites to the symbiotic Fungus associated with *Sirex* noctilio F. *Nature* 218: 189-190.

Madden, J.L. (1981) Egg and larval development in the woodwasp *Sirex noctilio*. *Australian Journal of Zoology* 29: 493-506.

McNair J.M. (1982) Optimal giving-up time and the marginal value theorem. *Am. Nat.* 119:511–529.

Mc Namara, J.M. and Houston, A.I. (1985) Optimal foraging and learning. *J. Theor. Biol.*, 117, 231-249.

Mc Namara, J.M. and Houston, A.I. (1987) Memory and the efficient use of information. *J. Theor. Biol.*, 125, 385-395.

Martínez, A., Fernández-Arhex ,V. and Corley, J.C. (2006) Chemical information from the fungus *Amylostereum aerolatum* and host foraging behaviour in the parasitoid *Ibalia leucospoides*. *Physiol. Entomol.* 31: 1-5.

Murdoch, W.W. and Stewart-Oaten, A. (1989) Aggregation by parasitoids and predators: effects on equilibrium and stability. *Am. Nat.* 134:288-310.

Pierre, J.S., van Baaren, J. and Boivin, G. (2003). Patch leaving decision rules in parasitoids: Do they use sequential decisional sampling? *Behav. Ecol. Sociobiol.* 54: 147– 155.

R Development Core Team (2008) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL http://www.R-project.org.

Shaltiel, L. and Ayal, Y. (1998) The use of kairomones for foraging decisions by an aphid parasitoid in small host aggregations. *Ecol. Entomol.* 23:319–329.

Stephens, D.W. and Krebs, J.R. (1986) Foraging theory. Princeton University Press, Princeton.

Godfray, H.C.J. (1994) Parasitoids, Behavioral and Evolutionary Ecology, Princeton University Press.

Spradbery, J.P. (1970a) Host finding by *Rhyssa persuasoria*, an ichneumonid parasite of siricid woodwasps. *Anim. Behav.* 18: 103-114.

Spradbery, J.P. (1970b) The biology of *Ibalia drewseni* Borries, a parasite of siricid woodwasps. Proceedings of the Entomological Society London B 45: 104-113.

Spradbery, J.P. (1977) The oviposition biology of siricid woodwasps in Europe. *Ecol. Entomol.* 2: 225-230.

Tentelier, C., Wajnberg, E. and Fauvergue, X. (2005) Parasitoids use herbivore-induced information to adapt patch exploitation behaviour. *Ecol. Entomol*. 30, 739–744.

Tentelier, C., Desouhant, E. and Fauvergue, X. (2006) Habitat assessment by parasitoids: mechanisms for patch use behavior. *Behav. Ecol.* 17, 515–521.

Tentelier, C. and Fauvergue, X. (2007) Herbivore-induced plant volatiles as cues for habitat assessment by a foraging parasitoid. *J. Anim. Ecol.* 76:1-8.

Thiel A. and Hoffmeister T.S. (2006) Selective information use in parasitoid wasps. *Anim. Biol.* 56, 233-245.

Turlings, T.C.J. and Wäckers, F.L. (2004) Recruitment of predators and parasitoids by herbivore-injured plants. *Advances in Insect Chemical Ecology* (eds R.T. Cardé & J.G. Millar), pp. 21–75. Cambridge University Press, Cambridge.

van Alphen, J.J.M., Bernstein, C. and Driessen, G. (2003) Information acquisition and time allocation in insect parasitoids. *Trends Ecol. Evol.* 18:81–87.

Vet, L.E .M. and Dicke, M. (1992) Ecology of infochemical use by natural enemies in a tritrophic context. *Annual Review Entomol*. 37, 141 – 172.

Vos, M., Hemerik, L. and Vet, L.E.M. (1998) Patch exploitation by the parasitoids *Cotesia rubecula* and *Cotesia glomerata* in multi-patch environments with different host distributions. *J. Anim. Ecol.* 67, 774-783.

Vos, M. and Hemerik, L. (2003) Linking foraging behavior to lifetime reproductive success for an insect parasitoid: adaptation to host distributions. *Behav. Ecol.* 14: 236–245.

Waage, J.K. (1979) Foraging for patchily-distributed hosts by the parasitoid, Nemeritis canescens. *J. Anim. Ecol*. 48:353–371

Wajnberg, E. (2006) Time allocation strategies in insect parasitoids: from ultimate predictions to proximate behavioral mechanisms. *Behav. Ecol. Sociobiol*. 60, 589 – 611.

Wang, X.G. and Keller, M.A. (2004) Patch-time allocation by the parasitoid Diadegma semiclausum (Hymenoptera: Ichneumonidae). III. Effects of kairomone sources and previous parasitism. *J. Insect Behav.* 17:761–776.

SECTION 3
GENERAL DISCUSSION

1. Main results and general discussion

Throughout this thesis I have explored how natural selection shapes several life history traits and behaviours, according to the environmental characteristics in which an organism lives. This study framework leads to research into specifics hypotheses concerning reproductive, dispersal and foraging processes in parasitoids.

My main results show that the female parasitoid of *Ibalia leucospoides* emerges with a high ovigeny index. In line with this, *I. leucospoides* shows low dependency on adult female feeding, not only for egg maturation but also for survival and flight (see below). Regarding host foraging, female wasps show the ability to accurately assess differences in host patch quality from a distance, without the need of displaying a sampling process. Following, I expand and discuss these central results as well as the perspectives for future work it opens.

