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Studies in *Ophrys* L. sectio *Pseudophrys* Godfery - I. *Ophrys forestieri* and *O. malacitana* spec. nov.

Keywords

Orchidaceae; Sectio *Pseudophrys*, *Ophrys fusca*, *Ophrys forestieri*, *Ophrys lupercalis*, *Ophrys malacitana*, *Colletes cunicularius*, *Andrena nigroaenea*, Flora of Spain, Flora of Portugal, Flora of France, Flora of Majorca, biometrics; taxonomy, distribution.

Summary

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In the first of a planned series of publications, biometric analysis is applied to two contentious issues within *Ophrys* sectio *Pseudophrys*. The affinities of *Ophrys fusca* s.s. from around Lisbon, Portugal to the *Andrena nigroaenea* pollinated *O. lupercalis* (*O. 'nigroaenea-fusca'* sensu Paulus) and to the *Colletes cunicularius* pollinated plants from the Province of Málaga in the south of Spain, known until now only by the working name of *O. 'colletes-fusca'*, have long been disputed. Biometric analysis of the three entities indicates that there are three species. The plants from Málaga are described as *Ophrys malacitana*. Secondly, following the designation of a lectotype for the name *O. forestieri* from the original material of Forestier gathered at L'Escaladieu in the French Pyrenees, a biometric analysis of plants from this region with the plants known as *O. lupercalis* from the south of France and eastern Spain concludes that they are one species. The name *O. forestieri* has priority and the name *O. lupercalis* becomes a synonym. Furthermore, disjunct populations in northern Spain show floral biometric differences, which may indicate pollinator shift, whilst the population on Majorca is very similar to that on the mainland of Spain and in southern France.

Zusammenfassung

Lowe, M. R. (2010): Untersuchungen an *Ophrys* L. sectio *Pseudophrys* Godfery - I. *Ophrys forestieri* und *O. malacitana* spec. nov.- J. Eur. Orch. 42(3/4): 541-562.

Die Ergebnisse der Bearbeitung der Sektion *Ophrys* sectio *Pseudophrys* soll in einer Reihe von Beiträgen mitgeteilt werden, hier wird über die Anwendung biometrischer Analysemethoden bei zwei bisher umstrittenen Fällen innerhalb dieser Sektion berichtet. Erstens werden *Ophrys fusca* s.s. aus der Umgebung von Lissabon (Portugal), die von *Andrena nigroaenea* bestäubte *O. lupercalis* (*O. 'nigroaenea-fusca'* sensu Paulus) und die von *Colletes cunicularius* bestäubten Pflanzen aus der Provinz Málaga in Süds Spanien (bisher nur unter dem Arbeitsnamen *O. 'colletes-fusca'* bekannt), deren verwandtschaftliche Nähe seit längerem kontrovers diskutiert wurde, vergleichend untersucht. Die mit diesen drei Sippen durchgeführten biometrischen Analysen weisen deutlich auf drei eigenständige Arten hin, die Pflanzen aus Málaga werden deshalb als *Ophrys malacitana* neu beschrieben. Zweitens wird *O. forestieri* anhand von Material aus L'Escaladieu (französische Pyrenäen, Terra typica entsprechend der Lectotypisierung des Namens auf von Forestier dort gesammeltem Originalmaterial) mit den unter *O. lupercalis* bekannten Pflanzen aus Südfrankreich und Ostspanien verglichen. Die biometrischen Analysen belegen deren konspezifische Identität. Da der Name *O. forestieri* Priorität besitzt, ist der Name *O. lupercalis* in dessen Synonymie zu stellen. Die disjunkten Teilpopulationen in Nordspanien zeigen Abweichungen der Blütenmaße, die auf einen Wechsel des Bestäubers hinweisen könnten. Die Teilpopulation von Mallorca hingegen kommt denen vom spanischen Festland und Südfrankreich sehr nahe.

* * *

Introduction

This paper is the first in a series that employs biometric analysis to the variation and taxonomic structure of *Ophrys* L. sectio *Pseudophrys* Godfery in a Mediterranean wide study. The results are intended to assist other methods of investigation and establish a provisional framework and working taxonomic model.

The *Ophrys fusca* - *O. lupercalis* controversy

Ophrys lupercalis J. & P. Devillers-Terschuren was published (DEVILLERS & DEVILLERS-TERSCHUREN 1994) as the name for the *Andrena nigroaenea* pollinated component of the *O. fusca* group known from the south of France, and eastern Spain, and distinct from *O. fusca* Link, later defined by the designation of a neotype (DELFORGE 1999: 188). PAULUS (2001: 137) disputed this distinction, considering the typical *O. fusca* from Lisbon, Portugal and *O. lupercalis* to be the same species. DELFORGE (1994, 1996) took the view that *O. fusca* s.s. is identical to the component of the *O. fusca* group pollinated by *Colletes cunicularius* (known by the working name *O. 'colletes-fusca'* discovered in Málaga, Spain (PAULUS & GACK, 1980, 1981 & 1983), whilst PAULUS (2001: 137) indicated a distinction between his *O. 'colletes-fusca'* and the typical *O. fusca* from Lisbon. However, hitherto no formal name has been given to *O. 'colletes-fusca'* in support of the view that it is a distinct species with its own pollinator, in accordance with the biological species concept which will be used throughout this series of papers.

Ophrys forestieri vs *O. lupercalis*

In a remarkable piece of detective work, DEVILLERS & DEVILLERS-TERSCHUREN (2006) identified the locality from which the type specimen of *Ophrys forestieri* (Rchb.f.) Lojac. was collected as being in the French département Hautes-Pyrénées. Based upon an assessment of various characters they considered the plant to be a species within the *O. obaesa* group and distinct from *O. lupercalis*, *O. vasconica* and *O. sulcata*. In my view the intra-specific variation of the form of the speculum and labellum patterns of species within the *O. fusca* group requires a cautious approach to species delimitation on such a basis. Indeed, the photographs used to illustrate *O. forestieri* from Bellegarde, Gers indicate a variation that is not atypical of that found more widely in southern France and eastern Spain within populations of *O. lupercalis*. Further, to my mind, the lectotype selected for *O. forestieri* and the other material collected by de Forestier from the abbey of L'Escaladieu are consistent with *O. lupercalis*.

Methods

Biometric analysis was conducted on material gathered from France, Spain and Portugal to examine these issues. Sites were selected to include the area of the type localities of *O. fusca*, *O. lupercalis*, *O. forestieri* and the Málaga

O. 'colletes-fusca' and localities from which pollinators have been identified. Summary data is provided in Table 1 and site locations are given at Table 2.

Twelve continuous dimensional floral characters were selected to examine variation as shown in Fig. 1. As the objective was to determine variation subject to genetic control and, so far as practical, to avoid environmental influences, vegetative characters were not used. A preliminary investigation indicated that floral characters are stable provided the sampling technique is subject to carefully controlled selection criteria, whilst vegetative characters are variable reflecting the nutritional state of the plant. Only fully expanded flowers were selected from vigorous plants. From within the middle of the spike a flower was selected above at least one fully expanded flower, whilst ensuring that the flower had not started to desiccate on the extremities. Flowers with any mutant characters, such as labelloidy, or subject to damage from herbivores were excluded from the sampling.

Excised labella and perianth segments were pressed after mounting with adhesive clear tape prior to measuring under 10X magnification ($\pm 0,1$ mm). Care was taken to unroll the edges of the outer perianth segments before mounting. This method ensures that shrinkage in the drying process is negligible. The labella were excised at the join with the stigmatic cavity. A colony sample of between 5 and 12 flowers, from within a group of at least 30 individual plants, was collected over a distance of less than 50 meters. A second sample was collected only from very numerous and extensive colonies, to ensure minimal conservation impact.

Univariate and multivariate statistical analysis and graphic presentations were conducted with the computer programmes Excel (Microsoft Corporation) and XLSTAT v2008.1.02 (Addinsoft Ltd.). Discriminant Analysis (DA), also known as Canonical Discriminant Function Analysis, was performed stepwise, (backward) and within-class covariance matrices were assumed to be equal. All analyses indicated multicollinearity to be within acceptable tolerance levels. Output figures of Discriminant Analysis indicate the centroids and 95% confidence limits. Multiple Dimensional Scaling was used to compress multidimensional data into two dimensions. Agglomerative Hierarchical Clustering was performed using the complete linkage method.

