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ORIGINAL ARTICLE

Synthesis, Surface Properties and Effect of an Amino Acid Head Group of 11-(2-Methoxy-4-vinylphenoxy)undecanoicacid-Based Anionic Surfactants

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Abstract Our present research describes the surface properties of three biobased anionic surfactant synthesized from vinylguaiacol and 11-bromo undecanoic acid. To further improve its hydrophobicity and bioavailability, amino acid head group incorporation was carried out. All these synthesized compounds were thoroughly characterized using NMR and mass spectroscopy. The performance properties such as foaming, wetting, emulsification value and calcium tolerance were evaluated. The studied surfactants possess excellent emulsion stability and moderate calcium tolerance as compared to commercially available surfactant sodium lauryl sulfate (SLS). The micelle formation and the thermodynamics involved at the air–water interface were estimated from surface tension measurements. These surfactants showed a higher tendency towards adsorption at the air–water interface than micellization. Dynamic light scattering and steady state fluorescence anisotropy study were carried out to shed light on the bulk micellization properties of the synthesized surfactant. Along with spherical micelles of <5 nm size, larger aggregates (35–84 nm) were observed with higher anisotropy values. FESEM images further confirmed the larger spherical micelles formed by these surfactants. The surfactants formed chiral aggregates above the critical micelle concentration as indicated by circular dichroism spectra. These surfactants may be suitable candidates for additives to detergents to improve their calcium tolerance especially in the case of hard water. Furthermore, a low foaming ability along with high emulsion stability may find these surfactants to be better replacement of the conventional surfactant used as emulsifiers in many industrial applications.

Keywords Vinylguaiacol · Undecanoic acid · Amino acid · Surface activity · Surface tension · Micellization · Dynamic light scattering · Anisotropy

Introduction

Surface-active agents or surfactants are widely used chemicals consumed in large quantities for various applications ranging from household to industrial. Biocompatibility or biodegradability of these materials is highly essential as these products create havoc in aquatic environments after end use. Hence biodegradable surfactants made from renewable resources are in high demand. Renewable raw material such as oils, proteins and carbohydrates are the alternatives to the petroleum resources for the preparation of surfactants [1, 2].

Surfactants assemble into various aggregate structures in aqueous solution such as micelles, vesicles, tubules and rods etc. above a certain concentration called the critical micelle concentration (CMC). The nature of these self-assemblies largely depends on geometric factors such as its hydrophobic head to hydrophobic tail ratio. Depending on the chemical structure and/or the surface active properties, surfactant molecule find application in industry or in common household products [3]. Apart from the biodegradability issue, surfactant should be stable and efficient at low concentration, so that minimum amount of

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surfactant can be used for various applications there by reducing the environment concern. One approach to stabilized self-assembled structures is polymeric surfactants, where the products require minimum concentration and are highly stable to external stimuli and are concentration independent [4–6]. Polymeric surfactants are used in all the major industries such as agriculture, coatings, cosmetics, biotechnology, medicine, water purification, electronic, and enhanced oil recovery [7].

Our group has engaged in the design of new surfactant molecules from renewable and cheap resources such as vegetable oil and isolated proteins from waste biomass [8–11]. In order to coin the concept of polymeric surfactant to our vegetable oil origin surfactant, we must design a surfactant having a polymerizable group, such as vinyl moiety, in its hydrophobic tail. Decarboxylated ferulic acid is one molecule which can be coupled with any fatty acid to generate a new class of polymerizable surfactants.

Ferulic acid (FA) is hydroxycinnamic acid, considered cheap, abundant and easily available from vegetables, wheat, maize, rice bran, fruits, coffee, and peanuts, etc. [12–14]. Biochemically or chemically degradation of FA produces a volatile phenol named 4-vinylguaiacol (VG). Used as a flavoring agent, VG is more economical than ferulic acid [15, 16]. Further the phenolic group of VG can be reacted with 11-bromo undecanoic acid to produce a new kind of biocompatible surfactant.

Our current research focused on the synthesis and self-assembly of purely biobased anionic surfactants with polymerizable groups. Decarboxylated FA introduces the polymerizable group, whereas the hydrophobic chain was undecanoic acid, a derivative of castor oil. To further enhance its performance along with biocompatibility; amino acids were introduced as a polar head group. The surface active properties, in particular foaming, wetting, emulsification and calcium tolerance were studied. The self-assembly properties such as critical micelle concentration, thermodynamics of micellization were studied in aqueous solution. Further insight was provided on the bulk micelle properties by steady state fluorescence anisotropy and light scattering.

Experimental

Chemicals

Ferulic acid (99 %), 11-bromoundecanoic acid (95 %) and 1,6-diphenyl-1,3,5-hexatriene (DPH) were purchased from Sigma-Aldrich, St. Louis, USA. 1-(3-Dimethylaminopropyl)-3-ethyl carbodiimide hydrochloride (EDC.HCl) (99 %), 1-hydroxybenzotriazole (HOBt) (98 %) were purchased from Spectrochem Pvt. Ltd. Mumbai, India. Glycine (99 %), L-alanine (99 %), sodium acetate (98 %), calcium acetate, potassium carbonate anhydrous AR (99.9 %), lithium hydroxide monohydrate (99 %) were purchased from M/s SD Fine Chem. Mumbai, India. All other common chemicals and solvents were purchased locally and used after purification.

Analytical Methods

Varian spectrometer (Model: INNOVA 400, Varian, Palo Alto, USA) was used to record all ^{1}H- and ^{13}C-NMR spectra. HRMS data were recorded on a Thermo Scientific Exactive Orbitrap Mass spectrometer (Germany). ESI–MS spectra were also recorded on a Waters LC–MS mass spectrometer (Palo Alto, USA) in the EI mode and are given in mass units (m/z). Melting points were determined using a Barnstead Electro Thermal melting point apparatus.

Surface tension measurement was performed on a Krüss tensiometer (K100MK2 Tensiometer, Hamburg, Germany). The surface tension (γ) of the synthesized surfactant molecule at different concentrations was measured at 27 °C. From the break point of the plot of γ *versus* ln C critical micelle concentration (CMC) was obtained.

