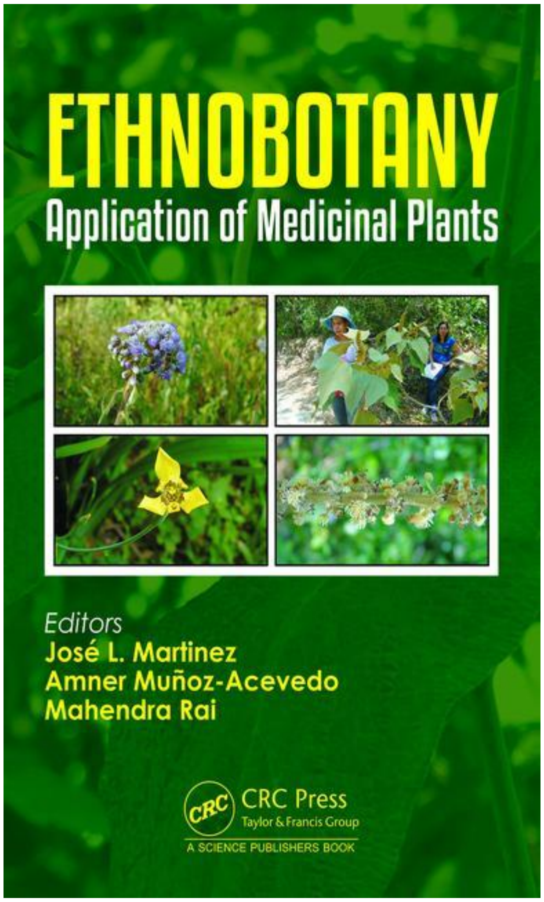

Ethnobotany: Application of Medicinal Plants

1st Edition

José L. Martinez, Amner Muñoz-Acevedo, Mahendra Rai.

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Summary

Ethnobotany includes the traditional use of plants in different fields like medicine and agriculture. This book incorporates important studies based on ethnobotany of different geographic zones. The book covers medicinaland aromatic plants, ethnopharmacology, bioactive molecules, plants used in cancer, hypertension, disorders of the central nervous system, and also as antipsoriatic, antibacterial, antioxidant, antiurolithiatic. The book will be useful for a diverse group of readers including plant scientists, pharmacologists, clinicians, herbalists, natural therapy experts, chemists, microbiologists, NGOs and those who are interested in traditional therapies.

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Paramela (*Adesmia boronioides* Hook.f.): From Popular Uses to Commercialization

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Introduction

Paramela (*Adesmia boronioides* Hook. f.) is a species with a long history of use among the native societies which inhabited in the past and inhabit today the Argentinean-Chilean Patagonia. Due to its cultural and symbolic value, it stands out as part of the biocultural heritage of the region. It is part of the knowledge and practices related mainly with the health and subsistence of Mapuche and Tehuelche communities since pre-hispanic times (Molares & Ladio, 2009a; Ciampagna & Caparelli, 2012). It is a species used for human consumption as medicine (Martínez-Crovetto, 1980; Campos et al. 1997; Montes & Milkomirsky, 2001) as well as ornamental and melliferous (Forcone & Muñoz, 2009; Green & Ferreyra, 2011).

Recently, it has raised an increasing commercial interest given its exceptional conditions and potentialities, specially due to its fragrant odor (Montes & Peltz, 1963; Bandoni et al., 2000). There is a clear distinction in views between rural areas where paramela is mainly used as a medicinal infusion, and urban areas where it is used as ornament and / or as an aromatic ingredient for the preparation of an alcoholic beverage. Since 2005, its essential oil has been used as a supply for the perfume industry.

This native plant of the Patagonian region inhabits low irrigation sites, is of slow growth, and its culture is of interest (Contardi et al., 2016, a, b). So far it is almost exclusively found in its natural state (Barthelemy et al., 2008). However, in a great part of the rural communities, mainly those of Mapuche-Tehuelche ascendance, the paramela is protected in family orchards either because it is tolerated and cared for with the rest of the plants or because it has been transplanted to these spaces, thus being possible to place it in an incipient domestication process (Ladio & Morales, 2017).

A successful and sustained development of products from native plants requires a domestication process of the species, starting from an improvement of the raw materials and allowing a standardization in the active principles' contents. Obtaining homogenous genetic material and quality plants is crucial for its production, processing and usage. Reproduction studies made from *A. boronioides* seeds, allowed for the development of the species propagation protocols and the production of plants in greenhouses (González et al., 2009; Sánchez et al., 2012; y Mazzoni et al., 2013). Since 2015, an experimental culture in the Andean region of the Argentinean Patagonia allows for the evaluation of the productivity and quality of the cultivated plant's essential oil in relation to the wild population.