Biometric analysis and discussion

Figures 2 & 3 compare the dissimilarity of floral biometric characters of 57 colonies (506 individual plants) of what previously have been regarded as *O. lupercalis* from southern France and eastern Spain, including the locality from which the type of *O. lupercalis* is based from La Clape and localities on the northern fringes of the Pyrenees for *O. forestieri*. Figure 2 illustrates the Mahalanobis Distances between each colony compressed into 2 dimensions by Multiple Dimensional Scaling. Figure 3 illustrates the same data expressed with Agglomerative Hierarchical Clustering. The colonies from Sub-Pyrenean Spain and Cantabria are strongly clustered, but not totally separated from the remaining colonies. The colony samples from near the type locality of *O. lupercalis* and from the region of the lectotype for *O. forestieri* are indicated in bold type. The results indicate that *O. forestieri* and *O. lupercalis* are part of a single cline of variation leading to the inevitable conclusion that they are the same species, the name *O. forestieri* having priority and rendering the name *O. lupercalis* as a synonym. Taking a journey from the coastal type locality of *O. lupercalis* at La Clape through the département of Pyrénées-Orientales (Rivesaltes, Salses, Pas de l'Escafe, Vingrau, Estagell, St. Paul-de-Fenouillet), from which *A. nigroaenea* has been determined as the pollinator, to the localities at Bugarach, Aude and along the French Pyrenees to the region of *O. forestieri* at Masseube and Puymaurin there are no discernible differences in biometric floral characters or in any other observed characters.

A comparison using Discriminant Analysis of five groups from; France (A1), eastern Spain (A2), Majorca (B), the Sub-Pyrenean area of northern Spain and the Basque Country (C) and, coastal dunes and sandy habitats from Cantabria and near Bilbao (D) is shown in Fig. 4. Further analyses, maximising the discriminatory differences between the sub-populations, are shown in Figs. 5 and 6. Figure 5 indicates that the sub-populations from France, eastern Spain (mainland) and Majorca are very similar, indicative of a minor degree of genetic drift. However, Fig. 6 indicates that the sub-populations C & D from northern Spain, which have disjunct distributions, show a more substantial degree of separation from the main Spanish sub-population (A2). The difference between the Spanish Sub-Pyrenean and Cantabrian populations is notable given the similarity between the populations from eastern Spain and Majorca.

A map of the distribution of the revised delimitation of *O. forestieri* is provided in Fig. 7. The pollinator is the first generation of *A. nigroaenea* as determined by ARNOLD (1981: 14, 1999: 122) from Catalonia, Spain and PAULUS & GACK (1999: 357) from the south of France. Coastal colonies

commence flowering in January, (very occasionally in late December) through February with a peak in March and have usually faded by early April. Inland colonies, especially in shady localities, flower several weeks later, all such dates being variable due to seasonal fluctuations. The populations from the Sub-Pyrenean area of northern Spain peak in late March and early April, whilst those from the Cantabrian dunes peak in mid March. No information is available to identify the pollinator for these Spanish Sub-Pyrenean and Cantabrian populations and, given the similarity of phenology and labellum dimensions, *A. nigroaenea* would appear to be the most likely candidate. Nonetheless, the possibility exists that pollinator shift has led to the formation of new species in these areas; a hypothesis that remains to be explored.

Discriminant Analysis of 68 colonies (596 individuals) from three populations: *O. fusca* from the area around Lisbon, including near the neotype locality; *O. forestieri* from Eastern Spain and France (samples A1 & A2) and; the *O. 'colletes-fusca'* from Málaga and neighbouring provinces is shown in Fig. 8. Figure 9 uses the same data standardised to the labellum length. Figure 10 shows the labellum characters alone, standardised to labellum length and Fig. 11 shows the perianth characters, standardised to the dorsal sepal length. The use of standardised data removes any discrimination by size. The differences between Figs. 8 & 9 indicate greater discrimination when size and shape are used compared to shape alone.

The results clearly demonstrate the existence of three taxa, with the *O. 'colletes-fusca'* and *O. forestieri* (pollinated by *A. nigroaenea*), being the most similar. The similarity of the perianth segment characters, which are not expected to be of any selective value in pollinator driven evolution, indicates that the *O. 'colletes-fusca'* and *O. forestieri* are likely to be closely related. The pollinator of *O. fusca* remains unknown (PAULUS 2001: 135). Its flower size and dark maroon colour show similarities with *O. iricolor* and the two species have more or less complementary distributions in the east and west Mediterranean. This raises the possibility, though speculative, that *A. morio* is the pollinator.

Ophrys 'colletes-fusca'

On the basis of the clear separation of the *O. 'colletes-fusca'* from *O. fusca* and *O. forestieri* and following extensive study of the distribution and phenology the well established plant known by the working name of *O. 'colletes-fusca'* is described below as a new species. The joint authorship of the name recognises the extensive assistance I have received from Ian Phillips in the study of

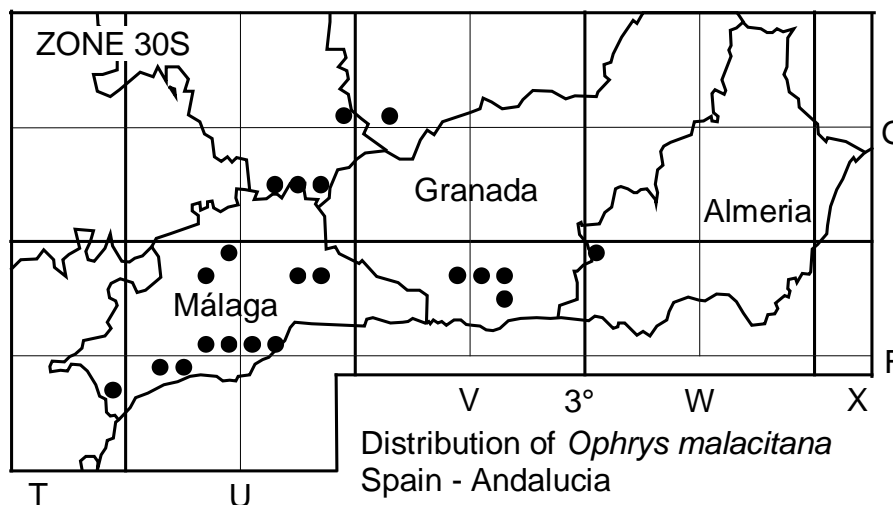
orchids in Málaga and the role of Hannes Paulus in discovering the pollinator. The epithet refers to the city of Málaga. Photographs are shown at Figs. 12-17 and are illustrated by PAULUS (2001: 175 in Abb. 6, 7 and 8).

***Ophrys malacitana* M. R. Lowe, I. Phillips & Paulus spec. nov.**

Planta a *Ophryde fuscae* et *O. forestieri* affinis sed characteribus floralibus eis manifeste distincta. Labellum 14,2-16,0 x 12,0-13,6 mm, colore atrobadio vel porphyreo, margine angusto glabroque interdum luteo, trilobatum, lobis lateralibus exterioribus 9,6-11,1 mm longis; speculum plerumque plumbeo-lazulinum, metallicum. Segmentum exterius dorsale perianthii 10,3-12,1 x 7,0-8,0 mm, segmenta exteriora lateralia 11,7-13,3 x 6,8-7,8 mm. Segmenta interiora perianthii 7,9-9,0 x 1,9-2,2 mm. Floret (Decembri) Januario-Martio. Pollinatio a *Collete cuniculario*. Habitat inter frutices herbosisque in stationes subrunderatis, finibus pinetium, fruticetibus rimosis et juxta sterilibus umbrosis, semper in solo calcareo.

Holotypus: Hs, Spain: Malaga, Sierra Mijas, Pichón de Jarapalo, 30SUF5656, ad 240 m.s.m., ubi 20.1.2002, M.R. Lowe & I. Phillips legit (asservatus in herbario ABH no. 51253).

Plant with affinities to *Ophrys fusca* and *O. forestieri*. Labellum 14,2-16,0 x 12,0-13,6 mm, colour dark reddish brown with narrow glabrous margin, occasionally yellow, outer lobes 9,6-11,1 mm long. Speculum usually metallic bluish grey. Dorsal outer perianth segment 10,3-12,1 x 7,0-8,0 mm. Lateral outer perianth segments 11,7-13,3 x 6,8-7,8 mm. Inner perianth segment 7,9-9,0 x 1,9-2,2 mm. Flowers (December) January-March. Pollinated by *Colletes cunicularius*. Grows in marginal habitats, edges of pine woods, garigues and shady bank sides on calcareous soils.