As per the Indian Standard specification (BIS-1185, Bureau of Indian Standards, New Delhi, 1957), the Draves-Clarkson method was used for estimating the wetting time. From unbleached grey carded Indian yarn of single 20s, skeins of 34 cm circumference weighing 5.0 ± 0.1 g were prepared. The sinking times of the skeins attached to the hook weighing 4.5 g carrying a lead anchor weighing 27.1 g were determined on surfactant solutions taken in a 500-mL measuring cylinder.

A Ross-Miles foam apparatus was used to determine the foaming power of the surfactant solutions at room temperature. The apparatus consisted of a cylindrical column of 90 cm height and 5 cm internal diameter surrounded by a jacket. The test solution was filled in a pipette (200 mL) with an orifice of 3 mm, which was supported in the above column. To the 50 mL of solution placed well in advance in the column, 200 mL solution from the pipette was poured into the same column. The foam height at different time intervals was measured using a scale attached to the column.

According to the method described by Prasad *et al*., the emulsifying power was measured for the water/liquid paraffin system [11]. To the 20 mL paraffin oil taken in a 100-mL stoppered graduated measuring cylinder, a surfactant solution of 20 mL was poured through the side wall of the measuring cylinder. For 1 minute the measuring cylinder was turned upside down 30 times. The emulsifying power was noted for separation of the phase for 10 and 20 mL respectively.

A modified Hart's method was used to determine calcium tolerance of the surfactant solution. The point of

turbidity just occurred during the titration of 0.1 % surfactant solution (50 mL) against 1 % calcium acetate solution was measured. Calcium tolerance was expressed as Ca^{2+} in ppm required to make 1 mL of the surfactant solution turbid.

The micelle size was determined by a dynamic light scattering technique using a Malvern Zetasizer Instrument (Nano-ZS90, Malvern, U.K.) having a He–Ne laser. All the surfactant solutions at a concentration of 5 times their CMC were prepared in de-ionized water. Before measurement, the solution was filtered through a 0.22-μm MF-Millipore MCE Membrane. The measurement was carried out at 25 °C with a scattering angle of 90°.

A fluorolog-3 spectrofluorometer (Horiba Jobin–Yvon, U.K.) equipped with a polarization accessory which uses the L-format instrumental configuration was used to measure the steady state fluorescence anisotropy. For the anisotropy measurement all the surfactant solutions were kept at the concentration 5 times of their respective CMC, and the DPH probe at 5 μL concentration. All the samples were kept in the dark overnight prior to their measurement. The excitation and emission wavelengths were kept at 350 and 450 nm respectively with a band pass of 5/5 nm.

The microscopic characterization was performed in a JEOL JSM-7610F, Field Emission Scanning Electron Microscopy (FESEM), operated at 10 kV. For FESEM the surfactant solutions were dropped onto small pieces of thoroughly washed glass slide and the film allowed to dry overnight in a vacuum desiccator. Prior to the measurement the glass slides were placed on a copper tape supported on a metal stub and sputter coated with gold to make it conducting. In order to obtain the particle size distribution the microscopic images were analyzed by a specialty image processing software named the Fiji version of ImageJ [17]. ImageJ was developed and maintained by the National Institute of Mental Health, Bethesda, MD.

The specific rotations were measured with an Anton Paar, MCP-200 digital polarimeter. The circular dichroism (CD) spectra were recorded with a Jasco J-850 spectropolarimeter using a quartz cell with a path length of 10 mm.

Synthesis

A new class of vinylguaiacol-based anionic surfactant was synthesized according to the synthetic scheme presented in Fig. 1.

Synthesis of 2-Methoxy-4-vinylphenol (1)

To the ferulic acid (33 g, 170 mmol) dissolved in 250 mL of DMF, sodium acetate (6.97 g, 85 mmol) was added (to facilitate decarboxylation) and the reaction mixture was refluxed for 1 h. After the completion of the reaction as monitored by TLC, the reaction mixture was diluted with ethyl acetate and washed with a saturated solution of NaCl, 5 % aq $NaHCO_3$, water and dried over anhydrous Na_2SO_4 and concentrated under vacuum. The crude product was purified by column chromatography using 2 % ethyl acetate in hexane (19.2 g, 76 %). ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.98–6.84 (m, 3H), 6.64 (dd, $J = 18.1$, 11.3 Hz, 1H), 5.64 (s, 1H), 5.59 (d, $J = 18.1$ Hz, 1H), 5.13 (d, $J = 10.5$ Hz, 1H), 3.92 (s, 3H). ESI MS: m/z 149 $[M\text{-}H]^+$.

Synthesis of Methyl 11-(2-methoxy-4-vinylphenoxy)undecanoate (2)

Methyl 11-bromoundecanoate (35 g, 126.6 mmol) and 2-methoxy-4-vinylphenol (19 g, 126.6 mmol) were dissolved in acetone (250 mL), K_2CO_3 (35 g, 253 mmol) was added to the reaction mixture and refluxed overnight. After the completion of the reaction as monitored by TLC, the solvent was removed under reduced pressure and the crude product was diluted with ethyl acetate (250 mL) and the organic layer was washed with saturated solution of NaCl and dried over Na_2SO_4. The organic layer was concentrated to dryness. Then the resulting mixture was purified by silica gel (eluent 1 % ethyl acetate in *n*-hexane) which afforded methyl 11-(2-methoxy-4-vinylphenoxy)undecanoate (35.26 g, 80 % yield) as a viscous liquid. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.00–6.89 (m, 2H), 6.82 (d, $J = 8.3$ Hz, 1H), 6.65 (dd, $J = 17.5$, 10.7 Hz, 1H), 5.61 (d, $J = 17.5$ Hz, 1H), 5.14 (d, $J = 10.7$ Hz, 1H), 4.01 (t, $J = 6.9$ Hz, 2H), 3.89 (s, 3H), 3.67 (s, 3H), 2.30 (t, $J = 7.5$ Hz, 2H), 1.90–1.77 (m, 2H), 1.69–1.59 (m, 2H), 1.51–1.21 (m, 14H). ESI MS: m/z 349.12 $[M + 1]^+$, 371.12 $[M + Na]^+$, 387.12 $[M + K]^+$.

