Host-related fitness trade-offs in a presumed generalist parasitoid, *Diaeretiella rapae* (Hymenoptera: Aphidiidae)

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Abstract. 1. Host ranges of parasitoid wasps are mediated by behavioural responses to hosts and their environment (infectivity), and development in hosts (virulence). Determinants of host range were measured in *Diaeretiella rapae* (Hymenoptera: Aphidiidae), which has been described as a generalist that attacks more than 60 species.

2. In northern Colorado, this wasp mainly attacks two hosts: cabbage aphid (*Brevicoryne brassicae*) and Russian wheat aphid (*Diuraphis noxia*). Here, laboratory experiments are described in which *D. rapae* originating from these two hosts were offered several hosts for oviposition. Both infectivity and virulence were measured.

3. Infectivity included host acceptance and handling time, while virulence was measured as productivity (number of progeny), survival of immatures within hosts, development time, and sex ratio.

4. Wasps had higher productivity and survival when attacking 'home' hosts than 'alternate' hosts, and trade-offs were found by quantitative genetic analyses to be genetically determined. Sex ratio and development times also showed trade-offs, but mainly related to the host environment in which females were reared.

5. In previous genetic studies in northern Colorado, populations were genetically subdivided on the scale of 1 km. The fitness differences described here could be strong enough to create populations adapted to different hosts, but it appears that gene flow is sufficient to prevent formation of separate lineages on the two hosts.

6. Rather than being a generalist with a broad host range, *D. rapae* is a serial specialist, attacking particular hosts according to availability in different seasons or in different geographical areas.

Key words. Aphid parasitoid, generalist, genetic trade-off, host range, local adaptation, quantitative genetics, reaction norms, specialist.

Introduction

For insect parasitoids, which consume their insect hosts during larval development, the question of host specificity is pervasive. Host ranges of parasitoids can affect biological control efforts and populations of non-target hosts after introduction (McEvoy, 1996; Strand & Obrycki, 1996; Henneman & Memmot, 2001; Symondson *et al.*, 2002; Louda *et al.*, 2003), and host ranges are influenced by localised genetic variation and whether natural selection leads to specialisation on particular hosts (Dres & Mallet, 2002; Lajeunesse & Forbes, 2002). For instance, local adaptation and limited gene flow could lead to trade-offs, compromising the ability to attack some hosts while adapting to

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others. Even with gene flow between populations, locally adapted populations could persist if the trade-offs are strong enough or if hosts rapidly evolve defences that parasitoids must overcome via new genetic variation brought in by gene flow (van Zandt & Mopper, 1998; Rausher, 2001; Gandon & Michalakis, 2002). Evolution towards specialisation on a single host will be even more rapid if fitness trade-offs favour the evolution of host preferences and host-associated mating preferences (Jaenike & Holt, 1991; Feder & Filchak, 1999; Via & Hawthorne, 2002). Thus, genetic differences between sub-populations may arise in parasitoids that exploit numerous host species in patchy environments (Fellowes & Kraaijeveld, 1998; Dupas *et al.*, 2003), a pattern already reported for insect herbivores that use several host-plant species (Boecklin & Mopper, 1998; Strauss & Karban, 1998).

When describing the behavioural and physiological traits affecting parasitoid attack, useful parallels are found in the description of infectivity and virulence of parasites and pathogens (Dybdahl & Storfer, 2003). For parasitoids, infectivity entails searching and acceptance of hosts, which is mediated by chemical signals from the insect hosts and host plants (semiochemicals), physical features of the plants the insect hosts feed upon, and a combination of the two (Godfray, 1994; Mackauer *et al.*, 1996; Vet *et al.*, 2002). Additionally, many parasitoids exhibit host preferences based upon larval conditioning and/or associative learning of semiochemicals from the host and host environment (van Emden *et al.*, 1996). Thus, female parasitoids often prefer the same host species from which they emerged or most recently encountered, rather than showing innate preference for a particular host species (Chow & Mackauer, 1992).

Parasitoid virulence is influenced by the internal environment of the host and entails successful development and performance (sex ratio and survival of immatures) on each host species. Host quality can vary considerably, especially for endoparasitoids living in close physiological contact within the host (Henter & Via, 1995; Kraaijeveld *et al.*, 2002; Dupas *et al.*, 2003; Hufbauer, 2002). Host resistance and parasitoid virulence function via genetic, biochemical, and immunological traits, and may include genetic elements (e.g. from polydnaviruses) incorporated into wasp genomes that aid in overcoming defences of the hosts' immune system (Strand & Pech, 1995). Resistance in some host species is influenced by bacterial symbionts living within the hosts (Oliver *et al.*, 2003; Ferrari *et al.*, 2004). The full spectrum of co-evolutionary interactions between parasitoids and hosts includes both virulence (wasps' ability to develop within hosts) and resistance (hosts' ability to disrupt parasitoid development). The focus of this study was one parasitoid species that attacks several aphid host species. Questions related to the hosts' view of parasitoid–host coevolution, and differential resistance within host species, are left to future studies.

Here are reported a series of laboratory experiments that examine infectivity and virulence of the parasitoid *Diaeretiella rapae* M'Intosh (Hymenoptera: Aphidiidae),

which has a worldwide distribution and is considered a generalist parasitoid of aphids (Baer *et al.*, 2004). First, traits that modulate infectivity and virulence on several aphid host species were experimentally tested. Second, host-specific fitness was tested in a series of laboratory transplant experiments, using wasps collected from field populations in northern Colorado, where *D. rapae* commonly attack both cabbage aphid, *Brevicoryne brassicae*, a worldwide pest of crucifer crops, and the cereal-feeding Russian wheat aphid (RWA), *Diuraphis noxia*. The fitness trade-offs described here suggest that local populations of *D. rapae* are adapted to either cabbage aphid or RWA, and that the adaptation works mainly through virulence rather than infectivity.

Materials and methods

Parasitoids, hosts, infectivity, and virulence

Diaeretiella rapae is considered a generalist endoparasitoid of aphids. The worldwide host list includes more than 60 aphid species, but only five or six are commonly attacked (Mackauer & Starý, 1967; Némec & Starý, 1994; Pike *et al.*, 1999). A ubiquitous host for *D. rapae* is the cabbage aphid, *B. brassicae*, which spread throughout the world from its presumed origin in central Europe. Despite a strong attraction to semiochemicals from crucifer plants (Vaughn *et al.*, 1996), *D. rapae* is not obligate on crucifer-feeding aphids and also parasitises cereal-feeding aphids like RWA. The distribution of RWA expanded in the past 30 years, moving from the western Mediterranean into South Africa and South America, then into North America in 1986 (Halbert & Stoetzel, 1998). Although *D. rapae* was already established in North America, biological control efforts begun in the 1980s by the U.S. Department of Agriculture and several state agencies included collection of *D. rapae* from the Mediterranean region, the Middle East, and Asia. Several hundred thousand wasps were subsequently released in the wheat-growing regions of western North America (Hopper *et al.*, 1998; Prokrym *et al.*, 1998), resulting in establishment and spread of new populations of *D. rapae* (Mohamed *et al.* 2000; Brewer *et al.*, 2001; Burd *et al.*, 2001). In northern Colorado, cabbage and cereal crops (wheat and barley) are grown in close proximity, so that *D. rapae* females have opportunities parasitise both RWA and cabbage aphid.

The life history of *D. rapae* is typical of aphid parasitoids. Adult females oviposit single eggs into the bodies of aphid hosts. Parasitised hosts become 'mummies' consisting of the hardened exoskeleton of the aphids; parasitoid larvae pupate inside and emerge as adults. Under laboratory conditions, egg-to-adult development ranges from 9 to 15 days. Adult females live for 10–15 days and have lifetime fecundity as great as several hundred offspring (Hafez, 1961; Reed *et al.*, 1992).

Although the traits measured differed slightly between the experiments described here, infectivity was measured as host acceptance and handling time, while virulence was measured as secondary sex ratio, development time of immatures, productivity, and progeny survival. Productivity was the number of adult wasps emerging from parasitised hosts, which is the product of fecundity and survival. Sex ratio also may be considered a part of infectivity, as the sex of progeny in Hymenoptera is determined by female decisions to fertilise eggs at the time of oviposition (Godfray, 1994). However, sex ratio was included as part of virulence because secondary sex ratio was counted among wasps that successfully emerged, which is the product of female behaviour and differential survival of male and female immatures. Further, a sex ratio skew toward females could lead to higher rates of parasitism in subsequent generations.