Descriptive Statistics

N = 166 mm	Mean \bar{x}	1st quartile	3rd quartile	Standard deviation σ	SD Mean
Labellum L (1)	15,1	14,2	16,0	1,26	0,10
Labellum W (2)	12,7	12,0	13,6	1,08	0,08
Outer lobe L (3)	10,4	9,6	11,1	1,10	0,09
Speculum L (4)	9,4	8,9	10,0	0,85	0,07
Mid lobe W max (5)	7,6	7,0	8,2	0,93	0,07
Mid lobe W min (6)	5,8	5,2	6,1	0,65	0,05
Dorsal OPS L (7)	11,3	10,3	12,1	1,14	0,09
Dorsal OPS W (8)	7,5	7,0	8,0	0,86	0,07
Lateral OPS L (9)	12,6	11,7	13,3	1,26	0,10
Lateral OPS W (10)	7,2	6,8	7,8	0,79	0,06
IPS L (11)	8,5	7,9	9,0	0,88	0,07
IPS W (12)	2,1	1,9	2,2	0,31	0,02

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Bibliography

- ARNOLD, J. E. (1981): Notas para una revisión del género *Ophrys* L. (*Orchidaceae*) en Cataluña.- *Collectanea Botanica* (Barcelona) **12**: 5-61.
- ARNOLD, J. E. (1999): La problématique des groupes d'*Ophrys fusca* et d'*Ophrys omegaifera* en Catalogne et dans le Pays Valencien (Espagne).- *Natural. belges* **80** (orchid 12): 120-140, 275.
- DELFORGE, P. (1994): *Guide des Orchidées d'Europe, d'Afrique du Nord et du Proche-Orient.*- Lausanne et Paris.
- DELFORGE, P. (1999): Contribution à la stabilisation de la nomenclature dans le groupe d'*Ophrys fusca*: désignation d'un néotype pour *Ophrys fusca* LINK in SCHRADER 1800, *Ophrys funerea* VIVIANI 1824, *Ophrys bilunulata*

- RISSO 1844 et *Ophrys forestieri* (REICHENBACH fil. 1851) LOJACONO 1909.- Natural. belges **80** (orchid 12): 179-229.
- DELFORGE, P. (2006): Orchids of Europe, North Africa and the Middle East.- London.
- DEVILLERS, P. & J. DEVILLERS-TERSCHUREN (1994): Essai d'analyse systématique du genre *Ophrys*.- Natural. belges **75** (Orchid 7 suppl.): 273-400.
- DEVILLERS, P. & J. DEVILLERS-TERSCHUREN (2006): *Ophrys forestieri* (Reichenbach fil.) Lojacono.- Natural. belges **87** (Orchid 19.): 36-62.
- PAULUS, H. F. (2001): Material zu einer Revision des *Ophrys fusca* s.str. Artenkreises 1. *Ophrys nigroaenea-fusca*, *O. colletes-fusca*, *O. flavipes-fusca*, *O. funerea*, *O. forestieri* oder was ist die typische *Ophrys fusca* Link 1799 (*Orchidaceae*).- J. Eur. Orch. **33**: 121-177.
- PAULUS, H. F. & C. GACK (1980): Beobachtungen und Untersuchungen zur Bestäubungsbiologie südspanischer *Ophrys*-Arten.- Jber. Naturwiss. Ver. Wuppertal **33**: 55-68.
- PAULUS, H. F. & C. GACK (1981): Neue Beobachtungen zur Bestäubung von *Ophrys* (*Orchidaceae*) in Südspanien, mit besonderer Berücksichtigung des Formenkreises *Ophrys fusca* agg.- Pl. Syst. Evol. **137**: 241-258.
- PAULUS, H. F. & C. GACK (1983): Untersuchungen zur Bestäubung des *Ophrys fusca*-Formenkreises in Südspanien.- Jber. Naturwiss. Ver. Wuppertal **36**: 65-72.
- PAULUS, H. F. & C. GACK (1999): Bestäubungsbiologische Untersuchungen an der Gattung *Ophrys* in der Provence (SO-Frankreich), Ligurien und Toscana (NW-Italien) (*Orchidaceae* und *Insecta, Apoidea*).- J. Eur. Orch. **31**: 347-422.

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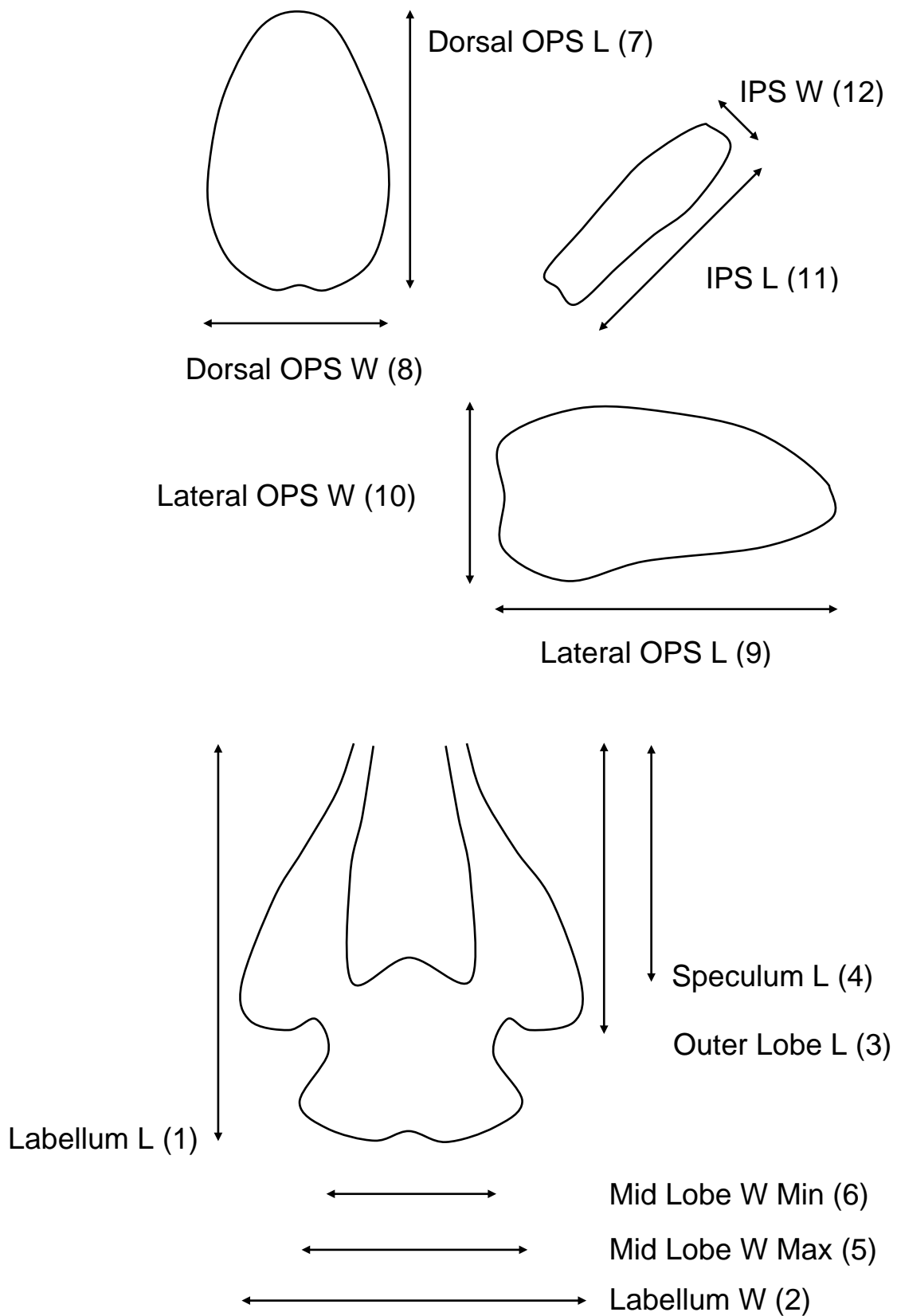


Figure 1: Explanatory diagram for the 12 morphological characters.

