Spatial and temporal escape from fungal parasitism in natural communities of anciently asexual bdelloid rotifers

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Sexual reproduction is costly, but it is nearly ubiquitous among plants and animals, whereas obligately asexual taxa are rare and almost always short-lived. The Red Queen hypothesis proposes that sex overcomes its costs by enabling organisms to keep pace with coevolving parasites and pathogens. If so, the few cases of stable long-term asexuality ought to be found in groups whose coevolutionary interactions with parasites are unusually weak. In theory, antagonistic coevolution will be attenuated if hosts disperse among patches within a metapopulation separately from parasites and more rapidly. We examined whether these conditions are met in natural communities of bdelloid rotifers, one of the longest-lived asexual lineages. At any life stage, these microscopic invertebrates can tolerate the complete desiccation of their ephemeral freshwater habitats, surviving as dormant propagules that are readily carried by the wind. In our field experiments, desiccation and wind transport enabled bdelloids to disperse independently of multiple fungal parasites, in both time and space. Surveys of bdelloid communities in unmanipulated moss patches confirmed that fungal parasitism was negatively correlated with extended drought and increasing height (exposure to wind). Bdelloid ecology therefore matches a key condition of models in which asexuals persist through spatio-temporal decoupling from coevolving enemies.

1. Introduction

The prevalence of sexual reproduction is a long-standing puzzle in evolutionary biology [1,2]. Lineages that abandon sex are relieved of the costs of making males, the genetic load of recombination, and the energy and health costs of finding and competing for mates [3]. Nevertheless, obligately asexual plant and animal lineages are very rare, and nearly always short-lived compared with the sexual clades in which they arise [4]. Various explanations have been suggested for the scarcity of asexuals, but current theoretical and empirical evidence supports a key role for antagonistic coevolutionary interactions [5–7]. This ‘Red Queen’ hypothesis posits that parasites and pathogens impose relentless, negative frequency-dependent selection against common host genotypes, which generates continuing advantages for novel, rare genotypes produced by sex, and limits the success of any common asexual clone [8–10].

Empirical support for the Red Queen hypothesis is derived chiefly from observations of relatively short-term dynamics in mixed systems of sexuals and asexuals [7,11–13]. However, there is a complementary way to examine the role of antagonistic coevolution in the long-term maintenance of sexuality. Some obligately asexual lineages are older than others by several orders of magnitude, and a small number have persisted for tens of millions of years without sex [4,14]. If clonality is typically kept in check by rapid coevolution, these long-standing asexuals ought to represent cases in which the selection imposed by biotic enemies has somehow been attenuated. Testing this prediction for specific cases will reveal whether the coevolution hypothesis offers unique insights on the correlates of stable asexuality across geological timescales.
consistent with the role of parasites and pathogens in maintaining sex during shorter-term studies.

The bdelloid rotifers are a class of microscopic freshwater invertebrates that comprises over 450 described species, distributed worldwide [15]. Molecular, fossil and microscopic data support the inference that bdelloids abandoned sexual reproduction at least 30 Ma, making them one of the oldest and best substantiated cases of ‘ancient asexuality’ [16–19]. Rotifers generally are tractable and versatile subjects for ecological and evolutionary inquiries [20], and bdelloids in particular provide an important test case for the prediction that long-standing asexuality should occur in groups with unusual ways to mitigate antagonistic coevolution with parasites and pathogens.

Early in the development of the Red Queen hypothesis, it was recognized that sex is not the only way host populations can keep pace with coevolving enemies [21]. Theoretical models demonstrated that migration can have similar effects under certain conditions [22–24]. Specifically, if hosts exist in metapopulations, and disperse among discrete patches rapidly and independently of parasites, then frequency-dependent selection is readily sustained in the absence of sex, with hosts persisting indefinitely as a cycling mixture of clones. Because these conditions decouple hosts from parasites in space and time, selection arising from antagonistic coevolution is weakened and no longer suppresses asexuality, even when other model parameters are favourable to sex [25].

