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Isolation, characterizaton and acute toxicity study of Terminalia elliptica (Saj) gum, Buchanania lanzan (Chironji) gum and Albizia lebbeck (Siris) gum

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***Abstract*---**The current investigation was focused on the collection, isolation, purification and acute toxicity study of Saj (*Terminalia elliptica*) gum, Chironji (*Buchanania lanzan*) gum and Siris (*Albizia labback*) gum. Each gums were collected from the bark of the tree, isolated and evaluated for various parameters. The results showed the total ash content 3.23± 0.61 %, 3.54± 0.69 % and 4.53 ± 0.02 %, loss on drying 10.31 ±0.7%, 6.8± 0.50% and 8.41 ± 0.31%, viscosity (4% w/v aqueous solution) 42±0.95cps , 38 ±.61 cps and 77 ± 0.23 cps , Swelling index 91.00± 1.69 %, 89.42± 1.93 % and 128.94± 2.02 %, hausner's ratio 1.23± 0.06, 1.18± 0.05 and 1.12± 0.03, compressibility index 19.04± 1.01 %, 15.73± 0.96 % and 12.86± 0.94 %, angle of repose 23.37±0.03 ^{0} , 16.04±0.15^0 and 13.37±0.21 ^{0} and pH 4.23 to 5.58 , 4.20 to 5.05 and 4.89 to 5.29 respectively for Saj gum, Chironji gum and Siris gum. Various qualitative photochemical tests confirmed the presence of carbohydrates and protein while absents of alkaloids, tannin, cardiac glycosides, terpenoids and steroids in all these gums. All these gums form a viscous gel in water and insoluble in organic solvents. The oral acute toxicity study on albino mice revealed that the LD_{50} of each gums were greater than

2000 mg/ kg body weight of animal, hence considered safe for oral use. FTIR spectral analysis pointed towards the presence of some common functional groups such as alcohol, carboxylic acids, esters, ethers, alkanes, alkynes, alkene phenols, amines in all these gums. The overall results concluded towards the good quality, purity, pH , compressibility (specially Saj and Chironji) , swelling index, viscosity, powder flow and safety profile of these gums, hence could be an alternative for various synthetic excipients in pharcaceutical sectors in future.

***Keywords*---**Terminalia elliptica (Saj), Buchanania lanzan (Chironji), Albizia lebbeck (Siris), natural gums, pharmaceutical adjuvant.

Introduction

In recent years ,there have been a significant improvements in designing of new medicinal dosage forms for the existing and new drugs. Due to its non toxic, biocompatible, bioerodable, inertness widely available and cost saving natures, natural gums are widely used as an excipients for the development of many pharmaceutical dosage forms. However it comes with many challenges such as such as microbial growth, batch to batch variation, alternation in rheology, swelling behavior, variation in collection source , time of collection, purity and reduced viscosity (Girish et al 2012 and Meka et al 2009). If all these lacunas are overcome, it could be the superior alternative to high cost , low compatible, non biodegradable synthetic polymers used in formulation of various dosage form. (Meka et al 2009) Terminalia elliptica Willd is a gum yielding species, commonly known as 'Saj" is belongs to genus Terminalia and family Combretaceae. Its Synonyms are T *alata* T *crenulata* T *tomentosa* etc In hindi it is called as Asana or Saj and in odia called as Sahaju. It is a large tree of of around 20-35 m hight common in the forests, specially in the humid regions of India, including the sub-HimalaSyan tracts of North West provinces,Sikkim and in many states including Odisha (Joshi et al 2013) . It has been reported that the plant contain phytoconstituents such as Oxalic acid, pyrogallol and catechol (Cock 2015). Many Previous literature reveled that since a long time many parts of the plant traditionally has been used in the conditions like dysentery, diarrhoea, pitta, ulcers, vata, fractures, haemorrhages, bronchitis cardiopathy, etc (Kirtikar 1991, Varier 1993) The plant parts have reported for many pharmacological properties like antidiarrhoeal, anti leucorrheal (Mahato et al 2005) antioxidant and anti microbial (Jain et al 2010) . Antifungal (Srivastava et al 2001, Shinde et al 2011) anti-hyperglycaemic (Alladi et al 2012) , antioxidant and antiepileptic activity of leaves exatract (Uddin et al at 2015). strong anti-oxidant , anti-secretory (Khan etal at 2017) anti inflammatory (Khatoon et al 2018) and antibacterial activity (Krishna et al 2020) of leaves extract, anti-infammatory anti arthritis activity of bark extracts (Jitta *at.al* 2019). *Buchanania lanzan* Spreng, is a gum yielding species commonly known as Char or Chironji in hindi and Charu in Oriya. It is belongs to genus Buchanania and family Anacardiaceae. Synonyms of the plant are Buchanania Cochinchinensis Lour, Buchanania latifolia Roxb etc (Rai et al 2015). This is a wild tree found in tropical formests up to 1200 m of Nort, west and central India in the states of Odisha, Chattisgarh, Jharkhand, Madhya

