COMPARATIVE MORPHOLOGICAL STUDY OF *TETRAMORIUM CAESPITUM* (LINNÉ, 1758) AND *TETRAMORIUM SEMILAEVE* ANDRÉ, 1881 (HYM., FORMICIDAE)

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Comparative morphological study of Tetramorium caespitum *(Linné, 1758) and* Tetramorium semilaeve *André, 1881 (Hym., Formicidae).*– The discrimination between *T. caespitum* and *T. semilaeve* is one of the many taxonomical problems within the genus *Tetramorium* in the Iberian peninsula. The aim of this work is to support the results obtained in previous studies on this genus, trying to make them to some extent objective and to offer new criteria of discrimination. The morphology of both species was quantified at individual and colonial levels. Correspondence Analysis and Stepwise Discriminant Analysis were applied to the data. The segregation results for the latter improve to perfect discrimination when using average data from colony samples.

Key words: *Tetramorium caespitum, Tetramorium semilaeve*, Morphometrics, Taxonomy, Iberian peninsula.

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INTRODUCTION

There exist many problems related to the taxonomy of the genus *Tetramorium* Mayr, 1855 in the Iberian peninsula, the majority of them basically associated with the identification of the worker caste. Some studies have been carried out on this question: first, a general structure of the genus into three species - groups was outlined (López, 1991a); and then, two of the more problematic taxa (*T. caespitum* and *T. semilaeve*) were compared by morphological criteria (López, 1991b); or by means of their distribution trends (López et al., 1990).

In spite of the acceptable degree of differentiation reached using the usual qualitative procedure (i.e., the pure observation without measurements), and due to the large variability spectrum in the morphology of the worker caste of *T. caespitum* and *T. semilaeve*, there are cases in which the application of the discrimination characters requires a considerable previous training, studying large numbers of specimens. This is not possible for an observer who wants to identify a single sample. In addition, quantification allows the statements to be verified by a different, and probably more objective, method.

Ants, like most social organisms, are a special kind of beings that show the property of being found in groups with the certainty of belonging to the same species. Several phenomena that take place in ants, as hybridization, social parasitism, slavery, xenobiosis and parabiosis (HOLLDOBLER & WILSON, 1990), are possible sources of confusion but, nonetheless, they do not alter the rule, specially if one knows whether or not the species (or the colonies) under study are affected by any of these phenomena, which is almost always detectable. This property was used in order to reach an absolute discrimination between the species studied in this work.

In this study, the morphological differences between the worker castes of *T. caespitum* and *T. semilaeve,* are analyzed quantitatively in order to provide criteria for the taxonomical discrimination of both species.

MATERIAL AND METHODS

Because of the necessity of including geographical variation and the virtual lack of polymorphism at the colony level, few individuals from many colonies were used, (as some authors have done like BRIAN & BRIAN, 1949 and ELMES & CLARKE, 1981, for example) rather than many individuals from one or few colonies (WILSON,1953; HIGASHI,1974; BARONI URBANI & KUTTER, 1979; ESPADALER & RIASOL, 1983; ACOSTA et al., 1986; among others). Information about a larger number of colonies offers the possibility of carrying out analyses considering each one of them as a unit (superorganism), which is a very interesting approximation, exclusive to the social organisms.

The series of specimens used for this study are listed in table 1. Each series (or sample) is composed by individuals (workers) belonging to the same colony. The collections of which the studied material comes from are: DBG. Collection of the Departamento de Biología Animal, Ecología y Genética of the Facultad de Ciencias of the Universidad de Granada (Dr. J.A. Tinaut); XE. Collection of Dr. X. Espadaler; FL. Collection of F. López.

The morphological measurements made in a fraction of the total population were (fig. 1): CEW. Cephalic width (×100); SCL. Scape length (×100); MTW. Maximum thorax width (×100); MTL. Maximum thorax length (×100); ESL. Epinotal spines length (×200); PEW. petiole width (×200); PEL. Petiole node length (×200) and PPW. Postpetiole width (×200). Numbers indicate the magnification used to make each measurement. The choice of these characters was based on previous works (BRIAN & BRIAN, 1949; BERNARD, 1956; BUSCHINGER, 1966; GRAY, 1973; BOLTON, 1976, 1977, 1979, 1980; among others), and on observations accumulated during the qualitative study of the specimens from the collections.

