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Twelve *Salpichroa* taxa have been phytochemically analyzed. From the aerial parts of *S. scandens*, the four known salpichrolides A, C, I, S, and an unreported withanolide named salpichrolide V (1), were isolated. In *S. dependens*, *S. gayi*, *S. glandulosa* subsp. *glandulosa*, *S. glandulosa* subps. *weddelii*, *S. leucantha*, *S. micrantha*, *S. microloba*, *S. proboscidea*, *S. ramosissima*, *S. tristis* var. *tristis*, and *S. weberbaueri*, no withanolides were found. The chemical content of ca. 85% of the *Salpichroa* taxa is in agreement with molecular studies, which suggest that *Salpichroa* and *Jaborosa*, a genus considered morphologically close to *Salpichroa*, are distant in the systematic of the Solanoidae subfamily.

Moreover, the *in vitro* cytotoxic activity of a set of natural salpichrolides and derivatives was examined against two prostate carcinoma cell lines (PC3 and LNCaP) and two human breast cancer cell lines (MCF-7 and T47D). Several compounds showed moderate activity ($IC_{50} = 64.91 - 29.97 \mu M$).

**Keywords:** *Salpichroa* • Withanolides • Structure elucidation • Chemotaxonomic markers • Cytotoxicity

Introduction

The genus *Salpichroa* Miers (Solanaceae) comprises around 17 species, restricted mainly to the highland Puna and Prepuna of Peru, Bolivia and northern Argentina. One single species (*S. tristis* Miers) occurs throughout the highland grasslands in the Andes, reaching as far as Venezuela, while *S. origanifolia* (Lam.) Baillon has a worldwide distribution. These species can take the forms of a scandent shrub (*Figure 1 A, D*), or a hanging shrub (*Figure 1 C, E*), or a procumbent shrub (*Figure 1 B, F*).

D’Arcy and Hunziker placed *Salpichroa* together with *Nectouxia* Kunth and *Jaborosa* Juss. in the tribe Jaborosae Miers, based mostly on morphological characters. Recent molecular phylogenies in Solanaceae proved that *Salpichroa* belongs

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to a different and distant clade from *Jaborosa*.\[^{4,5}\] Until now the phytochemical evidence was quite fragmentary, but in agreement with the molecular proposal to place *Salpichroa* and *Jaborosa* in independent clades.\[^{6}\]

Withanolides are steroidal lactones built on an ergostane skeleton of 28 carbons functionalized at carbons 1, 22, and 26, and commonly known as the withanolide skeleton, with their chemistry and occurrence having been the subject of several reviews.\[^{6-8}\] Over 750 withanolides have been described, principally, but not exclusively, from genera belonging to the Solanoideae subfamily. Some clades of the subfam. Solanoideae also contain withanolides with exclusive interesting structural arrangements, which can be considered at different hierarchical levels chemotaxonomic markers.\[^{6}\] Withanolides with a six-membered aromatic ring-D and a family of these related ergostane derivatives constitute an important group called salpichrolides. They have been isolated from *S. origanifolia* and *S. tristis* var. *lehmannii* (Dammer) Keel.\[^{9-14}\] Many of these compounds exhibit interesting biological activities such as antifeedant,\[^{15-17}\] antiproliferative,\[^{18}\] and cancer chemopreventive effects.\[^{19}\]

As part of an ongoing program aimed at to clarify the systematic position of *Salpichroa*, a phytochemical study of twelve taxa is herein reported. The species studied were: *S. dependens* (Hook.) Miers, *S. gayi* Benoist, *S. glandulosa* (Hook.) Miers (with two subspecies: subsp. *glandulosa* and subsp. *weddelii* (Benoist) Keel), *S. leucantha* Pereyra, Quip. & S. Leiva, *S. micrantha* Benoist, *S. microloba* Keel, *S. proboscidea* Benoist, *S. ramosissima* Miers, *S. scandens* Dammer (two samples), *S. tristis* Miers var. *tristis* (three samples), and *S. weberbauerii* Dammer.
Results and Discussion

Phytochemical study.

