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PHYTOCHEMICAL EVALUATION AND STANDARDIZATION OF POLYHERBAL ETHYL ACETATE EXTRACT AND ITS GEL-BASED FORMULATION OBTAINED FROM BERRIES AND FLAXSEED (EAPEG-BF)

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Keywords:

Berries, Flaxseed, Polyherbal extract, Standardization, Phytochemical evaluation

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ABSTRACT: Berries, fruits, and flaxseed have nowadays have increased in research studies. The berries such as *Rubus idaeus* (Red raspberry), *Vaccinium macrocarpon* (cranberry), *Vaccinium angustifolium* (blueberry) and *Fragaria x ananassa* Duch. (strawberry) have health beneficial compounds like anthocyanins, ellagitannins, ellagic acids, polyphenolic compounds and whereas in flaxseed the polyunsaturated fatty acids (PUFAs), α-linolenic acid, omega-3 fatty acids have a potential effect on health. This study aims to evaluate for standardization of polyherbal extract obtained from berries and flaxseed by qualitative phytochemical analysis, physicochemical analysis, Fourier transform infrared spectroscopy, UV analysis, thin layer chromatography. The maceration method is used for performing the extraction and plant-based edible polymer Acrypol 934 P for the preparation of the polyherbal ethyl acetate extract and its gel-based formulation. Different phytochemical tests and physicochemical evaluation procedures were followed for standardization of extract and afterwards gel formulation. The results confirmed the presence of various phytochemicals including flavonoids (rich in content in each), tannins, saponins, cardiac glycosides, phenolics. Different physicochemical studies including viscosity are some extent change due to gelation at room temperature, FTIR characterizes the presence of a particular functional group in the extract. Thin-layer chromato-graphy for the analysis of flavonoids with respective R_f value is determined. The study concludes that the berries and flaxseed have a potential effect on health benefits and they have a high range of flavonoids by phytochemical screening and stability studies are slight change due to conditions of climatic changes at room temperature.

INTRODUCTION: Herbal medicines and their products are increased day by day in a tremendous way in the last decades and because of this World Health Organization (WHO) takes broader steps for phytotherapy [1].

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Traditional medicines cover about 85% of the world population for their health needs and benefits. It is important and necessary to maintain the safety, quality, and efficacy of the plant and its products to avoid serious health problems [2].

Indian healthcare consists of medical facilities, and Ayurvedic medications or herbs remain dominant compared to allopathic medicines, particularly for the treatment of a variety of chronic disease conditions [3]. A numerous variety of plants have been evaluated for their therapeutic and effective potentials, which are found to be active against

various diseases in which the secondary metabolites especially the bioactive compounds present in them and provided the basis for several sophisticated traditional medicine systems like Ayurveda, Unani, Folk and Chinese [4]. In recent years, berry fruits have generated a lot of interest due to their low calories and high content of potential phytochemical or bioactive constituents such as (poly) phenols, fibre, minerals and vitamins.

As the polyphenols are secondary plant metabolites that are present in various berry family fruits and many more in which the major class is flavonoids (anthocyanins, flavan-3-ols, flavonols, flavanones, flavones, and isoflavones) [5]. Berries have different types of anthocyanins; for example, strawberries have pelargonidin, whereas blueberries contain cyanidin, malvidin conjugates [6].

Whereas flaxseed is also rich in antioxidants and oil, which contains linoleic and linolenic acids about 35-40% and 20-25% proteins, the seed contains cyanogenic glycosides as a bioactive compound which have potent pharmacological actions such as stimulating respiration, cardioprotective effects, anti-cancer effects, management of diabetes [7, 8].

The polyherbal extracts have greater potency rather than isolated constituents as polyherbal products are used for their medicinal and therapeutic application. It has also been recognized that polyherbal therapy or herb-herb interaction or combination can attain desired therapeutic and potential effects and decreased toxicity and show potent Pharma-cological action by exerting synergistic, antagonistic, and potentiate actions by various mechanisms [9, 10]. So, in the present study been made to evaluate the phytochemical evaluation and standardization parameters of polyherbal extract and gel formulation obtained from berries fruits family (Red raspberry, Cranberry, Blueberry and Strawberry) and flaxseed. The selection of these herbs depends on the presence of various flavonoids rich in antioxidants and helps in the treatment of various diseases. The objective of our study is to develop a new polyherbal formulation and phytochemical and standardization evaluation of polyherbal extract formulation.

