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COW GHEE AS A FUNCTIONAL MEDICINAL SUBSTANCE: A REVIEW OF ITS CHEMISTRY AND PHARMACOLOGICAL PROPERTIES

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1. ABSTRACT

Cow ghee, or clarified butter, was traditionally used in Ayurveda for food and curative purposes. This form of ghee is said to be rich in antioxidants with antibacterial and anti-inflammatory properties. The peculiar ability of ghee to penetrate the skin facilitates the absorption of other agents used for healing. Considering its medical uses, ghee is one of the most precious substances in Ayurvedic medicine and serves as a base or carrier for many other medicinal formulations. Recent studies conducted scientifically have investigated further properties of cow ghee to serve as an Ayurvedic ointment base. Lesser-known forms of traditional base materials have been compared, including Ceto stearyl alcohol, stearic acid, glyceryl monostearate, paraffin wax, white beeswax, and wool fat. The preparations were checked according to international quality standards (ICH guidelines) and tested for formulation stability by examining parameters such as color, consistency, pH, and solubility. This review article focuses on chemical composition, isolation and characterization of bioactive fractions, pharmacological activity and mechanism of action, comparative features – cow and buffalo ghee, regulatory and safety aspects as per FSSAI of cow ghee.

KEYWORDS: Conjugated linoleic acid [CLA], Saturated fatty acid [SFA], Free Fatty Acid[FFA], High Performance Liquid Chromatography [HPLC], Thin Layer Chromatography[TLC], FSSAI, Antioxidant, Anti-inflammatory.

2. INTRODUCTION

Cow ghee, a clarified butter obtained from cow's milk, has been an integral part of Indian culture and nutrition since the Vedic period. Traditionally, it is valued not only as a dietary fat but also as a component of Ayurvedic medicine, cosmetics, and religious rituals. Ghee is produced by heating butter or cream to remove milk solids, resulting in pure butterfat that is rich in energy, essential fatty acids, and fat-soluble vitamins (A, D, E, and K). From a nutritional perspective, cow ghee is considered one of the healthier fats due to its content of antioxidants, conjugated linoleic acid (CLA), and short- and medium-chain fatty acids that are efficiently metabolized by the liver.

In Ayurveda, cow ghee is described as a "yogawahi," capable of enhancing the delivery of nutrients and pacifying Pitta and Vata doshas. It is easily digestible, even for individuals with lactose or casein intolerance, due to the removal of milk solids during processing. The presence of naturally occurring antioxidants contributes to its long shelf life and therapeutic potential. As one of the most *sattvic* foods, cow ghee is believed to promote mental clarity, positivity, and spirituality. It also serves as a base for herbal infusions in Ayurvedic formulations, enhancing the absorption and efficacy of medicinal compounds.

Owing to its lipophilic nature, cow ghee facilitates nutrient transport across cell membranes, supporting mitochondrial and cellular functions. It promotes digestion, bowel regularity, immunity, and overall well-being.[1] Traditionally, ghee consumption is associated with delaying aging, improving vision, soothing skin conditions, and enhancing strength and endurance.[2] It also helps detoxify the body, imparts a natural glow to the skin, and maintains lipid balance by lowering LDL and increasing HDL cholesterol.[1,3]

The bioactive constituents of cow ghee contribute to its nutritional and medicinal value. Short-chain fatty acids like butyric acid strengthen the intestinal barrier and modulate inflammation, while medium-chain fatty acids are rapidly metabolized for energy and immune regulation. Long-chain fatty acids such as linoleic and oleic acids support cardiovascular health and lipid metabolism. Conjugated linoleic acid (CLA) provides anti-inflammatory, anti-carcinogenic, and cardioprotective benefits.[3,4,5]

aRich in fat-soluble vitamins (A, D, E, and K_2), cow ghee offers antioxidant protection, supports calcium absorption, and promotes bone health. Phenolic and flavonoid compounds further enhance its antioxidant potential. Overall, this unique blend of fatty acids, vitamins, and antioxidants establishes cow ghee as a valuable functional food with both traditional and modern therapeutic significance.[6,7]

3. Chemical Composition of cow ghee

3.1 Saturated Fatty Acids

Saturated fatty acids (SFA) constitute a major proportion of the total fatty acids present in cow ghee. The relationship between dietary SFA intake and the incidence of cardiovascular diseases (CVD), particularly coronary heart disease (CHD), has been widely studied. Overall, SFA are known to raise total and low-density lipoprotein (LDL) cholesterol, although individual saturated fatty acids exert different physiological effects. Myristic (C14:0) and palmitic (C16:0) acids have been associated with elevated serum LDL-cholesterol levels, while stearic acid (C18:0) is considered neutral.[8]

Lauric (C12:0) and myristic acids may exert stronger effects on plasma cholesterol than palmitic acid; however, results across studies vary. Recent evidence indicates that the overall balance of fatty acids in the diet may be more important than individual SFA levels. For instance, when dietary linoleic acid (C18:2 n-6) exceeds 5% of total energy, palmitic acid has little or no effect on LDL cholesterol. Moderate intake of myristic acid has even been associated with decreased plasma triglycerides and increased HDL cholesterol in healthy individuals.[8]

