Garcinia bark exudates – an important phytochemical source

Exudates are a complex mixture of organic compounds that ooze out of plants through pores, or as a result of injury/wound. These products are commonly called as 'sap' and are rich in carbon and hydrogen atoms. A flow of viscous liquid or bleeding from the underlying tissues (when living) of the bark, consists largely of gum, resin or latex, depending on the tree species[1]. These plant products have been collected since about 3000 BC, during the Egyptian civilization from *Acacia*[2].

The importance of products that are obtained from renewable sources is increasing day by day, as the safety of the synthetic pigments, food colour, latex, etc. has generated several controversial questions over the past decades. Consumer awareness has added significantly to the demand for natural products. Among the most commonly used natural polymers are the gums obtained from trees, also known as tree exudates. These are characterized by low toxicity, abundant availability and biocompatibility, are biodegradable, inert, and cheap compared to synthetic polymers[3].

Gum arabica, for example, is an *Acacia* tree exudate widely used in the food and drug industries[4]. Many plants of the genus *Garcinia* exude viscous gummy liquids through cuts in their bark. These exudates are white to yellow translucent masses, which get solidified when exposed to air.

Garcinia belonging to the family Clusiaceae are tropical evergreen trees, found in the Western Ghats, Andaman and Nicobar Islands, and NE Himalayas in India. *Garcinia* fresh and dry rind (exocarp) is used as a spice, condiment, garnish in several cuisines and also for preparing syrups and other food substance[5]. It is valued for the presence of (-)hydroxycitric acid, an anti-obesity compound in the fruit rind and leaves of several species.

For centuries, the dried *Garcinia* bark exudates known as gamboge are being used in the paint and lacquer industries and varnishes[6]. It is also used to prevent corrosion of metals from acids[7,8].

The dried exudates are used as a pigment in Indian murals and European water-paintings and dyeing clothes[9] and also for colouring wood, metal and leather. Gamboge is commonly extracted by tapping from the matured trees of *G. haburyi*, *G. morella* (India and Sri Lanka), and *G. elliptica*, *G. heterandra* (Myanmar).

The chemical composition and physical properties of such commercially important gums had to be extensively studied. Glicksman[4] studied the same two *Acacia* species, namely *A. xanthophloea* and *A. tortilis*. The present study deals with extraction and chemical composition of the bark exudates of three Garcinia species, namely *G. indica*, *G. gummigutta* and *G. xanthochymus*.

The barks were excised from the trunk of mature trees with a cut of a dimension of about 5–8 cm × 2–5 cm (Figure 1). The exudates were collected in clean vials, taped near the wound site. The liquid form solidifies into dry tears or irregular masses when in contact with air. The 10 g of exudates was taken in a measuring flask and 10 ml of ethyl acetate was added. The contents were mixed thoroughly and then ethyl acetate fraction was transferred to a clean beaker. The process was repeated twice so that resins, which are soluble in ethyl acetate, are extracted along with the solvent. The gum part remains as insoluble residue in the flask[10,11].

Resin content in the exudates was determined gravimetrically. The ethyl acetate extract was transferred to a preweighed evaporating dish. Under a fume hood, the ethyl acetate was then allowed to fade slowly. Once the solvent was completely removed, the dishes were weighed again. The difference between the initial and final weights gives the amount of resin present in the exudates[10].

Gum content was determined as total carbohydrates[12]. The insoluble residue obtained was then dissolved in water by gentle heating and thorough mixing. The carbohydrate (total sugar) content was estimated using anthrone reagent as described by Fairbain[13]. In this procedure, the carbohydrate moieties in the gum react with anthrone (9-oxo anthracene) in the presence of H_2SO_4 to form a green-coloured complex. The intensity of the colour formed is proportional to the carbohydrate content, which is measured in a spectrophotometer at 580 nm.

The amount of total phenols in the bark exudates of *Garcinia* was determined by Folin–Ciocalteau method[14]. The total flavonoids content of *Garcinia* bark exudates was determined by aluminium chloride method[15]. Xanthones from the exudates were studied by UV absorption method[16] at 243 nm wavelength in a UV-Vis spectrophotometer. The free-radical scavenging activity (antioxidant activity) of the extract was measured using 1,1-diphenyl-2-picrylhydrazyl (DPPH) as a free-radical model and the method of Magalhães *et al.*[17]. DPPH is a nitrogen-centred free radical, the colour of which changes from violet to yellow on reduction by the antioxidant compound present in the sample. Absorbance of the mixture was measured spectrophotometrically at 517 nm, and the free-radical scavenging activity was calculated by plotting the scavenging percentage of all samples. The final result was expressed as an IC_{50} value (the concentration of sample producing 50% scavenging of the DPPH radical (μg/ml))[18].