Infectivity or virulence?

In these experiments, the influence of infectivity and virulence on performance of wasps attacking different aphid host species was examined. Experience with the host environment during emergence is known to influence host preference in some parasitoids (Mackauer *et al.*, 1996; van Emden *et al.*, 1996). Therefore, both acceptance of aphid species that females previously experienced and progeny survival in the host of origin were tested.

In May 2000, a *D. rapae* colony was established in the laboratory for use in these experiments. The colony originated from RWA in Kazakhstan, and had been reared for more than 50 generations on RWA at the Colorado Department of Agriculture Insectary at Palisade, Colorado (Lester & Holtzer, 2002). In all experiments, female wasps were tested individually 24–32 h after emergence, mating, and feeding on a 10% honey solution.

(i) Experience with hosts. Two tests were conducted in which females were reared in one host but offered different hosts for oviposition. In the first rearing test, wasps were reared for a single generation on one of three hosts: RWA on wheat, and cabbage aphid or green peach aphid (*Myzus persicae*) on cabbage seedlings. Individual females were introduced into the centre of circular arenas (inverted Petri dishes, 5 cm diameter, 1 cm high) containing 10 second- and third-instar aphids of one of the three species on a filter paper disk. Oviposition strikes were counted until 10 min had passed or until five aphids had been stung. Each female was tested once, sample size for each combination of rearing host and test host ranged from 13 to 20 individuals. Females were scored as either accepting or rejecting the hosts, and data were fitted to a log-linear model with factors 'natal host' and 'host offered' using SPSS (SPSS Inc., 233 S. Wacker Drive, Chicago, Illinois.).

In a second rearing test that included host-plant cues, females were reared for a single generation on RWA or cabbage aphids, mummies were removed 2 days before emergence and held in vials for emergence. Females were placed individually into small cages (10 cm by 10 cm, 15 cm high) that contained either cabbage seedlings with cabbage aphid, or wheat seedlings with RWA (approximately 50 aphids in each cage). The measure of overall performance was the number of mummies after 10 days ($n = 10$ per treatment). Plants were grown in 10-cm pots in sterile potting soil.

(ii) Progeny survival within hosts. This experiment measured survival of wasp larvae within RWA, cabbage aphid, or green peach aphids. Aphids were removed from host plants with a fine paintbrush, confined with a female under a clear gelatin capsule until the parasitoid was observed to strike, and then returned to the host plant. Each wasp stung three to five aphids of one host species ($n = 18$–33 for each treatment combination). To control for mortality caused by handling, control aphids were handled but not parasitised, and their survival was recorded 3 days later. Parasitised RWA were reared on barley seedlings at the rate of five aphids per seedling. Parasitised cabbage aphid and green peach aphid were reared six at a time on potted cabbage seedlings. Aphid and host plants were kept in environmental cabinets at 22 °C, 60% RH. Under these conditions, *D. rapae* eggs hatch by the third day.

To measure survival of *D. rapae* immatures (eggs, larvae, and pupae) 3 and 6 days after stinging, sub-samples of hosts were dissected under a binocular microscope at 40× magnification. A third group of stung hosts was retained until 10 days after stinging when wasps mummify their hosts. Survival in the three aphid hosts was analysed initially in SPSS by a log-linear model, with factors 'host' and 'larval stage'. Survival differences between hosts at each developmental stage were compared by pairwise Fisher's exact tests, with critical values adjusted for multiple tests by the Dunn–Šidak correction (Sokal & Rohlf, 1995).

Transplant experiments

To examine whether multiple host use in *D. rapae* includes genetic trade-offs in performance on different hosts, two separate laboratory transplant experiments were conducted using field populations from RWA and cabbage aphids. The wasp populations were from an irrigated organic farm 35 km north of Fort Collins, Colorado (Grant Family Farms, formerly Piedmont Farms) where wheat fields infested with RWA and crucifer fields infested with cabbage aphid were found within 1 km of each other. Different fields were sampled in 1996 and 2001.

(i) Transplant experiment 1996. In this experiment, infectivity and virulence were confounded because behaviour of individual females was not observed. The goal was to measure overall performance of wasps originating from cabbage aphid and RWA in three aphid host environments in the laboratory: the original ('home'), the alternative host ('alternate'), and a third host ('novel'), represented by *M. persicae* (Fig. 1a). Laboratory cultures of RWA and cabbage aphid originated from several fields
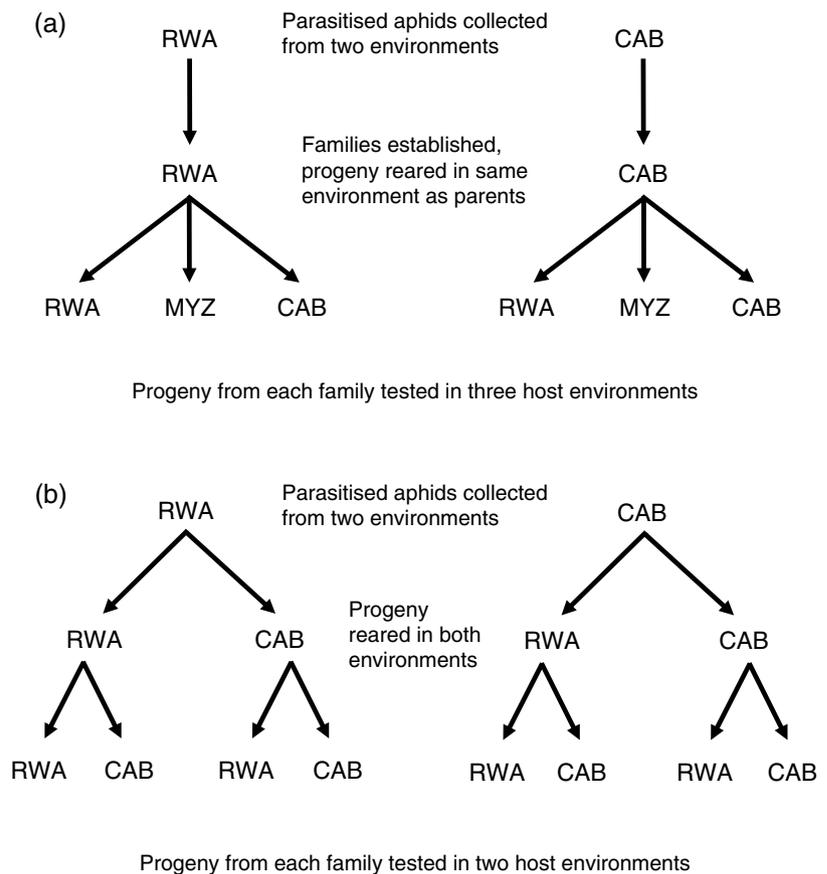
Fig. 1. Experimental design for transplant experiments in (a) 1996 and (b) 2001. Host designations: CAB, cabbage aphid (*Brevicoryne brassicae*); RWA, Russian wheat aphid (*Diuraphis noxia*); MYZ, green peach aphid (*Myzus persicae*).

in northern Colorado and were reared on potted wheat (TAM 107) and Brussels sprouts respectively. The third host, *M. persicae*, was found on pepper plants at a local nursery and was reared on potted peppers. Experiments were conducted in environment-controlled cabinets (22 °C, 60% RH).

Fields were sampled along transects, by searching plants for mummified aphids at 3-m intervals, then gently brushing a single mummy from each infested plant into a gelatin capsule. This procedure ensured that a large portion of each field was sampled while minimising sampling of related wasps. Within 2 h of emergence, males and females from each host environment were individually paired in 5-cm diameter Petri dishes for 24 h to mate. To establish full-sibling families, mated females were placed individually for 8 h in cages containing aphid-infested plants (60 aphids per plant) corresponding to their 'home' host. In this way 10 full-sib families were created from both cabbage aphid and RWA.

To test performance on each host, nine female offspring from each family were mated to unrelated males within 24 h of emergence. These females were released in individual cages with aphid-infested plants (30 aphids per plant); each family had three daughters tested on each of the 'home', 'alternate', or 'novel' hosts. After 3 h, females were removed and tibia length (third leg on the right side)

of each was measured to account for the effects of female body size on fitness. Productivity was counted as the number of second-generation offspring that emerged; sex ratio was the proportion of males among offspring and was arcsine-transformed for analysis. Development time (days from time of oviposition) and body size of second-generation progeny were also measured.

