RESEARCH PAPER

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Pharmaceutical Application of Starch Isolated from *Nelumbo nucifera* Gaertn (Fam. Nymphaeaceae)

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A study has been carried out to investigate the binding and disintegrating properties of starch isolated from rhizomes of *Nelumbo nucifera (Nelumbo* starch) along with the dissolution rate profiles. For this study the tablets of Paracetamol (500 mg), metronidazole (400 mg) and ibuprofen (400 mg) were prepared using maize, nelumbo and potato starch, each in batches of 200. All the products met the requirement of *in vitro* parameters such as uniformity of weight, assay, friability and hardness as per Pharmacopoeial requirements. These products also conform the dissolution specification of USP. The amounts of *Nelumbo* starch required as binding and disintegrating agent was half of the amount of maize and potato starch. Therefore *Nelumbo* starch can be effectively used in tablet technology.

NELUMBO nucifera Gaertn (Family Nymphaeaceae) commonly known as 'Padma', 'Kamala' or 'Pundarika' is an aquatic herb with stout creeping yellowish white rhizomes[1]. All most all parts of this plant are used medicinally by the traditional medical practitioners for curing various diseases[2]. Rhizomes commonly known as 'Kamal-Kakadi' measure 60-120 cm in length and 6-9 cm in diameter and show in cross section, a few large cavities surrounded by several small ones[3]. The powdered rhizomes are used as demulcents in piles and were found beneficial in dysentry and chronic dyspepsia[4]. They are also used as nutrient, diuretic and Cholagogue[5].

In spite of the fact that a large number of disintegrants and binders are now available; starches still occupy an important place in the technology of tablets. Usually maize starch is now used abundantly. Pharmacognostical investigations and other related work on rhizomes of *N. nucifera* has been reported earlier by Trivedi *et al*[6,7]. Estimation, Isolation and Characterization of *Nelumbo* starch along with scanning electron microscopy has already been reported by Pulok K. Mukherjee *et al*(1995)[8]. It was therefore thought necessary to carry out the preparation and *in vitro* testing of different tablets prepared with *Nelumbo* starch (N) and to compare them with those prepared with their maize (M) or potato starch (P). Tablets of paracetamol (500 mg), metronidazole (400 mg) and Ibuprofen (400 mg) were chosen for these investigations.

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EXPERIMENTAL

Plant Material

Rhizomes of *Nelumbo nucifera* were collected from Sandhipur, Midnapore district of West Bengal, India. Taxonomic identification of the plant was made from Botanical Survey of India, Shibpur, Howrah. Fresh rhizomes were stored in refrigerator until further use.

Isolation of Starch

Fresh rhizomes (1 Kg) were washed thoroughly to free them from soil, the outer fibrous covering

and dirt was removed. It was then pulped and minced for 2 minutes by adding 2 litres of distilled water in a mixture with fine teeth and strained through a linen. It was allowed to settle for one hour. Clear upper layer was decanted and the residue was washed with 0.1(N) NaOH solution repeatedly for 2-3 times. The residue was further washed with distilled water to make it free from alkali[8]. The starch so obtained was dried at 37-40°C and was treated further with Methanol : Chloroform (95:5) mixture, dried, powdered and passed through 100 mesh sieve then stored in a well closed container. The isolated starch complies with all the pharmacopoeial specifications[9].

Test for microbial contamination in *Nelumbo* starch

The isolated *Nelumbo* starch so far obtained was subjected to the test for the presence of *E.coli* and *Salmonella* as per the methods prescribed in Indian Pharmacopoeia.

Test for granule strength and flow property of granules prepared by *Nelumbo* starch

After preparation of the granules for different tablets i.e. paracetamol, metronidazole and ibuprofen, it was subjected to the test for determination of granule strength by the method developed by Carr et al. (1965)[10]. In this test a granule of each formulations was placed between anvils and force required to break the granule was measured.

To determine the flowability of granules prepared with different formulation of *Nelumbo* starch, angular properties of the granules were determined by measuring the angle of repose as described by Train *et al.* (1958)[11]. Here a funnel was secured with its tip at a given cone height, H, above a flat horizontal surface to which graph paper was attached, granules were carefully poured through the funnel until the apex of the conical pile just touched the tip of the funnel. Thus angle of repose was measured as $\tan \alpha = \frac{H}{R}$, where α is a angle of repose, R is the base length for the granules.

