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Received Date : 03-Feb-2016

Revised Date : 08-Jun-2016

Accepted Date : 21-Jun-2016

Article type : Standard Paper

Editor : Bethany Hoyer

**High adult mortality in disease-challenged frog populations increases vulnerability to drought**

**Running head:** High disease mortality reduces niche width

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This is the author manuscript accepted for publication and has undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the [Version of Record](#). Please cite this article as [doi: 10.1111/1365-2656.12569](https://doi.org/10.1111/1365-2656.12569)

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## Summary

1. Pathogen emergence can drive major changes in host population demography, with implications for population dynamics and sensitivity to environmental fluctuations. The amphibian disease chytridiomycosis, caused by infection with the fungal pathogen *Batrachochytrium dendrobatidis* (*Bd*), is implicated in the severe decline of over 200 amphibian species. In species that have declined but not become extinct, *Bd* persists and can cause substantial ongoing mortality. High rates of mortality associated with *Bd* may drive major changes in host demography, but this process is poorly understood.
2. Here, we compared population age structure of *Bd*-infected populations, *Bd*-free populations, and museum specimens collected prior to *Bd* emergence for the endangered Australian frog, *Litoria verreauxii alpina* (alpine tree frog). We then used population simulations to investigate how pathogen-associated demographic shifts affect the ability of populations to persist in stochastic environments.
3. We found that *Bd*-infected populations have a severely truncated age structure associated with very high rates of annual adult mortality. Near-complete annual adult turnover in *Bd*-infected populations means that individuals breed once, compared with *Bd*-free populations where adults may breed across multiple years.
4. Our simulations showed that truncated age structure erodes the capacity of populations to withstand periodic recruitment failure; a common challenge for species reproducing in uncertain environments.
5. We document previously undescribed demographic shifts associated with a globally emerging pathogen and demonstrate how these shifts alter host ecology. Truncation of age structure associated with *Bd* effectively reduces host niche width, and can help explain the contraction of *L. v. alpina* to perennial waterbodies where the risk of drought-induced recruitment failure is low. Reduced capacity to tolerate other sources of mortality may explain variation in decline severity among other chytridiomycosis-challenged species and highlights the

55 potential to mitigate disease impacts through minimising other sources of  
56 mortality.

57

## 58 **Key-words**

59 Amphibian declines, chytrid fungus, demography, environmental stochasticity, life-history,  
60 niche contraction, population dynamics, recruitment failure

61

## 62 **Introduction**

63 Pathogens are emerging as a key threat to biodiversity and have been implicated in  
64 recent severe declines in amphibians, bats, bees and corals (Daszak, Cunningham & Hyatt  
65 2000; Fisher *et al.* 2012). This threat is increasing with growth in international wildlife trade,  
66 and is amplified by biodiversity loss (Keesing *et al.* 2010; Martel *et al.* 2014). New pathogens  
67 can drive major shifts in host demography when their impact is variable across different host  
68 ages. For example, age truncation has been observed following disease emergence in perch  
69 (*Perca fluviatilis*), where maximum age decreased by approximately 15 years, along with a  
70 substantial reduction in mean age (Ohlberger *et al.* 2011). Similarly, Tasmanian devil facial  
71 tumour disease is associated with the loss of individuals older than two years in affected  
72 Tasmanian devil (*Sarcophilus harrisii*) populations, causing severe age structure truncation  
73 (Lachish, Jones & McCallum 2007; Lachish, McCallum & Jones 2009). Pathogens can also  
74 affect sex ratios, with Phocine Distemper virus causing higher male than female mortality in  
75 European harbour seals (*Phoca vitulina*) (Härkönen *et al.* 2007). In plants, fungal pathogens  
76 that cause seedling mortality, but do not kill mature individuals, can reduce conspecific  
77 recruitment, enabling other species to establish and changing community structure (Packer &  
78 Clay 2000). Disease-induced changes in host demography, such as those described above,  
79 can subsequently impact on host population dynamics (Saether & Bakke 2000), life-history  
80 (Gandon, Agnew & Michalakis 2002), genetic diversity (Romiguier *et al.* 2014), and  
81 sensitivity to environmental fluctuations (Rouyer *et al.* 2012). Thus, understanding the  
82 demographic impacts of diseases can help assess the vulnerability of populations to  
83 secondary threats.

