A strong association between immune responsiveness and natal dispersal in a songbird

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Because parasite faunas typically show considerable spatio-temporal variation, and because parasites can have important fitness consequences, host defence mechanisms, including the immune system, can be expected to coevolve with natal dispersal, i.e. the movement of a newborn individual from its site of birth to its first site of reproduction. We demonstrate that immigrant individuals show a significantly higher humoral immune response towards a novel antigen than do local recruits in two independent populations of the great tit (*Parus major*). There was no effect of age, sex, tarsus length or body mass on immune responsiveness. Our results are consistent with the idea that phenotype-dependent dispersal and/or dispersal-by-phenotype-dependent selection establish a relation between immune responsiveness and natal dispersal.

Keywords: great tit; natal dispersal; *Parus major*; immune responsiveness; ecological immunology

1. INTRODUCTION

Natal dispersal is thought to be an important life-history trait affecting both species persistence and evolution, but it is one of the least understood behavioural processes (Clobert *et al.* 2001). Previous research has mainly focused on factors that affect dispersal patterns such as sex biases, and on demographic and genetic consequences of natal dispersal at the population level (Stenseth & Lidicker 1992). However, few studies have addressed relationships between individual variation in dispersal and endocrine and physiological factors in animals (Dufty & Belthoff 2001). One aspect of an individual's phenotype that is important for coping with infectious agents, which have a strong impact on survival and/or reproductive success (Sheldon & Verhulst 1996), is the immune system.

Both a positive and a negative relation between immune responsiveness and natal dispersal can be predicted. Because infectious agents show spatial variation (Hudson *et al.* 2002), dispersing individuals are exposed to a broader range of environmental conditions than philopatric ones (Møller & Erritzøe 2001). Therefore, individuals that decide to disperse should have more efficient immune systems than philopatric individuals. Alternatively,

if their immune system is insufficiently effective, dispersing individuals should pay a higher survival cost. Overall, both scenarios will cause a positive relation between effective dispersal and immune responsiveness. Recently, Møller *et al.* (2004) found that nestlings of bird species with longer natal dispersal distances had stronger mean T-cell-mediated immune responses. By contrast, if mainly low-quality individuals are forced to disperse (de Laet 1985), and if quality is reflected in immune responsiveness (Sheldon & Verhulst 1996), one would predict that they have a lower immune responsiveness compared to philopatric individuals.

In this study, we examined whether natal dispersal explains any variation in immune responsiveness in two populations of the great tit (*Parus major*). The immune responsiveness of roosting great tits in winter, after the main dispersal phase has finished (Dingemanse *et al.* 2003), was assessed by challenging individuals with sheep red blood cells (SRBC), i.e. a novel multigenic antigen that elicits a thymus-dependent activation of the B cells (Kuby 1997). This is a well-established procedure in ecological immunology research, which yields a direct and reliable estimate of an individual's ability to elicit a humoral immune response towards a novel pathogen (Sheldon & Verhulst 1996).

2. MATERIAL AND METHODS

Natal dispersal and immune responsiveness were investigated in two natural great tit populations in northern Belgium. The great tit is a small, winter resident passerine bird in western Europe but an irregular migrant in northern parts of Europe. Great tits are hole-nesting passerines that readily accept nest-boxes for roosting and breeding. In a first, patchy population (study area 'Boshoeck'; see Matthysen *et al.* 2001), we sampled 10 woodlots of more than 250 years of age, of which some consist of only mature oak whereas others consist of poplar, beech and other deciduous trees. The second study area 'UIA' (see Van Duyse *et al.* 2002) consists of a more contiguous habitat with park-like vegetation created *ca.* 1975.

Birds were captured while roosting in nest-boxes in January 2002 in Boshoeck and in February 2001 in UIA. Sex and age (yearlings versus older) were determined and body mass and tarsus length were measured. Individuals were immunologically challenged by means of an intramuscular injection with 50 µl of a 40% solution of SRBC before they were returned to the nest-box. After 6 days (the time at which the antibody titre against SRBC has reached its maximum level in the great tit; T. Snoeijs, R. Pijten and M. Eens, unpublished data), individuals were recaptured and blood was sampled from the wing vein to quantify their humoral immune responses towards the SRBC. Samples were centrifuged and plasma was stored at -70 °C until immunological analyses (see electronic Appendix A, available on The Royal Society's Publication's Web site).

