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Research article

Sterculia foetida de-oiled seed cake proximate, mineral and secondary metabolite composition vary across different landscapes of southern Gujarat, India

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Abstract

We analysed variation in *Sterculia foetida* de-oiled seed cake (SFSC) proximate, minerals, secondary metabolites and volatile phytochemical composition across different landscapes of Gujarat, India. The sampling was done from eight landscapes falling in three districts, namely Narmada (lies in Satpura mountain ranges and Narmada river basin), Navsari and Valsad (Arabian Sea coast). Our findings revealed that de-oiled seed cake proximate attributes, namely crude protein (15.04), crude fiber (12.61), ether extract (6.38), acid insoluble ash (3.40), nitrogen-free extract (62.59) and total ash (3.31) varied significantly ($p \leq 0.05$), except organic matter (96.69), among the landscapes. Macro minerals, namely nitrogen, phosphorus, calcium and magnesium, with overall average percentage of 2.41, 0.43, 0.14 and 0.91%, respectively and micro minerals (ppm), namely zinc, iron, copper and manganese composition exhibiting 218.44, 547.80, 9.44 and 156.24 ppm content, respectively, in SFSC also varied significantly. Findings also revealed that the percentage of secondary metabolites, namely total phenols (1.70) and tannins (1.10), varied significantly, excluding saponins content (0.78). Seed cake gas chromatography mass spectrometry analysis revealed only two phytosterols, namely stigmasterol and β-sitosterol (51.73 and 48.21%, respectively). The study divulged that SFSC is a rich source of protein, contains good fat energy and soluble carbohydrates. Further, macro and micro mineral contents of seed cake were well above the critical level and total phenols and tannin contents were well below the toxic limit. Stigmasterol and β-sitosterol, detected in seed cake, exhibit various beneficial properties. Hence, SFSC could be utilized as an alternate feed source for livestock.

Keywords: De-oiled seed cake, Livestock feed, Metabolites, Minerals, Phytosterols, Proximate principles, Wild almond

Introduction

India, with only 2.29% of the world's land area, maintains more than 16% of livestock (more than 536 million heads), creating considerable pressure on land, water and other natural resources. Grasses, grazing on common lands, crop residues and agricultural byproducts, cultivated fodder, edible weeds, top feed resources from cultivated and uncultivated lands and agro-industrial byproducts are the major feed and fodder resources to cater to such a huge livestock. Feed cost accounts for about 70 to 75% of the total cost of livestock production, particularly in milch animals. India requires more than 85.78 million tons of concentrates; however, the estimated annual availability of total concentrate feed is only 61 million tons with a deficit of approximately 24.78 million tons or 28.9% of the demand (Anon, 2018; Roy *et al*., 2019). Concentrate feed consists of oilseed cakes, crushed pulses, grains, wheat and rice brans, mineral mixtures, *etc*. The Indian Council of Agricultural Research-National Institute of Animal Nutrition and Physiology estimated demand for concentrates at 56.2 million tons for 2011-12, of which grains constituted 30%. Accordingly, 16.9 million tons of grain were used as feed. Further, feed grain comprised 53% of maize, 5% of nutri-cereals, and 2% of rice, wheat and pulses (Anon, 2024).

The global and Indian scenario indicate that the oilseed cake demand significantly surpasses its supply, principally due to the country's large edible oil import dependence, leading to a high demand for oilseed cake as a byproduct of oil extraction, mostly used as animal feed,

