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Pharmaceutical excipient development from natural source

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ABSTRACT

Nature has provided us a wide variety of materials to help, improve and sustain the health of all living things either directly or indirectly. Gums and mucilage's are widely used natural materials for conventional and novel dosage forms. These natural materials have advantages over synthetic ones since they are chemically inert, nontoxic, less expensive, biodegradable and widely available. They can also be modified in different ways to obtain tailor-made materials for drug delivery systems. The USFDA also encourages innovation in excipients to develop new drugs or improve approved therapeutics and IPEC is helping to drive acceptance of these novel products into the market .The objective of the present research is to assess the use of constituents of *Cassia Roxburghii* as a novel excipient in the development of suitable pharmaceutical dosage forms. The identified excipients were prepared from the seeds of *C. Roxburghii* by solvent extraction. The final extracted mucilage was freeze dried to obtain a fine gum powder. The obtained novel excipients were characterized by various physical, phytochemical and structural identifications techniques; the ideal excipient characteristics such as Bulk density, Tapped density, Angle of repose and Carr's index results were comparable to the reference excipient (selected from IPEC - FDA). Further, the results from FT-IR, MALDI-TOF, MALDI-ESI and DSC confirmed the presence of saccharides as carbohydrates in multiple units and amino acids in variable amounts. It is common practice in the pharmaceutical industry to incorporate amino acids and saccharides into the final drug product formulation to enhance stability.

INTRODUCTION

We've all read about it: the US Food and Drug Administration approved 17 New Molecular Entities and 2 biologics in 2007, continuing the downward trend in drug approvals occurring over the last decade. The number of drug approvals in 2007 is the lowest number since 1983 and 68% lower than the number approved in 1996. Analysts have suggested many potential explanations for this trend, from tightening safety standards at the Agency to increasing costs and complexity of clinical trials and a shift in emphasis at pharmaceutical manufacturers away from truly ground-breaking treatments. One area that has not received much scrutiny is the lack of innovation in the area of pharmaceutical excipients.[1] The choice of excipients can spell the commercial and scientific success or failure of each active pharmaceutical ingredient (API): an API which performs unique and beneficial functions in biologic systems but cannot be formulated in a viable delivery system is essentially useless. Nevertheless, global excipient research and development are frozen in time, hamstrung by inconsistencies in the regulatory and safety framework which do not allow for approval of new excipients outside the context of a New Drug Application (NDA). Pharmaceutical manufacturers, wary of any additional risks to approval, are therefore reluctant to use new excipients in formulations under development, and excipient manufacturers are finding it difficult, if not impossible, to find customers for their innovative products[2,3].The International Pharmaceutical Excipients Council defines excipients as "substances, other than the active drug substances of finished dosage form, which have been appropriately evaluated

for safety and are included in a drug delivery system to either aid the processing of the drug delivery system during its manufacture; protect; support; enhance stability, bioavailability, or patient acceptability; assist in product identification; or enhance any other attributes of the overall safety and effectiveness of the drug delivery system during storage or use"[4].Novel excipients could represent a needed source of innovation for the pharmaceutical industry. The current regulatory system is a substantial barrier to such innovation. Since most excipients are designed to be biologically inactive, removing this barrier could provide a potentially immediate, low-risk solution to the current shortage of new drug formulations. The current system of excipient approval, in which approval in the first drug application provides automatic approvals for subsequent applications for similar routes of administration, may serve only to add an unnecessary barrier to the development of drug formulations containing novel excipient. The excipients used in pharmaceutical preparations are limited and from an academic point of view there is a clear requirement for new excipients. For an excipient to be approved as a part of a formulation its inclusion has to be justified, the compatibility with the active ingredient shown, and the quality (or grade) will have to be either justified or shown to be sufficient to fulfill the requirements for the final product. Furthermore, the suggested amount of excipient must be shown to be sufficient for the intended function of the excipient. No official lists are readily available to guide on the amount, types and use of excipient. Nevertheless, the Federal Drug Agency (FDA) has made a database and an Inactive Ingredients Guide from 1996 publicly available, in which the use of various excipients in registered products for the different delivery pathways is listed. A similar list was available at one point on the European Medicines Agency (EMEA) site [5]. In drug formulation, the safety of excipients is as important as the safety of the active product ingredient. For well-known excipients that have been recognized and used for a long time, the question of safety is mainly one of quality control of the products received from suppliers. For new excipients, thorough documentation is required including its safety, toxicity and immunogenicity by the EMEA and FDA. This is expensive as well as time-consuming and unfortunately is hampering the development of new excipients, but as the safety of the patients is paramount, this is a necessity. The concentration level of the excipients must be qualified with respect to safety before use in clinical trials. This is one of the main barriers to the use of new excipients in pharmaceutical products, as safety studies are time-consuming and expensive [6, 7].

