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Exploring the potentials of *Ziziphus mauritiana* Lam. seed kernel oil as pharmaceutical oil base: Physicochemical characterization and ketoconazole soap formulation

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ABSTRACT

Ziziphus mauritiana Lam. (Rhamnaceae) is a large spiny shrub or small tree, native to the Indian subcontinent that can reach a maximum height of 9–15 m. This plant stands as a renowned tropical fruit variety, commonly recognized as the Indian plum, Desert apple, or Indian Jujube. The objective of this study was to examine the standard physicochemical parameters of *Z. mauritiana* seed kernel oil and to formulate the ketoconazole soap (2 % w/w), using the obtained oil, as a base. The oil was obtained through hexane extraction from the seed kernels. To formulate pharmaceutical ketoconazole soap, *Z. mauritiana* seed kernel oil was subjected to a basic saponification reaction using potassium hydroxide. All the examined physicochemical parameters, namely acid value (4.71 mg KOH/g), saponification value (229.18 mg KOH/g), peroxide value (4.15 milliequivalents KOH/g), ester value (224.47 mg KOH/g), iodine value (11.19 mg KOH/g), refractive index (1.448), pH (5.93), viscosity (89 cP), and specific gravity (0.912 g/mL) were within the acceptable range for industrial purposes. The examination of quality control parameters, namely drug content (99.49 %), total fatty matter (71.13 %), foam retention time (17.21 min), foam height (18.56 cm), moisture content (9.14 %), and pH (7.16) indicated that the newly formulated ketoconazole soap complied with the acceptable limits. In summary, our research demonstrated the excellent physicochemical stability of *Z. mauritiana* seed kernel oil and its suitability as a soap base, supporting its promising prospects for cost-effective production of cosmetics, soaps, and shampoos in the pharmaceutical and cosmeceutical industries, reducing reliance on synthetic bases.

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1. Introduction

Ziziphus mauritiana Lam. (Family: Rhamnaceae) is a large evergreen spiny shrub or small tree with a height ranging from 9 to 15 m [1]. This plant is one of the popular tropical fruits native to the Indian subcontinent, commonly called Ber, Indian plum, Desert apple, Indian Jujube, or Jujube [2,3]. The plant is widely distributed throughout India, Malaysia, Nepal, Afghanistan, Pakistan, Australia, China, Japan, Philippines and the southern part of Africa [3,4]. It is renowned for its delicious, nutritious, crisp, and sugary-sweet drupe fruits (Fig. 1A) with an apple-like aroma [4]. The fruits are ethnomedicinally employed for the treatment of blood sickness, tuberculosis, vomiting, constipation, anorexia, sexual debility, and high blood pressure [5,6]. Within traditional Chinese medicine (TCM), the dried fruits are widely prescribed as a tonic with anti-tumor and styptic properties [4]. The seed kernels (Fig. 1C) extracted from the fruit possess beneficial properties for enhancing heart and brain functions. They are utilized in the treatment of morning sickness, wounds, aconite poisoning, abdominal pain during pregnancy, ophthalmic disorders, leucorrhea, cough, asthma, vomiting, diarrhea, burning sensation, anxiety, rheumatic disease, insomnia, and also serve as oral contraceptive [3,7–11]. In Ayurveda, the plant's root is recommended for alleviating cough, headache, nausea, biliousness, and as a cooling bitter tonic [11,12]. The roots are also utilized for managing gout, epilepsy, chronic fever, ulcers, and headaches [13]. The bark is employed as an astringent and for treating conditions such as boils, diarrhea, and dysentery [11]. The leaves have cooling, antipyretic, and anthelminthic capabilities that make them useful for managing asthma, typhoid fever, and stomatitis [11,14].

Scientific inquiries into *Z. mauritiana* fruit have unveiled a rich array of pharmacological properties, highlighting its prowess as a potent agent. These consist of anticancer effect [9], immunomodulatory potential [15], hypnotic effect [16], thrombolytic effect [17], analgesic effect [17], hypoglycemic effect [18], antiplasmodial effect [19], antidiarrheal effect [17], membrane stabilizing effect [17], and antibacterial effect [20]. Also, aqueous-ethanolic extract of the seed was reported to have anti-proliferative activity in human leukemia cell line (HL-60), cervical cancer cells (HeLa), acute lymphoblastic leukemia (Molt-4) cell lines, and ehrlich ascites carcinoma of Swiss albino mice [9]. Gas chromatography-mass spectroscopy (GC-MS) analysis of fatty acid methyl ester in saponified seed oil of *Z. mauritiana* had shown the presence of fatty acid methyl esters such as 7-octadecanoic acid (55.2 %), 9,12-octadecanoic acid (25.3 %), hexadecanoic acid (7.2 %), octadecanoic acid (6.9 %), eicosanoic acid (2.1 %), 11-ecosanoic acid (1.9 %), and docosanoic acid (1.5 %) [21]. However, the unsaponified fraction was dominated by stigmasterol (23.6 %), squalene (14.0 %), Δ 4- sitosterol-3-one (6.8 %), campesterol (5.8 %) and γ-tocopherol (4.3 %) [16]. The high presence of protein (~36 %), lipids (~27.4 %),carbohydrates (~21 %), dietary fiber (~11.04 %), minerals, and a wide range of fatty acids along with other metabolites, like tocopherol, sterols, and monounsaturated fatty acids indicate the potential benefits of this seed, in nutritional and medicinal purposes [21–25].

