

[note from authors: our answers to comments are in blue; line numbers refer to the revised version]

Dear Dr Phillips,

We warmly thank you and the 3 reviewers for your time and consideration of the manuscript, and for your thorough comments. We hope our revised version, and our answers to reviewer comments, will be well-received.

Before diving into the detailed replies, we would like to briefly summarise the main changes we made in response to reviewers:

- we addressed the key criticism about whether our use of the continuum v_F is appropriate for our simulations (with demographic stochasticity and discrete N/space/time) by referring to (Haond et al., 2018)'s results. Their figures show that *for the simulation specificities we used* and *at the carrying capacities K we used*, any bias is small (~1%, like we observed) and likely low enough to not lead to pushed/pulled misclassification at the treatment level.

- we now use the “weakly pushed” expansion concept from (Panja, 2004) to help interpret our simulation results. As a consequence, the discussion is simplified and extraneous details about founding population size at the edge are removed;

- For experimental data, we have added a new metric, the correlation between (a proxy of) K and the speed v , following (Haond et al., 2018). This metric adds to evidence that low-connectivity experimental expansions are pushed;

- We moved the “low connectivity + density-dependent dispersal” simulations to the main text, and use it to discuss how the interaction between DDD and connectivity may explain our experimental results. Whether this interaction is the result of plastic or evolved responses is the subject of another, in prep, manuscript.

- We corrected errors in the physical description of the experimental landscapes, and added a photograph.

We would also like to reaffirm that the “noisiness” or “dirtiness” of our results (i.e. the imperfect match to theoretical expectations) is a feature of our paper, not a bug: a key objective of ours was to see to what extent the “ideal” pushed expansion concept remains meaningful, even qualitatively, in ecologically realistic contexts: low K (relative to theory) and subsequent noise, dispersal and fecundity “noise”, discrete environments due to patchiness, incomplete information about the expansion profile (so only front position is available)... As we mention in the Discussion, our results show that the pushed expansion concept has the potential to be relevant and workable in practice, even beyond microbial examples in the lab or the portion of invasive species able to approximate these lab densities in the wild.

All in all, while we stand by our conclusions (indeed, the substance of our results has not substantially changed during revision), the reviewers' comments have forced us to sharpen our arguments and to think very carefully about each and every point we made, and how applicable they are. Our detailed answers to comments follow.

Maxime Dahirel, on behalf of the authors

Additional requirements of the managing board:
All requirements were already fulfilled, except the “Conflict of Interest Disclosure” one. Several authors are PCI recommenders; this has been added to the Conflict of Interest Disclosure paragraph

Reviewer 1

Comment 1

The authors investigate range expansions occurring within a set of patches or metapopulations. ‘Connectedness’ thereby refers to migration rate between patches (or structure facilitating migration in case of experiments). My main concern is that the parameter regime / setup the authors use is not suitable for a continuum description, around which key arguments are or at least might be based.

Specifically, I think this is a concern for computing v_f (around line 264 in the manuscript). Were r_0 or D_0 slightly higher, v_f would reach the maximum possible value of 1. I think there are two possibilities: Either I am wrong, in which case slight modification of the manuscript could prevent other readers from making the same erroneous conclusion. Or I am at least partially right, in which case a thorough discussion of connected metapopulations vs. continuum limit and discussion of applicability of results by Gandhi et al. and Birzu et al. to this study would be needed.

Answer to comment 1: yes, we agree in principle that we are translating a framework build around a continuum to an all-discrete scenario, and that this could be the source of some problems. However, we do not expect this to be a problem in our case. Indeed, a previous paper (Haond et al., 2018) showed that the continuum approximation of v_f we use only leads to minor error in our simulation context, errors that are unlikely to influence our inferences. See our answer to the introductory comment by reviewer 2 for more details, and for how we revised the manuscript to make this clearer.

Regarding the second point raised by the reviewer in this comment, namely “Were r_0 or D_0 slightly higher, v_f would reach the maximum possible value of 1”. Yes, this is true, and something that we have been trying to wrap our head around (we expect cutting off all $v_f > 1$ to 1 would be needed). In our opinion, this means nothing more than “if a lot of individuals are produced, the front is guaranteed to advance every generation” or “if all individuals are very dispersive, front advance is guaranteed as long as fecundity is high enough”. In any case, this is expected to happen whenever space/time discretization and stepping-stone dispersal are both used (as then no velocity can go higher than 1 patch/generation, independently of what the formula for v_f says), so it would also happen in the analytical frameworks used by Gandhi et al and Birzu et al. This is unrelated to our simulation setup, or to the fact we used discrete population densities, and did not prevent Gandhi et al. and Birzu et al.’s insights.

2) I struggle following the paragraphs in which different treatments are compared in simulations because sometimes both v and v_f change. Why did the authors not choose to keep one constant (compare to Gandhi et al., 2019)?

Answer to comment 2: We actually tried to keep things as constant as possible (as can be seen in the “reference” vs “density-dependent dispersal” comparison, where we worked things to not change d_0) but changes in both v and v_f simultaneously are an unavoidable consequence of our treatments. In particular, lowering connectivity lowers d_0 by definition, so has to change v_f at the same time as v . Note that the “ v and v_f both change” scenario is actually also present in Gandhi et al. (2019), contrary to what the reviewer mentions (see Gandhi et al.’s Figure 3). We already discuss the results in terms of v/v_f ratios rather than actual v (see lines 413-418, 512, 518, 543-546 in results and discussion), which should help with interpretation.

3) Given the high stochasticity in the system and the authors' aim to make inference from limited information (as is the case out in the field), the use of Bayesian inference appears reasonable. However, I am afraid that this approach makes the paper less accessible and 'less sticky', at least to readers like me, who understand the concept of Bayesian analysis but are not be able to scrutinize the details. This approach might also make the findings more vulnerable to misinterpretation than necessary. One example is the exponential approach to v . Is this assumption supported by the data? Is this assumption actually necessary to extract v ? I suggest the authors include more of the raw data (front profile, speed as function of time) in the manuscript without sacrificing any of their Bayesian inference. Fig. 3 is a good example of how statistical rigor (to the extent I can judge) and intuition can be combined.

