Ecology of an Improbable Association: The Pseudomyrmecine Plant-ant *Tetraponera tessmanni* and the Myrmecophytic Liana *Vitex thyrsiflora* (Lamiaceae) in Cameroon[1]

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ABSTRACT

In young individuals of the obligate myrmecophytic liana *Vitex thyrsiflora*, several species of ants and other arthropods compete for resources offered by the plant. In mature individuals, the only inhabitant is the ant species *Tetraponera tessmanni*, which is completely restricted to *Vitex* lianas as its sole host. Established colonies of this ant provide effective defense against herbivores. The association between *V. thyrsiflora* and *T. tessmanni* is unusual in two respects. First, the climbing life form is rare among myrmecophytes. Secondly, it is surprising that a pseudomyrmecine should be the obligate associate of a liana. Pseudomyrmecine plant-ants often prune vegetation contacting their host plant. This behavior functions in part to protect against invasion of the host by ecologically dominant ants. In contrast, *T. tessmanni* does not prune and is associated with a plant whose success, and thus that of its resident ant colony, depends on contacts with many other plants. Several traits of *V. thyrsiflora* and *T. tessmanni* combine to make the colonization of host plants by potential competitors very difficult. These include behavioral and morphological filters restricting entrance into the plant and exploitation of the resources it can supply; plant anatomical organization that enables *T. tessmanni* workers to carry out all activities, except leaf patrolling, within a single, branched private nesting space within which all food resources offered by the plant are produced; and polygyny, permitting the colony to monopolize a large, rapidly growing and long-lived territory.

RESUME

Dans des jeunes individus de la liane *Vitex thyrsiflora*, myrmécophyte obligatoire, plusieurs espèces de fourmis, ainsi que d'autres arthropodes, sont en compétition pour les ressources offertes par la plante. Les individus matures sont occupés uniquement par la fourmi *Tetraponera tessmanni*, strictement inféodée aux *Vitex* lianescents comme plantes-hôtes. Les colonies établies de cette fourmi protègent la plante efficacement contre les insectes phytophages. L'association entre *V. thyrsiflora* et *T. tessmanni* est remarquable pour deux raisons. Premièrement, les lianes sont rarement représentées parmi les myrmécophytes. Deuxièmement, il est surprenant que l'associée obligatoire de cette liane soit une Pseudomyrmécinae. Chez les fourmis de cette sous-famille, la plupart des espèces inféodées aux myrmécophytes éliminent la végétation qui contacte leur plante-hôte. Ce comportement participe à la protection des colonies contre l'invasion de l'hôte par des espèces écologiquement dominantes. Contrairement à ces espèces, *T. tessmanni* ne détruit pas la végétation voisine car elle est associée avec une plante dont le succès, et donc celui de la colonie résidente de fourmis, dépend de contacts nombreux avec d'autres plantes. Plusieurs traits de *V. thyrsiflora* et de *T. tessmanni* se combinent pour rendre très difficile la colonisation des plantes-hôtes par des compétiteurs potentiels. Ces traits incluent des filtres comportementaux et morphologiques restreignant l'entrée dans la plante et l'exploitation de ses ressources; l'organisation anatomique permettant aux ouvrières de *T. tessmanni* de conduire toutes leurs activités, à l'exception de la patrouille des feuilles, dans un seul espace privé, ramifié, à l'intérieur duquel toutes les ressources alimentaires offertes par la plante sont produites ; et la polygynie, qui permet à la colonie de monopoliser un territoire de grande taille dont la croissance est continue et qui persiste durant de très longues périodes.

NUMEROUS STUDIES DEMONSTRATE THE SPECIFICITY OF THE ASSOCI-ATION between specialist plant-dwelling ants and myrmecophytes, plants that offer to specialist ants lodging in the form of domatia and food, either directly (food bodies, extrafloral nectar) or indirectly (excretions of trophobiotic hemipterans). In these protection mutualisms, ants protect their host plants against herbivory, and sometimes from competition with other plants (Davidson &

McKey 1993, Gaume & McKey 1998, Moog *et al.* 1998, Renner & Ricklefs 1998). However, juvenile individuals of many obligate myrmecophytes are often inhabited by ants other than the specific associate(s) that completely depend on the plant, or by other insects. This suggests that young colonies of specialist plant-ants may compete with these other occupants. The virtually exclusive occupancy of mature plants by colonies of the specific associate(s) implies that they are strongly favored in such competition. What traits of specialist ants, or of their host plants, confer this advantage?

Competition among ant species for resources offered by myrmecophytes is of theoretical interest, because it may have shaped many features of the mutualistic interaction between ants and plants. Niche restriction through competition may have been the force, for example, that produced predictable associations between pairs of species, satisfying an important precondition for strong pairwise co-evolutionary interaction (Davidson & McKey 1993). Physical traits of the plant may be important in determining competitive superiority on a particular host. For example, the long dense trichomes typical of an entire guild of understory ant-plants exclude large, ecologically dominant ants and favor small subordinate ants that are the most effective mutualists of these plants (Davidson *et al.* 1989). Behavioral traits of ants may also prevent their competitive exclusion from particular hosts (Palmer *et al.* 2002). Some ant traits important in the functioning of mutualisms may even owe their evolutionary origin to a pre-existing function in competitive interactions. For example, the attacking of foreign vegetation contacting or surrounding the host, which enables many specialist plant-ants to act as allelopathic agents of their host plant (Janzen 1969), may be grounded in a trait whose initial function was to avoid competition. Some ant species ecologically subordinate in arboreal ant mosaics prune vegetation that touches their host, thereby eliminating points where competitive dominants could invade (Davidson *et al.* 1988). In specialist plant-ants, pruning behavior may continue to play a role in limiting competition with other ants (Davidson *et al.* 1988, Stanton *et al.* 1999).

Such traits of plants or ants that give specialists a competitive advantage on their host may be of particular importance in climbing myrmecophytes. By their very nature, lianas (woody vines) contact numerous other plants. The resulting ease with which many ants are recruited to plant resources may explain why climbing plants are well represented among plants involved in opportunistic mutualisms mediated by extrafloral nectaries (Bentley 1981). However, the liana life form also results in openness to invasion by numerous nonspecialist ants. Relatively few myrmecophytes are lianas or vines (Davidson & McKey 1993) and openness to invasion may be one reason why.

