



Research



Cite this article: West G, Pointer MD, Nash WJ, Lewis R, Richardson DS. 2025 Does genetic rescue disrupt local adaptation? An experimental test using thermally adapted *Tribolium castaneum* lines. *Proc. R. Soc. B* **292**: 20252036.

<https://doi.org/10.1098/rspb.2025.2036>

Received: 7 August 2025

Accepted: 10 October 2025

Subject Category:

Global change and conservation

Subject Areas:

genetics, ecology

Keywords:

genetic rescue, *Tribolium castaneum*, inbreeding depression, local adaptation, thermal adaptation, outbreeding depression

Author for correspondence:

George West

e-mail: gwest16@hotmail.com

Electronic supplementary material is available online at <https://doi.org/10.6084/m9.figshare.c.8105317>.

Does genetic rescue disrupt local adaptation? An experimental test using thermally adapted *Tribolium castaneum* lines

George West, Michael D Pointer, Will J Nash, Rebecca Lewis and David S Richardson

School of Biological Sciences, University of East Anglia, Norwich, Norfolk, UK

ORCID GW, 0000-0002-0489-1109; MDP, 0000-0002-7926-330X; RL, 0000-0003-4739-0280; DSR, 0000-0001-7226-9074

Anthropogenic drivers are restricting many species to small, genetically isolated populations. These are prone to inbreeding depression and are at an increased risk of extinction. Genetic rescue, the controlled introduction of genetic variation from another population, can alleviate inbreeding effects. A major conservation concern restricting the use of this technique is that such augmented gene flow may disrupt local adaptation crucial to a population's persistence. Using populations of the red flour beetle (*Tribolium castaneum*) experimentally adapted to reproduce at higher temperatures, we assess whether genetic rescue attempts disrupt thermal adaptation. Rescuers, drawn from populations adapted to either 30°C or 38°C, were introduced into populations adapted to 38°C, which had been inbred for two generations. We recorded population productivity for three generations post-rescue, in the adapted 38°C environment. Rescuers with and without local adaptation significantly increased the productivity of recipient inbred populations, but those sharing local adaptation to reproduction at 38°C provided greater increases in productivity. These results show that while rescue with non-locally adapted individuals may improve productivity, having the same adaptation in rescuing individuals and rescuee populations may be important in maximizing conservation outcomes.

1. Introduction

Climate change and habitat destruction are fragmenting species into increasingly small and isolated populations where gene flow is disrupted. This results in inbreeding, inbreeding depression and, consequently, increased risk of extinction [1–3]. Inbred individuals are more likely to be homozygous for any recessive, deleterious alleles present in the population, exposing their harmful effects [4,5]. This conversion of hidden genetic load, masked by dominance, into expressed load contributes to reduced fitness at both the individual and population levels. This increase in homozygosity may also lead such individuals to lose the benefits of heterozygote advantage [6]. Factors such as these interact with environmental drivers to push populations towards extinction [7–9].

Genetic rescue refers to the increase in fitness observed in an inbred population when novel genetic variation is introduced by a conspecific from another population, a 'rescuer' [10,11]. This process increases genome-wide heterozygosity within the target population, reducing expressed genetic load and improving fitness. Genetic rescue has been successfully implemented in

endangered populations [12–15], including the Florida panther (*Puma concolor cougar*) [16,17]. Experimental organisms have also extended our understanding of genetic rescue [18–22], including how genetic rescue can affect adaptation in changing environments [23]. Despite this body of evidence supporting its efficacy, genetic rescue remains controversial among conservation managers [24–26].

If populations in need of genetic rescue exhibit local adaptation, the input of novel genetic variation from other populations could disrupt beneficial gene complexes. Rescuers may introduce non-locally adapted alleles, the expression of which could disrupt adaptation and thus exacerbate reduced fitness in vulnerable recipient populations [27,28]. Additionally, deleterious alleles from the rescuer population can be introduced, lowering fitness and potentially leading to population crashes [29,30]. This effect, termed outbreeding depression, has been suggested to be a key risk of implementing genetic rescue as a conservation measure [31,32]. However, it has been argued that, if genetic rescue guidelines are followed, the risk of such a detrimental impact is overstated [24,33,34], relative to the potential benefits of rescue. Indeed, while there are various examples of outbreeding depression occurring, the general consensus is that in many situations the benefits of reducing inbreeding depression outweigh the risk of outbreeding depression [35–38].

Selecting the source population for genetic rescue attempts is key to avoiding outbreeding depression. Populations from across a species range may be locally adapted to different conditions, leading to the introduction of maladaptive traits reducing population fitness [39]. Increasingly, reintroductions from captivity are being considered to reinforce wild populations including in the context of genetic rescue [40]. Captivity, however, could promote maladaptation as natural selection is weakened if not absent [41], utilizing individuals from captivity for rescue could reduce the fitness of wild populations by introducing maladapted genotypes. Testing the effects of maladaptation on genetic rescue, from wild or captive populations, is vital to improve our ability to select the best source population for rescuers.

Climate change poses a significant challenge for endangered species [42], with genetically depauperate populations struggling to adapt [43]. The introduction of genetic variation into isolated populations via genetic rescue could expedite adaptation to rapidly evolving climate by supplementing standing genetic variation [32,44]. Seeding rescue attempts from populations with specific local adaptations, for example to high temperatures [45], could facilitate the introgression of beneficial adaptations into endangered populations [46,47]. Genetic rescue of inbred endangered populations that exhibit specific local adaptations is also a key consideration to protect such unique combinations of alleles to support future species-level resiliency.

Tribolium castaneum, a tenebrionid beetle, is a model organism [48] for population genetics, genetic rescue and thermal tolerance [18,49,50]. Here, we use *T. castaneum* populations experimentally selected for over 150 generations to reproduce at 38°C, compared to the ancestral population optimum of 30°C [51,52]. The thermally adapted populations maintain fecundity when developing at 38°C; they produce more eggs and have a greater hatching success than non-adapted populations developing at this temperature [53]. However, these thermally adapted populations produce fewer offspring than do non-adapted populations that have developed at 30°C, even when those non-adapted populations are transferred to mate and oviposit at 38°C as adults [53]. This suggests that adaptation to a higher temperature may carry a fitness cost. In order to test the impact of genetic rescue from rescuing individuals with and without local adaptations, we used replicated subpopulations of these adapted lines to generate inbred experimental populations. We assessed the impact of genetic rescue from individuals with different adaptive backgrounds on thermal tolerance adaptation. We show that genetic rescue by an adapted individual was the most effective treatment at increasing fitness in inbred populations.

