Isolation and Evaluation of Tamarind Seed Coat Mucilage as Pharmaceutical Suspending Agent

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Abstract

Natural polymers, specifically mucilages, have been used as a suspending agent for a long period of time. Natural excipients can serve as an alternative to synthetic products since they are less expensive, less toxic, and devoid of environmental pollution. The present study was undertaken to evaluate the mucilage isolated from Tamarindusindica (Fabaceae) seed coat, commonly named tamarind, as an innovative suspending agent. Paracetamol suspensions (10% w/v) were prepared using the T. indica seed coat mucilage as a suspending agent, and it was evaluated for parameters like physical stability, sedimentation profile, redispersibility, and flow property. Furthermore, it was assessed for its stability. The effect of the tested mucilage on the suspension was compared with commonly used suspending agents, i.e. sodium carboxymethyl cellulose (CMC) at concentrations of 0.5, 1.0, and 1.5% w/v. The results obtained indicated that the T. indica seed coat mucilage could be used as a suspending agent.

Keyword: Tamarind seed coat mucilage. Suspending agent, Suspension.

1. Introduction

Nowadays, a wide variety of gums and mucilage of natural origin, viz, starch, agar, alginates, acacia, pectin, gelatine, are used as pharmaceutical excipients. Natural materials do have advantages over synthetic materials, because they are non-toxic, less expensive, and more freely available. Furthermore, they can be modified to obtain tailor-made materials for the drug delivery system and they can compete with the synthetic agents available on the market as well as in public acceptance[1]. Among them, plant mucilageis well known, since ancient times, for their medicinal use. In recent years, plant mucilages have evoked tremendous interest due to its diverse applications in pharmacy, for the formulation of both solid and liquid dosage forms[2]. Plant mucilages are pharmaceutically important polysaccharides with a wide range of applications, such as thickeners, binding agents, water retention agent, emulsion stabilizer,

suspending agents, disintegrants, gelling agents, and film formers[3]. Thus, with the increased demand for these substances, it has been necessary to explore newer sources, to meet the industrial demands. Because of its geographical and environmental location, India has traditionally been a good source for such products among Asian countries.

Suspensions are dispersed systems where the solid state (internal phase) is dispersed into a liquid/semisolid dispersion medium or into an external phase. In the preparation of suspensions, it is important to consider the proper selection and application of additives, specifically suspending agents, to maintain the stability and accurate dosing of the preparations [4].A suspending agent that reduces the rate of settling and permits easy redispersion of any settled particulate matter, both by increasing the consistency of the suspending medium and by protective colloidal action[3]. There are reports about the successful use of various plant mucilages, as innovative suspending agents, like mucilages isolated from *Adansoniadigitata*L. leaves[5], *Grewiaferruginea*HochstEx A[6], *Aloe weloensis*Sebsebe[7],BananaPeels, *Cissusrubiginosa*Planch.fruit[8, 9],*Colocasiaesculenta*(L.) Schott[10],and so on. The purpose of this study is to search for ainexpensive and effective natural excipients that can be used as new alternative suspending agents for the formulation of pharmaceutical suspensions. Tamarind plants (*Tamarindusindica* L.) are widely available in the dry tracks of central and south India. Chemically, tamarind gums are branched carbohydrate polymers obtained from the endosperms of tamarind seeds[11].It possesses properties like high viscosity, broad pH tolerance, non-carcinogenicity, mucoadhesive nature, and biocompatibility. It is used as a stabilizer, suspending agent, thickener, gelling agent, andbinder in the food and pharmaceutical industries[12].However, no study has been reported so far on the isolation and use of mucilage isolated from *T. indica* L. seed coat, which is considered as waste. The present study is an attempt to isolate and investigate the suspending agent properties of the isolated mucilage from *T. indica* L. seed coat.

2. Materials and methods

2.1. Sample Collection and Mucilage Extraction

T. indica dried, mature, and healthy seeds were collected from a local market in Shirpur, Maharashtraand authenticated.The seed coats were separated from the seed kernels and used for the extraction of the mucilage. The extraction procedure of the mucilage isolation as per the published literature[13]. Briefly, the seed coats were sized reduced, then mixed with distilled water in a 0.3:1 ratio, soaked for 48 hours, and heated to 50°C for 1 hour before filtration. The extract was filtered using a clean muslin cloth to separate debris. Afterward, a sufficient amount of ethanol (96%) was added to the extract to precipitate mucilage. The procedure was repeated twice. The collected mucilage was further washed with ethanol to remove all traces of other phytochemical impurities. Finally, the mucilage was dried in an oven at 50°C for 24 hours, dried tamarind seed coat mucilage (TSCM) was powdered and used for characterization and suspension preparations.