Pradesh, Gujrat, Maharashtra, Uttar Pradesh, Bihar, Varanasi itc.These plants also found in Asian coundties like Australlia, Malasia, Pacific islands. Out of seven species the fruits of Buchanania lanzan an Buchananuua axillaries of angustifolia are eadible.(Siddiqui et al 2014, Neeraj et al 2020) .Since a long time all the parts of the plants has been used as traditional medicine as expectorant, blood diseases, aphrodisiac, purgative, blood purifier and cures digestive disorders and wound healing, fever. laxative etc *(Rajput etal 2018*). Bark Powder in infantile diarrhea (Amol et al 2010). Different Pharmacological studies using various animal models and antimicrobial studies revealed that all the parts of the plants like root, bark, leaf, fruit, seed, gum etc has potential towards antioxidant (Mehta et al 2009), analgesic, anti-inflammatory (Pattnaik et al 2011) . Anti-Diabetic, Anti-Hyperlipidemic (Sushma et al 2013) , anti-diarrhoeal, (Kodati et al 2010) , diuretic (Hullatti et al 2014) antistress (Mehta et al 2011) DNA protective (Shailasree et al 2012) memory enhancing (Neelakanth et al 2012) [18] Chemopreventive (Jain et al 2012) Anticancer (Sumithra et al 2013) Wound Healing(Chitra et al 2009) anti snake venom (Naseeb et al 2014) [22] hepatoprotective, anti-bacterial and anthelmintic (Nagulwar et al 2020) [23] activities. Isolated mucilage from the seeds shows sufficient mucoadhesive strength (Singh *at a 2014)* Albizia *lebbeck*, is an another gum yielding Indian species commonly known as Siris or Indian walnut. It is belongs to the family Fabaceae.It is widely cultivated in tropical and subtropical regions. in region of India (Yadav et al 2011) [57] Traditionally many parts of the plants has been used medicinally as astringent, cough ,ophthalmic infection, flu, gingivitis, lungs problems , abdominal tamers. inflammation etc (welth of india 2006). The above literatures states that these plants since a long time has been used by human being as the traditional medicines and various parts such as root, leaves, bark, fruits has been reported to shown various pharmacological effects. However a little work has been done to words the exploitation of the gum exudates of these plants. Keeping this in view the recent work is focused on the collection, isolation, purification and physicochemical rheological, and toxicological characterization of the Terminallia elliptica (Saaj), Buchanania lanzan (Chironji) and Albizia lebbeck (Siris) gums for the pharmaceutical use in future.

Material and Methods

Material

The fresh gums exudates were collected separately during the month of January – March. by hand tapping method from from the bark of the *Terminalia elliptica* (Saj*), Buchanania lanzan (Chironji)and Albizia labback (Siris)* in the forest of Narsinghnath of Bargarh District. based on the procedures adopted from the previous studies (Farooq *et al* 2013, Shahb et al 2009). All the other chemical used were of analytical grade.

Method
Authentication of Plants

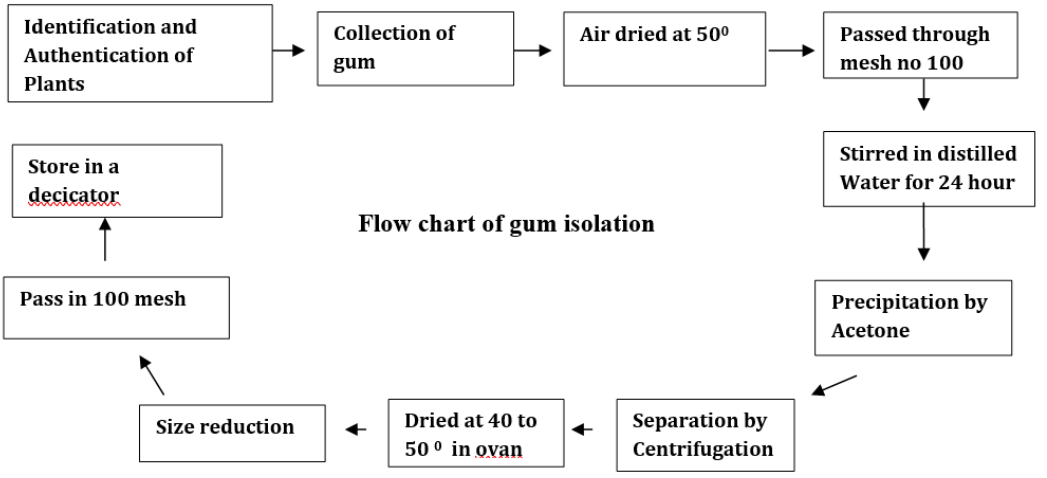
Identified plants of Saj (Terminalia elliptica), Chironji (Buchanania lanzan) and Siris (Albizia lebbech) were authenticated by Prof. Dr Surya kumar Badapanda, Head Department of Botany, Rampur college, Sonpur, Odisha.