Answer to comment 3:

On the first part of the comment: The use of Bayesian statistics has no bearing on the structure of the models we used; the exact same models could actually be fitted with classical frequentist statistics (in theory at least; it is likely they would not converge easily, given the complexity). Whether or not the reader is familiar with Bayesian statistics is a priori irrelevant for whether or not they can interpret the results, since they only need to get the model formula (which as we said is independent of whether the model is Bayesian or not) and compare the relevant 95% intervals. We note however that Bayesian methods are becoming mainstream in quantitative ecology/evolutionary biology, that we follow state-of-the-art methods, and that our technical paragraph (lines 270-285) provides to the interested reader the minimal information to judge the validity of our modelling strategy and the needed references.

On the second part of the comment:

If we understood correctly, the reviewer is asking why we used the model formula we used for velocities. First, the idea that the asymptotic velocity is not reached immediately but after some time is well supported by theory (see e.g. Panja, 2004). Once we agree that an asymptotic approach is needed at our time scales, a second question is "why an exponential decay specifically?". For that, we send the reviewer to our answer to comment 2b by reviewer 2, since they asked a similar question but with more details.

If the question is "is any model actually needed? why not just take the v at the end of the simulation?". First there were no guarantees that simulations would have converged to the asymptotic v by the end of the run. We could admittedly have re-run the simulations for longer, but (1) the trade-off in computing power between running the simulations longer vs running them for 100 generations + using statistical models was clearly (by at least an order of magnitude) in favour of the second approach; (2) we had to develop the statistical model to analyse empirical data anyway (ours or in general, since it's unlikely anyone would have experiments or observations on macroscopic organisms lasting for 1000s, or even 100s of generations). Finally, the use of the model allows us to predict *when* speed reaches the asymptotic v , which may be useful for some management applications (even if we don't use it in the present manuscript).

But since the simulated data show *a posteriori* that asymptotic convergence was more or less reached at generation 100, we have overlaid "observed" speeds at $t = 100$ to the "simulated speed" figure for illustration.

Finally, the reviewer asks why we do not provide more information (namely speed as a function of time and front profile). We have now added figures on how speed change with time in Supplementary Materials (Supplementary Material 6).

We explicitly chose not to investigate front profile shapes, to focus on indicators easy to get in natural expansions, still following our aim of increased applicability (and because this would have made an already long manuscript even longer and unwieldy). Indeed, quality information on population density along entire front profiles is likely impossible to get for many “real” range expansions. Even in our experiment, while we have profile information in theory, estimation noise in population densities (due to computer macro) would likely translate to very high uncertainties in profile widths, making this variable unusable. As our choices were made early in development, data to calculate front profiles were not collected during simulations. So, while we believe for these reasons profile shape details are not necessary, if reviewers and editor insist on their inclusion, we are willing to do the job in a later revision (for simulations), but that would involve re-running simulations from scratch.

4) Heterozygosity – experiments: I’d appreciate a quick introduction on what is measured and the consequences of wasp mating. Could the authors also share their interpretation of Table S4.2?

Answer to comment 4: Information about what is measured is already presented in the usual format in the main text (lines 259-268) and Supplementary Material 4, so we are not sure what the reviewer is asking here. We do not expect experimental wasps mating (vs simulated organisms reproducing asexually) would have any qualitative consequences on our genetic diversity results, unless female wasps consistently select males that would bear some alleles over others; and given microsatellites are a priori neutral, this is unlikely. The possibility of recombination brought by sexual reproduction could possibly slow down evolution (as discussed in Erm & Phillips, 2020 with respect to adaptive, not neutral, evolution) and lead to lower rates of genetic diversity decay across the board, but we would not expect this to change qualitative patterns.

About table S4.2: these are mostly standard population genetics descriptive stats, presented here since these loci were not previously described in the literature, and it is customary to present these along with at least the first description. If one wanted to add an interpretation to this table, one could say that it confirms all 3 stock populations are diverse at most if not all loci at the start of the experiment. For the locus with all $H_0 = 0$, we note that it does present variation in later generation samples (not included in the table, but a prerequisite for keeping the locus) so, barring widespread mutation, these 0 must simply represent sampling error at $t = 0$. We have added two sentences with these details to the Supplementary Material 4

5) Heterozygosity – simulations: Why did the authors choose to use only two alleles?

Answer to comment 5: This is a simplifying assumption, in line with experiments/simulations from Gandhi and Birzu et al. We do not expect increasing the number of alleles at the neutral locus would qualitatively change results (see supplementary material of (Birzu, Hallatschek, & Korolev, 2018; Birzu, Matin, Hallatschek, & Korolev, 2019)). It would make the calculation of allelic diversity more complex, since allelic variance cannot as easily be estimated in a multinomial setting.

6) Is non-neutral evolution of the wasp population a concern that can be ruled out?

Answer to comment 6: We are assuming here the reviewer only refers to the experiments, since the simulation does not allow for non-neutral evolution of any form. It is possible, as discussed in (Birzu et al., 2019), that dispersal and fecundity evolve during range expansion in a way that affects the long term “pushiness” of the expansion. The short answer is that it is neither a concern nor can be ruled out, and that we expanded and modified the Discussion to make this clear.

As is now made clear in this revised version (lines 599-631), our *experimental* results are best explained if we assume reducing connectivity also leads to a change in density-dependent dispersal.

That is, the best simulation-experiment match is reached if we compare “reference” with “reduced connectivity + DDD” simulations. This change in the dispersal-density reaction norm with connectivity/costs of dispersal is plausible, predicted by theory (Travis, Murrell, & Dytham, 1999) and has been seen empirically (Govindan, Feng, DeWoody, & Swihart, 2015).

However, this does not imply evolution: plastic responses to the perceived dispersal cost/benefit balance are equally plausible a priori, as we also discuss (lines 624-631)

To support these points, in addition to the changes in the Discussion detailed above, we moved the “Density Dependent Dispersal + low connectivity” simulation treatment to the main text.