Three categories of natal dispersal could be distinguished in the study area Boshoeck: (i) birds captured in their natal woodlot were defined as 'residents'; (ii) birds captured in a woodlot different from their natal woodlot were defined as 'dispersers'; and (iii) birds that were found in one of the woodlots but were not ringed as a nestling in the study area were defined as 'immigrants'. We can be sure that the immigrant group consisted mostly of individuals that dispersed over a longer distance than 'dispersers' because (i) the breeding density in the matrix between woodlots is low and, each year, nestlings were systematically ringed in the few nest-boxes provided by the public; (ii) the density of birds breeding in natural cavities within woodlots was very low (less than 10%; Matthysen *et al.* 2001). The dispersal distances of residents and dispersers ranged respectively between 0 and 370 m (mean $\pm$ s.d. = 172 ± 92 m) and between 146 and 1712 m (mean $\pm$ s.d. = 671 ± 416 m). In study area UIA, only two dispersal categories (residents and immigrants) could be distinguished.

3. RESULTS

The recapture probability of injected birds did not depend on dispersal status, nor on the interaction between dispersal status and age or sex (logistic regression:

Table 1. Results of a stepwise backward elimination procedure applied to a linear model with humoral immune responsiveness as the dependent variable.

(Non-significant independent variables are listed in the order in which they were removed from the starting model. A significance level of $\alpha = 0.05$ was used as the threshold for the variable to remain in the model. The F -statistics represent F -values from the statistical analysis using all covariates listed below the considered variable. The first number in the d.f. column refers to the degrees of freedom of the numerator of the F -ratio, whereas the second number refers to the degrees of freedom of the denominator of the F -ratio.)

independent variables	F	d.f.	p
dispersal category $\times$ sex	0.02	2,51	0.980
dispersal category $\times$ age	0.20	2,53	0.822
sex	1.28	1,55	0.262
age	1.26	1,56	0.267
dispersal category			
$\times$ vegetation type	1.20	2,57	0.309
dispersal category			
$\times$ tarsus length	2.56	2,59	0.086
tarsus length	1.17	1,61	0.285
dispersal category $\times$ body mass			
mass	2.55	2,62	0.086
body mass	0.10	1,64	0.757
vegetation type	5.69	1,65	0.020
dispersal category	9.74	2,65	< 0.001

Figure 1. Mean primary antibody titres ($\log_2$ -transformed), 6 days after immunization with SRBC per natal dispersal category in the study area Boshhoek. Triangles and bars represent parameter estimates from the statistical model indicating, respectively, means and 95% CIs for each dispersal category.

Boshhoek: all p -values greater than 0.25; UIA: all p -values greater than 0.09; see electronic Appendix A for sample sizes and statistical analyses). Winter dispersal reflected natal dispersal well (see electronic Appendix A for more details).

Dispersal category and vegetation type of the woodlot were the only two predictors of immune responsiveness for the Boshhoek population (see table 1; see table 2 in electronic Appendix A for statistical analyses). Immigrants showed a significantly higher humoral immune response than residents and dispersers ($F_{2,65} = 9.74$, $p < 0.001$), but there was no significant difference in immune responsiveness between residents and dispersers (figure 1).

Furthermore, individuals captured in the oak woodlots had a significantly lower humoral immune response compared with remaining woodlots ($F_{1,65} = 5.69$, $p = 0.02$). The fact that dispersal category was also significant in the full model ($F_{2,61} = 7.62$, $p = 0.001$) indicates that the difference could not be explained as an effect of body mass or tarsus length. There were also no indications for conditions at hatching to explain immune responsiveness (see electronic Appendix A for more details). We performed the same analysis to test the relation between immune responsiveness and natal dispersal distance within locally hatched birds, i.e. pooled residents and dispersers. Although in the same direction, the relation was not significant in the full model ($F_{1,37} = 1.89$, $p > 0.18$) nor after the deletion of all non-significant variables, including vegetation type ($F_{1,42} = 1.69$, $p = 0.20$; slope $\pm$ s.e. = 0.0007 ± 0.0005 m).

