
TRAMIL Ethnopharmacological Survey: Knowledge Distribution of Medicinal Plant Use in the Southeast Region of Puerto Rico

JOSÉ A. ALVARADO-GUZMÁN, MPH; JANNETTE GAVILLÁN-SUÁREZ, Ph D;
LIONEL GERMOSEN-ROBINEAU, MD, MPH

Background: TRAMIL network aims to understand, validate and expand health practices based on the use of medicinal plants in the Caribbean, a “biodiversity hotspot” due to high species endemism, intense development pressure and habitat loss.

Objectives: The purpose of this study was to document both the medicinal plants that are frequently used to treat health conditions prevalent in the southeastern region of the archipelago of Puerto Rico and the trends in their use among the study population.

Methods: An ethnopharmacological survey was conducted in the study region. The results were analyzed using univariate and multivariate statistical analyses.

Results: Overall, 118 medicinal plants were recorded as being used to treat depression, nervousness, chronic sinusitis, gastritis, gastroesophageal reflux disease, allergic rhinitis, rhinofaryngitis, asthma, arthritis and migraine. The plant species with significant use were

Citrus aurantium L., *Citrus aurantifolia* (Christm.) Swingle, *Pluchea carolinensis* (Jacq.) G. Don in Sweet, and *Mentha piperita L.* The use of medicinal plants is more frequent among single women with a high educational level, a trend similar to the use of CAM in the US.

Conclusion: Ethnopharmacological knowledge and the use of medicinal plants is decreasing in the study region due to an increase in the use of conventional medical care and to self-medication with over-the-counter pharmaceutical products. Four botanical species with significant uses that were not previously recorded in the Caribbean Herbal Pharmacopoeia have been identified. This report will be followed by the scientific validation and toxicity studies of these plant species and the TRADIF activities in the study region.

Key words: Puerto Rico, TRAMIL ethnopharmacological survey, Urban use of medicinal plants

The TRAMIL network (Traditional Medicines in the Islands) was founded in 1982 to understand, validate and expand health practices based on the use of medicinal plants in the Caribbean. At present, 48 TRAMIL ethnopharmacological surveys have been completed in 27 territories of the Caribbean (1). TRAMIL also organizes outreach activities (TRADIF) aimed at disseminating the results obtained from the efficacy and toxicity studies on the use of botanical remedies. TRADIF workshops have been offered at community, primary health care (providers and promoters) and governmental levels. At present, over 90 medicinal plants that have been evaluated by TRAMIL are recognized in Cuba,

Dominican Republic, Honduras, Nicaragua and Panamá as effective treatments that can be incorporated into primary health care programs (2-3). The TRAMIL program also promotes the protection and conservation of traditional knowledge that is now endangered by both the lack of verbal transfer to new generations and the degradation of natural resources (4-5). The Caribbean is one of the world’s centers of biodiversity and endemism identified as tropical mountainous “biodiversity hotspots” because “high species endemism combines with proportionally extensive habitat loss” (6). In Puerto Rico, a component of the Caribbean biodiversity hotspot, approximately 2,900 plant species have been identified, of which 236 are endemic, 135 are commonly used as medicinal plants and 170 plants are less well-known for their therapeutic value (7-8). In order to document and evaluate the traditional knowledge about medicinal plants use at the community level, Puerto Rico joined TRAMIL in 1994 by conducting a field survey in the southwestern region of the island. Among the 100 families surveyed, 159 (medicinal) plants were recorded and sixteen of these plants were identified as plants with “significant use” (reported in at least 20% of the interviews for the same health condition)

*Head, Census Information Center and Biostatistician, Institute of Interdisciplinary Research, †Affiliate Researcher, Institute of Interdisciplinary Research and Professor, Department of Chemistry, University of Puerto Rico at Cayey; ‡TRAMIL Scientific Coordinator, Department of Biology, Faculty of Science, University of French Antilles and Guyana, UAG UFR SEN BP 592, 97159 Pointe à Pitre, Guadeloupe (FWI) Guadeloupe

Address correspondence to: Jannette Gavillán-Suárez, Ph D, #205 Antonio R. Barceló Ave., Cayey, PR 00736. Tel: 787-738-2161 (2357) • Fax: 787-263-1625 • Email: jannette.gavillan@upr.edu

to treat common health problems identified by the study population (1). From December 2006 to January 2007, a TRAMIL ethnopharmacological survey was conducted in the mountainous and coastal southeastern region of the island to expand and report on the evaluation of medicinal plants use in the archipelago of Puerto Rico, The region includes eleven municipalities within the service area of the University of Puerto Rico at Cayey (UPR-Cayey). One important demographic characteristic of this region is its transformation from a rural to an urban area during the last two decades (9). This report describes the medicinal plants and botanical remedies with significant use to treat 10 health problems of high prevalence according to the Puerto Rico Continuous Health Study (10) and examines the trends in medicinal plant use in the region.

Methodology

Geographic coverage

Puerto Rico has three predominant physiographic regions: the mountainous interior, the coastal lowlands, and the karst area in the North. The mountainous interior (covering 60% of the island's territory) is formed by a central mountain range that transects the island from East to West. The northern coastal lowlands extend 13 to 19 km inward and the southern coastal lowlands 3 to 13 km. This study was conducted in the southeastern region of Puerto Rico that includes eleven municipalities located in the mountainous and coastal lands. Figure 1 shows the natural vegetation and land cover distribution of the region.