The dichloromethane extract of the aerial parts of *S. scandens* from Argentina (sample 1) was subjected to chromatographic purification to yield the new compound 1, and the known salpichrolides A,\(^1\) C,\(^{15}\) I,\(^{11}\) and S\(^{10}\) (Figure 2). HRESITOFMS of salpichrolide V (1) showed a quasi-molecular ion [M+Na]\(^+\) at m/z 509.2066, corresponding to an elemental formula of C\(_{28}\)H\(_{36}\)ClO\(_4\)Na. From the following observations, the NMR spectroscopic data of compound 1 revealed the typical signals corresponding to a salpichrolide-type withanolide characterized by the aromatic ring-D and a ß-lactol side chain: (i) the \(^1\)H NMR spectrum displayed the characteristic signals corresponding to aromatic protons at δ 7.14, δ 6.94, and δ 5.91, assigned to H-15, H-16, and H-18, respectively; (ii) the presence of an epoxy lactol was evidenced by the signal corresponding to the oxymethine proton at δ 4.92 (d, J = 9.9 Hz), assigned to the hydrogen H-26, and also by the resonances of the two methyl groups at C-24 and C-25 at δ 1.31 (H-24) and 1.29 (H-25), respectively; (iii) the signals at δ 65.2 [C (24)], δ 64.0 [C (25)], δ 91.7 [C (26)], δ 16.9 [C (27)], δ 19.1 [C (28)], observed in the \(^{13}\)C NMR spectrum, were in agreement with a side chain epoxy lactol arrangement. Regarding the A and B rings, the \(^1\)H NMR spectrum of 1 exhibited two olefinic proton signals at δ 5.91 (dd, J = 10.2 and 2.8 Hz) and 6.57 (ddd, J = 10.2, 5.1, and 2.2 Hz), typical of a 1-o xo-2-en system in ring A, while the signal at δ 4.07 (t, J = 2.9 Hz), bearing in mind that the molecule has a chlorine atom by HRESITOFMS, suggested the presence of a chlorine substituent at C (6) position. The substitution pattern in ring B was further confirmed by the signals at δ 77.5 and 64.0 in the \(^{13}\)C NMR spectrum, assigned to C (5) and C (6), respectively, and by the cross-correlation peaks between the signal of H-1C (6) with the signals of C (5), C (10) (δ 52.9), and C (8) (δ 32.9) in the HMBC spectrum. The \(\beta\) orientation of the chlorine group was established through the multiplicity and coupling constant value of H-1C (6) (J = 2.9 Hz), and by the cross-correlation peak observed between H-1C (6) and H-1C (9) (δ 2.16) in the NOESY experiment (see Supporting Information). Thus, the structure of compound 1 was established as (20S,22R,24S,25S,26R)-68-chloro-22,26,24,25-diepoxy-5c,26-dihydroxy-17(13 → 18) abeo-5c-ergosta-2,13,17-tetraen-1-one. Compound 1 is one of the examples of chlorinated withanolides found in other species of Solanaceae. To date, at least thirty-five withanolides with chlorine substituent on ring B have been reported from the *Jaborosa, Nicandra Adams., Physalis L., Tubocapsicum (Wettst.) Makino, and Withania Pauqy* genera.\(^{10,8,9,20}\) Following a similar procedure, the dichloromethane extract of *S. scandens* from Bolivia (sample 2) was analyzed and shown to yield the salpichrolides A, C, S, and V (1) by comparison with published spectroscopic and physical data.

![Figure 2. Withanolides isolated from Salpichroa species.](image)

The dichloromethane and ethyl acetate extracts of the remainder eleven Salpichroa taxa studied were analyzed by TLC and \(^1\)H NMR but no withanolides were detected from any of these. Withanolides have been reported in the majority of the genera within subfam. Solanoideae and in most species of some of these genera (eg. *Datura L., Jaborosa, Physalis,*...)

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Withania),[8] However, in other genera such as Solanum L. (ca. 1500 species), withanolides have been reported only in two species to date.[21-24] In addition to consulting the published papers related to phytochemical studies of the Solanum species, members of our working group conducted a phytochemical study in two Solanum species (S. commutum C.V. Morton and S. atropurpureum Schrank), but no withanolides were found in any of them.[25] Therefore, the absence of withanolides in some Salpichroa species would not be so strange.

Chemotaxonomical considerations.

The suprageneric position of Salpichroa within subfam. Solanoideae is still unresolved. Although the traditional position groups Salpichroa along with Jaborosa and Nectouxia in the tribe Jaboroseae,[1,3,26] molecular studies place Salpichroa and Nectouxia far from Jaborosa in the small and informal “Salpichroina clade”[4,5] These molecular studies encompassed the majority of the Solanaceae genera but only a few species of Jaborosa and Salpichroa. Recently, Moré et al.[27] proposed a phylogenetic reconstruction for Jaborosa; in their study eighteen Jaborosa species, three Salpichroa taxa (two species and one variety) and the unique species of Nectouxia (N. formosa Knuth) were included. Once again, Salpichroa and Nectouxia were placed in a well supported small clade, while Jaborosa was sister to the Eurasian genus Atropa L. (tribe Hyoscyameae Endl.).

