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LETTER

Alleles underlying larval foraging behaviour influence adult dispersal in nature

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Abstract

The dispersal and migration of organisms have resulted in the colonisation of nearly every possible habitat and ultimately the extraordinary diversity of life. Animal dispersal tendencies are commonly heterogeneous (e.g. long vs. short) and non-random suggesting that phenotypic and genotypic variability between individuals can contribute to population-level heterogeneity in dispersal. Using laboratory and field experiments, we demonstrate that natural allelic variation in a gene underlying a foraging polymorphism in larval fruit flies (*for*), also influences their dispersal tendencies as adults. Rover flies (for^R; higher foraging activity) have consistently greater dispersal tendencies and are more likely to disperse longer distances than sitter flies (for^S; lower foraging activity). Increasing *for* expression in the brain and nervous system increases dispersal in sitter flies. Our study supports the notion that variation in dispersal can be driven by intrinsic variation in food-dependent search behaviours and confirms that single gene pleiotropic effects can contribute to population-level heterogeneity in dispersal.

Keywords

Animal personalities, behaviour genetics, candidate gene approach, cGMP-dependent protein kinase, dispersal, foraging, movement ecology, pleiotropy, transgenic.

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INTRODUCTION

The movement of organisms in space and time is a complex phenomenon with major implications on the ecology and evolution of life (Kerr *et al*. 2002; Gros *et al*. 2006). Biodiversity itself is a product of the remarkable ability of organisms to disperse and adapt to new environments. A common finding in studies of animal dispersal, ranging from insects to humans, is that dispersal tendencies are naturally heterogeneous and non-random. Dobzhansky & Wright's (1943) pioneering efforts tracking the dispersal of drosophilid flies in nature was the first of many from various taxa to report that populations often consist of many individuals dispersing short distances and very few individuals dispersing extreme distances (Cavalli-Sforza 1959; Fraser *et al*. 2001; Meylan *et al*. 2009). For example, recent evidence from early hominid isotope and dental data suggests that while most individuals remained within their natal population, some individuals arrived from distant natal habitats (Copeland *et al*. 2011; Lalueza-Fox *et al*. 2011).

Genotypic and phenotypic variability between individuals can contribute to population-level variation in dispersal tendencies (Haag *et al*. 2005; Gurarie *et al*. 2010). Theoretical models assuming equal phenotypes (e.g. simple diffusion) for all individuals fail to accurately describe the commonly observed population-level variation in dispersal. Models incorporating individual variation are superior because the added complexity more accurately describes the distribution of movement distances from populations whose behaviour cannot be described by random walk or simple diffusion (Gurarie *et al*. 2010). Several researchers have proposed that variation in dispersal patterns may be driven by intrinsic differences in the behaviour of individuals (i.e. personalities, Réale *et al*. 2007; Cote *et al*. 2010) such as sociability (Réale *et al*. 2007; Seyfarth *et al*. 2012), shy-bold (Fraser *et al*. 2001; Edelsparre *et al*. 2013), and foraging phenotypes (Grinnell 1931; O'Riain *et al*. 1996; Meylan *et al*. 2009). Over 80 years ago Grinnell (1931) proposed that large-scale animal movements could arise from variation in routine behaviours such as foraging. For a non-migratory bird species, Grinnell calculated the prey search and foraging movements over a period of time and concluded that these distances together were similar to distances covered by migratory species during migration. The very idea that variation in the daily movement of animals gives rise to variation in dispersal presumes that both types of behaviours share a common mechanism (Woodbury 1941). To date, the physiological and genetic mechanisms underlying variation in dispersal tendencies are largely unknown (Gloria-Soria & Azevedo 2008; Wheat 2012). A link between dispersal tendencies and personality suggests a shared genetic basis wherein the genes underlying animal personalities may have pleiotropic effects on dispersal. Therefore, we employed a candidate gene approach (Fitzpatrick *et al*. 2005) addressing whether the *foraging* gene (*for*), known to influence the foraging behaviour of larval fruit flies and several other species, has pleiotropic effects on their dispersal as adults.

