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NIEMEYER, Hermann M.

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Quantitative screening for alkaloids of native vascular plant species from Chile: biogeographical considerations

[Muestreo cuantitativo de alcaloides en especies de la flora nativa vascular de Chile: consideraciones biogeográficas]

Hermann M. NIEMEYER

Departamento de Ciencias Ecológicas, Facultad de Ciencias, Universidad de Chile, Casilla 653, Santiago, Chile

Contactos | Contacts: Hermann M. NIEMEYER - E-mail address: niemeyer@abulafia.ciencias.uchile.cl

Abstract: A screening was undertaken of the native flora of Chile excluding Pteridophyta, Cactaceae and Poaceae the study included 396 species. Alkaloids were found in 189 species, the highest concentration being 3 mg/g dry tissue. A table was produced listing all species collected and specifying the subset containing alkaloids, the species with a particularly high alkaloid content and also the endemic species within this latter set. Alkaloid concentration as well as presence or absence data in different vegetational subregions correlated with mean collection latitude, under the limitations imposed by the relationship between alkaloid concentration and species lineage. In practical terms, the results indicate that in studies aimed at identifying alkaloids in the native flora of Chile, plant collections should take into consideration the promising species listed in the table and take place at the lowest possible latitude if the chance of finding alkaloids and the yield of isolated alkaloids are to be maximized.

Keywords: Alkaloids; native flora of Chile; potential sources of alkaloids; latitudinal distribution of alkaloids; alkaloid distribution in ecoregions

Resumen: Se muestreó un conjunto de 396 especies de la flora nativa de Chile (excluyendo Pteridophyta, Cactaceae y Poaceae), encontrándose alcaloides en 189 de ellas (concentración máxima: 3 mg/g de tejido seco). Se generó una tabla que contiene todas las especies analizadas y especifica aquellas que contienen alcaloides, aquellas que los contienen en altas concentraciones y, entre estas últimas, aquellas que son endémicas. La concentración de alcaloides y también los datos de presencia/ausencia correlacionaron con la latitud media de colecta, bajo las limitaciones impuestas por la relación entre concentración de alcaloides y linaje de la especie. En términos prácticos, los resultados señalan que en estudios que apunten a la identificación de alcaloides en la flora nativa de Chile la colecta de plantas debería tomar en consideración las especies promisoras incluidas en la tabla y ser realizada a la latitud más baja posible dentro del rango de distribución de la especie, maximizando así la probabilidad de encontrar alcaloides en altas concentraciones.

Palabras Clave: Alcaloides; flora nativa de Chile; fuentes potenciales de alcaloides; distribución latitudinal de alcaloides; distribución de alcaloides en regiones vegetacionales

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INTRODUCTION

The continental territory of Chile shows several peculiarities which suggest the occurrence of a unique native flora: i) it is geographically isolated by the Andean range, the Pacific Ocean, and the Atacama desert, ii) it includes vegetational regions which range from one of the driest deserts on Earth, the Atacama desert, to the cool southern temperate rain forests which receive an average annual rainfall exceeding 4.5 m, and iii) it comprises a large range of latitude (18 to 56°S) and altitude (0 to over 6000 m.a.s.l.). The continental vascular native flora is composed by 184 families, 1008 genera, and 5082 native species (Marticorena, 1990), whose degree of endemism (*ca.* 52 %) is higher than that expected from its surface area (Major, 1988). Thus, the Chilean flora provides an interesting and to a great extent unique set of plant species for phytochemical studies.

