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FORUM [FORO]

NUTRITIONAL EVALUATION AND ELIMINATION OF TOXIC PRINCIPLES IN WILD YAM (*Dioscorea* spp.)

[EVALUACIÓN NUTRICIONAL Y ELIMINACIÓN DE COMPUESTOS TÓXICOS EN *Dioscorea* spp. SILVESTRES]

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SUMMARY

Yam (*Dioscorea spp*.) is the staple of people in the tropics, but certain wild varieties are inedible because of the toxic substances present, some of which have pharmacological properties. The wild yam tubers consumed by the tribal Kanikkars of Kanyakumari district (*Dioscorea alata*, *D. bulbifera*, *D. esculenta*, *D. oppositifolia*, *D. pentaphylla*, *D. tomentosa* and *D. wallichi*) were evaluated for its nutritional quality. This paper reveals the proximate composition, mineral profiles, total protein, starch and sugar content of the wild yam tubers. The toxic principles like phenols, tannins, hydrogen cyanide, oxalate, amylase inhibitor activity and trypsin inhibitor activity were quantified. Treatments like soaking, cooking and autoclaving and their effects on changes in phenolics, tannins, hydrogen cyanide, oxalate, amylase inhibitor activity and trypsin inhibitor activity were investigated in the seven tuber samples. Soaking reduced the levels of antinutrients up to 80% whereas cooking and autoclaving (45 min.) reduced the toxic contents to a greater extent (< 80%). The amylase and trypsin inhibitor activity showed only slight reduction (up to 10%) in toxic principles after treatments. Of all the different water and hydrothermal treatments studied autoclaving seemed to be the most efficient method in eliminating the anti-nutrient present.

Key words: Anti-nutrients, hydrogen cyanide, amylase inhibitor activity, trypsin inhibitor activity.

RESUMEN

Dioscorea spp. es un alimento básico en el trópico, pero algunas variedades silvestres no son comestibles debido a la presencia de compuestos tóxicos, algunos de los cuales tiene propiedades farmacológicas. Se evaluó la calidad nutricional de los tubérculos de especies silvestres de *Dioscorea* (*Dioscorea alata*, *D. bulbifera*, *D. esculenta*, *D. oppositifolia*, *D. pentaphylla*, *D. tomentosa* y *D. wallichi*) consumidas por las tribus Kanikkars del distrito de Kanyakumari en la India. Se presentan los resultados de composición química, perfil mineral, proteína, almidón y azúcares. De igual manera, se cuantificaron los contenidos de fenoles, taninos, cianuro de hidrógeno, oxalato, inhibidores de amilasa e inhibidores de tripsina. El remojo redujo los niveles de estos compuestos hasta en un 80%, mientras que el cocido y autoclave (45 min) redujeron el contenido en una proporción mayor (<80%). La actividad de los inhibidores de tripsina y amilasa únicamente no se redujó más del 10% con estos tratamientos. Entre los métodos estudiados el uso de autoclave paracе ser el método más eficiente para eliminar la mayor proporción de los compuestos antinutricionales presentes.

Palabras clave: antinutrientes, cianuro de hidrógeno, inhibidor de tripsina, inhibidor de amilasa.

INTRODUCTION

India has one of the largest concentrations of tribal population in the world. The forest plays a vital role in the economy as well as daily needs of the tribals. In times of scarcity when the staple food is in short of supply tribals collect many types of wild roots and tubers to supplement their meagre food available at home (Vidyarthi, 1987). Nature has endowed plants with the genetic capacity to synthesize substances that are toxic and thus to ensure their survival against predators whether they be insects, fungi or animals including humans. Humans have learnt which foods are safe to eat or how such foods can be treated in order to destroy their toxicity.

Some wild yams like *Dioscorea alata*, *D. bulbifera*, *D. esculenta*, *D. oppositifolia*, *D. pentaphylla*, *D. tomentosa* and *D. wallichi* are used as food by the tribal Kanikkars of Kanyakumari district in times of food shortage but only after extensive detoxification processes. *D. alata* is recommended to diabetic patients as tolerable energy resources. The toxic principles of *Dioscorea* exhibit medicinal properties. Hence the biochemical basis of this is worthy of investigation. The study was made keeping in mind the nutritive value of yam tubers with food security orientation.

