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Pearl Millet Starch Films: A Comparative Study on Characteristics from Different Cultivars

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Abstract: The present study focuses on the use of underutilized coarse grain Pearl millet for the development of biodegradable films. Starch from two varieties of Pearl millet viz. HHB 67 and ProAgro 9444 was isolated and subjected to casting technique of film formation. Glycerol was used as plasticizer. Amylose content of HHB 67 and ProAgro 9444 was 15.05% and 20.21%, respectively. Both the varieties resulted in homogeneous films but ProAgro 9444 offered better films in terms of mechanical strength and other characteristic parameters owing to the high amylose content as compared to HHB 67. This study is crucial for moving towards biodegradable plastics because of the arising environmental concerns.

 Research Paper

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INTRODUCTION

Over the past three decades, considerable number of studies were carried out to eradicate the problem arising from the plastic or synthetic packaging, thus moving a step towards the biodegradable packaging systems [1]. The use of natural biopolymers such as polysaccharides, proteins and lipids have been explored as structural matrix materials for film formation and classified accordingly [2]. Among polysaccharides, starch, owing to its biodegradability, cost effectiveness, abundance and other advantages is a fascinating choice to act as a matrix forming material. Starch consists primarily of amylose and amylopectin, of these, amylopectin is branched and amylose is linear. Amylose has a smaller molecular weight, and amylopectin has a higher molecular weight. They are organized in alternating crystalline and amorphous lamellae in the granules. Amylose is in charge of the film-forming ability [3].

Pearl millet (*Pennisetum Glaucum*) is an underutilized coarse cereal grain and is included in the category of major millets. After rice and wheat it is the third most cultivated crop in India. Pearl millet accounts for 50% of the global millet production. In addition to this, India is the largest producer of Pearl millet in World [4]. Major producers of this crop in India are Rajasthan, Gujarat and Maharashtra. Owing to its short growing season and drought-resistance Pearl millet has an upper hand over the other major cereals [5].

Fundamentally this crop is grown for food and dry fodder [6], whereas it could act as a fascinating source of starch because of availability of different improved and hybrid varieties. Moreover, starch content of Pearl millet varies from 62.8 to 70.5% depending upon the variety.

The objective of the present study was to explore this underutilized grain for film forming capabilities of starch extracted from different varieties. The investigation also focused on mechanical characterization of the films developed thereof.

MATERIALS AND METHODS Materials

For starch isolation, two varieties of Pearl millet grains viz. HHB 67 and ProAgro 9444 were procured from M/s. Chaudhary Enterprises, Hisar (Haryana). Glycerol and other chemicals of analytical grade were obtained from Rankem, Mumbai (Maharashtra).

Methods

Extraction of starch

Starch from both varieties was extracted using wet milling approach. Pearl millet grains were soaked in distilled water containing 0.25% Potassium metabisulfite and 0.25 % citric acid for 24 hrs at room temperature (27°C). Then the grains were pulverized using a blender and screened through 100, 200 and 300 BSS mesh sieves. The slurry obtained thereafter was washed several times with distilled water and centrifuged at 5000 rpm for 15min. The upper layer was removed with a spatula. Starch was dried at 45°C for 24 hrs and stored in air tight containers for further analyses.

Amylose Content

Amylose content of starch was determined using the method of Juliano et al. [7] with slight modifications. 100mg starch from each variety was added to 100ml volumetric flask and then 1ml 95% Ethanol and 9ml 1N NaOH was added to it. After boiling for 10 min and cooling, volume was made upto 100ml. A sample of 5ml from the mixed solution was taken in another 100ml volumetric flask, 1ml of 1N Acetic acid and 2ml of iodide solution was added and made up the volume to 100ml. After 20minutes absorbance was recorded at 620 nm. The amylose content was calculated against the standard curve prepared from amylose' standard.

Preparation of Films

Casting technique was used for film formation. 5% starch and 2.5% glycerol were dispersed in distilled water and heated in water bath at 95°C for 30 minutes

with gentle and continuous stirring. Then the film forming solutions were poured in glass petri-plates and dried at 40°C for 12 hours. After drying, the dried films were peeled off and conditioned before carrying out further analysis at 53% RH at 25°C, maintained in desiccators using Magnesium Nitrate saturated solution.

Moisture content, Thickness and Solubility

Thickness of the dried films was evaluated at five random locations of films, using an analog micrometer (Mitutoyo-2046F) with an accuracy of ± 1 µm. Moisture content was determined gravimetrically in a hot air oven at 110°C and weight of films was observed at regular intervals until it got constant. Solubility of films was determined by method of Romero Bastida et al. [8]

Mechanical Properties

Tensile strength and Percentage elongation were measured using the reference method [S] with the help of Texture analyzer (Stable Micro Systems, Surrey, UK). The film width for the analysis was kept 24mm for all the samples.