There is a strong need and necessity to increase awareness about new excipient developments among pharmacy professionals and other technocrats. However, there is a strong need for additional information and guidelines regarding the development, characterization, and quality of new excipients and new applications of the current excipients. The pharmacopoeias and international councils have spearheaded some efforts to develop and harmonize the standards as well as to provide guidelines on good manufacturing and distribution practices for excipients. However, additional efforts are necessary to develop comprehensive and authoritative standards to promote innovation in the area of excipients in order to improve the understanding of the importance of excipients in the global market. [8] By exploring the usage of above stated product in pharmaceutical excipients will enhance the confidence about novel and or natural excipients and it will pave the way for further usage in pharmaceuticals.

MATERIALS AND METHOD

Novel excipients for the study were chosen from plant source of *Cassia roxburghii* due to the prior reported property to use as binding agent in tablets [9]. Red cassia is a graceful tree with spreading, drooping branches appearing to be over weighted by its wealth of clustering orange-red blossoms. The tree has alternate pinnate leaves in pairs with colored margins which further enhance the appearance. The leaves are about 1 ft. long and each has 15-20 pairs of oblong 2 in leaflets. Red cassia produces clusters of pink, rose or orange flowers in axillary and terminal, often branched, racemes. Sepal cup is hairy, with sepals ovate, 4-7 mm long. Petals are 1-1.5 cm long, oblong-obovate, hairy externally. Stamens are 10, 4 long, 3 medium sized and 3 small, not swollen in the middle. The fruit is a typical legume: it is cylindrical and indehiscent (does not split open by itself), 8-12 in long, less than 1 in in diameter, and bears many seeds separated by papery partitions [10].

The following sections briefly discuss various methods and materials utilized for this work.

The *C. roxburghii*, seeds were obtained from Department of Siddha, Tamil University, Thanjavur and Authenticated by G. V. S. Moorthy, Botanical Survey of India (BSI), Southern circle, Coimbatore, Tamil Nadu. The collected seeds were dried at room temperature (25^0C) for 24 hours and powdered by using grinder. The seeds of *C. roxburghii* were broken by mechanical impounding followed by powdering with elite grinder. The powder were defatted by soxhlet extraction using petroleum ether (60 ^{o}C - 80 ^{o}C) and repeatedly extracted with hot water till the complete mucilage was extracted. The mucilaginous solution was filtered through fine muslin cloth and precipitated with acetone. The extracted mucilage was freeze dried to get fine gum powder. The seed powder was soaked in sufficient water, kept over boiling water bath for 30 mins. With occasional stirring, left overnight and filter using muslin cloth. Then the mucilage obtained was freeze dried to get gum powder by using freeze drier (Fig 1). The obtained novel excipients were characterized by various physical, phytochemical and structural identifications techniques. The separated gum powder was evaluated for solubility, swelling index, loss on drying, bulk density, tapped density, angle of repose, carr's index. The extracted seed gum was subjected to some preliminary tests to confirm the nature of the obtained mucilage. The tests performed were to determine the presence of carbohydrates, starch, lipids, proteins and aminoacids, Gelatins, alkaloids, glycosides, terpenoids, volatile oils, tannins and resins [11]. To understand the novel excipients structural components FT-IR, MALDI and DSC were used. Fourier transform IR spectra were recorded on FT/IR-4100 type A. The spectra were recorded for *C. roxburghi*, and formulation made with the same. Samples were prepared in KBr disc (2 mg sample in 200 mg KBr).The scanning range was 400-4000 cm^{-1}, resolution was 4 cm^{-1}. The instrument was purged with nitrogen, and single-sided interferograms were measured without apodization. Absorbance was measured at a resolution of 4 cm-1, and a total of 1000 scans were co-added. Spectral noise was removed by smoothing using a 7-point function, and the absorbance was deconvoluted by the calculation of the second derivative using Grams/32 Spectral Notebase software (Thermo Galac-tic, NH). The absorbance of water was subtracted from that of protein using the criteria of a straight baseline from 2000 to 1720 cm^{-1} and no negative absorbance peaks. Solid-state protein spectra were measured using pressed potassium bromide discs containing 0.2% protein.

