Secondary metabolites as a survival strategy in plants of high mountain habitats

[Metabolitos secundarios como estrategia de supervivencia en plantas de hábitat de alta montaña]

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Abstract: This review work focuses on how the secondary chemistry could help in the survival of plants in high mountain habitats under extreme environmental conditions. The elevated levels of stress in high areas of the tropic and subtropic change dramatically not only by following the annual cycles of winter and summer but they also change in a single day. Some species, however, are able to successfully grow at heights more than 3000 m in the tropical mountains due, in part, to highly specialized physiological processes that affect their physical and chemical responses. In this study, it describes some strategies of how the secondary metabolites could help the plants to stand the high levels of stress in the high mountain ecosystems.

Keywords: Abiotic stress; Biotic stress; Secondary metabolites; Environments of high altitude.

Resumen: Este trabajo de revisión se centra en cómo la química secundaria podría ayudar en la supervivencia de plantas en hábitats de alta montaña en condiciones ambientales extremas. Los altos niveles de estrés en las zonas altas del trópico y subtrópico cambian dramaticamente no solo al seguir los ciclos anuales de invierno y verano, sino que también cambian en un solo día. Sin embargo, algunas especies pueden crecer con éxito a alturas superiores a 3000 m en las montañas tropicales debido, en parte, a procesos fisiológicos altamente especializados que afectan sus respuestas físicas y químicas. En esta revisión, se describen algunas estrategias de cómo los metabolitos secundarios podrían ayudan a las plantas a soportar los altos niveles de estrés en los ecosistemas de alta montaña.

Palabras clave: Estrés abiótico; Estrés biótico; Metabolitos secundarios; Ambientes de gran altura.
INTRODUCTION
A characteristic of most terrestrial plants is their sedentary nature, although some plant species develop vegetative in different directions from their anchoring place, remaining in the same place. This sedentary nature has a profound impact on the life history of individuals, as well as on the structure of the biome community associated with them. The germination, the settlement of the seedlings, their growth, their reproduction, and their propagation are determinants for the environmental characteristics, both in the physical aspect and in the biotic. Plants that grow at high altitude levels grow in extremely fragile ecosystems, usually as endemic species of short stature with some exceptional tree biological types such as the case of the genus Polylepis, whose growth is generally low. The plants are exposed to environmental conditions that change with altitude and latitude. In the high mountain (above 3500 m.a.s.l.), exist isolated or desert territories, this has led to the plants being affected by inbreeding and genetic drift (Ma et al., 2015). Therefore, it is foreseen that species whose ecological territories are placed in altitudinal gradients of considerable amplitude have a complex genetic structure and a combination of adaptive and plastic responses to the changing conditions of the environment (Oleksyn et al., 1998). Knowing the type of response, genetic versus plastic, allows to elucidate if the different phenotypes correspond to genotypes considered as result of the differential pressures of selection or, if the ecological responses are a result of the plasticity of the species, expressing different phenotypes in response to contrasting conditions of the altitudinal gradient (Premoli & Mathiasen, 2011).

To understand the responses of plants at high elevations, it is crucial to understand the differences between adaptation and acclimation. Acclimation, also known as phenotypic adaptation, could be presented as genetic progress that takes place on an evolutionary timescale, controlled by gradual modification of the genome in combination with the pressure of the environment that leads to natural selection according to the Darwinian model. Otherwise, adaptation is a phenomenon that occurs in ecological time, within the life cycle of an individual, therefore, much shorter and that depends on the capacity of intrinsic response of the species to the new climates in accordance with its ability to colonize new territories, this phenomenon is also known as genotypic adaptation.

