This is a fascinating paper about the new discovery of mixed-sex populations in a species of *Timema* stick insect that was formerly thought to reproduce only by parthenogenesis. The authors perform a series of field surveys, laboratory experiments, and genetic analyses to answer questions about the incidence of and capacity for parthenogenesis versus sex in these newly discovered populations. The study offers very compelling evidence that the species is not an obligate parthenogen, but is in fact facultatively parthenogenetic, with variation in reproductive mode observed across different populations, resulting in varying sex ratios. This lends support to the idea that facultative parthenogenesis is an important stepping-stone in evolutionary transitions from obligate sex to obligate parthenogenesis—an important question in evolutionary biology.

The paper is very nicely framed and beautifully communicated (both in the writing and the figures). My suggestions for changes are all rather minor, requiring only additional clarification, rewording, or elaboration. This is a very exciting paper! I can't wait to see it in its published form!

L48-49: "When mated, sexual females fertilize all their eggs, and population sex ratios are close to 50:50". Please provide a citation for this statement of fact.

L59-62: "Such variable population sex ratios could, for example, result from a mix between sexual and parthenogenetic females, or from facultative parthenogenesis ". Please reword this for clarity. Do you mean that female-only populations might be obligately parthenogenetic with no capacity for sex and mixed-sex populations might be obligately sexual with no capacity for parthenogenesis, or, alternatively, all populations might be facultatively parthenogenetic?

61-62: "Alternatively, if the newly discovered populations were sexual, variable sex ratios could also stem from genetic drive". Please reword to something like: "Alternatively, all the newly discovered populations could be obligately sexual with sex-ratio distortion determined by mechanisms other than parthenogenesis". Distortions in sex ratio can be mediated by any number of factors; genetic drive is only one of them. Other mechanisms include male-killing endosymbionts, sex-specific zygote mortality, sex-specific sperm mortality, temperature-dependent sex determination, etc. (see Krackow BiolRev. 1995). Some of these may be more likely in *Timema* than others. But given that you only measured the sex ratio and didn't attempt to assess the presence or absence of any of these competing mechanisms, I don't think it's possible to say anything about the mechanism involved.

62-64: Please cut this final sentence. You didn't assess whether X chromosomes exhibited genetic drive, at least I can't see it in your methods. It seems as though you only assessed what the sex ratio was.

90-91: "We aimed to characterise the reproductive mode of approximately 10 females from female-only populations and of approximately 20 females from populations with both sexes". Do you mean 10 females from *each* mixed-sex population, and 20 females from *each* female-only population? Or 10 and 20 females in total?

L97: Females were isolated in petri dishes?? How big were these petri dishes?

L98: Please add some brief words to explain why soil and cottonwool were included in the petri dishes.

L108-114: I think these sentences can be reworded to avoid repetition, something like: "To test whether the individuals collected in the field belonged to different genetic lineages, we genotyped 32 females and their mates (1 - 3 per female, 42 males in total). We also tested whether eggs produced before and after mating were fertilized or not. To do this, we genotyped 3 hatchlings from eggs laid before mating (when available) for each of the 32 genotyped females (77 hatchlings from 27 females in total; for the 5 remaining females, none of the eggs laid prior to mating hatched), and 7 hatchlings from eggs laid after mating (when available) for 24 of the 32 females."

L225-226: Measuring distortions in sex-ratio can't tell you what the mechanism for it is. As mentioned above, there are numerous possible mechanisms. What criteria did you use to assess that any possible distortion you observed was due to X chromosome drive and not some other mechanism?

L266: Because you calculated sex ratios in both parental and offspring generations, it would be good to orient readers as to which sex ratio you're talking about here.

L277-281: "The second lineage (yellow in Figure 1B) was found mostly in the eastern section of Manchester and is hereafter referred to as the "eastern Manchester lineage". Finally, the third lineage (red in Figure 1B) was mainly found in the western section of Manchester and is hereafter referred to as the "western Manchester lineage"". Perhaps it would make sense to justify the names by saying that there was a gradient of genotype incidences from east to west, with the red genotype common at the far west of the Manchester transect, and the yellow genotype common at the far east of the transect.

L334: Figure 3: Is it possible in this figure to also indicate the sex-ratio of the population from which each female originated? Some of the blue and yellow mothers have low relative heterozygosity indicating that they were produced parthenogenetically, but it would be interesting to know if they came from an all-female population or a mixed-sex population. It would be interesting to know whether the sexually produced mothers with higher relative heterozygosity always came from mixed-sex populations.

L361 (and throughout the results): Please provide test-statistics, model coefficients, and SEs for all models. It isn't sufficient to just cite p-values.

L393-394: "Finally, we tested whether X chromosome drive could be contributing to female-biased sex ratios by looking at the sex ratio of sexually produced offspring". I don't think merely testing for a skew in the sex-ratio is evidence for or against X chromosome drive. Sex-ratios of sexually produced offspring can be skewed by mothers (or fathers) in any number of ways. It might be worthwhile just reporting the sex ratio analysis here without mentioning X chromosome drive at all.

L405-406: "…and not by X chromosome drive". I would say "and not through sex-ratio distortion by other means".

L409: Add a comma after "strictly".

