

ORIGINAL RESEARCH ARTICLE

**Evaluation and FTIR Qualitative Analysis and of Gel Based Hand Wash Using *Camellia sinensis* (Green Tea) and *Myristica fragrans* (Nutmeg) Formulation**

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**ABSTRACT**

A gel based hand wash was formulated using *Camellia sinensis* (Green tea) and *Myristica fragrans* (Nutmeg), and evaluated by physical parameters like, colour, odour, spreadability, pH and overall appearance of the formulation. Identification of unknown materials, determination of quality, consistency, amount of components and detection of functional groups and characterization of covalent bonding information of the formulation was qualitatively analyzed using Shimadzu FTIR-8400S Fourier Transform Infrared Spectrometer instrument and spectra obtained for each sample was interpreted with a chart for interpreted with a chart for Characteristics IR absorption frequencies of organic functional groups and carbonyl containing functional group. The study showed that gel based hand wash containing mixture of *Camellia sinensis* (Green tea) and *Myristica fragrans* (Nutmeg) is rich in phyto-chemicals when compared to *Camellia sinensis* (Green tea) based hand wash gel and *Myristica fragrans* (Nutmeg) based hand wash gel.

**Key words:** *Camellia sinensis* (Green tea), *Myristica fragrans* (Nutmeg), hand wash gel, FTIR qualitative analysis

**INTRODUCTION**

Hand washings “remains the single most effective and cost-efficient method for preventing and reducing the transmission of nosocomial infections” [1]. Herbal companies all over the world produce a lot of cosmetics for one or the other purpose. The cosmetics are generally used externally like moisturizing lotion, fairness cream, and sunscreen lotions, anti ageing creams, face washes, hand and body washes etc. When an herbal cosmetic comes to market it is obvious that it had passed through several evaluation parameters direct from the crude drug to the finished product as per one or the other regulations. There are several guidelines for the efficacy evaluation of cosmetics [2]. Nature has been a source of medicinal agents for thousands of years and an impressive number of modern drugs have been isolated from natural sources, many based on their use in traditional medicine [3]. The extracts used in a formulation are concentrated preparations containing the active principles of vegetable or animal drugs. Chemical tests are carried out on the aqueous and hydro-ethanol extracts and on the powdered specimens using standard procedures to identify the constituents [4]. FTIR identifies chemical compounds in consumer

products, paints, polymer, coatings, which provides information about the chemical bonds and molecular structure of a materials whether organic or inorganic [5].

*Camellia sinensis* (Green tea) belongs to the family *Theaceae*, and is one of the most widely consumed beverages in the world, second only to water, and its medicinal properties have been widely explored. This plant has been traditionally useful in treating inflammations, asthma, heart diseases, lowering blood sugar and fights cancer. It is also useful in wound ulcers, coughs, bronchitis, burning sensation, diarrhea, dysentery, leprosy, fever, hair fall, greyness of hair and various skin diseases [6]. Green tea is prepared by picking, lightly steaming and allowing the leaves to dry [7]. Green tea extracts are utilized either in liquids (infusions) form or as dry extracts for further purification of the extract for its active constituents [8]. Due to the high antioxidant activity and potent antimicrobial activity of green tea extracts, it is useful as phyto-cosmeceutic, nutraceutical, additive, preservative, antioxidant and a promising solution to prevent apple juice and other foods from microbial contamination [9].

*Myristica fragrans* (Nutmeg) is an aromatic tree, 8 m or more tall with a dense crown. Leaves

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alternate, oblong 13 cm x 6.5 cm, dark green above and pale waxy beneath; Flowers dioeciously, small, creamy yellow; Fruit pear-shaped to globose drupe, 4-5 cm in diameter, yellowish, fleshy, splitting to reveal the seed (nutmeg) covered with a red, lacy, aril (mace)<sup>[10]</sup>. Its natural habitat is wet tropics and trees thrive with high, well-distributed annual rainfall, with little seasonal variation and temperatures over 25 – 35<sup>0</sup> C<sup>[11]</sup>. It is said to have stimulant, carminative and astringent properties. Its hallucinogenic properties are ascribed to the aromatic ethers myristicin, elemicin and safrole<sup>[12]</sup>.

Hence the present study was aimed to qualitatively analyse by FTIR and evaluate the efficacy of gel based hand wash formulations, which includes *Camellia sinensis* (Green tea) and *Myristica fragrans* (Nutmeg) extract in various concentrations.

### MATERIALS AND METHODS

#### Plant collection, extraction and formulation of gel based hand wash formulations using *Camellia sinensis* (Green tea) and *Myristica fragrans* (Nutmeg) extracts in various concentrations.