The majority of models developed to predict foraging behaviour (i.e. Sirot & Bernstein, 1996; Tenhumberg *et al.,* 2006, but see Bernstein & Jervis, 2008) and the incidence of time-limitation or egg-limitation (i.e. Rosenheim, 1996; Sevenster *et al.*, 1998, but see Ellers *et al.*, 2000) have mainly considered proovigenic parasitoids. However, empirical findings show that this reproductive strategy is very rare (< 2% of species, Jervis *et al.*, 2001), rendering many of the findings derived from the models, of limited generality. Although *Ibalia leucospoides* is not strictly proovigenic, this species has a high degree of eggs mature at emergence (OI = 0.77). This evidence enables us to place this species nearly the pro-ovigenic parasitoids and thus contrast our current findings with model predictions in order to improve the understanding of *I. leucospoides* life-history.

Corley *et al.* (2007) show that the host aggregated spatial distribution increases its aggregation with an increase in the number of attacked trees through time. *I. leucospoides* emerges within these clusters of attacked trees. Thus, as proovigenic parasitoid, this species has the ability to take advantage of high host encounter rates early in life, as females are ready to oviposit upon emergence. In accordance, I found that not only do *I. leucospoides* females emerge with almost all of their eggs completely mature, but that they also have an exceptional ability to detect and select the richest host patches within host patch aggregates. Therefore, this foraging mechanism plus the high ovigeny index would this parasitoid a rapid exploitation of host patches. Both the reproductive and foraging strategies of *I. leucospoides* may have evolved as a consequence of the variable abundance and distribution of *Sirex noctilio* in nature, due to their pulse-like outbreak dynamics.

The environmental stochasticity that female parasitoids face, implies *trade-off*s in the allocation of finite resources, which ultimately determine their life-history. Proovigeny is considered a more rigid reproductive strategy than synovigeny (Jervis *et al.*, 2001), being the former a one-time resource allocation strategy. Yet, proovigeny represents an adaptation to environments with low quality or scarce food availability (Boggs, 1997), as well as with significant stochasticity in host encounter (e.g. due to the distribution of host patches in space and the distribution of the number of hosts per parch, Ellers *et al.*, 2000, but see Ellers & Jervis; 2004).

In line with this, I found that *I. leucospoides* would emerge with sufficient teneral reserves to meet all their maintenance, reproductive and dispersal needs without feeding. Accordingly, nutrient reserves accumulated during larval development minimize the importance of adult female feeding. We observed that food supply increased longevity by 42% when it was provided *ad libitum*. This augmentation is not negligible but modest compared to other parasitoids (Siekmann *et al.*, 2001; Bezemer *et al.*, 2005; Desouhant *et al.*, 2005; Heping *et al.*, 2008; Lee & Heimpel; 2008). It must also be noted that an *ad libitum* regime is quite uncommon in field conditions but recurrent under laboratory setting (Siekmann *et al.*, 2001). When I attempted to mimic a more natural scenario providing females with a restricted food access, there were no major significant extensions in adult lifespan when comparing fed with starved wasps; regardless at which moment in the lifespan feeding occurs. This suggests that, when access to food is limited, due to either physiological or morphological constraints, wasps cannot acquire considerable quantities of nutrients, which result in different nutritional states. Therefore females may emerge with robust fat body reserves, enough to support maintenance function requirements without adult feeding. In order to add more information to these issues, we captured wasps in the wild and studied them with biochemical methods to seek for sugary food use during adult life. The partial results obtained suggest that sugary food acquisition is rare in *I. leucospoides* females.

Generally, for parasitoids, feeding is at the cost of time and energy investment, especially when hosts and food are found in different parts of the environment. *I. leucospoides* emerge within host patch aggregates inside pine forests, where the probability of finding food is low or else unavailable (personal observation). Sirot & Bernstein (1996) and Bernstein & Jervis (2008) models concerning food foraging in proovigenic parasitoids suggest that if food is scarce and not extremely needed, it should be more profitable not to search for it. *I. leucospoides* behaviour seems to be in accordance with this prediction in that this species is not likely to search for food. In addition, the search and utilization of metabolic resources observed in this species would fit well with proovigeny egg maturation strategy.

Furthermore, through laboratory experiments, I noted that egg maturation was not affected by adult female feeding. Apparently, wasps without feeding can deal with the potential catabolic drain that somatic maintenance exerts upon carried-over resources that fuel oogenesis; and therefore, stored reserves in a fat body (i.e. mainly glycogen and lipids) may still be allocated to mature the remaining eggs. In this way, I confirm that *I. leucospoides* has evolved low dependence on adult feeding for reproduction. This is represented by the high number of mature eggs at emergence and may be a consequence of the rareness of food presence or the limitation in the adult diet used (i.e. nectar or honey) of some nutrient type such as lipids and nitrogenous compounds.

Regarding the influence of environmental food availability on egg maturation strategies and resource allocation, Boggs & Ross (1993) pointed out the importance of taking into account the timing of resource depression relative to the specific developmental stage (i.e. larval vs. adult) and the actual state of the individual at a given moment (i.e. relative importance of the current capital vs. incoming resources). It is expected that the allocation to life-history traits under resource limitation, would be adjusted in evolutionary time, if the duration and intensity of resource depression is predictable. For example, when resource stress is of short duration with relation to the lifespan of an individual, resource depression may result in investment in survival at the expense of reproduction (i.e. eggs resorption); but when there is a long-term resource depression, the results may be the opposite, an investment in reproduction at the expense of survival.

