Chromosome number variation in two antipodean floras

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Abstract

Background and aims

We compared chromosome number (CN) variation in the nearly antipodean Italian and New Zealand floras to verify (i) whether patterns of variation reflect their similar latitudinal ranges or their different biogeographic/taxonomic contexts, (ii) if any differences are equally distributed across major taxa/lineages and (iii) if the frequency, number and taxonomic distribution of B-chromosomes differ between the two countries.

Methodology

We compared two datasets comprising 3426 (Italy) and 2525 (New Zealand) distinct cytotypes. We also compared a subset based on taxonomic orders and superimposed them onto a phylogeny of vascular plants. We used standard statistics, histograms, and either analysis of variance or Kruskal–Wallis tests to analyse the data.

Principal results

Mean CN of the vascular New Zealand flora is about twice that of Italy. For most orders, mean CN values for New Zealand are higher than those of the Italian flora and the differences are statistically significant. Further differences in CN variation among the orders and main clades that we studied, irrespective of geographical distinctions, are revealed. No correlation was found between chromosome and B-chromosome number.

Conclusions

Mean CN of the whole New Zealand dataset is about twice that of the Italian flora. This suggests that extensive polyploidization played a major role in the evolution of the New Zealand vascular flora that is characterized by a rate of high endemism. Our results show that the hypothesis of a polyploid increase proportional to distance from the Equator cannot be applied to territories with the same latitudinal ranges but placed in different hemispheres. We suggest that bioclimatic gradients, rather than in addition to latitudinal gradients, might account for a polyploidy increase. Our data also suggest that any adaptive role of B-chromosomes at geographic scale may be sought in their frequency rather than in their number.

Introduction

Italy and New Zealand are nearly antipodean countries, i.e. they are almost 180° apart in longitude but have a closely similar latitude, in the opposite hemisphere. Both countries share a strikingly similar shape, length, area, latitudinal and altitudinal range, and temperate climates. On the other hand, apart from being located in opposite hemispheres, the peninsular nature of Italy contrasts with the fully insular character of New Zealand. This accounts for the shorter length of the Italian coastline (Table 1). Myers et al. (2000) identified 25 biodiversity hotspots across the world where high concentrations of endemic species occur, together with an exceptional loss of habitats. They include the Mediterranean Basin (with the Italian Peninsula at its centre) and New Zealand.
endemism rate is reported at 52 % for the Mediterranean Basin, and 81 % for New Zealand (Table 2). It is therefore interesting to compare the biogeography of the two countries, one belonging to the Holarctic and the other to the Australian floral kingdoms (sensu Cox 2001).

Previous cytological studies show that polyploidy increases with distance from the Equator in both boreal (Löve and Löve 1957; Hanelt 1966) and austral hemispheres (Hair 1966; Hanelt 1966), and that the lowest percentages of polyploids occur in subtropical and warm-temperate regions (Stebbins 1971). However, this does not imply that polyploids are—in absolute terms—more cold tolerant than diploids within a particular region (Levin 2002). Such a trend has also been observed at a local scale for the Italian indigenous vascular flora, where mean chromosome number increases along a bioclimatic/latitudinal gradient, from warmer to colder areas (Bedini et al. 2011).

B-chromosomes (including supernumerary chromosomes or fragments; Battaglia 1964) may occur in addition to the usual chromosome complement of a species (e.g. for Italy, see Corsi and Garbari 1972). Some authors showed that in certain cases their number and frequency vary across a latitudinal gradient, suggesting a role in ecological adaptation of plants (e.g. for the orchid Listera ovata: Vosa 1969, 1983; Garbari 1972; Vosa and Barlow 1972). Moreover, Levin et al. (2005) showed that the occurrence of B-chromosomes is not randomly dispersed across angiosperms, while for the same plant group (angiosperms) Palestis et al. (2004) reported a negative correlation between chromosome and B-chromosome numbers. In contrast, such a correlation was not found in an extensive study of the Italian flora (Bedini et al. 2011).

Chromosome numbers are available for 35 % of Italian (Bedini et al. 2011) and 80 % of the New Zealand indigenous vascular flora (Dawson 2000; de Lange et al. 2006; de Lange and Rolfe 2010). Because of New Zealand’s oceanic and isolated geographic position, and much more recent settlement by people, there is a sharper distinction between indigenous (de Lange et al. 2006; de Lange and Rolfe 2010) and naturalized (Howell and Sawyer 2006) plants than for the flora of Italy. We consider these chromosome records to provide a representative sampling of the native florae of each country, and compared them by quantitative analyses, in order to address the following questions:

(i) Do floras with comparable extension, latitudinal and altitudinal range, but in opposite hemispheres, differ in chromosome number variation, thereby paralleling their biogeographic and taxonomic differences?
(ii) Is any difference equally distributed across major taxa/lineages (e.g. orders)?
(iii) Are there appreciable differences in the frequency, number and distribution of B-chromosomes across taxa in the two areas or in relation to chromosome number?