Extraction and Isolation gums
Steps1 Extraction

Some deep cuts were made in the bark and gums which ooze out from these cuts were collected. and cleaned by removing the external bark and other extraneous materials by hand. Finally dried in hot air oven at 50°C until it become sufficiently brittle. Then the dried gums were triturated properly in a high speed mechanical blender (Bajaj india) and passed through mess no. 100. The powdered gum dispersed in distilled water in a ratio of of 1 part to 10 parts of the water. Gums were extracted by dispersing the crude powdered gum in purified water under continuous stirring using rotary shaker (Remi instrument ltd,Mumbai, India) for 24 hrs at room. After that the supernatant was separated by filtration through a muslin cloth the remaining residue was further washed with water and the washings are mixed with the supernatant. The above procedure was repeated for 3 times (Sujitha et al 2012 an Malveya et al 2011)

Step 2- Isolation

Supernatants collected in the above procedure were treated with two times of its volume of acetone to isolates the gum in the form of precipitate. The precipitated were separated by the process of centrifugation. Then the precipitate was kept in hot air oven at 40-50^{o} till complete drying. Finally each isolated gums were powdered and passed through sieve number 100 and stored in desiccators for subsequent tests. **(**Kadajji et al 2009, Chatterjee et al 2019)



In-Vitro Characterization of isolated gum powder
Percentage yields

The percentage yields of the isolated gums are given in the Table 1 were calculated by following equation (Bhosle *et al* 2015).
% Yield = (W2/W1) x 100 -------[1]
Were W1= The weight of crude raw gum powder before dispersed in water

W2= Weight of the isolated gum powder found after final step of isolation procedure

Organoleptic properties

Organoleptic properties isolated gums powders were verified by visual inspection (Malvya *et al* 2011) [34] and results are given in Table 4.

Solubility studies

Around 10 mg of isolated gum powder was taken in 10 ml of ethanol, acetone, chloroform and water to observe the solubility of gum and results are given in Table 3 (Lala 1991)

Loss or loss on drying (LOD)

Tared weighed bottles were filled with 1 gm of the each gum separately and dried in a hot air oven at 105^0Centigrade. Weight of the dried gums were measured after 1 hour interval until a constant weight were obtained. The LOD was calculated for each gum by going through following equation (Das *et al* 2014 and Kulkarni *et al* 2002) and the results are summarized in Table 5.

$$\text{LOD} = \frac{\text{initial weight} - \text{final weight}}{\text{initial weigh}} \times 100 \text{ ----------eq.[2]}$$

Ash value

To determine the purity and quality of crude gums ash value is an important parameters.The residue obtained after the total incineration of the isolated gum powder called as taoal ash value. part which is insoluble in the dilute hydrochloric acid called acid insoluble ash and part is which is dissolve in water called water-soluble ash. The ash value can be calculated by following formula (Khandelwal *et al* 2010) and results are given in Table 5.

Total ash value=(weight of ash obtained by a gum/ weight of the powder gum)×100--- eq.[3]
Acid insoluble ash = (weight of acid insoluble ash of specific gum /weight of dried gum powder)×100--eq.[4[
Water soluble ash = [(weight of water-soluble ash) /weight of dried gum powder] ×100------eq.[5[

Swelling index

One gram of the each gums were taken in a 25 ml measuring cylinder and made up the volume up to 25 ml and shaken thoroughly in the gap of 10 minutes for around 1 hour. Then the cylinder was set aside of 24 hours at room and swelling volume of gums were measured and % swelling index were calculated using following equation Results are given in Table 5 (Malvya *et al* 2010).

Swelling index= [(Final volume -initial volume)/ Initial volume]×100------eq[6]

pH Determination

1% solution of each gums were prepard in water and tested for its pH in a digital pH meter at a specific temperature **a**nd results are given in Table 5 **(**Malvya *et al* 2010).

Viscosity

Viscosity of the each gums were determined using Brook field viscometers by taking 4% of aqeous solution at 10 rpm and 61 spindal at room temperature and results shown in Table 5 **(**Malvya *et al* 2010).

Determiantion of bulk characterization of gums (Srivastava *et al* 2010)

Bulk density and Tapped density

Tapped density and Bulk density determined by bulkdensity apparatus according to following formula and results were given in Table 5

Bulk Density of gum= [Measured weight of gum powder/ Volume of the powder before tapping]---eq[.7]
Tapped Density of gum =[Measured weight of gum powder /Volume of the powder after tapping .-----eq.[8[

Hausener's ratio

It is the ratio between tapped density and bulk density. It is the indirect way to measure the flow property of the powder and results are given in Table.5

Hausner's ratio = Tapped density/ Bulk density----------eq.[9]

Compressibility/Carr's Index Index

This gives the ideas asbout the compression characteristics as well as flow properties of the power. It was calculated by following formula and results are given in Table.5 .

% Compressibility index = [(Tapped Density-Bulk Density)/Tapped density] x 100---------eq [10]

Angle of repose

Angle of repose of the each gum powders are determined separately by fixed funnel method. Powders are accurately weighed were freely passed through a fixed funnel on to the surface. The funnel was arranged in such a way that the tip of the funnel is just touched the peak of the pile of gum powder. The diameter and hight of the piles or powder cones were measured separately for each gums. Angle of repose was calculated by following formul and results are summarized in Table 5.

