

My main comment is that the text as it stands is very long, especially part II, where a very long list of examples is cited and reviewed. Both reviewers have alluded to the same issue, but both have offered a very similar solution in the form of a summary table. I really like this idea and encourage you to explore it as an excellent way to make the text much more concise and easy to go through. The level of clarity and concision achieved in the Perspectives is ideal.

I also like the idea of reviewer #2 to present more clearly how to test the three macroevolutionary scenarios for a given dataset using the comparative toolbox currently available.

Thank you for considering our manuscript in *PCI Evolutionary Biology*. We sincerely appreciate the recommender's and the reviewers' efforts and comments, and have revised the manuscript following their suggestions. Notably, we added a table that synthesizes results of empirical papers (Table 2), resulting in a more concise paper ; we also added a figure (Figure 2) presenting some straightforward analyses to test the general predictions of the host-driven speciation hypothesis and the more specific predictions of the Oscillation hypothesis. (see answer to reviewer 2 for more details)

Reviewed by anonymous reviewer, 2019-03-26 14:23

Jousselin et al. give an interesting (and probably quite comprehensive) overview of hypotheses and studies testing phytophagous insect diversification in relation to their host plants. This is an interesting topic, and the review is generally clear and well written (could be a bit more concise in places, and needs in parts clarification, check for a couple of minor spelling mistakes, some I indicated below). I especially like the Perspectives part.

I have a couple of suggestions to improve the ms, indicated below. More generally, it may be useful to make a table indicating support for the different hypotheses (e.g. for different study groups/taxonomic groups [would be useful to use taxonomic symbols/silhouettes], and indicate the reference, and the method used, for example) (this would support the text with the sum-up of examples, which is quite long at the moment). The same (a table, or maybe better a figure) could perhaps be done for the Perspectives ideas.

As suggested by both reviewers and the recommender, instead of commenting the results of all the different studies out there in the main text, we now provide a table with the different predictions that have been derived from macro-evolutionary scenarios in the literature, how they have been tested and the conclusions of the authors. Taxonomic silhouettes have been used to ease reading (thanks for the suggestion). We chose not to use a Table for perspectives as this part was already quite concise.

We have also added a figure that presents a set of analyses to test host-driven speciation scenarios. This set of suggestion is not meant to be exhaustive but rather a set of relatively straight-forward analyses that test some predictions that are "uncontroversial" that is to say well accepted in the literature.

L48 Not clear whether hybrids will thus be selected against, or that this intermediate phenotype has the potential to become a new species through reproductive isolation. Or both. In case of both, perhaps some more information about the commonness of 'selection against hybrids' vs. 'hybrids become new species' can be presented.

We meant counter selection of hybrids. We added that “hybrids will be selected against” line 49

L76 This overview is interesting, but not particularly novel. To really move forward in this field, I guess it would be great to incorporate the available fossil evidence, to calibrate the radiations / estimate diversification rates, but perhaps also to obtain a better idea of insect-hostplant associations in the past, if such things fossilize (or co-occurrence of species through insect/plant fossil assemblages perhaps).

Yes, we admit that this is not particularly novel, but here the idea was to present the main approaches that have been used throughout the literature since the paper by Erlich and Raven and how they have evolved along with the development of phylogenetic methods, not really to present novel approaches. The incorporation of fossil evidence for each partner of the interaction is implicit when dating phylogenetic trees—and therefore estimating diversification rates (line 254). Unfortunately incorporating “fossilized associations” is actually not very feasible, as it is difficult to identify with certainty the host plants of fossilized insects and it would be difficult to actually conclude on “long-term”, specific associations based on that data. In order to better incorporate the use of fossil data in the review, we are now referring to investigating co-occurrence of fossil assemblages (referring to the work of Labandeira et al.) and advocate that more studies combining fossil information and phylogenetic information would be useful in the field (lines 267-271).