2.2 Phytochemical and phytochemical studies

The TSCM was subjected to qualitative chemical tests for confirmation of the purification. Physicochemical characteristics such as color, odour, taste, nature, solubility, ash value, acid

insoluble ash value, water-soluble ash value, loss on drying, and swelling factor of TSCM were studiedas per the prescribed procedure[13] and the results are given in Table 1.

2.3 Preparation of Paracetamol Suspensions

The paracetamol suspension was prepared as per the formula mentioned in Table 2. Three distinct suspensions containing various concentration polymers, including TSCM and sodium CarboxyMethylCellulose (CMC), were prepared. Each polymer suspension was made in three concentrations, 0.5%, 1%,and 1.50%, w/v.

Table 2 Compositions of paracetamol suspension formulations

Formulation ingredients	Composition (% w/v or % v/v)
Paracetamol	10
Suspending agent*	0.5, 1.0, 1.5
Methyl paraben	0.18
Propyl paraben	0.02
Propylene glycol	2 2
Tween 80	0.05
Sodium saccharine	0.07
Sucrose	15
Sorbitol (70%, w/v)	30 mL
Distilled water	Q.S. 100 mL

*The suspending agents used were CMC, TSCM; Q.S: quantity sufficient

The paracetamol suspension was prepared by triturating the suspending agent and paracetamol with 10 mL of a solution containing 15 g of sucrose, 0.07 g of sodium saccharin, 0.05 g Tween 80 to form a smooth paste. 30 mL of 70% sorbitol solution along with methylparaben and propylparaben dissolved in propylene glycol was added gradually with constant trituration. The mixture was transferred into a 100 mL amber coloured, stoppered measuring cylinder, made up to volume with distilled water and then shaken vigorously for 2 min[14].

2.4 Evaluation of Suspension

The TSCM and CMC suspension were evaluated for physical stability, redispersibility, sedimentation volume and viscosity.

2.4.1 Physical test

For four weeks, the prepared suspension was studied for any physical changes such as agglomeration, caking, or crystal growth development[5].

2.4.2 Redispersibility

The redispersibility of test suspensions was determined using the method described by Kumar et al [15], with minor modifications. A fixed volume (50 mL) of each suspension was placed in a 50 mL measuring cylinder and stored at room temperature. The measuring cylinder was gently turned at 180^{o} at regular intervals of 24 hours. The number of rotations taken for homogeneous redispersion of suspended particles was recorded. The recorded results are the average of three determinations.

2.4.3 Determination of sedimentation volume

Each suspension (50 mL) was kept at room temperature for 4 days in a 50 mL amber-colored, stoppered measuring cylinder. The sedimentation volume of each formulation was measured every after 24 hours, for four days. The sedimentation volumes were measured as the suspended particles settled and the hazy supernatant started to clear as it descended from the suspension's top surface. The percentage of sedimentation volume was calculated using the formula given below. The recorded results are the average of three determinations[16].

$$\%\ sedimentation\ volume = 100\frac{Vu}{Vo}$$

Where Vu is volume of the sediment and Vo is the total volume of suspension.

2.4.4 Determination of flow rate

Suspension flow rates were determined using a procedure published in the literature [15]. The flow rate was calculated by recording the time required for each 10 mL suspension sample to flow through a 10 mL pipette. The flow rates recorded are the mean of three measurements. The following equation was used to calculate it.

$$Flow\ rate = Vs/T$$

Where, Vs volume of suspension in the pipette (mL) and T is time (s) required for the 10 mL suspension to elute out of the pipette.

2.4.5 Stability study

The stability chamber was used to test the stability of TSCM and CMC paracetamol suspensions. The conditions maintained were as, the real-time condition of 30°C/65% Relative Humidity (RH), accelerated condition of 40°C/75% RH and in a refrigerator at 4^{o}C for 3 months. Samples were collected at 0, 30, 60 and 90 days. Visual examination, such as physical appearance, was used to assess the stability of the suspensions, and the pH was also recorded [17]. The recorded results are the averages of three determinations.

3. Results and Discussion

The yield of mucilage from *T. indica* L. seed coat, isolated from mature dried seeds, was 30.14% w/w. Various confirmation tests were carried out on the isolated TSC mucilage. The results of these tests on the isolated mucilage are summarized in Table 1.

In reaction toMolisch's reagent, a violet ring developed at the intersection of the two liquids, suggesting the presence of carbohydrates. When the mucilage was treated with ruthenium red, it became red, confirming the identification of the mucilage.

