



Extraction and Some Characteristics of Gelatin from Camel's Skins

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ABSTRACT

Background: Utilizing environmentally harmful waste materials remains a challenge. Porcine and bovine gelatins pose health risks, including swine flu (H1N1) and bovine spongiform encephalopathy (BSE) and disagree with *Halal* and *Kosher* dietary laws. Camel skins as byproducts offer a viable alternative. This study extracts and characterizes gelatin from camel skin.

Methods: Gelatin was extracted from camel skin using hot water (60°C, 6 h). Physicochemical properties (proximate composition, pH, clarity), amino acids (HPLC), gel strength, foaming capacity and FTIR functional groups were analyzed.

Result: Gelatin yield was 5.4±2.11%. Low turbidity (0.25±0.11 at 620 nm). Protein content: 95.31±1.94%, rich in glycine/proline (~28%). Gel strength (175.55±7.12 g), foaming expansion (164±5.65%) and FTIR confirmed Amide peaks (3444, 1628, 1559 cm⁻¹).

Key words: Camel's skin, Foaming expansion, FTIR, Gel strength, Gelatin.

INTRODUCTION

Gelatin is a natural protein originating from moderate hydrolysis of collagens. Collagen is obtained from skins, cartilage, bones and tendons from animals, such as cows (Abuibaid *et al.*, 2020; Al-Hassan, 2020a; Zhou *et al.*, 2022). Gelatin is a fat-free, cholesterol-free polymer originating from partially hydrolyzed collagen, where polypeptide bonds break down. It is widely used in food, pharmaceuticals and other industries for gelling, thickening, stabilizing and emulsifying. Its heat-reversible gel that melts near body temperature (Al-Hassan, 2020; Xu *et al.*, 2017; Zhou *et al.*, 2022). Major gelatin sources are extracted from pig's skins, bovine hides and bones, which represent about 98% of global marketed gelatin. The remaining percentage (2%) gelatin was sourced from fish (Ahmed *et al.*, 2020a; Al-Hassan, 2020). Gelatin types from the animal sources (*i.e.*, pigs and cows) have been extensively marketed (Fawale *et al.*, 2021; Kittiphattanabawon *et al.*, 2010). Religious certificates (*Halal* in Islam or Jewish *Kosher*) and ethical concerns restrict gelatin consumption from porcine or bovine sources in many regions (Al-Hassan, 2020a; Kittiphattanabawon *et al.*, 2010). Disease outbreaks, namely bovine spongiform encephalopathy (BSE), foot-and-mouth infections and swine flue have driven demands for safer gelatin sources (Ahmed *et al.*, 2020a; Xu *et al.*, 2017). Recently, fish gelatins are now growing alternatives from processing wastes. (Ahmed *et al.*, 2020a; Kittiphattanabawon *et al.*, 2010; Zhou *et al.*, 2022). In fact, fish gelatins had poorer quality, such as lower Bloom values (gel strength) and lower stability than the mammalian ones. Thus, they have limited applications (Ahmed *et al.*, 2020a; Kittiphattanabawon *et al.*, 2010). Mammalian gelatins have superior thermo-reversible gelling and water solubility and thus making them ideal for food and pharmaceutical applications (Abuibaid *et al.*, 2020; Fawale *et al.*, 2021).

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The U.S. gelatin market is expected to reach five billion USD by 2025, driving demand for nine hundred kilotons due to its use in functional foods and pharmaceuticals (Alipal *et al.*, 2019). The food and other industries are prompted to search for alternative sources for *halal* gelatin from traditional origins of gelatins. At the large production scale, few *halal* alternatives from mammal sources are

available as sources of gelatin production (Fawale *et al.*, 2021). To this end, camel skins could supply gelatins that meet religious needs. In Arabia and Africa, camels (one-hump, *Camelus dromedarius*) have been domestically raised for milk and meat sources and camel's skins could be alternative sources for gelatin (Al-Hassan, 2020a; Benyagoub *et al.*, 2022; Demlie *et al.*, 2023; Fawale *et al.*, 2021). Worldwide, the camel population is about thirty five million and the wet skin is a major by-product representing up to 15% of slaughtered camels (Al-Hassan, 2020a; Fawale *et al.*, 2021). In fact, scarce information has been published on camel's skin gelatin. Depending on the extraction method, there are the alkaline extracted gelatin (Type B) and the acid extracted type A gelatin from camel and other mammalian skins (Abuibaid *et al.*, 2020; Ahmad *et al.*, 2018, 2020; Al-Hassan, 2020b; Bessalah *et al.*, 2023; Mulyani *et al.*, 2017; Yu *et al.*, 2016). Alkaline treatments weaken gelatin gel strength and thermal stability, take longer time and yield lower purity gelatins. Acid treatment releases more soluble proteins and fats efficiently (Mulyani *et al.*, 2017).