Study population

The US Bureau of Census' definition for family households (11) was used to identify the number of families in each town/municipality. The Decennial Census of Puerto Rico (2000) was used to determine the number of family

households in each town (12). The total number of family households (sample unit) in the 11 municipalities was 123,855. Based on the total number of family households in the region, a study population of 270 households was calculated using the following equation: $n = NZ_{\alpha}^2 pq / d^2 (N-1) + Z_{\alpha}^2 pq$, where n = minimum sample size; N = total population size (123,855); $Z_{\alpha}^2 = 1.645^2$ ($\alpha = .10$); p = expected proportion (50%); $q = 1 - p = 1 - 0.5 = 0.5$ and $d =$

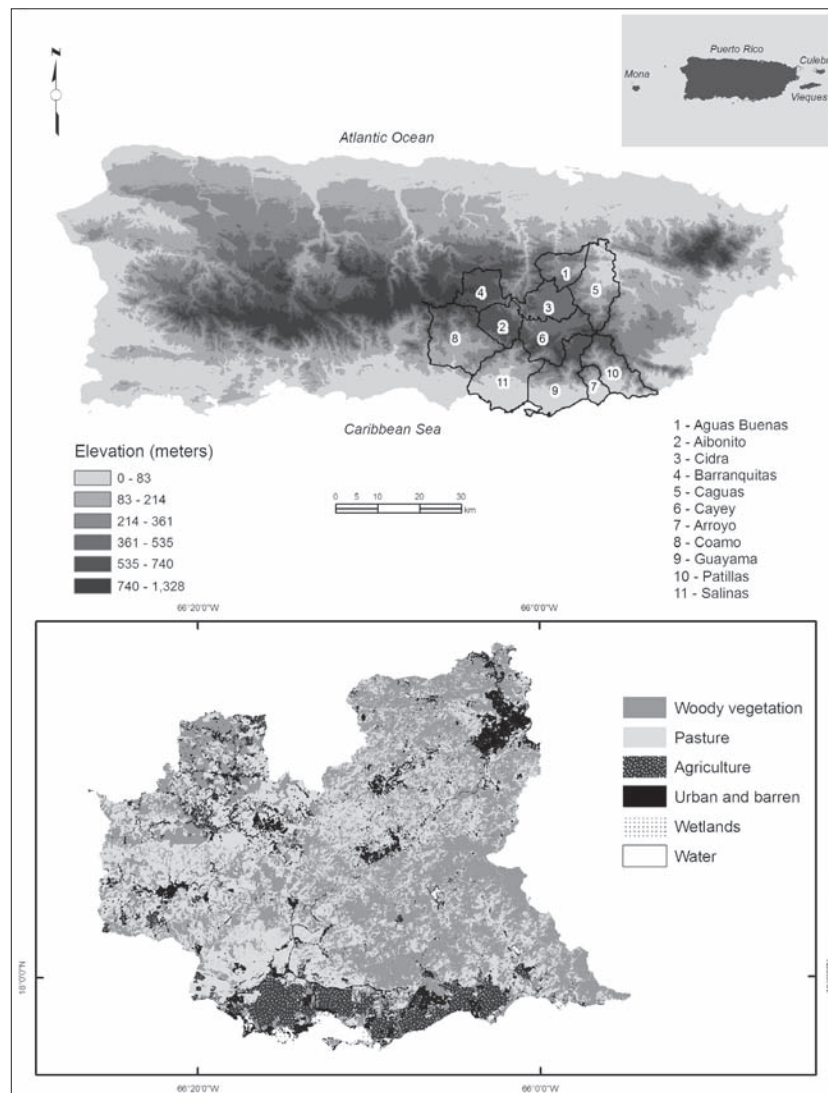


Figure 1. Archipiélago of Puerto Rico study region natural vegetation and land cover distribution: Woody vegetation (24%) includes lowland seasonal evergreen forests where elevations are high enough to experience rainfalls totals above 1200 mm per year and lowland dry semideciduous forests found at elevations generally below 300m with rainfall accumulation of 850 - 1100mm per year, Agriculture (3.2%), Pasture and grassland (54.5%), Wetlands and Urban and developed land cover (7.0%) (Helmer, et al., 2002). Municipalities surveyed: Aguas Buenas, Aibonito, Arroyo, Barranquitas, Caguas, Cayey, Cidra, Coamo, Guayama, Patillas and Salinas with an estimated total population of 467,339 (US Bureau of Census).

precision (5%). To obtain the desired level of precision and reduce the size of the sampling error, the sample size was increased by 30% resulting in a sample of 351 family households (13). Cluster, stratified and systematic sampling techniques were combined to select the family households (sample unit) in the survey. The barrios (town subdivisions) that were included in the survey were those that contained 10% or more of the family households reported for a given town. For example, in Cayey, three town subdivisions were selected (Pueblo, Rincón and Toíta) since they contain respectively 34%, 12.3% and 13.5% (more than 50% of the family households) of the 12,735 family households in the town. The selected town subdivisions are shown in Figure 2a. Each town subdivision selected was stratified by block groups to determine the sectors of the subdivision to be surveyed (14). The selection criterion in this case was that the block groups must be contained within the geographical limits of the town subdivision. Figure 2b illustrates an example of a selected block group in Barrio Pueblo, Cayey. Then, the block groups selected were stratified by median household annual income according to the 2000 Puerto Rico Census, using intervals of ten thousand (US\$). For each income strata, the block group with the largest number of family households was selected for the survey. Finally, using Geographic Information System (GIS) a point file was developed to enumerate all house units located on each of the block groups selected, starting at the house units closer to the main streets. Since the ratio of occupied house units to family households was 2 to 1, the systematic selection of family households included those households that were identified as multiples of three. This selection between clusters increases the surveyed area in a given block and reduces influences between close neighbors. Figure 2c illustrates this mapping. Maps were created using Maptitude Geographic Information System for Windows (version 4.8, Caliper Corp., Newton, Massachusetts, USA).



Figure 2. (a) Selected town subdivisions (“barrios”) in the region; (b) Selected block groups in “Barrio Pueblo”, Cayey and (c) House units that could be surveyed in a block group with id 720352608002 at Barrio Pueblo, Cayey. Maps were created using Maptitude Geographic Information System for Windows (version 4.8, Caliper Corp., Newton, Massachusetts, USA).

Prevalence of diseases surveyed

The Puerto Rico Continuous Health Study, which provides disease prevalence data for each of the seventy eight municipalities of Puerto Rico, was used to identify the specific health conditions surveyed (10). Only self-limiting conditions that might be treated with botanical remedies were considered. From the data for each town, the ten ailments (excluding diabetes and hypertension) that were prevalent in at least five of the towns were selected for the study. These conditions were asthma, arthritis, chronic sinusitis, allergic rhinitis, depression, rhinopharyngitis, gastritis, nervousness, migraine and gastroesophageal reflux disease. The International Classification of Diseases

(ICD-10; 15) provided the diagnostic codes for the health conditions that were discussed with the medical director of the Cayey Municipal Hospital. Information on the nosological entity, symptom description and clinical presentation of the ailments selected for the survey were obtained and used in the survey questionnaire to describe some of the ailments. For example, rhinopharyngitis was described as “nasal secretion with sore throat and hoarseness”, chronic sinusitis as “nasal congestion and inflammation”, allergic rhinitis as “nasal allergy with frequent sneezing”, gastroesophageal reflux disease as “reflux” and nervousness as “nerves”.

