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ISOLATION, ESTIMATION AND CHARECTERIZATION OF STARCH FROM SEEDS OF *ARTOCARPUS HETEROPHYLLUS*

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ABSTRACT

A study has been carried out to investigate the binding and disintegrating properties of modified starch, isolated for m the fresh seeds of "*Artocarpus heterophyllus"belonging* to the family "Moraceae". After modifications of Artocarpus starch (MAS) dissolution profiles were studied. Paracetamol (500mg) was used as marker drug and dissolution data of MAS compared with explotab (marketed modified rice starch) (ES) and rice starch (PS) in batches of 200. All the products met the requirement of *in vitro* parameters like uniformity of weight, assay, friability, and hardness as per pharmacopoeial requirements. These products also confirm the dissolution specification of USP. The amount of modified *Artocarpus* starch (MAS) required as binding and disintegrating agent was three fourth of the amount maize and rice starch. Therefore modified *Artocarpus* starch can be effectively used in tablet technology.

Key words: Modified *Artocarpus* starch, maize and rice starch, Paracetamol.

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INTRODUCTION

Almost all plant members is give the some different amount of starch. Starch is nothing but carbohydrate source. The some plants are gave the more quantity and some plants are gave high quality of the starch. Normally commercially used starch are mainly derived from following sources, Potato starch is derived from the tubers of *Solanumtuberosum,* Cassava starch is derived from the root parts of *Manihotutilissima,* arrowroot is obtained from the rhizomes of *Curcuma longa,* Wheat starches derived from the fruits of cereals of *Triticumaestivum,* Maize starch is derived from *Zea mays,* Rice starch is obtained from *Oryza sativa. Artocarpus* starch is important as prevention for people of all ages.

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Lysine has a unique meaning for small children (it supports the production of brain cells) and minerals, vitamins, unsaturated fatty acids, quality protein for sportsmen, which support growth of muscular matter. *Artocarpus* recovers cells and influences metabolism significantly (Nelson OE et al., 1978; Shannon JC, Garwood DL, 1984).

Artocarpus grains contain 7% of oil, where a high protein of essential fatty linoleic acid (70% unsaturated fatty acids) and essential squalene (6.5%) is present high protein content (17%) gluten free, dietary fibre (1.5%) and high contact of minerals (Ca, K, P, Fe) put *Artocarpus* on remarkable place in comparison to the other plants. Althouth a large number of disintegrants and binders are available. Starches have an important place in the technology of tablet. Usually maize starch is used abundantly. But there is always enough space for developing newer agents to be used in tablet technology for better result. In view of this the present study was carried out for isolation, modification and characterization of starch from *Artocarpus heterophyllus*. Studies on optimization of condition for carboxy methylation of

Artocarpus starch has been reported earlier by Bharttacharya *et al*., (1995).

MATERIALS AND METHOD

Plant material

Well identified seeds of *Artocarpus heterophyllus* were collected from local market, Bareilly. Seeds were identified according to the description of the wealth of India and macroscopic comparison was done for authentification of the samples at laboratory of college, where a voucher specimen is deposited.

Isolation procedure

The grains of *Artocarpus heterophyllus* were thoroughly washed with water to remove durt or adherent material. The grain were then dried and coarsely ground in a mixer grinder and passed through 22 – mesh sieve. These coarsely ground grains were extracted repeatedly with 0.25% NaOH in a ratio of 1:5, until seeds become free from protein (tested with biuret reagent).

The protein free grains were washed continuously for 2 hours with distilled water till free from NaOH and proteins. The residue was transfer to the grinder ground with to obtain a white paste of starch and fibre. Filtration was done though bolting cloth of 200-mesh size. Filtrate was collected and kept for sedimentation. The sediment obtained was subjected to centrifugation at 6000 rpm for 15 minutes. The upper brownish layer was scraped and remaining white layer was transferred the tray dryers and dried at 45^0c for 5 hrs. The dry starch obtained was passed through 60-mesh.

Modification of isolated starch

Sodium carboxy methyl starch (CMS) is prepared by the reaction of starch with sodium mono chloro acetate in presence of sodium hydroxide and organic solvent (isopropyl alcohol) based on williamson's Ether synthesis. And mechanics is essentially SN_2 (substitution nucleophilic bimolecular).

$$\text{Starch} - OH + Cl\,CH_2COONa + NaOH \longrightarrow$$
$$\text{Starch} - OCH_2COONa + NaCl + H_2O$$
$$\text{Starch mono chloro acetate} \longrightarrow$$
$$\text{Carboxy methyl starch}$$

Multistage carboxy methylation of the starch

The second stage carboxy methylation was done of the sodium CMS obtained from first stage as under the some condition (requirnments) as were in first stage. Similarly third stage carboxy methylation was carried out using starch obtained after second stage as a raw material but in the third stage, starch was getting gelatinized there for only double modification was done. The modified starch also complies with all the pharmacopoeial specifications except pH residue and ignition.