L78 an extra ‘and’

corrected

L88 Why only speciation rates? I would say diversification rates, which would include both speciation and extinction. I imagine certain host-plant relationships are less vulnerable to extinction (e.g. more generalized as opposed to specialized relationships?)

The reviewer is right, we tried to correct throughout the text when we meant “speciation” and “diversification” (i.e. speciation – extinction) . There was already a paragraph (line 575 in the previous version, from line 464 in the new version) commenting the difficulty of predicting the impact of specialization towards host-plants on extinction rates . The literature on plant-insect interaction is actually mainly oriented towards investigating speciation rates, without taking into account extinction rates -i.e. mostly setting extinction rates to 0.

L174 less fewer

corrected

L182 The musical chairs hypothesis needs a bit more detail. For example, the description does not really match the figure (1c), and it’s only indicated how it differs from the second hypothesis, not how it differs from the first, and more importantly, what makes it ‘unique’ .

We have now rephrased and underlined what makes the musical chair hypothesis unique and how it is different from the *Escape and Radiate* and the *Oscillation* hypothesis. This paragraph has been entirely rewritten. Also line 458, towards the end of the second section,

we underline that the *Musical chair* predictions are mainly rebuttals of the *Oscillation hypothesis* and, therefore, without undermining its validity, we think that is harder to validate.

L203 What about the methods that test for host switching and co-diversification – are those not required to test some of these hypotheses as well?

The cospeciation methods actually all assess whether the phylogenies of plants and insects are more congruent than expected by chance. The null hypothesis is cospeciation (i.e. more congruence than expected by chance in the phylogenetic trees). These methods cannot be applied to test for the importance of “host-switching” in alternative macroevolutionary scenarios. Both *Oscillation* and *Musical chairs* will yield a “not significant result when using cospeciation methods

L212 insect insects

corrected

L220 But a lack of support for coevolution has also been often found, or?

Yes the reviewer is right, we have entirely rewritten this paragraph giving a brief summary of the reviews by Winkler and Mitter 2008 and de Vienne *et al.* 2013. The previous version of the MS gave too much importance to cospeciation studies and studies supporting phylogenetic tracking though those are actually a minority in the literature. Furthermore the field has moved away from this type of investigation so we have now shortened the whole paragraph.

L230 and more generally – some predictions remain quite vague and could do with a bit more detail. For example, concepts/terms such as ‘high lability’ (in speciose lineages breadth) high compared to what? ‘Few transitions in host breadth’ – few compared to what? What is, and how does one measure ‘lability in host association’? ‘following the capture of a new host-plant lineage’ does that not directly indicate that there is host tracking? Or how can there be shifts to new host-lineages without tracking? ‘Conservatism of host-plants’ – compared to what? Conservatism in what? How does conservatism shift on a phylogeny – is it (or linked to) ‘intrinsic’ traits? ‘recurrent transitions in host breadth’ – what is recurrent, how often, how commonly?

In short – always make the comparison, i.e. larger (e.g. geographical range) than what. Make clear what ‘lability’ and ‘conservatism’ are – these are vague and difficult concepts.

We agree that these words and concepts are vague and vary according to authors. In this table we are paraphrasing the literature, which actually uses a lot of vague wording (that is something we are referring to line 289) and many studies do not agree on how to test the predictions laid out in the table. We have been more specific whenever possible and we have also defined phylogenetic tracking, conservatism and lability (which are all used extensively in ancestral character state reconstruction applied to phytophagous insect groups) in the glossary. We know these terms can be vague and can sometimes mean different things for different people. We hope that the glossary helps as well as the inclusion of Table 2 that provides details on how the predictions laid out in Table 1 were actually tested in the literature.

To answer more specifically, one measure ‘lability’ or “conservatism” in a character (host breadth or host use) using indicators of phylogenetic signal (this was indicated line 268 in the previous version, now line 280 citing specific methods), this is now in the glossary.

