Journal of Biodiversity, Bioprospecting and Development

Graf et al., J Biodivers Biopros Dev 2016, 3:1
DOI: 10.4172/2376-0214.1000156

Research Article Open Access

Discovering the Pharmacological Potential of Ecuadorian Market Plants using a Screens-to-Nature Participatory Approach

Brittany L Graf[1,2]*, Patricio Rojas-Silva[1] and Manuel E Baldeón[1]

[1]Centro de Investigación Traslacional, Universidad de Las Américas, José Queri y Av. Granados, Edificio 2, Quito, Ecuador
[2]Department of Plant Biology and Pathology, Rutgers University, 59 Dudley Rd, New Brunswick, NJ, USA

Abstract

Objective: Edible plants of medicinal value can serve as a resource for sustainable development in Ecuador, a country rich in agrobiodiversity and traditional ecological knowledge. This study surveyed the nematicidal, antimicrobial, and antioxidant potential of plants sold in local Ecuadorian markets through participatory scientific discovery workshops with local Ecuadorian students and researchers, while simultaneously enhancing the knowledge and technological capacity of workshop participants.

Methods: Edible plants were purchased from city markets at 3 distinct research sites in Ecuador - Cuenca, Quito, and Santa Elena. Botanical identification and traditional uses of each plant were assessed via herbarium specimen preparation and reference to ethnobotanical texts. Portable screens-to-nature (STN) extraction and assay technologies were employed to rapidly and qualitatively detect roundworm lethality, antibacterial, antifungal, and free radical scavenging activities of the plants during 3-day STN workshops at each research site. Participant learning was assessed through a retrospective pretest-posttest administered at the end of each STN workshop.

Results: A total of 50 plants were collected, representing 30 vascular plant families and a wide variety of traditional uses. Thirty-two participants among 3 STN workshops identified 1 plant with nematicidal activity, 14 plants with antibacterial activity, 20 plants with antifungal activity, and 41 plants with antioxidant activity. Nearly half of the plants (24 species) demonstrated both antimicrobial and antioxidant activities, correlating to their reported uses to treat both infectious and chronic/metabolic disorders in traditional Ecuadorian medicine. During the STN workshops, participant knowledge of pharmacological screening increased by 77%, whereas knowledge of biodiversity and conservation increased by 69%.

Conclusion: This study demonstrated that STN technologies, employed through a participatory research approach, are highly efficient in the detection of biochemical activities of traditionally used plants. Furthermore, edible Ecuadorian plants possess nematicidal, antimicrobial and antioxidant properties with potential for further development as functional foods, botanical supplements, or cosmetics.

Keywords: Agrobiodiversity; Bioactivity; Bioexploration; Ecuador; Functional food; Medicinal plants; Traditional use; Screens-to-nature

Abbreviations: ABTS-2,2'-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid); CBD-Convention on Biological Diversity; ESPOL-Escuela Superior Politécnia del Litoral; GIBEX-Global Institute for BioExploration; INSPI-National Institute of Public Health; MTT-3-[4,5-dimethylthazol-2-yl]-2,5-diphenyltetrazolium bromide; STN-Screens-to-nature; UCuenca-Universidad de Cuenca; UDLA-Universidad de las Américas; UPSE-Universidad Estatal Península de Santa Elena.

Introduction

Edible plants offer a cocktail of biologically active phytochemicals that can be harnessed to promote human health and wellness in the form of functional foods, botanical dietary supplements, or cosmetics. Commercialization of plant-derived health products can provide direct economic benefits to local agricultural communities and generate value in biodiversity conservation [1]. Meanwhile, food-based therapeutics serve as accessible, affordable forms of medicine, presenting a socioeconomic advantage in developing countries and low income communities [2]. The ethical research and development of food-based therapeutics can, therefore, mutually reinforce the three pillars of sustainable development: economic development, social development, and environmental protection [3].

Ecuador's biocultural diversity [3] is a strategic area of development. As a mega biodiverse country, Ecuador is comprised of over 17,000 species of vascular plants [4], nested within distinct geographic regions. Furthermore, 17 different cultural groups live in Ecuador, each holding a long history of traditional medicinal uses of edible plants [4]. In all major towns and cities, local markets contain specific stalls dedicated to the distribution of plants recognized for their therapeutic value. Due to Ecuador's long history of ethnobotanical research, competent inventories of the traditional uses of Ecuadorian plants have been compiled [4,5]. However, systemic categorization of the bioactive properties of a wide number of traditionally used Ecuadorian plants has not been conducted. A pharmacological assessment of edible plants available in local markets can provide baseline data for further development of phytomedicines that may drive sustainable development.

Participatory research approaches, which have been successful in various aspects of the social and natural sciences [6], may be an effective

***Corresponding author:** Brittany L. Graf, Ph.D, Department of Plant Biology and Pathology, Rutgers University, 59 Dudley Rd, New Brunswick, NJ, USA, Tel: 848-932-6342; E-mail: britgraf@gmail.com

Received October 12, 2015; **Accepted** January 05, 2016; **Published** January 10, 2016

Citation: Graf BL, Rojas-Silva P, Baldeón ME (2016) Discovering the Pharmacological Potential of Ecuadorian Market Plants using a Screens-to-Nature Participatory Approach. J Biodivers Biopros Dev 3: 156. doi:10.4172/2376-0214.1000156

way to perform pharmacological screening in Ecuador. Participatory approaches empower local people to acquire and use the knowledge generated by employing local people in the research activities [7]. Furthermore, participatory approaches are highly integrative, engaging multiple stakeholders across disciplines [8]. Lastly, these approaches utilize innovative technologies that enhance adaptive capacity within resource-limited settings, which can rapidly generate knowledge while transferring skills to research participants [7,8].

Screens-to-nature (STN) is a relatively new participatory approach to enable a broad-spectrum survey of the potential bioactivities of plants while providing community engagement [9]. STNs, developed by the Global Institute for BioExploration (GIBEX) [10], are rapid, cost-effective, robust, field-deployable pharmacological assays that are transferred to partnering research institutions through participatory workshops. STN workshops have been shown to enable rapid detection of biologically active natural products, highly engage students in science, build research and educational infrastructure, and enhance academic/community networks [11].

The objective of this study was to investigate the pharmacological potential of food plants in Ecuador using the STN participatory approach in parallel with an assessment of the educational impact of STN workshops. Plants were collected from local markets within three regions of Ecuador: Northern highlands (Quito), Southern highlands (Cuenca), and the Coast (Santa Elena). Three STN workshops each comprised of mainly university students, technicians, and professors were conducted to explore the antibacterial, antifungal, nematicidal, and antioxidant activities of these plants. At the conclusion of each workshop, participants conducted a written evaluation to assess the educational impact of the STN approach. Collectively, this work offers baseline data for a comprehensive study on the therapeutic potential of Ecuadorian market plants, while demonstrating the utility of the STN approach to effectively conduct pharmacological screening in resource-limited settings.

