Final Project Completion Report (PCR)

Project Title: Development of enzyme based extraction process for improving quality and recovery of starch from different varieties of *Colocasia esculenta* (Arbi) of Assam for food use

File No. 10/MFPI/R&D/2011

Submitted to

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Submitted by

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12.5.16.

ANNEXURE-C

FINAL PROJECT COMPLETION REPORT

1. **Title of the project:** Development of enzyme based extraction process for improving quality and recovery of starch from different varieties of *Colocasia esculenta* (Arbi) of Assam for food use

2. **Principal Investigator(s) and Co-Investigator(s):** Dr. Nandan Sit (P.I.) and Dr. Laxmikant S. Badwaik (Co P.I.)

3. **Implementing Institution(s) and other collaborating Institution(s):** Department of Food Engineering and Technology, Tezpur University, Assam

4. **Date of commencement:** 28/12/2011

5. **Planned date of completion:** 27/12/2013

6. **Actual date of completion:** 15/03/2015

7. **Objectives as stated in the project proposal:**
    - To examine the physico-chemical characteristics of starch from different varieties of *Colocasia esculenta* (Arbi) available in Assam.
    - To identify the varieties suitable for starch extraction for food use based on the physico-chemical characteristics.
    - To study the application of these starch in preparation of different food products.
    - To optimize the enzymatic starch extraction process using pure enzymes as well as crude enzymes extracted from microorganisms.
    - To compare the properties of the starch extracted by enzymatic process with that of conventional process

8. **Deviation made from original objectives if any, while implementing the project and reasons thereof:**

    All the objectives have been successfully completed.

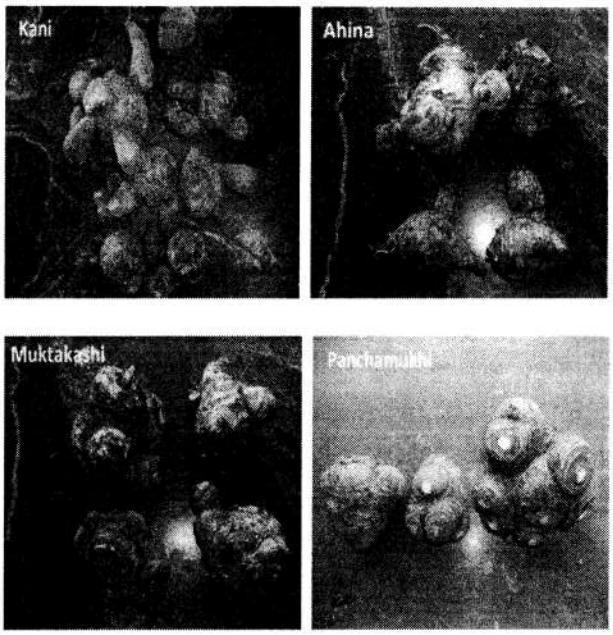
9. **Experimental work giving full details of experimental set up, methods**

adopted, data collected supported by necessary table, charts. diagrams & photographs:

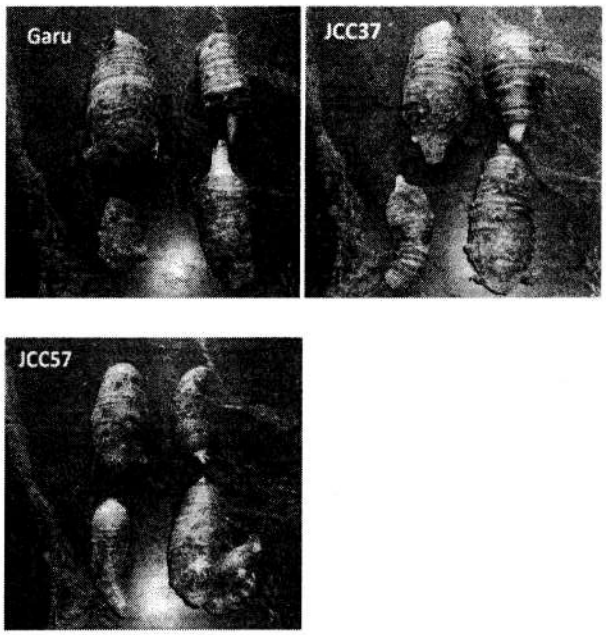
9.1 Physico-chemical characteristics of starch from different varieties of *Colocasia esculenta* (Arbi) available in Assam, its comparison to starches from other sources and identification of the variety suitable for food use

9.1.1 Sample collection

Tubers of seven cultivars of taro were used in the present investigation. The cultivars collected were: *Kani*, *Ahina*, *Muktakashi*, *Panchamukhi*, *Garu*, *JCC37* and *JCC57*. Among the seven cultivars *Kani*, *Ahina*, *Muktakashi* and *Panchamukhi* were "eddoe" type taro i.e. *Colocasia esculenta* var. *antiquorum*, and *Garu*, *JCC37* and *JCC57* were "dasheen" type taro i.e. *Colocasia esculenta* var. *esculenta* (Fig. 2.1). *Ahina*, *Muktakashi*, *Garu*, *JCC37* and *JCC57* were obtained from research farm of Assam Agricultural University, Assam, India, while *Kani* and *Panchamukhi* were collected from a local farm near Tezpur University, Assam, India. Freshly harvested tubers were collected after 8 months from cultivation. Starches from potato variety *Chandramukhi* (*Solanum tuberosum*) and rice variety *Mahsuri (Oryza sativa)*, locally known as *Aijong rice* were used for comparison. Maize starch was purchased from HiMedia Laboratory, India and also used for comparison.



A)



B)

Fig. 9.1 Images of tubers of various taro cultivars investigated: A) Cultivars belonging to 'eddoe' type taro, and B) Cultivars belonging to 'dasheen' type taro

9.1.2 Starch isolation

Starch from taro and potato was extracted as per the method described by Benesi et al. (2004). Tubers were washed under tap water, peeled and cut into cubes of approximately 1 cm. The cubes were ground using a high speed laboratory blender (Philips HL 1632, India) for two minutes. The slurry was mixed with 10 times its volumes of distilled water. The suspension was filtered through double fold cheese cloth and the filtrate was kept for sedimentation for 6 hours. The supernatant was discarded and the sediment thus obtained was washed with distilled water for two times. The final sediment was dried at 45 °C for 24 h in drying oven. The dried starch was ground and passed through 100 mesh sieve and kept in air tight plastic containers for further analysis. Starch from rice was extracted as per method described by Wang and Wang (2001).

9.1.3 Chemical composition and amylose content

The moisture, fat, ash and crude fibre content of the isolated starches were determined by AOAC (1990) methods. Protein content (N×6.5) was determined by Kjeldahl method (1990). The amylose content was determined by colorimetric method (McGrance et al., 1998). The standard curve was prepared using pure potato amylose type III (HiMedia, India).

9.1.4 Granule size and shape

Shape and size of starch granules were evaluated using scanning electron microscope (JEOL JSM 6390 LV, Singapore). A thin layer of starch granule was mounted on the aluminium specimen holder by double-sided tape. The samples were coated with platinum and examined under the microscope at an accelerating voltage of 15 kV with magnification of 4000X. Size of the starch granules were determined by measuring the diameters of 30 randomly selected granules from the micrographs.

9.1.5 XRD analysis

XRD analysis of the starch samples were carried out using X-ray diffractometer (Miniflex, Japan). The samples were exposed to X-ray beam at 15 mA and 30 kV. Data were recorded over a diffraction angle (2θ) range of 5° to 50° with a step angle

of 0.05°. Percent crystallinity was determined by calculating the percentage ratio of diffraction peak area to the total diffraction area.

9.1.6 Fourier transform infrared (FT-IR) analysis

In order to spectral features of the taro starches, FT-IR spectra were obtained using FT-IR (Spectrum 100, Perkin Elmer, SA). The spectra were recorded in absorbance mode from 4,000 to 400 cm^{-1} (mid-infrared region) at a resolution of 4 cm^{-1}. Samples were thoroughly grounded with exhaustively dried pure KBr (1:100, w/w) and pellets were prepared by compression and analyzed. Background value from pure KBr was acquired before each sample was scanned.

9.1.7 Swelling and solubility

Swelling power and solubility of the starches were determined by modified method of Torruco-Uco and Betancur-Ancona (2007). Starch (0.5 g) was dispersed in 20 ml distilled water in a pre-weighed 50 ml centrifuge tubes and kept in shaking water bath at 60, 70, 80 and 90 °C for 30 min. The suspension was then centrifuged at 12,000 × *g* for 10 min. The supernatant was carefully decanted in a Petri dish and dried at 103 °C for 12 h. After decantation the weight swollen granules were taken. The swelling power and percentage solubility were calculated using the following formulas:

Swelling Power = Weight of swollen granules × 100 / (Weight of sample – Weight of dissolved starch)

% Solubility = Weight of dried starch in Petri dish × 100 / Sample weight

9.1.8 Clarity and stability

Clarity and stability of the starches were measured following the method described by Sandhu and Singh (2007). Aqueous starch suspension was prepared by heating 0.2 g starch in 20 ml water in shaking water bath at 90 °C for 1 h. The starch paste was cooled to room temperature. The starch pastes were stored at 4 °C in refrigerator and the absorbance was measured at 640 nm in spectrophotometer (Spectrascan UV-2600, Thermo Fisher Scientific, India). The absorbance was measured after every 24 h for seven days to determine the stability of the pastes.

9.1.9 Freeze-thaw stability

The freeze-thaw stability was determined according to the method of Singhal and Kulkarni (1990). Starch (5% dry basis, w/v) was dissolved in distilled water at 95 °C

for 30 minutes with constant stirring. Ten ml of paste was transferred to weighed centrifuge tubes. The weight of the paste was then determined. This was subjected to alternate freezing and thawing cycles (22 h freezing at -20°C followed by 2 h thawing at 30°C) for 5 days, centrifuged at 5000 × g for 10 minutes after each cycle and the percentage syneresis was determined as weight of exudates to the weight of paste.

9.1.10 Thermal properties

Gelatinization properties of the starches were determined by differential scanning calorimeter (DSC-60, Shimadzu, Singapore) by the method described by Jiranuntakul et al. (2011) with some modifications. Starch (3 mg) was weighed in the aluminum pans and water was added in the ratio of 1:3 for starch: water. The pans were sealed and allowed to stand for 12 h at 4°C for moisture equilibration. The samples were scanned from 25 -120°C at 10°C/min. The onset temperature (T_o), peak temperature (T_p), conclusion temperature (T_c) and gelatinization temperature range (T_c-T_o) were noted from the graphs. The enthalpy of gelatinization was estimated as Joules per gram of dry starch.

9.1.11 Pasting properties

Pasting properties of the starches were evaluated using Rapid Visco-Analyzer (RVA), model StarchMaster2 from Newport Scientific, Australia. Viscosity profiles were recorded using 12.5 % starch slurry in distilled water (total weight 28 g). A heating and cooling cycle of 13 min was used where the samples were heated from 50°C to 95°C in 5 min, held at 95°C for 2 min, cooled from 95 °C to 50 °C in 4 min and held at 50°C for 2 min. Pasting temperature (PT), peak viscosity (PV), hold viscosity (HV), final viscosity (FV), breakdown viscosity (BV) and setback viscosity (SV) were recorded from the graph.

9.1.12 Texture analysis of starch pastes

Starch pastes were prepared by heating an aqueous suspension of starch (1 g starch in 50 ml distilled water) in a shaking water bath at 100 °C for 30 min. The starch pastes were cooled to 25 °C by keeping the starch pastes in cooling water bath maintained at 25 °C for 1 h. Textural properties such as firmness, consistency and cohesiveness of starch pastes were determined by back extrusion method in Texture Analyzer,

TA.HDplus (Stable Micro Systems, UK) using a cylindrical probe (P-35). The probe was allowed to penetrate 20 mm from the surface of the sample at a speed of 1 mm/s. Firmness, cohesiveness, consistency and index of viscosity were calculated from the graphs using the software Exponent Lite 32 provided with the instrument.

9.1.13 Colour analysis of starch pastes

Colour parameters of the starch pastes containing 2 % starch were measured using colorimeter (Ultrascan VIS, Hunterlab, USA). Results were obtained in terms of L* (lightness), ranging from 0 (black) to 100 (white), a* (redness), ranging from +60 (red) to −60 (green), and b* (yellowness), ranging from +60 (yellow) to −60 (blue) values.

9.1.14 Colour of starch (dry powder)

Colour of starch (dry powder) was measured using colorimeter (Ultrascan VIS, Hunterlab, USA). L, a* and b* values were noted. L is for lightness, a* for redness and b* yellowness.

9.1.15 Identification of the variety suitable for food use

Based on the physicochemical properties, particularly functional properties the variety suitable for food use was identified for further studies.

9.2 Effect of Incorporation of Taro starch on Various Quality Parameters of Tomato Ketchup and its comparison to ketchup prepared using maize starch

9.2.1 Materials

Panchamukhi taro was selected because the functional properties of *Panchamukhi* taro like solubility, freeze-thaw stability etc., were found to be better compared to the other cultivars of taro. *Panchamukhi* taro (*Colocasia esculenta* var. *antiquorum*) was collected from an agricultural farm near Tezpur University, Assam, India. Starch was isolated from the tubers by method of Benesi et al. (2004) as described earlier.

Maize starch was purchased from HiMedia Laboratory, India for comparison.

Tomatoes (*Lycopersicon esculentum*) of even ripeness and colour were purchased from local market. The initial total soluble solids (TSS) of the tomato pulp

was 3.5±0.29 °Brix. All other ingredient for preparation including spices was also purchased from local market of Tezpur University.

9.2.2 Preparation of tomato ketchup

Ketchup was prepared as per the method described by Srivastava and Kumar (1994). In brief, 500 g pulp was prepared from tomatoes by hot pulping method by heating the pieces of tomato at 70°C for 2-3 min. Sugar (7.5 % w/w of initial weight of pulp) and salt (1% w/w of initial weight of pulp) were added to the pulp and the pulp was cooked at 100±2°C till the TSS reached 25° Brix. Starch from taro and maize were added 10 min before the end of cooking at a rate of 1 and 2% by weight of the amount of initial pulp taken. The pulp was continuously stirred with stainless steel ladle during cooking. To prepare the control ketchup sample the pulp was continuously stirred till the end of cooking without adding any starch. Acetic acid (5 ml per 1 kg of tomato pulp) was added to the ketchup at the end of cooking. The total cooking time was approximately 40 min and the final volume of the ketchup samples obtained were approximately 220 g from 500 g of pulp.

9.2.3 Serum loss

Serum loss from the tomato ketchups prepared was measured according to the method described by Gujral et al. (2002). Tomato ketchup (20 g) was taken in a centrifuge tube and then centrifuged at 5000 rpm for 10 min. The supernatant was discarded and the remaining ketchup was weighed.

% Serum loss = (Weight of serum removed/ weight of ketchup taken) × 100

9.2.4 Textural properties of ketchup

The textural properties of the ketchup samples were analysed by a texture analyser TA.HDplus (Stable Micro Systems, UK). Back extrusion was done to determine textural properties such as firmness, consistency and cohesiveness with a back extrusion cell using a long probe adaptor along with a 35 mm compression disc. For measuring the textural properties, ketchup samples were filled into the glass beakers provided by the manufacturer up to a height of 5 cm. The probe was allowed to penetrate 20 mm from the surface of the sample at a speed of 1 mm / s. Firmness,

cohesiveness, consistency and index of viscosity were calculated from the graphs using the software 'Exponent Lite 32'.

9.2.5 Colour parameters of ketchup

The colour parameters of the ketchup samples were analysed by a colorimeter (Ultrascan VIS, Hunterlab, USA). L, a* and b* values were noted. L is for lightness, a* for redness and b* for yellowness.