*Camellia sinensis* (Green tea) was collected from Tea estates, Munnar, Kerala and *Myristica fragrans* (Nutmeg) was collected from the Changanacherry taluk of Kottayam District, Kerala and were authenticated in Tamil Nadu Agricultural University.

**Preparation of hydro-ethanolic plant extracts:** About 1.5 kg of fresh plants were collected in bulk, washed under running tap water to remove adhering dust, dried under shade and powdered. The hydro-ethanolic extract was prepared using water by simple maceration technique<sup>[14]</sup>. About 50 g of the plant materials was extracted with 250 mL of hydro-ethanol (1:1) with occasional shaking for about 48 hours at room temperature 22-24 °C, and filtered. The filtrate was evaporated to dryness.

*Camellia sinensis* (Green tea) and *Myristica fragrans* (Nutmeg) were used as active ingredients in the hand wash gel formulation.

#### Preparation of gel base<sup>[15, 16, 17]</sup>

S.No	Ingredients	Quantity taken
1	Carbopol-940	1g
2	Purified water	100 ml
3	Triethanolamine	q.s. to neutralize gel base

**Procedure:** Carbopol-940 was soaked in water overnight (12 hours). Then the swelled polymer was stirred using a mechanical stirrer to ensure the uniform dispersion of the polymer. The pH was adjusted to 7.0 using Triethanolamine. Then this base was used to incorporate medicaments or active ingredients *Camellia sinensis* (Green tea) and *Myristica fragrans* (Nutmeg).

#### Formula 1: *Camellia sinensis* (Green tea) based hand wash gel.

Ingredients	Quantity
Gel base	30g
<i>Camellia sinensis</i>	2%
Sodium Lauryl sulphate	0.2%
Methyl paraben	0.1%

#### Formula 2: *Myristica fragrans* (Nutmeg) based hand wash gel

Ingredients	Quantity
Gel base	30g
<i>Myristica fragrans</i>	2%
Sodium Lauryl sulphate	0.2%
Methyl paraben	0.1%

#### Formula 3: *Camellia sinensis* (Green tea) and *Myristica fragrans* (Nutmeg) based hand wash gel

Ingredients	Quantity
Gel base	30g
<i>Camellia sinensis</i>	1%
<i>Myristica fragrans</i>	1%
Sodium Lauryl sulphate	0.2%
Methyl paraben	0.1%

#### Determination of pH

pH of the prepared formulation was measured using digital pH meter

#### Determination of Spreadability

The spreading ability of the formulations was evaluated at ambient temperatures with the following conditions. The spreading diameter of 0.01 g of the formulations, placed between two glass plates (16 x 16) was measured after 1 minute. The mass of the upper plate 125 g. the following classification was adopted for gel.

#### Determination of Consistency of the formulation

Fluid gel: > 70 mm,  
Semi fluid gel: 70 mm ≥ 55mm,  
Semi stiff gel: 55mm ≥ 47mm,  
Stiff gel: 47mm ≥ 40mm,  
Very stiff gel: <40mm

**Organoleptic evaluation:** All three variations of *Camellia sinensis* (Green tea) and *Myristica fragrans* (Nutmeg) based hand wash gel were exposed to different temperature conditions of 4<sup>0</sup> C 25<sup>0</sup> C and 37<sup>0</sup> C for a period of four weeks. A known amount of stored samples were taken out aseptically at different time intervals (24 hours,

after seven days, after two weeks and after four weeks) and organoleptically evaluated for its overall appearance (color, odor, gel consistency). Bleeding test was also performed to evaluate semisolid preparations by keeping them alternatively in different temperature zones, and observed for bleeding of liquid. Stability of the product for all climatic conditions is determined by the absence of liquid phase to omit out.<sup>[18]</sup>

#### **Fourier transform infrared spectroscopy (FT-IR) Qualitative analysis of *Camellia sinensis* (green tea) and *Myristica fragrans* (nutmeg) based hand wash gel**

All three variations (F1, F2 and F3) of developed hand wash gel using *Camellia sinensis* (Green tea) and *Myristica fragrans* (Nutmeg) were subjected to FTIR analysis using Shimadzu FTIR-8400S Fourier Transform Infrared Spectrometer instrument and obtained spectra for all 3 variations of the product was comparatively analyzed and interpreted with a chart for Characteristics IR absorption frequencies of organic functional groups and carbonyl containing functional group. The sample analysis Process with instrument specifications is