Soaps are a long-standing cleaning agent that we use to prevent disease spread and maintain healthy skin condition [26]. They help us maintain hygiene by removing dust particles and microorganism from exposed parts of the body, such as the hand and face. Soaps are prepared through the saponification of free fatty acids by an alkaline base [27]. Despite the growing demand for various cleansing products, many commercially available options face multiple issues. These problems include the high production costs associated with synthetic bases, instability of the chemical base [28], base-induced skin hypersensitivity reactions [29,30], and the need for numerous additives to maintain base stability [28,31]. Recently, there has been a substantial surge in consumer interest for natural, organic, and

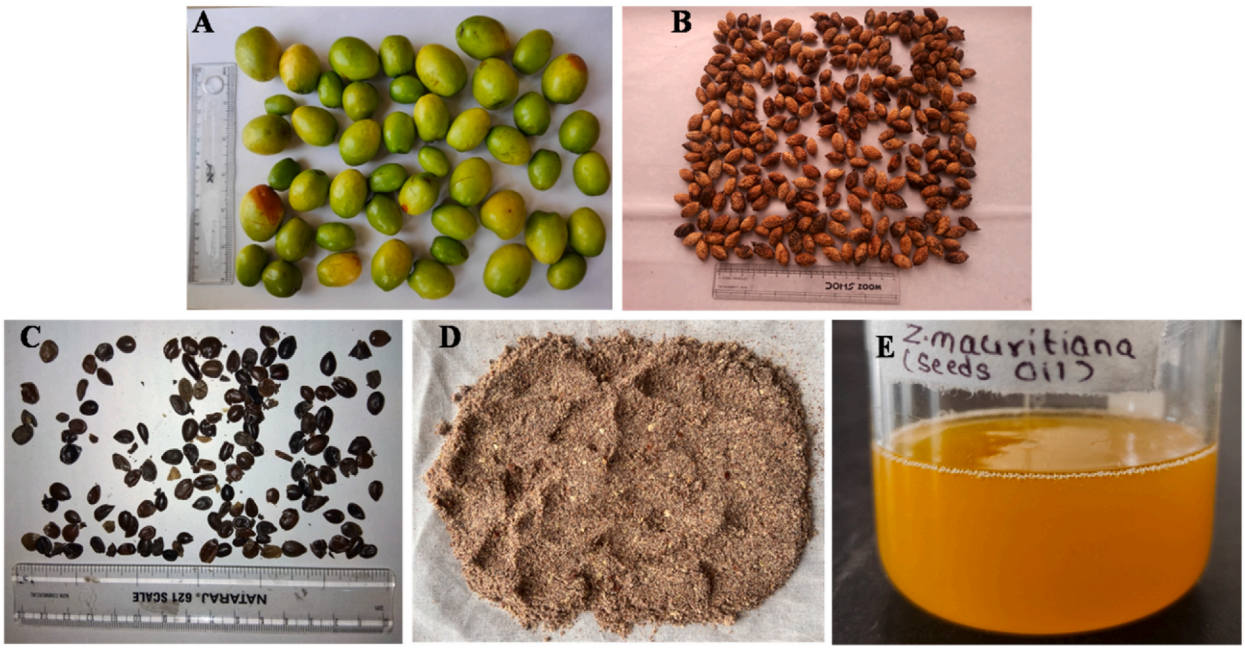


Fig. 1. Photographs of *Z. mauritiana* ripen fruits (A), dried seeds (B), dried seed kernels (C), dried powder of seed kernels (D), and hexane extract (E).

sustainable personal care items. The natural product sector has experienced consistent double-digit growth every year since 2007, with global sales reaching nearly $30 billion in 2014 [28]. Currently, the market of organic soap is projected to witness significant growth, increasing from USD 723.51 million to USD 1.1 billion by 2028, with a remarkable compound annual growth rate of 8.74 % [32]. As a result, various industries and research organizations are currently focusing on exploring the potential applications of different fixed oils derived from natural sources, such as plants, herbs, and seeds. Consequently, several underutilized seed oils are being effectively implemented in the production of cosmetic items, including soaps, creams, shampoos, toothpaste, hair conditioners, skincare products, beauty makeup products, hair care products, and deodorants [33]. The benefits of adding herbs to soaps have drawn increased attention, apart from the simple purpose of cleaning. For example, castor oil provides good lather in the soap, along with additional moisturizing, conditioning, and softening effects on the skin [26]. Neem oil has antibacterial, anti-scabies, anti-inflammatory, and analgesics properties [33]. Similarly, Mentha oil has antibacterial, antioxidant, and mind relaxation effects [27]. Furthermore, guava leaves, turmeric, aloe vera, nuts and rose petals, and honey are most often used for essence and/or other additional protective effects on skin, due to the phytoconstituents present in them [34,35].

Even though there is evidence of high nutritional value and medicinal use for this seed, the majority of people discard it as waste [14]. Therefore, the objective of this research is to evaluate physicochemical characteristics (including acid value, ester value, saponification value, iodine value, peroxide value, pH, refractive index, melting point, specific gravity, and viscosity) of *Z. mauritiana* seed kernel oil and to prepare 2 % w/w ketoconazole soap formulation by saponification.

2. Materials and methods

2.1. Chemical substances and organic solvents

Standard drug ketoconazole (99.89 % assay and 0.25 % loss on drying) was purchased from Biogain Remedies, Butwal, Nepal. Potassiumbromate iodine, sodium thiosulphate, iodine trichloride, and potassium iodide were purchased from Merck, India. Iodine monobromide and mercuric iodide were purchased from Thermo Fischer Scientific, India.