To distinguish the adaptability of high mountain plant species, the study of a series of characteristics related to the aptitude of the plant and the altitudinal adaptation is required. In general, it is recognized that phenotypic variation along altitudinal gradients are due to more difficult conditions with height gain (Körner, 2007). The higher the altitude, the smaller the plants, reduce the reproductive investment and increase the investment in energy to form denser layers of tissue on the leaf, helping in structural defense storage (Körner, 1989). The diversity of the secondary chemistry also plays an important role in the adaptation and acclimatization of plants to these high habitats, different types of secondary metabolites according to their structure could participate in the mediation of these parameters and some of them are analyzed in this review.

Stress factors on high environments
The different biotic and abiotic gradients that affect animal and plant life in an integrated manner are not well defined due to their complexity. The most observed components in an altitude gradient are shown in Figure No. 1.

The strong climatic and edaphic conditions affect the living environment. The severe stress conditions present in high elevation habitat negatively affect the growth and development of plants. Understanding the mechanisms through which plants transmit signals to the cellular machinery to activate adaptive responses is very important. Signal transduction pathways are the link between the detection mechanism and the genetic response. Plants face environmental changes by activating signal transduction cascades that control and coordinate the physiological and biochemical responses necessary for adaptation (Huang et al., 2012).

The regulation of gene expression, as part of stress responses, involves changes in the transcription levels of some plant genes (Shinozaki & Yamaguchi-Shinozaki, 2000). According to the presence of these mechanisms of tolerance to general and specific abiotic stress, plants are expected to have multiple ways of perceiving stress and, in response, transduction signals that can mitigate this effect (Chinnusamy et al., 2004).

The adaptability of high elevation plants could depend on different factors, among them we have: (i) genetic adaptation between populations with the flow of genes along the altitudinal gradient in the
long term. (ii) the phenotypic plasticity of the characteristics in the short-term selection and (iii) the variation of the gradient between the genetic and environmental influences in these characteristics (Gonzalo-Turpin & Hazard, 2009). In total, the number of plant species adapted to live at high elevations decreases as the percentage of specialized endemism increases, especially in isolated mountain ranges (Vetaas & Grytnes, 2002; Giménez et al., 2004).

**Stress factors in high mountain habitats**

- **Biotic factors**
  - Vegetal composition
  - Competition between plants
  - Herbivory
  - Acclimation and Adaptation
  - Genotype and Phenotype

- **Abiotic factors**
  - Air and soil temperature
  - Soil mineralization
  - Solar radiation
  - Air humidity and Water regime
  - Composition of atmospheric air

![Image of Andean páramos Mérida, Venezuela](image1)

**Figure No. 1**

Most relevant altitudinal components. Andean páramos Mérida, Venezuela

**An overall perspective of the altitudinal variation of secondary metabolites**

Some patterns of secondary metabolites have been established in the vegetation according to the elevation of their habitat, as shown in Figure 2. The biosynthesis of different types of secondary metabolites is influenced by the environmental factors present in these ecosystems. In high altitude plants, environmental factors present drastic changes, generating variations in their secondary chemistry.

![Image of altitudinal tendencies for different types of secondary metabolites](image2)

**Figure No. 2**

Altitudinal tendencies for different types of secondary metabolites of plants that shown different ecological functions (Zidorn, 2010)

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Below is a brief description of how these factors influence the presence of certain secondary metabolites in plants located at high elevations.

3.1. Biotic conditions as determinant:

If climate is an important factor, the selective pressure for herbivore is less pronounced. The climatic factors reduce the reproduction capability of insects besides, the acclimation difficulties, accessibility or abundance of resources according to the altitudinal pattern, it will give as a result of a decrease of the anti-herbivore response from low-altitude to high-altitude zones (Callis-Duehl et al., 2017; González-Reyes et al., 2017). For instance, a general decrease in plant palatability has been associated with a reduction in herbivore pressure at high elevation (Descombes et al., 2016). According to this criterion, high elevation plants that experience lower levels of herbivory are expected to have lower levels of defense against herbivores compared to their families at lower altitudes.