MATERIAL AND METHODS

Seven samples of wild yam tubers (*Dioscorea alata* L., *D. bulbifera* L., *D. esculenta* (Lour.) Burk, *D. oppositifolia* L., *D. pentaphylla* L., *D. tomentosa* J. Koniger Spreng. and *D. wallichi* Hook) grown in sandy loam soil consumed by the tribal Kanikkars were collected using multistage sampling technique in three consecutive rainy seasons during August and January from the South Eastern slopes of Western Ghats, Kanyakumari district, Tamil Nadu. Moisture content was determined by drying the samples in an oven at 80°C for 24 hrs and was expressed on a percentage basis. The samples were powdered in Willey mill 60 mesh size and stored in screw cap bottles at room temperature for further analysis. Nitrogen content was estimated by the micro-kjeldhal method (Humphries, 1956) and crude protein was calculated (N x 6.25).

The contents of crude lipid, crude fibre and ash were estimated by AOAC (1970) methods. Nitrogen free extract was obtained by difference method by subtracting the sum of the protein, fat, ash and fibre form the total dry matter (Muller and Tobin, 1980). The energy value of the corm was estimated (KJ) by multiplying the percentages of crude protein, crude lipid and NFE by the factors 16.7, 37.7 and 16.7 respectively. From the triple acid digested sample, sodium, potassium, calcium, magnesium, iron, copper, zinc and manganese were analysed using an atomic absorption spectrophotometer (Perkin Elmer Model 5000) (Issac and Johnson, 1975). Phosphorus was estimated colorimetrically (Dickman and Bray, 1940). The total soluble protein of the extract was estimated by the method of Lowry *et al*. (1951). The total starch and sugar content were determined by the titrimetric method of Moorthy and Padmaja (2002). The anti-nutritional factors, total free phenolics (Sadasivam and Manickam, 1996), tannins (Burns, 1971) hydrogen cyanide (Jackson, 1967); total oxalate (AOAC, 1984); trypsin inhibitor activity (Sasikiran and Padmaja, 2003) and amylase inhibitor activity (Rekha and Padmaja, 2002).

Processing methods

All the seven samples were subjected to treatments like soaking, cooking and autoclaving in three replications.

Soaking: The peeled yam tubers were chipped using hand operated chipping machine (1-1.5cm) and soaked in distilled water and 0.02% (w/v Sodium bicarbonate ($NaHCO_3$) solution (pH 8.6) for 3, 6 and 9 hours in the ratio of 1:10 (w/v). The water was drained off and the samples were dried at 55°C.

Cooking: Separate batches of the samples were cooked in distilled water (100°C) in the ratio of 1:10 (w/v) for 30, 60 and 90 minutes. The cooked samples were rinsed and dried.

Autoclaving**:** The samples were autoclaved at 15lbs. pressure (121°C) in distilled water (1:10 w/v) for 15, 30 and 45 min. The samples were rinsed with distilled water and dried at 55°C.

Statistical analysis

Data were analysed using the statistical analysis system SPSS (SPSS Software for windows release 10.0; SPSS Inc., Chicago, IL, USA). Analysis of variance and mean separations were calculated by the general linear model procedures.

RESULTS AND DISCUSSION

Table 1, shows the proximate and mineral composition of the tubers of *Dioscorea* spp. The proximate composition reveals that the crude protein, crude lipid, crude fibre, ash and NFE content is found to be higher when compared with the earlier reports (Oyenuga, 1968 and Rajyalakshmi and Geervani, 1994). Table 2. shows the mineral composition, total soluble protein, total starch and sugar content of the investigated yam tubers. The tubers are found to contain more than the adequate level of potassium compared to RDA's of infants and children (<800mg) (NRC/ NAS, 1980). *Dioscorea* tubers are rich sources of carbohydrate and protein. The protein content is rather low, ranging from 1-2% of the fresh weight (Mandal, 2006). Apart from being an item of food, yam is becoming increasingly important medicinally because of the toxic principles present.