2.2. Equipments

The instruments used in this study was as follow: UV Spectrophotometer (UV-1800 model; Shimdazu Corporation Pvt. Ltd, China), Brookfield viscometer (Brookfield AMETEK, DV plus model, USA), Abbe refractometer RFT-A1, Toledo, USA, Sonicator (INDOSATI Scientific Lab Equipments), PC9500 Benchtop digital pH meter, USA, Rotary evaporator (R-210/215, BUCHI Labor technok AG, Switzerland), Digital balance (ATX224, SHIMADZU Corporation, Philippines), Hot air oven (S. M. Scientific Instruments (P) Ltd., Delhi).

2.3. Collection and authentication of plant sample

Ripen fruits of *Z. mauritiana* were collected from the Bhairahawa, Rupandehi, Nepal (tropical region, 544 m above sea level) in April 2022. The collected plant materials underwent identification and authentication at the National Herbarium and Plant Laboratory Godawari, Nepal.

2.4. Extraction of Z. mauritiana seed kernel oil

Collected fruits were divided into pulp and hard seeds. The seed kernels were carefully separated from the tough outer shell and dried on clean filter paper in a well-ventilated laboratory room at a temperature of 25° Celsius (^{o}C) for a period of two weeks. After drying, the seed kernels were ground using a grinder, resulting in a coarse powder. The extraction method was adopted from the literature [36] with slight modification. Cold maceration method was employed for the extraction process, where 1 kg of the dried powder (in the ratio of 200 g powder per liter of hexane) was soaked with hexane in conical flasks, with intermittent shaking over a span of two days. The mixture was then filtered, and the remaining marc was pressed and filtered again. This process was repeated three times. The collected filtrate was combined and subjected to drying by using a rotary vacuum evaporator at a temperature of 40 °C, resulting in an orange-colored liquid hexane extract known as *Z. mauritiana* seed oil. The extract was further dried in a vacuum desiccator for a few days and stored in a refrigerator at 4 °C until further use. Fig. 1 illustrates photographs of the collected fruits, dried seeds, seed kernels, and hexane extract (oil) of *Z. mauritiana*. The summary of whole process is depicted in flowchart 1.

2.5. Analysis of physiochemical parameters of the Z. mauritiana seed oil

The physicochemical examination was conducted following the relevant literature [36] and guidelines outlined in the Indian Pharmacopeia (IP vol-II, 2018) [37], with sight modification and each experiment was repeated three times for accuracy.

2.5.1. Calculation of acid value

Around 10 g (g) of the *Z. mauritiana* seed oil was dissolved in a solution consisting of 50 mL of a neutralized mixture of ether and 95 % ethanol in a 1:1 volume-to-volume ratio. Subsequently, this sample solution was titrated with standardized potassium hydroxide to ascertain the acid value. Equation i, as mentioned in the study by Pandey et al. [36], was employed for this determination.

$$\text{Acid Value} = \frac{5.61n}{m} \quad \text{(i)}$$

where, the symbol 'n' represents the volume, measured in milliliters (mL), of 0.1 M potassium hydroxide that participated in the reaction. Similarly, the symbol 'm' represents the mass, measured in grams (g), of the sample used for the evaluation.

2.5.2. Determination of iodine value

The iodine value was determined using the Hanus method, as described by Pandey et al. [36]. Initially, 1 g of the sample was placed in a dry iodine flask with a capacity of 300 mL. Subsequently, 15 mL of chloroform were added to the flask, followed by the careful addition of 25 mL of iodine monobromide solution from a burette. The resulting mixture was then incubated in a dark location for 30 min, with intermittent shaking. Subsequently, 100 mL of distilled water and 10 mL of a 30 % w/v solution of potassium iodide were added to the sample mixture. The sample mixture was titrated with a standardized solution of 0.1 M sodium thiosulphate, with starch solution serving as an indicator during the final stages of titration. The volume of 0.1 M thiosulphate that reacted, denoted as 'x', was determined. To establish a baseline, the same titration procedure was repeated without the presence of a sample, and the volume consumed, labeled as 'y', was noted. Finally, the iodine value was determined using equation ii as specified in the study.

$$\text{Iodine value} = 1.269\ (y - x)/m \quad \text{(ii)}$$

in this context, the symbol 'm' represents the mass, measured in grams (g), of the sample used for the evaluation.

2.5.3. Measurement of saponification value

Approximately 2 g of the *Z. mauritiana* seed kernel oil was placed in a 250 mL round bottom flask and connected to a reflux condenser. A small amount of pumice powder and 25 mL of 0.5 methanolic potassium hydroxide were added, and the mixture was heated on a water bath for 30 min. After cooling for a few minutes, the resulting sample solution was titrated against 0.5 M HCl, with the addition of phenolphthalein as an indicator. Additionally, a blank titration was conducted without the inclusion of a sample. The saponification value was determined using equation iii, as described in the study by Pandey et al. [36].

$$\text{Saponification value} = 28.5\ (y - x)/m \quad \text{(iii)}$$

here, the symbol 'm' represents the mass, measured in grams (g), of the sample used for the evaluation. The symbols 'x' and 'y' represent the volumes, measured in milliliters (mL), of HCl consumed by the sample solution and the blank solution, respectively.