Tan θ = h/r, ----------eq.[11[

Where, 'h'is the height of the pile and 'r'isthe radius of the powder cone.

Qualitative tests for Phytochemical constituents

Standard procedure was followed for qualitative analysis of phytochemicals of 1% of each gums solution (Trease et al 1989, Harborne 1998 and Singh et al 2018)

Test for carbohydrate

Fehling's test: To each gum solutions Fehling's solution were added and boiled in water bath. If break red precipitated obtained, its confirms the presence of carbohydrate in these gums.Results are given in Table 4
Benedict's reagent: Benedict's solution were instilled to each gum solution and boiled in water bath. If red precipitation is obtained, presence of carbodydrate is confirmed. Results are given in Table 4

Test for proteins

Biuret test: To the 1% of each gum solutions 1 ml soluition of 40% NaOH and just 2 drops of 1% CuSO4 soluiton were added and checked wheather appearance of violet color for the confirmation of protein.Results are given in Table 4
Ninhydrin test: 2 drops of ninhydrine solution(10 mg of ninhydrin in 200 ml of acetone) were added to each gum solution and observed wheather purple colour is formed for the confirmation of protein. Results are given in Table 4

Test for alkaloids

Mayer's test: To the 1% of each gum solutions few drops of Mayer's reagent (KI + Hg2Cl2 solutions) were addedto check the appearance of cream cloured precipitation for he confirmation of alkaloids.Results are given in Table.
Dragondorff's test reagent: To the 1% of each gum solution few drops of Dragondorff"s (excess of KI +BiNO3 solutions) reagent were added. Checked of reddish brown precipitation for the confirmation of the presence of alkaloids. Results are given in Table
Hager's test: To the 1% of each gum solution few drops of Hager's reagent I,e picric acid were added. If yellow color precipitation takes place than alkaloids is present. Results are given in Table

Test for tannins and phenols

Ferric chloride test:
To the 1 % colloidal solutions of each gums, 1 ml of ferric chloride solution were added. If Bluish-black color formed it confirms the presence of tannins. Results are given in Table 4

Test for glycosides

Keller–Killiani test: To gum gums extract 1 ml of concentrated sulphuric acid and 1 ml of glacial acetic acied having ferric chloride traces were added. Formation of a reddish brown color in the junction of the two lalyers and bluish green color of upper layers confirms the presence of glycosides. Results are given in Table 4
Borntrager's test: To the each gum solution1 ml of benzene and half ml of dilutie ammonia solution were added . fommation of a pink reddish color indicates towards the presence of glycosides. Results are given in Table 4

Test for flavonoids

Alkaline reagent test: To the 1% gum solutions NaOH just few drops were added. Flavonoids present, If intencse yellow color is observed and further disappear with the addition of Concentrated HCl it indicates Results are given in Table 4

Test for steroids and terpenoids

Liebermann Burchard test: To each gums extract 1 ml chroloform and anhydrous acetic acid were added followed by cooling at 0 ^{0} centigrade. Than from the side of this testtube 1 ml sulphuric acid were added each mixture. Two layers are formed having a brown ring at the junction. lower layer becomes deep brown indicates the presence of terpenoids and green color formation in upper layer layer indicates that steroids is present. Results are given in Table 4
Salkowski test: To the 1% each gums solutions of 1ml of choloroform and sulphuric acid were added. Raddish brown color in the lower layer indicates the presence of steroids and if upper layer become yellow than terpenoids is present. Results were given in Table 4

Acute toxicity study of gums

Isolated gums of Terninalia elliptica (saaj), Buchanania lanzan (Chironji) and Albizia labback were tested for the acute toxicity studies based on the guide lines no 425 of organization for economic co- operative and development (OECD). The present study was approved by Institutional animal ethical committee (IAEC) having IAEC approval No. CPCSEA/ IAEC/JLS/17/03/22/041. LD $_{50}$ were determined taking healthy Albino mice of either sex. 35 numbers of animals weighing 160–200 g were selected for each gum and randomly divided into 7 groups comprising of five animals each. Among 7 groups, 1 group considered as controlled group and other six taken as test group. Each albino mice of control and test groups were fasted over night. The fasted body weight of each animal were determined. Dose of the gum was calculated for each animals according to the body weight. The control group received normal saline (10 mL/kg per oral) and other groups received 100, 200, 500, 1000, 1500 and 2000 mg/kg of gum dispersion in distilled water. The animals were observed continuously for certain behavioral changes and mortality for first 30 min after dosing and observed periodically for the next 24 hrs with a special attention given in first 4 hrs and then daily thereafter, followed for 14 days. The effect of gums on body weight after 14 days were observed. [1,2] and results were summarized in Table 6 and 7 (Divvela et al 2016)

FTIR analysis of gums

To detect the various functional grups present in these gums Forior Transform Infrared Spectroscopy (FTIR) was performed individually for each Saj gum, Chironji gum and Siris gum. Sample were analyzed using potassium bromide pellet technique were in the region of 3,500 and 500 cm -1 in a standazied IR spectrophotometer. Results are given in Table 8,9,and 10 and Fig.1,2 and 3 (Shankar et al 2010)