### Table 1 Geographic features of Italy and New Zealand.

| Feature          | Italy                  | New Zealand                |
|------------------|------------------------|----------------------------|
| Shape            | Long and narrow        | Long and narrow            |
| Length           | 1300 km                | 1500 km                    |
| Area             | 301 336 km²            | 268 680 km²                |
| Altitudinal range| 0–4810 m a.s.l.        | 0–3754 m a.s.l.            |
| Coastline        | 7456 km                | 15 134 km                  |
| Latitude         | 35° 29’ to 47° 05’N    | 34° 07’ to 47° 20’S        |
| Climate          | Temperate/Mediterranean| Temperate/oceanic          |
| Hemisphere       | Boreal                 | Austral                    |

### Table 2 Number of vascular plant species and endemism in the Mediterranean Basin, Italy and New Zealand.

Data are derived from Myers et al. (2000), updated for Italy with unpublished research of the authors and derived for New Zealand from the Landcare Research Plant Names Database. In the column for Italy, the percentage of species with respect to the Mediterranean Basin are reported in parentheses. In addition to these estimates, there are 1694 vascular plant species fully naturalized in New Zealand (Landcare Research Plant Names Database), and only 524 naturalized in Italy (Celesti-Grapow et al. 2009).

| Plant Type            | Mediterranean basin | Italy      | New Zealand |
|-----------------------|---------------------|------------|-------------|
| Indigenous plant species | 25 000              | 6100 (24 %) | 2019        |
| Endemic species       | 13 000              | 903 (7 %)  | 1639        |
| Endemism rate         | 52 %                | 15 %       | 81 %        |
Materials and methods

Data sources
Chrobase.it (Bedini et al. 2010 onwards) stores the available karyological information on the Italian vascular flora, including chromosome number (n and/or 2n) and B-chromosome occurrence, along with main geographic–administrative data and literature references (Bedini and Garbari 2003, 2004). As of 4 March 2011, the database consisted of 6756 count records, derived from 1279 literature references. They refer to 2785 accepted species and subspecies, according to the nomenclature of Conti et al. (2005, 2007). The New Zealand karyological dataset was derived from Dawson (2000, 2008) and Dawson et al. (2000): these data comprise 2525 count records, derived from 356 chromosome references of 2071 taxa. This dataset is comparable with the unreferenced chromosome numbers listed by de Lange and Rolfe (2010). Cytotypes were obtained by excluding replicated counts (i.e. the same chromosome number for the same taxon). Any n count was transformed to 2n and then included in the dataset.

Taxonomic and phylogenetic framework
The phylogenetic framework and taxonomic circumscription at order and family levels, as already described in G. Bedini, F. Garbari and L. Peruzzi (unpubl. data) for the Italian flora, was derived from APG III (2009) and Stevens (2008 onwards) for angiosperms, from Chaw et al. (2000) for gymnosperms, and from Pryer et al. (2001) and Smith et al. (2006) for euryphylyte cryptogams. This was done for both Italian and New Zealand datasets.

Data analysis
Analysis of variance (ANOVA) was used to test statistical differences in chromosome numbers (CNs) among considered groups, after a verification of normal distribution of data (Levene statistics). If data failed the normal distribution test, then a non-parametric test (Kruskal–Wallis) was used. The following data were also calculated for the entire dataset, and for each order: mean CN (± standard deviation) and boxplots, frequency of B-chromosome occurrence and their mean number (± standard deviation), and CN frequency profiles by histograms.

