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Effect of gamma irradiation on physicochemical properties of Indian Horse Chestnut (*Aesculus indica* Colebr.) starch

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ABSTRACT

Starch isolated from Indian Horse Chestnut (*Aesculus indica* Colebr.) was subject to irradiation at 0, 5, 10, 15 kGy doses. Effect of irradiation on physicochemical properties of native starch was studied. The result revealed increase in water absorption capacity from 0.94 to 1.00 g/g, carboxyl content from 0.00 to 0.06%, solubility from 0.15 to 0.53 g/g and freeze thaw stability. Syneresis, pasting properties and pH were reduced following irradiation treatment. Syneresis decreased from 3.47 to 0.64% after 120 h refrigerated storage. Peak viscosity reduced from 5156.5 to 1422.5 cP, setback viscosity from 1191.5 to 73.0 cP and final viscosity from 3232.0 to 410.5 cP. X-ray diffraction pattern showed A type of pattern in native as well as irradiated starches. Granule morphology of native and irradiated starches under scanning electron microscope revealed that granules were round, oval, irregular or elliptical with smooth surfaces. Pearson correlation studies revealed that irradiation dose was positively correlated with water absorption capacity, oil absorption capacity, and solubility index and negatively correlated with syneresis, swelling index, freeze thaw and pasting properties.

1. Introduction

Indian Horse Chestnut – IHCN (*Aesculus indica*) locally known as *Han dun* is found in temperate regions of Asia with elevations varying from 900 to 3600 m. In Asia, it is generally found in India, Nepal, Pakistan and Afghanistan. In India, the tree is found in rich, moist, shady ravines of Jammu and Kashmir, Himachal Pradesh and Uttar Pradesh (Singh, 2006; Zhang, Li, & Lian, 2010). It is present in planes as well as in hilly areas of Jammu and Kashmir. It is a large deciduous tree with an average height of 22.5 m and has a straight cylindrical bole with spreading crown. It produces multi-colored blossoms during May and June, which on fertilization yield seeds and it yields huge quantity of seeds every year. Seeds are consumed by wild animals namely, Kashmir stag (*Hangul*) in hilly areas of Kashmir. However, in plains the seeds mostly go waste.

Seed are present in the capsule and each capsule contains single seed. Seeds are about 3.5 cm in diameter with shining black hard rind from outside and lime white cotyledons inside (Parmar & Kaushal, 1982). Seeds ripen in October and these are rich in poisonous saponins. Saponins are poorly absorbed by human body and so pass through alimentary canal without harm. They can be removed by carefully leaching the seeds or flour in running water. Seeds possess therapeutic importance, e.g. in the treatment of fevers, for use in curing piles, wound healing, anti-inflammatory, antiviral, rheumatism, skin diseases and cardiovascular diseases (Kaul, 1997; Kaur, Joseph, & George, 2011). Seeds have also edible uses. They have been used as food during the times of famine by various tribes of North and North Eastern India. The seeds can be ground into powder and used as gruel (Singh & Kachroo, 1976). The seeds which constitute the edible portion of fruit contain 50.5% moisture, 5.85%, sugars, 0.39% protein, 1.93% ash (Parmar & Kaushal, 1982) and about 38.3% starch on dry weight basis (Singh, Katoch, Ram, & Aijaz, 2003). So starch is the major component of Indian Horse Chestnut seeds.

Starch is one of the most important functional biopolymers. The functional roles of starch in food products could be as a thickener, binding agent, emulsifier, clouding agent, or gelling agent. Starch is composed of two different glucan chains, amylose and amylopectin. These polymers have the same basic structure, but differ in their length and degree of branching, which ultimately affects the physicochemical properties. Native starches have certain limitations like low shear resistance, high

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retrogradation and syneresis thus, limiting their industrial use (Betancur & Chel, 1997). Therefore, starch is often modified by physical, chemical and enzymatic methods or a combination thereof, in order to obtain desirable functional properties for certain uses (Liu, Ramsden, & Corke, 1997). The most used way to obtain modified starch is by chemical methods which are usually complex, expensive and time consuming. Physical modification involving gamma irradiation are fast, low cost and environmentally friendly because they do not use pollutant agents, do not allow the penetration of some toxic substances in the treated products and do not generate undesirable residual products, do not require catalysts and laborious preparation of sample. Gamma irradiation is capable of hydrolyzing chemical bonds, thereby cleaving large molecules of starch into smaller fragments of dextrin. These changes may affect the physicochemical properties of irradiated foods, resulting in increased solubility of starch, decreased swelling power and viscosity of starch paste (Kang et al., 1999; Wu, Shu, Wang, & Xia, 2002). Since, no systematic study has been done on the isolation and modification of Indian Horse Chestnut starch, the aim of the present study was to evaluate the effect of gamma irradiation on physicochemical properties of starch isolated from the seeds of Indian Horse Chestnut.