Since the ovigeny index of *I. leucospoides* is high as compared to some other parasitoid species (Jervis *et al.*, 2001), this species has likely suffered long-term adult resource depression that has resulted in a greater investment in reproduction. Still, it can speculate that most likely, *I. leucospoides* females are not capable of completely discharging their egg complement over their adult life ($\approx$600 eggs). I draw this inference through taking into account several factors: the handling time that involved in each oviposition event (20-30 minutes approximately, personal observation), adult lifespan in the wild (10-15 days in the wild; Carvalho, 1993), potential mortality risks (i.e. due to weather, predation and/or capital reserves depletion), as well as low host density when the host is going through an endemic population phase. Nevertheless, females may have this overload of mature eggs to hold the chance of exploiting any opportunity to lay an unusual large number of eggs when woodwasp outbreaks occur. The benefits derived from the occasional match between egg load and host availability should offset the potential cost of an initial resource allocation to eggs and also the cost of reduced reproductive plasticity. It would be similar to winning the lottery.

Together with egg maturation, non reliance on feeding was reflected in our results about flight. Dispersal capacity was only dependent on the morphological characteristics of insects (size and wing loading). Larger females flew longer distances than smaller ones, probably due to the greater teneral reserves of the former ones. In addition, since there were no differences in flight parameters between fed and starved females, flight could be sustained by teneral reserves at least during one day of tethered flight display. I could not discern whether carbohydrates and/or lipids were used as primary flight fuel, the nutrient dynamics during flight was not measured here. Generally, both fat and carbohydrates stored in the fat body of insects are consumed during flights, and there can be a temporary variation in the use of fuel. That is, carbohydrates mainly fuel the earlier phases of flights, while there may be a switch from carbohydrates to fat oxidation during long lasting flights (Chapman, 1998). In our study, I could not rule out whether fed wasps use carbohydrates as the primary fuel, and unfed wasps a mixture of carbohydrates and lipids from stored reserves. Anyway, during trials no obvious differences in dispersal were noted.

If it is not feasible to obtain external resource input, internal resources may be the energy source for all demanding biological functions that share common resources, leading to *trade-offs*. An increase in resources allocated to one trait requires a decrease in resources allocated to another trait (the traditional "Y" model of resource allocation; van Noordwijk & de Jong, 1986). It still has to be investigated in *I. leucospoides* whether a potential *trade-offs* between longevity, oviposition activity, egg maturation and flight occur in the absence of adult food sources. Another interesting question is how a relative priority in resource allocation to competing functions is established under stressful conditions. Thus, a more thorough understanding of life-history variation among individuals requires a deeper study in energy budget dynamics. Only in this way, we shall be able to understand the physiology of internal resource allocation.

As above mentioned, various life-history traits of *I. leucospoides* are not affected by adult feeding. From an applied point of view, this finding can be very useful. It is a widely held view, that food supplementation in target areas and/or prior to parasitoid release should improve the impact on pest suppression (Jervis & Kidd, 1996; Lewis *et al.*, 1998; Bianchi *et al.*, 2006). It can be added, that the contribution of food supplementation to improve pest management may well be strongly reliant on the resource allocation pattern of the parasitoid species involved. One common characteristic of proovigenic parasitoids could be those individuals emerge with enough capital resources to meet all adult biological functions, even without feeding. If this is true, setting an effort to encourage adult feeding in the laboratory or in the field may be unwarranted.

Parasitoid fitness estimation can be carried out by measuring different individuals' life-history traits and by studying the contribution of each of these fitness components. It must be taken into account, firstly that not all life-traits actually constrain reproductive success. For example, in harsh environmental conditions – where host density can be very low and host distribution highly heterogeneous – fecundity may not be the most relevant fitness component when most of the individuals are mainly time-limited and die with eggs. However, dispersal and host finding abilities surely can be important fitness constraints (see below). It must also be considered that laboratory fitness measures such as longevity or fecundity can be non representative of true fitness in the field.

The lifetime reproductive success of *I. leucospoides* – surely prone to be a time-limited parasitoid – may possibly be constrained by a differential dispersal capability in relation to their hosts. In this thesis I show that larger parasitoid females fly longer distances than smaller ones probably due to their greater fat reserves. However, the potential advantage of the high dispersal capacity of this parasitoid is still does not match that of its host. *S. noctilio* can fly a maximum of nearly 50 km per day in flight mills (Bruzzone *et al.*, 2009) while *I. leucospoides* cannot exceed 28 km per day in similar conditions. In other words, and for the sake on managing invasive woodwaps populations, *I. leucospoides* may be unable to track the leading edge of the pest populations as has been recently shown for other natural enemies (Corley *et al.*, submitted). Still, further field studies are needed to actually learn whether *I. leucospoides* flight capacity acts as an obstacle for its reproductive success and thus constrains its ability to limit invasive woodwaps populations.

Dispersal strongly influences population dynamics and plays an important role in species interactions (Commins *et al.*, 1992; Hassell & May, 1973; Roland & Taylor, 1997; Briggs & Latto, 2000). Until now we had no information on flight capacity or dispersal in *I. leucospoides.* We believe that it is a relevant starting point to investigate the flight capacity of this wasp, even though we have no certainty yet if there is a direct link between "flight capacity" and "dispersal" in this species. Flying insects can disperse greater distances in the wild due to the use of wind currents as compared to their flight capacity measured in flight mills systems. Or, on the contrary, they could disperse less because there could be many attractive landing sites nearby. In any case, it remains of interest to investigate their flight capacity in order to know, among others things, whether *I. leucospoides* is potentially able to follow its host, and whether it is potentially capable of colonizing of new suitable habitats.