Of the few lianescent myrmecophytes, only *Spatholobus* associated with *Cladomyrma* has been investigated in any depth (Moog *et al.*, in press), and we are aware of no study that addresses competition among ants for these plants, or the traits conferring an advantage to their specialist plant-ant associates. We conducted a field study of *Vitex thyrsiflora* Gürke (Lamiaceae; formerly placed in Verbenaceae), a woody climber of up to 10 m high (but with trailing stems up to 50 m long) frequently found in secondary rain forests of southern Cameroon, Central/West Africa. Infrageneric

taxonomy of *Vitex* is not clearly established, and there is still much confusion in plant identification. The number of myrmecophytic liana species of *Vitex* is thus uncertain. Bequaert (1922) and Wheeler (1922) conducted extensive observations of lianescent *Vitex* myrmecophytes, but no detailed ecological study of this system has been done. Young individuals of *V. thyrsiflora* grow in forest clearings but mature individuals can also be found in old and almost undisturbed forests. With hollow, but not swollen stems, *V. thyrsiflora* is considered to be a specialized myrmecophyte because all mature lianas shelter the specific and obligate plant-ant species *Tetraponera tessmanni* (Stitz), formerly placed in its own genus, *Viticicola* (Bequaert 1922, Wheeler 1922, Schnell & Grout de Beaufort 1966, Ward 1991), and because provision of food resources for these ants by the plant involves an unusual mechanism in response to actions of *T. tessmanni* (Bequaert 1922). This ant patrols leaves, especially young leaves, of the host (Fig. 1a) and is highly aggressive in attacking insect herbivores of the plant (and in stinging human investigators). While early observations indicated effective host-plant protection (Bequaert 1922), no studies have demonstrated this.

Interestingly, this ant belongs to the subfamily Pseudomyrmecinae, a subfamily whose members are usually considered to be ecologically subordinate in arboreal ant mosaics (Davidson *et al.* 1988, Buschinger *et al.* 1994, Dejean *et al.* 1994, Palmer *et al.* 2000). For example, *Tetraponera penzigi*, a species closely related to *T. tessmanni* (Ward 1991) and associated with the African swollen-thorn *Acacia*, is ecologically subordinate in competition with three *Crematogaster* species for access to acacia trees and replaced by them during "succession" in individual trees (Young *et al.* 1997, Palmer *et al.* 2000). Some specialist plant-ants of this subfamily, such as *Pseudomyrmex* in Neotropical *Acacia*, *Tachigali*, *Triplaris*, and others (Ward 1999) and *Tetraponera aethiops* in *Barteria fistulosa* (Janzen 1972, Yumoto & Maruhashi 1999), are well known to prune vegetation surrounding their host plant. This behavior contributes to their ability to exclude other ants from their hosts (Davidson *et al.* 1988). We thus wondered how the pseudomyrmecine *T. tessmanni* maintains exclusive occupancy of mature individuals of a plant whose life form requires that it be in contact with numerous other plants. In this study, we ask the following questions: (1) What traits of *T. tessmanni* and/or its host enable this ant to monopolize all mature lianas? (2) What insects occupy *V. thyrsiflora* plants at different stages of their development, and how do they interact with *T. tessmanni*? and (3) Does *T. tessmanni* protect its host?

METHODS

Four surveys were carried out in a forest at 800–1100 m on the slopes of Mont Kala in southern Cameroon (3°50′N, 11°21′E) in 1996, 1997, and 2000. The vegetation of Mont Kala is described by Achoundong (1985). Young lianas were encountered in an abandoned plantain farm surrounded by remnants of old-growth forest. Complementary observations were made during the same period in undisturbed forest in the Dja Biosphere Reserve (3°25′N, 13°32′E; elevation about 400–500 m), in old-growth forest (at about
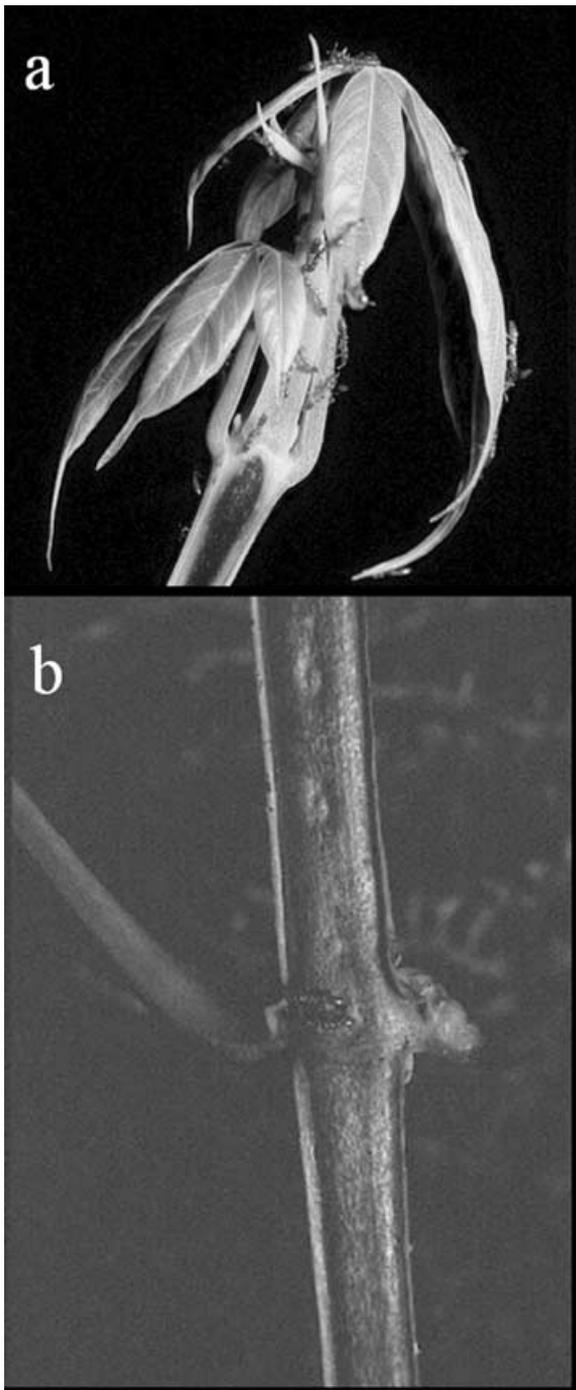
FIGURE 1. (a) Workers of *T. tessmanni* patrolling young leaves of *V. thyrsiflora*. Workers are *ca* 3.5 mm long. (b) A young stem (width *ca* 7 mm) of *V. thyrsiflora* showing the location of the entrance hole (partially obscured by two workers of *T. tessmanni*). Phyllotaxy is opposite and decussate. Entrance holes occur in pairs at each node, on the faces between the opposite leaf insertions. Above the entrance hole, and in a line with it, can be seen three roughly circular indentations, marking spots where *T. tessmanni* workers have excavated pits from the inner domatium wall to the vascular cambium, located in these young twigs just beneath a thin layer of cortex and wood. In each internode, these pits are always located on the same two sides of the stem as the entrance holes at the node just proximal.

800–1100 m) on the slopes of Mont Eloumden (3°33′N, 11°31′E) and at Ebeba (3°48′N, 11°13′E; elevation about 700 m). Sample sorting was performed at the Laboratory of Zoology of the University of Yaoundé I.