Data were analyzed by means of Correspondence Analysis (CA) (BENZECRI, 1973a, 1973b) and Stepwise Discriminant Analysis (SDA) from the BMDP7M statistical package (JENNRICH & SAMPSON, 1983).

RESULTS AND DISCUSSION

Despite having series of specimens coming from different geographical regions of the Iberian peninsula, this work does not attempt to study geographical variability within each species because there is not a clear *a priori* criterium to select different geographical units (areas) to compare the levels of variability once seen the variation showed by each one of the species throughout the whole territory comprised in this study (LÓPEZ, 1991a, 1991b). A similar study attemped by ELMES & CLARKE (1981), though having a clear categorization of areas by countries, failed to find geographical trends and to explain clearly the results obtained when comparing the morphological variability of a single ant species. On the other hand, the

Table 1. Series of specimens used in the present study. Tc. *T. caespitum*; Ts. *T. semilaeve*. (See text for collection acronyms).

Series de ejemplares empleados en el presente estudio. Tc. T. caespitum; *Ts.* T. semilaeve. *(Ver texto para siglas de las colecciones).*

No.	Province	Locality	Code	Altitude (m)	UTM	Collection	Specimens
Spain							
1	Alicante	Playa de Urbanova (Alicante)	AC.Tc	0	30SYH14	FL	25
2	Almería	Almería	AL.Tc	0	30SWF47	FL	25
3		Sierra Filabres	AL.Tc2	1600-2100	30SWG32	DBG	25
4			AL.Tc3	1600-2100	30SWG32	DBG	25
5	Ávila	Gavilanes	GA.Tc	800	30 TUK46	FL	25
6		Piedralaves	PI.Tc	800	30TUK56	FL	25
7	Cádiz	Faro del Carnero (Algeciras)	AG.Ts	0	30STE89	DBG	25
8		Sierra Grazalema	GR.Ts1	900-1600	30STF87	DBG	25
9	Ciudad Real	Tablas de Daimiel	DA.Tc	600	30SVJ33	DBG	20
10			DA.Ts	600	30SVJ33	DBG	25
11	Granada	Nechite	NE.Tc	1700	30SVG90	DBG	25
12			NE.Tc2	1700	30SVG90	DBG	25
13		Orjiva-Pampaneira	OR.Tc	1000	30SVF68	DBG	25
14		Prados de Otero (Sierra Nevada)	OT.Tc	2200	30SVG50	DBG	19
15		Sierra Baza	BA.Tc	1600-2000	30SWG02	DBG	25
16			BA.Tc2	1600-2000	30SWG02	DBG	25
17			BA.Tc3	1600-2000	30SWG02	DBG	25
18		Trevélez	TR.Tc	1500	30SVF79	FL	25
19	Guadalajara	Retiendas	RE.Tc	800	30TVL73	FL	25
20	Huesca	Parque de Ordesa	OR.Tc1	2000	30TYN42	DBG	18
21		Sariñena	SA.Tc	200	30TYM33	XE	25
22			SÑ.Ts	200	30TYM33	XE	25
23		Valle de Hecho	HE.Tc	800	30TXN83	DBG	25
24	Jaén	Andújar	AN.Ts1	300	30SVH01	FL	25
25			AN.Ts2	300	30SVH01	FL	25
26		Llanos de Hernán Perea (Sierra de Cazorla)	CZ.Tc	1800	30SWG09	DBG	19
27		Pico Cabañas (Sierra de Cazorla)	PC.Tc	2000	30SWG08	XE	25
28		Prado Llano (Sierra de Cazorla)	PR.Tc	1800	30SWG08	XE	25
29	León	Riaño	RI.Tc	1200	30TUN36	FL	19
30	Madrid	Aranjuez	A.Ts	500	30TVK43	FL	25
31			A.Ts2	500	30TVK43	FL	25
32		El Escorial	ES.Ts	1100	30TVK09	FL	20
33			ES.Ts2	1100	30TVK09	FL	25
34		Guadalix de la Sierra	G.Ts	800	30TVL41	FL	25
35		Hoyo de Manzanares	H.Tc	1000	30TVL29	FL	23
36			H.Ts	1000	30TVL29	FL	23
37		Madrid	M.Tc	700	30TVK47	FL	25
38			M.Ts1	700	30TVK47	FL	25
39			M.Ts2	700	30TVK47	FL	25
40			M.Ts3	700	30TVK47	FL	25
41		Pedrezuela	PE.Ts1	800	30TVL41	FL	25
42			PE.Ts2	800	30TVL41	FL	35
43	Málaga	Boquete Zafarraya	MA.Ts1	1200	30SVF09	DBG	18
44		Sierra de las Nieves	MA.Tc	1700	30SUF26	DBG	25
45	Oviedo	Lago Enol	EN.Tc	1000	30TUN39	DBG	22
46	Palencia	Cardaño de Arriba	CA.Tc1	1400	30TUN55	FL	25
47			CA.Tc2	1400	30TUN55	FL	18
48		Monte de Tabanera	TA.Tc	800	30TVM05	XE	25
49	Salamanca	Ledesma	LE.Tc	600	29TQF55	XE	17
50	Santander	Fuente Dé	FU.Tc	1800	30TUN57	FL	25
51	Teruel	Griegos	GR.Tc	1600	30TXK07	FL	25
52	Toledo	La Iglesuela	IG.Tc	400	30TUK55	FL	25
53	Valladolid	Valdunquillo	VA.Tc	600	30TUM05	XE	25
Portugal							
54	Algarve	Alvor	AV.Ts1	0	29SNB30	FL	25
55			AV.Ts2	0	29SNB30	FL	25
56		Hortas Do Tabual	HR.Ts	0	29SNB10	FL	25
57	Baixo Alenteço	Sines	SI.Tc	0	29SNC10	FL	25
France							
58	Languedoc-Roussillon	Banyuls-Sur-Mer	BA.Ts	0	31TEH10	XE	25