One of the most interesting types of plant secondary metabolites are the alkaloids, mostly on account of their involvement in ecological interactions (Harborne, 1990) and of their pharmacological activities which have led to their wide use as drugs (Cordell *et al.*, 2001). A recent review on alkaloids identified in the native flora of Chile reported that alkaloids had been isolated from only 12.5, 3.9 and 1.7% of families, genera and species of the native flora of Chile, respectively (Echeverría and Niemeyer, 2013). The study also found that most reports had been published more than 10 years ago, i.e., that the interest for isolating new alkaloids from native Chilean plants appeared to be vanishing. The paper argued for the study of some particular families which were more likely to yield alkaloids, e.g., Convolvulaceae, Ephedraceae, Lamiaceae, Liliaceae, Papaveraceae, Phytolaccaceae, Ranunculaceae and Zygophyllaceae. Given the importance of alkaloids in the cure of widespread diseases, more work on their isolation, structural characterization and biological activity should be undertaken. The present work aims at being a stimulus for undertaking such studies by providing a starting point in terms of promising species to analyze and defining environmental conditions for their efficient collection.

MATERIALS AND METHODS

Plant material

Aerial tissue was collected haphazardly from plants in different vegetational zones of Chile (Gajardo, 1994). Pteridophyta, Cactaceae and Poaceae were excluded

from the survey. The number of individuals sampled ranged from *ca.* five in the case of trees to *ca.* 100 in the case of small herbs. Material collected from each species was pooled, air-dried in the shade, ground, and stored at 15 °C ± 5 °C until extracts were prepared. Voucher specimens were lodged at herbaria at Universidad de Concepción, Museo Nacional de Historia Natural, or Universidad de Chile.

Extraction of alkaloids

Preparation of extracts and their analysis were performed as described (Bustamante *et al.*, 2006). In brief, acidic extracts were prepared with dry plant material and successive aliquots of Dragendorff reagent (Wagner *et al.*, 1984) were used as precipitating agent. Precipitates were isolated and dried; when no further precipitation occurred upon addition of an aliquot of the reagent the weight of the combined precipitates was used to extrapolate the amount of alkaloids in the sample from a regression line made with a set of standards. The regression line included alkaloids of different structural types and molecular weights. Interference by compounds such as coumarins, which give false positives when the Draggendorf reagent is used in color tests rather than as precipitating agent, was shown to be absent, and the method was shown to be highly reproducible (Bustamante *et al.*, 2006).

RESULTS AND DISCUSSION

The present work included 396 species (*ca.* 10% of total of the native Chilean flora, excluding Pteridophyta, Cactaceae and Poaceae) belonging to 200 genera (*ca.* 24% of total) and 75 families (*ca.* 48% of total). The level of endemism of the set of species studied was *ca.* 42% (165 endemic species). The subset of species collected was considered to be a fair representation of the Chilean flora as a whole (with the exception of Pteridophyta, Cactaceae and Poaceae) since the number of species collected was proportional to the number of species in the Chilean flora both at the family (Pearson correlation: $R=0.954$, $N=75$, $P<<0.001$) and genus levels (Pearson correlation: $R=0.758$, $N=200$, $P<<0.001$).

Table 1 shows the species collected specifying the subset found to contain alkaloids, the species with a particularly high alkaloid content and also the endemic species within this latter set. The occurrence of plant secondary metabolites has been shown to be under phylogenetic and environmental influence (e.g.,

Shonle and Bergelson, 2000). A seminal work testing the environmental effect on alkaloid occurrence showed that the proportion of plant species containing alkaloids within country floras was negatively correlated with the mean latitude of the country (Levin, 1976), a pattern thought to be a consequence of selective responses to insect pressure, which is higher in the tropics than at higher latitudes. In the

present work, this relationship was explored using vegetational subregions as spatial units (Gajardo, 1994). Figure 1 shows the robust relationship between the proportion of alkaloid bearing plants from 13 different vegetational subregions of Chile and the mean collection latitude. This correlation gives support to the idea of environmental effects of alkaloid occurrence in plants.

TABLE 1

Species of the native flora of Chile screened for alkaloids.

Normal type: species lacking alkaloids; bold type: species containing alkaloids; capital letters: species with a particularly high alkaloid content and absent from the review by Echeverría and Niemeyer (2012); underlining: endemic species within this latter set.