‘Following the capture of a new host-plant lineage’ does that not directly indicate that there is host tracking? Or how can there be shifts to new host-lineages without tracking?”

Phylogenetic tracking specifically implies phylogenetic congruence between insects and their host plants (now defined in the glossary). So, no, the capture of a new host plant lineage does not imply phylogenetic tracking in its strict definition.

L234 host-plant adapted traits – such as?

We meant, as defined in the glossary, “Host-plant adaptation: heritable trait that confers a selective advantage on a particular host-plant”. We are now more specific in the title line 341, “traits involved in host plant choice” (*e. g.* , detoxication enzymes, behavioural traits...).

L237 But I guess these studies have not found support for all predictions from the hypothesis, or? Maybe write something about the methods they used and how reliable their conclusions are.

Indeed, most of these studies show a less than perfect pattern of phylogenetic tracking. That’s why we had written in the previous version of the MS “partly mimicking”. Here we were only referring to papers that have deployed cospeciation analyses (of some sorts) and showed some level of congruence between the phylogenies of both interactors –i.e. the phylogenies were more congruent than expected by chance though the timing of speciation did not match. We have totally rewritten this paragraph, referring to the syntheses of Winkler and Mitter 2008 and de Vienne et al 2013. We did not add anything on the reliability of the conclusions of these studies as this is already discussed in the two reviews cited here. To make it brief some conclusions are not reliable because of a lack of resolution of the phylogeny (this affects most studies from the nineties), or sub-sampling or false positive.

L265 How can host history be mapped on a tree when there are multiple associations per species? Or how are host associations classified?

They are generally conducted at the host plant genus or family level (now specified in the text). Multistate characters can also be used. This has been added in the MS line 278.

L282 What if the host shift is to the nearest living relative of the current host plant? Or would that be called host tracking instead of host switching? Perhaps make clear.

Host shifts to the nearest living relative are best investigated through cospeciation methods: you expect to find phylogenetic tracking (now defined in the glossary) if shifts happen between the nearest relatives. If host shifts happen within one plant lineage you find host plant conservatism (at the genus or plant level).

L294-295 rephrase sentence

done

L300 few species of what? Plants? Angiosperms?

This has been deleted in the new version.

L326 I wonder whether the availability of (suitable, e.g. chemically matching) hosts also plays a role in the host-breadth evolution of insects. This therefore depends on the diversification of particular host plant lineages. Perhaps this could be mentioned here, or elsewhere.

This is an interesting point, however this has not been considered in the papers we reviewed here as we were focusing on studies that have investigated how host-switches have influenced insect speciation. The reviewer's comment refers more to the factors that determine an insect's host-breadth (the number of host plants an insect can feed on). This is of course related but we found it hard to include this comment in our narrative.

L333 Here, and more generally: what is specialized and what is generalized? Should that always be in comparison to something else (e.g. other species or previous/ancestral state?)

Specialists and generalists are defined in the glossary; specialized means in this context "being a specialist".

L349 Why would these be the result of range expansion? What is that based on?

We meant host breadth expansion, sorry, we have rephrased to make it clearer in the new version.

L378 disruptive selection to what - host plants? Make explicit, because disruptive selection could have still played a role during speciation.

As the studies are now summarized in table 2, this part referring to disruptive selection has been deleted.

L438 I wonder whether it may not be just phytophagy vs. non-phytophagy driving the high diversification rates, but phytophagy in interaction with other (extrinsic, or intrinsic) traits (such as host plant diversification / availability). This would require more sophisticated trait and interaction-dependent diversification models. Or, alternatively, a similar trait-based approach to Onstein et al. 2017 (Nature Ecology & Evolution) could be taken, in which the authors infer speciation rates based on important interaction-relevant traits (in that case of frugivory). Those traits would tell you something about the interaction (e.g. chemical traits, phytophagy and or host-breadth) as well as the consequence for speciation based on host-plant characteristics (e.g. host-plant diversity or diversification, or their variation in chemical traits).