Since ghee is derived from milk fat, it contains notable proportions of 12:0, 14:0, and 16:0 fatty acids, similar to other ruminant-derived fats. While excessive intake of such SFA can unfavorably alter lipid profiles, the overall impact of cow ghee on cardiovascular health may differ due to its unique composition, which includes bioactive lipids like conjugated linoleic acid (CLA), butyric acid, and natural antioxidants. Studies suggest that modifying the fatty acid profile of dairy fats—by reducing SFA and increasing cis-monounsaturated fatty acids—can lower total and LDL cholesterol without adversely affecting HDL levels.[8]

Thus, understanding the role of specific saturated fatty acids in ghee is essential for assessing its health implications. Although cow ghee is rich in SFA, the matrix in which these fatty acids are delivered, along with accompanying bioactive compounds, may influence their physiological outcomes differently than other animal fats.[8]

3.2 Butyrate

Cow ghee is a natural source of butyric acid (butyrate), a short-chain fatty acid known for its vital role in gut health and disease prevention. Butyrate exhibits anticarcinogenic, anti-inflammatory, and immune-modulating properties. It inhibits cancer cell growth, promotes differentiation, and induces apoptosis through mechanisms such as histone deacetylase inhibition, which regulates gene expression.[8]

Animal studies have shown that dietary butyrate or its derivatives (e.g., tributyrin) can reduce the incidence of chemically induced mammary and colon tumors. Although ghee consumption may not greatly raise plasma butyrate levels, its effects are likely enhanced by other bioactive components such as conjugated linoleic acid (CLA), vitamin D, and antioxidants. Overall, butyrate contributes significantly to the digestive, anticarcinogenic, and therapeutic potential of cow ghee, supporting its recognition as a functional food in traditional and modern nutrition.[8]

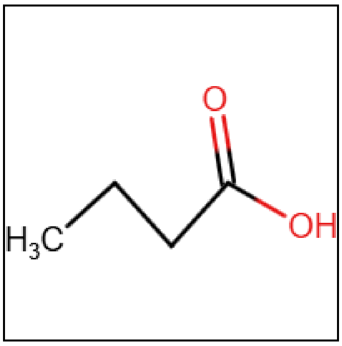


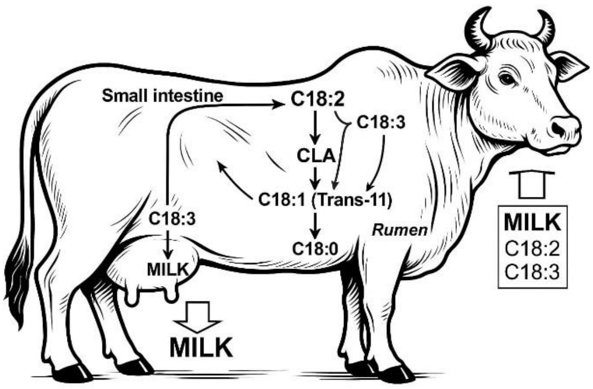
Fig 1: Chemical Structure of Butyrate.

3.3 Branched-Chain Fatty Acids (BCFAs)

Cow ghee contains a wide range of branched-chain fatty acids (BCFAs), including iso and anteiso isomers, typically with 15:0 and 17:0 anteiso being the most abundant. These fatty acids are known for their bioactive properties, including anticarcinogenic and cytotoxic effects against various cancer cell lines such as colon, liver, lung, and breast carcinoma. Studies indicate that BCFAs inhibit fatty acid synthesis in tumor cells by affecting fatty acid synthase activity. The cytotoxic potential of BCFAs is comparable to conjugated linoleic acid (CLA), highlighting their contribution to the health-promoting properties of cow ghee.[8]

3.4 Conjugated Linoleic Acid (CLA)

Conjugated linoleic acid (CLA) refers to positional and geometric isomers of octadecadienoic acid, with the *cis-9, trans-11* isomer being predominant in cow ghee. CLA exhibits a range of biological activities, including anticancer, anti-atherogenic, and immune-modulating effects. Experimental studies in animals and humans suggest that CLA can inhibit chemically induced tumor formation, improve lipid metabolism, enhance lean body mass, and reduce atherosclerosis. Thus, the presence of CLA contributes significantly to ghee's therapeutic and cardioprotective potential.[8]



Desi cow with cow ghee constituents

Fig 2: trans-11 octadecenoic acid is a common intermediate in the biohydrogenation of both α-linolenic acid and linoleic acid.[42]

3.5 Phylloquinones and Menaquinones (Vitamin K)

During traditional ghee preparation from fermented curd (*dahi*), probiotic bacteria generate phylloquinones (vitamin K_1) and menaquinones (vitamin K_2). These compounds have been inversely associated with the risk of type 2 diabetes and play a role in improving insulin sensitivity and maintaining bone and cardiovascular health. The fermentation step in traditional ghee-making therefore enhances its nutritional value.[9,10,11,12]

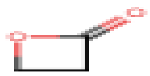
3.6 Cholesterol Oxidation Products (COPS)

Cholesterol oxidation products (COPS or oxysterols) can form when ghee is overheated or stored improperly. Although ghee naturally contains about 0.3–0.4% cholesterol, studies have shown that freshly prepared or properly stored ghee has negligible COPS content. Oxidized cholesterol compounds typically appear only after prolonged or repeated heating (e.g., frying beyond 60 minutes). Interestingly, ghee residues possess antioxidant activity that can inhibit cholesterol oxidation, making cow ghee relatively stable under normal cooking conditions.[13]

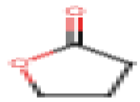
3.7 Lactones

Lactones are key aromatic compounds responsible for the characteristic sweet, nutty, and coconut-like flavor of cow ghee. Delta-lactones—particularly δ-octalactone, δ-decalactone, and δ-dodecalactone—are the dominant flavor components, with δ-octalactone having the lowest flavor threshold. The concentration of lactones increases with clarification temperature, which enhances ghee's aroma without affecting its quality.[14]

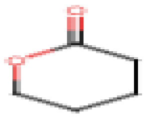
Alpha lactone



Beta Lactone



Gamma lactone



Delta lactone

Fig 3: Structure of Lactones.