METHODS AND MATERIAL:

Collection of Plant Material: The berries fruit Red raspberry (*Rubus idaeus*), Cranberry (*Vaccinium macrocarpon*), Blueberry (Vaccinium angustifolium), Strawberry (*Fragaria x ananassa*), and Flaxseed (*Linum usitatissimum*) were collected from the local market during the month of December-January. Berries were packed immediately into vacuum-sealed bags and stored in the freezer at -20 ºC until use, and the Flaxseeds are segregated from other extraneous material. The herbs were taxonomically identified by Dr. Naveen Kumar, Botanist of Christ Church College, CSA No. (PHARM/19-20/SPE/34).

Preparation of Polyherbal Extract: In the present study, the berries fruits were carefully selected, washed to remove impurities and shade dried. Individual fresh berries (100 gm) were macerated in a blender containing ethyl acetate (200 ml) for 30 minutes. After filtration, remove ethyl acetate extract (EAE) and dried under reduced pressure by Vaccum rotatory evaporator apparatus. Yields from 100 gm of fresh-weight berries are 17.476 gm. To remove moisture, the sample was kept in a desiccator for about 2-3 days. Then the sample is subjected to column chromatography for isolation of constituents by using solvents with varying polarity, i.e., methanol, distilled water (DW) (70:30 v/v), then the samples were collected in the glass beaker and kept on a water bath to evaporate and concentrate it. Simultaneously, thin layer chromatography (TLC) is also performed in the mobile phase solvent system, *i.e.*, chloroform: methanol: water in the ratio of (9:8:2 v/v/v) and (3:1:8 v/v/v).

Polyherbal Gel Extract Formulation: For this plant-based edible polymer is used. Accurately weighed 3 gm Acrypol 934 P polymer was taken in a beaker and dispersed in 75 ml of warm distilled, kept the beaker aside to swell the polymer for half an hour water, and then required quantity of methylparaben were dissolved by heating on the water bath, and homogenize with the homogenizer for about 15 min.

After this, sonicate it at 12000 rpm for 30 min with continuous stirring in one direction until the required consistency gel is not formed and adjust its pH (6.8-7). Take 1 gm of berries to extract each

in another beaker and mix with 3 ml of polymer solution with constant stirring and then incorporate 1 gm of flaxseed directly in the solution because after crushing or grinding it loses its ω-3 polyunsaturated fatty acids (PUFs). With these, we prepare three formulations with different variations.