True. We have now mentioned that other characters can explain high diversification rates – from line 5745 (as actually suggested in the studies cited, and mentioned in Table 2 in the "conclusion" column). The merits of studying interaction-relevant traits are mentioned in the perspective, and we have now added a sentence referring specifically to the use of *SSE methods in this context

L498 changed changes

done

L509 geography, or other factors. Explain why geography is likely in this case.

This has been deleted (now this study is mentioned in Table 2) and we refer specifically to concomitant shifts and geographic shifts that render interpretation difficult.

L591 Is such a phylogeny available?

No unfortunately, the deep nodes in the phylogeny of aphids are very difficult to solve we have added a sentence commenting on the availability of phylogenies for all these groups (line 490).

L735 Indicate which definition of adaptive radiation you follow – in some cases the radiation does not have to be rapid, just morphologically. Rapid refers to the necessity to also test for a shift in diversification rate at that point in time, which was not mentioned in the main text.

We refer to Schluter 2000, it is now stated in the Glossary. We do mention measuring shifts in diversification rates.

: Schluter, D. 2000. The Ecology of Adaptive Radiation. Oxford University Press, New York.

L747 What is a 'wide niche'? And what a 'narrow niche'?

Now defined in the glossary

Reviewed by Brian O'Meara, 2019-05-07 22:47

This paper is an important contribution: it goes beyond the hand waving arguments for diversification to get at which mechanisms may credibly lead to observed patterns.

Thanks

Major suggestions

A persistent problem throughout the paper is that diversification and speciation are treated as synonymous, but they are not. For example, one idea for escape and radiate is not that the plant speciation rate would go up (for what it's worth, I agree with the authors that this probably wouldn't happen) but extinction rate of plants could go down (less herbivore pressure). Both would increase diversification; only one relates to speciation. I would go through and any time there is diversification, change it to speciation if that is meant; if that is not meant, think about how varying extinction could lead to a diversification pattern.

Corrected, we usually meant speciation –as in most plant/insect interaction studies. We have corrected throughout the text when necessary. We also refer to the difficulty of making

predictions for diversification rates due to the lack of specific prediction on extinction rates
line 464

I would love another table: scenario on the rows, papers on the columns, and a symbol indicating which hypotheses were examined and which ones were supported. Much of the text is a verbal description of this, but doing a table like this would really help clarify the results: in situations where both hypothesis A and C were evaluated, C won 75% of the time, but comparing B and C, C won 17% of the time. One could lump the columns by focal taxon (butterflies, beetles, etc.). It would also help highlight which hypotheses have not been examined much, and which ones have been only looked at on their own without rigorous comparison to others.

We tried to do that table. The problem is that most papers investigate one scenario only and look at one specific prediction (which is actually derived and tested in only a couple of studies). In order to provide a more concise review, we built a Table (now table 2) where we have chosen to present predictions as rows and then list the studies that have investigated each prediction (in different ways with different study systems). As you'll see researchers have sometimes tested some of these predictions without referring to specific scenario and apart from the papers of Hardy et al 2014, no study have tested one scenario against another. Also the conclusions reached by each study were not always black or white, so it's hard to actually assess how many times scenario A won over scenario B or C... which is why we advocate for more standard approaches in this review.

Lines 289-313: an important caveat is that methods looking at host shifts and speciation shifts will miss some: a shift, then a shift back, for example. I suspect that missed speciation events are far more common than missed host shifts -- if anything, multiple missed speciation events and still few host shifts is even more support for the idea that most speciation doesn't involve a host shift, supporting the conclusion. I'd still mention this caveat. It also comes into the discussion on 321-325 -- one problem with these approaches is they assume all speciation events and host switches are on the tree, which is not true (it makes me wary of such methods).

Yes true, we added a comment (line 312) underlying this caveat.