Materials and Methods

Study sites

In order to broadly survey the food plants from the diverse agroecological regions of Ecuador, the present study was conducted in three distinct sites in Ecuador: Quito, Cuenca, and Santa Elena (Figure 1). Quito is situated 2850 m above sea level in Pichincha Province in the northern Sierra region (the Andes Mountains). Cuenca is located in Azuay Province in the southern Sierra at 2500 m above sea level. Meanwhile, Santa Elena city is located near sea level on the coast in Santa Elena Province in the southwestern lowlands of Ecuador [12]. The climate of Ecuador is characterized by two seasons: the wet season from October- May, and the dry season from June-September. Quito and Cuenca experience a temperate climate with average annual temperatures of 15°C, whereas Santa Elena has a hot tropical climate with average temperatures of 23°C. Each site is inhabited by a mix of ethnicities, among which Mestizo (mixed racial origin) is the most dominant group, though the Kichwa are prevalent among the Sierra region [12].

Plant collection, identification and uses

Plant samples were purchased from one of the largest, most diverse traditional markets within each of the study sites, including Mercado Iñaquito (Quito), Mercado 10 de Agosto (Cuenca) and Mercado Santa Elena (Santa Elena). Plants were co-selected by author BLG and one local workshop participant from each of the study sites based on association to traditional medicinal uses. Plants that are widely available outside Ecuador were excluded from the study. Prior permission/authorization to collect and study the plants was obtained from the Ecuadorian Ministry of Environment following national and international legislation. Collected plants were photographed, taxonomically identified, vouchered and archived at the National Herbarium of Ecuador (QCNE), Quito. Literature review of the traditional medicinal uses of each plant was conducted using reference texts [5] and [13]. Human ailments for which each plant has a history of use were grouped into 15 categories such as gastrointestinal, respiratory, dermatological and others, as indicated in Supp. Table 1 or were grouped as they relate to infectious versus chronic ailments (Supp. Table 2).

Screens-to-nature (STN) workshops

Workshops were carried out from September-November 2014 at the following locations: Universidad de Las Américas (UDLA)-Quito, Universidad de Cuenca (UCuenca) and Universidad Estatal Península de Santa Elena (UPSE). Between 8-17 workshop participants were hosted at each site. Representatives from neighboring universities and research institutions also attended, including the National Institute of Public Health (INSPI) in Quito and Escuela Superior Politécnia del Litoral (ESPOL) in Santa Elena. Workshop participants included students, technicians, professors and investigators from the following fields: agriculture, agroindustry and nutrition, biochemistry, biology, biomedical sciences, chemistry, marine biology, and pharmacy. Each STN workshop consisted of a 3-day program, implemented as follows: day 1 - introduction, plant voucher specimen preparation, plant extraction; day 2 - antibacterial, antifungal, nematicidal and antioxidant assays; day 3 - review of all results, completion of workshop evaluations. Plant extraction and biochemical assays were performed as described previously [9] with minor modifications, as detailed below. All workshop materials were provided to the collaborating institutions free-of-charge, and STN manuals illustrating each step-by-step procedure in Spanish were provided to every participant. The workshops afforded participants the opportunity to collaborate with the research team throughout the entire process, from the development of research questions and experimental design through to the execution of the study and analysis/discussion of the findings. Each participant

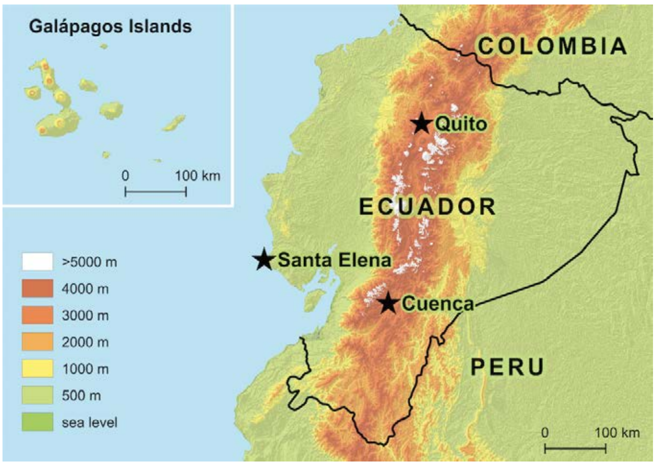

Figure 1: Map of research sites in Ecuador: Quito (northern highlands), Cuenca (southern highlands) and Santa Elena (coast). At each of these sites, plants were purchased from local markets, extracted, and screened for biological activity. Colors correspond with elevation (m above sea level).

was given the opportunity to speak about the relevance of the research to their life/work and the outcomes they hoped to obtain from it.

Plant extraction

An individual ethanolic-based extract was prepared from each fresh plant within 2 days of collection according to the procedure described previously [14] with modification. Two grams of plant material was extracted in 4 mL of 95% ethanol, thereby producing an extract with final concentration of roughly 500 mg/mL. Extracts were subjected to bioassay within 24-48 hours of extraction at final concentrations of 0.03-0.25 mg/mL, depending on the assay.

Antibacterial assay

Antibacterial properties were determined using the agar dilution test as described in [15] with modifications as detailed in [16]. Bacterial inoculums were collected from human saliva and incubated on a 48-well LB agar plates overnight together with plant extracts at 37°C. Fresh-squeezed garlic juice was used as a positive control.

Antifungal assay

Antifungal assays were performed as described earlier [16,17]. Yeast (*Saccharomyces cerevisiae*) was exposed to plant extracts for 12 h in 24-well plates and viability was determined using 3-[4,5-dimethylthazol-2-yl]-2,5-diphenyltetrazolium bromide (MTT) (Sigma). Fresh-squeezed lemon juice was used as a positive control.

Nematicidal assay

The free-living nematode *Panagrellus redivivus*, cultivated on oatmeal and baker's yeast medium, was used as a model organism for roundworm lethality via worm motility. The assay was performed as described previously [18] in a 96-well plate. Copper sulfate (15 mg/mL) was used as a positive control.

Antioxidant assay

Antioxidant activity was determined using a colorimetric assay similar to the method used earlier [19] in which 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid) (ABTS; Sigma) was converted to its radical cation by the addition of potassium persulfate. When reacted with antioxidants, the blue radical cation was immediately converted back to its colorless neutral form. Fresh-squeezed lemon juice and ascorbic acid (0.84 mg/mL) were used as positive controls.