The result that we found for the Boshhoek population was confirmed in the UIA population: immigrants showed a significantly higher immune responsiveness (mean $\pm$ s.d. of immigrants = 4.29 ± 2.26 and residents = 2.58 ± 2.02 ; Kruskal–Wallis test: $\chi^2_1 = 5.15$, $p = 0.023$).

4. DISCUSSION

In this study, we found that in two independent natural great tit populations immigrants showed a significantly higher humoral immune response towards SRBC than local recruits. Three mechanisms could cause an association between a phenotypic variable, quantified after natal dispersal, and dispersal itself: a phenotype-dependent dispersal decision, dispersal-dependent phenotypic plasticity and dispersal-by-phenotype-dependent survival. We interpret the observed pattern using indirect arguments for or against the possible mechanisms.

It has been shown at the interspecific level that immune function and natal dispersal can coevolve (Møller & Erritzøe 1998; Møller *et al.* 2004). In the same manner, a relation between immune function and natal dispersal at the intraspecific level could indicate the existence of a genetic polymorphism within one population; immigrants may be immunologically co-adapted to the increased chance to encounter a large variation in pathogens. Several lines of evidence suggest that a dispersal polymorphism linked to immunocompetence may exist in natural populations. Recently, not only has a genetic basis for dispersal (Hansson *et al.* 2003) been reported, but also large amounts of genetic variation for humoral immune responsiveness have been suggested in the great tit (J. Kilpimaa, T. Van de Castele, J. Mappes and R. Alatalo, unpublished data) and in a closely related species, the blue tit (*Parus caeruleus*; Råberg *et al.* 2003). Furthermore, the observation of differences in morphological parameters in the same Boshhoek study area between immigrants on the one hand and residents and dispersers on the other hand (Van de Castele 2002) suggests that dispersal covaries with other (morphological) characteristics that are often substantially heritable (Houle 1992). Nevertheless, the fact that two phenotypic traits show large genetic variance does not necessarily imply that a phenotypic correlation between these traits is genetically based. To reveal this, specific experimental designs are needed involving, for example, selection lines or breeding experiments.

Alternatively, a small-scale variation in environmental conditions, at the natal site or at the recapture site, could also cause the observed relation between immune responsiveness and natal dispersal, either directly or through phenotypic plasticity. However, based on the available information of our study, there is no evidence that the observed relation between immune responsiveness and natal dispersal would be environmental or the result of dispersal-dependent phenotypic plasticity. First, we found no effect of age, body mass or tarsus length on the immune responsiveness in the Boshhoek population, suggesting weak phenotypic plasticity, at least with respect to those variables. The observation of a significant effect of vegetation type on the immune responsiveness during the winter was rather unexpected and, at present, we have no obvious explanation for this. Second, there were no indications that hatching conditions influence the assessed immune responses. Third, we found a similar relation in two populations that differ strongly with respect to vegetation age and composition (see § 2).

Finally, if the relation between immune responsiveness and natal dispersal might be the result of dispersal-by-phenotype-dependent survival, this should be reflected in a negative relation between the variance in immune responsiveness and natal dispersal. Using data from the Boshhoek population, this was confirmed; the variance in immune responsiveness was smaller in immigrants than in pooled residents and dispersers (Bartlett's test for homogeneity of variances: one-tailed test, $\chi^2_1 = 3.47$, $p = 0.031$). For the UIA population there was no evidence of such a difference (one-tailed test: $\chi^2_1 = 0.17$, $p = 0.34$).