2. Methods

(a) Husbandry

Tribolium castaneum populations were maintained on standard fodder (90% white organic flour, 10% brewer's yeast and a layer of oats for traction) in a controlled environment of 30°C (unless otherwise stated) and 60% humidity with a 12:12 light:dark cycle. Populations were maintained following a standard cycle of virgin adults having 7 days of mating and oviposition followed by the removal of adult beetles, using 2 mm and 850 µm sieves, so that only eggs remain in the fodder. Each generation is initiated with a number of adults (line dependent, see below) that are given 7 days to mate and lay eggs before being removed. The eggs are left for 35 days to develop into mature adults.

(b) *Tribolium castaneum* lines

(i) Krakow super strain

Krakow super strain (KSS) is a combination of 14 laboratory strains that were bred to maximize genetic diversity in one population maintained at a census size of 600 individuals [54]. This line is highly productive at 30°C but has reduced fitness when developing at 38°C. This line is highly productive at 30°C but has reduced fitness when developing at 38°C. The number of eggs produced by a single KSS female developing at 38°C over 2 days declines from ~22 to ~11 and the proportion of eggs that hatch falls from ~40% to nearly 0% [53]. This line was used as the non-adapted rescuer population.

(ii) Thermal lines

Ten independent lines (census size = 100 adults) were founded from KSS and experimentally evolved for ~150 generations at an environmental temperature of 38°C [55]. This imposed selection for development and reproduction at this temperature, considerably above the thermal optimum for *T. castaneum* [56]. All other conditions were as described above aside from a shorter development period of 27 days, reflecting accelerated development at 38°C. A single thermally adapted female can produce more eggs over 2 days than KSS when developing at 38°C, around 18 eggs with around a 50% hatching proportion [53]. These were used as the thermally adapted rescuer populations.

(iii) Inbred lines

Ten inbred thermally adapted populations were created; each descended from one of the ten thermal lines described above. Adult beetles from each thermal line were housed individually for two weeks to ensure any fertilized eggs were laid. Single-pair matings were formed by housing together a previously isolated male and a female for 7 days of mating and oviposition, resulting in a single pair bottleneck for each thermal line. Full-sibling offspring resulting from this pairing were again paired for a second bottleneck. The following generation was initiated with 10 male and 10 female full-sibling offspring of full-sibling pairs. From the offspring of these groups, 6 inbred experimental populations (10 males and 10 females) were created from each of the 10 inbred thermal lines, to act as recipient populations for genetic rescue. One inbred population only produced 4 experimental populations, resulting in a total of 58 experimental populations split over two temporal blocks of 30 and 28. The two blocks were maintained one day apart for ease of handling but otherwise received identical treatment. Each population received a random ID number to blind the experiment and avoid bias. Experimental populations were initiated every generation using 10 males and 10 females sourced from the offspring of the previous generation to reduce density-dependent effects [57–59]. All experimental inbred recipient populations were kept at 38°C in A. B. Newlife 75 Mk4 forced air egg incubators (A. B. Incubators, Suffolk, UK); all other conditions were kept as described above.

(c) Genetic rescue protocol

Populations were kept in 125 ml tubs containing 70 ml of standard fodder. After 7 days, adults were discarded, and eggs were left to develop for ~21 days when 10 male and 10 female pupae were randomly selected to establish the next generation. Remaining individuals were maintained for 10 days following this and were then frozen and manually counted. Pupae taken at day ~21 were housed in plastic dishes containing 10 ml standard fodder in single-sex groups until they matured into adults after 10 ± 2 days, and the next generational cycle began with unmated adults, avoiding overlapping generations.

After a rest generation a single male from each inbred recipient population was removed and replaced with a single male rescuer to avoid demographic rescue effects (increased population fitness due to increased population size) [1,10,32]. Three treatments were created: (i) control—19 populations received no rescue (the male was not removed); (ii) locally adapted rescue—19 populations received a 38°C-adapted rescuer (a male from a different thermally adapted population); (iii) non-locally adapted rescue—20 populations received a non-thermally adapted rescuer (a KSS male, see above) (figure 1).

Population fitness was measured using productivity: the number of mature adult offspring the population produced each generation [60]. Populations were maintained for three non-overlapping generations following rescue. Two replicates were lost after two generations due to human error, resulting in a third generation with 19 control, 18 thermally adapted rescue and 19 non-thermally adapted rescue populations. Data for populations lost in generation 3 were included in the analysis of generations 1 and 2.

(d) Statistical analysis

R v.4.4.1 [61] was used with R Studio version 2024.04.2 + 764 [62]. Data management and exploration were performed with tidyverse [63], stats [61], Rmisc [64] and googlesheets4 [65]. ggplot2 [66] was used to visualize results. Data distribution was checked using the shapiro.test function [61]. The glmmTMB package [67] was used to fit generalized linear mixed models (GLMMs). DHARMA [68] was used to check model fit and the check_collinearity function from the performance package [69] to test variance inflation factor (VIF) scores. No overdispersion or collinearity ($VIF < 3$ for all variables) was found. R^2 was determined using the r.squaredGLMM function in MuMIn [70].

Counts of population productivity over all generations were analysed using a GLMM with a negative binomial errors and a log link function. Fixed explanatory variables were rescue treatment and generation as well as the interaction between these variables. To account for population variance and relatedness between replicate populations, a random factor was added, nesting individual ID within the stock thermal line from which the population descended. The control treatment was set as the baseline factor for comparison. The baseline was changed to non-adapted to compare between the two rescue treatments post-hoc.

GLMMs, constructed as described above, but excluding the generation variable, were then run post-hoc on each generation individually to test if there were significant differences between the treatments in each generation. The baseline was changed to non-adapted rescuers, to compare the two rescue treatments post-hoc.

