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Response to G.W. Sileshi's Letter to the Editor on AGEE13857 (2015): Exclusion of soil macrofauna did not affect soil quality – statistical artefact or true lack of effect?

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Response to G.W. Sileshi's Letter to the Editor on AGEE13857 (2015): *Exclusion of soil macrofauna did not affect soil quality – statistical artefact or true lack of effect?*

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Dear Editor,

We appreciate receiving the comments by Dr. G.W. Sileshi on our paper. We welcome the opportunity to respond to the points raised regarding our statistical methods and data interpretation, and the presumed recommendations regarding macrofauna management. In our response, we illustrate what appears to be a misinterpretation of our article, showing that we did follow accepted scientific practice for statistical analysis and inference. In the second part of our response we repeat and further elucidate our call for further research on the dual role of termites in SSA cropping systems and sustainable termite management strategies based on ecological knowledge.

Firstly, we disagree with Sileshi's claim that the data analysis does not match the experimental design. As rightfully observed, the analyzed trial is set up as split-plot design where macrofauna exclusion is nested within the different tillage and residue management combinations. Linear Mixed Models are well suited to deal with such cases of non-independence of data - in our case spatial autocorrelation through the split-plot design, and temporal autocorrelation through repeated measurements over 7 years. As stated in the Materials & Methods section 2.5 of our paper, "block and year were defined as random factors, and the autocorrelations of plot (tillage and residue treatments) and subplot (insecticide treatment) were accounted for" (p.78). This is reflected in the statistical model (Equation 1). The model shows that Dr. Sileshi's claim that "they entered block and year as the only random effects in the mixed linear model" is simply incorrect.

```
lmer(outcome_variable ~ til * res * ins + (1|block) + (1|plot:block)
      + (1|subplot:plot:block) + (1|year), termite, na.action = na.omit)
```

Equation 1: Statistical model in R language used in our article, including the spatial autocorrelation of tillage and residue (plot within block) and macrofauna exclusion (subplot within plot within block) as random effects

However, as there is never only one single correct solution in statistics, we compared variations in the structure of the random effects as suggested by Dr. Sileshi for the outcome variables soybean yield and soil carbon at 0-15cm soil depth (Equation 2). Both models rendered no or minimal differences in statistical results with no change in interpretation, underlining the robustness of our analysis (Table 1).

```
lmer(outcome_variable~ til * res * ins + (1|block) + (1|block:til) + (1|block:res)
      + (1|block:til:ins) + (1|block:res:ins) + (1|year), termite, na.action
      = na.omit)
```

Equation 2: Statistical model in R language as suggested by Dr. Sileshi, varying the formulation of random effects

Soybean yields

Factor	P value AGEE article	P value Sileshi model
Til	0.149	0.146
Res	0.531	0.664
Exc	<0.001	<0.001
Til*Res	0.051	0.049
Til*Exc	0.564	0.561
Res*Exc	0.38	0.376
Til*Res*Exc	0.079	0.076

Soil carbon 0-15cm depth

Factor	P value AGEE article	P value Sileshi model
Til	0.872	0.872
Res	0.146	0.146
Exc	0.618	0.618
Til*Res	0.312	0.312
Til*Exc	0.310	0.310
Res*Exc	0.550	0.550
Til*Res*Exc	0.759	0.759

Table 1: Comparison of P values resulting from statistical models of the original AGEE article (Equation 1) and a changed model based on Dr. Sileshi's suggestions (Equation 2). Treatments refer to combinations of reduced tillage (-) and conventional tillage (+); residue removal (-) and residue retention (+); without macrofauna exclusion (-) and with macrofauna exclusion (+). Levels of significance indicate single and interactive effects of tillage, residue and macrofauna exclusion.