Bioactivity scoring

Results of each STN test were scored qualitatively from 0 to 3, with score 0 representing no visible activity compared to the negative control and 3 representing the highest activity. For example, in the antibacterial assay, extract-treated wells containing a lawn of bacteria like the negative control wells received a "0," and extract-treated wells that appeared completely clear of bacterial growth like the positive control received a "3." In the antifungal assay, extract-treated wells which appeared dark purple in color like the negative control wells received a "0," whereas extract-treated wells which appeared yellow like the positive control wells received a "3." In the nematicidal assay, extract-treated wells in which all worms swam vigorously like the negative control received a "0," whereas extract-treated wells in which all worms appeared immobile like the positive control received a "3." In the antioxidant assay, extract-treated wells which appeared blue-green in color like the negative control wells received a "0," whereas extract-treated wells which appeared clear like the positive control received a "3." Extracts which exhibited low-moderate activity were scored "1" or "2," depending upon subjective assessment. Finally, the number and strength of activities per plant were compared.

Participant assessment of STN workshops

Permission to assess the educational impact of the STN workshops among participants was provided in the IRB #2014-1201. A longitudinal study using a retrospective pretest-posttest survey questionnaire at the conclusion of each workshop captured two time points simultaneously (before and after the workshop). The retrospective pretest method has been shown to be effective and not significantly different from a traditional pretest method in other studies [20]. Scale scores were compared across time points using two-way ANOVA for retrospective pretest-posttest. $P<0.05$ was considered significant.

Results and Discussion

Diversity and traditional uses of plants

The collected plants represented a wide range of biological and cultural diversity. Fifty plant species, belonging to 46 genera and 30 vascular plant families, were collected (Table 1 and Figure 2A). The dominant families were the Asteraceae and Solanceae (6 species each, 12%), followed by Fabaceae (5 species, 10%). Other families with represented by 2-3 species each include Amaranthaceae, Brassicaceae, Myrtaceae, Passifloreaceae, and Rosaceae, while the remaining 22 families had a single species representation. The wider utilization of species from Asteraceae, Solanaceae, and Fabaceae families might relate to the presence of bioactive chemical constituents effective in treating human ailments. Our results corroborate the findings of previous ethnobotanical surveys conducted in other countries such as Pakistan and Israel, where ethnomedicines are mostly prepared from Asteraceae, Solanaceae, and Fabaceae plants [9,21].

Most of species were native to Ecuador, but 40% of them were introduced from different origins (Table 1 and Figure 2B). Among the 50 plants selected, 90% are documented to be cultivatable (Figure 2C). These data indicate that development of functional products from a wide range of Ecuadorian food plants can lead to economic income and sustainability opportunities. Cultivatable plants can provide renewable sources of bioactive compounds and generate economic income among agricultural communities to promote sustainable production and conservation methods [1]. Meanwhile, geographical indications of native plants can be a valuable marking tool, offering improved market access and added value to niche products [22].

Literature review found over 150 medicinal indications recorded for these plants to treat a wide range of illnesses and ailments (Supp. Table 1). The highest number of species was reported for gastrointestinal ailments (34 species), followed by dermatological, respiratory and neuropsychological ailments (27 species each) (Table 1 and Figure 2D). These results are similar to those of ethnobotanical studies conducted in other countries, where gastrointestinal and dermatological disorders were among the most commonly treated by investigated plants [21]. The use of a wide number of plant species for the treatment of these ailments in Ecuador may be due to the high occurrence of these problems in the region as a result of inadequate sanitation, lack of clean drinking water and limited access to medical care [23].

Bioactivities of plant extracts

Plants displayed a range of bioactivities with 1, 14, 21 and 41 species showing nematicidal, antibacterial, antifungal and antioxidant activities, respectively (Table 1 and Figure 3A). Previous studies have shown that the investigation of plants with traditional medicinal use

Citation: Graf BL, Rojas-Silva P, Baldeón ME (2016) Discovering the Pharmacological Potential of Ecuadorian Market Plants using a Screens-to-Nature Participatory Approach. J Biodivers Biopros Dev 3: 156. doi:10.4172/2376-0214.1000156

Table 1: Edible plants selected from local markets in Ecuador and their functional properties, exhibited using screens-to-nature (STN) assays**.** References, if not obtained from Enciclopedia de las Plantas Utiles del Ecuador [5], are cited. Markets are listed as follows: C=Mercado 10 de Agosto, Cuenca; SE=Mercado de Santa Elena, Santa Elena; Q=Mercado Iñaquito, Quito. Activities in the STN assays were rated 0-3, in which 0=no activity, 1=low activity, 2=moderate activity, 3=high activity. STN results among all plants tested are summarized in Figure 3.

Scientific family and species name (voucher no.)	Common name(s)	Plant habit	Cultivated	Native	Traditional medicinal uses	Market	Plant part extracted	Antibacterial	Antifungal	Nematicidal	Antioxidant
Adoxaceae											
Sambucus nigra L. (0238994)	tilo	tree	x	-	nervous afflictions, kidneys, respiratory ailments, colds, cough, lung afflictions, fever, flu, diaphoretic	C	stems, flowers	0, 0	0, 0	0, 0	0, 1
Amaranthaceae											
Aerva sanguinolenta (L.) Blume (0238995)	escancel	herb	x	-	headache, cough, respiratory ailments, expectorant, circulatory problems, purgative, colic, digestive ailments, skin infections, lesions, mosquito bites, wounds, liver, inflammation, kidneys, fever	SE	stems, leaves	0, 0	0, 1	0, 0	0, 0
Amaranthus hybridus L. (0238996)	ataco	herb	x	x	clean the blood, treat the heart, postpartum afflictions, regulate menstruation, nervous attacks, pimples, carminative, clean digestive system, diarrhea	Q	leaves, inflorescences	0, 0	0, 0	0, 0	1, 0
Anacardiaceae											
Spondias purpurea L. (no voucher)	ovito, hobito	tree	x	-	vision problems, wounds, rash, astringent, kidneys, viral infection, body pain, prostate	Q	seeds, fruit casing, fruit pulp	0, 2, 3	0, 0, 0	0, 0, 0	3, 3, 1
Aquifoliaceae											
Ilex guayusa Loes. (0238997)	guayusa	tree	x	x	mouthwash, stimulating, awakening, headache, stress, hangover, depression, purify body, indigestion, child diarrhea, abdominal pain, stimulate stomach function, colic, female fertility, pregnancy, postpartum afflictions, flu, fever, rheumatism, arthritis, body pain, snakebite, kidney	Q	leaves	0	0	0	3
Asteraceae											
Artemisia vulgaris C. B. Clarke (0238998)	artemisia	herb	x[32]	x	antihelminthic, emmenagogue, antispasmodic	SE	stems, leaves	0, 0	1, 0	0, 0	1, 2
Bidens andicola Kunth (0238999)	ñachak	herb	-	x	excessive vaginal fluid, facilitate childbirth, postpartum, diarrhea, cholera, stomachache, nervous afflictions, liver, jaundice, skin problems, burns, bruises, colds, expectorant, asthma, eye inflammation, kidney, diuretic	C	leaves, flowers	0, 0	1, 1	0, 0	3, 3
Chuquiraga jussieui J. F. Gmel. (0239000)	chuquiragua	shrub	x	x	bad breath, clean blood, colds, cough, infections, rheumatism, menstrual discomfort, kidney, wounds, rashes, fever, flu, colic, gastrointestinal secretions, heart pain, inflammation, liver, jitters	Q	stems, leaves, inflorescences	0, 0, 0	0, 0, 0	0, 0, 0	1, 2, 3
Gamochaeta americana (Mill.) Wedd. (0239001)	lechuguilla	herb	-	x	flu, diarrhea, wound healing	C	leaves, flowers	0, 0	2, 1	0, 0	3, 1
Jungia rugosa Less. (0239002)	carne humana, fompo, guayombo	vine or shrub	-	x	bruises	C	stems, leaves	0, 0	0, 0	0, 0	1, 3
Matricaria recutita L. (0239003)	manzanilla	herb	x	-	stomachache, digestive ailments, carminative, antispasmodic, eye washes, eye irritation, colds, cough, child bronchitis, gargle, deafness, kidney, diuretic, bladder pain, pimples, skin problems, bruises, flu, pains, infections, tumors, headache, insomnia, inflammation, vaginal labial inflammation	C	stems, flowers	0, 0	0, 0	0, 0	0, 1
Basellaceae											
Ullucus tuberosus Caldas (no voucher)	melloco	herb	x	-	childbirth, toothache	Q	tubers	0	0	0	0
Boraginaceae											
Borago officinalis L. (0239004)	borraja	herb	x	-	nervous affliction, vitality, refreshing, postpartum, emmenagogue, galactogogue, menstrual problems, regulate menstruation, blood disorders, measles, fever, colds, cough, throat, lungs, whooping cough, bronchitis, diuretic, nephritis, skin irritation, stimulate cutaneous functions, abscesses, growths, diarrhea, spasms	C	stems, flowers	0, 0	0, 0	0, 0	0, 0
Brassicaceae											
Matthiola incana (L.) W. T. Aiton (0239005)	alhelí morado, flor de lilea morada	herb	x	-	purify blood, fever, feminine discomforts, liver, cough, heart, chest pain, digestive ailments, stomach pain, parasites, nervous afflictions, colds	SE	stems, leaves, flowers	0, 0, 0	0, 1, 0	0, 0, 0	0, 1, 2