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Foraging activity in *Drosophila melanogaster* is naturally polymorphic (Sokolowski 2001). Rover larvae cover larger areas within food patches and greater movement between patches than sitters (Sokolowski 1980) and adult rovers walk longer distances after feeding than sitters (Pereira & Sokolowski 1993; Kent *et al*. 2009). This polymorphism is largely attributed to allelic variation in *foraging* (*for*), a gene on the 2nd chromosomes and which encodes a cGMP-dependent protein kinase (PKG)(Osborne *et al*. 1997). Rovers (for^R/-) have higher *for*-mRNA levels and PKG activity levels than sitters (for^s/for^s)(Osborne *et al*. 1997). The kinase encoded by *for* is a key regulator of many downstream targets such that several pleiotropic effects of *for* have been documented (for review see Reaume & Sokolowski 2009). Given this, we believe *for* is an excellent candidate gene for research on dispersal.

The dispersal events of insects are particularly difficult to define because they often occur concomitantly with events including foraging, mate searching, egg laying and predator avoidance. Therefore, in such organisms, it is argued that any movement leading to a net displacement over time must be considered dispersal (Benton & Bowler 2012). Consequently, we investigate the influence of *for* on adult dispersal, defined in its simplest form as any movement of individuals leading to spatial spread with the potential for gene flow (Turchin 1998; Clobert *et al*. 2001; Ronce 2007).

Plots showing population-level variation in dispersal distances are typically leptokurtic in shape having a higher peak and longer tail than a normal distribution with comparable mean and variance (Dobzhansky & Wright 1943; Cavalli-Sforza 1959; Wallace 1966; Johnston & Heed 1975; Taylor 1978; Kot *et al*. 1996; Skalski & Gilliam 2000; Fraser *et al*. 2001; Rodríguez 2002; Gurarie *et al*. 2010). Consequently, if dispersal tendencies in adult fruit flies are influenced by rover-sitter behavioural differences then we predict rovers to contribute more to the long tail (i.e. long dispersers) and sitters to contribute more to the high peak (i.e. short dispersers). Furthermore, manipulations of *for* gene expression in flies would lead to changes in their dispersal tendencies.

MATERIALS AND METHODS

Fly lines and rearing

Naturally derived rover and sitter lines

The rover (for^R = +; for^R; +) and sitter lines (for^s = +; for^s; +) represent different individual *for* alleles obtained from nature (Sokolowski 1980). The rover line carries the for^R allele, whereas the sitter line carries the for^s allele on the 2nd chromosomes. To mediate the effect of genetic background, both lines share common for^R-derived third chromosomes (de Belle & Sokolowski 1987).

Sitter mutant line (for^{s2} = +; for^{s2}; +)

The for^{s2} mutation represents a laboratory-derived *for* mutant allele that was generated on the for^R genetic background described above. Therefore, the for^R and for^{s2} lines differ only at their *for* locus (Pereira & Sokolowski 1993; de Belle *et al*. 1993). for^{s2} is characterised as having sitter-like *for* mRNA expression, PKG activity levels, and foraging activity (Pereira & Sokolowski 1993; Osborne *et al*. 1997; de Belle *et al*. 1993). for^{s2} provides an important genomic background control for genetic variation outside of the *for* locus.

Transgenic lines used to manipulate for *expression*

The extensive use of *D. melanogaster* in research on genetics and developmental biology has led to the creation of numerous genetic tools including transgenic fly stocks. By capitalising on a molecular interaction specific to yeast (the binding of GAL4 protein to the UAS sequence), researchers have developed the ability to target the expression of specific genes to specific cells and tissues (Brand & Perrimon 1993). Here, this is conducted on a for^s genetic background since sitters naturally have lower *for* expression than rovers. In this study, we used the *elav*-GAL4 driver to artificially increase the expression of *for*, specifically the *for*T1a transcript, in the brain and central nervous system (CNS) of the flies. Briefly, the driver, *elav* was linked with GAL4 so that GAL4 protein is produced in cells that naturally express the *elav* gene (broad expression in the brain and CNS, Robinow & White 1988)(w^1; for^s; *elav*-GAL4). In a separate genetic line, UAS was linked with the T1a transcript of *for* (w^1; for^s; UAS-*for*T1a). This transgenic *for* sequence is only activated in cells containing GAL4 protein. Therefore, a cross between *elav*-GAL4 flies and UAS-*for*T1a flies will generate progeny that over-express *for* in the brain and CNS (w^1; for^s; *elav*-GAL4/UAS-*for*T1a). Altogether, by artificially increasing *for* expression in the nervous system, sitter larvae can be induced to exhibit rover-like feeding and foraging behaviours (Osborne *et al*. 1997; Kaun *et al*. 2007). Similarly, crossing a fly carrying either the GAL4 driver or the UAS activator to a fly containing a wild type 3rd chromosome (identified by +) produces progeny that represent effective controls (w^1; for^s; *elav*-GAL4/+) and (w^1; for^s; +/UAS-*for*T1a). The w^1; for^s; *elav*-GAL4, w^1; for^s; UAS-*for*T1a, and w^1; for^s; + lines were all obtained from M. Sokolowski.