Family	Species
Aetoxicaceae	<i>Aetoxicon punctatum</i>
Aizoaceae	TETRAGONIA ANGUSTIFOLIA, TETRAGONIA MARITIMA
Alliaceae	<i>Leucocoryne coquimbensis</i>
Amaryllidaceae	ALSTROEMERIA ANDINA, ALSTROEMERIA LIGTU, ALSTROEMERIA MAGNIFICA
Anacardiaceae	<i>Lithrea caustica, Schinus molle, Schinus montanus, Schinus polygamus</i>
Apiaceae	<i>Azorella compacta, Azorella cryptantha, Eryngium paniculatum, Gymnophyton flexuosum, Gymnophyton isatidicarpum, Gymnophyton spinosissimum, Laretia acaulis, Mulinum spinosum, Sanicula graveolens</i>
Aristolochiaceae	Aristolochia bridgesii, Aristolochia chilensis
Asteraceae	<i>Acrisione denticulata, Ageratina glechonophylla, Ambrosia artemisioides, Ambrosia chamissonis, Aristeguietia salvia, Baccharis boliviensis, BACCHARIS LINEARIS, Baccharis nivalis, Baccharis paniculata, Baccharis patagonica, Baccharis pingraea, Baccharis poeppigiana, Baccharis rhomboidalis, BACCHARIS SAGITTALIS, Baccharis santelicens, Baccharis scandens, Bahia ambrosioides, Centaurea chilense, Centaurea floccosa, Chaetanthera chilensis, Chaetanthera flabellifolia, Chaetanthera glandulosa, Chersodoma candida, CHERSODOMA JODOPAPPA, Chuquiraga kuschelii, Chuquiraga oppositifolia, CHUQUIRAGA SPINOSA, Chuquiraga ulicina, DIPLOSTEPHIUM CINEREUM, Diplostephium meyenii, ENCELIA CANESCENS, Erigeron fasciculatus, ERIGERON LEPTOPETALUS, Erigeron luxurians, FLOURENSIA THURIFERA, Gnaphalium gayanum, GNAPHALIUM VIRAVIRA, Gochnatia foliolosa var. fascicularis, Gochnatia foliolosa var. foliolosa, Grindelia glutinosa, Gutierrezia resinosa, Gypothamnium pinifolium, HAPLOPAPPUS CHRYSANTHEMIFOLIUS, Haplopappus deserticola, Haplopappus foliosus, HAPLOPAPPUS ILLINITUS, Haplopappus integerrimus, Haplopappus multifolius, HAPLOPAPPUS PINEA, Haplopappus schumannii, Haplopappus scrobiculatus, Haplopappus velutinus, HELENIUM AROMATICUM, Helogyne macrogyne, Hypochaeris scorzonerae, Leucheria floribunda, Leucheria rosea, Lophopappus tarapacanus, Madia sativa, Moscharia pinnatifida, Mutisia acerosa, Mutisia acuminata, MUTISIA ARAUCANA, Mutisia ilicifolia, Mutisia sinuata, Nardophyllum lanatum, Nassauvia cumingii, Nassauvia lagascae, Ophryosporus paradoxus, Oxyphyllum ulicinum, Pachylaena atriplicifolia, PARASTREPHIA LUCIDA, Parastrephia quadrangularis, Perezia multiflora, Perityle emoryi, Plazia daphnoides, Podanthus mitiqui, Polyachyrus poeppigii, PROUSTIA CUNEIFOLIA, PROUSTIA ILICIFOLIA FMA, BACCHAROIDES, SENEIO ADENOPHYLLUS, Senecio anthemidiphyllus, Senecio behnii, Senecio bipontini, Senecio bridgesii, Senecio candollii, Senecio chilensis, Senecio chionophilus, Senecio clarioneifolius, SENEIO DRIOPHYLLUS, Senecio eruciformis, Senecio fistulosus, Senecio hollermayeri, SENEIO HUMILLIMUS, SENEIO ILLAPELINUS, Senecio leucus, Senecio lorentziella, Senecio microphyllus, SENEIO MURINUS, Senecio nutans, SENEIO OLIVACEOBRACATEATUS, Senecio otaeguianus, Senecio otites, Senecio polygaloides, SENEIO</i>

