

Evaluation of Gelling Behavior of Natural Gums and their Formulation Prospects

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ABSTRACT

Purpose: Gums and polymers can be found abundantly in the nature from various plants, microbial and marine sources. Their physical and functional characteristics can be customized according to its application in product development. The purpose of this study is to determine the potentiality of natural gums obtained from plant based sources in development of gel formulation and its comparison with the commercially used polymers viz. Carbapol 934, Carboxy Methyl Cellulose, Xanthan, Sodium Alginate.

Methods: Gel formulations of varying ranges of polymeric concentrations were prepared and evaluated for its pharmaceutical parameters like pH, viscosity, homogeneity, tensile strength and antimicrobial activity. **Results:** The analysis of the developed gel formulations indicated that formulations developed using *Guar gum* possess much better gelling behavior i.e. viscosity and muco-adhesiveness than other selected gums and is more closer to its synthetic counterpart like Carbapol 934. Thus natural gum provides a comparatively broader range of characteristics and eco-friendly approach which enhances its applicability in formulation development. This research work rationalizes the importance of natural gums as gelling agents in development of pharmaceutical dosage forms, cosmetic products and nutraceutical formulations.

Key words: Eco-friendly, Gelling agents, Gel Formulations, Muco-adhesiveness, Antimicrobial activity.

INTRODUCTION

Greater success in developing the most advanced therapeutic systems today have been achieved due to the large scale availability of the natural polymers and additives, which provides an effective dosage of drug for prolonged periods. Natural polymers obtained from the plant gums and fibers are being utilized nowadays as gel forming agents to take a closer leap towards exploration of these gums and are being reported for their applications in formulation development. The development and selection of newer formulations for preparation of drug delivery system that controls the rate of drug release particular to the therapeutic needs with the help of hydrogels is a prime topic of investigation in the field of pharmaceutical sciences.¹ Hydrogel is a cross linked network, formed

from a macro-molecular hydrophilic polymer after absorbing huge volumes of water, variable range manifold of its own volume are being increasingly used in the drug delivery formulations.² The delivery systems utilizing hydrogels for controlled rate of drug release can be majorly classified into reservoir and matrix type. Drug release from a hydrogel based formulation is dependent on diffusion of water into matrix, dissolution and diffusion of the dissolved drug from the matrix. Usually, non-reactive polymer matrices are accepted to formulate this kind of delivery systems. Hydrogels are frequently used in clinical applications, including tissue engineering and regenerative medicine development³ diagnostics puposes,⁴ cellular and tissue immobilization,⁵ cells and bio-molecular

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separations,⁶ to optimize biological adhesions,⁷ as food source and cooking additive, cosmetic products, decoration. In development of medicinal formulations, they can act as depot formulation as drug steadily elutes and sustains a high concentration in the neighboring cells over a prolonged period.⁸ Natural hydrogels are generally biocompatible, as reflected in their successful use in the peritoneum and other sites *in vivo*.⁹ Biocompatibility is promoted due the large volume of water present in the gel matrix due to absorption and water carrying capacity of the polymeric gel. There is a physiochemical similarity of hydrogels to the native extracellular matrix in compositional (usually carbohydrate-based gels) and mechanical stability terms. Synthetic and semisynthetic polymers as Carbopol and Carboxymethyl cellulose used as thickening agents are commercially used in pharmaceuticals which have adverse effects on health due to its non-biodegradability not safe to be used directly. Carbopol is a high molecular weight, hydrophilic and cross-linked polyacrylic acid polymer. Due to high molecular weight and longer chain produce higher viscosity and higher viscous preparation are more difficult to administer, thus viscosity may retards the release of drug. The commercial gums such as *Guar*, *Tragacanth*, *Xanthan*, *Chitosan* and *Karaya* are widely used as emulsifying agent, adjuvant, gelling agents and hydrogels in food, pharmaceutical, agriculture and cosmetic for product development. Proteins, Enzymes, fibers, polysaccharides and gummy exudates obtained from nature are being used effectively in formulating the variety of pharmaceutical products. *Guar gum* is a high molecular weight polysaccharide obtained from *Cyamopsis tetragonoloba* (family Leguminosae) seed endosperm. The biopolymer has structure of β -1, 4 linked chain of mannose with α -1,6-linked galactose substituent at every second points. The ratio of galactose to mannose (M/G) in guar gum varies from 1.8:1.0 to 2.0:1.0. Guar gum forms good gel at lower concentration (0.5 and 1%) and achieves highest viscosity at low temperatures causing it to be widely exploited in the food industry as a gelling and emulsifying agent.