General fly rearing conditions

Stocks were maintained under typical rearing conditions and on standard yeast-sugar-agar media (Belay *et al*. 2007). For the laboratory analyses, lines were reared in sponge-topped 170-mL plastic *Drosophila* bottles (VWR) containing 40 mL of rearing media. For the field study, lines were reared in large population cages (Sterlite, model 1925, 27 L) each containing 9 open-ended culture bottles.

Laboratory dispersal assay

Dispersal in the laboratory was quantified in a simple tube-to-tube assay conducted in a confined room under ambient incandescent light (24 ± 2 °C, 70 ± 5% relative humidity). Arenas consisted of two 50-mL conical tubes (VWR) joined by a 1-mL pipette tip (VWR) midway down the length of the tubes (7 cm)(see Fig. 1a). The shape and placement of the pipette tip prevented flies from returning to the start tube. Two millilitre of either rearing media or agar were poured into the tubes and the open tops were capped with sponge plugs. The arenas were placed flat on a table and the location

and orientation was randomised for each trial. Trials commenced between 1000 and 1200 h with dispersal outcomes determined after 6 h. Dispersal was scored when a fly (or flies) had crossed the pipette tip and fully entered the adjacent tube. All flies were 5- to 7-day-old adults when tested.

We conducted several iterations of this general assay that were each aimed at asking specific questions. To ask whether rovers are more likely to disperse than sitters and sitter mutants, we placed food in both tubes and measured dispersal as the proportion of 32 flies per trial that moved to the adjacent tube (food–food, $N = 25$ replicates per genotype). These assays were conducted on males to avoid females altering the environment (e.g. eggs and larvae). To ask whether rover/sitter variation in dispersal is food dependent, we repeated the above scenario, but in half of the trials we placed agar in the start tube and food media in the adjacent tube (agar-food, $N = 20$ replicates per genotype). The other half contained agar in both tubes (agar–agar, $N = 20$ replicates per genotype). Together, these three assays assessed food-dependent decisions to disperse from a patch and attraction to a new patch. A replicate of the food–food group assay was conducted at a later date to address the effect of sex on rover/sitter dispersal patterns ($20 \leq N \leq 23$ replicates per genotype).