	<i>PUCHII</i>, <i>Senecio reicheanus</i>, <i>SENECIO SUBULATUS</i>, <i>Senecio tinctolobus</i>, <i>Senecio tristis</i>, <i>Senecio zoellneri</i>, <i>Solidago chilensis</i>, <i>Tagetes multiflora</i>, <i>Trichocline aurea</i>, <i>Trixis cacaloides</i>, <i>VIGUIERA PAZENSIS</i>, <i>Werneria glaberrima</i>, <i>Werneria pinnatifida</i>, <i>XENOPHYLLUM WEDDELLII</i>
Atherospermataceae	<i>Laurelia sempervirens</i>, <i>Laureliopsis philippiana</i>
Berberidaceae	<i>Berberis chillanensis</i>, <i>Berberis empetrifolia</i>, <i>Berberis microphylla</i>, <i>Berberis montana</i>
Bignoniaceae	<i>Argyria radiata</i>
Boraginaceae	<i>Cryptantha aprica</i> , <i>Cryptantha diffusa</i> , <i>Cryptantha involucrata</i> , <i>Heliotropium floridum</i> , <i>Heliotropium linariaefolium</i> , <i>Heliotropium megalanthum</i> , <i>Heliotropium myosotifolium</i> , <i>Tiquilia atacamensis</i> , <i>Tiquilia paronychioides</i>
Brassicaceae	<i>DESCURAINIA PIMPINELLIFOLIA</i>, <i>MENONVILLEA CUNEATA</i>, <i>Schizopetalon rupestre</i>, <i>Sisymbrium andinum</i>, <i>Sisymbrium lanatum</i>
Bromeliaceae	<u><i>PUYA COERULEA</i></u>
Buddlejaceae	<i>BUDDLEJA GLOBOSA</i>, <u><i>BUDDLEJA SUAVEOLENS</i></u>
Calyceraceae	<i>Gamocarpa alpina</i> , <i>Nastanthus caespitosus</i>
Campanulaceae	<i>Lobelia polyphylla</i>
Caryophyllaceae	<i>Pycnophyllum bryoides</i> , <i>Pycnophyllum molle</i> , <i>Pycnophyllum spathulatum</i> , <i>Silene andicola</i>
Celastraceae	<i>Maytenus boaria</i> , <i>Maytenus disticha</i>
Chenopodiaceae	<i>ATRIPLEX DESERTICOLA</i>, <u><i>ATRIPLEX MUCRONATA</i></u>
Cunoniaceae	<i>Caldcluvia paniculata</i> , <i>Weinmannia trichosperma</i>
Cupressaceae	<i>Austrocedrus chilensis</i>
Cuscutaceae	<i>Cuscuta chilensis</i>
Cyperaceae	<i>Carex andina</i>
Desfontainiaceae	<i>DESFONTAINEA SPINOSA</i>
Elaeocarpaceae	<i>Aristotelia chilensis</i>
Ephedraceae	<i>Ephedra breana</i> , <i>Ephedra chilensis</i>
Ericaceae	<i>Gaultheria pumila</i>
Euphorbiaceae	<i>ADENOPELTIS SERRATA</i> , <i>Colliguaja integerrima</i> , <i>Colliguaja odorifera</i> , <i>Colliguaja salicifolia</i> , <i>Euphorbia collina</i> , <i>Euphorbia portulacoides</i>
Fabaceae	<i>Acacia caven</i> , <i>ADESMIA AEGICERAS</i> , <u><i>ADESMIA ARGENTEA</i></u> , <i>Adesmia aspera</i> , <u><i>ADESMIA ATACAMENSIS</i></u> , <i>Adesmia echinus</i> , <i>Adesmia glutinosa</i> , <i>Adesmia hystrix</i> , <i>Adesmia jilesiana</i> , <i>Adesmia longipes</i> , <i>ADESMIA PEDICELLATA</i> , <i>ADESMIA RETUSA</i> , <i>ADESMIA SPUMA</i> , <i>Adesmia subterranea</i> , <i>Anarthrophyllum andicola</i> , <i>Anarthrophyllum cumingii</i> , <i>Anarthrophyllum gayanum</i> , <i>Astragalus coquimbensis</i> , <i>ASTRAGALUS CRUCKSHANKSII</i> , <i>ASTRAGALUS LOOSERI</i> , <i>Balsamorhiza brevifolia</i> , <i>Caesalpinia angulata</i> , <i>Calliandra chilensis</i> , <i>Dalea pennellii</i> , <i>Hoffmanseggia ternata</i> , <i>Lupinus microcarpus</i> , <i>LUPINUS SUBCAULIS</i> , <i>Otholobium glandulosum</i> , <i>SENNA BIROSTRIS</i> , <u><i>SENNA CANDOLLEANA</i></u> , <u><i>SENNA CUMINGII</i></u> , <i>Sophora macrocarpa</i> , <i>VICIA NIGRICANS</i>
Fagaceae	<i>Nothofagus alessandrii</i> , <i>Nothofagus alpina</i> , <i>Nothofagus antarctica</i> , <i>Nothofagus betuloides</i> , <i>Nothofagus dombeyi</i> , <i>Nothofagus glauca</i> , <i>Nothofagus macrocarpa</i> , <i>Nothofagus nitida</i> , <i>Nothofagus obliqua</i> , <i>Nothofagus pumilio</i>
Flacourtiaceae	<i>Azara integrifolia</i> , <i>Azara lanceolata</i> , <i>Azara microphylla</i> , <i>Azara petiolatis</i> , <i>Azara serrata</i>
Frankeniaceae	<i>Frankenia chilensis</i>
Geraniaceae	<i>Geranium sessiliflorum</i>
Gesneriaceae	<i>Mitraria coccinea</i>
Gunneraceae	<i>Gunnera tinctoria</i>
Hydrophyllaceae	<i>Phacelia cumingii</i> , <i>Phacelia pinnatifida</i> , <i>Phacelia secunda</i>

