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Analysis of Morphological Variation in a Field Sample of *Caladenia catenata* (Smith) Druce (Orchidaceae)

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Abstract

A morphometric analysis of the variation among 24 attributes of a field sample of 71 plants of the *Caladenia catenata* (Smith) Druce species complex from the Sydney region suggests that there are two phenetically distinct polythetic taxa within this orchid group. These taxa correspond with the traditionally circumscribed taxa *C. alba* R. Br. and *C. carnea* R. Br. but no exclusive or 'key' attribute could be found to discriminate between them. The morphological distinction between these taxa is correlated with habitat differences characterized by soil properties and species associations. However, some of the *C. alba* sites are ecologically intermediate between the 'extreme' *C. alba* sites and the *C. carnea* site, and these sites contain a number of *C. alba* individuals with some attributes traditionally thought to be diagnostic of *C. carnea*. We interpret this character distribution as being the result of introgression of some *C. carnea* attributes into a population of *C. alba*.

Introduction

Caladenia R. Br. (Orchidaceae) is a genus of about 70 named species, distributed throughout Australia, New Zealand, New Caledonia, New Guinea and Java. All the described species occur in Australia (Clements 1982) and most of these are endemic to south-western Western Australia (George 1971). However, the genus is also widespread in eastern Australia, covering a wide range of terrestrial habitats.

Caladenia has always presented taxonomic difficulties, as many of the taxa are hard to differentiate (cf. FitzGerald 1882; Nicholls 1931; Heberle 1982). In particular, the taxa allied to *Caladenia catenata* (Smith) Druce are often impossible to distinguish morphologically with confidence, and during the last 100 years there have been repeated calls for more detailed studies of this group (e.g. FitzGerald 1882; McGillivray 1969; Blaxell 1980).

C. catenata is the prior name for what, in the past, has been commonly known as *C. carnea* R. Br. (Hallé 1977; Blaxell 1980). This taxon is extremely variable, with at least 10 infraspecific epithets having been applied to the various morphological forms (Rupp 1946). When Robert Brown described the genus *Caladenia* in 1810 he recognized *C. alba* R. Br. as a distinct species, and most subsequent workers have followed suit (e.g. FitzGerald 1882; Nicholls 1931, 1969; Rupp 1943, 1946; Willis 1970; Cunningham *et al.* 1981; Jacobs and Pickard 1981; Clements 1982). However, Bentham (1873) could find very little difference between the two taxa and suggested that *C. alba* should be considered as merely a variety of *C. catenata*. This view has achieved a recent vogue (e.g. Hallé 1977; Weber and Bates 1978; Blaxell 1980; Jessop 1983). *C. catenata* var. *catenata* is a synonym of *C. carnea* var. *carnea* (Clements 1982), and Hallé (1977) has distinguished *C. catenata* var. *carnea* (R. Br.) N. Hallé as the name to be applied to the small pink form found in New

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Caledonia. No nomenclatural combination has been made in *C. catenata* for *C. alba* (cf. Beadle *et al.* 1982; Jessop 1983). Therefore, to simplify the discussion in our paper we will use the names *C. carnea* and *C. alba* to denote the two extreme morphological forms within the *C. catenata* complex.

These two taxa are traditionally distinguished by the presence of transverse red bands on the labellum and/or column of *C. carnea*. They have also been distinguished in the past as follows: *C. alba* has large white flowers and *C. carnea* smaller pink ones; *C. carnea* has more numerous calli than *C. alba*, and they are usually orange in colour and have clubbed heads; *C. alba* has longer stems; and *C. carnea* has a later flowering time (e.g. Brown 1810; Bentham 1873; FitzGerald 1882; Nicholls 1931, 1969; Rupp 1943, 1946; Willis 1970; Beadle *et al.* 1972; Weber and Bates 1978; Cunningham *et al.* 1981).

More recently, it has been recognized that orchid systematics needs to be based on a much broader foundation than in the past. In particular, greater attention should be paid to the direct morphological analysis of living plants (Dressler and Dodson 1960) and to the characterization of the environmental conditions at the same scale as that of the morphological variation in the plants (Sanford 1974).

With this in mind, the Sydney region provides an ideal area for analysing the morphological variation between *C. carnea* and *C. alba*. *C. carnea* is widespread and common throughout the tablelands and coastal region of eastern Australia, occurring from southern South Australia to north-eastern Queensland, as well as in Tasmania; while *C. alba* is found along the near-coastal region of New South Wales and south-eastern Queensland, and more rarely in southern Victoria and south-eastern South Australia (Clements 1982).