Result and Discussion

Table 1: %Yield of isolated gum powder

Name of the gums	Initial weight of raw gum powders (gm)	Final weight of isolated gum Powders (gm)	% Yield
Saj gum	500	36.64±2.10	7.32 ±1.17
Chironj gum	500	32.89 ±2.45	6.57±0.90
Sires gum	500	35.78 ±2.45	7.15±1.31

The symbol 'g'= Gram **,** Values given as mean ± Stadard Deviation (SD), (Number of observation, n=3)

Table2: Solubility of isolated gums

GUM → NAMEOF SOLVENT↓	SAJ	CHIRONJI	SIRIS
Warm water	Soluble	Soluble	Soluble
Cold water	Sparingly soluble to form gel	Sparingly soluble to form gel	Sparingly soluble to form gel
Benzene	Not soluble	Not soluble	Not soluble
Ethanol	Not soluble	Not soluble	Not soluble
Methanol	Not soluble	Not soluble	Not soluble
Acetone	Not soluble	Not soluble	Not soluble
Petroleum ether	Not soluble	Not soluble	Not soluble

Table 3: Organoleptic properties

Properties	Saj gum	Chironji gum	Indian walnut
Colour	Deep brown	Light brown	Beige
Odour	Characteristics	Characteristics	Characteristics
Taste	Sweetish sour	Sweetish sour	Sweetish sour
Texture	Irregular	Irregular	Irregular

Table 4. qualitative test for phytomemical constiturents of isolated gums powders

Sl. No.	Tests	Terminalia elliptica gum	Buchanania lanzan gum	Albuzia labback gum
1	Tests for Carbohydrates:			
	Fehling's test	+ ve	+ ve	+ ve
	Benedict's test	+ ve	+ ve	+ ve
2	Tests for proteins			
	Biuret test	+ve	+ve	+ve
	Ninhydrin test	+ve	+ve	+ve
3	Test for Alkaloids			
	Mayer's test	- ve	- ve	- ve
	Dragendorff 's test	- ve	- ve	- ve
	Hager's test	- ve	- ve	- ve
4	Test for Tannin			
	Ferric chloride test	- ve	- ve	- ve
5	Cardiac Glycocydes			
	Keller–Killiani test	-ve	- ve	- ve
	Borntrager's test	-ve	- ve	- ve
6	Tests for Flavonoids:			
	Alkaline reagent test	- ve	+ve	- ve
	Lead acetate solution test	- ve	+ ve	- ve
7	Tests for Steroids and terpenoids			
	Liebermann Burchard test	- ve	- ve	- ve
	Salkowski test	- ve	- ve	- ve

(+) sign indicates the presence and (-ve) sign indicates the absent of the particular phytoconstituent.

Table 5: Physicochemical & bulk charecterization of isolated gum powders

Properties of gums	Terminalia *elliptica* gum (Saj) gum	Buchanania *lanzan* (Chironji) gum	Albizia *lebbeck (Siris)* gum
Ph of gums (1% W/V solution)	4.23 - 5.58	4.20- 5.05	4.89-5.29
Swelling index %	91.00± 1.69 %	89.42± 1.93 %	128.94± 2.02 %
Viscosity of (1% W/V) cps	42±0.98% , and	38% ±0.61	77% ± 0.23%
Total ash %	3.23± 0.61 %	4.53 ± 0.02	3.54± 0.69
Acid insoluble ash %	1.21± 0.79	1.15 ± 0.63%	1.24± 0.81
Water soluble ash %	1.72± 0.31	1.61± 0.64 %	1.76 ± 0.27
Loss on Drying %	10.31 ±0.7	8.41 ± 0.31%	6.8± 0.50
Bulk Density (gm/cm^3)	0.476± 0.02	0.525± 0.03	0,501± 0.03
Tapped Density	0.588 ± 0.06	0.623± 0.02	0.575± 0.01

(gm/cm^3)			
Hausners Ratio (%)	1.23± 0.06	1.18± 0.05	1.12± 0.03
Compressibility index (%)	19.04± 1.01	15.73± 0.96	12.86± 0.94
Angle of repose (0)	23.37±0.03	16.04±0.15	13.37±0.21

Values given as mean ± Stadard Deviation (SD), (Number of observation, n=3)

Acute Oral toxicity studies

Table: 6 Effect of Gums on the body weight of albino mice at 2,000 mg/kg dose after 14 days.