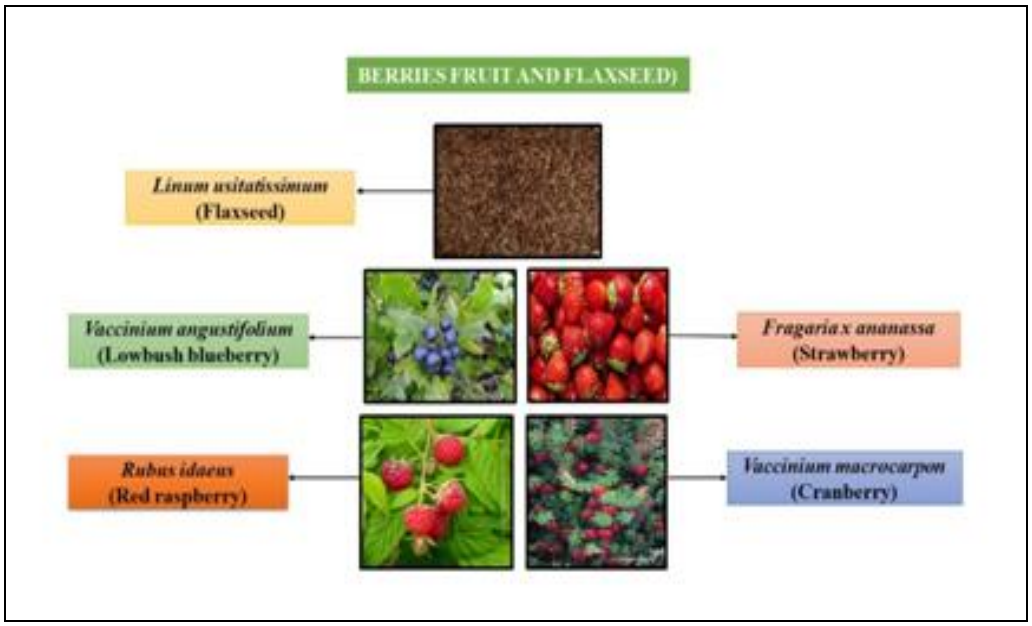

FIG. 1: BERRIES AND FLAXSEED PRESENT IN CURRENT RESEARCH STUDY FOR STANDARDIZATION

Chemicals and Reagents: Ethyl acetate, methanol, phosphoric acid, vanillin, silica gel G (for TLC) (Central Drug House (P) Ltd, New Delhi), chloroform, ethanol, distilled water, silica gel 60-120 mesh (for column chromatography) (Qualikems Fine Chem Pvt. Ltd., Vadodara) magnesium turnings, concentrated sulphuric acid, dilute hydrochloric acid, glacial acetic acid, ferric chloride, and acetic anhydride (Sigma-Aldrich, USA).

Qualitative Phytochemical Screening analysis: The extract was subjected to phytochemical screening investigation and was estimated or determined by a following standard procedure as these acts as templates for lead optimization programs.

The screening was carried out to identify the major natural chemical groups such as tannins, saponins, phenols, alkaloids, flavonoids, glycosides, coumarins, steroids, terpenoids, cardiac glycosides, and steroids. These general reactions adopted to analyses revealed the presence or absence of various constituents and compounds in the extract [11-13].

TABLE 1: PHYTOCHEMICAL SCREENING PROCEDURES FOR BERRIES EXTRACT EVALUATION

Chemical constituents	Standard Procedure	Appearance
Alkaloids	1 ml of crude extract + 5 ml of diluted HCL placed in a water bath at 60 ºC for about 15 min. 1 ml of Wagner's reagent was added into 1 ml of filtered suspension.	Reddish-brown precipitate
Flavonoids (Shinoda test)	Dry extract + 5 ml of 95% ethanol + few drops of concentrated HCL + 0.5 g magnesium turnings.	Pink colour observed
Saponins (Foam test)	A small amount of extract in a test tube with a small quantity of water. Shake vigorously.	The appearance of foam persisting for 10 min
Tannins and Phenolics	2 ml Extract in test tube + 3% $FeCl_3$ drop by drop.	The appearance of bluish-black precipitate
Cardiac glycosides	Extract + 1 ml glacial acetic acid (3%) + $FeCl_2$ + conc. H_2SO_4	Formation of brown ring
Phytosterols (Salkowski reaction)	Extract + 0.5 ml of chloroform, add 1 ml of conc. H_2SO_4 from sides of the test tube.	The appearance of reddish-brown colour in the chloroform layer

Thin Layer Chromatography (TLC) Analysis for Flavonoids: Thin layer chromatographic study was carried out on silica gel G plates in which 2 µg of extracts was loaded by using mobile phase chloroform: methanol: water in two different ratios *i.e.*, (9:8:2 v/v/v) and (3:1:8 v/v/v) to separate

flavonoid compounds of the extracts. the developed plate was air-dried. Than 1% vanillin in phosphoric acid was sprayed on the surface of the plate and incubate it for 15 min at room temperature. The present flavonoid compound of this extract was detected as a brown spot-on developed TLC plate. The R_f value of the bands was also determined [14].

Fourier Transform Infrared Spectrophotometer (FTIR): For the identification of types of chemical bonds or functional groups present in the extract is done by the most powerful tool, *i.e.*, Fourier Transform Infrared Spectrophotometer (FTIR). By interpreting the infrared absorption spectrum, the various chemical bonds and functional groups are determined and the wavelength of light absorbed is the characteristic feature of the chemical bonds which can be seen by the annotated spectrum. The extracted sample was loaded in FTIR spectroscope, *i.e.*, Perkin Elmer Spectrum 65, USA, with a scan range from 650-4000 cm^{-1} by using the Attenuated Total Reflection (ATR) Method. Results were obtained by using PerkinElmer Spectrum version 10.03.06 [15].

Estimation of Active Constituents by UV Analysis: Take two samples 10 ml of berries to extract in a 100 ml volumetric flask and made up to volume with distilled water and shaken well to dissolve the active constituents. Then the solution was filtered through Whatman filter paper and sonicate the sample. The various content of active constituents was determined by spectro-photometrically by using a standard curve plotted at different wavelengths 280 nm (λ_{max} of active constituents in the extracts).