Matrix-Assisted Laser Desorption/Ionization (MALDI) Analysis:

MALDI is a soft ionization technique used in mass spectrometry, allowing the analysis of biomolecules (biopolymers such as DNA, proteins, peptides and sugars) and large organic molecules (such as polymers, dendrimers and other macromolecules), which tend to be fragile and fragment when ionized by more conventional ionization methods.

DSC ANALYSIS (T_g):

T_g of the novel excipient with freeze-dried formulations was measured by heating approximately 5 mg of freeze-dried powder in a sealed sample tray using a DSC (Q200) differential scanning calorimeter (TA Instruments). Samples were equilibrated to 10°C and heated to 150°C at a rate of 1°C/min, and the transition temperatures were determined using the instrument software.

RESULTS AND DISCUSSION

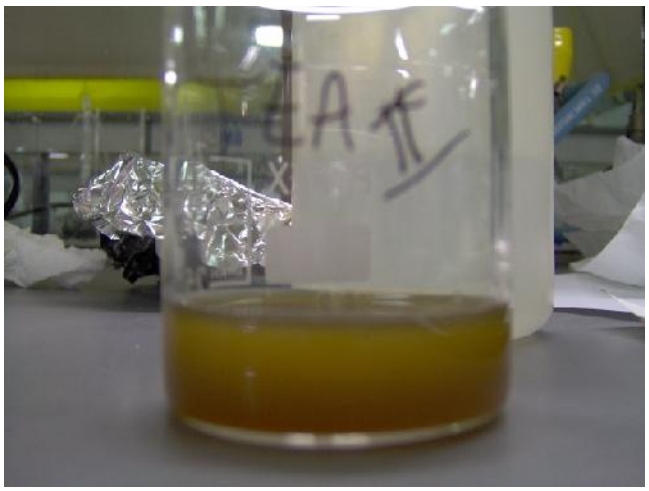
During the manufacturing process, the manufacturing yields were checked at different stages as in process checks. Defatted Seed Gum: yields were 24%; After Drying process: yields were 22%.

The excipient was White to off-white in color, Free flow powder, Non-sticky and non-adherent to the containers and closures. The manufacturing processes were consistent with yield and reproducible between batches. The qualities of novel excipients were constant between batches. All the process parameters were monitored for process optimization and observed to be reliable. The obtained novel excipient was soluble up to 25 grams/Liter in water. So according USP 23 (33 to 10 g/L),

Table 1: Excipient Properties

Parameters	Excipient Concentration					
	2%		4%		6%	
Formulation	Novel Excipient	Refer--ence	Novel Excipient	Refer--ence	Novel Excipient	Refer--ence
Bulk Density (g/cc)	0.47	0.48	0.42	0.42	0.46	0.48
Tapped Density (g/cc)	0.56	0.52	0.53	0.53	0.52	0.53
Angle of Repose (θ)	24.90	22.86	26.22	26.22	23.7	24.0
Carr's Index	16.07	9.615	13.20	13.20	10.04	9.43