Group	Treatment	Body weight (g)	
		Before treatment (Mean ± SD)	After treatment (Mean ± SD)
Control	Normal Saline (10 mL/kg per oral)	169 ±2.72	178 ± 2.45
Treated 1	Terminalia elliptica gum dispersion in distilled water (2000 mg/kg)	185 ± 1.89	192 ±1.67
Treated 2	Buchanania lanzan (Chironji) gum dispersion in distilled water (2000 mg/kg)	168 ±1.22	182 ±1.56
Treated 2	Albizia labback (Indian walnut) gum dispersion in distrilled water(2000 mg/kg)	172 ±1.37	179±2.39

Table 7: observations of behavioral parameters of the treated as well as the control animals

	30 Min				4 Hours				1 Day				2 day				7 days				14 Days			
	C	Te	Bl	Al	C	Te	Bl	Al	C	Te	Bl	Al	C	Te	Bl	Al	C	Te	Bl	Al	C	Te	Bl	Al
Mortality	0	0	0	0	0		0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Corneal reflex	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U
Pupils	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U
Salivation	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U
Lacrimation	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U
Hyper activity	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A
Pain	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A
Grooming	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A
Torch response	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U
Skin fur	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U
Alertness	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U
Tremors	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A	A
Pinna reflex	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U
Gripping strength	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U
Sleep	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U
Urination	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U	U

C-Control group given normal saline, A-Absent, U-Usual, Te-Terminalia *elliptica* (Saj)gum, Bl-Buchanania *lanzan(Chironji)* gum and Al-Albizia *lebbeck* (Siris) gum

Gums were collected from the of bark of the plants *Terimnalia elliptica* (Saj), *Buchanania lanzan* (Chironji) and *Ablizia lebbeck* (Siris) during summer season in the month of April and March. Gums were extracted from the raw gums using distilled water. After extraction pure gums were isolated by precipitation with acetone. Percentage yield, were found to be 7,32 %, 6.57% and 7.12 % for Saj, Chironji and Siris gums respectively (Table 1). Solubility data (Table 2) states that, all these gums were found to be sparingly soluble in cold water to form a gel like mass and convert in to colloidal solution in warm water but practically insoluble the organic solvent like ethanol , methanol, benzene , acetone and petroleum ether. Colors of the isolated Saj, Chironji and Indian Siris gums were found to be dark brown, light brown and beige in appearance respectively (Table 3). All these 3 gums were irregular in their texture with a characteristics odor and sweetish sour test. The presence of carbohydrate is confirmed in all the 3 gums by Fehling's test and Benedict's test (Table 4). The results obtained from protein test such as Ninhydrin and Biuret test confirmed the presence of protein in all these gums (Table 4).The results obtained in Dragendorff 's test, Mayer's test and Hager's test confirmed the absent of alkaloids in all the 3 gums (Table 4). As shown in Table 4, Tannin were found to be absent in all these gums confirmed by Ferric chloride test. The results obtained in Keller Killaine test and Borntrager's test confirmed the absent of cardiac glycosides in any of these gums (Table 4). As shown in Table 4, flovonoids were present in only Chironji gum, confirmed by various tests such as Alkaline reagent test and lead acetate solution test. The results of Liebermann - Burchard and Salkowaski test as given in Table 4 showed no shine of steroids or Tri-terpenoids in these gums.The pH of the 1% (W/V) solution of Saj, Chironji and Siris gums were found to be in the rang of 4.4 -5.9, 4.3-4.9 and 4.3-4.8 respectively (Table 5), that indicates towards the slightly acidic nature of all these isolated gums. Hence the pH may need to be adjust while preparing these gums for Oral or Bucal drug delivery system. Swelling index of Saj gum, Chironji gum and Siris gums were found to be 91.00± 1.69 %, 89.42± 1.93 % and 128.94± 2.02 % respectively after the 24 hours of study (Table 5), which states that, all the gums were excellent swelling properties. hence could be used as a drug release modifiers in pharmaceutical dosage form designing. The swelling index of Sirs (Albizia lebbeck) gum was found to be highest among three. Though it had a very good swelling index but the gell strength of the same was not upto the mark as other two gums such as Saj and Chironji gum. Viscoctiy of 4% W/V solutions with distilled water was found to be 42±0.98 cps , 38% ±.61cps and 77% ± 0.23cps respectively for Saj, Chironji and Siris gum. The total ash, acid insoluble and water soluble ash was found to be 3.23± 0.61 % , 1.21± 0.79% and 1.72± 0.31% for Saj gum, 4.53 ± 0.02%, 1.15 ± 0.63% and 1.61± 0.64 % for Chironji gum, 3.54± 0.69%,1.24± 0.81% and 1.76 ± 0.27% Siris gum respectively (Table 5). The results of ash values indicated towards the good quality and purity of all these gums. The loss on drying for Saj,Chironji and Siris gums (Table 5) were found to be 10.31 ±0.73, 8.41 ± 0.31%, 6.8± 0.50% which suggest towards the slight hygroscopic nature of these gums, hence should be stored in an air tight container. Reffered table 5 The bulk density and tapped density of 3 gums (Table 5) were calculated as respectively 0.476± 0.02 gm/cm^3 gm/cm^{3}and 0.588 ± 0.06 gm/cm^3 for Saj gum, 0.525± 0.03 gm/cm^3 and 0.623± 0.02 gm/cm^3 for Chironji gum and 0,501± 0.03 gm/cm^{3}and 0.575± 0.01 gm/cm^3 for Siris gum. The Hausner's ratio (Table 5) of all Saj,