This study investigates camel's skin gelatin, covering extraction methods, composition and potential applications in food and pharmaceuticals. It highlights its properties, contributing to research on alternative gelatin sources.

MATERIALS AND METHODS

Samples and other materials

Fresh camel skins were obtained from a slaughterhouse in Al-Omran, 23 km northeast of the university campus. Adult male camels (2-year-old) were selected, as preferred for meat. Samples were stored at -20°C before analysis. Analytical-grade chemicals were used and experiments ran from 2023-2024 in the Food and Nutrition Department, College of Agricultural and Food Sciences.

Gelatin extraction

Skin preparations and gelatin extraction were done as reported previously (Chen *et al.*, 2014; Ma *et al.*, 2018; Yu *et al.*, 2016). Thawed skin pieces (1 kg) were rinsed under tap water to remove dirt, lightly squeezed, then soaked in 10% NaCl (5 L) for six h at room temperature.

After NaCl treatment, skin pieces were washed with running tap water, then soaked in 3500/ ml of 2% Ca(OH)₂ for fifteen/ h. Hair was manually removed and residual fat and meat were trimmed using a knife and scraper. Washing continued until the pH of squeezed water reached ~6.7. Un-haired skins were cut into ~2×1×1/ cm pieces, placed in polyethylene bags and frozen at -20°C for fifteen/ h. After thawing at 5°C, pieces were soaked in 1% HCl (1:10) for fifteen min. Skin pieces became thicker (swollen) and lighter in color after acid treatments. They were washed thoroughly with water until near neutrality (pH 6.8). Gelatin was extracted from skin in DDW (1:10) at 60°C for six/ h in a digital water bath, with occasional stirring. The extract was filtered through a five-layer cheesecloth to remove residues. Extracted clear gelatin was lyophilized at -55°C

(1.3 mBar) using a FreeZone Triad Freeze Dry System (Labconco, MO, USA), then oven-dried at 55°C until constant weight (<2% moisture). Final gelatin weight was recorded.

Extracted gelatin from skin

Gelatin (hereafter CG) yields from camel's skin were calculated using the formula below:

$$\text{Yield (\%)} = \frac{W2 \text{ (g)}}{W1 \text{ (g)}} \times 100$$

Where,

W1 = Original weight of skin in g.

W2 = Powdered gelatin in g.

Proximate composition of CG

Moisture, fat and ash of GC were analyzed by AOAC methods 942.05 and 920.39. Protein was determined by the Kjeldahl method using a nitrogen factor of 5.55 (Al-Hassan, 2020a; Sompie *et al.*, 2015). All tests were done in triplicate.

pH of CG

pH of triplicate 1% gelatin solutions at 55°C was measured using an ATC pH meter (ThermoScientific, USA) following US Pharmacopoeia (GMIA, 2019). The pH meter was calibrated with pH 4 and 7 buffers.

UV-Visible spectrum of CG

A 1% gelatin solution was prepared in DDW at 40°C and scanned at 200-400/ nm against a DDW blank using an Evolution 201 spectrophotometer (Thermo Scientific, USA). Commercial bovine gelatin was used for comparison.

Amino acid composition by HPLC analysis

Gelatin (80/ mg) was hydrolyzed with 10/ ml 6N HCl and 600 µl 1% phenol using a microwave digestion system at 150°C for 90/ min (Messia *et al.*, 2008; Tran Jeong 2017). Filtered samples were diluted prior to HPLC injections. Protein digestates were analyzed following a reported protocol (Henderson *et al.*, 2000). HPLC analysis was performed using an Agilent 1260 Infinity system with a quaternary pump and auto-sampler (Agilent Technologies, Santa Clara, CA, USA). A Zorbax Eclipse AAA column (3.5 µm, 4.6 × 150 mm) and OPA/FMOC derivatives of digested gelatin were used. Data were analyzed using Agilent ChemStation3 (Model G1656B). Amino acid concentrations were quantified with an Agilent amino acid standard (250/ pmol µl in 0.1 M HCl).

