Appendix S2

Scavenging in two mountain ecosystems: Distinctive contribution of ants in grassland and non-ant invertebrates in forest

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Caveats about ant suppression bait

We chose imidacloprid and sulfonamide as ant suppression agents due to their high specificity to insects (Bai et al. 1991), the low concentration required (Rust et al. 2004, Parr et al. 2016), and their rapid degradation in the environment (Anhalt et al. 2007). Also, the method described here has already been tested in the studies by Parr et al. (2016) in African savannas, and Griffiths et al. (2018) carried out in Bornean forests. These studies demonstrated that this method has no detrimental effect on other groups of invertebrates and was effective in suppressing 76% of ants in savannas and 90% in Borneo tropical forests. In addition, we sought to minimise or remove any possible effect on other non-target organisms and the environment, following the precautions: I) Imidacloprid and sulfonamide are neocotinoids that specifically affect the receptors of insect motor neurons (Bai et al. 1991), and therefore have low toxicity to non-target organisms like vertebrates and plants. II) The doses used are very low (100 p.p.m and 300 p.p.m); for example, the Imidacloprid dose is four times lower than the lowest recommended dose for controlling pests that attack crops such as cauliflower, broccoli, and citrus (Wang et al. 2015, Bayer 2019). III) The baits are designed to be highly attractive to ants that usually remove them quickly from the soil surface and carry them to their nests where consumption by other organisms is unlikely (Parr et al. 2016). IV) The application timetable was defined based on the increased activity of the ants, maximising the removal speed by these organisms (Hölldobler and Wilson 1990). V) The active ingredient Imidacloprid was chosen because it rapidly degrades microorganisms in the soil, reducing the persistence of this chemical in the environment (Anhalt et al. 2007). VI) We carried out periodic monitoring of ant abundance and only repeated the application of baits if necessary (Appendix S1: Figure S1). The chemicals used, therefore, present a very low risk to other non-target organisms (Parr et al. 2016, Griffiths et al. 2018). We found no significant detrimental effects on the overall abundance of non-target epigeic invertebrates among treatments (Appendix S5: Figure S1).

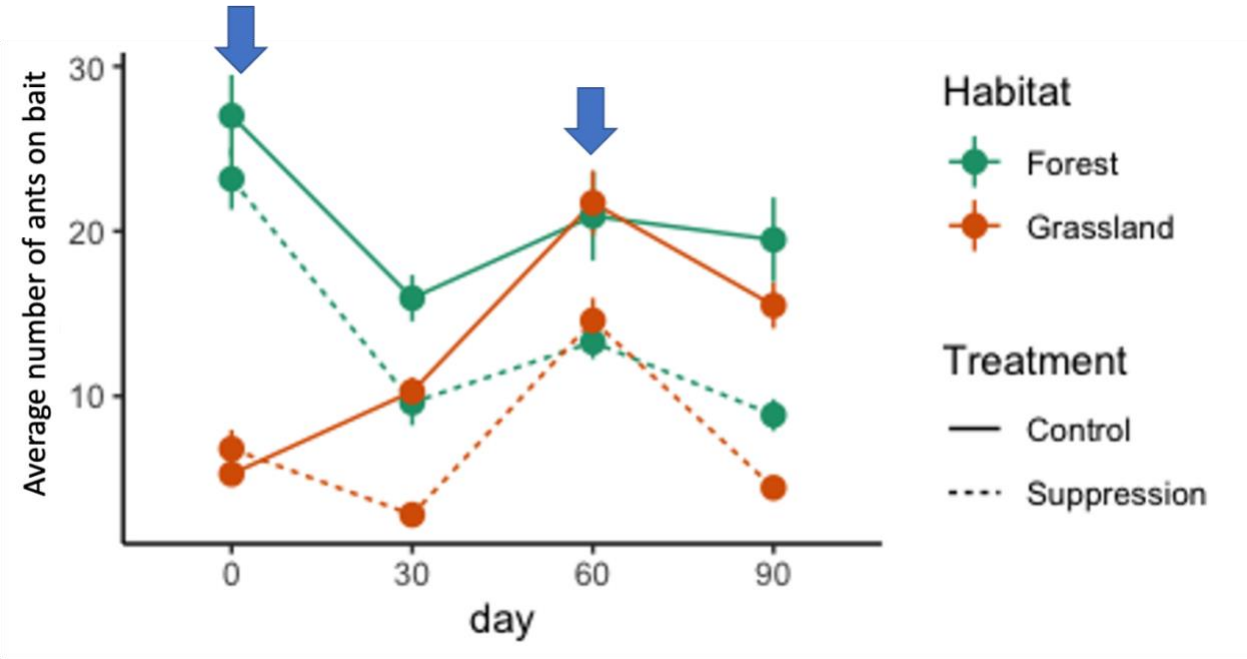

Figure S1: Monthly ant activity in baits (cat food soaked in sugary water) from December 2019 to March 2020. Points are the average ants per bait in each plot, and lines represent the standard error. Blue arrows showed when we applied the ant suppression baits.