(Data that can disentangle evolutionary from plastic explanations have been collected on F2 descendants of the individuals studied here, and their analysis is ongoing. However, these data fall outside the scope of the present manuscript, and will actually take an entire manuscript (in prep.) for themselves.)

A number of minor comments:

7) Line 82: Is the statement that disaccharides lead to an Allee effect correct in this generality?

Answer to comment 7: We rephrased the sentence to instead explicitly say which sugar leads to an Allee effect, as we don't know whether the monosaccharide/disaccharide difference is actually generalizable. (lines 82-84).

8) Line 146: Visualise the function?

Answer to comment 8: We replaced Table 1 (which was redundant with the text) with a new Figure 2 showing the dispersal and fecundity functions

9) Line 154: Is there a prefactor N missing?

Answer to comment 9: No, as μ^* is the per-capita fecundity, not the population-level fecundity. We have rewritten slightly to make that clearer (lines 169-172).

* previously λ , changed to avoid confusion with other metrics named λ in-text

10) Line 189: I would have enjoyed a picture of the setup. For the general reader, this might be the first paper with this experimental setup they come across. Similarly, a flow diagram of the experiment might help visualize the experiment.

Answer to comment 10: We have added an annotated photograph of the setup (now Fig. 3).

11) Line 197: What does “16:8 L:D” mean?

Answer to comment 11: This is standard notation for the duration of light and dark phases in experimental studies

12) Line 199: Is there a concern of infecting the same egg twice?

Answer to comment 12: There is indeed relatively frequent superparasitism in *Trichogramma* wasps, but this is not a concern. While each host may occasionally be “infected” several times, in most cases only 1 individual survives within-host competition (Corrigan, Laing, & Zubricky, 1995). We estimate densities and photograph hosts late in development, a priori after this competition phase has ended. We have added a sentence to make this clear (lines 247-250).

13) Line 208: ‘until emergence of first adults’. Why not wait for all adults?

Answer to comment 13: The “until the emergence of first adults” refers, in the sentence, to the moment where we come back at the start of the cycle and add fresh eggs; if you follow the cycle, we did give 48 hours to all adults to emerge. This is admittedly confusing, we have removed “first” from the sentence (lines 250-251).

14) Lines 248: With this definition of v_t , *approaching v might take a long time. Why not define a moving average of the local speed which would allow one to disregard the initial phase of approaching v?*

Answer to comment 14: We believe the answer to this is in answer to comment #3.

15) Line 270: *How much does v_F change when changing from r_0 to r_1 and r_2 and D_0 to D_1 and D_2 ?* (See also next point.)

Answer to comment 15: We would first like to reassert using r_1 is *needed* in the case of Allee effects, since our formulation leads to $r_0 = -\infty$. Putting that aside, it would of course depends on the dispersal and fecundity functions, but in our specific example:

- the dispersal rate of reference changes at most, by 0.36% ($D_0=0.2$ versus D_1 with density-dependence and $K=225 = 0.2007121$; $D_1 > D_0$)

- the growth rate of reference changes at most by ~0.45% ($r_0 = 1.609438$ versus r_1 with $K=225 = 1.602285$; $r_1 < r_0$)

- so even using a worst scenario assumption (setting both the dispersal and growth “errors” to 0.45% and both into the same direction rather than cancelling themselves as they actually do), we get at most $v_F(1) \sim 1.0022$ times $v_F(0)$. This is a very minor “error”, and in any case much lower than the threshold for “true” pushed expansions (about 1.06 times $v_F(0)$, so this unlikely to affect any of our interpretations.

The fact r_1 and r_0 are virtually identical (same for d_1 and d_0) can be grasped from the newly added Figure 2, that shows the dispersal and growth functions.

16) Line 315: *As the reference scenario’s speed is very close to v_F , the dependence on K is a bit surprising. A brief comment might be helpful. Relatedly, what do the authors attribute the fact to that v_F for $K=225$ (v_F estimate, v estimate, others)?*

17) Line 375: Dependence on K : Can it be ruled out that it’s an indirect effect of changing K ?

Answer to comments 16 and 17: Our results regarding K in general largely replicate a previous article (Haond et al., 2018), hence why do not expand on them too much on the possible causes in the paper itself.

To answer comment 16, we now detail why there could a K-v relationship present even in the simulated reference (lines 530-536).

As for comment 17, Haond et al also analyse this and show that the relationship between K and v still persists when a new growth function, that does not change when K is changed, is introduced. This rules out, to our opinion, the “indirect effect of changing K” hypothesis.

18) Line 450: I unfortunately cannot follow the argument in this paragraph.

Answer to comment 18: we deleted this argument (which is of minor importance for the wider discussion compared to the other ones)

19) Supplementary Figure S7.1: What is the rationale behind using Bayesian inference here? Aren't the data directly available from simulations?

20) Supplementary Material S7: Is ‘based on our second model’ referring to ‘reduced connectedness + DDD’?

Answer to comments 19 and 20: this Supplementary Material has been deleted, as we do not use the argument it underpinned in the Discussion anymore.

Reviewer 2

The manuscript entitled “Shifts from pulled to pushed range expansions caused by reduction of landscape connectedness” presents the results of both individual-based simulations and experiments with minute wasps to investigate the role of “reduced connectedness” on populations undergoing a range expansion. The main messages are: (i) in addition to densitydependent growth and/or dispersal (studied previously), “reduced connectedness” can also lead to “pushed” expansions; (ii) the “pushed” expansions that can occur do not always match the current descriptions of pushed waves, in that they travel faster than pulled waves, as expected, but also lose genetic diversity faster, in contrast with current descriptions/understanding.

While the manuscript introduces a very interesting hypothesis, I am left doubtful on whether the results are sufficient to prove this hypothesis. More specifically, while the word connectedness is used in both titles and in the main text, which suggests a relationship with topology of the structured environment, the way in which it is implemented in the simulations is by reducing the basal diffusion coefficient of a sFKPP equation. As previously shown (see Hallatschek and Korolev, Fisher Waves in the Strong Noise Limit, PRL, 2009), this results in an increased relevance of population noise compared to the deterministic component of the reaction-diffusion equation (also seen in simulations), which breaks the assumptions that lead to the traditionally used Fisher velocity v_F as estimated for the “pulled” front velocity. In this reference, it is shown that the actual front velocity becomes slower than the predicted v_F as diffusion decreases, leaving me to wonder how the results presented in fig. 2 have been produced and whether they are an artifact of the way the simulations are analysed or v_F is estimated.