**Abiotic conditions as determinant**

Extreme climatic conditions are factors of abiotic stress in plants regarding radiation and temperature. While it is true that UV-B radiation may directly cause damages to DNA, the harmful effect of low temperature is indirect, both factors in plant cells produce reactive oxygen species (ROS) increased oxidative damage (Zlatev & Lidon, 2012). Consequently, compounds that improve the impact of, for example, low temperatures, UV-B radiation or generation of reactive oxygen species would increase in high altitudes (Körner, 2003).

Both biotic and abiotic factors have been presented for experimental verification. First, the influence of biotic factors could be supported by different studies reported in the literature. For instance, Carey & Wink (1994) showed a decrease in the content of total quinolizidine alkaloids in seeds and leaves of populations of *Lupinus argenteus* Pursh, a perennial herbaceous found in 7 places of high altitude (2776 - 3600 m.a.s.l.) in the western Rocky Mountains of North America. Although authors did not define the content of nitrogen (N) in soil or in the leaf that could have influenced the accumulation rate of total alkaloids, it was observed that this difference in the content of alkaloids is at least partially heritable and therefore, genetically determined. It would be therefore the responsibility of the case of acclimation that is attributable to a constitutive response. However, other plants of high elevation like *Lupinus meridanus* and *L. eremonomo* that grow above 4000 m a.s.l. in the Venezuelan páramo accumulate big quantities of alkaloids, mainly sparteine and lupin (Rojas, 1998). Other species of the South American high páramo like *Senecio formosus* H.B.K accumulates 0.52% of pyrrolizidine alkaloids divided into 7 different compounds. These substances are highly hepatotoxic (Toro-Gonzales et al., 1997) even though the pressure of herbivores and phytopathogens is not important in the zone. Nevertheless, its relation with phenology or with environmental conditions is still unknown since there is no systematic studies as to confirm or deny the hypothesis of Carey and Wink (Alonso-Amelot, 2008). It has neither been analyzed from the perspective of the carbon/nitrogen (C/N) nutrient balance theory (Hamilton et al., 2001), despite it is known that C/N rate in soil may directly influence the quantity and rate of secondary metabolites in plants (Ibrahim et al., 2011).

However, there are situations in which such a decrease in the population of herbivores is not verified, as in the case of abundant populations of herbivore Ortopthera, in the tropical Andean páramo above 3500 m a.s.l. (Calcagno-Pissarelli et al., 2010). *Polylepis* trees distributed along the Andean Mountains at 3000-5000 m a.s.l., lost about 20% of the total surface of the leaves in some locations in Colombia and Venezuela (Velez et al., 1998) where some Ortopthera species (e.g., *Meridacris diabólica*, *Chibchacris carrikeri*), and Coleopthera (*Dyscolus sp*), are potential specialists insects of *Polylepis* genus (Díaz et al., 1997). The chemical compounds isolated for this genus are mainly triterpenes oxygenated and flavonoids glycosidated (Catalano et al., 1995; Neto et al., 2000; Lozano et al., 2017), which indicates that its secondary chemistry could be oriented to other defense strategies. This suggests that the selective forces of abiotic conditions (e.g. cold hardiness) might be stronger than biotic conditions (e.g. resistance to herbivores) along elevation gradients, although this might be organ-specific, depending on the trait analyzed. Due to environmental pressure exposed at high elevations, the plants could lead to an increased protection (chemical and physics) of the highest value organs according to the optimal defense theory (Rasmann et al., 2014).