Table 1Phytochemical analysis of TSCM

Test	TSCM
Test for carbohydrates (Molisch's test)	Present
Test for tannins (Ferric chloride test)	Absent
Test for proteins (Ninhydrin test)	Absent
Test for alkaloids (Wagner's test)	Absent
Test for glycosides (Comparative test A and B)	Absent
Test for mucilage (Ruthenium red test)	Present
Test for flavonoids (Shinoda test)	Absent

A physicochemical analysis of isolated mucilage revealed that it was brown in color, has a mucilaginous taste, and a distinct odor. In hot water, it is quickly soluble, while forming a neutral, viscous colloidal solution in warm water and is sparingly soluble in cold water, but is insoluble in ammonia, methanol and chloroform. The mucilage was also examined for its total ash value, acid in soluble ash, water insoluble ash, loss of drying, surface tension, melting point, pH and swelling index. The results obtained are summarized in Table 2.These parameters were developed as standards for future analysis.

Table 2: Organoleptic, Solubility behaviour, and physicochemical properties of TSCM

Organoleptic Properties		Solubility Behaviour		Physicochemical Properties	
Parameters	***Observation***	***Solvent***	***Observation***	***Parameters***	***Observation***
Color	Brown	Cold water	Sparingly soluble	pH	6.2
Odour	Characteristics	Hot water	Quickly Soluble	LOD (%)	3.8
Taste	Mucilaginous	Warm water	Soluble	Surface Tension (dynes/cm)	88.12
		Ammonia	Insoluble	Ash value (%)	4.56
		Methanol	Insoluble	Acid insoluble ash (%)	2.58
		Chloroform	Insoluble	Swelling index (mL)	4
				Melting Point (°C)	252

To evaluate the TSCM suspending capabilities, Suspensions were produced with a fixed concentration of paracetamol (10%, w/v)and varied concentrations of tamarind seed coat mucilage (0.25, 0.50, 1.0% w/v) and standard suspending agents, CMC.

The physical stability of the suspension formulations was assessed throughout the first 48 hours. In early observations, no particle aggregation, caking, or crystal growth development was seen, and all suspension formulations were found to be stable. Later, the redispersibility, sedimentation volume, viscosity, and flow rate of these suspensions were measured and compared.

As the suspension produces sediment on storage, it must be readily dispersible so as to ensure the uniformity of the dose. If the sediment remains even after shaking vigorously for the specified time, the system is described as cake[18]. Thus, redispersibility is an essential property of pharmaceutical quality[19]. To assess the redispersibility, the number of

inversion cycles required to completely redisperse the suspension at the end of 24 h was measured (Table 3). The test was performed consecutively for four days every after 24 h. It was observed that the number of inversions required to redisperse the suspension formulations was concentration-dependent. The formulations prepared with the highest concentration of suspending agent, i.e. TSCM and CMC required fewer inversion cycles i.e. 11 ± 1.53 and 4 ± 1.15, respectively. Similar results were reported in earlier research [7, 14].

Table 3Redispersibility of the suspension formulations after 24 h

Suspending agent concentration (%)	No. of inversion cycles (mean ± S.D.)	
	TSCM	CMC
0.5	21 ± 2.52	18 ± 0.58
1.0	16 ± 2.08	12 ± 2.52
1.5	11 ± 1.53	4 ± 1.15

Values are mean ± S.D.; n = 3

Suspensions are routinely evaluated for their rate of separation which indicates its suspending property. The sedimentation volume profile of the suspensions prepared with sodium CMC andTSCM mucilage is shown in the figure 1.