I also have major reservations regarding the experimental data, whose results in front speed are too qualitative, somewhat in disagreement with the theoretical predictions, and also not well controlled, since variations in growth rate are not accounted for. As a result, I don't believe the manuscript should be recommended for publications. I list all my other minor concerns in details below:

Answer to introductory comment: we would like to thank the reviewer for forcing us to think harder about stochasticity, by pointing us towards papers about the sKPP. Our answer is in two steps.

First, as far as we can determine, our model does not actually match 100% a sKPP, contrary to what the reviewer says, in one critical aspect. Indeed, the noise term in a “classical” sKPP only refers to demographic/ growth rate stochasticity, not dispersal stochasticity (e.g. “This decomposition implies that the front diffuses with diffusivity D independent of the noise strength b ” ; Hallatschek & Korolev, 2009). This interpretation is supported explicitly by (Giometto, Altermatt, & Rinaldo, 2017) among others in an ecological context, and indirectly by (Reluga, 2016; Schreiber & Beckman, 2020), as they found, like us, that dispersal stochasticity/variability should lead speed to increase, in direct contradiction with the sKPP predictions in (Brunet & Derrida, 1997; Hallatschek & Korolev, 2009). We note however, that our results can be seen as a new way to produce “weakly pushed” expansions sensu (Panja, 2004; Panja & van Saarloos, 2002). This led us to reframe the paper, in the Introduction and Discussion, around this concept (e.g. lines 105-112, 522-529).

Second, the question remains, of course, as to whether our use of the continuum version of v_f is justified, given the stochasticity (whatever its source). We believe it is, at least within the range of K we used. In an IBM structurally very close to the one in the present paper (stepping-stone dispersal in a 1D lattice, discrete time, dispersal and growth stochasticity), (Haond et al., 2018) showed that the use of the continuum v_f actually leads to only negligible error for $K >$ roughly 150 to 200. Based on their figures, at $K=225$, the deviation between the continuum approximation of v_f and mean speed in discrete pulled simulations is expected to be $\sim 1\%$, 2% at most, which is in line with what we found in reference scenarios. We have added this justification in the Methods where we present which v_f we use (lines 326-333).

1. There are several instances where key concepts or parameters were not well defined or explained, which made it initially difficult to understand the work, for example:

[Note from authors: we have taken the liberty to split comment 1 in several parts, to make our answer clearer]

o “(Reduced) Connectedness” – Despite being the titular concept in the paper, I initially found it quite difficult to understand what this actually meant in the context of this work. When it is first introduced (Ln. 90) the idea of “physical links” is used, which relates well to the experiments discussed later, but not to the simulations used, where “reduced connectedness” in practice means that the base dispersal rate D_0 is lower. This made it difficult initially to appreciate the significance of Fig. 1, which with hindsight does actually motivate the work reasonably well.

Answer to 1a: The original use of “connectedness” over “connectivity” stemmed from the original motivation of the study, highlighted in the introduction (lines 91-94): to find an experimental manipulation that would tend to generate pushed expansions without (too much) confounding. The reviewer is however right that our underlying argument is based on reduced connectivity/dispersal more generally. To make the manuscript clearer, we have thus changed all mentions of “connectedness” to “connectivity” throughout the manuscript.

o A more minor point, but the parameters discussed in the simulations (such as K , r_0 and D_0) are described as being biologically plausible (with citations), but this is

not obvious to me given that we don't know how large a "patch" is, or how long a "step" is, and all of the parameters have no units.

Answer to 1b: We have added more details about the sources of our D_0 , r_0 and K value choices. For dispersal, we rewrote the existing sentence to make it clear the rate is a typical rate based on data spanning many taxa (lines 186-189). For fecundity, we clarify that it is based on existing syntheses of insect data (line 195). For K , this is complicated by the fact natural densities are often reported in N per area, rather than N per patch; we nonetheless provide evidence that our K values fall within one order of magnitude of values observed in nature (lines 200-203), which is in any case much closer than most previous models of pushed expansions. In addition, we clarify an existing sentence explaining we also based K on previous experiments and models (lines 198-200).

However, we are not 100% sure we fully understand the reviewer's comment, especially the part about units, since most parameters' units are given implicitly or explicitly in the text (dispersal rates, growth rate, carrying capacity K ...). If we misunderstood and the reviewer is wondering about the physical area of the patch and the physical dispersal distance in the simulation, we believe they are actually irrelevant here, as they are "abstracted out". Assuming a patchy environment as we do, the only relevant patch characteristic for our purpose is K , which is explicitly given. And as we are using nearest-neighbour/ stepping-stone dispersal, the "dispersal length" implied by the model is by definition "the typical dispersal length of the species, whatever the species is".

2. I found it generally quite difficult to follow the methods section, particularly all of the statistical analyses that are used throughout the work.

[Note from authors: we have taken the liberty to split comment 2 in several parts, to make our answer clearer]

o In the simulations, the maximum dispersal in a timestep is limited to one patch, whereas in the experiments this is not true. Is this difference not important, and if not, why impose the condition in the simulations?

Answer to comment 2a: We do not believe the difference is qualitatively important for our results. Indeed, the fact that v_f can be estimated from D_0 and r_0 is independent from the dispersal kernel underlying D_0 . The formula for v_f may have to be adjusted (Lutscher, 2019), but the fact that a v_f exists _ and so that $v/v_f > 1$ are possible _ is independent from our choice of dispersal kernel. Indeed, qualitative agreement between an IBM with stepping stone dispersal and an wasp experiment with a more complex dispersal kernel has already been found, for instance, by (Haond et al., 2018).