84 Amphibians are the vertebrate taxon most threatened by disease (Skerratt *et al.* 2007;  
85 Wake & Vredenburg 2008), however, research on the demographic effects of pathogen  
86 emergence in frogs remains limited. Chytridiomycosis, caused by infection with the fungal  
87 skin pathogen *Batrachochytrium dendrobatidis* (*Bd*) (Berger *et al.* 1998), is the worst  
88 recorded disease afflicting biodiversity (Skerratt *et al.* 2007; Wake & Vredenburg 2008). The  
89 global emergence of chytridiomycosis has been implicated in the apparent extinction of 113  
90 species and major declines of a similar number of additional species (Skerratt *et al.* 2007;  
91 Fisher, Garner & Walker 2009). The emergence of *Bd* in susceptible amphibian communities  
92 is associated with rapid increases in prevalence and infection intensity, followed by mass  
93 mortality (Lips *et al.* 2006; Rachowicz *et al.* 2006; Vredenburg *et al.* 2010). However, while  
94 *Bd* is implicated in species extinctions, populations of many susceptible, declined species  
95 now persist with the pathogen in an endemic state (Retallick, McCallum & Speare 2004;  
96 Briggs, Knapp & Vredenburg 2010; Scheele *et al.* 2014a). Endemic *Bd* infection remains a  
97 major source of mortality in populations of some declined amphibian species across multiple  
98 continents (Murray *et al.* 2009; Pilliod *et al.* 2010; Muths, Scherer & Pilliod 2011; Phillott *et*  
99 *al.* 2013; Scheele *et al.* 2015).

100 Despite the widespread occurrence of *Bd*, the effects of *Bd* infection on amphibian  
101 population age structure have not been investigated. High rates of ongoing mortality could  
102 result in age structure truncation, compromising a species' capacity to tolerate other sources  
103 of mortality, leading to decreased population stability and increased extinction risk (Berkeley  
104 *et al.* 2004). Many amphibian species reproduce in uncertain environments and are reliant on  
105 relatively high adult survival to buffer against year-to-year variation in recruitment success  
106 (Wilbur & Rudolf 2006). In particular, loss of individuals capable of reproducing over  
107 multiple years may erode the capacity of species to tolerate drought-induced recruitment  
108 failure, which is a common stressor in amphibian populations (Wilbur 1980; Wilbur &  
109 Rudolf 2006). Similarly, a disproportionate loss of adults may increase vulnerability to other  
110 threats, such as tadpole predation by introduced predators (Gillespie 2001). An analogous  
111 example comes from heavily exploited fisheries, where age structure truncation (Olsen *et al.*  
112 2004), predisposes populations to collapse through increased dependence on recruitment,  
113 which can be strongly influenced by environmental fluctuations (Rouyer *et al.* 2012).

114 Here, we address two knowledge gaps: (1) how endemic *Bd* affects amphibian  
115 population age structure, and (2) how changes in population age structure influence the  
116 capacity of species to tolerate other sources of mortality; namely, periodic recruitment failure

117 associated with drought. We studied the endangered frog *Litoria verreauxii alpina* (alpine  
118 tree frog; Watson, Loftus-Hills and Littlejohn 1971), a species that was historically common  
119 throughout sub-alpine regions of Australia, but experienced major declines in the 1980s  
120 associated with the emergence of *Bd* (Osborne, Hunter & Hollis 1999; Scheele *et al.* 2015).  
121 In *L. v. alpina*, endemic *Bd* is associated with high annual adult mortality, but importantly,  
122 adults breed before succumbing to chytridiomycosis and recruitment remains sufficient to  
123 facilitate population persistence in some locations (Scheele *et al.* 2015).

124 We examined how chytridiomycosis affects adult population age structure by  
125 comparing long exposed *Bd*-infected *L. v. alpina* populations, extant *Bd*-free populations and  
126 museum specimens collected from now extinct populations prior to *Bd* emergence in the  
127 1980s. We then used matrix population models to investigate how age structure truncation  
128 associated with chytridiomycosis affects population resilience to drought-induced  
129 fluctuations in recruitment. We hypothesised that age structure truncation would result in  
130 reduced population viability in response to drought, because periodic recruitment failure  
131 associated with El Niño weather cycles is an important stressor in amphibian populations in  
132 eastern Australia (Osborne 1989). Our study provides a novel perspective on the  
133 demographic and ecological consequences of emerging pathogens, and can help increase our  
134 understanding of these processes in other species affected by emerging diseases worldwide.

135

## 136 **Materials and methods**

### 137 **Study species**

138 *Litoria v. alpina* is a spring breeding amphibian that reproduces in seasonal and  
139 perennial ponds in sub-alpine regions of south-eastern Australia. Following snow melt, male  
140 breeding aggregations form and females arrive throughout the spring. Eggs are deposited  
141 below the water surface attached to aquatic vegetation. During the non-breeding season,  
142 adults are widely dispersed from breeding habitat and occupy grassland and woodland. Sub-  
143 adults also occupy terrestrial habitat following dispersal from maternal ponds in late summer.  
144 *Litoria v. alpina* experienced major declines in the 1980s associated with *Bd* emergence. It is  
145 now restricted to a small number of perennial ponds, and is absent from previously occupied  
146 ephemeral pond habitat (Osborne, Hunter & Hollis 1999) where El Niño-associated droughts  
147 can result in early pond drying and reproduction failure (Osborne 1989).

