A model on the evolution of cryptobiosis

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Cryptobiosis is an ametabolic state of life entered by some lower organisms (among metazoans mainly rotifers, tardigrades and nematodes) in response to adverse environmental conditions. Despite a long recognition of cryptobiotic organisms, the evolutionary origin and life history consequences of this biological phenomenon have remained unexplored. We present one of the first theoretical models on the evolution of cryptobiosis, using a hypothetical population of marine tardigrades that migrates between open sea and the tidal zone as the model framework. Our model analyses the conditions under which investments into anhydrobiotic (cryptobiosis induced by desiccation) functions will evolve, and which factors affect the optimal level of such investments. In particular, we evaluate how the probability of being exposed to adverse conditions (getting stranded) and the consequences for survival of such exposure (getting desiccated) affects the option for cryptobiosis to evolve. The optimal level of investment into anhydrobiotic traits increases with increasing probability of being stranded as well as with increasing negative survival effects of being stranded. However, our analysis shows that the effect on survival of being stranded is a more important parameter than the probability of stranding for the evolution of anhydrobiosis. The existing, although limited, evidence from empirical studies seems to support some of these predictions.

Introduction

Cryptobiosis is the collective name for an ametabolic state of life utilized by some organisms to overcome periods of unfavourable environmental conditions (Keilin 1959, Crowe 1975, Wright *et al.* 1992, Kinchin 1994). Cryptobiotic organisms are known from both the plant and animal kingdom, but in animals only among invertebrates (Wright *et al.* 1992). The main factors inducing the cryptobiotic state are desiccation (anhydrobiosis), freezing (cryobiosis) and oxygen deficiency (anoxybiosis, Wright *et al.* 1992). Organisms with cryptobi-

otic abilities are often separated into those in which the cryptobiotic state may be entered only within a specific (ontogenetic) life stage, and those that may enter cryptobiosis over the entire life cycle (Crowe 1971, Wright *et al.* 1992). The first category includes species from the taxa Arthropoda, Crustacea, Brachiopoda, Insecta, spores of various fungi and bacteria, and pollen and seeds of some plants, while the second category mainly includes species from Protozoa, Rotifera, Nematoda, Tardigrada, and various species of mosses, lichens and algae, as well as some higher plants (Keilin 1959, Wright *et al.* 1992).
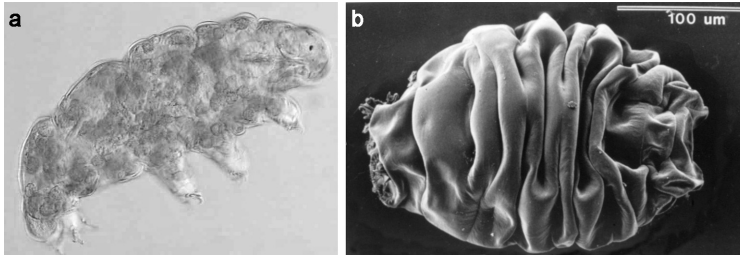
Fig. 1. Tardigrade (**a**) in a hydrated active state (light microscopy photo by K. I. Jönsson) and (**b**) in a desiccated anhydrobiotic state (SEM photo by R. M. Kristensen).

In the cryptobiotic state, several fundamental biological characteristics are affected. Since all metabolic processes have stopped, reproduction, development, and repair are prevented. Therefore, as a life history component cryptobiosis is mainly characterized by survival. However, to successfully achieve a high survival in the cryptobiotic state, the organism has to make biochemical preparations of the body, such as the production of carbohydrates to replace the lost water in anhydrobiotic organisms (e.g., Crowe 2002). Such processes are energetically costly (Jönsson & Rebecchi 2002), and will withdraw resources from potential use in other life history functions, e.g., reproduction. Thus, most likely there are trade-offs between investment in cryptobiotic functions and investment in other life history functions.

Tardigrades (phylum Tardigrada, Fig. 1) are well known for their cryptobiotic capacity (e.g., Wright *et al*. 1992) and occur in a variety of ecosystems, some of which represent the most extreme natural habitats on earth (e.g., continental Antarctica; Sømme & Meier 1995). They represent an important component of the meiofauna in habitats exposed to desiccation, and may survive for several years in a cryptobiotic state (Jönsson & Bertolani 2001, Guidetti & Jönsson 2002).