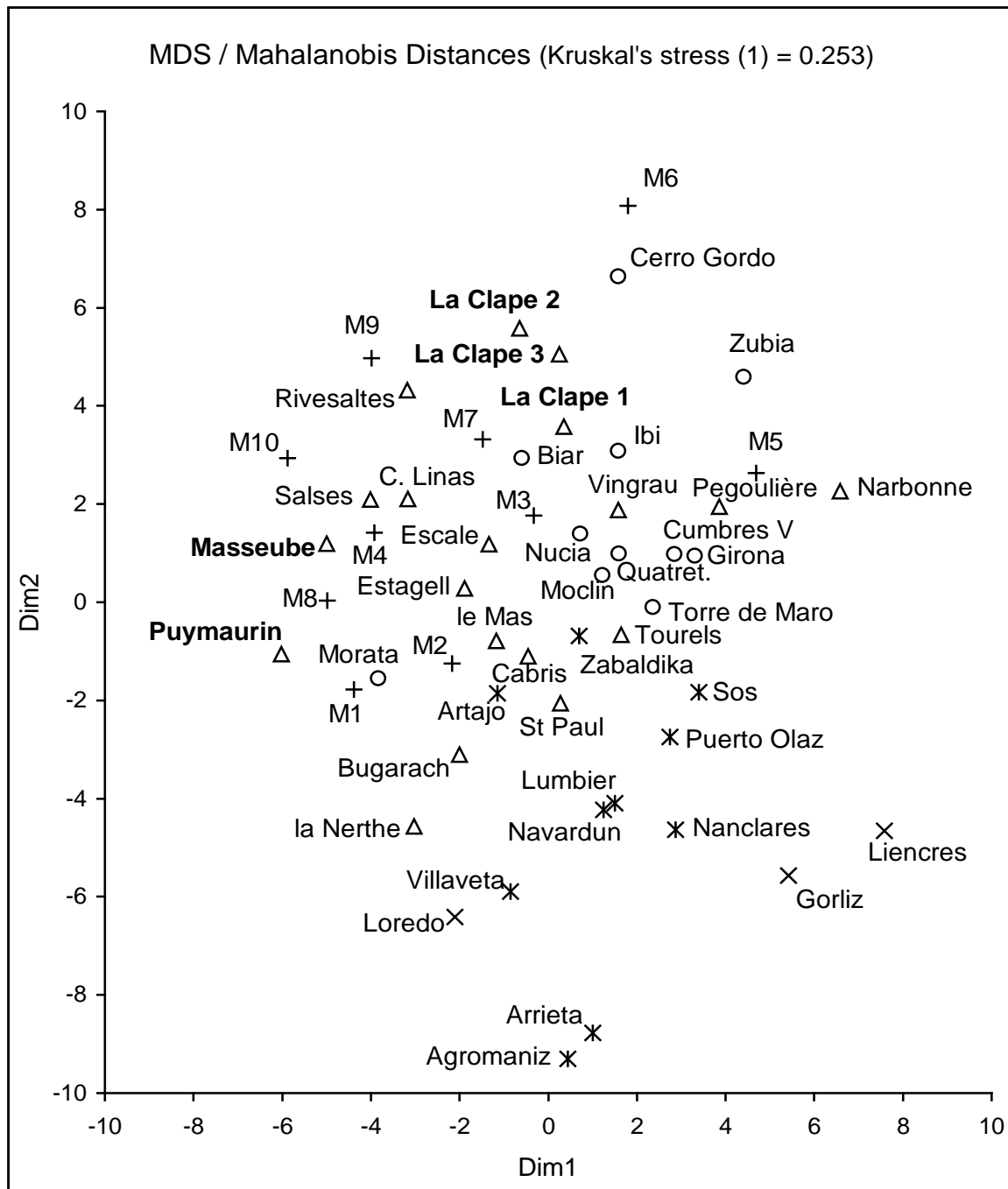


Figure 2: Multiple Dimensional Scaling of Mahalanobis Distances of *Ophrys lupercalis* and *O. forestieri* sample populations from France and Spain.

Key to sample localities: Δ A1 France, \circ A2 Eastern Spain, $+$ B Majorca (M), $*$ C Sub-Pyrenean Spain, \times Cantabrian coast.

Bold type indicates the localities from Le Clape, the locus classicus for *O. lupercalis*, and Masseube and Puymaurin from west French Pyrenees localities for *O. forestieri*.

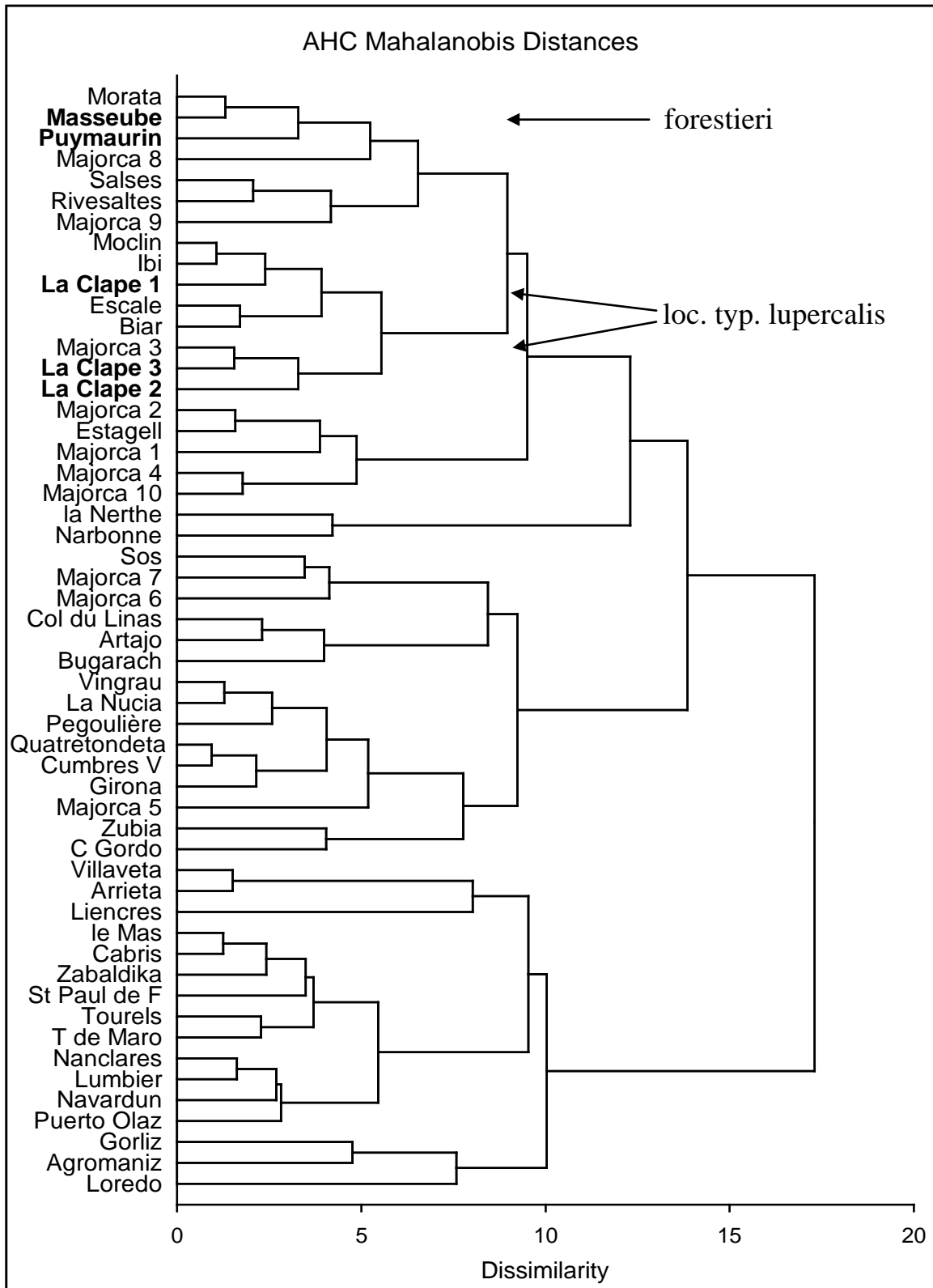


Figure 3: Agglomerative Hierarchical Clustering of Mahalanobis Distances of *Ophrys lupercalis* and *O. forestieri* sample populations from France and Spain.

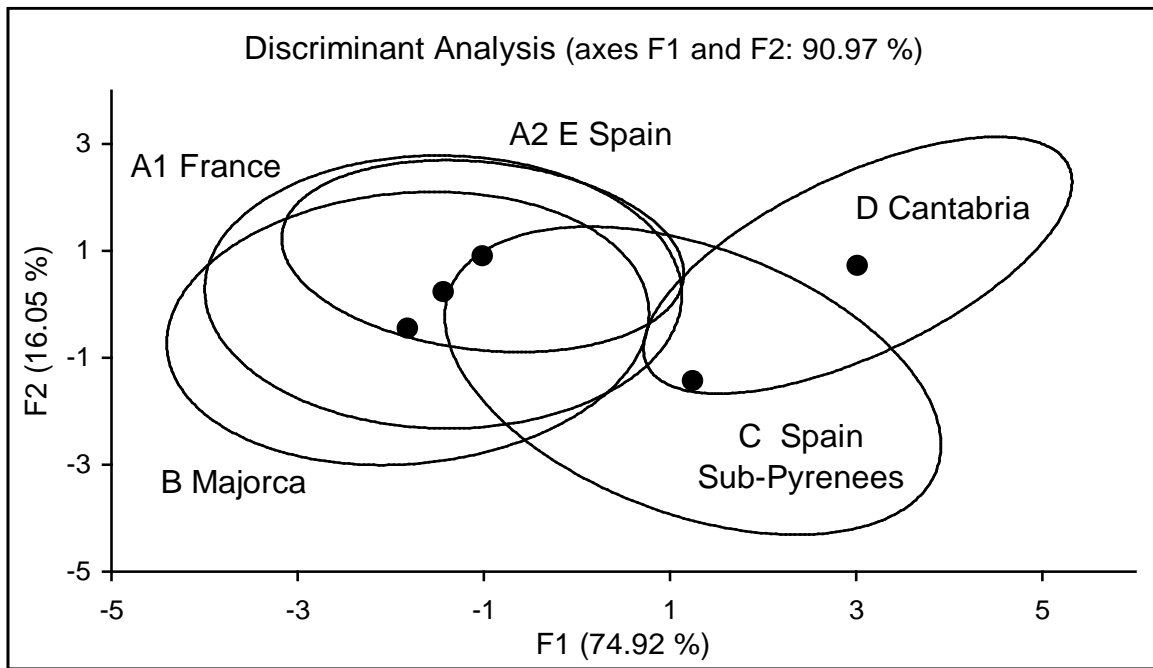


Figure 4: Discriminant Analysis of 5 sub-populations of *Ophrys forestieri* (including *O. lupercalis*).