3.8 Free Fatty Acids (FFA)

Free fatty acids contribute both to the flavor and quality of ghee. Ghee contains several FFAs such as C6, C8, C10, C12, C14:1, C15, C16, C18, and C18:2. Although excessive short-chain FFAs can cause rancidity, moderate levels are essential for ghee's characteristic taste. Cow ghee, unlike buffalo ghee, contains certain unique FFAs such as C18:2, contributing to its distinct aroma and flavor profile. The absence of volatile short-chain FFAs (C2–C4) is due to their loss during heating in the clarification process.[14]

Table 1: Fatty acids profile of ghee.[43]

Fatty acid	Cow ghee (%)	Buffalo ghee (%)
Butyric acid (C4:0)	1.772±0.004^{b}	1.958±0.001^{a}
Caproic acid (C6:0)	1.437±0.001^{a}	0.861±0.002^{b}
Caprylic acid (C8:0)	0.986±0.001^{a}	0.410±0.002^{b}
Capric acid (C10:0)	2.542±0.001^{a}	0.858±0.002^{b}
Lauric acid (C12:0)	3.144±0.001^{a}	2.030±0.003^{b}
Tridecylic acid (C13:0)	0.085±0.001^{a}	0.055±0.002^{b}
Myristic acid (C14:0)	10.268±0.001^{a}	8.516±0.003^{b}
Pentadecylic acid (C15:0)	0.963±0.001^{a}	1.027±0.003^{a}
Palmitic acid (C16:0)	23.954±0.015^{b}	28.751±0.009^{a}
Margaric acid (C17:0)	0.492±0.009^{b}	0.784±0.004^{a}

Stearic acid (C18:0)	9.331±0.007b	13.993±0.007a
Arachidic acid (C20:0)	0.120±0.003b	0.227±0.003a
Behenic acid (C22:0)	0.038±0.001b	0.087±0.001a
Tricosylic acid (C23:0)	0.018±0.001b	0.095±0.002a
Lignoceric acid (C24:0)	0.026±0.001b	0.078±0.002a
SFA	55.175	59.729
Myristoleic acid (C14:1)	0.937±0.000a	0.346±0.003b
Palmitoleic acid (C16:1)	1.177±0.006a	1.124±0.011a
Oleic acid (C18:1)	19.980±0.013a	18.584±0.213b
MUFA (cis)	22.094	20.055
Linoleic acid (C18:2)	1.635±0.001a	0.920±0.011b
a-Linolenic acid (C18:3)	0.661±0.001a	0.482±0.005b
Dihomo-g-linolenic acid (C20:3)	0.069±0.000a	0.055±0.001b
Eicosatrienoic acid (C20:3)	0.020±0.001a	0.000±0.000b
Arachidonic acid (C20:4)	0.105±0.000a	0.072±0.002b
Eicosapentaenoic acid (C20:5)	0.065±0.001a	0.056±0.001b
Docosahexanoic acid (C22:6)	0.032±0.003a	0.000±0.000b
PUFA (cis)	2.586	1.586
Petroselinic acid (18:1, t-6)	0.000±0.000b	0.171±0.006 a
Elaidic acid (18:1, t-9)	0.000±0.000b	0.215±0.005 a
Vaccenic acid (18:1, t-11)	2.125±0.009a	2.227±0.006 a
Octadecadienoic acid (18:2, c-9, t-12)	0.124±0.001a	0.000±0.000b
Octadecadienoic acid (18:2, t-9, c-12)	0.171±0.003a	0.000±0.000b
Conjugated linoleic acid (18:2, c-9, t-11)	0.995±0.000a	0.765±0.004b
Octadecatrienoic acid (18:3, t-9, t-12, t-15)	0.012±0.001a	0.000±0.000b
Octadecatrienoic acid (18:3, t-9, c-12, c-15)	0.124±0.013a	0.091±0.002b
rTFA	3.550	3.468
Total FA	83.405	84.838
n-3/n-6	0.43	0.51
AI	2.76	3.00
TI	2.62	3.00

3.9 Esters

Esters formed by the reaction of short-chain alcohols with FFAs also play a role in ghee's flavor development. These compounds are produced by bacterial esterases during fermentation or heating and contribute to the pleasant aroma of cow ghee.[14]

4. Isolation and Characterization of Bioactive Fractions from Cow Ghee

4.1 Saponification and Extraction of Unsaponifiable Matter (USM)

Before any useful chemicals can be extracted from ghee, triglycerides must be removed. This is achieved through the process of saponification, which entails heating the ghee sample with alcoholic potassium hydroxide, also called KOH, at 60 to 80 degrees Celsius. This process breaks down triglycerides into soap and glycerol. Following saponification, the product of the process gets diluted with water, or the inert fraction—which contains hydrocarbons, vitamins, and sterols—is separated many times via petroleum ether or hexane. The organic layers are combined, washed to neutralize them, dried with the help of the anhydrous form of sodium sulfate, and ultimately diluted under low pressure.