Previous phytochemical studies involving thirteen Jaborosa species and two Salpichroa taxa (S. origanifolia and S. tristis var. lehmannii) have supported the molecular proposal.[8,9,14] Now, with new knowledge concerning approximately 85% of the Salpichroa species, more accurate conclusions can be made. In Jaborosa, all the analyzed species contained withanolides with exclusive rearrangements (trechonolides, sativolides, spiranoid-γ-lactones, spiranoid withanolides at C-22, aromatic ring-A withanolides, and 15,21-cyclowithanolides).[9] In contrast, only 17.5% of the Salpichroa taxa contained aromatic or oxygenated ring-D withanolides, with these latter compounds possibly being involved in the biosynthetic pathway leading to ring-D expansion.[28] Based on these different chemical contents found for each genus, we reaffirm the phytochemical support mentioned above in agreement with the molecular position of maintaining Jaborosa and Salpichroa distant in independent clades.

Citotoxic activity.

Considering the selective antiproliferative activity toward hormone-dependent ER(+) breast cell lines when compared to ER(-) negative cell lines shown by salpichroline A, the in vitro cytotoxic activity of a set of natural salpichrolides (Figure 3) and derivatives was examined on two human prostate carcinoma cell lines, PC-3 (androgen independent) and LNCaP (androgen dependent), and on two human breast cancer cells that possessing the estrogen receptor, MCF-7 and T47D. The salpichroline derivatives tested (2 – 11) were obtained by biotransformation of salpichrolides A, C, and G with filamentous fungi Rhizomucor miehei (CECT 2749) and Cunninghamamella elegans (CECT 2113), (Figure 4).[29] The results, expressed as half maximal inhibitory concentration (IC50), are summarized in Table 1. Cell lines were exposure for 48 hours with the compounds at a range 10-65 μM. MTS one step colorimetric assay was done to test the cytotoxic potential and to determine IC50 doses. Doxorubicin was used as reference anticancer drug. Overall, the data obtained showed that the LNCaP and T47D cell lines were more sensitive to the compounds tested. The compounds salpichrolides A, D, G, M, V, 2,3-dihydrosalpichrolide B, and the derivatives 4 and 11 showed cytotoxic activity against at least one of the assayed cell lines (IC50 = 64.91 – 29.97 μM). Comparison of salpichrolide A and salpichrolide C showed that the cleavage of the 5,6-epoxy-group to give the 5α,6β-diol abolished the cytotoxic activity, in agreement with results previously reported in the literature.[15,30] The presence of a hydroxyl group at C-12 position (α or β) led to a loss of activity (LNCaP and T47D), this is observed when comparing the compounds salpichrolide A and salpichrolide T and compound 6, in agreement with results previously reported for compounds with similar structural features.[31] Finally, no loss of activity was observed when the 5α,6α-epoxy functionality was replaced by 5α-chloro,6β-hydroxy system (salpichrolide A and 1). On the contrary, this structural difference enhanced cytotoxic activity against PC-3 cell line.
Table 1. In vitro cytotoxic activity of salpichrolides A, C, D, G, M, S, T, and 2,3-dihydrosalpichrolide B and derivatives 1–11 against human solid breast and prostate tumor cell lines.\(^a\)