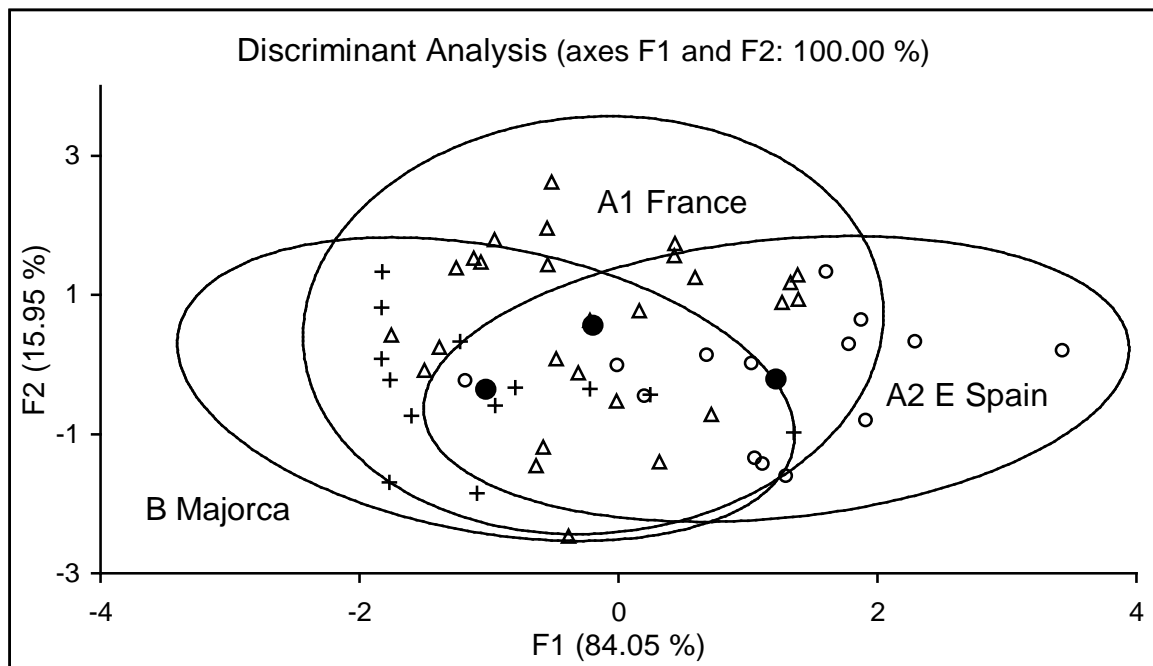


Figure 5: Discriminant Analysis of 3 sub-populations of *Ophrys forestieri* (including *O. lupercalis*).

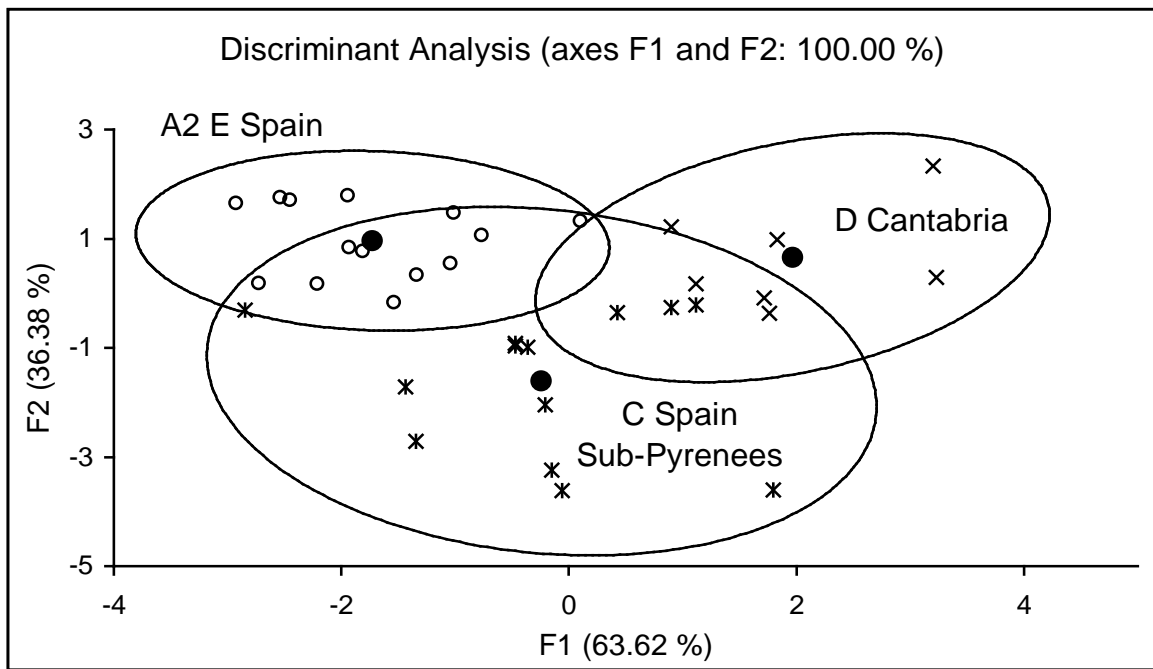


Figure 6: Discriminant Analysis of 3 sub-populations of *Ophrys forestieri* (including *O. lupercalis*).