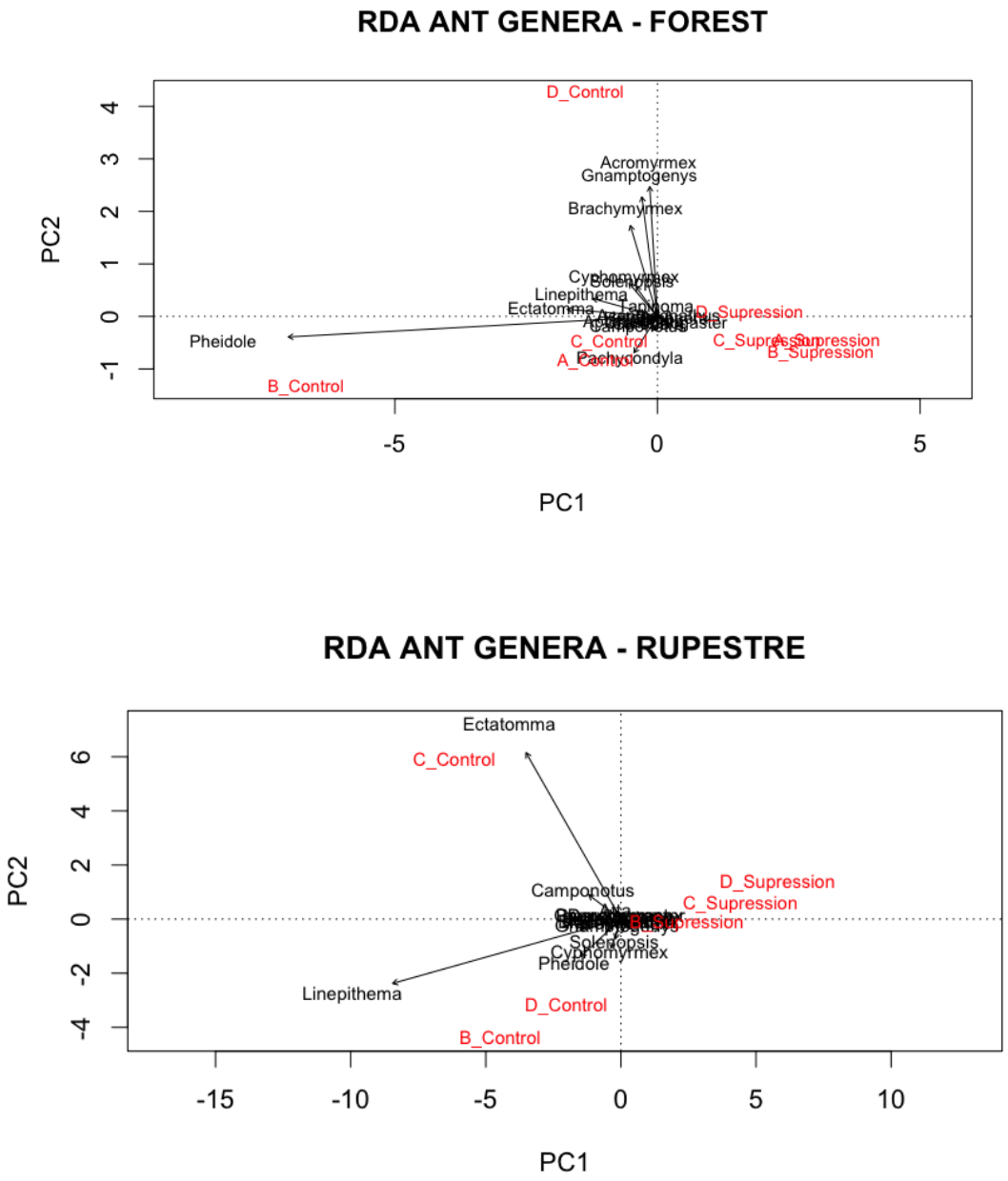

Figure S2: Redundance Analysis (RDA) of the ant genera collected from pitfall traps post-treatment (March 2020) on ant suppression plots (suppression) and control plots set in two mountainous habitats in southeast Brazil: **A)** forest (montane rainforest forest) and **B)** Grassland (*campo rupestre*). Red words represent the plot's position, control plots on the left and suppression on the right.