Preparation of Tablets

Wet granulation method was employed for preparation of paracetamol, metronidazole and ibuprofen tablet granules[10]. Three different batches of each of paracetamol, metronidazole and ibuprofen were prepared by using maize, nelumbo and potato starch at the specific quantity as mentioned in **Table-1**. 2% talc was added as lubricant to the granules. Half of the disintegrant (starch) which was to be added before compression was also mixed thoroughly with the granules. The tablets of 200 batch size each were punched using a single punch machine.

Uniformity of Weight

The weight variation of the tablets were performed in accordance with the official recommendation. 20 tablets of each batch weighed and the average weight was calculated[9].

Test for Hardness

Hardness of the tablets was measured with the help of Monsanto hardness tester. Five tablets were chosen at random for this test from all formulations and the mean values were recorded[12].

Friability Test

Friability of the tablets was determined with the help of Campbell friability test equipment. 20 tablets of each batch were used for the test and the percentage of powder eroded during 3 min was recorded[12].

Disintegration Time

Disintegration time of the tablets were examined using the method prescribed in I.P. for uncoated tablets[13].

Table 1 : Granule strength as a function of granule size of different formulations prepared with *Nelumbo* starch

Granules prepared with *Nelumbo* starch for different formulations	Breaking Load (g)	Midrange or granule size (mm)
Paracetamol (500 mg)	710 ± 4.5	1.0
	850 ± 3.8	1.5
	975 ± 5.2	2.0
Metronidazole (400 mg)	650 ± 3.7	1.0
	790 ± 4.2	1.5
	879 ± 6.0	2.0
Ibuprofen (400 mg)	760 ± 3.5	1.0
	870 ± 4.2	1.5
	991 ± 3.4	2.0

Uniformity of Dosage Units

For the assay of each batch of tablets, 20 tablets from each batch were selected at random. They were pulverized well in mortar and an amount equivalent to the specific quantity stated in the individual monograph of Indian Pharmacopoeia was subjected to know the uniformity of content as follows :

For Paracetamol tablets colorimetric assay at 395 nm wavelength was performed using 5% w/v solution of vanillin in isopropyl alcohol as suggested by Sethi[14] using Beckman DU-64 spectrophotometer.

For the assay of metronidazole tablets titrimetric method using 0.1(N) perchloric acid as prescribed in I.P. was followed[15].

Estimation of ibuprofen tablets were performed titrimetrically in accordance with the method prescribed in I.P., with 0.1(N) Sodium hydroxide, using phenolphthalein solution as indicator[15].

Dissolution Studies

The test was carried out using Dissolution Rate Test Apparatus 1 (Basket stirring element) or USP[16] as follows.

For paracetamol tablets the test was performed using 900 ml of pH 5.8 phosphate buffer as dissolution medium, a speed of 50 r.p.m. and temperature of 37° ± 0.5°C was maintained all round. The sampling time specified in USP[16] was modified i.e. instead of withdrawing a single sample at 30 minute interval, serial sampling was done at 5, 10, 15, 20 and 30 minute intervals. This provided additional data about the dissolution profile. After each withdrawal of a sample, an equal volume of dissolution medium was added to the dissolution vesel. The filtered samples were suitably diluted and assayed clorimetrically at 395 nm[14]. Percent drug dissolved in 30 minutes (as per requirement of USP[16]) was found out. Values of cumulative percent drug dissolved at various time intervals were also found out and ploted against time (**Fig.1**). Values of t_{50} (time for 50% dissolution), t_{70} (time for 70% dissolution)

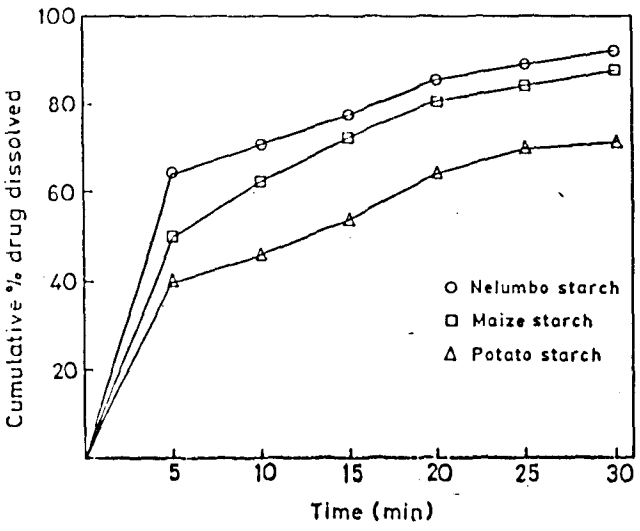

Figure 1: Dissolution profiles of various Paracetamol tablets containing different starches as diluent.