FAUNA OF PLANT INTERNODES.—Three classes of internodes of *V. thyrsiflora* were distinguished. The first included young internodes that were filled with fresh pith. The second included mature internodes in which the pith had dried up and/or had been excavated by insects, leaving only the internode wall. In open mature internodes the cavities were often used as nest sites by ants and various other insects. When the plant was inhabited by a large ant colony, the pith cavities of successive internodes were connected, with no nodal septa. Cavities remain open as long as occupants maintain entrance holes, located at nodes (see below), which are otherwise closed by callus growth. *T. tessmanni* appears to be the only occupant capable of maintaining these openings. Some open internodes were not currently occupied, but bore evidence of recent occupation by ants or beetle larvae. The third class included mature internodes with no central cavity. Such internodes, when present, were always found at the base of a young liana that had been cut one to many times. It was impossible to determine whether these internodes had ever been occupied by insects, but we believe that most had not been.

After preliminary observations on many young lianas and older individuals, the survey was organized as follows. Entire above-ground parts of a total of 115 young individuals of *V. thyrsiflora* were collected and transported individually in labeled plastic bags to the laboratory for immediate dissection. Here, the leaves were removed and counted, and damage to them was scored on a semi-quantitative scale (see below). For scoring occupancy of stem internodes, we took into consideration only those potentially occupiable, *i.e.*, the first two internode classes distinguished above, those full of pith (fresh or dry) or with pith cavities hollowed, forming domatia, which are current or potential feeding and/or nesting sites for the different insects. Internodes with pith cavities secondarily closed (and thus useless for animals) were counted, but not included in the analysis.

Internodes were opened and their contents (fresh or dry pith, living insects, beetle frass, ant wastes [characterized by the presence of various insect cuticular remains]) noted. All living inhabitants were collected and preserved in 70 percent ethanol, then identified to the morpho-species level where possible. Voucher specimens were retained at the Laboratory of Zoology of the University of Yaounde I. The plants examined were classed into one of four categories: (1) those completely lacking *T. tessmanni* ($N = 27$ plants); (2) those with only one or more foundresses of *T. tessmanni* ($N = 53$); (3) those with incipient colonies ($N = 22$), and (4) those with established colonies of *T. tessmanni* ($N = 13$).

Categories 2–4 represented lianas with three developmental stages of colonies of *T. tessmanni*. Lianas with one or more "foundation" stage colonies (category 2) had one or more completely closed domatia, each containing a founding queen with brood and/or a few workers. Foundation is claustral, *i.e.*, the foundress remains in an enclosed place, the entrance hole being reopened only by the first cohort of workers. Plants with one or more "incipient colonies" (category 3) were defined as those in which the colony occupied a small number of domatia. Plants occupied by incipient colonies

had other internodes that were either unoccupied or contained other arthropods (often including other ants). In incipient colonies, and in contrast to foundations, entrance holes of domatia were opened and workers had already begun to patrol the plant's surfaces. Each plant with an "established colony" (category 4) had large numbers of workers that actively patrolled the plant's surfaces, particularly young leaves. To test whether the ants in a large *V. thyrsiflora* liana constitute a single colony or more than one, we conducted worker-transfer experiments between different plants ($N = 10$ replications $\times$ 6 different pairs of plants $\times$ 2 tests, one in each direction, for a total of 120 tests) and between two branches of the same individual liana ($N = 10$ replications $\times$ one pair of branches on each of 6 individual plants $\times$ 2 tests, one in each direction, for a total of 120 tests).

For statistical comparison of the number of domatia per plant in relation to category of occupation by *T. tessmanni*, we performed an ANOVA for global comparisons, followed by post hoc tests (lsmeans, with Bonferroni adjustment) using multiple pairwise comparisons (Systat 7.0.1 [SYSTAT 1997]).

PLANT PROTECTION BY ANTS.—To evaluate the influence of *T. tessmanni* on the activity of phytophagous insects on *V. thyrsiflora*, we compared the extent of defoliation by herbivores on the leaves of plants according to the categories established above: those without any colony of *T. tessmanni* or with only foundation-stage colonies of this species (plants not patrolled by *T. tessmanni*) (categories 1 and 2 above pooled; 80 plants and 568 leaves examined); those with one or more incipient colonies of *T. tessmanni* (category 3 above; 22 plants and 156 leaves examined); and those with an established colony of this ant (category 4; 13 plants and 302 leaves examined).

Leaves of *V. thyrsiflora* are palmately compound with five (rarely four) leaflets. Scoring for leaf damage was carried out by subjectively assigning each leaf to four defoliation levels (Dejean & Djiéto-Lordon 1996): level 1 (less than 25% of the leaf surface removed), level 2 (25–50% of the leaf surface removed), level 3 (50–75% of the leaf surface removed), and level 4 (over 75% of the leaf surface removed). As a conservative measure, missing leaflets were not taken into account in assessing extent of herbivory, as the cause of their absence could not be determined. We conducted a MANOVA (PROC GLM, SAS [SAS 1996]) to compare the distribution of leaves in the four defoliation levels among the three groups of plants (categories 1 and 2 pooled, category 3, and category 4). This analysis was then followed by a multiple comparison of means among groups for each of the defoliation levels (lsmeans and Tukey–Kramer tests).

To examine the relationship between occupation of plants by *T. tessmanni* and the presence of larval Coleoptera and Diptera in the internodes, we compared among the four occupation categories (1 = *T. tessmanni* absent; 2 = foundation stage only; 3 = incipient colonies; 4 = established colonies) the proportion of plants in which these occupants were present in one or more internodes, or absent (PROC GENMOD, SAS [SAS 1996], with a binomial distribution). We tested for an overall effect of occupation category on probability of presence of each of these insects, followed by multiple pairwise comparisons (lsmeans).

RESULTS

TRAITS OF PLANTS IN RELATION TO INTERACTION WITH ANTS.—As already noted by Bequaert (1922), *V. thyrsiflora* does not produce food bodies or extrafloral nectar. Dissection of numerous plants confirmed the absence of hemipterans (*i.e.*, Sternorrhyncha, formerly included in Homoptera, now known to be paraphyletic [von Dohlen & Moran 1995]) that could potentially act as trophobionts for resident ant colonies. We also confirmed earlier observations that ant entrance holes are located only at the nodes, on the sides of the stem (which is square in cross-section) between the opposite leaf insertions (two entrance holes per node; see Fig. 1b). Entrance holes must be opened by the ants, although this location appears pre-adapted or adapted to facilitate such entry (Bailey 1922). In unoccupied plants, the central stem cavity is filled with pith. In plants with foundation-stage or incipient colonies, occupied domatia are interrupted by sections of stem containing intact pith. In large plants with established colonies (see Fig. 2a), the central cavity is continuous, and the entire branching structure of the plant is connected in one highly branched system of internal passageways. All the hollow stems of a plant are connected in one great branching gallery, with no nodal septa or other divisions. Ants in an established colony can thus move between any two points anywhere on the plant, in the protected gallery, without ever having to go onto the twig surface. Entrance holes are maintained open in stems of all sizes (Fig. 2b), and ants can also quickly exit hollow stems from any location in the plant.