Although the ability of tardigrades to enter cryptobiosis has been known for a long time, and has been subject to numerous investigations (*see* Wright *et al*. 1992, Wright 2001), the evolutionary background to this ability has not been studied much. May (1951) suggested that marine tardigrades were the ancestors of terrestrial and limnic tardigrades, and that the transition from marine to terrestrial forms was accompanied by the acquisition of a tolerance to anoxia and desiccation, and ultimately of cryptobiosis. If this scenario is correct, the initial evolutionary steps towards cryptobiotic tardigrades were prob-

ably taken in populations living in seashore and tidal habitats, where great variation in the water availability created a strong selection for desiccation tolerance. Some tardigrade populations of the genus *Echiniscoides* and *Archechiniscus* inhabiting littoral marine environments show a capacity to survive desiccation (Grøngaard *et al*. 1990, Wright *et al*. 1992), whereas other marine tardigrades cannot do so.

In this paper, we present a theoretical model on the evolution of anhydrobiosis (cryptobiosis induced by desiccation). The model describes a hypothetical population of an ancient marine tardigrade inhabiting the littoral zone, where the risk of exposure to desiccation creates a force of selection for anhydrobiotic ability. The main purpose of the analysis is to evaluate two general aspects of anhydrobiotic selection: the probability of being exposed to a time period of dry environmental conditions, and the consequences on survival of such exposure. Based on our analysis, we propose a set of conditions that should favour energy investments in traits that improve the animal's anhydrobiotic capacity.

The model

General model scenario

Our starting point is a non-anhydrobiotic population of ancient marine tardigrades that migrates (actively or passively) between the tidal zone and permanent water. Such migratory behaviour could arise from generally better foraging conditions in the tidal zone. Consequently, the organisms forage in the tidal zone during high tide, and return to the non-tidal zone when the water withdraws in order to avoid stranding and subsequent desiccation. We assume that egg laying in these marine tardigrades takes place in open

water and that eggs are not able to migrate to the tidal zone during high tide (in tardigrades, eggs are either laid freely in the substrate or within the moulted cuticle (Bertolani 1983)). The reproductive cycle of our model population then begins with a prebreeding period of foraging in the tidal zone followed by migration to open water. During the period of foraging and resource accumulation, eggs are developed. If the population succeeds in migrating back to open water, the animals moult and lay their eggs, and in the following postbreeding period they migrate back to the tidal zone to start a new period of foraging. However, the population runs the risk of being exposed to a dry period (low tide) during which the animals cannot return to open water and which exposes them to desiccation stress. The model assumes no parental care.

The model scenario corresponds well to a marine environment with tidal changes in water availability, e.g., the supposed environment under which the first steps towards anhydrobiosis in tardigrades may have been taken (May 1951). However, the scenario could just as well represent the conditions under which anhydrobiosis evolved in any other metazoan. Also, because the general life history effects should be similar regardless of the factor (desiccation, freezing, anoxia) inducing a cryptobiotic state, our analysis could well be adopted as a general analysis on the evolution of cryptobiosis.

Fitness function

Let us denote survival over the prebreeding period by P_1, survival over the postbreeding period P_2, and reproductive output, i.e. the number of surviving offspring at each reproductive occasion, by R. Given our ecological scenario, and assuming that fertility and adult survival are constant over adult age classes, the fitness (λ) can be approximated by the function

$$\lambda = P_1(R + P_2) \qquad (1)$$

This fitness function is an extension of the one proposed by Charnov and Krebs (1974), and is generally applicable for reproductive cycles in which two temporal phases, each with a separate

survival parameter, may be distinguished (*see* Jönsson *et al.* 1995a, 1995b, 1998). A deduction of function (Eq. 1) from the Euler–Lotka equation is provided in the Appendix. The point of offspring release, which in the present case is equivalent to egg-laying, separates the two phases.

Now assume the existence of within population genetic variation in the ability to survive partial or complete desiccation, generating continuous variation in traits related to this ability. Such traits may include storage of energy, synthesis of protectant molecules or enzymes, cuticular structures reducing transpiration, aggregation behaviours, or any other trait that increase the stress tolerance of the animal (Wright *et al.* 1992, Jönsson 2001). We call these traits "anhydrobiotic traits" and assume that the ability to survive a period of desiccation depends on the amount of resources invested into such traits. The magnitude of these investments is here denoted by K, which is the phenotypic trait that we evaluate below.

