Stoffel et al map segregation distortion loci in a pedigreed sheep population.  Three loci were found from a significant deficiency of homozygotes for the putative (semi)lethal alleles among progeny of ‘carriers.’  The deleterious alleles are estimated to reduce viability by 25-50% when homozygous.

The authors argue that the deleterious allele is declining in frequency through time at two of these loci but not for the third.  The last locus may exhibit overdominance with positive fitness effects in the heterozygote.  It is very interesting that alleles with strongly negative homozygous effects segregate at fairly high frequency in a population.  I enjoyed reading this paper but think it would be improved by clarification of the second analysis and a bit more follow-up on the results.  
  
There are two main parts to the analysis: (1) mapping the loci with semi-lethal alleles, and (2) estimating allele frequency change at these loci through time in relation to evolutionary models.  The method for (1) is clear and compelling – here reading the Methods and then rereading the main text is essential to understanding what is going on.  For (2), the paper would benefit from a better explanation of how the ‘allele dropping method’ generates the correct distributions for hypothesis testing.  If I understand the algorithm, allele dropping simulates neutral allelic segregation while holding the pedigree as fixed.  Certainly, most forms of natural selection will alter the pedigree (some families would expand or contract contingent on the genotypes of the parents).  However, maybe this is not an issue because allele dropping is used to generate a distribution of outcomes <b>under neutrality</b> (e.g. Fig 2).  Of course, if you ran neutral evolution independently in two populations, you would get different pedigrees.  Does this not matter?  Perhaps these statistical issues have been resolved previously but then a bit more explanation would help those unfamiliar with the logic.

R1: Thank you for this comment.

The reviewer is correct that the allele dropping method generates a neutral allelic segregation under the same pedigree structure with the same founder allele frequencies. This approach allows a direct comparison of what we observe with what we expect in this scenario under Mendelian sampling alone. As the reviewer implies, the pedigree will reflect some element of drift, but also some effects of selection (i.e. fitter individuals have fitter offspring). This is why we make a comparison against the same pedigree – as this combination of drift and selection in determining *who* gets to reproduce can rapid allele frequency changes at loci that are not necessarily under selection, precisely because they go down a “fitter” branch of the pedigree. What our analysis asks is – given that selection will shape who got to reproduce, what is the neutral scenario for all loci following those same paths down the pedigree?

We occasionally have questions on the best way to do gene-drop, and we have thought long and hard about this problem over the years (and indeed continued to do so since receiving this comment which is partly why we were slow to resubmit). After ruminating on this, we believe it makes sense to make a direct comparison against the observed pedigree as we have done in the first submission. There are a few further points we’d like to make to justify our decision:

1. Our approach is very conservative, as that allele frequency changes have to be substantial to be significantly higher or lower than what is expected under neutrality, given the pedigree structure it had to follow.
2. If we have interpreted correctly, the reviewer may be concerned that the locus itself may affect reproductive success, then it will be responsible for the structure of the pedigree. This is only likely to be an issue when reproductive success is substantially heritable and has a large effect on reproductive success. However, we know that the heritability of fitness is low and is likely to be highly polygenic, and so the focal locus will have a negligible effect on the pedigree structure itself.
3. The reviewer rightfully points out that if you run neutral evolution in two populations, you get two different pedigrees. With this in mind, we have tested some approaches that “break” the selection aspect of a pedigree, such as resampling offspring between parents of the same cohort (for example, all offspring of parent A will be reassigned to parent B). However, this means that we can no longer make a *direct* comparison between our observed and expected slopes as the pedigree effect of selection is no longer present. This effect is precisely what we are trying to overcome.

To clarify this slightly, we have modified the start methods text to read as follows: “We used gene-drop simulations to directly compare whether haplotype frequency changes across time are potentially the result of selection, or are in line with the neutral expectation given the structure of the Soay sheep pedigree (i.e. thought genetic drift). Simulations were run in genedroppeR v0.1.0 (code available at <https://github.com/susjoh/genedroppeR>).”