Botanical and ecological information of the species

Botanical description

A. boronioides is a perennial bush belonging to the Leguminosae family (Fabaceae). Its common name is "paramela". Its habitat ranges from 0 to 2,200 m a.s.l. It has a medium size, varying between 0.40-2 m. It is highly ramified, with glandular branches, fragrant and very resinous and sticky to the touch. It has an axonomorph root. The leaves are ± 3-6 cm, shortly petiolate, 10-20-leaflets, leaf rachis with erect, brief hair; obovate, fleshy, glabrous, toothed, shiny, 4-6 mm leaflets with crateriform glands especially in the edge; short, amplexicaul, glabrous, glandular stipules. Clusters of 4-7 cm, densiflorous, sessile, ovate, acute, glandular, glabrous bracts. The flowers are 7-10 mm, colorful, yellow, perfumed, with campanulate chalice, pubescent, glandulous, with short teeth, serice-pubescent in their interior; glabrous vexil (banner), glabrous wings and keel shorter than vexil. Ovary with some marginal hair. Narrow, pubescent, glandular, 3-5-articulate loments; semicircular, dehiscent 4.5-6 mm trusses.

Figure 1: Detail of the glands in the bundle and the back of the leaflets (10x)

Its anatomy was initially studied by Nájera et al. (2000). Subsequently, in an evaluation of leaves and stems of diverse origins in their Patagonian distribution, very similar microscopic characters were found. They all have cyclocytic stomata on the leaves' epidermis. The secretory pores are located, in greater number, on the abaxial surface, although some pores can be observed on the adaxial surface. Secretion pores at the end of each leaf semilimbo were found on some samples (Los Antiguos, El Calafate, Bariloche, Villa La Angostura) (González et al., 2014).

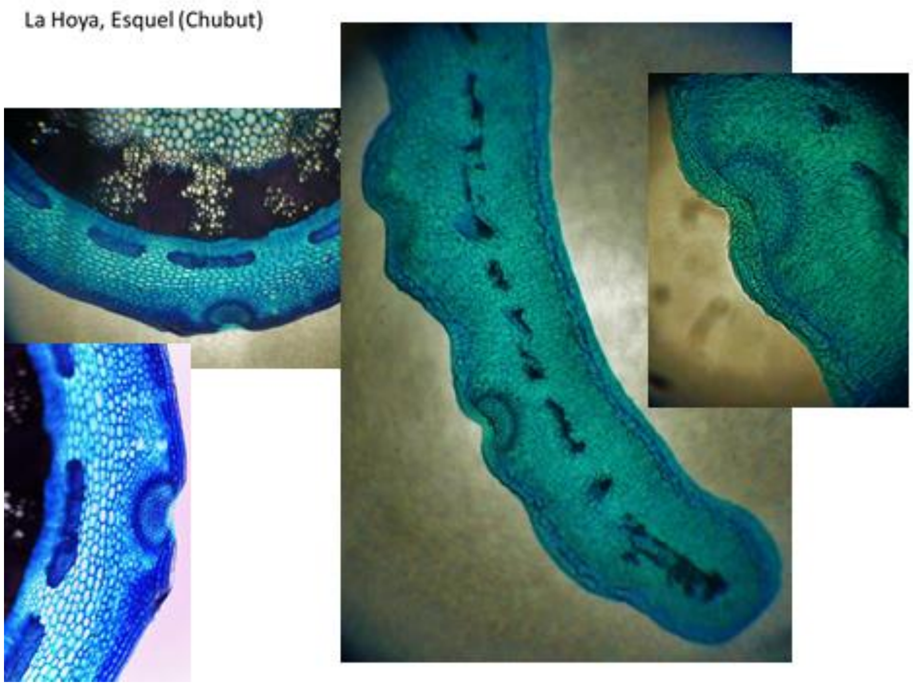

Figure 2: Microscopic structure of the glandular structures in stems and leaves (40x).

Geographic distribution

A. boronioides's distribution ranges from Mendoza to Tierra del Fuego provinces, in Argentina, including also Neuquén, Rio Negro, Chubut, and Santa Cruz, and the XI and XII Regions of Chile (http://www2.darwin.edu.ar/Proyectos/FloraArgentina/fa.htm, Burkart, 1997; Ulibarri & Burkart, 2000).

It inhabits sunny areas, shrubs, river sides, roads and ravines, mainly in the Patagonian steppe, shrubland areas and steppe-forest transition zones (https://www.sib.gov.ar/ficha/PLANTAE*adesmia*boronioides). The forest areas where paramela can be found consist mainly of *lenga* (Nothofagus pumilio), *ñire* (Nothofagus antarctica) and cypress (Austrocedrus chilensis) (Molares & Ladio, 2012b). It has also been found in the atlantic littoral, in Santa Cruz province, in the Rio Gallegos area (González et al., 2014). In slopes, near Los Molles, Mendoza, it grows in shrubland vegetation (Fortunato, com. pers.).