Interview methods

The main focus of the TRAMIL survey was to document the use of botanical remedies to treat common health conditions and to assess the perceptions of these ailments by the study population. Following the TRAMIL methodology, the mother or woman in the family was the preferred interview subject over the other members of the family, and informed consent was always sought. Studies on gender roles in relation to the knowledge about medicinal plant use support the idea that the utilization and responsibility of transferring this knowledge is traditionally a women’s domain (16-18). To be eligible for participation in the study, participants had to be at least 18 years of age. Prior to the field work, an interdisciplinary team of undergraduate research assistants at the UPR-Cayey were offered workshops in ethnobotany, voucher preparation and how to conduct interviews. Interviewers were guided during the household selection by protocols that established the working definition of family and established the condition that at least one family member had suffered from one or more of the ten ailments included in this study.

The research assistants approached potential participants in their homes, determined their willingness to participate in the study and administered the survey. The study was divided into three phases to monitor the quality and consistency of the interviews. During each phase, the assistants submitted the research, study instruments (questionnaires, informed consent forms, maps) and vouchers. Field work was supervised and monitored at all times.

Survey instrument

The questionnaire used was approved by the Internal Review Board at the UPR-Cayey and field-tested in a pilot study in a nonparticipating block group, and individual items were refined as needed. The questionnaire was adapted from the one published by TRAMIL (www.tramil.net), which has two sections: the first section collects

specific demographic and socio-economic information from a given community, and the second follows a structured interview, asking participants to provide information about the botanical remedies used by the family as the first treatment for the ailments included in the survey (Table 1). When the participant stated the use of a medicinal plant as the first treatment, open-ended questions were used to obtain a detailed description of the treatment and of the form of preparation and application, including combinations with other plants, dosage and contraindications or side effects for adults and children. The place where the plant was collected or obtained was also identified. The data collected in this section allowed researchers to compare the use of botanical remedies reported in the study with the use in other Caribbean locations as reported in the Caribbean Herbal Pharmacopoeia (1).

Collection of plants

Appropriate vouchers were collected and numbered during the interviews, and photographs were taken when the medicinal plants were obtained from the family’s or neighbors’ backyards. Classification of the botanical species was performed by José Sustache, Botanist and Head of the PR Department of Natural and Environmental Resources Herbarium. The vouchers were deposited at the George Proctor Herbarium (SJ) in Puerto Rico.

Statistical analysis

Data from the questionnaires was entered into an Access (Microsoft Office 2003 Suite) database and imported into SPSS 13.0 (SPSS Inc. Chicago, Ill.) for Windows. Frequencies and cross tabulation tables were used to describe the socio-demographic characteristics of the study participants. Family income was excluded from the analysis since one-third of the families refused to offer this information.

The Continuity Correction of the Chi Square Test was applied to evaluate associations with medicinal plant use (1 if the family used medicinal plants for at least one of the conditions and 2 if not) and the independent variables: age (less or equal to 50 years and more than 50 years), marital status (single, which includes divorced and widowed; and married, which includes cohabiting or living in common-law marital union), education (“below college degree” which includes 6th to 12th grades and technical degrees; and “college degree” which includes 2-year college, baccalaureate and higher degrees), and employment status (unemployed and employed, which includes both part time and full time employment). To measure the strength of the statistically significant associations, odds ratios (OR) and 95% confidence intervals (95% CI) were estimated from

Table 1. Ethnopharmacological survey questionnaire

Part I. Socioeconomic Information		
1. Age		
2. Gender		
a. female	b. male	
3. Marital status:		
a. single	d. widow	
b. married	e. living with a partner	
c. divorce		
4. Education:		
a. first to six grade	d. technical degree (specify):	
b. seventh to nine grade	e. college/university degree	
c. tenth to twelve grade (High School diploma)	f. None	
5. Monthly Family Income		
6. Employment Status		
a. housewife	c. full time employee	
b. student	d. part time employee	
Part II. Ethnopharmacological Survey		
1. Have you or anyone in your family has suffered from one of the following health problems?		
a. asthma		
b. migraine		
c. nasal allergy with frequent sneeze		
d. gastritis		
e. depression		
f. reflux		
g. nasal secretion with sore throat and hoarseness		
h. nasal congestion and inflammation (sinusitis)		
i. nerves		
j. arthritis		
2. Give a brief description of the problem		
3. What was the first treatment you used the last time that you or a member in your family suffered the health problem?		
a. medicinal plant or "home remedies"		
	b. physician consultation	
	c. botanical supplements	
	d. selfmedication with pharmaceutical drugs	
	(If the participant answered b – d the interview is completed for that particular condition.	
4. What plants or combination of plants were used the last time that you suffered the condition? If several plants are mentioned complete Part II for each plant.		
5. What part(s) of the plant did you use to prepare the remedy?		
a. leaves	e. flower	
b. bark	f. fruit	
c. root	g. seed	
d. pulp		
6. Describe how you prepared the treatment?		
a. decoction	d. juice	
b. infusion	e. raw	
c. aqueous steeping		
7. Describe how the treatment was administered:		
a. oral	c. inhalation	
b. bath	d. poultice, compress	
8. In what quantity or dosage did you use the treatment?		
9. For how long?		
10. Where do you get the plant(s)?		
a. garden around the house		
b. in the market		
c. forest		
d. other (specify):		
Take photographs and request a botanical sample if the plant is obtained in the garden. Prepare voucher on site.		
11. Have you used this treatment?		
a. Yes	b. No	
12. What results did you obtained?		
13. Are there any precautions/contraindications when using this remedy?		
14. Do you use this remedy for children? What is the dosage? Are there any precautions/contraindications?		