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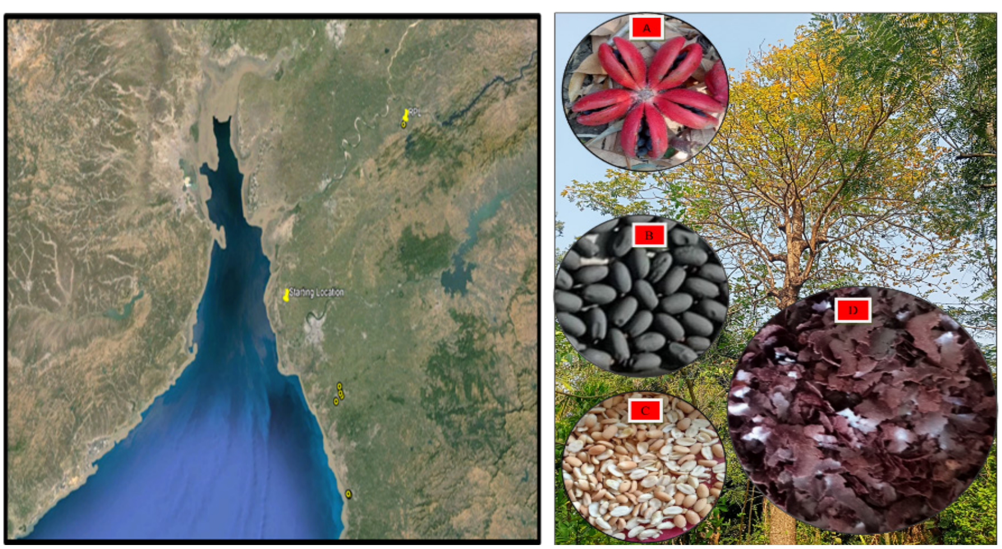
while domestic production struggles to meet this demand due to low oilseed yields compared to other producing nations. The gap is further augmented by swelling livestock populations and growing awareness of protein-rich feed stuff. In summary, the foremost reasons for these bottlenecks include high import dependency, low domestic production, growing demand for protein feed, price variability, *etc*. Oilseed cakes are rich in protein and contain various antioxidants, fibers, and vitamins and therefore, are preferred as supplement feed for animals and sometimes beneficial for humans. In addition, major bakery products are being produced with sunflower oilseed cakes (Sunil *et al.,* 2015). Oilseed cakes are also used for the production of bioenergy, biopolymers, pesticides and bioelectricity (Naik *et al.*, 2018). Most of the oilseed cakes are currently used as animal feed or plant fertilizers. It represents an attractive feedstock for the development of a new biorefinery. Protein-added-value products can also be generated by oilseed cake. The proteins from oilseed cake could be turned into renewable sources for producing bioplastics. Oilseeds are cultivated over 2836 million hectares annually across the world and 29 million hectares in India (Yang *et al.*, 2019). This indicates that there is a need to explore more natural resources of oilseed cake and concentrates to bridge the gap between demand and supply and look for improvement opportunities in these existing resources/agricultural or industrial byproducts (Thakur *et al.,* 2015; Sukhadiya *et al.,* 2019; Jangir *et al.,* 2020).

There are numerous plant species including perennial trees which have been evaluated for edible seed oil and after oil extraction cakes are used as a concentrate for livestock feeding or other purposes like manuring (Konuskan *et al.,* 2017; Hegde *et al.,* 2019). In the current scenario, perennial species like Jatropha (*Jatropha curcas*), Karanja (*Pongamia pinnata*) are used for biofuel production (Hashemi and Mirmohamadsadeghi, 2020) and oil seed cake of *Madhuca latifolia* has been considered are nutrient rich feed resource (Hegde *et al.,* 2024). One such potential species could be *Sterculia foetida* L., commonly known as Hazel sterculia and wild almond. It is found from eastern tropical Africa to north Australia, through Malaysia, Burma, Bangladesh, India, Sri Lanka and Malacca (Mujumdar *et al.,* 2000). The generic name is based on the Latin word 'stercus', meaning 'manure', which refers to the smell of the flowers and leaves of some species. Fruits ripen in February, nearly 11 months after the 1st appearance of the flowers. It produces approximately 200 to 350 kg of seed per year (Orwa *et al.,* 2009). *S. foetida* seeds are safe to eat and contain 30 to 36% fat and 11.4% protein. Seeds yield a considerable amount of oil (58.7 g/100 g) and have been found to be a good biofuel (Bose *et al.,* 2021). Due to low moisture content, the seeds of *S. foetida* have a prolonged shelf life. The seeds contain significant amounts of crude oil, protein, lipids and minerals. *S. foetida* oil is a good source of carbohydrates, protein, lipids, and macro and micronutrients (Bose *et al.,* 2021). There is a dearth of information on its nutritive value. Therefore, the present study was intended to find out *S. foetida* seed cake proximate, minerals, secondary metabolites and volatile phytochemical composition across landscapes of south Gujarat extending from the Arabian coast to Satpura mountain ranges, India.



a b

Fig 1. a) Geographic location of *S. foetida* landscapes and b) *S. foetida* tree in arboreal landscape [A) fruit, B) seed, C) seed kernel and D) de-oiled seed cake]

Materials and Methods

***Study area description and fruit collection*:** *S. foetida* fruits were collected from distantly located eight different landscapes falling in three districts, namely Narmada (falls in Satpura ranges and Narmada River basin), Navsari and Valsad (coastal parts of Arabian Sea) of south Gujarat. The detailed geographical location of landscapes and growth attributes of selected trees were recorded (Table 1; Fig 1). Landscape Rajpipla (RPL)-L_1 falls under the Narmada district and is characterized by prominent ridge and valley topography oriented in the East-West direction. The soil type of this region is black cotton and stony. The region experiences average annual rainfall of 924.8 mm and annual maximum and minimum temperatures of this region is 34.7 and 20.7°C, respectively. This landscape is arboreal in nature, *i.e., S. foetida* trees were growing in association with other tree species.