Second, the influence of abiotic factors could also be supported by different studies published in the literature. For example, the studies on *Pteridium*...
Different types of kaurene (Usubillaga & 1, 2017). Beside was called a (Usubillaga & 1, 2017). Kaurenic acids of these high ecosystems. An outstanding case is present in these ecosystems. A work revealed that picrosides levels in natural population of Picrorhiza kurroa revealed that picrosides levels in natural population of Picrorhiza kurroa revealed that both kaurenic acid (1), grandifloric acid (2) and grandiflorenc acid (3) were found in 72 species of this subtribe (Usubillaga et al., 2003; Padilla-González et al., 2017). Beside was reported an increase in the accumulation of kaur-16-ene (1) acid, from 0.83% (m / m) to 4.4% in leaves of Coespeletia timotensis, in samples taken in June and December at 4100 m a.s.l. in the tropical Andes. These environments - the minimum temperatures and precipitations decrease significantly (Rojas & Usubillaga, 1998). The presence of these secondary metabolites could act as an adaptive response to low temperatures or drought in these Andean ecosystems. The 19-carboxy-kaurene and its derivatives could give more flexibility to the lipid membranes of the plant, helping it to tolerate low temperatures as mentioned above. This assessment is confirmed by two works. First, Padilla-González et al. (2017), through studies of metabolomics, showed the presence of different types of kaurene (4-9, see Figure No. 3), these compounds were found in different concentrations in 72 different species of Espeletiinae subtribes collected at heights of over 3000 m.a.s.l. in the páramo of Colombia and Venezuela, between December of 2007 and August of 2011. In addition, an increase in the amount of kaur-16-in-19-al (10) was reported in five different species of Ruilopezia (giant rosettes), located in the high Andean north (Aparicio et al., 2001).

Allelopathy is a well-known strategy of competition between plants, and it is frequently regulated by the secondary metabolism by which plants interact in situations of competition due to lack of soil resources. The dynamics of allelopathic compounds in plant species that grow along the altitudinal gradient, where there could be an inclination of intra-plant competition, would undoubtedly be of great interest. To our knowledge, there is a lack of data on the systematic development of such information. However, Minthostachys mollis located in rough zones, rocky soils and without slopes drained in the Andean sub-páramo, has a slow growth in its natural environment. This species is used by farmers to preserve potato crops in a way that it prevents sprouts. Plants of “Muna” (name commonly used in Peru and Ecuador) are put inside potato bags. This helps to preserve the product for longer time. It undoubtedly presents an allelopathic approach of acclimation and adaptation

Secondary chemistry is also a response to the acclimatization and adaptation of these high-altitude environments, which together with other responses, help plants overcome the types of stress present in these ecosystems. An outstanding case is present in Espeletiinae subtribe rosettes, which is capable of biosynthesizing a variety of diterpenoids with a kaurane skeleton (Figure No. 3) and represents one of the main classes of natural products in plant species such as: Ruilopezia, Espeletiopsis, Espeletia, Libanothamus and Coespeletia from the tropical Andes of Colombia and Venezuela (Usubillaga & Morales, 1972; Padilla et al., 2017). Kaurenic acids such as: kaurenec acid (1), grandifloric acid (2) and grandiflorenc acid (3) were found in 72 species of this subtribe (Usubillaga et al., 2003; Padilla-González et al., 2017). Beside was reported an increase in the accumulation of kaur-16-ene (1) acid, from 0.83% (m / m) to 4.4% in leaves of Coespeletia timotensis, in samples taken in June and December at 4100 m a.s.l. in the tropical Andes. These environments - the minimum temperatures and precipitations decrease significantly (Rojas & Usubillaga, 1998). The presence of these secondary metabolites could act as an adaptive response to low temperatures or drought in these Andean ecosystems. The 19-carboxy-kaurene and its derivatives could give more flexibility to the lipid membranes of the plant, helping it to tolerate low temperatures as mentioned above. This assessment is confirmed by two works. First, Padilla-González et al. (2017), through studies of metabolomics, showed the presence of different types of kaurene (4-9, see Figure No. 3), these compounds were found in different concentrations in 72 different species of Espeletiinae subtribes collected at heights of over 3000 m.a.s.l. in the páramo of Colombia and Venezuela, between December of 2007 and August of 2011. In addition, an increase in the amount of kaur-16-in-19-al (10) was reported in five different species of Ruilopezia (giant rosettes), located in the high Andean north (Aparicio et al., 2001).