Host searching ability can also be an important limiting factor of lifetime reproductive success especially when habitat characteristics are adverse. While we lack precise knowledge of the conditions under which *I. leucospoides* evolved, it is likely that *S. noctilio* in their native habitat displays highly unpredictable population dynamics as currently observed in invaded, cultivated forests. In this thesis, I have given evidence that *I. leucospoides* is able to deal with several tasks during host foraging. Firstly, wasps were able to accurately assess differences in host patch quality from a distance without the need of a sampling process. Secondly, patch exploitation times depended both on the richness of the current patch visited and on the surrounding context. Hence, female parasitoids seem to have evolved, both a very sensitive ability to detect chemical cues specific to *S. noctilio* and a high responsiveness to host presence while a patch is being exploited. It is worthwhile noting that while this parasitoid accounts for a "rigid" egg maturation strategy by being proovigenic, skillfully finding the host can be translated in fitness gains.

Females, in the experimental study carried out here, were host deprived for seven days; however, they showed motivation for searching hosts. They made a real choice given they selected the richest patch and exploited it thoroughly despite the fact that there were alternative patches to go to. Females' possessed high number of mature eggs ready to use when 7 days old and egg resorption was not observed. All the above factors may strongly contribute to immediate host patches exploitation when appropriate circumstances are present. Again, the behavioural outcome may represent adaptations to the original host ecological traits (i.e. low host availability and stochastic distribution of host).

It is desirable that future efforts are addressed to improve our knowledge on those biological parameters related to both the parasitoid and the habitat in order to reconstruct the life-history of a nearly proovigenic parasitoid such as *I. leucospoides*. The research described here is a contribution to the understanding of the potential ecological conditions under which high ovigeny index is possibly a selected egg maturation strategy. Gaining an insight into how egg production strategy interacts with host foraging strategies, and how both of them affect the dynamics of the hosts-parasitoid interactions is called for topic. This is attractive not only from fundamental standpoint, but also from a practical point of view, and would help to deeply understand success and failure impact of parasitoids used in biocontrol programs.

Natural selection is expected to result in reproductive strategies that approach a quantitative match between egg load and host availability in the habitat. In line with this, natural selection may shape aspects of the life history of individuals, surely involving decision making processes in response to the ecological surroundings experienced by animals. The results of this thesis show how parasitoids might have evolved according the environment they inhabit. *I. leucospoides* may focus their foraging effort on host searching, by aiming at the site where reproductive opportunities probably match better with the number of eggs. That is, females display an efficient mechanism of patch choice resulting in the selection of patches bearing the highest numbers of hosts. In line, *I. leucospoides* displays a flight capacity non reliant on adult feeding but with the potential to move amongst trees. In turn, the robust capital reserves, this inferred through the negligible effects that food has on egg maturation, maintenance and flight, may allow parasitoids to search for hosts rather than food. Finally, a high degree of mature eggs occurs at female emergence. All these traits and mechanisms displayed by *I. leucospoides* females may well have evolved in response to habitat characteristics, including host and food abundance and distribution.

REFERENCE

Amat, I., Castelo, M., Desouhant, E. and Bernstein, C. (2006) The influence of temperature and host availability on the host exploitation strategies of sexual and asexual parasitic wasps of the same species. *Oecologia* 148: 153–161.

Baker, H.G. and Baker, I. (1983) A brief historical review of the chemistry of floral nectar. In: Bentley, B. and Elias, T. (eds.) *The Biology of Nectaries*. Columbia University Press, New York, pp. 126– 52.

Bernstein, C. and Jervis, M.A. (2008) Food-searching in parasitoids: the dilemma of choosing between 'immediate' or future fitness gains. *Behavioral Ecology of Insect Parasitoids: from theoretical approaches to field applications* (ed. by E. Wajnberg, C. Bernstein & J.J.M. Van Alphen), pp. 129-171. Blackwell Publishing, Oxford, U.K.

Bezemer, T.M., Harvey, J.A. and Mills, N.J. (2005) Influence of adult nutrition on the relationship between body size and reproductive parameters in a parasitoid wasp. *Ecol. Entomol.* 30: 571–80.

Boggs, C.L. (1981) Nutritional and life-history determinants of resource allocation in holometabolous insects. *Am. Nat.* 117: 692-709.

Boggs, C.L. and Ross, C.L. (1993) The effects of adult food limitation on life history traits in *Speyeria mormonia* (Lepidoptera: Nymphalidae). *Ecology* 74: 433-441.

Boggs, C.L. (1997) Reproductive allocation from reserve and income in butterfly species with differing adult diets. *Ecology* 78: 181-191.

Boggs, C.L and Freeman, K.D. (2005) Larval food limitation in butterflies: effects on adult resource allocation and fitness. *Oecologia* 144: 353-361.

Boivin, G., Fauvergue, X. and Wajnberg, E. (2004) Optimal patch residence time in egg parasitoids: innate versus learned estimate of patch quality. *Oecologia* 138: 640–647.

Briggs, C.J. and Latto, J. (2000) The effect of dispersal on the population dynamics of a gall-forming midge and its parasitoids. *J. Anim. Ecol.* 69: 96–105.

Bruzzone, O.A., Villacide, J.M., Bernstein, C. and Corley, J.C. (2009) Flight variability in the woodwasp *Sirex noctilio* (Hymenoptera: Siricidae): an analysis of flight data using wavelets. *J. Exp. Biol.* 212: 731-737.

Carvalho, A.G. (1993) Aspectos biológicos de *Ibalia leucospoides* (Hochenwarth) (Hymenoptera: Ibaliidae). *Conferencia Regional da vespa-da-madeira, Sirex noctilio, na América do Sul.*

Carter, C., Shafir, S., Yehonatan, L., Palmer, R.G. and Thornburg, R. (2006) A novel role for proline in plant floral nectars. *Naturwissenschaften.* 93: 72–79

Casas, J., Pincebourde, S., Mandon, N., Vannier, F., Poujol, R. and Giron, D. (2005) Lifetime nutrient dynamics reveal simultaneous capital and income breeding in a parasitoid. *Ecology.* 86: 545–54.