Gums can form a hydrogel due to the formation of numerous inter and intra molecular three dimensional network, which entraps the water molecule within it. This network can be tailored by treating the gel with various physical parameters such as change in pH, temperature of processing or by addition of any chemical agent such as EDTA into the gel. Synergistic interaction of Galactomannan of Xanthan gum and Carrageenan can improve the elasticity of gel formed from their mixture.¹⁰

The utilization of known polymers including natural gum provides a more rational and cheaper approach, as it showcases a strategy to utilize different polymers with distinct functional properties that can be modulated, avoiding the high expenses involved in the synthesis and development of a completely new material.

MATERIALS AND METHODS

Materials

Guar (Merck), *Tragacanth* and Butylated Hydroxyl Toluene (HPLC Pvt. Ltd.), Sodium Alginate (Loba Chemie Pvt. Ltd), *Xanthan* (SD fine chem. Ltd.), CMC (Merck), Carbopol 934 (Molychem) including other additives such as preservative viz. Methyl paraben, Propyl paraben, Propylene glycol were commercially purchased from Molychem Pvt. Ltd.

Methods of preparation of gel formulations

Gel solutions were prepared by adding required amount of gum polymers in luke warm distilled water (not more than 50°C; 50 % weight of the batch size) with stirring (2000rpm) and avoiding air entrapment. Gel was allowed to soak overnight for better absorption and removal of entrapped air bubbles.

Required amount of methyl paraben (0.2gm) and propyl paraben (0.1gm) were weighed separately and dissolved into 5ml of propylene glycerol, poured into the gel solutions with continuous stirring. 0.1gm of sodium metabisulphite was dissolved in 1 ml of distilled water and poured into the gel solution with continuous stirring. pH of the gels was maintained (pH-7.0) by adding triethanolamine to it. Finally remaining quantity of distilled water was added with stirring (2000 rpm) to maintain the volume of mixtures which were stirred again until homogenous gels were formed. The gels were then transferred into clear glass vials and centrifuged at 1000 – 3000 rpm for 10 min.

Total eighteen formulations were prepared by varying the concentrations (1-3%) of six polymers.¹¹ All the prepared gels were then subjected to various evaluation tests in order to identify their gelling characteristics. The composition of gel formulations is listed in Table 1.

Evaluation

Evaluation and optimization of formulations at different concentrations of natural gum was conducted out as follows:

Appearance

Gel formulations were tested for clarity, color, homogeneity, consistency, presence of particles and aggregates. Homogeneity and clarity were examined

using compound microscope (CETI). Small quantity of gel was pressed between the thumb and the index finger and its consistency and behavior was observed.¹² Formulations were also centrifuged at 2000 rpm by using a centrifuge (REMI, Model- R8C) to check the presence of any settled down particle after centrifugation, if present.

Determination of pH

The pH of the gel formulations was determined by using a calibrated digital pH meter (SYSTEM 361) at constant room temperature range ($24 \pm 2^\circ\text{C}$).¹³

Moisture content

Moisture content in the gel formulations was determined using oven dry method (AOAC, 1990). Weighed amount of formulation was kept in a petridish and kept in an oven at 105°C . The sample was continuously dried in oven until weight of the formulation becomes constant.¹⁴

Extrudability determination of formulations

Extrudability of the gel formulations was determined after filling it in a capped collapsible aluminum tubes. The weights of the tubes were recorded after filling it with gel and sealing at the ends. The tubes were placed

between two glass slides and were clamped on which a weight of 500 gm was placed over the slides and then the cap was removed. The gel extruded was collected and weighed to determine the percent of the extruded gel.¹⁵ The comparative extrudability of the gel formulations is shown in Table 2.