2. Material & methods

2.1. Materials

The seeds of Indian Horse Chestnut were harvested from the trees located in the main campus of University of Kashmir, Hazratbal, Srinagar–190 006, India during the month of November, 2011. Seeds were dehulled and stored at 5 °C until further use. All the reagents used in the study were of analytical grade.

2.2. Starch isolation

Seeds were manually deshelled and the cotyledons were chopped into small pieces of approximately 2 × 2 cm. The pieces were pulverized along with water for 5 min in a domestic mixer blender. The slurry obtained was then diluted to ten times (volume/volume) with distilled water, and the pH was adjusted to 9 using 0.5 m NaOH. The slurry was continuously mixed using magnetic stirrer for 1 h and then filtered through a 75 μm-mesh sieve to separate the fiber. The filtered slurry was then centrifuged at 3000× *g* for 30 min at 10 °C (5810R, Eppendorf, Hamburg, Germany). The aqueous phase obtained on centrifugation was discarded, whereas the sediment obtained was scraped off from the surface and the lower white portion was washed three times with double distilled water and recovered as starch. The starch was dried at 40 °C in a hot air oven (NSW-143; Narang Scientific Works Pvt. Ltd., New Delhi, India).

2.3. Starch irradiation

The starch samples were packed in two layers of polyethylene bags and were irradiated in a cobalt-60 (^{60}Co) source irradiator at room temperature (20 ± 2 °C). Samples were irradiated with absorbed doses of 0 (unirradiated), 5, 10 and 15 k Gy at the dose rate of 83 Gy/min. All the starch materials were kept at 20–40% relative humidity under 0.1 M Pa pressure. The irradiation treatments were performed at the Inter University Accelerator Center, New Delhi, India. Irradiations were performed in duplicate.

2.4. Physico-chemical properties

2.4.1. Composition

Moisture (925.10), protein (920.87), fat (920.85) and ash (923.03) contents were determined according to the methods of AOAC (1990).

2.4.2. Color

Color of the starch was determined using Color Flex Spectrocolorimeter (Hunter Lab Colorimeter D-25, Hunter Associates Laboratory, Ruston, USA) after being standardized using Hunter Lab color standards and their Hunter *L* (lightness), *a* (redness to greenness) and *b* (yellowness to blueness) values were measured.

2.4.3. Apparent amylose content

Apparent amylose contents of the starch samples were determined by the method of Williams, Kuzina, & Hlynka (1970). Starch sample (20 mg) was taken, 10 mL of 0.5 M KOH was added and the suspension was mixed thoroughly. The dispersed sample was transferred to a 100 mL volumetric flask and the volume was made up to the mark with distilled water. An aliquot of the test starch solution (10 mL) was pipetted into a 50 mL volumetric flask and 5 mL of 0.1 M aqueous HCl was added followed by 0.5 mL of iodine reagent. The volume was diluted to 50 mL and allowed to stand for 5 min. The absorbance was measured at 625 nm (UV Spectrophotometer, U-2900, Hitachi, Tokyo, Japan). The content of amylose was determined from a standard curve developed using standard amylose and amylopectin blends from potato starch.

2.4.4. pH of starch suspension

The pH of each starch suspension was determined using a digital pH meter (Hanna, USA). Starch samples for pH measurements were prepared by suspending 1 g of starch in 25 mL of water at 25 °C and agitating for 5–10 min (Sokhey & Chinnaswamy, 1993).

2.4.5. Carboxyl content

The carboxyl content of oxidized starch was determined according to the procedure of Chattopadhyay, Singhal, and Kulkarni (1997).

2.4.6. Water and oil absorption capacity

2.5 g starch on dry weight basis (db) was mixed with 20 mL distilled water or mustard oil and then stirred for 30 min at 25 °C. The slurry was then centrifuged at 3000× *g* for 10 min (5810R, Eppendorf, Hamburg, Germany) and the supernatant was decanted. The gain in weight was expressed as percentage of water/oil absorption capacity.

2.4.7. Bulk density

Bulk density was measured according to the method of Wani, Sogi, and Gill (2013). A 10-mL graduated cylinder previously tarred was gently filled up to the 10 mL mark with starch. The sample was then packed by gently tapping the cylinder on the bench top from a height of 5 cm until there was no further diminution of the sample level. The weight of the filled cylinder was taken, and the bulk density was calculated as the weight of sample per unit volume of sample (g/mL).

2.4.8. Swelling & solubility index

Starch sample (0.2 g db) were taken in pre-weighed centrifuge tubes with 10 mL of distilled water. The starch suspensions were then incubated in a water bath for 30 min at 50, 60, 70, 80 and 90 °C with vortexing after every 5 min. After cooling the samples to room temperature, the tubes were centrifuged at 5000× *g* for 15 min. Supernatant was decanted in pre weighed moisture dishes. The