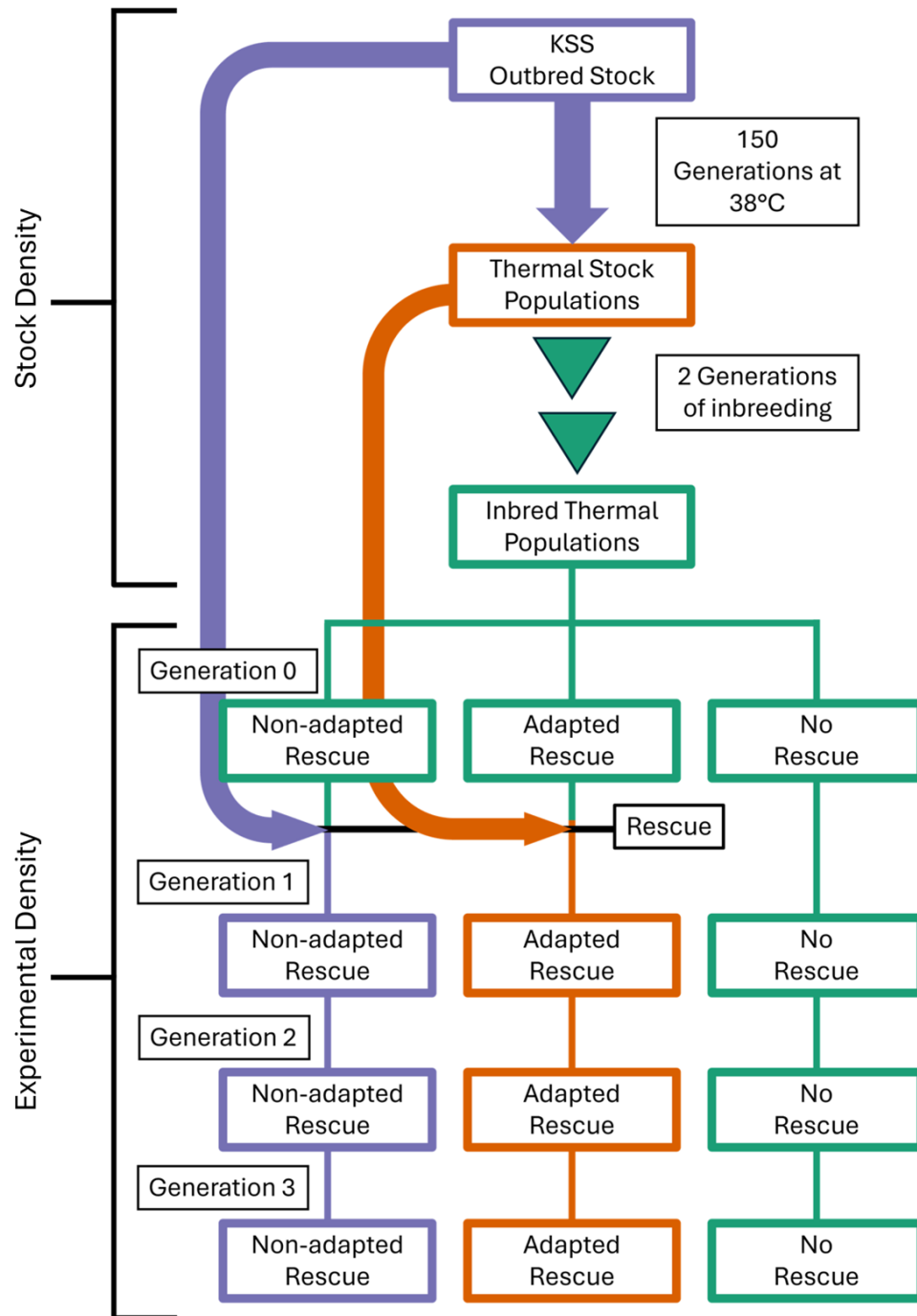


Figure 1. Experimental design for the attempted genetic rescue of inbred thermally adapted *T. castaneum* populations by a single thermally adapted or non-thermally adapted rescuer. Inbred, thermally adapted lines were created by inbreeding lines thermally adapted to 38°C over ~150 generations [55] with two generations of full-sibling matings, before being kept for three generations at $n = 20$ (10 females and 10 males) during the experiment. The 10 inbred thermal lines were replicated to be represented in each experimental treatment twice. The final sample size was 56 experimental populations (see main text).

3. Results

The interaction between treatment and generation was significant for the thermally adapted rescue ($\beta = 0.093 \pm 0.039$, $z = 3.38$, $p = 0.017$) but not for the standard rescue ($\beta = 0.055 \pm 0.039$, $z = 1.41$, $p = 0.16$) when each was compared with the no-rescue control (table 1, figure 2). A post-hoc contrast between the two rescue treatments showed no difference in the gradient of productivity change over generation ($\beta = 0.038 \pm 0.037$, $z = 1.01$, $p = 0.31$, 95 % CI = -0.035 to 0.111).

In generation 1, no significant differences existed between any of the treatments (non-adapted—adapted: estimate = 0.015, s.e. = 0.057, $z = 0.26$, $p = 0.794$; table 2). In generations 2 and 3, productivity in the rescue treatments differed significantly from the no rescue control, were not significantly different in generation 2 (estimate = 0.095, s.e. = 0.057, $z = 1.67$, $p = 0.095$), but exhibited a significantly difference in generation 3 (estimate = 0.097, s.e. = 0.045, $z = 2.17$, $p = 0.030$).