Turbidity of CG

Gelatin solution (6.67%) turbidity was measured at 620 nm (GMIA, 2019) using a ThermoScientific Evolution spectrophotometer.

Gel strength (bloom)

Bloom strength was tested (6.67% gelatin, 60°C and cooling to 10°C for 17h) per GMIA (2019) using a texture analyzer (TA Texture Analyser XT plus, Stable Micro System, England, UK). Triplicate experiments were conducted.

Foam expansion (FE) and stability (FS)

FE of CG (1%) was assessed (40°C, 35,000 rpm, 1 min) and compared to that of commercial bovine. FE measured volume increase and was recorded after 60 min (Abuibaid *et al.*, 2020; Shahidi *et al.*, 1995).

Fourier Transform Infrared Spectroscopy (FTIR)

FTIR spectra (4000-500 cm^{-1} , 4 cm^{-1} resolution) of CG were obtained using KBr pellets (Abuibaid *et al.*, 2020; Dai *et al.*, 2020). The FTIR Affinity-1S (Shimadzu Corporation, Japan) was used for that purpose.

Data analysis

The results obtained of triplicate tests were processed using an Excel of MS Office 365.

RESULTS AND DISCUSSION

Gelatin yield

Skin pretreatments yield two gelatin types; Type A (acid-soaked) and Type B (alkaline-soaked) (Al-Hassan, 2020a; Bahar *et al.*, 2018; GMIA, 2012).

As shown in Table 1, the translucent gelatin (type A) constituted 5.4% ± 2.11 of camel skin. The yield of CG extracted at 60°C for 6h was closer to that mentioned by other researchers (Bessalah *et al.*, 2023). Actually, there are very few reports on gelatin extraction from camel skins, by either acidic or alkaline processing of skins (Abuibaid *et al.*, 2020; Ahmed *et al.*, 2020b; Al-Hassan, 2020b; Bessalah *et al.*, 2023). Depending on extraction temperatures and time, gelatin yield range was 9.5-28.6% (Ahmed *et al.*, 2020). It was reported that 9.5% gelatin (camel skin) yield was obtained at 60°C for five hours (Abuibaid *et al.*, 2020). A similar yield (~9%) was also obtained under extraction temperature of 60°C for six hours (Bessalah *et al.*, 2023). An extreme gelatin yield (42%) from camel's skin was reported (Al-Hassan, 2020b). It was stated that extraction conditions directly affected polypeptide chain length, functional properties and other quality parameters of gelatin (Al-Hassan, 2020). Harsh extraction

conditions increased gelatin yield but reduced gel quality, while milder conditions improved gel properties despite lower yields (Abuibaid *et al.*, 2020). In fact, in the present investigation a conventional protocol (Al-Hassan, 2020b; Alipal *et al.*, 2019; Chen *et al.*, 2014; Yu *et al.*, 2016) of gelatin extraction at 60°C for six hours was adopted. Regarding other sources, % yield of skin gelatin were 16.11, 15.95, 8.49 and 23.1%, from cows (Ahmad *et al.*, 2018), goats (Fawale *et al.*, 2021), rabbits (Yu *et al.*, 2016) and fish (Sae-Leaw *et al.*, 2016), respectively.

pH of CG

The pH of gelatin solution was 3.89 $\pm$ 0.13 (Table 1). The pH of gelatin from camel skins was 5.26 recorded during extraction at 71.9°C for 3 m (Ahmed *et al.*, 2020b). The pH values of other camel skin gelatins were 7.1 to 7.6 which were higher compared to bovine skin gelatin (pH 5.45) (Al-Hassan, 2020).

UV-Visible pattern of CG

Fig 1 shows the spectrum (200-400 nm) of CG solution. The bovine one also exhibited a maximum peak at 240 nm. The chromophore groups showed absorption at 210-240 nm indicated presence of peptide bond in gelatins (Das *et al.*, 2017).