9.2.6 Sensory evaluation

Sensory evaluation of ketchup samples were conducted using a 9-point hedonic scale. The colour was judged visually. The smoothness was judged through the sense of touch. The flavour was judged by the sense of smell and taste. The mouth feel was judged by the tactile character and eating quality. The samples were presented individually to 15 semi-trained panellists. Water was used for mouth rinsing before and after testing of each sample.

9.3 Optimization of Starch Isolation from Taro Using Enzymes and Comparison of Properties of Starches Isolated by Enzymatic and Conventional Methods

9.3.1 Starch isolation using enzymes

Taro tubers locally known as *Panchamukhi* (*Colocasia esculenta* var. *antiquorum*) was collected from an agricultural farm near Tezpur University, Assam, India. Tubers were washed under tap water, peeled and cut into cubes of approximately 1 cm. Cubes (100 g) were weighed and ground using a laboratory blender (Philips HL 1632, India) for 1 min 30 s. The slurry was mixed with 100 ml distilled water and transferred to a 250 ml beaker. The slurry was subjected to enzymatic treatment using cellulase from *Aspergillus niger* (Sigma-Aldrich, 0.3 U/mg) and xylanase from *Thermomyces lanuginosus* (Sigma-Aldrich, 2500 U/g) and their combination. The slurry was incubated with different concentrations of cellulase and xylanase for varying time at different temperatures as per the experimental design in incubator shaker (CERTOMAT® IS, Sartorius Stedim Biotech, Goettingen, Germany) at 150 rpm. After incubation the suspension was filtered through double fold cheese cloth and the filtrate was centrifuged at 3000 × g for 10 min. The

supernatant was discarded and sediment was washed twice with distilled water. The final sediment was dried at 45°C for 24 h in hot air drying oven. The dried starch was ground and passed through 100 mesh sieve and kept in air tight plastic containers for further analysis. For conventional method the incubation step with enzymes was omitted. Starch slurry after grinding was mixed with 100 ml distilled water and was filtered through double fold cheese cloth and the filtrate was centrifuged at 3000 × g for 10 min. The rest of the procedure was similar to the enzymatic process.

Moisture and starch contents of the dried samples were determined to calculate the yield of pure starch. Moisture content was determined by hot air oven method[12] and starch content was determined by acid hydrolysis method using perchloric acid which hydrolysed the starch to glucose and dehydrated it to hydroxylmethyl furfural which was then measured by Anthrone reagent (Sadasivam and Manickam, 2011). The starch content of the taro tubers were 21.96±0.42 g (n=5) per 100 g fresh tuber (21.96 % wb).

Yield was obtained by calculating the amount of pure starch (db) recovered from 100 g of fresh taro sample.

9.3.2 Experimental design

A central composite rotatable design (CCRD) with four numerical factors was employed to design the experiments. The numerical factors were cellulase concentration (C), xylanase concentration (X), temperature of incubation (T) and incubation time (t). The cellulase and xylanase concentration were varied from 0 to 400 U/ 100 g taro tuber, incubation time was varied from 1 to 5 h and temperature was varied from 30°C to 50°C. A total of 30 experiments were performed (Table 4.1). Six replicates at the centre points of the design were performed to allow the estimation of pure error. All experiments were carried out in a randomized order to minimize the effect of external factors (Wanasundara and Shahidi, 1998).

9.3.3 Data analysis and optimization

Design Expert version 8 was used for analysis of data for starch yield and optimization. Experimental data were fitted to a second order polynomial model as follows:

$$Y = \beta_o + \sum_{i=1}^{3} \beta_i X_i + \sum_{i=1}^{3} \beta_{ii} X_i^2 + \sum \sum_{i<j=1}^{3} \beta_{ij} X_i X_j \qquad \text{Eqn. 1}$$

Where Y represents the response i.e. starch recovery, β_o is the constant, β_i, β_{ii} and β_{ij} are the regression coefficients and X_i and X_j are the independent variables in coded values.

Significant terms in the model were found by analysis of variance (ANOVA). Model adequacy was checked by lack of fit test, R^2, predicted R^2, adequacy precision and predicted residual sum of squares (PRESS). A non-significant ($p > 0.05$) lack of fit, predicted R^2 comparable to fitted R^2, low PRESS and adequacy precision higher than 4, shows that the model fitted is adequate to predicting (Corzo et al., 2008; Erbay and Icier, 2009; Mohapatra and Bal, 2010). Response surfaces were generated to study the effect of interactions on starch recovery. The optimization of the extraction process was done using desirability function.

9.3.4 Properties of starches isolated by enzymatic and conventional methods

After optimization of the enzymatic method, starch was isolated from taro tubers using the optimized condition. The functional properties of the starch isolated by the optimized enzymatic method were compared with the starch isolated by conventional method. The functional properties studied were freeze-thaw stability, pasting properties, swelling and solubility, and clarity of starch pastes. The methods were already described earlier.

9.4 Statistical analysis

The data were subjected to single factor analysis of variance (ANOVA) using 'Data Analysis Tool' of 'Microsoft Excel'. Fisher's 'Least Significant Difference (LSD)' method was used to determine the statistical difference between the results obtained.

10. Detailed analysis of results indicating contributions made towards increasing the state of knowledge in the subject:

10.1 Physicochemical properties of starches from various cultivars of taro and its comparison to maize, rice and potato starches

10.1.1 Chemical composition and amylose content

The chemical composition and amylose content of the isolated starches are shown in Table 10.1. The moisture content of the starches varied from 8.96 to 11.93% which were within safe limit for storage of starches without deterioration in quality of starches (Mweta et al. 2010). The ash contents of the starch samples varied from 0.07 to 0.77%. The lowest ash content was observed for maize starch whereas the highest was observed for potato starch. The higher ash content of potato starch might be attributed to the presence of phosphates in potato starch. The ash content of the starches from various taro cultivars varied from 0.09 to 0.71%. The lowest ash content was observed for *Panchamukhi* starch. Nand et al. (2008) found that the ash contents of starch obtained from the cassava and taro samples ranged between 0.08 - 0.20 %. Mweta et al. (2008) also found that ash content of the taro and cassava starches ranged from 0.10-0.20%. The protein content of the taro starches ranged from 0.33 to 0.71%, and that of rice, maize and potato starches were 1.26, 0.64 and 0.39% respectively. The reason for higher protein content of rice starch might attributed to the structure of the starch granules in cereals which are embedded in protein matrix, so it is difficult to separate protein from the granules and might have contributed to the protein content of rice starch. The results were in accordance with the results obtained by Peroni et al. (2006) for cassava, arrowroot and sweet potato starches whose protein content varied from 0.08-0.35%. The lipid content of taro starches ranged from 0.11 to 0.43% for the various taro cultivars. Jane et al. (1992) reported that the lipid content in taro starches ranged between 0.08 to 0.12 % (dry basis) which is in agreement with the present findings. Peroni et al. (2006) also found that tuber starches contain lipids in the range 0.10-.24%. Lower lipid content of starches indicates a high purity of extracted starches. The differences in the composition of the starches among the taro cultivars might be attributed to the differences in mucilage content of the taro tubers. Tubers with higher mucilage contributed to the impurities of the starch samples as separation of starch granules from other components of the starch slurry with higher mucilage was difficult.

Table 10.1 Chemical composition and amylose content of the isolated starches from various taro cultivars, rice, maize and potato

Sample[1,2,3]	Moisture, % wet basis	Protein, % dry basis	Fat, % dry basis	Fibre, % dry basis	Ash, % dry basis	Starch, % dry basis	Amylose, % dry basis of pure starch
Kani	11.27±1.02^{ab}	0.71±0.23^{b}	0.30±0.05^{abc}	0.15±0.02^{ab}	0.71±0.20^{ab}	96.25±1.15^{ab}	16.19±1.52^{cd}
Ahina	9.36±2.12^{bc}	0.62±0.12^{bc}	0.33±0.09^{abc}	0.21±0.06^{ab}	0.52±0.21^{ab}	94.18±1.62^{b}	15.79±2.23^{cde}
Muktakashi	10.12±1.36^{abc}	0.54±0.21^{bcd}	0.21±0.20^{cd}	0.19±0.09^{ab}	0.43±0.15^{b}	95.95±1.32^{ab}	14.78±1.27^{de}
Panchamukhi	9.22± 0.10^{bc}	0.34±0.05^{d}	0.11± 0.007^{d}	0.09±0.03^{b}	0.09± 0.002^{c}	95.87± 1.52^{ab}	18.14 ± 1.23^{c}
Garu	10.36±0.89^{abc}	0.52±0.06^{bcd}	0.41±0.10^{ab}	0.19±0.05^{ab}	0.65±0.11^{ab}	96.01±1.25^{ab}	13.21±0.98^{ef}
JCC37	9.57±1.54^{bc}	0.33±0.09^{d}	0.43±0.14^{a}	0.25±0.11^{a}	0.57±0.17^{ab}	96.69±0.94^{ab}	11.87±1.24^{f}
JCC57	8.96±0.98^{c}	0.42±0.15^{cd}	0.36±0.07^{abc}	0.17±0.08^{ab}	0.43±0.25^{b}	95.12±2.12^{ab}	13.18±0.57^{ef}
Rice	9.96±1.54^{abc}	1.26±.26^{a}	0.31±0.04^{abc}	0.23±0.03^{a}	0.58±0.09^{a}	95.69±1.56^{ab}	21.57±2.08^{b}
Maize	11.93±0.23^{a}	0.64±0.07^{b}	0.24±0.006^{bcd}	0.16±0.08^{ab}	0.07±0.004^{c}	96.30±1.96^{ab}	24.20±1.51^{ab}
Potato	10.64±1.23^{abc}	0.39±0.11^{cd}	0.19±0.09^{cd}	0.10±0.02^{b}	0.77±0.36^{a}	97.13±0.88^{a}	25.75±2.12^{a}

[1]Values reported as Mean ± S. D. of three replications and expressed in dry basis except for moisture which is expressed in wet basis
[2]Meanss followed by same superscript small letters within a column are not significantly different ($p>0.05$)
[3]Amylose content calculated as amount of amylose present in pure starch; and amylopectin, % = (100 – amylose %)

The purity of the isolated starches from taro cultivars varied from 94.18 to 96.69%, which were less as compared to potato starch with a purity of 97.13%, but were closer to the purity of rice and maize starches with purity of 95.69 and 96.30%. This might be due to high amount of mucilage in taro tubers compared to potato, and separation of starch from starch water containing mucilage becomes difficult, but the values were not significantly different from each other.

The amylose content of the taro starches ranged between 11.87 to 18.14%, which were significantly lower than rice, maize and potato starches with amylose content of 21.57, 24.20 and 25.75% respectively. The amylose content of *Kani*, *Ahina*, *Muktakashi* and *Panchamukhi* cultivars were found to be more compared to *Garu*, *JCC37* and *JCC57* cultivars. This might be due to the fact that *Kani*, *Ahina Muktakashi* and *Panchamukhi* belonged to same variety i.e. *antiquorum* whereas *Garu*, *JCC37* and *JCC57* belonged to var. *esculenta*. The highest amylose content was observed for *Panchamukhi* starch. The results were in agreement with the findings of Mweta et al. (2010) for different varieties of taro starches which varied from 10.6 to 21.0 %. Singh et al. (2004) reported amylose content of native potato starches from 25.6 to 30.4 % and for rice starch, amylose content varied from 21.88 to 31.6 % (Ashogbon and Akintayo, 2012; Wang et al., 2010). The amylose content of the taro starches was found to be lower than potato and rice starches reported by other authors (Singh et al., 2004; Ashogbon and Akintayo, 2012). The low level of amylose makes taro starches more hydrolysable as starch with high level of amylose hydrolyzed more slowly than starch with low level of amylose owing to the double helical form of amylose molecule which is not easily accessible by enzymes.

10.1.2 Granule size and shape

Scanning electron micrographs (Fig. 10.2) of the taro cultivars clearly showed a mixture of various shapes and sizes of the starch granules. The starch granules were dome shaped, polygonal, split and irregular in shape. The average size of the starch granules varied from 2.22 to 3.29 μm for the different taro cultivars (Table 10.2). Significant differences were not observed between the sizes of the starch granules, although the *JCC37* was found to have largest granule size amongst the seven cultivars investigated. Similar values for granule sizes of taro starch from different regions were observed by, Mweta et al. (2010), Jane et al. (1992), Mepba et al. (2009), and Agama-Acevedo et al. (2011). The average sizes of rice, maize and

potato starch granules were 4.31, 11.71 and 20.50 μm. The shape of rice starch granules was similar to the taro starch granules. Maize starch was found to have irregular, round or oval shapes, while potato starch was found to have round and oval shapes. It was evident from the micrographs that taro starch granules were much smaller than starch granules of maize or potato, and were comparable with that of rice starch in size and shape of granules. Maize and potato starch granules also presented broader range of the sizes of the starch granules. Shape and size of starch granules influence many physicochemical and functional properties of starches like paste viscosity (Agama-Acevedo et al., 2011) and swelling volume and solubility (Torruco-Uco and Betancur-Ancona, 2007; Moorthy, 2003).

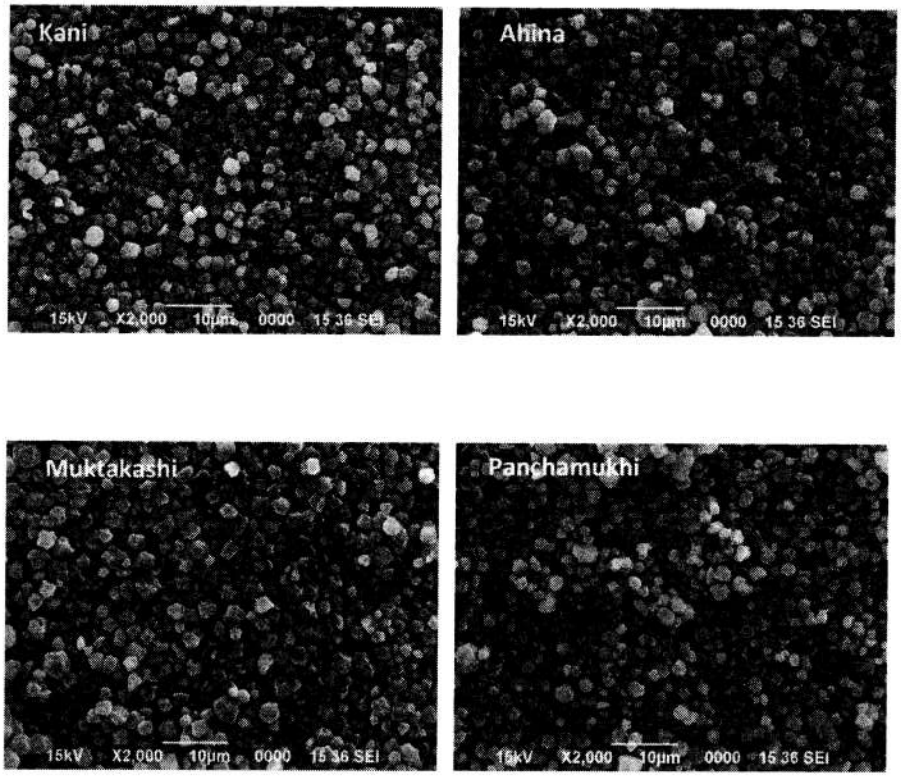


Fig. 10.2 a) Scanning electron micrographs of starch granules of taro cultivars *Kani*, *Ahina*, *Muktakashi* and *Panchamukhi* belonging to *Colocasia esculenta* var. *antiquorum*