Filtered seed gum



Freeze dried gum powder

Figure 1 : Isolation of Seed gum

Table 2: Phytochemical Results

S.No	Components	Name of test	Results*	Comments
1	Carbohydrates	Fehling solution test	-	Absence of reducing sugars
		Benedict's test	-	Absence of reducing sugars
		Molish test	+	Presence of carbohydrates.
		Seliwanoff's test	-	Presence of aldoses
2	Starch	Jelly test	+	Presence of starch
3	Lipids	Grease spot test	-	Absence of lipids
		Emulsification test	-	
4	Proteins and amino acids	Biuret test	+	Presence of peptide linkage
		Millons test	+	Presence of phenolic hydroxyl group
		Xanthoprotic test	+	presence of aromatic ring in amino acids or aromatic ring containing amino acid in proteins
5	Gelatin	Solubility test	-	Absence of gelatin
		Soda lime test	-	
6	Alkaloids	Dragendorff's test	+	Presence of alkaloids
		Mayer's Test	+	
		Tannic Acid Test	+	
7	Glycosides	Bomtragor's Test	+	Presence of Anthraquinone Glycosides
		Haemolysis test	-	Absence of Saponin Glycosides
		Libermann Bruchard test	+	Presence of steroid and triterpenoid glycosides
		Keller Killiani test:	+	Presence of Cardiac glycosides
		FeCl3 test	-	Absence of Chemical tests for Coumarin glycosides
		Sodium picrate test	+	Presence of Cynophoric glycoside
		Ammonia test	-	Absence of flavonoid glycosides
8	Terpenoids	Chemical test	-	Absence of Terpenoids
9	Volatile oils	Chemical test	-	Absence of Volatile oils
10	Resins	Turbidity test	-	Absence of Resins
11	Tannins	Test with Iron salts	-	Absence of Tanins

these substances fall under sparingly soluble category. There was no volume changes observed in both the solutions containing novel excipients. Results showed that, swelling index of novel excipient were constant before and after the experimental duration. This shows that, swelling capability of novel excipients is less and it may be due to poor wetting properties and less viscous nature. Loss on drying was 4 %. The LOD were observed to be constant between bathes.

Pre compression Parameters for novel excipients like Bulk density, Tapped density, Angle of repose and Carr's index were measured and compared with reference excipient at particular concentrations (Table 1). The results shown that, there were marginal differences observed in Carr's index only at 2% level of concentrations between novel excipient and reference. This could