Spreadability

Spreadability was expressed in terms of time (seconds) taken by two slides (10cm x 10cm) to slip off from gel placed in between the slides under the direction of certain load (25gm), spreadability was calculated by using the formula.

$$S = (M.L/T)$$

Where, S = Spreadability, M = Weight tied to upper slide, L = Length of glass slides and T = Time taken to separate the slides.^{16,17}

Viscosity

Viscosity of formulated gels was determined using Brookfield viscometer (Brookfield Viscometer V6.5 RV) at $25^\circ\text{C} \pm 2^\circ\text{C}$ at the rpm 0.3.¹⁸

Tensile strength

Tensile strength of the gel formulations was determined by Tensiometer (Krusk K20S Easy Dyne) using DuNouy

Table 1: Composition of different prepared gel formulations.

F. N.	Guar(gm)	Tragacanth (gm)	Xanthan (gm)	S. A. (gm)	CMC (gm)	Carbapol 934 (gm)	Sodium Meta Bisuphlite (gm)	Methyl paraben (gm)	Propyl Paraben (gm)	PG (ml)	Water (100 ml)
F1	1.0	-	-	-	-	-	0.1	0.2	0.1	5	q.s
F2	2.0	-	-	-	-	-	0.1	0.2	0.1	5	q.s
F3	3.0	-	-	-	-	-	0.1	0.2	0.1	5	q.s
F4	-	1.0	-	-	-	-	0.1	0.2	0.1	5	q.s
F5	-	2.0	-	-	-	-	0.1	0.2	0.1	5	q.s
F6	-	3.0	-	-	-	-	0.1	0.2	0.1	5	q.s
F7	-	-	1.0	-	-	-	0.1	0.2	0.1	5	q.s
F8	-	-	2.0	-	-	-	0.1	0.2	0.1	5	q.s
F9	-	-	3.0	-	-	-	0.1	0.2	0.1	5	q.s
F10	-	-	-	1.0	-	-	0.1	0.2	0.1	5	q.s
F11	-	-	-	2.0	-	-	0.1	0.2	0.1	5	q.s
F12	-	-	-	3.0	-	-	0.1	0.2	0.1	5	q.s
F13	-	-	-	-	1.0	-	0.1	0.2	0.1	5	q.s
F14	-	-	-	-	2.0	-	0.1	0.2	0.1	5	q.s
F15	-	-	-	-	3.0	-	0.1	0.2	0.1	5	q.s
F16	-	-	-	-	-	1.0	0.1	0.2	0.1	5	q.s
F17	-	-	-	-	-	2.0	0.1	0.2	0.1	5	q.s
F18	-	-	-	-	-	3.0	0.1	0.2	0.1	5	q.s

(S. A.-Sodium alginate, CMC-Carboxy methyl cellulose, PG-Propylene glycol)

ring method to evaluate the muco-adhesive properties of prepared formulations.

Electrical conductivity

The electrical conductivity (κ) was determined at 25°C using a bench top conductometer (Lovibond, Model No.- Con110).¹⁹

Evaluation of microbial growth

The formulations prepared were evaluated to check its microbial resistance by taking 100 mg of gel and dissolving it in 1ml of distilled water and later spreading it over an agar plate. The plates were incubated at 37°C±1°C for 24 days to check the presence of microbial growth.

Stability studies

Stability studies were carried out according to ICH guideline at different temperature conditions (4°±2°C, 75±5% RH, 25°C±2°C, 75±5% RH and 37°C±2°C, 75±5% RH) for 6 months in a borosilicate vessel. All of the formulation parameters were performed periodically and formulations were evaluated for their stability and gelling behavior.²⁰ Effect of centrifugation on stability in terms of uniformity and solubility of formulation was also studied by Centrifuge device (Eltek TC4100F)

at 2000rpm for 60 minute (recorded after consequent 5, 15, 30 and 60 min respectively).

Statistical Analysis

Pearson co-relation and analysis of variance were applied in triplicate on the data obtained from evaluation tests using SPSS16.0 software. Multiple comparison study was performed at different concentrations using Post hoc Tukeys *t*-tests at $p<0.05$ to compare two responses of natural gum formulations.

RESULTS AND DISCUSSION

Appearance

Formulations were prepared by changing the concentration of natural gums and polymers which were later evaluated in terms of the gelling characteristics. Formulations showed variations in color and textures (Table 2). No aggregate and insoluble particles were found to be settled down after applying strong centrifugal force (2000 rpm).

pH studies of gel formulations

pH values lie in the normal pH range which is acceptable for skin and varied from 6.5 to 7.5, shown in Table 2.