Composition of CG

Table 1 lists proximate composition of CG. Proteins were the major constituent (95.31% $\pm$ 0.94) in CG (Table 2). Their

Table 1: Yield, proximate composition, turbidity (A) and pH of camel skin gelatin (CG).

Yield (%)	5.4 $\pm$ 2.11
Composition (%)	
Protein	95.31% $\pm$ 1.94
Fat	1.19 $\pm$ 0.57
Ash	1.07% $\pm$ 0.28
Absorbance (A)	0.25 $\pm$ 0.11
pH (1% solution)	3.89 $\pm$ 0.13

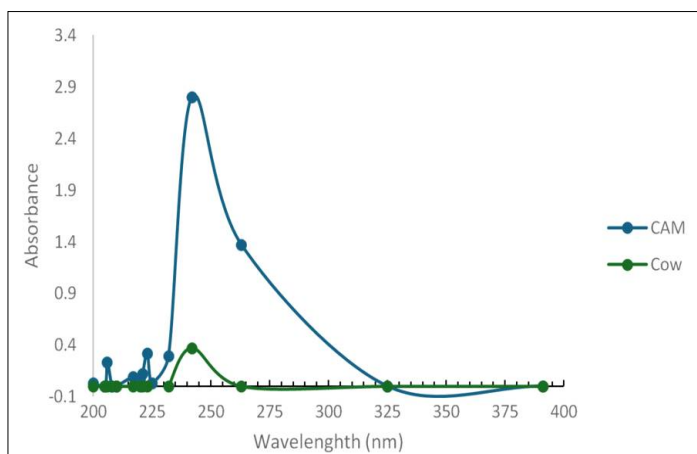


Fig 1: UV-Visible spectrum of gelatin solutions (1%) from camel's skin and a commercial bovine gelatin.

concentration was slightly higher than those reported by a previous publication where proteins ranged from 88.21 to 92.53% (Al-Hassan, 2020). From other animal species, protein contents were 89.8-91.3% for bovine, porcine and fish skin gelatins (Al-Hassan, 2020). Pretreatments of skin and extraction conditions would contribute to differences in protein contents of gelatin. Fat and ash were 1.19 and 1.07%, respectively (Table 1).

Amino acid composition by HPLC analysis

Amino acid concentrations in CG are listed in Table 2. The amino acid glycine (15.2%) was the predominant one. That corroborated previous reports where glycine was the major amino acid in gelatin extracted from camel's skin (Ahmed *et al.*, 2020; Al-Hassan, 2020). Meanwhile, glycine was also the major amino acid in gelatin from cow's skin (18.18%) and in rabbit skins (22.2%) (Ahmad *et al.*, 2018; Yu *et al.*, 2016). Besides glycine, proline represented 12.26% of CG (Table 2). It was reported that proline was the second most abundant amino acid in gelatin from different sources (Ahmed *et al.*, 2020b; Al-Hassan, 2020a; Yu *et al.*, 2016). It is noteworthy to mention that functional properties of gelatins (e.g., gel strength) were associated to amino acid composition of proteins (Al-Hassan *et al.*, 2021; Chen *et al.*, 2014; Yu *et al.*, 2016).

Clarity of gelatin

Turbidity of gelatin was related to insoluble and foreign matters (GMIA, 2012). The absorbance (A) of CG solution was 0.25 ± 0.11 (Table 3) and thus gelatin of our study was more transparent than those previously reported (Abuibaid *et al.*, 2020; Al-Hassan, 2020a). Gelatine turbidity resulted from colloid particles or aggregates (GMIA, 2012). Additionally, higher temperature and longer extraction time elevated carbonyl-amino reactions in non-enzymatic browning (Abuibaid *et al.*, 2020).