148

**149 Field sampling**

150 We sampled populations across the species' entire range; our data set included seven  
151 extant *Bd*-infected populations, two extant *Bd*-free populations and preserved museum  
152 specimens from three now extinct populations housed at the Museum Victoria. Information  
153 on population location, sample size and date of sampling are provided in Table S1 (Appendix  
154 S1). Snout-urostyle length (SUL) was measured for all individuals with dial calliper (0.1 mm  
155 precision) and a single toe removed at the base of the third phalange for age determination.  
156 The majority of individuals sampled were adult males (Table S1, Appendix S1). Sex was  
157 determined by the presence or absence of male nuptial pads, gravidness and/or size;  
158 individuals >32 mm SUL without nuptial pads were recorded as female. Three individuals  
159 <32 mm SUL, but larger than the smallest gravid female were conservatively classified as  
160 sub-adults, but may represent non-gravid females. Across all populations and years, eighteen  
161 sub-adults were sampled; all of which were one year of age, with the exception of one two  
162 year old museum specimen.

163 In 1996, toe samples were collected from individuals from two extant *Bd*-infected  
164 populations and frogs were individually marked to allow identification. Recaptures were  
165 attempted in 1997 (six nights of survey per population) and new individuals were also  
166 sampled. In 2011, six extant *Bd*-infected populations and two extant *Bd*-free populations were  
167 sampled. The two *Bd*-free populations sampled are the only known naïve populations and  
168 occur in geographically isolated areas. In 2012, we conducted surveys to recapture  
169 individuals from which a toe was removed in 2011 and sample new individuals. In total, four  
170 nights of survey were undertaken in each population in 2012, with each survey involving two  
171 or three experienced personnel searching all potential habitat within a site for five to seven  
172 hours (see Scheele *et al.* 2015 for details).

173 Museum specimens originate from three populations. Each of the museum  
174 populations were sampled in a single night in breeding habitat. Although detailed collection  
175 notes are not available, it is unlikely that any size bias was present in this sample (e.g. larger  
176 males targeted for collection) because males, females and sub-adults were collected. Museum  
177 specimens were fixed in formalin and stored in ethanol. Identification numbers for museum  
178 specimens, along with age and size data for all individuals, are provided in Scheele *et al.*  
179 (2016).

180

181

182

### 183 ***Batrachochytrium dendrobatidis* sampling**

184 In 2011 and 2012 we sampled all individuals (except the museum specimens and four  
185 additional individuals) for *Bd* infection using sterile swabs (Medical Wire & Equipment Co.  
186 MW 100–100). Sampling was conducted in a standardised way with three strokes on each  
187 side of the abdominal midline, the inner thighs, hands and feet. A new pair of disposable  
188 powder-free nitrile gloves was used for each frog. To prevent *Bd* spread we washed and dried  
189 waders, boots and clothing between populations, and sterilised all field equipment. Samples  
190 were analysed using real-time quantitative polymerase chain reaction following the  
191 methodology of Boyle et al. (2004), with the exception that the highest concentration  
192 standard used was 16667 zoospore equivalents (*zse*), restricting maximum *zse* values to  
193 16667 as we did not extrapolate *zse* values outside the range of our standards. In 2012, a  
194 Qiagen master mix was used instead of Taqman and *zse* estimates were not constrained.  
195 Samples were analysed in triplicate and were considered positive if all three wells returned a  
196 positive reaction (Hyatt *et al.* 2007). Standard histological methods were used to confirm *Bd*  
197 presence in both populations sampled in 1996 and 1997 (Berger *et al.* 1998). Museum  
198 specimens were not sampled for *Bd* as they were collected (1969-1974) prior to amphibian  
199 declines in the study region during the 1980s (Osborne, Hunter & Hollis 1999). The earliest  
200 retrospective detection of *Bd* in Australia is from 1978 (Australian Government 2006) and  
201 testing of museum *Pseudophryne corroboree* specimens – a species that was sympatric with  
202 *L. v. alpina* – collected from the study region failed to detect *Bd* prior to 1980 (Hunter *et al.*  
203 2010). We refer to the populations represented in the museum collection as ‘pre-decline’, but  
204 suggest that they are also *Bd*-free and accordingly combine them with the extant *Bd*-free  
205 populations in our analyses and simulations.

