PROXIMATE COMPOSITION AND BIOLOGICAL ACTIVITY OF FOOD PLANTS GATHERED BY CHILEAN AMERINDIANS[1]

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Guillermo Schmeda-Hirschmann, Ivan Razmilic, Margarita I. Gutierrez, *and* **Jose I. Loyola** *(Instituto de Química de Recursos Naturales, Universidad de Talca, Casilla 747, Talca, Chile).* PROXIMATE COMPOSITION AND BIOLOGICAL ACTIVITY OF FOOD PLANTS GATHERED BY CHILEAN AMERINDIANS. *Economic Botany 53(2):177–187, 1999. The proximate composition and biological activity of food plants and mushrooms gathered by Chilean Amerindians were assessed. The gathered plants served primarily as sources of carbohydrates with highest values for* Dioscorea *tubers,* Prosopis alba *pods meal and* Bromus catharticus *seeds. The mushrooms* Clavaria coralloides *and* Boletus loyus *proved to be the best protein sources in our survey, but deficient in the amino acids methionine and cysteine. Some extracts of the plants and mushrooms under study showed biological activity as free radical scavengers, enzyme inhibitors, hypotensive or DNA binding effect. Free radical scavenging activity was detected in* Cryptocarya alba *fruit extract, while* Typha angustifolia *showed a strong DNA binding effect at 0.50 mg/ml. Methanolic extracts of the Apiaceae* species Sanicula graveolens *and* Apium australe *were moderately active as ß-glucuronidase inhibitors at 50 µg/ml.*

Composición proximal y actividad biológica de plantas alimenticias recolectadas por Amerindios chilenos. *Se determinó la composición proximal y la actividad biológica de plantas y hongos alimenticios recolectados por amerindios de Chile. Las plantas colectadas fueron en conjunto una fuente de carbohidratos, con valores mayores en los tubérculos de* Dioscorea, *la harina de frutos de* Prosopis alba *y los frutos de* Bromus catharticus. *Los hongos* Clavaria coralloides *y* Boletus loyus *resultaron ser las mejores fuentes proteicas de este estudio, siendo ambos, sin embargo, deficitarios en los aminoácidos esenciales metionina y cisteína. Algunos extractos de las plantas y hongos en estudio mostraron actividad biológica como atrapadores de radicales libres, inhibidores enzimáticos, efecto hipotensor o unión al ADN. Se detectó actividad atrapadora de radicales libres en el extracto de frutos de* Cryptocarya alba, *así como un fuerte efecto de unión al ADN del extracto de* Typha angustifolia *a 0.50 mg/ml. Extractos metanólicos de las Apiaceae* Sanicula graveolens *y* Apium australe *fueron moderadamente activos como inhibidores de la enzima ß-glucuronidasa a 50 µg/ml.*

Key Words: Native food plants; mushrooms; proximate composition; biological activity; *Bromus catharticus*; *Dioscorea* spp.; *Prosopis* spp.; *Clavaria coralloides.*

FOOD USE AND BIOLOGICAL ACTIVITY OF GATHERED PLANTS

Partial information about the use of natural resources as food and medicine among Amerindians can be traced through descriptions by early chroniclers, ethnographic literature, and by archeological findings. In recent times, a human settlement dated 13 000 years BP and belonging to the late Pleistocene was discovered in southern Chile (Dillehay et al. 1982). This site, Monte Verde, contained identifiable remains of at least 68 plant species, the relative abundance and characteristics of which suggest their use as food and medicine by the paleoindians (Ramírez 1989: 147–170). Considering that most of the plants were gathered as a food source, the diet of paleohunthers and gatherers should have consisted of a high number of plants. Pollen analysis of the archeological site of Cuchipuy as well as at the Tagua-Tagua lagoon revealed the occurrence of 12 edible and 9 potentially edible taxa (Rojas 1991). A significant number of the identifiable species at Monte Verde, Tagua-Tagua, and Chuchipuy have had, or still have a con-

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temporary use as food or medicine among the Mapuche (Mösbach 1991:28, 29, 52, 79, 80, 84; Muñoz, Barrera and Meza 1981:5, 6, 12, 13, 47).

At Monte Verde, fruits and seeds of the Cyperaceae *Scirpus californicus* (C. A. Mey.) Steud. were abundant. The rhizomes of *S. californicus* too were abundant and are edible. Several Amerindian cultures depended on *S. californicus,* particularly in the Andean Altiplano (Heiser 1978). The plant is known by the Mapuche as tahua-tahua or taguatagua (Mösbach 1991:65). Achenes of *Madia sativa* L. were present in large quantities in the settlement. The plant was cultivated by the Mapuche Amerindians to obtain an edible oil (Schmeda-Hirschmann 1995).

