

Estimation of worker population size-density by nest counting in the Asian weaver ant, *Oecophylla smaragdina* (Formicidae: Formicinae) and its dynamic in oil palm plantations industry

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Supplementary Materials

Sup 1. Chronological task performance of nests collection (counting of major workers)

All the preliminary methods are explained in the main text of the manuscript. The following table shows (reviewer 2 request in page 4, line 39) the step by step tasks performed to carefully and efficiently capture *O. smaragdina* nests in order to keep their integrity and reduce bites nuisance. This procedure should be carried out after raining to avoid windy nights period (upon raining in the evening, weather conditions are more stable by late night). The wind would disturb during the use of the pyrethrin toxic by returning on the team members faces and move away weaver ants individuals fallen on the large transparent plastic (5 m x 5 m) under the tree. It is a team of 8 persons. Collecting nests of one colony per day is more suitable to avoid damaging them.

Equipment:

1. 4 wheel drive field cars (2 Toyota Hilux pickup trucks)
2. Field motorbikes (2)
3. Pyrethrin bottles (2)
4. Protective mask and eyes glasses
5. Safety industrial hand gloves for the person holding the palm tree frond and surgical gloves for others.
6. Large hard plastic bags with wide opening to seal the nests inside and conserve integrity (fertilizers overused big bags were solid enough and satisfactory).
7. Cotton
8. Chloroform
9. Large transparent square figure plastics (5 m x 5 m) to collect falling individuals in case of emergency safety pyrethrin knockdown.
10. Industrial sharp scissors, cutters
11. Plant hanging tags waterproof plastic labels reusable.
12. Wide bag neck sealers
13. Powerful torch lights (dual function of focus zoom and wide coverage open angle)
14. Night vision device help (counting resting major workers on palm tree fronds) but not compulsory item.
15. Large freezers

Wearing protective masks, eyes glasses is recommend because of repeated usage of toxic gaz for safety and preventives reasons.

Dispatching task to team members:

- (i) 1 person using a telescopic palm tree saw bring down the tagged fronds.

- (ii) 1 person is in charge to hold the tip end of each tagged palm tree frond and bring it down at the lowest as possible to bend the palm tree frond at the maximum. The same person spray pyrethrin above the hold area (along leaflets and rachis at both adaxial-abaxial side) to create a barrier stopping the major workers from attacking to bite.
- (iii) Another person is in standby mode to spray pyrethrin in case of need.
- (iv) 1 person count and record all the major workers resting along the rachis of the frond (adaxial-abaxial side).
- (v) Drivers (2) always stand by in case of need whenever the nest is on high position 9-12 m)
- (vi) 1 person in charge to perform all measurements of nest size as per method. It is better to perform this procedure just before cutting the nests because the different size parameters change back to laboratory (once inside sealed bags and transportation).
- (vii) 1 person to cut carefully the nests at the base of all the side leaflets comprising the nest architecture. Cutting the frond would trigger an emergency alarm
- (viii) 1 person in charge to open the large fertilizer bags (wet cottons with chloroform is put inside just before nests capture).
- (ix) The person who cut the nest help to store them in the fertilizer bags and pyrethrin is sprayed inside the bags to trap *O. smaragdina* individuals between the chloroform and the toxic barrier. It is impossible for the ants to escape.
- (x) For each nest collection procedures, a large transparent plastic (5 m x 5 m) is laid on the ground (just under the frond which is kept at lowest knees level) to receive eventual falling or escaping ants, neutralized by pyrethrin knockdown.
- (xi) 2 persons with motorbikes are responsible to bring back the sealed bags to laboratory and dispose them carefully above the long lab tables.
- (xii) By morning, all bags were opened to evacuate the strong formic acid, toxic gas smell in the open field air.
- (xiii) After 2 hours, all bags are put back in the laboratory (doors, windows open due to reluctant aggressive smell). Each nest is carefully dissected to sort out all individuals for direct counting with accurate counter RS Pro mechanical surface mount RS 4 digit display type.
- (xiv) All different *O. smaragdina* individuals caste are separated and recorded for each nest.
- (xv) All individuals ants are spread by their respective caste on different white long tables used in the laboratory to proceed to direct counting and make temporary records tables on A4 white papers.
- (xvi) All measurements recorded are transferred into Microsoft Excel version 2013 on lap top files to make a dataset to conduct further statistical analysis.