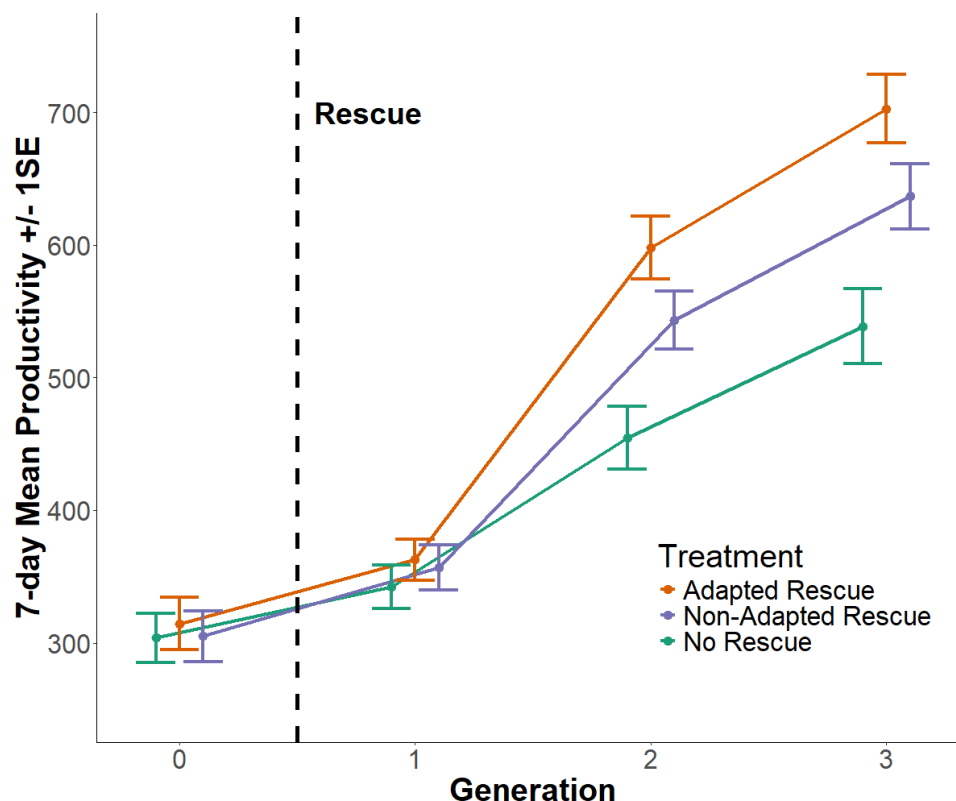


Figure 2. The effect of introducing a (1) thermally adapted (orange) or (2) non-adapted (blue) rescuer, compared to control populations (green) on the mean productivity of inbred thermally adapted populations of *T. castaneum* over three generations after a single introduction event (dotted vertical line) while maintained at 38°C (population size = 20; number of experimental populations = 58 in generations 1 and 2, 56 in generation 3). Generation 0 was not included in statistical analysis. Plot is jittered to aid in visualization, error bars represent ± 1 s.e.

Table 1. Summary of a GLMM fitted to model the productivity of small, inbred, thermally adapted *T. castaneum* populations (population size = 20, number of populations = 58 or 56) after receiving a rescue by a thermally adapted, or non-adapted, male rescuer or no rescue over three generations. Predictors in bold are significant ($p < 0.05$). Marginal $R^2 = 0.637$, conditional $R^2 = 0.740$. The treatment (no rescue) was the baseline for comparison in the model, the values in the table are relative to that treatment.

predictor	estimate	s.e.	z	p
intercept	5.626	0.0756	47.43	<2e–16
treatment (no rescue)				
thermally adapted rescue	0.024	0.096	0.25	0.803
non-adapted rescue	0.028	0.096	0.29	0.771
generation	0.224	0.031	7.33	<0.001
treatment (no rescue) $\times$ generation				
treatment (thermally adapted rescue) $\times$ generation	0.091	0.041	2.21	0.027
treatment (non-adapted rescue) $\times$ generation	0.055	0.041	1.33	0.185
random	172 observations		variance	
ID:thermal line	58		0.002	
thermal line	10		0.007	

4. Discussion

We tested the disruptive effect of genetic rescue in locally adapted populations, by introducing thermally adapted or outbred, non-adapted genetic rescuers to inbred thermally adapted populations of *T. castaneum* and then measuring productivity as an estimate of population fitness. We show that populations receiving thermally adapted rescue recovered fitness following two generations of inbreeding at a faster rate than populations that did not receive new genetic variation. When looking at each generation individually, both rescue treatments improved fitness in the second and third generations, when compared to the no-rescue control. In the third generation, fitness in populations that received thermally adapted rescue improved over both the no-rescue control and non-adapted rescue treatment. We also observed increased productivity in the no rescue treatment, this was seen in previous experiments [71] and is probably due to the change from highly dense stock populations to low density

Table 2. Composite table of three GLMM results for each generation of the productivity of small, inbred, thermally adapted *T. castaneum* populations (population size = 20, experimental populations = 58 or 56) after receiving a rescue by a thermally adapted, or non-adapted, male rescuer or no rescue. Predictors in bold are significant ($p < 0.05$).

	gen 1 estimate	s.e.	z	p	gen 2 estimate	s.e.	z	p	gen 3 estimate	s.e.	z	p
intercept (no rescue)	5.810	0.055	105.84	<2e-16	6.100	0.057	107.29	<2e-16	6.263	0.051	122.93	<2e-16
thermally adapted rescue	0.061	0.058	1.05	0.291	0.284	0.058	4.90	<0.001	0.277	0.045	6.20	<0.001
non-adapted rescue	0.056	0.057	0.81	0.420	0.188	0.057	3.28	0.001	0.180	0.044	4.07	<0.001
	gen 1 observation		gen 1 variance		gen 2 observation		gen 2 variance		gen 3 observation		gen 3 variance	
random	58				58				56			
ID:thermal	58		<0.001		58		0.030		56		0.017	
thermal	10		<0.001		10		0.005		10		0.016	

experimental populations effecting all populations equally [57]. This makes the inclusion of no rescue control populations vital, comparisons before and after rescue would be misleading without the data they provide.