The fruiting body of *Clavaria coralloides* L. was also identified at Monte Verde. The mushroom can be found during late winter and sometimes early spring. The fruiting bodies of loyo, *Boletus loyus* Espinosa (Agaricaceae), changle, *Clavaria coralloides* L. (Clavariaceae) and gargal, *Grifola gargal* Singer are still widely consumed in rural communities of Central and Southern Chile, where they are collected and sold in the markets (Muñoz, Barrera and Meza 1981:5, 6; Mösbach 1991:28, 29, 52).

The ripe fruits of *Cryptocarya alba* (Mol.) Looser (Lauraceae) were eaten by the Mapuche either boiled in water or raw. The leaves of *Apium australe* Thouars (Apiaceae) are edible (Muñoz, Barrera and Meza, 1981:47, 70; Mösbach 1991:79, 80, 99). The young leaves and sprouts of *Typha angustifolia* L. (Typhaceae) were also consumed. The plant is kwnon as batru or batu by the Mapuche (Mösbach 1991:60–61).

Early chroniclers refer to mangu (*Bromus mangu* Desv., Poaceae) as a native cereal cultivated by the Mapuche and now extinct. *Bromus catharticus* (Mol.) Vahl is a grass known as lanco whose leaves are used as an expectorant and purgative (Mösbach 1991:63). The grains can easily be collected and were included in the present study because of their potential use as a cereal.

The tubers of several Dioscoreaceae species have long been used as a food source and are still occasionally collected as food (Muñoz, Barrera and Meza 1981:12–13; Mösbach 1991:69). Plants of the Dioscoreaceae flourish in central Chile during early spring and the aerial parts die during the summer.

Sanicula graveolens Poepp. ex DC. (Apiaceae) is known as wild coriander and is used as a spice (Muñoz, Barrera and Meza 1981:73). The herb is common in the Andean slopes and has an intense aroma closely resembling that of coriander (*Coriandrum sativum* L.)

Prosopis alba Griseb. and *P. tamarugo* Phil. (Mimosaceae) are typical trees of the northern desert of Chile. The pods are used as a feedstock for sheep, goats and cattle (Rodriguez, Matthei, and Quezada 1983:291–292) and were eaten by the Amerindians who settled in the northern part of the country (Briones 1985:49–51; Mösbach 1991:84; Muñoz, Barrera, and Meza 1981:49).

Food Plants and Biological Activity

The relationship between diet and pharmacologically active compounds is frequently overlooked (Etkin and Ross 1991). The exposure to nutritional and non-nutritional plant constituents affects the health of the consumers. Food plants may contain secondary metabolites with a direct effect on cardiovascular system. Ness and Powles (1997) reviewed the relationship between fruit and vegetables intake and cardiovascular diseases. The results are consistent with a strong protective effect of fruit and vegetables for stroke and a weaker protective effect on coronary heart disease. Murkies, Wilcox, and Davis (1998) concluded that phytoestrogens present in food plants exhibit physiological effects in humans including benefits in cases of hypercholesterolaemia. Data obtained in epidemiological, animal, and in vitro studies suggest further research of the role of phytoestrogens in cancer prevention.

The life span of hypertensive rats increased by food intake of garlic and linseed oil (Brandle et al. 1997). Chiu and Fung (1997) examined the effect of mung beans (*Vigna aureus* [=*V. radiata* (L) Wilcz]), common rue (*Ruta graveolens* L), and kelp (*Laminaria japonica* Areach) in rats and showed that these plants contained cardiovascular active substances that had a direct effect on the cardiovascular system and that these substances further interacted to modify their cardiovascular effects.

Among various physiological disorders, excessive free radical production and lipid peroxidation are known to cause several pathological conditions including atherosclerosis, ageing, rheumatic diseases, cardiac and cerebral ischemia (Wong et al. 1987).

In our studies (see Materials and Methods) in vitro and in vivo assays were used to obtain information on the biological activity of secondary

metabolites present in the extracts. Free radical scavenging activity and antioxidant activity of our samples were assessed by the xanthine oxidase inhibition assay as well as by the decoloration of a methanolic solution of the diphenyl picryl hydrazyl radical (DPPH) according to Joyeux et al. (1995). The enzyme xanthine oxidase (XO) catalyses the oxidation of hypoxanthine to xanthine and finally uric acid, the excess of which is considered to produce the symptomatology of gout in humans (Cao, Sofic, and Prior 1997; Cos et al. 1998). Several natural products present in plants acts as xanthine oxidase inhibitors and some of them have been reported to have cancer chemopreventive activity (Ohnishi et al. 1996; Murakami, Ohigashi, and Koshimizu 1996).