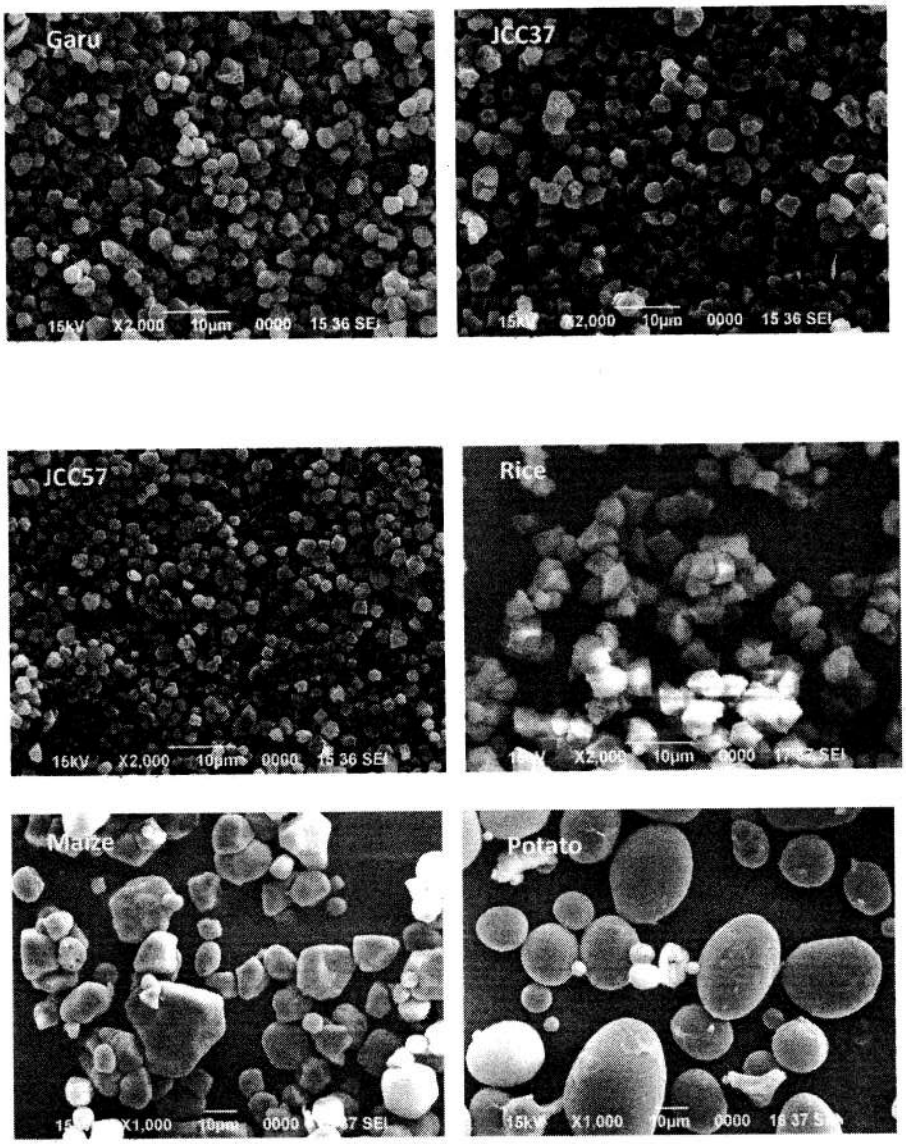


Fig. 10.2 b) Scanning electron micrographs of starch granules of taro cultivars *Garu*, *JCC37* and *JCC57* belonging to *Colocasia esculenta* var. *esculenta* and of rice, maize and potato

10.1.2 XRD pattern

The position of strong and weak peaks and % crystallinity of the starches of the taro cultivars investigated were presented in Table 2.2. The positions of the peaks were found to be similar for the taro starches. It was observed that all the taro cultivars and rice and maize displayed A-type XRD pattern (Fig. 10.3), which is a characteristic of cereal starches. Potato starch presented B-type pattern. Generally tuber starches have B or C-type pattern (Hernandez-Lauzardo et al., 2004). Strong peaks were observed near 15°, 17°, 18° and 23° 2θ for starches isolated from the different taro cultivars and the starches isolated from rice and maize. For potato starch strong peaks were observed near 6° and 17° 2θ, which is a characteristic of B-type starches. The % crystallinity of the taro starches were higher than rice or potato starch and were closer to that of maize starch. Many properties like digestibility and retrogradation are affected by XRD pattern and the degree of crystallinity of the starches (Jane et al., 1992; Han and Bemiller, 2007).

10.1.4 FT-IR spectra

FT-IR spectra of the starch samples in the 4,000–400 cm^{-1} region are shown in Fig. 2.4. The spectra obtained for the ten starch samples were similar in the form and intensity of the major peaks. The spectra showed high absorption near the wave numbers 574, 930, 1016-1022, 1048, 1080, 1154, 1365, 1420, 1644, 2930 and 3380cm^{-1} confirming the carbohydrate nature of the samples. The peaks at 3,400 cm^{-1} and 2,930 cm^{-1} could be attributed to O–H and H-C-H bond stretching, respectively, while the peak at 1644 may be attributed to COO- stretching vibration in a carbohydrate group (Zeng et al., 2011; Fan et a., 2012). Peaks at 1,420 cm^{-1} and 1,365 cm^{-1} were attributable to the bending modes of H–C–H and C–H symmetric bending of CH_3 (Zeng et al., 2011; Van Soest et al., 1994; Kauráková and Mathlouthi, 1996; Kizil et al., 2002). C-C and C-O

Table 10.2 Granule size distribution, % crystallinity and position of peaks in X-ray diffractogram of starch samples

Sample	Granule size distribution		% Crystallinity	Position of strong peaks (2θ)	Position of weak peaks (2θ)
	Range, µm	Average granule size[1, 2], µm			
Kani	1.07 – 3.33	2.22±0.73[b]	40.26	15.4, 17.25, 18.00, 23.4	26.80, 30.45, 33.45
Ahina	1.19 – 4.17	2.51±0.79[b]	34.97	15.15, 17.45, 18.15, 23.35	26.35, 30.85, 32.20
Muktakashi	1.43 – 3.87	2.95±0.73[ab]	39.24	15.30,17.4,18.05, 23.25	26.40,31.65,33.05
Panchamukhi	2.19 – 4.28	2.77±1.12[a]	44.73	15.15, 17.15, 17.80, 23.10	26.35, 30.35, 33.05
Garu	1.91 – 4.17	3.12±0.75[b]	39.14	15.45, 17.35, 18.1, 23.30	26.45, 31.1, 33.65
JCC37	2.14 – 4.88	3.29±0.86[a]	42.13	15.35,17.55,18.35,23.25	26.30, 31.45, 33.10
JCC57	1.43 – 3.45	2.49±0.53[b]	45.11	15.60,17.49, 18.6,23.70	26.75,30.75,33.55
Rice	2.26 – 6.55	4.31±1.15[b]	31.55	15.25, 17.05, 17.95, 23.15	26.70, 30.45, 33.95
Maize	7.38 - 16.67	11.71±3.93[b]	43.59	15.30, 17.60, 18.45, 23.10	26.15, 30.50, 33.75
Potato	7.65 – 47.63	20.50±11.57[a]	23.57	5.60, 17.15	11.65, 15.00, 22.30, 24.00, 34.35

[1]Values reported as Mean ± S. D. of 30 granules
[2]Meanss followed by same superscript small letters within the column are not significantly different ($p>0.05$)

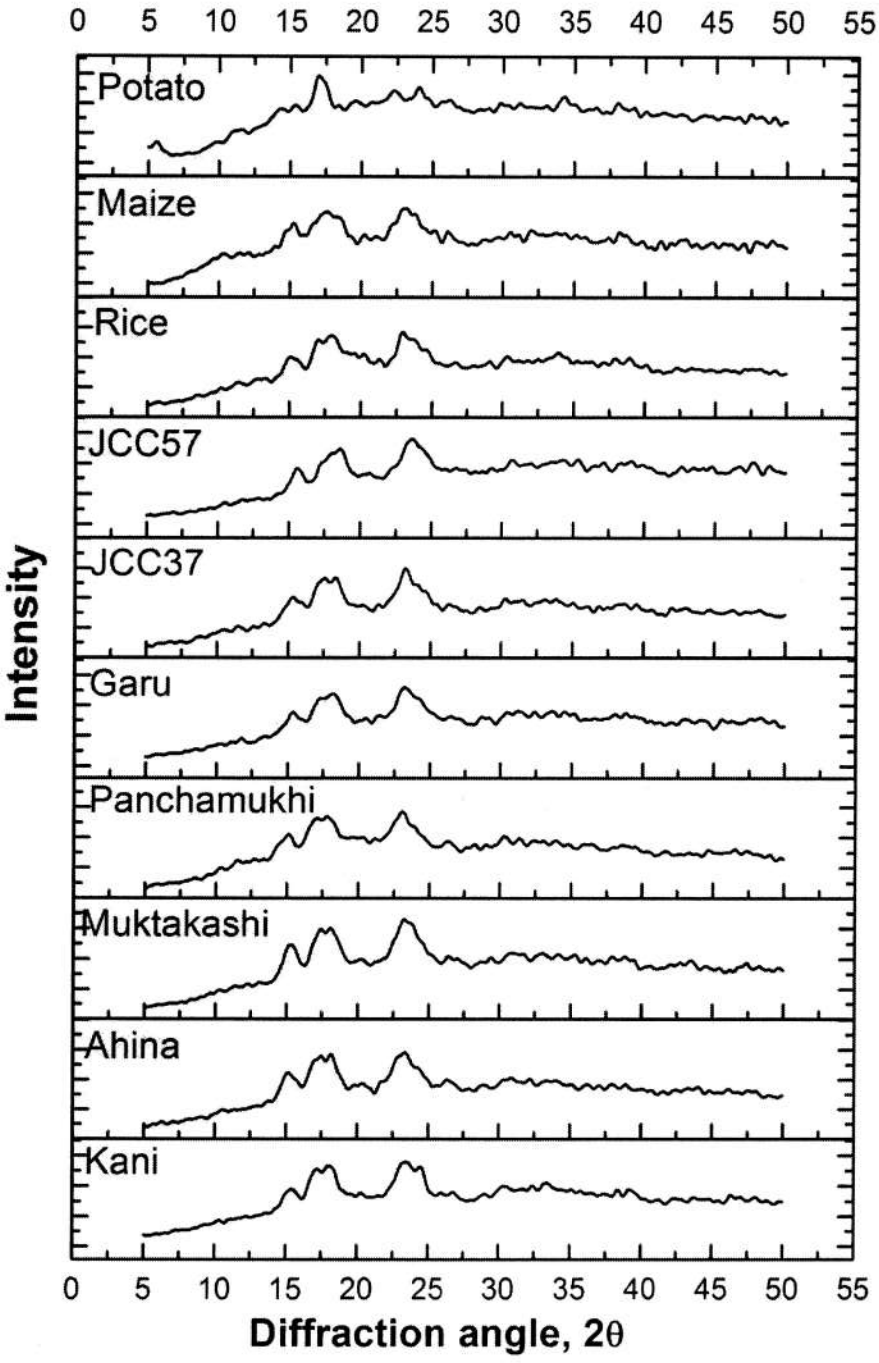


Fig. 10.3 XRD pattern of starches of various taro cultivars, rice, maize and potato

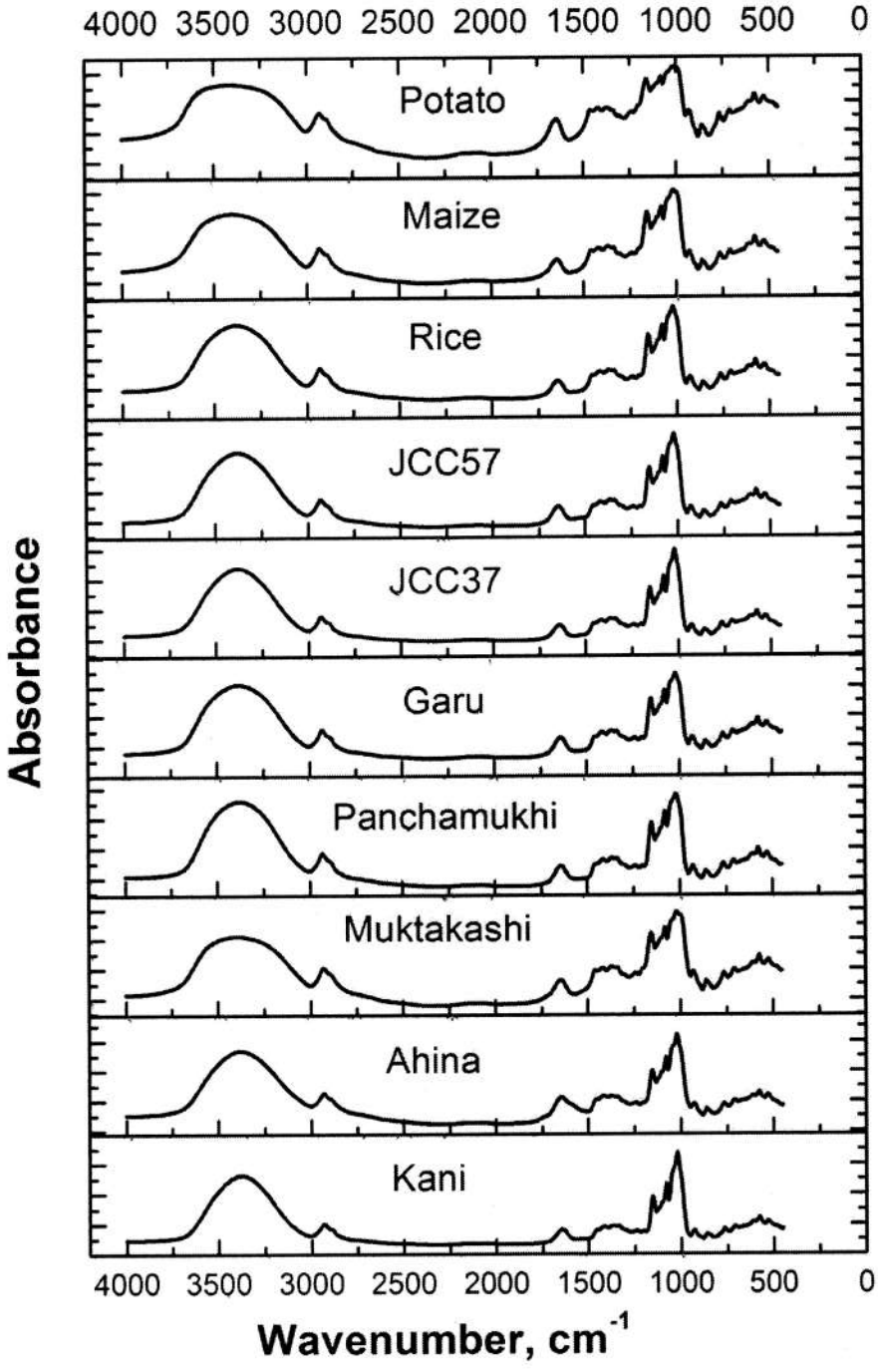


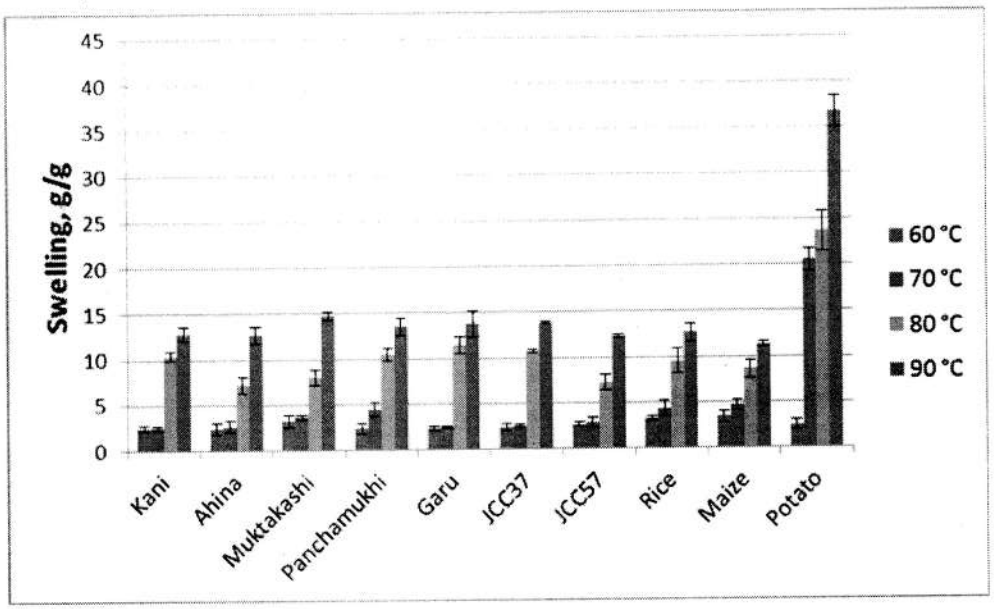
Fig. 10.4 FT-IR spectra of starches from various taro cultivars, rice, maize and potato

bond stretching could be assigned to the peak at 1154 cm^{-1} (Zeng et al., 2011; Van Soest et al., 1994). The peak at 1080 cm^{-1} was attributed to C–O–H bending.[49] The bands at 1,047 and 1,022 cm^{-1} were associated with the crystalline and amorphous structures of starch, respectively (Zeng et al., 2011; Fan et a., 2012; Van Soest et al., 1995). The ratio of absorbance at 1047 and 1022 cm^{-1} indicate the degree of order in starch samples (Van Soest et al., 1995; Koksel et al., 2008; Sevenou et al., 2002). The bands at 930 cm^{-1} and 574 cm^{-1} describe the D-glucopyranosyl ring vibrational modes and skeletal modes of pyranose ring, respectively. The FT-IR spectra obtained in the present work are not much different from the spectra obtained by Aboubakar et al. (2008) for Cameroonian taro starches and that of rice starches reported by Fan et al. (2012).

