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ORA- Analytical Study

Quality Control and GC-MS Study of an Ayurvedic Linctus Formulation - *Kalyanaka Guda*

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ABSTRACT:

Background: *Kalyanaka Guda* (KG) is a polyherbal linctus preparation of Ayurveda widely used in the management of anemia, hemorrhoids, skin diseases etc., Such commonly practiced formulations lack the preliminary quality control (QC) data. Hence this study aimed to test KG's QC parameters and subjected it to analysis with Gas Chromatography Mass Spectrometry (GC-MS). **Methodology:** Following the classical method of preparation of KG, analysis such as organoleptic characters, loss on drying, total ash, acid insoluble ash, total fat content qualitative inorganic elemental analysis were performed. Thin Layer Chromatography (TLC) for KG was developed experimentally using reflux extraction by n-hexane, chloroform and methanol with Toluene, Ethyl acetate, Formic acid and Methanol (3:3:0.8:0.2) as mobile phase. All the three extracts were then subjected to analysis in GC-MS. **Results:** Results revealed characteristic colour, odour and appearance of KG. Whereas pH, loss on drying, total ash, acid insoluble ash and total fat content were found as 3.74, 9.34%, 3.09%, 0.46% and 4.2% respectively. Elements such as sodium, sulphate and chloride were found in KG. TLC of n-hexane and chloroform had one band in short wave and three bands in long wave each. The second trial of methanol extract displayed good separation with three and six bands in short wave and long wave respectively with the corresponding band in standard gallic acid. GC-MS of methanolic extract identified 13 compounds. **Conclusion:** The study successfully rendered QC parameter findings and also developed methodology for TLC for important and commonly used formulation like *Kalyana Guda*. The findings of GC-MS helped identify compounds that had formed as a secondary metabolite during the pharmaceutical processing of *Kalyanaka Guda*.

KEYWORDS: *Avaleha*, confections, Ayurvedic medicine, chromatography, *Guda kalpana,* QC.

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1. INTRODUCTION

Ayurveda describes wide varieties of dosage forms catering specific needs. Among them, *Avaleha kalpana*, considered a secondary dosage form to *kwatha kalpana* (decoctions) in Ayurveda, is widely used due to its advantages, such as sweetness, convenient packaging, and therapeutic efficacy. [1] *Avaleha* are those prepared by usually adding sweetening agents like sugar, sugarcane juice or even jaggery. The processing is made in such a way that the end-product results in a thick, semi-solid state, thus making the preparation an easy one to consume by licking. This gave the preparation the name "*Avaleha*" whichs means "to lick". It is essential to differentiate *Avaleha* from *Guda kalpana* (jaggery confections). While both use jaggery as a sweetening agent, Avaleha allows the use of any of the mentioned sweetening agents, whereas *Guda kalpana* solely relies on jaggery. Classical texts describe two primary methods for preparing *guda kalpana*: the first involves using fire for confectioning jaggery (*Sagni*), and the second method involves pounding jaggery with other finely powdered herbal ingredients in a stone mortar and pestle (*Niragni*). [2]

