SYSTEMATICS, MORPHOLOGY AND PHYSIOLOGY

Morphological Caste Studies In The Neotropical Swarm-Founding Polistinae Wasp *Angiopolybia pallens* (Lepeletier) (Hymenoptera: Vespidae)

LUIZ F.F. GELIN[1], JUCELHO D. DA CRUZ[2], FERNANDO B. NOLL[1], EDILBERTO GIANNOTTI[3], GILBERTO M. DE M. SANTOS[2] AND CARLOS C. BICHARA-FILHO[2]

[1]*Lab. Vespas Sociais, Depto. Zoologia e Botânica, Univ. Estadual Paulista "Júlio de Mesquita Filho", Instituto de Biociências, Letras e Ciências Exatas, Rua Cristóvão Colombo 2265, Jardim Nazareth 15054-000, São José do Rio Preto, SP; fernandogelin@yahoo.com.br, noll@ibilce.unesp.br*
[2]*Lab. Entomologia, Depto. Ciências Biológicas, Univ. Estadual de Feira de Santana, km 3 BR 116, Campus Universitário 44031-460, Feira de Santana, BA; jucelho@uefs.br, gmms@uefs.br, bichara@uefs.br*
[3]*Depto. Zoologia. Instituto de Biociências de Rio Claro, Univ. Estadual Paulista "Júlio de Mesquita Filho", Avenida 24-A, 1515 Bela Vista 13506-900, Rio Claro, SP; edilgian@rc.unesp.br*

Neotropical Entomology 37(6):691-701 (2008)

Estudos Morfológicos das Castas da Vespa Polistinae Enxameadora Neotropical, *Angiopolybia pallens* (Lepeletier) (Hymenoptera: Vespidae)

RESUMO - As vespas neotropicais enxameadoras possuem colônias poligínicas e uma variação muito grande em relação à diferenciação das castas, desde espécies em que rainhas e operárias possuem formas e tamanhos semelhantes até aquelas em que grande diferença na forma e no tamanho são observados. Estudos morfométricos utilizando oito medidas corporais em nove colônias de *Angiopolybia pallens* (Lepeletier) foram realizados a fim de aumentar o conhecimento sobre diferenciação de castas em Epiponini, empregando-se análise canônica discriminante e teste *t* de Bonferroni. Os indivíduos foram dissecados para determinar o nível de desenvolvimento ovariano e as espermatecas foram removidas para verificar a ocorrência de inseminação e assim definir os três grupos de fêmeas: rainhas (ovários desenvolvidos e espermateca fecundada), intermediárias (ovários desenvolvidos e espermateca não-fecundada) e operárias (ovários não-desenvolvidos). Apesar de serem encontradas diferenças significativas para algumas medidas em sete das nove colônias, a análise multivariada (lambda de Wilks) mostrou que as castas são de difícil distinção apenas pela morfologia. É possível concluir que diferenças morfológicas entre castas de *A. pallens* são pequenas ou ausentes, o que reflete o plano basal para os Epiponini, i.e. padrão pós-imaginal de diferenciação castas.

PALAVRAS-CHAVE: Inseto social, morfometria, Epiponini, diferenciação de castas

ABSTRACT - Neotropical swarm-founding wasps present polygynic colonies and a great variation in relation to caste differentiation, ranging from species in which queens and workers are similar in shape and size to those where variation in shape and size is conspicuous. Canonical discriminant analysis and Bonferroni *t*-test analysis on morphometric data collected from eight body parts of specimens from nine colonies of *Angiopolybia pallens* (Lepeletier) were undertaken as a step towards to a better understanding on caste differenciation in Epiponini. All specimens were dissected to verify the ovary developmental stage, and the spermatheca was removed to check for the mating status. Females were then grouped as queens (mated with developed ovaries), intermediates (virgin with developed ovaries) and workers (virgin with undeveloped ovaries). Even though differences were found for some measurements in seven out of the nine colonies evaluated, multivariate analysis (Wilks' lambda values) indicated castes could not be reliably distinguished only by morphology. We conclude that morphological differences among *A. pallens* castes are low or absent, reflecting a possible groundplan for the Epiponini, i.e. a post-imaginal pattern of caste differentiation.

KEY WORDS: Social insect, morphometry, Epiponini, caste differentiation

Eusocial insects with simple societies have no typical morphological castes of totipotent females, and colony social environment determines whether they will become workers or queens (Solís & Strassmann 1990). In complex societies like honeybees, workers and queens differ morphologically, with their caste being determined before adulthood, generally by differential feeding (Bourke 1999).

The swarm-founding polistine belongs to the Neotropical tribe Epiponini (Carpenter 1997). In Epiponini, species with or without caste morphological differences can both be found (Richards 1978, Jeanne 1980). In Polistinae, five main caste forms can be identified: (i) morphological differences between queens and workers are absent in the whole colony cycle; (ii) morphological differences between queens and workers are absent in the whole colony cycle but young females would develop ovaries only in some phases of colony cycle; (iii) morphological and physiological discontinuity between castes are subjected to colony cycle; (iv) castes differences increase according to the colony cycle, and virgin, egg-laying females appear in some phases of colony cycle; and (v) morphological differences between castes are constant and queens are always distinct from workers during colony cycle.