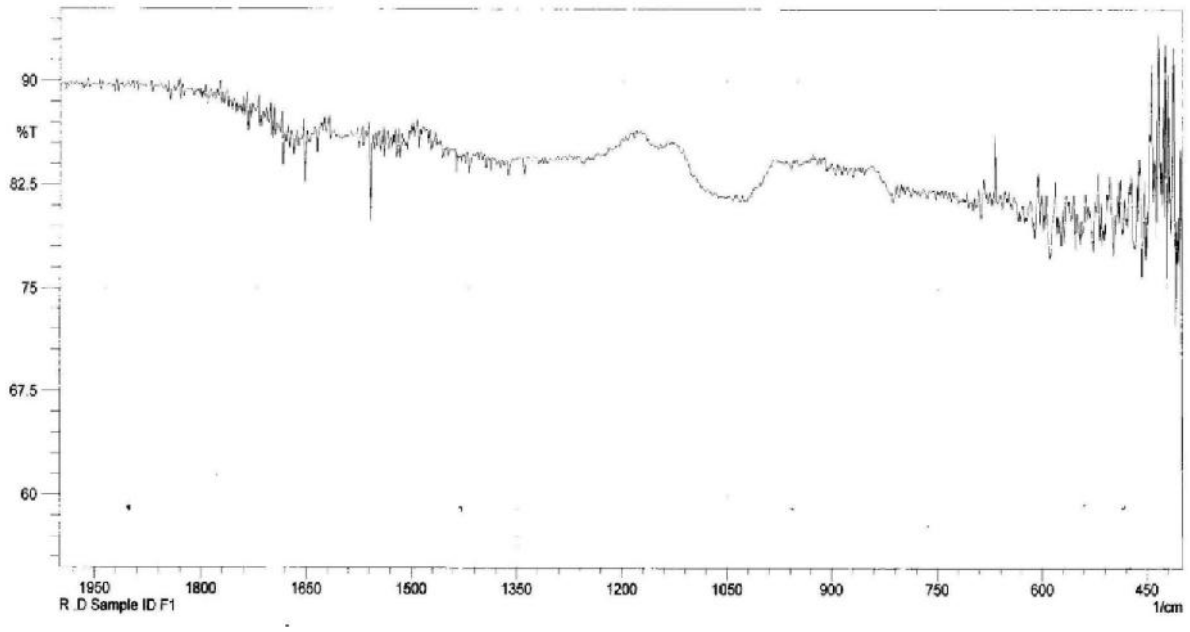


Figure 2 : FT-IR Profile of Novel Excipients

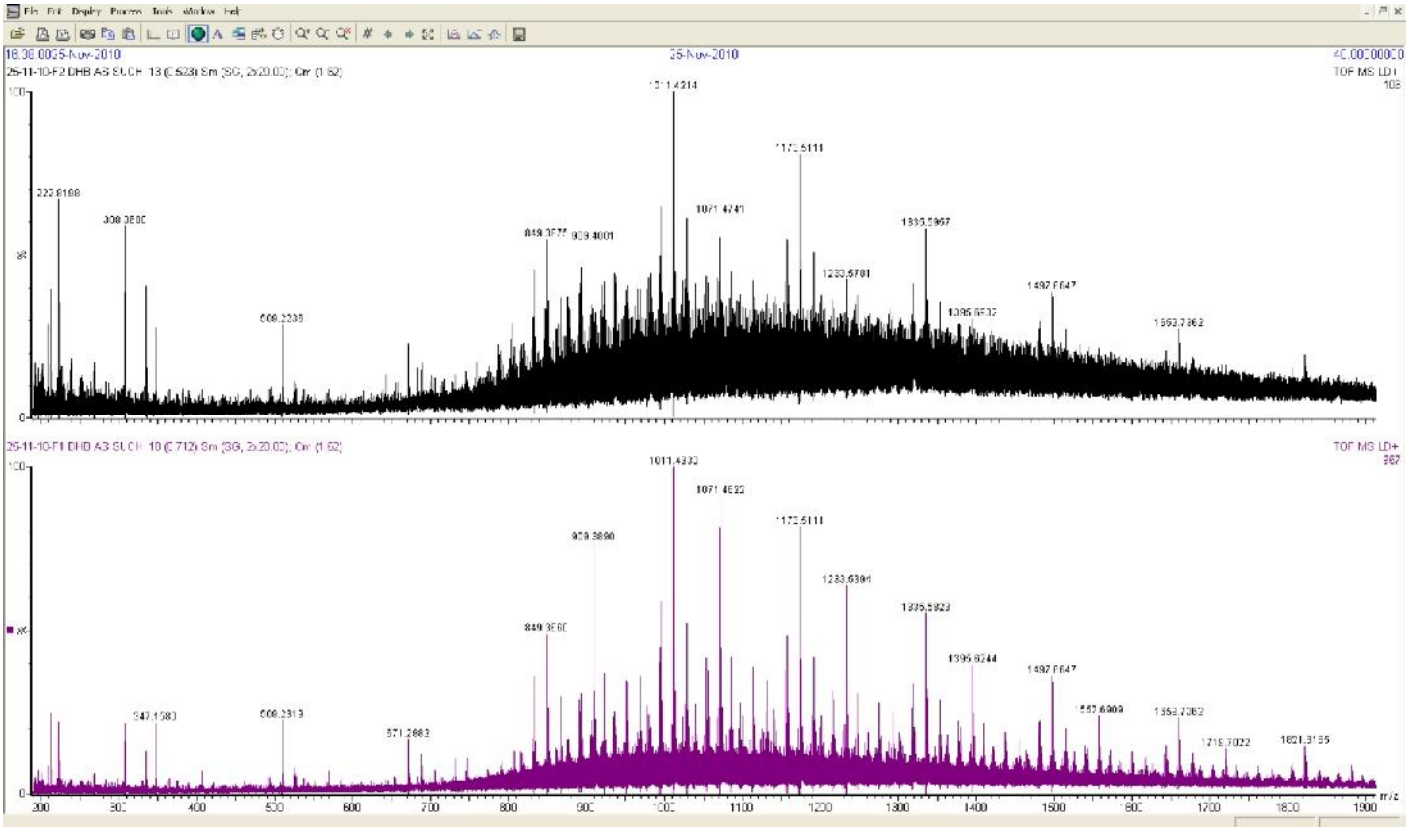


Figure 3 : MALDI-TOF profile for Novel Excipients

be due to the inherent properties of the novel excipients at this particular concentration. During any kind of compressibility calculations with novel excipients, these results need to be considered. Remaining parameters observed to be comparable between the components and concentrations.