(ii) Transplant experiment 2001. In this experiment, both infectivity (host acceptance, handling time) and virulence (sex ratio, development time, productivity, and progeny survival) were examined. A half-sibling mating design was implemented to separate additive genetic from environmental effects (Fig. 1b, see Rossiter, 1998). These experiments required two levels of between-host manipulation: wasps from both RWA and cabbage aphid populations were reared on both hosts in the laboratory, and then tested on both hosts. The complication was necessary to cleanly separate genetic differences between populations from the effects of conditioning on females while they develop and emerge from hosts (cf. Mackauer *et al.*, 1996; van Emden *et al.*, 1996).

To begin, *D. rapae* mummies were collected from RWA and cabbage aphid in fields at the same organic farm described above. Collections were made as before, mummies were held individually in gelatin capsules, and only a single mummy from each infested leaf was taken. Newly

emerged males (sires) were isolated with three females (dams) that emerged on the same day and held in a 5-cm diameter Petri dish with honey for 24 h to mate. In this way families of half-sibling were created that shared fathers but had different mothers (Fig. 1b)

For laboratory rearing, field-collected females were placed individually into Petri dishes with RWA or cabbage aphid for 3 h. Immediately afterward, each female was placed into a fresh Petri dish with the alternate host for another 3 h. Parasitised aphids were transferred to either Brussels sprout or wheat seedlings, by allowing aphids to move off cut leaves. When mummies developed 9 days later, they were individually isolated in gelatin capsules and held until adult emergence.

Infectivity (host acceptance, handling time) was measured in four of the females from each family and from each host: two females were tested on RWA and two on cabbage aphid (Fig. 1b). All females were inexperienced at the time of testing. Females were placed individually in 15 cm diameter Petri dishes with one of the two hosts, chosen at random, and observed for 15 min, when handling times of the first host and number of hosts stung were recorded. Handling time was measured to the nearest second as the time from when the wasp first curled her abdomen into the strike position after encountering a host until she left the host. Because wasps often required several attempts to pierce the cuticle of cabbage aphid, multiple strikes on the same host were included in handling time if the wasp did not leave the host between strikes. After the observation period, females were placed in Petri dishes with honey and held overnight. Hosts were transferred to caged plants and held for 9 days, when mummies were isolated in gelatin capsules for wasp emergence.

Parasitoid virulence was measured as progeny survival (adult progeny divided by the number of aphids that had been stung in behavioural trails), development time, sex ratio among progeny, and overall productivity. To measure productivity, sex ratio, and development time, females were tested the day following behavioural observations by placing them into Petri dishes with one of the hosts for 3 h, then switching to fresh Petri dishes with the other host for another 3 h. The order of offering cabbage aphid or RWA was alternated between females from the same family. After 9 days on host plants, mummies were collected and held until adult emergence, when productivity, development time, and sex ratio were scored.

All experiments and rearing were under broad-spectrum light at 22 °C, 60% RH. The laboratory culture of RWA was a mixture of aphids from the Colorado Department of Agriculture insectary at Palisade, Colorado and several fields in northern Colorado, and was maintained on wheat (Tam 107). Cabbage aphids were reared on Brussels sprouts seedlings, and the laboratory colony originated from a vegetable garden in Salt Lake City, Utah, a laboratory colony from the University of Kentucky, and several fields in northern Colorado.

(iii) Data analysis. Nested ANOVA with Type III sums of squares was implemented in SYSTAT (Wilkinson, 1992) in 1996 and SPSS in 2001. In the 1996 experiment, main effects were fixed (population origin, laboratory host environment) or random (family), and the analyses included female body size as a covariate to correct for potential maternal effects arising from differences in body size. For the 2001 experiment, main effects were fixed (population origin, initial host environment, host environment of progeny) or random (family). ANOVA included interactions between host population origin, families, and host environments, which measure trade-offs in performance between hosts. Effect sizes (SS_{effect}/SS_{error}) were estimated for statistically significant main effects and interactions to determine relative influence of each (Rosenthal & Rosnow, 1985).

To calculate narrow-sense heritabilities from the half-sibling analysis in 2001, separate ANOVA of sires, and of dams nested within sires, were performed using replicates in which the initial rearing host was the same as the original host, using methods in Lynch and Walsh (1998). Under the haplo-diploid inheritance of Hymenoptera, variances among males (sires) provide estimates of additive genetic variance ($\sigma^2_{sire} = \frac{1}{2}V_A$), while variance among females (dams) may also include dominance and maternal effects ($\sigma^2_{dams(sire)} = 1/4V_A + \frac{1}{2}V_D + V_M$) (Killick, 1971). Variances were estimated by restricted maximum likelihood (REML) in SPSS, and heritability was estimated by sire variance and by sire plus dam variances (assuming that V_D and V_M are minor). Standard errors were calculated using methods for unbalanced designs (Lynch & Walsh, 1998).

Results

Infectivity or virulence?

(i) Experience. In the insectary population, which had been maintained on RWA for >50 generations, all experiments showed that wasps favoured RWA for oviposition despite exposure to or rearing on other hosts. In the first rearing experiment (Fig. 2a), wasps reared on RWA accepted the highest proportion of hosts ($G_2 = 9.95$, $P = 0.007$) and all females showed discrimination against green peach aphid (host offered: $G_2 = 8.72$, $P = 0.013$). Females did not, however, show a predisposition to accept the host in which they developed, as the interaction between rearing host and host offered was not significant ($G_4 = 2.14$, $P = 0.71$). In the second rearing experiment that included both hosts and host-plant cues (Fig. 2b), the greatest number of mummies were recovered from the RWA host environment ($F_{1,36} = 6.2$, $P < 0.02$), whether females were reared on RWA or cabbage aphid ($F_{1,36} = 0.36$, $P > 0.50$).

(ii) Progeny survival. Survival of immatures differed within the three host species (host × stage interaction: $G_4 = 13.83$, $P = 0.008$), with the highest survival in RWA. To determine where differences lay, survival in each host was compared at each time point (Fig. 3). Three days after stinging, fewer larvae were found in green peach
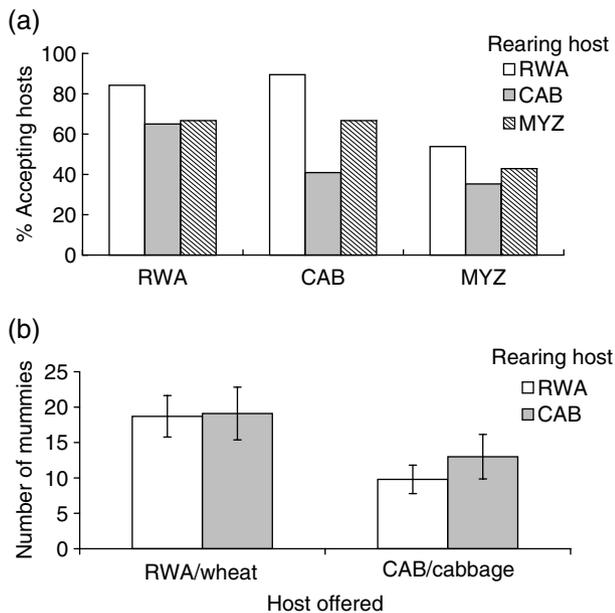
Fig. 2. Effect of experience and rearing on different hosts on oviposition and productivity of *Diaeretiella rapae* females from a laboratory colony originating from Russian wheat aphid (RWA). (a) The percentage of each host species accepted for oviposition by *D. rapae* females that had been reared on RWA, cabbage aphid (CAB), or green peach aphid (MYZ). (b) The number of mummies recovered from plants with RWA or cabbage aphid, after females had been reared on RWA or cabbage aphid.

aphid than cabbage aphid. Survival in cabbage aphid and RWA was not statistically different, nor was survival in RWA and green peach aphid different. Six days after stinging, survival in RWA was higher than green peach aphid. Survival in cabbage aphid and green peach aphid was not statistically different, nor did survival differ between RWA and cabbage aphid. By the mummy stage, differential survival between hosts increased; mummification was similarly low in cabbage aphid and green peach aphid, while RWA aphids had more
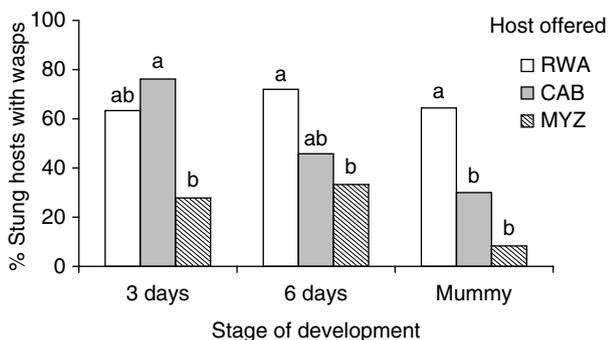

Fig. 3. Percentage of stung aphids found to contain a developing parasitoid immature after 3 days, 6 days, and after 10 days when hosts were mummified. Bars with the same letters were not statistically different at the $P < 0.05$ level.

mummies that either of the other hosts. Mortality of the three aphid host species did not differ because of aphids being handled during the experiments or because of stinging by D. rapae (both $G_2 > 0.25$, $P > 0.61$).