the corresponding 2 x 2 tables. Based on the previous studies (19-20) showing significant relationships between medicinal plant use and age or employment status, these variables were analyzed as confounding variables of the significant associations according to the Mantel-Haenszel Method (21). A multivariable logistic regression analysis was conducted using the reference variables: age less or equal to 50 years, below college degree, married, and unemployed. The Backward Stepwise method was used to eliminate non-significant factors from the model (22). A Post Hoc Power Analyses (23) was conducted for every association evaluated using G*Power 3.0.10 (Franz Faul, University of Kiel, Germany 1992 - 2008). The Index of Agreement on Remedies (IAR) was calculated for the ten health conditions surveyed in this study based on the following equation: $IAR = (na - nr) / (na + nr - 1)$, where na is the citation frequency of the health condition and nr is the number of different plant remedies cited to treat that health condition (24). The IAR values offer a consensus index of participants about the use of these remedies for a given health condition. IAR values vary between "0"

and "1", representing no consensus and total consensus, respectively. Due to the limitation of the index, IAR values were calculated for health conditions where the use of medicinal plants was mentioned at least ten times.

Results

Description of the study population

The total number of family households in the study region was 150,986 families with an average of three individuals per household. From a population of 433,648 individuals living in family households, 86.4% (267,704) identified themselves as the children or spouse of the head of the family and 13.3% (42,089) reported to be related as grandchildren, siblings or parents. Married couples and single women comprise 68.7% (85,084 families) and 26% (32,245 families) of the family households, respectively. Overall, 76,576 families (ca 50% on each group) reported having children less than eighteen years of age. A total of 351 families were surveyed in the study region. The women in the household were

interviewed in all families except one, where a male was interviewed. Median age was 47 years old (± 1.4 years). Approximately half of the study population interviewed (51.5%) had less than a college education, and 64.8% were married. Approximately 42% of the participants had full or part-time jobs, while 58% were either unemployed or studying. Consulting a physician (48%), self-medication with pharmaceutical products (32%) and use of botanical remedies (14%) were described by the study population as the first treatments used for the health conditions surveyed.

Description of the population using medicinal plants

In total, 118 families (33.6%) used medicinal plants as the first treatment for at least one of the health problems surveyed. The characteristics of the women using medicinal plants are summarized in Table 2. The median age of the participants who used medicinal plants was 47 years (± 15.4 years). Participants in the age group ≤ 50 years used more medicinal plants than older participants (>50 years) with 67.0% and 48.0% usage, respectively. The majority of the participants reported having a college degree and being single (65.0 and 62.0 percent, respectively). Roughly half of the study population that used medicinal plants reported being employed. Thus, women in family households that reported the use of medicinal plants in the region were mostly single, younger than 50 years of age and had a college education.

Factors associated with the use of medicinal plants

In this study, socio-demographic variables demonstrating a significant univariate association ($p < 0.05$) with

medicinal plant use were education ($p = .036$, power = 0.71) and marital status ($p = .002$, power = 0.91; Table 2). Significant relations were not found between medicinal plant use and age or employment status. Based on the OR values (OR = 1.67, 95% CI, 1.10 – 2.62), the odds of families who treated a disease with medicinal plants if the woman held a college degree was 67.0% higher than the odds for families where the women had less than a college degree. The odds ratio for families that use medicinal plants when the woman was “single” was twice the OR for families where the woman was “married” (OR 2.09, 95% CI, 1.37 – 3.31). Families where the woman was single and held a college degree were three times more likely than the rest of the study population to use medicinal plants. A stratified analysis was conducted to measure the association and to estimate the OR in each category by age and employment status. Statistical analyses showed that there is no difference in the odds ratios between the strata of the two variables (age and employment status), although employment status weakens the association between education and the use of medicinal plants (Crude OR = 1.67, Adjusted OR = 1.57). In the multivariate logistic regression analyses (results not shown), education (OR = 1.61, S.E. = 0.27) and marital status (OR = 2.22, S.E. = 0.24) remained significant predictors of medicinal plant use ($p < 0.05$).

Medicinal Plant Use

Two hundred thirteen different plant remedies were used among the study population the last time that one of their family members suffered one of the surveyed health problems. The number of ailments treated with botanical remedies varied from one to seven in a given

Table 2. Descriptive characteristics of the families (n=118) that reported the use of herbal remedies

Variables	Women in families using herbal remedies (n=118)	Percentage within herbal remedies use	χ^2 p-value	OR	95% CI	Power ^a (1 – β)
Age Groups						
<=50	67	58.6	.522	1.19	0.758-1.87	0.19
>50	48	41.4				
Marital Status ^b						
single	62	46.6	.002	2.09	1.37-3.31	0.93
married	54	53.4				
Employment Status						
employed	56	49.6	.061	1.59	1.01-2.50	0.63
unemployed	57	50.4				
Level of Education						
college degree	65	57.0	.036	1.67	1.10-2.62	0.71
< college degree	49	43.0				

a) Probability of rejecting the null hypothesis H0 (hypothesis that state no association between variables) given that in fact it is false; b) Single includes women that are single, divorced or widow; Married includes women living with a partner or married. The distinction between “married” and “living with a partner” was made by the respondent.

family, with an average of two per family. Table 3 lists the citation frequency of plant remedies, IAR-values for the health conditions surveyed, and the number of different (medicinal) plants that were reported for the treatment of a given health condition. The ailments most frequently cited affect the respiratory (36% of families used botanical remedies for asthma, rhinopharyngitis, allergic rhinitis and sinusitis) and gastrointestinal systems (28% used plants for gastritis and reflux). Plant remedies were also used for nerves (13.6%), migraine (12.2%), depression (5.2%) and arthritis (4.7%). Fifty-eight different medicinal plants were used for the preparation of botanical remedies. The average consensus values for the use of medicinal plants were obtained for rhinopharyngitis (0.58), nervousness (0.54), depression (0.50), gastritis (0.49), sinusitis (0.40) and migraine (0.40). Most of the treatments (78%) were

reported just once for a given health problem and are, therefore, not described in this TRAMIL-based report. Most of the plants were obtained at a store or market (65.3%). Other significant sources of (medicinal) plants included house gardens and the forest (23.4%).