206

### 207 **Age determination**

208 Skeletochronology was used to determine the age of 594 individuals from seven *Bd*-  
209 infected and two *Bd*-free extant populations and three pre-decline populations preserved in

210 museum collections (Table S1, Appendix S1). A rotary microtome was used to section  
211 decalcified bone embedded in paraffin wax. Sections were stained using haematoxylin and  
212 lines of arrested growth were counted using a microscope (see Scheele *et al.* 2015 for  
213 details). Skeletochronology is a reliable method for aging amphibians in regions that  
214 experience strong, consistent seasonal variations in climate, leading to the formations of  
215 clearly discernible lines of arrested growth (Smirina 1994), and has effectively been used in  
216 *L. v. alpina* (Scheele *et al.* 2015).

217

### 218 **Statistical analysis**

219 To investigate whether population *Bd* status and elevation were associated with adult male  
220 age we used a generalised linear mixed model with a Gamma distribution in the R package  
221 lme4 (Bates, Maechler & Bolker 2012; R Development Core Team 2014). The dependent  
222 variable was age, with population *Bd* status (presence-absence; museum and extant *Bd*-free  
223 populations combined) and elevation as fixed effects and population as a random effect. We  
224 constructed a set of candidate models arising from all combinations of the explanatory  
225 variables, including an interaction between population *Bd* status and elevation. We then used  
226 an information-theoretic model selection process to rank models based on their Akaike's  
227 Information Criterion value with a correction for small sample size (AICc) using the R  
228 package MuMIn. We considered models with  $\Delta\text{AICc}$  values  $<2$  of the best ranked model  
229 (Burnham & Anderson 2002).

### 230 **Population modelling**

231 We used Lefkovich matrix models to investigate the effects of age structure  
232 truncation on the capacity of populations to tolerate recruitment failure. We combined our  
233 demographic data across sampled populations to create two generalised scenarios to examine  
234 responses of 1) *Bd*-infected and 2) *Bd*-free/pre-decline *L. v. alpina* populations to drought-  
235 induced reproduction failure. We used pre-breeding female based models and assumed a 1:1  
236 sex ratio and equivalent vital rates between sexes. We first created a matrix model based on  
237 demographic information from *Bd*-free and pre-decline populations. The matrix contained  
238 seven stages; pre-sub-adult, sub-adult and five breeding adult stages (Table S2, Appendix  
239 S1). Because age to maturity in *L. v. alpina* varies, pre-sub-adults were able to transition  
240 either into sub-adult or breeding adult one. Sub-adults were able to transition into breeding



241 adult stages two and three (Table S2, Appendix S1). Adult survival rates were calculated  
242 from the combined age structure data of the *Bd*-free and pre-decline populations. This method  
243 involved comparing the proportion of individuals of age  $x$  in a given year with the proportion  
244 of individuals in the same cohort in the subsequent year (aged  $x + 1$ ) (Caughley 1977). Using  
245 this method it was not possible to estimate the survival of one and two year old adults due to  
246 variation in age at maturity. Thus, we assumed the same rate of annual survival between one  
247 and two, and two and three year olds as three and four year olds. The two *Bd*-free and three  
248 pre-decline populations used to calculate adult survival estimates were sampled over four  
249 different years, reducing the likelihood that stochastic events in any one year would have a  
250 substantial influence on age structure. The pre-sub-adult stage incorporates clutch size, egg,  
251 tadpole and metamorph survival to year one. Clutch size was obtained from the literature  
252 (Anstis 2002). Egg to adult survival was unknown and was incrementally adjusted to achieve  
253 a stable age distribution that matched the observed age structure of *Bd*-free/pre-decline  
254 populations (Table S2, Appendix S1). We then created a second matrix model to emulate the  
255 effect of age structure truncation resulting from *Bd* infection in these populations. This  
256 involved enforcing 100% mortality after first breeding, consistent with no recorded  
257 recaptures across years in *Bd*-infected populations (see Results section) (Table S3, Appendix  
258 S1).

259 In our study region, El Niño-associated droughts can result in early pond drying and  
260 complete reproductive failure of spring breeding amphibians (Osborne 1989). Previous  
261 research documented a contraction of *L. v. alpina* to habitat with perennial ponds, with the  
262 species absent from previously occupied ephemeral pond habitat (Osborne, Hunter & Hollis  
263 1999). Ephemeral pond habitat is vulnerable to drought-induced recruitment failure, while  
264 perennial ponds retain water throughout droughts. Our simulations investigated whether this  
265 habitat contraction could be explained by age structure truncation associated with *Bd*  
266 emergence decreasing the capacity of *L. v. alpina* to persist in ephemeral habitats that  
267 experience recruitment failure.