Synthesis of 11-(2-Methoxy-4-vinylphenoxy)undecanoicacid (3)

To a solution of methyl 11-(2-methoxy-4-vinylphenoxy) undecanoate (35 g, 100.57 mmol) in THF: H_2O (7:3), $LiOH.H_2O$ (1.07 g, 25.57 mmol) was added and the reaction mixture was stirred at room temperature for 4 h. After completion of reaction, the solvent was removed under reduced pressure, diluted with ethyl acetate and neutralized with 2 N HCl and the organic layer was separated and washed with water, dried using anhydrous Na_2SO_4. The solvent was evaporated under reduced pressure and the crude product was subjected for column chromatography using silica-gel (10–30 % ethyl acetate: *n*-hexane) to obtain the pure compound (30.23 g, 90 %, m.p. 40 °C). ^{1}H NMR (500 MHz, $CDCl_3$) δ 6.97 (d, $J = 1.98$ Hz, 1H), 6.92 (dd, $J = 8.2$, 1.9 Hz, 1H), 6.82 (d, $J = 8.2$ Hz 1H), 6.65 (dd, $J = 17.7$, 10.9 Hz, 1H), 5.61 (dd, $J = 17.5$, 0.7 Hz, 1H), 5.14 (dd, $J = 10.8$, 0.6 Hz, 1H), 4.01 (t,

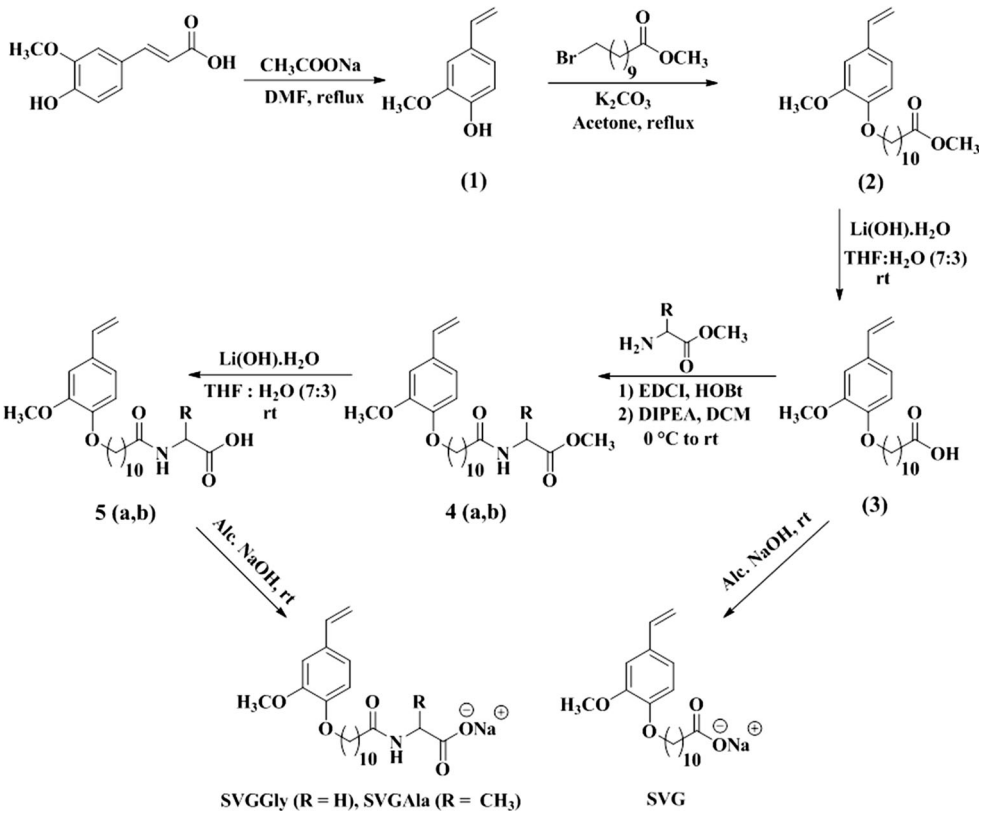

Fig. 1 Schematic representation of synthesis of the 11-(2-methoxy-4-vinylphenoxy)undecanoic acid-based surfactant

$J = 7.01$ Hz, 2H), 3.89 (s, 3H), 2.35 (t, $J = 7.4$ Hz, 2H), 1.87–1.80 (m, 2H), 1.66–1.59 (m, 2H), 1.48–1.40 (m, 2H), 1.39–1.24 (m, 12H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 179.96, 149.39, 148.57, 136.56, 130.64, 119.48, 112.68, 111.70, 109.05, 69.06, 55.98, 34.04, 29.48, 29.37, 29.23, 29.16, 29.06, 25.95, 24.70. ESI MS: *m/z* 332.99 $[M\text{-}H]^+$. HRMS (*m/z*) calculated for $C_{20}H_{31}O_4$, $[M]^+$ 335.2216, found 335.2212.

General Procedure for the Synthesis of Peptide Coupling Reactions (4)

To a solution of 11-(2-methoxy-4-vinylphenoxy) undecanoic acid (5 g, 15 mmol) in dry CH_2Cl_2 (50 mL) at 0 °C, EDCI (3.16 g, 16.47 mmol), HOBt (2.22 g, 16.47 mmol) and DIPEA (2.87 mL, 16.47 mmol) were added at the same temperature and the reaction mixture was stirred for 10 min. Then a solution of pre-neutralized amino acid methyl ester (16.47 mmol) in dry CH_2Cl_2 was added to the reaction mixture at 0 °C and stirred. After completion of the reaction as monitored by TLC, the reaction mixture was diluted with CH_2Cl_2 and washed with dil. HCl (100 mL ×2), 10 % Na_2CO_3 (100 mL ×2), water and saturated solution of NaCl, dried over anhydrous Na_2SO_4 and concentrated under vacuum. The crude product was subjected to column chromatography using silica-gel (1–5 % methanol: chloroform) to obtain the pure compound.