The enzyme β-glucuronidase (β-gluc) is a glycosidase that hydrolyses β-D-glucuronides to an alcohol and D-glucuronate. It has been reported that hepatotoxic compounds produce an important increase of β-gluc activity and that endogenous biliary β-gluc might be responsible for bilirubin deconjugation leading to gallstones (Ho et al. 1986). The action mechanism of many antitumoral drugs involves interaction of small molecular weight ligands with DNA and can be assessed by RP-HPLC (Pezzutto et al. 1991). We determined the effect of our extracts on the enzyme β-glucuronidase and their DNA binding. The activity of fungal and *Typha angustifolia* extracts on the blood pressure of rats was assessed. The doses used were selected according to previous work on bioactive constituents in medicinal and food plants (Pezzutto et al. 1991; Schmeda-Hirschmann et al. 1992; Schmeda-Hirschmann et al. 1994).

Despite the existance of many chemical studies on Chilean medicinal plants, very little has been done on the chemical composition and biological activity of plants gathered as food by Chilean Indians. Continuing our ethnopharmacological studies on South American economic plants, we report here the proximate composition and biological activity of some plants and mushrooms gathered by Chilean Amerindians.

Materials and Methods

Plant Material

Most of the material studied was collected in Region VII of Chile. Voucher herbarium numbers correspond either to José I. Loyola (JIL) or José San Martín (SM) collections and have been deposited at the Herbario de la Universidad de Talca and were identified by José San Martín.

Apium australe Thouars (Apiaceae) (JIL 0154), Rio Lircay, Altos de Vilches, (35°54′S; 71°59′W, 200 m.o.s.l.) 12. 1995. *Bromus catharticus* (Mol.) Vahl (Poaceae) (JIL 0150), University Campus, Talca, 11.1995. *Dioscorea bridgesii* Griseb. ex Kunth (Dioscoreaceae) (JIL 0145), Cuesta La Chepica, Pencahue, 16.11.1994 and 25.10.1995. *Dioscorea humifusa* Poepp. var. *humifusa*: Cuesta La Chépica, Pencahue (35°24′S; 71°48′W), VII Región, 16.11.1994. (SM 1503). *Dioscorea humifusa* Poepp. var. *gracilis* (H. et A.) Navas. San Rafael (35°18′S; 71°31′W, 145 m.o.s.l.), VII Región, 20.10.1994. (SM 1504).

Cryptocarya alba (Mol.) Looser (Lauraceae) (JIL 185): Plaza de Armas, Talca city (35°26′S; 71°39′W, 90 m.o.s.l.), 30.04. 1996. *Prosopis alba* Griseb. (Mimosacae) (JIL 179) and *Prosopis alba* Griseb. var. *panta* Griseb. (JIL 187), Pampa del Tamarugal (20°24′S; 69°44′W), 01.1997. *Prosopis tamarugo* Phil. (Mimosaceae) (JIL 0153), Pampa del Tamarugal, I Región, 10.1995. *Sanicula graveolens* Poepp. ex DC (Apiaceae)(JIL 137, 175–177), slopes of Volcán Chillán (36°54′S; 71°29′W, 1240 m.o.s.l.), VIII Región, 14.11.1996.

Scirpus californicus (C.A. Mey.) Steud. (Cyperaceae) (JIL 029), Llico (34°46′S, 72°05′W, 15 m.o.s.l.), Provincia Curicó, VII Región, 03.03.1994. *Typha angustifolia* L. (Typhaceae)(JIL 0139), University Campus, VII Región (Chile), 23.02.1995.

Boletus loyus Espinosa (Basidiomycetes) (JIL H-1), *Clavaria coralloides* L. (Clavariaceae) (JIL H-2) and *Grifola gargal* Singen (Polyporaceae) (JIL H-3) were collected at Cordillera Pelada, Monumento Nacional Alerce Costero, CONAF, Provincia Valdivia, X Región, 40°12′S, 73°26′W, 14–16.04, 1996.

The plant parts analyzed were: tubers (*Dioscorea* spp.), seeds (*Bromus catharticus*), fruits (*Cryptocarya alba*), pods (*Prosopis* spp.), rhizome (*Scirpus californicus*; *Thypha angustifolia*) and young leaves (*Thypha angustifolia*). For the fungal samples, the fruiting bodies were studied.

Proximate Analysis

Crude lipid content of the samples was estimated by exhaustive Soxhlet extraction of a known weight of dried sample with petroleum

TABLE 1. PROXIMATE ANALYSIS OF EDIBLE PLANTS AND FUNGI IN G/100 G DRY WEIGHT.