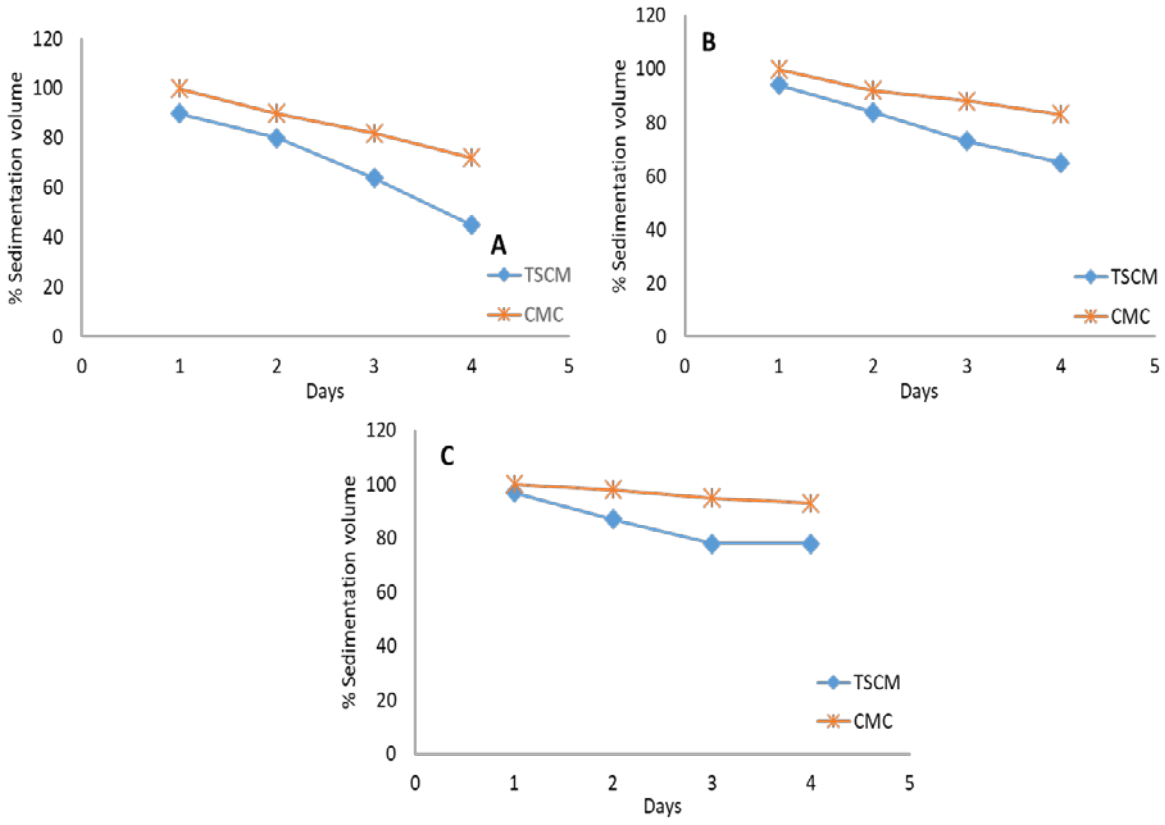

Figure 1.Comparative sedimentation volume (%) of suspensions using TSCM and CMC, (a) 0.5% (b) 1.0% and (c) 1.5% of TSCM CMC

The results revealed that increasing the concentration of the suspending agent increased the sedimentation volume of formulations, which could be attributed to an increase in the viscosity of the medium with concentration.

The uniformity of dosing is dependent upon flowability of the suspension. The flowability of the suspension should be optimum to suspend the drugs for a longer period of time. This will yield a better distribution of the drug throughout the formulated suspension. The TSCM and CMC prepared suspension formulations decreased the flow rate with the increase in the concentration of suspending agents (Table 4). This led to a better distribution of drug throughout the formulated suspension.

Table 4Flow rate of the suspension formulations

Suspending agent concentration (%)	Flow rate (mL/sec) (mean ± S.D.)	
	TSCM	CMC
0.25	1.47 ± 0.09	0.57 ± 0.06
0.5	1.55 ± 0.08	0.21 ± 0.03
1	1.04 ± 0.10	0.15 ± 0.02

Values are mean ± S.D.; n = 3;

In stability testing, colour, odour, and pH of all suspension formulations were tested. During and after the 3 months' different conditions of stability testing.The visual appearance and odour of all suspension formulations remained constant (creamy-white and odourless, respectively). The fact that the organoleptic characteristics and pH range (5.0 - 6.0) of paracetamol suspension formulations were constant under all storage settings and found within the acceptable range, indicates that the formulations were stable.

4. Conclusion

The present investigation is a primary platform to indicate the suitability of the tamarind seed coat mucilage as a suspending agent. As such, the tamarind seed coat is considered to be a waste product. Based on these findings, it is possible to conclude that the isolated mucilage from the tamarind seed coat has the potential to be used as a suspending agent with a high parentage yield, as well as a pharmaceutical adjuvant, given the ease with which tamarind seed can be obtained.

5. References

[1] *D. Harpaz, Colloidal silicon dioxide Handbook of pharmaceutical excipients, 2nd edition ed., Washington, D.C.: American Pharmaceutical Association* ***(1994)***.

[2] *G. Rao, P. Prasad, K. Srinivas, and M. Rao, "Application of Moringa oleifera as a suspending agent in the formulation of sulphamethoxazole,"The Indian Pharmacist* vol. 5, **(2005)**, pp. 75-77.