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effect. Alonso-Amelot et al. (2006) discovered that this species accumulates an unusual big quantity of (+) pulegone (13) (> 83 % of the essential oil, Figure No. 3) in exudates of glandular trichomes that have a strong inhibitory effect against seeds and weed plants potentially competitive. Labdane diterpene (14-17) and clerodane (18-20) (Figure No. 3) have been identified in the high páramo endemic species like Blakiella bartsiifolia and Oxylobus glanduliferus. These compounds show a strong inhibition against commercial plants (Amaro & Adrian, 1982; Valero, 2010; Oliveros-Bastidas et al., 2011; Rodriguez-Hernandez et al., 2014; Rodriguez-Hernandez et al., 2017). However, there is only the possibility of developing their allelopathic capacity in these habitats, since they proved to be soluble in hydrocarbons, but this would make their bioavailability much more difficult when transferred to the soil. It is unknown whether these or other compounds located in the exudates of the glandular trichomes have an adaptive role in mediation.

Figure No. 3
Secondary metabolites of some high altitude plants

The species of Espeletiinae subtribe, as well as other several high-altitude species produce a significant amount of volatile mono- and sesquiterpenes (Table No. 1). Among them, there are some of taxonomic relevance for this subtribe. In some less-pubescent plants of páramo these compounds seem to be concentrated in the cuticular wax and glandular trichomes that use to cover all the adaxial surface of the leaves, giving it a sticky appearance (e.g. Minthostachys mollis; Rojas & Usubillaga, 1995). Studies on the composition of essential oils in an altitudinal gradient as well as stages in the vegetative cycle in to species of the Andes Venezuelans (Coespeletia moritziana and Espeletia schultzii) show a change in the composition of these secondary metabolites with the altitude
Espeletiinae has not been systematically studied although the presence of high quantities of monoterpenes (Table No. 1), known as insecticides suggests, a possible potential defensive role.