Nine medicinal plants species belonging to five genera and five families were identified as important herbal treatments in the region. None of these species were endemic to Puerto Rico. The families best represented were Lamiaceae and Rutaceae, with two species each. Table 4 presents the common name, parts of the plant used for a given remedy, form of preparation and administration and frequency of use for each of these important species. The preparation methods included decoction of fresh or dried leaves in water, poultice or juice. Dosages were variable, with most families reporting the use of one

Table 3. Index of Agreement on Remedies (IAR) Values for Common Health Conditions

Health Condition (ICD-10 Code) ^a (Description of symptoms in spanish)	Number of times the ailment was cited (n = 118) ^b	Percent	Number of Medicinal plants cited for the ailment	IAR-value ^c
Gastritis (ICD – K29) dolor/ardor en el estómago, dolor/ardor en la boca del estómago, acidez, vómitos	42	19.7	22	0.49
Nervousness (ICD – R45) susto, ansiedad, asfixia, calores repentinos, temblor, intranquilidad, coraje	29	13.6	14	0.54
Migraine (ICD – G43) presión en la cabeza, dolor de cabeza bien fuerte, latidos en la cabeza, punzadas en los ojos, pesadez y presión en la cabeza	26	12.2	16	0.40
Asthma (ICD – J45) fatiga, presión en el pecho, asfixia, pito en el pecho	24	11.3	24	0.0
Rhinopharyngitis (ICD – J31) sangrado nasal, gotereo, secreción, ardor y congestión nasal, inflamación, picor y ardor de garganta, estornudo, flema, tos, ronquera, dolor en la cara	20	9.4	9	0.58
Gastroesophageal reflux disease (ICD – K21) calentón en el esófago, saliva agria, acidez, suben jugos gástricos, ardor que sube y baja en la garganta	18	8.5	15	0.18
Allergic Rhinitis (ICD – J30) Gotereo nasal, picor en ojos y nariz	17	8.0	14	0.19
Chronic Sinusitis (ICD – J32) dolor de cabeza, hinchazón en cavidad nasal, calambres, corriente en la nariz	16	7.5	10	0.40
Depression (ICD – F32) ansiedad, nerviosismo, asfixia, dificultad respiratoria, ganas de irse del mundo, hablar	11	5.2	6	0.50
Arthritis (ICD – M05) dolor en coyunturas, huesos, articulaciones, inflamación coyunturas, hinchazón	10	4.7	10	0.0

a) International Classification of Diseases (ICD-10) classifies diseases and other health problems recorded on health and vital records including hospital records. Source <http://www.who.int/classifications/apps/icd/icd10online/>

b) Out of 351 interviews, 118 mentioned the use of medicinal plants as the first treatment.

c) IAR values range from 0 to 1, with 0 representing no consensus, 0.5 average consensus and 1 total consensus.

Table 4. Plants with medicinal use in the Southeast Region of Puerto Rico

Scientific Name (voucher specimen)	Local name	Parts used/ Preparation	Administration	Diseases treated	Frequency ^a (%)
Annonaceae <i>Ammona muricata</i> L. (JGS024)	guanábano	Fresh leaves/ Decoction	Oral	Gastritis	14
Asteraceae <i>Pluchea carolinensis</i> (Jacq.) G. Don in Sweet (JGS025)	salvia	Fresh leaves/ Cataplasm	Topical	Migraine	27
Laminaceae <i>Mentha piperita</i> L. (JGS040)	menta	Fresh leaves/ Decoction	Oral	Sinusitis	25
Laminaceae <i>Mentha nemorosa</i> Willd. Ex L. (JGS011)	yerbabuena	Fresh leaves/ Decoction	Oral	Gastritis	14
Rutaceae <i>Citrus aurantium</i> L. (JGS027)	naranja	Fresh leaves/ Decoction Fresh or dry leaves/ Decoction	Oral Oral	Depression Nervousness	33 20
Rutaceae <i>Citrus aurantifolia</i> (Christm.) Swingle (JGS019)	limón	Fresh pulp/Juice	Oral	Rhinopharyngitis	40
Verbenaceae <i>Lippia stoechadifolia</i> (L.) Kunth (JGS017)	poleo	Fresh leaves/ Decoction	Oral	Gastritis	10

^aPercentage of the families using this plant for this health problem

cup of tea (decoction) from one to three times a day over several days. Plants were used without restrictions, even for children, except for the use of the fresh juice of *Citrus aurantifolia* (Christm.) Swingle, where the dosage was reported to be half of that used by adults to treat the symptoms of rhinopharyngitis.

Discussion

Relative importance of the species with significant use

TRAMIL defines a remedy with significant use as the combination of plant species, plant part and form of preparation that is identified by 20% or more of the study population as a primary treatment resource for a given disease the last time it affected a family member. Based on the TRAMIL methodology, only six medicinal plant species (9.0% of the total medicinal plants recorded) qualified as having significant use for treating the symptoms of depression, nervousness, rhinofaryngitis, migraine and sinusitis (Table 4). The plants, in decreasing order of use, were *Citrus aurantifolia* (Christm.) Swingle (40.0%), *Citrus aurantium* L. (33.0%), *Pluchea carolinensis* (Jacq.) (30.0%) and *Mentha piperita* L. (25.0%). The large number of responses obtained for use

of these species suggests a high degree of popular belief in their therapeutic properties (25). *Citrus aurantifolia* is used for rhinopharyngitis, *C. aurantium* leaves have significant use for depression and nervousness, *M. piperita* is used for sinusitis, and *P. carolinensis* is used for migraine. The use of *C. aurantifolia* juice for cold, flu and cough has been reported in the TRAMIL surveys of Colombia, Honduras, Panamá, Guyana, Dominica and Puerto Rico (1). TRAMIL reports that the essential oil obtained from the leaf, flower, fruit and bark of *C. aurantifolia* is rich in the monoterpene derivatives limonene and linalool, and that the fresh juice of *C. aurantifolia* exhibits antimicrobial activity against *Staphylococcus aureus* and *Candida albicans* and stimulates gastric secretion. Naturally occurring (-)-Linalool has been reported to reduce carrageenin-induced edema in rats at 25 mg/Kg, suggesting its potential anti-inflammatory activity (26). These biological activities have been associated with the antiseptic, antitussive and expectorant action of essential oil-rich herbs and their efficacy in clinical phytotherapy to treat respiratory ailments (27). *Citrus aurantium* has been described as having a sedative effect on the central nervous system (CNS), and as a result, it has been used to treat anxiety, hysteria, and cases of depression. The