L416-417: “However, lower hatching success of post-mating eggs could also be due to females being older”. But wouldn't you then expect to see a decline in emergence in eggs produced by ALL females after mating? Or do you mean that females from the Orr lineage might suffer reproductive ageing to a greater degree than females from the other lineages? This seems unlikely. I'd cut this sentence.

L419: “Females of the two facultatively parthenogenetic lineages”. Are only two of the lineages facultatively parthenogenetic?? I thought you found that all populations showed a capacity to reproduce parthenogenetically, and mating resulted in at least some sexually produced offspring in all lineages. That would seem to suggest that all three lineages are, by definition, facultatively parthenogenetic but vary in their ability to reproduce sexually versus parthenogenetically. I suggest rewording this for clarity and accuracy.

L420: “…the largely obligate one…”. Characterising the third population this way doesn't seem correct either, based on your results. Yes, the Orr lineage mostly *reproduced* parthenogenetically, but females of this lineage also produced some offspring sexually when they mated (which, by definition, means the lineage cannot be *obligately* parthenogenetic—it’s a facultative lineage. It’s just that females show variation in their facultative capacity). The wording should be revised accordingly.

L425: “Five females reproduced via obligate parthenogenesis…”. Just because these females *reproduced* only parthenogenetically doesn't mean they are necessarily obligately parthenogenetic. You don't know whether these females would have produced offspring sexually if they had mated more times with more males. You don't even know whether the matings that these females received resulted in sperm transfer. They may have received poor quality sperm, or incompatible sperm. I think it makes more sense to just say that these females only reproduced parthenogenetically, even after mating. Given that sex and parthenogenesis occur in all three lineages, it makes more sense to characterise all three of them as being facultatively parthenogenetic, but that there is population and individual variation in the incidence of sex and parthenogenesis.

L425: What was the phenotypic similarity? Just that they reproduced only parthenogenetically? Or was there some other aspect of the phenotypes that was similar?

L426: “…”obligately” parthenogenetic females of the Orr lineage”. Again, I would avoid this kind of characterisation.

L430-431: “…(facultative and largely obligate parthenogenesis) …”. Again, I don't think it makes sense to make this distinction. Your results suggest that all populations are facultatively parthenogenetic, but the capacity of individual females to reproduce sexually versus parthenogenetically differs.

L432: Delete “as”.

L439-440: “However, variable selection pressures along the transect could also play a role, favouring respectively sexual or parthenogenetic reproduction”. One such pressure that’s expected to generate these kinds of spatial mosaics of sex-ratio in facultatively parthenogenetic species is sexual conflict (see Burke & Bonduriansky, PhilTrans, 2018). It might be a good idea to mention that here.

L485-486: “Repeated transitions towards parthenogenesis are not surprising if the ancestor of these species was already capable of reproducing via facultative or spontaneous parthenogenesis”. I would add: "..., and if selection has the opportunity to increase parthenogenetic capacity".

L491-492: “Widespread tychoparthenogenesis capacity could thus serve as a stepping stone for the repeated evolution of more successful obligate parthenogenesis”. More successful than what? Tycoparthenogenesis is just a rare capacity for parthenogenesis in organisms that otherwise and typically reproduce sexually. Sex with a capacity for tycoparthenogenesis may in fact be more successful, especially depending on the environmental or ecological conditions. I'd cut "more successful".

L492-493: “…which could help explain why obligate parthenogenesis evolves so frequently in the *Timema* genus”. In an ultimate sense, I don't think widespread tychoparthenogenesis explains why obligate parthenogenesis evolves so frequently in Timema. Tychoparthenogenesis is only a pre-adaptation. What's required for obligate parthenogenesis to evolve from tychoparthenogenesis is some selective pressure (such as persistent mate scarcity). Greater clarity here would be welcome.

L514-515: “However, the loss of sex in facultative parthenogens could in theory be driven by sexual conflict (if mating always reduces female fitness; Burke & Bonduriansky, 2017), but this has never been demonstrated in nature”. But your results provide some support for this, no? Females from the most parthenogenetic lineage (the Orr lineage) showed a significant reduction in hatching success after they mated, and very few of their eggs were actually fertilised, suggesting that mating and sex might be more costly for these females. This contrasts with the other two lineages which both increased in hatching success following mating. Obviously, comprehensive fitness estimates would be required to properly assess this sexual conflict hypothesis. But still, there seems to be some quite suggestive evidence for it in your results. It might be worthwhile highlighting that here.

L518-581: “In this case, whether obligate strategies are likely to replace facultative ones will depend on local ecological conditions favouring sex or parthenogenesis, and on the fluctuations of such conditions”. I think it would be good to provide an assessment of the likelihood of this explanation, given your results. I'm not sure this explanation makes a lot of sense given that you found sharp differences in the incidence of and capacity for sex vs. parthenogenesis over just a few hundred meters. Are ecological conditions likely to fluctuate so greatly across such a short distance that the selective advantage of one reproductive mode over the other would switch so completely? Seems unlikely. I suggest adding something at the end of this paragraph like: “This explanation is unlikely in our case, since populations showed large differences in sex ratio over very short distances but did not differ greatly in ecology”.

L528: Delete “other” and “would”.