We assume that anhydrobiotic traits incur costs in terms of a reduction in survival under non-desiccating conditions by withdrawing resources from this life history trait. Survival may then be modelled by a function P_i that depends on K so that

$$\frac{dP_i(K)}{dK} < 0$$

Moreover, investment into anhydrobiotic traits is assumed to divert resources from reproduction, making reproductive output, R, a decreasing function of K so that

$$\frac{dR(K)}{dK} < 0$$

Inserting the functions of survival and reproductive output into fitness function (Eq. 1) gives the following fitness for a migrating animal

$$\lambda = P_1(K)[(R(K) + P_2(K)] \qquad (2)$$

However, this is the fitness for animals that succeed in returning to the non-tidal area after foraging. If they fail in this process, they become stranded and must wait until the high tide returns before they can migrate to open water. If the population is stranded, survival decreases sig-

nificantly. Still, those individuals with anhydrobiotic traits will gain an advantage since survival for stranded animals, P_1', increases with K so that

$$P_1'(K) = 1 - \frac{c}{\alpha K + 1} \qquad (3)$$

Equation 3 describes a survival function that increases asymptotically towards 1 as K increases towards infinity. Here, c is a parameter between 0 and 1 indicating the survival cost for a stranded non-anhydrobiotic animal. A high value of c implies that being stranded is devastating for the stranded individual, and for $c = 1$ it will die if no investments are made into anhydrobiosis ($K = 0$). On the other hand, a low value of c implies that being stranded does not affect survival very much. Note that stranding does not necessarily mean that the animal gets desiccated, only that it cannot return to the moist area. Thus, c reflects the probability of being desiccated for a stranded non-anhydrobiotic organism. The parameter α converts K to a dimensionless entity in order to make survival a probability. We assume in the analysis below, for the sake of simplicity, that $\alpha = 1$. Note also that we do not take the length of the period in anhydrobiosis into account, implying either that this period is a constant, or that the length of the period is short enough not to influence survival.

When water returns in the tidal area, anhydrobiotic individuals rehydrate and migrate to the non-tidal area where they reproduce. The survival of an animal that forages in the tidal area, gets stranded, and returns to the area of permanent water is then

$$P_1 P_1' = P_1(K)\left(1 - \frac{c}{\alpha K + 1}\right) \qquad (4)$$

As a consequence, fitness of an animal that becomes stranded is

$$\lambda = P_1(K)\left(1 - \frac{c}{\alpha K + 1}\right) \times \left[R(K) + P_2(K)\right] \qquad (5)$$

Environmental heterogeneity

If the probability of being stranded is q, the chance of safe return to open water is $(1 - q)$, i.e., a proportion q of all visits to the tidal area

ends up in stranding for the population. Since we consider a temporal heterogeneity that affects the whole population and that spans several reproductive cycles we believe that the geometric mean fitness is the proper fitness measure in this type of model (for discussion on geometric mean fitness, *see* Philippi and Seger (1989)). The use of an arithmetic mean would imply that the populations of tardigrades experience spatial heterogeneity, i.e. a proportion of the population gets stranded whereas the other individuals do not. In this model, however, we are only focusing on temporal heterogeneity and hence we use the geometric mean fitness. The expected fitness in our hypothetical population is then

$$\lambda = \left\{ P_1(K)\left(1 - \frac{c}{\alpha K + 1}\right)\left[R(K) + P_2(K)\right]\right\}^q \\ \times \left\{ P_1(K)\left[R(K) + P_2(K)\right]\right\}^{(1-q)} \qquad (6)$$

Under these circumstances, a phenotype with no anhydrobiotic investments ($K = 0$) that gets stranded will have a zero fitness as long as death is certain to a stranded non-anhydrobiotic individual, i.e. $c = 1$. In the long run, it also implies that when $c = 1$, a population of non-anhydrobiotic individuals ($K = 0$) will become extinct when there is a risk of being stranded. This seems to be a plausible consequence of the system outlined above.

Below we analyse the above fitness function for decreasing functions of $P_i(K)$ and $R(K)$ with three curvatures, i.e. linear, concave and convex functions. We also assume that pre- and post-breeding survival is equally affected by K, i.e. $P_1(K) = P_2(K)$. Our analysis should constitute a sufficient basis for drawing general conclusions about the conditions for the evolution of anhydrobiosis. For linear and concave functions, we present graphical analyses because analytical analyses become exceedingly complex and difficult to interpret.