Kalyanaka Guda (KG) is a widely practiced *Guda kalpana* utilized in managing various health conditions, including hypothyroidism, female infertility, leucoderma, and lifestyle disorders. [3-6] Its mention can be traced back to the *Ashtanga Hridaya*, that includes the formulation for purgation in *Kalpasthana*. [7] The ingredients used in KG are as follows: *Vidanga (Embelia ribes* Burm. F.*), Pippali moola (*Root of *Piper longum* L.*), Triphala (Terminalia chebula* Retz.*, Terminalia bellirica* (Gaertn.) Roxb.*, Emblica officinalis* Gaertn.*), Dhanyaka (Coriandrum sativum* L.*), Chitraka (Plumbago zeylanica* L.*), Maricha (Piper nigrum* L.*), Indrayava* (seeds of *Holarrhena antidysenterica* Wall.*), Ajaji (Carum carvi* L.*), Pippali* (Fruit of *Piper longum* L.*), Gaja pippali (Syndapsus officinalis* Schoott.*), Yavani (Trachyspermum ammi* (L.) Sprague.*), Souvarchala lavana* (*Unaqua sodium chloride*), *Saindhava lavana* (*Sodi chloridium*), *Vida lavana* (*Ammonium chloride*), *Audbidha lavana* (Sulphate of soda with chloride of sodium), *Samudra lavana* (*Sodi muris*), *Tila taila (Sesamum indicum* L.*), Trivrut (Operculina turpethum* (L.) Silva Manso*), Amalaki rasa (*Juice of *Emblica officinalis* Gaertn.) and Jaggery. Subsequent texts such as *Vangasena Samhita*, *Gadanigraha*, and *Brihat Yoga Tarangini* also discuss KG but with some alterations in few ingredients. [8] Despite the significance and widespread use of KG, there is a lack of research work on preliminary quality control analysis or analytical studies to analyze the chemical composition of the formulation to date.

Gas chromatography-mass spectrometry (GC-MS) is a versatile analytical technique extensively applied to detect herbal compounds in complex fractions. There are two precision techniques with this analysis - the gas chromatography helps section the compounds from the given sample mixture whereas the mass spectrometry identifies and analyzes the detected compounds by chromatography individually. GC-MS is ideal when compounds tend to vaporize without breaking down more under moderate heat and still be chemically stable and unaltered while inside the system. GC-MS is particularly valuable in separating and identifying

analytes that exist solely in the gas phase at temperatures below 100°C, a feat not achievable with other techniques. Moreover, it can expand the number of possible analytes by forming stable, volatile derivatives of compounds unsuitable for GC-MS in their natural state. [9] Taking these factors into account, the current study aims to assess the quality control of *Kalyanaka Guda* and analyze through GC-MS, its bioactive herbal components responsible for its therapeutic effects on various diseases.

2. MATERIALS AND METHODS:

The ingredients were procured from GMP certified pharmacy and KG was prepared in the Department of Rasashastra and Bhaishajya Kalpana, KAHER's Shri B. M. K. Ayurveda Mahavidyalaya, Belagavi adopting the classical method. [7] Table 01 shows the ingredients and their proportion used in the preparation of KG. All of the physico-chemical analysis of KG was done at AYUSH approved Drug Testing Laboratory of KAHER's Shri BMK Ayurveda Mahavidyalaya, Belagavi, Karnataka.

Table 01: Ingredients along with Ratio of Kg

Sl. No	Drug name	Part used	Proportion used
1	*Vidanga*	Dried fruit	1 *karsha* (12 g)
2	*Pippali moola*	Root bark	1 *karsha* (12 g)
3	*Triphala*	Dried fruits	1 *karsha* (12 g)
4	*Dhanyaka*	Seed	1 *karsha* (12 g)
5	*Chitraka*	Root bark	1 *karsha* (12 g)
6	*Maricha*	Dried fruit	1 *karsha* (12 g)
7	*Indrayava*	Seed	1 *karsha* (12 g)
8	*Ajaji*	Dried fruit	1 *karsha* (12 g)
9	*Pippali*	Dried fruit	1 *karsha* (12 g)
10	*Gaja pippali*	Dried fruit	1 *karsha* (12 g)
11	*Yavani*	Seed	1 *karsha* (12 g)
12	*Saindhava lavana*	Salt	1 *karsha* (12 g)
13	*Souvarchala lavana*	Salt	1 *karsha* (12 g)
14	*Samudra lavana*	Salt	1 *karsha* (12 g)
15	*Vida lavana*	Salt	1 *karsha* (12 g)
16	*Audbidha lavana*	Salt	1 *karsha* (12 g)
17	*Tila taila*	Oil	8 *pala* (384 g)
18	*Trivrut*	Root	8 *pala* (384 g)
19	*Amalaki swarasa*	Fresh juice of fruit	3 *prastha* (2.304 l)
20	*Guda*	Jaggery	½ *tula* (2.4 kg)

