Short Communication

# Evaluation of phytonutrients and vitamin contents in a wild yam, *Dioscorea belophylla* (Prain) Haines

Poornima G.N. and Ravishankar Rai V.\*

Department of Studies in Applied Botany and Biotechnology, University of Mysore, Manasagagotri Mysore-570006 India.

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The species studied was found to contain bioactive compounds comprising saponins (18.46 mg  $100^{-1}$  g), alkaloids (0.68 mg  $100^{-1}$  g), flavonoids (8.84 mg  $100^{-1}$  g), tannins ( $4.2 \times 10^2$  mg  $100^{-1}$  g) and phenols ( $2.8 \times 10^3$  mg  $100^{-1}$  g). This yam contained vitamins such as ascorbic acid, riboflavin and thiamin. The importance of these chemical constituents is discussed with respect to the role of this *Dioscorea* species in herbal medicine.

Key words: Bioactive compounds, *Dioscorea*, herbal medicine, saponin, vitamins.

# INTRODUCTION

The yams (*Dioscorea* species) are the most important tuber crops in West Africa. They are among the root and tuber crops which are widely distributed through out the tropics with only a few members in the temperate regions of the world (Eka, 1998; FAO, 1985)

Yams are consumed as staple food. Apart from food, yams are mainly used for medicinal purposes for the sapogenins, aglycons of yam. Saponins are important mainly because of their steroid structure. They are precursors for the hemisynthesis of birth control pills (with progesterone and estrogen) as well as similar hormones and corticosteroids (Crabbe, 1979)

Yams like higher plants have a complex phytochemical profile. The most predominant phytochemical characteristic of yam is the presence of dioscorine alkaloid and diosgenin saponin. Although dioscorine and diosgenin are traditionally considered as toxic, such toxicity is removed by washing, boiling and cooking (Eka, 1998)

Some yam cultivars cannot be eaten raw because of itchiness, bitterness or toxicity of the raw tuber. The bitterness or acute toxicity in yams may be due to its alkaloid content while the saponins and sapogenins may constitute the pharmaceutical agent. This study investigates the fundamental scientific basis for the use of *Dioscorea belophylla* in herbal medicine. The contents of bioactive compounds and vitamins present in the species were determined.

## MATERIALS AND METHODS

## **Collection of plant material**

The tubers were collected from the Bisle Ghat region of Hassan District, Karnataka.

## Sample preparation

The tubers were washed and air dried. After drying, the samples were ground into powder and stored in airtight bottles before analysis.

#### Saponin determination

20 g of plant sample was dispersed in 200 ml of 20% ethanol. The suspension was heated over a hot water bath for 4 h with continuous stirring at about 55°C. The mixture was filtered and the residue re-extracted with another 200 ml of 20% ethanol. The combined extracts were reduced to 40 ml over water bath at about 90°C. The concentrate was transferred into a 250 ml separating funnel and 20 ml of diethyl ether was added and shaken vigorously. The aqueous layer was recovered while the ether layer was discarded. The purification process was repeated. 60 ml of n-butanol extracts were washed twice with 10 ml of 5% aqueous sodium chloride. The

<sup>\*</sup>Corresponding author. E-mail: rrai33@hotmail.com. Tel: 9845950155.

remaining solution was heated in a water bath. After evaporation the sample were dried in the oven into a constant weight. The saponin content was calculated in percentage (Nahapetian and Bassiri, 1975).

#### Alkaloid determination

5 g of the sample was weighed into a 250 ml beaker and 200 ml 20% acetic acid in ethanol was added and covered to stand for 4 h. this was filtered and the extract was concentrated using a water bath to one quarter of the original volume. Concentrated ammonium hydroxide was added drop wise to the extract until the precipitation was complete. The whole solution was allowed to settle and the precipitation was collected by filtration and weighed (Obadoni and Ochuko, 2001; Harborne, 1973).