This fraction can then be thoroughly studied. The presence of β-sitosterol or other plant sterols indicates that vegetable lipids have contaminated the ghee.[15]

Table 2: Saponification Value of Cow Ghee.[43,44]

Milk fat content	Minimum 99.5% (m/m)
Moisture	Maximum 0.5% (m/m)
Free fatty acids (as oleic acid)	Maximum 2.0% (m/m).
Reichert Meissl value	Minimum 24
Polenske value	Minimum 24
Butyro-refractometer reading	40 to 44.
Colour	Golden yellow, uniform, no artificial colouring
Odour and flavour	Characteristic of fresh ghee, free from rancid or foreign odours.

4.2 Chromatographic Techniques

A variety of chromatographic techniques are used to separate and identify the bioactive substances in the insoluble fraction.

a. Reversed-Phase TLC (RP-TLC) and Thin Layer Chromatography (TLC): TLC provides a quick way to screen for sterol and hydrocarbons. RP-18 plates and a petroleum ether and acetonitrile solvent combination can detect β-sitosterol in contaminated samples even at 1%.

b. High-Performance Thin Layer Chromatography (HPTLC): HPTLC offers higher resolution and permits densitometric chemical measurement. It is widely used to profile sterols, vitamins, and carotenoids.

c. Column Chromatography: This method helps separate the USM into fractions that contain sterols, hydrocarbons, and carotenoids.

d. High-Performance Liquid Chromatography (HPLC/RP-HPLC): HPLC makes it possible to measure certain elements like cholesterol, β-sitosterol, and tocopherols precisely.[15]

4.3 Gas Chromatography (GC) and Gas Chromatography–Mass Spectrometry (GC–MS)

Gas chromatography (GC) and gas chromatography–mass spectrometry (GC–MS) are two of the best techniques for identifying and quantifying bioactive compounds in the unsaponifiable fraction. Before analysis, the USM is derivatized, usually with silylation agents like BSTFA, to make the sterols volatile and thermally stable. Every component in GC-MS produces a unique mass spectrum that serves as a fingerprint, and spectral libraries such as NIST are used to confirm its authenticity.

Comprehensive profiles of n-alkanes, phytosterols (β-sitosterol, campesterol, and stigmasterol), carotenoid degradation products, and cholesterol are produced by this method. Unmistakable indicators of vegetable oil adulteration include plant sterols or strange hydrocarbon peaks in ghee analysis.[16]

4.4 Supercritical CO_2 Extraction (SC–CO_2)

Supercritical CO2 extraction is a solvent-free, clean technique for separating nonpolar bioactive materials. When carbon dioxide reaches its critical point (31 °C and 73.8 bar), it transforms into a supercritical fluid with properties of both a gas and a liquid. Because the process operates at low temperatures, it lessens the thermal deterioration of fragile materials like vitamins and carotenoids.[16]

4.5 Nuclear Magnetic Resonance (NMR) Spectroscopy

Nuclear magnetic resonance (NMR) spectroscopy is used to confirm the structures of purified bioactive compounds. After samples are dissolved in deuterated solvents like DMSO-d^2 or chloroform-d, both ^1H and ^{13}C NMR spectra are obtained. NMR can help distinguish authentic ghee components from those that have been adulterated by identifying specific structural traits of sterols, tocopherols, and carotenoids.[17]

4.6 Integrated Analytical Workflow

1. Sample Preparation
2. Saponification (Alcoholic KOH)
3. Extraction of Unsaponifiable Matter (Hexane / Petroleum Ether)
4. Chromatographic Screening (TLC, HPTLC, HPLC)
5. GC or GC–MS for Identification and Quantification
6. Optional SC–CO_2 Extraction for Cleaner Recovery

7. NMR for Structural Confirmation.[15,18]

5. Pharmacological activity of cow ghee

5.1 Antioxidant activity

Cow ghee has a high level of antioxidant activity because it contains bioactive elements such conjugated linoleic acid (CLA), vitamins A and E, carotenoids, and phenolic antioxidants. These compounds reduce oxidative stress in biological systems by scavenging free radicals and neutralizing reactive oxygen species (ROS). The fat-soluble antioxidants in ghee, particularly α-tocopherol and β-carotene, protect cellular membranes from lipid peroxidation, while CLA enhances endogenous antioxidant defenses and prevents oxidative lipid modification. According to research, taking ghee supplements can improve the body's defenses against oxidative damage by increasing the activity of crucial antioxidant enzymes like glutathione peroxidase (GPx), catalase (CAT), and superoxide dismutase (SOD).[38,39]

Mechanism of action

Ghee naturally contains high levels of the powerful antioxidants vitamin A and vitamin E. Vitamin E protects our cells and lipids from the damaging effects of free radicals, which are harmful chemicals that accelerate aging and disease. Vitamin A promotes strong immune function, tissue repair, and healthy skin. These vitamins work together to protect the body from oxidative stress while preserving the health of our organs and ensuring that our cells are functioning properly.[33]

5.2 Anti-inflammatory activity

Cow ghee has long been used in traditional medicine for its soothing and healing properties, and modern studies now support its anti-inflammatory effects. One of the key components responsible for this activity is butyrate, a short-chain fatty acid that naturally helps calm inflammation in the body. Butyrate is known to reduce the activity of NF-κB, a major pathway that triggers the release of inflammatory chemicals like TNF-α, IL-1β, and IL-6. By slowing this pathway, ghee helps reduce swelling and tissue irritation. Another compound present in ghee, conjugated linoleic acid (CLA), also contributes by regulating eicosanoids and lowering oxidative stress, both of which play a role in long-term inflammation. Research has shown that regular consumption or external application of ghee can help reduce edema, support wound healing, and protect delicate tissues such as the stomach and intestinal lining. These combined benefits explain why cow ghee is often recommended for digestive problems, respiratory irritation, and conditions involving chronic inflammation.[33,38]