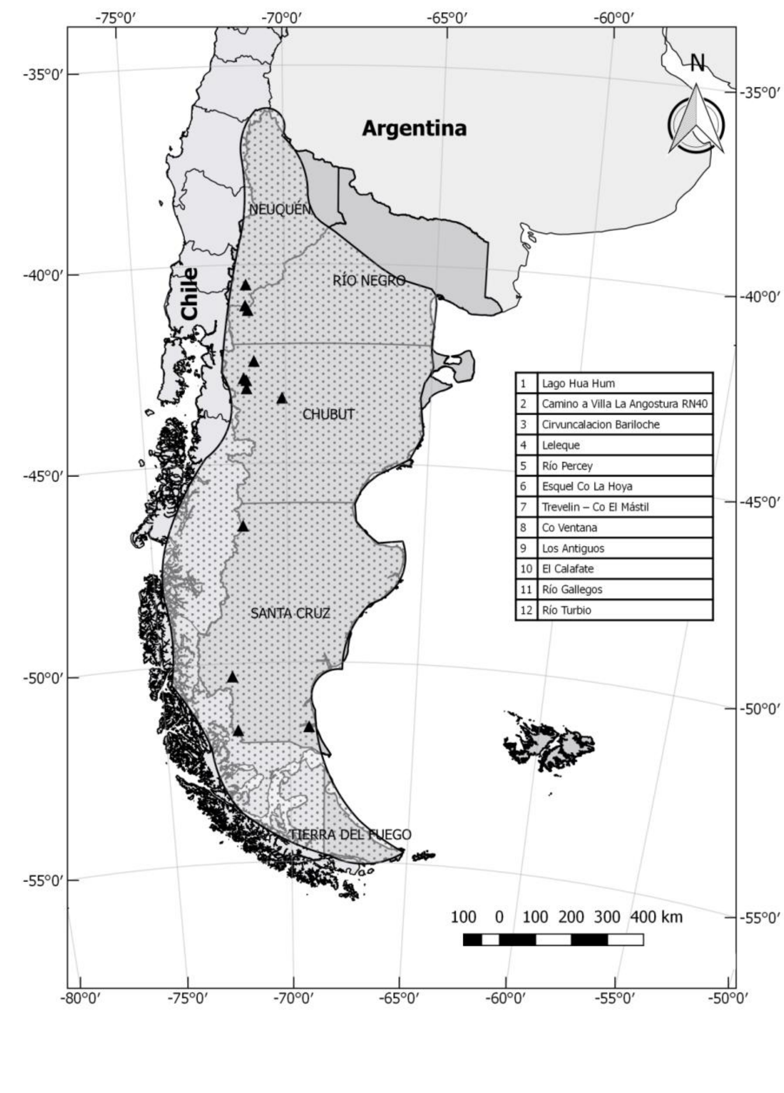

Figure 3: *A. boronioides* distribution map.

Generalities of the Adesmia genus

The *Adesmia* DC genus is exclusively South American. It has approximately 240 species distributed mainly in centre of Chile and south and west of Argentina. In our country, more than 100 representatives have been quoted, which makes it the most numerous genus of the Leguminosae, Papilionoideae. There are about 55 species in the Patagonian region (Burkart, 1984; Ulibarri & Burkart, 2000; Ulibarri & Simpson 2010).

A. boronioides stands out as the only species of this highly glandulous-resinous genus (Burkart, 1997).

Background of human use

Its common name is paramela, but it is also known as *té silvestre* (wild tea), *yerba carmelita* (Carmelite herb), *éter* (ether), *pega pega*, *yagneu* and *lonkotrevo* (González, 2002; Molares & Ladio, 2012a, b). Native people of Patagonia value this plant greatly, and have used it with both medical and symbolic purposes. Its use must be framed in a holistic health system in which different participants, elements and unique worldviews that have no equivalent in Western science are involved (Ladio & Molares, 2014; 2017). For this reason, a "translation" of certain uses from a Western perspective must be done carefully since they could be oversimplified or interpreted as therapeutically inaccurate.

This plant is used as medicine by a large part of the Patagonian rural population. Its medicinal use has been registered in ethnobotanical works by communities very distant from each other (Neuquén, Rio Negro and Chubut provinces, as well as the south regions of Chile) (González et al., 2005; Molares & Ladio, 2009a). In most studies conducted in Mapuche-Tehuelche communities, the species has high levels of consensus among inhabitants, in many cases greater than 75%, that is to say, their knowledge and use of the plant is widely shared and disseminated among the rural population (Ladio, 2006; Molares & Ladio, 2014).

The species has been most recurrently cited as digestive, antirheumatic, diaphoretic and antiemetic (Campos et al., 1997; González et al., 2004, 2005; Toledo & Kutschker, 2012; Molares & Ladio, 2014). Today, it is a substantial part of these communities' home medicine, being part of the first-aid kit of rural families that use it to cope with health problems autonomously (Richieri et al., 2010). Traditionally, it is collected in autumn-winter, while the animals are taken care of, and kept dry in paper bags in dark places to use it as medicine during the whole year (Richeri et al., 2013). The people of the countryside distinguish it by its "perfumed" character and it is listed in the group of plants with “magical soul" and "sweet smell and bitter taste” (Molares & Ladio, 2009b; Ladio & Molares, 2014).