Nasturtium officinale W. T. Aiton (0239006)	berro	herb	x	-	purify blood, chest, stop hemorrhages, liver, eliminate excess bile, gallbladder, stimulant, kidneys, lungs, throat, expectorant, dropsy, skin afflictions, facial scars, iron deficiency, digestion	C	leaves	0	0	0	0
Cactaceae											
Hylocereus megalanthus (K. Schum. ex Vaupel) Ralf Bauer (no voucher)	pitajaya		x[33]	x[33]	no recorded information	SE	seeds, fruit pulp	0, 0	1, 1	0, 0	0, 0
Caricaceae											
Vasconcellea stipulata (V. M. Badillo) V. M. Badillo (no voucher)	siglalón, chamburo, jigacho de monte, toronche	shrub-tree	x[34]	x	no recorded information	C	seeds, fruit casing, fruit pulp	0, 0, 0	0, 0, 0	0, 0, 0	0, 0, 1
Equisetaceae											
Equisetum giganteum L. (0239007)	cola de caballo	polypodiaphyta (non-seed plant)	x[35]	x	clean blood, hemorrhages, pimples, irritation, astringent, wash wounds, skin, wound healing, liver, kidneys, urinary system, diuretic, headache, emmenagogue, dropsy, cystic ulcerations, lungs, chronic cough, stomach acid, purgative, digestive ailments, mouth and gum inflammation	Q	stems, leaves	0, 0	0, 0	0, 0	2, 2
Ericaceae											
Vaccinium floribundum Kunth (no voucher)	mortiños	shrub	x	x	diabetes, lung, kidney, liver, rheumatism, fever, flu, weakness, nerves, hangover, stomach pain, colic	Q	entire fruit	0	0	0	3
Fabaceae											
Spartium junceum L. (0239008)	retama	shrub	x[36]	x[37]	laxative, heart, kidney, liver, stomach pain, urine retention[13]	Q	aerial parts	0	0	0	2
Lupinus mutabilis Sweet (no voucher)	chocho, lupino	shrub	x	x	wash and soften the face, rheumatism, fever, cholesterol, vermifuge, hypoglycemic agent	Q	seeds	0	0	0	0
Otholobium mexicanum (L. f.) J. W. Grimes (0239009)	trinitaria	shrub	-	x	contraceptive, postpartum afflictions, sheep ticks, diarrhea, stomach acid, stomach pain, indigestion, gas, intestinal infections	C	leaves, inflorescences	0, 0	0, 0	0, 0	3, 0
Tamarindus indica L. (no voucher)	tamarindo	tree	x	-	cholera, laxative, purgative	Q	fruit pulp	2	3	0	2
Vicia faba L. (no voucher)	haba	herb	x	-	inflammation, headache, fever, pus-filled wounds, tumors	SE	fruit casing, fruit pulp	0, 0	2, 0	0, 0	3, 0
Gentianaceae											
Centaurium quitense (Kunth) B. L. Rob (0239010)	canchalagua	herb	x[38]	x[39]	purify blood, cure fright, stomachache, fever, tonic, diaphoretic, regulate menstruation, kidney, malaria, pimples, rash, scurvy, inflammation, liver	SE	roots, stems, leaves, flowers	2, 0, 0, 0	0, 1, 1, 1	0, 0, 0, 0	1, 1, 2, 3
Lamiaceae											
Mentha pulegium L. (0239011)	menta	herb	x	-	colic, gas, nervous afflictions	C	stems, leaves	0, 0	2, 0	0, 0	1, 2
Malvaceae											
Malva pusilla Sm. (0239012)	malva	herb	x	-	diarrhea, inflammation	Q	stems, leaves, flowers	0, 0, 0	0, 0, 0	0, 0, 0	0, 2, 0
Myrtaceae											
Eucalyptus globulus Labill. (0239013)	eucalypto	tree	x	-	rheumatism, toothache, mouth disorders, throat disorders, colds, bronchitis, asthma, flu, convalescents	SE	stems, leaves	2, 1	2, 0	0, 0	3, 2
Myrcianthes hallii (O. Berg) McVaugh (0239014)	arrayán	shrub-tree	x	x	whiten teeth, cavities, bleeding gums, toothache, colds, lung afflictions, wound healing, heal the navel, colic, rheumatism, postpartum, night sweat, liver, child diarrhea	Q	stems, seeds	0, 1	0, 0	0, 0	3, 1
Psidium guajava L. (no voucher)	guayaba	shrub-tree	x	x	difficulty breathing, colds, rheumatism, diarrhea, stomachache, wounds, childbirth, vaginal washes, jaundice	C	fruit casing, fruit pulp	0, 0	2, 0	0, 0	3, 1
Onagraceae											
Fuchsia loxensis Kunth (0239015)	pena pena	shrub	x	x	fever, heart afflictions	C	flowers	3	0	0	3
Passifloraceae											
Passiflora edulis Sims (no voucher)	maracuyá		x	-	parasites	SE	fruit casing, fruit pulp, seed embryo	3, 0, 0	2, 0, 0	0, 0, 0	0, 3, 0