effects of this plant also include hypnotic, anticonvulsant and hypothermic properties (28-29). A sedative activity (as measured by sleeping time induced by sodium pentobarbital; SPB: 40 mg/Kg i.p.) has also been reported for the extracts and essential oil from *Citrus aurantium* (28). The hexane and dichloromethane fractions from the hydroethanolic extract of the leaves and the essential oil from the peel have both been shown to enhance the sleeping time induced by barbiturics at a dose of 1.0 g/Kg. In this study, the route of administration (i.p.) and the form of preparation were different from the traditional oral route in humans. TRAMIL applied research addresses the need to validate the biological activity according to the traditional use of those (medicinal) plants with significant use in order to determine if the plant's use in primary medical care is warranted. Therefore, the extent of the effects reported cannot be extrapolated to validate the traditional use since the pharmacokinetics of the active components and the therapeutic doses may be affected by the preparation and administration (29). The sedative effects of flavonoids, quercetin, chrysin and apigenin and the flavonoid glycoside isoquercitrin in *Citrus aurantium* have also been reported (29). The chemical composition of the leaf oils of *Pluchea carolinensis* was reported by Pino, et al. (30), which identified juniper camphor as the major component. No antinociceptive effect of the leaves of this plant has been reported, although this effect was reported for the essential oil *Hyptis fruticosa*, which also contains juniper camphor (31). Other authors have suggested that flavonoids account for the anti-inflammatory activity in tinctures of *Pluchea carolinensis* (32). Ethnobotanical accounts in Puerto Rico have previously documented *C. aurantifolia* and *C. aurantium* as treatments for hoarseness and restlessness, which are the key characteristics of rhinopharyngitis and nervousness, respectively, and *P. symphytifolia* (Mill.) Gillis for migraine (7, 33-34). Hernández, et al. (35) reported that *C. aurantium* is the plant most frequently used as a sedative and for gastrointestinal disorders among the patients visiting outpatient clinics on the island. One of the most frequently found plants in botánicas or herbal shops in southern Florida is *P. carolinensis*, which is employed to alleviate headaches (36). Contrary to mainstream beliefs that botanical remedies are often harmful or toxic, the medicinal plants with higher frequencies of use in this report are common, edible food plants. These plants are designated by the US Food and Drug Administration (FDA) as "generally recognized as safe" (GRAS) (37). The significant levels of use found in this study region for the decoction of the fresh or dry leaves of *Citrus aurantium* L. to treat depression and nervousness, *Mentha piperita* to treat sinusitis, and the topical use of *Pluchea*

carolinensis to treat migraine expand on the uses for these plants reported by TRAMIL.

In this study region, a second group of plants with less than 20% frequency of use, including *Mentha nemorosa* Willd. Ex L., *Lippia alba* (Miller) N.E. Brown and *Annona muricata* L., were reported for the treatment of gastritis, the ailment where the use of medicinal plants was most cited (42 citations; IAR 0.49). This group of plants, specifically the decoctions of *A. muricata* leaves, are reported in the ethnobotanical literature as the most recommended for disorders of the gastrointestinal system including gastric ulcers, diarrhea, vomiting and indigestion. However, this study did not find significant use of these species as defined by TRAMIL. This stringency of definition reflects TRAMIL's strategy, which is designed to select medicinal plant uses that will be validated and promoted for use in community primary health care practices. Nevertheless, the decoction of leaves of *Mentha spp.* (yerbabuena) to treat some of the symptoms described for gastritis has been documented in Venezuela and Martinique and validated by TRAMIL (1). The effect of the infusion of *L. alba* on the rat gastric mucosa has been evaluated and compared to ranitidine (100 mg/Kg, p.o.) as the reference antiulcer drug (38). Oral treatment with the infusion (12.5 g dry plant/Kg) did not cause gastric irritancy in rats treated over five consecutive days. In addition, the oral administration of *L. alba* was found to be effective at preventing gastric ulceration induced by indomethacin (50 mg/Kg, p.o.) in rats in both short and long term studies. The chemical composition, pharmacological activity and traditional uses of 52 species attributed to the genus *Lippia* shows that these species are predominantly used to treat gastrointestinal and respiratory disorders. The essential oil or the flavonoids from the plant extracts have been suggested to be responsible for the reported activities (39).

Trends in medicinal plant use

Overall, about 14% of the families in the southeastern region of Puerto Rico used at least one herbal remedy on the last occasion that one of the ten surveyed ailments was suffered. Unlike other reports (18, 40), this survey did not find a significant relationship between age and medicinal plant use, suggesting a consensus among the population in the study region about the herbal species used to treat the ailments surveyed. This data shows that knowledge of the traditional use of medicinal plants to treat self-limiting ailments is lower at the community level than knowledge of conventional medical treatments. The process of medicalization and integration of exogenous technologies into the Puerto Rico health care system, which started during the 1950s as a result of the island's economic

expansion and industrialization, has been described as a significant social factor leading to the abandonment of traditional practices in the health system (41). Some aspects of this report are consistent with data from the National Health Interview Survey (NHIS), which reported the use of herbal medicine (9.6%; 42) or natural products among US adults, including herbal medicine and functional food supplements (18.9%; 20). These reports also describe the use of complementary and alternative medicine (CAM) as most prevalent among women, persons with education of ≥ 16 years and persons aged 35 to 54. One limitation of our study is that participants were not asked about their reasons for using a specific botanical remedy, whether they used it concomitantly with prescription medications, or their degree of satisfaction with conventional medical treatments. Despite these limitations, the participants reported, when interviewed, a desire to receive accurate information on herbal medicines, which reflects a wish to find alternative treatments that fit their values and beliefs (43) and to learn about traditional knowledge on medicinal plant use. These findings suggest that future studies are warranted to develop and test TRADIF materials to understand the socioeconomic differences in medicinal plant use and to educate the communities in the study region about evidence-based herbal use. The survey will also be expanded to include Vieques (one of the islands of the archipelago of Puerto Rico) and the US Virgin Islands.