It has been used since ancient times by the native people of the region mainly to relieve bruises, sprains, cramps, and joint and muscle pains (González, 2002; Estomba et al., 2006; Igon et al., 2006). It is also valued for having the property of “heating the body" when prepared in steam inhalations and baths with the plant submerged in hot water (González, 2005). These baths are mainly used for children not to urinate in bed when it is cold (Igon et al., 2006). Also, the steam inhalations are used against cold and cough discomfort, in communities of Neuquén, Río Negro and Chubut provinces (Igon et al., 2006; Lys et al., 2007; Eyssartier et al., 2011; Richeri et al., 2013). According to Richeri (2016) families in the Chubut plateau look for paramela in order to help asthmatic people or for bronchitis cases. This author also records the use of the plant as incense (burning) to perfume and cleanse the houses of evil spirits. According to Ochoa (2005), the species is also used in ointment as a sedative for rheumatic problems and to heal wounds.

The tea or infusion of paramela is widely used as a digestive (Igon et al., 2006). It is also included in the *mate*, a traditional drink of the region that is mainly composed by *mate* herbs (Ilex paraguariensis) (Weigandt et al., 2004). Its digestive use is the most important reputed use for which it is marketed in businesses dedicated to the sale of medicinal plants in S.C. de Bariloche (Cuassolo, 2009). The infusion is also used to wash the hair and kill lice. It is considered to strengthen the hair (Martínez-Crovetto, 1980, Conticello et al., 1997, Igon et al., 2006). Ochoa (2005) describes that, in Arroyo Las Minas (Rio Negro), the infusion is used for kidney problems, and women drink the infusion after childbirth to recover faster, as an invigorating drink. The use for kidney problems has also been cited by Kutschker et al. (2002). Some studies have also identified it as an aphrodisiac (Igon et al., 2006, Muñoz et al., 2001). In Comallo and Pilcaniyeu (Rio Negro), people emphasize the use of the plant’s decoction to ease the flu, stomach pains, diarrhea, and fever (Eyssartier et al., 2009; 2011; 2013).

Different ways of preparing paramela: popular uses

Infusion (tea): In most local recipes it is prepared using a tablespoon of leaves per cup. The dose is one cup per day.

Mate: Fresh/dry leaves are added to the mate infusion.

Decoction or cooking: The twigs are boiled in water and eaten with burnt sugar or alone.

Plaster: The leaves are crushed and embedded in a gauze or rag that is placed over the sore area.

Baths and steam inhalations: A large bowl is filled with hot water and branches of the plant are added.

Incense: Branches are placed on the kitchen stove or burned slowly to release its fragrance.

Other local uses

It is cited as an excellent forage (Green & Ferreyra, 2011). In his work on the Chubut plateau, Castillo (2014) found that it is a plant used as forage for goats and sheep, even though it is considered by the shepherds as a problematic plant, as it gives a bad taste to the meat of the animal that eats it. Also, Green and Ferreyra (2011) registered its moth repellent properties.

Chemical constituents

The first chemical analysis of essential oils and resinoids obtained from leaves and stems was made in 1963 by Dr. A.L. Montes (Montes, 1963). He described physicochemical properties of the essential oils and chromatograms, but without mentioning any specific compounds due to the instrumental limitations of the time.

Agnese et al. (1989; 1992; 1993; 1995) made phytochemical studies on some *Adesmia* species (*A. grandiflora, A. bicolor, A. retrofracta, A. trijuga, A. horrida, A. incana, A. aegiceras*). They analysed hydrophilic compounds including flavonoids, pinitol, vainillin and glucose, and some lipophilic compounds such as alkanes, carboxylic acids and wax.

Faini et al. (1995) reported three new triterpene glycosides of the malabaricane type in *A. aconcaguensis*.

Essential oil composition of *A. boronioides*

Essential oil yields obtained by hydrodistillation are 0.5% in average. This oil is yellow-greenish with a pleasant sweet odor, specific gravity 0,9690 and refraction index 1,4972 at 20°C, [α]= +6 (0,02; hexane). The essential oil has a high content of sesquiterpenoids. The main compounds have a novel bisnor sesquiterpene structure (González et al. 2002) and were named Esquelenona and Isoesquelenona . The other important compounds have cadinane and eudesmane skeletons, the first one belonging to α-copaen-11-ol and the other to Eudesmol-10-epi-γ.

The relative amounts of main components is directly influenced by the recollection sites, phenological stage, drying time, season, among others.

The main compound might derive biogenetically from MW 236, an intermediary compound, precursor of furopelargones, proposed by Lukas et al. (1964). In turn, this hypothetical structure might derive from a guaiane skeleton (6,9-guaiadiene ; Fig. 4).