Gel strength of CG

Bloom measures gelatin strength and serves as a key quality indicator. As seen in Table 3, gel strength of CG was about 176 g. The gel strength is low (<150 g), medium (>150-220 g) and high (>220-300 g) (Ahmed *et al.*, 2020). The gel strength of gelatins from camel skin extracted under different conditions were 196-293 g (Abuibaid *et al.*, 2020). Despite the difference in the extraction conditions of camel skin, the bloom value of skin from the present study (Table 3) was very similar to that reported by Abuibaid *et al.* (2020). A highest gel strength (239 g) took place at 60°C for 5 hours. Higher temperatures and longer times reduced Bloom values (Abuibaid *et al.*, 2020). Lowest gel strength (72-122) of camel skin gelatin extracted at 75°C for 3 h (Al-Hassan, 2020b) corroborated a previous finding on detrimental effects of higher temperature on gel strength of camel's skin gelatin (Abuibaid *et al.*, 2020). The gel strengths of gelatin from other skin sources were 554.90 for bovine (Ahmad *et al.*, 2018), 320 for porcine (Chen *et al.*, 2014) and 410 for rabbit (Liu *et al.*, 2019). For commercial

demands, gelatins with gel strengths of 50-260 g are used in some food, beverages, pharmaceuticals and other applications (Ahmed *et al.*, 2020b; Alpal *et al.*, 2019).

Foaming expansion (FE) and stability (FS)

The FE of CG solution (1%) was $134\% \pm 3.5$ (Table 3). It was higher than those extracted at 60°C for 5-7h (Abuibaid *et al.*, 2020; Bessalah *et al.*, 2023). Camel skin gelatins showed higher FS values than bovine and porcine ones (Abuibaid *et al.*, 2020). A very low FE (10%) was reported for camel skin gelatins (Ahmed *et al.*, 2020). The FS of CG was $130\% \pm 14.1$ (Table 3). Lower FS values (<110) were mentioned for camel skin gelatins (Abuibaid *et al.*, 2020; Bessalah *et al.*, 2023). Previous reports mentioned % FS of 43% for bovine gelatin and 155% for goat skin gelatin (Abuibaid *et al.*, 2020). Both FE and FS were associated with protein ability to migrate, adsorb and reorient at the air-water interface (Abuibaid *et al.*, 2020). They are linked to their hydrophobic characteristics of proteins. Hydrophobic protein group increases may improve their functional properties (Ahmed *et al.*, 2020b). Commercial bovine gelatin showed higher FE (164%) than CG but lacked FS (Table 3).

Table 2: Amino acid composition of camel skin gelatin (CG).

Amino acid	Concentration (%)
Glycine (GLY)	15.3
Proline (PRO)	12.3
Glutamic acid (GLU)	10
Tyrosine (TYR)	8.6
Histidine (HIS)	7.5
Threonine (THR)	6.8
Phenylalanine (PHE)	5.2
Alanine (ALA)	4.7
Asparagine (ASP)	4.1
Lysine (LYS)	2.6
Arginine (ARG)	2.1
Cysteine (CY2)	2.1
Serine (SER)	1
Methionine (MET)	0.9
Isoleucine (ILE)	0.8
Valine (VAL)	0.7
Leucine (LEU)	0.5

Table 3: Gel strength (bloom), foaming expansion (FE) and foaming stability (FS) of CG.

Property	Camel skin gelatin (CG)	Bovine gelatin
Gel strength (bloom, g)	175.55 ± 7.12	NT ^a
Foaming expansion (%)	134 ± 3.51	164 ± 5.65
Foaming stability (%)	130 ± 14.13	- ^s

^aNot tested.

^sNo foam stability.

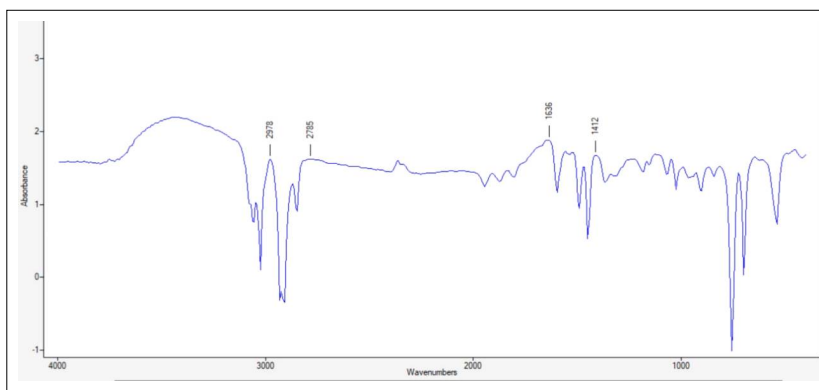


Fig 2: FTIR spectrum of gelatin from camel skins.