### Table No. 1
Composition of the essential oils of Andean plants selected

<table>
<thead>
<tr>
<th>Species</th>
<th>Elevation (m)</th>
<th>% (m/m)</th>
<th>Main components (%)</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Coespeletia timotensis</td>
<td>4000</td>
<td>0.45</td>
<td>β-phellandrene (46), α/β-pinene (39), kaur-16-ene (0.9)</td>
<td>Rojas et al., 1999</td>
</tr>
<tr>
<td>C. moritziana</td>
<td>3750</td>
<td>0.58</td>
<td>α/β-pinene (56), β-phellandrene (18)</td>
<td>Aparicio et al., 2002</td>
</tr>
<tr>
<td>C. spicata</td>
<td>3850</td>
<td>0.02</td>
<td>α/β-pinene (57), α-tujene (16)</td>
<td>Aparicio et al., 2002</td>
</tr>
<tr>
<td>C. thyrsiformis</td>
<td>3000</td>
<td>0.2</td>
<td>α/β-pinene (30), germacrene-D (15), selinene (6)</td>
<td>Aparicio et al., 2002</td>
</tr>
<tr>
<td>Espeletia semiglobulata</td>
<td>3800</td>
<td>0.2</td>
<td>α-tujene (73), α/β-pinene (10), Kaur-16-ene (0.5)</td>
<td>Usubillaga et al., 1999</td>
</tr>
<tr>
<td>E. weddelli</td>
<td>4080</td>
<td>0.18</td>
<td>p-cimene (20), α/β-pinene (19), β-phellandrene (18), Kaur-16-ene (0.2)</td>
<td>Khouri et al., 2000</td>
</tr>
<tr>
<td>E. nana</td>
<td>3000</td>
<td>0.08</td>
<td>α/β-pinene (55), mircene (15)</td>
<td>Peña et al., 2012</td>
</tr>
<tr>
<td>E. batata</td>
<td>4200</td>
<td>0.2</td>
<td>α/β-pinene (48), α-tujene (13), limonene (9) (-)-kaur-16-en-9-al (1.6)</td>
<td>Usubillaga et al., 2001a</td>
</tr>
<tr>
<td>E. grandiflora</td>
<td>3328</td>
<td>-</td>
<td>α-pinene (69.7), β-pinene (3.4)</td>
<td>Padilla-González et al., 2016</td>
</tr>
<tr>
<td>E. killipi</td>
<td>3717</td>
<td>-</td>
<td>α-pinene (61.8), Sabine (6.5), β-pinene (4),</td>
<td>Padilla-González et al., 2016</td>
</tr>
<tr>
<td>Espeletiopsis angustifolia</td>
<td>2870</td>
<td>0.18</td>
<td>α-pinene (30), β-caryophyllene (14), α-gurjunene (9.9), β-pinene (9.6), (-)-kaur-16-en-9-al (5.3)</td>
<td>Meccia et al., 2007</td>
</tr>
<tr>
<td>Rallopezia marcesans</td>
<td>3000</td>
<td>0.03</td>
<td>α/β-pinene (35), germacrene-D (34), (-)-kaur-16-en-9-al (0.5)</td>
<td>Aparicio et al., 2001</td>
</tr>
<tr>
<td>R. lindenil</td>
<td>3100</td>
<td>0.04</td>
<td>α/β-pinene (35), germacrene-D (34), (-)-kaur-16-en-9-al (2.5)</td>
<td>Aparicio et al., 2001</td>
</tr>
<tr>
<td>R. atropurpurea</td>
<td>3400</td>
<td>0.03</td>
<td>limonene (49) α/β-pinene (12), mircene (12), (-)-kaur-16-en-9-al (11)</td>
<td>Aparicio et al., 2001</td>
</tr>
<tr>
<td>R. floccosa</td>
<td>3800</td>
<td>0.08</td>
<td>α/β-pinene (31), limonene (24), (-)-kaur-16-en-9-al (4.4)</td>
<td>Aparicio et al., 2001</td>
</tr>
<tr>
<td>R. bracteosa</td>
<td>3085</td>
<td>0.23</td>
<td>mircene (34.2), α/β-pinene (32), 7-epi-α-selinene (9.1), kaur-16-en-9-al (3.0)</td>
<td>Alarcón et al., 2015</td>
</tr>
<tr>
<td>Libanothamus occultus</td>
<td>2800</td>
<td>0.11</td>
<td>α/β-pinene (44), mircene (17), p-cimene (11), (-)-kaur-16-en-9-al (0.2)</td>
<td>Usubillaga et al., 2001b</td>
</tr>
<tr>
<td>L. nerifolia</td>
<td>2800</td>
<td>0.13</td>
<td>sabine (26), limonene (19), α-tujene (19), α-phellandrene (11), α/β-pinene (6)</td>
<td>Usubillaga et al., 2001b</td>
</tr>
<tr>
<td>L. humbertii</td>
<td>3200</td>
<td>0.05</td>
<td>limonene (19), α-tujene (18), α-phellandrene (17), α/β-pinene (15), (-)-kaur-16-en-9-al (0.2)</td>
<td>Usubillaga et al., 2001b</td>
</tr>
<tr>
<td>L. lucidus</td>
<td>3400</td>
<td>0.10</td>
<td>α-tujene (30), α/β-pinene (28), α-phellandrene (10) (-)-kaur-16-en-9-al (0.5)</td>
<td>Usubillaga et al., 2001b</td>
</tr>
</tbody>
</table>
There are different secondary metabolites capable of smoothing the strong incidence of UV lights in these habitats. Plants whose leaves are protected by woolly masses usually have low UV chemical filters. On the contrary, other plants accumulate significant quantities of these compounds. Some secondary metabolites such as stilbenes, flavonoids, lignans, tannins and others are able to absorb the UV radiation between 210 and 350 nm. This optical characteristic could be a key factor in the development of these compounds as substances of adaptation to the UV radiation of the plant with the evolutionary time (Schnitzler et al., 1996, Bornman et al., 1997, Kolb et al., 2001; Padilla-Gonzalez et al., 2017). In fact, some tropical mountain species that grow on bare slopes accumulate unusual amounts of condensed phenolics and tannins in their aerial parts (Alonso-Amelot et al., 2007).