Figure 1. Extraction of *Garcinia gummigutta* bark exudates.

Figure 2. Dry exudates.

Table 1. Biochemical composition of exudates

Exudate parts	*G. gummigutta* (%)	*G. indica* (%)	*G. xanthochymus* (%)
Resin	68.3	60.4	40.0
Total sugars	14.2	20.3	35.1
Insoluble part	17.5	19.3	24.9

Table 2. Biochemical composition of resin part of the exudates

Biochemical content	*G. gummigutta*	*G. indica*	*G. xanthochymus*
Total phenol (g/100 g)	56.37	53.43	67.13
Total flavonoids (g/100 g)	16.64	18.80	37.61
Xanthone content (g/100 g)	35.57	32.42	20.12
Antioxidant activity (IC_{50} µg/ml)	20.4	18.2	21.7

Fresh exudates were sparingly soluble in water, but it became insoluble when dry. Colour varied from white (*G. xanthochymus*) to deep yellow (*G. gummigutta*). If kept in the open, it forms an opaque, brown, solid, irregular mass.

Biochemical composition of the exudates showed that resin percentage is high in *G. gummigutta* (68%), it is 60.4% in *G. indica*, but less in *G. xanthochymus* (40%; Table 1).

The total sugar percentage (carbohydrate) is high in *G. xanthochymus* (35.1%) and the colour of exudates is also white, while it is lowest in *G. gummigutta* only 14.2% and in *G. indica* it is 20.3%. The fresh colour of the exudates is yellowish-orange in *G. gummigutta* and in *G. indica* (Figure 2).

The resin is smooth, conchoidal fracture of a waxy lustre and orange-red in colour. The powder is bright yellow and sometimes used as drug adhesive, for yellow appearance of the drug. Taste is mild but acrid; odour is irritating. The cake or lump of gamboge is sold in masses weighing two or three pounds. The mass is less uniform, less brittle, and is called as 'coarse gamboge'. Pure gamboge is completely soluble by successive treatment with ether or alcohol and then water.

Total phenol percentage is high; it is 56.37, 53.43 and 67.13 g/100 g respectively, in *G. gummigutta*, *G. indica* and *G. xanthochymus*. Flavonoids (a class of plant secondaty metabolites) are highest in *G. xanthochymus* while they are low in *G. gummigutta* and *G. indica*. The name of these phyto-nutrients is actually derived from their colour-related chemistry; the Latin word *flavus* meaning 'yellow'[19].

Xanthones are also phyto-nutrients. Apart from the rich nutritional value, the phyto-nutrients are antioxidants as well. Xanthon content is considerably high in *G. gummigutta* and *G. indica* (35.57 and 32.42 g/100 g respectively) in *G. xanthochymus*, it is 20.12 g/100 g. Xanthones are a class of polyphenolic compounds that commonly occur in plants and have been shown to have extensive biological and pharmacological activities[20].

The present study indicates that *Garcinia* exudates contain useful phytochemicals. Bio-affinity as a tool for the study of useful biological compositions and valuable nutrients is gaining increasing importance in the present era. Alternative products are being obtained from renewable sources, such as the bark exudates, gum, bark, flower and leaf of plants which grow wild in many tropical and subtropical countries, which are underutilized but are highly valuable. Lewington[9] reported that *Garcinia* exudates were used to dye the robes of Buddhist monks. In the pigment industry, this powder is ground or mixed with a variety of binders in order to make paints and varnishes[21]. In ancient India, gamboge had an important place among artists, herbalists and spiritual communities[22]. Prasarset[23] reported its use in Thailand mural paintings. It is used in traditional medicine for the treatment of ulcers, skin infection, appetite suppression and to lower blood pressure[24]. As it has antioxidant properties and high flavonoid contents, it can be used as food and drug colour. Though there are records of its use in pigements and in other colouring materials, this study shows that it is harmless and contains all high-valued ingredients for using it in any food or drug industry.

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Why should we preserve wetlands?