We also confirmed earlier observations that colonies of *T. tessmanni* are highly polygynous. As already noted by Wheeler (1922), females are dimorphic. One morph possesses fully functional wings. We observed that claustrally founded colonies were without exception founded by dealate (*i.e.*, wings cut off) females of this morph. The other morph has wings reduced to functionless nubs, and a narrower alitrunk (the alitrunk is the middle part of the body of ants, comprising the thorax and adjacent segments of the abdomen). *Tetraponera tessmanni* is the only pseudomyrmecine known to show such queen dimorphism (P. Ward, pers. comm.). Established colonies include numerous apparently functional egg-laying queens of both morphs (B. Schatz, pers. comm.).

The 25 adult individuals surveyed were all occupied by *T. tessmanni*. Transfer of workers between two plant individuals always (in all 120 cases) resulted in full attack on the alien by resident workers, while transfer of workers from two branches of the same liana, at points separated in some cases by more than 50 m, always (all 120 cases) resulted in acceptance of introduced individuals. We conclude that the ants occupying a plant constitute a single colony. From the moment when foraging workers appear, the colony of *T. tessmanni* grows as the host plant grows, rapidly colonizing all new internodes.

ANT FAUNA OF *V. THYRSIFLORA*.—Ants were by far the main occupants of the internodes of *V. thyrsiflora*, being present in 44.4 percent of all internodes examined (Table 1). Eight species of ants were identified from the 115 young *V. thyrsiflora* individuals examined (Fig. 3 and Appendix 1). Among those ants, *T. tessmanni*, the obligate

FIGURE 2. (a) Basal portion of the stem of a large liana of *V. thyrsiflora*, showing the size typically reached by mature individuals. This stem (diam *ca*. 12 cm) bore an open pith cavity, occupied by *T. tessmanni*. (b) Cross-section of a stem of *V. thyrsiflora* (diam *ca* 2 cm), showing the central cavity formed in excavated pith and, in oblique section, an entrance hole of *T. tessmanni*. Many entrance holes are maintained open in old stems and continually extended as the stem increases in diameter.

TABLE 1. *Occupation of domatia of* V. thyrsiflora *by other arthropods, as a function of the status of* T. tessmanni *in the plant*.

Status of *T. tessmanni* in the plant (category)	Total number of plants[a]/domatia	Contents of internodes of *V. thyrsiflora* (% of all plants or all domatia in the category)						
		T. tessmanni	Other ants	Other arthropods	Ant wastes	Beetle[b] frass	Pith	Empty
1. Absent	27 (23.5%)	0	21 (77.8%)	27 (100%)	6 (22.2%)	19 (70.4%)	17 (63.0%)	21 (77.8%)
	289 (17.6%)	0	67 (23.2%)	67 (23.2%)	15 (5.2%)	49 (17.0%)	29 (10.0%)	62 (21.5%)
2. Foundation stage	53 (46.1%)	53 (100%)	38 (33.0%)	53 (100%)	25 (47.2%)	40 (75.5%)	37 (69.8%)	48 (92.5%)
	766 (46.6%)	153 (20.0%)	104 (13.6%)	110 (14.4%)	93 (12.1%)	89 (11.6%)	61 (8.0%)	156 (20.4%)
3. Incipient colonies	22 (19.1%)	22 (100%)	18 (81.8%)	10 (45.5%)	12 (54.6%)	8 (36.4)	8 (36.4)	17 (77.3%)
	318 (19.3%)	109 (34.3%)	50 (15.7%)	10 (3.1%)	38 (12.0%)	18 (5.7%)	20 (6.3)	73 (23.0%)
4. Established colonies	13 (11.3)	13 (100%)	1 (7.7%)	0	6 (46.2%)	0	1 (7.7%)	5 (38.5%)
	272 (16.5%)	247 (90.8%)[c]	1 (0.4%)	0	14 (5.2%)	0	1 (0.4%)	9 (3.3%)
Total	115	88 (76.5%)	57 (49.6%)	90 (78.3%)	43 (37.4%)	67 (58.3%)	75 (65.2%)	91 (79.1%)
	1645	509 (30.9%)	222 (13.5%)	187 (11.4%)	160 (9.7%)	156 (9.5%)	111 (6.8%)	300 (18.2%)

[a]Figures for "plants" indicate number (and %) of plants having at least one internode with the type of contents noted. Single domatia also sometimes had mixed contents. Percentages thus total more than 100%.

[b]*Ischnolanguria concolor* (Languriidae).

[c]Some older internodes of these plants, probably produced before they had acquired established colonies, remained unoccupied.

plant-ant, was by far the most frequent and the most abundant species (Table 1). In most of the immature lianas surveyed, *T. tessmanni* was represented either by colonies in the foundation stage, not yet large enough so that workers foraged externally (53 of the 115 surveyed plants [46.1%]), or by active colonies whose workers permanently patrolled the young stems and leaves of the host plant (35 of the surveyed plants, 22 with incipient colonies, and 13 with established colonies [Table 1]).

The probability of occupation by *T. tessmanni* varied with plant size. Unoccupied plants had significantly fewer internodes than occupied plants (Fig. 4). There was also a tendency (not significant in our sample) for increased degree of occupation with increasing

plant size. The average size of category 4 plants (harboring established colonies of *T. tessmanni*), and perhaps others, is greatly underestimated in our sample, which only considered juvenile plants. Mature lianas can reach stem basal diameters of 12 cm or more (Fig. 2a), with total stem length in the tens or hundreds of meters, bearing thousands of internodes and harboring colonies of enormous size.