2.5.4. Determination of peroxide value

Approximately 5 g of the test sample were introduced into a 250 mL conical flask, which was equipped with a glass stopper. A mixture of glacial acetic acid and chloroform in a ratio of 3:2 was added slowly while continuously shaking the flask. Subsequently, 0.5 mL of saturated potassium iodide was added to the mixture. The resulting sample mixture was thoroughly mixed for precisely 1 min, with frequent shaking. Following this, 20 mL of distilled water was added and the titration of the resulting mixture was performed using a standardized solution of 0.01 M sodium thiosulphate, continuing until the yellow color completely disappeared. Furthermore, 0.5 mL of a 5 % w/v starch solution was added drop by drop, and the titration was resumed with vigorous shaking until the blue color completely vanished (measured as 'x' mL). Similarly, a blank titration was conducted without the presence of a sample (measured as 'y' mL). Finally, the peroxide value was determined using equation iv as outlined in the study by Pandey et al. [36].

$$\text{Peroxide value} = 10\ (x - y)/m \quad \text{(iv)}$$

in this context, the symbol 'm' represents the weight, measured in grams (g), of the sample used for the evaluation.

2.5.5. Calculation of ester value

The ester value was calculated using equation v [36].

$$\text{Ester value} = \text{Saponification value} - \text{Acid value} \quad \text{(v)}$$

2.5.6. Measurement of specific gravity

Initially, the weight of a clean and dried pycnometer with a volume capacity of 50 mL, along with its cap, was measured and recorded as (x). Subsequently, the pycnometer was filled with the sample until it reached the point of overflow. The pycnometer was then sealed with a stopper, and the combined weight of the pycnometer and the sample was noted as (y). After cleaning and drying, the same pycnometer was filled with water, and its weight was recorded as (z). Finally, the specific gravity was calculated using equation vi, as described in the study by Muhammad et al. [38].

$$\text{Specific gravity} = \frac{y - x}{z - x} \quad \text{(vi)}$$

2.5.7. Evaluation of pH and refractive index

To measure the pH, a clean and dry beaker was filled with 100 mL of oil. The pH was measured three times using a digital pH meter. Similarly, the refractive index was determined at room temperature using the Abbey refractometer, following the methods described in

the studies by Pandey et al. [36] and Muhammad et al. [38].

2.5.8. Determination of viscosity

For the measurement of viscosity, a DV-III ULTRA Brookfield viscometer was used. For this, approximately 20 g of the oil was poured in a dry 250 mL beaker. Viscosity was determined by using spindle no. 64. The test sample was subjected to rotation at 10 rpm with 16.1 dyne-cm torque, for 1 min. The operation was carried out at the temperature of 25 °C. The experiment was conducted in triplet and data was presented in centipoises unit (cps) [36,37,39,40].

2.6. Formulation of ketoconazole soap (2 % w/w) by using Z. maurtiana seed oil as a base

The determination of the saponification value confirmed the quantity of potassium hydroxide (KOH) in milligrams (mg) needed to saponify 1 g (g) of the *Z. mauritiana* seed oil sample. The soap formulation involved a basic saponification reaction, where the *Z. mauritiana* seed oil was reacted with KOH to produce soap. In this process, exactly 200 g (g) of seed oil was placed in a beaker and heated in a water bath at 55 °C. Simultaneously, in another beaker, the required amount of KOH to saponify the 400 g of oil (specifically 45.84 g KOH) was dissolved in an appropriate volume of deionized water and allowed to cool to a temperature range of 35–40 °C. Subsequently, the oily and aqueous solutions were combined and stirred continuously for approximately 30 min until the oil completely transformed into a homogeneous solution. The solution was then allowed to cool and filtered using a Buchner funnel and Whatman No. 1 filter paper. Following the filtration process, 300 mL of saturated sodium chloride solution was added to the filtrate in order to precipitate the soap. The obtained precipitate was separated and placed into a clean beaker. To create a 25 g medicated soap, 24.5 g of the soap and 0.5 g of ketoconazole were combined with gentle heating and continuous stirring for a duration of 20 min. Finally, the medicated soap mixture was poured into an appropriate mold and left to solidify for 5–6 h, resulting in the desired hard soap product [36,41,42].

2.7. Quality control evaluation of the 2 % w/w ketoconazole soap

2.7.1. pH measurement

A digital pH meter was employed to determine the pH of a 100 mL aqueous solution with a concentration of 10 % w/v [33,40].

2.7.2. Evaluation of foam retention time and foaming index

Exactly 1 g of soap was dissolved in approximately 50 mL of water using a 100 mL graduated measuring cylinder. The mixture was then vigorously shaken for a period of 2–3 min and left undisturbed for 10 min. Afterward, the height of the resulting foam was measured using a measuring scale. The foam retention time was recorded as the duration it took for the soap solution to completely eliminate foam [33].

2.7.3. Quantification of ketoconazole content

At first, 5 soap pieces measuring 4.75 cm in length, 2.20 cm in width, 0.79 cm in height, and weighing 25 g were ground to a smaller size using a mortar and pestle. Following that, an amount of sample equivalent to 50 mg of ketoconazole (2.5 g) was weighed and transferred to a 100 mL dry volumetric flask. The sample was then dissolved in methanol. To ensure complete solubilization of ketoconazole, the sample solution was subjected to sonication for duration of 30 min. After filtration, the sample was diluted to obtain a final solution with a concentration of 25 parts per million (ppm). Likewise, a standard solution was prepared by dissolving 50 mg of ketoconazole standard in a 100 mL volumetric flask using methanol. Through appropriate dilution, a standard ketoconazole solution with a concentration of 25 ppm was prepared. Subsequently, the prepared final solutions were analyzed at a wavelength of 225 nm using a UV–Visible spectrophotometer [43,44]. The drug content was calculated using the pharmacopeial method, employing equation vii as a calculation tool [40,43].

$$\frac{\text{Absorbance of sp}}{\text{Absorbance of std}} \times \frac{\text{Wstd}}{100} \times \frac{5}{100} \times \frac{100}{\text{Wsp}} \times \frac{100}{5} \times \frac{\text{Purity of std}}{100} \times \frac{(100 - \text{LOD})}{100} \times 100 \quad \text{(vii)}$$

in the context of the given abbreviations, the abbreviations “sp” represents the sample, “std” represents the standard, “Wsp” denotes the weight of the sample soap taken (2.5 g), “Wstd” represents the weight of the standard drug taken (50 mg), and “LOD” stands for loss on drying for the standard.