10.1.5 Swelling and solubility

The swelling and solubility of the starches increased with the increase in temperature (Fig. 10.5). The solubilities at 60°C were lower and were not significantly different among the seven cultivars and the starches from rice maize and potato, although the solubility of *Panchamukhi* starch was highest at this temperature. At 70°C and 80°C the solubility of *Panchamukhi* taro starch was significantly higher than the other starches. At 90°C highest solubility was observed for *Kani* starch and was found to be close to that of *Panchamukhi* starch and the differences in solubility of these two starches were not significantly different. It was further observed that *Ahina* and *Muktakashi* which belonged to same variety as that of *Kani* and *Panchamukhi* i.e. var. *antiquorum*, showed lower solubilities compared to *JCC37* and *JCC57* at 80 and 90°C, but were higher than that of *Garu* starch at 60, 70 and 80 °C. The low solubility of starches at low temperatures might be due to the semi-crystalline structure of the starch granules and the hydrogen bonds formed between hydroxyl groups within the starch molecules. As the temperature increased, the solubility increased due to breaking of starch granules and exposure of hydrophilic groups to water (Eliasson and Gudmundsson, 1996). Swelling power indicates the water holding capacity of starch granules and is affected by the extent of chemical cross bonding within the granules (Chen et al., 2003). It was observed that at 60 °C the differences in swelling power of all the starches were not significantly different.

But as the temperature was increased potato starches swelled significantly more as compared to other starches. Swelling of taro starches were not significantly different from each other at all temperatures and were comparable with that of rice and maize starch. The reason for higher swelling power of potato starch may be attributed to the large size of the starch granules and higher amount of phosphate present in potato starch (Kim et al., 1996).



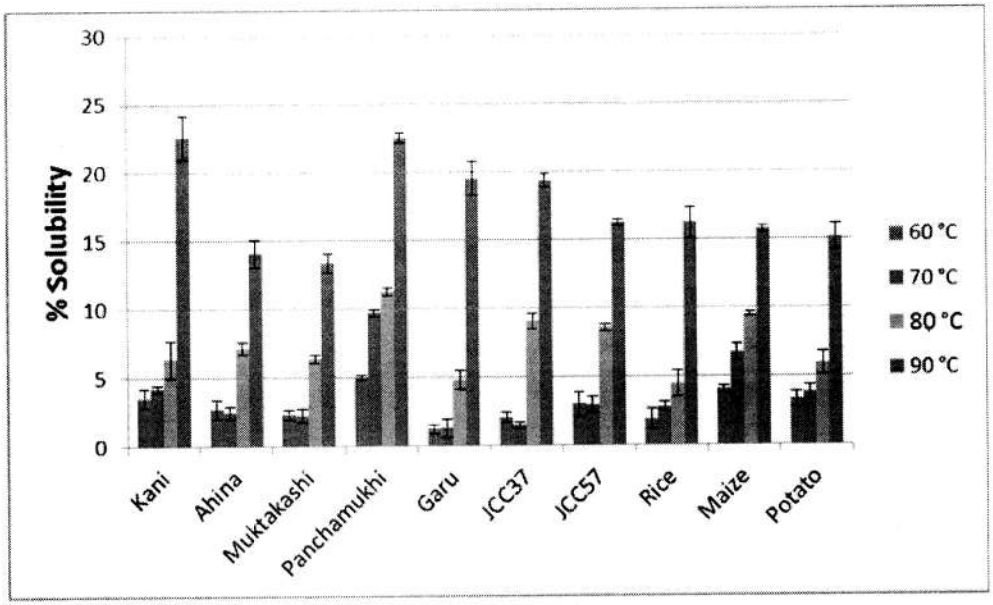


Fig. 2.5 Swelling and solubility of starches of various taro cultivars, rice, maize and potato

Vertical error bars above the columns represent standard deviation (S. D.) of three replications

10.1.6 Paste clarity and stability

Clarity (% transmittance) of the starch pastes of the starches from the different taro cultivars and that of rice, maize and potato for a period of seven days are shown in Fig. 10.6. The clarity of starch paste from *Panchamukhi* was significantly higher than other taro starches during the initial and final days. The stability of *Panchamukhi* starch paste was also found to be better in comparison to the starch pastes from other taro cultivars as the variation in clarity during 7 days of storage was less. Potato starch showed the highest light transmittance as compared to other starch pastes. The clarity of the rice and maize starch paste was also significantly more than the taro starches. The stability of potato starch paste was also more than other starches as % light transmittance did not differ significantly for the storage period. Clarity of the taro starch pastes were the lowest and found least stable except for *Panchamukhi* taro which was closer to clarity and stability of rice starch paste. The higher clarity and stability of potato starch paste may be due to the large size of the potato starch granules because of which less number of starch particles was present in the solution thereby scattering less amount of light (Singh et al., 2004). Taro starches being smaller in size scattered more amount of light. In addition, potato starch has B-type crystallinity and B-type starches exhibit better clarity than A-type starches (Moorthy, 2002). The lower stability of taro starch paste can be attributed to lower swelling of taro starch granules, as starch with higher swelling power are less susceptible to retrogradation which determines the stability of starch pastes (Nwokocha et al., 2009). The clarity of the starch pastes increased up to the 3rd day, remained stable till 5th day, and then started decreasing. Similar pattern of starch paste stability was observed by Mweta et al. (2008) for taro and cassava starches. The reason for increase in clarity in the initial period might be settling of the larger molecules or impurities present in the starch paste, thereby increasing the clarity. The decrease in clarity after a certain period of storage may be attributed to retrogradation, where molecules comprising gelatinized starch reassociate into an ordered structure to retrieve a crystalline order, and during this process, the starch paste becomes more opaque (Thomas and Atwell, 1999).

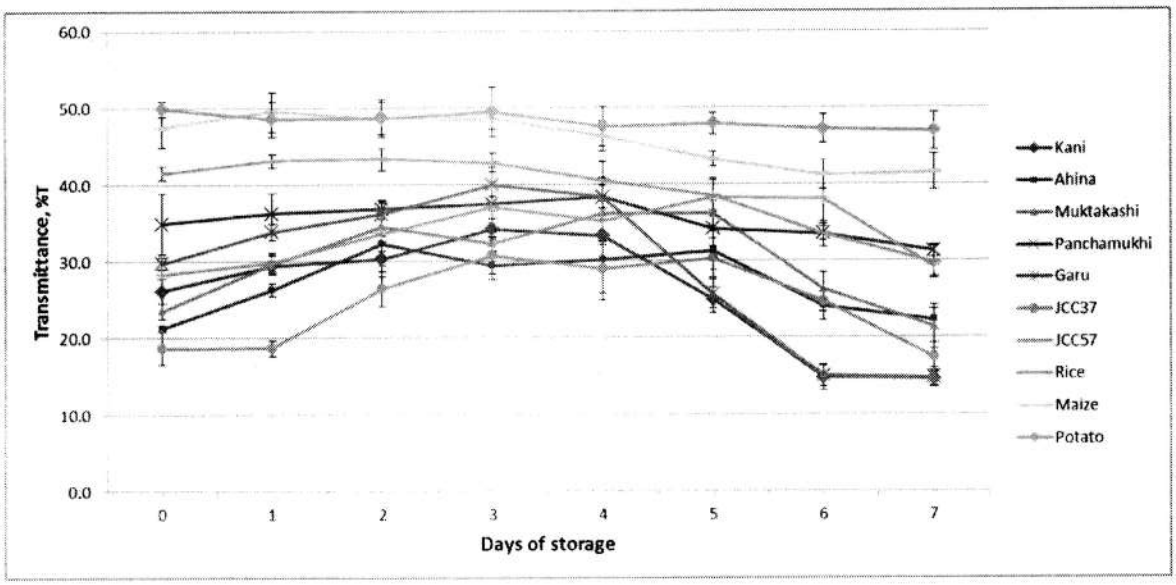


Fig. 10.6 Clarity and stability of starch pastes from various taro cultivars, rice, maize and potato

Vertical error bars represent standard deviation (S. D.) of three replications

10.1.7 Thermal properties

The gelatinization properties of the starches are presented in Table 2.3. The peak gelatinization temperatures (T_p) of the taro cultivars were significantly different from each other. The T_p of starches for the taro cultivars ranged from 70.61 and 83.33°C. The T_p of *JCC37* was the highest while that of *Muktakashi* starch was the lowest among the taro cultivars.

The T_p for rice and maize starches were 72.53 and 69.70°C and was found to be closer to the taro starches except for *JCC37* and *JCC57* starches. The T_p of potato starch was observed to be 65.55°C which was much lower compared to taro and rice starches. Wang et al. (2010) and Park et al. (2007) reported similar values for rice starches, Jiranuntakul et al. (2011) for rice and potato starches, and Mweta et al. (2010) for native taro starches of South Africa. The enthalpy of gelatinization (ΔH) of potato starch (15.27 J g^{-1}) was the highest among the starches followed by maize starch with ΔH of 15.24 J g^{-1}. The difference in the thermal behavior of the starches might be attributed to the amylose content (Bao et al., 2004) and crystallinity of the starch granules (Tester, 1997) as taro, maize and rice starches presented A-type XRD patterns. Potato starches also exhibited broader range of gelatinization compared to taro and rice starches, which might be attributed to the higher amylose content of the potato starches (Sandhu et al., 2005). It was observed that the gelatinization behaviour of the taro starches were comparable to rice and maize starch, but differed much from potato starch.

10.1.8 Pasting properties

The pasting profile of the starches from the different taro cultivars, rice, maize and potato is presented in Table 10.4. Pasting temperature is the temperature at which the viscosity of the starch pastes begins to rise. The pasting temperatures of all the taro cultivars were found to be high and ranged from 82.1 to 88.3°C. *Kani*, *Ahina*, *Muktakashi* and *JCC57* showed higher pasting temperature than the other three cultivars. The pasting temperatures of the North-East Indian varieties were found to be higher than the Mexican taro variety (Agama-Acevedo et al., 2011) or the Australian variety reported by Aprianita et al. (2009). The pasting temperatures of the taro starches were observed to be higher than the T_p values in DSC study, except for *JCC37* where pasting temperature and T_p values were closer. This might be attributed to the moisture equilibration process carried out before scanning the samples in DSC.

Starch granules of taro being smaller in size, absorbed moisture at faster rate during equilibration which facilitated in reaching the gelatinization temperature at an early stage. Starch granules of *JCC37* being larger in size as compared to the other cultivars might not have absorbed moisture at the same rate unlike other cultivars, thereby showing closer values of pasting temperature and T_p. Pasting temperatures were also observed to be higher than rice, maize and potato starches investigated. The results corroborates with the findings of other authors (Ashogbon and Akintayo, 2012; Chávez-Murillo et al., 2012). The pasting temperatures obtained in the present study were found to be closer to corn starches reported by others (Nwokocha et al., 2012; Sandhu and Singh, 2007). The peak, hold, final, breakdown and setback viscosities of *Garu*, *JCC37* and *JCC57* were much higher compared to the viscosities of other cultivars (Fig. 10.7) particularly *Kani* and *Muktakashi*. These differences might be due to amylose content of the starches and the differences in variety of taro. The viscosities observed in the present study were much higher than those observed by Lu et al. (2008) and were similar to those observed by Agama-Acevedo et al. (2011). It was seen that for all the starches the final viscosity was lower than peak viscosity. These results support the observations made by Mepba et al. (2009) and Agama-Acevedo et al. (2011) but were different than those observed by Nwokocha et al. (2009) and Aprianita et al. (2009) where final viscosity was found to be higher than peak viscosity. The viscosities of rice and maize starch were found to be closer to that of the taro starches particularly *Garu*, *JCC37*, *JCC57* and *Panchamukhi* starches. The final and setback viscosities of taro starches were lower than rice and maize starches, which makes them better suited for application requiring constant stirring during heating and cooling. It was further observed that the peak, hold and final viscosities of potato starch were much higher than the other starches studied. This might be due to the large size of the starch granules and higher swelling of the potato starch. The viscosity values of the taro starches measured by RVA were comparable to rice starches of Indian varieties investigated by Gani et al. (2013) and starch from Chinese rice varieties (Yu et al., 2012) and were found to be lower compared to potato, arrowroot and canna starches (Srichuwong et al., 2005).