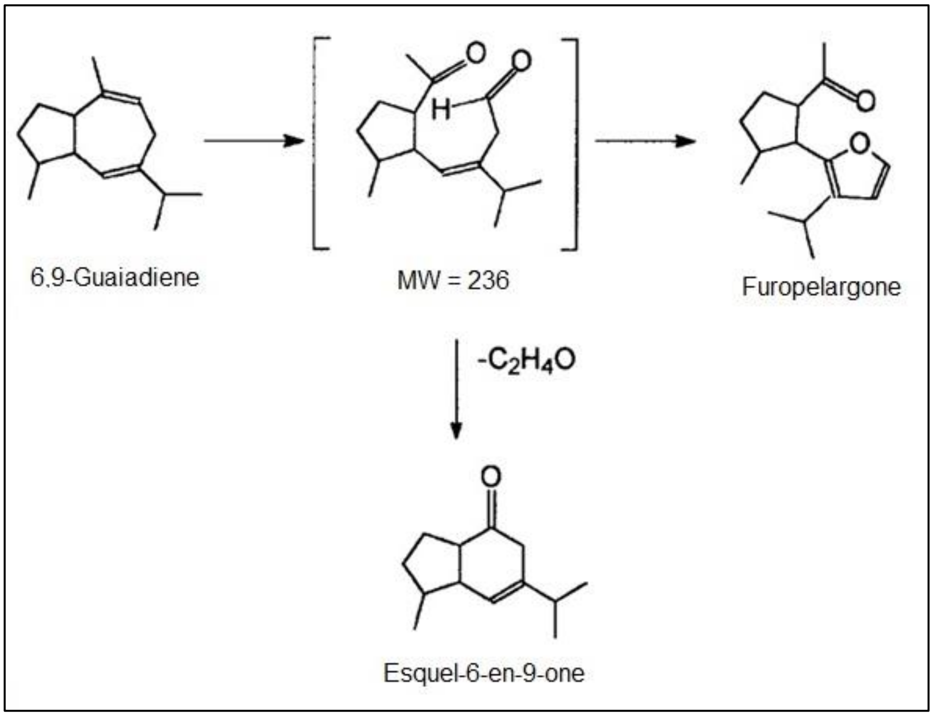

Figure 4:Esquelenone proposed biosynthesis

The olfactory profile of this essential oil is uncommon and interesting, with a sweet, woody and spiced odor, with strong tenacity and fixation properties. (González et al. 2002).

Structural elucidation of the main compound was performed through NMR (mono and bidimensional), vibrational circular dichroism (VCD) and its absolute configuration was recently reassigned (González et al. 2002; Cerda-García-Rojas et al. 2015).

Essential oil composition includes not only novel structural components (Fig. 5) but also new terpene skeletons not reported previously in other species (González et al., 2004).

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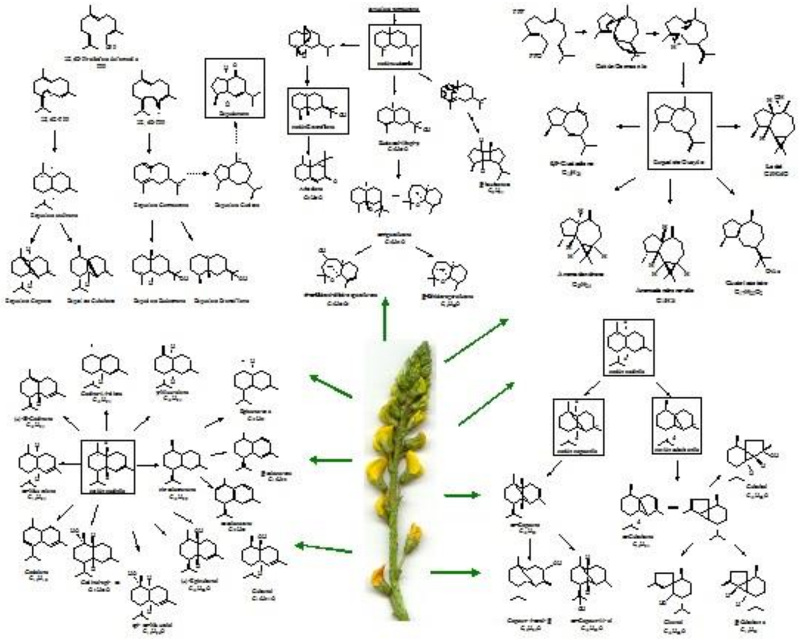

Figure 5: Main sesquiterpenes skeletons in *A. boronioides's* essential oil (González, 2002).

The main component structure is a bisnor sesquiterpene called "esquelenona", named after the city Esquel, in Patagonia Argentina, where paramela grows naturally.

Taking into account the stereochemistry of the compound, the name is (1S,4R,5S)-esquel-6-en-9-ona, whereas the IUPAC name is (3aS,1R,7aS)-1,2,3,3a,5,7a-hexahydro-1-methyl-6-(1-methylethyl)-4H-inden-4-one.

The essential oil, some extracts and the main component, esquelenona, all have nice, fruity odor suitable for perfumery production. A dermal sensitivity test showed it is harmless to the skin (González, 2002). Also, some essays showed high stability of the essential oil at room temperature, even with solar exposition. These findings are promising in relation to the possibility of using this natural product in fragrance industries. When the essential oil was exposed to heat and air, the esquelonena compound went through a process of isomerization and oxidation, respectively, which affected the fresh and fruity odor.

The esquelenona compound shares structural features with products used currently in the perfumery industry such as 6,7-dihydro-1,1,2,3,3-pentamethyl 4-(5H)-indanona (Cashmeran®).