Mechanism of Action

One of the main health advantages of ghee is butyric acid, a short-chain fatty acid that reduces inflammation. It does this by preventing NF-κB, the protein that triggers inflammation, from doing its job. Controlling NF-κB reduces the body's production of inflammatory chemicals like TNF-α, IL-6, and IL-1β. This not only reduces intestinal inflammation but also reduces inflammation throughout the body. Ghee is therefore commonly suggested for digestive and bowel health problems.[33]

5.3 Anticancer activity

In a study of feeding cow ghee versus soyabean oil on 7,12-dimethylbenz(a)-anthracene (DMBA) induced mammary carcinogenesis and expression of cox-2 and peroxisome proliferators activated receptors-γ(PPAR-γ) in mammary glands of rats revealed anticancer potential of cow ghee. In the DMBA (a carcinogen) treated groups, the animals given soybean oil had higher tumour incidence (65.4%), tumour weight (6.18 g) and tumour volume (6285 mm3) compared to those fed with cow ghee (26.6%, 1.67g, 1925 mm3, respectively). Tumour latency period was increased to 27 week on cow ghee as compared to 23 week on soyabean oil.[21]

Histological analysis of tumours showed more rapid progression of carcinogenesis on soybean oil than on cow ghee fed rats. The expression of cyclooxygenase-2 was observed only in DMBA treated rats and it was significantly less on cow ghee than on soybean oil. The expression of PPARγ was also significantly more on cow ghee than on soybean oil. The study proved that dietary cow ghee opposed whereas soybean oil attenuates mammary carcinogenesis induced by DMBA. Also, the effect is mediated by decreased expression of cyclooxygenase-2 and increased expression of PPAR-γ in the former group.[21]

Mechanism of action

Colostrum contains cytokines such as interleukins 1, 6, 10, interferon G, and lymphokines that are used in cancer treatment. It has been discovered that colostrum lactalbumin can induce the selective death (apoptosis) of cancer cells while having no effect on the surrounding non-cancerous tissues. Similar reports of lactoferrin's anti-cancer properties have been made. Colostrum's combination of growth and immunological factors can stop cancer cells from spreading.[40]

5.4 Analgesic activity

In a study of cow urine and its distillate to assess the analgesic effect using rat tail immersion method it was observed that the animals received cow urine showed comparable analgesic effects to standard drug group received Diclofenac Sodium thereby strengthening the recommendations of Ayurvedic texts to use cow urine as reasonably safe, easily available and economic natural analgesic. Their analgesic activity is attributable to the steroidal moieties and some volatile fatty acids whose presence in cow urine is established through other parallel studies involving chemical and instrumental analysis.[22]

Mechanism of Action

It is believed that an Autoimmune process, potentially triggered by an allergic response to the protein GAD present in cow's milk, causes juvenile diabetes (Type I, insulin dependent). This and other allergies can be counteracted by a number of substances found in colostrum.IgF-1 efficiently treats acute hyperglycemia and reduces a Type II diabetic's reliance on insulin, according to human trials.[40]

5.5 Antidiabetic Activity

The effect of cow urine formulation (Gomutra ark, GoA) on experimental alloxan-induced diabetes in rats was studied. Wistar albino rats of either sex weighing 200-250 g were used. The biochemical parameters observed were blood sugar, vitamin C and malondialdehyde (MDA) release. GoA significantly lowers blood glucose in diabetic rats although the observed effect was found to be less than standard antidiabetic, glibenclamide. It is suggested that GoA might have a significant protective effect against alloxan-induced type I Diabetes Mellitus. GoA contains volatile fatty acids like acetic acid 2 propenyl ester, acetic acid methyl ester, 2 2 3 trichloro propionic acid, Butanoic acid3methyl, propyl ester, 1H indol-3-acetate, acetic acid phenyl ester, quinoline, which act as an antioxidant. The antioxidant potential might be contributing for the antihyperglycemic effect, by preventing formation of the free radicals which cause damage to the beta cells of pancreas. It significantly lowers the level of malondialdehyde and vitamin C in diabetic rats. No toxicity was observed even when cow urine was given 32 times of the study dose in acute toxicity and no significant change were observed when it was used chronically, suggesting that cow urine is having a very high therapeutic index.[23]

The findings of the study supported the traditional use of cow urine in diabetes and have a high therapeutic index and safety profile for chronic use.[23] In a study of use of cow urine distillate in diabetes rats, the diabetes was induced by administration of streptozotocin (50 mg/kg bw., i.p) dissolved in citrate buffer (0.1 M, pH 4.5). The anti diabetic effect of the (three different doses) and a standard drug Glibenclamide (0.25 mg/kg, p.o) was studied in these diabetic rats. The parameters employed in the study included assessment of fasting blood glucose levels, serum lipid profiles, liver glycogen levels and initial and final changes in body weight. The cow urine distillate produced a significant reduction of elevated blood glucose, serum cholesterol and serum triglycerides level when compared with diabetic control.[24]