Table 1. Proximate and Mineral composition of *Dioscorea alata*, *D. bulbifera*, *D. esculenta*, *D. oppositifolia*, *D. pentaphylla*, *D. tomentosa* and *D. wallichi* [a]

	D. alata	*D. bulbifera*	*D. esculenta*	*D. oppositifolia*	*D. pentaphylla*	*D. tomentosa*	*D. wallichi*
Proximate composition (g /100g)							
Moisture	87.72	92.48	86.67	88.93	90.13	87.60	71.06
CP	10.73±0.13	15.75±0.08	10.50±0.05	13.54±0.11	5.68±0.11	5.25±0.04	10.50±0.06
CL	2.50±0.02	8.13±0.01	2.58±0.01	6.43±0.12	4.01±0.04	2.86±0.01	1.18±0.01
CF	6.48±0.12	3.92±0.03	7.82±0.05	8.47±0.11	7.10±0.07	3.21±0.03	9.23±0.15
Ash	8.50±0.04	2.86±0.02	6.17±0.11	6.09±0.03	2.68±0.11	2.48±0.14	8.42±0.07
NFE	71.79	69.34	72.93	65.47	80.53	86.20	70.67
GE	1472.33	1727.50	1490.55	1561.88	1590.89	1635.04	1400.03
Mineral composition (mg/100g)							
Na	32.00±0.06	63.38±0.01	51.68±0.08	113.00±0.14	83.24±0.11	25.54±0.10	54.36±0.04
K	155.00±1.21	1548.00±0.82	1638.00±0.13	1460.41±0.13	1230.60±0.62	1335.42±0.54	1241.60±0.12
Ca	338.10±0.12	228.15±0.08	238.10±0.01	124.00±0.03	560.10±0.12	220.30±0.11	660.00±0.09
Mg	566.10±0.14	440.17±0.10	330.00±0.12	548.33±0.11	430.10±0.10	219.08±0.04	618.10±0.03
P	110.10±0.03	120.50±0.06	126.30±0.12	98.40±0.08	136.00±0.08	78.68±0.06	96.40±0.04
Zn	1.26±0.01	1.30±0.01	1.48±0.02	1.20±0.06	3.32±0.03	5.20±0.01	4.44±0.01
Mn	5.36±0.03	10.60±0.03	4.54±0.04	8.84±0.24	2.22±0.01	1.10±0.01	2.58±0.01
Fe	33.10±0.02	3.90±0.12	9.38±0.09	28.10±0.12	78.10±0.11	22.00±0.01	18.12±0.09
Cu	8.30±0.02	2.64±0.02	1.34±0.03	6.58±0.03	14.10±0.09	1.34±0.01	3.54±0.01

CP: Crude protein, CL: Crude lipid, CF: Crude fibre, NFE: Nitrogen free extract, GE: Gross energy, $KJ.100^{-1}$ DM
a: values are mean of triplicate determinations expressed on dry weight basis. b: ± standard error.

Table 2. Soluble protein, starch and sugar content of *Dioscorea alata*, *D. bulbifera*, *D. esculenta*, *D. oppositifolia*, *D. pentaphylla*, *D. tomentosa* and *D. wallichi* (g/100g)[a]

Name	Soluble protein	Starch	Total Sugar
D.alata	1.29±0.14[b]	64.29±2.23	1.19±0.11
D.bulbifera	2.16±0.31	42.86±1.13	2.70±0.03
D. esculenta	1.12±0.04	60.00±0.48	5.0±0.12
D. oppositifolia	1.67±0.18	64.29±0.51	5.49±0.48
D. pentaphylla	1.18±0.04	61.26±0.52	3.28±0.14
D. tomentosa	1.22±0.14	56.26±0.14	6.02±0.04
D. wallichi	1.22±0.09	56.25±0.34	3.57±0.14

a: values are mean of triplicate determinations expressed on dry weight basis.
b: ± standard error.