Chironji and Siris gums were calculateld as 1.23± 0.06 %, 1.12± 0.03 %, 1.18± 0.05 % respectively which indicates for the good flow behavior of these gums. The compressibility Index were calculated as 19.04± 1.01 %, 15.73± 0.96 % and 12.86± 0.94 % respectively for Saj, Chironji and Siris gum (Table 5), which states that the Saj gums were more compressible than other two gums but the compressibility properties of Indian Siris gum was less than other tow gums. The angle of Repose were determined as 23.37±0.03 ^{0} for Saj gum, 16.04±0.15 ^{0} for Chironji gum and 13.37±0.21 for Siris gum (Table 5) which results indicates for the good to excellent flow properties by these isolated gum powders. The FTIR profile of Saj (Terminalia *elliptica*) gums (Table 8 and Figure 1) showed peaks at 3772.1, 3432.9, 2929.7, 2359.4, 1628.8, 1423.8, 1021.3 and 782.7 cm^{-1} which indicates towards the presence of functional groups such as alcohol , carboxilic acid, alkynes, alkanes, alkene, amines, amides, esters, ethers, aromatic rings, phenols etc in Saj gum. Chirongi (Buchanania *lanzan*) gum (Table 9 and Figure 2) showed the the peak at 3447.8, 2929.7, 2143.2, 1621.4, 1423.8, 1073.5, 771.6 and 704.5 cm^{-1} which indicates that functional groups such as amines, amide, phenol, alkanes, nitriles, alkynes, esters, ethers, carboxilic acids, alcohols, akenes and akenes in this gum. The characteristics peaks of Siris (Albizia *lebbeck*) gum (Table 10 and Figure 3) were found at 3429.2, 2929.7, 2363.1, 1625.1, 1423.8, 1073.5, 771.6 and cm^{-1-} which reflects towards the presence of Monomeric Alcohol, Carboxilic acid, Hydrogen bonded alcohol, Phenol, Amine , amide, Alkanes, Alkenes, Esters,Ethers, Aromatic rings, Alkenes in this gums . Acute toxicity study of all the 3 gums were performed based on guide lines number '425' of Organization for Economic Co- operative and Development (OECD) by orally feeding each gums to albilno mice. The results are summurised in Table 6 and Table 7. The effect of gums on the body weights of the animals, showed no significant observational adverse effect. All the test group animals were showed a normal increase in body as compared with control group of animals (Table 6). The observational health parameters & behavioral changes like corneal reflex ,pupils, salivation, lacrimation, skin fur ,pain, Grooming ,Torch response ,Hyper activity, sleep , Alertness ,Pinna reflex Tremors, Gripping strength etc of both test and control groups were recorded(Table 7). From the observational data it was found that there is no significant difference between Control and treated groups, which indicates towards the un harmfull nature of these gums. Not even a single mortality during the period of study has been observed (Table 7) which indicates towards the nontoxic and safe characteristics of these gums. There is no mortality observed at a higher dose of 2000 mg or 2 g/ kg body weight (Table 7) so it might be concluded that LD50 value of all the 3 gums were more than 2 g/kg body weight of animal.

FTIR Studies of gums

Table 8: FTIR profile of Terminlallia elliptica (Saj) gum

Serial No	Wave numbers (Cm^{-1})	Range	Functional groups
1	3772.1	3590-3650 3500-3650	Monomeric Alcohol , Monomeric Carboxilic acid
2	3432.9	3200-3600	Hydrogen bonded alcohols, Phenols

3	2929.7	2850-2970	alkanes
4	2359.4	2500-2700	Alkynes , Carboxilic acid
5	1628.8	1610-1680	Alkene
6	1423.8	1340-1470	alkanes
7	1237.5	1180-1360	Amines, Amides
8	1021.3	1050-1300	Esters,Ethers, Carboxilic acids, Alcohols
9	782.7	675-995	Aromatic rings,

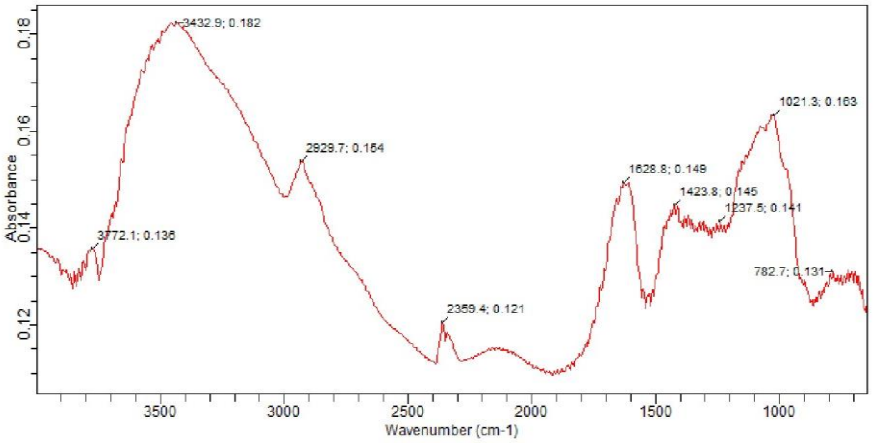


Figure 1. FTIR profile of Terminalia elliptica (Saj) gum

Table 9: FTIR profile of Buchanania *lanzan* (Chironji) gum

Serial No	Wave numbers (Cm^{-1})	Frequency cm-1	Functional groups
1	3447.8	3300- 3500 3200-3600	Amines, Amide Phenol
2	2929.7	2850-2970	alkanes,
3	2143.2	2010-2280 2100-2260	Nitriles Alkynes
4	1621.4	1610-1680	Alkenes
5	1423.8	1340-1470	Alkanes
6	1073.5	1050-1300	Esters,Ethers, Carboxilic acids, Alcohols
7	771.6	675-995	Akenes
8	704.5	675-995	Aromatic ring, Alkynes