Sample	Moisture	CP[1]	CL[2]	Crude Fiber	Ash	NNE[3]	P[4]
Bromus catharticus							
Seeds	8.5	14.00	1.60	3.40	2.10	78.90	337
Cryptocarya alba							
Fruit, peel	5.6	6.88	32.30	20.40	1.80	38.62	124
Fruit, pulp and seed	5.3	6.25	16.15	5.60	2.30	69.70	130
Dioscorea humifusa var. *humifusa*							
Tubers	9.9	3.63	0.89	7.30	7.23	80.95	124
Dioscorea humifusa var. *gracilis*							
Tubers whole	9.6	5.32	0.82	12.40	4.10	77.36	154
Tubers peeled	9.5	5.61	0.54	3.60	2.35	87.90	150
Dioscorea bridgesii							
Tubers	8.5	6.00	0.63	16.00	6.30	71.07	146
Prosopis alba	7.1	6.90	0.29	6.60	3.10	83.11	129
Prosopis alba var. *panta*	8.1	7.90	0.50	7.20	3.20	81.20	109
Prosopis tamarugo							
Pods 1996	7.6	8.40	0.72	31.40	2.60	56.88	125
Prosopis tamarugo							
Pods 1997	7.3	12.50	0.84	24.00	4.30	58.36	214
Scirpus californicus							
Rhizome	14.1	8.93	1.80	18.90	10.60	69.87	347
Typha angustifolia							
Young leaves	11.4	7.00	1.50	22.50	9.00	60.00	300
Rhizome	10.0	5.81	0.90	17.20	8.80	67.29	158
Fungi							
Boletaceae							
Boletus loyus							
Fruiting bodies	12.0	22.00	1.05	21.00	6.10	49.9	280
Clavariaceae							
Clavaria coralloides							
Fruiting bodies	11.2	26.00	2.30	12.70	6.50	52.5	400
Grifola gargal							
Fruiting bodies	10.0	5.00	2.50	10.00	1.10	90.4	28

[1] CP: crude protein.
[2] CL: crude lipids.
[3] NNE: non-nitrogentated compounds (carbohydrates).
[4] P: phosphate (mg %).

ether (bp 40–60°C). The defatted residue was analyzed for protein, fiber, ash and non-nitrogenated compounds (carbohydrates). Crude protein and fiber content were determined by standard Kjeldahl conversion and acid detergent fiber techniques (Goering and Van Soest). The carbohydrate content (excluding fiber) was obtained by substracting the sum of protein, ash, fiber and crude lipids from the total dry matter (Heldrich, 1990,: 32, 80, 330, 781; Schmidt-Hebbel, 1981,: 24, 30–31, 39–41). Phosphate levels were determined colorimetrically by the ammonium molybdate complex method (Heldrich 1990:56). Results are summarized in Table 1.

The fatty acid composition of the oil obtained from the fruits of *Cryptocarya alba* and the

TABLE 2. GAS CHROMATOGRAPHY ANALYSIS OF METHYL ESTERS OF FATTY ACIDS FROM *BROMUS CATHARTICUS* AND *CRYPTOCARYA ALBA* SEEDS.

	Percent of total fatty acid methyl esters					
	16:0	16:1	18:0	18:1	18:2	18:3*
Bromus catharticus						
Seeds	23.15	—	1.11	13.40	51.36	2.23
Cryptocarya alba						
Fruits	15.73	9.51	2.18	44.50	25.03	2.93
Fruit peel	10.00	17.15	1.52	56.92	9.52	3.23

* Number of carbons and unsaturated bonds, respectively.

seeds of *Bromus catharticus* was determined by gas chromatography of the corresponding fatty acid methyl esters (FAME). Some 100 mg of the petroleum ether solubles were saponified with 15 ml of alcoholic KOH for 20 min. After addition of 15 ml of 20% BF_3 in methanol, the reaction mixture was refluxed for two hours. To this, water and 5 ml of hexane were added. The methyl esters dissolved in the hexane fraction. The mixture was analyzed by gas chromatography on a Shimadzu GC-9A with FID detector using a fused silica capillary column SUPELCO SP-2330 (30 m, 0.25 mm ID, 0.20 μm film thickness). The FAME were identified by comparison of the retention times and peak enhancement with known FAME mixtures as well as by GC/MS using a Hewlett-Packard 5890 equipment with a HP-5 capillar column (Heldrich 1990:963–965). Results are presented in Table 2.

To assess the quality of the protein contained in the surveyed plants and mushrooms, the amino acid composition of acid hydrolizates was determined. After treatment with hydrochloric acid, hydrolized amino acids from samples were derivatized with phenylisothiocyanate and the resulting phenylthiocarbamyl (PTC) derivatives were separated and quantitated by reversed-phase HPLC using a Merck-Hitachi equipment consisting of a L-6200 pump, a L-4000 UV detector and D-2500 chromato-integrator. The column used was a Superspher 60 RP-Select B 250–4 mm (Bidlingmeyer, Cohen and Tarvi 1984; Elkin and Wasynczuk 1987; Bidlingmeyer et al. 1987; Heldrich 1990: 1096–1097). Alpha-aminobutyric acid (AABA) was used as an internal standard. Values were corrected by a normalized factor of 95% hydrolysis. Results are shown in Tables 3 and 4.