Mechanism of action

It is belived that autoimmune process, potentially triggered by an allergic response to the protein GAD present in cow's milk causes juvenile diabetes [Type I, insulin dependent] this and other allergies can be counteracted by a number of substance found in colostrum. IgF-1 efficientyl treats acute hyperglycemia and reduces a type II diabetic's reliance on insulin, according to human trials.[40]

5.6 Anti-hemorroids activity

Hemorrhoids are a common anorectal condition defined as the symptomatic enlargement and distal displacement of the anal mucosa and classified as Grade I, II, III and IV on the basis of symptoms and severity. An inflammatory reaction and vascular hyperplasia may be present in Hemorrhoids. Ayurvedic texts have an abundant literature regarding anti-microbial, bio-availability enhancer, free-radical scavenging properties of Cow-urine. Sushruta samhita states that Arsha can be treated by the four ways viz. Bheshaja (Internal medication), Kshara (Local application), Agnikarma (Cauterization), Shastrakarma (Surgery). This work is based on the Ayurvedic principle of Aushadhi Chikitsa. Cow-urine is a known appetizer (Agnivardhaka). Since Agnimandya is responsible for all types of the diseases, Gomutra through its Agnivardhaka effect relieves the Agnimandya. Cow-urine acts on the large intestine through its Mala-bhedana effect. This results into the smooth excretion of stool, thereby providing a greater relief to the patients of hemorrhoids e.g. relief from pain during defecation, bleeding, perianal itching.[25]

Mechanism of action:- the antihumeroid activity mechanism of cow ghee includes: Anti-inflammatory action by modulating cytokines and inflammatory mediators. Antioxidant activity through fat-soluble vitamins that protect tissues from oxidative damage. Lipophilic property facilitating cellular absorption and delivery of active components. Promoting tissue healing and reducing rectal damage related to hemorrhoidal inflammation. Thus, cow ghee acts as both a therapeutic agent and a carrier, aiding in reduction of inflammation and promotion of healing in hemorrhoidal tissues.

5.7 CNS Effects

5.7.1 Antiepileptic activity

Panchgavya Ghrita(PG) was screened for some neurological parameters in rats and it was found that PG protected rats from maximal electroshock (MES) induced convulsions, increased the spontaneous motor activity as measured by actophotometer and inhibited the pentobarbitone induced sleep time in rats without much influence on the general behavior of the rats except increase in the general activity. PG appears to possess anti convulsant property but the degree of protection might not be sufficient to use it as single antiepileptic agent. It is concluded that PG can be used as adjuvant in treatment of epilepsy.[27]

Mechanism of action:- Ghee is rich in omega-3 and omega-9 fatty acids, which are vital for the brain and hormone balance. By making the membranes of brain cells more elastic, these lipids help nerve signals travel through the body more smoothly. They also aid in the synthesis of dopamine and serotonin, two neurotransmitters that influence mood, memory, and focus. Along with supporting the nervous system and helping to maintain hormonal balance, these fatty acids also play a part in the biosynthesis of hormones.[18]

5.7.2 Antistress Activity

Panchgavaya Ghrita (PG) along with ethanolic extracts of Aloe barbadensis Mill was evaluated for antistress activity using Tail suspension model in mice against Alprazolam as standard. The combination was found to possess significant antistress potential at the level $p<0.01$ as compared with standards and controls. The synergistic action of Panchgavya ghrita and Aloe extract was attributed to the increased levels of GABA and decreased levels of dopamine and plasma corticosterone levels.[28]

Mechanism of Action:- The immune system can be momentarily depressed by intense exercise and athletic competiton, which lowers the quantity of T-lymphocytes and NK cells. As a result, illnesses, including chronic Fatigue Syndrome, are more common in athletes. Numerous immunological components found in colostrum can significantly lower the frequency and severity of infections brought on by both mental and physical stress.[40]

5.8 Wound Healing and Antiulcer activity

In a case study on a wound in a buffalo which did not respond to variable treatment that ranges from simple herbal preparation like turmeric to modern medicines (antibiotics) responded very well to the treatment by a formulation containing cow's ghee. Cow's ghee has been reported to exert significant wound healing activity. Its antifungal activity has also been shown to be independent of any antibiotic or antifungal agent, which may be included into the formulation. Ghee contains several saturated and unsaturated fatty acids which are capable of taking part in metabolic processes involved in any wound healing. It seems therefore worthwhile that the cow's ghee is explored further as an effective clinical agent. A study of wound healing activity of preparation containing Aegle marmelos leaves and cow ghee showed enhanced and rapid healing. The effects produced by topical application of combination of Aegle marmelos leaves extract and cow ghee with reference to wound contraction, wound closure, decrease in surface area of wound and tissue regeneration at the wound site were studied. The Kaushik et al, IJPSR, 2016; Vol. 7(4): 1383-1390. E-ISSN: 0975-8232; P-ISSN: 2320-5148 International Journal of Pharmaceutical Sciences and Research 1389 wound healing activity was found to significant as the wound was healed completely in eight days. Study of wound healing activity of cow urine in wistar albino rats by excision wound model revealed significant healing activity of cow urine. The parameter studied was the rate of wound contraction. The studies on excision wound healing revealed that there was a decrease in wound area. External application of urine showed significant increase in wound healing in male and female rats after Day 4 as compared to all other groups. However till the end of 14th day animals showed that only 0 % and 0.40.4 % of healing was left, which may be due to normal immunity of the animals whereas nitrofurazone, standard drug treated animals showed 0 % and 0.50.3 % healing. Study revealed that the cow urine on external application to the wound fastened the healing process. In a study to evaluate the cow ghee containing formulation of Aloe vera for wound healing potential, histological examinations revealed good keratinization, epithelization, fibrosis and collagenation indicative of good healing process. The results were comparable with Framycetin sulphate cream (1% w/w). Incision wound for tensile strength, excision wound contraction and histological observations of regenerated tissues were used to investigate the healing potential of the formulation.[41,42,43,44]