Introducing new genetic diversity into an inbred population is predicted to improve population fitness, and such genetic rescue effects have been observed in applied, unreplicated conservation studies [72,73]. Here, we provide experimental support for this suggestion, as well as showing that the fitness of locally adapted, inbred populations recovered at the greatest rate following the introduction of a rescuing individual from a population with a similar selective background. Importantly, we show that non-adapted rescuers also improved fitness compared with no rescue, despite the potential to disrupt adaptation, though the magnitude of the fitness increase was smaller than when using a locally adapted rescuer. The benefits of rescue were also clear even utilizing only a single rescuer in a single rescue event in the experiment, which has also been observed in *Drosophila* [74], supporting the one-migrant-per-generation rule for preventing inbreeding depression [75]. Recent experimental studies have also shown that the benefits of rescue can be long-lasting, far beyond the three generations followed here [22].

The ancestral population in our experiment is highly outbred and should represent an ideal source of variation for use in genetic rescue [33]. Our findings show that local adaptations, evolved over 150 generations, considerably increased the efficacy of genetic rescue. Previous studies have suggested that the reinitiation of gene flow between populations following more than 20 generations of environmental divergence may risk outbreeding depression [24]. However, there is a growing body of evidence that the risk of outbreeding depression in genetic rescue may be exaggerated [34,76,77]. In our study, the relatedness of the lines, benign conditions and experimental set-up make outbreeding depression unlikely to occur, excepting the disruption of adaptation to increased temperature.

In our study, genetic rescue by a single individual bearing local thermal adaptation was more effective than rescue by an outbred rescuer not bearing this adaptation. Our findings support recommendations that genetic rescue should utilize source populations inhabiting similar environments, reducing the risk of disrupting local adaptation [28]. We used 10 independent thermally adapted lines in this study, all originating from the same genetically diverse ancestral population. As these populations adapted to 38°C independently [51], they may represent differing subsets of genetic variation, providing alternative substrates for genetic rescue to act on. This design provides a proxy for population fragmentation, with subpopulations diverging from a larger outbred population. As the experimental evolution for thermal adaptation probably resulted in bottlenecks of the adapting populations [55], we predict these populations to be less genetically diverse than the outbred ancestral population.

In the thermally adapted populations we study, the key adaptation is the capacity to remain fertile while developing at 38°C [51,53,78]. Our findings suggest that the introduction of outbred variation may have disrupted this adaptation. If the increase in fitness were solely attributable to the introduction of genetic variation, an outbred rescuer would generate the most impactful rescue effect [33]. We show that the addition of locally adapted variation was more important to rapid recovery from inbreeding effects, supporting the importance of rescue from populations with similar adaptive backgrounds [25]. Using translocations from locally adapted populations could be important in a conservation context as they may improve population resilience to a changing climate [76].

It is beyond the scope of this study to identify the causative genetic variation that conferred local adaptation and that allowed locally adapted individuals to act as the most effective rescuers. Future work should aim to test if effective rescue is mediated by the transfer of beneficial alleles directly contributing to adapted traits [46], or by the purging of genetic load, mildly deleterious mutations, in genes associated with the adaptation [79]. Despite this lack of resolution, we use a powerful, highly replicated, experimental design to answer a question difficult to test in wild populations. Our findings are highly relevant to conservation scenarios and help highlight the utility of experimental evolution in contributing to applied questions. We provide support for the consideration of local adaptation in genetic rescue programmes showing that single rescuer can provide clear fitness benefits and adapted populations are a better source of rescuers.

Ethics. No ethics approval was required for this study as experiments were conducted on an unregulated invertebrate species.

Data accessibility. All data and scripts are available at Dryad [60].

Supplementary material is available online [80].

Declaration of AI use. We have not used AI-assisted technologies in creating this article.

Authors' contributions. G.W.: conceptualization, data curation, formal analysis, investigation, methodology, visualization, writing—original draft, writing—review and editing; M.P.: investigation, supervision, writing—review and editing; R.L.: conceptualization, supervision, writing—review and editing; D.R.: conceptualization, funding acquisition, project administration, resources, supervision, writing—review and editing; W.N.: supervision, writing—review and editing.

All authors gave final approval for publication and agreed to be held accountable for the work performed therein.

Conflict of interest declaration. We declare we have no competing interests.

Funding. This work was supported by the Natural Environment Research Council, including an ARIES DTP PhD [NE/S007334/1] to G.W., and a Research Grant (Understanding heatwave damage through reproduction in insect systems) [NE/T007885/1] to Matt Gage. The authors also acknowledge support from the Biotechnology and Biological Sciences Research Council (BBSRC), part of UK Research and Innovation, Core Capability Grant BB/CCG2220/1 at the Earlham Institute (EI) and its constituent work packages (BBS/E/T/000PR9818 and BBS/E/T/000PR9819), and the Core Capability Grant BB/CCG1720/1 and the National Capability BBS/E/T/000PR9816 (NC1—Supporting EI's ISPs and the UK Community with Genomics and Single Cell Analysis), BBS/E/T/000PR9811 (NC4—Enabling and Advancing Life Scientists in data-driven research through Advanced Genomics and Computational Training), and BBS/E/T/000PR9814 (NC 3—Development and deployment of versatile digital platforms for 'omics-based data sharing and analysis). Also support from BBSRC Core Capability Grant BB/CCG1720/1 and the work delivered via the Scientific Computing group, and the physical HPC infrastructure and data centre delivered via the NBI Computing infrastructure for Science (CIS) group.

Acknowledgements. We thank the members of the UEA Tribolium Lab for assistance with line maintenance and data collection.