Physico-chemical Evaluation of Polyherbal Extract GelFormulations:

Physical Appearance and Homogeneity Evaluation: The physical evaluation is done by physical appearance and homogeneity of the prepared polyherbal gel extract by visual perception.

Measurement of pH: The pH of the polyherbal gel extract formulations was measured by a digital glass electrode pH meter.

Spreadability: For determining Spreadability, we use two sets of glass slides which are of standard dimensions. The polyherbal gel extract formulation was placed over one of the slides. The other slide was placed on the top of the gel, in a way that the gel was sandwiched between the two slides in an area occupied by a distance of 7.5 cm along with slides. About 1 gm weight of gel was placed on the upper slides so that the gel was between the two slides was pressed uniformly to form a thin layer. Then the weight was removed and the excess of gel adhering to the slides was scratched off. The two slides were fixed in a position and to stand without the slightest disturbance and in such a way so that upper slides to slip freely by the force of weight tied on it. A 10-gm weight was tied to the upper slide carefully. The time taken by the upper slide to travel the distance of 7.5 cm and separate away from the lower slide under the influence of the weight was noted. This procedure was repeated three times, and then the mean was taken for calculation 16. Spreadability was calculated by using the formula below:

$$S = m \times 1/t$$

Where S= Spreadability, m = weight tied to upper slides (10 gm), l = length of the glass slide (7.5 cm), t= time taken to separate the slide each other (sec)

Viscosity: The viscosity of polyherbal gel extract is measured by using Brookfield viscometer with the spindle. And the polyherbal gel extract was poured into the container and the proper spindle (no. 74) was attached and after that viscosities were measured in 25 ºC and 50-250 rpm [16].

Stability Studies: For ensuring the quality, safety and efficacy, stability study was performed as per ICH guidelines for different polyherbal extract gel formulations, which were prepared by using Acrypol 934 P edible polymer as to exhibited better quality characteristics. The stability study of the most satisfactory formulation was sealed in a glass vial and kept at 25º ± 2 ºC, 30º ± 2 ºC and 40 ºC ± 2 ºC at RH 60% ± 5%, 65% ± 5% and 75% ± 5% for 3 months. At the end of the study, various parameters are analyzed.

RESULTS AND DISCUSSION: The composition of various polyherbal for the preparation of extract of berries is showed in **Table 2**.

The various ingredients used in the preparation of polyherbal extract gels and various components of its polyherbal gels formulations are showed in Table 3 and **Table 4**. In this flaxseed were directly incorporated because if they are crushed, it loses its polyunsaturated fatty acids. And the organoleptic properties of polyherbal gels formulations is showed in **Table 5**. The phytochemical screening analysis for different ethyl acetate extract for strawberry, red raspberry, cranberry, and blueberry is shown in **Table 6**. TLC analysis for flavonoids identification of ethyl acetate extracts of berries is showed in **Table 7**. The physicochemical evaluation of polyherbal gels formulations showed in **Table 8**, as low viscosity because of difference due to gelation temperature as the presence of polymer and its concentration. FTIR characterization of different bonds and functional group present in extracts as showed in Table 9 and depicted in **Fig. 2** with peak ranges. UV analysis is showed by the **Fig. 3,** which estimates the active constituent's wavelength in the extract. Stability studies of polyherbal extract gel formulation (F1) with different variations of temperature and relative humidity which depicts the pH, colour, odour, homogeneity of the formulation as shown in **Table 10**.

The presently prepared formulations are used to treat several disorders related to cardiovascular and metabolic syndromes. As these formulations have potential bioactive compounds which undergo various mechanism of actions and produced a pharmacological action in a level of humankind. The phytoconstituents were rich in anthocyanins, flavonoids, ellagitannins, and cardiac glycosides, which have numerous activities in accordance to treat the diseases. These act as an antioxidant, inhibiting the reactive oxygen species and inhibit the free radical generation by free radical scavenging activity. Matairesinol, lariciresinol and pinoresinol are also present but in the small range. Numerous studies suggested that Alpha-Linolenic Acid (ALA) may be responsible and the potent bioactive which produces the antihypertensive action of flaxseed [17].