Passiflora ligularis Juss. (no voucher)	granadilla	bejuco	x	x	pimples, kidneys, urinary system, fever, indigestion, intestinal inflammation, nervous afflictions, liver, stop bleeding	C	seeds, fruit casing, fruit pulp	0, 0, 1	0, 0, 0	0, 0, 0	3, 0, 0
Piperaceae											
Peperomia inaequalifolia Ruiz & Pav. (0239016)	congona	herb	x	x	earache, deafness, heart afflictions, stimulate heart, headache, female fertility, menstrual cramps, postpartum, kidney, liver	C	stems, leaves	1, 1	0, 0	0, 0	0, 0
Plantaginaceae											
Plantago major L. (0239017)	llantén	herb	x	-	toothache, mouth and tooth inflammation, purify, colds, cough, tuberculosis, throat problems, parasites, dysentery, ulcers, stomach pain, burning eyes, wound healing, pimples, bruises, hemorrhoids, blood pressure, heart afflictions, tumors, liver, kidney, diuretic, urinary retention, menstruation, tiredness, fever, infections	Q	roots, leaves, seeds	0, 0, 1	0, 0, 0	0, 0, 0	0, 3, 1
Poaceae											
Zea mays L. purple variety (no voucher)	maíz morado	herb	x	-	tumors, wounds, bruises, ulcers, diarrhea, liver, kidney, measles, tiredness, nerves, weakness of heart, hemorrhages, postpartum	Q	seeds	0	0	0	3
Proteaceae											
Oreocallis grandiflora (Lam.) R. Br. (0239018)	gañal	shrub-tree	-	x	hernia, astringent, burns, menstrual afflictions, ovarian inflammation, eye problems, kidney, uterine flow, diuretic, liver, flu, fever, constipation, intestinal afflictions, gastric ulcer, diabetes, cholesterol, inflammation	C	leaves, flowers	0, 0	1, 0	0, 0	3, 2
Rosaceae											
Sanguisorba minor subsp. *muricata* (Spach) Briq. (0239019)	pimpinela	herb	x	x	purify blood, cleansing, bad teeth, pimples, astringent, heart pain, haemostatic, nervousness, emmenagogue	C	leaves	2	1	0	3
Prunus domestica L., golden variety (no voucher)	reina claudia	shrub	x	-	constipation, cough, throat problems	SE	fruit casing, fruit pulp	0, 2	3, 2	0, 0	3, 3
Rutaceae											
Ruta graveolens L. (0239020)	ruda	shrub	x	-	stomach cramps, stomach ache, colic, purging, antihelminthic, antispasmodic, nerves, vertigo, headache, nervous afflictions, stimulant, hysteria, epilepsy, rheumatism, arthritis, earache, pneumonia, fever, flu, plague killer, scabies, bruises, regulate menstruation, menstrual colic, childbirth, abortive, stimulate menstruation, heart problems, damaged blood, eye inflammation, cramps	SE	roots, leaves, flowers	0, 2, 0	0, 0, 0	0, 0, 0	1, 3, 3
Solanaceae											
Brugmansia sanguinea (Ruiz & Pav.) D. Don (0239021)	guanto rojo, floripondio rojo	shrub-tree	x	x	hallucinogen, heachache, analgesic, calming, promotes childbirth, menstrual disorder, asthma, varicose veins, inflammation, wounds, fractures, rheumatism, fever, colic	Q	flowers	0	0	0	2
Physalis peruviana L. (no voucher)	uvilla	herb-shrub	x	x	increase talking process of children, fever, diuretic, kidneys, bladder pain, expulse coagulated blood in the stomach, styes, upset stomach, child diarrhea, colic, infections, asthmatic cough, cough, skin afflictions, ringworm, infected wounds	Q	entire fruit, fruit casing	0, 0	0, 0	0, 0	0, 1
Solanum betaceum Cav. (no voucher)	tomate de árbol	tree	x	x	anemia, tonsillitis, throat pain, liver inflammation	C	fruit casing, fruit pulp	0, 0	0, 0	0, 0	1, 0
Solanum muricatum Aiton (no voucher)	pepino dulce	shrub	x	x	headache, cancer, pimples, rheumatism, arthritis, intestinal ulcers, purgative	SE	fruit pulp	0	1	0	1
Solanum nigrum L. (0239022)	hierba mora	herb-shrub	x[40]	x	insecticide, cloudy eye, eye inflammation, colds, infections, gastritis, intestinal cleaning, colon, cholera, purgative, stomach ache, antispasmodic, flu, fever, mumps, smallpox, liver, nerves, anger attack, hangover, narcotic, kidney, diaphoretic, lice, bruises, skin irritation, growths, rash, wounds, erysipelas, infected wounds, toothache, pains after childbirth	C	leaves, entire fruits	0, 0	1, 0	0, 0	3, 1
Solanum quitoense Lam. (no voucher)	naranjilla	shrub	x	x	angina, throat inflammation, diarrhea	C	fruit casing, fruit pulp	0, 0	0, 3	0, 0	0, 0
Tropaeolaceae											
Tropaeolum tuberosum Ruiz & Pav. (no voucher)	mashua, mashwa, isaño	herb	x	x	rheumatism, scurvy, repels insects and other invertebrates	Q	fruit casing, fruit pulp	2, 1	0, 0	3, 2	1, 1
Urticaceae											

Urtica dioica L. (0239023)	ortiga	herb	x[41]	-	cramps, nerves, varicose veins, liver, kidneys, colds, headaches, back pain, arm pain, knee pain, muscle pain	SE	roots, stems, leaves	0, 0, 0	2, 2, 0	0, 0, 0	0, 0, 2
Verbenaceae											
Aloysia citriodora Palaú (0239024)	cedrón	shrub	x	x	carminative, antispasmodic, purgative, improve digestion, stomach pain, burping, flatulence, diarrhea, intestinal gas, heart problems, chest pain, blood pressure, nerves, fainting, sedative, liver, diabetes, colds	C	stems, leaves	0, 0	0, 0	0, 0	0, 3
Violaceae											
Viola odorata L. (0239025)	violeta	herb	x	-	antispasmodic, dysentery, constipation, stomach and intestinal abscesses, ulcers, foot and mouth disease, burns, wounds, external inflammation, cough, colds, eliminate phlegm, bronchial problems, throat problems, smallpox, flu, chest pain, haemostatic, intoxications, cancerous ulcers	C	flowers	0	0	0	0

(ethno-directed research) consistently leads to higher discovery of bioactivities than that of randomly selected plants [24,25]. Therefore, the high number of bioactivities observed in our study was expected.