Moisture content or Weight Loss on drying

Moisture content of different prepared gel formulations varied from 92.82-98.72% respectively. Maximum moisture content was found in F1 (1%) which was comparable to F13 (1%). F3 (3%) have minimum moisture and comparable to F18 (3%). Highly viscous gels such as those made up of *Guar gum* and Carbapol 934, were found to entrap large quantity of water leading to high moisture retaining gels (Figure 1). From the Figure it was observed that moisture content of F2, F4 was comparable to F8 and F5 was comparable to F11. The percentage moisture content of different formulations decreased with the increase in the concentration of the gum polymers added in the gels because formulations were prepared according to w/v and due to the increase in the concentration of gum polymer the volume of water added was lesser so to make up the volume to 100 ml as a result the water content in the formulation was reduced which was further quantified by loss on drying method. It may be due to the increase in absorption and complexation property of gels which decreases the rate of removal of water from it. It is better to use viscous and more strongly linked gel as moisture retainers in topical formulations as it has capacity to hold more solvent into it for a long time.

Statistical analysis indicates that $P<0.05$, so it was concluded that observed data is significant and shown

Table 2: Evaluation of colour, extrudability and pH of different gel formulations.

Batch no.	Color	Extrudability	pH
F1	Transparent	++	7.33
F2	Creamish white	++	6.57
F3	Creamish white	+	7.33
F4	Light ivory	+++	7.06
F5	Ivory	++	7.50
F6	Ivory	++	7.48
F7	White	++	7.44
F8	Off White	++	7.44
F9	Off white	+	7.50
F10	Creamish white	+++	7.05
F11	Creamish yellow	++	6.35
F12	Creamish yellow	++	7.0
F13	Transparent	+++	7.30
F14	Transparent	++	7.44
F15	Transparent	++	7.35
F16	Transparent	++	7.45
F17	Transparent	+	7.25
F18	Transparent	+	7.52

(*+++ indicates > 90%, '++' indicates >80%, '+' indicates < 70% extrudability)

a relationship between polymers and concentration variable with the LOD value.

Extrudability determination of formulations

It is a test that measures the force required to extrude the material from tube filled with formulation. Viscous formulations prepared from Carbapol and *Guar* gum were found to have lower extrudability than other formulations (Table 3). Formulations F3 (3%), F9 (3%), F17 (2%), F18 (3%) showed ambient extrudability in moisture range of 92-95.78% respectively. Better extrudability is observed in formulations (F2, F5, F6, F7, F8, F11, F12, F14-16) with moisture ranging from 94.68 (F6) to 97.84% (F7). Best extrudability is correlated with moisture range of 97.28 (F3) to 98.30% (F13). It was also noticed that extrudability of the formulation decreased with the rise in concentration of the gum and as the concentration increases, a complex cross-linked structure is formed that require a large amount of pressure on it to extrude it out of the collapsible tube.

Spreadability

Spreadability of different gel formulations varied from 2.01 to 40.00 g.cm /sec. Sodium-alginate had maximum spreadability and least of Carbapol. Spreadability of F1, F2, F3 and F16, F17, F18 were having least spreadability than others gel formulations (Figure 2). Result, based on all evaluated parameters, had coefficient of determination is found to be in range of 0.901-0.999. From graphical representation (Figure 2) of the spreadability of various natural gums, it is evident that gel formulation i.e. *Guar* and *Tragacanth* bear coefficient of determination, range from 0.980-0.999 (Figure 1), which is comparable to bacterial, synthetic and *Tragacanth* polymer, is at par with marine based plant and semi-synthetic gums. Spreadability denotes the extent to which the gel on application get easily spreads to skin or a membrane. A good gel takes less time to spreads and will have high spreadability. Formulations prepared by carbapol 934 (F16, F17, F18) and *guar gum* (F1, F2, F3) were not having higher spreadability as it decreases with increase in viscosity due to high cohesive density causing greater resistance to flow.

Statistical analysis of the spreadability indicates that P value < 0.05 , which is significant and there exist a relationship between polymer and concentration variable with the spreadability value of the polymeric formulations.