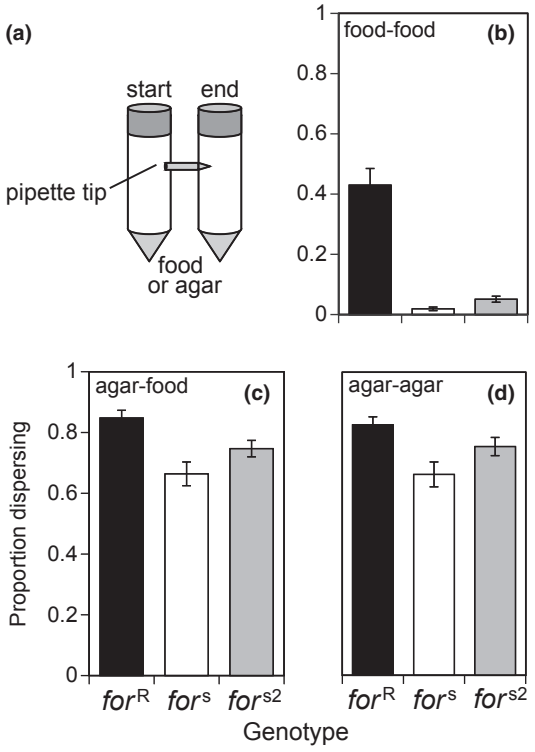

Figure 1 Rover/sitter dispersal tendencies in the laboratory. (a) Schematic of the assay setup. (b–d) Dispersal tendencies of groups of 32 adult male for^R (dark bars), for^s (white bars) and for^{s2} (grey bars) flies. Panels b–d differ in the availability and arrangement of food: (b) food media in both tubes, (c) agar in the start tube and food in the end tube, (d) agar in both tubes. When food is present in the start tube (b), for^R flies tend to disperse from the patch, whereas for^s and for^{s2} tend to remain. The dispersal tendencies of all three genotypes are similarly high when food is absent from the starting tube (c, d). Values represent the mean proportion of flies dispersing ± SEM, (b) $N = 25$ replicate trials per genotype, (c & d) $N = 20$ replicate trials per genotype.

To ask if rover/sitter variation in dispersal persists in the absence of conspecifics, we repeated the food–food assay on single flies (male or female)($32 \leq N \leq 36$ replicates per genotype). To ask if the dispersal tendencies of rovers and sitters are repeatable, we conducted the food–food assay on individual male flies at three different ages during their life: 3, 7 and 11 day post-eclosion ($19 \leq N \leq 21$ replicates per genotype). Finally, to address whether the manipulation of *for* gene expression influences dispersal tendencies we conducted the food–food assay on the individual male and female progeny from three genetic crosses (see Fly Lines and Rearing above) ($62 \leq N \leq 97$ replicates per genotype).

Mark-recapture field experiment

Dispersal was quantified in a 400 m × 400 m open field plot at the University of Toronto's Koffler Scientific Reserve, King City, Ontario, Canada (44°03′ N, 79°29′ W). Baited traps consisted of a small plastic urine cup [90 mL, Starplex (Starplex Scientific, Toronto, ON, Canada)] containing 20 mL of the rearing media described above. Traps were inserted into a wire ring mounted on a 5 cm long bamboo stick. A 2.5 cm wide strip of double-sided tape was wrapped around the stick below the trap mount and the stick was inserted leaving the trap approximately 1 cm above the ground. The tape prevented crawling invertebrates from entering the trap while still being accessible to the fruit flies. A single trap was placed at the epicentre (release trap) and additional traps were distributed along each transect as follows: 2.5, 5, 10, 15, 20, 25, 30 and 40 m from the centre trap. Beyond 40 m, traps were placed every 10 m until they reached a distance of 200 m. In total, there were four 200 m transects radiating in the four cardinal directions from the epicentre. Prior to release and during the entire experiment all traps were continuously exposed. Replacement traps were temporarily inserted while traps containing flies were briefly removed and taken to a counting station located 25 m from the centre trap midway between two transect arms. Media was replaced twice daily or as necessary based on evaporation or soiling. Marking involved shaking the flies in a small amount of dry fluorescent pigment (DayGlo, Cleveland, OH, USA). The flies maintained a pigment badge on their ventral and dorsal thoraces following a grooming period of 24 h. A portable black light was used to visualise the pigments in the field. Pigments were randomly and blindly assigned as for^R (Horizon Blue, A-19), for^s (Saturn Yellow, AX-17-N), and for^{s2} (Aurora Pink, AX-11-5). At 1300 h on 09 September 2010 we released 5971 marked and groomed flies at the epicentre of the plot. All flies were 5- to 8-day-old adults when released. Monitoring of the traps commenced at 0700 h on 10 September 2010 and continued for three consecutive days. Monitoring entailed inspecting each trap every 30 min until dusk (ca. 1930 h). Captured flies were retained to ensure independence in daily recaptures and were released at dusk at their capture trap (Dobzhansky & Wright 1943; Johnston & Heed 1975). The number of recaptures at each trap was summed across days and the four transects were collapsed along a single axis (Dobzhansky & Wright 1943; Johnston & Heed 1975; Kot *et al.* 1996; Rodríguez 2002).

Analyses

The effect of genotype on the proportion of flies dispersing in our group laboratory assay was analysed using ANOVA. Specific differences between genotypes were assessed using a Tukey *post hoc* analysis. Two-Way ANOVA was used to assess the effects of genotype × sex or genotype × food treatment. Arcsine square root transformations were used to normalise the data on the proportion of flies dispersing (Zar 1999); however, we plot the untransformed proportions for ease of interpretation. For laboratory analyses where the unit of replication is the individual fly, we utilised the Chi-Square probability test to analyse the effects of genotype or transgene expression on the propensity to disperse from the start tube.