Kim et al., (2018) used proteomic tools to study the tolerance mechanism of Herpetospermum pedunculosum at high altitudes, 2800-3300 m.a.s.l. The results indicated that the chloroplast pathway, as well as signaling production, the ROS uptake path and the Calvin cycle pathway can collectively mediate adaptation to high altitudes in H. pedunculosum. In general, they showed that altitude affects some key stress and defense proteins to maintain the balance of ROS, and therefore, it could be considered that the increase in these proteins contribute to the greater tolerance of H. pedunculosum to a large altitude. High levels of some antioxidative enzymes and metabolites secondary antioxidant could be of significance for the response to high-altitude environments. Two compounds with structure of sesqui-norlignan, herpetone and dehydrodiconiferyl alcohol, were reported for this species, both aromatic compounds are able to absorb the UV radiation between 210 and 350 nm (Zhang et al., 2006). Blakilla bartisifolia, a species of the Andean páramos at 4200 m.a.s.l. has a foliar surface are almost completely covered by glandular trichomes with accumulation of a colorless liquid at the edge of the vas. These drops may act as incidental sunlight refraction lens to modulate its impact on the underlaying parenchyma. Their cuticular waxes protecting the epidermis, represent to 9% of the leaf weight on a FW basis. Accumulation of cuticular exudate in such quantity has scarcely been described. Therefore, cuticular investment in B. bartisifolia must be adaptive. The composition of this exudate includes a high content of flavones (21-22, see Figure No. 4) with absorbance in the UV-B zone that influences the vegetation at those elevations. Although the atmosphere at 3500-4200 m.a.s.l. filters off radiation below 280 nm, the molecular structure of many flavonoids includes chromophores with important molar extinction coefficients above the cutoff wavelength, thus providing UV-B protection. (Calcagno-Pissarelli et al, 2010). In Arnica montana cv. ARBO was reported that the ratio of 3’,4’-dihydroxylated flavonoids to flavonoids without that substitution pattern, generate a positive correlation in the proportion of quercetin derivatives to kaempferol derivatives to the altitude of the growing site. The data revealed that the DPPH radical scavenging potential of flowering stems from plants grown at higher altitudes was significantly higher than that of lowland plants (Spitaler et al., 2008). It is known that quercetin derivatives are three to four times more potent radical scavenging, than kaempferol derivatives (Rice-Evans et al., 1996). Padilla-Gonzalez et al., (2017), based on a study with metabolomics approach showed that in species from Espeletia genus collected at heights more than 3000 m.a.s.l. from the páramos of Colombia and Venezuela, biosynthesized high quantities of the flavonoids (Figure 4). For example, species located in páramos of Boyacá (Colombia) at 3000-4200 m showed high quantities of the flavonoids 3-methoxy quercetin and hesperetin (23-24), whereas species from páramos of Cundinamarca at 3200-4100 m a.s.l. were characterized by high quantities of dimeric flavonoids such as 8,8’-methylene-bisquercetin (25). Lastly, the species located in the páramos of Venezuela at 3000-4500 m a.s.l. were characterized by the presence of high quantities of glycosylated flavonoids, such as quercetin-3-O-galactoside (26). The accumulation of flavonoid in high quantities in most of the species of Espeletia genus from the páramos of Venezuela and Colombia, suggest that the increase of UV-B filtration compounds may converge towards thermal protection against low temperatures and act on the cells that produce reactive oxygen species (ROS) (Chalker-Scott & Scott, 2004; Spitaler et al., 2008).
Andean species of *Hypericum*, located in the altitudinal range of 3000-5000 m.a.s.l., have been reported the presence of different types of aromatic compounds (Crockett *et al.*, 2010). Tocci *et al.*, (2018) based on presence or absence of 34 phenolic compounds, identified and quantified from seven of Andean *Hypericum* species (*H. cardonae*; *H. cuatrecasii*; *H. myricariifolium*; *H. humboldtianum*; *H. carinosum*; *H. laricifolium* and *H. garciae*, all native to the páramo ecosystem in Colombia), and using tools of chemical clustering, reported that these species were divided into two chemical groups, the results reflected an evolutionary relationship, especially as chemical profiles are often diverse or homoplastic, and thus, largely independent of phylogeny at the species level (e.g. Farag *et al.*, 2013). Of the 34 compounds identified, 22 were flavonoids, quercetin-3-glucuronide (27), quercetin-3-glucoside (28), kaempferol-3-glucoside (29), isorhamnetin-3-glucoside (30), procyanidin B2 (31), epicatechin (32) and catechin (33) compounds (Fig. 4) were reported in higher amounts. Others
Hypericum species (H. irazuense; H. gnidioides and H. laricifolium) from Andean mountains, has been reported the presence of xanthones derivatives compounds (Crockett et al., 2010; Ramirez-Gonzalez et al., 2013). Several dimeric acylphloroglucinols derivatives (Figure No. 5), hyperbrasilol B (35), isohyperbrasilol B (36), uliginosin B (37), isouliginosin B (38), and uliginosin A (39), were identified in different Hypericum species (H. andinum; H. laricifolium; H. brevistylum and H. silenoides) collected from Peruvian Andean páramos at 3100-3600 m.a.s.l. (Ccana-Ccapatinta et al., 2015) besides, these species are rich in flavonoids principally hyperoside (34) that has been found in high quantities (Ccana-Ccapatinta et al., 2014).