Angiopolybia Araujo is a basal genus on the Epiponini phylogeny (Wenzel & Carpenter 1994) that occurs on the rain forests of Ecuador, Colombia, Venezuela, Trinidad, Peru, and Brazil (Richards 1978, Andena *et al*. 2007). Cruz *et al*. (2006a, b) studied some aspects of the biology and foraging activities of these wasps, like nest site selection, flying capacity, daily activity and resource collection. *Angiopolybia* presents no morphological differences associated with ovary development, which is probably due to a post-imaginal caste determination (Noll *et al.* 2004). A similar pattern was found in some species of *Parachartergus* Ihering, *Pseudopolybia* Dalla Torre, *Chartergellus* Bequaert, *Brachygastra* Perty, *Synoeca* Saussure, and *Metapolybia* Ducke (Mateus *et al*. 2004, Noll *et al*. 2004). However, comprehensive studies on caste differentiation on *Angiopolybia pallens* (Lepeletier) are lacking, and they are made interesting as some species of epiponines can alter the level of caste dimorphism in some species (Noll & Zucchi 2000, 2002). Therefore, we aim at a detailed analysis of caste morphological and physiological differentiation in nine colonies of *A. pallens*.

Materials and Methods

Specimens of *A. pallens* were taken from nine colonies. Three colonies were collected in Presidente Figueiredo (Amazon Rain Forest), north Brazil on June 2004 (colonies G18, G36 and G39), two in Ilhéus (14º 47' S; 39º 03' W), Bahia state, northeast Brazil (G1 e G2) on July 2005, and the last four (J1, J2, J3 and J4) were collected between March and June 2003 in Vera Cruz (12º 58' S; 38º 36' W) and Ilhéus, Bahia state.

In order to detect caste morphological differences, the head width (HW), minimum interorbital distance (PML), mesoscutum width (MSW), basal width of tergite II (T2BW), alitrunk length (AL), basal height of basal tergite I (T1BH), length of gastral tergite I (T1L) and partial forewing length (WL) of females were measured under a stereomicroscope. Female mating status and ovary development were examined for all evaluated specimens to allow for caste identification. Female mating status was evaluated by removal of the spermatheca and verification of sperm presence under a microscope. Numerical data were analyzed statistically in relation to the caste, which were determined by the ovarian and spermathecal conditions. Statistical analyses including the canonical discriminant analyses based on discriminant function analyses using the stepwise procedure (CDA: Rao 1973) and comparisons among means (Bonferroni *t*-test) were performed with Statistica for Windows Software version 7.0.

Nest descriptions were made by counting and measuring the combs, the position of the nest in relation to the substrate, material used in nest construction, position of entrance, number of pedicels, orientation of the fibers and type of substrate.

Results

Nest architecture and colony phase. Four nests were used to study their architecture. All nests were found under leaves, and the entrance was oriented downwards taking the shape of a funnel (Fig. 1A), and the combs were suspended on the leaves by a central pedicel (Fig. 1B). The central axis of the nest was in a diagonal to the leaves. The single envelop was directly attached to the leaf skirted the combs. Other combs were suspended by pedicels, both from those above and from the envelope. Two nests had three combs, one had five combs, and another one had six. The largest reached 12 cm in length and 8 cm in width, while the others were 6 cm x 9 cm, 4,5 cm x 8 cm and 5,5 cm x 8 cm in size. Combs were connected to one another by a strong central pedicel and by some auxiliary pedicels. Nest material was a mixture of vegetal fibers and salivary secretions. The lines of construction were long and compact, forming parallel lines of fiber.

Colony J1 presented only one comb, few eggs and larvae, and we assumed it was in the pre-emergence phase. The colonies G1, G2, G18, G36, G39 and J2 were in worker-production phase as they had many eggs, larvae and pupae, while colonies J3 and J4 were in male-production phase.

Ovarian development and contents of the spermatheca. Four different ovary developmental stages were recognized in the colonies analyzed (Fig. 2): stage A - ovaries at the pre-vitellogenic stage, stage B - ovarioles with early vitellogenic oocytes, stage C - ovarioles with the late vitellogenic oocytes, and stage D - ovarioles with post-vitellogenic oocytes. Females with ovaries in stages A, B and C were all virgin, as indicated by their empty spermathecae. Therefore, only females with ovaries at stage D were mated and regarded as queens, while females of ovaries at stage B and C were classified as intermediates, and females at stage A as workers. The frequency distribution of each ovary type is shown in Table 1.

Morphological differences between castes. Seven colonies differed on some morphological measurements among the

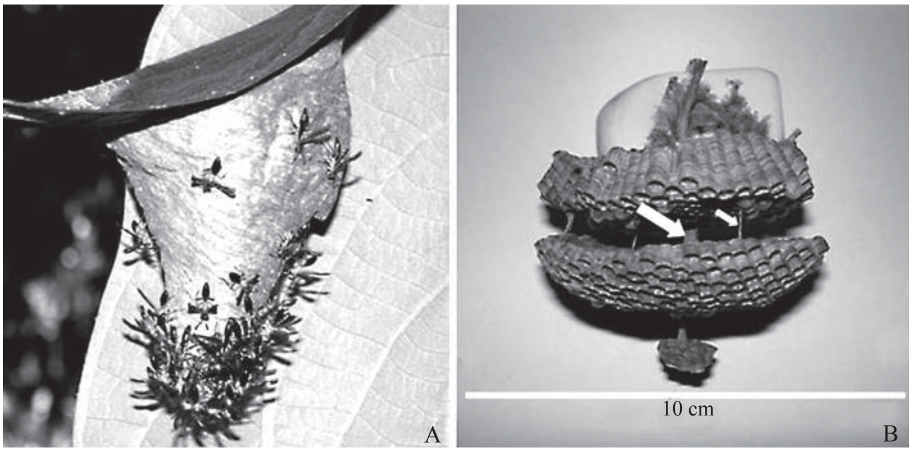

Fig. 1. Nest of *A. pallens*. A – General view of a nest. B – The combs suspended from those above by pedicels. The large arrow indicates the main pedicel, the small one indicates an auxiliary pedicel.