Nevertheless, there is very little local sympatry between these taxa; *C. alba* is restricted to fertile soils while *C. carnea* is found on poorer sandy soils (FitzGerald 1882; Wallace 1981). However, on the Hornsby Plateau, north of Sydney, where Hawkesbury Sandstone and Wianamatta Shale are interbedded and the sandstone valleys accumulate the runoff from the shale caps (Walker 1960), the two taxa are sometimes found in close proximity. When this happens, morphological intergradation is occasionally observed and this is commonly interpreted as hybridization (e.g. Nicholls 1931; Rupp 1946).

Our study sought to answer two basic questions: (1) what is the nature of the morphological intergradation between *C. alba* and *C. carnea*; and (2) given their apparent ecological separation, does this morphological variation correlate with any environmental factors?

Materials and Methods

Sites

A bushfire in November 1976 in the recreation reserve south of Leuna Ave in Fox Valley, a northern suburb of Sydney, stimulated flowering in a number of orchid species. Several *Caladenia* species have been reported to flower profusely after fires (e.g. Erickson 1965; Willis 1970; Stoutamire 1974; Bates 1984), and the abundant flowering of *C. carnea* and *C. alba* during September–October 1977 allowed a quantitative study of the morphological intergradation between these taxa on a larger scale than would normally be possible.

We sampled five subjectively chosen sites in the recreation reserve, each roughly 6 m in diam. Three of the sites were chosen to represent the morphological extremes apparent within the *C. catenata* complex, and the remaining sites cover the range of the observable morphological intergradation. Using Nicholls (1969) and Beadle *et al.* (1972), the individuals at sites 1 and 2 were all identified *a priori* as predominantly possessing attributes diagnostic of *C. alba*, and those at site 5 were identified as having predominantly *C. carnea* attributes. The plants at sites 3 and 4 showed a combination of attributes typical of both of these taxa. *C. carnea* individuals were uncommon, and we could not find a replicate sampling site for this taxon.

The area sampled was in the valley of a small creek, with the vegetation grading from tall open-forest near the creek to woodland and tall heath near the ridge tops. Sites 1, 2, 4 and 5 were along an 80 m transect running up the hillside from the creek while site 3 was 200 m away at the headwater of the creek.

Data Collection

All flowering *Caladenia* plants were sampled at each site and scored for a total of 24 morphological attributes (see Table 1). These attributes include all those used by other workers to separate these taxa. Only one flower was scored per plant; if more than one flower was open on a raceme, only the lowest flower was scored. In total, 71 plants were sampled (Table 1).

The potential ecological correlations were tested in two ways: (1) analyses of the soils on which the plants occur; and (2) association of the *Caladenia* individuals with the other species occurring in the area. For the soil analyses, two random soil samples, each from the top 17 cm of the profile (excluding the A_0 horizon), were collected from each site with an auger. Three replicate analyses were performed on each soil sample for each of three characteristics estimating soil fertility and water content (see Table 3), using the methods of Allen *et al.* (1974). For the species association, every other vascular plant species within 50 cm of each *Caladenia* sampled was recorded.

Statistical Analyses

A range of univariate and multivariate statistical procedures was used to analyse the data.

For the morphological data, an ordination technique, principal components analysis with varimax rotation (Kim 1975), was used to analyse the pattern of variation among all of the sites simultaneously. Discriminant function analysis (Klecka 1975) was then used to find the maximum degree of morphological separation that exists between the plants at the extreme sites. This technique derives a small number of linear functions which weight the original variables so as to maximize the separation of the total scores of a set of reference samples summed over all of the variables. The two reference samples that we used were: (1) all the plants from sites 1 and 2 (*C. alba* group); and (2) the plants from site 5 (*C. carnea* group). This approach assumes that the two reference samples are distinct groups, and this was confirmed by the principal components analysis. The intermediate plants (sites 3 and 4) were then individually allocated to one of the two groups, using the discriminant function derived from the analysis of the reference samples.