Iridaceae	<u>SISYRINCHIUM CUSPIDATUM</u>, <u>SOLENOMELUS PEDUNCULATUS</u>
Krameriaceae	<i>Krameria cistoidea</i>
Lamiaceae	<i>Satureja gilliesii</i> , <i>Sphacele salviae</i> , STACHYS ALBICAULIS , <i>Stachys grandidentata</i> , <i>Stachys philippiana</i> , <u>TEUCRIUM BICOLOR</u>
Lauraceae	<i>Cryptocarya alba</i> , <i>Persea lingue</i>
Ledocarpaceae	<i>Balbisia microphylla</i> , <i>Balbisia peduncularis</i> , <i>Wendtia gracilis</i>
Loasaceae	<i>Caiphora coronota</i> , <u>LOASA HETEROPHYLLA</u> , <u>LOASA PALLIDA</u> , <i>Loasa tricolor</i>
Lythraceae	<i>Pleurophora pungens</i>
Malesherbiaceae	<i>Malesherbia lanceolata</i> , <i>Malesherbia linearifolia</i>
Malvaceae	<i>Cristaria glaucophylla</i> , <i>Cristaria integerrima</i> , <i>Cristaria viridilutiola</i> , <i>Nototriche compacta</i> , TARASA OPERCULATA
Monimiaceae	<i>Peumus boldus</i>
Myrtaceae	<i>Luma apiculata</i> , <i>Luma chequen</i> , <i>Myrceugenia planipes</i> , <i>Myrcianthes coquimbensis</i>
Nolanaceae	<i>Nolana crassulifolia</i> , <i>Nolana incana</i> , <u>NOLANA LEPTOPHYLLA</u> , <u>NOLANA PARADOXA</u> , <i>Nolana peruviana</i> , <u>NOLANA SALSOLOIDES</u> , <i>Nolana sedifolia</i>
Nyctaginaceae	<i>Mirabilis elegans</i>
Onagraceae	<i>Epilobium ciliatum</i> , <i>Epilobium glaucum</i> , <i>Fuchsia lycioides</i> , <i>Fuchsia magellanica</i> , <i>Gayophytum micranthum</i> , <i>Oenothera acaulis</i> , <i>Oenothera coquimbensis</i> , <i>Oenothera picensis</i>
Oxalidaceae	<i>Oxalis gigantea</i> , <i>Oxalis lineata</i> , <i>Oxalis magellanica</i> , <i>Oxalis squamata</i>
Philesiaceae	<u>LUZURIAGA POLYPHYLLA</u>
Podocarpaceae	<i>Podocarpus saligna</i> , <i>Prumnopitys andina</i> , <i>Saxegothaea conspicua</i>
Polemoniaceae	<i>Gilia crassifolia</i>
Polygonaceae	<i>Muhlenbeckia hastulata</i>
Portulacaceae	CALANDRINIA COMPACTA , <i>Cistanthe grandiflora</i> , <i>Montiopsis potentilloides</i> , <i>Montiopsis sericea</i>
Proteaceae	<i>Embothrium coccineum</i> , <i>Gevuina avellana</i> , <i>Lomatia dentata</i> , <i>Lomatia ferruginea</i>