A total of 99 extracts were prepared from the 50 plants because, in most cases, multiple plant parts (i.e., leaves, flowers, stems, and roots) were extracted. Different parts of the same plant showed differential activities (Table 1). This pattern has been shown in many other plant species, in which the antioxidant and biological potency of extracts derived from different plant parts correlates to the quantity and quality of active phytochemicals within them [26].

Nearly half of the plants (24 species) exhibited multiple activities (Table 1 and Figure 3B), comprised of a combination of antimicrobial and antioxidant activities (Figure 4B). Most plants included in this study (78%) have been reported for treatment of both infectious diseases and chronic/metabolic disorders (Figure 4A). By simultaneously combating both the viability of microbial pathogens and the production of inflammation-related free radicals in diseased organisms, these plants may offer a pharmacologically valuable cocktail of co-acting phytochemicals worthy of further study and development.

A significant portion of plants (34%) demonstrated antioxidant activity alone. The intake of antioxidant-rich plants has been correlated to a decrease in the risk of developing chronic, metabolic disorders such as diabetes, cardiovascular disease, and cancer [27]. Natural antioxidants, such as vitamins, carotenoids, polyphenols and organosulfur compounds, scavenge reactive oxygen species, or free radicals, thereby inhibiting oxidative damage that leads to inflammatory and degenerative diseases [27-30]. Antioxidants may also be particularly beneficial during infections, since these molecules can maintain immune cell integrity by attenuating the cellular damage induced by the acute oxidative stress produced during immune response to pathogen invasion [27]. Overall, this study initiates the scientific work necessary to validate the traditional uses and ethnopharmacological claims of 50 edible Ecuadorian plant species.

Among the plants which exhibited bioactivity in our study, only a few showed high activity, indicated by a qualitative score of “3” (Table 1). For example, the only species to exhibit high nematicidal activity was *Tropaeolum tuberosum* (mashua). Two plants showed high antibacterial activity: *Spondias purpurea* (ovito) and *Passiflora edulis* (maracuyá). Three plants showed high antifungal activity: *Tamarindus indica* (tamarindo), *Prunus domestica* golden variety (reina claudia amarilla), and *Solanum quitoense* (naranjilla). Meanwhile, twenty-four plants showed high antioxidant activity. These plants include *Spondias purpurea* (ovito), *Ilex guayusa* (guayusa), *Bidens andicola* (ñachak), *Chuquiraga jussieui* (chuquiragua), *Gamochaeta americana* (lechuguilla), *Jungia rugosa* (carne humana), *Vaccinium floribundum* (mortiño), *Genista tinctoria* (trinitaria), *Vicia faba* (haba), *Centaurium quitense* (canchalagua), *Eucalyptus globulus* (eucalypto), *Myrcianthes hallii* (arrayán), *Psidium guajava* (guayaba), *Fuchsia loxensis* (pena pena), *Passiflora edulis* (maracúya), *Passiflora ligularis* (grandilla), *Plantago major* (llantén), *Zea mays* purple variety (maíz morado), *Oreocallis grandifolium* (gañal), *Sanguisorba minor* subsp. *muricata* (pimpinela), *Prunus domestica* golden variety (reina claudia amarilla), *Ruta graveolens* (ruda), *Solanum nigrum* or *Solanum americanum* (hierba mora), and *Aloysia citriodora* (cedrón). These plants demonstrate the highest potential for further study and development of functional foods, botanical supplements, and cosmetics.

Five plants did not exhibit any activity in our assays, including *Ullucus tuberosus* (melloco), *Borago officinalis* (borraja), *Nasturtium officinale* (berro), *Lupinus mutabilis* (chocho) (Latin name), and *Viola odorata* (violeta) (Table 1 and Figure 3B). These plants may be useful for their nutritional characteristics (i.e., vitamin and mineral content) or physical properties (i.e., mucilage), which were not measurable in our assays. Alternatively, these plants may contain antimicrobial agents that were not effective against the specific microbial organisms utilized in our study. A final consideration is that one of these plants (*Lupinus mutabilis*) is processed before distribution in local markets by leaching out toxic alkaloids in water for several hours, thereby affecting the bioactivity of the raw material [31].

Participant learning

We examined the change in self-reported knowledge of workshop participants as a result of a 3-day scientific training program using STN technologies. A total of 32 participants completed a retrospective pretest-posttest survey to score the educational impact of the STN workshops, including 13 professors or postdoctoral researchers and 19 university students or technicians. Scores improved from the retrospective pretest when compared to the posttest on both measures: (1) knowledge of pharmacological screening (2.69 ± 0.20 vs. 4.75 ± 0.08, representing an increase of 77%, $P<0.0001$), and (2) knowledge of biodiversity and conservation (2.59 ± 0.24 vs. 4.38 ± 0.15, an increase of 69%, $P<0.0001$) (Figure 5). Therefore, there was a significant positive change in reported knowledge after the workshop was completed. These results were complimented by positive comments provided by the participants in the open-ended sections of the evaluation form.

Overall, results from the workshop participant survey indicated that the STN participatory approach is highly effective in rapidly increasing the knowledge of multiple aspects of bioexploration. Hence, this approach is useful for both education and start-up research. Long-term surveys will assess whether or not long-term cross-department and cross-institutional collaborations were forged between workshop participants, and if the STN technologies were utilized in coursework and research projects after the workshop.

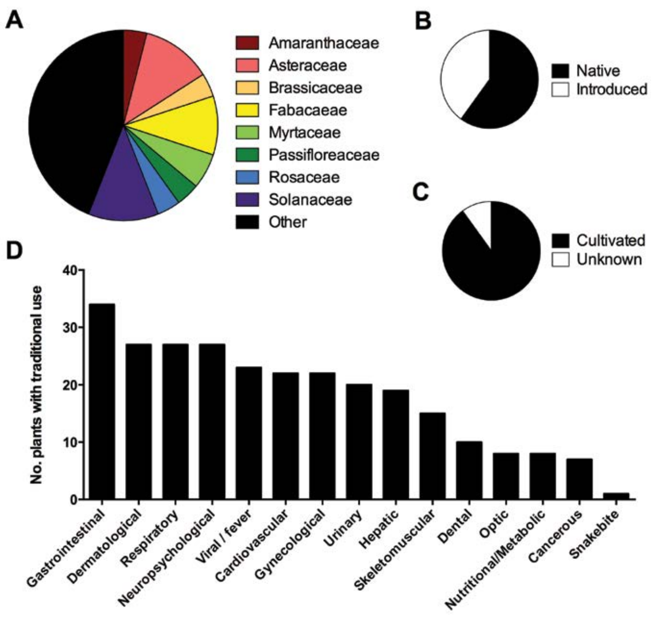

Figure 2: Profile of the 50 Ecuadorian plants collected from local markets. **A**: Distribution of plants among taxonomic plant families. **B:** Number of native and introduced plants. **C:** Number of cultivated plants. **D:** Distribution of plants among categories of reported treatments for ailments associated to the indicated organs and systems. Traditional use categories were determined as specified in Materials and Methods and indicated in Table 1.