Results

Linear and exponential effects

Assuming that survival as well as reproduction decrease linearly with investment of resources in
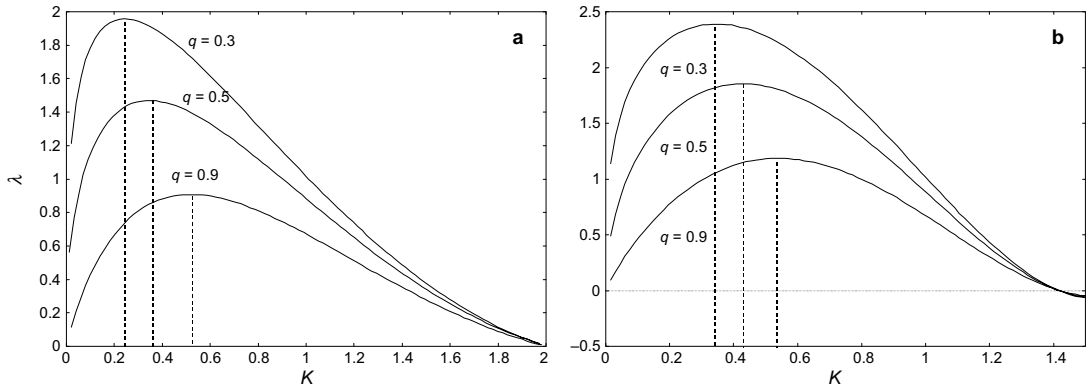
Fig. 2. Fitness (λ) plotted against investments in anhydrobiotic structures (K) for three values of the probability of getting stranded (q) when (**a**) survival and reproduction functions are linearly decreasing ($r = 3$, $\alpha = 1$, $\beta = 0.5$, $\gamma = 1$, $c = 1$), and (**b**) survival and reproduction functions are concavely decreasing ($r = 3$, $\alpha = 1$, $\beta = 0.5$, $\gamma = 1$, $z = 2$, $c = 1$).

anhydrobiotic structures, the simplest case may be described by functions of the sort

$$P_i(K) = 1 - \beta K$$

and

$$R(K) = r - \gamma K$$

where β (the marginal decrease in survival due to investment into anhydrobiosis) converts K into a dimensionless entity, and γ is a parameter that converts K into the number of surviving young. In the analysis, we restrict our case to the situation when $\beta = 0.5$, i.e. we will not here analyze variations in the survival cost function. We also assume that $\gamma = 1$, i.e. one unit of K represents one surviving young less. The parameter r determines the reproductive output of an animal with no investments into anhydrobiosis whatsoever and this parameter, together with β and γ, determines how the effects of anhydrobiotic investments affect reproduction in comparison to the effects on survival. If β and γ are held constant then a higher value of r means less impact on reproduction from investment into anhydrobiotic structures as compared with the impact on survival.

We also evaluate the possibility that anhydrobiotic investments will have an exponentially increasing negative effect on survival as well as on reproduction. The functions will then be concave and the easiest way of obtaining such curvature is by adding an exponential to K so that

$$P_i(K) = 1 - \beta K^z$$

and

$$R(K) = \mathrm{r} - \gamma K^z.$$

In the equations above, β, γ and r have the same meaning as in the case of linear effects whereas the parameter z determines the curvature of the functions (the higher value of z, the more dramatic are the effects of an increase in K). As in the linear case, we will let $\beta = 0.5$, $\gamma = 1$ while we let $z = 2$.

Under these assumptions the fitness function (Eq. 6) may be plotted against K, which will reveal the optimum investments into anhydrobiosis. Figure 2 shows fitness as a function of K for three values of the probability of getting stranded, q, and for linear and concave survival/reproduction functions, respectively. The panels indicate that optimal investment in anhydrobiotic structures should increase as q increases. They also suggest that the probability of getting stranded does not have to be particularly high for anhydrobiosis to evolve, as long as the effects of getting stranded are devastating ($c = 1$ in the panels).