Rhamnaceae	<i>Colletia hystrix</i> , <u>DISCARIA TRINERVIS</u> , <u>RETANILLA TRINERVIA</u> , <i>Trevoa quinquinervia</i>
Rosaceae	<i>Acaena magellanica</i> , <i>Acaena pinnatifida</i> , <i>Acaena splendens</i> , <i>Kageneckia angustifolia</i> , <u>KAGENECKIA OBLONGA</u> , <i>Polylepis rugulosa</i> , <i>Polylepis tarapacana</i> , <i>Quillaja saponaria</i> , <i>Tetraglochin alatum</i> , <i>Tetraglochin cristatum</i>
Rubiaceae	<u>CRUCKSHANKSIA PALMAE</u> , <u>CRUCKSHANKSIA PUMILA</u> , GALIUM SUFFRUTICOSUM
Salicaceae	SALIX HUMBOLDTIANA
Santalaceae	<i>Arjona patagonica</i> , <i>Quinchamalium chilense</i>
Sapindaceae	BRIDGESIA INCISIFOLIA , <i>Guindilia trinervis</i> , <i>Llagunoa glandulosa</i>
Saxifragaceae	ESCALLONIA ALPINA , <u>ESCALLONIA ILLINITA</u> , <i>Escallonia myrtoidea</i> , <u>ESCALLONIA PULVERULENTA VAR. GLABRA</u> , <u>ESCALLONIA PULVERULENTA VAR. PULVERULENTA</u> , <u>ESCALLONIA RUBRA</u> , <i>Ribes cucullatum</i> , <i>Ribes gayanum</i>
Scrophulariaceae	ALONSOA MERIDIONALIS , <i>Calceolaria ambigua</i> , <i>Calceolaria andina</i> , <i>Calceolaria arachnoidea</i> , <i>Calceolaria cana</i> , <i>Calceolaria filicaulis</i> , <i>Calceolaria hypericina</i> , <i>Calceolaria integrifolia</i> , <i>Calceolaria lanceolata</i> , <i>Calceolaria petiolaris</i> , <i>Calceolaria pinifolia</i> , <i>Calceolaria polyfolia</i> , <i>Calceolaria purpurea</i> , <i>Calceolaria segethii</i> , <i>Calceolaria thyrsoflora</i> , <i>Euphrasia flavicans</i> , MELOSPERMA ANDICOLA , <u>MIMULUS DEPRESSUS</u> , <i>Mimulus luteus</i>
Solanaceae	<i>Cestrum parqui</i> , <i>Dunalia spinosa</i> , <i>Lycium chilense</i> , <i>Lycium deserti</i> , <i>Nicotiana acuminata</i> , <i>Nicotiana undulata</i> , <i>Phrodus microphyllus</i> , <u>REYESIA JUNIPEROIDES</u> , <i>Schizanthus candidus</i> , <i>Schizanthus grahamii</i> , <i>Schizanthus hookeri</i> , <i>Solanum chilense</i> , <i>Solanum ligustrinum</i> , <u>SOLANUM MARITIMUM</u>
Thymelaeaceae	OIDIA ANDINA