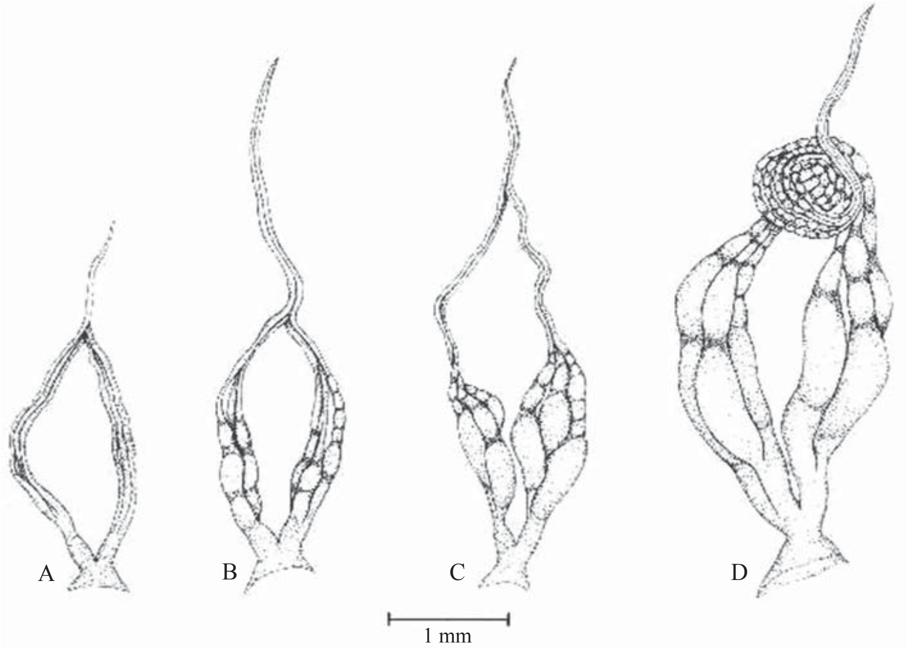

Fig. 2. The four types of ovaries in *A. pallens* showing workers ovaries (A and B), intermediates ovaries (C) and queens ovaries (C and D).

Table 1. Frequency of the four types of ovaries within the colonies of *A. pallens*.

Colony	Cycle phase	Types of ovarian %			
		A	B	C	D
J1	Pre-emergence	29	9	58	3
J2	Work production	42	21	33	4
J3	Male production	19	20	59	2
J4	Male production	30	32	33	4
G1	Work production	46	16	29	9
G2	Work production	57	5	7	31
G18	Work production	35	21	29	15
G36	Work production	37	24	26	13
G39	Work production	39	15	35	11

different castes ($P < 0.05$), with no differences being detected for colonies J2 and G1. In colonies where differences among caste were detected, queens were larger than workers and intermediates in a few measurements (Table 2). For the other colonies, intermediates and workers did not differ, with exception of G2, in which workers were larger than intermediates in a few measurements (Table 2). No specific body part was significantly different in most of the colonies (Table 3).

Higher Wilks' lambda values found in all colonies revealed that each character was not a good discriminator, and only a combination of variables could detect any difference (Table 3). Therefore, despite univariate statistics being significant, the values of Wilks' lambda were always above 0.7, indicating a very low level of discrimination. In fact, Eigen values for the canonical analyses (Table 4) indicate a

Table 2. Univariate analysis and Bonferroni *t*-test for the nine analyzed colonies of *A. pallens*.

		Means ± SD							
Colony #	Characters	Queens	Intermediates	Workers	ANOVA	P	Q/W	Q/I	W/I
G18		n = 27	n = 46	n = 63	(F)				
	HW	2.10 ± 0.53	2.08 ± 0.50	2.08 ± 0.44	3.46	0.03	Q > W*		
	PML	0.88 ± 0.03	0.87 ± 0.03	0.87 ± 0.03	1.38	0.25			
	MSW	1.31 ± 0.06	1.29 ± 0.05	1.28 ± 0.04	2.37	0.1			
	T2BW	0.81 ± 0.05	0.79 ± 0.04	0.78 ± 0.04	3.89	0.02	Q > W*		
	AL	2.93 ± 0.13	2.86 ± 0.10	2.85 ± 0.15	3.61	0.03	Q > W*		
	T1BH	0.30 ± 0.02	0.30 ± 0.02	0.30 ± 0.03	0.28	0.75			
	T1L	1.67 ± 0.11	1.65 ± 0.07	1.62 ± 0.08	4.63	0.01	Q > W*		
	WL	3.42 ± 0.13	3.37 ± 0.13	3.38 ± 0.13	1.5	0.23			
G36		n = 16	n = 22	n = 48	(F)				
	HW	2.20 ± 0.05	2.18 ± 0.03	2.15 ± 0.06	6.11	<0.01	Q > W#		I > W*
	PML	0.95 ± 0.02	0.95 ± 0.03	0.94 ± 0.03	2.67	0.07			
	MSW	1.40 ± 0.05	1.40 ± 0.04	1.38 ±0.06	0.54	0.58			
	T2BW	0.85 ± 0.03	0.86 ± 0.03	0.85 ± 0.03	0.38	0.68			
	AL	3.00 ± 0.12	3.06 ± 0.07	3.02 ± 0.11	1.54	0.22			
	T1BH	0.34 ± 0.03	0.34 ± 0.03	0.34 ± 0.02	0.24	0.78			
	T1L	1.84 ± 0.09	1.86 ± 0.07	1.85 ± 0.09	0.17	0.84			
	WL	3.75 ± 0.12	3.74 ± 0.07	3.73 ± 0.14	0.17	0.85			
G39		n = 19	n = 15	n = 66	(F)				
	HW	2.18 ± 0.05	2.16 ± 0.08	2.13 ± 0.09	3.51	0.03	Q > W*		
	PML	0.93 ± 0.03	0.93 ± 0.04	0.91 ± 0.04	2.45	0.09			
	MSW	1.38 ± 0.06	1.38 ± 0.08	1.33 ± 0.07	4.51	0.01	Q > W*		
	T2BW	0.84 ± 0.56	0.82 ± 0.53	0.80 ± 0.51	3.97	0.02	Q > W*		
	AL	3.12 ± 0.08	3.10 ± 0.13	3.00 ± 0.15	6.91	<0.01	Q > W#		
	T1BH	0.31 ± 0.03	0.32 ± 0.04	0.30 ± 0.03	3.07	0.05			
	T1L	1.80 ± 0.09	1.82 ± 0.11	1.76 ± 0.10	2.71	0.07			
	WL	3.82 ± 0.11	3.78 ± 0.18	3.68 ± 0.20	4.7	0.01	Q > W*		
G1		n = 14	n = 70	n = 73					
	HW	2.30 ± 0.06	2.28 ± 0.06	2.28 ± 0.07	0.71	0.49			
	PML	0.98 ± 0.03	0.97 ± 0.04	0.97 ± 0.03	1.53	0.22			
	MSW	1.52 ± 0.03	1.50 ± 0.05	1.51 ± 0.05	0.43	0.65			
	T2BW	0.9 ± 0.04	0.9 ± 0.04	0.9 ± 0.04	0.001	1.00			
	AL	3.29 ± 0.1	3.27 ± 0.13	3.27 ± 0.12	0.21	0.81			
	T1BH	0.36 ± 0.02	0.35 ± 0.03	0.36 ± 0.03	0.25	0.78			
	T1L	1.98 ± 0.09	1.95 ± 0.13	1.98 ± 0.08	2.56	0.08			
	WL	4.03 ± 0.16	4.02 ± 0.17	4.03 ± 0.13	1.31	0.27			