<table>
<thead>
<tr>
<th>Compound</th>
<th>LNCaP</th>
<th>PC-3</th>
<th>MCF-7</th>
<th>T47D</th>
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</thead>
<tbody>
<tr>
<td>salpichrolide A</td>
<td>46.95 ± 1.06</td>
<td>&gt; 65</td>
<td>&gt; 65</td>
<td>44.61 ± 1.03</td>
</tr>
<tr>
<td>2,3-dihydrosalpichrolide B</td>
<td>&gt; 65</td>
<td>47.87 ± 1.03</td>
<td>&gt; 65</td>
<td>&gt; 65</td>
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<tr>
<td>salpichrolide C</td>
<td>&gt; 65</td>
<td>&gt; 65</td>
<td>&gt; 65</td>
<td>&gt; 65</td>
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<tr>
<td>salpichrolide D</td>
<td>52.72 ± 1.04</td>
<td>&gt; 65</td>
<td>&gt; 65</td>
<td>58.68 ± 1.03</td>
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<tr>
<td>salpichrolide G</td>
<td>48.05 ± 1.08</td>
<td>51.16 ± 1.05</td>
<td>&gt; 65</td>
<td>41.81 ± 1.08</td>
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<tr>
<td>salpichrolide M</td>
<td>55.69 ± 1.03</td>
<td>61.82 ± 1.04</td>
<td>&gt; 65</td>
<td>41.79 ± 1.08</td>
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<tr>
<td>salpichrolide S</td>
<td>&gt; 65</td>
<td>&gt; 65</td>
<td>&gt; 65</td>
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<tr>
<td>salpichrolide T</td>
<td>&gt; 65</td>
<td>&gt; 65</td>
<td>&gt; 65</td>
<td>&gt; 65</td>
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<tr>
<td>salpichrolide V (1)</td>
<td>40.23 ± 1.08</td>
<td>54.16 ± 1.03</td>
<td>&gt; 65</td>
<td>45.25 ± 1.07</td>
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<tr>
<td>2</td>
<td>64.91 ± 1.03</td>
<td>&gt; 65</td>
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<td>4</td>
<td>51.45 ± 1.06</td>
<td>&gt; 65</td>
<td>&gt; 65</td>
<td>29.97 ± 1.02</td>
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<tr>
<td>11</td>
<td>64.63 ± 1.05</td>
<td>&gt; 65</td>
<td>&gt; 65</td>
<td>44.06 ± 1.08</td>
</tr>
<tr>
<td>doxorubicin</td>
<td>1.63 ± 0.01</td>
<td>1.89 ± 0.01</td>
<td>6.11 ± 0.02</td>
<td>10.02 ± 0.01</td>
</tr>
</tbody>
</table>

\(^a\) IC\(_{50}\) values given in μM (mean ± S.D.) were obtained on the basis of triplicate assay.
Figure 3. Salpichrolides assayed against human prostate and breast cancer cell lines.
Figure 4. Salpichrolide derivatives obtained by biotransformation\textsuperscript{19} assayed against human prostate and breast cancer cell lines.
Conclusions

The chemical content of ca. 85% of the *Salpichroa* taxa reaffirms that *Jaborosa* and *Salpichroa* are distant in the systematic of the Solanoideae subfamily. Moreover, salpichrolides should be considered to be relative taxonomic markers for *Salpichroa* at the generic level for two reasons: the aromatic ring-D withanolides are not exclusive of *Salpichroa* genus since they have also been isolated from *Nicandra physalodes* (L.) Gaertn. (Ray and Gupta, 1994)[8] and *Physalis hispida* (Waterf.) Cronquist[32] and because these metabolites are only present in 17.5% of the *Salpichroa* species.

Regarding the bioactivity of the salpichrolides and derivatives, although the cytotoxic activity observed for the tested compounds in the present work was moderate (IC₅₀ ≥ 29.97 μM), these results contribute to deeper structure–activity relationship studies for this type of withanolides.

Experimental Section

General Experimental Procedures

Optical rotation was measured on a JASCO P-1010 polarimeter. The UV spectrum was obtained using a Shimadzu-260 spectrophotometer, and the IR spectrum was produced using a Nicolet 5-SXC spectrophotometer. NMR spectra were recorded on a Bruker AVANCE II AV-400 NMR spectrometer operating at 400.13 MHz for ¹H and 100.63 MHz for ¹³C, while 2D spectra (COSY, HSQC, HMBC and NOESY) were obtained using standard software. Chemical shifts are given in ppm (δ) downfield from the TMS internal standard. HRESIQTOFMS was determined on a Micro TOF II Bruker Daltonics spectrometer. The chromatographic separations were performed by column chromatography on silica gel 60 (0.063-0.200 mm) and Sephadex LH-20, and preparative TLC was carried out using silica gel 60 F₂₅₄ (0.2 nm thick) plates. Preparative TLC separations were performed under the following conditions: (i) the amount of sample applied was approximately 15 mg for 20 cm plate; (ii) the bands were visualized using ultraviolet light; (iii) compounds were eluted from the silica using CH₂Cl₂:MeOH (8:2).