Despite the recurrence of this ‘spatio-temporal escape’ scenario in several models [22–25], to our knowledge no instance has yet been demonstrated in nature. However, it has been suggested that bdelloid rotifers may fit the requirements [22,26,27]. They are found in almost every freshwater and limno-terrestrial habitat patch worldwide, however tiny, remote or ephemeral. This is due to their small size (typically less than 0.3 mm) and an ability to withstand the complete loss of cellular water at any life stage [28]. When a patch dries out, bdelloids contract to form desiccated ovoid ‘tuns’, which remain dormant until water returns. During an individual’s lifetime, it can withstand multiple episodes of anhydrobiosis, typically lasting days or weeks, but up to 9 years in extreme cases [29,30]. Importantly, tuns are readily transported considerable distances by wind, attached to tiny substrate particles such as fragments of moss or soil [26,31–33]. Each tun represents an independently reproductive propagule that can colonize any suitable habitat in which it lands. Consequently, bdelloid ecology is likely to be characterized by metapopulations spanning considerable areas, with vast numbers of patches and a mechanism enabling rapid dispersal among them [32].

Models of escape depend critically on the ability of hosts to disperse independent of and more rapidly than biotic enemies. It has been suggested that the physically and physiologically stressful processes of extended anhydrobiotic dormancy and dispersal by wind could separate bdelloid rotifers from their parasites in both time and space [22,26,27]. If rotifers survive desiccation better than their enemies, an episode of drought could purge coadapted parasites from a habitat patch, and store host genotypes in a temporal ‘bank’ of tuns, to be re-cycled against different parasites in future wet periods. In addition, if dispersing tuns leave sympatric parasites behind, they could colonize patches where their rare, immigrant genotype is unfamiliar to local enemies, or perhaps establish populations in new patches where parasites are absent. In theory, a combination of temporal and spatial decoupling erodes the advantage of sex under antagonistic coevolution [22–27,34–36]. Indeed, if spatio-temporal escape is very frequent, drought-sensitive parasites may not meet the conditions needed to sustain coevolution with bdelloid rotifers at all.

Evidence to evaluate these possibilities has so far been limited to laboratory studies. The most commonly recorded enemies of bdelloid rotifers are lethal fungal and oomycete parasites (ca 50 species: [37]), chiefly belonging to the genus Rotiferophthora, which preys specifically upon them. It has been demonstrated experimentally that Rotiferophthora angustispora cannot withstand desiccation or disperse by wind to the degree as its bdelloid host, Habrotrocha clausa [26]. Desiccation for 35 days eliminated the fungus from over 90% of cultured rotifer populations, and when infected cultures were desiccated for just 7 days, but also subjected to dispersal in a wind chamber, the prevalence of parasites in newly founded populations was reduced by 60% relative to water-dispersed controls. These results have now been replicated for three bdelloid species in two genera, and five species of fungal parasites [27].

We conducted a study to test whether desiccation-mediated escape also occurs in free-living communities of bdelloid rotifers. First, we carried out manipulative field experiments to determine the ecological relevance of the spatial and temporal escape processes identified in the laboratory. We predicted that desiccating samples collected from the field would reduce the incidence of fungal parasitism in the associated bdelloid communities, and that bdelloids in samples recently blown by wind to elevated habitat patches (mimicking moss on a rock or tree trunk) would have fewer parasites than those colonizing patches at ground level via water. Subsequently, we surveyed unmanipulated habitat patches (moss from different heights on tree trunks) to determine whether spatial and temporal patterns of parasitism in bdelloid communities were consistent with the operation of the hypothesized escape processes. Specifically, we predicted lower prevalence of parasites higher up on trees, where immigration is largely by wind and desiccation is more frequent; we also predicted reduced parasitism at all heights when sampling was preceded by an extended natural drought versus copious rainfall. In both study phases, we compared the frequencies of bdelloid rotifers with those of nematodes, tardigrades and monogonont rotifers, to test the extent to which these other microfaunal groups also can withstand desiccation and colonize elevated microhabitats.

2. Material and methods

(a) Field experiments

Field experiments were conducted at Cornell University’s Experimental Ponds Facility, north of Ithaca, NY, USA (42°30′09″N, 76°26′14″W), between September and October 2009. Forty shallow ponds (20 × 20 × 2.4 m deep) are arranged in a 4 × 10 grid over a flat, treeless area of approximately 150 × 350 m. Roughly 1.5 m inland from the western bank of each pond, a disc of soil ca 150 mm in diameter and 35 mm deep was excavated (figure 1a) and transported to the laboratory along with associated vegetation (moss, lichen, grass, etc.). A 0.1 g sample from each site was plated on a Petri dish and screened microscopically as an initial control to establish the baseline prevalence of bdelloid and monogonont rotifers, nematodes and tardigrades, and the incidence of fungal parasites of bdelloids.
Details of sample preparation and screening are presented in the electronic supplementary material.