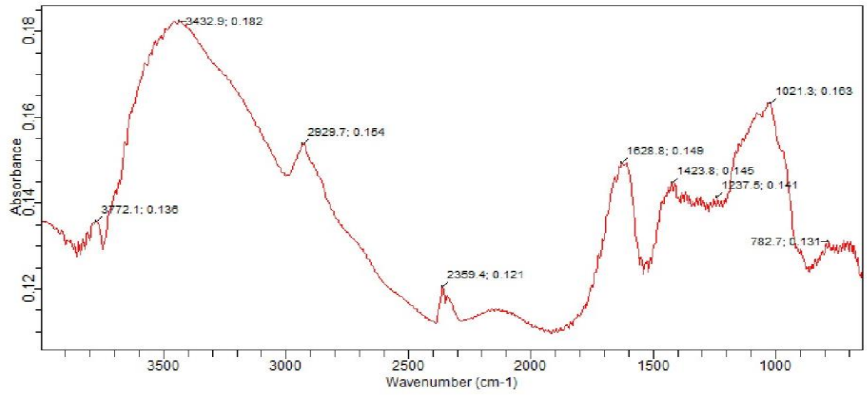


Figure 2.FTIR profile of Buchanania lanzan(Chironji) gum

Table 10 FTIR profile of Albizia lebbeck (Siris) gum

Serial No	Wave numbers (Cm^{-1})	Range	Functional groups
	3772	3590-3650	Monomeric Alcohol , Monomeric Carboxilic acid
1	3429.2	3200-3600 3300-3500	Hydrogen bonded alcohol, Phenol Amine , amide
2	2929.7	2850-2970	Alkanes,
3	2363.1	2500-2700	Hydrogen bonded Carboxilic acid
4	1625.1	1610-1689	Alkenes
5	1423.8	1340-1470	Alkanes
6	1073.5	1050-1300	Esters,Ethers, Carboxilic acids, Alcohols
7	771.6	675-995	Alkenes, Aromatic rings
8	670.9	675-995	Alkenes

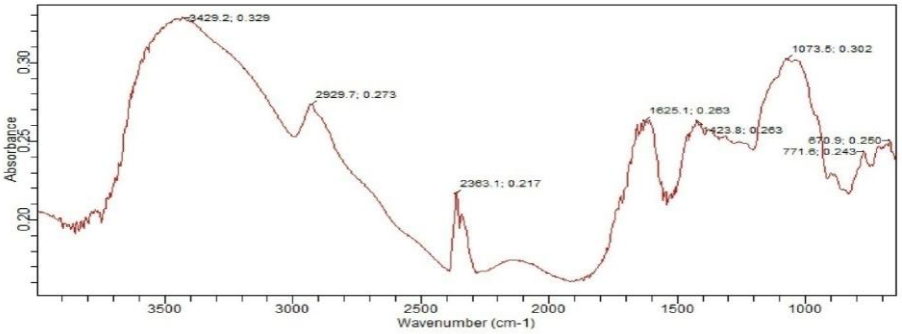


Fig 3.FTIR profile of Albizia lebbeck (Siris) gum

Conclusion

From the above investigation it was concluded that, all the isolated gums such as *Termianalia elliptica* (SaJ), *Buchanania lanzan* (Chironji) and *Albizia lebbeck* (Siris) contain carbohydrats and protein in it. The organoleptic properties looks to be acceptable for all these gums. The Swelling index and viscosity of gums were found to be Siris >Saj >Chironji. The pH of all these gums were slightly acidic. The Hausner's ratio, Compressibilty index and angle of repose states towards the good to excellent flow propetis of these gums. The total ash content and Loss on drying values were under specified limit. The IR spectral study indicates towards the presence of functional group such as alcohol , carboxilic acid, alkynes, alkanes, alkene, amines, amides, esters, ethers, phenols etc in Saj gum, amines, amide, phenol, alkanes, nitriles, alkynes, esters, ethers, carboxilic acids, alcohols and akenes in Chironji gum and monomeric alcohol, carboxilic acid, hydrogen bonded alcohol, phenol, amine , amide, alkanes, alkenes, esters,ethers , alkenes in sires gum. The Acute toxicity study of all the 3 gums revealed no mortality at a higher dose of 2000 mg or 2 g/ kg body weight so it might be concluded that LD50 value of all the 3 gums were more than 2 g/kg body weight of animal.. All these results revealed that the gums were safe for the oral administration and have enough potential to be used as excipients such as release modifiers, suspending agent, binding agents etc in formulation of various pharmcaceutical dosage form in future.

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