Chemical variability in the essential oil of *A. boronioides*

A study of the volatile compounds chemical variability along the Patagonian distribution was conducted, including Neuquén, Río Negro, Chubut and Santa Cruz provinces in Argentina.

Contents and regulation of secondary metabolites are very sensitive to environmental influences and the presence of pathogens and predators. The chemical variability in natural populations of paramela in Patagonia has been confirmed. It is specially remarkable the hemiterpene concentration in the most southern sites and the quantitative difference of cadinane skeletons which are preponderant in some sites (La Hoya, Chubut 41% in 2015) and almost absent in others (Los Antiguos, Santa Cruz 0 to 3% in 2013-2015). Esquelanes/Guaianes are quantitatively important in all sites and years studied, varying the percentage of total essential oils contents from 27,4% to 60,4%. (González et al. 2016). Low molecular weight compounds were detected in Santa Cruz, Los Antiguos and El Calafate. The most remarkable of these compounds is 2-methylbutanenitrile, precursor of cyanogenic glycosides, present in numerous species of plants and insects (González et al. 2014).

These findings show the importance of establishing the origin of the plant material in order to assure the essential oil quality.

Cyanogenic glycosides

The most southern populations of *A. boronioides* analyzed (Los Antiguos y el Calafate) were the only ones which showed a positive reaction to Guignard test to detect cyanogenic glycosides. This is consistent with the presence of 2-methylbutanenitrile, lotaustralin precursor, in volatile compounds of *A. boronioides* in these locations (González et al. 2014).

According to the consulted bibliography, cyanogenic plant populations are associated with lower altitude locations, which means there is a negative correlation between altitude and cyanogenic plant number for a given species (de Araújo, 1976; Richards & Fletcher, 2002). This previous work is consistent with our findings, where the populations in altitudes lower than 300 m a.s.l showed a positive reaction in the Guignard test (Silva Sofrás, 2016).

Paramela might be a polymorphic species regarding cyanogenic glycosides production (Kakes, 1990) and its function would be related to different endogenous or exogenous factors, for example the presence of seed-feeding insects, very common in studied populations (Delfino et al. 2009).

The quantitative assay gave a maximum value of 0,47 µg de HCN/g per plant. Taking into account that the lethal dose for an adult human is 30-120 mg de HCN, it would be necessary to ingest 65 kg of this plant to reach this level. *A. boronioides* is traditionally used in infusions, with barely 5g in 100ml of hot water, and therefore it is safe for human consumption.

Phenolic compounds content

There is only one previous study regarding paramela phenolic compounds (Silva Sofrás, 2016). In this work, twelve populations from different altitudes and latitudes in Patagonia were collected and analysed in two seasons: spring and autumn. The antioxidant variability found by Gastaldi et al. (2016) is coherent with the total phenolic contents in each location.

The sites with altitudes of less than 500 m a.s.l had a higher value of total phenolic compounds in autumn and the ones above 500 m a.s.l had a higher value of flavonoids in spring.

Total phenolic contents and antioxidant activity in *A. boronioides* are higher in autumn than in spring. There was a positive correlation between the number of compounds found and the site's latitude. Seventeen (17) constituents were detected through qualitative analysis: 3 phenolic acids and 14 flavonoids.

The largest amount of compounds was detected in Rio Turbio (Santa Cruz), including a phenolic acid exclusive of this site. Three (3) flavonoids were detected in Bariloche and Villa La Angostura which are exclusive of these sites.

There was no difference in the number of compounds detected in autumn and spring for the sites analysed in both seasons.

The phenols and flavonoids qualitative analysis was coherent with total flavonoid content quantitative analysis (Silva Sofrás, 2016).

In vitro biological activity studies

Antioxidant activity

The aerial parts of the plant exhibit antioxidant activity, most likely due to the presence of phenolic compounds and flavonoids (Gastaldi et al., 2016; Silva Sofrás et al., 2016). Estomba et al. (2010) studied the antioxidant activity and pigments of *A. boronioides* with micropropagated 60-day seedlings from sterile cultured seeds. A low amount of total chlorophyll was observed with chlorophyll "a" reduction at the expense of the "b" (chlorophyll a / b: 2.98). Catalase activity (EC1.11.1.6) was low. The authors concluded in the possibility of applying these in vitro cultures as a source of bioactive metabolites.

Trypanocidal activity

The concentration of *A. boronioides* that inhibits the growth of parasites by 50% is lower than the benzimidazole IC 50, and its research in future assays is very promising. In the inhibition curve, the trend line adjusts to a polynomial equation, which would indicate that the growth of epimastigotes is affected by more than one variable. Villagra et al. (2008) concluded that new tests are required to support this hypothesis and suggested that it is possible to encourage the search for active compounds against Chagas disease from these essential oils.