Navsari (NVS), L_1 to L_4 landscapes fall under the Navsari district, situated near the Arabian Sea coast. Soils of this region are black rock outcrops, shallow black, brown and alluvial soils of recent origin. The region contains Orthents-Ochrepts formed and Orthents-Rock-Outcrop types of soils. The average annual rainfall in the district ranges from 1500 to 3000 mm. The average annual temperature is about 26ºC and the maximum temperature may shoot up to 46ºC and winter temperatures are 22ºC. The selected trees of these locations represent Agroforestry (Navsari-L_2) and city landscape, *i.e.*, roadside plantation (Navsari-L_3, Navsari-L_4 and Navsari-L_5). Valsad (VLD; L_6–L_8), locations fall under city landscapes situated along the Arabian Sea coast. Geologically, it is a northern extension of the Deccan Plateau of Central India, belonging to the late Cretaceous-early Eocene age and followed by Quaternary sediments. The region has black cotton soils. The region experiences maximum and minimum temperatures of 41.2 and 9.9°C, respectively. The trees selected in these landscapes are avenue plantation (Valsad-L_6 and Valsad-L_7) and arboreal (Valsad-L_8), thus representing the city and arboreal landscape, respectively.

***Seed collection and seed cake proximate composition*:** From each landscape, five trees were marked and fruits were collected in bulk (January 2024) from each landscape and brought to the laboratory and seeds were extracted. After oil extraction in expeller seed cake was collected in bulk from each landscape. Seed cake proximate parameters namely moisture content (MC%), dry matter (DM%), crude protein (CP%), crude fibre (CF%), ether extract (EE%), total ash content (TAC%), organic matter (OM%), acid insoluble ash (AIA%) and nitrogen-free extract (NFE%) of seed kernel were determined as described by AOAC (2016).

Seed cake micro and macro minerals and secondary metabolites: All the macro (nitrogen, calcium, phosphorus and magnesium) and micro minerals (iron, zinc, copper and manganese) were estimated following standard procedures given by AOAC (2016). Total phenolic, tannin and saponin contents were determined by the Folin-Ciocalteu colorimetric method (Malick and Singh, 1980), the Folin-Denis method (Schanderl, 1970) and the method of Obadoni and Ochuko (2002), respectively. Volatile phytochemicals were detected through GC-MS, partly following Sukhadiya *et al.* (2021).

Statistical analysis: All respective data sets were analyzed as per standard statistical procedure using a complete randomized design (CRD). F-test was done and ANOVA was constructed with following model Yij = μ + ti + eij, where Yij = an observation, μ = overall mean effect, ti = true effect of the (Ti) treatment, eij = error term of the (Yj) unit receiving (i) treatment, i = a particular treatment, j = a particular replicate as described by Snedecor and Cochran (1980). Treatment means were compared at

Table 1. Geographical location and average growth attributes of selected *S. foetida* trees in different landscapes of south Gujarat India

Landscapes (L)	Altitude (m)	Latitude (N)	Longitude (E)	Tree height (m)	DBH (cm)
RPL-A-L_1	67	21°85′81′′	73°51′87′′	22.50	45.06
NVS-AF-1-L_2	33	20°92′23′′	72°90′52′′	21.20	68.30
NVS-RP-2-L_3	34	20°97′03′′	72°93′42′′	10.60	32.80
NVS-RP-3-L_4	33	20°93′67′′	72°94′01′′	15.30	49.90
NVS-RP-4-L_5	39	20°94′63′′	72°93′49′′	18.80	51.70
VLD-RP-1-L_6	30	20°60′42′′	72°91′38′′	12.40	46.60
VLD-RP-2-L_7	33	20°60′82′′	72°90′83′′	8.60	34.19
VLD-A-3-L_8	32	20°60′46′′	72°91′37′′	21.50	36.00

DBH: Diameter at breast height; RPL: Rajpipla; NVS: Navsari; VLD: Valsad; A: Arboreal; AF: Agroforestry; RP: Roadside plantation

P≤0.05. Further, Duncan's multiple range test (DMRT) was adopted to compare the sets of means of each factor (Sheoran *et al.*, 1998).