Urticaceae	<i>URTICA MOLLIS</i>
Valerianaceae	<i>VALERIANA FLORIBUNDA</i> , <i>VALERIANA STRICTA</i>
Verbenaceae	<i>ACANTHOLIPPIA DESERTICOLA</i> , <i>Glandularia origenes</i> , <i>Junellia arequipensis</i> , <i>JUNELLIA CONNATIBRACTEATA</i> , <i>Junellia minima</i> , <i>Junellia selaginoides</i> , <i>Junellia uniflora</i> , <i>Rhaphithamnus spinosus</i> , <i>Verbena gynobasis</i> , <i>VERBENA RIBIFOLIA</i>
Violaceae	<i>Viola frigida</i> , <i>Viola volcanica</i>
Vivianiaceae	<i>Viviania marifolia</i>
Winteraceae	<i>Drymis winteri</i>
Zygophyllaceae	<i>FAGONIA CHILENSIS</i> , <i>PORLIERIA CHILENSIS</i>

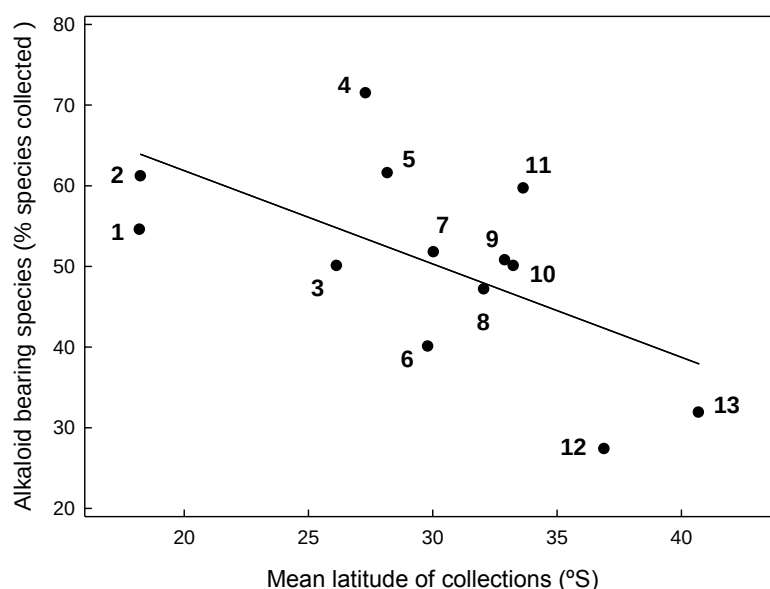


FIGURE 1

Effect of latitude on the proportion of alkaloid bearing plants, in 13 biogeographical subregions of Chile: 1 = prealtiplanic scrubby steppe, 2 = high Andean altiplanic steppe, 3 = coastal desert of TalTal, 4 = flowering desert of the plains, 5 = coastal desert of Huasco, 6 = high Andean steppe of Coquimbo, 7 = coastal scrubby steppe, 8 = Andean sclerophyllous scrubland, 9 = high Andean steppe of Santiago, 10 = coastal sclerophyllous forest, 11 = Andean sclerophyllous forest, 12 = deciduous high Andean forest of Chillán, and 13 = evergreen forest of the lakes region (Gajardo, 1994). The figure included only subregions where 10 or more species were collected; total number of species included was 347. Pearson correlation: $R = -0.613$, $N = 13$, $P = 0.026$.

The effect of latitude was also tested using alkaloid concentration instead of alkaloid presence or absence. The correlation between alkaloid content and latitude for all collected species was statistically significant (Pearson correlation: $R = -0.122$, $N = 396$, $P = 0.015$); however, the proportion of variation explained was extremely low (ca. 1%). On the other hand, a

correlation similar to that of figure 1 using mean alkaloid concentration instead of proportion of alkaloid bearing species and mean collection latitude of sets of species from different vegetational subregions did not reach the level of significance (Pearson correlation, $R = -0.431$, $N = 13$, $P = 0.141$). These correlations may be confounded by the fact that