Plant Material

A voucher specimen for all the *Salpichroa* taxa studied has been deposited at the Museo Botánico Córdoba (CORD), Universidad Nacional de Córdoba (Argentina), and was identified by G. Barboza (GB) and S. Leiva González. The following samples were collected: *Salpichroa dependens* (Hook.) Miers in Pariahuanca, 11°59'28.68''S, 74°58'18.11''W, Junín, Perú, in January 2013 (voucher under GB 3949); *S. didierana* Jaub. in Urubamba, 13°06'51.5''S, 72°20'50''W, Cuzco, Perú, in January 2012 (voucher under GB 3417); *S. gayi* Benoist in Urubamba, 13°17'07.8''S, 72°02'58.6''W, Cuzco, Perú, in January 2012 (voucher under GB 3428); *S. glandulosa* (Hook.) Miers subsp. *glandulosa* in Urubamba, 13°17'07.8''S, 72°02'58.6''W, Cuzco, Perú, in January 2012 (voucher under GB 3411); *S. ramosissima* Miers in Matucana, 11°50'42.7''S, 76°22'57.2''W, Lima, Perú, in January 2012 (voucher under GB 3411); *S. scandens* Dammer (sample 1) in Tafi, km 75/76, Tucuman, Argentina, in December 1995 (voucher under GB 3042); *S. scandens* (sample 2) in Quillacollo, 17°27'06.6''S, 66°21'11.6''W, Cochabamba, Bolivia, in February 2012 (voucher under GB 3615); *S. tristis* Miers var. *tristis* (sample 1) in Tafi, km 75/76, Tucuman, Argentina, in December 1995 (voucher under GB 3042); *S. tristis* var. *tristis* (sample 2) in Tafi, Mirador del Infiernillo, Tucuman, Argentina, in March 2006 (voucher under GB 1711); *S. tristis* var. *tristis* (sample 3) in Carrasco, 17°28'30.4''S, 65°38'14.9''W, Cochabamba, Bolivia, in March 2012 (voucher under GB 3621); *S. weberbauerii* Dammer in Quello-Quello, 13°23'30.3''S, 71°49'01''W, Cuzco, Perú, in January 2012 (voucher under GB 3415).

*Solanum comptum* and *S. atropurpureum* were collected in Corrientes, Argentina, in December 2008. A voucher specimen for these two species has also been deposited at CORD, and was identified by G. Barboza (GB); *S. comptum* (voucher under GB 2078) and *S. atropurpureum* (voucher under GB 2125).

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**Extraction and Isolation of compounds from Salpichroa species**

The fresh aerial plants of *S. scandens* from Argentina (590 g) (sample 1) were triturated with EIOH (1000 mL) at room temperature immediately after collection. After 24 h. the sample was filtered and the solvent was evaporated at reduced pressure. The residue obtained (22.9 g) was defatted by partition in n-hexane–EIOH–H$_2$O (10:3:1), with the resultant EIOH–H$_2$O phase being washed with n-hexane (3 x 250 mL) and EIOH evaporated at reduced pressure. The resulting aqueous phase was extracted with CH$_2$Cl$_2$ (3 x 250 ml). The CH$_2$Cl$_2$ extract was dried (Na$_2$SO$_4$), filtered, and evaporated to dryness under reduced pressure, with the resulting residue (1.85 g) being initially fractionated by column chromatography over Sephadex LH-20, and elution with n-hexane–CH$_2$Cl$_2$–MeOH (2:1:1) affording thirty four fractions. All fractions with similar TLC profiles were combined and analyzed by $^1$H NMR, resulting in three fractions containing withanolides (I–III). Fraction I (316.1 mg) was column chromatographed over silica gel 60 G, using CH$_2$Cl$_2$–MeOH mixtures of increasing polarity (100:00–85:15) for the elution, to afford salpichrolide A (185.9 mg) and an impure fraction (17 mg) which was further purified by preparative TLC with CH$_2$Cl$_2$–MeOH, 94:06 to give salpichrolide A (3.5 mg) and salpichrolide I (7.4 mg). Fraction II (89.7 mg) was subjected to silica gel 60 G. Elution with CH$_2$Cl$_2$–MeOH (100:00–85:15) afforded salpichrolide S (8.1 mg) and a mixture (26.2 mg) which was fractionated by TLC (CH$_2$Cl$_2$–MeOH 98:02) to yield 8.0 mg of salpichrolide A and 9.6 mg of compound 1. Fraction III (340.7 mg) was chromatographed on a silica gel column, using AcOEt as the eluent to afford salpichrolide C (25.2 mg).