Transplant experiment 1996

Wasps had the highest productivity on cabbage aphid, and overall performance differed between families (Table 1). Host-related trade-offs were found in both productivity and sex ratio: wasps originating from cabbage aphid produced more offspring on the 'home' cabbage aphid than either the 'alternate' RWA or 'novel' green peach aphid (Fig. 4a). Wasps originating from RWA had similar productivity on RWA and cabbage aphid, but low productivity on the 'novel' green peach aphid host. Sex ratios were most female biased on each 'home' host (Fig. 4b). Body size of ovipositing females had no effect on either productivity or sex ratio (Table 1).

Development of progeny was faster when wasps were reared on their 'home' host than on the 'alternate' or 'novel' hosts (Fig. 5a), and the trade-off (population origin × host interaction) had a large effect (Table 1). Development times were correlated with maternal body size; the offspring of larger mothers developed faster ($r = 0.74$, $P < 0.05$), but this was a relatively small effect.

Overall, body size of progeny was greatest for wasps emerging from cabbage aphid (Fig. 5b, Table 1). The population origin × host environment interaction was also significant, as wasps developing in their 'home' host were relatively larger. Effect sizes of host, and population origin × host interactions were similar.

Transplant experiment 2001

In this experiment, both infectivity (host acceptance, handling time) and virulence (sex ratio, development time, survival of immatures, and productivity) were considered. Overall, genetic trade-offs and significant heritabilities for virulence traits were found, but infectivity was mostly influenced by the host environment. In most cases, interactions that included genetic effects were of the same size or larger than environmental effects (Tables 2 and 3).

(i) Infectivity. Host acceptance and handling time of hosts showed no trade-offs between females that were reared on RWA or cabbage aphid (Table 2). Host acceptance differed between the cabbage aphid and RWA populations, and females originating from cabbage aphid had highest host acceptance overall. However, acceptance of RWA (mean: 25 aphids stung in 15 min) was higher than acceptance of cabbage aphid (14 hosts) for both populations. Host acceptance differed between families, indicating genetic variability for the trait.

The only factor that influenced handling time was the host species (Table 2). Handling time for oviposition was longer for cabbage aphid (7 s) than for RWA (2 s). This

Table 1. Transplant experiment 1996: mixed-model ANOVA of productivity and sex ratio of females offered Russian wheat aphid, cabbage aphid, or green peach aphid MYZ, and development time and size of their progeny. Based upon a split-plot design (see Fig. 4), fixed factors: Population origin, Host environment; random factor: Family. Effect sizes were calculated as SS_{effect}/SS_{error} (Rosenthal & Rosnow, 1985).

Source	Productivity			Sex ratio			Development time			Body size		
	d.f.	Mean square	Effect size	d.f.	Mean square	Effect size	d.f.	Mean square	Effect size	d.f.	Mean square	Effect size
Population origin (A)	1	11.3		1	0.008		1	20.6		1	0.01	
Family within population origin (B)	9	5.8*	0.018	9	0.023		9	5.6		9	0.01	
Host offered during testing (C)	2	9.3*	0.059	2	0.194**	0.075	2	49.9***	0.048	2	1.44***	0.0004
A × C	2	10.7*	0.067	2	0.379***	0.149	2	34.1***	0.033	2	1.09***	0.0003
B × C	18	2.9		18	0.010		18	2.1		18	0.05	
Female body size	1	3.4		1	0.006		1	7.6*	0.018	1	0.02	
Error	112	2.8		112	0.046		864	2.4		864	8.67	

$*P < 0.05$, $**P < 0.01$, $***P < 0.001$, F-ratios for tests are: $F_A = MS_A/MS_B$, $F_B = MS_B/MS_{Error}$, $F_C = MS_C/MS_{B\times C}$, $F_{A\times C} = MS_{A\times C}/MS_{B\times C}$, $F_{B\times C} = MS_{B\times C}/MS_{Error}$.

was true for wasps originating from RWA or cabbage aphid, and whether wasps were reared on RWA or cabbage aphid.

(ii) Virulence. Overall productivity did not differ between wasps from cabbage aphid and RWA populations (Table 4). Nonetheless, performance differed
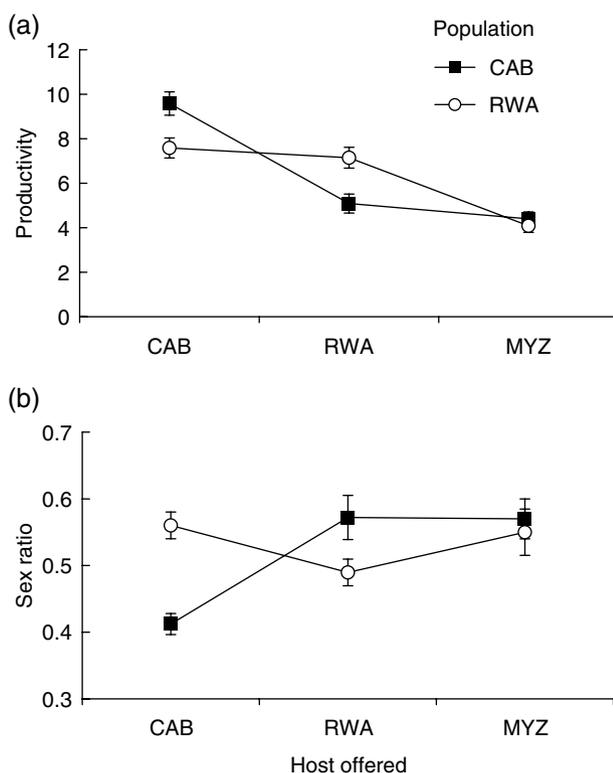
between families, and trade-offs were found for wasps attacking the 'home' vs. 'alternate' host (Fig. 6a; Table 3: A × D interaction). Wasps from RWA showed much higher productivity on RWA than cabbage aphid, while females from the cabbage aphid population showed similar productivity on both hosts. Further, productivity differed between families, and families varied in the extent to which the host of rearing influenced productivity [Table 3: B × C(A) interaction].

Progeny survival was similar between wasps from the RWA and cabbage populations, and was similar when

Fig. 4. Transplant experiment 1996: productivity and sex ratio (proportion males ± SE) of laboratory-reared females from two populations [cabbage aphid (CAB) and Russian wheat aphid (RWA)], tested in three hosts. See Table 1 for statistical analysis. MYZ, green peach aphid.
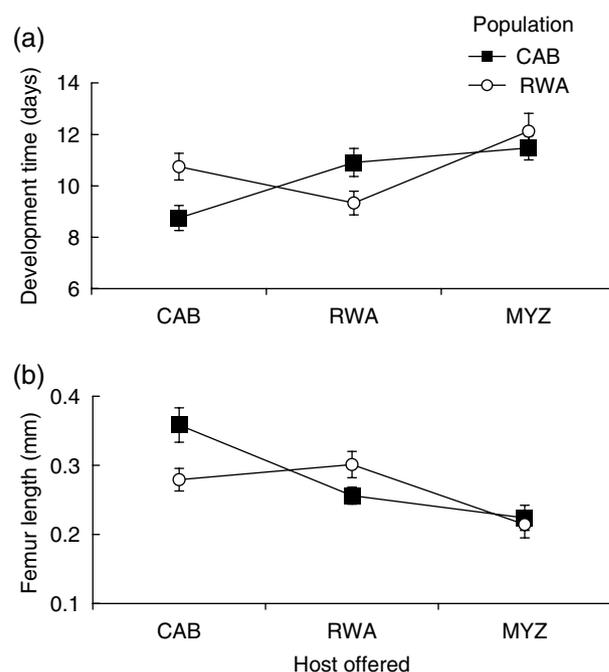

Fig. 5. Transplant experiment 1996: development time and body size (± SE) of progeny reared in three hosts. See Table 1 for statistical analysis. CAB, cabbage aphid; RWA, Russian wheat aphid; MYZ, green peach aphid.