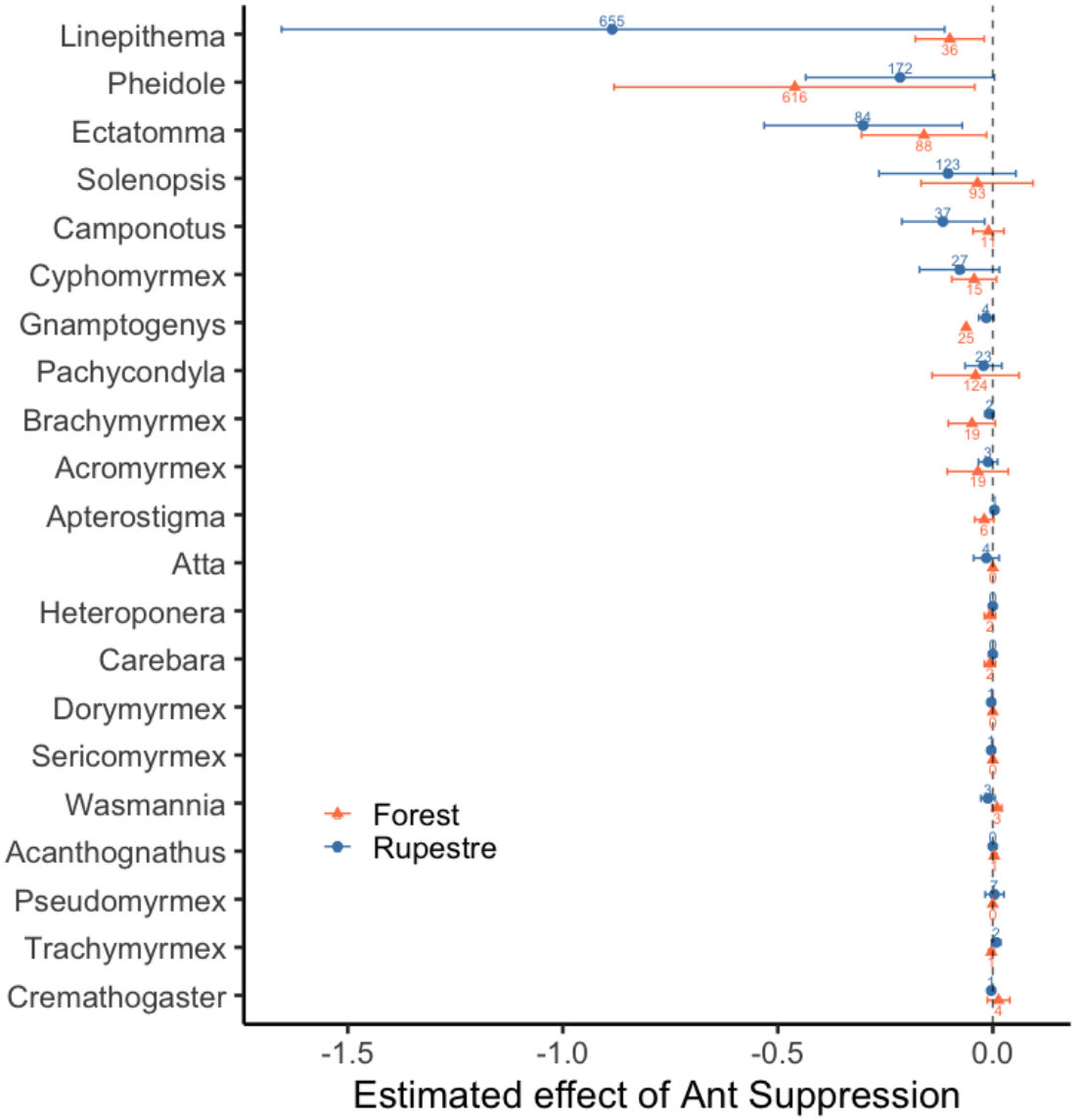

Figure S3: The estimated effect of ant suppression on forest (montane forest) and grassland (*campo rupestre*). Negative values are when the ant suppression negatively affects the genera and a positive value is when the ant suppression positively affects the group. If the interval bar overlaps the 0 the effect is not significant.

References

Anhalt, J. C., T. B. Moorman, and W. C. Koskinen. 2007. Biodegradation of imidacloprid by an isolated soil microorganism. Journal of Environmental Science and Health, Part B 42:509–514.

Bai, D., S. C. R. Lummis, W. Leicht, H. Breer, and D. B. Sattelle. 1991. Actions of imidacloprid and a related nitromethylene on cholinergic receptors of an identified insect motor neurone. Pesticide Science 33:197–204.

Bayer, S. A. 2019. EVIDENCE 700 WG. MAPA 006294.

Griffiths, H. M., L. A. Ashton, A. E. Walker, F. Hasan, T. A. Evans, P. Eggleton, and C. L. Parr. 2018. Ants are the major agents of resource removal from tropical rainforests. Journal of Animal Ecology 87:293–300.

Hölldobler, B., and E. O. Wilson. 1990. The Ants. Page The Belknap Press of Harvard University Press.

Parr, C. L., P. Eggleton, A. B. Davies, T. A. Evans, and S. Holdsworth. 2016. Suppression of savanna ants alters invertebrate composition and influences key ecosystem processes. Ecology 97:1611–1617.

Rust, M. K., D. A. Reierson, J. H. Klotz, and J. H. Klotz. 2004. Delayed Toxicity as a Critical Factor in the Efficacy of Aqueous Baits for Controlling Argentine Ants (Hymenoptera : Formicidae) Delayed Toxicity as a Critical Factor in the Efficacy of Aqueous Baits for Controlling Argentine Ants (Hymenoptera : Formic 97:1017–1024.

Wang, L., L. Zeng, and J. Chen. 2015. Sublethal Effect of Imidacloprid on *Solenopsis invicta* (Hymenoptera: Formicidae) Feeding, Digging, and Foraging Behavior. Environmental Entomology 44:1544–1552.