alkaloid concentration is a function of lineage both at family and genus levels (ANOVA using families in which more than one species was analyzed: $H = 138.81$, g.l.= 50, $P < 0.001$; ANOVA using genera in which more than one species was analyzed: $H = 147.03$, g.l.= 74, $P < 0.001$). The problem of lineage

may be avoided if pairs of samples of a given species collected at different latitudes are analyzed. Table 2 shows that in 12 out of the 13 cases studied, the alkaloid concentration was higher in the sample collected at lower latitude.

TABLE 2
Comparison of alkaloid content in species collected at two different latitudes.

Species	Family	Collection 1		Collection 2	
		Latitude (°S)	Alkaloids (mg/g)	Latitude (°S)	Alkaloids (mg/g)
<i>Adesmia aegiceras</i>	Fabaceae	29.80	0.78	31.76	0.95
<i>Anarthrophyllum cumingii</i>	Fabaceae	32.83	0.30	33.34	0.15
<i>Baccharis linearis</i>	Asteraceae	30.00	0.85	33.73	0.25
<i>Berberis microphylla</i>	Berberidaceae	36.92	0.73	40.78	0.38
<i>Berberis empetrifolia</i>	Berberidaceae	32.83	0.36	36.93	0.10
<i>Encelia canescens</i>	Asteraceae	28.14	0.95	30.03	0.47
<i>Escallonia alpina</i>	Saxifragaceae	36.92	1.15	40.78	0.67
<i>Fuchsia lycioides</i>	Onagraceae	31.46	0.07	33.27	0.04
<i>Guindilia trinervis</i>	Sapindaceae	31.80	0.55	33.31	0.23
<i>Nolana crassulifolia</i>	Nolanaceae	27.19	0.04	33.27	0.01
<i>Nolana paradoxa</i>	Nolanaceae	29.07	0.67	33.27	0.56
<i>Schizanthus grahamii</i>	Solanaceae	34.96	0.97	36.92	0.38
<i>Senecio zoellneri</i>	Asteraceae	18.11	0.18	18.33	0.01

The study in which low-latitude-country floras showed higher proportions of alkaloid containing species (Levin, 1976) was supported by the analysis of the native Chilean flora at the ecoregion level; moreover, the idea was shown to apply to alkaloid concentration in addition to presence or absence data, under the limitations imposed by the relationship between alkaloid concentration and species lineage. In practical terms, these results indicate that in studies aimed at identifying alkaloids in the native flora of Chile, plant collections should take into consideration the promising species listed in Table 1 and take place at the lowest possible latitude if the chance of finding alkaloids and the yield of isolated alkaloids are to be maximized.

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