Results and Discussion

Proximate composition: Perusal of results deduced that percentage of the proximate attributes namely crude protein (CP), crude fiber (CF), ether extract (EE), acid insoluble ash (AIA), nitrogen free extract (NFE) and total ash (TA) in *S. foetida* seed cake (SFSC) varied significantly [($p \leq 0.05$); except organic matter (OM)] among the studied 8 landscapes (Table 2; Fig 2). Maximum CP (16.74%), EE (8.13%), AIA (3.97%) and TA percentage (3.99%) were in VLD-3-L_8 landscape seed cake. Maximum CF (13.60%), NFE (64.90%) and OM (97.05%) were found in VLD-2-L_7, RPL-L_1 and NVS-3-L_4 landscape seed cake. Minimum CP (13.27%) and AIA (2.90%) were recorded in RPL-L_1 and VLD-3- L_8 landscape seed cake showed minimum NFE (58.56%). Lowest CF (11.29%), EE (5.28%) and TA (2.95%) were in the seed cake of NVS-4-L_5, NVS-1-L_2 and NVS-3-L_4 landscapes. Overall, in all the landscapes, an average of 15.04, 12.61, 6.38, 3.40, 62.59, 3.31 and 96.69% CP, CF, EE, AIA, NFE, TA and OM, respectively, were recorded in seed cakes.

The results of the present study indicated that, except for OM, all other proximate constituents of SFSC varied significantly ($p \leq 0.05$) across 8 landscapes. Findings deduced that *S. foetida* crude protein (CP) content ranged from 13.27 to 16.73%, which was higher than reported earlier elsewhere (Bera *et al.*, 1995) in this species. The commonly used cottonseed meal contains about 21% CP (Gul *et al.*, 2010), whereas olive cake was reported with 6.51%. SFSC can be considered as a rich source of CP as described for most of the seed cakes being fed to small ruminants and livestock (Anon, 2012).

In the present investigation, SFSC crude fiber content ranged from 11.29 to 13.60%, which was lower than that reported by Bera *et al.* (1995) in this species. However, present CF values were in agreement with those of *Azadirachta indica* seed cake (Gowda and Sastry, 2000). Even the CF content in cottonseed meal and olive cake was reported to be higher (Gul *et al.*, 2010). The present study expressed that EE content in seed cake ranged from 5.28 to 8.13%, which was in line with the range reported earlier (Bera *et al.*, 1995) in this species. Further, EE reported in cotton seed meal and olive cake (Gul *et al.*, 2010) and *A. indica* seed cake (Gowda and Sastry, 2000) was also comparable with *S. foetida* EE content in the present findings. A slightly higher EE content in *Calophyllum inophyllum* seed oil cake was reported by Rath *et al.* (2017). Thus, in general, SFSC possessed higher EE compared to other seed cakes (Abdeltawab and Khattab, 2018), which indicated that SFSC contained fairly good fat energy (Anon, 2012).

Our results indicated that NFE content in SFSC ranged from 58.56 to 64.90% and was in line with values reported by Bera *et al.* (1995) in this species and that of *Calophyllum inophyllum* seed cake (Rath *et al.*, 2017). Cottonseed meal and olive cake NFE content were recorded with lower NFE content (Gul *et al.*, 2010) as compared to that reported in *S. foetida* in present findings. The results indicated that total ash (TA) content ranged from 2.95 to 3.99% in seed cake, which was lower than that obtained by Bera *et al.* (1995) in the same species. However, *S. foetida* cake TA content was at the same level as *Pongamia glabra* cake and cottonseed meal and olive cake (Vinay and Kanya, 2008; Gul *et al.*, 2010). Organic matter content in seed cakes ranged from 96.01 to 97.05% and was slightly higher

Table 2. Variation in *S. foetida* de-oiled seed cake proximate composition among different landscapes of south Gujarat India

Landscape (L)	CP (%)	CF (%)	EE (%)	AIA (%)	NFE (%)
RPL-A-L_1	13.27^{c}	12.95^{a}	5.46^{cd}	2.90^{c}	64.90^{a}
NVS-AF-1-L_2	14.76^{bc}	12.64^{ab}	5.28^{d}	3.67^{ab}	64.11^{ab}
NVS-RP-2-L_3	14.84^{bc}	12.52^{ab}	5.46^{cd}	3.20^{bc}	64.12^{ab}
NVS-RP-3-L_4	14.60^{bc}	12.81^{ab}	6.68^{bc}	3.48^{abc}	62.96^{b}
NVS-RP-4-L_5	15.72^{ab}	11.29^{b}	6.20^{cd}	3.07^{bc}	64.11^{ab}
VLD-RP-1-L_6	14.40^{bc}	12.85^{ab}	6.14 ^{cd}	3.75^{ab}	63.07^{ab}
VLD-RP-2-L_7	15.99^{ab}	13.60^{a}	7.68^{ab}	3.20^{bc}	58.91^{c}
VLD-A-3-L_8	16.74^{a}	12.25^{ab}	8.13^{a}	3.97^{a}	58.56^{c}
Mean	15.04	12.61	6.38	3.40	62.59
SEM (±)	0.56	0.71	0.44	0.14	0.98