TABLE 2: COMPOSITION OF POLYHERBAL FOR PREPARATION OF EXTRACT OF BERRIES

S. no.	Berry fruits	Botanical name	Family	Parts used	Quantity
1	Red raspberry	*Rubus idaeus*	Rosaceae	Fruit (drupelets)	100 gm
2	Lowbush blueberry	*Vaccinium angustifolium*	Ericaceae	Fruit (drupelets)	100 gm
3	Cranberry	*Vaccinium macrocarpon*	Ericaceae	Fruit (drupelets)	100 gm
4	Strawberry	*Fragaria x ananassa*	Rosaceae	Fruit (drupelets)	100 gm

As such the results indicate the presence of compounds which acts as nutraceuticals supplements for preparation of dosage forms of the formulations along with its vital gel properties and stability conditions. Thus, the prepared formulations required further clinical and mechanistic basis for the manufacturing of folk medicines for suffering patients from hypertension, cardiovascular disorders related to diabetes, metabolic syndrome.

TABLE 3: INGREDIENTS FOR POLYHERBAL EXTRACT GELS

S. no.	Ingredients	Quantity
1	EAE of *Rubus Idaeus*	0.02%
2	EAE of *Vaccinium angustifolium*	0.02%
3	EAE of *Vaccinium macrocarpon*	0.02%
4	EAE of *Fragaria x ananassa*	0.02%
5	Flaxseed *(Linum usitatissimum)*	0.01%
6	Acrypol 934 P	0.03%
7	Methylparaben	0.2%
8	Distilled water	75 ml

TABLE 4: COMPOSITIONS OF POLYHERBAL EXTRACT GELS FORMULATIONS

S. no.	Formulation	Acrypol 934 P (%)	Extract (%)	Methylparaben (%)	Flaxseed (%)
1	F1, Gel (2%)	0.03%	0.02%	0.2%	0.01%
2	F2, Gel (3%)	0.03%	0.03%	0.2%	0.01%
3	F3, Gel (4%)	0.03%	0.04%	0.2%	0.01%

TABLE 5: ORGANOLEPTIC PROPERTIES OF POLYHERBAL GELS FORMULATIONS

Formulation	Appearance	Colour	Odour	Taste
F1, Gel (2%)	Soft, Smooth	Purplish pink	Fruity	Sweet taste
F2, Gel (3%)	Soft, Smooth	Purplish pink	Fruity	Sweet taste
F3, Gel (4%)	Soft, Smooth	Purplish pink	Fruity	Sweet taste

TABLE 6: PHYTOCHEMICAL ANALYSIS OF DIFFERENT ETHYL ACETATE EXTRACTS OF RED RASPBERRY, LOWBUSH BLUEBERRY, CRANBERRY AND STRAWBERRY

Phytochemical screening Analysis	Ethyl acetate extract of *Rubus idaeus*	Ethyl acetate extract of *Vaccinium angustifolium*	Ethyl acetate extract of *Vaccinium macrocarpon*	Ethyl acetate extract of *Fragaria x ananassa*
Alkaloids (Wagner's reagent test)	-	-	-	-
Cardiac glycosides	++	+++	+++	++
Flavonoids (Shinoda test)	+++	+++	++	+++
Saponins (Foam test)	+	+	+	+
Tannins and phenolics (Ferric chloride test)	+	+	+	+
Phytosterols (Salkowski test)	-	-	-	-

Note: Presence/Absence (+ Presence and -Absence)

TABLE 7: THIN-LAYER CHROMATOGRAPHY (TLC) FOR FLAVONOIDS ANALYSIS OF A POLYHERBAL EXTRACT OF BERRIES (RED RASPBERRY, CRANBERRY, BLUEBERRY AND STRAWBERRY)

Extracts	Solvent System	Ratios	Revealing reagent	No. of spots	Rf value		
Ethyl acetate extract of berries	Chloroform: methanol: water	9:8:2 (v/v/v)	1% vanillin in phosphoric acid	3	0.66	0.50	0.17
	Chloroform: methanol: water	3:1:8 (v/v/v)	1% vanillin in phosphoric acid	2	0.7	0.5	