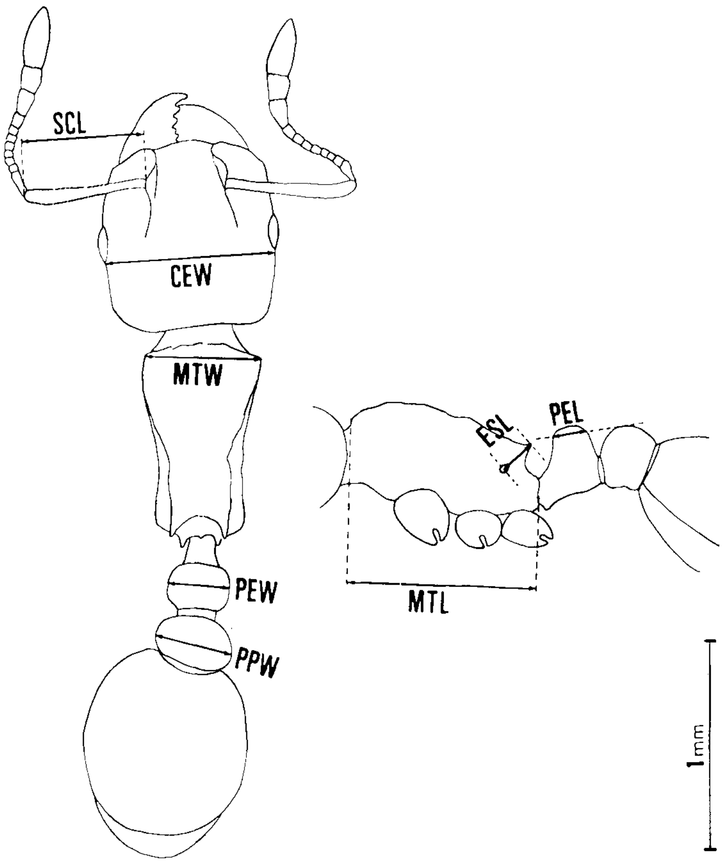

Fig. 1. Measurements made in the workers of *T. caespitum* and *T. semilaeve* (see the text for abbreviations).

Medidas efectuadas en las obreras de T. caespitum *y* T. semilaeve *(ver el texto para abreviaturas).*

goal of this work was rather to use the variability comprised in the different samples taken to compare the two species under study.