Figure 2: A Texture Analyzer measuring mechanical properties of films

Water Vapor Permeability

Water vapor permeability of films was determined by ASTM E96 (2015) [9] method with slight modifications. Circular film samples were placed on cup mouth and sealed. The cup was previously filled to threefourth its volume with anhydrous calcium chloride. The cups were placed in desiccators at 25°C containing saturated sodium chloride solution (75% RH). After regular intervals (2hrs), weight of cups was recorded. Weight change was plotted against time and from the Linear Regression curve, the slope was calculated as the Water Vapor Transmission Rate (WVTR). WVP was calculated using the formula:

$$WVP = \frac{WVTR}{S\left(R_1 - R_2\right)} \times d$$

Where, WVP was water vapor permeability (g.m/Pa.s.m²), S, the saturation vapor pressure of water (Pa) at the test temperature 25°C, R_1 , the percent relative

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humidity in desiccators, R_2 , the percent relative humidity in cup and 'd' is the film thickness (m).

Statistical Evaluation

Statistica-log software version 7 (M/s StatSoft Inc. USA) was used for all the statistical analysis. All the analyses were determined in triplicates. Significant differences were obtained by using One-way ANOVA followed by the Duncan's multiple range test at significance level of <0.05.

RESULTS AND DISCUSSION

Yield and amylose content of starch

Starch yield and amylose content of both varieties is shown in Table 1. Significant variation ($p \le 0.05$) was observed between the varieties. The difference in yield may be accounted to varietal difference. Amylose content has significant effects on mechanical properties of films [10]. Botanical origin, Growth and harvesting conditions and other environmental constraints affects the variability in amylose content within the same species [11].

Table 1. Starch	vield and	amvlose	content fron	n two varieties	of Pearl millet
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Variety	Starch yield (%)	Amylose content (%)
HHB 67	$41.23^{\text{a}} \pm 0.09$	$15.05^a\pm0.18$
ProAgro 9444	$45.66^b\pm0.16$	$20.21^b\pm0.08$

Mean values in the same column with different superscript differ significantly ($p\leq0.05$). Mean \pm standard deviation are from triplicate readings.

Moisture content, thickness and solubility

Starch films from both the varieties were translucent in appearance. Moisture content, thickness and solubility are listed in Table 2. Moisture content of films from ProAgro 9444 was lower than HHB 67. This may be attributed to higher amylose content in former variety than HHB 67. Similar trends have been observed for Elephant Foot yam and Lotus starch films by Sukhija et al. [12]. There was no significant difference in

thickness of films for both the varieties. Water solubility of films from ProAgro 9444 was significantly lower than the other variety. As the plasticizer content was same in film formulations so the difference in solubility may be due to difference in amylose content. From this we can conclude that starch network is stronger in films developed from ProAgro 9444 variety due to interactions between amylose molecules. Similar trends have been reported for Pea and Rice starch by Mehyar et al. [13]



Figure 1: Films formed from a) ProAgro 9444 b) HHB 67

Table 2: Parameters calculated for films from HHB 67 and ProAgro 9444 variety of Pearl millet

Parameter	HHB 67	ProAgro 9444
Moisture content (%)	24.23 ^a ±0.08	23.18 ^b ±0.04
Solubility (%)	38.12 ^a ±0.12	45.71 ^b ±0.06
Thickness (mm)	22±0.01	21±0.02
WVP (x 10 ¹¹) (g/Pa.s.m)	6.07 ^a ±0.12	7.63 ^b ±0.05
Tensile strength (MPa)	$245^{a}+010$	$3.90^{b}+0.12$

Mean values in the same row with different superscript differ significantly ($p \le 0.05$). Mean \pm standard deviation are from triplicate readings.

Water vapor permeability

Water vapor permeability was high for the starch films than the synthetic counterparts such as LDPE films [14], this may be due to hygroscopic nature of starch polymeric network. But among the varieties, films from HHB 67 presented a higher WVP than ProAgro 9444 (Table 2). The reason behind this could be

the lower percentage of amylose in the former. Similar trends have been observed by Stading et al [15].

Tensile strength and percent elongation

Tensile strength of the films was significantly higher for the variety ProAgro 9444 than HHB 67 (Table 2). Due to higher percentage of amylopectin in HHB 67,

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the films formed were frail because of higher degree of entanglement arising from extensive branching. However, in case of ProAgro 9444 variety films, the phase separation was possibly prevented by the amylose gelation resulting in films with comparatively better mechanical properties [16-18]

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