Mechanism of action:- Healing of wounds Numerous components of colostrum promote the healing of wounds. By directly affecting DNA and RNA, nucleotides, EGF, TGF, and IGF-1 promote skin growth, cellular growth, and repair. These growth factors aid in the healing of tissues injured by burns, ulcers, trauma, surgery, or inflammatory diseases. Skin, muscle, cartilage, bone, and nerve cells are the tissues that benefit from colostrum's wound-healing qualities. After being washed and sterilized, powdered colostrum can be administered topically to burns, cuts, abrasions, aphthous ulcers, gingivitis, and sensitive teeth.[40]

5.9 Immunomodulatory activity

Cow urine distillate was found to have immunomodulatory effect as it enhances proliferation of T and B lymphocytes, blastogenesis and increases levels of IgG in mice and chicks (avian species). Distillate and redistillate of Cow's urine was studied for protective effects on Human Polymorphonuclear Leukocytes challenged with established genotoxic chemicals. Actinomycin-D (0.1µmol/L) and hydrogen peroxide (150 µmol/L) were used for inducing DNA strand break with 0.1% DMSO as negative control. The antioxidant status and volatile fatty acid levels were determined. The study showed that both actinomycin-D and peroxide caused statistically significant DNA unwinding of 80% & 75% respectively ($P<0.001$) as revealed by fluorimetric analysis of DNA unwinding (FADU) but the damage could be protected with the redistilled cow's urine distillate (1, 50 & 100 µL). The redistillate of cow's urine was found to possess total antioxidant status of around 2.6 mmol, contributed mainly by volatile fatty acids (1500 Kaushik et al, IJPSR, 2016; Vol. 7(4): 1383-1390. E-ISSN: 0975-8232; P-ISSN: 2320-5148 International Journal of Pharmaceutical Sciences and Research 1388 mg/L) as revealed by the GC-MS studies. These fatty acids and other antioxidants might cause the observed protective effects. The redistilled cow's urine distillate(RCUD) was found to posseses strong antigenotoxic and anticlastogenic properties against human polymorphonuclear leukocytes(HPNLs) and human peripheral lymphocytes(HLC) in-vitro when treated with Cr+6 and MnO2. Manganese dioxide and hexavalent chromium are established genotoxicants and clastogens which could cause induction of DNA strand break, chromosomal aberration and micronucleus. Three different levels of RCUD- 1 µL/mL, 50 µL/mL and 100 µL/mL were used in the study. RCUD showed statistically significant against DNA strand break, chromosomal aberration and micronucleus formation. The effects can be contributed to the antioxidants present in RCUD.[45,46,47,48]

Mechanism of action:- The fat-soluble vitamins in ghee, especially A, D, E, and K, boost immunity in a number of ways. Vitamin D fortifies the body's defenses by enhancing the activity of immune cells. The lungs, stomach, and other internal body surfaces are kept healthy by vitamin A, which prevents. CLA also improves the immune response by encouraging the production of healthy T cells and antibodies. Together, these nutrients support an efficient immune system that is ready to fight off infections.[18]

6. Comparative Features – Cow and Buffalo Ghee

Ghee, the clarified fat derived from milk, shows notable compositional differences depending on the originating species. While both cow and buffalo ghee are dominated by triglycerides (about 98%), the relative proportions of minor lipid groups and individual fatty acids vary and have meaningful nutritional and functional consequences.[29,32]

6.1 Fatty-Acid Composition

The main fatty acids detected in ghee from both species are myristic (C14:0), palmitic (C16:0), stearic (C18:0) and oleic (C18:1). Quantitative profiles diverge: cow ghee typically contains a marginally higher share of unsaturated fatty acids (≈32.21%) compared with buffalo ghee (≈28.73%). Buffalo ghee tends to be richer in certain short- and long-chain saturated acids (e.g., C4:0, C6:0, C16:0, C18:0), whereas cow ghee more often contains higher levels of mono- and polyunsaturated species such as oleic, linoleic (C18:2), linolenic (C18:3) and eicosanoic (C20:0) acids.[30,31]

6.2 Physicochemical Implications

These compositional distinctions affects the physical and chemical behavior. A greater proportion of long-chain saturated fatty acids elevates the melting point and solid fat fraction in buffalo ghee, making it firmer at room temperature. The cow ghee, with relatively more unsaturated and medium-chained components, softens and hence melts at the lower temperatures, which can further influence the digestibility. The lower unsaturation in buffalo ghee confers improved resistance to oxidation and enzymatic hydrolysis; by contrast, cow ghee often includes higher concentrations of endogenous antioxidants (carotenoids, tocopherols) and bioactive lipids such as conjugated linoleic acid (CLA), especially when produced from pasture-fed animals.[29,31,32]

6.3 Seasonal and Extrinsic Factors

Species-specific differences are further modulated by the extrinsic factors. Feed composition, seasonal grazing patterns, lactation stage and processing practices alter fatty-acid distribution. Empirical data indicate that the samples collected in summer (May–June) commonly shows increased unsaturation relative to monsoon and winter samples, demonstrating that the environmental and management variables can accentuate or attenuate interspecies contrasts.[32]

Table 3: Proximate composition and physicochemical characteristics of buffalo and cow ghee.