1. The source: IR energy is emitted from a glowing black-body source. The beam is passed through an aperture which controls the amount of energy presented to the sample (and, ultimately to the detector).
2. The Interferometer: The beam enters the interferometer where the “spectral encoding” takes place. The resulting interferogram signal then exist the interferometer.
3. The sample: the beam enters the sample compartment where it is transmitted through or reflected off to the surface of the sample, depending on the type of analysis being accomplished. This is where specific frequencies of energy, which are uniquely characteristic of the sample, are absorbed.
4. The detector: the beam finally passes to the detector for final measurement. The detectors used are specially designed to measure the special interferogram signal.
5. The computer: the measured signal is digitized and sent to the computer where the Fourier transformation takes place. The final infrared spectrum is then presented to

the user for interpretation and any further manipulation

6. Advantages of FTIR over dispersive technique
7. Speed: because all the frequencies are measured simultaneously, most measurements by FTIR are made in a matter of seconds rather than several minutes. This is referred to as the Fellgett advantage.
8. Sensitivity: is dramatically improved with FTIR for many reasons. The detectors employed are much more sensitive, the optical throughput is much higher which results in much lower noise levels, and the fast scans enable the co-addition of several scans in order to reduce the random measurement noise to any desired level (referred to as signal averaging).
9. Mechanical simplicity: the moving mirror in the interferogram is the only continuously moving part in the instrument. Thus there is very little possibility of mechanical breakdown.
10. Internally calibrated: The instrument employs a He Ne laser as an internal wavelength. Calibration standards referred to as Connes Advantage. These instruments are self calibrating and never needs to be calibrated by the user<sup>[19]</sup>.

#### **RESULT AND DISCUSSION**

Among the three formulations, Formula 3: Mixture of *Camellia sinensis* (Green tea) and *Myristica fragrans* (Nutmeg) based hand wash gel exhibited good overall appearance, good spreadability and appropriate pH suitable for utility.

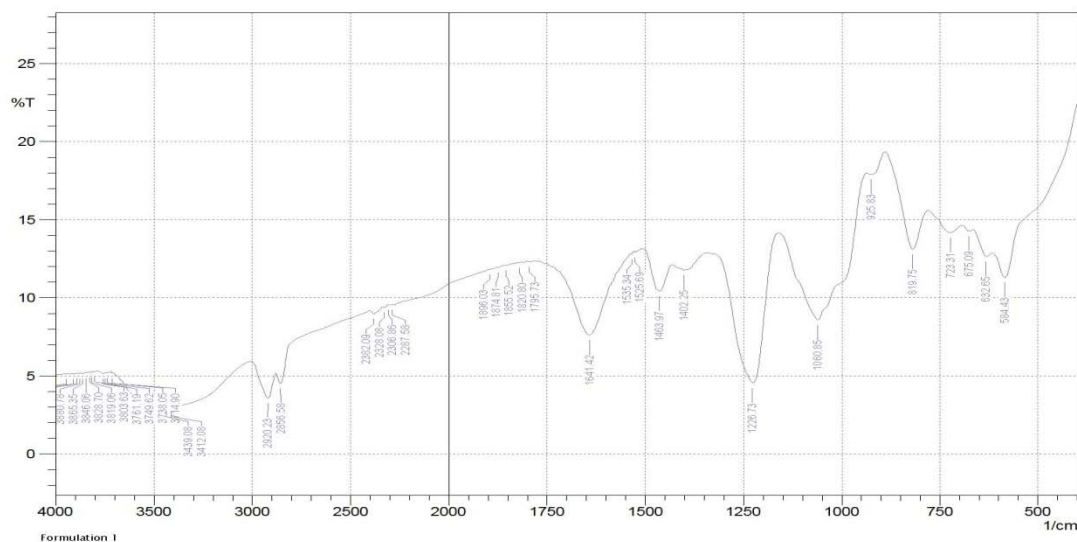
Comparing the results in (Tables 2, 3 & 4), Formulation 3 contains more functional groups such as aromatic alkenes, alcoholic groups, amines and alkanes when compared to the formulation 1 and 2. This shows that the product is rich in many phytochemicals. The presence of carboxylic acid group might be due to the oxidation of primary alcohol and also due to oxidation of few of the aldehyde sites. Based on the results from the organoleptic evaluation and FTIR analysis of F1, F2, F3 and the F3 with 2% concentration of active ingredients mixed with gel base to form gel based hand wash was considered as the best formulation.