CP: Crude protein; CF: Crude fibre; EE: Ether extract; AIA: Acid insoluble ash; NFE: Nitrogen free extract; RPL: Rajpipla; NVS: Navsari; VLD: Valsad; A: Arboreal; AF: Agroforestry; RP: Roadside plantation; Letter different in same vertical column are significantly different according to Duncan's multiple range test ($p \leq 0.05$)

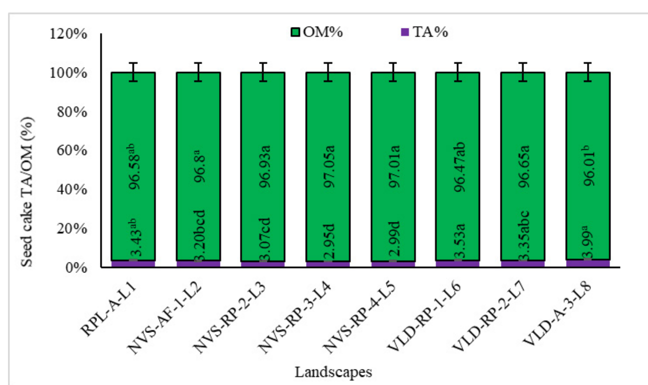


Fig 2. Variation in *S. foetida* de-oiled seed cake total ash (TA%; SEM=±0.09) and organic matter (OM%; SEM=±0.09) among different landscapes of south Gujarat, India; L: Landscape; RPL: Rajpipla; NVS: Navsari; VLD: Valsad; Letters different in vertical bars are significantly different according to Duncan's multiple range test ($p \leq 0.05$)

than reported in cottonseed meal and olive cake (Gul *et al.*, 2010). It was reported that de-oiled seed cake is rich in protein (28–89%) that can be used as animal and fish food supplements (Oliveira *et al.*, 2000; Shamsundar and Paramjyothi, 2010).

Macro and micro mineral composition: The percentage of macro mineral namely nitrogen (N), phosphorus (P), calcium (Ca) and magnesium (Mg) and micro minerals (ppm) namely zinc (Zn), iron (Fe), copper (Cu) and manganese (Mn) composition in SFSC varied significantly (*p ≤0.05*) among the different landscapes (Fig 3).

Amongst macro minerals, maximum N (2.73%), P (0.46%), Ca (0.18%) and Mg content (1.05%) were recorded in seed cake of NVS-4-L_5, VLD-3-L_8, NVS-3-L_4, and LD-1-L_6 landscapes, respectively (Fig 3A). N (1.86%), P (0.39%) and Mg (0.73%) contents were minimum in seed cake from RPL-L_1 landscape and minimum Ca content (0.10%) in VLD-2-L_7 landscape. Overall, average N, P, Ca, and Mg contents were 2.41, 0.43, 0.14 and 0.91%, respectively in all landscapes.