BIOLOGICAL ACTIVITY

The dry plant material and mushrooms fruiting bodies were macerated in MeOH, chopped and re-extracted with MeOH and EtOAc. After filtration, the combined extracts were concentrated and partitioned with dichloromethane (DCM) and H_2O affording a DCM and a water-soluble fraction that were concentrated under reduced pressure and then lyophilized.

Xanthine Oxidase Activity

Xanthine oxidase (XO) derived from cow's milk, xanthine and the standard inhibitor allopurinol were purchased from Sigma Chemical Co. (St. Louis, MO, U.S.). The XO activities with xanthine as substrate were measured spectrophotometrically as previously reported using a Shimadzu UV-160A equipment (Schmeda-Hirschmann et al. 1992: 1996). The crude extracts were evaluated at 50 μg/ml.

β-Glucuronidase Activity

β-Glucuronidase (β-gluc) activity was measured spectrophotometrically as previously reported (Schmeda-Hirschmann et al. 1992; 1994), using p-nitrophenyl β-D-glucuronide as substrate. The inhibitor glucosaccharo-1:4-lactone was used as a control. All reagents were obtained from Sigma Chemical Co. Extracts were tested at 50 μg/ml.

DNA Binding Activity

The DNA binding activity was assessed by HPLC as described by Pezzutto et al. (1991) with some modifications (Klinar et al. 1995). Calf thymus DNA was purchased from Sigma Chemical Co. The HPLC equipment used was a Merck-Hitachi with a L-4000 absorbance detec-

TABLE 3. AMINO ACID COMPOSITION OF AMERINDIAN FOOD PLANTS.[1]

	Bromus catharticus	*Dioscorea humifusa*	*Prosopis alba*	*Prosopis alba* var. *panta*	*Prosopis tamarugo*
Aspartic acid	2.65	4.61	0.83	1.24	6.22
Glutamic acid	13.23	6.38	0.80	1.10	10.38
Serine	2.54	2.28	0.20	0.50	4.54
Glicine	1.77	0.84	0.31	0.51	2.40
Histidine	1.11	0.94	0.13	1.03	3.19
Arginine	2.57	7.57	0.40	—	6.50
Threonine	1.89	1.67	0.25	0.52	2.57
Alanine	3.10	4.01	0.50	2.30	5.47
Proline	9.14	1.92	1.90	2.50	36.80
Phenylalanine	4.00	0.90	0.24	1.80	2.74
Tyrosine	2.42	4.87	0.40	0.63	2.29
Valine	4.14	2.40	0.35	0.88	3.85
Methionine	—	—	0.24	—	—
Cysteine	—	—	0.80	—	—
Isoleucine	2.67	0.89	0.08	0.81	1.42
Leucine	4.27	1.61	0.53	1.06	2.58
Lysine	3.16	2.20	0.15	0.40	2.80

[1] g Amino acid/16 g N, corrected.

tor set at 254 nm and a D-2500 recorder/data module. Column: C 18 RP (Lichrospher 100, 5 μ particle size). Mobile phase: water to methanol (100:0 to 0:100). Under these conditions, free DNA eluted with a R_t = 0.5 min. Test samples and DNA solutions were pre-mixed and incubated at room temperature for 30 min. before injection. Extracts were assayed at 0.50 mg/ml.

TABLE 4. AMINO ACID COMPOSITION OF NATIVE EDIBLE FUNGI.[1]

	Boletus loyus	*Clavaria coralloides*	*Grifola gargal*	WHO
Aspartic acid	11.85	35.50	2.40	
Glutamic acid	18.42	36.40	3.50	
Serine	10.61	29.20	3.66	
Glicine	4.68	9.10	0.90	
Histidine	3.98	7.80	0.36	
Arginine	6.17	15.00	0.93	
Threonine	7.26	18.90	1.51	4.0
Alanine	10.70	19.86	2.66	
Proline	6.72	15.30	1.81	
Phenylalanine	3.73	8.36	2.85	6.0
Tyrosine	2.74	10.00	3.98	
Valine	6.99	12.70	2.70	5.0
Methionine	3.26	—	—	3.5
Isoleucine	5.50	12.00	2.82	4.0
Leucine	8.44	15.30	2.14	7.0
Lysine	5.85	8.80	1.63	5.5

[1] g Amino acid/16 g N, corrected. WHO: World Health Organization ideal values for essential amino acids.

Doxorubicine and vinblastine were used as standard inhibitors.