To investigate dispersal at ground level, each soil disc was autoclaved and, 5 days later, returned to the field site and embedded at its original location in a plastic dish (150 mm diameter × 35 mm deep) flush with the ground surface. Holes in the bottom and sides of each dish connected the sterilized patch with surrounding groundwater. For the next five weeks, a sample (approx. 16 cm²) of surface material was collected weekly and returned to the laboratory, where a subsample (0.1 g) was screened for microfauna and parasitic fungi (figure 1a). On the fifth week, we also collected undisturbed surface material from 30 cm away, and screened a 0.1 g subsample as a terminal control, to check whether the background prevalence of any groups had changed during the experiment.

To determine the effect of extended desiccation without dispersal, 0.1 g subsamples of material were reserved from the initial and terminal control samples from each site. These were dried and maintained at 40% relative humidity. After 35 days, they were rehydrated with sterile distilled water and screened. Replicate subsamples were screened specifically for fungal parasites using dishes that had been ‘baited’ [37] with laboratory-reared cultures of bdelloid rotifers, so that parasites could be detected even if no bdelloids were naturally present in the rehydrated material (see the electronic supplementary material for further details).

To investigate desiccated dispersal by wind, when the sterilized soil patches were returned to the field, we also installed an elevated, barren habitat patch at each of the 40 sites. A plastic filter funnel was mounted atop a stake, 30 cm above the soil surface and 30 cm away from the sterilized soil disc (figure 1a). The catchment area of the funnel (150 mm in diameter) was equal to that of the soil patch, and open to wind and rain. Into each funnel, we recessed a lidless 88 mm Petri dish of known mass, that of the soil patch, and open to wind and rain. Into each catchment area of the funnel (150 mm in diameter) was equal to the mass of the soil patch, and open to wind and rain. Into each catchment area of the funnel (150 mm in diameter) was equal to the initial and terminal control samples from each site. These were rehydrated with sterile distilled water and screened. Of each sample and used to prepare two mass-standardized 0.1 g subsamples, which were screened for microfauna and fungal parasites in the presence and absence of ‘bait’ rotifers (see the electronic supplementary material for details). Daily temperature and precipitation records were obtained from a weather station 200 m distant.

The 40 days prior to sampling in May 2011 were unusually dry: there was no rain for 21 days, then 16 mm fell in a single day, followed by a further 15 days with minimal precipitation. To assess the effect of this natural period of desiccation, we repeated the height-stratified sampling in September 2011, following a 40 day period with similar temperatures, but nearly five times more rainfall, and no more than four consecutive days without rain. Seven additional trees were sampled in September, to boost the power to detect spatial and temporal patterns (n = 32 trees, 96 samples).

(b) Natural patch surveys
To test whether ‘escape’ dynamics operate in the longer term, and without experimental intervention, surveys of unmanipulated habitat patches were conducted in a protected area of deciduous woodland on the Silwood Park estate near Ascot, Berkshire, UK (51°24′34″N, 0°38′41″W). In May 2011, three samples of moss (approx. 16 cm²) were gathered from each of 25 English oak trees (Quercus robur) within a 250 × 250 m area. Tree trunks were divided into three height zones: low (within 30 cm of the ground), medium (30–70 cm) and high (70–130 cm); and trees were sampled only if they had patches of the moss Hypnum cupressiforme in all zones (figure 1b). Samples were collected at or near the centre of each height zone. Dry mass and water content were recorded for a portion of each sample and used to prepare two mass-standardized 0.1 g subsamples, which were screened for microfauna and fungal parasites in the presence and absence of ‘bait’ rotifers (see the electronic supplementary material for details). To assess the effect of this natural period of desiccation, we repeated the height-stratified sampling in September 2011, following a 40 day period with similar temperatures, but nearly five times more rainfall, and no more than four consecutive days without rain. Seven additional trees were sampled in September, to boost the power to detect spatial and temporal patterns (n = 32 trees, 96 samples).