Univariate statistical analyses were used to assess the contribution of each attribute to the distinctiveness of each site. Ten random individuals were chosen from each site for the analysis of each attribute. Those attributes with obviously non-normal data (attributes 2, 14–17, 19, 22 and 23) were analysed using the Kruskal–Wallis test followed by the Simultaneous Test Procedure (Sokal and Rohlf 1981). The remaining attributes were analysed using one-factor analyses of variance followed by Student–Newman–Keuls tests (Underwood 1981). Cochran's test was used to assess the homogeneity of the variances in all cases.

For the soil data, each characteristic was analysed by using two-factor nested analyses of variance and Student–Newman–Keuls tests, after first testing the homogeneity of the variances with Cochran's test.

Results

There is great variation apparent both within and between the populations at each site for all the morphological attributes, and the ranges of the attributes overlap considerably between sites (Table 1).

The principal components analysis separated the *Caladenia* individuals into two distinct groups, with all the individuals from site 5 (*C. carnea*) in one group and the remaining plants in the other group (Fig. 1). This confirms our original subjective assessment of the individuals at sites 1, 2 and 5 as representing morphological extremes. However, the intermediate plants are very similar to the *C. alba* plants even though they were originally assessed as having at least some attributes not diagnostic of this taxon. In fact, some of the plants from the intermediate populations are more dissimilar to the *C. carnea* plants than are the *C. alba* plants. This is particularly true of the plants at site 3, which show a large degree of variability among themselves along both components I and II. The population at site 4, on the other hand, is more homogeneous. Similarly, among the *C.*

Table 1. Attribute measurements and results of analyses of variance and Kruskal–Wallis testing of data from the morphological analysis

Attributes 2, 14–17, 19, 22, and 23 were analysed by Kruskal–Wallis tests and the Simultaneous Test Procedure; the remaining attributes were analysed by analyses of variance and Student–Newman–Keuls tests. For the analyses of variance, d.f. in all cases are 4,45; for the Kruskal–Wallis tests, d.f. in all cases are 4. n.s., Not significant at $P < 0{\cdot}05$. ** Significant at $0{\cdot}01 < P < 0{\cdot}025$. **** Significant at $P < 0{\cdot}005$. S1–S5, means for sites 1–5 respectively. Underlined values are not significantly different at $P = 0{\cdot}05$

Attribute	Site means and ranges					Statistical significance	Multiple comparisons test
	C. alba		Intermediate		*C. carnea*		
	Site 1 ($n = 12$)	Site 2 ($n = 12$)	Site 3 ($n = 19$)	Site 4 ($n = 10$)	Site 5 ($n = 18$)		
1 Stem length (cm)	16·0	13·7	8·9	12·6	7·3	****	S5 S3 S4 S2 S1
	9·5–21·0	8·0–24·0	3·8–13·6	7·3–18·7	3·4–13·7		
2 Column length (mm)	9·7	9·2	9·3	9·3	6·2	****	S5 S2 S4 S3 S1
	9·0–11·0	7·0–11·0	8·0–10·5	8·5–10·0	5·0–7·1		
3 Column width (mm)	3·9	3·6	3·9	3·6	2·9	****	S5 S4 S2 S3 S1
	3·0–4·9	3·0–4·0	3·0–5·0	3·0–4·5	1·7–4·0		
4 Labellum length (mm)	9·2	9·1	9·9	9·7	6·1	****	S5 S1 S2 S4 S3
	8·0–10·5	7·0–12·0	8·0–11·7	8·5–10·5	5·0–8·0		
5 Labellum width (mm)	6·9	6·1	6·3	6·7	6·1	n.s.[C]	
	3·0–8·5	5·0–7·5	5·0–7·3	6·0–8·0	4·8–7·5		
6 Labellum midlobe width (mm)	3·3	2·6	3·1	3·3	1·7	****	S5 S2 S4 S1 S3
	2·0–4·0	2·0–3·0	2·0–4·0	2·7–3·7	1·0–2·0		
7 Dorsal sepal length (mm)	15·7	14·8	14·6	15·4	11·0	****	S5 S2 S3 S4 S1
	11·0–19·0	10·0–19·5	4·0–17·0	13·5–17·0	7·2–14·0		
8 Dorsal sepal width (mm)	3·5	3·0	3·2	3·0	2·3	****	S5 S2 S4 S3 S1
	2·5–4·0	2·0–4·0	2·5–4·1	2·3–3·5	1·2–3·0		
9 No. of flowers per raceme	1·3	1·2	1·0	1·0	1·3	n.s.	
	1–2	1–2	1	1	1–3		
10 No. of calli fringing labellum midlobe	9·2	7·5	9·3	11·7	6·5	****	S5 S2 S1 S3 S4
	6–13	3–11	5–16	8–17	3–9		
11 No. of large clubbed calli at labellum base	6·4	4·2	4·8	5·5	3·0	****	S5 S3 S2 S4 S1
	4–8	2–8	4–8	4–7	2–6		
12 No. of rows clubbed calli on labellum lamina	2·3	2·3	2·3	2·0	2·1	n.s.[C]	
	2–4	2–4	2–4	2	2–3		
13 No. of clubbed calli on labellum lamina	21·3	24·3	25·7	20·9	17·4	**	S5 S1 S4 S2 S3
	8–31	17–42	18–44	14–32	12–26		