Although there are very clear-cut differences in immune responsiveness between immigrants and the other two dispersal categories, one could criticize the observed positive relation between immune responsiveness and natal dispersal status for two reasons. First, one could argue that the difference in immune responsiveness between immigrants versus residents and dispersers is due to a misclassification of unringed recruits hatched in the study area. However, if birds of the immigrant group with the 10% highest immune responsiveness values (i.e. the expected proportion of misclassified immigrants, see § 2) are removed from the dataset, the difference between immigrants and residents and dispersers remains significant ($F_{2,58} = 3.99$, $p = 0.02$). Second, there was no significant effect of dispersal distance at the local scale. However, this could be explained by the fact that the selective advantage of immunocompetence operates at relatively larger scales, possibly linked to the spatial scale of variation in parasite fauna.

In conclusion, to our knowledge this is the first study reporting a positive association between immune function and natal dispersal in natural populations. Clearly, more observational and experimental studies are now necessary to confirm our results and to investigate which of the three mechanisms underlies the observed pattern.

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- Clobert, J., Danchin, E., Dhondt, A. A. & Nichols, J. D. 2001 *Dispersal*. Oxford: Clarendon.
- de Laet, J. V. 1985 Dominance and aggression in juvenile great tits, *Parus major major* L., in relation to dispersal. In *Behavioral ecology—ecological consequences of adaptive behaviour* (ed. R. M. Sibly & R. H. Smith), pp. 375–380. Oxford: Blackwell Scientific.
- Dingemanse, N. J., Both, C., van Noordwijk, A. J., Rutten, A. L. & Drent, P. J. 2003 Natal dispersal and personalities in great tits (*Parus major*). *Proc. R. Soc. Lond. B* **270**, 741–747. (DOI 10.1098/rspb.2002.2300.)
- Duffy Jr, A. M. & Belthoff, J. R. 2001 Proximate mechanisms of natal dispersal: the role of body condition and hormones. In *Dispersal* (ed. J. Clobert, E. Danchin, A. A. Dhondt & J. D. Nichols), pp. 217–229. Oxford: Clarendon.
- Hansson, B., Bensch, S. & Hasselquist, D. 2003 Heritability of dispersal in the great reed warbler. *Ecol. Lett.* **6**, 290–294.
- Houle, D. 1992 Comparing evolvability and variability of quantitative traits. *Genetics* **130**, 195–204.
- Hudson, P. J., Rizzoli, A., Grenfell, B. T., Heesterbeek, H. & Dobson, A. P. 2002 *The ecology of wildlife diseases*. Oxford: Clarendon.
- Kuby, J. 1997 *Immunology*, 3rd edn. New York: Freeman.
- Matthysen, E., Adriaensen, F. & Dhondt, A. A. 2001 Local recruitment of great and blue tits (*Parus major*, *Parus caeruleus*) in relation to study plot size and degree of isolation. *Ecography* **24**, 33–42.
- Møller, A. P. & Erritzøe, J. 1998 Host immune defence and migration in birds. *Evol. Ecol.* **12**, 945–953.
- Møller, A. P. & Erritzøe, J. 2001 Dispersal, vaccination and regression of immune defence organs. *Ecol. Lett.* **4**, 484–490.
- Møller, A. P., Martín-Vivaldi, M. & Soler, J. J. 2004 Parasitism, host immune defence and dispersal. *J. Evol. Biol.* (In the press.)
- Råberg, L., Stjernman, M. & Hasselquist, D. 2003 Immune responsiveness in adult blue tits: heritability and effects of nutritional status during ontogeny. *Oecologia* **136**, 360–364.
- Sheldon, B. C. & Verhulst, S. 1996 Ecological immunology: costly parasite defences and trade-offs in evolutionary ecology. *Trends Ecol. Evol.* **11**, 317–321.
- Stenseth, N. C. & Lidicker Jr, W. Z. 1992 *Animal dispersal: small mammals as a model*. London: Chapman & Hall.
- Van de Castele, T. 2002 Ecological and genetic aspects of natal dispersal in the great tit. PhD thesis, University of Antwerp, Belgium.
- Van Duyse, E., Pinxten, R. & Eens, M. 2002 Effects of testosterone on song, aggression, and nestling feeding behavior in male great tits. *Parus major*. *Horm. Behav.* **41**, 178–186.

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