#### Preparation of fat free sample

2 g of the plant sample was defatted with 100 ml of n-hexane using a soxhlet apparatus for 2 h.

#### **Determination of total phenols**

For the extraction of the phenolic component, the fat free sample was boiled with a 50 ml of ether for 14 min. 5 ml of the extract was pipette into a 50 ml flask, then 10 ml of distilled water was added. 2 ml of ammonium hydroxide solution and 5 ml of concentrated amyl alcohol were also added. The sample was made up to mark and left to react for 30 min for colour development. The absorbance of the solution was read using spectrophotometer at 505 nm wavelength (Obadoni and Ochuko, 2001; Harborne, 1973).

#### Flavonoid determination

10 g of the sample was extracted repeatedly with 100 ml of 80% aqueous methanol at room temperature. The whole solution was filtered through Whatmann filter paper No.1. The filtrate was later transferred into a crucible and evaporated to dryness over a water bath and weighed (Boham and Kocipai, 1994).

#### Tannin determination

500 mg of the sample was weighed into 100 ml bottle; 50 ml of distilled water was added and shaken for 1 h in a shaker. This was filtered into a 50 ml volumetric flask and made up to the mark. Then 5 ml of the filtrate was pipette out into a tube and mixed with 3 ml of 0.1 M FeCl<sub>3</sub> in 0.1 N HCl and 0.008 M potassium ferrocyanide. The absorbance was measured in a spectrophotometer at 120 nm wavelength within 10 min. A blank sample was prepared and read at the same wavelength. A standard was prepared using tannin acid to get 100 ppm and measured (Van-Burden and Robinton, 1981).

#### **Determination of thiamine**

50 g of the sample was homogenized with ethanolic sodium hydroxide (50 ml). It was filtered into a 100 ml flask. 10 ml of the filtrate was pipette and the colour was developed by addition of 10 ml of potassium dichromate and read at 360 nm. A blank sample was prepared and read at the sample wavelength.

#### Determination of riboflavin

5 g of the sample was extracted with 100 ml of 50% ethanol solution and shaken for 1 h. This was filtered into a 100 ml of the extract that was pipette into 50 ml volumetric flask. 10 ml of 5% potassium permanganate and 10 ml of 30% H<sub>2</sub>O<sub>2</sub> were added and allowed to stand over a hot water bath for about 30 min. 2 ml of 40% sodium sulphate was added. This was made up to 50 ml mark and the absorbance measured at 510 nm in a spectrophotometer.

#### Determination of ascorbic acid (vitamin C)

5 g of the sample was weighed in to a bottle and 100 ml of EDTA / TCA (2:1) extracting solution were mixed and the mixture shaken for 30 min. This was transferred into a centrifuge tube and centrifuged at 3000 rpm for about 20 min. It was transferred into a 100 ml volumetric flask and 1% starch indicator was added. These were added and titrated with 20% CuSO<sub>4</sub> solution to get a dark end point (Barakat et al., 1993).

# **RESULTS AND DISCUSSION**

The saponin and alkaloid content are considered important due to their toxicity in yam. These toxic metabolites occur in varying concentrations in yam tubers. *D. belophylla* has the saponin content of 18.46 mg  $100^{-1}$  g. A good amount of alkaloid was also found apart from saponins with 0.68 mg  $100^{-1}$  g. Other phytonutrients important in the pharmacological characteristics relating to yams include the flavonoids. 8.84 mg  $100^{-1}$  g of flavonoid was found. Tannins and phenolic compounds are in meager quantities than other phytonutrients determined in the yam tubers.

The tubers are relatively rich in ascorbic acid, riboflavin and thiamin. *D. belophylla* contained 1.67 mg  $100^{-1}$  g of ascorbic acid, 0.70 mg  $100^{-1}$  g of thiamin and 0.43 mg  $100^{-1}$  g of riboflavin.