Results
Chromosome number variation of entire datasets
Chromosome numbers were available for taxa belonging to 41 Italian (3426 cytotypes) and 51 New Zealand (2525 cytotypes) orders. There were a total of 56 different orders, 64% (36) of them being shared in the two datasets. However, only 34% (60/171) of families, <7% (68/989) of genera and only 0.1% of species/subspecies (5/4850: Aspelenium trichomanes subsp. quadrivalens, Calystegia soldanella, Montia fontana subsp. fontana, Schoenoplectus tabernaemontani and Zannichellia palustris) were shared by both datasets. A total of 185 different CNs were found, 46.5% of them being shared by Italy and New Zealand, ranging from 2n = 6 (in both datasets) to 2n = 1080 (in the New Zealand dataset only). However, CNs are apparently distributed in different proportions in the two geographical areas (Fig. 1): the most frequent (modal) CN in Italy is 2n = 18; in New Zealand it is 2n = 42. Mean CN in the Italian indigenous vascular flora is 2n = 30.56 ± 22.06, but 2-fold larger (2n = 60.90 ± 55.58) in New Zealand. This difference is supported by ANOVA (F = 840.280; P < 0.000). If data are taken together, irrespective of geographical areas, each order is significantly different from others, in terms of CN variation (χ² = 1104.841, df = 55, P < 0.001).

Mapping CN variation on vascular plant phylogeny
We mapped only 38 orders (or equivalent lineages, e.g. Boraginaceae) on vascular plant phylogeny, by excluding those with 1–5 counts only (considering both datasets). The higher CN values of the New Zealand indigenous vascular flora are largely maintained across the phylogenetic tree of vascular plants, where Italy and New Zealand share a comparable variation pattern among orders, but with CN values for New Zealand offset with respect to those of Italy (Fig. 2). Despite a relatively high number of CN counts available, Polypodiales ferns share similar CN variation in the two areas, completely overlapping. If data are taken all together, irrespective of geographical areas, the clade including the orders from Saxifragales to Brassicales (N = 1200) has CN variation significantly different from the clade (N = 2677) including the orders from Santalales to Apiales (ANOVA: F = 94.050, P < 0.001). A similar picture (data not shown) emerges from the comparison among sister clades within ‘core dialypetalous’ (Fabales–Malpighiales vs. Geraniales–Brassicales) and ‘core gamopetalous’ (Gentianales–Lamiales vs. Asterales–Apiales) orders.

Pairwise CN variation among shared orders
A total of 36 out of the 56 considered orders were shared by both datasets, but only 30 of them possessed a sufficient number of cytotypes (i.e. >1 in both datasets) to infer some kind of variation (Table 3). Sixteen of these 36 orders were statistically different in terms of CN variation between Italian and New Zealand indigenous vascular flora (Table 3). In all these cases, paralleling
the results for the entire datasets, CNs were always higher in the New Zealand flora (see also Fig. 2). For the only three orders in which mean CN is lower in New Zealand than in Italy (Alismatales, Liliales and Eri-cales), the results were not statistically significant.

B-chromosome frequency, number variation and taxonomic distribution

B-chromosomes occur in 246 cytotypes (5.3 % of the dataset) of the Italian vascular flora and in 44 cytotypes only (1.7 % of the dataset) of the New Zealand vascular flora. Among the taxa showing B-chromosomes, their mean number is 2.03 ± 1.75 in Italy and 2.54 ± 1.56 in New Zealand. Since the data on B-chromosome numbers did not follow a normal distribution, we performed the non-parametric Kruskal–Wallis test, which failed, however, to find significant differences between the numbers of B-chromosomes among the two geographical areas. However, it is apparent that the frequency of B-chromosome occurrence in Italy is more than 4-fold higher than in New Zealand.

Table 4 shows the orders and families involved, with their number of taxa/cytotypes showing B-cytotypes for each area: only Asteraceae (Asterales), Orchidaceae (Asparagales) and Ranunculaceae (Ranunculales) are shared among the datasets. Consequently, each of the two geographical groups showed exclusive orders and families with B-chromosomes (Table 4). In the Italian dataset, no significant correlation was detected between CN and B-chromosomes, while in the New Zealand dataset only a weak positive correlation (r = 0.548, P < 0.001) was found.