**Table 1: Organoleptic evaluation of *Camellia sinensis* (green tea) and *Myristica fragrans* (nutmeg) based hand wash gel**

S.No	Formulations	Parameters studied	Organoleptic evaluation ( 4 <sup>0</sup> C, 25 <sup>0</sup> C and 37 <sup>0</sup> C for four weeks)
1	Formula 1	Overall appearance	Blackish green color, pungent odor, stiff gel
		Spreadability	47mm ≥ 40mm
		pH	4.1 – 6.7
2.	Formula 2	Overall appearance	Orangish green color, spicy odor, stiff gel
		Spreadability	47mm ≥ 40mm
		pH	4.1 – 6.7
3.	Formula 3	Overall appearance	Green color, pleasant odor, semi-fluid to semi-stiff gel, negative bleeding test
		Spreadability	Between 70 mm ≥ 55mm and 55mm ≥ 47mm,
		pH	4.1 – 6.7

**Table 2: FTIR spectral analysis results of *Formula 1***

S.No	Wave number (absorptions) (cm <sup>-1</sup> )	Functional groups	Type of vibration	Intensity
1	675.09	=C-H (Alkene)	Bending	14.279
2	723.31	=C-H (Alkene)	Bending	14.175
3	819.75	=C-H (Alkene)	Bending	13.124
4	925.83	=C-H (Alkene)	Bending	17.913
5	1060.85	(C-O) alcohol, (C-O) Ester,	Stretch	8.605
6	1402.25	(C=C) aromatic	Stretch	11.777
		(-C-H) alkane	Bending	
7	1463.97	(C=C) aromatic	Stretch	10.423
		(-C-H) alkane	Bending	
8	1525.69	(N-O) Nitro compounds	Stretch	12.93
9	1535.34	(N-O) Nitro compounds	Stretch	12.785
10	1641.42	(C=C) alkene	Stretch	7.63
		(C=O)) Amide	Bending	
11	1795.73	(C=O) carbonyl	Stretch	12.315
12	1820.8	(C=O) anhydride	Stretch	12.261
13	2856.58	(C-H) alkane	stretch	4.53
14	2920.23	(C-H) alkane	stretch	3.58
15	3412.08	(O-H) hydroxyl group	Stretch	3.053
16	3439.08	Ketone	Stretch	3.044



**Table 3: FTIR spectral analysis results of *Formula 2***

S.No	Wave number (absorptions) (cm <sup>-1</sup> )	Functional groups	Type of vibration	Intensity
1	615.29	=C-H (Alkene)	Bending	6.5411
2	711.73	=C-H (Alkene)	Bending	7.036
3	763.81	=C-H (Alkene)	Bending	7.5396
4	815.89	=C-H (Alkene)	Bending	8.51174

5	1037.7	(C-O) Ester,	Stretch	3.4757
6	1087.85	(C-O) alcohol, (C-N) amine	Stretch	3.5833
7	1244.09	(C-O) Acid	Stretch	5.3199
8	1386.82	(-C-H) alkane	Bending	3.7883
9	1423.47	(C=C) aromatic	Stretch	3.2716
10	1510.26	(C=C) aromatic	Stretch	4.2841
11	1583.56	(C=C) aromatic (N-H) amide	Stretch Bending	2.3429
12	1635.64	(C=C) Alkene	Stretch	2.5427
13	1730.15	(C=O) carbonyl	Stretch	5.3059
14	2856.58	(C-H) alkane	stretch	2.6476
15	2922.16	(C-H) alkane	stretch	1.7493
16	3375.43	(N-H) Amine, (O-H) alcohol	Stretch	1.6079