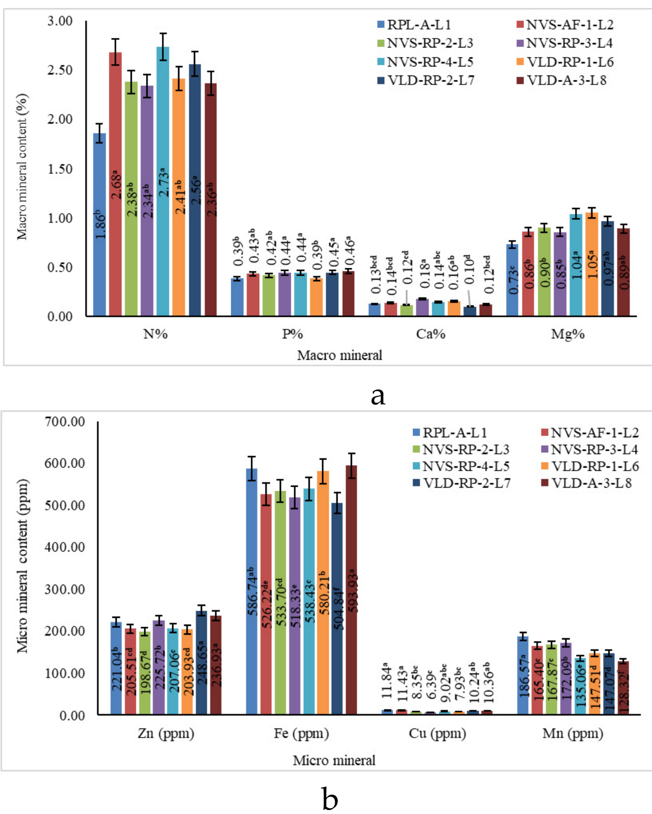


Fig 3. Variation in *S. foetida* de-oiled seed cake A) macro minerals composition *viz.*, nitrogen (N; SEM=±0.10), phosphorous (P; SEM= ±0.02), calcium (Ca; SEM= ±0.01) and magnesium (Mg; SEM=±0.03) and B) micro mineral *viz.*, zinc (Zn; SEM=±2.32), iron (Fe; SEM=±2.91), copper (Cu; SEM=±0.90) and manganese (Mn; SEM=±4.23) among different landscapes of south Gujarat, India; L: Landscape; RPL: Rajpipla; NVS: Navsari; VLD: Valsad; Letter different in vertical bars are significantly different according to Duncan's multiple range test (p ≤0.05)

Amongst micro minerals, the highest Zn (248.65 ppm) and Fe content (593.93 ppm) were obtained in the seed cake of VLD-2-L_7 and VLD-3-L_8 landscapes, respectively (Fig 3B). Whereas, maximum Cu (11.84 ppm) and Mn (186.57 ppm) contents were recorded in the seed cake of RPL-L_1 landscape. Minimum Zn, Fe, Cu and Mn contents were in NVS-2-L_3, VLD-2-L_7, VLD-1-L_6 and VLD-3-L_8 landscapes. Overall, average Zn, Fe, Cu and Mn contents of 218.44, 547.80, 9.44 and 156.24 ppm were observed across 8 landscapes.

Minerals are vital for normal growth, reproduction, health and proper functioning of the animal body.

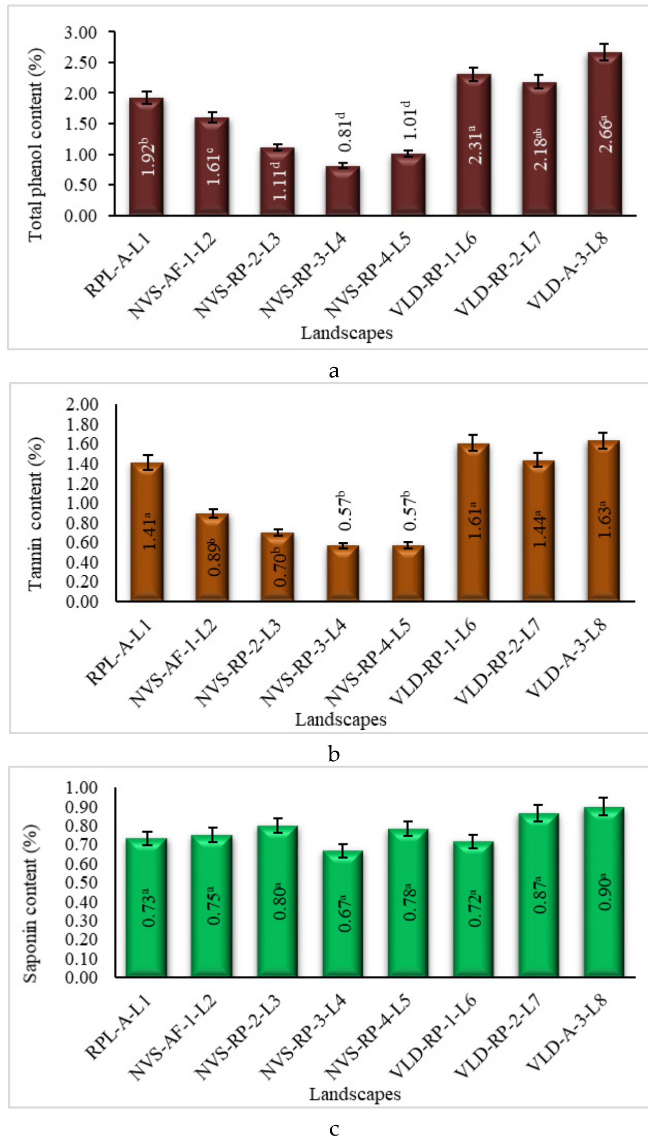


Fig 4. Variation in *S. foetida* de-oiled seed cake A) total phenols content (SEM=±0.11) B) tannin content (SEM=±0.04) and C) saponin content (SEM=±0.09) among different landscapes of south Gujarat; L: Landscape; RPL: Rajpipla; NVS: Navsari; VLD: Valsad; Letter different in vertical bars are significantly different according to Duncan's multiple range test (*p* ≤0.05)