Discussion

The New Zealand indigenous vascular flora shows a 2-fold mean CN with respect to the Italian flora. A similar pattern of higher mean CN for New Zealand was observed in most of the orders shared by the two areas. A noteworthy exception is represented by the fern order Polypodiales, whose (high) CN values are very similar and completely overlapping in the two areas. This could be explained by the extensive and ancient polyploidization (palaeopolyploidy) of the order (Chiarugi 1960). Our results support an extensive higher polyploid nature of New Zealand compared with
Fig. 2 Clustered boxplots (ITA = Italy, NZ = New Zealand) illustrating the variability of CN, ranging from \(2n = 6\) to \(2n = 1080\), superimposed onto a phylogenetic framework. The outlined central box depicts the middle 50% of the data extending from upper to lower quartile; the horizontal bar is at the median. The ends of the vertical lines indicate the minimum and maximum data values, unless outliers are present in which case the lines extend to a maximum of 1.5 times the inter-quartile range. Circles indicate outliers, unless extreme outliers are present in which case the circles extend to a maximum of three times the inter-quartile range and the extreme outliers are indicated as asterisks. Taxa are arranged by phylogenetic (ordinal) grouping (according to the phylogenetic tree at the bottom of the graph).
Table 3  Number of cytotypes (N) and mean CN ± standard deviation (SD) among orders shared by the Italian and New Zealand CN datasets. Asterisks mark those orders not mapped onto a phylogeny in this study because of the low number of counts available in the datasets; in bold are those orders in which differences in CN variation between Italy and New Zealand are statistically significant. Italics and square brackets show the results of those statistical tests that were significant.

|          | Italy |          |          |          | New Zealand |          |          |          |          |
|----------|-------|----------|----------|----------|--------------|----------|----------|----------|----------|
|          | N     | Mean     | (±) SD   | N        | Mean        | (±) SD   | N        | Mean     | (±) SD   |
| Isoëtales| 10    | 69.2     | 46.5     | 2        | 22           |          | /        |          |          |
| Ophioglossales| 2 | 165      | 106.1    | 5        | 368          | 318.2    |          |          |          |
| Osmundales*| 1   | /        | /        | 5        | 43.6         | 0.9      |          |          |          |
| Salviniales*| 1   | /        | /        | 2        | 32           | 17       |          |          |          |
| Polypodiales| 22  | 126.45   | 52.4     | 164      | 140.8        | 71.9     |          |          |          |
| Pinales   | 5     | 23.6     | 0.9      | 27       | 27.4         | 7.2      |          |          |          |
| Piperales | 13    | 15.7     | 6.8      | 7        | 31.1         | 8.8      |          |          |          |
| [Kruskal–Wallis, $x^2$ = 10.863, $P < 0.001$] |
| Alismatales| 43  | 36.6     | 21.4     | 14       | 26.1         | 9        |          |          |          |
| Liliales  | 68    | 53.7     | 39.7     | 3        | 23.3         | 5.8      |          |          |          |
| Asparagales| 483 | 31.95    | 15.7     | 186      | 52.5         | 40.1     |          |          |          |
| [ANOVA, $F = 91.116$, $P < 0.001$] |
| Arecales*| 1     | /        | /        | 3        | 32           | /        |          |          |          |
| Poales    | 231   | 34.6     | 23.7     | 453      | 53.3         | 32.2     |          |          |          |
| [ANOVA, $F = 60.787$, $P < 0.001$] |
| Ranunculales| 153| 26.8     | 15.85    | 67       | 42.7         | 22.4     |          |          |          |
| [Kruskal–Wallis, $x^2$ = 38.290, $P < 0.001$] |
| Proteales*| 1     | /        | /        | 2        | 28           | /        |          |          |          |
| Saxifragales| 64  | 36.9     | 25.9     | 27       | 47.4         | 27.5     |          |          |          |
| Fabales   | 308   | 25.1     | 18.6     | 38       | 32.2         | 16.4     |          |          |          |
| [Kruskal–Wallis, $x^2$ = 26.050, $P < 0.001$] |
| Rosales   | 43    | 30.1     | 11.6     | 56       | 41.3         | 19.3     |          |          |          |
| [Kruskal–Wallis, $x^2$ = 12.674, $P < 0.001$] |
| Fagales   | 9     | 24       | /        | 4        | 26.5         | 1        |          |          |          |
| [Kruskal–Wallis, $x^2$ = 11.700, $P < 0.001$] |
| Celastrales*| 1   | /        | /        | 1        | /            | /        |          |          |          |
| Oxalidales| 1     | /        | /        | 13       | 28.5         | 3.4      |          |          |          |
| Malpighiales| 118 | 30       | 20.1     | 36       | 44.9         | 23.9     |          |          |          |
| [Kruskal–Wallis, $x^2$ = 12.760, $P < 0.001$] |
| Geraniales| 16    | 25.6     | 8.2      | 19       | 51.6         | 18.1     |          |          |          |
| [Kruskal–Wallis, $x^2$ = 19.585, $P < 0.001$] |
| Myrtales  | 7     | 28       | 10.3     | 114      | 28.6         | 7        |          |          |          |
| Crossosomatales*| 1 | /        | /        | 1        | /            | /        |          |          |          |
| Sapindales| 11    | 31.1     | 15.1     | 10       | 37.6         | 16.5     |          |          |          |
| Malvales  | 34    | 30.6     | 18       | 30       | 36.9         | 23.6     |          |          |          |
the antipodean Italian flora, especially concerning angiosperms. This higher polyploidy is also maintained across most of the orders. Polyploidy (including allopolyploidy) must have been an important evolutionary factor in New Zealand (see also Hair 1966), given that 82% of that flora is endemic (Myers et al. 2000; de Lange et al. 2006; de Lange and Rolfe 2010; Murray and de Lange 2011). This is further confirmed by the low taxonomic affinities at the species level among Italian and New Zealand CN datasets (which we assume here to be representative of the total indigenous vascular floras); only 0.1% of the studied species are shared by the two areas. This is in good agreement with the assignment of the two floras to distinct floral kingdoms (Cox 2001). The distribution of CNs that we obtained for the New Zealand dataset (Fig. 1) is in good agreement with a similar histogram presented by a New Zealand-only study (Murray and de Lange 2011).