INTERACTIONS BETWEEN DIFFERENT ANT SPECIES IN DOMATIA OF *V. THYRSIFLORA*.—*Tetraponera tessmanni* most frequently shared plants with the myrmicine *Tetramorium tabarum*. The frequency of *T. tabarum* was usually low in the domatia of *V. thyrsiflora*, but when
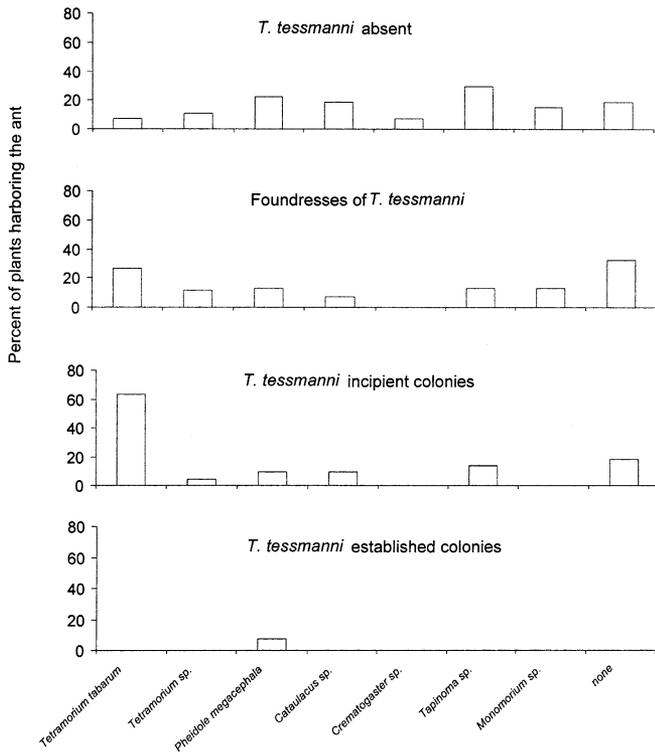
FIGURE 3.   Percentage of *V. thyrsiflora* plants in which ant species other than *T. tessmanni* were present, in relation to the status of *T. tessmanni* colonies in the plant. Some plants harbored more than one species of ant (see Appendix 1). Sample sizes: *T. tessmanni* absent, $N = 27$ plants; foundresses, $N = 53$; incipient colonies, $N = 22$; established colonies, $N = 13$.
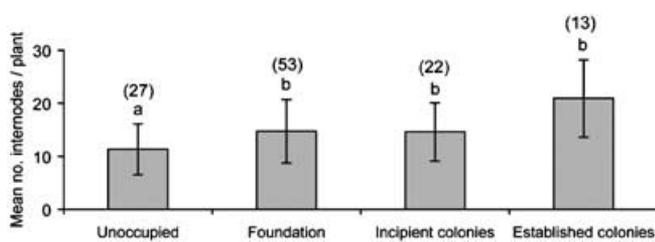

FIGURE 4.   Variation in the size of *V. thyrsiflora*, estimated by the number of internodes per plant (mean ± SD), as a function of the status of occupation by *T. tessmanni*. Statistical comparisons: Global test (ANOVA): $F_{3,111} = 6.04$, $P = 0.0008$. Different letters indicate significantly ($P < 0.05$) different means in a multiple pairwise comparison with Bonferroni adjustment. Sample sizes (number of plants) are given in parentheses above each bar.

present, this ant was frequently associated either with incipient colonies of *T. tessmanni* (63.6% of the 22 lianas with incipient colonies) (Fig. 3), or less frequently, with plants sheltering ant species other than *T. tessmanni* (or only foundations of *T. tessmanni*) (Fig. 3; Appendix 1).

Furthermore, dead individuals and brood of *T. tessmanni*, as well as those of other ant species, were regularly found in *T. tabarum*
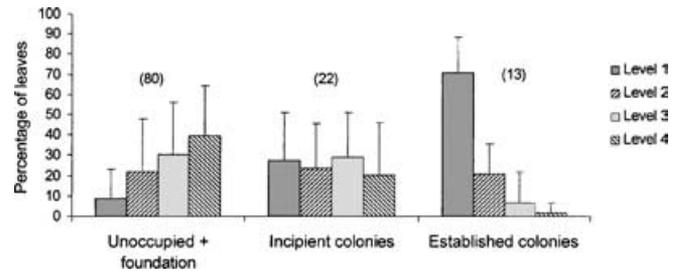

FIGURE 5.   Variation in herbivory to *V. thyrsiflora* plants as a function of occupancy by *T. tessmanni*. Percentages (mean ± SD) of all leaflets in defoliation levels 1 (<25% herbivory), 2 (25–50%), 3 (50–75%), and 4 (>75%). Sample sizes (number of plants) for each occupation category are given in parentheses above each set of histograms. See text for statistical comparisons.

nests and in the domatia in which they packed their wastes. These observations confirm the myrmecophagous habits of *T. tabarum* (Dejean & Djiéto-Lordon 1996). As *T. tabarum* nests in juvenile *V. thyrsiflora* and feeds on incipient colonies of *T. tessmanni* and other ants, it may constitute a limiting factor to the successful establishment of incipient colonies. Nevertheless, *T. tabarum* was completely absent from plants with large *T. tessmanni* colonies.

The other ant species represented, also not obligate inhabitants of *V. thyrsiflora,* were either ants of small size that occupied some internodes (*e.g.*, *Monomorium* sp. and *Pheidole* cf. *megacephala*) or incipient colonies and founding queens (*e.g.*, *Crematogaster* sp. and *Cataulacus* sp.) that were generally replaced by *T. tessmanni* as the colony of this species grew and occupied an increasing proportion of the plant's domatia. These opportunistic ants were more frequent in plants without *T. tessmanni* or with only foundations than in those with incipient colonies, and were almost completely absent in those with established colonies (Fig. 3; Appendix 1).

INFLUENCE OF ANTS ON DAMAGE BY LEAF-EATING INSECTS.—Leaves of plants with established colonies were much less damaged than those of plants with incipient colonies and plants lacking a *T. tessmanni* colony; plants with incipient colonies showed damage levels intermediate between those with established colonies and those without colonies (Fig. 5). As a result, plant category had a highly significant effect on the amount of herbivory sustained by leaves (Wilks' lambda; $F_{6,220} = 21.55$; $P < 0.0001$). Differences among plant categories were most pronounced for defoliation levels 1 (<25% removed; $F_{2,112} = 78.51$; $P < 0.0001$) and 4 (>75% removed; $F_{2,112} = 17.76$; $P < 0.0001$). Differences among categories were less pronounced for defoliation level 3 ($F_{2,112} = 5.23$; $P < 0.007$), and were not significant for defoliation level 2 ($F_{2,112} = 0.06$).

OCCUPATION BY *T. TESSMANNI* AND PRESENCE OF NON-ANT INHABITANTS OF DOMATIA.—Apart from ants, the larvae of the beetle *Ischnolanguria concolor* Kraatz (Languriidae, subfamily Languriinae) were the most frequent occupants of *V. thyrsiflora* internodes. Larvae of *I. concolor* live in the internodes of *V. thyrsiflora* and feed on the
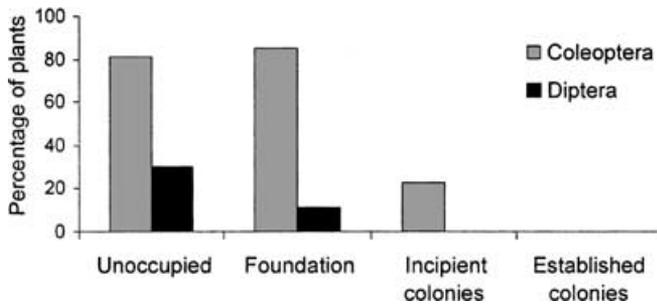
FIGURE 6. Proportions of *V. thyrsiflora* plants harboring larvae of Coleoptera (*Ischnolanguria concolor* Kraatz, Languriidae) and Diptera in young twigs (plants containing only frass of these insects are not included), in relation to the status of *T. tessmanni* colonies in the plant. Sample sizes as in Figure 4.