FTIR properties of CG

FTIR spectroscopy analyzes material structures, like gelatin, by identifying functional groups through infrared spectrum bands (Abuibaid *et al.*, 2020; Bessalah *et al.*, 2023). Proteins consist of amino acids linked by amide bonds, with polypeptides exhibiting nine characteristic IR absorption bands, amide-A, B and I-VII (Nur Hanani *et al.*, 2012). Fig 2 shows Amide A (3444 cm^{-1}), 3291 cm^{-1} , 1628 cm^{-1} and 1559 cm^{-1} bands. Gelatins from various sources showed IR absorption bands in the Amide region. The bands at 3285 , 1632 , 1550 and 1238 cm^{-1} corresponded to Amide-A, glycine/water molecules, Amide I, II and III, respectively (Nur Hanani *et al.*, 2012). Amide-A signified NH-stretching with hydrogen bonding; amide I, C=O stretching/hydrogen bonding with COO; amide II, N-H bending and C-N stretching; amide III, in-plane C-N, N-H, or CH_2 vibrations (Nur Hanani *et al.*, 2012). FTIR spectrum (Fig 2) was very similar to those reported for camel skin gelatin. They were Amide A ($3400\text{--}3300\text{ cm}^{-1}$), band at 3309.38 cm^{-1} (O-H stretching), Amide I at 1647.98 cm^{-1} resulting from C-O stretching and band at 1543.13 cm^{-1} representing amide II (-NH twisting mode) (Al-Hassan, 2020). Additionally, the presence of amide III band based on spectral features such as CN stretching vibration, NH deformation from amide linkages and wagging vibration from CH_2 groups from amino acids glycine and proline (Al-Hassan, 2020). As reported by for camel skin gelatin (Bessalah *et al.*, 2023), the absorption for hydrogen bonded N-H associated to amid A amide was at 3362 cm^{-1} . FTIR showed the absorption band at 1656 cm^{-1} for C=O stretching (Amide I). GC (Fig 2) shows camel skin gelatin with similar FTIR spectra for Amide I, II, III and Amide-A to those of a previous report (Bessalah *et al.*, 2023). Camel skin gelatins extracted under different conditions displayed a similar FTIR spectra (Abuibaid *et al.*, 2020) to those of CG (Fig 2). The Amide A band in camel gelatin ($3307\text{--}3315\text{ cm}^{-1}$) occurred from N-H stretching with H-bonding. Gelatins generally show a broad Amide A band ($3300\text{--}3400\text{ cm}^{-1}$) with free N-H stretching (Abuibaid *et al.*, 2020). The Amide-B band (2924 cm^{-1}) in camel gelatin indicated C-H and - NH^3+ stretching (Abuibaid *et al.*, 2020). The Amide I (1629

cm^{-1}) and II (1548 cm^{-1}) wavenumbers reported earlier (Abuibaid *et al.*, 2020) were very similar to those obtained from the present study on CG (Fig 2). The wavenumbers between $1238\text{--}1244\text{ cm}^{-1}$ were associated with Amide III (Abuibaid *et al.*, 2020). The Amide III indicated the α -helical transformation to a random coil structure which will result in the loss of triple helix structure. Furthermore, the amide-III peak represented a blend of N-H deformation and C-N stretching vibrations originating from amide linkages along with a response due to vibrations caused by CH_2 groups from the glycine back-bone and proline side chains (Abuibaid *et al.*, 2020). Amid A bands ($3286\text{--}3291\text{ cm}^{-1}$) and water molecules in beef, pork and Tilapia fish gelatins were reported (Nur Hanani *et al.*, 2012).

CONCLUSION

CG as an underutilized resource, offers sustainable potential for food, pharmaceutical and packaging applications. Its unique properties (clarity, gel strength, foaming) and cultural acceptability address limitations of traditional sources. CG-based films show promise for food preservation against oxidative degradation.

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Disclaimers

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Informed consent

All animal procedures for experiments were approved by the Committee of Experimental Animal care and handling

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Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this article. No funding or sponsorship influenced the design of the study, data collection, analysis, decision to publish, or preparation of the manuscript.

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