The prepared KG was analyzed for the Organoleptic parameters (Colour, Odour, Taste and Consistency) and Physico-Chemical analysis - pH, Loss on drying, Total ash, Acid insoluble ash and Total fat content. Qualitative confirmation for the presence of Sodium, Calcium, Iron, Sulphate, Carbonate, Chloride and Nitrate were carried out with dilute Hydrochloric acid (HCl) which was used in Acid insoluble ash procedure and analyzed using Ayurvedic Pharmacopoeia of India methods. [10]

Chromatographic analysis: All the solvents - n-Hexane, Chloroform and Methanol and mobile phases for the TLC were of analytical grade.

Since, there was no existing methodology to perform chromatography for KG; the mobile phase was prepared by permutation and combination of different solvents.

Sample preparation: In absence of specific method of TLC for KG, the standardized procedures mentioned in Ayurvedic Pharmacopoeia of India for *Kalyanaka avaleha* and *Chyvanaprasha avaleha* were referred. 5 g of KG was taken with 75 ml of n-Hexane under reflux on a water bath for 30 minutes.[11] Consequent extraction

was done similarly using Chloroform and Methanol with the same marc using reflux condenser. The marc was dried after each extraction. The three extracts were filtered and concentrated to 10 ml and stored in labeled containers for TLC and GCMS analysis.

Thin Layer Chromatography: The TLC was carried out using silica gel 60 F254 aluminum backed TLC plates (Supelco, Sigma-Aldrich Canada Co. Ltd. Canada).**Procedure:** Using capillary, 10 µl of all the three extracts were spotted separately on three TLC Plates and placed in the chamber containing specific mobile phase (Table 02).[11] It was made to run till the 8 cm distance and then removed from the chamber to dry. The plates were carefully observed in short (254 nm) and long (366 nm) waves in the UV chamber. Gallic acid standard was prepared by dissolving 17 mg of standard Gallic acid in 1 ml of Methanol and was spotted in the TLC plate and was made to run along with Trial 2. [12]

Gas chromatography-Mass spectrometry:

Procedure: The same extracts prepared for TLC using n-Hexane, Chloroform and Methanol were sent to VIT-SIF Lab, SAS, Vellore, Tamilnadu for the GCMS analysis.

Instrument: Perkin Elmer Clarus 680 GC with fused silica column, packed with Elite- 5MS (Dimensions: 30 m ×0.25 mm ID × 250µm df).

Carrier gas: Helium.

Procedure: The injector temperature was set at 260 °C during the chromatographic run. The 1µL extract sample was injected with a split ratio of 10:1 into the instrument when the oven temperature was as follows 60 °C (2 min); followed by 300 °C, elevated at the rate of 10 °C per min; and 300 °C, where it was held for 6 min. The clarus 600 (EI) mass detector conditions were transfer line temperature 240 °C; ion source temperature 240 °C; and ionization mode electron impact at 70 eV. The fractions of 40 to 600 Da were detected. The spectrums of the components were compared with the database of spectrum of given components stored in the GC- MS NIST (National Institute of Standards and Technology – Mass spectral Database 2008) library.

Table 02: Mobile Phase used in each Trial of Kg Extracts

Extracts of KG	Trial 1	Trial 2
n-Hexane	Toluene : Ethyl acetate (8.5 : 1.5)	-
Chloroform extract	Toluene : Ethyl acetate: Methanol (9:1:1)	-
Methanol extract	Toluene : Ethyl acetate (8:2)	Toluene : Ethyl acetate: Formic acid : Methanol (3:3:0.8:0.2)