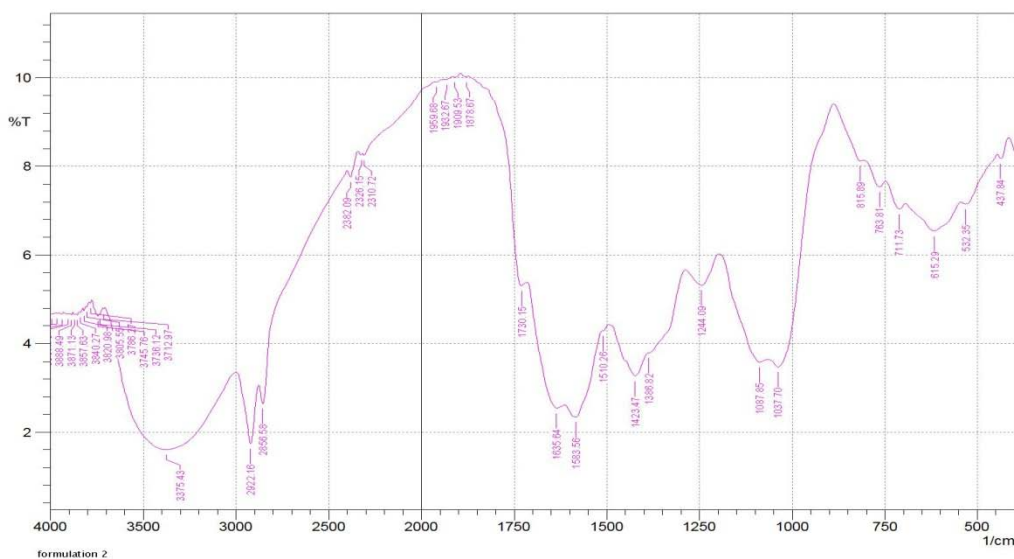
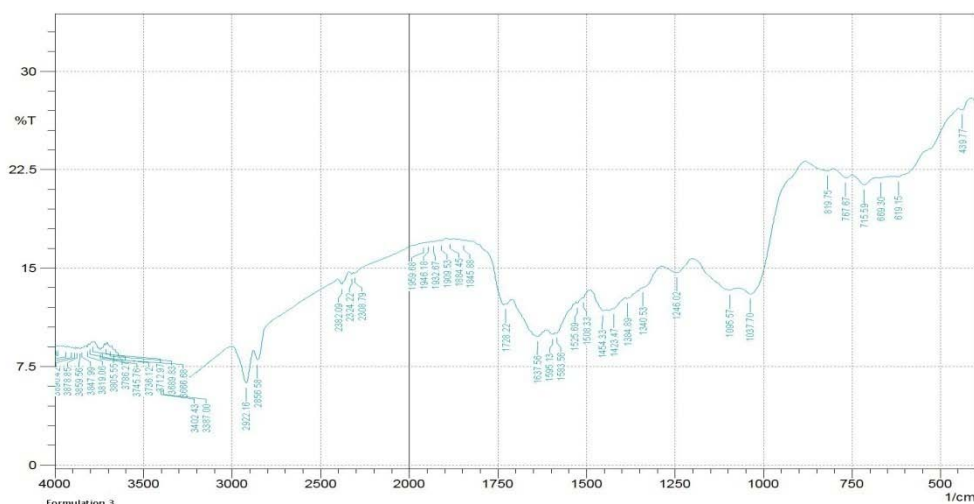


Table 4: FTIR spectral analysis results of Formula 3

S.No	Wave number (absorptions) (cm <sup>-1</sup> )	Functional groups	Type of vibration	Intensity
1	715.59	=C-H (Alkene)	Bending	21.367
2	669.3	=C-H (Alkene)	Bending	21.891
3	767.67	=C-H (Alkene)	Bending	21.875
4	819.75	=C-H (Alkene)	Bending	22.411
5	1037.7	=C-H (Alkene)	Bending	13.026
6	1095.57	(C-N) amine,(C-O)Alcohol	Stretch	13.337
7	1423.47	(-C-H) alkane, (C=C) aromatic	Bending	11.879
8	1454.33	(C=C) aromatic	Stretch	11.737
9	1508.33	(-C-H) alkane, (C=C) aromatic	Bending Stretch	12.743
10	1525.69	(C=C) aromatic, (N-O) Nitro compounds	Stretch	12.327
11	1583.56	(C=C) aromatic	Stretch	10.036
12	1595.13	(C=C) aromatic (N-H) amide	Stretch Bending	9.942
13	1637.56	(C=C) Alkene	Stretch	9.796
14	1728.22	(C=O) carbonyl	Stretch	12.246
15	2856.58	(C-H) alkane	stretch	8.009
16	2922.16	(C-H) alkane	stretch	6.261
17	3387	(N-H) Amine, (O-H) alcohol	Stretch	6.097
18	3402.43	(N-H) Amine, (O-H) alcohol	Stretch	6.094



## CONCLUSION

General hand wash gel containing *Camellia sinensis* (Green tea) and *Myristica fragrans* (Nutmeg) was formulated into three variations containing varied concentration of active ingredients. Organoleptic evaluation and FTIR analysis of the three formulations viz. Formula 1, 2 and 3 produced hand cleansing formula with therapeutic properties in a gel based carrier. Among them, Formula 3 containing mixture of *Camellia sinensis* (Green tea) and *Myristica fragrans* (Nutmeg) is the best preparation. For further studies, the formulation was subjected to *invitro* anti-microbial assays and *invitro* cell line studies.

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