References

- Haddad NM *et al.* 2015 Habitat fragmentation and its lasting impact on Earth's ecosystems. *Sci. Adv.* **1**, e1500052. (doi:10.1126/sciadv.1500052)
- Wright S. 1990 Evolution in mendelian populations. *Bull. Math. Biol.* **52**, 241–295. (doi:10.1007/BF02459575)
- Wright S. 1969 Evolution and the genetics of populations. In *Theory of gene frequencies*, vol. **2**. Chicago, IL: University of Chicago Press.
- Crnokrak P, Roff DA. 1999 Inbreeding depression in the wild. *Heredity* **83**, 260–270. (doi:10.1038/sj.hdy.6885530)
- Charlesworth D, Willis JH. 2009 The genetics of inbreeding depression. *Nat. Rev. Genet.* **10**, 783–796. (doi:10.1038/nrg2664)
- Hedrick PW, García-Dorado A. 2016 Understanding inbreeding depression, purging, and genetic rescue. *Trends Ecol. Evol.* **31**, 940–952. (doi:10.1016/j.tree.2016.09.005)
- Soule M, Gilpin M. 1986 *Conservation biology: the science of scarcity and diversity*. Sunderland, MA: Sinauer Associates.
- Blomqvist D, Pauliny A, Larsson M, Flodin LA. 2010 Trapped in the extinction vortex? strong genetic effects in a declining vertebrate population. *BMC Evol. Biol.* **10**, 33. (doi:10.1186/1471-2148-10-33)
- Palomares F, Godoy JA, López-Bao JV, Rodríguez A, Roques S, Casas-Marce M, Revilla E, Delibes M. 2012 Possible extinction vortex for a population of *Iberian lynx* on the verge of extirpation. *Conserv. Biol.* **26**, 689–697. (doi:10.1111/j.1523-1739.2012.01870.x)
- Ingvarsson PK. 2001 Restoration of genetic variation lost: the genetic rescue hypothesis. *Trends Ecol. Evol.* **16**, 62–63. (doi:10.1016/S0169-5347(00)02065-6)
- Hedrick PW, Adams JR, Vucetich JA. 2011 Reevaluating and broadening the definition of genetic rescue. *Conserv. Biol.* **25**, 1069–1070. (doi:10.1111/j.1523-1739.2011.01751.x)
- Clarke JG, Smith AC, Cullingham CI. 2024 Genetic rescue often leads to higher fitness as a result of increased heterozygosity across animal taxa. *Mol. Ecol.* **33**, e17532. (doi:10.1111/mec.17532)
- Frankham R. 2016 Genetic rescue benefits persist to at least the F3 generation, based on a meta-analysis. *Biol. Conserv.* **195**, 33–36. (doi:10.1016/j.biocon.2015.12.038)
- Frankham R. 2015 Genetic rescue of small inbred populations: meta-analysis reveals large and consistent benefits of gene flow. *Mol. Ecol.* **24**, 2610–2618. (doi:10.1111/mec.13139)
- White SL, Rash JM, Kazyak DC. 2023 Is now the time? Review of genetic rescue as a conservation tool for brook trout. *Ecol. Evol.* **13**, e10142. (doi:10.1002/ece3.10142)
- Pimm SL, Dollar L, Bass OL. 2006 The genetic rescue of the Florida panther. *Anim. Conserv.* **9**, 115–122. (doi:10.1111/j.1469-1795.2005.00010.x)
- Onorato DP *et al.* 2024 Multi-generational benefits of genetic rescue. *Sci. Rep.* **14**, 17519. (doi:10.1038/s41598-024-67033-6)
- Hufbauer RA, Szűcs M, Kasyon E, Youngberg C, Koontz MJ, Richards C, Tuff T, Melbourne BA. 2015 Three types of rescue can avert extinction in a changing environment. *Proc. Natl Acad. Sci. USA* **112**, 10557–10562. (doi:10.1073/pnas.1504732112)
- Fitzpatrick SW, Bradburd GS, Kremer CT, Salerno PE, Angeloni LM, Chris Funk W. 2019 Genetic rescue without genomic swamping in wild populations. *bioRxiv* (doi:10.1101/701706)
- Zajitschek SRK, Zajitschek F, Brooks RC. 2009 Demographic costs of inbreeding revealed by sex-specific genetic rescue effects. *BMC Evol. Biol.* **9**, 289. (doi:10.1186/1471-2148-9-289)
- Bijlsma R, Westerhof MDD, Roekx LP, Pen I. 2010 Dynamics of genetic rescue in inbred *Drosophila melanogaster* populations. *Conserv. Genet.* **11**, 449–462. (doi:10.1007/s10592-010-0058-z)
- Pérez-Pereira N, Kleinman-Ruiz D, García-Dorado A, Quesada H, Caballero A. 2025 A test of the long-term efficiency of genetic rescue with *Drosophila melanogaster*. *Mol. Ecol.* e17690. (doi:10.1111/mec.17690)
- Durkee LF, Olazuaga L, Melbourne BA, Hufbauer RA. 2024 Immigration delays but does not prevent adaptation following environmental change: experimental evidence. *J. Evol. Biol.* **37**, 665–676. (doi:10.1093/jeb/voae031)
- Frankham R, Ballou JD, Eldridge MDB, Lacy RC, Ralls K, Dudash MR, Fenster CB. 2011 Predicting the probability of outbreeding depression. *Conserv. Biol.* **25**, 465–475. (doi:10.1111/j.1523-1739.2011.01662.x)
- Edmands S. 2007 Between a rock and a hard place: evaluating the relative risks of inbreeding and outbreeding for conservation and management. *Mol. Ecol.* **16**, 463–475. (doi:10.1111/j.1365-294X.2006.03148.x)
- Fitzpatrick SW, Mittan-Moreau C, Miller M, Judson JM. 2023 Genetic rescue remains underused for aiding recovery of federally listed vertebrates in the United States. *J. Hered.* **114**, 354–366. (doi:10.1093/jhered/esad002)
- Kawecki TJ, Ebert D. 2004 Conceptual issues in local adaptation. *Ecol. Lett.* **7**, 1225–1241. (doi:10.1111/j.1461-0248.2004.00684.x)
- Lenormand T. 2002 Gene flow and the limits to natural selection. *Trends Ecol. Evol.* **17**, 183–189. (doi:10.1016/S0169-5347(02)02497-7)
- Pérez-Pereira N, Caballero A, García-Dorado A. 2022 Reviewing the consequences of genetic purging on the success of rescue programs. *Conserv. Genet.* **23**, 1–17. (doi:10.1007/s10592-021-01405-7)