Antimicrobial and antifungal activity

For in vitro susceptibility and diffusion tests on solid medium, extracts in methanol, ethyl acetate, dichloromethane, hexane and water were prepared . The extracts were challenged with microorganisms (CCMA-29: Collection of Microbial Cultures of Argentina No. 29), impregnating filter paper discs of 5.5 mm up to a total load of 0.25 mg / ml. The culture media were at two pH (Antibiotic Medium I pH 6.6 and II and pH 7.9). The antimicrobial and antifungal activities were zero under the assay conditions, except for the extract in ethyl acetate, in medium at pH 6.6, which showed activity on Staphylococcus aureus (González, 2002). In more recent studies (Blengini et al., 2016), antibacterial activity against Gram-positive bacteria of clinical importance such as Staphylococcus aureus ATCC 25923 and Enterococcus faecalis ATCC 11198 was detected, with MIC values of 62.5 μg / ml for both bacteria. These studies were conducted with an experimental technique adapted from those suggested by the Clinical Laboratory Standards Institute (CLSI), using a panel of Gram positive and negative bacteria and assaying the oil. For the antifungal evaluation the broth microdilution method was used, in front of a panel of standardized fungi. The oil showed antifungal activity against Candida glabrata and Candida parapsilosis, with MIC values of 1,000 μg / ml against both yeasts.

Anti-inflammatory activity

A methanolic extract, an infusion and the essential oil obtained by hydro distillation were assayed, according to the Argentine National Pharmacopoeia VII Edition.

The polar fractions (methanolic and aqueous) and the essential oil of *A. boronioides* were assayed at concentrations of 15 and 50 μg / ml for their eicosanoid generation effect (TBX2, PGE2 and LTB4) in rat peritoneal leukocytes. The methanolic extract and the essential oil showed a strong inhibition of LTB4 generation, whereas the aqueous extract was comparatively inactive. The methanolic extract showed potent TXB2 inhibition, but the essential oil and aqueous extract were much less active. The effects on PGE2 production were lower, implying that the greatest effect is on thromboxane synthase. The essential oil showed a significant LDH release in rat peritoneal leukocytes suggesting substantial toxicity to the cells; the other two extracts were not harmful.

Its anti-inflammatory activity has been tested *in vitro*, which would support one of the properties attributed to the plant, that is of a "remedy" for rheumatic pains (González et al., 2003).

Acute toxicity studies

The potential acute toxicity of *A. boronioides* infusion with *Artemia salina* model was recently studied. A 5% infusion was prepared from aerial parts of the plant following the standards of the Argentine National Pharmacopoeia VII Edition to make an infusion. A lyophilisate was obtained from the infusion. The *A saline* model organism was exposed to different concentrations of the lyophilisate in order to obtain concentration-response curves and to determine lethal concentrations, 50 (LC50) in mg / ml.

A LC50 $\leq$ 1mg / ml in the toxicity bioassay with *A. salina* is considered an acute toxicity indicator for an aqueous vegetal extract, extrapolable to animals and humans. The value obtained for *A. boronioides* in that assay was 5.16 mg / ml, which would indicate that the infusion of this species would not present a risk of acute toxicity for humans (Mongelli et al., 1995; Perez & Lazo 2010; Gastaldi Et al., 2016).

Allergenicity: dermal irritability test

Tests performed with *A. boronioides* essential oil included the analysis of erythema (non-pruritic rash, bright red and slightly raised skin) and edema (accumulation of interstitial fluid in large amounts) in albino rabbit skin. The results showed the safety of the essential oil under the test conditions, according to the methodology of Draize et al. (1944) and Gonzalez (2002).

Background of the species conservation state

In Argentina, the state of conservation of the paramela has not been systematically evaluated. According to IUCN its status is "not evaluated" (NE). However, in Chile it is in the red book of CONAF (Corporación Nacional Forestal, www.conaf.cl/) as a vulnerable species. In Argentina, there are no reliable studies so far, except for the fact that it is protected in all Patagonian National Parks (https://www.sib.gov.ar/ficha/PLANTAE*adesmia*boronioides). There is also no official information on the volumes that are subject to commercial exploitation to date, so we consider that their conservation status should be addressed and studied in more detail.

According to national and international standards (National Biodiversity Strategy and Plan of Action 2015-2020, IUCN and CBD), the use of goods should not affect the functioning and sustainability of ecosystems of flora, fauna, biodiversity, water, cultural values, landscape, among others, according to the criteria set forth in the special law. Consequently, in the case of commercial use of this species, it is necessary to provide the necessary mechanisms for natural resources to be used in the context of sustainable development that includes the rights and values of local communities. This is the current conservation perspective, where the protection of biodiversity is intimately related to socio-cultural and economic components through the concept of sustainable development.

Additionally, it is important to consider the protection of the different wild populations of this species, given that, due to its geographical extent, it shows morphological and chemical variations, particularly in its essential oil (González, 2016).

Commercial use

Today, the species is commercialized in three ways: 1) as an ingredient for an alcoholic beverage; 2) as a medicinal herb, mainly in infusions and decoctions; and 3) its essential oil as a raw material for perfumery. (Fig. 6)

Figure 6: Alcoholic beverage and perfume with *A. boronioides* as an ingredient.

In the first case, used in an alcoholic beverage, it is a product called Estepvka, made in El Calafate, Santa Cruz. The labeling of the product shows that its ingredients are water, alcohol and paramela. In addition, it is detailed that the alcoholic concentration is of 40% (http://www.latiendagourmet.com.ar/bebidas/espirituosas/vodka-estepvka/).