Polyploidy was repeatedly shown to increase with latitude in the boreal hemisphere (Löve and Löve 1957; Hanelt 1966; Levin 2002; Brochmann et al. 2004), and a similar (symmetrical) relationship was also suggested for the austral hemisphere (Hair 1966; Hanelt 1966; Levin 2002). Consequently, one would expect that the CN variation in the New Zealand flora be comparable to that of the Italian flora, as the two countries share comparable areas and are situated approximately at the same latitude and altitudinal range. However, our results clearly show that this is not the case. This discrepancy could be explained by the different climate currently found in the two areas: temperate Mediterranean in Italy and temperate oceanic in New Zealand. According to the results presented in Bedini et al. (2011), on a more local scale (see Introduction), we can suggest that CN variation could follow a bioclimatic rather than just a latitudinal gradient. The different polyploidy levels of the floras may also reflect the differing geological histories of the respective landmasses. Italy has had a long history of connection with the European landmass and (during much of this time) plant dispersal is likely to have been more continuous and less restricted. On the other hand, the area now occupied by New Zealand has had a long history of geographic isolation with disruption of the flora through glaciations and a possible marine inundation during the Oligocene. These disruptions have created opportunities for long-distance dispersal and establishment of propagules to the landmass, with recent and rapid species radiations occurring in new habitats (Dawson and Winkworth

| Table 3 Continued |
|-------------------|-----------------|-----------------|
|                   | Italy           |               |
|                   | Mean (±) SD     | New Zealand    |
|                   | N               | Mean (±) SD    |
| Brassicales       | 196             | 23             | 28             |
|                   | 14.5            | 63             | 35.1           |
| Santalales        | 4               | 17.5           | 8              |
|                   | 4.4             | 25.25          | 2.4            |
| Caryophyllales    | 316             | 32.1           | 62             |
|                   | 25.3            | 55.2           | 31.2           |
| Ericales          | 33              | 42.6           | 68             |
|                   | 22.1            | 30.7           | 17             |
| Gentianales       | 93              | 31.9           | 111            |
|                   | 12.2            | 56.3           | 36.3           |
| (Boraginaceae)    | 82              | 32.7           | 18             |
|                   | 26              | 45.2           | 3.9            |
| Solanaceae        | 17              | 30.6           | 16             |
|                   | 13.6            | 37.4           | 23.1           |
| Lamiales          | 270             | 29.6           | 250            |
|                   | 18.1            | 55.2           | 26.6           |
| Asterales         | 614             | 26.9           | 430            |
|                   | 16.4            | 72.7           | 51.2           |
| Apiales           | 84              | 22.8           | 132            |
|                   | 11              | 36.3           | 27.2           |

[ANOVA, F = 117.043, P < 0.001]
[ANOVA, F = 39.943, P < 0.001]
[ANOVA, F = 34.438, P < 0.001]
[Kruskal–Wallis, $\chi^2 = 19.071, P < 0.001$]
[ANOVA, F = 166.379, P < 0.001]
[ANOVA, F = 427.010, P < 0.001]
[ANOVA, F = 18.454, P < 0.001]
These conditions are likely to favour extensive hybridism, allopolyploidy and polyploidy. Examples of polyploidy among New Zealand genera are discussed in the review by Murray and de Lange (2011).