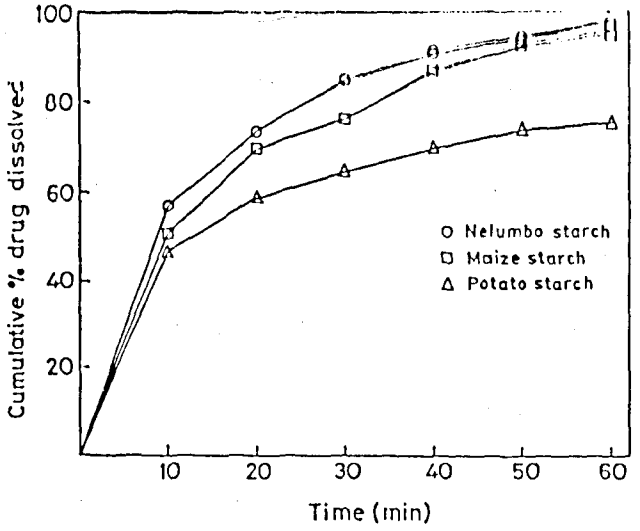

Figure 2: Dissolution profiles of various Metronidazole tablets containing different starches as diluent.

and t_{90} (time for 90% dissolution) were determined from this plot (Table 2).

For metronidazole tablet 900 ml of 0.1(N) hydrochloric acid was used as dissolution medium. A speed of 100 r.p.m. and temperature of 37 ± 0.5°C was maintained. Serial sampling was done at 10, 20, 30, 40, 50 and 60 min intervals, excepting the single sampling at 60 min as specified in USP[16]. The filtered samples were suitably diluted and assayed[15]. Percent drug dissolved in 60 minutes (as per requirement of USP[16]) was found out. Values of cumulative percent drug dissolved at various time intervals were also found and ploted against time (**Fig.2**). Values of t_{50}, t_{70} and t_{90} were determined from this plot (Table 2).

For determining the dissolution profile of ibuprofen tablet 900 ml of pH 7.2 phosphate buffer was used as dissolution medium. A speed of 150 r.p.m. and temperature of 37 ± 0.5°C was maintained. Sampling was done at 5, 10, 15, 20 and 30 min interval by a little modification of the USP[16] method. The filtered samples were suitably diluted and assayed titrimetrically[15]. Values of cumulative percent drug dissolved at various time intervals were found out and plotted against time (**Fig.3**) and from this t_{50}, t_{70} and t_{90} were calculated using the same procedure as stated before in paracetamol tablets (Table 2).

RESULTS AND DISCUSSIONS

Formulations and manufacturing processes have significant effects on the disintegration, dissolution and other physico-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[17].

Nelumbo starch met the requirement of the test for the absence of *E.coli* and *Salmonella* as per pharmacopoeial specifications[9].

The measurement of granule strength are aimed at estimating the relative magnitude of attractive forces seeking to hold the granules together. The resultant strength of a granule depends upon the base material, the kind and amount of ganulating agent used and the granulating equipment. Granule strength affect the changes in particle size distribution of granulations and consequently compressibility

Table 2 : *In vitro* evaluation of various categories of tablets using different starches as diluent

Product	Uniformity of Weight Average wt (mg)	maximum % Deviation	Assay (Percent of labelled amount)
Paracetamol (500mg)			
M	573	+ 3.8 - 3.1	102.8%
N	566	+ 2.3 - 2.8	99.76%
P	564	+ 4.1 - 3.7	97.81%
Metronidazole (400mg)			
M	465	+ 2.1 - 3.6	98.6%
N	467	+ 3.2 - 3.6	100.3%
P	461	+ 4.2 - 3.8	96.7%
Ibuprofen (400mg)			
M	547	+ 3.1 - 2.8	101.67%
N	538	+ 3.6 - 2.2	98.97%
P	542	+ 3.45 - 1.25	97.5%

M = Tablets prepared using Maize starch as diluent.
N = Tablets prepared using Nelumbo starch as diluent.
P = Tablets prepared using Potato starch as diluent.

into cohesive tablets[18]. In case of *Nelumbo* starch, granule strengths of different formulations as a function of granule sizes are shown in Table I.

Several factors and granule characteristics have been studied for their effect on the angle of repose, such as, particle size, use of glidants, moisture effects and particle shape[18]. In our case in all the

Table 3 : Hardness and Disintegration time of different tablets containing different starches as diluent.