14 No. of red bands on mid line of column dorsal surface	0	0	1·1	0	6·1	****	S1 S2 S4 S3 S5
	0	0	0–9	0	1–13		
15 No. of red bands on column latero–dorsal surface	2·3	0	10·1	0	16·2	****	S2 S4 S1 S3 S5
	0–21	0	0–26	0	11–21		
16 No. of red bands on labellum undersurface	0	0	0·7	0	3·9	****	S1 S2 S4 S3 S5
	0	0	0–8	0	0–15		
17 No. of red bands on lamina and labellum side lobes	1·7	0	2·4	0	22·8	****	S2 S4 S1 S3 S5
	0–20	0	0–20	0	18–30		
18 *Colour of labellum tip*[A]	3·2	3·1	3·1	3·1	1·9	****[C]	S5 S1 S2 S4 S3
	3–4	3–4	2–4	3–4	1–3		
19 Colour of perianth inner surface[B]	1·2	1·0	1·7	1·5	2·1	****	S1 S2 S4 S3 S5
	1–2	1	1–4	1–2	1–3		
20 Background colour of labellum side lobes[B]	1·4	1·0	2·1	1·7	1·6	**	S2 S1 S4 S5 S3
	1–3	1	1–4	1–3	1–3		
21 Colour of anther cap[B]	1·3	1·2	1·3	1·6	3·2	****	S1 S2 S3 S4 S5
	1–3	1–2	1–2	1–3	2–5		
22 Colour of calli fringing labellum mid lobe[A]	1·2	1·0	1·2	1·0	2·0	****	S2 S3 S4 S1 S5
	1–2	1	1–3	1	2		
23 Colour of clubbed calli on labellum mid lobe[A]	1·1	1·0	1·0	1·0	2·1	****	S2 S3 S4 S1 S5
	1–2	1	1	1	2–3		
24 Colour of large basal calli on labellum[A]	3·1	2·5	2·5	2·6	2·2	****	S5 S2 S4 S3 S1
	3–4	2–3	2–4	2–3	2–3		

[A] Colour graded subjectively on a scale of 1 (white) to 4 (orange).
[B] Colour graded subjectively on a scale of 1 (white or cream) to 5 (deep pink or red).
[C] Variance homogeneous only at $P = 0{\cdot}01$, therefore significance accepted only at $P < 0{\cdot}01$.

alba plants, those at site 1 are more variable than are those at site 2, indicating that even within a population there can be a large degree of morphological heterogeneity.

The discriminant function analysis gave a very good separation of the two reference samples, with an eigenvalue (or canonical root) of 63·08, and there was an extremely high association between the discriminant function and the original variables (canonical correlation coefficient = 0·99). Based on our samples, the morphologically extreme populations thus form two discrete groups. The attributes contributing most (i.e. with the