2.7.4. Measurement of total moisture content

Initially, the weight of a dried and clean crucible (referred to as “x") was measured and tarred. Subsequently, approximately 6 g of soap (referred to as “y") was weighed and placed in the crucible. The content of the crucible was then heated at a temperature of 101 °C for duration of 2 h. Afterward, the crucible was placed inside a desiccator to cool, and the weight of the crucible containing the sample (referred to as “z") was recorded. The moisture content was determined using equation viii [33].

$$\text{Moisture content} = \frac{y - (z - x)}{y} \quad X\ 100 \quad \text{(viii)}$$

2.7.5. Calculation of total fatty matter (TFM) present in ketoconazole soap

Approximately 10 g of soap (denoted as "m") was dissolved in 150 mL of distilled water. The solution was then subjected to heat while treating it with 20 % sulfuric acid until it became clear. After a few minutes, a film of fatty acid appeared on the surface. Subsequently, approximately 7 g of wax (denoted as "x") was added to the solution and heated again. Upon cooling, a cake formed (as shown in Fig. 2C), which was then separated and weighed (referred to as "a"). Finally, the Total Fatty Matter (TFM) was calculated using equation ix as described by Sindhu et al. [33].

$$\%\ \text{Total Fatty Matter} = \frac{(a - x)}{m} \quad X\ 100 \tag{ix}$$

2.8. Data analysis

All the values were expressed as mean ± SD and each experiment was replicated three times. Microsoft excel 2021 was used for the analysis of data.

3. Results and discussion

3.1. Extractive yield of Z. mauritiana seed kernel extract

The extractive yield of hexane extract from *Z. mauritiana* seed was determined to be 29.24 %. Comparatively, previous studies have reported that extractive yields of commercial oils, including olive oil (14 %) [45], coconut oil (61.3 %) [46], *Princepia utilis* seed oil (26.43 %) [36], and palm oil (59.32 %) [47]. These findings indicate that the oil yield from *Z. mauritiana* seed kernel is of moderate quantity.

3.2. Analysis of the physicochemical parameters of Z. mauritiana seed kernel oil

Various physicochemical properties of *Z. mauritiana* seed kernel oil were examined to assess its quality and state (Table 1). The table presents a comparison of the physicochemical parameters of the examined oil with olive oil, a widely used vegetable oil [45], and *Diploknema butyracea* seed fat, also known as chyuri fat [40]. The oil had an orange-yellow color, as depicted in Fig. 1E.

As shown in Table 1, the acid value of *Z. mauritiana* seed kernel oil is lower than chyuri fat and similar to olive oil. This indicates the sample has a low amount of free fatty acids. The acid value signifies oil's susceptibility to degradation from a range of factors, such as temperature, light, and lipase enzyme activity [40,51,52]. Additionally, it provides information about the suitability of the oil for edible purposes. Oils or fats with an acid value exceeding 4 mg/g are considered unhealthy for consumption [53].

The iodine value serves as a scientific measure of the oil's unsaturation level. A higher degree of unsaturation (especially double bonds) indicates a greater likelihood of oxidative degradation. Therefore, the iodine value consistently provides a quantitative indication of the oil sample's susceptibility to oxidative decomposition [54]. The lower iodine value of *Z. mauritiana* seed kernel oil (11.19 g I_2/100 g) suggested that the amount of unsaturated fatty acids is relatively low in this oil sample. A previous research work, which supports our finding, claims that the saturated to unsaturated ratio of fatty acids, obtained by dividing the total percentage of saturated fatty acids by the percentage of unsaturated fatty acids, was determined to be 5.3 [23]. This ratio is comparable to that of olive oil, which has a similar unsaturated to saturated fatty acids ratio of 4.7 [23]. Major saponifiable fatty acids identified in *Z. mauritiana* seed kernel oil are the ester forms of saturated docosanoic acid (1.5 %), eicosanoic acid (2.1 %), octadecanoic acid (6.9 %), hexadecanoic acid (7.2 %), and octadecenoic (55.2 %). Also, it contains diunsaturated 9,12-octadecenoic acid (25.3 %) and monounsaturated