Free Radical Scavenging Activity

The free radical scavenging effect of the crude extracts was assessed by the decoloration of a methanolic solution of the diphenyl picryl hydrazyl radical (DPPH) according to Joyeux et al. (1995). Extracts were tested at 50 and 10 μg/ml.The degree of decoloration indicates the free radical scavenging efficiency of the substances. A methanolic solution of DPPH served as a control. The percentage of DPPH decoloration was calculated as follows:

$$\text{Decoloration percentage} = 1 - \frac{(\text{Absorbance with compound})}{\text{Absorbance of the blank}} \times 100.$$

Results are presented in Table 5.

Hypotensive Effect in Normotensive Rats

The effect of mushrooms and *Thypha angustifolia* extract on the blood pressure of normotensive rats was assessed by intravenous administration of the samples. Animals were anaesthetized with urethane (1.2 g/kg, i.p.). The blood pressure was measured by means of a catheter implanted into the left carotid artery and connected to a P-23 XL Gilson pressure transducer. Crude extracts were administered intravenously

TABLE 5. BIOLOGICAL ACTIVITY OF PLANTS AND MUSHROOMS USED AS FOOD BY CHILEAN AMERINDIANS.

Taxon and extract name	Enzyme inhibition % at 50 µg/ml		Free radical scavenging DPPH[2] % decoloration		DNA binding 0.50 mg/ml
	β-Gluc[3]	XO[4]	50 µg/ml	10 µg/ml	
Apium australe PE	33	29	0	0	0
Apium astrale MeOH	58	6	0	0	26
Bromus catharticus Seeds, PE	5	2	0	0	0
Bromus catharticus Seeds, MeOH	24	15	0	0	18
Cryptocarya alba Fruit, MeOH	5	32	70	8	57
Dioscorea bridgesii Aerial parts, MeOH	50	4	35	6	0
Dioscorea bridgesii Tubers, MeOH	65	6	9	0	11
Prosopis alba	31	43	0	0	0
Prosopis alba var. *panta* Pods, MeOH	34	1	0	0	2
Prosopis tamarugo Pods, MeOH 1996	2	5	10	1	34
Prosopis tamarugo Pods, MeOH 1997	42	0	0	0	18
Sanicula graveolens Aerial parts, PE	13	9	0	0	0
Sanicula graveolens Aerial parts, MeOH	43	2	51	6	0
Typha angustifolia Aerial parts, MeOH	0	0	0	0	87
Typha angustifolia Rhizome, MeOH	10	3	7	0	69
Boletaceae					
Boletus loyus MeOH	4	14	3	0	16
Clavariaceae					
Clavaria coralloides MeOH	2	3	15	0	24
Polyporaceae					
Grifola gargal MeOH	2	2	3	1	41

[1] Extract: PE = petroleum ether-soluble; MeOH = methanol-soluble.
[2] DPPH = free radical scavenging activity.
[3] β-Gluc = β-Glucuronidase.
[4] XO = Xanthine oxidase.

TABLE 6. CHANGES IN MEAN BLOOD PRESSURE INDUCED BY CRUDE AQUEOUS EXTRACTS AFTER INTRAVENOUS ADMINISTRATION (5 MG/KG) IN NORMOTENSIVE RATS.

Taxon	ME% P ± SD[1]	Maximum effect (min.)	Lasting time (min.)	N
Boletus loyus	−5.4 ± 1.6	0.60	2.0	4
Clavaria coralloides	−19.3 ± 5.2	0.60	1.9	6
Grifola gargal	−17.8 ± 4.1	0.17	1.7	5
Typha angustifolia	−18.7 ± 0.9	0.16	2.3	6
Acetylcholine (300 ng/kg)	−32.5 + 0.8	0.10	1.6	3

[1] ME% P ± SD: percent variation in the mean blood pressure ± standard deviation, in mm Hg.

at 5 mg extract/kg body weight in a volume of 0.1 mL. The samples were dissolved in saline solution. The mean basal pressure (BP) was 120 ± 0.5 mm Hg (n = 24). After recording the BP the extract was administered and the effect was observed until recovery of the initial values. Each extract was assayed in at least three different animals and the experiment was repeated twice with each rat. The values are reported as percentage variation of the basal blood pressure in mm Hg as mean values ± standard deviation. The maximum effect time as well as the duration (min) were also assessed. Under the same conditions, the vehicle (saline solution) did not elicit changes in the blood pressure. Acetylcholine (300 ng/kg) was used as a reference hypotensor. Statistical analysis was performed according to paired Student's two-tailed t-test (See Table 6). p values <0.05 were considered significant (Schmeda-Hirschmann et al. 1992).

Results and Discussion

The proximate analysis and biological activity of thirteen edible higher plant and mushroom taxa have been asessed in order to relate their use as food with their potential as a source of bioactive products.