[3] *N. Yeole, P. Sandhya, P. Chaudhari, and P. Bhujbal, "Evaluation of Malva sylvestris and Pedalium murex mucilage as suspending agent,"International Journal of PharmTech Research*, vol. 2, no. 1, **(2010)**, pp. 385-389.

[4] *A. K. Kulshreshtha, O. N. Singh, and G. M. Wall, Pharmaceutical suspensions: from formulation development to manufacturing: Springer,****(2009)***.

[5] *S. Deshmukh, Y. Katare, S. Shyale, S. Bhujbal, S. Kadam, D. Landge, D. Shah, and J. Pawar, "Isolation and evaluation of mucilage of Adansonia digitata Linn as a suspending agent,"Journal of pharmaceutics*, vol. 2013, **(2013**.

[6] *T. G. Haile, G. G. Sibhat, E. Tadese, D. Tesfay, and F. Molla, "Evaluation of grewia ferruginea hochst ex A. Rich mucilage as suspending agent in metronidazole benzoate suspension,"BioMed Research International*, vol. 2020, **(2020**.

[7] *Y. Mengesha, A. Tuha, Y. Seid, and A. A. Adem, "Evaluation of Aloe weloensis (Aloeacea) Mucilages as a Pharmaceutical Suspending Agent,"Advances in Pharmacological and Pharmaceutical Sciences*, vol. 2021, **(2021**.

[8] *S. Anyebe, Y. Apeji, and O. Olayemi, "The suspending properties of Cissus rubiginosa fruit mucilage in paracetamol suspension formulation,"Nigerian Journal of Pharmaceutical Research*, vol. 13, no. 1, **(2018)**, pp. 19-25.

[9] *S. Olayemi, Y. Anyebe, and O. Apeji, "The suspending properties of Cissus rubiginosa fruit mucilage in paracetamol suspension formulation,"Nigerian Journal of Pharmaceutical Research*, **(2019**.

[10] *C. Alalor, and K. Augustine, "Assessment of Colocasia esculenta Mucilage as Suspending Agent in Paracetamol Suspension,"Saudi journal of medical and pharmaceutical sciences*, vol. 3, no. 7, **(2017)**, pp. 752-755.

[11] *R. Deveswaran, S. Bharath, S. Furtado, S. Abraham, B. Basavaraj, and V. Madhavan, "Isolation and evaluation of tamarind seed polysaccharide as a natural suspending agent,"Int J Pharm Biol Arch*, vol. 1, no. 4, **(2010)**, pp. 360-363.

[12] *B. Panda, "Effect of Tamarind seed mucilage on rheological properties: evaluation of suspending properties,"International Research Journal of Pharmaceutical Sciences*, vol. 1, no. 1, **(2010**.

[13] *B. Divekar Varsha, and K. Mohan, "Isolation and characterization of mucilage from Lepidium sativum Linn. seeds,"Internaional journal of pharma research and development*, vol. 2, **(2010)**, pp. 1-5.

[14] *M. G. Kalaskar, R. E. Mutha, A. U. Tatiya, S. D. Firke, S. J. Surana, K. A. Dhoka, and K. Heda, "Purification and modification of neem gum for enhancement of its suspending property,"Future Journal of Pharmaceutical Sciences*, vol. 7, no. 1, **(2021)**, pp. 1-8.

[15] *R. Kumar, M. Patil, S. R. Patil, and M. S. Paschapur, "Evaluation of Abelmoschus esculentus mucilage as suspending agent in paracetamol suspension,"International Journal of PharmTech Research*, vol. 1, no. 3, **(2009)**, pp. 658-665.

[16] *E. Jarald, E. Sheeja, D. Shweta, A. Raj, and G. Smita, "Application of hibiscus leaves mucilage as suspending agent,"Indian Journal of Pharmaceutical Education and Research*, vol. 41, no. 4, **(2007)**, pp. 373-375.

[17] *A. Rignall, "ICHQ1A (R2) stability testing of new drug substance and product and ICHQ1C stability testing of new dosage forms,"ICH quality guidelines: an implementation guide*, vol. 3, **(2017**.

[18] *B. Anroop, S. Bhatnagar, B. Ghosh, and V. Parcha, "Studies on Ocimum gratissimum seed mucilage: evaluation of suspending properties,"Indian journal of pharmaceutical sciences*, vol. 67, no. 2, **(2005)**, pp. 206.

[19] *A. Deicke, and R. Süverkrüp, "Dose uniformity and redispersibility of pharmaceutical suspensions 2: assessment of three commercial erythromycin ethyl succinate oral liquids,"European journal of pharmaceutics and biopharmaceutics*, vol. 49, no. 1, **(2000)**, pp. 73-78.