Table 10.3 Gelatinization properties of starch samples

Sample[1,2]	T_o, °C	T_p, °C	T_c, °C	T_c-T_o, °C	ΔH, J/g
Kani	66.13±0.08^{e}	74.67±0.09^{c}	77.94±0.12^{e}	11.81±0.09^{c}	13.36±0.15^{de}
Ahina	66.23±0.09^{e}	73.33±0.11^{f}	78.05±0.03^{e}	11.82±0.07^{c}	12.58±0.19^{f}
Muktakashi	64.18±0.15^{f}	70.61±0.13^{h}	75.27±0.07^{g}	11.08±0.06^{c}	13.59±0.23^{cd}
Panchamukhi	66.94±0.08^{d}	74.36±0.08^{d}	78.96±0.13^{c}	12.02±0.11^{b}	13.98±0.15^{bc}
Garu	67.25±0.13^{c}	74.10±0.07^{b}	78.59±0.14^{d}	11.34±0.07^{d}	12.24±0.27^{f}
JCC37	77.24±0.21^{a}	83.33±0.22^{a}	86.99±0.05^{a}	9.75±0.12^{g}	14.25±0.41^{b}
JCC57	74.29±0.11^{b}	80.01±0.14^{b}	84.67±0.08^{b}	10.38±0.13^{f}	13.15±0.24^{e}
Rice	66.31±0.21^{e}	72.53±0.12^{g}	77.67±0.09^{f}	11.36±0.17^{d}	14.94±0.18^{a}
Maize	62.19±0.07^{g}	69.70±0.11^{i}	74.87±0.14^{h}	12.68±0.09^{a}	15.24±0.18^{a}
Potato	58.62±0.12^{h}	65.55±0.09^{j}	71.32±0.17^{i}	12.70±0.15^{a}	15.27±0.21^{a}

[1] Values reported as Mean ± S. D. of three replications

[2] Means followed by same small letter superscripts within a column are not significantly different (p 0.05)

Table 10.4 Pasting properties of the starch samples

Sample[1, 2]	Pasting Temperature, °C	Peak Viscosity, cP	Hold Viscosity, cP	Final Viscosity, cP	Breakdown Viscosity, cP	Setback Viscosity, cP
Kani	88.3±1.02^{a}	2129±32.3^{i}	1575±15.6^{h}	2203±98.6^{f}	554±46.3^{i}	628±94.3^{g}
Ahina	87.20±0.21^{ab}	3379±22.2^{h}	1875±34.1^{g}	2871±16.4^{c}	1504±22.2^{h}	996±29.2^{d}
Muktakashi	87.00±0.67^{ab}	2175±10.4^{i}	1553±21.3^{h}	2034±59.6^{g}	622±31.1^{i}	481±34.4^{h}
Panchamukhi	84.1±1.12^{c}	4301±31.3^{f}	2114±18.6^{f}	2940±48.6^{e}	2187±46.3^{f}	826±24.3^{ef}
Garu	82.1±1.15^{d}	4736±45.6^{c}	2217±23.2^{e}	2982±103.2^{e}	2519±54.3^{d}	765±106.4^{f}
JCC37	83.30±1.08^{cd}	4868±31.2^{d}	2241±15.3^{e}	3160±29.6^{d}	2627±21.3^{c}	919±25.9^{de}
JCC57	86.10±0.34^{b}	5704±16.9^{b}	2680±11.7^{c}	3834±47.2^{c}	3024±17.6^{b}	1154±54.4^{c}
Rice	78.4±1.06^{e}	5130±36.4^{c}	2757±21.1^{b}	4741±69.4^{b}	2373±36.5^{e}	1984±82.2^{a}
Maize	78.1±0.75^{e}	3974±25.6^{g}	2331±26.5^{d}	3878±56.9^{c}	1661±52.6^{g}	1529±29.4^{b}
Potato	68.8±0.96^{f}	10571±29.2^{a}	6082±16.3^{a}	6380±156.3^{a}	4489±78.2^{a}	298±54.2^{i}

[1] Values reported as Mean ± S. D. of three replications

[2] Means followed by same small letter superscripts within a column are not significantly different (p 0.05)

10.1.9 Freeze-thaw stability

Freeze-thaw stability of a starch gel is an important parameter in determining the industrial application of a starch (Huang et al., 2007). The amount of water separated (% syneresis) from the starch gels of the seven taro cultivars and rice, amize and potato up to 5 freeze-thaw cycles is shown in Fig. 10.8. It was observed that percent syneresis gradually increased up to 4 freeze thaw cycle and then decreased after the 5th cycle for *Kani*. *Ahina*, *Muktakashi*, *JCC37* and *JCC57* starches. In case of *Panchamukhi* starch %syneresis decreased up to the 3rd cycle and then gradually increased till 5th cycle. Syneresis in *Garu* and potato starch pastes initially increased up to the 3rd cycle and then decreased in the fourth cycle and then again increased in the 5th cycle. For rice starch syneresis decreased in the second cycle and then gradually increased in the subsequent cycles till the 5th cycle. Syneresis in maize starch paste increased in the second cycle, then decreased in the 3rd cycle and again an increase in the syneresis was observed till the 5th cycle. This decrease in syneresis was likely from the changes in gel structure to sponge-like structure. This structure easily reabsorbed most of the extruded water if the water separation was too slow (Charoenrein et al., 2008). Similar observations for amaranth, maize, cowpea, chickpea and rice starch gel have been reported (Nwokocha et al., 2012; Huang et al., 2007; Baker and Rayas-Duarte, 1998; Yuan and Thompson, 1998). The %syneresis was highest for *Muktakashi*, whereas it was lowest for *Panchamukhi*. Although, all the starch gels exhibited poor freeze-thaw stability, *Panchamukhi*, *JCC37* and *JCC57* starch pastes were more stable to freeze-thaw compared to *Kani*, *Ahina*, *Muktakashi* and *Garu* starch pastes. Freeze-thaw stability of *Panchamukhi*, *JCC37* and *JCC57* was found to be better compared to rice starch and was comparable to maize starch. Potato starch showed best freeze-thaw stability among the starches, which might be due to the higher swelling of potato starch granules and presence of phosphate groups in the amylopectin molecules. Among the taro cultivars *Panchamukhi* starch showed least syneresis and best freeze-thaw stability. Modification of the taro starches would be required if they are to be used in foods meant for refrigeration.

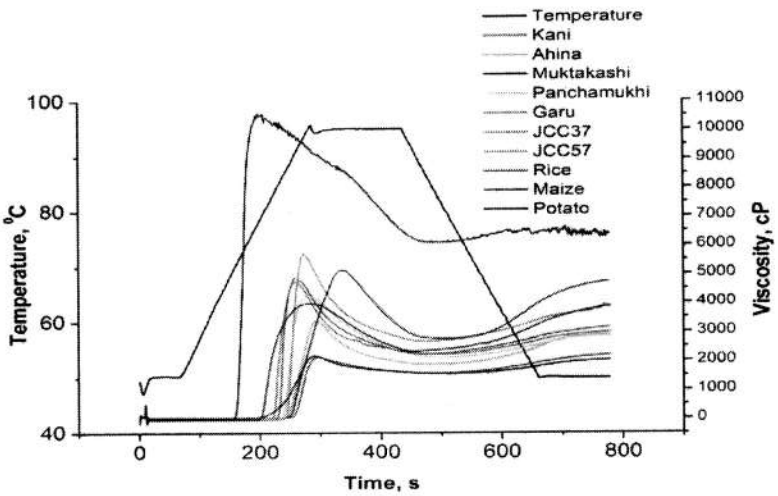


Fig. 10.7 Pasting profiles of starches from various taro cultivars, rice, maize and potato

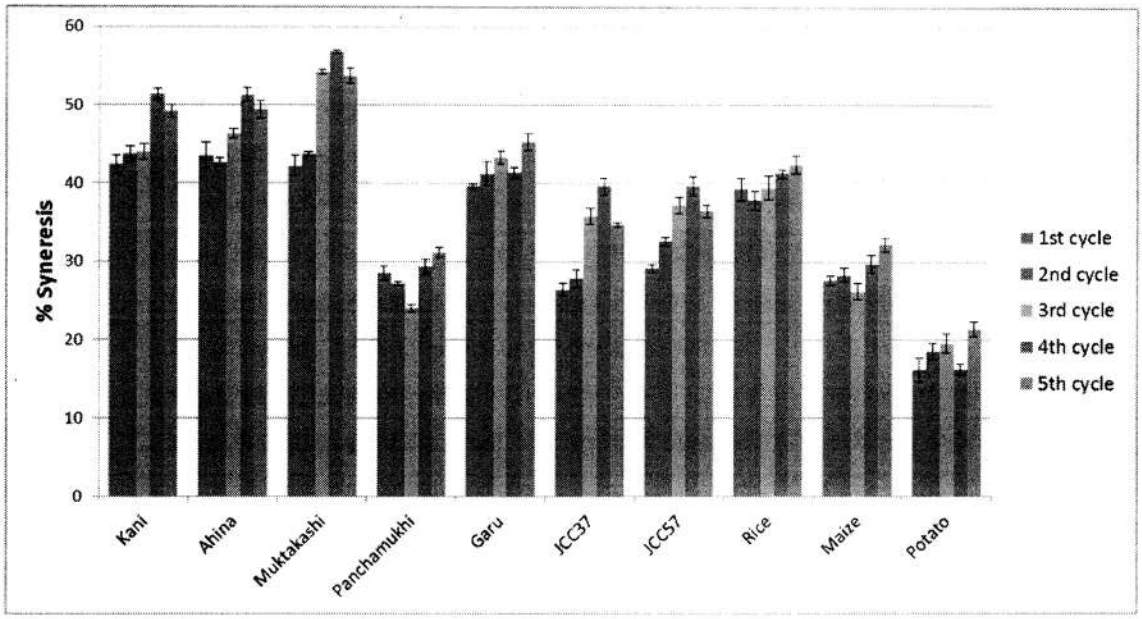


Fig. 10.8 Freeze-thaw stability of starch pastes from various taro cultivars, rice, maize and potato

Vertical error bars above the columns represent standard deviation (S. D.) of three replications

10.1.10 Texture properties

The texture parameters of the starch pastes are shown in Table 10.5. The firmness of the starch pastes from the taro cultivars varied from 11.17 to 11.93 g. The firmness of taro starch pastes did not differ significantly from each other. The firmness of the taro starch pastes was also not significantly different from the starch pastes of rice and maize with firmness of 11.38 and 11.71 g respectively. The firmness of potato starch paste was considerably higher than the other starch pastes. This might be due to higher swelling of potato starch granules. Similar trend was observed with consistency and cohesiveness of the starch pastes, where potato starch paste had the highest values of consistency and cohesiveness compared to the starch pastes of various taro cultivars and that of rice and maize. This might be due to larger granule size and higher viscosity of the potato starch as observed in SEM and pasting property analysis respectively. The indexes of viscosity of all the starch pastes were low signifying that the viscosities of the starch pastes change rapidly with change in temperature. The texture of the starch pastes are an important parameter in determining the application of a starch. It was observed that the firmness of the starch pastes were lower which will give a smooth texture to the products in which the taro starches will be incorporated.

10.1.11 Colour properties

The L*, a* and b* values of the starch pastes are shown in Table 10.5. It was observed that *Panchamukhi* and *Garu* starch pastes were significantly lighter than starch pastes of other taro cultivars. This might be due to the difference in composition and sizes of the starch granules. *Panchamukhi* starch had lesser impurities compared to other starches whereas the granule size of *Garu* and *JCC37* starches were larger and the lightness of the pastes are related to granule size and swelling power of the starches (Moorthy, 2002; Nwokocha et al., 2009). Potato starch paste exhibited highest lightness as the granules of potato starch are significantly larger than taro, rice or maize starches and the swelling power of the granules was also very high. The a* and b* values of all the starch pastes were not significantly different from each other and the values were positive, indicating that the starch pastes were slightly reddish and yellowish in nature. Starch paste colour is an

important property of starch for industrial application and light coloured pastes are preferred (Radley, 1976). The colours of the taro starch pastes were found to be comparable to the colour of rice and maize starch pastes. The colour quality of the starch pastes can be improved by application of certain chemicals which affects other functional properties of the starch (Rani et al., 1998). However, in certain food applications where light colour is not desirable like ketchups or sauces, native taro starches might find useful applications.

10.1.12 Colour of starch (dry powder)

Colour of dry starch is an important characteristic for determination of its quality. The colour parameters of the starch (dry powder) samples are presented in Table 10.6. The isolated starches had high L* values and lower a* and b* values which confirms the high purity of the starches. The L* value of *Panchamukhi* starch (95.88) was the highest and *Ahina* starch (92.29) recorded the lowest. Pérez Sira and Amaiz (2004) estimated that a value greater than 90 gives a satisfactory lightness for the purity of starch. Present results revealed that a* and b* values of the starches varied from 1.54 to 1.95 and 2.27 to 4.79 respectively for the various taro cultivars. Significant differences were not observed in the a* values. The b* values of *Kani, Ahina, Muktakashi* and *Garu* starches were higher compared to *Panchamukhi, JCC37 and JCC57* and the differences were significant indicating that *Kani, Ahina, Muktakashi and Garu* starches were slightly yellowish. The difference in the colour parameters among the taro starches might be attributed to the difference in the chemical composition of the starches. The colour of the dry starches of the taro cultivars were less lighter than the dry starches from rice and maize but were similar to that of potato starch. The high L* values combined with low a* and b* values of the taro starches evinced that it could be conveniently used in products requiring clear and uniform colour.

Table 10.5 Texture and colour parameters of starch pastes

Sample[1,2]	Texture properties				Colour parameters		
	Firmness (g)	Consistency (g.s)	Cohesiveness (g)	Index of viscosity (g.s)	L*	a*	b*
Kani	11.93±0.11^{b}	194.07±1.21^{b}	7.15±0.12^{c}	0.87±0.20^{b}	22.29±2.58^{def}	1.34±0.52^{abc}	3.83±0.85^{bc}
Ahina	11.57±0.56^{bc}	189.67±1.29^{cdc}	6.76±0.34^{f}	0.84±0.37^{b}	17.23±1.56^{f}	1.69±0.58^{ab}	2.03±0.54^{d}
Muktakashi	11.17±0.24^{c}	188.87±2.21^{dc}	7.88±0.12^{c}	1.04±0.21^{ab}	23.17±2.34^{dc}	1.13±0.12^{abc}	3.97±0.23^{b}
Panchamukhi	11.37±0.15^{bc}	188.36±1.04^{c}	7.66±0.18^{cd}	1.02±0.15^{ab}	26.31±2.31^{cd}	1.84±0.96^{a}	3.56±0.24^{bc}
Garu	11.93±0.56^{b}	195.33±0.96^{b}	7.42±0.21^{dc}	0.92±0.35^{ab}	27.04±3.21^{cd}	1.69±0.12^{ab}	5.02±0.54^{a}
JCC37	11.64±0.28^{bc}	190.61±2.12^{cdc}	7.57±0.26^{cd}	0.98±0.25^{ab}	22.19±3.47^{def}	1.34±0.47^{abc}	3.83±0.71^{bc}
JCC57	11.56±0.09^{bc}	189.93±1.08^{cdc}	8.46±0.17^{b}	1.25±0.19^{a}	18.84±2.54^{ef}	1.74±0.36^{ab}	4.32±0.11^{ab}
Rice	11.38±0.11^{bc}	191.59±0.58^{cd}	8.54±0.13^{b}	1.24±0.26^{a}	32.91±1.88^{b}	0.79±0.41^{c}	2.95±0.69^{cd}
Maize	11.71±0.57^{bc}	188.65±1.12^{c}	7.89±0.21^{c}	1.10±0.26^{ab}	29.51±3.14^{bc}	0.93±0.25^{bc}	4.25±0.37^{ab}
Potato	13.49±0.23^{a}	220.44±1.16^{a}	10.32±0.09^{a}	0.99±0.12^{ab}	44.49±5.02^{a}	1.09±0.36^{abc}	5.00±0.81^{a}

[1] Values reported as Mean ± S. D. of three replications

[2] Means followed by same small letter superscripts within a column are not significantly different (p 0.05)

Table 10.6 Colour parameters of starch (dry powder)

Sample[1, 2]	L*	a*	b*
Kani	94.38±0.54^{bc}	1.96±0.38^{a}	4.32±0.14^{bc}
Ahina	92.29±0.64^{d}	1.86±0.28^{a}	4.17±0.21^{c}
Muktakashi	92.41±0.51^{d}	1.95±0.54^{a}	4.58±0.15^{ab}
Panchamukhi	95.88±0.91^{b}	1.54±0.42^{a}	2.27±0.24^{g}
Garu	93.26±0.62^{cd}	1.85±0.45^{a}	4.79±0.12^{a}
JCC37	93.27±1.07^{cd}	1.67±0.23^{a}	2.95±0.09^{de}
JCC57	92.89±0.85^{cd}	1.81±0.27^{a}	3.22±0.18^{d}
Rice	97.64±1.56^{a}	1.67±0.43^{a}	2.67±0.19^{ef}
Maize	98.26±1.04^{a}	1.24±0.26^{a}	2.43±0.31^{fg}
Potato	95.19±0.25^{b}	1.71±0.77^{a}	3.13±0.17^{d}

[1]Values reported as Mean ± S. D. of three replications

[2]Means followed by same small letter superscripts within a column are not significantly different (p>0.05)

10.2 Quality parameters of tomato ketchup prepared using taro and maize starch

10.2.1 Effect of starch concentration and storage time on serum loss from tomato ketchup

The stability of tomato ketchup during storage is indicated by the amount of serum separated from the ketchup after storage. Lower serum loss i.e. ability to hold more water signifies better stability of ketchup during storage. Ketchups prepared by addition of starch from *Panchamukhi* taro and maize at 1 and 2% concentrations significantly lowered the serum loss as compared to the control sample throughout

the storage period (Fig. 10.9). The serum loss increased with increase in storage period for all the ketchup samples. Significant differences were observed in the values of serum loss in the control sample after 15 and 30 days of storage. Ketchups prepared using 1% *Panchamukhi* taro starch had higher serum loss after 15 and 30 days than ketchup prepared by incorporating 1% maize starch, although significant differences were not observed on 0 day i.e. before storage. Stability of ketchups containing 2% starches were found to be better than ketchups containing 1% starches. The stability of ketchup sample prepared using 2% *Panchamukhi* starch was found to be better compared to that prepared by using 2% maize starch, as the difference in serum loss in ketchup containing 2% *Panchamukhi* starch at 0, 15 and 30 days after storage were very less and statistically non-significant.