We estimated the repeatability of dispersal by assessing: (1) whether rovers and sitters differed in their dispersal tendency and (2) whether individuals were repeatable within each genotype. We fit a general linear mixed effects model with a binomial error distribution for each genotype separately including day as a fixed effect and individual ID as a random effect. Day was included to account for changes in movement over time (e.g. habituation or learning effects). The residual variation was used to determine the repeatability (R) for all individuals within genotypes (Nakagawa & Schielzeth 2010).

In the mark-recapture study, we quantified dispersal in nature for each genotype over 3 days by summing recaptures at particular distances radiating from the release site. We fitted a nonlinear least square regression model (Eqn 1) to the cumulative recapture at each distance. In this model, Y is the number of flies, X is the distance travelled, a is the intercept, and b is the slope.

$$Y = a - b\sqrt{log(x)} \quad (1)$$

In spite of its simplicity, this equation sufficiently characterises the dispersal data from several drosophilid species (Wallace 1966; Johnston & Heed 1975; Taylor 1978). We estimated the slope of the fitted distribution as a measure of the dispersal tendency of each of our three fly genotypes. We used the confidence limits for slope estimates to inspect whether the distribution of dispersal distances differed between for^R, for^s and for^{s2} in the direction we predicted. We also used a Tukey test to assess differences in the slopes of the three genotypes. We assessed genotypic differences in the number of flies dispersing short vs. long distances using a one-tailed Fisher's Exact test.

Analyses were conducted using R (v. 2.12.1; R Development Core Team, Vienna, Austria) and JMP 9.0.2 (SAS Institute, Cary, NC, USA).

RESULTS

In the laboratory, we found that male for^R flies dispersed to the new tube in much higher proportions (43 ± 5.5%, mean ± SEM, $N = 25$ replicates) than for^s (1.9 ± 0.6%, $N = 25$) and for^{s2} flies (5.1 ± 1.0%, $N = 25$) (Fig 1b, ANOVA, $F_{2,72} = 50.696$, $P < 0.0001$, Tukey *post hoc*). This reflects genotype-dependent decisions to disperse from a food patch rather than attraction to new patches since assays without food in the start tube yielded high proportions of dispersing flies for all three genotypes (Fig. 1b vs. c,d)(Two-Way ANOVA, Treatment: $F_{2,210} = 300.107$, $P < 0.0001$, Tukey *post hoc*) although overall there was still a genotypic effect across all treatments (Genotype: $F_{2,210} = 51.804$, $P < 0.0001$). This suggests that all three genotypes are highly mobile in the absence of food, but when food is present, movement is inhibited, and inhibited more in for^s and for^{s2} than in for^R flies (Interaction: $F_{4,210} = 8.774$, $P < 0.0001$).

We replicated the food–food assay to now include groups of either males or females. Once again, we found that movement was genotype-specific ($F_{5,119} = 7.131$, $P < 0.0001$) and we were unable to detect an effect of sex ($F_{1.119} = 1.295$, $P = 0.257$) or an interaction ($F_{1,119} = 1.251$, $P = 0.290$). These rover/sitter dispersal differences persisted in the absence of conspecifics. In the food–food assay, individual for^R flies were also more likely to disperse to the new tube (56.3% of $N = 32$ trials) compared to individual for^s (15.1%, $N = 33$) and for^{s2} (16.7%, $N = 36$) (Fig. 2, $\chi^2 = 17.372$, df = 2, $P = 0.0002$).

The dispersal tendencies of individual rovers and sitters were highly repeatable. When individual flies were assayed at 3, 7 and 11 days post-eclosion, for^R flies had a higher tendency to disperse compared to for^s and for^{s2} and these differences were repeatable within individuals (for^R: $R = 0.76$, $N = 21$; for^s: $R = 0.96$, $N = 19$). Due to exceptionally high repeatability in for^{s2} ($N = 20$) resulting in negligible variation between individuals, we were unable to calculate an R-value for this genotype (Nakagawa & Schielzeth 2010). When we statistically controlled for the effect of learning and used AIC to identify the best models, dispersal tendency was consistently genotype-dependent since models not including genotype had greater AIC values (Δ AIC = 17).