Viscosity studies

Viscosity of different gel formulations varied from 21.33-914853 cps at 0.3 rpm spindle no.1. F18 (3%) had maximum and F13 (1%) had least. Viscosity of F1

(1%) was found to be nearly comparable to F15 (3%). Viscosity of F5 (2%) was found to be equivalent to sodium-alginate (F11) and CMC (F14). Viscosity of F7 (1%) was comparable to F10 (1%) (Figure 3). Viscosity of gel formulations increased with increase in gum concentration. Increase in viscosity with concentration of gums/polymers is probably due to molecular association in fluids, increase in number of high molecular weight polymeric chains of the gums per unit volume and increased interaction between these chains of gums in hydrogels.

Statistical analysis of the Viscosity data indicates that P value < 0.05 , so it was concluded that observed data is significant and there exist a relationship between natural gum and concentration variable with the viscosity value of the polymeric formulations.

Tensile strength of formulations

Muco-adhesive property of the gel can be determined using surface tension. Higher surface tension liquids are more resistant to wetting and produce a smaller (and taller) bead of liquid than do low surface tension liquids. Similarly, lower surface tension liquids will tend to absorb into membranes more readily.

Tensile strength of formulations prepared by *Guar* and Carbapol were found to be higher than others. F3 was found to be having maximum tensile strength while F10 have least. Tensile strength of F7 and F8 was comparable to F4. Natural *Guar gum* had six fold tensile strength in comparison to commercially available marine source sodium-alginate. (Figure 4) Natural *Tragacanth* had two fold higher tensile strength as compared to commercially available sodium-alginate.

It was also observed that, as concentrations of natural gums increases, the value of surface tension decreases and vice-versa. So, for preparing an easily spreadable gel formulation with lower surface tension may be preferred.

Electrical conductivity measurements

The conductivity of different gel formulations varied from 1.75-12.06 mS. It was observed that as the concentration of the gum increased in the gel, the value of conductivity (χ) increased, which may be due to increase in free ion content. Formulations prepared by sodium alginate (Figure 4) were having higher conductivity than other formulations. When compared it was examined that concentration of gum is having a direct influence on conductivity and increases proportionally with gum concentration (Figure 5). Conductivity (or specific conductance) of an electrolyte solution measures the ability of the prepared formulation to conduct electricity and to determine the ionic content in it.²⁰ Wide variation

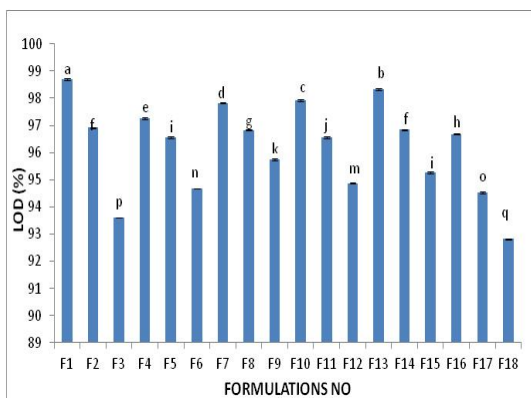


Figure 1: % Loss on drying of different formulations.

Abbreviation: F1- F3 (1%- 3%)-Guar F4-F6 (1-3%)-Tragacanth F7-F9 (1-3%)-Xanthan F10-F12 (1- 3%)-Sodium alginate, F13-F15 (1-3%)-CMC, Carbapol-F16-18 (1-3%)

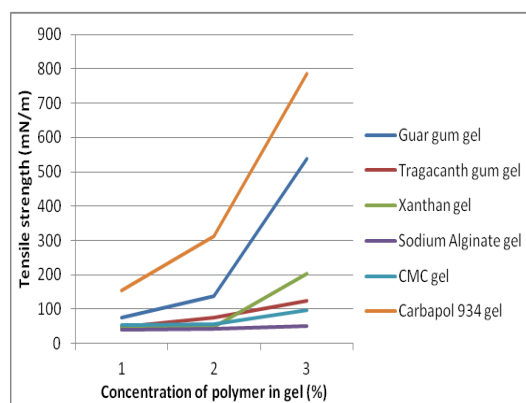


Figure 4: Tensile strength of gel formulations prepared from different gums at different concentrations.