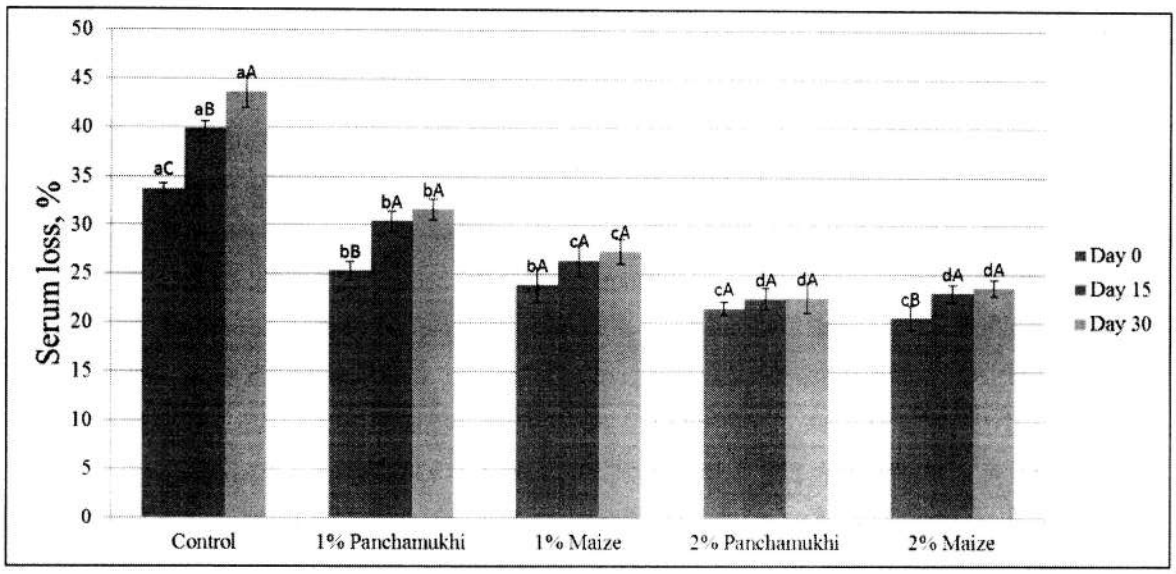


Fig. 10.9 Serum loss during storage from the ketchup samples prepared by incorporation of Panchamukhi and maize starch at different concentrations

Vertical error bars above the columns represent standard deviation (S. D.) of three replications

Similar small letters above the bars denote values are not significantly different ($p>0.05$) between different ketchup samples for same storage period

Similar capital letters above the bars denote values are not significantly different ($p>0.05$) between different storage period for the same ketchup sample

10.2.2 Effect of starch concentration and storage time on texture properties of tomato ketchup

The maximum force experienced by the probe during penetration is taken as the measurement of firmness of the sample and is used as an index of textural quality (Bourne, 2002). The firmness of the different ketchup samples prepared by incorporating *Panchamukhi* and maize starch is given in Fig. 10.10. The firmness of the ketchups significantly increased due to incorporation of starch which acts as a thickening agent. The firmness also increased as the concentration of the starch in the ketchup samples were increased from 1 to 2%. The firmness of the ketchups containing maize starch was much higher compared to that of *Panchamukhi* starch at same concentration. This might be due to the higher final viscosity of maize starch as observed in RVA profile. Not much difference was observed in the firmness of the ketchups containing starch as when compared to the control sample. The firmness of the control sample decreased significantly after 15 days of storage, which might be due to loss of serum from the ketchup as in the absence of any binding agent (stabilizer or thickener) the solids in the control sample were not able to hold the water and it separated out (Gujral et al., 2002). No significant differences were observed up to 30 days of storage with the ketchups prepared with starch, indicating that both *Panchamukhi* and maize starch can be used as a thickener in ketchups.

Consistency is another important parameter in determining the quality of ketchup. If the consistency is very high the ketchup might not flow properly from the container, and if it is very low ketchup might not give the desired mouth feel. Fig.10.11 shows that consistencies of the ketchups prepared using starch were significantly higher than that of control. Consistency increased with increase in concentration for ketchups prepared using *Panchamukhi* starch, but for maize starch both the concentration gave similar consistencies and was close to that of ketchup containing 2% *Panchamukhi* starch. The consistency of the control ketchup decreased significantly after 15 days, but significant differences were not observed in the other ketchup samples.

The cohesiveness and viscosity of the ketchup samples also increased due to incorporation of starch indicating that more force is required to overcome the

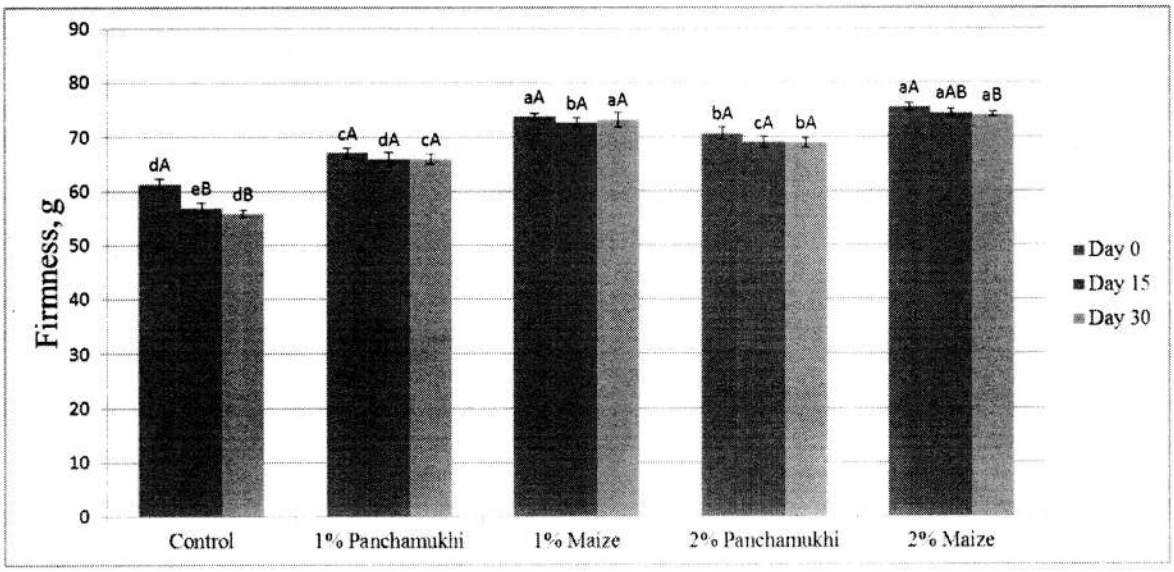


Fig. 10.10 Variation in firmness of different ketchup samples with days of storage
Vertical error bars above the columns represent standard deviation (S. D.) of three replications
Similar small letters above the bars denote values are not significantly different ($p>0.05$) between different ketchup samples for same storage period
Similar capital letters above the bars denote values are not significantly different ($p>0.05$) between different storage period for the same ketchup sample

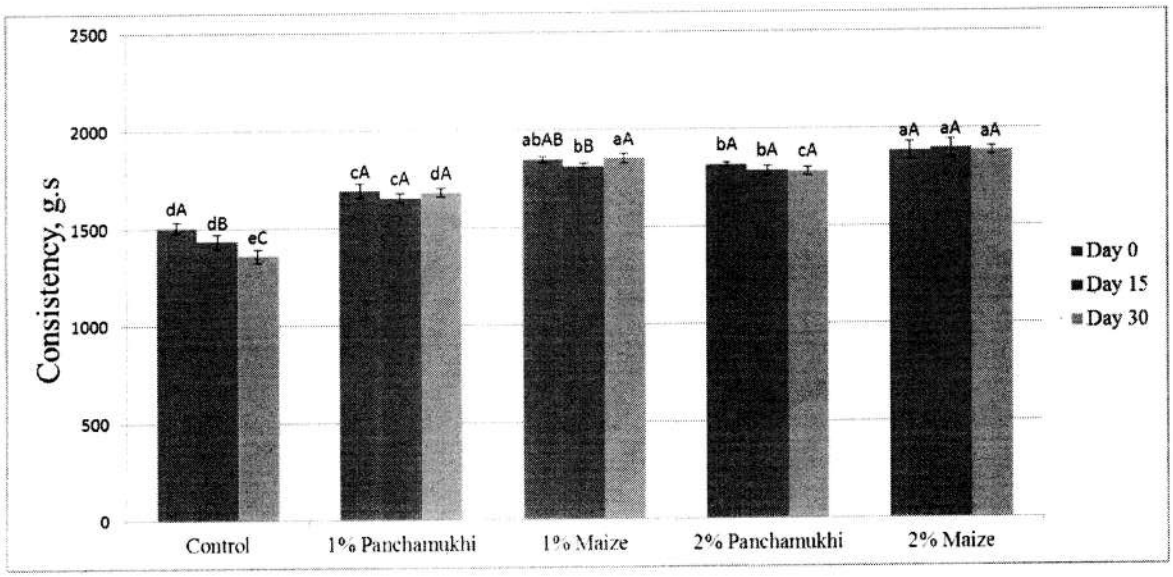


Fig. 10.11 Variation in consistency of different ketchup samples with days of storage

Vertical error bars above the columns represent standard deviation (S. D.) of three replications

Similar small letters above the bars denote values are not significantly different ($p>0.05$) between different ketchup samples for same storage period

Similar capital letters above the bars denote values are not significantly different ($p>0.05$) between different storage period for the same ketchup sample

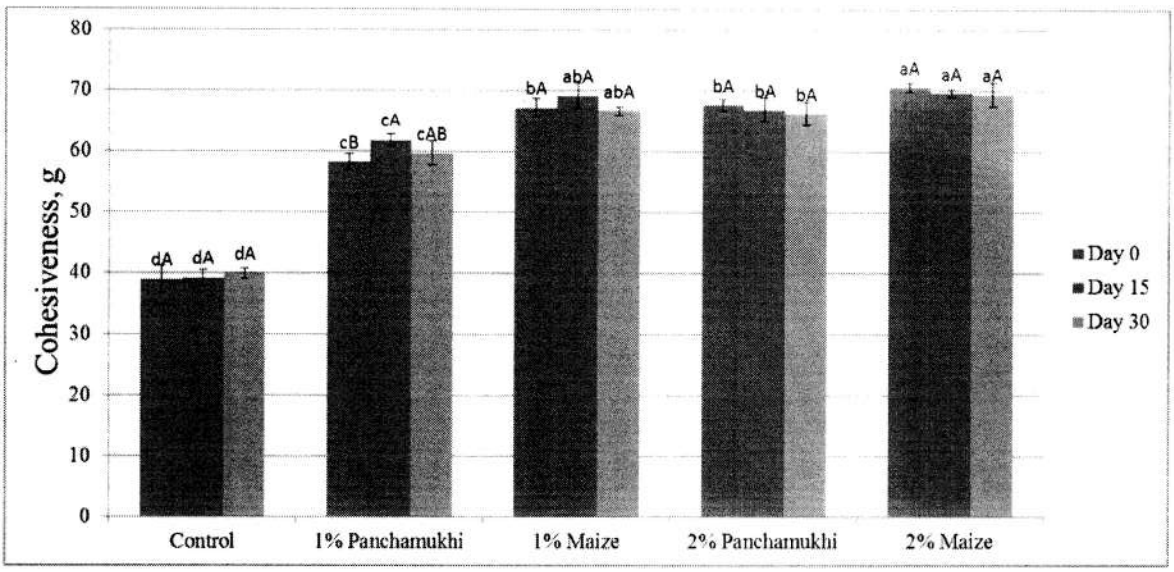


Fig. 10.12 Variation in cohesiveness of different ketchup samples with days of storage
Vertical error bars above the columns represent standard deviation (S. D.) of three replications
Similar small letters above the bars denote values are not significantly different ($p>0.05$) between different ketchup samples for same storage period
Similar capital letters above the bars denote values are not significantly different ($p>0.05$) between different storage period for the same ketchup sample

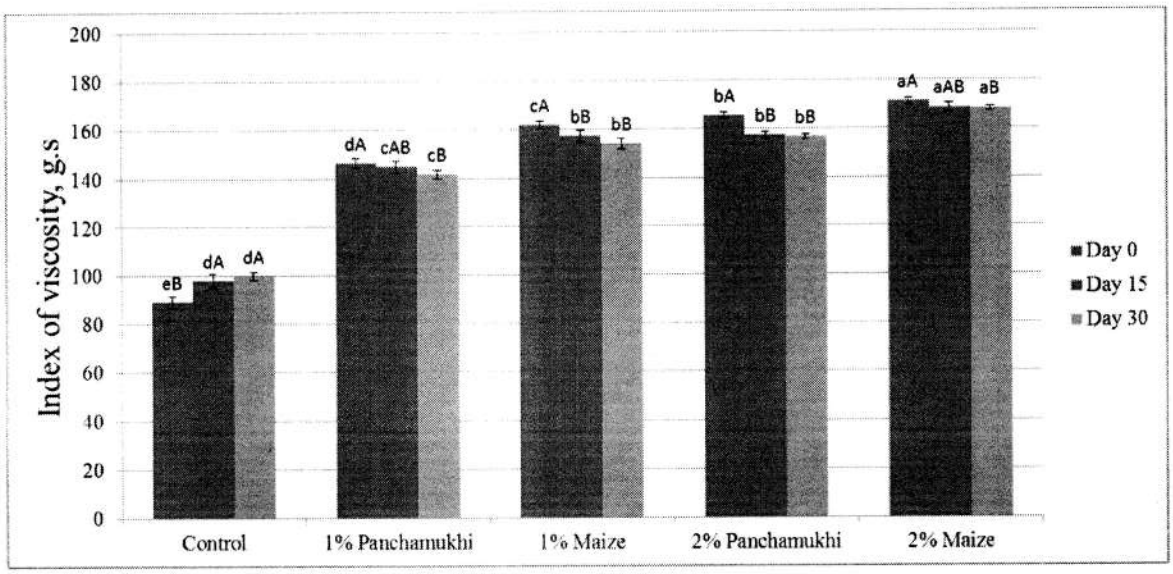


Fig. 10.13 Variation in index of viscosity of different ketchup samples with days of storage
Vertical error bars above the columns represent standard deviation (S. D.) of three replications
Similar small letters above the bars denote values are not significantly different ($p>0.05$) between different ketchup samples for same storage period
Similar capital letters above the bars denote values are not significantly different ($p>0.05$) between different storage period for the same ketchup sample

resistance during pulling out the sample (Liu et al., 2007). The variation in cohesiveness and index of viscosity with storage time for the different ketchup samples is given in Fig. 10.12 and Fig. 10.13 respectively. Cohesiveness values of the ketchup samples prepared by incorporation of the starches were significantly higher compared to the control sample for all storage periods. The cohesiveness of the of the ketchups containing 2% starch were higher than that containing 1% starch for a particular type of starch, although the differences between ketchups containing 1% and 2% maize starch were less as compared to between 1% and 2% *Panchamukhi* starch incorporated ketchups. Further it was observed that cohesiveness of ketchup containing 2% *Panchamukhi* starch was not significantly different form that containing 1% and 2% maize starch. The results suggest that increasing the concentration of *Panchamukhi* starch is more effective in controlling the texture of tomato ketchup compared to maize starch. No significant differences were observed in the cohesiveness of a particular ketchup sample during storage. Significant increase in index of viscosity was observed for the control sample after 15 days. This might be attributed to higher serum loss from the control sample and aggregation of the solids which resulted in higher force during back extrusion. The index of viscosities of ketchup samples containing 2% starches were more compared to those containing 1% starches, which shows that change in viscosity with temperature is more in ketchups containing 2% starches.