Methyl2-(11-(2-methoxy-4-vinylphenoxy) undecanamido)acetate (**4a**).Yield 82 %, (m.p. 82 °C)

^{1}H NMR (300 MHz, $CDCl_3$) δ 7.01–6.89 (m, 2H), 6.82 (d, $J = 8.1$ Hz, 1H), 6.65 (dd, $J = 17.5$, 10.7 Hz, 1H), 5.94 (br, s, 1H), 5.61 (d, $J = 17.5$ Hz, 1H), 5.14 (d, $J = 10.7$ Hz, 1H), 4.06 (d, $J = 5.1$ Hz, 2H), 4.01 (t, $J = 6.9$ Hz, 2H), 3.89 (s, 3H), 3.77 (s, 3H), 2.24 (t, $J = 7.3$ Hz, 2H), 1.89–1.77 (m, 2H), 1.71–1.55 (m, 2H), 1.51–1.21 (m, 14H). ESI MS: *m/z* 406.41 $[M + 1]^+$, 428.36 $[M + Na]^+$.

Methyl2-(11-(2-methoxy-4-vinylphenoxy) undecanamido)propanoate (**4b**), Yield 80 % (m.p. 66 °C) ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.97 (d, $J = 1.8$ Hz, 1H), 6.93 (dd, $J = 8.3$, 2.0 Hz, 1H), 6.82 (d, $J = 8.3$ Hz, 1H), 6.65 (dd, $J = 17.7$, 10.7 Hz, 1H), 5.98 (d, $J = 5.8$ Hz, 1H), 5.61 (d, $J = 17.7$ Hz, 1H), 5.14 (d, $J = 11.1$ Hz, 1H), 4.67–4.55 (m, 1H), 4.01 (t, $J = 6.7$ Hz, 2H), 3.89 (s, 3H), 3.75 (s, 3H), 2.20 (t, $J = 7.3$ Hz, 2H), 1.89–1.77 (m, 2H), 1.69–1.53 (m, 2H), 1.51–1.24 (m, 17H). ESI MS: *m/z* 420.49 $[M + 1]^+$, 442.44 $[M + Na]^+$, 458.4 $[M + K]^+$.

General Procedure for the Hydrolysis of Ester to Acid (5)

To a solution of compound **4** (5 g, 12.78 mmol) in THF: H_2O (7:3), $LiOH.H_2O$ (1.07 g, 25.57 mmol) was added and the reaction mixture was stirred at room temperature for

4 h. After the completion of reaction, the solvent was removed under reduced pressure, diluted with chloroform and neutralized with 2 N HCl and the organic layer was separated and washed with water, dried using anhydrous Na_2SO_4. The solvent was evaporated under reduced pressure and the crude product was subjected to column chromatography using silica-gel (5–10 % methanol: chloroform) to obtain the pure compound.

2-(11-(2-Methoxy-4-vinylphenoxy) undecanamido)acetic acid (**5a**) (VGGly) Yield 95 %, (m.p. 96 °C) ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.01–6.88 (m, 2H), 6.82 (d, $J = 8.1$ Hz, 1H), 6.64 (dd, $J = 17.5$, 10.7 Hz, 1H), 6.39 (s, 1H), 5.61 (d, $J = 17.5$ Hz, 1H), 5.14 (d, $J = 10.7$ Hz, 1H), 4.05 (d, $J = 5.1$ Hz, 1H), 4.00 (t, $J = 6.9$ Hz, 2H), 3.88 (s, 3H), 2.26 (t, $J = 7.3$ Hz, 2H), 1.90–1.74 (m, 2H), 1.71–1.54 (m, 2H), 1.50–1.21 (m, 13H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 174.65, 172.57, 149.33, 148.53, 136.53, 130.68, 119.53, 112.72, 111.76, 109.10, 69.10, 55.99, 41.54, 36.28, 29.42, 29.35, 29.23, 29.14, 25.94, 25.54. ESI MS: *m/z* 390.41 $[M\text{-}H]^+$. HRMS (*m/z*) calculated for $C_{22}H_{34}O_5N$, $[M]^+$ 392.2431, found 392.2428.

2-(11-(2-Methoxy-4-vinylphenoxy) undecanamido)propanoicacid (**5b**) (VGAla), Yield 80 %, (m.p. 86 °C) ^{1}H NMR (500 MHz, $CDCl_3$) δ 6.97 (d, $J = 1.9$ Hz, 1H), 6.93 (dd, $J = 8.2$, 1.8 Hz, 1H), 6.82 (d, $J = 8.2$ Hz, 1H), 6.65 (dd, $J = 17.5$, 10.8 Hz, 1H), 6.11 (s, 1H), 5.61 (dd, $J = 17.5$, 0.6 Hz, 1H), 5.14 (dd, $J = 10.9$, 0.6 Hz, 1H), 4.58 (p, $J = 7.1$ Hz, 1H), 4.01 (t, $J = 6.8$ Hz, 2H), 3.89 (s, 3H), 2.4 (t, $J = 7.6$ Hz, 2H), 1.89–1.79 (m, 2H), 1.67–1.59 (m, 2H), 1.49–1.39 (m, 5H), 1.38–1.24 (m, 11H). ^{13}C NMR (75 MHz, $CDCl_3$) δ 175.58, 174.02, 149.34, 148.54, 136.54, 130.67, 119.51, 112.71, 111.74, 109.10, 69.09, 55.98, 48.29, 36.43, 29.40, 29.34, 29.22, 29.12, 25.93, 25.52, 18.01. ESI MS: *m/z* 404.53 $[M\text{-}H]^+$. HRMS (*m/z*) calculated $C_{23}H_{34}O_5N$, $[M]^+$ 404.2431, found 404.2432.