Secondly, Dr. Sileshi raises concern about the high spatial variability of soil macrofauna presence and activity. While we do agree with the commonly observed patchy distribution of termites and ants across and within habitats, we disagree committing a “failure to recognize” the same. In the Discussion section 4.2 of our paper we acknowledge that macrofauna data was characterized by high variability in space and time, emphasizing the challenges of accurately estimating soil macrofauna abundances, especially in small agricultural plots (p.81). Further research into method development is definitely needed. Although Dr. Sileshi notes that “the high variance in density estimates make it difficult to demonstrate statistically significant differences”, macrofauna exclusion *did* result in a significant impact on macrofauna abundance, but *not* tillage and residue. Nevertheless, given the large standard errors we deliberately did not emphasize this lack of effect of tillage and residue in the paper, e.g. we did not mention it as a key conclusion in the title or abstract. Please note that we reject the notion of “inflated standard error” as this suggests data manipulation. Moreover, Dr. Sileshi cites absence of baseline data as reason to doubt the success of insecticide application in macrofauna exclusion. While it is correct that we do not present data on macrofauna prior to exclusion (pre-2005), we do not consider it necessary to substantiate our conclusions. We do *not* claim to test the effectiveness of macrofauna exclusion in comparison to the natural undisturbed situation, but strictly between macrofauna exclusion treatments (+ vs. -) in otherwise

similarly treated agricultural plots. For example, the 86% reduction of termites at 0-15cm refers to the exclusion treatment compared to the non-exclusion control, *not* to a situation before trial establishment. The effect of macrofauna was also visible in the field where macrofauna exclusion resulted in higher residue retention (Figure 1). Sileshi's last point in this paragraph refers to the choice of square-root transformation of macrofauna data. Square-root or log transformation of macrofauna count data for data normalization is a commonly employed method in this field, and was for example also used in Ayuke *et al.* (2011) and Kihara *et al.* (2014). However, we agree that there are more elegant and sophisticated solutions available for analysis of count data (Bolker *et al.*, 2009).



Figure 1: Experimental plots (-T+R) without insecticide application (left) and with insecticide application (right)

Thirdly, Dr. Sileshi criticizes “incorrect inference” in our paper, pointing to a long discussed argument in statistics – the (arbitrary) drawing of thresholds or levels of significance. It is well known and inherent to formal statistical hypothesis testing that $P > 0.05$ does not necessarily equal a lack of biological effect. However, we see this discussion beyond the scope of our paper as we are simply adhering to commonly accepted scientific practices in the field of experimental studies. On appropriate occasions, we do mention ‘marginally significant’ results (< 0.07), when these also correspond to a large enough difference between treatments means, e.g. with regard to soil aggregation and soybean yields (p.79). Further, Dr. Sileshi questions our conclusion that macrofauna exclusion resulted in a 34% increase of maize yields over 7 years, while differences might not be significant in single years. Statistical analysis needs to correspond amongst other to the research questions. Since we were mainly interested in the overall impact of treatment factors, we included year as random factor in the statistical model. While we insist that the overall yield effect analysis results is an ecologically, economically and statistically sound number, it is indeed *not* suited nor intended to make practical recommendations to farmers. Testing seasons separately, as Dr. Sileshi suggests, is especially relevant for questions regarding risks that farmers face, and an interesting research question for further study. Moreover, we want to reiterate a limitation from paragraph 4.4 of the Discussion section of our article: Although our study establishes a link between exclusion of (termite dominated) macrofauna (community) using pesticides and crop yield increases, it cannot be attributed with certainty to termite pests. As it is clearly outlined in our article, alternative explanations include indirect soil and moisture effects through enhanced residue retention in the absence of termites (see also Fig. 1), or possible side effects through exclusion of other pest organisms as the insecticides used are not specific to termites (p.83).

To conclude, we want to emphasize again that we certainly do *not* intend to advocate any particular pest control method to smallholders based on our paper, and we regret that this may be misinterpreted by readers of our paper. When we report the success of insecticide application in this study, we mean its success to create +/- macrofauna treatments. On the contrary, we strongly agree with Sileshi *et al.* (2009) who call for more research into sustainable termite management strategies. Termites are perceived by many smallholder farmers as a severe production challenge and different local control measures are used in practice, as reported for example by Sileshi *et al.* (2008) for Malawi, Mozambique and Zambia, and Ayuke (2010) for Western Kenya. Our study suggests that a crucial entry point lies in the macrofauna community composition and dominance of opportunistic pest species, which may proliferate especially in degraded and nutrient depleted farming systems. Ecologically sound methods to restore macrofauna communities to enhance self-regulation and beneficial impacts might be more useful than short-term (and possibly counterproductive) remedies such as insecticides. We therefore stress again the statement made in the abstract of our paper: We need more research to better understand factors that influence detrimental pest and potentially beneficial ecological effects of termites, because the limited number of publications available on this topic indicate that those differ between agro-ecologies and farming systems.

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