Table 2. Transplant experiment 2001: mixed-model ANOVA of host acceptance and host handling time, based upon a split-plot design. Fixed factors: Population origin (Russian wheat aphid or cabbage aphid), Host environment during development, Host environment during testing; random factor: Family.

Source	Host acceptance			Handling time		
	d.f.	Mean square	Effect size	d.f.	Mean square	Effect size
Population origin (A)	1	493.0*	0.25	1	3.0	
Family within population origin (B)	14	103.2**	0.72	14	16.2	
Host during rearing (C)	1	99.1		1	26.8	
Host during testing (D)	1	292.7		1	670.7*	1.12
A × C	1	172.6		1	14.3	
A × D	1	120.6		1	0.6	
B × C(A)	14	38.0		14	20.7	
B × D(A)	14	48.1		14	13.5	
C × D	1	40.9		1	23.6	
A × C × D	1	167.0*	0.08	1	44.8	
B × C × D(A)	14	26.4		14	17.5	
Error	53	37.8		53	11.3	

*$P < 0.05$, **$P < 0.01$, ***$P < 0.001$, F-ratios for tests are: $F_A = MS_A/MS_B$, $F_B = MS_B/MS_{Error}$, $F_C = MS_C/MS_{AxC}$, $F_D = MS_D/MS_{AxD}$, $F_{A×C} = MS_{A×C}/MS_{B×C}$, $F_{A×D} = MS_{A×D}/MS_{B×D}$, $F_{B×C} = MS_{B×C}/MS_{Error}$, $F_{B×D} = MS_{B×D}/MS_{Error}$, $F_{C×D} = MS_{C×D}/MS_{A×C×D}$, $F_{A×C×D} = MS_{A×C×D}/MS_{B×C×D}$, $F_{B×C×D} = MS_{B×C×D}/MS_{Error}$.

females were reared on RWA or cabbage aphid (Table 3). However, the trade-off in survival between the population of origin and the larval environment of wasps was clear (A × D interaction). Wasps from both the cabbage aphid and RWA fields survived best in the 'home' host, regardless of the host in which they were reared (Fig. 6b). Again, survival differed between families, and survival within some families differed between hosts [Table 3: B × C(A) and B × D(A) interactions].

There was little variation in development time. Nonetheless, development time differed between families and development time within some families differed on the two hosts [Table 3: B × C(A) interaction].

Overall, sex ratio was male biased in this laboratory environment, but sex ratios differed between the two populations (Fig. 6c). Sex ratio also differed between families, again indicating genetic variability for the trait (Table 3). Wasps tended to produce a more female-biased sex ratio in the host in which they developed. However, this did not occur in all families, as the effect was significant only for interactions that included families [Table 3: B × C(A), B × C × D(A) interactions]. In these experiments,

Table 3. Transplant experiment 2001: mixed-model ANOVA of development time of females, their productivity, survival of their progeny, and sex ratio of their progeny, based on a split-plot design.

Source	Productivity			Survival of immatures			Development time			Sex ratio		
	d.f.	Mean square	Effect size	d.f.	Mean square	Effect size	d.f.	Mean square (× 100)	Effect size	d.f.	Mean square	Effect size
Population origin (A)	1	351.6		1	6.7		1	2.5		1	0.64***	0.48
Family within population origin (B)	14	89.4***	0.51	14	4.2***	1.27	14	1.8**	0.18	14	0.02*	0.18
Host during rearing (C)	1	0.001		1	0.13		1	4.3		1	0.03	
Host during testing (D)	1	581.2		1	12.5					1	0.004	
A × C	1	19.6		1	0.05		1	1.0		1	0.02	
A × D	1	261.8***	0.52	1	60.1***	1.30				1	0.01	
B × C(A)	14	34.3*	0.20	14	2.1*	0.64	14	2.1***	0.22	14	0.02*	0.18
B × D(A)	14	20.1		14	2.6**	0.77				14	0.01	
C × D	1	0.08		1	1.9					1	0.10	
A × C × D	1	8.1		1	1.6					1	0.04	
B × C × D(A)	14	17.0		14	1.9*	0.58				14	0.024**	0.26
Error	151	16.1		53	0.87		86	0.72		147	0.01	

*$P < 0.05$, **$P < 0.01$, ***$P < 0.001$, F-ratios for tests are: $F_A = MS_A/MS_B$, $F_B = MS_B/MS_{Error}$, $F_C = MS_C/MS_{AxC}$, $F_D = MS_D/MS_{AxD}$, $F_{A×C} = MS_{A×C}/MS_{B×C}$, $F_{A×D} = MS_{A×D}/MS_{B×D}$, $F_{B×C} = MS_{B×C}/MS_{Error}$, $F_{B×D} = MS_{B×D}/MS_{Error}$, $F_{C×D} = MS_{C×D}/MS_{A×C×D}$, $F_{A×C×D} = MS_{A×C×D}/MS_{B×C×D}$, $F_{B×C×D} = MS_{B×C×D}/MS_{Error}$.

Table 4. Estimates of heritability for six traits. A half-sib mating design was used and variance components were estimated using restricted maximum likelihood. Shown are estimates from the two populations from Russian wheat aphid and cabbage aphid, tested on both hosts. Estimates in bold are significantly different from zero, negative estimates are indicated as zero.

| | | Wasps from RWA | | | | Wasps from cabbage aphid | | | |
| | | Tested on cabbage aphid | | Tested on RWA | | Tested on cabbage aphid | | Tested on RWA | |
		h^2	SE×2	h^2	SE×2	h^2	SE×2	h^2	SE×2
Host acceptance	Sire	0.67	1.23	0	0.51	0.23	0.49	0	0.68
	Sire + Dam	0	0.61	0	0.92	0	0.41	**1.67**	**0.74**
Handling time	Sire	0	0.51	0.27	0.88	0	0.30	0	0.41
	Sire + Dam	0	0.92	0	0.79	0	0.46	0.50	0.55
Productivity	Sire	0.11	0.52	**0.89**	**0.68**	**0.72**	**0.61**	**1.29**	**0.61**
	Sire + Dam	**0.76**	**0.54**	0.23	0.26	0	0.24	0.03	0.15
Survival of immatures	Sire	0.93	1.52	0.42	1.13	**1.61**	**0.66**	0.68	1.14
	Sire + Dam	0.43	0.60	**1.29**	**0.69**	**0.55**	**0.22**	**1.18**	**0.58**
Development time of tested females	Sire	–		0	0.17	0	0.25	–	
	Sire + Dam	–		0.20	0.23	**0.47**	**0.31**	–	
Sex ratio	Sire	0	0.29	0.03	0.29	0	0.30	0	0.28
	Sire + Dam	0	0.44	0	0.39	0.24	0.43	0	0.42

secondary sex ratio was measured, which reflects both behavioural decisions by females at the time of oviposition and differential survival of male and female immatures (Godfray, 1994). However, no correlation was found between sex ratio and either productivity ($r = -0.26$) or progeny survival ($r = -0.27$, both $P > 0.05$), indicating the sex ratio shift on 'home' hosts resulted from female behaviour rather than differential survival.