Given we don't expect the dispersal kernel to make a qualitative difference, our use of a stepping-stone dispersal model in our simulation is by simplicity, and to be able to tie our work more directly to previous work (Birzu et al., 2018, 2019; Gandhi et al., 2019; Haond et al., 2018). We now mention the connection with these previous studies more explicitly (lines 150-151)

o Why using a statistical analysis for the simulations? The data should be plenty to be able to give the average front velocity. How is this front velocity determined (position of the front vs position of the half point)? Why using an exponential decay to fit the asymptotic velocity when it's known to approach it via power-law (see Birzu, 2019).

Answer to comment 2b. The use of statistical inference to extract key parameters from simulations is not infrequent in ecological studies (see e.g. Fournier, White, & Heard, 2019; Morel-Journel, Assa, Mailleret, & Vercken, 2019), as unless population N are very large (which they are not, by design, in our virtual experiments), it is very helpful to partition variation into its different sources. In addition, there were no a priori guarantee that asymptotic front velocity would have been reached by the end of runs; the use of a statistical model allows us to get the right speed even if the simulation is not yet at the asymptotic speed (for more on this, see our answer to comment 3 from reviewer 1). Finally, this allows for consistency of methods between simulation and experiments, and between metrics (since even in the simulation, the genetic metric of interest `_decay rate _` can only be known by statistical modeling).

But since the simulated data show *a posteriori* that asymptotic convergence was more or less reached at generation 100, we have overlaid “observed” speeds at $t = 100$ to the “simulated speed” figure for illustration.

As we mention in the text, front velocity is determined here by the position of the most advanced occupied patch. While “front halfpoint” is indeed standard in studies that are based on continuum models, a true edge exists in naturally discrete populations. In addition, data from the most advanced patch(es) are easy to obtain from natural invasions, while data for the entire expansion density profile (needed to know the front halfpoint) would be much more complicated to get in many cases. Results that are based on the most advanced patch are therefore much more ecologically relevant and transferrable.

We added a comparison of power-law decay vs exponential decay models in our code. For experimental data, we find that the power law decay model has a slight predictive advantage indeed, but not large enough to affirm with certainty it is better. In any case, the qualitative conclusion (no difference in asymptotic speed between treatments) remains unchanged. For simulation data, the exponential model actually has a large advantage over the power model. This advantage can be seen in Supplementary Material 6, where the power model consistently misses the fact velocities have converged, leading to underestimations towards the end of the simulations.

Based on these, we decided to go with the exponential model in both scenarios (for consistency). We have added a sentence to the methods briefly explaining that a power law fit could have been justified, and why we chose an exponential fit instead (lines 302-307). Both models are present, commented, in the published analysis code.

o I don't understand the description in S3 about determining whether the reduced connectedness did limit movement/dispersal. Additionally, it seems like the difference is (averaged over everything) significantly less than one patch. Given that eggs can only be laid in integer patches, is this difference significant, especially given that no difference is within the error bounds? Also, was the 'growth rate' in the experiments unaffected, i.e. did they get the same number of new wasps with each generation in the two regimes? These points are important, because it's used in the discussion to justify why the reduced connectedness expansions must be pushed – see later comment.

Answer to comment 2c.

On dispersal: As seen in our answer to comment 1 from reviewer 3, our experimental protocol here relies on the fact that dispersal distance is, almost by definition, the distance between parental starting patch and offspring birth patch (see there for changes we made to clarify this point). So any

protocol that measures how far from the starting point, on average, eggs are laid is measuring dispersal. Regarding the difference between treatments: first, the reviewer is mistaken here; the 95% compatibility interval of the difference in dispersal distances between the two treatments does not actually include zero (see right panel of supplementary figure S3.1). More generally, we fail to see the problem with average distances lower than 1, even with a discrete variable (it is perfectly possible for the average of a sequence of 0s, 1s and 2s to be <1 if zeroes are numerous enough). To answer the larger point, the treatment reduces dispersal distances by 20%, which is not a small effect size by any means (it is less than 1 patch, yes, but only because the average distance dispersed in the reference is 1 patch).

On growth rate: Our wasps in both treatments are sampled from the same source populations, so there is no reason to think the low-density growth rate/fecundity differs between treatments at the start of the experiment (the same holds for the base dispersal rate, independently of the effect of the connectivity treatment itself). One may still wonder if random sampling may have created artificial differences: but the probability of multiple $N \approx 300$ samples (N probably $\approx 150-200$ if we only count females) to deviate in their mean from the population average significantly, and systematically enough to create artificial treatment differences that would affect our inferences, seems in our opinion negligible.

In any case, we actually have collected low-density fecundity data (which would be a proxy for r_0) after the experiment, from descendants of the wasps that colonized the landscapes (for another, in prep., article). Based on preliminary analyses, there are actually no detectable differences between the two treatments, or between core and front patches, confirming our assumption is valid.

We have updated the discussion to make it more explicit our “ r_0 equal” assumption relies on the fact both treatments come from the same stock populations (lines 548-550).

o Specifically, on how the experiments were carried out, it says when the first adults start to emerge from the previous generation's eggs, new eggs are provided, and after 48 hours, the adults and old eggs are removed. My question is how do you know the new adults didn't infect some of the uninfected old eggs, which are then removed 2 days later? This could affect effective growth rates etc, which again could impact the argument later about VF in the discussion.

Answer to comment 2d: First, we want to point out that this is an unavoidable constraint of the experimental protocol in this species, as it is not logistically feasible to individually remove eggs without disrupting the vials and the developing parasitoids that have not yet emerged.

Second, host acceptability and parasitoid fitness generally decrease with host egg age, including when hosts are irradiated (Kishani Farahani, Ashouri, Zibae, Abroon, & Alford, 2016; Ksentini, Jardak, & Zeghal, 2014; Pandir, Sahingoz, & Ercan, 2013), so we would not expect newly emerged *Trichogramma* to try and infect the (limited number of) old eggs first when there are plenty of new eggs to infect first.

3. A few thoughts on the results section:

[Note from authors: we have taken the liberty to split comment 3 in several parts, to make our answer clearer]

o One of the reference velocities ($K=225$) is lower than VF, which isn't addressed

other than to say it's "very close," but I would argue that this statement may or may not be justified depending on the error. From the figure, the result doesn't seem statistically compatible with the Fisher velocity predicted.