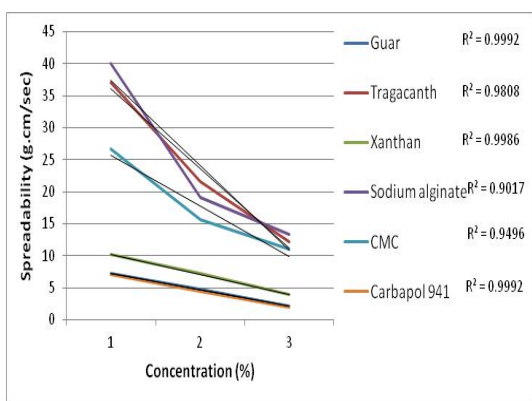


Figure 2: Spreadability of gel formulations prepared from different gums at different concentrations.

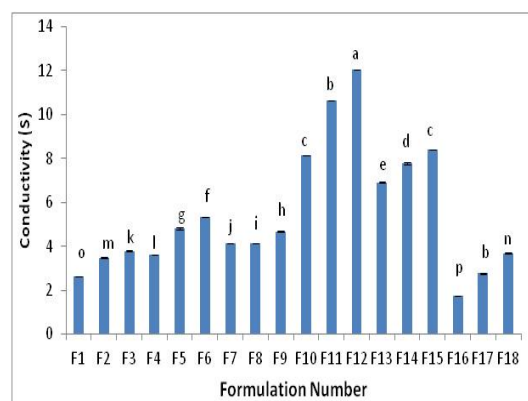


Figure 5: Electrical Conductivity data of different formulations.

Abbreviation: F1- F3 (1%- 3%)-Guar F4-F6 (1-3%)-Tragacanth F7-F9 (1-3%)-Xanthan F10-F12 (1-3%)-Sodium alginate, F13-F15 (1-3%)-CMC, Carbapol-F16-18 (1-3%)

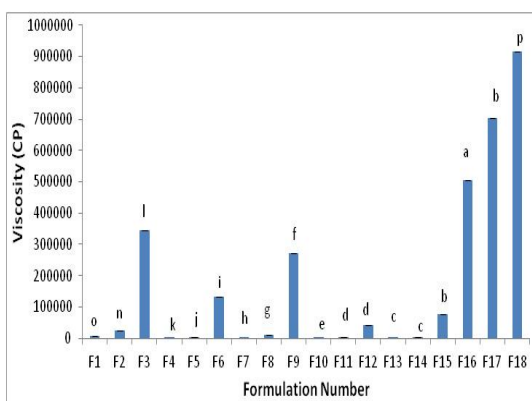


Figure 3: Viscosity of different formulations at 0.3 rpm.

Abbreviation: F1- F3 (1%- 3%)-Guar F4-F6 (1-3%)-Tragacanth F7-F9 (1-3%)-Xanthan F10-F12 (1-3%)-Sodium alginate, F13-F15 (1-3%)-CMC, Carbapol-F16-18 (1-3%)

was observed in conductivity data from graphical representation (Figure 5). Maximum conductivity was observed in F12 and minimum in F16. Conductivity of F1 was comparable to F17, F3 to F7 and F8.

Statistical analysis of the Conductivity data indicates that P value < 0.05 , so it was concluded that observed data is significant and there exist a relationship between

gum polymer and concentration variable with the conductivity value of the polymeric formulations.

Multiple comparison studies of natural gums with other polymers at different concentration using *Post hoc* tests shows that the data is significant.

Evaluation of microbial growth resistance property

No microbial growth was found to be present on the culture media after seven days of incubation. It may be due to the presence of preservative agent interaction with gelling agent making it unsuitable for microbial growth.

Stability studies

Slight variation in formulation was observed after 6 months of stability testing, the color of the gel was slightly darkened, while the transparency of some gels prepared from natural gums was also reduced. Settling of aggregates was observed in formulations prepared from sodium alginate and CMC (F10, F11 and F13)

Table 3: Pearson correlation among different parameters.				
S.N.	Formulation No	Viscosity and Spreadability	Viscosity and Tensile strength	Spreadability and Tensile strength
1.	F1 F2 F3	-0.959	0.997*	-0.978
2.	F4 F5 F6	-0.876	0.937	-0.989
3.	F7 F8 F9	-0.825	0.999*	-0.806
4.	F10 F11 F12	-0.770	0.967	-0.908
5.	F13 F14 F15	-0.720	0.999*	-0.749
6.	F16 F17 F18	-0.999*	0.966	-0.951

*Correlation is significant at the 0.05 level (2-tailed)

which may be due to the hydrolysis of natural gums. No sign of microbial contamination was observed after 6 months of stability testing.