3. RESULTS:

Results of preliminary organoleptic characters and physico-chemical parameters of the prepared KG are furnished in Table 03 and 04 respectively. Inorganic elemental analysis detected the presence of sodium, sulphate and chloride salts whereas the other elements remained undetected in present study (Table 05). With the trials performed for TLC, only one compound was spotted at Rf 0.92 in 254 nm in KG's n-hexane extract (Figure 01). Whereas, 366 nm showed three spots corresponding to Rf 0.52, 0.62 and 0.66 (Figure 02). The chloroform extract of KG separated four bands totally (one in 254 nm at Rf 0.88 and three in 366 nm at Rf 0.4, 0.5 and 0.78) (Figures 03 and 04). Out of the two trials

performed for methanolic extract of KG, the first trial did not show any compound detected at 254 nm. But when observed at 366 nm, the sample showed two spots corresponding to Rf 0.65 and 0.75 (Figure 05). Trial 02 with methanolic extract fetched a total of nine band separations - three in 254 nm (Rf - 0.13, 0.46 and 0.6) and six in 366 nm (Rf - 0.13, 0.45, 0.6, 0.71, 0.82 and 0.93) along with the standard gallic acid whose Rf at 0.61 was spotted in both 254 nm and 366 nm (Table 06 (Figures 06 and 07). The GC-MS of methanolic extract yielded 13 peaks (Figure 08). Of them, compounds with RT – 10.17, 10.37, 10.55 and 10.73 showed significant spikes in the GC-MS chromatogram. Table 07 is provided with complete results of GC-MS done for KG's methanolic extract.

Table 03: Organoleptic Characters of KG

Parameter	Obtained result	PSAF result
Colour	Brown	Brownish black
Odour	Sweet aromatic smell	Fragrant
Taste	Sweet and Savoury	Sweetish and astringent
Consistency	Semi-solid	Sticky

Table 04: Physico-Chemical Parameters of KG

Parameter	Obtained result	PSAF result
pH	3.74 (5% solution)	4.1-5.3
Loss on drying	9.34 %	5.02-15% w/w
Total ash	3.09 %	Not found
Acid insoluble ash	0.46 %	Not found
Total fat content	4.20 %	3.755-11.32% w/w

Table 05: Inorganic Qualitative Analysis of KG

Element	Result
Sodium	Detected
Calcium	Not detected
Magnesium	Not detected
Potassium	Not detected
Iron	Not detected
Sulphate	Detected
Carbonate	Not detected
Phosphate	Not detected
Chloride	Detected
Nitrate	Not detected

Table 06: RF values of KG extracts in TLC

n-Hexane		Chloroform		Methanol				Gallic acid standard	
				Trial 1		Trial 2			
Short wave	Long wave	Short wave	Long wave	Short wave	Long wave	Short wave	Long wave	Short wave	Long wave
0.92	0.52	0.88	0.4	NIL	0.65	0.13	0.13	0.61	0.61
	0.62		0.5		0.75	0.46	0.45		
	0.66		0.78			0.6	0.6		
							0.71		
							0.82		
							0.93		

Table 07: GC-MS analysis of Phytocompounds of KG Methanolic Extract

Sl. No.	RT	Compound	Peak Area	Area%	Metabolite class
1	8.952	4H-pyran-4-one,2,3-dihydro-3,5-dihydroxy-6-methyl-	193,890,016.0	5.176	Pyranones
2	9.042	4H-pyran-4-one,2,3-dihydro-3,5-dihydroxy-6-methyl-	71,736,544.0	1.915	Pyranones
3	10.172	2-furancarboxaldehyde, 5-(hydroxymethyl)-	439,902,272.0	11.744	Furan derivatives
4	10.232	2-furancarboxaldehyde, 5-(hydroxymethyl)-	700,856,064.0	18.711	Furan derivatives
5	10.377	2-furancarboxaldehyde, 5-(hydroxymethyl)-	788,825,408.0	21.059	Furan derivatives
6	10.552	2-furancarboxaldehyde, 5-(hydroxymethyl)-	818,053,056.0	21.840	Furan derivatives
7	10.827	2-furancarboxaldehyde, 5-(hydroxymethyl)-	158,685,840.0	4.236	Furan derivatives
8	10.932	2(5H)-furanone, 5,5-dimethyl-	104,434,872.0	2.788	Furanones
9	11.007	4-Hepten-3-one, 4-methyl-	63,162,120.0	1.686	Ketones
10	11.127	2,7-nonadien-5-one, 4,6-dimethyl	52,701,300.0	1.407	Ketones
11	14.094	Xylose	178,345,344.0	4.761	Monosaccharides (Pentoses)
12	14.149	D-Mannoheptulose	119,302,848.0	3.185	Monosaccharides (Pentoses)
13	16.385	Endo-2,3-O-Ethylidene-β-D-erythrofuranose	55,848,048.0	1.491	Furanose sugars