Answer to 3a: As the reviewer pointed themselves in their introductory comment, demographic stochasticity can lead to v_f lower than the "nominal" v_f we used. As mentioned in our reply to that comment, based on (Haond et al., 2018), the 1% difference is indeed fully explainable by demographic stochasticity and discreteness. As mentioned in our answer to that introductory comment, we have detailed the Methods, and the Discussion to make it clear that this discrepancy is expected, why it is expected, and that it does not affect our conclusions (lines 326-333, 514-516). Indeed, it is easy to see from Fig. 4 that our main result (low connectivity leads to pushed expansion) holds whether or not we correct for that 1% discrepancy in the reference speed.

o I think it would be interesting, if possible, to see the diversity decay rates in terms of an effective population size, and how these related to the actual population sizes that are in the simulations/experiments. This concept is alluded to later in the discussion, but not actually shown for most of the data/rates in the paper.

Answer to 3b: in simulations, the effective population size is (approximately) the inverse of our decay rate λ , $N_e=1/\lambda$. In diploid individuals, $N_e= 0.5/\lambda$. However, it is not clear to what extent the $0.5/\lambda$ estimate is valid for our experiment, since wasps are not diploid but haplodiploid. We therefore chose to not explicitly analyse the data in terms of N_e , since (a) this is not needed if the N_e - λ relationship is made clear (b) this would risk introducing error for the experiment scenario.

If the reviewers judge it useful (and not source of confusion), we can easily add a secondary y axis on the genetic decay plots, showing the corresponding effective sizes N_e , in the next revision. This would need to be accompanied by a warning in the figure caption that this is for illustrative purposes only, especially for the haplodiploid wasps in experimental data. In the meantime we have added a sentence in the Methods highlighting how decay rate and effective population size are connected (lines 370-372).

We cannot go further as, despite intuition, the ratio N_e/N or N_e/K cannot be used here to determine whether an expansion is pushed. Indeed, the relationship between N or K and N_e is generally nonlinear with the shape /exponent both system-specific and depending on the degree of "pushiness" itself (Birzu et al., 2018), and anyway our K are too low to use the scaling relationships defined by Birzu et al (2018, 2019)(they mention in their supplementary material that the scaling breaks down at $K < 10000$).

4. I have a few issues with the argument outlined in the discussion about why the experimental expansions in the reduced connectedness regime must be "more pushed." The basis of the argument is that the velocity is the same in both cases, and that V_F must be lower in the reduced connectedness case, leading to a larger V/V_F ratio in the reduced connectedness case, meaning it must be "more pushed." My issues are:

[Note from authors: we have taken the liberty to split comment 4 in several parts, to make our answer clearer]

o The velocities are the same, but with very large error bounds (Fig. 3b)

Answer to 4a: Yes, but that uncertainty will be inevitable in ecological « realistic » contexts, and ties to our broader point about the usability of the pushed/pulled framework in practice. But we do see the point the reviewer is making and modified the discussion to make the uncertainty clearer (lines 553-555).

o They say that VF must be lower in the reduced connectedness case because the initial dispersal is lower.
§ As I mentioned earlier, I'm not sure how significant the lower dispersal is when the difference is much less than one patch, and has comparably large errors to those on the velocities (indeed, the error bounds include zero difference).

§ This also assumes, as they say, that everything else, such as the growth rate at low densities remains the same. I'm surprised this doesn't appear to have been checked given how fundamental it is to the argument. Given the variability/errors in all the other measurements, I wouldn't be surprised if this also varied significantly, and may cancel out any reduction in dispersal.

Answer to 4b: see answer to comment 2c

5. I also don't completely understand the argument put forward to explain why the genetic diversity decays *faster* in the reduced connectedness case, even though velocities would indicate that it is pushed, and therefore should decay slower.
o Basically, I'm not convinced as to why the cases where there is no movement into the first unpopulated patch ($N_{X+1,t+1}=0$) should be removed. Their argument for this is that there is no "new" population in this case, and so it is not relevant, whereas I would argue for every step where this is true, the previous new population becomes larger. Consider the toy example below

t=1 $N_X=1$ =lots $N_{X+1}=0$ $N_{X+2}=0$

t=2 $N_X=1$ =lots $N_{X+1}=5$ $N_{X+2}=0$

t=3 $N_X=1$ =lots $N_{X+1}=20$ $N_{X+2}=0$

t=4 $N_X=1$ =lots $N_{X+1}=30$ $N_{X+2}=2$

In this example going to the t=2 step would give a new population of size 5, and to the t=4 step would give you a new population of size 2, with the step to t=3 being discarded. I think though, given that no movement occurred in step from t=2 to t=3, then the "new" population size at position x=2 is in the end 20, because the 5 that were there first did not advance the front, and so going from step t=3 to t=4 those 5 no longer have any advantage over the other 15. At t=3, I think the effective population size at the front is therefore 20, so I think by removing these steps the effective population size measured is being artificially reduced, which is key to the argument used to explain why diversity decreases faster (smaller population=faster loss of diversity).

Answer to comment 5: while we see the point the reviewer is making, our initial argument relied here on what happens at the **foundings** of new populations at the edge. Hence, using front populations that are generations old (as would be the case if we kept the $N_{x=2,t=2}$ and $N_{x=2,t=3}$ populations, in the reviewer's argument) would be inappropriate. Our use of that metric is not new: it has been proposed by e.g. (Williams, Hufbauer, & Miller, 2019).

In any case, alterations to the Discussion mean we do not rely on the argument these data helped sustain anymore; this entire Supplementary Material has been deleted.

REVIEWER 3:

Major points

I would like clarification about the degree to which changing the number of tubes between the vials in the experimental landscapes reduced dispersal between vials. I was unsure of how to interpret the reduction in mean egg laying distance between the reduced connectedness landscape and the reference landscape in terms of a reduction in dispersal. I found it more difficult then to compare the results of the experiment and simulation without knowing the degree to which dispersal was reduced in the experimental landscapes. I do not have the expertise to comment on whether the IBM model setup or statistical analysis framework were appropriate. They were extensively described.