The anti-nutritional factors like total free phenolics, tannins, hydrogen cyanide, total oxalate, amylase inhibitor and trypsin inhibitor activities are presented in Table 3. Phenolic compounds inhibit the activity of digestive as well as hydrolytic enzymes such as amylase, trypsin, chymotrypsin and lipase (Salunkhe *et al*., 1982). The total free phenolics content in the tubers of *Dioscorea alata* and *D. oppositifolia* is lower than that of the earlier studies in the tubers of *D. alata* (Osagie and Opoku, 1982). Recent researches report that the phenolic compound is the main human dietary antioxidant and has a decreased incidence of chronic diseases (Padmaja *et al*., 2005).

The level of tannins, hydrogen cyanide and total oxalate is found to be lower when compared with the earlier reports of the tubers of *Dioscorea alata*, *D. cayenensis*, *D. rotundata* and *D. esculenta* (Esuabana, 1982). The tubers of *D. oppositifolia* contain more trypsin inhibitor activity when compared with earlier reports in the tubers of *Dioscorea dumetorum* and *D. rotundata* (Sasikiran *et al*., 1999).

Many foods including the root crops cannot be digested in their natural state and hence requires cooking, which increases the palatability, the keeping qualities and the safety of the foods from potentially toxic substances (Bradbury and Holloway, 1988). Various methods of processing treatments tend to reduce the level of anti-nutritional factors like total free phenolics, tannins, hydrogen cyanide, total oxalate, amylase inhibitor and trypsin inhibitor activity in the yam tubers (Tables 4 and 5).

A number of polyphenolic compounds are present in plants, which contribute towards the defense mechanism of plants. Although these are considered earlier as antinutritional compounds, under the present nomenclature phenols fall under the category of nutraceuticals, offering many nutritional advantages to man (Padmaja *et al.*, 2005). Soaking of tubers of *Dioscorea bulbifera* and *D. esculenta* in distilled water and sodium bicarbonate solution (0.02%w/v for 9 hrs.) showed only 6-10% of reduction in the level of phenolics favouring health benefits. However, excess of phenolics and tannins should be removed. Pressure cooking resulted in maximum loss of phenolics, tannins and total oxalate ($P<0.01$ in *D. oppositifolia* and *D. bulbifera* and $P<0.05$ in *D. alata*) followed by cooking and soaking.

Table 3. Anti-nutritional factors of the tubers of *Dioscorea alata*, *D. bulbifera*, *D. esculenta*, *D. oppositifolia*, *D. pentaphylla*, *D. tomentosa* and *D. wallichi* (g/100g)[a]

Botanical Name	Total free pheolics	Tannin	Hydrogen cyanide mg/100g	Total oxalate	Amylase inhibitor AIU	Trypsin inhibitor TIU
D. alata	0.49±0.003[b]	0.51±0.015	0.15±0.006	0.63±0.002	8.11	2.75
D. bulbifera	1.40±0.012	1.59±0.014	1.12±0.003	0.98±0.001	1.01	0.87
D. esculenta	0.56±0.011	0.11±0.003	0.19±0.001	0.23±0.001	14.50	2.92
D.oppositifolia	0.28±0.006	0.15±0.009	0.11±0.003	0.31±0.003	1.69	23.70
D. pentaphylla	0.41±0.002	0.06±0.001	0.17±0.004	0.33±0.001	1.85	2.56
D. tomentosa	0.61±0.005	0.10±0.001	0.14±0.002	0.30±0.004	3.04	1.47
D. wallichi	0.22±0.003	0.02±0.03	0.15±0.012	0.10±0.002	5.07	2.10

a: values are the mean of triplicate determinations expressed on dry weight basis b: ±standard error

Table 4. Effect of soaking, cooking and autoclaving on the levels of total free phenolics, tannins, hydrogen cyanide, total oxalate, amylase inhibitor activity and trypsin inhibitor activity in *Dioscorea alata*, *D. bulbifera*, and *D. esculenta*.