pith and associated soft tissues of the internal wall; the adults feed on the leaves of *V. thyrsiflora*. This beetle appears to be the only occupant other than *T. tessmanni* that opens entrance holes (at the nodes, in the same location where this ant constructs its openings). Larvae of *I. concolor* were present in 72 of 115 plants examined, but the probability of their presence varied with the stage of development of the colony of *T. tessmanni* in the host plant (Table 1; Fig. 6). They were found in over 80 percent of plants that lacked *T. tessmanni* or in which only foundresses of this ant were present, but were found in fewer than one-fourth of the plants with incipient colonies, and were absent in plants with established colonies of this species (Fig. 6; $X^2 = 57.6$; df = 3; $P < 0.0001$). There was no significant difference in the probability of beetle presence between plants lacking *T. tessmanni* and those with only foundation-stage colonies, but both of these were different (lsmeans tests, $P < 0.0001$) from plants with either incipient or established colonies, which did not significantly differ from each other. Also, some plants unoccupied by *T. tessmanni* in which beetles were absent contained their frass, indicating their presence previously. Diptera were much less frequently present, in 13 of 115 plants (Table 1; Fig. 6). There was an overall effect of category on probability of their presence ($X^2 = 12.86$; df = 3; $P < 0.005$), with completely unoccupied plants being more likely to have flies than those with established colonies of *T. tessmanni* (lsmeans, $P < 0.0001$). Only a few other arthropods used the domatia as shelter for reproduction (spiders) or pupation (Lepidoptera).

DISCUSSION

INTERACTIONS BETWEEN *T. TESSMANNI* AND OTHER OCCUPANTS OF JUVENILE *V. THYRSIFLORA*.—Our results suggest that the ant species associated specifically with this plant, *T. tessmanni*, competes with other arthropods for the occupancy of juvenile host plants. Hollow internodes of young lianas of *V. thyrsiflora* represent suitable habitat for many ant species incapable of constructing their own nests, and permit multiple colonization of individual plants by foundresses of a diversity of ant species. This situation seems common in specialized

myrmecophytes (Davidson *et al.* 1990, Fiala & Maschwitz 1992). However, most potential occupants do not open entrance holes into the cavity, and only enter as secondary colonists after *T. tessmanni*, or after the beetle *I. concolor*, the only other occupant observed to bore entrance holes.

In the young lianas censused at our study site, those with colonies of *T. tessmanni* at the foundation stage usually harbored other ant species, not specific to this plant, or larvae of the beetle *I. concolor*, which may be a specific parasite of the plant. In contrast, lianas with established colonies of *T. tessmanni*, in which large numbers of workers patrolled on the plant's surface, were occupied by a single colony to the exclusion of all other arthropods, including other ant species. The worker-transfer experiments clearly suggested that workers distinguished between members of the same and of other colonies, and that workers from the same plant were treated as con-colonial, even when transferred to points on the plant 50 m away from the point where they were captured. The host plant thus appears to be occupied by a single colony, in whose interest it is to efficiently protect the plant's entire crown.

In young individuals, incipient colonies must usually compete with other occupants. Nonspecialist ants that benefit from the nest-site resources offered by *V. thyrsiflora* may forage in the soil litter, on neighboring plants for prey and liquid food, or on *Vitex* for prey. In particular, *T. tabarum*, which feeds mainly on ants (Dejean & Djiéto-Lordon 1996), may find abundant prey in the domatia of this plant. The relative importance of interspecific competition and of predation by *T. tabarum* as factors influencing establishment of *T. tessmanni* colonies is unknown.

The specialist *T. tessmanni* appears to find all its food on the plant (Bequaert 1922). We have never found workers foraging on neighboring plants, and despite the absence of food bodies or nectaries on the plant, the ants never attended hemipterans for honeydew. This specialist ant is reported to feed mainly on wound callus produced by the cambium in response to chewing of the internal stem walls by the ants (Bequaert 1922). In the laboratory, we noted in domatia from which these ants had recently been removed the small depressions in the internal walls of the domatia indicated by Bequaert (1922) and the presence at the surfaces of these depressions of a poorly organized mass of plant tissue of undetermined nature. Such tissue, presumably the wound callus described by Bequaert (1922), was observed only in plants occupied by *T. tessmanni* and appears to be produced exclusively in response to their chewing of the internal walls of the domatia. Whether or not these ants eat the arthropods they encounter on the plant, or simply discard the insects they capture, as reported for many specialist plant-ants that provide highly effective protection of their host plants (Gaume & McKey [1998] and references therein) is not known with certainty. Wheeler and Bailey (1920) surmised from studies of contents of infrabuccal pouches that *T. tessmanni* is not insectivorous. However, they were prudent about the interpretation of their results. Some specialist plant-ants do eat insects encountered on the host plant (Gaume & McKey 1998, Dejean *et al.* 2001a,b). We have no direct evidence concerning this point for *T. tessmanni*. It is also unknown whether *T. tessmanni* workers glean pollen, spores, or other food from the leaves of the host,

as do some other pseudomyrmecines (Baroni Urbani & Andrade 1997).

Another arthropod frequent in the domatia was the larvae of *I. concolor*, a beetle that may be a specific parasite of the plant. Neither the original description of *I. concolor* (Kraatz 1899) nor the most recent revision of African Languriidae (Villiers 1961) includes any information on host plants, and the larvae appear not to have been previously collected. Larvae of the subfamily Languriinae develop in stems of herbaceous angiosperms while adults are found on leaves (Wegrzynowicz 2002), and the few species studied appear to be oligophagous. *I. concolor* is known from southern Cameroon, Equatorial Guinea, Gabon, and the Congo Republic, but there is also one record from Togo, much further west (Villiers 1961). These sites are all within the reported range of *Vitex thyrsiflora*, which extends from Guinea to the Congo basin (Hutchinson & Dalziel 1963), although whether populations from the West African forest block (Nigeria to Liberia) are myrmecophytes appears to be unknown. The feeding habits of the larvae of *I. concolor* (eating tender tissues on the internal stem walls) suggest that they may exploit the plant in a way similar to the obligate mutualist ant. However, our observations show that the plant does not respond to attack of this beetle by producing wound callus.

PLANT PROTECTION BY *T. TESSMANNI*.—The evidence we present for the protective role of *T. tessmanni* against phytophagous insects on *V. thyrsiflora* confirms the observations of Wheeler (1922) and Bequaert (1922). As described by these two authors, when the host plant is slightly disturbed, workers rush out of the hollow stems in large numbers and actively explore the plant. In addition to this, activity censuses (C. Djiéto-Lordon, pers. obs.) showed that workers of *T. tessmanni* patrolled principally the plant's young shoots, which are most vulnerable to phytophagous insects (see Fig. 1a). The relationship between ant and plant can thus be considered a protection mutualism. Ants appear to protect not only against leaf-eating insect herbivores, but also against the beetle and fly larvae that exploit pith in stems.