Following operational principles, the main objective was to separate both species (*T. caespitum* and *T. semilaeve*) as clearly as possible with the minimum number of parameters. To begin the process of selection, a sub-sample of 50 specimens (25 of each species) was taken from the series, trying to include the maximum intercolonial variability observed.

A multidimensional ordination like the CA produces a grouping taking into account simultaneously all the quantified variables which can be compared with the one produced by qualitative criteria, which is to some extent a way of checking its subjectivity. The first three axes of the CA (fig. 2) absorb 74.1% of the variance and offer a neat separation of both groups, coinciding almost perfectly with the *a priori* classification. In detail, axis I and III are the best separators with a few "displaced" individuals. Axis II shows more deficiencies in its discriminant power, because the individuals of both species are intermingled along this axis. The three variables associated with these axis are PEL (I), ESL (II) and PPW (III).

Once seen the results of the confrontation of the two discrimination methods, a SDA was performed with the same 50 individuals to obtain a combination of variables, selected from the ones initially measured. PEL, PPW and CEW were selected by the analysis, allowing a perfect separation of *T. caespitum* and *T. semilaeve*. It is interesting to note that the first two variables also directed to a great extent the ordination of the individuals in the CA.

With the objective of obtaining a discriminant function from a large quantity of individuals, and to test the differentiation power of the three selected parameters for such a large range of variation, a new SDA was performed on the total of specimens of both species (1385) using only these three variables. The discriminant equation obtained was: y = – 3.517 – 26.686 CEW + 79.265 PEL + 40.750 PPW; and the limits of the variation intervals (obtained taking the individuals with the extreme coordinates in the analysis) were: – 2.139 to 6.114 for *T. caespitum*, and – 2.665 to 0.260 for *T. semilaeve*. The results on discriminant power are shown in table 2 and figure 3A. The certainty of separation is very near to 100%, although there exists a little zone of overlap-