TABLE 8: PHYSICO-CHEMICAL EVALUATION OF POLYHERBAL GEL EXTRACT FORMULATION

Formulation	Concentration (%)	pH	Spreadibility (gm cm/sec)	Viscosit (cm Poise)		Physical Appearance
				At 0 Time	After 3 Months	
F1, Gel (2%)	0.02%	6.23	18.12	19000	22000	Smooth, Soft Clear, Purplish pink
F2, Gel (3%)	0.03%	6.26	19.19	25000	28000	Rough,Dull, Purplish pink
F3, Gel (4%)	0.05%	6.56	17.33	30000	33000	Lumps, Purplish pink

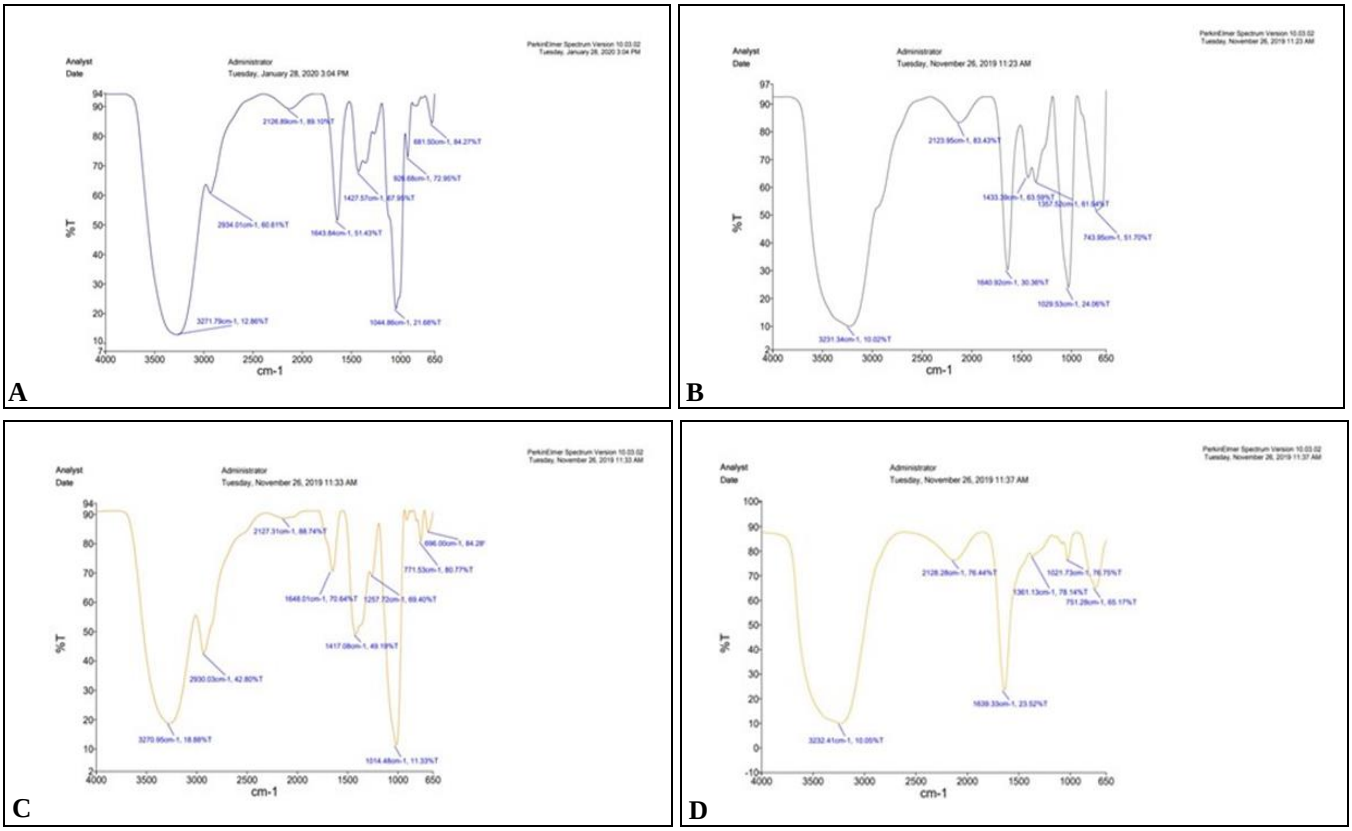

FIG. 2: FTIR SPECTRUM OF POLYHERBAL EAE OF BERRIES (A) RED RASPBERRY (B) CRANBERRY (C) STRAWBERRY (D) BLUEBERRY

TABLE 9: FTIR PEAK RANGES WITH THEIR ASSIGNED RESPECTIVE FUNCTIONAL GROUPS OF POLYHERBAL ETHYL ACETATE EXTRACT OF BERRIES