The different class of aromatic compounds reported for some Andean Hypericum species, could indicate an important role in ecological niche differentiation; an argument in line with the hypothesis that adaptive radiation potentially explains the high richness in the Andean paramos of Hypericum species (Nürk et al., 2013).

Figure No. 5
Some dimeric acylphloroglucinols reported for Hypericum Andean species

These aromatic natural compounds could be one of the strategies that Hypericum species use for their adaptation and acclimatization in these Andean ecosystems. No doubt, the secondary chemistry has an impact on acclimatization and adaptation, helping plants together with some physical defense strategies to withstand stressful conditions in high altitude ecosystems.

Perspectives
Considering that plants use different strategies, physical and chemical, to survive in environments with high levels of stress as those found in high mountain ecosystems. These fragile ecosystems extremes have been getting smaller by man as result of the implementation of agriculture. In this regard, by the benefit of these habitats is necessary the creating protected areas especially in tropical páramos that have been more affected by human activity. Exist are many questions to unravel in the complex relationships that occur in these ecosystems, the plants are a rich source of secondary metabolites, and it play a key role in the ecology of these environments. The plants localized at high altitude, most are endemic species very little studied, with a huge phytochemical potential that should be a source of inspiration for further research (pharmacological, agrochemical, ecological), which will result in a better understanding the different interactions that characterized in these high mountain environments.

The new investigations could be directed for example: What abiotic and biotic factors have been changing in the last decade as a result of climate change? How has this impacted and influenced the biosynthesis of secondary metabolites in plants? Has the phenotype and genotype been altered? Has secondary chemistry been altered as a strategy for plants to continue their survival in these high-altitude ecosystems? These are questions that still do not have a coherent answer and that should be considered, among others.

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Secondary metabolites as survival in plants of high altitudes


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