Resumen

Transfondo: La Red TRAMIL tiene como objetivos entender, validar y ampliar las prácticas sanitarias basadas en el uso de plantas medicinales en el Caribe, centro (“hotspot”) de alta biodiversidad y amenaza en el planeta debido al alto grado de endemismo de especies, la intensa presión de desarrollo y la pérdida de hábitats. Este estudio documenta las plantas medicinales que se utilizan para tratar las condiciones de salud de mayor prevalencia en la región sudeste del archipiélago de Puerto Rico y las tendencias en el uso de plantas medicinales entre la población estudiada. Método: Se realizó una encuesta etnofarmacológica en la región de estudio y se completó el análisis univariado y multivariado de variables asociadas al uso de plantas medicinales. Resultados: Se documentaron 118 plantas medicinales que se utilizan para tratar la depresión, el nerviosismo, la sinusitis crónica, la gastritis, el reflujo gastroesofágico, la rinitis alérgica, la rinofaringitis, el asma, la artritis y la migraña. Las especies de plantas con uso significativo en la región son *Citrus aurantium* L., *Citrus aurantifolia* (Christm.) Swingle, *Pluchea carolinensis* (Jacq.) G. Don Sweet y *Menta*

piperita L. El uso de plantas medicinales es más frecuente entre las mujeres solteras con un alto nivel de escolaridad, una tendencia similar a la utilización de la MAC en los EE.UU. Conclusión: El conocimiento etnofarmacológico y el uso de plantas medicinales está disminuyendo en la región de estudio debido a un aumento en el uso de la atención médica convencional y la auto-medicación con productos farmacéuticos. Se identificaron cuatro especies botánicas de nuevo uso no identificados previamente en la Farmacopea Vegetal del Caribe. Los resultados de la encuesta informan a la Red sobre la validación científica y los estudios de toxicidad de estas especies y las actividades TRADIF futuras en la región de estudio.

Acknowledgments

The authors wish to thank all the families from the eleven municipalities served by the University of Puerto Rico at Cayey who participated in this survey. Special thanks to the UPR-Cayey students who conducted the interviews: Carlos Marzant, José I. López, Melissa Guzmán, Melissa Olivieri, Mercedes López, Orly Santos, Rosángela Rosario, Yahaira Rosario and Yasmín Pérez; and to the students who completed the data entry: Dalixis Rivera and María del C. Rodríguez. Thanks to Mr. José Sustache, botanist and Director of the Puerto Rico Department of Natural and Environmental Resources, who helped in the identification of the plants collected and provided the herbarium facilities to maintain the vouchers, to Mrs. Nora Alvarez, Director of the Puerto Rico Department of Natural and Environmental Resources Geoinformatics Division for creating the natural vegetation and land cover distribution maps of the study region, to Dr. Enrique López for his comments about the use of Maptitude applications during the design of the sampling methodology and to Jolene Yurkes for reviewing this manuscript. This work was conducted with partial support from the UPR-Cayey Institutional Research Fund and the National Institutes of Health RIMI Program #1-P2MD001112-0. The authors have no conflict of interest to disclose.

References

1. Germosén-Robineau L, Delens M, García-González M, Herrera J, Morón F, Sáenz-Campos D, Solís P. editors. Farmacopea Vegetal Caribeña, 2da Ed; León, Nic; Editorial Universitaria, UNAN-León, 2005: p. 485.
2. Farnsworth NR, Akerele O, Bingle ES, Soejarto DD, Guo Z. Medicinal plant in therapy. Bulletin of the World Health Organization 1985;63:965-981.
3. DaSilva EJ, Hoareau L. Medicinal plants: a re-emerging health aid. Electron J Biotechnol 1999;2:56-70.
4. Longuefosse J-L, Nossin E. Medical ethnobotany survey in Martinique. J Ethnopharmacol 1996;53:117-142.