In Fig. 3, fitness is plotted against K for three values of c. The probability of getting stranded is in both cases 0.7, i.e. 70% of the visits to the tidal zone result in stranding. Still, this is not enough to promote anhydrobiotic investments when survival and reproduction decrease linearly with K if c is sufficiently low (Fig. 3a). These effects are not so obvious when we assume concave functions. However, in both cases optimum K increases as c increases (Fig. 3).
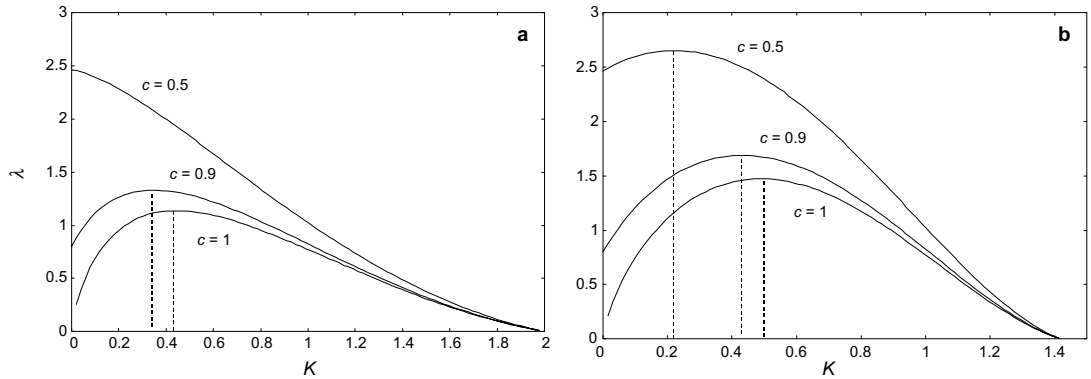
Fig. 3. Fitness (λ) plotted against investments in anhydrobiotic structures (K) for three effects on survival if not entering anhydrobiosis when stranded (c) when (**a**) survival and reproduction functions are linearly decreasing ($r = 3$, $\alpha = 1$, $\beta = 0.5$, $\gamma = 1$, $q = 0.7$), and (**b**) survival and reproduction functions are concavely decreasing ($r = 3$, $\alpha = 1$, $\beta = 0.5$, $\gamma = 1$, $z = 2$, $q = 0.7$).
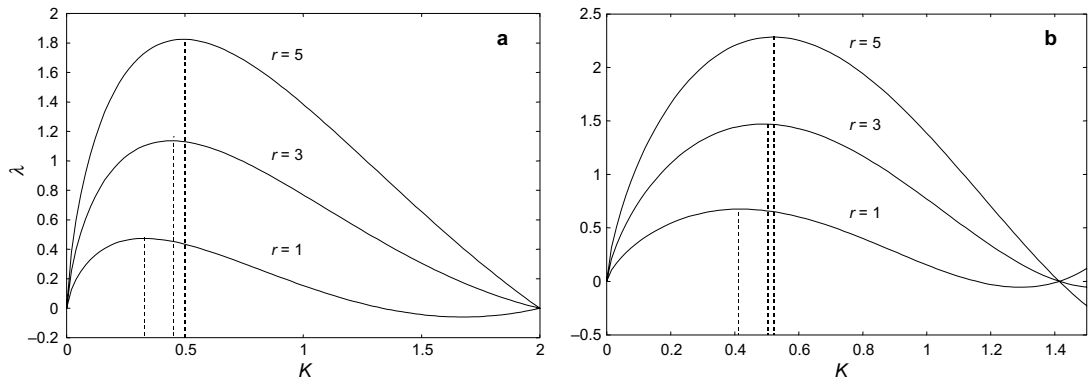
Fig. 4. Fitness (λ) plotted against investments in anhydrobiotic structures (K) for three values of the effects on reproductive output (r) when (**a**) survival and reproduction functions are linearly decreasing ($\alpha = 1$, $\beta = 0.5$, $\gamma = 1$, $c = 1$, $q = 0.7$), and (**b**) survival and reproduction functions are concavely decreasing ($\alpha = 1$, $\beta = 0.5$, $\gamma = 1$, $z = 2$, $c = 1$, $q = 0.7$).

The effects of reproductive output, r, are presented in Fig. 4. Here there is a tendency for increased investment into anhydrobiosis as its relative effects on potential reproductive output decreases (K increases as r increases). Optimal anhydrobiotic investment should thus be higher in phenotypes with high original reproductive output.