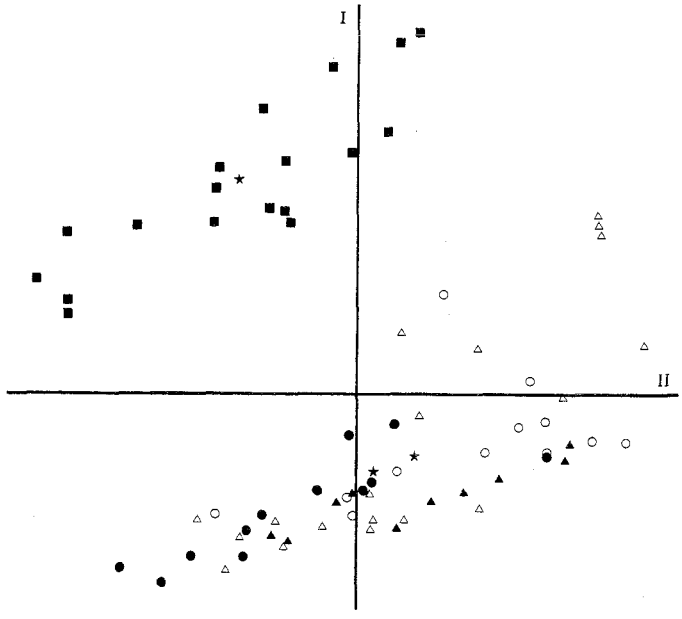

Fig. 1. Projection of the *Caladenia* individuals onto axes representing the first two components from the principal components analysis with varimax rotation of the morphological data. The attributes with the highest correlations with component I are the number of red bands and the colour of the calli (attributes 17, 14, 22, 15 and 23), while component II correlates best with the length and width measurements (attributes 3, 8 and 5). The eigenvalues associated with these two components account for 52·2% of the total variation of the original data set. *C. alba*: ○ Site 1, ● Site 2; Intermediate: △ Site 3, ▲ Site 4; *C. carnea*: ■ Site 5; ★ component means for each of the morphological groupings.

largest weighting coefficients) to the discriminating power of this discriminant function (Table 2) were the colour of the anther cap and the number of red bands on the undersurface of the labellum and on the mid line of the dorsal surface of the column (attributes 14, 16 and 21). Most of the other attributes contributed relatively little to the ability of the function to separate the two groups (Table 2).

All the individuals from the two intermediate samples were identified as having a high statistical probability of belonging to the *C. alba* group. Thus, each intermediate plant is more closely related morphologically to the *C. alba* plants when all 24 attributes are considered simultaneously (cf. Fig. 1), despite the overlap in the range of each of the attributes with those of the *C. carnea* plants (cf. Table 1).

For the analyses of variance of the morphological data (Table 1), all the attributes had homogeneous variances at $P = 0{\cdot}05$ except attributes 5, 9, 12 and 18. To overcome this, attribute 9 was square root-transformed before the analysis of variance, and attributes 5, 12 and 18 were tested for significance in their analyses only at $P = 0{\cdot}01$. The results of the Student–Newman–Keuls tests (Table 1) suggest that the *C. carnea* and *C. alba* populations are distinct for most of the attributes measured, with the intermediate populations generally more similar to the *C. alba* populations than to the *C. carnea* population (e.g. attributes 3, 4, 6–8, 11, 18 and 21).

Table 2. Results of the discriminant function analysis of the morphological data

Attribute codes as in Table 1. The weighting coefficients of the attributes on the discriminant function have been standardized to zero mean and unit variance, thus making all the coefficients directly comparable

Attribute	Discriminant function coeff.	Attribute	Discriminant function coeff.
1	−0·070	13	−0·164
2	−0·160	14	0·930
3	0·061	15	−0·610
4	−0·565	16	−1·167
5	0·477	17	0·543
6	−0·263	18	−0·546
7	−0·340	19	0·587
8	−0·448	20	−0·319
9	0·487	21	1·218
10	0·051	22	0·653
11	0·214	23	0·127
12	0·398	24	0·011

For the Kruskal–Wallis tests (Table 1), all of the attributes had heterogeneous variances at $P = 0{\cdot}01$. Therefore, the results of these analyses should be interpreted with caution (Underwood 1981), although all the analyses showed extremely significant results. The Simultaneous Test Procedures (Table 1) suggest that the *C. alba* and *C. carnea* populations are distinct for all the attributes measured and that, again, the intermediate populations are generally more similar to the *C. alba* populations (e.g. attributes 2, 14, 17, 22 and 23).

Table 3. Characteristics and results of analyses of variance of data for the soils from each site sampled for the morphological analysis

For the analyses of variance, d.f. in all cases are 4,5. * Significant at $0{\cdot}025 < P < 0{\cdot}05$. ** Significant at $0{\cdot}01 < P < 0{\cdot}025$. *** Significant at $0{\cdot}005 < P < 0{\cdot}01$. S1–S5, means for sites 1–5 respectively. Underlined values are not significantly different at $P = 0{\cdot}05$