30. Hedrick PW, Peterson RO, Vucetich LM, Adams JR, Vucetich JA. 2014 Genetic rescue in Isle Royale wolves: genetic analysis and the collapse of the population. *Conserv. Genet.* **15**, 1111–1121. (doi:10.1007/s10592-014-0604-1)
31. Tallmon DA, Luikart G, Waples RS. 2004 The alluring simplicity and complex reality of genetic rescue. *Trends Ecol. Evol.* **19**, 489–496. (doi:10.1016/j.tree.2004.07.003)
32. Bell DA, Robinson ZL, Funk WC, Fitzpatrick SW, Allendorf FW, Tallmon DA, Whiteley AR. 2019 The exciting potential and remaining uncertainties of genetic rescue. *Trends Ecol. Evol.* **34**, 1070–1079. (doi:10.1016/j.tree.2019.06.006)
33. Ralls K, Sunnucks P, Lacy RC, Frankham R. 2020 Genetic rescue: a critique of the evidence supports maximizing genetic diversity rather than minimizing the introduction of putatively harmful genetic variation. *Biol. Conserv.* **251**, 108784. (doi:10.1016/j.biocon.2020.108784)
34. Powell DM. 2023 Losing the forest for the tree? On the wisdom of subpopulation management. *Zoo Biol.* **42**, 591–604. (doi:10.1002/zoo.21776)
35. Hedrick PW, Robinson JA, Peterson RO, Vucetich JA. 2019 Genetics and extinction and the example of Isle Royale wolves. *Anim. Conserv.* **22**, 302–309. (doi:10.1111/acv.12479)
36. Turček FJ, Hickey JJ. 1951 Effect of introductions on two game populations in Czechoslovakia. *J. Wildl. Manag.* **15**, 113–114.
37. Loope KJ, DeSha JN, Aresco MJ, Shoemaker KT, Hunter EA. 2025 Common-garden experiment reveals outbreeding depression and region-of-origin effects on reproductive success in a frequently translocated tortoise. *Anim. Conserv.* **28**, 249–261. (doi:10.1111/acv.12977)
38. Whitlock R, Stewart GB, Goodman SJ, Piertney SB, Butlin RK, Pullin AS, Burke T. 2013 A systematic review of phenotypic responses to between-population outbreeding. *Environ. Evid.* **2**, 13. (doi:10.1186/2047-2382-2-13)
39. Bachmann JC, Jansen van Rensburg A, Cortazar-Chinarro M, Laurila A, Van Buskirk J. 2020 Gene flow limits adaptation along steep environmental gradients. *Am. Nat.* **195**, E67–E86. (doi:10.1086/707209)
40. Al Hikmani H, van Oosterhout C, Birley T, Labisko J, Jackson HA, Spalton A, Tollington S, Groombridge JJ. 2024 Can genetic rescue help save Arabia's last big cat? *Evol. Appl.* **17**, e13701. (doi:10.1111/eva.13701)
41. Frankham R. 2008 Genetic adaptation to captivity in species conservation programs. *Mol. Ecol.* **17**, 325–333. (doi:10.1111/j.1365-294X.2007.03399.x)
42. Hoegh-Guldberg O *et al.* 2018 Impacts of 1.5°C global warming on natural and human systems. In *Global warming of 1.5°C: an ipcc special report on the impacts of global warming of 1.5°C above pre-industrial levels and related global greenhouse gas emission pathways, in the context of strengthening the global response to the threat of climate change, sustainable development, and efforts to eradicate poverty* (eds V Masson-Delmotte *et al.*), pp. 175–311. Cambridge, UK: Cambridge University Press. In Press.
43. Bellard C, Bertelsmeier C, Leadley P, Thuiller W, Courchamp F. 2012 Impacts of climate change on the future of biodiversity. *Ecol. Lett.* **15**, 365–377. (doi:10.1111/j.1461-0248.2011.01736.x)
44. Bell G, Gonzalez A. 2009 Evolutionary rescue can prevent extinction following environmental change. *Ecol. Lett.* **12**, 942–948. (doi:10.1111/j.1461-0248.2009.01350.x)
45. Macadam A *et al.* 2025 Assessing the potential for 'assisted gene flow' to enhance heat tolerance of multiple coral genera over three key phenotypic traits. *Biol. Conserv.* **306**, 111155. (doi:10.1016/j.biocon.2025.111155)
46. Kelly E, Phillips BL. 2019 Targeted gene flow and rapid adaptation in an endangered marsupial. *Conserv. Biol.* **33**, 112–121. (doi:10.1111/cobi.13149)
47. Rudin-Bitterli TS, Evans JP, Mitchell NJ. 2021 Fitness consequences of targeted gene flow to counter impacts of drying climates on terrestrial-breeding frogs. *Commun. Biol.* **4**, 1195. (doi:10.1038/s42003-021-02695-w)
48. Pointer MD, Gage MJG, Spurgin LG. 2021 *Tribolium* beetles as a model system in evolution and ecology. *Heredity* **126**, 869–883. (doi:10.1038/s41437-021-00420-1)
49. Sales K, Vasudeva R, Gage MJG. 2021 Fertility and mortality impacts of thermal stress from experimental heatwaves on different life stages and their recovery in a model insect. *R. Soc. Open Sci.* **8**, 201717. (doi:10.1098/rsos.201717)
50. Sokal RR, Sneath RH. 1968 The ecology of selection in hybrid populations of *Tribolium castaneum*. *Ecol. Monogr.* **38**, 345–379.
51. Vasudeva R, Sutter A, Sales K, Dickinson ME, Lumley AJ, Gage MJ. 2019 Adaptive thermal plasticity enhances sperm and egg performance in a model insect. *eLife* **8**, 49452. (doi:10.7554/eLife.49452.001)
52. Skourt A, Kallieratos NG, Papanikolaou NE. 2022 Demographic responses of *Tribolium castaneum* (Coleoptera: Tenebrionidae) to different temperatures in soft wheat flour. *J. Therm. Biol.* **103**, 103162. (doi:10.1016/j.jtherbio.2021.103162)
53. Lewis RC. 2020 Thermal adaptation in a model pest insect. Thesis, University of East Anglia, Norwich, UK. <https://ueaeprints.uea.ac.uk/id/eprint/79830>.
54. Laskowski R, Radwan J, Kuduk K, Mendrok M, Kramarz P. 2015 Population growth rate and genetic variability of small and large populations of red flour beetle (*Tribolium castaneum*) following multigenerational exposure to copper. *Ecotoxicology* **24**, 1162–1170. (doi:10.1007/s10646-015-1463-3)
55. Dickinson M. 2018 The impacts of heat-wave conditions on reproduction in a model insect, *Tribolium castaneum*. Thesis, University of East Anglia, Norwich, UK. <https://ueaeprints.uea.ac.uk/id/eprint/67673>.
56. Howe RW. 1962 The effects of temperature and humidity on the oviposition rate of *Tribolium castaneum* (Hbst.) (Coleoptera, Tenebrionidae). *Bull. Entomol. Res.* **53**, 301–310. (doi:10.1017/S0007485300048148)
57. Duval C, Park T, Miller EV, Lutherman CZ. 1939 Studies in population physiology. IX. The effect of imago population density on the duration of the larval and pupal stages of *Tribolium*. *Ecology* **20**, 365–373.
58. King CE, Dawson PS. 1972 Population biology and the tribolium model. *Evol. Biol.* **5**, 133–227.
59. Janus MC. 1989 Phenotypic diversity of *Tribolium confusum* pupae in heterogeneous environments. *Entomol. Exp. Appl.* **50**, 281–286. (doi:10.1111/j.1570-7458.1989.tb01203.x)
60. West G, Pointer M, Nash W, Lewis R, Richardson RD. Data from does genetic rescue disrupt local adaptation? An experimental test using thermally adapted *Tribolium castaneum* lines. Dryad Digital Repository. ()
61. R Core Team. 2024 R: a language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing.
62. PositTeam. 2024 RStudio: Integrated Development for R. Boston, MA: Posit Software.
63. Wickham H *et al.* 2019 Welcome to the tidyverse. *JOSS* **4**, 1686. (doi:10.21105/joss.01686)
64. Hope R. 2022 Rmisc: Ryan miscellaneous. <https://cloud.r-project.org/web/packages/Rmisc/Rmisc.pdf>.
65. Bryan J. 2023 googlesheets4: access google sheets using the sheets API V4. <https://rdrr.io/cran/googlesheets4/man/googlesheets4-package.html>.
66. Wickham H. 2016 *Ggplot2: elegant graphics for data analysis*. New York, NY: Springer. (doi:10.1007/978-3-319-24277-4_9)
67. Brooks ME, Kristensen K, Benthem KJ, Magnusson A, Berg CW, Nielsen A, Skaug HJ, Mächler M, Bolker BM. 2017 glmmTMB balances speed and flexibility among packages. *The R Journal* (doi:10.32614/RJ-2017-066)
68. Hartig F. 2022 DHARMA: residual diagnostics for hierarchical (multi-level / mixed) regression models. See <https://github.com/florianhartig/dharma>.
69. Lüdtke D, Ben-Shachar M, Patil I, Waggoner P, Makowski D. 2021 performance: an R package for assessment, comparison and testing of statistical models. *J. Open Source Softw.* **6**, 3139. (doi:10.21105/joss.03139)
70. Bartoń K. 2024 MuMIn: multi-model inference. See <https://CRAN.R-project.org/package=MuMIn>.