General Procedure for the Preparation of Sodium Salts of Compounds

The compounds **3** and **5(a, b)** were dissolved in ethanol and equivalent ethanolic NaOH was added. The content was stirred for overnight at room temperature and the solvent was evaporated under vacuum to get corresponding sodium salt: Sodium 11-(2-methoxy-4-vinylphenoxy) undecanoate (SVG), Sodium 2-(11-(2-methoxy-4-vinylphenoxy)undecanamido)acetate (SVGGly), Sodium 2-(11-(2-methoxy-4-vinylphenoxy)undecanamido)propanoate(SVGAla). The specific rotation of the chiral SVGAla is found to be $[\alpha]_D = -0.12°$ (H_2O).

Results and Discussion

A new class of biobased anionic surfactant was synthesized taking vinylguaiacol (decarboxylated ferulic acid) and 11-bromoundecanoic acid as starting materials. To further improve its solubility and bioavailability amino acid was coupled to its head group. The synthesized surfactants were well characterized through ^{1}H- and ^{13}C-NMR, mass spectrometry (All the spectra are available as electronic supplementary material).

Performance Properties

In order to check the industrial feasibility of these vinylguaiacol-based anionic surfactants as replacements for petroleum and/or other commercial surfactants, performance properties such as wetting ability, emulsion stability, foaming and calcium tolerance were evaluated and these are presented in Table 1.

One of the major criteria for choosing a surfactant to be used in household or in industrial process is its wetting ability. Hence we study the wetting properties of the synthesized vinylguaiacol-based surfactants and compared with commercially available sodium lauryl sulfate (SLS). All of the vinylguaiacol-based surfactants show poor wetting properties as compared with SLS. Among the three surfactants, SVG showed better wetting ability. Introduction of amino acids head group further decreases its wetting ability.

Superior emulsification of oil in water makes them suitable candidates for the food/pharmaceutical/petroleum industries. Better emulsion stability was observed comparing the emulsion stability of these synthesized surfactants with SLS. For separation of 10 mL emulsions, SVG took almost double the time taken for SLS. Amino acid head group further strengthen the emulsion stability. Increase in hydrophobic nature of the amino acid head group further boosted the emulsion stability as was observed in case of SVGAla (243 s for 20-mL separation).

The surfactants possess very low foam heights and these are also unstable. Hence we could not measure any foam height for SVG and SVGGly. The more hydrophobic SVGAla shows little foaming, which is slightly stable. Low foaming surfactants are really useful in emulsion polymerization, waste water treatment and even in industrial cleaning [18].

Cleaning/washing in variable environments is related to the resistance of the surfactant to the hardness of the water. Hence the calcium tolerance of these surfactants was studied. All these surfactants showed better calcium tolerance value as compared to the commercial surfactant. It

Table 1 Surface active properties of the surfactants

Surfactant	Wetting time (s)	Emulsion stability time (s)		Foam height (cm)			Calcium tolerance (ppm)
		10 mL	20 mL	0 min	5 min	10 min	
SVG	69	99	181	–	–	–	18.98
SVGGly	293	97	230	–	–	–	18.98
SVGAla	191	111	243	9.5	6	5	82.3
SLS	14	53	130	13.5	13	12.5	11.39

is interesting to observe that the SVGAla showed much higher calcium tolerance value.

Surface Properties

Monolayer formation at the air–water interface and micelle formation in the bulk aqueous solution were determined by surface-tension measurement. Figure 2 shows the plots of surface tension (γ) *versus* log (concentration) of the synthesized surfactants. The critical micelle concentrations (CMC) were obtained from the γ *versus* the ln C plot and are presented in Table 2. SVG (7.56 mM) showed a similar CMC value to the commercially available surfactant SLS (~8 mM) whereas SVGGly (3.08 mM) showed lower CMC values.

As was observed from the performance property studies, all the surfactants showed moderate surface activity. The observed surface tension values at the CMC (γ_{CMC}) were in the range of 38.45–43.79 mN/m, which was not highly surface active in nature. Introduction of an amino acid head group enhanced the surface activity as evidenced by their higher (>3) pC_{20} value [19]. pC_{20} is defined as the negative logarithm of the surfactant concentration required to reduce the surface tension of water by 20 units. While comparing the amino acid head group, the more polar glycine showed

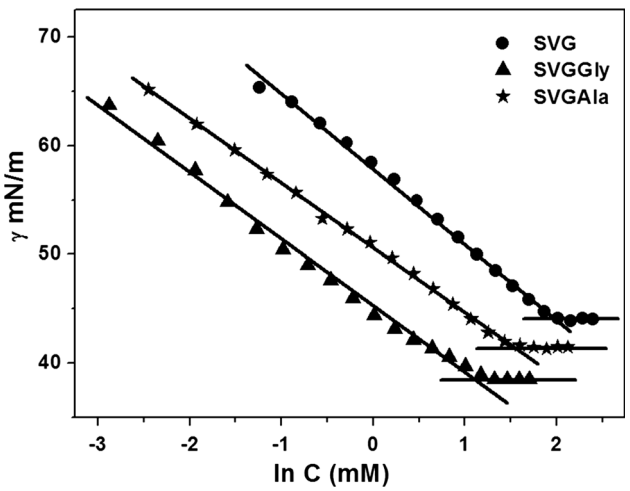

Fig. 2 Surface tension *versus* concentration of surfactants in aqueous solution

better surface activity ($\gamma_{CMC} = 38.45$ mN/m and $pC_{20} = 3.48$) than alanine ($\gamma_{CMC} = 41.36$ mN/m and $pC_{20} = 3.11$). [20, 21].

Using Gibb's adsorption isotherm (Eq. 1) two important thermodynamic parameter such as; the maximum surface excess concentration (Γ_{max}) and minimum surface area per surfactant molecule (A_{min}) at the air–water interface was calculated from the linear part of surface tension plot [22].

$$\Gamma_{max} = -(1/nRT)(d\gamma/d\ln C) \quad (1)$$

$$A_{min} = 1/N \cdot \Gamma_{max} \quad (2)$$

where R is the gas constant (8.314 J/mol.K), T is the absolute temperature, γ is the surface tension, C is the surfactant concentration and N is Avogadro's number. As there is one counter ion that is associated with one ionic head group, the value of n is taken to be 2 [19]. The degree of packing and the orientation of the surfactant molecule at the air–water interface were obtained from the Γ_{max} and A_{min} values. SVGAla showed lower Γ_{max} and higher A_{min} whereas SVG showed higher Γ_{max} and lower A_{min} at the air–water interface area. This might be due to the bulkier group (alanine) causing less order packing in the air–water interface during the formation of Gibb's monolayer.