Product	% binder	%Disintegrant	Distinte-gration time (sec)	Friability %	Hardness units (Kg/sq.cm)
Paracetamol					
M	4	5	120 ± 1.03	0.98 ± 1.01	7.3 ± 1.02
N	2	3.5	90 ± 2.01	0.83 ± 1.08	7.5 ± 1.06
P	5	7	140 ± 3.36	1.11 ± 2.03	8.1 ± 2.01
Metronidazole					
M	4	5	90 ± 2.1	0.85 ± 1.05	6.3 ± 1.03
N	2	3.5	80 ± 1.05	0.82 ± 1.02	6.5 ± 1.02
P	5	7	110 ± 3.05	1.05 ± 2.02	7.2 ± 1.05
Ibuprofen					
M	4	5	118 ± 1.67	1.04 ± 1.06	7.1 ± 1.02
N	2	3.5	112 ± 1.32	0.98 ± 1.02	7.3 ± 1.05
P	5	7	139 ± 2.17	1.35 ± 2.01	8.2 ± 2.02

M = Tablets prepared using Maize starch as diluent.

N = Tablets prepared using Nelumbo starch as diluent.

P = Tablets prepared using Potato Starch as diluent.

Table 4 : Dissolution parameters of Paracetamol, Metronidazole and Ibuprofen tablets containing different starches as diluent

Product	Starch	t_{50} (min)	t_{70} (min)	t_{90} (min)
Paracetamol				
	Nelumbo	4.0	9.5	26.5
	Maize	5.0	14.0	> 30.0
	Potato	12.5	25.5	> 30.0
Metronidazole				
	Nelumbo	9.0	18.0	39.0
	Maize	10.0	20.0	45.5
	Potato	13.0	40.0	< 60.0
Ibuprofen				
	Nelumbo	4.75	12.0	22.5
	Maize	6.50	16.5	< 30.0
	Potato	8.75	24.5	< 30.0

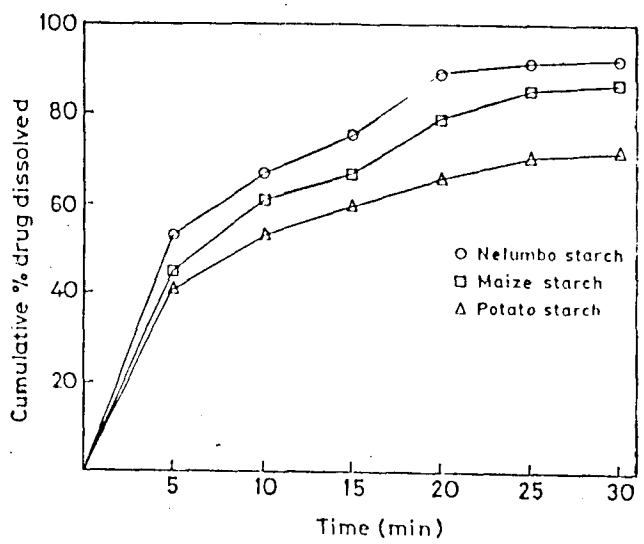

Figure 3: Dissolution profiles of various Ibuprofen tablets containing different starches as diluent.

formulations the values for angle of repose was ≤ 30° which indicate that the granules prepared with *Nelumbo starch as well as other starches used in the experiment were free flowing.*

The study of the physical parameters of the tablets reveals that all the categories of tablets prepared with different starches met the Pharmacopoeial requirement of uniformity of weight (I.P.)[9] (Table-2). Values of maximum percent deviation were well within the pharmacopoeial limit. All the products conformed to the requirement of assay (Table 2) as prescribed in their individual monogrpah[15].

Hardness of the tablets were within pharmacopoeial limit (**Table 3**) in all cases. Percentage of binder (starch) required for tabletting with Nelumbo starch was half of the amount required for other starches. The friability study shows that the firability was with in the order of N < M < P.

The study on the disintegrating property of all the tablets prepared with different starches revealed that the disintegration time in the tablets prepared with *Nelumbo* starch was less than that of Maize and Potato starch (**Table 4**). So the results of these studies showed the *Nelumbo* starch has got good disintegrating and binding property.