5. Katewa SS, Chaudhry BL, Jain A. Folk herbal medicines from tribal area of Rajasthan, India. *J Ethnopharmacol* 2004;92:41-46.
6. Helmer EH, Ramos O, López T del M, Díaz W. Mapping the Forest Type and Land Cover of Puerto Rico, a Component of the Caribbean Biodiversity Hotspot. *Caribb J Sci* 2002;38:165-183.
7. Nuñez E. Plantas Medicinales de Puerto Rico. Puerto Rico; Editorial de la Universidad de Puerto Rico, 1989: p. 1-444.
8. Martínez TT, Martínez RR. Medicinal Herbs from the Caribbean National Forest (El Yunque), Puerto Rico. *Proc West Pharmacol Soc* 2002;45:20-22.
9. US Census Bureau, Puerto Rico 2000. Resumen de características Sociales, Económicas y de Vivienda: In 1980, 56.2% of the study region population lived in rural areas. This number decreased to only 7.3% in 2000. Urban area is define as a densely populated territory with 2,500 to 50,000 habitants or more. USA Department of Commerce, 2001: p. 13.
10. Ramos-Valencia G. Estudio Continuo de Salud para los Municipios de Puerto Rico, Public Health School, RCM-UPR; 2003.
11. US Census Bureau, Puerto Rico 2000. Resumen de características Sociales, Económicas y de Vivienda: Family's Households: Habitual residence of two or more persons who are related through blood (birth), marriage or adoption. USA Department of Commerce, 2001: p. 17-18.
12. US Census Bureau, Puerto Rico 2000. USA Department of Commerce, 2002: p. 127-145.
13. Aday LA, Cornelius L. Designing and Conducting Health Surveys: A Comprehensive Guide, 3rd Ed; Jossye-Bass, 2006: p. 154-194.
14. US Census Bureau, Puerto Rico 2000. Resumen de características Sociales, Económicas y de Vivienda: Block Group is define as an area with a population between 600 and 3,000 inhabitants, with an optimum population of 1,500 inhabitants. USA Department of Commerce, 2001: p. 13-14.
15. World Health Organization (WHO). International Statistical Classification of Diseases and Related Health Problems (ICD-10), 2007. Accessed 04/28/08. Available from: URL: <http://www.who.int/classifications/apps/icd/icd10online/>.
16. Arango-Caro S. Ethnobotanical studies in the Central Andes (Colombia): Knowledge distribution of plant use according to informants' characteristics. *Lyonia* 2004;7:89-104.
17. Singhal R. Medicinal plants for primary health care. *J Health Manag* 2005;7:277-293.
18. Quinlan MB, Quinlan RJ. Modernization and Medicinal Plant Knowledge in a Caribbean Horticultural Village. *Med Anthropol Q* 2007;21:169-192.
19. Kuo GM, Hawley ST, Weiss LT, Balkrishnan B, Volk RJ. Factors associated with herbal use among urban multiethnic primary care patients: a cross-sectional survey. *BMC Compl Alternative Med* 2004;4:18-26.
20. Nahin RL, Dahlhamer JM, Taylor BL, Barnes, PM, Stussman BJ, Simile CM, Blackman MR, Chesney MA, Jackson M, Miller H, McFann KK. Health behaviors and risk factors in those who use complementary and alternative medicine. *BMC Public Health* 2007;7:217-225.
21. Szklo M, Nieto JF. Identifying Noncausal Association: Confounding. In *Epidemiology: Beyond the Basics*, 1st Ed; Sudbury, MA; Jones & Bartlett; 2003: p. 190-197.
22. Pagano M, Gauvreau K. Principles of Biostatistics, 2nd Ed, Duxbury, 2000: p. 196-213, 470-487.
23. Faul F, Erdfelder E, Lang A, Buchner A. G*Power 3: A flexible statistical power analysis program for the social, behavioral, and biomedical sciences. *Behav Res Methods* 2007;39:175-191.
24. Vandebroek I, Balick MJ, Yukes J, Durán L, Kronenberg F, Wade C, Ososki A, Cushman L, Lantigua R, Mejía M, Robineau L. Use of Medicinal Plants by Dominican Immigrants in New York City for Treatment of Common Health Problems – A Comparative Analysis with Literature Data from the Dominican Republic. In: *Traveling Cultures and Plants. The Ethnobiology and Ethnopharmacy of Human Migrations*; Volume 7. Studies in Environmental Anthropology and Ethnobiology; Oxford, UK; Berghahn Books, 2007: p. 39-63.
25. Carrillo-Rosario T, Moreno G. Importancia de las plantas medicinales en el autocuidado de la salud en tres caseríos de Santa Ana Trujillo, Venezuela. *Revista de la Facultad de Farmacia* 2006; 48:21-28.
26. Peana AT, D'Aquila PS, Panin F, Serra G, Pippia P, Moretti MDL. Anti-inflammatory activity of linalool and linalyl acetate constituents of essential oils. *Phytomedicine* 2002;8:721-726.
27. Delens, M. ed. Cuaderno de Fitoterapia Clínica (Afecciones Respiratorias y Digestivas), 1era Ed; Mérida, Venezuela; Comisión Nacional para el Aprovechamiento de Plantas Medicinales (CON-APLAMED); 2000: p. 11-86.
28. Costa M, Carvalho-Freitas MIR. Anxiolytic and sedative effects of extracts and essential oils of *Citrus aurantium* L. *Biol Pharm Bull* 2002;25:1629-1633.
29. Emamghoreishi M, Heidari-Hamedani G. Sedative-hypnotic activity of extracts and essential oil of coriander seeds. *Iran J Med Sci* 2006;31:22-27.
30. Pino JA, Marbot R, Payo A, Chao D, Herrera P, Martí MP. Leaf Oils of Two Cuban Asteraceae Species: *Pluchea carolinensis* Jacq. and *Ambrosia hispida* Pursh. *J Essent Oil Res* 2005;17:318-320.
31. Menezes IAC, Marques MS, Santos TC, Dias KS, Silva ABL, Mello ICM, Lisboa ACCD, Alves PB, Cavalcanti SCH, Marçal RM, Antonioli AR. Antinociceptive effect and acute toxicity of the essential oil of *Hyptis fruticosa* in mice. *Fitoterapia* 2007; 78:192-195.
32. Rosales-Clares VP, Gross Fernández MC, Rosales Clares RA, García Díaz RC, León Sarabia JE, Vidal M. Evaluación farmacológica de *Pluchea carolinensis* Jacq. (salvia de playa) en animales de experimentación. *Rev Cubana Plant Med* 1999;2:65-67.
33. Benedetti MD. ¿Hasta los baños te curan! Plantas Medicinales, remedios caseros y sanación espiritual en Puerto Rico; 3era Ed; Puerto Rico; Verde Luz, 2001: p. 208-209, 211.
34. Benedetti, MD. Sembrando y sanando en Puerto Rico; 1era Ed; Puerto Rico, Verde Luz, 1996: p. 258-259, 266-268.
35. Hernández L, Muñoz RA, Miró G, Martínez M, Silva-Parra J, Chávez PI. Use of medicinal plants by ambulatory patients in Puerto Rico. *Am J Hosp Pharm* 1984;41:2060-2064.
36. Hodges S, Bennett BC. The ethnobotany of *Pluchea carolinensis* (Jacq.) G. Don (Asteraceae) in the botánicas of Miami, Florida. *Economic Botany* 2006;60:75-84.
37. FDA Electronic Code of Federal Regulations (eCFR) GRAS status information Title 21 Parts 182 and 184; 1988.
38. Pascual ME, Slowing K, Carretero ME, Villar A. Antitumorogenic activity of *Lippia alba* (Mill.) N. E. Brown (Verbenaceae). *Farmacology* 2001;56:501-504.
39. Pascual ME, Slowing K, Carretero ME, Sánchez Mata D, Villar A. *Lippia*: traditional uses, chemistry and pharmacology: a review. *J Ethnopharm* 2001;76:201-214.
40. Nolan JM. The Roots of Tradition: Social Ecology, Cultural Geography, and Medicinal Plants Knowledge in the Ozark – Ouachita Highlands. *J Ethnobiol* 1998;18:249-269.
41. Parrilla-Rodríguez AM, Gorrín-Peralta JJ. La Lactancia Materna en Puerto Rico: Patronos Tradicionales, Tendencias Nacionales y Estrategias para el Futuro. *PR Health Sci J* 1999;18:223-228.
42. Ni H, Simile C, Hardy AM. Utilization of Complementary and Alternative Medicine by United States Adults. *Med Care* 2002; 40:353-358.
43. Haller CA. We should always ask our patients about unconventional treatments. *West J Medicine* 2001;175:164.