The standard free energy of micellization ($\Delta G°_{mic} = RT$ ln CMC) in the bulk aqueous solution and the standard free energy of adsorption ($\Delta G°_{ads} = \Delta G°_{mic} - (\Pi_{CMC}/\Gamma_{max})$ at the air–water interface were also calculated [23, 24]. Where R is gas constant 8.314 J/mol K and T is absolute temperature and the surface pressure at CMC; Π_{CMC} ($\Pi_{CMC} = \gamma_{water} - \gamma_{CMC}$). The adsorptions at the air–water interface as well as the micellization in the bulk aqueous phase are a spontaneous process at 27 °C as observed by the negative sign (Table 2). All these three surfactant showed higher tendency towards adsorption than micellization. The comparatively lower CMC surfactant, SVGGly showed higher standard free energy of micellization ($\Delta G°_{mic} = -28.85$ kJ/mol).

Dynamic Light Scattering

The surfactant in aqueous solution above the CMC forms micelles of various sizes and shapes. The aggregated

Table 2 Thermodynamic properties and anisotropy value of the surfactants

Surfactant	CMC (mM)	γ_{CMC} (mN/m)	pC_{20}	$\Gamma_{max} \times 10^{12}$ mol/mm^2	A_{min} (nm^2/molecule)	ΔG°_{mic} (kJ/mol)	ΔG°_{ads} (kJ/mol)	Anisotropy (r)
SVG	7.56	43.79	2.64	2.77	0.59	−24.37	−34.55	0.074
SVGGly	3.08	38.45	3.48	2.45	0.67	−28.85	−42.54	0.106
SVGAla	4.31	41.36	3.11	2.38	0.69	−27.17	−40.04	0.144

micelle size can be obtained through dynamic light scattering (DLS) technique. In order to shed light on the various morphologies formed by the synthesized surfactant in aqueous solution, concentration dependent aggregate size measurement were carried out and presented in Table 3. All the surfactants showed mostly 3 kinds of aggregates in the solution. The first category; size ≤5 nm (around 10–25 % abundance) was found which corresponds to the typical spherical micelle. The second category is the most populated (63–84 %) aggregates around 35–84 nm in size, which is slightly bigger than the conventional spherical micelle. A similar kind of bigger spherical micelle of 50–200 nm size was reported for lauryl ester of L-phenylalanine and lauryl ester of L-tyrosine in aqueous solution [25]. The third category is the ill aggregates formed by the synthesized surfactant (>100 nm) in aqueous solution whose population is ∼10 %. As expected, incorporation of amino acid head group increases the size of the aggregates. While comparing among the two amino acid head group surfactant, SVGAla has lower aggregate size than SVGGly. The size reduction of SVGGly to SVGAla may be due to the bulkiness of the amino acid side chain and the spatial orientation of the stereogenic centers, which affect the extent of close packing of the surfactant molecule during self-assembly formation. Similar size reduction was observed from 151 nm to 135 nm for sodium *N*-[4-(*n*-dodecyloxy)benzoyl]-L-leucinate to sodium *N*-[4-(*n*-dodecyloxy)benzoyl]-L-isoleucinate surfactant [26].

In order to investigate the packing of the hydrophobic chain of these surfactants steady state anisotropy measurements were carried out using DPH as an anisotropy probe. The fluorescence depolarization of a probe molecule can be expressed in terms of anisotropy (r) as follows:

$$r = (I_{VV} - GI_{VH}) / (I_{VV} + 2GI_{VH}) \quad (3)$$

where I_{VV} and I_{VH} are the fluorescence intensities polarized parallel and perpendicular to the excitation light, and G is the instrumental correction factor ($G = I_{VV}/I_{VH}$). The anisotropy values obtained for this anionic surfactant were presented in Table 2. It has been observed that the rigidness increases with the incorporation of the amino acid head group, thus by increasing the anisotropy value. The higher anisotropy value obtained for SVGAla (0.144) is may be due to the compact packing of the hydrophobic chain in a single micelle. The bulky head groups having stereogenic centers may repeal each other and force the hydrophobic tail to have compact packing.

The morphology of the aggregates formed by the surfactant was observed by field emission electron microscopy. The microscopic images along with the particle distribution analysis were presented in Fig. 3. Spherical micelle of little bigger size was observed for all the three surfactant, which further confirms the findings of DLS experiment. The average micelle size of SVGAla (22.9 nm) is comparatively smaller than the SVGGly (37.38 nm) micelle. The size reduction in the of case

Table 3 The size of the surfactant micelles at various concentrations

Sample	Concentration (mM)	Size (d, nm)		
	15.0	2.99 (15.7 %)	37.23 (67.3 %)	179.65 (17.0 %)
SVG	22.5	3.46 (19.9 %)	43.40 (72.1 %)	306.6 (8.0 %)
	30.0	3.37 (25.3 %)	34.84 (64.0 %)	470.5 (10.7 %)
	37.5	3.25 (23.0 %)	43.05 (63.1 %)	243.7 (13.9 %)
	6.0	3.40 (19.7 %)	82.13 (63.5 %)	103.5 (16.8 %)
SVGGly	9.0	5.18 (16.4 %)	78.01 (66.1 %)	249.7 (17.5 %)
	12.0	2.53 (17.5 %)	83.92 (65.9 %)	100.06 (16.6 %)
	15.0	3.07 (24.8 %)	80.50 (75.2 %)	
	8.6	2.70 (15.6 %)	57.99 (84.4 %)	
SVGAla	12.9	2.83 (9.6 %)	59.88 (82.4 %)	894.7 (8.0 %)
	17.2	2.77 (16.3 %)	56.06 (83.7 %)	
	21.5	2.37 (17.5 %)	56.32 (82.5 %)	

SVGAla is correlated with the stereogenic center present in the surfactant head group, which affect the aggregation formation.