Minerals protect and maintain the structural components of the body, organs and tissues, and are constituents of body fluids and tissues as electrolytes (Ali, 2023). Among macro minerals, P and Mg content in SFSC were well above the critical level, whereas Ca was below the critical limit (Anon, 2001; Garg *et al.*, 2009). Nitrogen is a major nutritional requirement in the diets of food-producing animals. Protein metabolism of domestic (and wild) animals from the aspects of food consumption, digestion, absorption, and eventual utilization by the animal body is largely synonymous with nitrogen metabolism. In a sense, animals consume protein to obtain nitrogen (Johnson *et al.*, 1979). The present study revealed that nitrogen content in SFSC varied significantly between landscapes. It ranged from 1.86 to 2.73%, with an overall average of 2.41% of all the studied landscapes in southern Gujarat. The results of the study evinced that there was significant variation in P content in SFSC from eight different landscapes. P content varied between 0.39 to 0.46%, having an overall average of 0.43% in the studied landscapes. P content observed in *A. indica* seed cake (0.45%; Gowda and Sastry, 2000) was in line with the range found in the present study. Seed cakes of *Moringa peregrina* and *Ailanthus excelsa* (Al-Harthi *et al.*, 2022; Pal *et al.*, 2023) were reported to have lower P content compared to SFSC. In our investigation, Ca content varied significantly in eight different landscapes and ranged from 0.10 to 0.18%, with an overall average percentage of 0.14. Commonly used palm, sunflower and *A. indica* seed cakes (0.27, 0.65 and 0.75%, respectively) were found to contain higher Ca content compared to *S. foetida* obtained in the present study (Gowda and Sastry 2000; Abdeltawab and Khattab 2018). In earlier studies, higher P and Ca contents were reported in *S. foetida* (Bera *et al.*, 1995).

The range of Mg content recorded in the present study varied from 0.73 to 1.05%, with an overall average percentage of 0.91. Much lower Mg content (0.025 and 0.16%) was reported in *S. foetida* and palm kernel cake than in the present study (Bera *et al.*, 1995; Abdeltawab and Khattab, 2018). However, slightly lower Mg content (0.43 and 0.44%) was recorded in *M. peregrina* and *A. indica* seed cake (Gowda and Sastry, 2000) as compared to that recorded in *S. foetida* in our findings. Interestingly, the results deduced that the range of all micro minerals reported in SFSC in our study was well above the critical level (Anon, 2001; Garg *et al.*, 2009). In the present study, SFSC Zn content ranged from 198.67 to 248.65 ppm, with an overall average of 218.44 ppm in the studied landscapes. Zn content in SFSC reported in the present study was higher compared to *Moringa peregrina*, flaxseed, groundnut and mustard seed cake (Kaur *et al.*, 2021; Al-Harthi *et al.*, 2022). Fe content ranged from 504.84 to 593.93 ppm, having an overall average of 547.80 ppm among the studied landscapes and was higher compared to mustard, nigella and groundnut seed cake (Kaur *et al.*, 2021). SFSC Cu content varied between 7.93 to 11.84 ppm, with an overall average of 9.44 ppm. Cu content reported in *S. foetida* was in line with some species like flaxseed and sesame seed cake and was higher as compared to mustard, nigella and groundnut seed cake (Kaur *et al.*, 2021). Mn content ranged from 128.32 to 186.57 ppm, having an overall average of 156.24 ppm. Mn content reported in *S. foetida* was higher compared to seed cake of species like flaxseed, sesame, mustard, nigella and groundnut (Kaur *et al.*, 2021). However, Zn, Fe, Cu and Mn contents in the present study were lower than earlier reports in the same species (Bera *et al.*, 1995; Abdeltawab and Khattab, 2018).

Secondary metabolite: The results expressed that the percentage of secondary metabolites, namely total phenols and tannins, varied significantly ($p \leq 0.05$) but there was no significant variation in saponin content in SFSC among the studied 8 landscapes (Fig 4a-4c). Significantly maximum total phenols (2.66%) and tannin content (1.63%) were in VLD-3-L_8 and minimum values for these attributes (0.81 and 0.56%, respectively) were in the seed cake of NVS-3-L_4 landscape. Saponin content, however, ranged from 0.58-0.82%. Overall, average total phenols, tannin and saponins were 1.70, 1.10 and 0.78%, respectively, in selected landscapes. Results revealed that total phenol content (TPC) ranged from 0.81 to 2.66%. Tannin content ranged from 0.56 to 1.63% in seed cake.