With regard to its medicinal use, there is fragmentary information about the volumes used for commercial sales, the existence of collection centers, and the number of marketing intermediaries involved. Empirical works carried out in the city of S.C. Bariloche show that it is distributed mainly dry, sometimes fresh, in very variable quantities in bulk, including branches, leaves and flowers. The main sales destinations are Patagonian herbalists, pharmacies and houses selling naturist products, where the product is fractionated in the selling points (Cuassolo, 2009). *Adesmia boronoides* appears as one of the most commercialized native medicinal plants in the urban centers of the region (Cuassolo, 2009).

Regarding its use as an essential oil, it has been used in perfumery since 2005, and to this purpose it is marketed and exported. The extraction and harvesting of the material comes only from natural populations, and, according to unverified sources, would add to more than 300 tonnes to date, processed by the distillation plants. There are audio-visual records of the recollection in Lago Buenos Aires, Santa Cruz province, where weekly truck shipments to the city of Esquel are mentioned (https://www.youtube.com/watch?v=mxSeFGrs30U).

According to the internet (https://www.iucn.org/node/16897), in Chile it is promoted and sold as the "mapuche viagra", as part of a herbal preparation called Palwen that increases sexual vigor.

In 2008, the Natura cosmetic company started commercializing a perfume with a fragrance containing essential oil of paramela. It is the "Amor América" line, inspired by plants from The Andes and Patagonia, including not only the paramela but also the *palo santo* of Ecuador (Bursera graveolens (Kunth) Triana & Planch.).

It is worth mentioning that the species has begun to be sold as ornamental in some regional fairs and nurseries of Patagonia because of the beauty of its flowers and its perennial character.

Domestication experiences and propagation

The species' background indicates that *A. boronioides* arouses much interest in companies on a commercial scale, and that the material is obtained only from natural populations.

Germination studies developed by the National University of Patagonia S.J.B (UNPSJB), INTA and the National University of Río Negro (UNRN) allowed to identify effective and practical methods of propagation. To achieve success in the germination of *A. boronioides* it is necessary to break the physical barrier with previous treatments that weaken the seed coat. From seeds collected in natural populations, pre-germination treatments at 80 ° C allow for 85% germination (González et al., 2009), and previous scarification for 83.7% germination (Mazzoni et al. 2014).

These experiences and results suggest that it would be feasible to produce large-scale seedlings to establish future crops as a productive alternative for the region.

Container cultivation was evaluated by the UNRN Tecnicatura en Viveros (Plant Nursery Technicature): a production of 120 plants in one-liter containers with substrate mixture of volcanic ash, peat and soil was obtained from seeds (Sánchez & Riat, 2012). This plant material was transplanted in 2015 to a nursery on outdoor soil, located at INTA Bariloche to continue with future evaluations (Fig. 7).

Figure 7: Natural populations and stonemasons with specimens obtained from seeds of that origin.

In addition, this exploratory cultivation test generated vegetal material that was harvested and is being characterized chemically in relation to its contents of essential oils in the UNPSJB. In this way the domestication studies carried out in the region try to make technical contributions that allow us to think about productive alternatives, reduce the collection pressure of the environment and generate products of commercial quality (Mazzoni et al., 2016, Contardi et al., 2016a, b) .

Conclusions

The previously detailed background confirms that *A. boronioides* has a long history of use as an aromatic and medicinal plant in the original Mapuche-Tehuelche communities. Consistent with the commitment made by Argentina to the Convention on Biological Diversity and the Nagoya Protocol, it is necessary to develop by-laws to regulate and protect the access to traditional knowledge associated with this plant.

To a great extent, the results of this research are promising. To date, no evidence of toxicity or allergenicity has been found in traditional forms of use in terms of preparation and dosage. However, there are no high-dose safety studies, nor for long-term use. There are chemical studies that show variability in the chemical composition (qualitative and quantitative) of the essential oil and in the content of phenols in different populations of *A. boronioides* throughout Patagonia, which means it is of major importance to analyze the plants used in the production of beverages (populations-chemotypes) and thus ensure a homogeneous quality of the composition and eventually recommend its consumption. In turn, it would be important to develop systems for quality control and authentication of the raw material used as an additive in edible products.

It is of particular interest to point out the negative consequences of unreasonable commercial exploitation of the wild populations of the species and the substantial importance of their cultivation. In the latter case, further studies are needed regarding the selection of chemotypes and their population dynamics, as well as major research on the possible management and sustainable use of wild populations. This is essential in order to be able to establish manuals of good practices for those who are engaged in their collection. Studies show that propagation of *A. boronioides* is possible from seeds. The plants produced could be used to establish field crops in the future, and not rely on natural populations as the only alternative for obtaining plant material intended for the production of commercial products.

Finally, in order to analyze the uses, potentialities and threats of this species, it is necessary to adopt a multicultural and transdisciplinary approach in order to bring together the perspectives of the academy with those of the local communities that have been the main heirs and makers of this valuable Patagonian heritage.

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