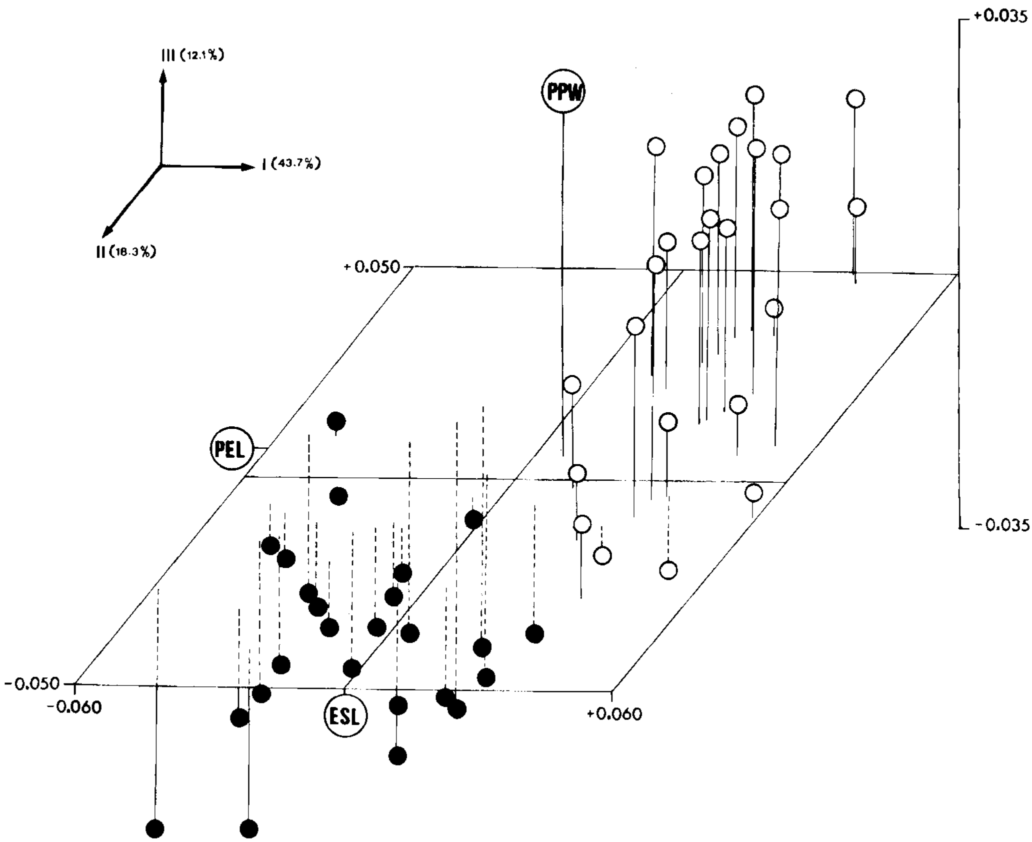

Fig. 2. Representation of the three first axis (I, II and III) of the Correspondence Analysis performed with the morphometrical parameters under study; percentages indicate the absorbed variance by each axis. ● *T. caespitum*; ○ *T. semilaeve*.

Representación de los tres primeros ejes (I, II y III) del Análisis de Correspondencias realizado con los parámetros morfométricos estudiados; los porcentajes indican la varianza absorbida por cada eje. ● T. caespitum; ○ T. semilaeve.

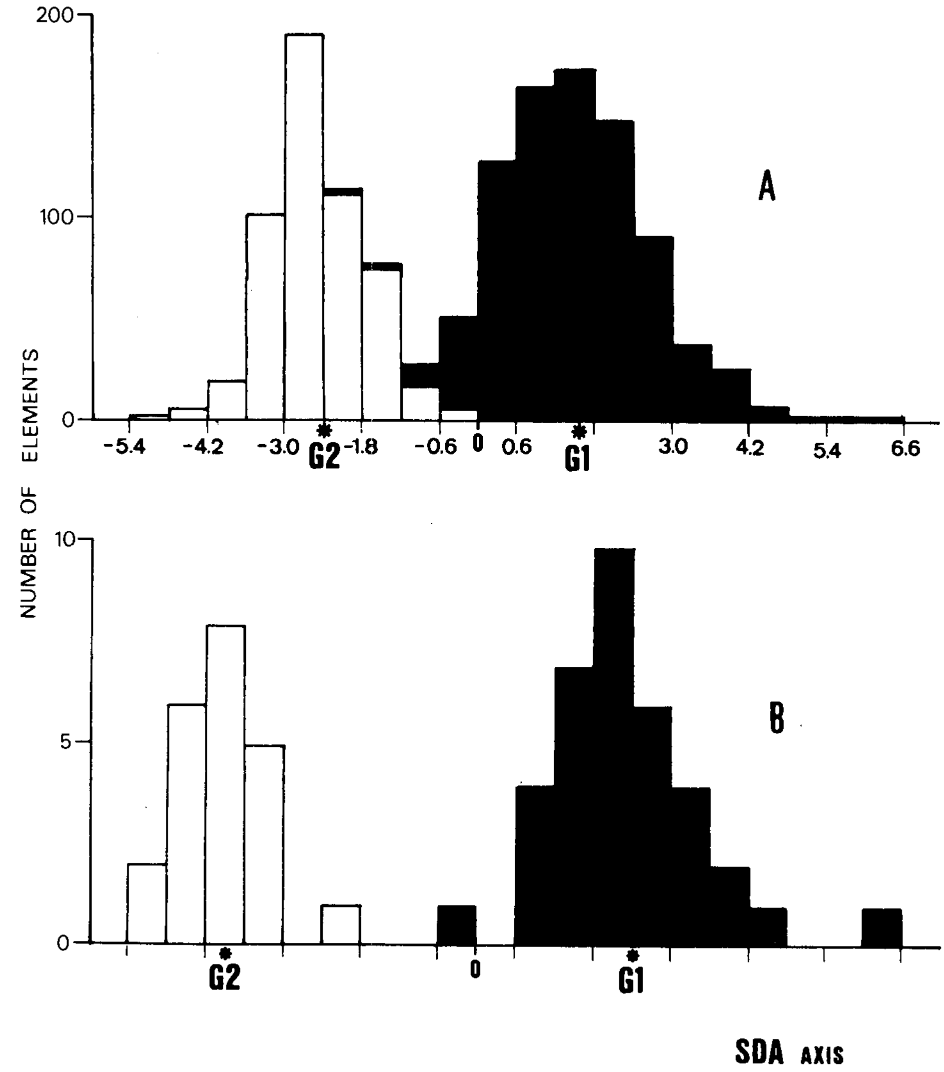

Fig. 3. Histograms of frequency of elements over the coordinates axis of the Stepwise Discriminant Analysis for the isolated individuals (A) and for the series (B). G1. Gravity center of group 1 (*T. caespitum*); G2. Idem of group 2 (*T. semilaeve*); ■ *T. caespitum*; □ *T. semilaeve*.