***Volatile phytochemicals*:** The volatile phytochemicals detected through gas chromatography mass spectrometry (GC-MS) in SFSC (Table 3) and chromatogram showing retention time and their relative abundance were also recorded (Fig 5). Only two phytochemicals, namely stigmasterol and β-sitosterol, were identified with relative area 51.73 and 48.21%, respectively.

Plant secondary metabolites are important components of the human and animal diet as they have many important biological properties that are needed for body growth and development and may be of interest in the pharmaceutical sector (Chanyawiwatkul *et al.*, 2018; Haleshappa *et al.*, 2022). Total phenol content refers to the amount of phenolic compounds present in feedstuffs and is a class of phytochemicals that are known for their antioxidant properties (Xuan *et al.*, 2018). Tannins are a type of

Table 3. Phytochemicals detected in *S. foetida* de-oiled seed cake

Sr. No.	Compound name	Chemical formula	Molecular weight	Retention time	Relative area (%)
1.	Stigmasterol	$C_{29}H_{48}O$	412.70	64.895	51.73
2.	β-Sitosterol	$C_{29}H_{50}O$	414.70	65.653	48.21

compound found in plants, known for their astringent, bitter taste and ability to bind to proteins and other molecules. Tannin binds to proteins including enzymes of the digestive tract thereby affecting the utilization of protein. Anti-nutritional factors such as tannins decrease feed intake due to their astringent properties (Ani *et al.,* 2011). However, in our study, *S. foetida* showed quite less and is well below the toxic limit to ruminants, *i.e*. more than 5% has been reported to be toxic for ruminants (Bhat *et al.,* 2013). The tannin content of TBOs like *P. glabra* seed cake (Vinay and Kanya, 2008) and *M. longifolia* seed meal (Ramadan *et al.,* 2016) is in the range found in SFSC in the present investigation. Higher saponins, which are often bitter, can decrease the palatability of plants for animals, making them less appealing as a food source. Saponins have been reported to cause depression in feed intake (Njidda and Ikhimioya, 2012). Ramadan *et al.* (2016) and Singh and Singh (1991) reported 9.8% saponin content in *M. longifolia* and *M. indica*, which was much higher than that found in SFSC in the present study. A number of flavonoids, phenolics, steroids, coumarins, phenylpropanoids and cerebrosides have been found in the roots, leaves, seeds and barks of this species (Cuong *et al.,* 2019). The *S. foetida* phytochemical survey studies focused on flavonoids as the main ingredients revealed triterpenoids, steroids, and fatty acids (Pham *et al.,* 2021).

More than 200 different phytosterols have been identified from different species, and stigmasterol and β-sitosterol are the most commonly occurring ones (Piironen *et al.,* 2000). Phytosterols (stigmasterol, β-sitosterol and squalene) exhibit numerous beneficial properties, including analgesic, immune-regulating, anti-inflammatory, antioxidant, anticancer, and anti-aging effects, as well as the ability to prevent cardiovascular diseases (Feng *et al.,* 2019; Zhang *et al.,* 2022; Dos Santos *et al.,* 2024). β–Sitosterol is one of the main dietary phytosterols belonging to the class of organic compounds known as stigmastanes (Sharma *et al.,* 2021). These have been reported in *Sacha Inchi* and *Prinsepia* oil (Cheng *et al.,* 2022) and are a potential source of natural oil rich in essential polyunsaturated fatty acids and have various nutraceutical and health benefits (Ramos-Escudero *et al.,* 2021). It was revealed that the incorporation of gallic acid into typical phytosterols (β-sitosterol and stigmasterol) and galloyl β-sitosterol had slightly better antioxidant activity at ambient temperature and better cholesterol-reducing activity (Wang *et al.,* 2019). Hence, *S. foetida* de-oiled cake would be beneficial for biological activities if fed to livestock.

Conclusion

Our study revealed significant variation in SFSC proximate, minerals and secondary metabolites among different landscapes. Proximate attributes deduced that seed cake is a rich source of protein, contains fairly good fat energy, *i.e.,* ether extract and the readily digestible, soluble carbohydrates like sugars and starches, as represented by nitrogen-free extract. Further, macro and micro mineral contents of seed cake were in a good range and among macro minerals, P and Mg content were well above the critical level, whereas Ca was below the critical limit. Similarly, total phenols and tannin content were quite low and well below the toxic limit to ruminants, which indicates that cake could be helpful in providing antioxidant properties. Phytosterols, *i.e.,* stigmasterol and β-sitosterol, were detected in seed cake, which exhibit numerous beneficial properties. Hence, SFSC could be utilized as an alternate feed source for livestock.

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