(iii) Heritability. Heritabilities within each of the populations were estimated only from replicates where the initial rearing host was the same as the original host, for several reasons. First, this provided the most balanced data in terms of sample size. Second, estimates from different hosts and populations could not be combined because of the genetic trade-offs in traits on RWA or cabbage aphid. Third, initial behavioural discrimination of field-caught females limited the sample size of some treatment combinations. In 2001, females collected in the field from one host often refused to oviposit in the alternate host. Six of the 14 field females collected from cabbage refused oviposition in RWA. Similarly, four of the 13 females from RWA failed to oviposit in cabbage aphid. Their daughters, however, showed much less discrimination: only five of the 48 wasps from the cabbage aphid population and four out of 46 from the RWA population failed to oviposit in both hosts. Thus, heritability estimates were from half-sibling families of five sires from RWA (10 families) and seven sires from cabbage aphid (12 families).

Heritability was estimated using variance between sires (males) and combined sire and dam variances (Table 4). Only one heritability estimate for infectivity, host acceptance by the cabbage aphid population, was greater than zero. On the other hand, most estimates for heritability of virulence were positive, and nine estimates were significantly greater than zero. Productivity and larval survival, which showed the clearest evidence of trade-offs in host

use, also showed the most consistently significant heritability estimates. Most of the significant heritabilities came from the cabbage aphid population, where more sires were tested, resulting in greater statistical power.

Discussion

These three experiments demonstrate the parasitoid *D. rapae* originating from one aphid host species suffered reduced fitness on an 'alternative' or 'novel' host. This pattern was repeatable in populations from two different sources (a laboratory colony and field populations near Fort Collins, Colorado) and at different times (1996, 2001). Reactions to different hosts were not equally strong in each of the experiments: the RWA population showed a lesser difference between hosts in the 1996 transplant experiment, and the cabbage aphid population showed a lesser effect in the 2001 transplant experiment. Nonetheless, host-related trade-offs for virulence (sex ratio, development time, productivity, and survival) were found to have large effects in all experiments, and trade-offs in productivity and survival were genetically based. On the other hand, trade-offs in sex ratio and development time were most conditioned by the host environment in the laboratory. Females produced more female-biased sex ratios on the host from which they emerged, as previously noted by Hafez (1961).

Transplant experiments demonstrated genetic variation within the populations. In the 1996 transplant experiment, full-sibling families differed in productivity. In 2001, half-sib families were found to differ for all traits except handling times (Table 3), and this was reflected in heritability estimates (Table 4). Further, families differed in their responses to the two hosts (Table 3, Fig. 6). This implies the existence of genetic variation in the ability to successfully attack both hosts (plasticity), as opposed to
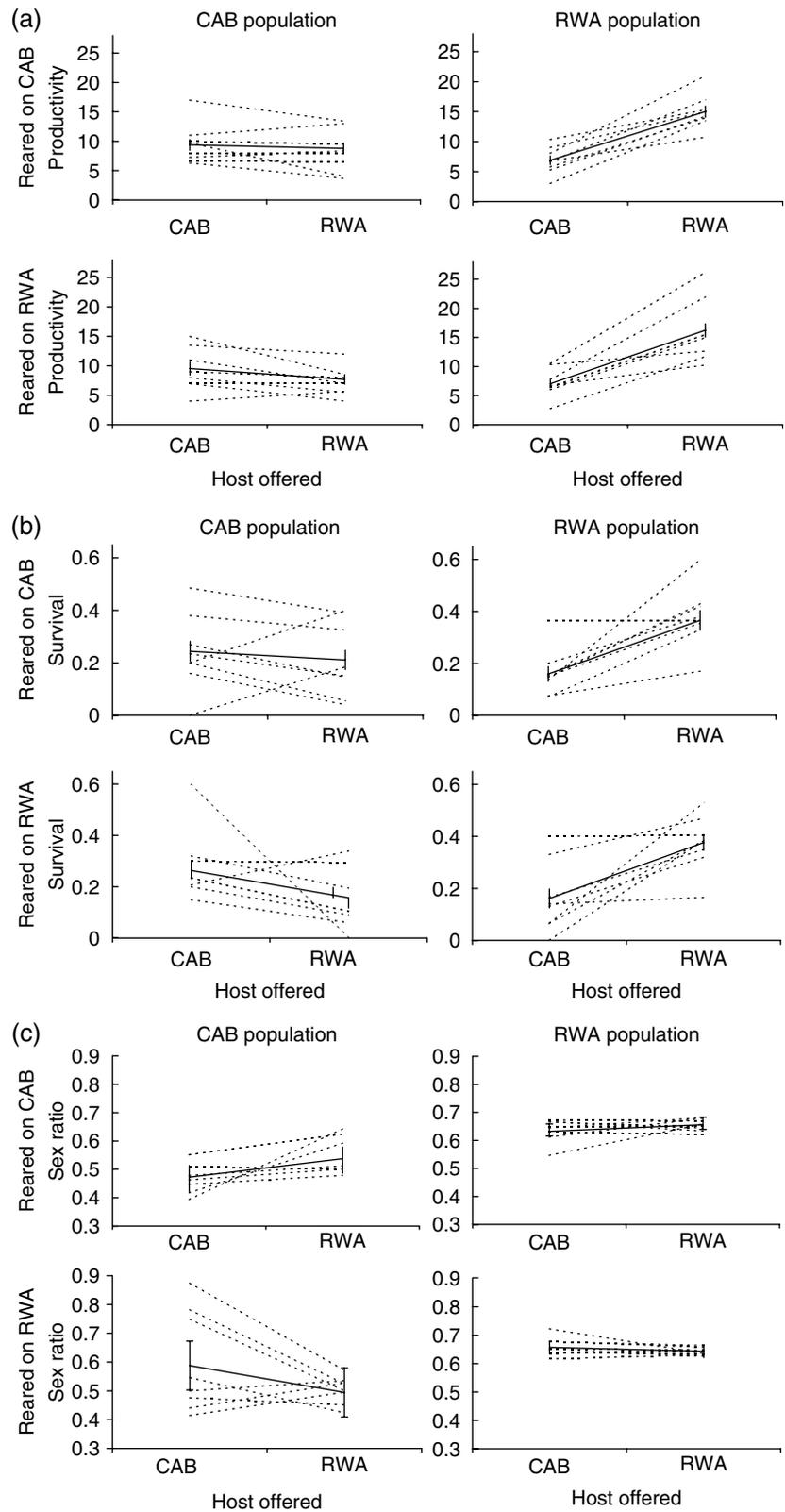
Fig. 6. Transplant experiment 2001: productivity of female wasps, survival of their progeny, and sex ratio of their progeny, from two populations [cabbage aphid (CAB) and Russian wheat aphid (RWA)], reared and tested in two host environments. Means of individual half-sibling families are plotted (broken lines) along with the average of all families (solid line ± SE). (a) Overall productivity, (b) survival of immatures, (c) sex ratio (proportion males). See Table 3 for statistical analysis.

genetically fixed trade-offs. Genetic variation in host use by aphid parasitoids has been described previously. Henter and Via (1995) found that virulence in *Aphidius ervi* populations that attack the pea aphid was heritable. Subsequent studies failed to find differences in virulence on pea aphids from clover and alfalfa, even though pea aphids from clover and alfalfa differ in resistance to parasitism (Hufbauer, 2001, 2002). Other studies of several *Aphidius* species, *Ephedrus* species, and *D. rapae* have also pointed to a genetic basis for differences in performance on different aphid species (Powell & Wright, 1988, 1992; Pennacchio *et al.*, 1994; Pike *et al.*, 1999), although larval/pupal conditioning could not be ruled out in those experiments.

Infectivity differed between experiments. Host-specific preferences were found in wasps collected in 2001 from both RWA and cabbage aphid in the field; wasps rejected the 'alternative' host at a high rate, while their laboratory reared progeny were much less discriminating. Some innate host preferences and differences in host searching were apparent: wasps always discriminated against the 'novel' host (green peach aphid) and time needed to attack cabbage aphid was longer than time for attacking RWA. Further, wasps from the long-term laboratory colony always preferred their 'home' host, RWA, regardless of the rearing host (Fig. 2). In previous studies of *D. rapae*, a consistent attraction to volatiles from crucifer plants and cabbage aphids was found in wasps from both RWA and cabbage aphid in northern Colorado, and the attraction did not change with host experience (Vaughn *et al.*, 1996). Differences between field-caught and laboratory-reared wasps are not surprising, because all of the stimuli use by wasps to guide attack of hosts cannot be controlled in a laboratory setting. On the other hand, differences in infectivity could also represent incomplete behavioural adaptation to particular hosts. In some parasitoids, populations differ in how much host experience affects subsequent host acceptance (Bjorksten & Hoffman, 1995; Vos & Vet, 2004).