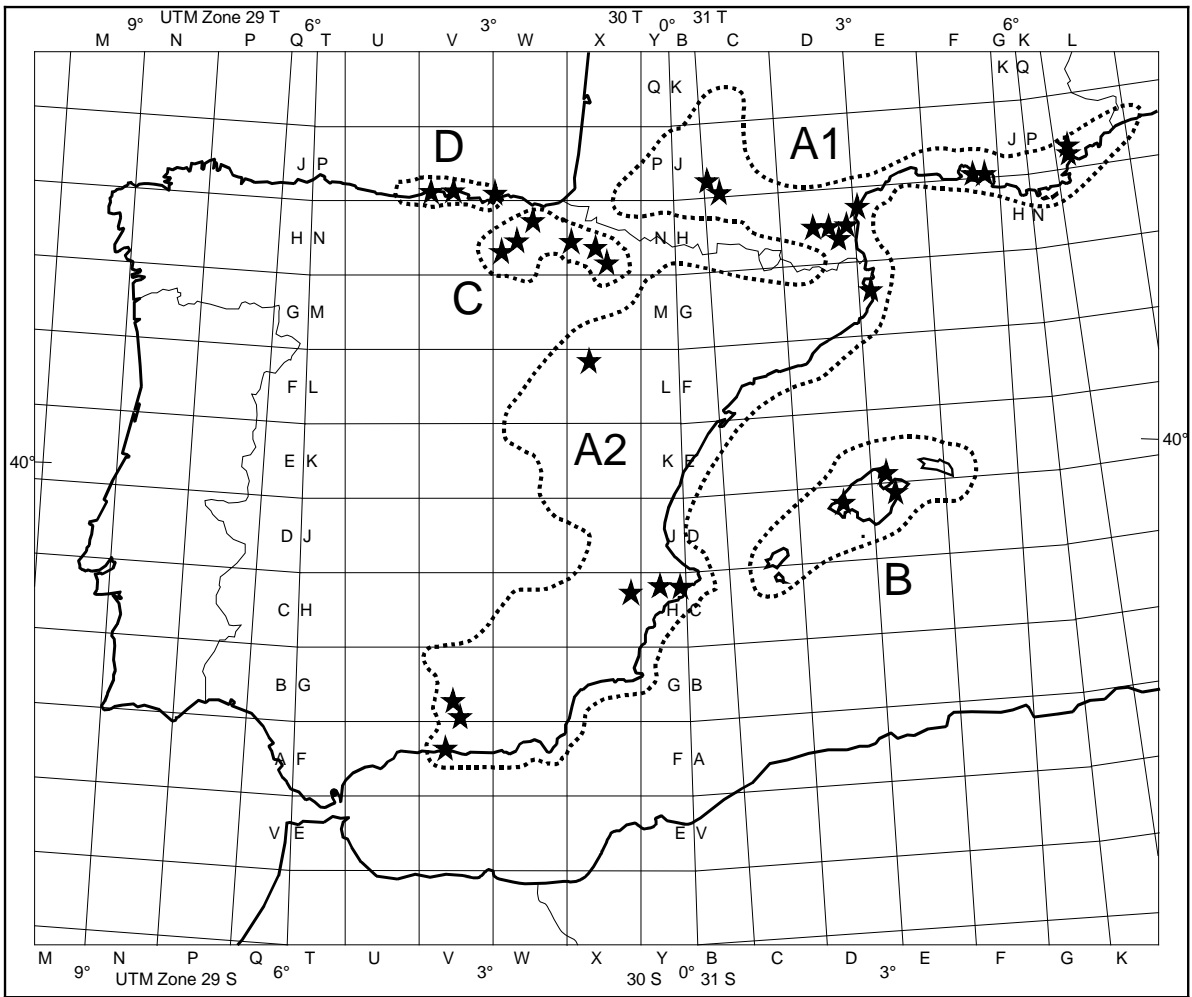


Figure 7: Distribution of *Ophrys forestieri* showing sub-populations: A1 (France), A2 (Eastern Spain), B (Mallorca), C (Sub-Pyrenees) and D (Cantabrian Coast). Sampling points indicated by the symbol ★.

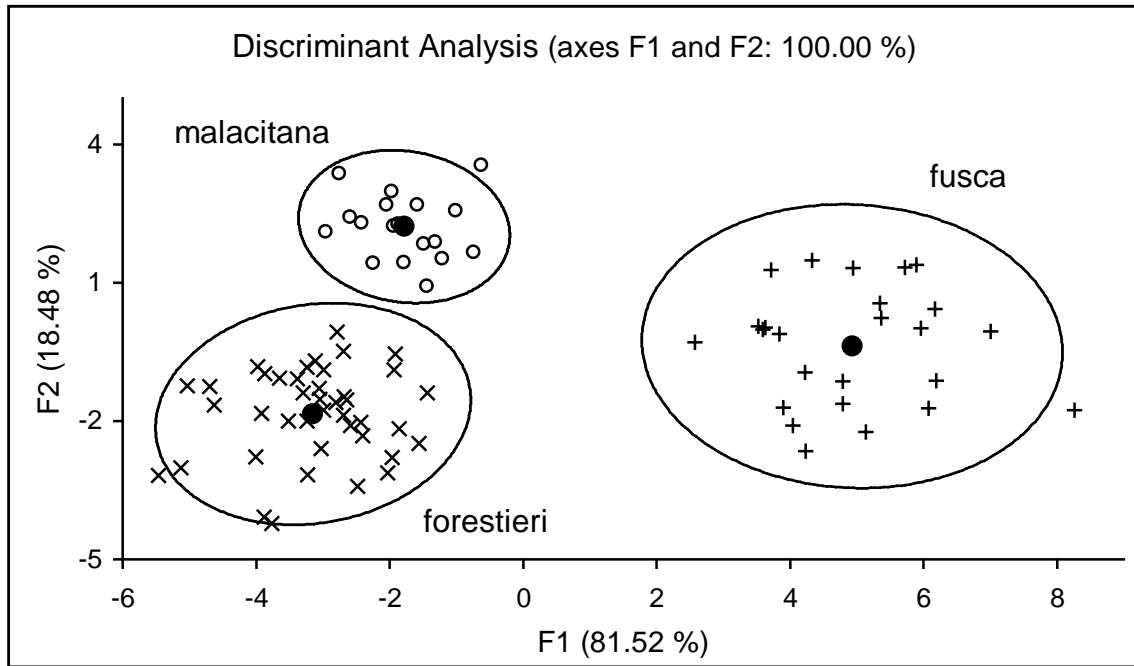


Figure 8: Discriminant Analysis of populations of *Ophrys fusca* (Lisbon population), *O. forestieri* (including *O. lupercalis*) samples A1 & A2 from Spain and France and *O. malacitana*. Data from all 12 characters shown in Fig. 1 - indicating differences in size and shape.

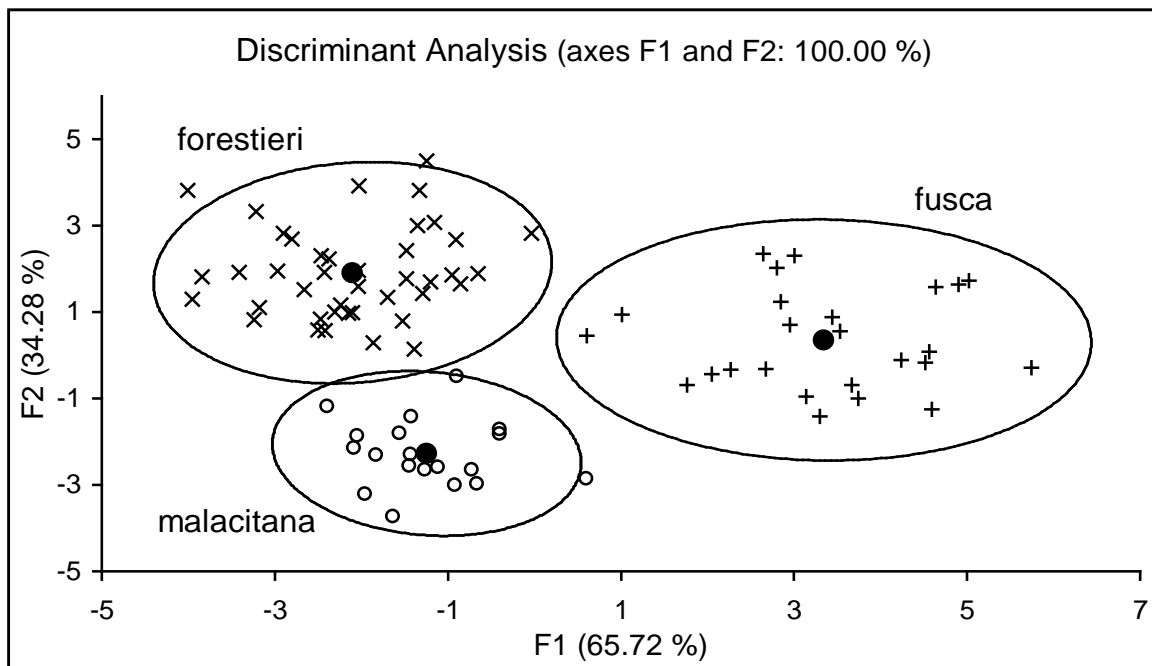


Figure 9: As Fig. 8 with data standardised to labellum length - indicating differences in shape.

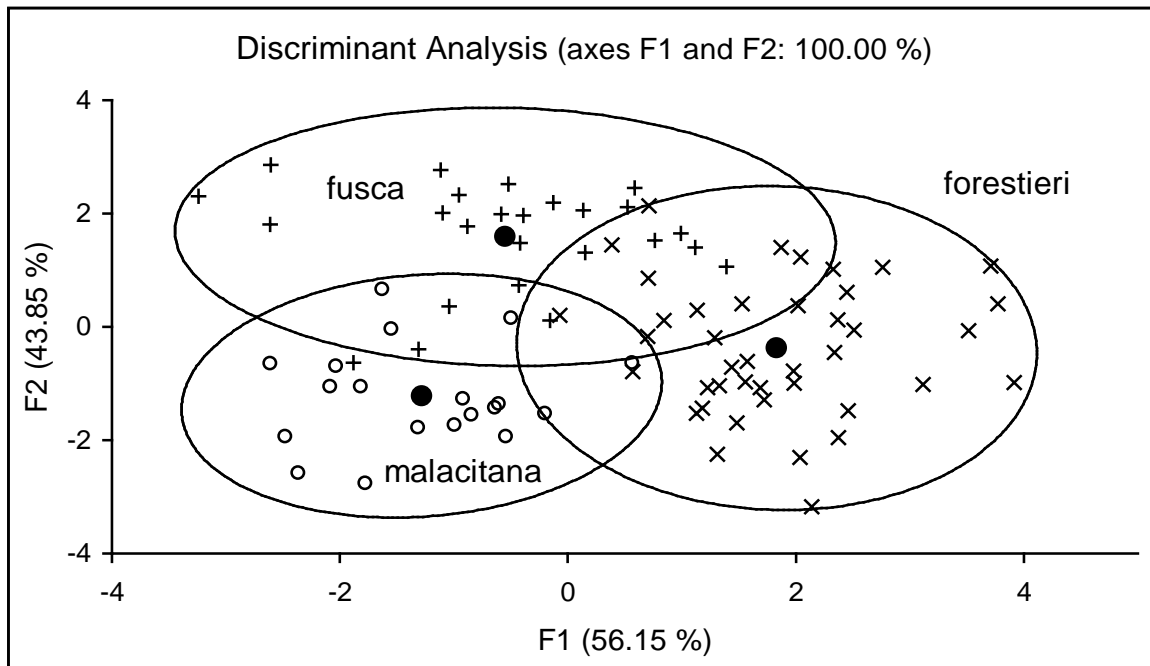


Figure 10: As Fig. 9 but only labellum characters - indicating differences in labellum shape.