The effect of the stereogenic center present in the surfactant head group on the aggregation behavior in aqueous solution was monitored by circular dichroism (CD) spectra measurement. The CD spectra of the aqueous surfactant solutions were recorded above and below its CMC. The CD spectra plotted as [θ] *versus* wavelength for 42.6 mM SVG, 17.1 mM SVGGly, 21.9 mM SVGAla and 0.87 mM SVGAla (below CMC) are presented in Fig. 4. We also recorded the CD spectra for 1.7 mM SVG and 0.68 mM SVGGly (both are below the CMC), but for clarity purpose the spectra were excluded from the figure. Two distinct CD bands were appeared at 218 nm and 236 nm for concentration above the CMC of SVGAla. No such CD band appeared for SVGAla, when the measurement was performed below its CMC as shown in the figure. Hence it is

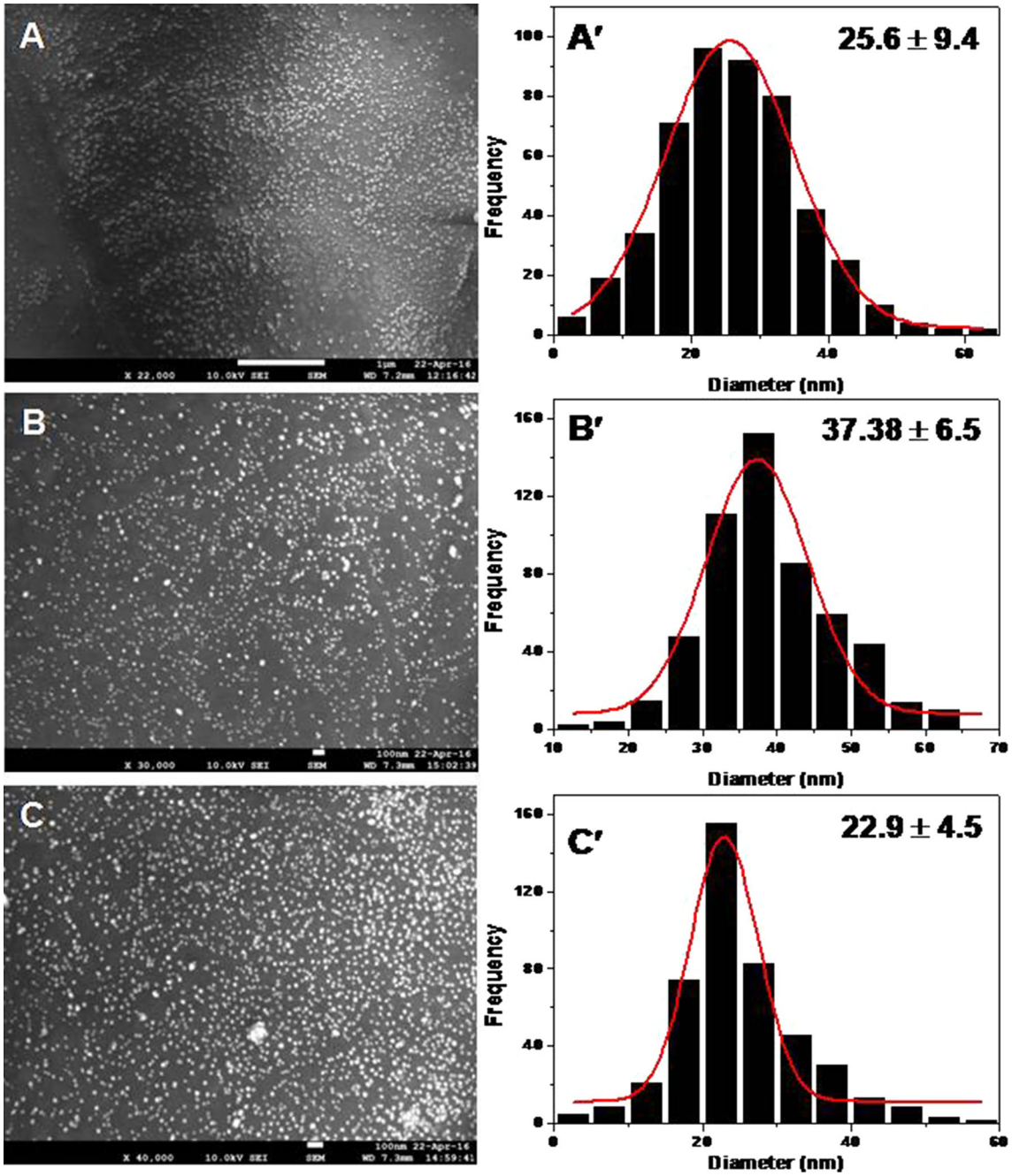

Fig. 3 FESEM images and their corresponding ImageJ analyzed histogram plot for 42.6 mM SVG (A and A′), 17.1 mM SVGGly (B and B′), 21.9 mM SVGAla (C and C′)

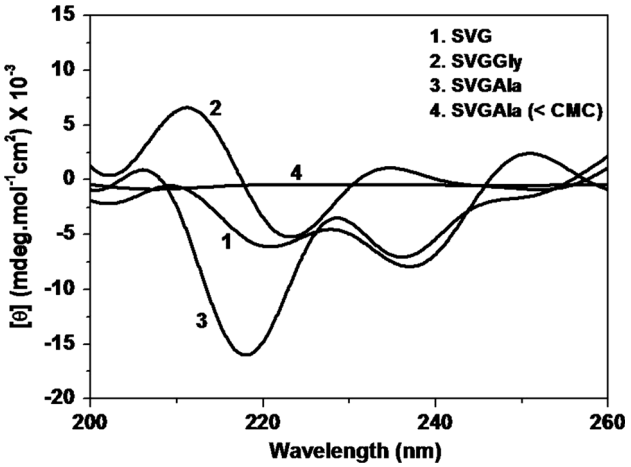

Fig. 4 Circular dichroism spectra of 42.6 mM SVG (*1*), 17.1 mM SVGGly (*2*), 21.9 mM SVGAla (*3*) and 0.87 mM SVGAla in aqueous solution

noteworthy to mention the CD band that appeared for SVGAla is due to chiral aggregation formed in aqueous solution well above its CMC. Similarly for higher concentrations, SVG showed CD bands at 221 and 237 nm whereas SVGGly displayed a band at 223 nm. These CD bands were not observed, when the spectra recorded for the concentration below its CMC.