Chapman, R.F. (1998) *The Insects: Structure and Function*. Cambridge, UK: Cambridge Univ. Press.

Charnov, E.L. (1976) Optimal foraging: the marginal value theorem. *Theor. Popul. Biol.* 9: 129-136.

Chrystal, R.N. (1930) Studies of the Sirex parasites. Oxford Forestry Memories No 11. Oxford University Press, London, U.K.

Clark, C.W. and Mangel, M. (2000) Dynamic state variable models in ecology. Methods and applications. Oxford University Press.

Commins, H.N., Hassell, M. and May, R.M. (1992) The spatial dynamics of host-parasitoid systems. *J. Anim. Ecol.* 61: 735-748.

Corley, J.C. (1999). Host density signal and aggregation in an insect parasitoid. *Netherlands Journal of Zoology.* 49: 97-109.

Corley, J.C., Capurro, A.F. and Bernstein, C. (2004) Prolonged diapause and the stability of host-parasitoid interaction. *Theor. Popul. Biol.* 65: 193-203.

Corley, J.C., Villacide, J.M. and Bruzzone, O.A. (2007) Spatial dynamics of a *Sirex noctilio* woodwasp population within a pine plantation in Patagonia. *Ent. Exp. Appl.* 125: 231-236.

Corley, J.C. and Bruzzone, O.A. (2009) Delayed emergence and the success of parasitoids in biological control. *Biol. Control*. 51: 471-474.

Corley, J.C., Villacide, J.M. and Liebhold, A. Can entomophagous nematodes slow the spread of invasive insect populations? Biological Invasions (submitted)

Cortesero, A.M., Stapel, J.O. and Lewis, W.J. (2000) Understanding and manipulating plant attributes to enhance biological control. *Biol. Control* 17: 35-49.

Cook, R.M. and Hubbard, S.F. (1977) Adaptive searching strategies in insect parasites. *J. Anim. Ecol.* 46: 115-125.

Coutts, M.P. and Dolezal, J.E. (1969) Emplacement of fungal spores by the woodwasp, *Sirex noctilio*, during oviposition. *Forest Science* 15: 412-416.

Desouhant, E., Driessen, G., Lapchin, L., Wielaard, S. and Bernstein, C. (2003) Dispersal between host populations in field conditions: navigation rules in the parasitoid *Venturia canescens. Eco.l Entomol.* 28: 257-267.

Desouhant, E., Driessen, G., Amat, I. and Bernstein, C. (2005) Host and food searching in a parasitoid wasp *Venturia canescens:* a trade-off between current and future reproduction. *Anim. Behav.* 70: 145-52.

Desouhant, E., Lucchetta, P., Giron, D. and Bernstein, C. (2010) Feeding activity pattern in a parasitic wasp when foraging in the field. *Ecol. Res.* 25: 419-428.

Driessen, G. and Hemerik, L. (1992) The time and egg budget of *Leptopilina clavipes*, a parasitoid of larval *Drosophila. Ecol. Entomol.* 17: 17-27.

Driessen, G., Bernstein, C., van Alphen, J.J.M. and Kacelnik, A. (1995) A count-down mechanism for host search in the parasitoid *Venturia canescens*. *J. Anim. Ecol.* 64:117-125.

Driessen, G. and Bernstein, C. (1999) Patch departure mechanisms and optimal exploitation in an insect parasitoid. *J. Anim. Ecol*. 68: 445-459.

Dudley, R. and Srygley, R.B. (2008) Airspeed adjustment and lipid reserves in migratory Neotropical butterflies. *Func. Ecol.* 22: 264-270.

Ellers, J. (1996) Fat and eggs: an alternative method to measure the trade-off between survival and reproduction in insect parasitoids. *Netherlands Journal of Zoology* 46: 227-35.

Ellers, J. and van Alphen, J.J.M. (1997) Life history evolution in *Asobara tabida*: Plasticity in allocation of fat reserves to survival and reproduction. *J. Evol. Biol.* 10: 771-85.

Ellers, J., Sevenster, J.G. and van Alphen, J.J.M. (1998) A field study of size–fitness relationships in the parasitoid *Asobara tabida. J. Anim. Ecol.* 67: 318–324.

Ellers, J., Sevenster, J.G. and Driessen, G. (2000) Egg load evolution in parasitoids. *Am. Nat.* 156: 650–665.

Ellers, J. and Jervis, M.A. (2003) Body size and the timing of egg production in parasitoid wasps. *Oikos* 102:164-72.

Elliott, C.G., and Evenden, M.L. (2009) Factors influencing flight potential of *Choristoneura conflictana. Physiol. Entomol.* 34: 71-78.

Fernández-Arhex, V. and Corley, J.C. (2005) The functional response of *Ibalia leucospoides* (Hymenoptera: Ibaliidae), a parasitoid of *Sirex noctilio* (Hymenoptera: Siricidae). *Biocontrol Science and Technology.* 15: 207-212.

Fernández-Arhex, V. and Corley, J.C. (2009) The effects of patch richness on con-specific interference in the parasitoid *Ibalia leucospoides* (Hymenoptera, Ibaliidae). *Insect Science*. 17: 379-385.

Flanders, S.E. (1950) Regulation of ovulation and egg disposal in the parasitic Hymenoptera. *Canadian Entomologist.* 82: 134-140.

Frechette, B., Dixon, A., Alauzet, C. and Hemptinne, J.L. (2004) Age and experience influence patch assessment for oviposition by an insect predator. *Ecol. Entomol.* 29:578-583.