Soil characteristic	Site means (±s.e.)					Statistical significance	Student–Newman–Keuls test
	Site 1	Site 2	Site 3	Site 4	Site 5		
Total water content (%)	30·0 (2·62)	20·1 (0·43)	13·4 (1·58)	11·7 (0·66)	10·9 (1·07)	*	S5 S4 S3 S2 S1
Loss on ignition (%)	12·4 (1·10)	10·2 (0·78)	6·6 (0·27)	4·9 (0·36)	5·3 (0·35)	***	S4 S5 S3 S2 S1
Total extractable cations (m-equiv./100 g)	2·7 (0·19)	2·6 (0·27)	1·1 (0·10)	0·7 (0·04)	0·7 (0·07)	**	S4 S5 S3 S2 S1

For the analyses of variance of the soil data (Table 3), all the characteristics had homogeneous variances at $P = 0{\cdot}05$. The results of the Student–Newman–Keuls tests (Table 3) suggest that the fertility and moisture content of the soils from the *C. alba* and

C. carnea populations are distinct, and that the soils from the intermediate populations are more similar to that of the *C. carnea* population than to those of the *C. alba* populations.

For the species association, there was a strong tendency for certain species to be associated with each of the morphological groupings. Of the 80 species recorded, 13 were locally endemic to the *C. alba* sites, 13 to the intermediate sites, and 16 to the *C. carnea* site. The intermediate sites have vegetation that is slightly more similar to that of the *C. carnea* site than to that of the *C. alba* sites (51% common species compared to 45%).

Discussion

The results of our study demonstrate that, at least in our study area, there are two distinct phenetic groups within the *C. catenata* complex, corresponding to the traditional concepts of *C. carnea* and *C. alba* (Fig. 1). The discriminant function analysis suggests that these groups are good polythetic taxa, as they do not overlap when all 24 attributes are considered simultaneously, and all those plants that were considered *a priori* to be 'intermediate' could be assigned confidently to one group (*C. alba*).

This indicates that these taxa represent two distinct genotypes; however, the large within-group variation in many of the attributes suggests there is a low degree of genotypic constancy within each taxon. Nevertheless, despite considerable variability within each population, the taxa do not lose their diagnostic features when cultivated under the same conditions (personal observation), and so are not merely phenotypic variants of one gene pool. The 'intermediates' thus seem to be the result of introgression of *C. carnea* attributes into a population of *C. alba*. The position of some of the individuals on the ordination is also suggestive of back-crossing of the F_1 hybrids to the *C. carnea* parents.

No exclusive or 'key' attribute could be found to discriminate between the two taxa (Table 1), owing to the large morphological variation within each group. Instead, these taxa can only be distinguished by using a combination of attributes; these include column size (attributes 2 and 3), dorsal sepal size (attributes 7 and 8), the number of red bands on the labellum and on the mid line of the column (attributes 14, 16 and 17) and the colour of the labellum tip and the anther cap (attributes 18 and 21). Attributes previously used to discriminate *C. carnea* from *C. alba* but which we found to be inadequate include stem length, labellum size, number and colour of the flowers, number of red bands on the edges of the column, and the number, number of rows and colour of the calli.

The fact that no exclusive attribute could be found explains the difficulties encountered in the past in distinguishing consistently between these two taxa. Any of the traditional diagnostic attributes may break down in a particular plant, and this makes it difficult to create a simple key that will allow plants to be easily assigned to one taxon or the other. Nevertheless, the two taxa are probably best distinguished as follows:

C. carnea: always with red bands on the mid line of the dorsal surface of the column and/or on the undersurface of the labellum, and the anther cap pink or red;

C. alba: usually without red bands on either the column or labellum but, if so, the anther cap white or cream.

These two taxa are clearly separated ecologically. The *C. alba* individuals occur with species commonly found in gullies and sheltered damp places, often on shale, with an overstorey consisting of *Pittosporum undulatum*, *Syncarpia glomulifera* and *Eucalyptus gummifera*, and an understorey dominated by *Platylobium formosum*, *Smilax glyciphylla*, *Haloragis heterophylla*, *Lindsaea microphylla*, *Dianella caerulea* and *Pteridium esculentum* (all species nomenclature follows Beadle *et al.* 1982). The *C. carnea* plants occur with species common in low fertility areas on sandstone, often near ridge tops, with a canopy of *E. gummifera* and *E. haemastoma*, and an understorey of e.g. *Pimelea linifolia*, *Dillwynia retorta*, *Dampiera stricta*, *Actinotus minor*, *Acacia suaveolens*, *Casuarina torulosa*, *Banksia ericifolia*, *Pultenaea elliptica*, *Rhytidosporum procumbens*, *Stackhousia viminea* and *Phebalium squamulosum*.