Continue

Table 2. Continuation.

		Means ± SD							
Colony #	Characters	Queens	Intermediates	Workers	ANOVA	P	Q/W	Q/I	W/I
G2		n = 24	n = 63	n = 116					
	HW	2.22 ± 0.05	2.22 ± 0.05	2.23 ± 0.05	1.09	0.34			
	PML	0.93 ± 0.03	0.94 ± 0.03	0.94 ± 0.03	1.26	0.29			
	MSW	1.43 ± 0.04	1.44 ± 0.05	1.47 ± 0.05	9.83	<0.01	Q < W*		I < W*
	T2BW	0.89 ± 0.04	0.86 ± 0.04	0.88 ± 0.05	5.00	0.01	Q > W*		
	AL	3 1 ± 0 1	3.12 ± 0.14	3.13 ± 0.12	0.81	0.45			
	T1BH	0.34 ± 0.02	0.33 ± 0.02	0.34 ± 0.02	8.70	<0.01		I < W#	
	T1L	1.87 ± 0.07	1.95 ± 0.09	1.86 ± 0.09	0.85	0.43			
	WL	3.77 ± 0.12	3.80 ± 0.14	3.85 ± 0.16	3.26	0.04			
J1		n = 35	n = 145	n = 75					
	HW	2.29 ± 0.07	2.26 ± 0.06	2.25 ± 0.06	4	0.02	Q > W*		
	PML	0.96 ± 0.03	0.95 ± 0.03	0.95 ± 0.03	4.7	0.01	Q > W#	Q > I*	
	MSW	1 5 ± 0 13	1.46 ± 0.09	1.44 ± 0.05	7.29	<0.01	Q > W#	Q > I*	
	T2BW	0.86 ± 0.03	0.84 ± 0.03	0.85 ± 0.03	4.5	0.01		Q > I*	
	AL	3.26 ± 0.16	3.18 ± 0.13	3.16 ± 0.11	7.4	<0.01	Q > W#	Q > I#	
	T1BH	0.38 ± 0.03	0.38 ± 0.03	0.37 ± 0.03	1.58	0.21			
	T1L	1.64 ± 0.11	1.56 ± 0.09	1.54 ± 0.13	2.83	0.06			
	WL	3.61 ± 0.18	3.52 ± 0.14	3.47 ± 0.14	11.7	<0.01	Q > W#	Q > I#	I > W*
J2		n = 19	n = 66	n = 62					
	HW	2.27 ± 0.04	2.27 ± 0.04	2.24 ± 0.04	0.5	0.6			
	PML	0.97 ± 0.01	0.96 ± 0.02	0.96 ± 0.02	0.8	0.46			
	MSW	1.49 ± 0.05	1.47 ± 0.05	1.47 ± 0.05	1.35	0.26			
	T2BW	0.89 ± 0.03	0.89 ± 0.03	0.89 ± 0.02	0.1	0.86			
	AL	3.25 ± 0.09	3.24 ± 0.11	3.22 ± 0.14		0.61			
	T1BH	0 4 ± 0 04	0.4 ± 0.04	0.39 ± 0.04	0.77	0.46			
	T1L	1.63 ± 0.07	1.63 ± 0.1	1.65 ± 0.09	0.78	0.46			
	WL	3.46 ± 0.11	3.48 ± 0.10	3.47 ± 0.09	0.7	0.49			
J3		n = 11	n = 59	n = 16					
	HW	2.35 ± 0.04	2.33 ± 0.05	2.3 ± 0.04	4.5	0.01		Q > I*	
	PML	0.98 ± 0.02	0.97 ± 0.03	0.95 ± 0.02	5.11	<0.01	Q > W*		I > W*
	MSW	1.55 ± 0.04	1.54 ± 0.05	1.50 ± 0.05	5.99	<0.01	Q > W*		I > W#
	T2BW	0.90 ± 0.02	0.89 ± 0.04	0.86 ± 0.04	6.13	<0.01	Q > W#		I > W#
	AL	3.37 ± 0.08	3.31 ± 0.11	3.26 ± 0.07	3.82	0.02	Q > W*		
	T1BH	0.38 ± 0.02	0.38 ± 0.02	0.38 ± 0.02	0	0.99			
	T1L	1.64 ± 0.11	1.62 ± 0.10	1.58 ± 0.08	1.71	0.19			
	WL	3.74 ± 0.11	3.71 ± 0.09	3.64 ± 0.11	4.26	0.02	Q > W*		I > W*

Continue

Table 2. Continuation.