Diminishing effects

Finally we evaluate the case when survival and reproduction functions are slightly convex. This could be accomplished by simply setting the above parameter z to a value below one. How-ever, we are interested in finding an analytical solution to the problem so instead we chose the following set of functions.

$$P_i(K) = 1 - \frac{\beta K}{\beta K + 1}$$

$$R(K) = \frac{r}{\gamma K + 1}$$

In these equations, both β and γ convert K to a dimensionless entity in order to make P_i and R currencies for survival and reproductive output, respectively. As before, r is reproductive output when no investments are made into anhydro-biosis. A great advantage with these functions is that they allow analytical investigation when

assuming that $\gamma = 1$ and $\beta = 1$. Differentiating Eq. 6 with respect to K and solving for optimum K yields

$$\hat{K} = 0.5qc - 1 + c \qquad (7)$$

Inserting $\hat{K}$ into the second derivative of Eq. 7 gives us the following expression

$$\frac{d^2\lambda}{dK^2} = -\frac{32}{81}\frac{r+1}{q^4 + c^4} \qquad (8)$$

which is always negative, implying that $\hat{K}$ represents a maximum.

Equation 7 provides a number of interesting results. First, if $c = 1$ and $q > 0$, i.e. if there is any probability of being stranded and stranding is equivalent to death to a non-anhydrobiotic organism, anhydrobiotic investment will always be selected for ($\hat{K} > 0$). This result was indicated also for linear and concave functions. Second, $\hat{K}$ increases as the risk of being stranded increases. Third, as the survival cost of being stranded (c) increases, optimal investment into anhydrobiotic traits ($\hat{K}$) increases. Fourth, the probability of being stranded (q) together with the effect of being stranded (c) must be high enough to satisfy the inequality $c(0.5q + 1) > 1$ in order for any anhydrobiotic investments to be advantageous. In conclusion, for anhydrobiosis to evolve at all, i.e. $\hat{K} > 0$, either both q and c have to be rather high, or $c = 1$ and $q > 0$. Even with a risk of being stranded equal to 1, i.e. all visits to the tidal zone end up with stranding, anhydrobiosis will only be an optimal strategy if the reduction in survival when being stranded is relatively high, i.e. $c \geq 2/3$ in the model above.

We may plot $\hat{K}$ as a function of both q and c as in Fig. 5. The largest investment into anhydrobiotic traits is found under circumstances where stranding implies death for a non-anhydrobiotic individual ($c = 1$) and stranding is inevitable ($q = 1$). At the other extremes, i.e. when $c = 0$ and $q = 1$, or when $c = 1$ and $q = 0$, we expect no anhydrobiotic investments at all (Fig. 5). Another important message from Eq. 7 and Fig. 1 is the confirmation that c is the major determinant of the magnitude of anhydrobiotic investments. This result is indicated by the steeper slope of $\hat{K}$ as a function of c, as compared with $\hat{K}$ as a function of q (Fig. 2 and Eq. 9)
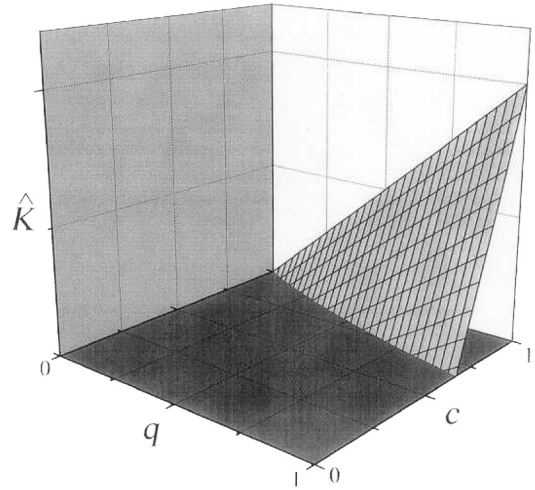

Fig. 5. Optimum anhydrobiotic investment, $\hat{K}$, as a function of the probability of being stranded, q, and the reduction in survival when stranded, c.

$$\frac{d\hat{K}}{dc} = 0.5q + 1 > 0.5c = \frac{d\hat{K}}{dq} \qquad (9)$$

Discussion

Given the assumptions of our model, it is clear that anhydrobiosis will evolve under two conditions: (i) when both the probability of stranding and the survival cost paid by a stranded individual is relatively high, or (ii) under low probability of stranding if the cost of being stranded is detrimental. In addition, our analysis suggests that the effect, rather than the risk, of being stranded is the crucial parameter for the evolution of anhydrobiosis. Formulated more generally, this suggests that cryptobiosis may evolve (i) when both the probability of experiencing adverse conditions and the survival cost of such conditions are high, or (ii) when survival costs are very high despite a low probability of adverse conditions. These results emerged from the analysis with convex functions of survival and reproduction, and under rather specific assumptions ($\alpha = 1$, $\beta = 1$, $\gamma = 1$). However, also in the case of linear functions, in spite of a risk of 70% of getting stranded ($q = 0.7$) even a 50% risk of dying if not entering anhydrobiosis ($c = 0.5$) will not suffice to promote any investments into anhydrobiotic structures (Fig. 3a).