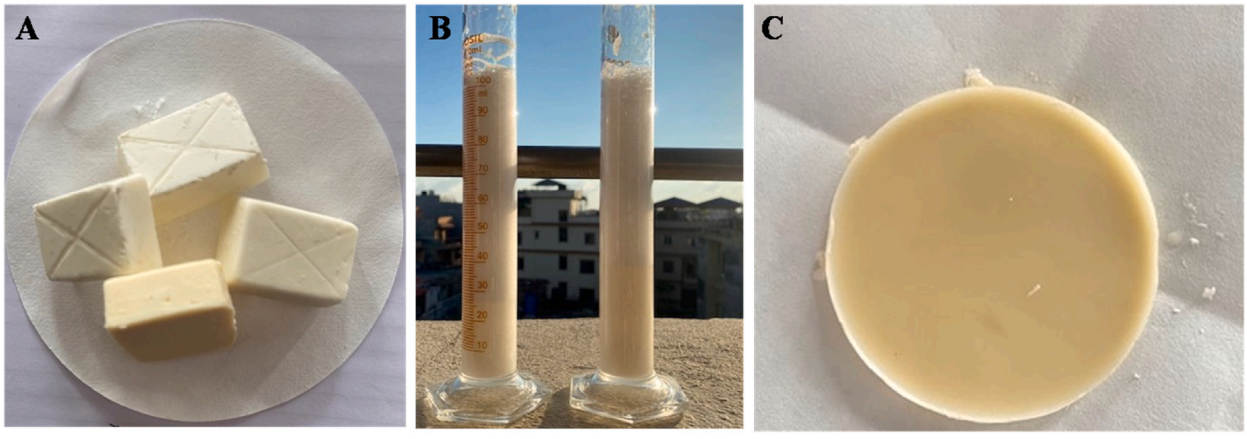
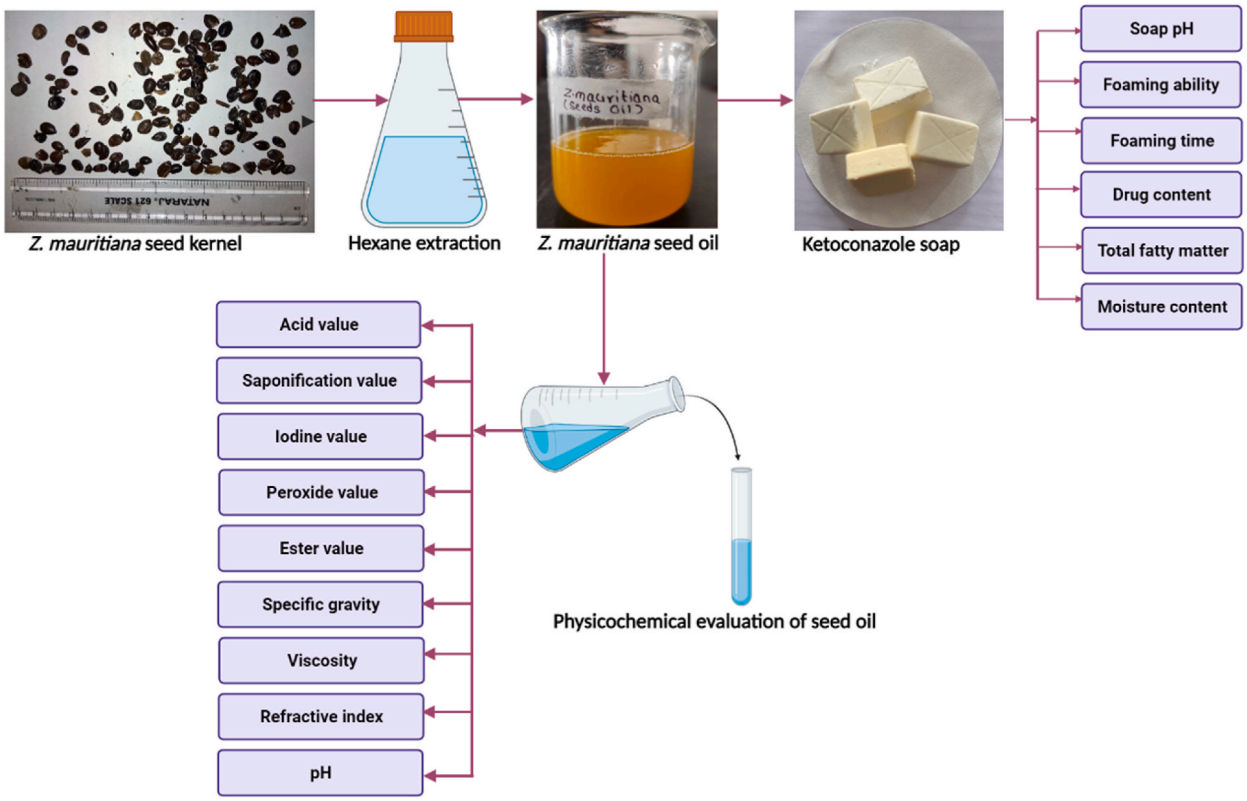


Fig. 2. Photographs of 2 % w/w ketoconazole soap formulated by using *Z. mauritiana* seed kernel oil as a base (A), foam-forming ability of newly formulated soap (B), and total fatty matter present in the newly formulated soap (C).



Flowchart 1. Summarizing the overall steps of investigations conducted for *Z. mauritiana* seed kernel.

Table 1
Results depicting physicochemical parameters of the *Z. mauritiana* seed kernel oil compared with a commercial vegetable oil (olive oil) and *D. butyracea* seed kernel fat (chyuri fat).