(c) Statistical analyses
Screening yielded binary data on the presence or absence of various groups of microfauna and on the incidence of fungal parasitism in samples that contained bdelloid rotifers (see the electronic supplementary material, data supplement). Occurrence rates in the field experiments were compared using pairwise contingency tests, adjusting for multiple comparisons using Tukey’s tests of honestly significant differences (HSD; [38, p. 564]) when appropriate. The change in parasitism over time during ground-level recolonization was analysed using binomial logistic regression. Patch surveys involved sampling the same trees at two different times and three different heights. Results were analysed using generalized linear mixed models (GLMMs) with a logit link function, binomial error structure and tree as a random factor. Results from
unbaited and baited screening were analysed separately. Water content of patch samples was analysed using ANOVA after arcsine transformation. Analyses were conducted in R (v. 2.14.2), using the lme4 package to fit GLMMs [39].

3. Results

(a) Field experiments

Desiccation of field-collected material for 35 days significantly reduced the incidence of fungal parasites of bdelloid rotifers (\(p < 0.05\), Fisher’s exact test). Error bars indicate ± 95% Wilson score confidence intervals. Total bar length in the lower panel indicates the number of samples that contained bdelloid rotifers; shaded portions indicate samples in which fungal parasites also were detected. Replicate ‘baited’ screening was conducted for a subset of samples, by adding laboratory-cultured rotifers to enhance parasite detection; again, desiccation significantly reduced parasitism.

There was no change in the background prevalence of bdelloids or their fungal parasites at the field site over the course of the five-week experiment; initial and terminal control samples were almost identical (figure 3). Sterilization of the excavated habitat patches was effective: none of the autoclaved samples contained living animals (data not shown). When the sterilized discs were returned to the field site at ground level, bdelloid rotifers and their parasites returned quickly. Bdelloids reached their initial frequency within two weeks, and their frequency at five weeks (45%) matched that in the undisturbed terminal control samples. The odds of finding parasites approximately doubled for each week the patches remained in place (figure 3, binomial logistic regression: \(\log (p/(1-p)) = -4.69 + 0.755 \times \text{week}; \text{odds ratio: 2.13, 95\% confidence interval (CI) = [1.15-3.94, d.f.} = 1, n = 90, z = 2.4, p = 0.016)\); by week five, parasitism in the sterilized patches had returned to the level seen in the control samples (27.8%). Thus, rates of dispersal via groundwater were not detectably different between hosts and parasites over 35 days.

Results from the corresponding elevated habitat patches differed markedly (figure 3). After five weeks, bdelloids were present in 87.5% of dishes, which was significantly higher (by nearly two times) than in any ground-level sample (Zar’s test of multiple proportions, using Tukey’s HSD, wind versus controls, \(q_{3,157} = 6.76, p < 0.001\); wind versus week 5 wet, \(q_{3,157} = 5.85, p < 0.001\)). However, only one elevated sample (2.9%) contained parasites. This is lower by nearly an order of magnitude than parasite incidence in the ground-level samples—including initial and terminal control samples (HSD, \(q_{3,86} = 3.94, p = 0.018\) and samples collected after five weeks of wet recolonization (HSD, \(q_{3,86} = 3.6, p = 0.034\)). The incidences of fungal parasitism in the elevated wind dispersal and static desiccation treatments both were significantly lower than in ground-level control samples (figure 4; desiccation: HSD, \(q_{3,128} = 3.46, p = 0.041\); wind: HSD, \(q_{3,128} = 3.94, p = 0.017\), but the two treatments did not differ significantly from each other (HSD, \(q_{3,128} = 0.96, p = 0.776\)). Reduced parasitism in the elevated habitat patches was probably due in part to the effects of desiccation: bdelloids must have arrived as anhydrobiotic tunns, and the new populations experienced further bouts of desiccation \textit{in situ} because rainfall was intermittent over the course of the experiment (for weather data, consult the electronic supplementary material, figure S1). Although there were never more than seven consecutive dry days in the field, the parasite reduction in elevated samples matched that observed after 35 days of static experimental desiccation (figure 4). This reinforces the results of earlier laboratory experiments in which even short bouts of desiccation, combined with the physical stress of wind transport, generated a substantial dispersal asymmetry in favour of bdelloids, and ensured that most newly founded populations were parasite-free [26,27].