Geervliet, J.B.F., Ariens, S., Dicke, M. and Vet, L.E.M. (1998) Long-distance assessment of patch profitability through volatile infochemicals by the parasitoids *Cotesia glomerata* and *C. rubecula* (Hymenoptera: Braconidae). *Biol. Control* 11: 113-21.

Giron, D., Rivero, A., Mandon, N., Darrouzet, E. and Casas, J. (2002) The physiology of host feeding in parasitic wasps: implications for survival. *Func. Ecol.* 16:750-57.

Giron, D. and Casas, J. (2003) Lipogenesis in an adult parasitic wasp. *J. Insect Physiol.* 49:141-47.

Giron, D., Pincebourde, S. and Casas, J. (2004) Lifetime gains of host-feeding in a synovigenic parasitic wasp. *Physiol. Entomol.* 29: 436-42.

Godfray, H.C.J. (1994) *Parasitoids. Behaviour and Evolutionary Ecology*. Princeton University Press, Princeton.

Green, R.F (1984) Stopping rules for optimal foragers. *Am. Nat.* 123: 30-40.

Goubault, M., Outreman, Y., Poinsot, D. and Cortesero, A.M. (2005) Patch exploitation strategies of parasitic wasps under intraspecific competition. *Behav. Ecol.* 16:693-701.

Hassell, M.P. and May, R.M. (1973) Stability in insect host–parasite models. *J. Anim. Ecol.* 42: 693-736.

Hassell, M.P., Comins, H.N. and May, R.M. (1991) Spatial structure and chaos in insect population dynamics. *Nature.* 353: 255–258.

Hassell, M.P. and Waage, J.K. (1984) Host-parasitoid population interactions. *Annual Review Entomol.* 29: 89-114.

Hassell, M.P. (2000) Host-parasitoid population dynamics. *J. Anim. Ecol*. 69: 543-566.

Haugen, D.A. and Underdown, M.G. (1993) Reduced parasitism of *Sirex noctilio* in radiata pines inoculated with the nematode *Beddingia siricidicola* during 1974 – 1989. *Australian Forestry.* 56: 45-48.

Haugen , D.A., Bedding, R.A., Underdown, M.G. and Neumann, F.G. (1990) National strategy for control of *Sirex noctilio* in Australia . *Australian Forest Grower.* 13, 8.

Heimpel, G.E. and Collier, T.R. (1996) The evolution of host-feeding behaviour in insect parasitoids. *Biological Reviews.* 71: 373-400.

Heimpel, G.E., Rosenheim, J.A. and Kattari, D. (1997) Adult feeding and lifetime reproductive success in the parasitoid *Aphytis melinus*. *Ent. Exp. Appl.* 83:305–315.

Hoebeke, E.R., Haugen, D.A. and Haack, R.A. (2005) *Sirex noctilio*: Discovery of a Palearctic siricid woodwasp in New York. *Newsletter of the Michigan Entomological Society.* 50: 24-25.

Hurley, B.P., Slippers, B. and Wingfield, M.J. (2007) A comparison of control results for the alien invasive woodwasp, *Sirex noctilio*, in the southern hemisphere. *Agr. Forest Entomol.* 9: 159-171.

Hubbard, S.F. and Cook, R.M. (1978) Optimal foraging by parasitoid wasps. *J. Anim. Ecol.* 47: 593-604.

Houston, A., Clark, C., McNamara, J. and Mangel, M. (1988) Dynamic models in behavioural and evolutionary ecology. *Nature*. 332: 29-34.

Hougardy, E., Bezemer, T.M. and Mills, N.J. (2005) Effects of host deprivation and egg expenditure on the reproductive capacity of *Mastrus ridibundus*, an introduced parasitoid for the biological control of codling moth in California. *Biol. Control.* 33: 96–106.

Hougardy, E. and Mills, N.J. (2006) The influence of host deprivation and egg expenditure on the rate of dispersal of a parasitoid following field release. *Biol. Control.* 37: 206–13.

Hougardy, E. and Mills, N.J. (2007) Influence of host deprivation and egg expenditure on the patch and host-finding behavior of the parasitoid wasp *Mastrus ridibundus*. *J Insect Behav.* 20: 229-46.

Iede, E.T., Penteado, S.R. and Bisol, J.C. (1988) *Primeiro registro de ataque de Sirex noctilio em Pinus taeda no brasil* . *Circular técnica* . EMBRAPA – CNPF, Curitiva, Brasil.

Iwasa, Y., Higashi, M. and Yamamura, N. (1981) Prey distribution as a factor determining the choice of optimal foraging strategy. *Am. Nat.* 117:710-723.

Jervis, M.A. and Kidd, N.A.C. (1986) Host-feeding strategies in hymenopteran parasitoids. *Biological Reviews.* 61: 395–434.

Jervis, M.A., Heimpel, G.E., Ferns, P.N., Harvey, J.A. and Kidd, N.A.C. (2001) Life-history strategies in parasitoid wasps: a comparative analysis of 'ovigeny'. *J. Anim. Ecol.* 70: 442-458.

Jervis, M.A., Ferns, P.N. and Heimpel, G.E. (2003). Body size and the timing of egg production in parasitoid wasps: a comparative analysis. *Funct. Ecol.* 17:375–83.

Jervis, M.A. and Ferns, P.N. (2004) The timing of egg maturation in insects: ovigeny index and initial egg load as measures of fitness and of resource allocation. *Oikos.* 107: 449-460.

Jervis, M.A., Boggs, C.L. and Ferns, P.N. (2005) Egg maturation strategy and its associated trade-offs: a synthesis focusing on Lepidoptera. *Ecol. Entomol.* 30: 359-75.

Jervis, M.A. and Heimpel, G.E. (2007) Phytophaghy. *Insects As Natural Enemies: A Practical Perspective* (ed. by M.A. Jervis), pp 543. Springer Publishing, The Netherlands.