Experiments have shown that artificially increasing *for* expression in sitter neurons elicits rover-like PKG levels and behaviour (Osborne *et al.* 1997; Belay *et al.* 2007). When we investigated these same genetic lines and performed the same crosses (see Methods) we found that adult sitter dispersal increased significantly when *for* was artificially over-expressed

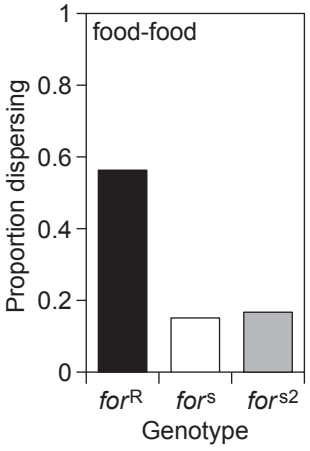

Figure 2 Rover/sitter dispersal tendencies persist in the absence of conspecifics. In single fly trials, a higher proportion of adult for^R flies (dark bars, 18/32 trials) dispersed to the adjacent tube than for^s (white bars, 5/33 trials) and for^{s2} (grey bars, 6/36 trials). Values represent the proportion of trials in which individual flies dispersed.

in neurons (w^1; for^s; *elav*-GAL4/UAS-*for*T1a, 34.8% of $N = 92$ trials) relative to either the GAL4 (w^1; for^s; *elav*-GAL4/+, 21.6%, $N = 97$) or UAS control (w^1; for^s; +/UAS-*for*T1a, 17.7%, $N = 62$)(Fig. 3.; $\chi^2 = 6.894$, df = 2, $P = 0.032$).

We released a similar number of adult flies representing the three genotypes (for^R $N = 1993$, for^s $N = 1991$, for^{s2} $N = 1987$) at the field site (see Methods). As predicted, and similar to our laboratory findings, we found differences in the dispersal of rovers compared to sitters and sitter mutants in nature. The slopes generating curves for for^R, for^s and for^{s2} each significantly differ from zero and provide significant fits to the data (for^R: t = 6.3, df = 5, $P = 0.001$, for^s: t = 7.9, df = 5, $P < 0.0001$, for^{s2}: t = 7.9, df = 5, $P < 0.0001$; Fig. 4). Most importantly, however, the slopes predict different dispersal outcomes that are consistent with our predictions. A Tukey test revealed that the slope of for^R (31 ± 12, 95% C.I.) was significantly different from that of for^s (90 ± 30, $q = 7.44$, $P < 0.01$) and for^{s2} (72 ± 23, $q = 5.163$, $P < 0.01$) but the slopes of for^s and for^{s2} were statistically indistinguishable ($q = 2.277$, $P > 0.05$). Thus, the model for for^R predicted a curve with a lower peak and longer tail than those for for^s and for^{s2}.

Over 3 days, we recaptured fewer for^R flies within 5 m of the central trap (98 recaptures) than for^s (243 recaptures) and for^{s2} (208 recaptures), whereas at distances > 20 m we recaptured significantly more for^R flies (7 recaptures) than for^s (0 recaptures) and for^{s2} (1 recapture)(Fisher's Exact test, $P < 0.0001$)(Fig. 4).