The three most speciose bdelloid families occurred in all treatment groups: Habrotrochoidea was the most common, followed by Adinetidae and Philodiniidae (see the electronic supplementary material, figure S2). Relative representation of these families did not differ significantly among treatments (\(\chi^2 = 11.9, n = 325, p = 0.064\)). Five genera of fungal parasites were recorded (see the electronic supplementary material, figure S3). Rotiferophthora was most common (42.6% of all parasite records), followed by Pseudomeria, Harposporium, Triactitus and Pochonia. All five are lethal,
and bdelloid abundance in screening dishes rapidly decreased once epidemics began. No significant differences were found in the relative representations of fungal genera among treatments.

Other microfauna were also recorded during the field experiments (figure 4). Nematodes were present in 63.8% of ground-level control samples, but their frequency was significantly lower (37%) in the desiccated samples (HSD, \( z_{3,194} = 4.63, p = 0.004 \), and significantly lower again (7.5%) in the elevated (wind-dispersed) samples (HSD, \( z_{3,194} = 5.35, p < 0.001 \)). Nematodes were significantly more prevalent than bdelloids in control samples (FET, \( n = 160, p = 0.026 \)), and returned rapidly to sterilized, ground-level habitat patches (data not shown). However, nematodes were significantly less prevalent than bdelloids in the desiccated (FET, \( n = 154, p < 0.001 \)) and elevated samples (FET, \( n = 80, p < 0.001 \)). Monogonont rotifers and tardigrades were recorded too rarely to judge their response to the treatments, except that their prevalence was always significantly lower than that of bdelloids (\( p < 0.001 \) in all contrasts).

(b) Patch surveys

Because of heavy precipitation in the summer of 2011 (figure 5a), the water content of moss patches on tree trunks was significantly higher in September than in May (figure 5b, ANOVA, \( F_{1,167} = 70.45, p < 0.001 \)). In both periods, water content was significantly lower in moss patches that were higher up on trees (ANOVA, \( F_{2,167} = 16.53, p < 0.001 \)), because they were more exposed to wind and farther removed from ground-level moisture. The effects of date and height did not interact significantly. Neither the mean daily temperature (\( t = 1.58, p = 0.12 \)) nor the variance in temperature (\( F_{40,40} = 0.59, p = 0.1 \)) differed significantly between the 40 day periods preceding the two sampling dates (figure 5b).

Bdelloids were less frequently parasitized in samples from moss patches higher up trees than lower down, in both unbaited and baited screens (figure 5c). For each nominal step down the tree trunk (approx. 30 cm), the odds of finding parasites in unbaited screens approximately doubled (see the electronic supplementary material, table S1; GLMM binomial, odds ratio: 2.00, 95% CI = 1.08–3.70, d.f. = 1, \( n = 160, z = 2.21, p = 0.027 \)), and the odds for baited screens increased even more steeply (GLMM binomial, odds ratio: 3.44, 95% CI = 1.89–6.26, d.f. = 1, \( n = 171, z = 4.05, p < 0.001 \)). The odds of finding parasites also increased significantly in September relative to May (figure 5c) in unbaited (GLMM binomial, odds ratio: 5.64, 95% CI = 1.81–17.54, d.f. = 1, \( n = 160, z = 2.99, p = 0.003 \)) and baited screens (GLMM binomial, odds ratio: 129, 95% CI = 21.1–789, d.f. = 1, \( n = 171, z = 5.26, p < 0.001 \)). There was no significant interaction between height and date. Five genera of fungal parasites were recorded in the moss samples from tree trunks (see the electronic supplementary material, figure S3). Once again, Rotiferophthora was the most common (72.5% of records), followed by Harposporium,
**Pseudomeria, Pochonia** and the oomycete genus *Haptoglosa*. The relative representation of genera did not differ significantly among heights or between dates, suggesting that escape is effective against multiple genera of fungal and oomycete parasites, including the two most common and speciose groups affecting bdelloids (*Rotiferophthora* and *Harposporium*).