Jervis, M.A., Ellers, J. and Harvey, J.A. (2008) Resource Acquisition, Allocation, and Utilization in Parasitoid Reproductive Strategies. *Annual Review Entomol.* 53: 361-385.

Johnson, C.G. (1969) *Migration and Dispersal of Insects by Flight.* Methuen, U.K.

Kaeslin, M., Pfister-Wilhelm, R. and Lanzrein, B. (2005) Influence of the parasitoid *Chelonus inanitus* and its polydnavirus on host nutritional physiology and implications for parasitoid development. *J. Insect Physiol.* 51:1330-39.

Kidd, N.A.C. and Jervis, M.A. (2007) Population Dynamics. *Insects As Natural Enemies: A Practical Perspective* (ed. by M.A. Jervis), pp 515. Springer Publishing, The Netherlands.

Krivan, V. and Sirot, E. (1997) Searching for food or hosts: The influence of parasitoids behavior on host-parasitoid dynamics. *Theor. Popul. Biol.* 51: 201-9.

Lee, J.C. and Heimpel, G.E. (2008) Floral resources impact longevity and oviposition rate of a parasitoid in the field. *J. Anim. Ecol.* 77: 565-572.

Lewis, W.J., Stapel, J.O., Cortesero, A.M. and Takasu, K. (1998) Understanding How Parasitoids balance food and host needs: importance to biological control. *Biol. Control.* 11: 175-183.

Li, C., Roitberg, B.D. and Mackauer, M. (1997) Effect of contact kairomone and experience on initial giving-up time. *Entomol. Exp. Appl.* 84:101-104.

Liebhold, A.M. and Tobin, P.C. (2008) Population ecology of insect invasions and their management. *Annu Rev. Entomol.* 53: 387-408.

Madden, J.L. (1968) Behavioural responses of parasites to the symbiotic Fungus associated with Sirex noctilio F. *Nature.* 218: 189-190.

Madden, J.L. (1974) Oviposition behaviour of the woodwasp *Sirex noctilio* F. *Australian Journal of Zoology* 22: 341-351.

Madden, J.L. (1981) Egg and larval development in the woodwasp *Sirex noctilio*. *Australian Journal of Zoology,* 29: 493-506.

Madden, J.L. (1988) *Sirex* in Australasia. Dynamics of forest insect populations: patterns, causes and implications (ed. By A.A. Berryman), pp. 407-427. Plenum Press.

Martínez, A., Fernández-Arhex, V. and Corley, J.C. (2006) Chemical information from the fungus *Amylostereum aerolatum* and host foraging behaviour in the parasitoid *Ibalia leucospoides. Physio.l Entomol.* 31: 1-5.

McArthur R.H. and Pianka, E.R. (1966) An optimal use of a patchy environment. *Am. Nat.* 100: 603-609.

McNair J.M. (1982) Optimal giving-up time and the marginal value theorem. *Am. Nat.* 119:511-529.

McNamara, J.M. and Houston, A.I. (1990) State-dependent ideal free distribution. *Evol. Ecol.* 9: 298-311.

Mills, N.J. and Wajnberg, E. (2008) Optimal foraging behavior and efficient biological control methods. *Behavioral Ecology of Insect Parasitoids: from theoretical approaches to field applications* (ed. by E. Wajnberg, C. Bernstein & J.J.M. Van Alphen), pp. 3-30. Blackwell Publishing, Oxford, U.K.

Murdoch, W.W., Briggs, C.J., Nisbet, R.M., Gurney, W.S.C. and Stewart-Oaten, A. (1992) Aggregation and stability in metapopulation models. *Am. Nat.* 140: 41-58.

Nicolson, S.W. and Thornburg, RW. (2007) *Nectar chemistry. In: Nicolson SW, Nepi M, Pacini W, editors. Nectaries and nectar. Dordrecht: Springer-Verlag;* p. 215-264.

Nonacs, P. (2001) State dependent behavior and the Marginal Value Theorem. *Behav Ecol* 12:71-83.

Olson, D.M., Fadamiro, H., Lundgren, J.G. and Heimpel, G.E. (2000) Effects of sugar feeding on carbohydrate and lipid metabolism in a parasitoid wasp. *Physiol. Entomol.* 25:17–26.

O´Brien, D.M. (1999) Fuel use in flight and its dependence on nectar feeding in the hawkmoth *amphion floridensis. J. Exp. Biol.* 202: 441-451.

Pelosse, P. (2008) Rôle des compromis évolutifs dans la spécialisation et la coexistence d’espèces compétitrices. Développements théoriques et expérimentaux chez les insectes parasitoïdes. PhD thesis, Lyon 1 University, France.

Pennacchio, F. and Strand, M.R. (2006) Evolution of developmental strategies in parasitic Hymenoptera. *Annual Review Entomol.* 51: 233–58.

Pierre, J. S., van Baaren, J., and Boivin, G. (2003). Patch leaving decision rules in parasitoids: Do they use sequential decisional sampling? *Behav. Ecol. Sociobiol.* 54: 147-155.

Rivero, A. and Casas, J. (1999) Incorporating physiology into parasitoid behavioural ecology: the allocation of nutritional resources. *Res. Popul. Ecol.* 41: 39-45.

Rivero, A., Giron, D. and Casas, J. (2001) Lifetime allocation of juvenile and adult nutritional resources to egg production in a holometabolous insect. *Proceedings of the Royal Society, Biological Sciences,* 268: 1231-1238.

Rivero, A. and West, S.A. (2005) The costs and benefits of host feeding in parasitoids. *Anim. Behav*. 69:1293–301.