This differentiation in vegetation among the populations presumably reflects the differences in soil properties between the sites. Both *C. alba* sites were near the base of the valley and the soils had a higher moisture content and fertility level than the *C. carnea* site further towards the ridge top (Table 3). However, comparative cultivation and transplant experiments would be needed to identify the causal agent involved in this ecological separation; this would be difficult as caladenias are not easy to raise in culture (personal observation).

Both the intermediate populations occur at sites with vegetation intermediate between that of the typical *C. alba* and *C. carnea* populations, and a soil similar to that of the *C. carnea* site. The overstorey and understorey are basically similar to those of the *C. carnea* site but the understorey at site 3 also shows evidence of disturbance, with a large number of grasses such as *Entolasia marginata*, *Cyathochaeta diandra*, *Ptilanthelium deustum*, *Microlaena stipoides* and *Eragrostis brownii* as well as introduced weeds such as *Hypochoeris radicata* and *Andropogon virginicus*. Some species characteristic of damp places are also present in small numbers, e.g. *Lindsaea microphylla*, *Drosera peltata*, *Haloragis heterophylla*, *Smilax glyciphylla* and *Pteridium esculentum*.

No experimental evidence has been presented for natural hybridization between *C. carnea* and *C. alba* but interspecific hybridization among *Caladenia* species is extremely common (Heberle 1982; S. D. Hopper personal communication), with the main barrier to cross-pollination among species apparently being phenological (Heberle 1982). Phenological isolation can be ruled out for *C. alba* and *C. carnea* as, although there is some small differentiation, the flowering times overlap considerably. Similarly, there is no geographical isolation as the distribution of *C. alba* is completely within that of *C. carnea*. The morphology of the flowers is similar, suggesting that both species are probably pollinated by a range of nectar- and pollen-feeding Diptera and Hymenoptera (cf. Stoutamire 1983), although *C. alba* has only been reported to be pollinated by syrphid flies (Uhlherr 1967) and *C. carnea* by a native bee (Cleland 1960). Moreover, the intermediate plants in our study were observed to set seed. There is thus apparently no genetic, mechanical or ethological barrier isolating *C. carnea* from *C. alba*.

These results suggest that, in our study area, gene flow between *C. alba* and *C. carnea* is restricted by ecological isolation. Where this ecological separation breaks down and the habitats intergrade, the 'intermediates' are often encountered, particularly if the area has been disturbed. A testable generalization prompted by this conclusion is that these taxa are ecologically isolated throughout the area in which they are broadly sympatric and that morphological intermediates are always restricted to ecotones and/or disturbed areas. The restriction of morphological intermediates to ecologically intermediate and/or disturbed zones is relatively common (cf. Briggs 1962), and within *Caladenia* a similar situation has been observed, e.g. between *C. patersonii* var. *longicauda* and *C. hueglii* in Western Australia (S. D. Hopper personal communication).

One test of this generalization would be a broad-scale analysis of morphological and/or genotypic variation within the *C. catenata* species complex. *C. carnea* shows considerable variation throughout its geographic range, and some forms of this taxon apparently resemble *C. alba* more closely than does the *C. carnea* population examined in our study. For example, some populations known as *C. carnea* var. *gigantea* from the north coast of New South Wales resemble *C. alba* in flower size, the colour of the labellum tip and the colour of the calli at the base of the labellum. It is thus possible that the polythetic distinction between these taxa reported for our study area might break down elsewhere. A second test of the generalization would be the ecological analysis of other populations showing apparent introgression to see whether the relationship we have demonstrated is replicated in different areas. Our experience suggests that the situation we have reported is likely to be replicated in many areas.

The taxonomic implications of our conclusions are debatable, and their acceptance would depend on the species concept that one holds. Under a biological species concept

(e.g. Mayr 1969), where evidence of gene flow between two taxa indicates their conspecificity, *C. alba* and *C. carnea* would correctly be treated as subspecies of one polymorphic species. Under an evolutionary species concept (e.g. Wiley 1978), however, in which species lineages are only required to have their own evolutionary tendencies and fate, the existence of some gene flow between species is accepted as long as the individual identities of the lineages are not lost. *C. alba* and *C. carnea* would thus be distinct evolutionary species if they are ecologically isolated to some extent throughout their zone of sympatry.

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