Wetlands are transitional zones between terrestrial and aquatic systems, and remain inundated or saturated due to high groundwater or surface water during a part or all through the year[1]. Wetlands in different parts of the world have been used for agriculture because of their natural fertility and water availability[2]. Livelihood, food security, income and nutrition of the people living in and around the wetlands in Asia and Africa are strongly affected by their management. Wetlands are amongst the most environmentally sustainable systems, but produce low yield due to traditional systems of management[3]. Therefore, prudential intensification of wetland agriculture in the absence of holistic approach has disintegrated wetland ecosystem services along with adverse impacts on environment quality[4]. Moreover, indiscriminate and intensive use of wetlands, without considering the preservation of ecological integrity has converted a large pedologic soil organic carbon (SOC) sink into a net source[5]. Therefore, it is important to document the SOC pool of wetlands under changing climate, because carbon management in any terrestrial pool is one of the priority actions of national and international policy goals. Keeping this in view, the present study was undertaken in the Chatla wetland (90°45′N and 24°45′E) of Barak Valley, North East India, with an objective to explore its standing organic carbon stock. Chatla is the catchment of River Ghagra, the tributary of River Barak. The topography of the area is low-lying with numerous small hillocks in between that are inhabited by the villagers. The geographical area of Chatla is ~10 km^2 (ref. 6). The major ethnic group is the 'Kaivartas', a fisher community. Paddy cultivation is the primary farming system. To achieve the desired goal of estimating SOC stock of Chatla wetland, soil samples were collected from three principal eco-zones of wetlands, viz. (i) littoral (interface between land and water basin), (ii) sub-littoral (shallow water zone) and (iii) deep water zone during the winter season (January 2014). However, during winter months deep water zone was inundated. Therefore, for this zone, square-sized mudden boundary was prepared to remove the water. After drying, soil samples were collected up to a depth of 1 m from three strata, viz. 0–10, 10–30 and 30–100 cm. Three pits were dug in each zone to collect soil samples and average of the three zones was used as the representative SOC value for the wetland. The SOC concentration was determined by Walkley and Black's rapid titration method[7]. The SOC stock (Mg ha^{-1}) of each eco-zone was computed following the method of Blanco-Canqui and Lal[8]. The study revealed that the magnitude of SOC stock was in the order deep water > sub-littoral > littoral zone (Figure 1). Higher SOC stock in deep water can be attributed to permanent high water table, which slows down organic matter decomposition and allows accumulation of more organic carbon[9]. In the case of littoral and sub-littoral zones, they shrink as they dry and swell as they become moist, creating deep and wide cracks that potentially enhance organic matter oxidation, leading to loss of SOC from such eco-zones[10]. Total SOC stock of the wetland was 220 Mg ha^{-1}, which is higher than any tropical land uses (Table 1)[11–17]. Hence, preserving wetlands is important, so that the carbon stored is not released to the atmosphere. However, such a large SOC pool has been disintegrated through intensification of agricultural practices. It has been estimated that ~70% of total global wetland area and almost similar amount of SOC have been lost since the industrial

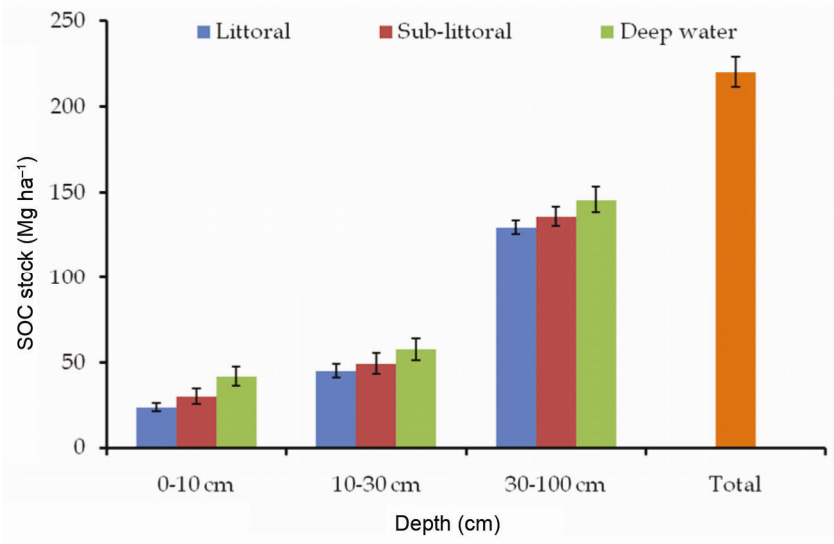

Figure 1. Soil organic carbon (SOC) stocks of different eco-zones of the Chatla wetland, Barak Valley, North East India. Line on each bar represents standard error of the mean. Bar on total represents the standard error of mean calculated from SOC stocks (0–100 cm) of littoral, sub-littoral and deep water zones.