Test for granule strength and flow property of the granules prepared by modified Artocarpus starch

After preparations of paracetamol granules with MAS, ES and PS granule strength were measured as method described by Carr RL *et al*. (1965).Flow ability and angular properties of the granules were determined by as described by Train *et al*., (1958).

Preparation and evaluatin of tablet

Paracetamol tablet using different starches (MAS,ES, PS) were prepared by wet granulation method (Train *et al*., 1958). The tablet of 200 batch size each was punched using single punch machine (cadmach, U.K). The weight variation, test for hardness, friability, disintegration time, uniformity of content and *in vitro* release pattern tests were performed according to pharmocopial specification (Kohli DPS, 1991; Dhayagude CT, Sharma AK, 2004).

In vitro release pattern

In vitro release pattern in phosphate buffer (pH 7,4) were determine by using apparatus 1 according to USP XXII and procedure followed as described in USP XXII values of cumulative percent drug dissolved as various time intervals were also found out and plotted against time. Values of t_{50} (time for 50% dissolution) were determined form this plot. (Mukherjee PK *et al*., 1996; Shure M *et al*., 1983; United State Pharmacopoeia, 1990).

Statistical analysis

The data were analysed using non parametric mann – whitney U-test. The pearson chi – square test was used for comparing the granule strength uniformity of weight, hardness, dissolution rate between all the groups where rice starch used as positive control the data was expressed as mean + S.D and a p-value of < 0.05 was considered statistically significant.

RESULTS AND DISCUSSION

Formulation and manufacturing processes have significant effects on the disintegration, dissolution and other physic - chemical characteristics of the dosage form. It should be noted however that the rates of the process of dissolution are all dependent upon the composition and method of preparation of dosage form. Modified *Artocarpus* starch met the requirement of the test for the options of E.coli and salmonella as per pharmocopoeial specification [12]. The estimations of granule strength is aimed at estimating the relative magnitude of attractive forces seeking to hold the granules together. The resultant strength of granule depends upon the base material, the kind and amount of granulating agent used and granulating equipment. Granules affect the changes in particle size distribution of granulations and consequently compressibility in to cohesive tablets. In the case of modified *Artocarpus* starch, granule strengths of different

formulation as a function of granule size are shown in table – 2. Several factors and granule characteristic have been studied for their effect on the angle of repose, such as, particle size, use of glidant, moisture effect and particleshape (Tsai JS, Kuo CG, 1980; Frydman KB, 1963; Basak Sc et al., 2003). In our case in all the formulations the value for angle of repose was less than 30^0 which indicate that the granules prepared with *Artocarpus* starch as well as other starches used in the formulation were free flowing. Study of the physical parameter of the tablets reveals that all the categories of the tablets prepared with different starches met the pharmacopoeial requirement of uniformity of weight (Indian Pharmacopoeia, 1985).(Table - 3)

Values of maximum percent deviation were will within the pharmacopoeial limit. All the products confirmed to the requirement of assay (table -3) as prescribed in the monograph. Hardness of the tablets was within pharmacopoeial limit (table -3) in all cases. The friability was within the order of MAS<MES<PS. The study on the disintegrating property of all the tablets prepared with different starches revealed that the disintegration time in the tablets prepared with MAS was less than that of Explotab and rice starch (table - 3). So the result of these studies showed that modified *Artocarpus* starch has got good disintegrating and binding property. One point dissolution data (as per requirement of USP) of all the products are shown in fig.1, 2 and 3.

All the products met the dissolution requirement of USP i.e., each product of paracetamol tablet shows not less than 80% dissolution at stage S_1 of dissolution test. Values of t_{50}, t_{70} and t_{90} of all products are indicated in (table-4) from these values it is clear that tablets prepared with modified *Artocarpus* starch showed fastest dissolution rate. However as mentioned above, all the products confirmed to the one point dissolution test specified in USP. The from the results show for obtained it can be concluded that modified *Artocarpus* starch has got better binding, disintegrating property and dissolution characteristics therefore can be exploited for commertial use.