268 Simulations were conducted in the software package RAMAS GIS version 5. We  
269 used four different scenarios to investigate the effect of age structure truncation on population  
270 response to periodic recruitment failure. Each scenario was run for a 100 year period with  
271 1000 iterations (stochastic models) and an initial population size of 2000 and a population  
272 ceiling of 4000. The first two scenarios were deterministic with one based on the *Bd*-free/pre-  
273 decline matrix (Table S2, Appendix S1) and the other on the *Bd*-infected matrix (Table S3,

274 Appendix S1) described above. The next two scenarios were stochastic and incorporated  
275 periodic recruitment failure into the *Bd*-free/pre-decline and *Bd*-infected matrices to simulate  
276 the effects of drought. Probability of recruitment failure was stochastic (0.125 chance of  
277 complete recruitment failure in any one year) and was based on observations of premature  
278 pond drying over the period 1997-2013 (premature pond drying occurred in 2002 and 2006).  
279 In years with recruitment failure, no individuals were recruited into the population, but adult  
280 survival remained unchanged.

281

## 282 **Results**

### 283 **High burden of disease in *Bd*-infected populations**

284 *Batrachochytrium dendrobatidis* infection prevalence was 70% (95% confidence  
285 intervals = 64.5-75.2%) in 292 individuals sampled across seven infected populations. The  
286 mean infection intensity in infected individuals was 3774 (SE = 578) *zse*. *Batrachochytrium*  
287 *dendrobatidis* was absent from 103 individuals sampled in two extant *Bd*-free populations.

288

### 289 **Reduced adult longevity in *Bd*-infected populations**

290 Males from *Bd*-infected populations had reduced longevity (mean age = 1.97; SE  $\pm$   
291 0.01) compared with *Bd*-free/pre-decline populations (mean age = 3.07; SE  $\pm$  0.08) (Fig. 1).  
292 The best supported model demonstrated a strong negative association between male age and  
293 population *Bd* status (Table S4, Appendix S1). The second ranked model included population  
294 *Bd* status and elevation and had a  $\Delta$ AICc value of 2.03. The addition of elevation had  
295 minimal effect on maximized log-likelihood values (-351.684 versus -351.683; see Table S4,  
296 Appendix S1), indicating that population elevation was not associated with age. Consistent  
297 with reduced longevity in *Bd*-infected populations, no individuals were recaptured between  
298 years (two populations in 1996-1997 and three populations 2011-2012). In contrast, *Bd*-free  
299 populations demonstrated relatively high inter-annual adult survival, with eight of twenty  
300 individuals recaptured across two consecutive years.

301

### 302 **Truncated age structure in *Bd*-infected populations**

303 We identified severe age structure truncation in *Bd*-infected populations for both adult  
304 males and females (Figs. 1 & 2). Most males were two years old, with no individuals older  
305 than three. In contrast, *Bd*-free/pre-decline populations contained males ranging from one to  
306 seven years old (Table S5, Appendix S1; Fig. 1). No females older than three were recorded  
307 from *Bd*-infected populations, while the majority of females in *Bd*-free populations were  
308 older than three years (Table S6, Appendix S1).

309

### 310 **Age structure truncation reduces population size and resilience**

311 Using simulation models, we demonstrated that *Bd*-infected populations with  
312 truncated age structure were smaller and less resilient to drought-induced recruitment failure.  
313 In the deterministic (Fig. 3 *Bd*-free/pre-decline) and stochastic (Fig. 3 *Bd*-free/pre-decline &  
314 drought) *Bd*-free/pre-decline models, average abundance remained stable. In contrast,  
315 average abundance experienced major declines in both *Bd*-infected models, with the  
316 deterministic model stabilising at a small population size (Fig. 3 *Bd*-infected), and the  
317 addition of stochastic recruitment failure driving the population to extinction (Fig. 3 *Bd*-  
318 infected & drought).

319

## 320 **Discussion**

321 Our study documents a previously unrecognised demographic impact associated with  
322 *Bd*-infection; population age structure truncation. We found that *Bd* presence was associated  
323 with a major reduction in inter-annual adult survival, reduced mean adult age and thus the  
324 loss of individuals capable of reproducing over multiple years. Our simulations highlight that  
325 increased adult mortality and age structure truncation can increase the probability of  
326 extinction for amphibians that reproduce in environments where climatic fluctuations result  
327 in occasional recruitment failure.