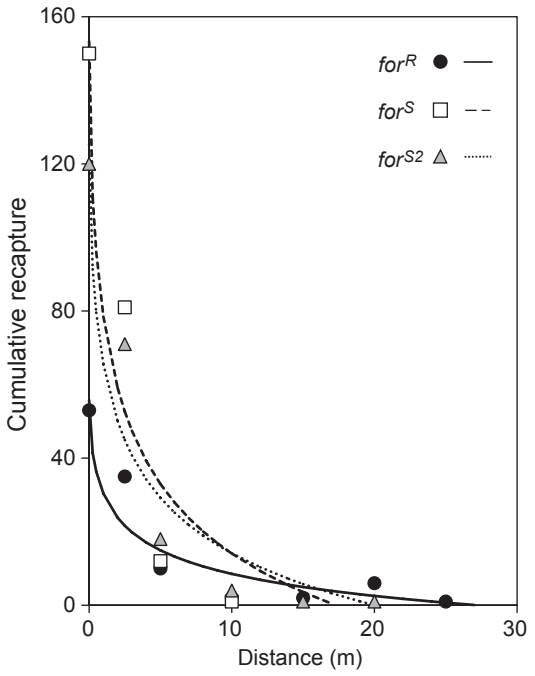

Figure 4 Dispersal heterogeneity in nature is influenced by *for*. The field recapture of adult rovers (for^R, dark circle), sitters (for^s, open square), and sitter mutants (for^{s2}, grey triangle) summed across 3 days. The predicted movement of for^R (solid line), for^s (dashed line), and for^{s2} (dotted line) shows that for^s and for^{s2} contribute mostly to the high peak, whereas for^R contributes most to the tail. A Tukey test revealed that the slope of for^R is significantly different from for^s and for^{s2} while the slopes generated for for^s and for^{s2} are statistically indistinguishable.

DISCUSSION

Congruence between our laboratory and field findings solidifies our interpretation of the effect of *for* on dispersal and supports our hypothesis of a common genetic basis for both foraging and dispersal. A higher level of *for* expression (for^R, rovers) leads to higher foraging activity (Osborne *et al*. 1997). In our study, this scenario led to adult flies with a higher propensity to disperse and to disperse longer distances. The lower *for* expression inherent to sitters (for^s) leads to lower larval foraging activity (Osborne *et al*. 1997) and our study shows that sitter adults also have a lower dispersal propensity and they disperse shorter distances. These differences were both highly repeatable within individuals and persisted in social and non-social contexts (e.g. groups vs. single flies). The for^{s2} mutant line, characterised by sitter-like *for* expression levels and larval foraging behaviour (Osborne *et al*. 1997) also behaved like sitters with respect to adult dispersal. Furthermore, the transgenic over-expression of *for* increased the dispersal tendencies of adult sitter flies. Taken together, our study provides multiple lines of evidence all linking *for* with dispersal.

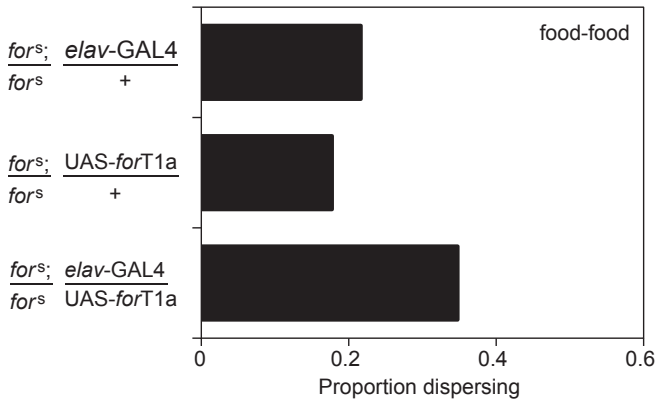

Figure 3 Over-expression of *for* in adult sitter neurons induces rover-like dispersal. The propensity to disperse is increased by transgenic over-expression of *for* in the nervous system of sitter flies using the pan-neuronal *elav*-GAL4 driver (for^s; *elav*-GAL4/UAS-*for*T1a; 32/92 moved). Control crosses containing only GAL4 (for^s; *elav*-GAL4/+; 21/97) or UAS (for^s; +/UAS-*for*T1a; 11/62) had lower dispersal tendencies compared to flies carrying both the GAL4 driver and UAS activator. Values represent the proportion of trials in which individual flies dispersed.

The dispersal distances we observed over 3 days in the field are comparable to those previously reported for *D. melanogaster* (Dobzhansky & Wright 1943; Johnston & Heed 1975). We acknowledge that our experiment was neither intended to understand the specific outcomes of the dispersal we observed nor to measure the range of absolute lifetime dispersal distances for *D. melanogaster*. However, the 3 days of movement we observed provides a 'snapshot' of the variation of dispersal tendencies and distances of rover and sitter flies.