CN per se bears little (if any) phylogenetic and systematic significance, since it is well known that many taxa share the same (unrelated) CN across different groups (Stace 2000; Guerra 2008; Stuessy 2009). However, recent work by G. Bedini, F. Garbari and L. Peruzzi (unpubl. data) emphasized that quantitative analysis of CN variation can be very useful for taxonomic and, in some cases, phylogenetic characterization of taxa: major superordinal clades, all orders, some families and genera of the Italian vascular flora indeed show distinctive CN variation patterns. The results of G. Bedini, F. Garbari and L. Peruzzi (unpubl. data) are largely confirmed in the present study, also including new data from the New Zealand vascular flora.

Conclusions and forward look

Our results clearly support previous data showing that B-chromosome frequency and distribution do not vary randomly in angiosperms (Levin et al. 2005; Bedini et al. 2011). B frequencies, either in Italian or in New Zealand indigenous vascular flora, are higher than the value that can be calculated from the data reported by Jones (1995; ~0.5% of vascular land plants), even if that author estimates a frequency of 10–15% to be more realistic. Bedini et al. (2011) have already pointed out that the negative correlation between chromosome and B-chromosome numbers, previously found by Palestis et al. (2004), is not confirmed for the Italian vascular flora. Concerning New Zealand, only a weak (but positive!) correlation resulted; this further confutes the hypothesis of Palestis et al. (2004). On the other hand, the lower frequency of B-

### Table 4

Orders and families showing B-chromosomes in Italy and New Zealand, with respective number of taxa (cytotypes) and mean number of B-cytotypes ± SD. In bold are those orders shared among Italian and New Zealand CN datasets; / = not applicable.

| Order   | Family            | Italy   | New Zealand |
|---------|-------------------|---------|-------------|
|         |                   | B-cytotypes | Mean ± SD | B-cytotypes | Mean ± SD |
| Apiales | Apiaceae          | 2       | 1.50 ± 0.71 | 0          | /          |
| Asparagales | Amaryllidaceae   | 14      | 1.00 ± 0.68 | /          | /          |
| Asparagales | Asparagaceae     | 44      | 3.29 ± 2.20 | 0          | /          |
| Asparagales | Iridaceae        | 5       | 2.20 ± 1.64 | /          | /          |
| Asparagales | Orchidaceae      | 7       | 1.28 ± 0.95 | 5          | 1.80 ± 0.45 |
| Asterales | Asteraceae       | 28      | 1.82 ± 1.16 | 44         | 3.06 ± 1.67 |
| (Boraginaeae) | Boraginaceae     | 11      | 1.73 ± 1.42 | 0          | /          |
| Brassicales | Brassicaceae     | 16      | 1.68 ± 1.30 | 0          | /          |
| Caryophyllales | Caryophyllaceae | 0       | /          | 1          | 2          |
| Ericales | Primulaceae       | 1       | 1          | 0          | /          |
| Fabales | Fabaceae          | 53      | 1.49 ± 1.39 | 0          | /          |
| Gentianales | Gentianaceae     | 1       | 2          | 0          | /          |
| Lamiales | Lamiaceae         | 6       | 1.50 ± 0.55 | /          | /          |
| Lamiales | Orobanchaceae    | 1       | 4          | /          | /          |
| Lamiales | Plantaginaceae   | 3       | 1.33 ± 0.58 | 0          | /          |
| Malpighiales | Euphorbiaceae   | 1       | 1          | 0          | /          |
| Poales | Poaceae           | 45      | 2.18 ± 1.42 | 0          | /          |
| Ranunculales | Ranunculaceae   | 5       | 3.80 ± 5.21 | 5          | 1.4 ± 0.55 |
| Rosales | Rhamnaceae       | 0       | /          | 2          | 1.0 ± 0 |
| Saxifragales | Crassulaceae   | 1       | 1          | 0          | /          |
| Solanales | Solanaceae       | 1       | 2          | 0          | /          |
chromosome occurrence in the New Zealand indigenous vascular flora (also noted by Murray and de Lange 2011) compared with Italy further supports the observations of Bedini et al. (2011) in suggesting that any adaptive role of B-chromosomes at geographic scale is more likely to be found in their frequency of occurrence (much higher in Italy than in New Zealand) rather than in their number. This is because where B-chromosomes occur, their number is largely overlapping among the two countries.

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Contributions by the authors
L.P. conceived the project, analysed the data and wrote the first draft of the manuscript. M.I.D. furnished digitalized data of New Zealand chromosome number knowledge, and contributed to parts of the Introduction and Discussion dealing more specifically with New Zealand. G.B. contributed by furnishing and handling Italian chromosome number information and contributed substantially to successive versions of the manuscript.

Conflicts of interest statement
None declared.

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