Bdelloid rotifers were found in 100% of samples at all heights, except samples collected near ground level in September when 41% of samples did not contain bdelloids. This implies that rotifer abundance was significantly reduced in September than in May, but mean and variance of daily temperatures did not differ significantly. (See the electronic supplementary material, figure S4; HSD, $\eta_{9,171} > 5.35$, $p < 0.004$ versus any other sample set). There were no significant differences in the relative representations of bdelloid families among heights or between dates (see the electronic supplementary material, figure S5; $\chi^2_{10} = 6.86$, $n = 297$, $p = 0.74$).

Nematodes were present in 57% of samples overall and were significantly less common than bdelloids in all samples except those collected near ground level in September (see the electronic supplementary material, figure S4; FETs, $p < 0.05$). Nematode occurrence did not differ significantly between the two sampling dates but was significantly lower in higher patches (binary logistic regression; $\log(p/(1-p)) = 1.07–0.398 \times \text{height}$; odds ratio: 0.67, 95% CI = 0.46–0.98, d.f. = 1, $n = 171$, $z = -2.07$, $p = 0.039$). There was no evidence that the presence of nematodes affected bdelloid frequencies. Nematodes were equally frequent near ground level in May and September, but only in September, when fungal parasitism was at a maximum, did the frequency of bdelloids drop significantly. Monogonont rotifers and tardigrades were recorded too rarely to detect spatial or temporal patterns (see the electronic supplementary material, figure S4).

### 4. Discussion

Studies conducted over relatively short timescales have indicated that intense coevolutionary interactions with parasites and pathogens facilitate the maintenance of sexual reproduction in host organisms [7,12,13]. A corollary prediction of this hypothesis is that the few obligately asexual lineages that have persisted across substantial evolutionary timescales should possess unusual characteristics that attenuate the impact of antagonistic coevolution [4,40,41]. Theoretical models have highlighted a scenario in which hosts become spatio-temporally decoupled from parasites by dispersing rapidly among discrete habitat patches [21–25]. Our study evaluated whether natural communities of anciently asexual bdelloid rotifers meet a critical condition for these models: dispersal in time and space that is independent of major parasites.

Experimental desiccation of field samples for 35 days decreased fungal parasitism in associated bdelloid communities by a factor of four, and new populations founded by wind dispersal of desiccated propagules were nine times less likely to contain parasites than those founded simultaneously by dispersal in groundwater. Temporal and spatial escape also was evident in surveys of unmanipulated habitat patches on tree trunks. Following a protracted natural drought, fungal...
parasites were rarely detected, except in moss near the base of trees where the moisture content was highest, and rotifer immigration occurs readily by water, without desiccation. Patches higher up on trees, which receive bdelloid immigrants as wind-blown tuns but not through groundwater, were virtually parasite-free. Later the same year, following a period with equivalent temperatures but fourfold higher rainfall, parasites were five times more common overall, and their occurrence again was negatively related to height.

Taken together, these results indicate that extended desiccation of habitat patches provides natural communities of bdelloid rotifers with a temporal escape from fungal parasites, which are more sensitive to drought. This enables recovering rotifers to avoid renewing interactions with the same antagonists. Wind dispersal offers a further escape in space, so that desiccated bdelloids can leave infested habitats and disperse independently of their parasites to found healthy populations elsewhere. Habitat patches 30–130 cm above ground are readily colonized and desiccate frequently; they therefore serve as refugia from the fungal parasites that afflict bdelloid populations at ground level. Suitable habitat patches are available at considerably greater heights in most biomes, and bdelloids have been recovered from wind socks at least 16 m above the ground [31]. Our data encompass communities containing three major bdelloid families, whose relative representation did not vary significantly among experimental treatments, nor among the natural patch samples. This indicates that the ability to survive desiccation and disperse rapidly on the wind is widespread among bdelloids rather than restricted to a specialist subset, and reinforces other studies concluding that desiccation tolerance is an ancient and nearly ubiquitous trait in the class Bdelloidea [30,42].