10.2.3 Effect of starch concentration and storage time on colour of tomato ketchup

The L*, a* and b* values of the ketchups prepared using *Panchamukhi* and miaze starch were not found to be significantly different from the control ketchup (Table 10.7). Although, L*, a* and b* values increased when both the starches were incorporated at 2% concentration. Statistically significant differences were not observed in the ketchup samples up to 30 days of storage. The results revealed that addition of starch up to 2% concentration might not affect the colour of tomato ketchups.

10.2.4 Effect of starch concentration on sensory quality of tomato ketchup

Sensory scores (Table 10.8) revealed that statistically significant differences from control were not present in colour, and flavour of tomato ketchup prepared by incorporation of *Panchamukhi* taro or maize starch. Ketchup containing 1%

Table 10.7 Change in colour of ketchups incorporated with *Panchamukhi* taro and maize starch during storage

Ketchup Sample[1, 2, 3]	L*			a*			b*		
	0 Day	15 Day	30 Day	0 Day	15 Day	30 Day	0 Day	15 Day	30 Day
Control	31.63±1.26^{aA}	31.64±1.56^{aA}	31.58±0.99^{aA}	12.68±0.96^{aA}	12.84±1.06^{aA}	12.90±0.98^{aA}	9.95±0.59^{aA}	9.65±0.57^{aA}	9.72±0.62^{aA}
1% *Panchamukhi*	31.91±1.23^{aA}	31.82±1.12^{aA}	31.88±1.07^{aA}	12.50±0.59^{aA}	12.28±0.78^{aA}	12.30±0.54^{aA}	9.85±0.87^{aA}	9.56±0.97^{aA}	9.03±0.49^{aA}
2% *Panchamukhi*	32.79±0.69^{aA}	32.87±0.98^{aA}	32.80±0.84^{aA}	12.95±0.96^{aA}	13.09±.83^{aA}	13.74±1.12^{aA}	10.02±1.02^{aA}	10.21±0.67^{aA}	10.12±0.96^{aA}
1% Maize	31.68±0.98^{aA}	31.64±0.59^{aA}	31.54±0.67^{aA}	12.09±1.02^{aA}	12.47±0.42^{aA}	12.53±0.58^{aA}	9.57±0.49^{aA}	9.52±0.59^{aA}	9.21±0.59^{aA}
2% Maize	32.72±1.09^{aA}	32.51±1.42^{aA}	32.30±1.16^{aA}	13.18±0.77^{aA}	13.16±0.69^{aA}	13.25±1.07^{aA}	10.11±0.67^{aA}	10.04±1.02^{aA}	9.71±0.52^{aA}

[1]Values reported as Mean ± S. D. of three replications
[2]Means followed by same superscript capital letters within a row are not significantly different for a particular colour parameter ($p>0.05$)
[3]Means followed by same superscript small letters within a column are not significantly different ($p>0.05$

Table 10.8 Sensory scores of ketchups prepared using starch

Ketchup Samples[1,2]	Colour	Smoothness	Flavor	Mouthfeel
Control (Without Starch)	7.43±0.16[a]	7.24±0.21[b]	7.20±1.05[a]	7.28±0.06[d]
1% *Panchamukhi*	7.35±0.13[a]	7.59± 0.17[a]	7.15±0.60[a]	8.34±0.12[a]
1% Maize	7.31±0.10[a]	7.54±.19[a]	7.19±01.25[a]	8.29±0.13[a]
2% *Panchamukhi*	7.39±0.17[a]	7.14±0.15[b]	7.09±0.09[a]	8.26±0.[15ab]
2% Maize	7.11±0.09[b]	7.20±0.08[b]	7.12±0.29[a]	7.75±0.18[c]

[1]Scores reported as Mean ± S. D. of 15 semi-trained panelists

[2]Means followed by same superscript capital letters within a column are not significantly different ($p<0.05$)

Panchamukhi taro and maize starch were smoother compared to control or ketchup containing 2% starch from *Panchamukhi* and maize. The scores for mouthfeel of tomato kethchups containing 1 and 2% *Panchamukhi* taro starch and 1% maize starch were significantly higher compared to control sample and ketchup prepared using 2% maize starch and the differences were statistically significant. Ketchup containing 2% maize starch was felt more firm by the panellists and was not liked in terms of mouthfeel. *Panchamukhi* starch might be used in preparation of tomato ketchup for improving the texture and mouthfeel without affecting the acceptability.

10.3 Optimization of starch extraction from taro tubers and comparison of properties of enzymatically and conventionally isolated starches

10.3.1 Model fitting

The values of starch yield for different experimental combinations are shown in Table 10.9. Multilinear regression analysis of the data yielded second order polynomial equations for starch yield. Analysis of variance (ANOVA) was performed to determine the significant effects of the process variables on starch yield. Regression coefficients of the different terms in the equations for starch yield in coded factors were obtained (Table 10.10). Model adequacy was checked by lack of fit test and by considering fitted R^2, predicted R^2, PRESS and adequacy precision. A

non- significant ($p > 0.05$) lack of fit, predicted R^2 comparable to fitted R^2, low PRESS and adequacy precision higher than 4, implies that the model fitted is adequate to predicting (Corzo et al., 2008; Erbay and Icier, 2009).

It can be observed from Table 2 that the probability (p) values of the model, all the main factors, quadratic term for only temperature (T^2) and interaction of t×C, t×X, and C×X for the response significant and the model has non- significant lack of fit ($p>0.05$) with *p-value* of 0.1404, which is good, and also the adequacy precision for the model was more than 4. The R^2 value of the model was 0.97, whereas the adjusted R^2 (0.95) and predicted R^2 (0.86) were comparable indicating that the model fitted provided appropriate approximation of the true process.

The final equations in terms of coded factors are as follows:

Starch yield, $Y = 15.20 + 0.21t + 0.32T + 0.70C + 0.49X - 0.05t^2 + 0.19T^2 + 0.08C^2 + 0.03X^2 + 0.08t\times T - 0.20t\times C - 0.23t\times X - 0.10T\times C - 0.10T\times X - 0.25C\times X$ Eqn. 2

10.3.2 Effect of interaction of various factors and their interactions on starch yield

The variation in starch yield with time and temperature is shown in Fig. 10.14 a). Starch yield increased with incubation period but decreased as the temperature was increased. Shah (2007) also reported similar findings for incubation period in extraction of litchi juice using enzymes and Guan and Yao (2008) observed similar changes with temperature for extraction of protein from oat bran using enzymes. The decrease in yield of starch with increase in temperature might be attributed to the loss of activity of the enzymes at higher temperature and resulted in lower breakdown of the cell wall components and thereby releasing lower amount of starch. Another possible reason might be that, at higher temperatures some amount of starch might became solubilised in the water and could not be recovered during centrifugation.

Table 10.9 Starch yield for different combinations of experimental conditions

Experiment No.	Time (t), h	Temp (T), °C	Cellulase (C), U/100g tuber	Xylanase (X), U/100g tuber	Yield,%
1	1(-2)[1]	40(0)	200(0)	200(0)	14.46
2	2(-1)	35(-1)	100(-1)	100(-1)	13.56
3	2(-1)	35(-1)	100(-1)	300(1)	15.67
4	2(-1)	35(-1)	300(1)	100(-1)	15.99
5	2(-1)	35(-1)	300(1)	300(1)	17.49
6	2(-1)	45(1)	100(-1)	100(-1)	13.17
7	2(-1)	45(1)	100(-1)	300(1)	15.16
8	2(-1)	45(1)	300(1)	100(-1)	15.48
9	2(-1)	45(1)	300(1)	300(1)	15.94
10	3(0)	30(-2)	200(0)	200(0)	16.57
11	3(0)	40(0)	0(-2)	200(0)	13.99
12	3(0)	40(0)	200(0)	0(-2)	14.29
13	3(0)	40(0)	200(0)	200(0)	15.19
14	3(0)	40(0)	200(0)	200(0)	15.31
15	3(0)	40(0)	200(0)	200(0)	15.11
16	3(0)	40(0)	200(0)	200(0)	14.96
17	3(0)	40(0)	200(0)	200(0)	15.24
18	3(0)	40(0)	200(0)	200(0)	15.38
19	3(0)	40(0)	200(0)	400(2)	16.02
20	3(0)	40(0)	400(2)	200(0)	16.69
21	3(0)	50(2)	200(0)	200(0)	15.05
22	4(1)	35(-1)	100(-1)	100(-1)	14.78
23	4(1)	35(-1)	100(-1)	300(1)	16.02
24	4(1)	35(-1)	300(1)	100(-1)	16.45
25	4(1)	35(-1)	300(1)	300(1)	16.61
26	4(1)	45(1)	100(-1)	100(-1)	14.65
27	4(1)	45(1)	100(-1)	300(1)	15.54
28	4(1)	45(1)	300(1)	100(-1)	15.96
29	4(1)	45(1)	300(1)	300(1)	16.03
30	5(2)	40(0)	200(0)	200(0)	15.21

[1]Numbers in parentheses correspond to coded values of different levels of factors

Table 10.10 Analysis of variance (ANOVA) for fitted model of starch yield

Source	Sum of Squares	DF	Mean Square	F-Value	p-value
Model	25.45	14	1.82	37.83	< 0.0001*
t	1.08	1	1.08	22.38	0.0003*
T	2.46	1	2.46	51.15	< 0.0001*
C	11.76	1	11.76	244.75	< 0.0001*
X	5.88	1	5.88	122.39	< 0.0001*
t^2	0.07	1	0.07	1.42	0.2527
T^2	1.03	1	1.03	21.47	0.0003*
C^2	0.16	1	0.16	3.34	0.0877
X^2	0.03	1	0.03	0.52	0.4816
t×T	0.10	1	0.10	2.13	0.1650
t×C	0.67	1	0.67	13.99	0.0020*
t×X	0.86	1	0.86	17.81	0.0007*
T×C	0.16	1	0.16	3.41	0.0845
T×X	0.16	1	0.16	3.33	0.0880
C×X	1.02	1	1.02	21.23	0.0003*
Residual	0.72	15	0.05		
Lack of Fit	0.61	10	0.06	2.72	0.1404
Pure Error	0.11	5	0.02		
Correlation Total	26.17	29			
R^2	0.97				
Adjusted R^2	0.95				
Predicted R^2	0.86				
Adequate Precision	26.21				
PRESS	3.67				

t – Time: T – Temperature: C – Cellulase concentration: X – Xylanase concentration

* Terms significant at $p<0.05$

Starch yield increased with increase in concentration of cellulase/xylanase for all incubation periods [Fig. 10.14 b) and Fig. 10.14 c)]. No significant variation was observed in starch yield with change in time at higher concentration of the enzymes, but at lower

concentration starch yield increased with increase in incubation time, indicating that less time is required to hydrolyze the cell wall components when concentration of the enzymes were high. The starch yield was constant with incubation time at higher concentration of the enzymes. However, it was always found to be higher at high concentration of enzymes than at low concentration, even at higher incubation times. Similar findings were reported for cassava starch yield by Dzogbefia et al. (2008) using pectinase enzyme and Shah (2007) for litchi juice extraction. Starch yield was found to be higher when the concentration of the enzymes was high. This was observed regardless of incubation time. This shows that higher yield of starch similar to that when enzyme concentrations were high, cannot be achieved by keeping low concentration of the enzymes even when the incubation time is increased. This might be due to inhibition of the enzymes by the degradation products of the enzymes i.e. glucose and xylose. When the enzymes were present at lower concentrations most of the enzymes might be inhibited by the products and therefore could never give similar yield comparable with higher concentration of enzymes even when the incubation time is increased.

From Fig. 10.14 d) and Fig. 10.14 e) it can be observed that starch yield decreased with increase in temperature. At lower temperature the variation in starch yield was more with change in concentration of enzyme as compared to higher temperature. This might be attributed to inactivation of the enzymes or solubilization of some starch at higher temperatures thereby decreasing the yield.

It has already been observed that starch yield increased with increase in concentration of both the enzymes and combination of the two enzymes significantly increased the yield of starch from the tubers [Fig. 10.14 f)]. The effectiveness of xylanase in increasing the recovery of starch from taro tubers clearly indicates that xylan is an important component of the cell walls of taro tubers and degradation of xylan is necessary for releasing starch granules from the cells of taro.

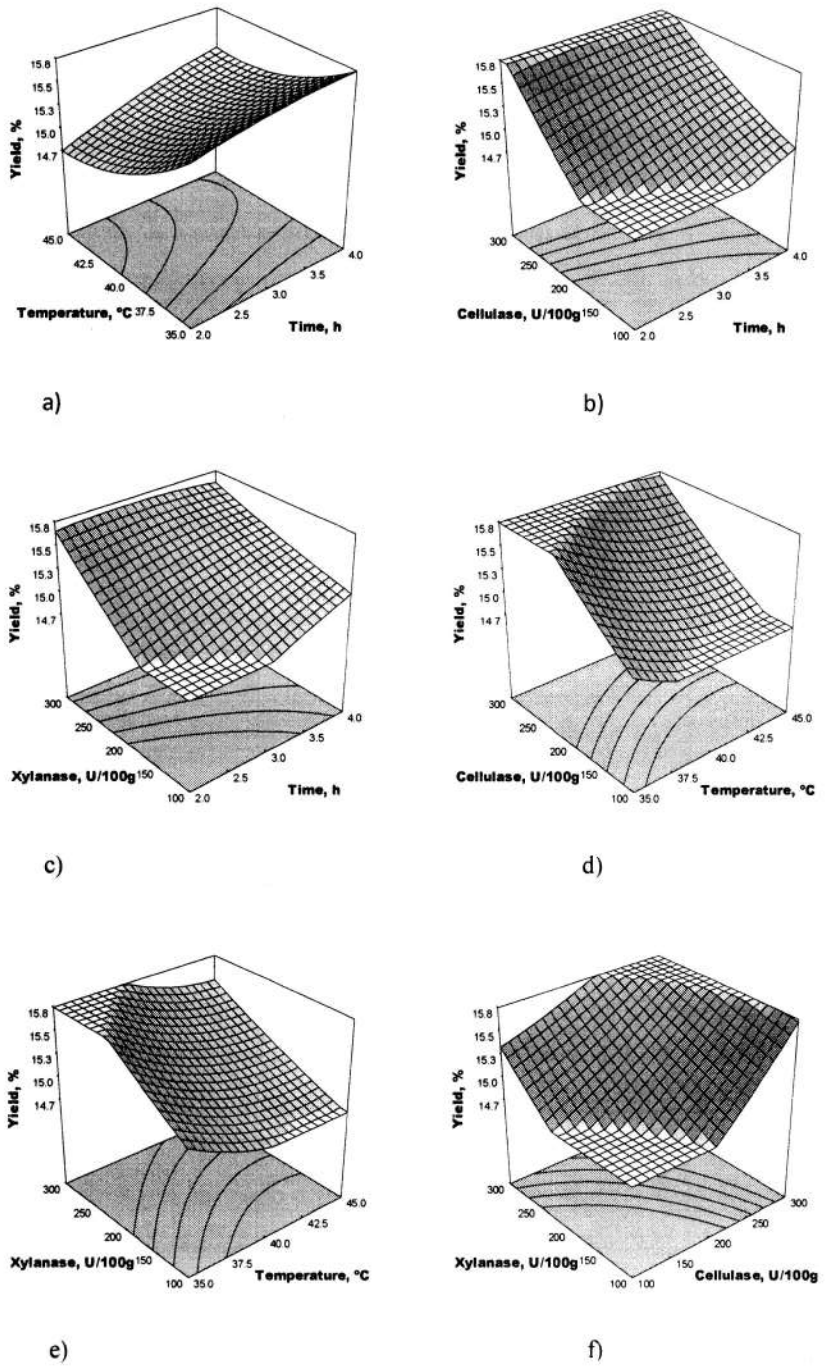


Fig. 10.14 Variation in starch yield due to interaction of a) temperature and incubation time, b) concentration of cellulase and incubation time, c) concentration of xylanase and incubation time, d) concentration of cellulase and temperature, e) concentration of xylanase and temperature, and f) concentration of xylanase and cellulase

10.3.3 Optimization of enzymatic starch isolation process from taro tuber

Optimization of starch isolation process from taro tubers using cellulase and xylanase was done to obtain maximum yield of starch. The independent parameters selected were cellulase concentration (C), xylanase concentration (X), temperature of incubation (T) and incubation time (t). The cellulase and xylanase concentration were varied from 0 to 100 U/ 100 g taro tuber, incubation time was varied from 1 to 5 h and temperature was varied from 30 °C to 50 °C. Optimization was done using desirability function. One possible solution with highest desirability was selected (Table 10.11). The optimum condition with highest yield of 17.22% was obtained when cellulase and xylanase concentration were 299.86 and 300 U/100g tuber respectively, temperature was 35 °C and incubation time was 2 h.