HOW *T. TESSMANNI* MONOPOLIZES MATURE LIANAS.—Numerous traits of plant and ant give a competitive advantage to *T. tessmanni*. First, this appears to be the only ant that opens entrance holes in stems of the plant, giving this ant a priority advantage. The narrow, elongate bodies of this pseudomyrmecine allow it to drill entrance holes small enough to exclude most other ants, and to defend each entrance hole against other ants small enough to enter (cf. McKey *et al.* 1999, Brouat *et al.* 2001, Palmer *et al.* 2002). Because the plant grows continuously, the pith of young stems can be continuously opened, with no septa marking periods of intermittent growth. Thus all of the plant's stems are connected in a single branched system of passageways, in which ants can move to any part of the plant without having to venture on its surface. All food produced by the plant is located within this private space. Furthermore, the food resource appears to be produced only in response to a specific behavior of *T. tessmanni*. The protection provided by *T. tessmanni* increases the likelihood that the plants it occupies grow and persist. Finally, the social structure of *T. tessmanni* may confer a strong competitive

advantage. All the occupants of a mature liana appear to constitute a single integrated colony, with multiple reproductive females that should confer capacity for very rapid growth, enabling the colony to occupy and defend a territory that is itself growing. Polygyny should also increase longevity and size of the colony (McKey *et al.* 1999, Feldhaar *et al.* 2000), further increasing its competitive ability.

Our study suggests an interest in detailed studies of other myrmecophytic climbers. Available data suggest some parallels. The hosts of *Cladomyrma* spp. (Formicinae) in tropical Asian forests, for example, include several vines (*Spatholobus, Millettia, Strychnos*) (Moog *et al.* 2003). In all of these, ant food resources appear to be completely absent from plant surfaces and restricted to trophobiotic hemipterans within stem cavities (Moog *et al.* 2003). *Korthalsia*, *Daemonorops*, and other rattans (lianescent palms) might also offer interesting counterpoints. However, several features of the system described here appear to be unique. These include the presence of a single interconnected, branching private space throughout the entire host plant, and the restriction of food production to this private space and apparently only in the presence of the mutualist.

ACKNOWLEDGMENTS

LITERATURE CITED

ACHOUNDONG, G. 1985. Etude écologique et floristique de la végétation des collines de Yaoundé au dessus de 1000 m. Thèse de Doctorat, Université de Yaoundé. 301 pp.

BAILEY, I. W. 1922. Notes on Neotropical ant-plants. I. *Cecropia angulata*, sp. nov. Bot. Gazette 74: 369–391.

BARONI URBANI, C., AND M. L. DE ANDRADE. 1997. Pollen eating, storing and spitting by ants. Naturwissenschaften 84: 256–258.

BENTLEY, B. L. 1981. Ants, extrafloral nectaries, and the vine life-form: An interaction. Trop. Ecol. 22: 127–133.

BEQUAERT, J. 1922. Ants and their diverse relations to the plant world. Bull. Am. Mus. Nat. Hist. 45: 333–583.

BROUAT, C., N. GARCIA, C. ANDARY, AND D. MCKEY. 2001. Plant lock and ant key: Pairwise coevolution of an exclusion filter in an ant-plant mutualism. Proc. R. Soc. Lond. B 268: 2131–2141.

BUSCHINGER, A., R. W. KLEIN, AND U. MASCHWITZ. 1994. Colony structure of a bamboo-dwelling *Tetraponera* sp. (Hymenoptera: Formicidae: Pseudomyrmecinae) from Malaysia. Insectes sociaux 41: 29–41.

DAVIDSON, D. W., R. B. FOSTER, R. R. SNELLING, AND P. LOZADA. 1990. Variable composition of some tropical ant-plant symbioses. *In* P. W. Price, T. M. Lewinsohn, G. W. Fernandez, and W. W. Benson (Eds.). Plant-animal interactions: Evolutionary ecology in tropical and temperate regions, pp. 145–162. John Wiley, New York.

———, J. T. LONGINO, AND R. R. SNELLING. 1988. Pruning of host plant neighbors by ants: An experimental approach. Ecology 69: 801–808.

———, AND D. MCKEY. 1993. The evolutionary ecology of symbiotic ant-plant. J. Hymenoptera Res. 2: 13–83.

———, R. R. SNELLING, AND J. T. LONGINO. 1989. Competition among ants for myrmecophytes and the significance of plant trichomes. Biotropica 21: 64–73.

DEJEAN, A., A. AKOA, C. DJIÉTO-LORDON, AND A. LENOIR. 1994. Mosaic ant territories in an African secondary rain forest (Hymenoptera: Formicidae). Sociobiology 23: 275–292.

———, AND C. DJIÉTO-LORDON. 1996. Ecological studies on the relationships between ants (Hymenoptera: Formicidae) and the myrmecophyte *Scaphopetalum thonneri* (Sterculiaceae). Sociobiology 28: 91–102.

———, P. J. SOLANO, M. BELIN-DEPOUX, P. CERDAN, AND B. CORBARA. 2001a. Predatory behavior of patrolling *Allomerus decemarticulatus* workers (Formicidae; Myrmicinae) on their host plant. Sociobiology 37: 571–578.

———, P. J. SOLANO, J. ORIVEL, M. BELIN-DEPOUX, P. CERDAN, AND B. CORBARA. 2001b. The spread-eagling of prey by the obligate plant-ant *Pheidole minutula* (Myrmicinae): Similarities with dominant arboreal ants. Sociobiology 38: 675–682.

FELDHAAR, H., B. FIALA, R. bin HASHIM, AND U. MASCHWITZ. 2000. How to maintain an ant-plant symbiosis: Secondary polygyny in the *Macaranga triloba-Crematogaster* sp. association. Naturwissenschaften 87: 408–411.

FIALA, B., AND U. MASCHWITZ. 1992. Domatia as most important adaptations in the evolution of myrmecophytes in the Paleotropical tree genus *Macaranga* (Euphorbiaceae). Plant Syst. Evol. 180: 53–64.

GAUME, L., AND D. MCKEY. 1998. Protection against herbivores of the myrmecophyte Leonardoxa africana (Baill.) Aubrèv. T3 by its principal ant inhabitant *Aphomomyrmex afer* Emery. Comptes Rendus de l'Académie des Sciences de Paris. Sciences de la Vie/Life Sciences 321: 593–601.

HUTCHINSON, J., AND J. M. DALZIEL. 1963. Flora of West Tropical Africa. Vol. 2. Crown Agents for Overseas Governments and Administrations, London. 544 pp.

JANZEN, D. H. 1969. Allelopathy by myrmecophytes: The ant *Azteca* as an allelopathic agent of *Cecropia*. Ecology 50: 147–153.