71. West G, Pointer M, Nash W, Lewis R, Gage MJG, Richardson DS. 2025 Sexual selection matters in genetic rescue, but productivity benefits fade over time: a multi-generation experiment to inform conservation. *Proc. R. Soc. B* **292**, 20242374. (doi:[10.1098/rspb.2024.2374](https://doi.org/10.1098/rspb.2024.2374))
72. Madsen T, Loman J, Anderberg L, Anderberg H, Georges A, Ujvari B. 2020 Genetic rescue restores long-term viability of an isolated population of adders (*Vipera berus*). *Curr. Biol.* **30**, R1297–R1299. (doi:[10.1016/j.cub.2020.08.059](https://doi.org/10.1016/j.cub.2020.08.059))
73. Johnson WE *et al.* 2010 Genetic restoration of the florida panther. *Science* **329**, 1641–1645. (doi:[10.1126/science.1192891](https://doi.org/10.1126/science.1192891))
74. Spielman D, Frankham R. 1992 Modeling problems in conservation genetics using captive *Drosophila* populations: improvement of reproductive fitness due to immigration of one individual into small partially inbred populations. *Zoo Biol.* **11**, 343–351. (doi:[10.1002/zoo.1430110506](https://doi.org/10.1002/zoo.1430110506))
75. Mills LS, Allendorf FW. 1996 The one-migrant-per-generation rule in conservation and management. *Conserv. Biol.* **10**, 1509–1518. (doi:[10.1046/j.1523-1739.1996.10061509.x](https://doi.org/10.1046/j.1523-1739.1996.10061509.x))
76. Fitzpatrick SW, Reid BN. 2019 Does gene flow aggravate or alleviate maladaptation to environmental stress in small populations? *Evol. Appl.* **12**, 1402–1416. (doi:[10.1111/eva.12768](https://doi.org/10.1111/eva.12768))
77. Fitzpatrick SW, Gerberich JC, Kronenberger JA, Angeloni LM, Funk WC. 2015 Locally adapted traits maintained in the face of high gene flow. *Ecol. Lett.* **18**, 37–47. (doi:[10.1111/ele.12388](https://doi.org/10.1111/ele.12388))
78. Sales K *et al.* 2018 Experimental heatwaves compromise sperm function and cause transgenerational damage in a model insect. *Nat. Commun.* **9**, 4771. (doi:[10.1038/s41467-018-07273-z](https://doi.org/10.1038/s41467-018-07273-z))
79. Grossen C, Guillaume F, Keller LF, Croll D. 2020 Purging of highly deleterious mutations through severe bottlenecks in *Alpine ibex*. *Nat. Commun.* **11**, 1001. (doi:[10.1038/s41467-020-14803-1](https://doi.org/10.1038/s41467-020-14803-1))
80. West G, Pointer M, Lewis R, Richardson D, Nash W. 2025 Supplementary material from: Does genetic rescue disrupt local adaptation? An experimental test using thermally adapted *Tribolium castaneum* lines. Figshare. (doi:[10.6084/m9.figshare.c.8105317](https://doi.org/10.6084/m9.figshare.c.8105317))