Pearson correlation of formulations among different parameters

Formulations of different concentrations of natural polymers were co-related to each other to determine the effect of variation in polymer concentration on pharmaceutical properties i.e viscosity, tensile strength and spreadability of the formulation and identify the *in-vitro* correlation between them. The Pearson correlation value (r) between the parameters viscosity and spreadability of guar gels ($r = -0.959$) was found to be comparable with the r value of carbapol 934 gels (-0.999^*). It was found that there is a very high co-relation between the viscosity of a gel and its tensile strength as the evident from the values ranging from (-0.937 - 0.999^*) of all the formulations prepared. The r value between tensile strength and spreadability was very high of formulations prepared using guar gum, tragacanth gum and carbapol 934 ranging from -0.951 to -0.989 . Using Pearson correlation method, it was found that there is a negative correlation between viscosity and spreadability and as the value of viscosity increased with concentration, the value of spreadability reduced. There was a positive co-relation between viscosity and tensile strength which proves that the values of both parameters will increase with increase in concentration.

CONCLUSION

Gels prepared from the natural gums possess good spreadability, extrudability and bio-adhesive strength. Natural gums have good gelling properties and are comparable to the commonly used synthetic and semi-synthetic polymer i. e carbapol 934 based gel. A proportionality relationship based on *in-vitro* co-relation test is established that showcases the influence of one pharmaceutical parameter on the other. On increasing the concentration, viscosity of gel increases and extrudability decreases on applying same stress. Also it was explored that highly concentrated liquid due to more viscosity was difficult to spread then the low concentration less viscous gel, also by analyzing the data we can say that viscosity is inversely proportional to the spreadability and extrudability of the gels but directly proportional to the surface tension, i.e., $\text{Viscosity} \propto 1/E \propto 1/S \propto S.T.$ Guar gum is found to bear good viscosity and spreadability, hence it can be as a cost effective, eco-friendly alternative of carbapol, xanthan and other commonly known gelling agents. The work justifies the utilisation and usage of environmental friendly natural gums as potential gelling agents in preparing gel based formulations and natural products for pharmaceutical, cosmetic and nutraceutical applications.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

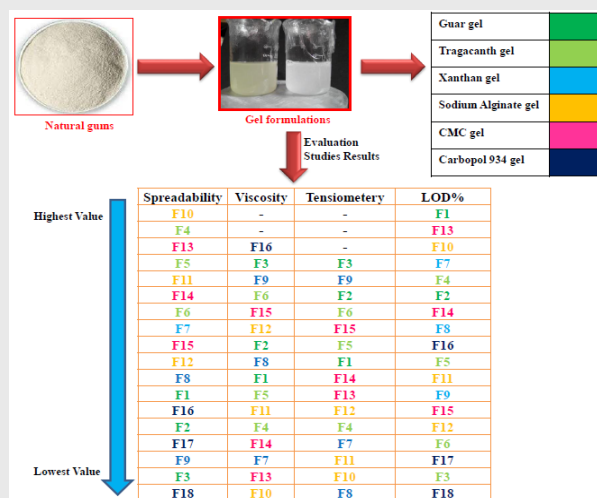
ABBREVIATIONS

rpm: Rotation per minute; **g.cm/s:** Gram centimeter per second; **°C:** Degree centigrade; **mS:** Simense; **CMC:** Carboxy methyl cellulose; **cps:** Centipoise; **E:** Extrudability; **S:** Spreadability; **S.T:** Surface tension

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PICTORIAL ABSTRACT



SUMMARY

Gel formulations developed at different concentrations using plant gums (Guar & Tragacanth) and compared with commercially used natural, semisynthetic and synthetic polymers. Formulations were evaluated for pharmaceutical parameters and antimicrobial activity was tested. A proportionality relationship on correlation was established among different parameters as viscosity, tensile strength and spreadability. Further studies revealed guar gum was found to have better gelling characteristics due to viscosity and mucoadhesiveness, which could thus be selected as most suitable to be used as effective eco-friendly alternative of Carbopol, Xanthan and other commonly known gelling agents.

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