*Histogramas de frecuencia de elementos sobre el eje de coordenadas del Análisis Discriminante Paso a Paso para los individuos aislados (A) y para las series (B). G1. Centro de gravedad del grupo 1 (*T. caespitum*); G2. Idem del grupo 2 (*T. semilaeve*);* ■ T. caespitum; □ T. semilaeve.

ping between both groups, due to the increase of variation. This variability is greater in the case of *T. caespitum*, as can be seen in all the results of the SDA. This fact was also evident in the qualitative study, and is reflected in other way by the frequency distributions of the three variables measured in all specimens (fig. 4).

It is very interesting to note that the extreme individuals in the analysis (which are

Table 2. Results on discriminant power of the Stepwise Discriminant Analysis performed over the isolated individuals (I.I.) and over the series (S.) using the average value. (For other abbreviations see table 1).

Resultados sobre el poder discriminante del Análisis Discriminante Paso a Paso realizado sobre los individuos aislados (I. I.) y sobre las series (S.) utilizando el valor medio. (Para otras abreviaturas, ver tabla 1).

	% Well classified	Total number of elements
	I.I./S.	I.I./S.
Tc	96.7/100	849/36
Ts	99.1/100	536/22
Total	97.6/100	1386/58

neither the biggest nor the smallest ones) are extreme for the combination of variables selected but not necessarily for the other characteristics. In fact, these workers can be differentiated by means of qualitative criteria without problems. And the opposite situation is also present: those "problematic" specimens before the quantification become easy to separate because they are placed within the corresponding group in inner zones of the variation range of the coordinates.

When a species is morphologically studied, an ideal abstraction of it, that could be

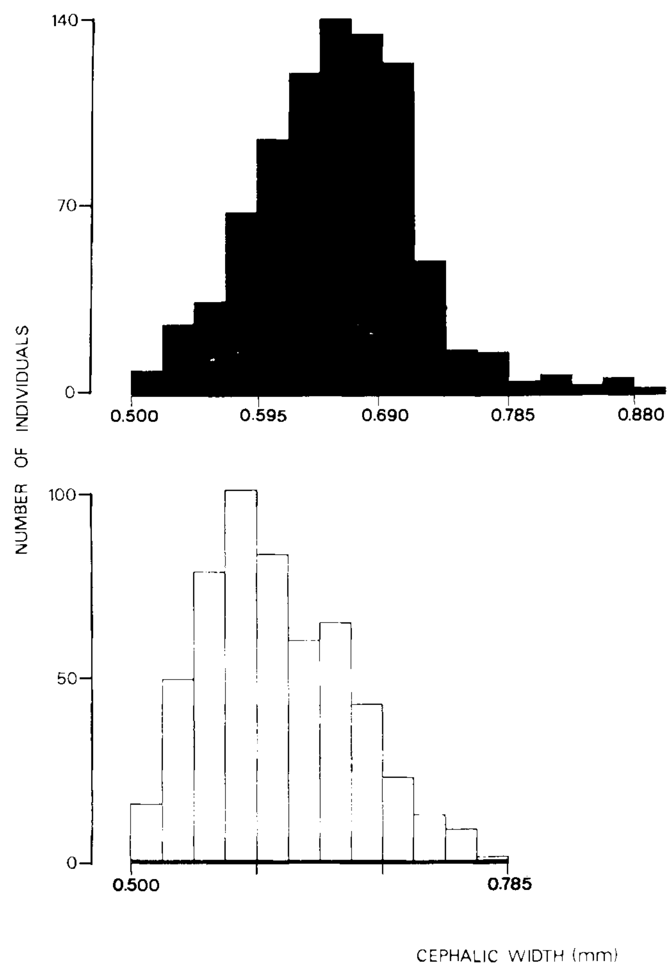

Figure 4. Frequency distributions in millimeters for the cephalic width of *T. caespitum* (■) and *T. semilaeve* (□).