328 Across regions where *Bd* has emerged, the pathogen commonly persists in an endemic  
329 state, in both remnant populations of declined species and resistant species. To date, endemic  
330 *Bd* infection has been associated with high ongoing rates of adult mortality in susceptible  
331 species (Murray *et al.* 2009; Pilliod *et al.* 2010; Muths, Scherer & Pilliod 2011; Phillott *et al.*  
332 2013), including in *L. v. alpina* (Scheele *et al.* 2015) (although see Briggs *et al.* (2010) for an

333 example of an endemically infected system dominated by juvenile mortality). Our results  
334 extend our understanding of the demographic impacts of endemic *Bd* infection by  
335 demonstrating that persistent, high adult mortality results in age structure truncation. We  
336 suspect that similar demographic impacts to those we document in *L. v. alpina* may  
337 commonly occur in other amphibian species where endemic *Bd* infection is associated with  
338 reduced adult survival.

339         The loss of older individuals may have important implications for population  
340 dynamics. In other vertebrate groups, reproductive output and success commonly increases  
341 with age (until senescence), with older individuals contributing more, and/or higher quality  
342 offspring, compared to first time breeders (Berkeley *et al.* 2004). In amphibians, clutch  
343 volume (number and size of eggs) is generally correlated with female size (Morrison & Hero  
344 2003). In *Bd*-infected *L. v. alpina* populations, all females were two or three years old  
345 compared to *Bd*-free/pre-decline populations that were dominated by females four years or  
346 older. Young females were substantially smaller than older females (B. Scheele unpublished  
347 data), potentially resulting in smaller average clutch volumes in *Bd*-infected *L. v. alpina*  
348 populations compared to *Bd*-free/pre-decline populations. The loss of old females may also  
349 drive temporal variation in the timing of reproduction, with smaller, younger females  
350 attaining sufficient size and energy reserves for clutch development later in spring; extending  
351 the breeding season into less suitable conditions during summer. Similarly, due to the  
352 energetic costs associated with frequent calling (Ryan 1988), the loss of larger, older males  
353 may result in reduced calling intensity. However, recent research indicates that this could be  
354 off-set by increased calling effort of *Bd*-infected frogs (An & Waldman 2016), highlighting a  
355 need for future research in this area.

356         *Batrachochytrium dendrobatidis*-infected populations had a higher proportion of  
357 young, sexually mature adults compared to *Bd*-free/pre-decline populations; suggesting  
358 earlier maturation could be occurring in diseased populations. Life-history theory predicts  
359 that increased mortality that disproportionately impacts adults can result in earlier maturation  
360 (Stearns 1992; Stearns 2000). Earlier maturity is a documented evolutionary response in fish  
361 and mammals to elevated adult mortality caused by disease (Jones *et al.* 2008; Ohlberger *et*  
362 *al.* 2011). Earlier maturation may reduce the risk of death before reproduction in *Bd*-infected  
363 populations and is an important area requiring targeted investigation.

364 In species that reproduce in uncertain environments, high adult survival and multiple  
365 reproduction opportunities, can provide a buffer against environmental conditions that result  
366 in recruitment failure (Wilbur & Rudolf 2006). However, truncation of age structure and loss  
367 of individuals capable of reproducing across multiple years erodes this capacity and increases  
368 vulnerability to stochastic events (Olsen *et al.* 2004; Rouyer *et al.* 2012). Populations with a  
369 truncated age structure, similar to what was observed in *Bd*-infected populations, are much  
370 more sensitive to periodic recruitment failure (Fig. 3). This is consistent with the observed  
371 spatial pattern of *L. v. alpina* decline, whereby the species has been extirpated from  
372 ephemeral wetland habitat – where periodic drought-induced recruitment failure occurs – but  
373 persists with *Bd* in a small number of perennial wetlands that provide reliable breeding  
374 grounds (Osborne, Hunter & Hollis 1999). This inability to tolerate recruitment failure  
375 effectively results in a major reduction in niche width, restricting the species to habitats  
376 where risk of recruitment failure is low. Thus, the emergence of chytridiomycosis in *L. v.*  
377 *alpina* populations has essentially redefined the habitat requirements of the species; shifting it  
378 from a habitat generalist, to a species dependent on perennial wetlands. This reduction in  
379 breeding habitat heterogeneity increases extinction risk for the species.

380 The population level impacts of *Bd* emergence can be highly variable within a species  
381 (Fisher, Garner & Walker 2009). Our results indicate that spatial variation in population  
382 outcome from introduction of chytridiomycosis may reflect underlying variability in other  
383 sources of mortality between populations (e.g. frequency of periodic recruitment failure).  
384 *Batrachochytrium dendrobatidis*-induced age structure truncation may lead to an extinction  
385 debt, whereby, some populations survive the initial emergence of the pathogen only to be  
386 extirpated when next challenged by a secondary source of mortality. More broadly, pathogens  
387 are well-known to increase extinction risk in host populations (de Castro & Bolker 2005), and  
388 truncation of age structure may be an underlying mechanism that could predispose diseased  
389 populations to extinction. Our simulations highlight the importance of considering multiple  
390 factors when evaluating extinction risk in disease-challenged populations.