Treatment	Botanical Name	Total free phenolics g/100g	Tannin g/100g	Hydrogen cyanide mg/100g[1]	Total oxalate g/100g	Amylase inhibitor AIU	Trypsin inhibitor TIU
Raw Sample	*D. alata*	0.49±0.003[b]	0.51±0.015	0.15±0.006	0.63±0.002	8.11	2.75
	D. bulbifera	1.40±0.012	1.59±0.014	0.12±0.003	0.98±0.001	1.01	0.87
	D. esculenta	0.56±0.011	0.11±0.003	0.19±0.001	0.23±0.001	14.20	2.92
Soaking in distilled water 9 h	*D. alata*	0.39±0.003 (-20)	0.32±0.010 (-37)	0.10±0.001 (-35)	0.58±0.001 (-8)	8.00	2.60
	D. bulbifera	1.33±0.003 (-6)	1.38±0.006 (-14)	0.08±0.001 (-34)	0.86±0.001 (-12)	0.91	0.79
	D. esculenta	0.51±0.009 (-9)	0.08±0.002 (-27)	0.11±0.002 (-41)	0.19±0.000 (-18)	14.29	2.49
Soaking in sodium bicarbonate solution 9 h	*D. alata*	0.35±0.002 (-29)	0.43±0.013 (-10)	0.09±0.001 (-42)	0.56±0.001 (-11)	8.01	2.59
	D. bulbifera	1.28±0.003 (-9)	1.45±0.004 (-9)	0.09±0.003 (-24)	0.87±0.001 (-11)	0.95	0.80
	D. esculenta	0.48±0.008 (-8)	0.05±0.000 (-56)	0.09±0.001 (-52)	0.18±0.001 (-22)	14.25	2.54
Cooking 90 min.	*D. alata*	0.19±0.002 (-61)**	0.36±0.010 (-30)**	0.04±0.001 (-74)	0.52±0.002 (-17)**	7.89	2.51
	D. bulbifera	0.38±0.001 (-13)*	1.42±0.012 (-11)*	0.03±0.007 (-15)	0.82±0.001 (-16)*	0.81	0.78
	D. esculenta	0.16±0.006 (-71)	0.03±0.010 (-65)	0.09±0.001 (-49)	0.09±0.001 (-61)	14.38	2.75
Autoclaving 45 min.	*D.alata*	0.16±0.001 (-67)	0.31±0.010 (-39)	0.08±0.001 (-87)	0.55±0.002 (-13)	7.91**	2.45**
	D. bulbifera	0.35±0.001 (-75)	1.39±0.001 (-13)	0.008±0.001 (-93)	0.80±0.003 (-18)	0.80*	0.75*
	D. esculenta	0.19±0.010 (-65)	0.01±0.000 (-91)	0.03±0.001 (-84)	0.08±0.000 (-15)	14.36*	2.78*

a: values are mean of triplicate determinations expressed on dry weight basis. b: ± standard error ; Values in the parenthesis indicate percent loss from raw samples. * significant at $P<0.01$; ** significant at $P<0.05$

Table 5. Effect of soaking, cooking and autoclaving on the levels of total free phenolics, tannins, hydrogen cyanide, total oxalate, amylase inhibitor activity and trypsin inhibitor activity in *Dioscorea oppositifolia*, *D. pentaphylla*, *D. tomentosa* and *D. wallichi*.[a]