Physicochemical Parameters	Samples		
	Z. mauritiana seed kernel oil	Olive Oil	Chyuri fat
Saponification value (mg KOH/g)	229.18 ± 3.56	186.33 [38]	225.05 ± 1.98 [40]
Peroxide value (Meq KOH/g)	4.15 ± 0.23	17.00 [38]	3.14 ± 0.17 [40]
Iodine value (g I_2/100 g)	11.19 ± 0.72	76.0 [38]	36.26 ± 0.89 [40]
Acid value (mg KOH/g)	4.71 ± 0.23	4.53 [38]	61.86 ± 1.16 [40]
Ester value (mg KOH/g)	224.47 ± 3.45	181.77 [38]	163.19 ± 2.75 [40]
Specific gravity (g/mL)	0.912 ± 0.019	0.91 [48]	0.92 [40]
pH	5.93 ± 0.15		
Viscosity(centipoise)	89 ± 0.15	62 [48]	310 ± 0.61 [40]
Refractive index	1.448 ± 0.01	1.471 [49]	1.45–1.46 [40][50]

11-eicosenoic acid (1.9 %) [21,23]. Oils with an iodine value below 100 are categorized as non-drying oils, while those with values ranging from 100 to 130 are referred to as semi-drying oils. On the other hand, oils having iodine values above 130 are classified as drying oils [55]. Scientific studies have suggested non-drying oils are chemically stable for various industrial applications [40,56–58], including the cosmetic, margarine, bakery, and drug industries [56,57]. According to American Society for Testing and Materials standard (D6751), oil samples having iodine value less than 115 are considered suitable for biodiesel and lubricant applications [58]. Moreover, non-drying oils exhibit water resistance and excellent color retention characteristics. As a result, they are utilized in the paint industry for producing paints and varnishes [59].

A low iodine value of this examined oil sample suggests that it is resistant to rancidity and oxidation, when used in the production of related products [57,60]. Furthermore, the iodine value of *Z. mauritiana* seed kernel oil closely resembles that of commonly consumed coconut oil [61,62]. Also, saturated fatty acids present in this sample are of high nutritional value [21].

The peroxide value of an oil or fat is a useful parameter for assessing its susceptibility to rancidity and auto-oxidation [57]. When the peroxide value exceeds 10 meq/kg, there is potential for auto-oxidation due to the presence of peroxidase and lipoxygenase enzymes, as well as moisture content or trace elements [63]. In the case of *Z. mauritiana* seed kernel oil, its lower peroxide value indicates a greater resistance to peroxidation [64] and so, it may be less prone to oxidative degradation induced rancidity. Typically, unrefined

oils tend to have higher peroxide values compared to refined oils [64]. Consequently, conducting a comparative study on various extraction techniques for *Z. mauritiana* seed kernel oil could serve as a valuable scientific investigation aimed at further enhancing the peroxide value of this oil sample.

The saponification value provides insights into the molecular weight and type of fatty acids present in a sample. A low saponification value indicates a larger proportion of high molecular weight fatty acids in the sample. For *Z. mauritiana* seed kernel oil, the high saponification value and ester value indicated a larger quantity of low molecular weight short-chain fatty acids and easily saponifiable triglycerides, respectively, in the oil composition. Oils with higher saponification values, such as *Z. mauritiana* seed kernel oil (229.64), are considered favorable choices for industrial, pharmaceutical, and cosmetic applications [63,65]. Comparatively, the saponification value of the examined oil sample is higher than that of commonly used oils like palm oil (192.64) [66], olive oil (186.33) [40], and sunflower oil (182.23) [67]. This suggests that *Z. mauritiana* seed kernel oil may possess desirable characteristics for various commercial applications in these industries.

The measurement of specific gravity is a valuable quality control parameter when analyzing oil samples, as it helps detect any potential adulteration by cheaper oils. Changes in specific gravity can indicate variations in the composition and quality of the oil. In our research, the specific gravity of the oil sample was determined to be 0.912 g/mL, which closely resembled the specific gravity of olive oil (0.91 g/mL) [68]. This similarity in specific gravity suggests a comparable quality between the two samples. The refractive index of oil is a useful parameter that can indicate its susceptibility to rancidity. Higher refractive index values are associated with a greater likelihood of rancidity [69]. In our study, the refractive index of *Z. mauritiana* seed kernel was determined to be 1.448, which is better than the value of olive oil (1.471) reported by Muhammad et al. [40]. Additionally, oils with refractive index values ranging from 1.475 to 1.485 are classified as drying oils [70]. Based on the observed refractive index, *Z. mauritiana* seed kernel oil falls under the category of non-drying oils. Non-drying oils exhibit higher chemical stability and do not form a layer when exposed to air, unlike drying oils [71]. In our study, *Z. mauritiana* seed kernel oil demonstrated a higher viscosity (89 cp) compared to other commonly used oils, such as pumpkin oil (48.09 cp) [72], coconut oil (29 cp) [73], and it is comparable to the viscosity of olive oil (84 cp) [68]. Oils with higher viscosity are well-suited for lubrication purposes [68].

3.3. Evaluation of the 2 % w/w ketoconazole soap

After confirming its physicochemical suitability as a soap base, 2 % w/w ketoconazole soap (Fig. 2A) was formulated using the basic saponification method, with a total weight of 25 g, for each soap. Quality control parameters, namely, total drug content, total fatty matter (TFM), moisture content, total foam forming ability, and pH, were assessed, as outlined in Table 2. It was observed that all measured parameters fell within the limits specified by pharmacopeia standards and existing literature.

3.3.1. Evaluation of soap pH

The pH of the soap was determined to be within the optimal range for application on human skin. Previous studies have indicated that the normal pH condition for human skin typically falls within the range of 5.5–7.5. Therefore, the pH of the soap formulation aligns with the recommended pH range for maintaining healthy skin [33].

3.3.2. Evaluation of foam retention time and foaming index

The formulation exhibited a foaming index of 18.56 ± 1.60 cm, indicating its satisfactory lather-producing ability. Furthermore, the soap demonstrated a foam retention time of 17.21 ± 1.57 min, indicating good stability of the lather. These findings were consistent with previous studies [74,75]. Fig. 2B visually represents the foam-forming ability displayed by the newly formulated 2 % w/w ketoconazole soap.