		Means ± SD							
Colony #	Characters	Queens	Intermediates	Workers	ANOVA	P	Q/W	Q/I	W/I
J4		n = 21	n = 48	n = 30					
	HW	2.36 ± 0.05	2.35 ± 0.05	2.32 ± 0.14	1.64	0.2			
	PML	1.00 ± 0.03	0.98 ± 0.03	0.97 ± 0.04	3.78	0.03	Q > W*		
	MSW	1.58 ± 0.03	1.56 ± 0.04	1.54 ± 0.07	4.11	0.02	Q > W*		
	T2BW	0.92 ± 0.03	0.91 ± 0.03	0.91 ± 0.04	0.56	0.57			
	AL	3.40 ± 0.12	3.32 ± 0.18	3.33 ± 0.16	1.54	0.22			
	T1BH	0.39 ± 0.02	0.38 ± 0.02	0.37 ± 0.02	3.52	0.03	Q > W*		
	T1L	1.73 ± 0.09	1.68 ± 0.09	1.73 ± 0.20	1.99	0.14			
	WL	3.78 ± 0.09	3.71 ± 0.10	3.70 ± 0.18	3.06	0.05			
							#P < 0.01	*P < 0.05	

Table 3. Discriminant morphometric variables between queens and workers in *A. pallens*.

Colony #	Variable	Wilks' Lambda	F	P
G18				
	T1L	0.86	3.28	0.04
	HW	0.84	2.16	0.12
	AL	0.83	1.34	0.26
	WL	0.85	2.73	0.07
	T2BW	0.83	1.42	0.25
	MSW	0.83	1.33	0.27
G36				
	HW	0.93	11.52	<0.01
	AL	0.78	3.52	0.03
	WL	0.76	2.14	0.12
	T1BH	0.74	1.03	0.36
G39				
	AL	0.94	5.41	0.01
	T1BH	0.88	1.72	0.19
G1				
	T1L	1.00	2.56	0.08
G2				
	MSW	0.84	9.72	<0.01
	T1BH	0.82	6.65	<0.01
	T2BW	0.82	6.26	<0.01
	HW	0.79	3.29	0.04
	T1L	0.78	1.31	0.27
J1				
	T2BW	0.88	4.71	<0.01
	WL	0.88	5.80	<0.01
J2				
	HW	0.94	0.19	0.82
	PML	0.94	0.30	0.74
	MSW	0.95	0.96	0.39
	T2BW	0.95	1.27	0.28
	AL	0.93	0.02	0.98
	T1BH	0.94	0.49	0.62
	T1L	0.94	0.49	0.62
	WL	0.95	1.22	0.30
J3				
	HW	0.78	0.06	0.94
	PML	0.78	0.23	0.80
	MSW	0.80	1.42	0.25
	T2BW	0.80	1.24	0.30
	AL	0.78	0.35	0.71
	T1BH	0.79	0.83	0.44
	T1L	0.78	0.45	0.64
	WL	0.78	0.14	0.87
J4				
	HW	0.83	1.08	0.35
	PML	0.82	0.71	0.49
	MSW	0.85	2.05	0.14
	T2BW	0.82	0.58	0.56
	AL	0.82	0.40	0.67
	T1BH	0.83	1.23	0.30
	T1L	0.82	0.54	0.58
	WL	0.83	1.36	0.26

Table 4. Canonical scores for the nine colonies of *A. pallens*.

	Eigenval	
Colony #	Root1	Root2
G1	0,073	0,033
G2	0,162	0,121
G18	0,132	0,082
G36	0,298	0,070
G39	0,163	0,018
J1	0,117	0,034
J2	0,035	0,001
J3	0,187	0,040
J4	0,112	0,043

very low separation of the groups because they recover only 29% of the variation in colony G36 and under 18% for the remaining colonies. Plotting the canonical scores, all groups of females determined a priori were collapsed, indicating measurements overlap (Fig. 3). This indicates the absence of a clear morphological differentiation between the castes. In despite of differences among single means, the capacity of separating queens from workers was almost null. Similarly, the classification analyses (Table 5) were imprecise in discriminating among castes, suggesting that the separation of the three previously defined groups (workers, queens and intermediates) cannot be made by size alone.

Discussion

Low caste differentiation is a common trait in the Epiponini, characterizing several genera such as *Protopolybia*,