Yams have been well respected by the herbalist community for generations due to their potency in enhancing fertility in males. This may be due to the presence of steroidal sapogenins such as diosgenin which have been isolated from yams. Diosgenin from yams have been used as precursors for the synthesis of hormones and corticosteroids which improve fertility in males (Crabbe, 1979; Oliver-Bever, 1989). It should be noted that toxic saponins are removed by washing the tubers before consumption (Eka, 1998).

Properties of saponins include formation of foams in aqueous solution, hemolytic activity and cholesterol binding properties and bitterness. Saponins natural tendency to ward off microbes makes them good candidates for treating fungal and yeast infections. These compounds served as natural antibiotics, which help the body to fight infections and microbial invasion (Sodipo et al., 2000). These compounds also appear to greatly enhance the effectiveness of certain vaccines. Plant saponins help humans to fight fungal infections, combat microbes and viruses, boost the effectiveness of certain vaccines and knock out some kinds of tumor cells, particularly lung and blood cancers (Barakat et al., 1993). They also lower blood cholesterol there by reducing heart disease. The most outstanding and exciting prospects for saponins are how they inhibit or kill cancer cells. They may also be able to do it without destroying normal cells on the process, as is the mode of some cancer fighting drugs. Cancer cells have a more cholesterol type compounds on their membranes than normal cells. Saponins therefore bind cholesterol and thus interfere with cells growth and division (Ryam and Shattuck, 1994).

Alkaloids in *Dioscore*a species have been reported to contain dihydrodioscorine. This compound is a convulsant alkaloid and it causes central nervous system parlaysis in animals. An extract of dihydrodioscorine produces a long lasting hypotension and contraction of the smooth muscle fibers of the intestine both *in vivo* and *in vitro* when administered to animals (Oliver-Bever, 1989). This explains the use of *Dioscorea* species for the preparation of poison bit for fishing, hunting and preparation of insecticides.

Flavonoids are widely distributed group of polyphenolic compounds, characterized by a common benzopyrone ring structure, that have been reported to act as antioxidants in various biological systems. The biological functions of flavonoids apart from its antioxidant properties include protection against allergies, inflammation, free radicals, platelet aggregation, microbes, ulcers, hepatoxins, viruses and tumors (Barakat et al., 1993, Okwu and Omodamiro, 2005). Flavonoids reduced cancers by interfering with the enzymes that produce estrogen for e.g. Flavonoids inhibit estrogen synthesase, an enzyme that binds estrogen to receptors in several organs (Okwu and Omodamiro, 2005; Farquer, 1996). Some flavonoids behave as a powerful protective agent against inflammatory disorders. They reduce edema formation and prevent platelet stickiness and hence platelet aggregation.

The trace quantities of phenolic compounds help to prevent the death of the crop, since phenolic compounds from plant extracts act as antimicrobial agent (Ofokansi et al., 2005). In some species of yam tubers, browning reactions occur when the tissues are injured and exposed to air. This type of browning is due to the oxidation of phenolic constituents, especially o-hydroxy or trihydroxy phenolics, by a phenol oxidase present in the tissue (Martin and Rubeste, 1976). The presence of phenols (Farquer, 1996) indicates that *Dioscorea* species could act as antiinflammatory, anti-clotting, antioxidant, immune enhancers and hormone modulators (Okwu and Omodamiro, 2005).

The bitter principle may be due to the presence of tannins in them. The trace quantities of tannin available in yam tubers act as a repellant against rot in yam.

Ascorbic acid activates the functions of all the cells. It is a powerful antioxidant. It favours the absorption of iron in the intestine, protects against infections. Neutralizes blood toxins and intervenes in the healing of wounds

# (Roger, 1999).

It is significant to note that *Dioscorea* species, which have been regarded as non-edible food by people as a result of culture, religion, belief from folk stories or due to chemical constituents of such yams have been from this study to be edible. These yams not only contain vitamins but also phytonutrients that help to fight against most diseases of man.

*Dioscorea* species contain important nutritive health promoting substances, which prompted their use as food and drug in herbal medicine.

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