Answer to comment 1: Dispersal is often defined as “movement leading to gene flow in space” or “movement between your own site of birth and your offsprings” (both definitions meaning nearly the same thing). By that definition, the average distance between the release site and the egg-laying site should be the average dispersal distance, so any reduction of the former is a reduction of the latter. We've explicated the above clarification in the main text (lines 252-258)

Minor points

(...) The language in general was clear. There were a few instances of vague pronoun references ex. line 96 “This can increase the influence of population density on dispersal success,...” It was unclear to me based on the previous sentence what “this” was referring to. Ex. line 100 “They are however much lower than those considered in most pushed expansion models...”; line 469 “This is not without precedent...”; line 473 “this shows more studies...”

Answer to comment 2 : These have been explicated (“Reduced dispersal rate”, line 98, “These are however much lower densities than...”, line 104, [sentence deleted in revision] and “...our results highlight more studies...”, line 647, respectively)

It may be useful to add a figure illustrating the experimental setup

Answer to comment 3: We have added a photograph of the setup as Figure 3

For clarity, I would recommend adding “(core patch $t=0$)” after “origin” in the Figure 5 key

Answer to comment 4: We have added ($x = 0, t = 0$) instead, as technically, the origin patch is both the core and the edge patch when the experiment starts.

Line 14: replace “these phenomena” with “range expansions”

[Answer to comment 5: This is done](#)

Line 83: Does “changing the resource” refer to changing the quality of the resource to increase the strength of the density dependence of dispersal?

[Answer to comment 6: Yes, the sentence has been rewritten to make this explicit \(lines 85-87\).](#)

Line 311: Grammatical construction is a bit confusing. Perhaps “Relative velocities in the reference landscapes were close to those thus expected for pulled expansions”

[Answer to comment 7: This has been corrected following the reviewer’s suggestions \(lines 413-414\)](#)

Line 413-417: Am I correct in my understanding that the relative number of individuals contributing to a new population is higher under the reduced-connectedness scenario because the source populations in these landscapes are smaller (because of reduced dispersal) so the ratio of individuals that disperse from a source patch to the source patch population is higher? If I am correct, I would recommend that the authors clarify this sentence.

[Answer to comment 8: Yes. But as is obvious from the multiple comments from the reviewers, and from discussions during writing, this is a concept we have struggled to make clear even for ourselves. Because of this, and because the revision has shown this demonstration was not needed for the purpose of this article, we have deleted these sentences and the accompanying analyses.](#)

Line 444: I think it would be helpful to expand upon how the simulated expansions corresponded better to the results of the experiment. Is the result of this simulation presented in one of the figures?

[Answer to comment 9: Following a reorganization of the manuscript, we have moved the 5th “DDD + low connectivity” treatment \(the result in question here\) from supplementary material to the main text and figures, so the discussion could also be easier to follow. We have expanded the discussion sentence to detail more explicitly how adding density-dependence brings the patterns in the simulated “reduced connectivity” treatment closer to its experimental counterpart \(relative to their “reference” treatments\) \(lines 599-631\).](#)

Line 449: “spatial behavior in response, which evidence suggests they do...”

[Answer to comment 10: this has been changed accordingly \(lines 622-624\)](#)

Line 460: I would recommend moving the explanation for why the authors do not believe that differences in equilibrium population sizes drove differences in genetic diversity losses between the experimental and control landscapes from the supplementary material to the body of the manuscript. My a priori assumption would be that this alternative explanation could drive substantial differences in the rate of genetic diversity loss and I think it would be useful to expand the discussion of this alternative explanation in the manuscript.

Answer to comment 11: Following changes in the structure of the manuscript, the analysis of equilibrium population sizes has been moved to the main text. At the same time, we've redone the analysis with more stringent criteria for model acceptance (as the data moved from "anecdotal supplementary data" to "key part of the main text"): although the pattern in that direction remains, there is no statistically clear evidence that low connectivity leads to an increase in equilibrium density in our system. This alternative explanation to genetic diversity patterns can thus be excluded even more strongly (it could already be, based on simulation results, but this is further evidence).

Line 469: clarify that "in these contexts" refers to "under reduced landscape connectivity"

Answer to comment 12: this sentence has been clarified ("when (weakly) pushed expansions are caused solely by reduced connectivity.", line 635-636).

Line 472: I think it would be helpful for the authors to clarify what they mean in advising empirical researchers to "be careful about which questions they ask" in light of their results.

Answer to comment 13: in an ecological context, the "pushed expansion" concept maps to changes in population genetics, expansion speeds and population density profile along the expansion. It is often implicitly assumed these 3 things are linked; our results show they aren't necessarily (at least for speed and genetics). This means researchers cannot use e.g. speed-based evidence of pushed expansion to make inferences about population genetics; if their question of interest is about population genetics, they need to either measure what they need directly, even if the other metric would be easier/cheaper, or provide more solid evidence that the two metrics are linked. We have added a sentence to the discussion to make this point clearer (lines 641-646)

Line 474: Does "types of pushed expansions" refer to types resulting from different drivers of pushed expansions?

Answer to comment 14: Yes, we've rewritten the sentence to make this clearer (lines 646-648)

Line 480: Perhaps it is a little strong to conclude that reducing connectivity may benefit expanding species based on a slowing of the decline in neutral genetic diversity. Perhaps this concern is addressed in the following citations; however, I think this argument could benefit from explicitly stating the evidence provided in these citations if that is the case.

Answer to comment 15: Our argument is based on the fact lower genetic diversity/ effective population size has well documented negative effects (inbreeding, loss of adaptive potential, higher risk of extinction...), so our results imply that "in some cases", reducing connectivity might reduce these negative effects, providing relative benefits. We have hedged further by replacing "may" by "might", and by expliciting the benefits of more genetic diversity (less inbreeding, more adaptive potential), with citations (lines 649-654).

Line 486: I think this sentence would be clearer if the structure were flipped. "We call for... because..."