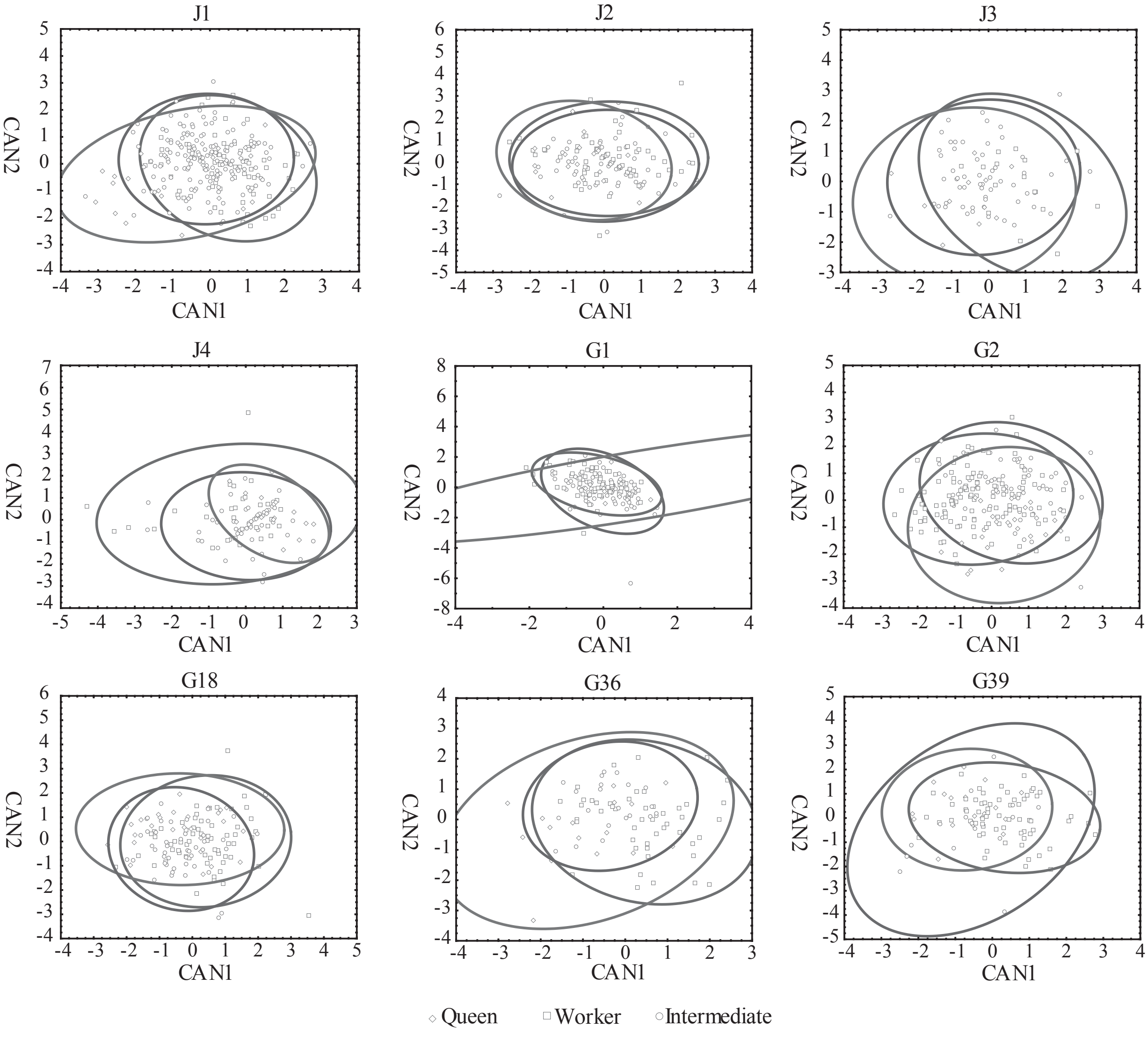

Fig. 3. Caste discrimination in the nine analyzed colonies of *A. pallens* based on the first two canonical roots. Each ellipse encompasses 95% of the variation.

Table 5. Classification analyses of the castes in the analyzed colonies of *A. pallens*.

		Predicted classification			
Colony #	Observed Classification	Worker	Queen	Intermediate	Correctly classified females (%)
G18					
	Worker	51	0	12	80.9
	Queen	16	7	4	25.9
	Intermediate	21	2	23	50
G36					
	Worker	40	3	5	83.3
	Queen	8	7	1	43.7
	Intermediate	13	3	6	27.3
G39					
	Worker	65	1	0	98.5
	Queen	19	0	0	0
	Intermediate	12	0	3	20
G1					
	Worker	50	0	23	68.5
	Queen	9	0	5	0
	Intermediate	35	0	35	50
G2					
	Worker	98	2	16	84.5
	Queen	16	4	4	16.7
	Intermediate	37	4	22	34.9
J1					
	Worker	21	54	0	28
	Queen	3	26	6	17
	Intermediate	14	129	2	89
J2					
	Worker	26	34	2	42
	Queen	7	12	0	0
	Intermediate	20	46	0	70
J3					
	Worker	4	12	0	25
	Queen	0	11	0	0
	Intermediate	3	55	1	93
J4					
	Worker	9	20	1	30
	Queen	2	18	1	4.76
	Intermediate	5	42	1	87.5

Parachartergus, *Pseudopolybia*, *Polybia, Angiopolybia*, *Chartergellus,* and *Brachygastra* (Mateus *et al*. 2004, Noll *et al*. 2004). The analyzed colonies of *A. pallens* showed that a few measurements were statistically significant to separate the groups previously defined. However, the high values of Wilks' lambda show that the overall differences are not enough to create a model able to classify the females as workers, intermediates or queens based only on morphology, as previously proposed by Noll *et al.* (2004).

In addition, the values of the canonic analysis show an overlapping of queens, workers and intermediates in the nine analyzed colonies (Fig. 3). Colonies with pre-imaginal determination show clear caste separation in the values of CAN1 (Baio *et al*. 1998; Noll & Zucchi 2000, 2002; Shima *et al*. 2000).

Based on the five patterns of caste differences established by Noll & Zucchi (2002), *A. pallens* belongs to the first type: queen-worker differences are practically absent and the castes are probably not determined during pre-imago development, but during adulthood instead.