Figure 01: Short wave of n-hexane extract of KG

Figure 02: Long wave of n-hexane extract of KG

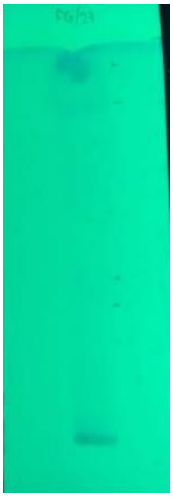

Figure 03: Short wave of chloroform extract of KG

Figure 04: Long wave of chloroform extract of KG

Figure 05: Long wave of methanolic extract of KG (Trial 01)

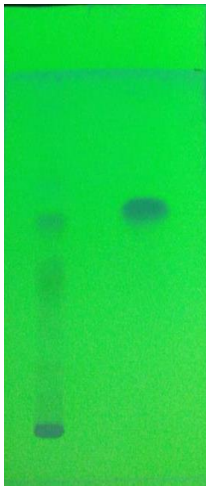

Figure 06: Short wave of methanolic extract of KG along with Gallic acid standard

Figure 07: Long wave of methanolic extract of KG along with Gallic acid

(Trial 02) **standard (Trial 02)**

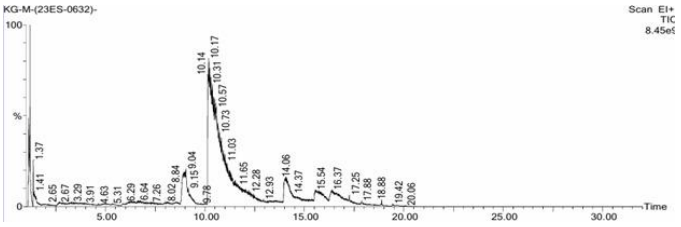

Figure 08: GC-Ms peaks of methanolic extract of KG

4. DISCUSSION:

Guda kalpana in Ayurvedic pharmaceutics, stands as one of its kind to have *Guda* (jaggery) as its main ingredient. This can serve in aiding proper pharmaceutical processing as well as render desired therapeutic benefits. In the present study, preparation of *Kalyanaka Guda* followed the classical way of *sagni gudapaka* (processing of jaggery based confections) followed by addition of drugs. [7]

The electron ionization (EI) process, a widely used ionization technique in GC-MS, induces extensive fragmentation, producing unique patterns that aid in unequivocal identification when combined with gas chromatographic retention-time data. [13] Notably, GC-MS offers a significant advantage due to its high reproducibility in generating mass spectra using EI. This hard ionization process yields very consistent mass spectra across different instruments, contributing to the technique's reliability and broad applicability. [14] Due to these advantages, the present study aimed to analyze KG in GC-MS rather than in LC-MS.