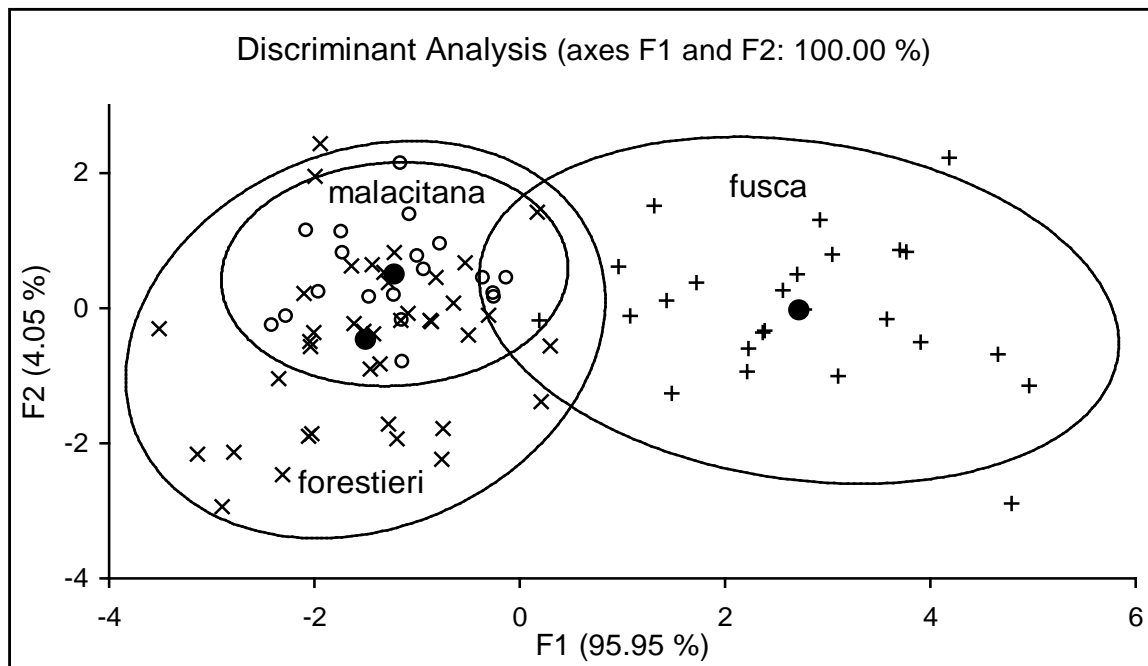


Figure 11: As Fig. 9 but only perianth segment characters - indicating differences in perianth segment shape.

Table 1: Summary data from biometric samples indicating mean (\bar{X}) dimensions and standard deviation (σ).

mm		forestieri A1 France	forestieri A2 E Spain	forestieri B Majorca	forestieri C Spain Sub-Pyrenees	forestieri D Cantabria	fusca Lisbon	malacitana Andalusia
N		179	107	79	93	48	144	166
Labellum characters								
1.	Length (L) σ	15,1 1,08	15,5 1,34	14,7 1,23	15,6 0,97	16,8 1,62	17,6 1,34	15,1 1,26
2.	Width (W) σ	12,1 1,07	12,3 1,17	11,8 0,95	12,5 1,02	13,1 1,52	14,3 1,21	12,7 1,08
3.	Outer lobe length (OLL) σ	11,3 0,98	11,5 1,13	10,8 1,01	11,3 1,06	12,3 1,55	12,7 1,23	10,4 1,10
4.	Speculum length (SP) σ	9,7 0,81	10,0 0,79	9,4 0,97	10,0 0,74	10,7 1,22	10,9 0,95	9,4 0,85
5.	Mid lobe width max. (MA) σ	7,2 0,89	6,9 0,78	6,9 0,80	7,9 0,81	8,0 1,17	9,4 0,96	7,6 0,93
6.	Mid lobe width min. (MI) σ	5,6 0,67	5,5 0,59	5,3 0,54	6,1 0,62	6,1 0,70	6,7 0,63	5,8 0,65
Perianth characters								
7.	Dorsal outer length (DL) σ	10,4 0,92	10,9 0,97	10,1 0,94	11,0 1,07	12,4 1,34	12,1 1,20	11,3 1,14
8.	Dorsal outer width (DW) σ	7,0 0,83	7,0 0,79	6,7 0,73	7,5 0,95	8,2 1,12	7,8 0,96	7,5 0,86
9.	Lateral outer length (LL) σ	11,6 1,02	12,2 1,26	11,1 0,94	12,2 1,25	13,5 1,49	13,4 1,40	12,6 1,26
10.	Lateral outer width (LW) σ	6,7 0,66	6,9 0,73	6,6 0,67	7,2 0,69	7,6 0,97	7,4 0,87	7,2 0,79
11.	Inner length (IL) σ	7,7 0,76	8,1 0,82	7,4 0,76	8,2 0,78	9,1 1,06	9,6 1,00	8,5 0,88
12.	Inner width (IW) σ	2,0 0,29	2,0 0,34	1,9 0,28	2,3 0,38	2,3 0,50	2,7 0,45	2,1 0,31

Table 2: List of *Ophrys* populations sampled for biometric studies.

Sample identifier	n	Colony locality	UTM grid WGS 84	altitude	date
<i>Ophrys fusca</i>					
04LuF001	8	Portugal, Estremadura, Aldeia de Irmãos	29SMC 972 616	90	10.03.2004
04LuF002	11	Portugal, Estremadura, Serra de S. Francisco	29SNC 018 649	130	11.03.2004
04LuF003	14	Portugal, Estremadura, Moinho da Cruz	29SNC 014 645	175	11.03.2004
04LuF004	14	Portugal, Estremadura, Palmela	29SNC 076 689	180	11.03.2004
09LuF026	10	Portugal, Estremadura, Sesimbra, Santana	29SMC 917 567	220	22.03.2009
09LuF027	10	Portugal, Estremadura, Sesimbra Castelo	29SMC 908 562	185	22.03.2009
09LuF028	5	Portugal, Estremadura, Sa. da Arrábida	29SNC 053 605	130	22.03.2009
09LuF029	5	Portugal, Estremadura, Cascais, Charneca	29SMC 603 870	65	23.03.2009
09LuF030	6	Portugal, Estremadura, Bucelas	29SMD 892 049	90	23.03.2009
09LuF031	5	Portugal, Estremadura, Santiago dos Velhos	29SMD 904 066	160	23.03.2009
09LuF032	5	Portugal, Estremadura, Santiago dos Velhos	29SMD 905 081	185	23.03.2009
09LuF033	5	Portugal, Estremadura, Ericeira	29SMD 643 152	90	23.03.2009
09LuF037	6	Portugal, Estremadura, Mato de Cruz	29SMD 929 080	315	31.03.2009
09LuF034	5	Portugal, Ribatejo, Ind. Estate do Campores	29SNE 518 177	300	25.03.2009
09LuF035	5	Portugal, Beira Littoral, Rabaçal - Pombalinho	29TNE 460 306	205	25.03.2009
09LuF036	7	Portugal, Estremadura, Alvorge	29SNE 470 249	260	29.03.2009
09LuF038	6	Portugal, Estremadura, Sa. de Montejunto	29SMD 929 362	440	24.03.2009
09LuF039	17	Portugal, Estremadura, Sa. de Montejunto	29SMD 926 363	400	24.03.2009
<i>Ophrys malacitana</i>					
02HsF007	22	Spain, Málaga, Sa. Blanca, Matagallar	30SUF 440 540	400	20.01.2002
02HsF008	9	Spain, Málaga, Sa. Blanca, Puerto Matagallar	30SUF 439 539	420	20.01.2002
02HsF009	7	Spain, Málaga, SW Monda	30SUF 353 549	550	20.01.2002
02HsF010	10	Spain, Málaga, Monda	30SUF 363 553	420	20.01.2002
02HsF011	8	Spain, Málaga, Sa. de Mijas, Pichón de Jarapalo	30SUF 569 569	240	20.01.2002
05HsF002	8	Spain, Málaga, Coín, Los Llanos	30SUF 414 558	320	28.01.2005
05HsF003	5	Spain, Málaga, Pantano del Chorro	30SUF 395 877	380	03.03.2005
05HsF004	7	Spain, Málaga, Sa. de Alcaparaín	30SUF 357 810	550	03.03.2005
05HsF005	8	Spain, Málaga, Torremolinos	30SUF 631 543	305	05.03.2005
05HsF006	8	Spain, Málaga, Las Lomas de Mijas	30SUF 554 528	480	05.03.2005
05HsF007	7	Spain, Málaga, Casabermeja	30SUF 721 835	835	04.03.2005
05HsF008	8	Spain, Málaga, Riogordo	30SUF 851 875	495	04.03.2005
02HsF012	12	Spain, Córdoba, Sa. de Araceli	30SUG 699 376	800	12.03.2002
04HsF006	13	Spain, Córdoba, Zamoranos	30SUG 973 581	380	13.03.2004
04HsF007	10	Spain, Jáen, Castillo de Locubín	30SVG 138 535	700	13.03.2004
04HsF008	7	Spain, Granada, Restábal	30SVF 485 862	860	14.03.2004
04HsF009	9	Spain, Granada, Río Alhayón	30SVF 668 790	790	14.03.2004
06HsF003	8	Spain, Granada, Orgiva	30SVF 631 818	325	03.03.2006
<i>Ophrys forestieri</i>					
Sample A1 - France					
02GaF013	8	France, Aude, La Clape, les Tinas	31TEH 090 765	60	22.03.2002
02GaF014	15	France, Aude, La Clape, le Pech Rouge	31TEH 104 766	30	22.03.2002
02GaF015	14	France, Aude, La Clape, la Garde	31TEH 093 766	65	22.03.2002