Nutrients

The plants gathered as a whole were a source of carbohydrate, with higher values for the tubers of *Dioscorea* spp., *Prosopis alba* pod meal and *Bromus catharticus* seeds. The protein and carbohydrate content of our *B. catharticus* collection is close to local wheat samples (14% crude protein and 76% NNE) (Schmidt-Hebbel and Pennachiotti 1985). Aconitic acid has been reported to occur in *B. catharticus* (Hegnauer 1986, vol. 7: 635).

Among the surveyed *Dioscorea* species, the carbohydrate content ranges between 71.07 and 80.95%, increasing after peeling. The lowest values for crude protein was found in *D. humifusa* var. *humifusa* (3.63%), while the values for *D. humifusa* var. *gracilis* and *D. bridgesii* were higher (5.32–6.00%). For Chilean potatoes (*Solanum tuberosum*), the carbohydrate content ranges between 80–84% with 9–13% crude protein. Sweet potatoes (*Ipomoea batatas*) tubers contain 87% of carbohydrates and 4% protein (Schmidt-Hebbel and Pennachiotti 1985). According to Hegnauer (1963, Vol. II:146–147), chelidonic acid has been isolated from *Dioscorea humifusa.*

The edible pod meal of *Prosopis alba* and *P. alba* var. *panta* has a pleasant, sweet taste and is still consumed in some small villages in northern Chile. The proximate composition of Argentinian samples of *Prosopis chilensis, P. argentina, P. nigra, P. flexuosa, P. caldenia* and *P. alba* pods was reported by Lamarque et al. (1994). Crude protein, fatty acids and sterols content were investigated. On a dry weight basis, the crude protein content ranged between 242 and 356 g/kg. The oil content was 107–172 g/kg. The seed lipids contained linoleic and oleic acids as main constituents. The main sterol in all samples was ß-sitosterol. The amino acid composition was not reported. *Prosopis tamarugo* is an important foraging species in northern Chile. Their pod meal is astringent and is not used as an human food. References to edible "algarrobo" pods in Chile refer most appropiately to *P. alba* and their varieties.

Low protein content was observed for most samples, except the seeds of *Bromus catharticus* (14%) and fruiting bodies of *Boletus loyus* and *Clavaria coralloides* (22 and 26%, respectively). According to the amino acid profile and the WHO reference, the best protein sources were *Clavaria coralloides* and *Boletus loyus.* Both were, however, deficient in methionine and cysteine. *Bromus catharticus* protein proved to be deficient in threonine, valine, methionine, cysteine, isoleucine, leucine and lysine. PTC analysis, however, could account for decreased amounts of cystine. *Prosopis tamarugo* pods showed a very high proline content. This fact is in agreement with the ecological response to the saline environment where the tree grows (Pahlich 1990).

The carbohydrate-rich rhizomes of *Scirpus californicus* and *Typha angustifolia* as well as the *Prosopis tamarugo* pods are also characterized by high fiber content (17.2–31.4%). In our study, however, ripe *P. tamarugo* pods were used. The rhizomes of *Scirpus grossus* contained 77.1% carbohydrates, 8.9% of total proteins, 0.57% lipids, 2.6% fiber and 4% ash (Hegnauer 1986:600–609).

The unsaturated fatty acids acids, oleic, linoleic and linolenic account for 67% of the total fatty acids present in the lipid fraction of *Bromus catharticus.* For *Cryptocarya alba* fruits, the main fatty acid components are the unsatu-

rated C 18:1 to C 18:3, as determined by GC. From the lipid fraction of *Scirpus californicus,* sitosterin and stigmasterin were identified (Schmeda-Hirschmann et al. 1996).

Biological Activity

Eighteen extracts from thirteen higher plant and mushroom taxa were assayed as inhibitors of the enzymes xanthine oxidase (XO) and β-glucuronidase (β-gluc) as well as for their DNA-binding activity and free radical scavenging effect in vitro. Under our assay conditions, the IC_{50} of the standard inhibitors were as follows: allopurinol (XO) 0.11 mM; glucosaccharo-1:4-lactone (β-gluc), 0.40 mM; doxorubicine and vinblastine at 0.50 mg/ml: 100 and 70% DNA binding, respectively.

The fruits of *Cryptocarya alba* proved to be the most active in our bioassays, inhibiting by 70% the DPPH decoloration at 50 μg/ml and with a strong DNA binding effect in vitro. The marginal effect of the fruit extract towards the enzyme β-glucuronidase at 50 μg/ml contrast with the IC_{50} value of 7.0 μg/ml reported for an hydroalcoholic leaf extract (Schmeda-Hirschmann et al. 1992).

The seeds of *Bromus catharticus* as well as the extracts of *Typha angustifolia* were weakly active or inactive as enzyme inhibitors, the latter showing a strong effect in the DNA binding assay at 0.50 mg/ml.