One point dissolution data (as per the requirement of USP[16]) 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, Metronidazole and Ibuprofen tablets showed not less than 80%, 85% and 70% dissolution at stage S_1 of dissolution test[16]. Values of t_{50}, t_{70} and t_{90} of all products are indicated in Table IV. From these values it is clear that tablets prepared with *Nelumbo* starch showed fastest dissolution rate. However as mentioned above, all the products conformed to the one point dissolution test specified in USP. Thus from the results so far obtained it can be concluded that *Nelumbo* starch has got better binding and disintegrating property and as all the formulations met the requirements of USP[16]. So from this study it can be concluded that *Nelumbo* starch being used as half of the amount of other starches, have better binding, disintegrating and dissolution characteristics and can be exploited for commercial use.

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REFERENCES

1. Chopra, R.N., Chopra I.C. and Handa, K.L., Indigenous Drugs of India, 2nd Ed., U.N. Dhur and Sons Pvt. Ltd., Calcutta, 1958, 679.

2. Raghunathan, K. and Mitra, R., Pharmacognosy of Indeginous Drugs, Vol. 2, Central Council for Research in Ayurveda and Siddha, New Delhi, 1982, 806.

3. The Wealth of India, Vol.III, Publication and Information Directorate, C.S.I.R., New Delhi, 1966, 7.

4. Chatterjee, A. and Pakrashi, S.C., The Treatise on Indian Medicinal Plants, Vol. I, Publication and Information Directorate, C.S.I.R., New Delhi, 1991, 94.

5. Kirtikar, K.R. and Basu, B.D., Indian Medicinal Plant, Bishen Singh and Mahendra Pal Singh, Dehradun, 1975, 116.

6. Trivedi, A., Srivastava, M.P. and Misra, P., **Eastern Pharmacist.**, 1980, 23, 187.

7. Trivedi, A., Srivastava, M.P. and Misra, P., **Eastern Pharmacist.**, 1982, 25, 199.

8. Mukherjee, P.K., Giri, S.N., Saha, K., Pal, M.P. and Saha, B.P. **Indian Drugs**, 1995, 32, 392.

9. Pharmacopoeia of India, Vol.2, 3rd Ed., Ministry of Health and Family Welfare, Government of India, Controller of Publications, Ministry of Health, Govt. of India, Delhi, 1985, pp.480. 501.

10. Carr, R.L., **Chem. Engg.**, 1965, 72, 163.

11. Train, D., **J. Pharm. Pharmacol.**, 1958, 10, 127.

12. Lachman, L., Lieberman, H.A. and Kanig, J.L., The Theory and Practice of Industrial Pharmacy, 3rd Ed., Varghese Publishing House, Bombay, 1987, pp.297, 323.

13. Pharmacopeia of India, Vol.2, 3rd Ed., Ministry of Health and Family Welfare, Government of India, Controller of Publication, Delhi, 1985, A-122.

14. Sethi, P.D., Quantitative Analysis of Drugs in Pharmaceutical Formulations, 2nd Ed., CBS Publishers and Distributors, Delhi, 1995, 50.

15. Pharmacopoeia of India, Vol.1, 3rd Ed., Ministry of Health and Family Welfare, Government of India, Controller of Publication, Delhi, 1985, pp. 251, 320.

16. United States Pharmacopoeia, XXII, United State Pharmacopoeial Convention, INC, 1990, pp.14, 891, 683, 1578.

17. Levy, G., Physico-chemical Aspects of Drug Action, E.J. Ariens, Ed., Pergamon, N.Y., 1968, 48.

18. Lieberman, A.H., Lachman, L. and Schwartz, B.J., Pharmaceutical Dosage Forms, Tablets, Vol.2, 2nd Ed., Marchel Dekker, Inc, 1990, pp.300, 309.

(References continued from page no. 58)

10. Fiero, G.W., **J. Am. Pharm. Assoc., Sci. Ed.,** 1940, 29, 18.

11. Draizer, J.H., Woodward, G. and Calvery, H.O.,**J. Pharmacol. Exp. Therap.**, 1944, 82, 377.

12. Pharmacopoeia of India, 3rd Ed., Controller Publications, Ministry of Health, Govt. of India, Delhi. 1985, Vol. II, A-96.

13. Supplement 1975 to the Pharmacopoeia of India, Govt. of India Press, Nasik, India, 1966, 206.

14. The United States Pharmacopoeia XXII and the National Formulary, XVII, United States Pharmacopoeia Convention, Inc., Rockville, Md. 20852, 1990, 1479.

15. Bhalla, H.L., Menon, M.R., Gopal N.G.S., **Indian J. Pharm. Sci.**, 1983, 45, 203.