3.3.3. Determination of total moisture content in soap

Assessing the total moisture content is a simple method to estimate the shelf life of soap. When soap contains excessive moisture, even under normal storage conditions, the water present can react with unsaponified oil or fat, leading to the liberation of glycerol and free fatty acids through a hydrolysis reaction. According to the Encyclopedia of Industrial Chemical Analysis, the typical moisture content range for soap is less than 15 % [74]. Hence, the developed ketoconazole soap displayed favorable outcomes in terms of its moisture content, measuring at 9.14 %.

Table 2
Results for the different quality control parameters of the 2 % w/w ketoconazole soap formulated by using *Z. mauritiana* seed kernel oil as a base.

Evaluation	Result	Acceptable range
Soap Assay (%)	99.49 ± 1.38	95–105
Total fatty matter (%)	71.13 ± 1.07	70-75 = grade 2 ≥76 = grade 1
Total moisture content (%)	9.14 ± 0.76	0–15
Foam retention time (minutes)	17.21 ± 1.57	≥15
Foam forming ability (cm)	18.56 ± 1.60	≥15
pH	7.16 ± 0.47	5.5–7.5

3.3.4. Evaluation of total fatty matter (TFM)

The assessment of Total Fatty Matter (TFM) (Fig. 2C) plays a vital role in ensuring soap quality, as it directly affects its moisturizing properties on the skin. A lower TFM value in soap is associated with a higher likelihood of causing dryness to the skin [74–76]. A higher total fatty matter content is desired in soap as it contributes to its cleansing and moisturizing properties. Soaps with a higher proportion of saponifiable fatty acids tend to have a richer and more nourishing lather, making them more effective for cleansing and moisturizing the skin [77]. Soaps are categorized into different grades based on their Total Fatty Matter (TFM) values. Grade-1 soaps have a TFM above 76 %, while grade-2 soaps fall within the TFM range of 70–75 % [76]. The *Z. mauritiana* seed kernel oil-based soap in this study had a TFM of 71.13 %, indicating that it belongs to grade-2 and ensures good cleaning and leathering properties [76]. This TFM value is comparable to commercially available soaps such as Lifebuoy (63.4 %) [78], Dettol (65.4 %) [78], Liril lime and tea tree oil soap (70.4 %),[79], and Lux beauty (60.7 %) [79].

3.3.5. Determination of drug content in the soap

The assay conducted as per the pharmacopeial standards confirmed that the ketoconazole content in the soap fell within the acceptable range of 95–105 % as specified by the US Pharmacopeia 2020 (USP 2A; pp. 2506–2507). Thus, using *Z. mauritiana* seed kernel oil as a base for manufacturing a medicated soap could serve as a viable alternative to synthetic bases. Nevertheless, thorough investigations encompassing multiple aspects including real-time stability, accelerated stability, drug release profiles, and skin irritation tests are crucial to determine the commercial viability and acceptability of the soap.

4. Conclusions

In summary, this study investigated various physicochemical parameters of *Z. mauritiana* seed kernel oil. Moreover, we were able to formulate ketoconazole soaps by utilizing *Z. mauritiana* seed kernel oil as a natural base. Desirable parameters of saponification value (229.18 mg KOH/g), ester value (224.47 mg KOH/g), iodine value (11.19 mg KOH/g), and peroxide value (4.15milliequivalents KOH/g) suggest that this oil is chemically stable and well-suited for cosmetic, pharmaceutical, and industrial applications. Additionally, the low iodine value signified that the oil is suitable for biodiesel, lubricants, painting, and varnishing purposes. Similarly, satisfactory results of refractive index (1.448), specific gravity (0.912 g/mL), and viscosity (89.15 cp) further confirmed the physical and chemical stability of *Z. mauritiana* seed kernel oil for commercial applications. The low acid value, 4.71 (similar to olive oil), indicates that it can be utilized as a commercial cooking oil. The peroxide value of *Z. mauritiana* seed kernel oil can be further enhanced through purification methods. Moreover, the quality control examination of *Z. mauritiana* seed kernel oil derived ketoconazole soap revealed that its drug content (99.49 %), total fatty matter (71.13), foam retention time (17.12 min), foam height (18.26 cm), moisture content (9.14 % 0, and pH (7.16) are within the acceptable range. These results were comparable to grade-2 commercial soaps available in the market.

Antifungal soaps formulated solely with natural oil may provide several additional benefits, such as moisturization, skin soothing effects, hair and skin rejuvenation, antioxidant properties, antiseptic effects, and the absence of carcinogenic risk associated with synthetic soaps. However, further advanced research is crucial to enable the commercialization of *Z. mauritiana* seed kernel oils as a viable alternative to synthetic oils and fats. Indeed, the utilization of *Z. mauritiana* seed kernel oils in cosmetic production not only creates greater value to these otherwise discarded seeds but also provides a viable income-generating option for people living in rural areas.

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Data availability statement

Data will be made available on request.

CRediT authorship contribution statement

Jitendra Pandey: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Project administration, Resources, Supervision, Validation, Writing – original draft, Writing – review & editing. **Sushan Gaire:** Investigation, Resources. **Kamal Sharma:** Investigation, Resources. **Dila Pun:** Investigation, Resources. **Anjali Gyawali:** Investigation, Resources. **Gopal Lamichhane:** Writing – original draft. **David Budean:** Writing – review & editing. **Hari Prasad Devkota:** Writing – review & editing.

Declaration of competing interest

Not applicable.

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