FTIR Range	Peak Origin Groups
3200-3400 cm^{-1}	O-H group in alcohol and phenol
2800-3000 cm^{-1}	CH stretching bond of alkane or alkyl groups
1600-1800 cm^{-1}	Carbonyl group of amide and esters
1000-1200 cm^{-1}	Primary amines

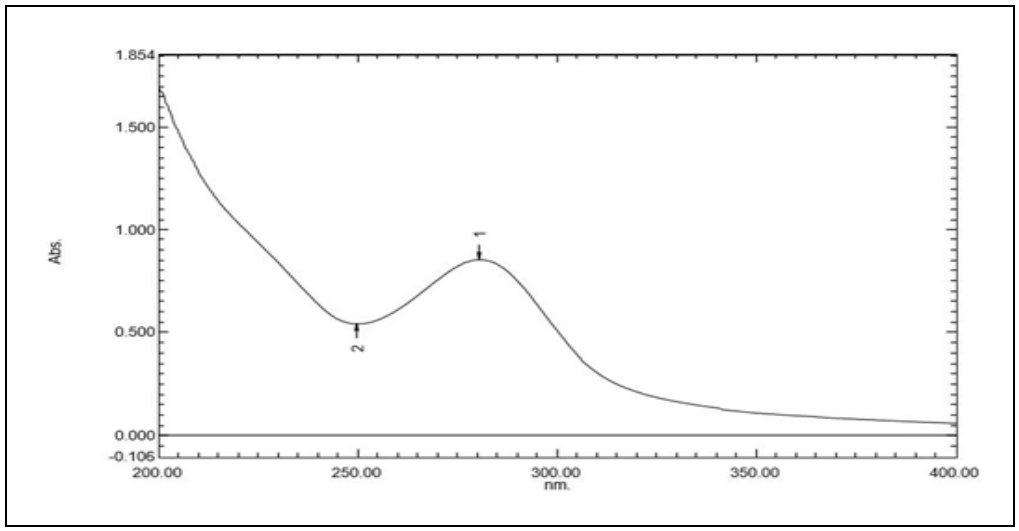

FIG. 3: UV SPECTRUM OF EAE OF BERRIES AT 280 nm

TABLE 10: STABILITY PROFILE OF EAPEG-BF

S. No.	Study	EAPEG-BF gel formulation (F1 Gel 2%) Storage condition											
		25ºC ± 2 ºC/60% ± 5%RH Months				30ºC ± 2 ºC/65% ± 5%RH Months				40ºC ± 2 ºC/75% ± 5%RH Months			
		0	1	2	3	0	1	2	3	0	1	2	3
1	Colour	No change in colour				No change in colour				Change in colour			
2	Odour	Fruity				Fruity				Fruity			
3	Homogeneity	Good and smooth				Good and smooth				Good and smooth			
4	pH	6.4	6.4	6.3	6.3	6.4	6.4	6.3	6.50	6.3	6.4	6.3	6.3

CONCLUSION: Herbs and their type of formulations are more potent than any other system of medications because they have lesser side effects, toxicity, low cost, and a higher range of suitability and availability among the community. The polyherbalism leads to a high range of therapeutics effects towards the various chronic as well as mild diseases and disorders due to their synergistic effects, and due to this, the berries and flaxseed have a higher content of flavonoids which act as antioxidants and helps in the ailment of various diseases by the use in daily lifestyle dietary intake of an individual. Their phytochemical bioactive contents such as anthocyanins, ellagic acid, ellagitannins, polyunsaturated fatty acids deal with various mechanisms of the human body to reduce the effects of any type of disease. This study also concludes that berries, fruit, and flaxseed consumption is good for health and any type of formulation prepared either in gel, syrup, extract jams, jellies were prepared with respective procedures and methods.