The post-imaginal determination is given by the conflict between the individuals of the colony (West-Eberhard 1978, 1981), and not by their feeding during larval development (Strassman *et al.* 1991). For this reason caste morphological differences are practically absent. Colonies of *A. pallens* are characterized by the presence of intermediates during all the colony cycle and this was verified in all analyzed colonies. The presence of intermediates is related to the low degree of colony dimorphism (Richards 1978, Noll *et al*. 1996, Mateus & Noll 1997, Shima *et al*. 1998). Noll & Zucchi (2000) verified that for *Polybia occidentalis*, *P. scutellaris* and *P. paulista*, the presence of intermediates is related to the cycle of the colony, appearing in the phase of male production. This does not apply to *A. pallens*, because potentially reproductive females were found in all analyzed colonies. According to West-Eberhard (1978) they will produce their offspring in a next swarm with other queens. Richards (1971) related the intermediates with the production of trophic eggs. The presence of intermediates in all phases of the cycle was verified in *Protopolybia exigua* and *P. sedula* (Noll & Zucchi 2002). In these genera oophagy occurs and intermediates are in charge of laying such eggs (Naumman 1970).

Richards (1971) proposed and indirect correlation between the number of queens and the ovarian development in workers. In other words, when there are a few queens we might expect to find more workers with their ovaries developed and when there are several queens the workers are basically sterile. However, the presence of intermediates in colonies with low degree of morphological differentiation is common. In these groups, queens are incapable to regulate workers ovarian development (Noll 2005).

Phylogenetic analyses of Polistine genera place *Angiopolybia* as a sister group of *Agelaia* (Wenzel & Carpenter 1994). Studies on caste differentiation of *Agelaia vicina* (Sakagami *et al*. 1996, Baio *et al*. 1998), *A. multipicta* and *A. pallipes* (Noll *et al*. 1997) have shown morphological caste differences, being queens smaller than workers in some parts (usually in the head) and larger in others (usually abdomen), with no intermediate females. Clear differences were seen when comparing individuals of the two castes of *A. vicina* (Fig. 4), but there were no differences among castes of *A. pallens* (Fig. 5).

Fig. 4. Queen (A) and worker (B) of *A. vicina.*

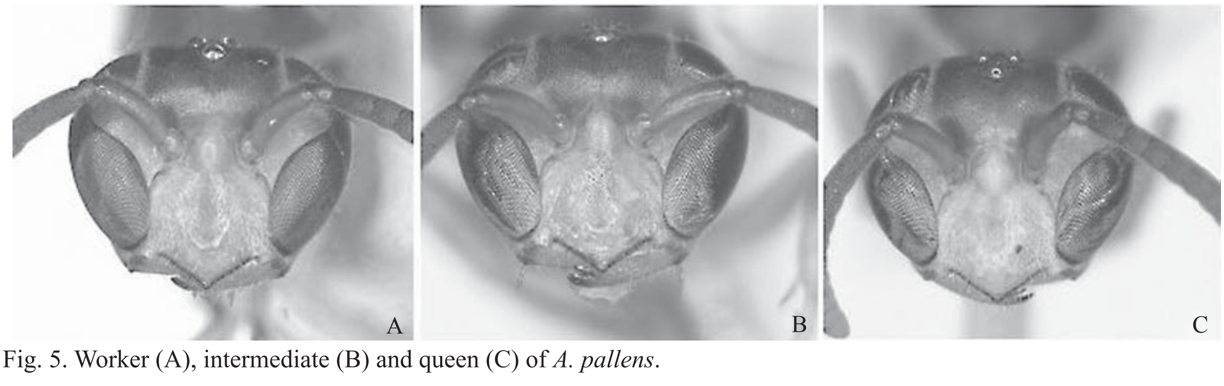

Fig. 5. Worker (A), intermediate (B) and queen (C) of *A. pallens*.

The pattern described for *Agelaia* was also observed in *Apoica* (Richards 1978, Shima *et al*. 1994). The presence of morphological castes in both *Agelaia* and *Apoica* suggests that pre-imaginal differentiation in these genera originated independently (Noll *et al*. 2004) and that *Angiopolybia* keeps the ancestral character of post-imaginal differentiation.

We can conclude that *A. pallens*, which belongs to a basal genus of the tribe Epiponini, preserves the post-imaginal caste determination which is the ancestral condition of the social behavior in Epiponini wasps. The post-imaginal determination of castes implies the absence of morphological differences between workers, queens and intermediates.

Acknowledgments

The authors thank J. R. Somera for the illustration of the ovaries of *A. pallens*; G. M. Tanaka Jr. for the photos of *Agelaia vicina*; to S. Mateus and O. A. L. Oliveira for collecting some of the nests used in this study; and P. C. B. Fernandes for helping in the laboratory. The authors also thank two anonymous reviewers for valuable comments. The authors acknowledge the financial support by FAPESP (grants 04/14646-0, 05/03569-8 and 07/0866-1) and CNPq (grants 300312/2004-0 and 479790/2004-3). Specimens were collected under permits from IBAMA (10739-1).

References

Andena, S.R., F.B. Noll & J.M. Carpenter. 2007. Phylogenetic analysis of the Neotropical social wasps of the genus *Angiopolybia* Araujo, 1946 (Hymenoptera, Vespidae, Polistinae). Zootaxa 1427: 57-64.

Baio, M.V., F.B. Noll, R. Zucchi & D. Simoes. 1998. Non-allometric caste differences in *Agelaia vicina* (Hymenoptera, Vespidae, Epiponini). Sociobiol. 32: 465-476.

Bourke, A.F.G. 1999. Colony size, social complexity and reproductive conflict in social insects. J. Evol. Biol. 12: 245-257.

Carpenter, J.M. 1997. A note on the names of paper wasp tribes (Insecta: Hymenoptera: Vespidae). Nat. Hist. Bull. Ibaraki Univ. 1: 15-16.