Distribuciones de frecuencias en milímetros para la anchura cefálica de T. caespitum *(■) y* T. semilaeve *(□).*

subjective, arises from the integration of the specimens observed, the degree of abstraction varying with the amount of variability exhibited by the individuals under study (see a similar comment on the morphological concept of species in ELMES & CLARKE, 1981). The abstraction can be quantified, and the social organisms allow to do this in the special and exclusive level of the superorganism. There is discussion about the value of this concept based on its supposed uselessness (WILSON, 1971) but, using an analogy with system dynamics, the absence of knowledge about the emergence properties of the system (BERTALANFFY, 1968) does not invalidate the concept. In fact, recent research on colony self-organization (PASTEELS et al., 1987; DENEUBOURG et al., in press; GOSS & DENEUBOURG, in press a, in press b; for example) can be viewed as uses of it with a contribution to that knowledge. In the present case, the concept is used for practical purposes in two different ways: as a confirmation of the ownership of the individuals to a specific category (as was explained above) and as a successful level of discrimination between the categories, as it is going to be seen now.

Taking the average of each of the three variables for each series (which represents a colony) a new SDA was performed. Using the averages this way, superindividuals are created, whose characteristics are the result of the interactions of all the individuals that integer them. This is simply an application of the central limit theorem for a favorable case (in the sense of the known assignment to one category or another of a group of individuals). The discriminant equation obtained was: $y = -1.468 - 54.557\ CEW + 107.671\ PEL + 86.998\ PPW$; and the limits of the variation intervals were: -0.152 to 6.127 for *T. caespitum*, and -4.906 to -1.965 for *T. semilaeve*. The results on discriminant power are shown in table 2 and figure 3B. The certainty in the classification is total, without overlap, which to some extent indicates a proper entity of this organization level that allows each colony to be characteristic of one group or another in spite of containing non-characteristic individuals. This fact is almost self evident given that there really are two distinct populations, but it is shown here to see the degree of improvement and discriminant equation obtained when using colony means.

This procedure (very simple, on the other hand) is only one of a kind of possible grouping analysis that could be made using the colony level. The size of the group of individuals that embrace the morphological colony definition (with the possible associated questions of cellularity), its relation with the colony size (BRIAN & BRIAN, 1951), the inclusion of the reproductive forms in relation with it, the development of the colony morphology, the relation of the latter with the individual morphology, etc., could be other approaches. Its usefulness for other applications has yet to be proved but it seems reasonable to expect good results.

Finally, from what has been shown above, some recommendations can be made on the taxonomic differentiation between *T. caespitum* and *T. semilaeve*. The sexual forms of both species are clearly differentiated (EMERY, 1925; KUTTER, 1977; TINAUT, 1981) but in most cases require a lot of additional work for their capture. It is also better to use a large sample of workers but this is not always possible, so it seems clear that a procedure combining qualitative and quantitative criteria for the identification of *T. caespitum* and *T. semilaeve* offers an almost total security in the decision to be taken. It is important to emphasize that it is not necessary to have a large series of specimens to apply the discriminant equation, since, even with a single individual, the assumed certainty in the clasification is near to the 100%

(as shown in table 2). Moreover, it is also essential to take into account the fact that the studied specimens were not always found to be simultaneously "problematic" for qualitative and quantitative criteria and, therefore, their discrimination becomes easier if both criteria are considered. Although subjectivity is a problem of difficult resolution in taxonomy, the contribution with added criteria for differentiation could contribute to a less exclusive use of this tool for those not interested in the object-studying but in the object-using.

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SUMMARY

Estudio morfométrico comparativo de Tetramarium caespitum *(Linné, 1758) y* Tetramorium semilaeve *André, 1881 (Hym., Formicidae).*

Se estudia la casta obrera de *Tetramorium caespitum* y *Tetramorium semilaeve* (fig. 1, tabla 1), con el fin de ofrecer nuevos criterios de diferenciación taxonómica entre ambas especies. Los datos fueron analizados por medio de técnicas de ordenación (fig. 2) y clasificación (fig. 3) multivariantes. El poder discriminador de estos análisis mejoró al emplear valores medios coloniales de las medidas tomadas (tabla 2), lo que sugiere muchas posibilidades en relación con una posible "morfología colonial" como unidad de estudio, mucho más definitoria que la individual.

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