The organoleptic characters of the final product have fetched satisfactory results indicating proper preparation method. 3.74 pH of KG determined in the present study was done using 5% solution. Whereas, as per API the similar ingredient containing formulations *kalyanaka avaleha* and *Chyavanaprasha avaleha* pH range between 4.82-5.3 with 1 % aqueous solution. Hence, considering on the above said grounds the obtained pH of 3.74 seems to be a little nearer to the value mentioned in API. Loss on drying refers to the total moisture content in any given sample. The semi-solid consistency was corroborated with the loss on drying of 9.34 % as opposed to the study of Pravin RF et al., where the *Manibhadra guda* granules yielded 7.05% but the consistency is noted as crispy. [15] Total ash value represents the amount of minerals contained in any given sample. With KG, due to the addition of five kinds of salts during preparation, it is obvious to expect a total ash value of 3.09%. Acid insoluble ash is a test that indicates the level of silica impurities present in any substance. [16] This justifies the non-contamination of prepared KG with any silica impurities. The obtained total fat % of 4.20 is found satisfactory within the limits outlined by Pharmacopoeial standards for Ayurvedic formulations. Inorganic analysis detected sodium, sulphate and chloride salts from KG. Salts used in the preparation of KG - *like saindhava lavana, sauvarchala lavana, vida lavana, samudra lavana* and *audbidha lavana* might have contributed towards the detection of sodium in inorganic elemental analysis.

Sulphate content in KG might have probably been reflected from barium sulphate in *audbidha* and *samudra lavana*. Chloride is supposed to be a lucid element found in almost all of the salts included in KG. [17-21] Despite the availability of ample evidence to indicate the presence of other inorganic elements in different types of salts, the undetected state of other elements in present study might be due to their lower concentration that falls below the detection limit of qualitative analysis. Qualitative methods do not guarantee the absence, as the analytes may be having these elements in a trace amount below the defined limits of detection. [22]

For chromatographic analysis the sample was prepared through successive solvent extraction where it was subjected to reflux with solvents of increasing polarity in sequential manner. [23] The polarity of n-hexane, chloroform and methanol is 0.9, 25.9 and 76.2 respectively. [24] During the process of extraction, the polarity of the solvents determines the type of the compounds that will be extracted. N-Hexane, being a non-polar solvent, is effective at extracting hydrophobic compounds like fatty acids and esters. [25] While chloroform can extract alkaloids, flavonoids, and phenolic compounds and methanol extract sugars, amino acids, and glycosides. [26, 27] The sequential use of non-polar n-hexane to moderately polar chloroform, and finally to polar methanol was used for the extraction of a broad spectrum of compounds.

Due to the absence of an established TLC methodology specific to the formulation, we referred to the standardized procedures utilized for *Chyawanaprasha* and *Kalyanaka Avaleha* formulations in Ayurvedic Pharmacopoeia of India (API) due to presence of almost similar ingredients. [11] This approach, while not tailor made for our formulation, provides a reasonable insight for compound identification.

Prepared methanolic extract was subjected to two trials following the API method and also the method reported by Sawant L. et al., [12] to observe the standard gallic acid band. When gallic acid standard was run alongside the methanolic extract, distinct bands were observed at Rf values of 0.61 for the gallic acid standard and 0.60 for the methanolic extract under both short-wave (254 nm) and long-wave (366 nm) UV light. The close similarity in Rf values under both wavelengths suggests the presence of gallic acid in the methanolic extract. The difference may be attributed to few factors such as human error and matrix. [28] This method also yielded more bands indicating it was more efficient in partitioning.

The TLC of methanolic extract resulted in separation of more bands than other extracts tested; hence, methanolic extract was subjected to GC-MS testing. Also, it was noted that, though few bands were detected in the TLC, no compounds were obtained in the GC-MS analysis. The observed discrepancy likely arises from a couple of key factors.