Dispersal tendencies are intimately linked to *for* expression levels in the brain; however, there are likely other contributors including additional genes and the environment. The role of

other candidate genes for dispersal such as *pgi* (lepidopterans, Haag *et al*. 2005) and *npr-1* (nematodes, Gloria-Soria & Azevedo 2008) in *D. melanogaster* warrants further investigation. Environmental factors also affect rover/sitter dispersal tendencies. We observed high dispersal tendencies of for^R, for^s, and for^{s2} in the absence of food suggesting a high basal propensity for dispersal. However, in the presence of food we observed a decrease in dispersal across the genotypes but a drastic decrease in for^s and for^{s2} (ca. 14-fold). Thus, sitter flies have a markedly reduced tendency to disperse from a food patch and our mark-recapture results suggest this could lead to shorter dispersal distances in nature. This food-dependent dispersal provides validation for the conceptual link between foraging and dispersal and also the role of personalities in modulating context-dependent dispersal (Clobert *et al*. 2009; Cote *et al*. 2010). Key future experiments aimed at understanding environmental factors such as patchiness (distance, size, quality), and hunger level are necessary as are those investigating links between dispersal and other personality traits.

Our study sheds light on some of the mechanisms underlying dispersal (i.e. *for*, food-dependence) in *D. melanogaster* but it is difficult at this stage to determine the specific details of rover/sitter movement patterns that lead to the observed differences in the lab and field. Genotypic differences in turning rate, step distance and directionality of movement could explain our observed differences and should be the topic of future study in this system. The movement patterns of rovers and sitters on nutritive and non-nutritive substrates have been characterised previously (for review see Sokolowski 2001; Reaume & Sokolowski 2009); however, these studies focused on net displacement over time. More detailed information on rover/sitter movement patterns in the laboratory could be useful for gaining a better understanding of the magnitude of movements in our field experiment.

Differential effects of marking pigments on survival or recapture could have affected our field results, however, pigments did not affect movement, fecundity, longevity or dispersal in other *Drosophila* studies (Johnston & Heed 1975). In our preliminary analyses, all pigments were clearly visible and unambiguous for over 14 days (laboratory and field enclosures) and maintained through heavy rain (> 3 cm, field enclosure). Although the different colours might have affected visibility to predators, the similarity between our laboratory and field data suggests that any effect of predation is minimal or equal across the colours.

Key aspects of population dynamics (i.e. density and frequency) are important determinants of fitness among genotypes of *D. melanogaster*. Evidence from the rover/sitter system shows that *for* allele frequencies can be shifted by density-dependent selection (Sokolowski *et al*. 1997). Negative frequency-dependent selection via larval competition during food-limited conditions can act as a mechanism to maintain rover/sitter allelic variation in *for* (Fitzpatrick *et al*. 2007). Inherent variation in dispersal tendencies could influence these population dynamics and could play an important role in the maintenance of the rover/sitter polymorphism. For example, we might predict newly established populations, with low connectivity, to have a higher initial representation of for^R. Over time, we would predict an increase in for^s in these populations due to their frequency-dependent advantage in rover-dominated patches (Fitzpatrick *et al*. 2007).

In this study, we demonstrate a clear genetic association between foraging and dispersal that validates theoretical models (Grinnell 1931; Woodbury 1941; Réale *et al*. 2007; Cote *et al*. 2010; Gurarie *et al*. 2010) and we directly implicate *for* as contributor to dispersal tendencies in *Drosophila*. More generally, our findings contribute to the rising interest in merging mechanistic and genetic data with movement ecology (Nathan *et al*. 2008; Wheat 2012). It is possible that the role of *for* in dispersal extends beyond the fruit fly. *for* is known to be involved in the food-related behaviours of several other organisms (reviewed in Reaume & Sokolowski 2009). Furthermore, food-specific changes in the behaviour of dispersal morphs in lizards (Meylan *et al*. 2009) and naked mole rats (O'Riain *et al*. 1996) have been documented. Critically, our work shows that *for*, a gene known to influence foraging behaviour, has pleiotropic effects on dispersal.

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AUTHOR CONTRIBUTIONS

A.H.E. and M.J.F. conceived and designed the field experiment. A.H.E., M.J.F., and A.V. conducted the field experiment. M.J.F. designed the laboratory assay. A.H.E. conducted the transgenic and repeatability experiments. J.H.L. conducted the group lab trials (Fig. 1b-d). M.A. conducted the lab trial with single flies (Fig. 2). M.J.F. and A.H.E. analysed the data and wrote the manuscript. All authors discussed and commented on the manuscript during revisions.

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