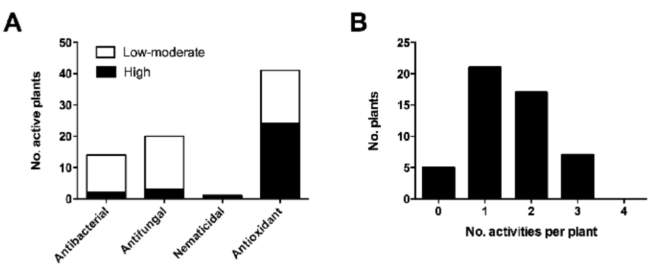

Figure 3: Bioactivities exhibited by edible Ecuadorian plants in screens-to-nature (STN) assays. **A:** Number of edible Ecuadorian plants that exhibited low-moderate activity and high activity in the antibacterial, antifungal, nematicidal, and antioxidant STN assays. Plants were collected from local markets in three different regions of Ecuador, as shown in Figure 1. **B:** Number of plants demonstrating no, one, or more than one bioactivity.

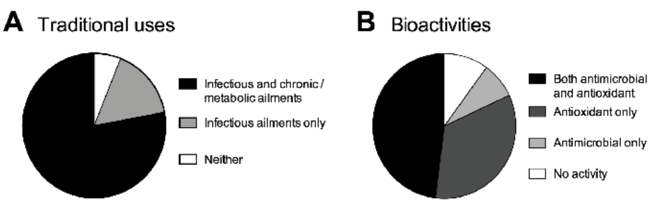

Figure 4: Correlation of traditional uses to bioactivities observed using screens-to-nature (STN) assays. **A:** Portion of plants recorded to treat either (a) both infectious and chronic/metabolic ailments, defined as specified in Table 2, (b) infectious ailments only, or (c) neither infectious nor chronic/metabolic ailments. **B:** Portion of plants that demonstrated (a) Both antimicrobial and antioxidant activities, (b) antioxidant activity only, (c) antimicrobial activity only, or (d) no activity. Antimicrobial activity is defined as either antibacterial or antifungal activity.

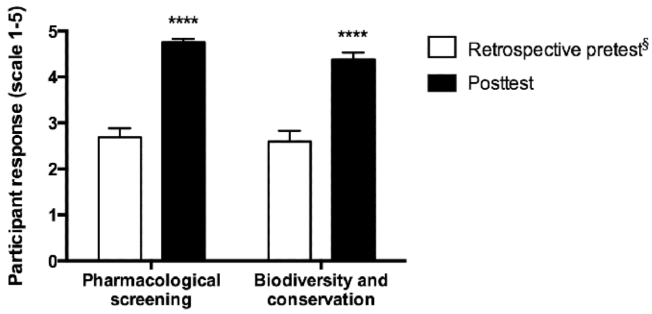

Figure 5: Knowledge acquired by screens-to-nature (STN) workshop participants, determined by participant responses according to time using a retrospective pretest and posttest. §Retrospective pretest: given at end of workshop simultaneously with posttest evaluations. Survey questions based on a 5-point Likert scale where 1 indicates little to no knowledge and 5 indicates high quality of knowledge, i.e., higher number reflects a more positive attitude/change. Data are the mean ± SEM (n=32). *****P* < 0.0001, Sidak 2-way ANOVA.

Conclusion

While there is significant knowledge regarding the traditional medicinal uses of plants in Ecuador, these plants lack scientific and clinical data. We found that the STN approach, based on rapid, portable extraction and bioassay technologies, deployed through participatory research, enabled the simultaneous analysis of four pharmacological activities among a large number of market-collected plants. This project has identified and validated a subset of pharmacologically active Ecuadorian food plants that possess potential to drive sustainable development through scientific discovery, product development and education. Long-term follow-up work will include bioassay-guided isolation and identification of bioactive phytochemicals, extract optimization, toxicity testing and animal model experiments.

Acknowledgment

This study was funded by the 2014-2015 U.S. Fulbright Student IIE Fellowship and the Garden Club of America's Anne S. Chatham Fellowship in Medicinal Botany. The authors thank all screens-to-nature workshop participants for their time and enthusiasm in this project. Special thanks to Cristina Quiroga (UDLA) who played an important role in establishing our research collaboration with the National Herbarium of Quito and mounted the herbarium voucher specimens. We thank Christoph Fink (Universität Saizburg) for assistance in map making. The authors also thank all members of the Global Institute for BioExploration (GIBEX) who played a key role in developing the screens-to-nature assays and workshop format, especially Drs. Gili Joseph, Mary Ann Lila, Josh Kellogg and Ilya Raskin. Lastly, we thank Dr. Ilya Raskin and Lindsey Gohd for editing the manuscript draft.

References

1. Johns T, Eyzaguirre PB (2006) Linking biodiversity, diet and health in policy and practice. Proc Nutr Soc 65: 182-189.
2. Verpoorte R (2012) Good practices: the basis for evidence-based medicines (editorial). J Ethnopharmacol 140: 455-457.
3. Johns T, Sthapit BR (2004) Biocultural diversty in the sustainability of developing-country food systems. Food Nutr Bull Pp: 25.
4. De la Torre L, Cerón CE, Balslev H, Borchsenius F (2012) A biodiversity informatics approach to ethnobotany: meta-analysis of plant use patterns in Ecuador. Ecol Soc 17: 15.
5. De la Torre L, Navarrete H, Muriel MP, Macía MJ, Balslev H (2008) Enciclopedia de las Plantas Utiles del Ecuador. Herbario QCA & Herbario AAU Quito.
6. Probst K, Hagmann J, Fernandez M, Ahby JA (2003) Understanding participatory research in the context of natural resource management-paradigms, approaches and typologies. Agren-Network. Agricultural Research & Extension Network pp: 130.

7. Minore B, Boone M, Katt M, Kinch P, Birch S (2004) Addressing the realties of health care in northern aboriginal communities through participatory action research. J Interprofessional Care 18: 360-368.

8. Sayer JA, Campbell B (2004) Research to integrate productivity enhancement, environmental protection and human development. Currents 33: 4-10.