First, the compounds separated and visualized on TLC may not always be detected by GC-MS due to difference in sensitivity and compound characteristics. Some compounds may become thermally unstable and eventually degrade during GC-MS analysis. [29] Additionally, the mobile phase can influence detection. [30]

The methanolic extract showed the presence of two compounds—4H-pyran-4-one and 2,3-dihydro-3,5-dihydroxy-6-methyl appearing at 8.95 and 9.04 minutes, respectively. These compounds are likely products of the Maillard reaction, which typically occurs when glucose and amino acids undergo thermal interaction. [31] Additionally, 5-hydroxymethylfurfural (HMF) was detected at several retention times (10.172, 10.232, 10.377, 10.552, and 10.827), which suggests its formation through the dehydration of glucose and fructose during the Maillard reaction, particularly under elevated temperatures and in the presence of acids such as ascorbic acid. Similarly, 5,5-Dimethyl-2(5H)-furanone (RT 10.932) appears to be another probable Maillard reaction byproduct. [32] Maillard reaction is a non-enzymatic reaction that occurs with reducing sugar on reaction with amino acids, polypeptides or proteins. It creates a distinct brown colour and aroma that could have probably resulted in the characteristic features of the end product KG.[33] The compound 4-Hepten-3-one, 4-methyl- (RT 11.007), is a ketone body that seems to have formed as a product of oxidation of free fatty acids formed as a result of thermal degradation of fatty acids. It is noted that *tila taila* used in the preparation of KG might have been a source of fatty acids. [34] Xylan is a polysaccharide that is important for the cell wall structure present in the plant. [35] Xylan might have disintegrated into xylose (RT 14.049) during the hydrothermal process.[36] D-mannoheptulose, a prominent 7-carbon soluble sugar, has been identified in various plant parts, including phloem sap, leaf petiole exudates, seeds, and mesocarp. Research indicates its prevalence in Primulaceae and Euphorbiaceae family plants, notably including *Vidanga* and *Amalaki*, key ingredients in our formulation, KG. [37] Mannoheptulose exhibits unique potential as a Caloric Restriction Mimetic (CRM) substance. Inhibiting hexokinase disrupts glucose metabolism, effectively simulating a state of energy restriction within the cell. This metabolic interruption prompts a cascade of cellular responses; leading to the activation of genes associated with improved metabolic efficiency and enhanced protective functions. Notably, mannoheptulose-enriched herbal feed has demonstrated anti-obesity effects in high-fat diet-fed animal models, suggesting potential applications for KG in obesity management while endo-2,3-O-Ethylidene-β-D-erythrofuranose is known function to as anaesthetic, beta-blocker, anti-cancer, nitric oxide inhibitor, CNS depressant, diaphoretic, and dopaminergic.[38, 39] This justifies the rational use of KG in classically mentioned indications like *gulma* (phantom disorder), *jwara* (fever), *kushta* (skin diseases), *arsha* (hemorrhoids), etc.,

This study lays the groundwork for quality control standards in KG, a widely used formulation, by establishing essential parameters. A novel Thin Layer Chromatography (TLC) method was developed to effectively separate KG's metabolites across a range of non-polar to polar media. Furthermore, Gas Chromatography-Mass Spectrometry (GC-MS) analysis identified various compounds resulting from KG's pharmaceutical processing, adhering to established protocols.

Building on these findings, future research directions include employing Liquid Chromatography-Mass Spectrometry (LC-MS) to detect additional non-volatile compounds. Frankly, if the pharmaceutical industry truly prioritizes proper standardization, we might finally achieve reliable medication consistency and efficacy. Implementing enhanced protocols is overdue. As for in-silico analysis and clinical trials - these are essential, not just procedural steps. They offer real opportunities to identify KG's therapeutic qualities, expand its possible applications, and genuinely validate its safety and effectiveness. It's time to move beyond theoretical claims and actually substantiate them with rigorous evidence.

5. CONCLUSION:

This study established the quality control profile and GC-MS fingerprint of KG, verifying the satisfaction of its incorporated ingredients. Inorganic elemental analysis revealed the presence of sodium, sulfate and chloride. The methanolic extract demonstrated notably better separation in the TLC experiments than the other extracts. GC-MS analysis backed this up, showing consistent results. So, it is clear that the methanolic extraction method is effective and offers solid insight into the chemical profile of KG.

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