From the phytochemical results (Table 2), the novel excipient may contains variable amounts of following components like Carbohydrates in aldose in nature, Aromatic amino acids Atypical or typical Alkaloids Anthraquinone, steroid and triterpenoid, Cardiac and Cynophoric glycosides. The characterization studies were done by FTIR studies i.e. by using

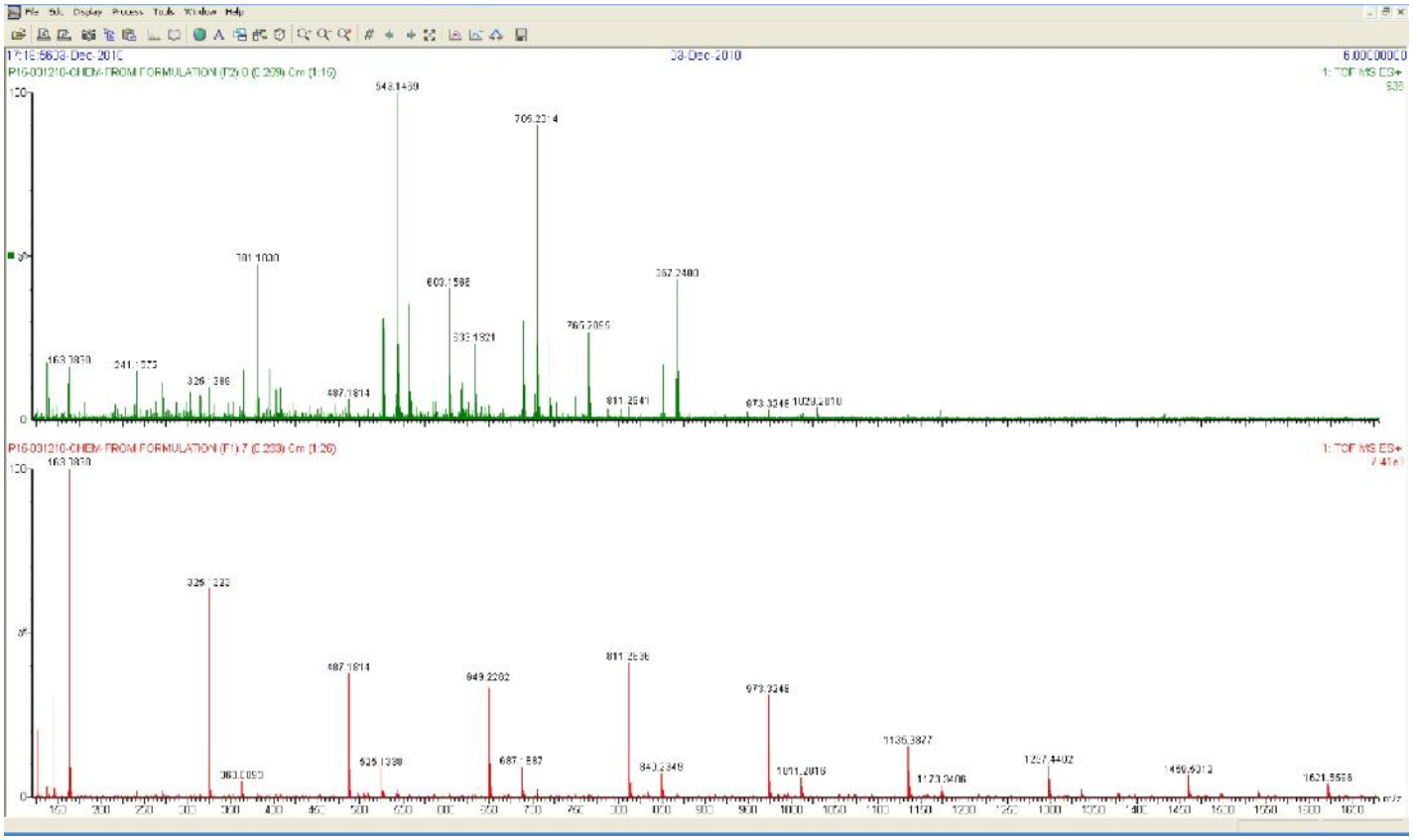


Figure 4 : MALDI- ESI profile for Novel Excipients

KBr pellet method. The FT-IR profiles were shown (Fig 2). Further, the results from FT-IR, MALDI-TOF (Fig 3), MALDI-ESI (Fig 4) and DSC confirmed the presence of saccharides as carbohydrates in multiple units and amino acids in variable amounts.

CONCLUSION

The suitability of the novel excipient for using in pharmaceutical dosage forms were evaluated in comparison to a USFDA approved excipient in that category. The identified excipient was prepared from the seeds of *C. Roxburghii* by solvent extraction. The final extracted mucilage was freeze dried to obtain a fine gum powder. The selected novel excipient was evaluated for ideal excipient characteristics such as Bulk density, Tapped density, Angle of repose and Carr's index and the results were comparable to the reference excipient (selected from IPEC - FDA). Further, by using FT-IR, MALDI-TOF, MALDI-ESI and DSC, the novel excipient was characterized as a mixed population of polymers and monosaccharides in variable amounts. It was found that the novel excipient performed on par with the proven USFDA approved excipient. All study results support the usage of novel excipient in pharmaceutical dosage forms. To explore the possibilities for using the identified excipient in pharmaceutical formulations, further studies need to conduct the explorative pharmacokinetic and pharmacodynamics studies in suitable animal model as per the ICH guidelines [12]; studies to be planned with API to ascertain its role in pharmaceutical formulations and structural level identifications to be explored with suitable physio-chemical methods in order to establish exact chemical structure.