Table 10.11 Results of optimization of starch recovery by desirability function

Sl. No.	Time, h	Temperature, °C	Cellulase, U/100g tuber	Xylanase, U/100g tuber	Yield, %	Desirability
1	2.00	35.00	299.86	300.00	17.22	0.94

10.3.4 Functional properties of starch isolated by enzymatic and conventional methods

The data for swelling, solubility and clarity of the starch pastes are presented in Table 10.12 and pasting properties is presented in Table 10.13 for starches isolated by enzymatic and conventional methods. It was observed that starch isolated by enzymatic method swelled significantly more as compared to conventional method. On the contrary, starch isolated by conventional method had higher solubility compared to enzymatic method. Dzogbefia et al. (2008) reported lower swelling and higher solubility for cassava starch using pectinase as opposed to the present finding, and the variation might be due to the use of different kind of enzyme. Correia et al. (2012) and Puchongkavarin et al. (2005) also observed similar increase in swelling for chestnut starch and rice starch respectively with enzymatic methods. The higher swelling and lower solubility in the present investigation might be attributed to loss of amylose in enzymatic method which might have leached out during incubation. Amylose hinders swelling and contributes to the soluble portion of the starch and loss of amylose affected the swelling and solubility.

The clarity of the starch pastes extracted by enzymatic method was found to be more as compared to conventional method. The increase in clarity of the starch pastes due to

enzymatic treatment might be attributed to the breakdown of the cell wall components like cellulosic and hemicellulosic fibres into soluble fragments which were removed during centrifugation or else it can contribute to the impurities of the isolated starch.

Table 10.12 Swelling, solubility and clarity of starch samples isolated by conventional and enzymatic methods

Isolation method[1,2]	Swelling, g/g	Solubility, %	Clarity, %T
Conventional	13.32±0.43[b]	20.21±0.12[a]	32.67±1.21[b]
Enzymatic	14.95±0.31[a]	19.08±0.27[b]	38.94±1.74[a]

[1]Values reported as Mean ± S. D. of three replications

[2]Means followed by same small letters within a column are not significantly different ($p>0.05$)

The pasting properties of the starches isolated by the two methods evinced no significant differences in pasting temperature, PV, HV, FV, SV and BV values, although the pasting temperature was slightly higher and viscosities were lower in the starch pastes isolated by enzymatic method. This might be due to loss of amylose in enzymatic method which might have increased the viscosity of the starch pastes. Dzogbefia et al. (2008) also did not observe significant differences in pasting properties in cassava starch isolated by enzymatic and conventional methods. The results indicate that enzymatic treatment has no adverse effect on the pasting properties of the starch which could be used to achieve higher recovery of starch.

Table 10.13 Pasting properties of starch isolated by conventional and enzymatic methods

Isolation method[1,2]	Pasting Temperature, °C	Peak Viscosity, cP	Hold Viscosity, cP	Final Viscosity, cP	Breakdown Viscosity, cP	Setback Viscosity, cP
Conventional	84.4±0.30[a]	4333±120[a]	2218±134[a]	2868±97[a]	2115±89[a]	650±34[a]
Enzymatic	84.6±0.40[a]	4272±147[a]	2126±156[a]	2697±108[b]	2146±67[a]	571±53[b]

[1]Values reported as Mean ± S. D. of three replications

[2]Means followed by same small letters within a column are not significantly different ($p>0.05$)

The freeze-thaw stability of the starch gels were evaluated up to three freeze-thaw cycles and are shown in Figure 10.15. The freeze-thaw stability of the starch pastes isolated

by enzymatic method was more stable as the % syneresis was lower and the differences were less among the three freeze-thaw cycles. Differences in freeze-thaw stability among different types of starches might be due to a variety of factors, most notably, amylose content (Baker and Rayas-Duarte, 1998). The better freeze thaw stability of the enzymatically isolated starch might be attributed to loss of amylose during incubation, and lower amylose corresponds to lower retrogradation tendency, thereby lowering syneresis (Ciacco and D'Appolonia, 1977).

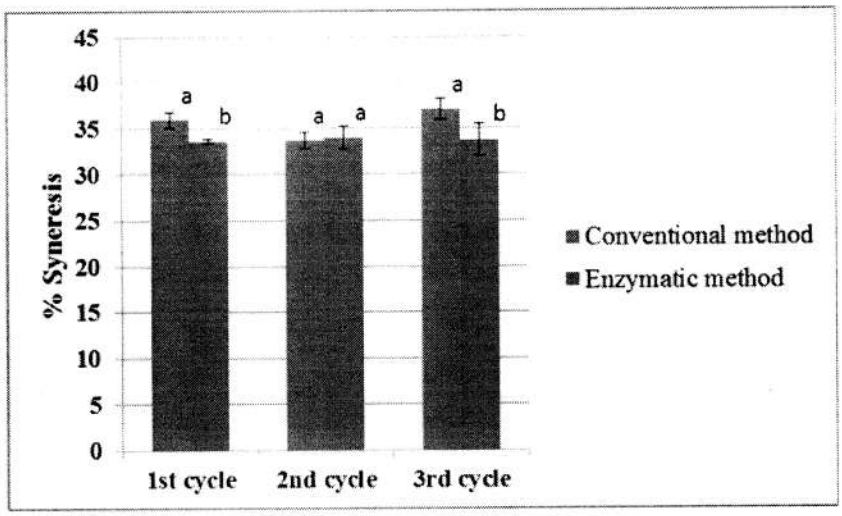


Fig. 10.15 Freeze-thaw behaviour of starches isolated by conventional and enzymatic methods

Vertical error bars above the columns represent standard deviation (S. D.) of three replications

Columns separated by same small letters within a cycle are not significantly different ($p>0.05$)

11. Conclusions summarising the achievements and indication and scope for future work:

11.1 Conclusions

The study revealed that the physicochemical, functional, textural and colour properties of starches of various taro cultivars of Assam from North-East India are different from taro varieties available in other regions of the world. The properties of taro starch were found to be closer to rice and maize starch in many respects, evincing its potential as an ingredient in food processing and other industries. The high gelatinization

and pasting temperatures coupled with low viscosity make them suitable for applications in food products which are subjected to heating at higher temperature, wherein change in viscosity is not desirable during heating and cooling. However, the poor freeze-thaw stability of these starches makes them inadequate as thickener and stabilizer for refigerated and frozen foods. It addition it would be prudent to mention that starch from *Panchamukhi* taro has better functional properties like better clarity and stability, pasting properties, and freeze-thaw stability as compared to the starches from the other taro cultivars investigated.

Incorporation of *Panchamukhi* taro and maize starch as thickeners in preparation of tomato ketchup significantly reduced the serum loss compared to control sample, thereby increasing the stability of the tomato ketchups. Texture, colour and sensory evaluation of the ketchups incorporated by *Panchamukhi* taro and maize starch showed significant improvement in the texture properties like firmness and consistency without affecting the colour. The ketchup containing 2% *Panchamukhi* taro starch was found to be more acceptable to the consumers than the other ketchup samples indicating that *Panchamukhi taro* starch has potential for other food and non-food applications also.

The study revealed that higher yield of starch from taro tubers could be achieved using cellulase and xylanase. However, combination of both the enzymes yielded significantly higher amount of starch. Effectiveness of xylanase indicated that xylan is an important component of the cell walls of taro tubers and removal of xylan from the cell walls of taro is necessary in achieving higher yield of starch from taro. The highest yield of starch was achieved when both time and temperature were low and concentrations of the enzymes were high. It indicated that no major modification of the properties of starch is possible as both time and temperature are low. The study further revealed that higher yield of starch from taro tubers could be obtained using enzymes without much affecting the properties of native starch.

11.2 Scope for future work

- Other enzymes may be used for extraction of starch.
- Enzymatic treatments might be combined with other treatments like microwave, ultrasound etc. to further increase the efficiency.
- Structural changes in the amylose or amylopectin molecules due to ultrasonication or enzymatic treatment might be investigated.

- The effect of incorporation of starches extracted by different methods might be examined on the quality parameters of tomato ketchup.

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12. S&T benefits accrued:

i. List of Research publications

Sl. No.	Authors	Title of paper	Name of the Journal	Volume	Pages	Year
1	Nandan Sit, Sudip Misra, Sankar Chandra Deka	Physicochemical, functional, textural and colour characteristics of starches isolated from four taro cultivars of North-East India	Starch/Starke	65	1011-1021	2013
2	Nandan Sit, Sudip Misra, Devastuti Baruah, Laxmikant S. Badwaik, Sankar Chandra Deka	Physicochemical properties of taro and maize starch and their effect on texture, colour and sensory quality of tomato ketchup	Starch/Starke	66	294-302	2014
3	Nandan Sit, Sudip Misra, Sankar Chandra Deka	Characterization of Physicochemical, Functional, Textural and Color Properties of Starches from Two Different Varieties of Taro and Their Comparison to Potato and Rice Starches	Food Science and Technology Research	20	357-365	2014
4	Nandan Sit, Sankar Chandra Deka, Sudip Misra,	Optimization of starch isolation from taro using combination of enzymes and comparison of properties of starches isolated By enzymatic and conventional methods	Journal of Food Science and Technology	52	4324-4332	2015

ii. Manpower trained on the project

a) Research Scientists or Research Associates

b) No. of Ph.D, produced

c) Other Technical Personnel trained – Two (02) SRFs

iii. Patents taken, if any – None

13. Financial Position:

No.	Financial Position/ Budget Head	Funds Sanctioned	Expenditure	% of Total Cost
I	Salaries/Manpower	Rs. 529200/-	Rs. 223833/-	8.71%
II	Equipment	Rs. 1888425/-	Rs. 1878594/-	73.13%
III	Consumables	Rs. 450000/-	Rs. 450138/-	17.53%
IV	Contingencies	NIL	NIL	N.A.
V	Travel	Rs. 16157/-	Rs. 16157/-	0.63%
VI	Institutional Charges	NIL	NIL	N. A.
	Total	Rs. 2613782/-	Rs. 2568772/-	100%

14. Procurement/Usage of equipment

a)

S No.	Name of Equipment	Make/Model	Cost (FE/Rs.)	Date of Installation	Utilization Rate (%)	Remarks regarding maintenance/ breakdown
1.	Starch Viscosity Analyzer	LAB-KITS, NDJ-5S	Rs. 948500/-	25/02/13	60%	Working
2.	Vacuum Oven	K&K, KVO64	Rs. 292000/-	22/01/13	80%	Working
3.	Muffle Furnace	K&K, KMF03	Rs. 196000/-	22/01/13	80%	Working
4.	Refrigerated Centrifuge	Sigma-SVI, 3K15	Rs. 442094/-	28/01/13	95%	Working

b) Plan for utilizing the equipment facilities in future

The equipment will be utilized for carrying out research of post graduate students of the department.

Name and Signature with Date

a. 23.12.15. (Nandan Si

(Principal Investigator)

b. LS Badwaik 23.12.15 (L.S. Badwaik)

(Co-Investigator)

Date:

Dean, Research & Development,
Tezpur University

संकायाध्यक्ष, शोध एवं विकास
तेजपुर विश्वविद्यालय
Dean, Research & Development
Tezpur University

ANNEXURE- D

PROFORMA FOR GFR 19-A

(See Government of India's Decision (1) below rules 150) Form of Utilization Certificate

Sr. No.	Letter No. and Date	Amount
1	10/MFPI/R&D/2011 Dated 28th Dec. 2011	Rs. 22,82,425/-
2	10/MFPI/R&D/11 Dated 13th May, 2013	Rs. 3,31,357/-
		Total= Rs. 26,13,782/-

Certified that out of Rs. 26,13,782/- of Grant-in-aid sanctioned during the year 2011-13 in favour of the Registrar, Tezpur University, Napaam-784028, Tezpur/ Department of Food Engineering & Technology, Assam under this Ministry letter no. 10/MFPI/R&D/2011 Dated 28th Dec. 2011 and 13th May, 2013 given in the margin, a sum of Rs. 25,68,722/- has been utilized for the purpose of purchase of equipments, chemicals, glasswares, raw materials, and expenses of manpower for which it was sanctioned and that the balance of Rs. 45,060/- remaining unutilized at the end of the year has been surrendered to the government will be adjusted towards the grant-in-aid payable during the next year 2015.

1. Certified that I have satisfied myself that conditions on which the grant-in-aid was sanctioned have duly been fulfilled/ are being fulfilled and that I have exercised the following checks to see that the money was actually utilized for the purpose for which it was sanctioned.

Kinds of checks exercised:

1. Accounts audited by qualified chartered accountant appointed by this University as Internal Auditor.
2. All the equipments, chemicals, glasswares and consumables etc. purchased from the grant are entered in the stock register.

Signature:
Designation:
Date:

21.5.15

22/5/15

Statement of Expenditure

Project Title: Development of enzyme based extraction process for improving quality and recovery of starch from different varieties of *Colocasia esculenta* (Arbi) of Assam for food use

File No. 10/MFPI/R&D/2011

Duration: 28/12/2011 to 15/03/2015

Grant-in-aid received:

1st Instalment- Rs. 2282425/-

2nd Instalment- Rs. 331357/-

Toal Amount Received - Rs. 2613782/-

Amount spent- Rs. 2568722/-

Details of expenditure

Sl. No.	Particulars	Purchase order nos.	Amount
1	Starch Viscosity Analyzer	TU/11-15/Pur/FPT/2010-755	Rs. 948500/-
2	Refrigerated Centrifuge	TU/11-15/Pur/FPT/2010-725	Rs. 442094/-
3	Vacuum Oven	TU/11-15/Pur/FPT/2010-726	Rs. 292000/-
4	Muffle furnace	TU/11-15/Pur/FPT/2010-726	Rs. 196000/-
5	Consumables	----------	Rs. 450138/-
6	Fellowship	----------	Rs. 223833/-
7	Travel	----------	Rs. 16157/-
		Total	**Rs. 2568722/-**

Amount Returned: Rs. 45060/-

21.12.15

(Nandan Sit)

Associate Professor and Principal Investigator

21-12-15

Signature of Competent Authority with Seal

Finance Officer
Tezpur University

Comparative statement for the equipments purchased

Sr. No.	Items/Equipments as approved by PAC	Cost as approved by PAC	Actual cost as per bill/invoice	Bill no./ Invoice no. attached
1	Table top centrifuge	Rs. 6,09,572/-	Rs. 4,42,094/-	23-SUP-116
2	Vacuum drying oven	Rs. 5,50,000/-	Rs. 2,92,000/-	B/058/12-13/TB
3	Viscometer	Rs. 3,28,853/-	Rs. 9,48,500/-	011
4	Muffle furnace	Rs. 4,00,000/-	Rs. 1,96,000/-	B/058/12-13/TB