One caveat about diversification models that could be more strongly emphasized is that under any of these models where phytophagy affects diversification rate, we don't expect that is the ONLY factor (this could be relevant to bring up around line 451). Gaining or losing flight likely has a substantial effect on diversification rate, for example, and that this happens sometimes does not mean phytophagy does or does not also matter.

We agree, and in the previous version we had already mentioned this issue (l. 476-480 in the previous version). We have now added a couple of sentences to further clarify this issue (l. 405).

The interaction network approach is compelling -- I am glad it is in this paper.

thanks

Table 1 is a good contribution.

thanks

The paper would be stronger if there were a section explaining more clearly how to compare the models in Table 1 in one study (this related to line 707: the paper argues for a standardization of these predictions, but show better how this can be used in practice). Say I have a fairly well sampled insect phylogeny where I have host families for all of them. What parameters, model selection, would help me select which macroevolutionary scenario best explains my data? Some of the predictions in table 1 are vague ("high lability", "few transitions", "conservatism") -- how do these work operationally?

We agree that the field is affected by “vagueness”, because scenarios started by ad hoc narratives for phylogenetic reconstructions

The way the prediction work operationally are now laid out in table 2 –where more details are given on the methodologies used.

We also added a figure presenting some tests that can be applied to tests some relatively straightforward predictions of the different scenarios, namely

- partitioning of plant resources at speciation events (under any scenarios of host-plant induced speciation)
- lability of host breadth (under oscillation)
- adaptive radiation patterns following major host shifts (any host-plant driven speciation scenarios)
- increase in speciation rates following shifts in host breadth.

We do not give specific recommendation of models and parameters for all these analyses, these will be greatly dependant of the sampling, the taxonomic scale etc... We have tried throughout the text to underline the limitations of each method and to encourage caution in their use.

Minor suggestions

Line 162: Figure misspelled

corrected

Line 553: "hwas" typo.

corrected

Line 557: Shouldn't be parentheses around author names

corrected

I would say BiSSE and related methods are referred to more as SSE or *SSE rather than -SSE methods.

corrected

Line 613: This is a good point, but it's even worse than that: there could be many changes and it's still not robust to say a state led to a diversification rate increase. See Beaulieu & Donoghue (2013) <https://doi.org/10.1111/evo.12180> -- a single change in one clade that correlates with diversification increase could make it seem like that change always increases rate, but it's not true.

We totally agree. We have included a reference to this work (line 540)

615: I'm not sure sister group comparisons are "very" sensitive to sampling biases in the way SSE models are. Sensitive, yes -- but when one is comparing a clade of 1000 vs one with 50, you'd have to do some pretty extreme sampling to get the wrong sign.

We removed 'very' to tone down this statement

Closing on transcriptomes was odd -- it's not bad, but I could see counter reasoning (selection in the present doesn't necessarily correlate with selection when switching hosts; this does not get at the speciation genes). It does not mean cut this, only that I might move this somewhere less prominent -- it's an approach, not the ultimate approach.

We have inverted section II.2 and II.3, so we do not end on the transcriptomic approach and we now also emphasize its limitations. Line 559. We agree that it is not "THE" approach.

Line 717: should be sister lineages.

Done thanks

Left align the contents of Table 1: justified looks terrible

Done thanks, yes it did look awful...sorry.

References

de Vienne, DM, G Refregier, M Lopez-Villavicencio, A Tellier, ME Hood, T Giraud. 2013. Cospeciation vs host-shift speciation: methods for testing, evidence from natural associations and relation to coevolution. *New Phytologist* 198:347-385.

Schluter, D. 2000. *The Ecology of Adaptive Radiation*. Oxford University Press, New York.

Winkler, IS, C Mitter. 2008. The phylogenetic dimension of insect-plant interactions: a review of recent evidence. In: K Tilmon, editor. *Specialization, Speciation and Radiation: the Evolutionary Biology of Herbivorous insects*: University of California Press p. 240-263