Characteristics / components	Buffalo ghee	Cow ghee
Moisture (%)	0.3 ± 0.016 a	0.3 ± 0.022 a
Lipid (%)	98.9 ± 0.50 a	98.9 ± 0.80 a
Protein (%)	0.78 ± 0.026 a	0.81 ± 0.045
Ash (%)	0.03 ± 0.002 b	0.09 ± 0.028 a
Energy (kcal/kg)	9305 ± 230.5 a	9483 ± 44.5 a
Acid value (mg/NaOH)	0.03 ± 0.01 b	0.21 ± 0.02 a
Free Fatty Acids (%)	0.01 ± 0.005 b	0.1 ± 0.01 a
Saponification Value (mg KOH/g)	233.9 ± 38.5 a	217 ± 9.2 a
Iodine value (g iodine/100g)	22.6 ± 1.58 b	50.6 ± 1.59 a

Data are expressed as mean ± standard deviation. Different letter within a raw indicates statistically difference ($p < 0.05$)

7. Future Prospects of Cow Ghee

Cow ghee, until recently considered only as a traditional fat for cooking, has now emerged as a multi-functional lipid of scientific, nutritional, and industrial interest. Due to overwhelming evidence on its health-promoting potential, there is an increased demand for cow ghee both in domestic and global markets. Newer processing technologies and interest in traditional foods on the part of consumers are providing new opportunities for its use.[49]

1. The Functional and Nutraceutical Applications

The cow ghee is characterized by the presence of short and medium chained fatty acids, butyric acids, and the CLA is responsible for anti-inflammatory, antioxidant, and gut-protective activities. It hence acts as a carrier for lipophilic bioactive compounds and fat-soluble vitamins A, D, E, and K, enhancing their absorption and stability. The future nutraceutical products may include fortified ghee formulations, herbal ghee, and encapsulated ghee powders suitable for the functional foods and the clinical nutrition.

2. Technological Advancements

Recent technological advances comprising microencapsulation, nanoemulsion formation, and enzymatic modification have improved the oxidative stability, flavor, and shelf life of cow ghee. These advancements also allow the creation of low-cholesterol, probiotic, and omega-3-enriched ghee products, expanding its functional and therapeutic applications. On the other hand, bio-fortified and the herbal-infused ghee products are gaining research attention owing to their enhanced nutraceutical potential.[50]

3. Industrial and Cosmetic Applications

Cow ghee contains rich amounts of natural triglycerides, antioxidants, and sterols, finding greater usage in cosmetic and pharmaceutical formulations. It possesses emollient, moisturizing, and wound-healing properties, making it suitable for use in herbal creams, ointments, and medicated oils. Ghee improves skin hydration, enhances collagen synthesis, and helps in the healing of minor burns and wounds, strengthening its role in modern cosmeceuticals.[51]

4. Market and Sustainability Outlook

The ghee market is fast-growing worldwide, and demand for natural, organic, and lactose-free dairy fats is expected to continue growing. The market is expected to grow significantly between 2024 and 2030, especially in North America and Europe, where consumers have started adopting traditional fats as healthier versions of processed oils. Attention to sustainable production of dairy, eco-friendly packaging, and standardized quality control will define the long-term growth and export potential of cow ghee originating from India and other dairy-rich regions.[52]

8. CONCLUSION

This article review discusses the overall importance of the cow ghee, describing its biochemical composition, pharmacological potentials, industrial uses, and regulatory standards. The cow ghee is a class of clarified lipids derived from the cow's milk hence representing a perfect blending of the traditional Ayurvedic principles with modern nutrition science. Rich in short-, medium-, and long-chain fatty acids, butyric acid, CLA, tocopherols, carotenoids, and fat-soluble vitamins A, D, E, and K, it exerts a wide range of nutritional, antioxidant, anti-inflammatory, and therapeutic activities.

Chemically, bioactive lipids include butyrate and CLA, which improve gut health, lipid metabolism, and immune function, and reduce oxidative stress and inflammation. Overall, the cow ghee due to a higher proportion of unsaturated and bioactive fatty acids, offers better benefits to cardiovascular and metabolic health compared to buffalo ghee.

Advanced techniques such as TLC, HPLC, GC–MS, and NMR have identified the major unsaponifiable constituents responsible for the functional characteristics of ghee, thus assuring quality control and detection of adulteration. It has also been pharmacologically validated to possess hepatoprotective, neuroprotective, antidiabetic, and immunomodulatory activities. The Panchgavya Ghrita formulation enhances its Ayurvedic therapeutic applicability with the CNS and anti-stress activities.

Its applications are further extended in modern food and nutraceutical systems through various industrial innovations, such as fractionation, encapsulation, and microencapsulation. On the other hand, the regulatory bodies make sure that purity, compositional, and safety standards are maintained to uphold authenticity and consumer trust in it. One of them is FSSAI.

Thus, cow ghee is a traditional but scientifically endorsed multifunctional lipid nutriment with established nutritional and pharmacological attributes. It promotes the cardiovascular, hepatic, gastrointestinal, and neural health in moderation and standardized hygienic production. Continued interdisciplinary research combining the Ayurvedic knowledge with the molecular and clinical approaches will further enhance its potential as a functional food and therapeutic nutraceutical.

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