The fresh aerial plants of *S. scandens* from Bolivia (1.0 kg) (sample 2) were triturated with EIOH (2000 mL) at room temperature immediately after collection. After 24 h. the sample was filtered and the solvent was evaporated at reduced pressure. The residue obtained (31.1 g) was defatted by partition in n-hexane–EIOH–H$_2$O (10:3:1), with the resultant EIOH–H$_2$O phase being washed with n-hexane (3 x 500 mL) and EIOH evaporated at reduced pressure. The resulting aqueous phase was extracted with CH$_2$Cl$_2$ (3 x 500 ml). The CH$_2$Cl$_2$ extract was dried (Na$_2$SO$_4$), filtered, and evaporated to dryness under reduced pressure. This resulting residue (6.2 g) was initially fractionated by column chromatography over Sephadex LH-20. Elution with MeOH afforded twenty three fractions. All fractions with similar TLC profiles were combined and analyzed by $^1$H NMR to give two fractions containing withanolides (fractions I and II). Fraction I (738.5 mg) was chromatographed over silica gel 60 G CC. Elution and Isolation of compounds from Salpichroa species

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C-20), 38.8 (CH, C-9), 37.1 (CH₂, C-4), 35.0 (CH₂, C-7), 34.0 (CH₂, C-23), 32.9 (CH, C-8), 31.0 (CH₂, C-12), 26.0 (CH₂, C-11), 19.1 (CH, C-28), 17.6 (CH₂, C-21), 16.9 (CH, C-27), 16.3 (CH₂, C-19). HRESITOFMS \textit{m/z} [M+Na] + 509.2066 (calcd for C₂₈H₃₅ClO₅Na, 509.2065).

Cell culture and reagent

LNCaP, PC3, MCF-7 and T47D cells were obtained from the American Type Culture Collection (Manassas, VA, USA) and maintained at 37 °C in a humidified incubator with a 5% CO₂/95% air atmosphere in RPMI 1640 medium (EMEVE Medios, Lab MicroVet, Buenos Aires, Argentina) supplemented with 10% fetal bovine serum (Natocor, Córdoba, Argentina) and antibiotics (Invitrogen). Doxorubicin was kindly provided as a gift by Dr. Gonzalo Sequeira (IBYME) and was purchased from Sigma-Aldrich Chemical Co. (St. Louis, MO, USA).

Biological evaluation

Treatment of cancer cells. Optimum seeding density for 96-well plates was determined for both cell lines. LNCaP cells (10⁴ cells/100 µl), PC3, MCF-7 and T47D cells (5x10³ cells/100 µl) were seeded into each well of a 96-well microtiter plate. After incubation for 24 h, the media were aspirated and replaced with 100 µl of serum free medium containing different concentration of each compound and incubated for 48 h. In parallel, cells were treated with doxorubicin as standard anticancer drug. Wells with serum free medium and wells with an equal amount of DMSO were used as negative controls. Triplicate wells were prepared for each individual dose.

Cytotoxicity assay. MTS cell viability analysis was performed 48 h post treatment using the Cell Titer 96 AQueous One Solution Proliferation Assay System (Promega Corporation, Madison, USA), in which viable cells convert MTS tetrazolium into a formazan-colored product (A 490 nm). Following the manufacturer’s instructions, 20 µl of MTS reagent were added to each well and cells were incubated at 37 °C for 1 h. Absorbance was detected at 490 nm on a Thermo Scientific Multiskan FC plate reader. The IC₅₀ value was defined as the concentration of compound yielding 50% cell survival.

Statistical Analysis. Statistical Analysis was conducted using the Graph Pad Prism 5.0 software (Graph Pad Software Inc., San Diego, CA, USA). The data were recorded as means ± standard deviations from triplicates.

Supplementary Material

Supporting information for this article is available on the WWW under http://dx.doi.org/10.1002/MS-number.

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Author Contribution Statement

A.V.B. carried out the phytochemical analysis and participated in field trips; S.L.G. organized and participated in the majority of the field trips in Peru and identified some Salpichroa species; V.P.C and P.A.S. designed biological experiments; P.A.S. performed biological determinations; V.P.C., P.A.S., and J.C.C. interpreted biological data; G.E.B. provided and identified all the species studied in this work; V.E.N. participated in the design and coordination of the study.

V.E.N. and G.E.B. have equal contribution. Both participated to writing, and all the authors read and approved the final manuscript.

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References and notes


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