Evolution of host range

Whether local selection could eventually lead to *D. rapae* completely specialising on different hosts will depend on the genetic architecture of the traits involved. Via and Hawthorne (2002) suggested that host specialisation is most likely when trade-offs in performance on alternative hosts become genetically correlated with mate choice. Female *D. rapae* produce a sex pheromone to attract mates (Vaughn *et al.*, 1996), but the effective range of attraction and other aspects of habitat or mating preferences remain to be explored. Similarly, host specialisation would be expected to evolve rapidly if infectivity and virulence became genetically correlated, as oviposition decisions by females would result in greatest fitness on the 'correct' host (Jaenike & Holt, 1991; Feder & Filchak, 1999).

Currently available evidence argues against pure host specialisation by *D. rapae* (Baer *et al.*, 2004). Instead, it is

likely that *D. rapae* engages in serial host specialisation, where a particular host species is used while it is available. At the temporal and spatial scales of agricultural fields, *D. rapae* females may be limited to opportunistically attacking whichever aphid hosts are present during their short lifespans. It appears that some traits (host acceptance, sex ratio) are influenced by environmental cues derived from the host in which wasps develop, and may be learned. Host preferences, however, will be followed by selection for virulence in that host species. Use of a single host will be favoured by both infectivity and virulence as long as that host remains locally abundant; switching to an alternate host will only be favoured if wasp populations can rapidly respond to selection in the new environment. Thus, the effective host range of a parasitoid will be influenced both by infectivity and virulence, and in *D. rapae* it appears that infectivity maintains some behavioural plasticity, while virulence is genetically determined.

Three other lines of evidence point to incomplete adaptation to hosts in *D. rapae*. First, trade-offs were asymmetric and varied slightly between transplant experiments in 1996 and 2001. Second, each population harboured considerable genetic variation, which would not be expected if strong selection drove the populations to endpoints. Third, in a previous study of fine-scale population genetic structure conducted at the same organic farm used in these experiments, modest but consistent differentiation was found ($F_{st} \approx 0.07$) between adjacent RWA and cabbage aphid fields (Vaughn & Antolin, 1998). In both years of that study, populations were least differentiated in early summer, and became more differentiated in samples taken in middle and late summer (Vaughn & Antolin, 1998). A subsequent genetic study of *D. rapae* based on mtDNA variation further suggested that gene flow was high (Baer *et al.*, 2004). Thus, it is likely that gene flow prevents *D. rapae* populations from becoming completely isolated, and that local selection rather than isolation creates genetic subdivision between populations on different hosts. Year-to-year variation in trade-offs in these transplant experiments may result from year-to-year variation in genetic composition of the populations, an expected result when local selection and gene flow work in opposition.

Generality of these results beyond northern Colorado remain to be demonstrated, as local adaptation has not been found in all cases of several well-studied host–parasitoid systems (Hufbauer, 2001, 2002; Kraaijeveld *et al.*, 2002; Dupas *et al.*, 2003; Vos & Vet, 2004). RWA originated in central Asia and invaded the wheat-growing regions of the western North America in 1986 (Halbert & Stoetzel, 1998). Since then, *D. rapae* from wheat-growing regions of the Mediterranean and Asia were released for biological control in western North America and became established. It is possible that the host adaptation seen in Colorado is the result of RWA-adapted genotypes released at that time (Baer *et al.*, 2004). Whether infectivity and virulence on different hosts continue to evolve in *D. rapae*, and how they

relate to adaptation to hosts and host range on a larger scale will require further study.

References

Baer, C.F., Tripp, D.W., Bjorksten, T.A. & Antolin, M.F. (2004) Phylogeography of a parasitoid wasp (*Diaeretiella rapae*): no evidence of host-associated lineages. *Molecular Ecology*, **13**, 1859–1869.

Bjorksten, T.A. & Hoffmann, A.A. (1995) Effects of pre-adult and adult experience on host acceptance in choice and non-choice tests in 2 strains of *Trichogramma*. *Entomologia experimentalis et applicata*, **76**, 49–58.

Boecklin, W.J. & Mopper, S. (1998) Local adaptation in specialist herbivores: theory and evidence. *Genetic Structure and Local Adaptation in Natural Insect Populations: Effects of Ecology, Life History, and Behavior* (ed. by S. Mopper and S. Y. Strauss), pp. 64–88. Chapman & Hall, New York.

Brewer, M.J., Nelson, D.J., Ahern, R.G., Donahue, J.D. & Prokrym, D.R. (2001) Recovery and range expansion of parasitoids (Hymenoptera: Aphelinidae and Braconidae) released for biological control of *Diuraphis noxia* (Homoptera: Aphididae) in Wyoming. *Environmental Entomology*, **30**, 578–588.

Burd, J.D., Shufran, K.A., Elliot, N.C., & French, B.W. & Prokrym, D.R. (2001) Recovery of imported Hymenopterous parasitoids released to control Russian wheat aphids in Colorado. *Southwest Entomologist*, **26**, 23–31.

Chow, A. & Mackauer, M. (1992) The influence of prior ovipositional experience on host selection in four species of aphidiid wasps. *Journal of Insect Behavior*, **5**, 99–108.

Dres, M. & Mallet, J. (2002) Host races in plant-feeding insects and their importance in sympatric speciation. *Philosophical Transactions of the Royal Society of London, Series B*, **357**, 471–492.

Dupas, S., Carton, Y. & Poirie, M. (2003) Genetic dimension of the coevolution of virulence-resistance in *Drosophila*–parasitoid wasp relationships. *Heredity*, **90**, 84–89.

Dybdahl, M.F. & Storfer, A. (2003) Parasite local adaptation: Red Queen versus Suicide King. *Trends in Ecology and Evolution*, **18**, 523–530.

van Emden, H.F., Sponagl, B., Wagner, E., Baker, T., Ganguly, S. & Douloumpaka, S. (1996) Hopkins' 'host selection principle': another nail in its coffin. *Physiological Entomology*, **21**, 325–328.

Feder, J.L. & Filchak, K.E. (1999) It's about time: the evidence for host plant-mediated selection in the apple maggot fly, *Rhagoletis pomonella*, and its implications for fitness trade-offs in phytophagous insects. *Entomologia experimentalis et applicata*, **91**, 211–225.

Fellowes, M.D.E. & Kraaijeveld, A.R. (1998) Coping with multiple enemies–the evolution of resistance and host–parasitoid community structure. *Ecology Letters*, **1**, 8–10.

Ferrari, J., Darby, A.C., Daniel, T.J., Godfray, H.C.J. & Douglas, A.E. (2004) Linking the bacterial community in pea aphids with host-plant use and natural enemy resistance. *Ecological Entomology*, **29**, 60–65.

Gandon, S. & Michalakis, Y. (2002) Local adaptation, evolutionary potential and parasite–host coevolution: interactions between migration, mutation, population size, and generation time. *Journal of Evolutionary Biology*, **15**, 451–462.

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

Hafez, M. (1961) Seasonal fluctuations of population density of the cabbage aphid *Brevicoryne brassicae* (L.) in the Netherlands, and the role of its parasite, *Aphidius (Diaeretiella) rapae* (Curtis). *Tijdschriften Plantenziekten*, **67**, 445–548.

Halbert, S.E. & Stoetzel, M.B. (1998) Historical overview of the Russian wheat aphid (Homoptera: Aphididae). *Response Model for an Introduced Pest–the Russian Wheat Aphid. Proceedings: Thomas Say Publications in Entomology* (ed. by S. S. Quisenberry and F. B. Peairs), pp. 12–30. Entomological Society of America, Lantham, Maryland.

Henneman, M.L. & Memmott, J. (2001) Infiltration of a Hawaiian community by introduced biological control agents. *Science*, **293**, 1314–1316.

Henter, H.J. & Via, S. (1995) The potential for coevolution in a host–parasitoid system. I. Genetic variation within a population of wasps in the ability to parasitize an aphid host. *Evolution*, **49**, 427–438.