Sample identifier	n	Colony locality	UTM grid WGS 84	altitude	date
06GaF026	6	France, Bouches-du-Rhone, la Pegoulièr	31TFJ 768 042	60	01.04.2006
06GaF027	7	France, Bouches-du-Rhone, S les Tourels	31TFJ 700 046	80	02.04.2006
06GaF029	7	France, Bouches-du-Rhone, la Nerthe	31TFJ 845 065	140	03.04.2006
06GaF025	6	France, Alpes-Maritimes, Cabris	32TLP 290 363	500	04.04.2006
06GaF010	7	France, Var, Narbonne, la Colle Noire	32TLP 223 295	250	04.04.2006
08GaF005	6	France, Gers, Masseube	31TCJ 052 149	255	12.04.2008
08GaF006	5	France, Haute-Garonne, Puymaurin	31TCJ 182 039	220	12.04.2008
08GaF007	35	France, Aude, Bugarach, le Mas	31TDH 444 481	440	14.04.2008
08GaF008	6	France, Aude, Bugarach, S le Mas	31TDH 446 478	440	14.04.2008
08GaF009	6	France, Aude, Bugarach	31TDH 477 475	530	14.04.2008
08GaF010	6	France, Aude, Col du Linas	31TDH 496 475	670	14.04.2008
08GaF012	6	France, Pyrénées-Orient., St.Paul-de-Fenouillet	31TDH 605 403	235	17.03.2008
08GaF013	10	France, Pyrénées-Orientales, Estagell	31TDH 765 359	80	17.03.2008
08GaF014	6	France, Pyrénées-Orientales, Rivesaltes	31TEH 887 375	30	16.03.2008
08GaF015	10	France, Pyrénées-Orientales, Pas de l'Escale	31TDH 833 439	280	17.03.2008
08GaF016	6	France, Pyrénées-Orientales, Vingrau	31TDH 824 443	165	17.03.2008
08GaF066	7	France, Pyrénées-Orientales, Salses	31TDH 925 423	30	17.03.2008
Sample A2 - E Spain (mainland)					
01HsF001	12	Spain, Alicante, Quatretondeta, Tossal de Lloses	30SYH 320 886	650	20.03.2001
01HsF002	5	Spain, Alicante, W Quatretondeta	30SYH 327 896	590	19.03.2001
02HsF016	9	Spain, Girona, Urb. Riells	31TEG 127 618	50	24.03.2002
02HsF017	7	Spain, Granada, Moclín	30SVG 307 277	990	13.03.2002
02HsF018	11	Spain, Granada, Cumbres Verde	30SVG 520 054	1225	13.03.2002
03HsF001	16	Spain, Alicante, Biar, Santuari Verge de Gràcia	30SXH 959 782	780	22.03.2003
04HsF010	9	Spain, Granada, Pinares de Zubia	30SVG 497 069	970	14.03.2004
04HsF011	7	Spain, Granada, Cerro Gordo	30SVF 312 669	150	15.03.2004
04HsF012	8	Spain, Málaga, Torre de Maro	30SVF 261 683	80	15.03.2004
06HsF008	8	Spain, Alicante, La Nucía, Ermita Sant Vicent	30SYH 520 772	160	01.02.2006
06HsF009	7	Spain, Alicante, Ibi - Bañerea, Alto de Biscoy	30SYH 092 812	950	28.03.2006
08HsF089	8	Spain, Zaragoza, Morata de Jalon, Millarroya	30TXL 311 899	530	09.04.2008
Sample B - Majorca					
05BIF015	7	Majorca, Serra de Cans	31SDD 621 829	270	18.03.2005
05BIF016	5	Majorca, Serra de Cans	31SDD 637 844	190	18.03.2005
05BIF017	12	Majorca, Bosc de Bellver	31SDD 670 800	70	18.03.2005
05BIF018	6	Majorca, Galilea - Puigpuñent	31SDD 582 847	390	18.03.2005
05BIF019	8	Majorca, Coll de Sa Gremola	31SDD 492 840	840	16.03.2005
05BIF020	6	Majorca, Paguera - Capdellá	31SDD 539 804	90	16.03.2005
05BIF022	11	Majorca, Platja de Muro	31SEE 111 040	5	14.03.2005
06BIF013	6	Majorca, Platja de Muro	31SEE 112 040	5	21.03.2006
06BIF011	6	Majorca, Son Bauló	31SEE 147 012	10	21.03.2006
06BIF012	12	Majorca, Pinar de Cala Agulla	31SED 388 965	30	20.03.2006
Sample C - Spain, Sub-Pyrenees					
08HsF017	12	Spain, Euskadi, Alava, Nanclares de la Oca	30TWN 155 391	510	10.04.2008
08HsF018	6	Spain, Euskadi, Alava, Agrómaniz	30TWN 375 468	570	10.04.2008

Sample identifier	n	Colony locality	UTM grid WGS 84	altitude	date
08HsF021	19	Spain, Navarra, Zabaldika	30TXN 156 454	455	10.04.2008
08HsF019	7	Spain, Euskadi, Alava, Arrieta	30TWN 411 711	560	10.04.2008
08HsF022	7	Spain, Navarra, Villaveta	30TXN 305 367	475	10.04.2008
08HsF023	6	Spain, Navarra, Artajo	30TXN 358 325	450	10.04.2008
08HsF024	6	Spain, Navarra, ENE Lumbier	30TXN 431 260	470	11.04.2008
08HsF025	6	Spain, Navarra, ENE Lumbier	30TXN 409 243	460	11.04.2008
08HsF026	6	Spain, Navarra, SW Lumbier	30TXN 372 232	455	11.04.2008
08HsF027	6	Spain, Navarra, Puerto Olaz	30TXN 352 202	710	11.04.2008
08HsF029	6	Spain, Zaragoza, Sos del Rey Católico	30TXN 462 064	560	11.04.2008
08HsF031	6	Spain, Zaragoza, Navardún	30TXN 527 086	510	11.04.2008
Sample D - Cantabria/Euskadi coastal					
06HsF017	34	Spain, Cantabria, Dunas de Liencres	30TVP 220 114	5	22.03.2006
06HsF018	6	Spain, Cantabria, Loredo	30TVP 414 127	5	23.03.2006
06HsF019	8	Spain, Euskadi, Gorliz	30TWP 045 076	20	23.03.2006

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12	13
14	15
16	17

Figs. 12-16 *Ophrys malacitana* (photos M. R. Lowe)

Fig. 12: Spain, Málaga, Pichón de Jarapalo UTM 30SUF5656 20.01.2002 (Type). (www.herbariovirtual.ua.es/herbariotipos.htm)

Fig. 13: Spain, Málaga, Pichón de Jarapalo UTM 30SUF5656 20.01.2002.

Fig. 14: Spain, Málaga, Matagallar UTM 30SUF4353 20.01.2002.

Fig. 15: Spain, Málaga, Casares UTM 30STF9631 25.02.1995.

Fig. 16: Spain, Málaga, Casares UTM 30STF9932 25.02.1995.

Fig. 17: *Ophrys malacitana* with pollinator *Colletes cunicularius*, Spain, Málaga, Pichón de Jarapalo 04.03.1980 (photo H. F. Paulus).