Answer to comment 16: We believe ending the article by the “call” itself rather than a string of citations is more “impactful”. But we see the point the reviewer makes, and split the sentence in two to make it clearer (lines 657-663):

“Finally, density-dependent dispersal (...) and Allee effects (...) themselves may evolve during range expansions, and the effects of habitat fragmentation on dispersal ecology and evolution are abundantly documented (...). For these reasons, we call for more systematic eco-evolutionary studies of context-dependent dynamics during range expansions and shifts.”

We are open to altering the conclusion further if needed.

REFERENCES CITED IN REPLIES

Birzu, G., Hallatschek, O., & Korolev, K. S. (2018). Fluctuations uncover a distinct class of traveling waves. *Proceedings of the National Academy of Sciences*, 115(16), E3645–E3654. doi:

10.1073/pnas.1715737115

Birzu, G., Matin, S., Hallatschek, O., & Korolev, K. S. (2019). Genetic drift in range expansions is very sensitive to density dependence in dispersal and growth. *Ecology Letters*, 22(11), 1817–1827.

doi: 10.1111/ele.13364

Brunet, E., & Derrida, B. (1997). Shift in the velocity of a front due to a cutoff. *Physical Review E*, 56(3), 2597–2604. doi: 10.1103/PhysRevE.56.2597

Corrigan, J. E., Laing, J. E., & Zubricky, J. S. (1995). Effects of parasitoid to host ratio and time of day of parasitism on development and emergence of *Trichogramma minutum* (Hymenoptera: Trichogrammatidae) parasitizing eggs of *Ephestia kuehniella* (Lepidoptera: Pyralidae). *Annals of the Entomological Society of America*, 88(6), 773–780. doi: 10.1093/aesa/88.6.773

Erm, P., & Phillips, B. L. (2020). Evolution transforms pushed waves into pulled waves. *The American Naturalist*, 195(3), E87–E99. doi: 10.1086/707324

Fournier, A. M. V., White, E. R., & Heard, S. B. (2019). Site-selection bias and apparent population declines in long-term studies. *Conservation Biology*, 00(0), 10.

Gandhi, S. R., Korolev, K. S., & Gore, J. (2019). Cooperation mitigates diversity loss in a spatially expanding microbial population. *Proceedings of the National Academy of Sciences*, 116(47), 23582–23587. doi: 10.1073/pnas.1910075116

- Giometto, A., Altermatt, F., & Rinaldo, A. (2017). Demographic stochasticity and resource autocorrelation control biological invasions in heterogeneous landscapes. *Oikos*, *126*(11), 1554–1563. doi: 10.1111/oik.04330
- Govindan, B. N., Feng, Z., DeWoody, Y. D., & Swihart, R. K. (2015). Intermediate disturbance in experimental landscapes improves persistence of beetle metapopulations. *Ecology*, *96*(3), 728–736. doi: 10.1890/14-0044.1
- Hallatschek, O., & Korolev, K. S. (2009). Fisher waves in the strong noise limit. *Physical Review Letters*, *103*(10), 108103. doi: 10.1103/PhysRevLett.103.108103
- Haond, M., Morel-Journel, T., Lombaert, E., Vercken, E., Mailleret, L., & Roques, L. (2018). When higher carrying capacities lead to faster propagation. *BioRxiv (Peer-Reviewed and Recommended by Peer Community In Ecology)*, 307322. doi: 10.1101/307322
- Kishani Farahani, H., Ashouri, A., Zibae, A., Abroon, P., & Alford, L. (2016). The effect of host nutritional quality on multiple components of *Trichogramma brassicae* fitness. *Bulletin of Entomological Research*, *106*(5), 633–641. doi: 10.1017/S000748531600033X
- Ksentini, I., Jardak, T., & Zeghal, N. (2014). Could *Trichogramma* species be provided with old UV-killed *Ephestia kuehniella* eggs during mass rearing programs? *Bulletin of Insectology*, *67*(2), 6.
- Lutscher, F. (2019). *Integrodifference equations in spatial ecology*. Springer International Publishing. doi: 10.1007/978-3-030-29294-2
- Morel-Journel, T., Assa, C. R., Mailleret, L., & Vercken, E. (2019). Its all about connections: Hubs and invasion in habitat networks. *Ecology Letters*, *22*(2), 313–321. doi: 10.1111/ele.13192
- Morel-Journel, T., Girod, P., Mailleret, L., Auguste, A., Blin, A., & Vercken, E. (2016). The highs and lows of dispersal: How connectivity and initial population size jointly shape establishment dynamics in discrete landscapes. *Oikos*, *125*(6), 769–777. doi: 10.1111/oik.02718

- Pandir, D., Sahingoz, R., & Ercan, F. S. (2013). Mediterranean flour moth *Ephestia kuehniella* eggs and larvae exposed to a static magnetic field and preference by *Trichogramma embryophagum*. *Biocontrol Science and Technology*, 23(12), 1402–1411. doi: 10.1080/09583157.2013.835789
- Panja, D. (2004). Effects of fluctuations on propagating fronts. *Physics Reports*, 393(2), 87–174. doi: 10.1016/j.physrep.2003.12.001
- Panja, D., & van Saarloos, W. (2002). Fronts with a growth cutoff but with speed higher than the linear spreading speed. *Physical Review E*, 66(1), 015206. doi: 10.1103/PhysRevE.66.015206
- Reluga, T. C. (2016). The importance of being atomic: Ecological invasions as random walks instead of waves. *Theoretical Population Biology*, 112, 157–169. doi: 10.1016/j.tpb.2016.06.002
- Schreiber, S. J., & Beckman, N. G. (2020). Individual variation in dispersal and fecundity increases rates of spatial spread. *AoB PLANTS*, 12(3). doi: 10.1093/aobpla/plaa001
- Travis, J. M. J., Murrell, D. J., & Dytham, C. (1999). The evolution of density–dependent dispersal. *Proceedings of the Royal Society of London B: Biological Sciences*, 266(1431), 1837–1842. doi: 10.1098/rspb.1999.0854
- Williams, J. L., Hufbauer, R. A., & Miller, T. E. X. (2019). How evolution modifies the variability of range expansion. *Trends in Ecology & Evolution*, 34(10), 903–913. doi: 10.1016/j.tree.2019.05.012