When functions are concave, the requirements for investing in anhydrobiotic traits seem to be less restrictive. This is not surprising since concavity implies that minor initial investments impose low costs on survival and reproduction. The result that the effect, rather than the probability, of experiencing adverse conditions is the more important parameter in promoting the evolution of cryptobiosis has an important implication. It suggests that cryptobiosis may be promoted also in environments with relatively infrequent events of adverse conditions. According to this prediction anhydrobiosis could, therefore, be widespread among organisms even in terrestrial habitats that dry out relatively rarely. We know of no data evaluating this prediction.

A quantitative result of our analysis is that the optimal investment into cryptobiotic traits should increase with increasing probability of adverse conditions as well as with increasing negative survival effects of such conditions. As a prediction from this result we would expect cryptobiotic organisms in different environments to allocate different relative amounts of energy to cryptobiotic functions. We may also expect differences in cryptobiotic capacity (e.g. in the ability to survive rapid desiccation) among organisms adapted to different environments. In line with these predictions, Grøngaard *et al.* (1990) showed a qualitative difference in anhydrobiotic capacity in the littoral tardigrades *Echiniscoides sigismundi* and *Echiniscoides hoepneri*. Both species are found in the upper littoral zone, but the former species inhabits more exposed sites (e.g., among barnacle plates) and survives desiccation, while the latter species inhabits more sheltered sites (e.g. within barnacles) and cannot survive desiccation. The first attempts by marine non-cryptobiotic tardigrades to forage in the tidal zone were probably made fairly close to the border of the open sea. In this environment, the risk of being stranded is low but still exists, whereas the effect of being stranded would be just as devastating as further up on the shore. This circumstance would be enough for a mutant tardigrade with some investment in cryptobiotic traits to gain a selective advantage over non-cryptobiotic individuals. Figure 2 shows that as the probability of being stranded increases, there is a gradual increase in optimal anhydrobiotic investment. Hence, forag-

ing attempts further and further into the dry area would select for phenotypes with increasingly higher investment into anhydrobiotic traits.

In our analysis we have concentrated on the initial stage in the evolution of cryptobiosis, when anhydrobiotic populations evolved from non-anhydrobiotic populations. Once anhydrobiosis had evolved, the transition towards populations inhabiting pure terrestrial habitats exposed to occasional or regular periods of desiccation may have been relatively rapid. To some extent, our results should apply also to this phase in the evolution of anhydrobiotic behaviour, with the probability of being stranded turning into a general risk of dehydration. Our results will then indicate that terrestrial organisms living in habitats that always dry out within a reproductive cycle ($q = 1$, $c = 1$) will evolve a high anhydrobiotic capacity, whereas organisms living in habitats that dry out only occasionally (e.g., $q = 0.5$, $c = 1$) will evolve a lower capacity of anhydrobiosis. In fact, according to our analysis we would expect a gradual increase in investments to anhydrobiotic traits as the risk of desiccation increases (Fig. 5 and Eq. 7). To the extent that anhydrobiotic capacity is directly related to energy invested into this trait, this prediction is supported by studies on tardigrades (Wright 1991) and nematodes (Williams 1978, Solomon *et al.* 1999) showing that anhydrobiotic capacity is related to the desiccation conditions in the natural habitat of a species. We may also expect that optimal energy investment into anhydrobiotic traits over a reproductive cycle increases with the frequency of dry periods expected within the cycle. Thus, in habitats where periods of desiccation occur almost on a daily basis, investment into anhydrobiotic traits (e.g., protectant molecule production; Westh & Ramløv 1991, Wright *et al.* 1992) should be high.

In the analysis with linear and concave survival/reproduction functions, an increase in r yielded an increase in optimum K (Fig. 4). This would suggest that an organism with high original fecundity should invest more in cryptobiotic functions than an organism with low original fecundity subject to the same environmental conditions. Hence, the general reproductive strategy (high or low fecundity) should influence the optimal allocation to cryptobiotic functions.