REFERENCES

1. Girish K Jani, Gums and mucilages, Asian Journal of Pharmaceutical Sciences 2009, 4:5, 309-323.
2. Christopher C. DeMerlis, Jay M. Goldring, Ranga Velagaleti, William Brock, Robert Osterberg, Regulatory Update: The IPEC Novel Excipient Safety Evaluation Procedure, Pharmaceutical Technology, 2009, 33(11), 72-82.
3. Dr. Jay Goldring, Novel Excipients: The Next Pharmaceutical Frontier? Drug Delivery, American pharmaceutical review, 2009, 12.
4. Ogaji IJ, Nep EI, Audu-Peter JD, Advances in Natural Polymers as Pharmaceutical Excipients, Pharm Anal Acta, 2012, 3:146
5. EMA, scientific guideline for Novel excipient, 2003
6. Guidance for Industry, Nonclinical Studies for the Safety Evaluation of Pharmaceutical Excipients, 2009.
7. Guideline on Excipients in the Dossier for Application for Marketing Authorization of a Medicinal Product, EMEA/CHMP/QWP/396951/2006.
8. Lene Jorgensen, Susanne Hostrup, Eva Horn Moeller, and Holger Grohganz, Recent trends in stabilising peptides and proteins in pharmaceutical formulation considerations in the choice of excipients; Expert Opinion on Drug Delivery, 2009, 6(11), 1219-1230.

9. Kundlik Girhepunje, Arulkumaran, Ranju Pal, Nitin Maski, N Thirumoorthy, A Novel Binding agent for pharmaceutical formulation from Cassia roxburghii seeds, IJPS, 2009, 1, 1-6.

10. Website: http://www.flowersofindia.net/catalog/slides/Red%20Cassia.html.

11. Sayeed Ahmad, Introduction of Plant Constituents and their Tests, Pharmacognosy, 2008, 3-40.

12. EMA, Scientific guideline for Novel excipient, 2003.