R Development Core Team (2008) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL http://www.R-project.org.

Roland, J. and Taylor, P.D. (1997) Insect parasitoid species respond to forest structure at different spatial scales. *Nature*. 386: 710-713.

Rosenheim, J.A. (1996) An evolutionary argument for egg limitation. *Evolution*. 50: 2089-2094.

Schmale, I., Wäckers, F.L., Cardona, C. and Dorn, S. (2001) Control potential of three hymenopteran parasitoid species against the bean weevil in stored beans: the effect of adult parasitoid nutrition on longevity and progeny production. *Biol. Control.* 21: 134-139.

Sevenster, J.G., Ellers, J. and Driessen, G. (1998) An evolutionary argument for time limitation. *Evolution*. 52: 1241-1244.

Siekmann, G., Tenhumberg, B. and Keller, M.A. (2001) Feeding and survival in parasitic wasps: sugar concentration and timing matter. *Oikos*. 95: 425-430.

Sirot, E. and Bernstein, C. (1996) Time sharing between host searching and food searching in solitary parasitoids: State dependent optimal strategies. *Behav. Ecol.* 7: 189-94.

Sisterton, M.S. and Averill, A.L. (2002) Costs and benefits of food foraging for a braconid parasitoid. *J. Insect Behav.* 15: 571–88.

Spradbery, J.P. (1970a) Host finding by *Rhyssa persuasoria*, an ichneumonid parasite of siricid woodwasps. *Anim. Behav.* 18: 103-114.

Spradbery, J.P. (1970b) The biology of *Ibalia drewseni* Borries, a parasite of siricid woodwasps. *Proceedings of the Entomological Society London B,* 45, 104-113.

Spradbery, J.P. (1977) The oviposition biology of siricid woodwasps in Europe. *Ecol. Entomol.* 2: 225-230.

Stearns, S. C. (1989) Trade-Offs in Life-History Evolution. *Functional Ecology* 3: 259-268.

Stephens, D.W. and Krebs, J.R. (1986) Foraging theory. Princeton University Press, Princeton

Strand, M.R. and Casas, J. (2008) Parasitoid and host nutritional physiology in behavioral ecology. *Behavioral Ecology of Insect Parasitoids: from theoretical approaches to field applications* (ed. by E. Wajnberg, C. Bernstein & J.J.M. Van Alphen), pp. 113-128. Blackwell Publishing, Oxford, U.K.

Takasu, K. and Lewis, W.J. (1995) Importance of adult food sources to host searching of the larval parasitoid *Microplitis croceipes*. *Biol. Control.* 5: 25-30.

Tenhumberg, B., Siekmann, G. and Keller, M.A. (2006) Optimal time allocation in parasitic wasps searching for hosts and food. *Oikos.* 113: 121-131.

Tentelier, C., Desouhant, E. and Fauvergue, X. (2006) Habitat assessment by parasitoids: mechanisms for patch use behavior. *Behav. Ecol.* 17: 515–521.

Thiel, A and Hoffmeister, T.S. (2004) Knowing your habitat: linking patch-encounter rate and patch exploitation in parasitoids. *Behav. Ecol.* 15:419-425.

Thiel, A., Driessen, G. and Hoffmeister, T.S. (2006) Different habitats, different habits? Response to foraging information in the parasitic wasp *Venturia canescens*. *Behav Ecol Sociobiol*. 59: 614–623.

Tylianakis, J.M., Didham, R.K. and Wratten, S.D. (2004) Improved fitness of aphid parasitoids receiving resource subsidies. *Ecology.* 85: 658–66.

Turlings, T.C.J. and Wäckers, F.L. (2004) Recruitment of predators and parasitoids by herbivore-injured plants. *Advances in Insect Chemical Ecology* (eds R.T. Cardé & J.G. Millar), pp. 21–75. Cambridge University Press, Cambridge.

van Alphen, J.J.M., Bernstein, C. and Driessen, G- (2003) Information acquisition and time allocation in insect parasitoids. *Trends Ecol. Evol.* 18:81-87.

van Noordwijk, A.J. and de Jong, G. (1986) Acquistion and allocation of resources: their influence on variation in life history tactics. *Am. Nat.* 128:137-42.

Villacide, J.M. and Corley, J.C. (2008) Parasitism and dispersal potential of *Sirex noctilio*: implications for biological control. *Agr. Forest Entomol.* 10: 341-34.

Waage, J.K. (1979) Foraging for patchily-distributed hosts by the parasitoid *Nemeritis canescens*. *J. Anim Ecol*. 48: 353-371.

Wajnberg, E. (2006) Time allocation strategies in insect parasitoids: from ultimate predictions to proximate behavioral mechanisms. *Behav. Ecol. Sociobiol.* 60: 589-611 .

Wäckers, F.L. (1994) The effect of food deprivation on the innate visual and olfactory preferences in the parasitoid *Cotesia rubecula*. *J. Insect Physiol.* 40: 641-9.

Wäckers, F.L. (2004) Assessing the suitability of flowering herbs as parasitoid food sources: Flower attractiveness and nectar accessibility. *Biol. Control* 29: 307-14.

Wanner, H., Gu, H. and Dorn, S. (2006) Nutritional value of floral nectar sources for flight in the parasitoid wasp, *Cotesia glomerata*. *Physiol. Entomol.* 31:127-133.

Weisser, W.,Volkl, W. and Hassell, M. P. (1997) The importance of adverse weather conditions for behaviour and population ecology of an aphid parasitoid*.* *J. Anim. Ecol.* 66: 386-400.

Zera, A.J. and Harshman, L.G. (2001) The physiology of life history trade-offs in animals. *Annu. Rev. Ecol. Syst.* 32: 95-126.