391 Wildlife diseases have been documented to cause age structure truncation in other  
392 vertebrate taxa, including fish (Ohlberger *et al.* 2011) and mammals (Härkönen *et al.* 2007;  
393 Lachish, McCallum & Jones 2009). In addition to increasing extinction risk, high mortality in  
394 adults can have direct flow-on effects, such as reduced reproductive output (Coltman *et al.*  
395 2003) and can disrupt social hierarchies (Rogers *et al.* 1998). However, indirect effects, such  
396 as shifts in host species ecological requirements as seen here, appear less well documented.

397 Our results indicate that this perverse, indirect impact can render species maladapted to  
398 previously occupied habitat. Thus, areas previously considered high quality may be  
399 unsuitable due to secondary impacts of pathogens on environmental tolerance.

## 400 **Conclusion**

401 *Batrachochytrium dendrobatidis* is a devastating pathogen that is now endemic in  
402 many amphibian communities globally (Skerratt *et al.* 2007; Wake & Vredenburg 2008;  
403 Olson *et al.* 2013). While populations can persist with *Bd*, we have shown that  
404 chytridiomycosis can result in severe age structure truncation, which increases vulnerability  
405 to other sources of mortality. However, our results illustrate that amphibian populations can  
406 persist despite high disease impact when other sources of mortality, such as drought-induced  
407 recruitment failure, are low. This highlights a major opportunity to develop new management  
408 strategies that focus on increasing resilience in chytridiomycosis-challenged populations  
409 (Scheele *et al.* 2014b). In the case of *L. v. alpina*, this may be achieved through habitat  
410 manipulation to reduce recruitment failure during drought years. More broadly, a thorough  
411 understanding of how wildlife diseases influence the interaction of host demography and  
412 environmental stressors is needed to identify strategies to mitigate the threat posed by  
413 emerging diseases.

414

## 415 **Data Accessibility**

416 Data are available from Dryad Digital Repository:  
417 <http://dx.doi.org/10.5061/dryad.d3m1j> (Scheele *et al.* 2016).

418

## 419 **Acknowledgements**

420 Funding was provided by a Taronga Zoo Field Conservation Grant, the New South  
421 Wales Office of Environment and Heritage and Australian Research Council grant  
422 LP110200240. C. Scheele and S. Kearney provided field assistance and K. Smith and L.  
423 Brannelly helped facilitate museum sampling. C. Foster provided useful comments on an  
424 earlier draft of this manuscript. Research was conducted under scientific permits SL100436  
425 and SL10006052 issued by the New South Wales Office of Environment and Heritage and

426 ethics approval from the Australian National University (A2011/19) and the University of  
 427 Canberra (CEAE 98/7).

428

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596

597

### 598 **Figure captions**

599 Figure 1. Proportion of adult male *Litoria verreauxii alpina* in each age cohort for each  
600 population. The number of individuals sampled in each population is given at the top of each  
601 bar.

602

603 Figure 2. Proportion of adult female *Litoria verreauxii alpina* in each age cohort (populations  
604 combined). The total number of individuals sampled in *Bd*-infected and *Bd*-free/pre-decline  
605 and populations is given at the top of each bar.

606

607 Figure 3. Predictions from Lefkovich matrix models of population abundance through time.  
608 Simulations were run for *Bd*-free/pre-decline and *Bd*-infected (truncated age structure)  
609 *Litoria verreauxii alpina* populations, both with and without periodic recruitment failure  
610 (drought).