Among other microfauna recorded in our study, the facultatively sexual monogonont rotifers can survive desiccation only as resting eggs. Adults require stable aquatic habitats, and their low prevalence in our surveys suggests they do not thrive in the wind-dispersed limno-terrestrial metapopulations favoured by bdelloids. Only tardigrades and some nematode species share the capacity for anhydrobiosis at any life stage [43], and they do not disperse as readily as bdelloids on the wind [31]. Sohlenius & Bostrom [44,45] sampled isolated limno-terrestrial habitat patches in the Antarctic, where dispersal is primarily by wind and extended desiccation is commonplace. Results from this remote and inhospitable region are especially informative, as tardigrades conjectured to mitigate the costs of asexuality and help bdelloid rotifers avoid the typical fate of clonal lineages [46]. Most of these invoke consequences of desiccation at the genetic level, such as incorporation of exogenous DNA or removal of deleterious elements during repair of chromosome damage after rehydration [18]. Our results indicate that the ecological consequences of desiccation also bear importantly on bdelloid asexuality, in the context of theoretical treatments linking sex, antagonistic coevolution and dispersal asymmetry. If an unusual combination of spatial and temporal escape has reduced the selective impact of parasites on bdelloid rotifers, their exceptionally long-lasting asexual reproduction could be accommodated within the explanatory domain of the Red Queen hypothesis, reinforcing a link between coevolutionary dynamics and the maintenance of sexual reproduction.

The small size of bdelloid rotifers and their tolerance for extreme physical stress allows them to reach and exploit isolated, miniscule and ephemeral habitat patches, and to disperse among these patches repeatedly and more rapidly than other microfauna. However, their most commonly recorded fungal parasites are not able to tolerate desiccation for the same duration, and they rarely accompany dormant bdelloids that travel on the wind to colonize new habitats. This ecology satisfies a key condition of models for the long-term maintenance of asexuality under antagonistic coevolution, in which hosts are disengaged from parasites in time and space [22–25]. In these models, parasites are present in all habitat patches, and impose frequency-dependent selection on a mixture of host clones with differing resistance alleles. If dispersal is independent and asymmetric, as shown here for bdelloids and fungal parasites, host diversity is preserved because a temporarily disfavoured clone can escape locally adapted parasites and establish in a new patch where parasite virulence alleles are poorly matched or where no parasites exist. Indeed, our results also indicated that habitat patches suitable for bdelloid rotifers are sometimes free of fungal parasites entirely, probably owing to recent extended desiccation or isolation by height. The effect of such refugia was not examined in the original models, but their presence should extend the range of conditions under which asexuality is favoured. For instance, even if parasites have broader host ranges than envisioned in escape models based on ‘matching alleles’ [22], asexual hosts may persist if they have ready access to parasite-free patches. Further theoretical and empirical work will be necessary to fully resolve the coevolutionary mosaic in the rotifer–fungus system. In particular, the population dynamics of rotifer clones should be examined within and among patches to gather direct evidence of frequency-dependent selection [11–13], and to test whether it is indeed attenuated in patches where rotifers are ‘high and dry’.

Desiccation tolerance has been linked with several mechanisms conjectured to mitigate the costs of asexuality and help bdelloid rotifers avoid the typical fate of clonal lineages [46]. Most of these invoke consequences of desiccation at the genetic level, such as incorporation of exogenous DNA or removal of deleterious elements during repair of chromosome damage after rehydration [18]. Our results indicate that the ecological consequences of desiccation also bear importantly on bdelloid asexuality, in the context of theoretical treatments linking sex, antagonistic coevolution and dispersal asymmetry. If an unusual combination of spatial and temporal escape has reduced the selective impact of parasites on bdelloid rotifers, their exceptionally long-lasting asexual reproduction could be accommodated within the explanatory domain of the Red Queen hypothesis, reinforcing a link between coevolutionary dynamics and the maintenance of sexual reproduction.

Acknowledgements. We thank G. Masters, R. I’Ansen Price, S. Frost and N. Brito-Rivera for laboratory assistance; R. Johnson for field site management; O. Moore for moss identification and N. G. Hainston Jr, H. Kern Reeve, T. Barralough, D. Fontaneto, T. Cooper and several anonymous reviewers for helpful comments.

Funding statement. Financial support was provided by an EMBO Long-Term Fellowship (ALTF 733–2010) to C.G.W., the Department of Neurobiology and Behavior at Cornell, Sigma Xi and a Stephen H. Weiss Presidential Fellowship to P.W.S.
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