9. Joseph G, Faran M, Raskin I, Lila MA, Fridlender B (2014) Medicinal plants of Israel: a model approach to enable an efficient, extensive and comprehensive field survey. J Biodivers Bioprospect Devel 1: 10000134.

10. Dushenkov V, Raskin I (2008) New strategy for the search of natural biologically active substances. Russ J Plant Physiol 55: 564-567.

11. Kellogg J, Joseph G, Andrae-Marobela K, Sosome A, Flint C, et al. (2010) Screens-to-nature: opening doors to traditional knowledge and hands-on science education. NACTA Journal 54: 41-48.

12. A Country Study: Ecuador (1991) Library of Congress Country Studies. Washington, DC: Library of Congress.

13. Rios M, Koziol MJ, Pedersen HB, Granda G (2007) Useful Plants of Ecuador: Application, Challenges and Perspectives. Ediciones Abya-Yala: Quito PP: 652.

14. Dey M, Ripoll C, Poulev R, Dorn R, Aranovich I, et al. (2008) Plant extracts from central Asia showing antiinflammatory activities in gene expression assays. Phytother Res 22: 929-934.

15. NCCLS (1988) Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically. National Committee for Clinical Laboratory Standards (NCCLS), Villanova, PA, USA.

16. Andreae-Marobela K, Ntumy AN, Makobela M, Dube M, Sosome A, et al. (2012) “Now I heal with pride” - The application of screens-to-nature technology to indigenous knowledge systems research in Botswana: implications for drug discovery. In: Chubale K, et al. (eds.) Drug Discovery in Africa: Impacts of Genomics, Natural Products, Traditional Medicines, Insights into Medicinal Chemistry and Technology Platforms in Pursuit of New Drugs. Springer-Verlag: Berlin Pp: 239-264.

17. Meletiadis J, Mouton JW, Meis JF, Bouman BA, Donnelly JP, et al. (2001) Colorimetric assay for antifungal susceptibility testing of Aspergillus species. J Clin Microbiol 39: 3402-3408.

18. Simpkin KG, Coles GC (2007) The use of Caenorhabditis elegans for antihelmintic screening. J Chem Technol Biotechnol 31: 66-69.

19. Walker RB, Everette JD (2009) Comparative reaction rates of various antioxidants with ABTS radical cation. J Agric Chem Environ 57: 1156-1161.

20. Swindle S, Baker SS, Auld GW (2007) Operation frontline: assessment of longer-term curriculum effectiveness, evaluation strategies and follow-up methods. J Nutr Educ Behav 39: 205-213.

21. Khan I, AbdElsalam NM, Fouad H, Tariq A, Ullah R, et al. (2014) Application of ethobotanical indices on the use of traditional medicines against common diseases. J Evid Based Complementary Altern Med pp: 1-21.

22. Bramley C, Kirsten JF (2007) Exploring the economic rationale for protecting geographical indicators in agriculture. Agrekon 46: 69-93.

23. Taylor JLS, Rabe T, McGaw LJ, Jager AK, van Staden J (2001) Towards the scientific validation of traditional medicinal plants. Plant Growth Regulation 34: 23-37.

24. Khafagi IK, Dewedar A (2000) The efficiency of random versus ethno-directed research in the evaluation of Sinai medicinal plants for bioactive compounds. J Ethnopharmacol 71: 365-376.

25. Balick MJ (1990) Ethnobotany and the identification of therapeutic agents from the rainforest. In: Chadwick DJ and Marsh J (eds.), Bioactive Compounds from Plants. John Wiley & Sons Ltd. New York, NY Pp: 22-31.

26. Zhu Q, Nakagawa T, Kishikawa A, Ohnuki K, Shimizu K (2015) In vitro bioactivities and phytochemical profile of various parts of the strawberry (Fragaria x ananassa var. Amaou). J Func Foods 13: 38-49.

27. Hughes DA (2002) Antioxidant vitamins and immune function. In: Calder PC, Field CJ, Gill HS (eds.) Nutrition and Immune Function. CABI: New York, NY pp: 171-191.

28. Oyagbemi AA, Azeez OI, Saba AB (2009) Interactions between reactive oxygen species and cancer: the roles of natural dietary antioxidants and their molecular mechanisms of action. Asian Pacific J Cancer Prev 10: 535-544.

29. Soory M (2012) Nutritional antioxidants and their applications in cardiometabolic diseases. Infect Disord Drug Targets 12: 388-401.

30. Rajendran P, Nandakumar N, Rengaranjan T, Palaniswami R, Gnanadhas EN, et al. (2014) Antioxidants and human diseases. Clinica Chimica Acta 436: 332-347.

31. Fornasini M, Castro J, Villacrés E, Narváez L, Villamar MP, et al. (2012) Hypoglycemic effect of Lupinus mutabilis in health volunteers and subjects with dysglycemia. Nutr Hosp 27: 425-433.

32. Wagner WL, Herbst DR, Sohmer SH (1919) Manual of the flowering plants of Hawaii. Revised edition. Bernice P Bishop Museum Special Publication. University of Hawai'i Press/Bishop Museum Press, Honolulu.

33. Ortiz-Hernández Y, Carrillo-Salazar JA, Pitahaya (2012) (Hylocereus spp.) A short review: Comunicata Scientiae 3: 220-237.

34. Restrepo MT, d'Eeckenbrugge GC, Jiménez D, Veja J (2005) Morphological divesity of cultivated mountain papayas (Vasconcellea spp.) in Ecuador. Proceedings of the Interamerican Society for Tropical Horticulture 48: 119-123.

35. Husby CE (2009) Ecophysiology and biomechanics of Equisetum giganteum in South America. FIU Electronic Theses and Dissertations pp: 200.

36. Carruthers SP (1986) Alternative Enterprises for Agriculture in the UK. Centre for Agricultural Strategy, University of Reading.

37. Gilbert KG, Cooke DT (2001) Dyes from plants: past usage, present understanding and potential. Plant Growth Regulation 34: 57-69.

38. Hooker WJ (1834) The Journal of Botany: Containing Figures and Descriptions of Such Plants as Recommended Thems. by Their Novelty, Rarity, History. Longman, Rees, Orme, Brown, Green & Longman: London.

39. Jorgensen PM, León-Yánez S, Cat Vasc Pl (1999) Ecuador. Monogr. Syst. Bot. Missouri Bot. Gard 75: 1-1181.

40. Edmonds JM, Chweya JA (1997) Black nightshades. Solanum nigrum L. and related species. Promoting the conservation and use of underutilized and neglected crops. Institute of Plant Genetics and Crop Plant Research. Gatersleben/International Plant Genetic Resources Institute, Rome, Italy. 92: 9043-9321.

41. Pagliarulo CL, Hayden AL, Giacomelli GA (2004) Potential for greenhouse aeroponic cultivation of Urtica dioica. Acta Hort 659: 61-66.