611

612

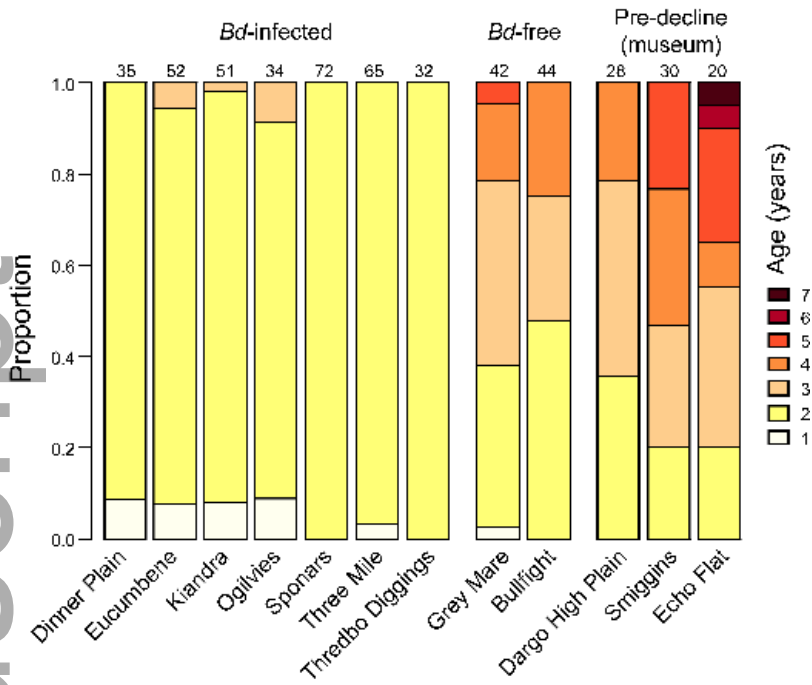
### 613 **Supporting Information**

614 The following Supporting Information is available for this article online:

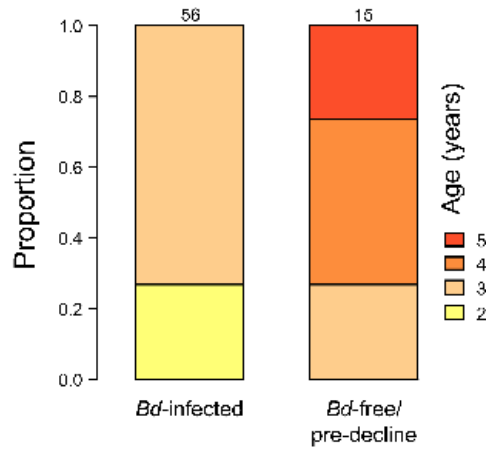
615 **Appendix S1. Table S1.** Population locations and sample sizes. **Table S2.** Population matrix  
616 with a stable age distribution that approximated the observed *Bd*-free age structure. **Table S3.**  
617 Matrix from Table S2 modified to incorporate complete post-breeding adult mortality. **Table**  
618 **S4.** Full model summary for different candidate models investigating impact of population

619 *Bd*-status and elevation on adult male frog age. **Table S5.** Number of adult males in each age  
620 class. **Table S6.** Number of adult females in each age class.

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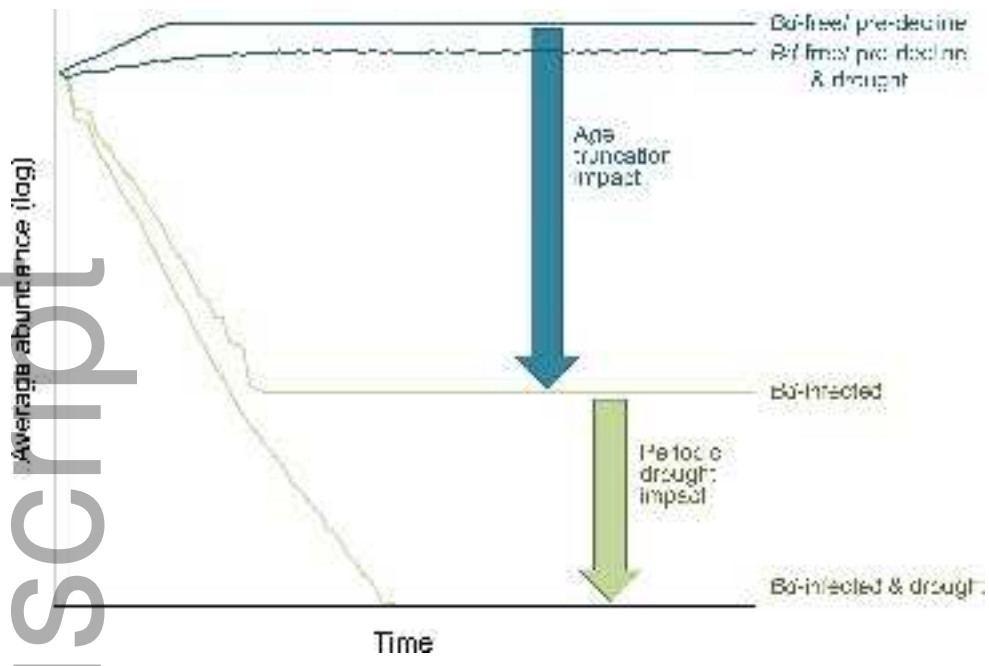


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**Title:**

High adult mortality in disease-challenged frog populations increases vulnerability to drought

**Date:**

2016-11-01

**Citation:**

Scheele, B. C., Hunter, D. A., Banks, S. C., Pierson, J. C., Skerratt, L. F., Webb, R. & Driscoll, D. A. (2016). High adult mortality in disease-challenged frog populations increases vulnerability to drought. *JOURNAL OF ANIMAL ECOLOGY*, 85 (6), pp.1453-1460. <https://doi.org/10.1111/1365-2656.12569>.

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