However, this was not the case for the specific convex functions that we used, where *r* did not appear in the optimality condition (Eq. 7). Thus, the curvature of the trade-off functions between cryptobiotic investment and reproduction and survival in the hydrated state may be important for predicting optimal investments into cryptobiosis. The shape of these trade-offs has to our knowledge never been investigated in cryptobiotic organisms, but linear or concave functions may seem more likely to expect than convex functions. With convex functions, the marginal effects of cryptobiotic investment on reproduction and survival in the hydrated state will decline the more the organism invests in cryptobiotic functions, which is probably not a generally expected condition. One could, however, envisage a system where the initial investment involves forming of new structures that have large effects on organism morphology/physiology or behaviour. Such initial investments could reduce survival and reproduction significantly. In such a case, additional investments into cryptobiotic capacity would only imply an increase in some quantitative trait or adjustments of the system that may have a less adverse effect on survival and reproduction. Consequently, convex trade-off functions should not be ruled out completely. A concave relationship between *K* and anhydrobiotic survival seems plausible though, since investment in cryptobiotic functions (e.g. production of protectant molecules) above a certain level will have smaller and smaller effects on survival.

From our assumed trade-off between cryptobiotic and reproductive investments we would also expect an inverse relationship between cryptobiotic capacity and reproductive investments. Organisms living in exposed habitats will thus be expected to invest a large proportion of available resources into cryptobiotic functions and thus have low fecundity as compared to non-cryptobiotic organisms living in constantly favourable (moist) environments. At present, there is no data available to evaluate these predictions. Overall, it would be of great interest to compare frequencies of adverse conditions, cryptobiotic capacity and life history patterns in cryptobiotic organisms inhabiting different habitats, to the results obtained from this study.

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Appendix

Assume that survival between two breeding events contains two components, post- and prebreeding survival, and that the point at which offspring survival become independent of parent survival demarcate these two periods. With the notations P_1 and P_2 for pre- and postbreeding survival, respectively, the Euler–Lotka equation may be expressed as follows (*see also* Jönsson *et al.* 1995)

$$\sum_{x=1}^{n} \lambda^{-x} m(x) \left(\prod_{i=0}^{x} P_{1i} \right) \left(\prod_{j=0}^{x-1} P_{2j} \right) = 1 \tag{A1}$$

where x is age class and $m(x)$ is fecundity at age class x. Let us further assume that our organism reproduces after a juvenile stage plus a prebreeding period so that survival before first breeding is $P_{10} P_{20} P_{11}$, and that fecundity is constant over age-classes. The Euler–Lotka equation above then converges to

$$m P_{10} P_{20} P_{11} \sum_{x=1}^{n} \lambda^{-x} \left(\prod_{i=0}^{x-1} P_{1i} \right) \left(\prod_{j=0}^{x-1} P_{2j} \right) = 1 \tag{A2}$$

Assuming that adult survival is constant over ages, i.e. $P_{11} = P_{12} = \ldots = P_{1n}$ and $P_{21} = P_{22} = \ldots = P_{2n}$, equation A2 becomes equivalent to

$$m P_{10} P_{20} P_1 \sum_{x=1}^{n} \lambda^{-x} P_1^{x-1} P_2^{x-1} = 1 \tag{A3}$$

which is equal to

$$\frac{m P_{10} P_{20} P_1}{P_1 P_2} \sum_{1}^{n} \sum_{x=1} \lambda^{-x} P_1^x P_2^x = 1 \tag{A4}$$

Here the term $m P_{10} P_{20}$ is the reproductive output at each reproductive event, in our model denoted by R. Equation A4 can be rewritten as

$$\frac{R}{P_2} \sum_{x=1}^{n} \lambda^{-x} P_1^x P_2^x = 1 \tag{A5}$$

The geometric sum of A5 can be simplified as

$$\frac{R}{P_2} \left[\frac{1 - \left(\frac{P_1 P_2}{\lambda} \right)^n}{1 - \left(\frac{P_1 P_2}{\lambda} \right)} - 1 \right] = 1 \tag{A6}.$$

Now let n approach infinity — recalling that we assume constant age-specific survival — and the equation A6 can be simplified and rewritten as

$$\frac{R}{P_2} = \frac{\lambda}{P_1 P_2} - 1 \tag{A7}$$

which gives our fitness function, $\lambda = P_1(R + P_2)$.