Dioscorea extracts were active towards the ß-glucuronidase with an IC_{50} of 40–50 μg/ml, the aerial parts being more active than the edible tubers. The biological activity of *Scirpus californicus* has been recently reported. Bioassay-guided isolation leads to the stilbenes piceatannol, scirpusin A and B as the main xanthine oxidase inhibitors of the rhizome extract (Schmeda-Hirschmann et al. 1996).

Extracts of *Prosopis alba* and *P. alba* var. *panta* pods displayed some activity towards the enzyme β-gluc at 50 μg/ml. A sample of *P. alba* was also active in the XO assay. Some DNA binding activity was detected in *P. tamarugo* pods.

Apium australe and *Sanicula graveolens* extracts were moderately active as β-glucuronidase inhibitors and showed a very weak inhibition towards the xanthine oxidase. The methanolic extract of *Sanicula graveolens,* however, displayed some activity as a radical scavenger at 50 μg/ml. Bioassay-guided isolation led to caffeic acid derivatives and flavonoids as the main free radical scavengers of *S. graveolens.* After hydrolysis, caffeic acid and quercetin proved to be the bioactive principles of the plant (Viturro, Molina and Schmeda-Hirschmann 1999). The occurrence of a furocoumarin and myristicin from the aerial parts of *Apium australe* has been reported (Hegnauer, vol. 9, 1990:683).

Biological evaluation of the samples in our in vitro test systems shows that the edible fungi are almost inactive at the assayed concentrations. A moderate effect in the DNA binding assay was demostrated for *Grifola gargal* (41%), while *Clavaria coralloides* and *Boletus loyus* (24% and 16%, respectively) were less active.

At 5 mg/kg, the water-soluble extracts of *Clavaria coralloides, Grifola gargal* and *Typha angustifolia* elicit a strong hypotensive response in normotensive rats (−17.8% to −19.3%) (Table 6).

Conclusions

Thirteen edible plant and mushroom taxa formerly gathered by the Mapuche or other Chilean Amerindians were choosen for proximate composition and biological activity studies based on ethnobotanical information or current use.

The plants and fungi presented in this study were probably seasonal food sources, except *Scirpus californicus* and *Typha angustifolia,* which were available all year. *Dioscorea* species and the reported fungi can be gathered at the end of winter and/or early spring, while *Bromus* and *Madia* seeds are available during late summer. *Prosopis* species were a main food supply in northern Chile and their fruits ripen in summer. The consumption of biologically active fruits of *Cryptocarya alba* was (as in present days) mainly as a sweet, being chewed. Tubers, rhizomes, pods and fruits complemented nutrition where the main protein intake probably consisted of animal meat and/or cultivated plants. Some of the surveyed plants and mushrooms also contributed biologically active secondary metabolites that might have contributed to maintenance of health.

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Book Review

Plant Breeding Systems, ed. 2. A. J. Richards. 1997. Chapman & Hall, 115 Fifth Avenue, New York, NY 10003. xii + 529 pp. (hardcover). $ 49.95 (paperback), no price found for hardcover. ISBN 0-412-57440-3.

This remarkable book is the most detailed and unbiased discussion of this topic that has appeared recently. Some of the newest books on breeding systems are so biased in their discussions that one wonders if the authors realize that they are studying individuals of living organisms or "virtual nature." Richards is completely aware of the lives of the organisms he discusses, and the variables they must content with to survive. All those interested in how reproduction in the plants they study interacts with other organisms, human or otherwise, should read this book.

Surprisingly, this book goes from the very basics of flower parts and how the function, to amazing depth in the complexities of genetics and population ecology. The *Introduction* discusses plant vs. animals breeding systems, and the roles of breeding. *Sexual theory in seed plants* covers the fundamentals, distribution, advantages, and disadvantages of sexual reproduction, plus other things. These discussions are followed by chapters entitled *Sexual reproduction in flowering plants,* and *Floral diversity and pollination.* A change takes the topic next *to Pollination biology and gene flow,* then *Multi-allelic self-incompatibility.* Four more chapters include *Heteromorphy, Dicliny, Self-fertilization and inbreeding,* and *Agamospermy.* The book is concluded with a *Conclusion* chapter, a *Glossary, References* and an *Index.*

The author has made every effort to update the original 1986 version with mostly new references (pp. 471–512) and dramatically changed information in some of the chapters. Clearly not all aspects of plant reproduction can be covered in a book of this size, although it is not small (over 530 pages). Richards points out that the buzz-word "Biodiversity," now so common, did not even exist when the first edition of his book appeared. I wish he had referenced the statement (p. xi) that the rate of destruction of tropical forests has declined by more than 80% since 1986. Even if that is true, and I doubt it, there remains too much unknown about the World humans scandalously ravage.

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