Hopper, K.R., Coutinout, D., Chen, K., Kazmer, D.J., Mercadier, G., Halbert, S.E. *et al.* (1998) Exploration of natural enemies to control. *Diuraphis noxia* (Homoptera: Aphididae) in the United States. *Response Model for an Introduced Pest–the Russian Wheat Aphid. Proceedings: Thomas Say Publications in Entomology* (ed. by S. S. Quisenberry and F. B. Peairs), pp. 166–182. Entomological Society of America, Lantham, Maryland.

Hufbauer, R.A. (2001) Pea aphid–parasitoid interactions: have parasitoids adapted to differential resistance? *Ecology*, **82**, 717–725.

Hufbauer, R.A. (2002) Evidence for nonadaptive evolution in parasitoid virulence following a biological control introduction. *Ecological Applications*, **12**, 66–78.

Jaenike, J. & Holt, R.D. (1991) Genetic variation for habitat preference: evidence and explanations. *American Naturalist*, **137**, S67–S90.

Killick, R.J. (1971) Sex-linkage and sex-limitation in quantitative inheritance. I. Random mating populations. *Heredity*, **27**, 175–188.

Kraaijeveld, A.R., Ferrari, J. & Godfray, H.C.J. (2002) Costs of resistance in insect–parasite and insect–parasitoid interactions. *Parasitology*, **125**, S71–S82.

Lajeunesse, M.J. & Forbes, M.R. (2002) Host range and local parasite adaptation. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, **269**, 703–710.

Lester, P.J. & Holtzer, T.O. (2002) Patch and prey utilization behaviors by *Aphelinus albipodus* and *Diaeretiella rapae* (Hymenoptera: Aphelinidae and Aphidiidae) on Russian whea aphid (Homoptera: Aphididae). *Biological Control*, **24**, 183–191.

Louda, S.M., Pemberton, R.W., Johnson, M.T. & Follett, P.A. (2003) Nontarget effects–the Achilles' heel of biological control? Retrospective analyses to reduce risk associated with biocontrol introductions. *Annual Review of Entomology*, **48**, 365–396.

Lynch, M. & Walsh, B. (1998) *Genetics and Analysis of Quantitative Traits*. Sinauer Associates, Sunderland, Massachusetts.

Mackauer, M., Michaud, J.P. & Völkl, W. (1996) Host choice by Aphidiid parasitoids (Hymenoptera: Aphidiidae): host recognition, host quality, and host value. *Canadian Entomologist*, **128**, 959–980.

Mackauer, M. & Starý, P. (1967) Hym. Ichneumonoidea, World Aphidiidae. *Index of Entomophagous Insects* (ed. by V. Delucchi and G. Remaudiere). Le Frabcois, Paris.

McEvoy, P.B. (1996) Host specificity and biological pest control – how well is research on host specificity addressing the potential risks of biological control? *BioScience*, **46**, 401–405.

Mohamed, A.H., Lester, P.J. & Holtzer, T.O. (2000) Abundance and effects of predators and parasitoids on the Russian wheat aphid (Homoptera: Aphididae) under organic farming conditions in Colorado. *Environmental Entomology*, **29**, 360–368.

Némec, V. & Starý, P. (1994) Population diversity of *Diaeretiella rapae* (M'Int) (Hym, Aphidiidae), an aphid parasitoid in agroecosystems. *Zeitschrift ang Entomologisches*, **97**, 223–233.

Oliver, K.M., Russell, J.A., Moran, N.A. & Hunter, M.S. (2003) Facultative bacterial symbionts in aphids confer resistance to parasitic wasps. *Proceedings of the National Academy of Sciences of the United States of America*, **100**, 1803–1807.

Pennacchio, F., Digilio, M.C., Trembay, E. & Tranfaglia, A. (1994) Host recognition and acceptance behavior in two aphid parasitoid species: *Aphidius ervi* and *Aphidius microlophi* (Hymenoptera: Braconidae). *Bulletin of Entomological Research*, **84**, 57–64.

Pike, K.S., Starý, P., Miller, T., Alison, D., Boydston, L., Graf, G. et al. (1999) Host range and habitats of the aphid parasitoid *Diaeretiella rapae* (Hymenoptera: Aphidiidae) in Washington state. *Environmental Entomology*, **28**, 61–71.

Powell, W. & Wright, A.F. (1988) The abilities of the aphid parasitoids *Aphidius ervi* Halliday and *A. rhopalosiphi* De Stephani Perez (Hymenoptera: Braconidae) to transfer between different known host species and the implications for the use of alternative hosts in pest control strategies. *Bulletin of Entomological Research*, **78**, 683–693.

Powell, W. & Wright, A.F. (1992) The influence of host food plants on host recognition by four aphidiine parasitoids (Hymenoptera: Braconidae). *Bulletin of Entomological Research*, **81**, 449–453.

Prokrym, D.R., Pike, K.S. & Nelson, D.J. (1998) Biological control of *Diuraphis noxia* (Homoptera: Aphididae): implementation and evaluation of natural enemies. *Response Model for an Introduced Pest – the Russian Wheat Aphid. Proceedings: Thomas Say Publications in Entomology* (ed. by S. S. Quisenberry and F. B. Peairs), pp. 163–208. Entomological Society of America, Lantham, Maryland.

Rausher, M.D. (2001) Co-evolution and plant resistance to natural enemies. *Nature*, **411**, 857–864.

Reed, H.C., Reed, D.K. & Elliot, N.C. (1992) Comparative life table statistics of *Diaeretiella rapae* and *Aphidius matricariae* on the Russian wheat aphid. *Southwestern Entomologist*, **17**, 307–312.

Rosenthal, R. & Rosnow, R.L. (1985) *Contrast Analysis: Focused Comparison in the Analysis of Variance*. Cambridge University Press, Cambridge.

Rossiter, M.C. (1998) Assessment of genetic variation in the presence of maternal or paternal effects in herbivorous insects. *Genetic Structure and Local Adaptation in Natural Insect Populations: Effects of Ecology, Life History, and Behavior* (ed. by S. Mopper and S. Y. Strauss), pp. 113–138. Chapman & Hall, New York.

Sokal, R.R. & Rohlf, F.J. (1995) *Biometry*, 3rd edn. W.H. Freeman, New York.

Strand, M.R. & Obrycki, J.J. (1996) Host specificity of insect parasitoids and predators. *Bioscience*, **46**, 422–429.

Strand, M.R. & Pech, L.L. (1995) Immunological basis for compatibility in parasitoid–host relationships. *Annual Review of Entomology*, **40**, 31–56.

Strauss, S.Y. & Karban, R. (1998) The strength of selection: intraspecific variation in host-plant quality and the fitness of herbivores. *Genetic Structure and Local Adaptation in Natural Insect Populations: Effects of Ecology, Life History, and Behavior* (ed. by S. Mopper and S. Y. Strauss), pp. 156–177. Chapman & Hall, New York.

Symondson, W.O.C., Sunderland, K.D. & Greenstone. M.H. (2002) Can generalist predators be effective biocontrol agents? *Annual Review of Entomology*, **47**, 561–594.

Vaughn, T.T. & Antolin, M.F. (1998) Population genetics of an opportunistic parasitoid in an agricultural landscape. *Heredity*, **80**, 152–162.

Vaughn, T.T., Antolin, M.F. & Bjostad, L.B. (1996) Behavioral and physiological responses of *Diaeretiella rapae* to semiochemicals. *Entomologia experimentalis et applicata*, **78**, 187–196.

Vet, L.E.M., Hemerik, L., Visser, M.E. & Wäckers, F.L. (2002) Flexibility in host-search and patch-use strategies of insect parasitoids. *The Behavioural Ecology of Parasites* (ed. by E. E. Lewis, J. F. Campbell and M. V. K. Sukhdeo), pp. 39–64. CABI Publishing, New York.

Via, S. & Hawthorne, D.J. (2002) The genetic architecture of ecological specialization: correlated gene effects on host use and habitat choice in pea aphids. *American Naturalist*, **159**, S76–S88.

Vos, M. & Vet, L.E.M. (2004) Geographic variation in host acceptance by an insect parasitoid: genotype versus experience. *Evolutionary Ecology Research*, **6**, 1021–1035.

Wilkinson, L. (1992) *SYSTAT: the System for Statistics*. Systat Inc., Evanston, Illinois.

van Zandt, P.A. & Mopper, S. (1998) A meta-analysis of adaptive deme formation in phytophagous insect populations. *American Naturalist*, **152**, 595–604.