JANZEN, D. H. 1972. Protection of *Barteria* (Passifloraceae) by *Pachysima* ants (Pseudomyrmecinae) in a Nigerian rain forest. Ecology 53: 885–892.

KRAATZ, G. 1899. Ueber die Languriiden-Arten von Kamerun nebst einigen verwandten Formen. Deutsche Entomologische Zeitschrift: 307–315.

MCKEY, D., L. GAUME, AND A. DALECKY. 1999. Les symbioses entre plantes et fourmis arboricoles. L'Année Biologique 38: 1–26.

MOOG, J., B. FIALA, M. WERNER, A. WEISSFLOG, L. G. SAW, AND U. MASCHWITZ. 2003. Ant-plant diversity in peninsular Malaysia, with spe-cial reference to the Pasoh Forest Reserve. *In* T. Okuda, N. Manokaran, Y. Matsumoto, K. Niiyama, S. C. Thomas, and P. S. Ashton (Eds.). Pasoh: Ecology of a lowland rainforest in southeast Asia, pp. 459–494. Springer Press, Tokyo.

MOOG, J., T. DRUDE, AND U. MASCHWITZ. 1998. Protective function of the plant-ant *Cladomyrma maschwitzi* to its host *Crypteronia griffithii*, and the dissolution of mutualism (Hymenoptera: Formicidae). Sociobiology 31: 105–129.

PALMER, T. M., T. P. YOUNG, AND M. L. STANTON. 2002. Burning bridges: Priority effects and the persistence of a competitively subordinate acacia-ant in Laikipia, Kenya. Oecologia 133: 372–379.

PALMER, T. M., T. P. YOUNG, M. L. STANTON, AND E. WENK. 2000. Short-term dynamics of an acacia ant community in Laikipia, Kenya. Oecologia 123: 425–435.

RENNER, S. S., AND R. E. RICKLEFS. 1998. Herbicidal activity of domatia-inhabiting ants in patches of *Tococa* guianensis and *Clidemia heterophylla*. Biotropica 30: 623–631.

SCHNELL, R., AND F. GROUT DE BEAUFORT. 1966. Contribution à l'étude des plantes à myrmécodomaties de l'Afrique intertropicale. Mémoires de l'IFAN 75: 1–66.

SAS 1996. SAS/STAT user's guide, release 6.12 edition. SAS Institute, Cary, North Carolina.

STANTON, M. L., T. M. PALMER, T. P. YOUNG, A. EVANS, AND M. L. TURNER. 1999. Sterilization and canopy modification of a swollen thorn acacia tree by a plant-ant. Nature 401: 578–581.

SYSTAT. 1997. SYSTAT version 7.0.1, SPSS Inc., Chicago, Illinois.

VILLIERS, A. 1961. Révision des Coléoptères Languriidés africains. Annales du Musée Royal de l'Afrique Centrale, Sciences Zoologiques, no. 98. 385 pp.

VON DOHLEN, C., AND N. A. MORAN. 1995. Molecular phylogeny of the Homptera: A paraphyletic taxon. J. Mol. Evol. 41: 211–223.

WARD, P. S. 1991. Phylogenetic analysis of pseudomyrmecine ants associated with domatia-bearing plants. *In* C. R. Huxley and D. F. Cutler (Eds.). Ant-plant interactions, pp. 335–352. Oxford University Press, Oxford.

———. 1999. Systematics, biogeography, and host plant associations of the *Pseudomyrmex viduus* group (Hymenoptera: Formicidae), *Triplaris*- and *Tachigali*-inhabiting ants. Zool. J. Linn. Soc. 126: 451–540.

WEGRZYNOWICZ, P. 2002. Morphology, phylogeny and classification of the family Erotylidae based on adult characters (Coleoptera: Cucujoidea). Genus 13: 435–505.

WHEELER, W. M. 1922. The ants collected by the American Museum Congo expedition. Bull. Am. Mus. Nat. Hist. 45: 39–269.

WHEELER, W. M., AND I. W. BAILEY. 1920. The feeding habits of pseudomyrmine and other ants. Trans. Am. Phil. Soc. Philadelphia (NS) 22: 235–279.

YOUNG, T. P., C. H. STUBBLEFIELD, AND L. A. ISBELL. 1997. Ants on swollen-thorn acacias: Coexistence in a simple system. Oecologia 109: 98–107.

YUMOTO, T., AND T. MARUHASHI. 1999. Pruning behaviour and inter-colony competition of *Tetraponera* (*Pachysima*) *aethiops* (Pseudomyrmecinae, Hymenoptera) in *Barteria fistulosa* in a tropical forest, Democratic Republic of Congo. Ecol. Res. 14: 393–404.

APPENDIX 1.   *Associations between ant species occupying internodes of* V. thyr-siflora. *All samples were taken from Mont Kala.*

Occupants	No. of plants	Percent of total
Unoccupied by ants	5	4.35
One species		
Cataulacus sp.	4	3.48
Tapinoma sp.	6	5.22
Pheidole megacephala	1	0.87
Crematogaster sp.	2	1.74
Tetramorium tabarum	1	0.87
Monomorium sp.	1	0.87
Tetraponera tessmanni*	17	14.78
T. tessmanni**	4	3.48
T. tessmanni***	12	10.43
Total	48	41.74
Two species		
Monomorium sp. + P. megacephala	1	0.87
Cataulacus sp. + Tetramorium sp.	1	0.87
Crematogaster sp. + Tetramorium sp.	1	0.87
T. tessmanni* + P. megacephala	5	4.35
T. tessmanni* + Cataulacus sp.	2	1.74
T. tessmanni* + Tet. Tabarum	11	9.57
T. tessmanni* + Tetramorium sp.	4	3.48

APPENDIX 1.   *Continued.*

Occupants	No. of plants	Percent of total
T. tessmanni* + Tapinoma sp.	2	1.74
T. tessmanni* + Monomorium sp.	7	6.09
T. tessmanni** + P. megacephala	1	0.87
T. tessmanni** + Tet. Tabarum	11	9.57
T. tessmanni** + Tapinoma sp.	1	0.87
T. tessmanni** + Crematogaster sp.	1	0.87
T. tessmanni*** + P. megacephala	1	0.87
Total	53	46.09
Three species		
T. tessmanni* + T.tabarum + Cataulacus sp.	2	1.74
T. tessmanni* + Tetramorium sp.* + Tapinoma sp.	1	0.87
T. tessmanni* + T.tabarum* + Tapinoma sp.	1	0.87
T. tessmanni* + P. megacephala + Tetramorium sp.	1	0.87
T. tessmanni** + P. megacephala + Tetramorium sp.	1	0.87
T. tessmanni** + Cataulacus sp. + Tet. Tabarum	1	0.87
T. tessmanni** + Tapinoma sp. + Tet. Tabarum	2	1.74
Total	9	7.83
Overall Total	115	100

* = foundations; ** = incipient colonies; *** = established colonies.