Treatment	Botanical Name	Total free phenolics g/100g	Tannin g/100g	Hydrogen cyanide mg/100g	Total oxalate g/100	Amylase inhibitor AIU	Trypsin inhibitor TIU
Raw sample	*D. oppositifolia*	0.28±0.006[b]	0.51±0.009	0.11±0.003	0.31±0.003	1.69	93.73
	D. pentaphylla	0.41±0.002	0.06±0.001	0.17±0.004	0.33±0.001	1.85	2.56
	D. tomentosa	0.61±0.005	0.10±0.001	0.14±0.002	0.30±0.004	3.04	1.47
	D. wallichi	0.22±0.003	0.02±0.003	0.15±0.012	0.10±0.002	5.07	2.10
Soaking in distilled water 9 h	*D. oppositifolia*	0.19±0.006 (-31)	0.08±0.002 (-48)	0.05±0.003 (-53)	0.24±0.002 (-23)	1.59	93.52
	D. pentaphylla	0.37±0.003 (-9)	0.01±0.001 (-82)	0.10±0.001 (-42)	0.28±0.002 (-15)	1.62	2.48
	D. tomentosa	0.53±0.001 (-14)	0.02±0.001 (-79)	0.08±0.001 (-43)	0.22±0.003 (-27)	2.86	1.38
	D. wallichi	0.09±0.002 (-54)	0.007±0.003 (-57)	0.09±0.010 (-38)	0.07±0.002 (-29)	4.95	1.91
Soaking in sodium bicarbonate solution 9 h	*D. oppositifolia*	0.15±0.005 (-46)	0.08±0.003 (-48)	0.01±0.001 (-90)	0.25±0.002 (-19)	1.56	93.60
	D. pentaphylla	0.29±0.003 (-29)	1.01±0.001 (-82)	0.10±0.003 (-41)	0.26±0.001 (-21)	1.61	2.48
	D. tomentosa	0.52±0.002 (-15)	0.01±0.002 (-88)	0.08±0.001 (-43)	0.21±0.001 (-31)	2.82	1.32
	D. wallichi	0.10±0.003 (-54)	0.005±0.002 (-70)	0.08±0.011 (-44)	0.08±0.001 (-21)	4.45	1.95
Cooking 90 min.	*D. oppositifolia*	0.05±0.001 (-82)*	0.06±0.002 (-61)*	0.009±0.002 (-90)	0.19±0.001 (-39)*	1.52**	93.61**
	D. pentaphylla	0.24±0.002 (-41)	0.008±0.002 (-84)	0.02±0.002 (-87)	0.20±0.001 (-39)	1.70	2.45
	D. tomentosa	0.44±0.005 (-28)	0.01±0.001 (-89)	0.05±0.002 (-63)	0.20±0.001 (-34)	2.91	1.38
	D. wallichi	0.03±0.001 (-86)	0.002±0.002 (-83)	0.05±0.004 (-67)	0.007±0.002 (-91)	4.97	2.00
Autoclaving 45 min.	*D. oppositifolia*	0.06±0.001 (-79)	0.05±0.000 (-69)	0.006±0.001 (-94)	0.18±0.000 (-42)	1.59**	93.58**
	D. pentaphylla	0.15±0.001 (-63)	0.06±0.002 (-87)	0.03±0.002 (-81)	0.21±0.002 (-36)	1.60**	2.41**
	D. tomentosa	0.32±0.007 (-47)	0.001±0.000 (-90)	0.02±0.001 (-85)	0.18±0.001 (-40)	2.98	1.35
	D. wallichi	0.03±0.002 (-86)	0.001±0.003 (-83)	0.01±0.001 (-93)	0.009±0.003 (-88)	4.91	1.98

a. values are mean of triplicate determinations expressed on dry weight basis. b. ± standard error; Values in the parenthesis indicate percent loss from raw samples. * significant at $P<0.01$; ** significant at $P<0.05$

The decrease in the content of total free phenolics and tannins during soaking may be due to leaching out of the phenolic substances in soaking under the influence of concentration gradient. A lot of HCN (known to inhibit the respiratory chain at the cytochrome oxidase level) is lost during soaking and cooking (Kay *et al.*, 1977) so that its content in the tubers poses no danger of toxicity. When the yam tubers were subjected to soaking, cooking for 90min and autoclaving for 45min significant reduction ($P<0.05$) has been observed in the levels of hydrogen cyanide and total oxalate. Inhibitors of alpha amylases and protein digesting enzymes interfere with the digestion of starch and protein. Hence, attempt has been made to eliminate these inhibitors. Boiling for sufficient time makes the tubers soft enough and inactivates all the trypsin inhibitor (Bradbury and Holloway, 1988).

CONCLUSION

Based on the nutritive evaluation studies on the wild edible yam tubers consumed by the tribal Kanikkars it can be summarized that most of them are found to be a good source of protein, lipid, crude fibre, starch and minerals. Various processing methods showed that autoclaving seems to be the best for the removal / inactivation of anti-nutritional factors as there is significant reduction ($P<0.05$) in amount of antinutrients thus improving digestibility. The potential of this crop has to compete with other alternatives under recognition of economical factors.

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