Cruz, J.D., E. Giannotti, G.M.M. Santos, C.C. Bichara-Filho & A.A. Rocha. 2006a. Nest site selection and flying capacity of Neotropical wasp *Angiopolybia pallens* (Hymenotera: Vespidae) in the Atlantic Rain Forest, Bahia State, Brazil. Sociobiology 47: 739-749.

Cruz, J.D., E. Giannotti, G.M.M. Santos, C.C. Bichara-Filho &. & J.J. Resende. 2006b. Daily activity resources collection by the swarm-founding wasp *Angiopolybia pallens* (Hymenotera: Vespidae). Sociobiology 47: 829-842.

Jeanne, R.L. 1980. Evolution of social behavior in the Vespidae. Annu. Rev. Entomol. 25: 371-396.

Mateus, S. & F.B. Noll. 1997. *Nectarinella xavantinensis*, a new Neotropical social wasp (Hymenoptera : Vespidae; Polistinae). J. N. Y. Entomol. Soc. 105: 45-49.

Mateus, S., F.B. Noll & R. Zucchi. 2004. Caste flexibility and variation according to the colony cycle in the swarm-founding wasp, *Parachartergus fraternus* (Hymenoptera: Vespidae: Epiponini). J. Kansas Entomol. Soc. 77: 281-294.

Naumann, M.G. 1970. The nesting behavior of *Protopolybia pumila* (Saussure, 1863) (Hymenoptera: Vespidae) in Panama. Doctoral Dissertation, University of Kansas, Kansas.

Noll, F.B. 2005. Reproductive caste in neotropical swarming wasps (Hymenoptera: Vespidae; Epiponini): A comprehensive analysis. Trends Entomol. 4: 59-66.

Noll, F.B., D. Simões & R. Zucchi. 1997. Morphological caste differences in Neotropical swarm-founding Polistinae wasps. *Agelaia m. multipicta* and *A. p. pallipes* (Hymenoptera: Vespidae). Ethol. Ecol. Evol. 9: 361-372.

Noll, F.B., J.W. Wenzel & R. Zucchi. 2004. Evolution of caste in Neotropical swarm-founding wasps (Hymenoptera: Vespidae; Epiponini). Am. Mus. Novit. 3467, 24p.

Noll, F.B. & R. Zucchi. 2000. Increasing caste differences related to life cycle progression in some neotropical swarm-founding polygynic polistine wasps (Hymenoptera Vespidae Epiponini). Ethol. Ecol. Evol. 12: 43-65.

Noll, F.B. & R. Zucchi 2002. Castes and the influence of the colony cycle in swarm-founding polistine wasps (Hymenoptera: Vespidae; Epiponini). Ins. Soc. 49: 62-74.

Noll, F.B., S. Mateus & R. Zucchi. 1996. Morphological caste differences in Neotropical swarm-founding Polistinae wasps. 5. *Protopolybia exigua* (Hymenoptera: Vespidae). J. N. Y. Entomol. Soc. 104: 62-69.

Rao, C.R. 1973, Linear statistic inference. Wiley, New York, 260p.

Richards, O.W. 1971. The biology of social wasps (Hymenoptera; Vespidae). Biol. Rev. 46: 45.

Richards, O.W. 1978. The social wasps of the Americas excluding the Vespinae. British Museum (Natural History), London, 580p.

Sakagami, S.F., R. Zucchi., S. Yamane., F.B. Noll & J.M.P. Camargo. 1996. Morphological caste differences in *Agelaia vicina*, the Neotropical swarm-founding Polistine wasp with the largest colony size among social wasps (Hymenoptera: Vespidae). Sociobiology 28: 208-223.

Shima, S.N., F.B. Noll. & R. Zucchi. 2000. Morphological caste differences in the Neotropical swarm-founding polistine wasp, *Brachygastra lecheguana* (Hymenoptera: Vespidae, Polistinae, Epiponini). Sociobiology 36: 41-52.

Shima, S.N., F.B. Noll, R. Zucchi & S. Yamane. 1998. Morphological caste differences in the Neotropical swarm-founding polistine wasps IV. *Pseudopolybia vespiceps*, with preliminary considerations on the role of intermediate females in social organization of the Epiponini (Hymenoptera, Vespidae). J. Hym. Res. 7: 280-295.

Shima, S.N., S. Yamane & R. Zucchi. 1994. Morphological caste differences in some Neotropical swarm-founding Polistinae wasps I. *Apoica flavissima* (Hymenoptera, Vespidae). Jap. J. Ent. 62: 811-822.

Silveira, O.T., M.C. Espósito, J.N. Santos Jr. & F.E. Gemaque Jr. 2005. Social wasps and bees captured in carrion traps in a rainforest in Brazil. Entomol. Sci. 8: 33-39.

Solis, C.R. & J.E. Strassmann. 1990. Presence of brood affects caste differentiation in the social wasp, *Polistes exclamans* Viereck (Hymenoptera: Vespidae) Funct. Ecol. 4: 531-541.

Strassmann, J. E., D.C. Queller, C.R. Solis & C.R. Hughes. 1991. Relatedness and queen number in the neotropical wasp, *Parachartergus colobopterus*. Anim. Behav. 42: 461-470.

Wenzel, J.W. & J.M. Carpenter. 1994. Comparing methods: Adaptive traits and tests of adaptation, p.79-101. In P. Eggleton & R.I. Vane-Wright (eds.), Phylogenetics and ecology: London: Academic Press, 376p.

West-Eberhard, M.J. 1978. Temporary queens in *Metapolybia* wasps: non-reproductive helpers without altruism? Science 200: 441-443.

West-Eberhard, M.J. 1981. Intragroup selection and the evolution of insect societies, p.3-17. In R.D. Alexander & D.W. Tinkle (ed.), Natural selection and social behaviour. Chiron Press, New York, 532p.

Received 24/IX/07. Accepted 27/XI/08.