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REFERENCES:

1. Che CT, Wang ZJ, Chow MSS and Lam CWK: Herb-herb combination for therapeutic enhancement and

advancement: Theory, practice and future perspectives. Molecules 2013; 18(5): 5125-41.
2. Waxler-Morrison NE: Plural medicine in India and Sri Lanka: do ayurvedic and Western medical practices differ? Social Science Medicine 1988; 27: 531-44.
3. Mohd Fuat AR, Aidoo KE and Calvert TW: G. Mycoflora, Cytotoxicity, and DNA Interaction of Polyherbal Products from Malaysia. Pharma Biology 2006; 44(1): 23-31.
4. Gurib-Fakim A: Medicinal plants: Traditions of yesterday and drugs of tomorrow. Molecular Aspects Medicine. 2006; 27(1): 1-93.
5. Rio DD, Rodriguez-Mateos A, Spencer JP, Tognolini M, Borges G and Crozier A: Dietary (poly)phenolics in human health: structures, bioavailability and evidence of protective effects against chronic diseases. Antioxidant Redox Signal 2013; 18: 1818-92.
6. CrozierA, Yokota T, JaganathIB, Marks S, Salthmarsh M and Clifford MN: Secondary metabolites as dietary components in plant-based foods and beverages. Blackwell Publishing: Oxford, UK 2006; 208-02.
7. Nahrin A, Junaid M, Afrose SS, Alam MS, Hosen SM, Akter R and Sharmin T: A Review of *Saurauia roxburghii* Wall. (Actinidiacaea) as a Traditional Medicinal Plant, Its Phytochemical Study and Therapeutic Potential. Mini-Reviews in Medicinal Chemistry 2020; 20(19): 2036-51.
8. Dupasquier CM, Weber AM, Ander BP, Rampersad PP, Steigerwald S, Wigle JT, Mitchell RW, Kroeger EA, Gilchrist JS, Moghadasian MM, Lukas A and Pierce GN: Effects of dietary flaxseed on vascular contractile function and atherosclerosis during prolonged hypercholesterolemia in rabbits. American Journal of Physiology-Heart and Circulatory Physiology2006; 291: H2987-H2996.
9. Wachtel-Galor S, Yuen J, Buswell JA and Benzie IF: *Ganoderma lucidum* (Lingzhi or Reishi). In Herbal Medicine: Biomolecular and Clinical Aspects,CRC Press/Taylor & Francis, Edition 2, 2011.
10. Wang H, Wang J, Qiu C, Ye Y, Guo X, Chen G, Li T, Wang Y, Fu X and Liu RH: Comparison of phytochemical profiles and health benefits in fibre and oil flaxseeds *Linum usitatissimum* L. Food chemistry2017; 214:227-233.
11. Alabri TH, Al Musalami AH, Hossain MA, Weli AM and Al-Riyami Q: Comparative study of phytochemical screening, antioxidant and antimicrobial capacities of fresh and dry leaves crude plant extracts of Datura metel L. Journal of King Saud University-Science 2014; 26(3): 237-243.
12. Mandal S, Patra A, Samanta A, Roy S, Mandal A, Mahapatra TD, Pradhan S, Das K and Nandi DK: Analysis of the phytochemical profile of *Terminalia arjuna* bark extract with antioxidative and antimicrobial properties. Asian Pacific journal of tropical biomedicine 2013; 3(12): 960-66.
13. Kokate CK: Practical Pharmacognosy. Vallabh Prakashan, 2001:45-49.
14. Amalraj A and Gopi S: Medicinal properties of *Terminalia arjuna* (Roxb.) Wight & Arn.: a review. Journal of traditional and complementary medicine 2017;7(1):65-78.
15. Eberhardt TL, Li X, Shupe TF and Hse CY: Chinese tallow tree *Sapium sebiferum* utilization: characterization of extractives and cell-wall chemistry. Wood Fiber Science 2007; 39(2): 319-24.
16. Nayak SH, Nakhat PD and Yeole PG: Development and evaluation of cosmeceutical hair styling gels of ketoconazole. Indian Journal of Pharmaceutical Science 2005; 52: 231-33.
17. Barbeau PA, Holloway TM, Whitfield J, Baechler BL, Quadrilatero J, van Loon LJ, Chabowski A, and Holloway GP: α-Linolenic acid and exercise training independently, and additively, decrease blood pressure and prevent diastolic dysfunction in obese Zucker rats. The Journal of Physiology 2017; 595(13): 4351-64.
18. Babu TM and Kumar CR: Standardization of Atasi beeja to physicochemical and phytochemical evaluation. IAMJ 2016; 11(4): 3311-15.

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