Table 1. Tablet formulation using starch as disintegrant

Formulations	Ingredients (mg/tablets)				
	Paracetamol	**Lactose**	**PMAS**	**PRS**	**PES**
$PMAS_1$	500	7.0	53.0	-	-
$PMAS_2$	500	33.5	26.5	-	-
$PMAS_3$	500	46.75	13.25	-	-
PRS_1	500	7.0	-	53.0	-
PRS_2	500	33.5	-	26.5	-
PRS_3	500	46.75	-	13.25	-
PES_1	500	7.0	-	-	53.0
PES_2	500	33.5	-	-	26.5
PES_3	500	46.75	-	-	13.25

PAMS is tablet using modified artocarpus starch as disintegrant, *PRS* is tablet using rice starch as a disintegrant, and *PES* is tablet using explotab as a disintegrant.

Table 2. Granule strength as a function of granule size prepared with modified artocarpus starch

Granules prepared with artocarpus starch for tablet	Breaking load (g)	Midrange or granule size (mm)
Paracetamol (500mg)	700 ± 4.5^a	1.0 ± 0.030^a
	830 ± 4.1^a	1.5 ± 0.048^a
	941 ± 5.6^a	2.0 ± 0.091^a

Mean ± SEM, n=6 [a]significantlydifferent from control ($p<0.01$); [b] significantlydifferent from starch ($p<0.01$).

Table 3. *In vitro* evaluations of various tablets using different starches as disintegrant

Formulation	Uniformity of weight		Assay (Persent of labled amount)	Hardness (kg/cm^2)±SD	Friability %±SD	Disintegration time (s)
	Average wt (mg)	Maximium% deviation				
$PMAS_1$	598.3 ± 3.56^a	3.6 ± 0.030^a	102.6 ± 7.98^a	4.8 ± 0.09^a	0.2 ± 0.01^a	113 ± 5.05^a
$PMAS_2$	596.5 ± 17.89^a	3.8 ± 0.29^a	101.33 ± 6.23^a	4.6 ± 0.06^b	0.2 ± 0.03^b	121 ± 4.87^b
$PMAS_3$	597.3 ± 24.98^a	4.1 ± 0.39^b	98.6 ± 4.98^a	4.3 ± 0.05^a	0.12 ± 0.01^b	129 ± 3.23^b
PRS_1	593.4 ± 25.27^b	4.2 ± 0.40^a	101.33 ± 2.19^b	4.5 ± 0.02^s	0.5 ± 0.04^a	159 ± 7.09^a
PRS_2	596.3 ± 13.49^b	4.2 ± 0.33^a	104.0 ± 3.85^b	4.1 ± 0.02^a	0.4 ± 0.03^a	173 ± 3.33^b
PRS_3	595.5 ± 23.91^b	4.5 ± 0.51^a	98.6 ± 3.97^a	4.3 ± 0.02^a	0.7 ± 0.02^b	183 ± 4.20^a
PES_1	583.6 ± 17.98^a	3.7 ± 0.29^a	102.6 ± 3.25^a	3.8 ± 0.06^b	0.3 ± 0.00^a	138 ± 6.12^a

PES_2	586.7±23.43[b]	3.9±0.41[a]	97.81±3.45[b]	4.6±0.08[b]	0.36±0.03[b]	151±3.98[b]
PES_3	579.5±19.98[b]	4.0±0.59[a]	102.8±3.45[a]	4.1±0.05[a]	0.38±0.01b	162±4.16[a]

*Mean ± SEM, n=6 [a]Significantly different from control (P<0.01); [b]Significant different from starch (P<0.01).

Table 4. Dissolution parameter of paracetamol tablets containing different starches as disintegrant

Formulation	T_{50}(min)	T_{70}(min)	T_{90}(min)
$PMAS_1$	13.0±0.28[a]	24.3±0.41[b]	38.9±0.74[b]
$PMAS_2$	14.2±0.56[a]	25.9±0.87[b]	45.8±0.65[a]
$PMAS_3$	17.2±0.45[a]	28.1±0.58[b]	44.7±0.52[b]
PRS_1	5.6±0.78[a]	10.9±0.81[a]	27.9±0.69[a]
PRS_2	7.0±0.59[a]	12.9±0.44[a]	29.9±0.71[b]
PRS_3	8.5±0.25[a]	13.8±0.65[a]	33.5±0.98[a]
PES_1	6.5±0.41[b]	15.9±0.55[a]	41.6±0.47[a]
PES_2	9.0±0.32[b]	17.3±0.63[a]	36.5±0.55[a]
PES_3	10.9±0.49[b]	20.0±0.87[a]	34.4±0.51[a]

*Mean±SEM, n=6. [a]Significantly different from control (P<0.01), [b]Significant different from starch(P<0.01).

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None

CONFLICT OF INTEREST

No interest.

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