The CD band that appeared for this achiral glycine bearing SVGGly and the precursor SVG surfactants may be due to the secondary structure obtained in solution well above its CMC. The intermolecular hydrogen bonding between amide groups may be the solely responsible factor for this kind of chiral aggregate formation. As expected, the extent of molar ellipticity is observed to be higher in the case of SVGAla as compared to SVG and SVGGly, due to the presence of stereogenic center. The spatial orientation of the stereogenic centers affects the close packing of the SVGAla molecule during aggregate formation, and hence reduced the size of the micelle as observed in DLS and FESEM experiments.

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References

1. Nnanna IA, Xia J (1998) Protein based surfactants: synthesis, physicochemical properties, and applications. surfactant science series, vol. 101, Marcel Dekker, New York, USA
2. Hill K (2007) Industrial development and application of biobased oleochemicals. Pure Appl Chem 79:1999–2011
3. Zana R (2005) Dynamics of surfactant self-assemblies: micelles, microemulsions, vesicles, and lyotropic phases. Surfactant Science Series, vol. 125, CRC Press, Taylor & Francis Group, USA
4. Nayak RR, Roy S, Dey J (2006) Characterization of polymeric vesicles of poly(sodium 11-acrylamidoundecanoate) in water. Colloid Polym Sci 285:219–224
5. Roy S, Nayak RR, Dey J (2006) Stable vesicle formation through intra- and inter-chain aggregation of poly[sodium N-(11-acrylamidoundecanoyl)-l-valinate] in aqueous solution. Colloids Surf A 290:62–69
6. Nayak RR, Roy S, Dey J (2005) Characterization and self-assembly of poly [sodium N-(11-acrylamidoundecanoyl)-L-alaninate] in water. Polymer 46:12401–12409
7. Raffa P, Wever DAZ, Picchioni F, Broekhuis AA (2015) Polymeric surfactants: synthesis, properties, and links to applications. Chem Rev 115:8504–8563
8. Sreenu M, Prasad RBN, Sujitha P, Kumar CG (2015) Synthesis and surface properties of sodium *N*-acyl phenylalanines and their cytotoxicity. Ind Eng Chem Res 54:2090–2098
9. Sreenu M, Rao BVSK, Prasad RBN, Sujitha P, Kumar CG (2014) Synthesis, surface and biological properties of sodium *N*-acyl isoleucines. Eur J Lipid Sci Tech 116:193–206
10. Kamalakar K, Satyavani T, Mohini Y, Prasad RBN, Karuna MSL (2014) Synthesis of thumba, castor and sal fatty ethanolamide-based anionic surfactants. J Surfact Deterg 17:637–645
11. Adewuyi A, Oderinde RA, Rao BVSK, Prasad RBN (2012) Synthesis of alaknolamide: a Nonionic surfactant from the oil of gliricidia sepium. J Surfact Deterg 15:89–96
12. Rosazza JPN, Huang Z, Dostal L, Volm T, Rousseau B (1995) Review: biocatalytic transformations of ferulic acid: An abundant aromatic natural product. J Ind Microbiol 15:457–471
13. Walton NJ, Narbad A, Faulds CB, Williamson G (2000) Novel approaches to the biosynthesis of vanillin. Curr Opin Biotechnol 11:490–496
14. Mariod AM, Adamu HA, Ismail M, Ismail N (2010) Antioxidative effects of stabilized and unstabilized defatted rice bran methanolic extracts on the stability of rice bran oil under accelerated conditions. Grasas Aceites 61:409–415
15. Mathew S, Abraham TE (2006) Bioconversions of ferulic acid, an hydroxycinnamic acid. Crit Rev Microbiol 32:115–125
16. Terpinc P, Polak T, Šegatin N, Hanzlowsky A, Ulrih NP, Abramovic H (2011) Antioxidant properties of 4-vinyl derivatives of hydroxycinnamic acids. Food Chem 128:62–69
17. Schindelin J, Arganda-Carreras I, Frise E et al (2012) Fiji: an open-source platform for biological-image analysis. Nat Methods 9:676–682
18. Azira H, Tazerouti A, Canselier JP (2008) Study of foaming properties and effect of the isomeric distribution of some anionic surfactants. J Surf Deterg 11:279–286
19. Rosen MJ (2004) Surfactants and interfacial phenomena, 4th edn. Wiley Interscience, New York
20. Gad EAM, El-Sukkary MMA, Ismail DA (1997) Surface and thermodynamic parameters of sodium *N*-acyl sarcosine surfactant solutions. J Am Oil Chem Soc 74:43–47
21. Negm NA, Aiad IA (2007) Synthesis and characterization of multifunctional surfactants in oil-field protection applications. J Surfact Deterg 10:87–92
22. Chu B, Zhou Z (1996) Physical chemistry of polyalkylene block copolymer surfactants. In: Nace VM (ed) Surfactant science series. Nonionic Surfactants, Marcel Dekker, pp 60–67
23. Mukherjee P (1967) The nature of the association equilibria and hydrophobic bonding in aqueous solutions of association colloids. Adv Colloid Interface Sci 1:242–275

24. Moulik SP, Haque ME, Jana PK, Das AR (1996) Micellar properties of cationic surfactants in pure and mixed states. J Phys Chem 100:701–708
25. Vijay R, Baskar G, Mandal AB, Polavarapu PL (2013) Unprecedented relationship between the size of spherical chiral micellar aggregates and their specific optical rotations. J Phys Chem A 117:3791–3797
26. Mohanty A, Dey J (2007) Effect of the headgroup structure on the aggregation behavior and stability of self-assemblies of sodium *N*-[4-(*n*-dodecyloxy)benzoyl]-L-aminoacidates in water. Langmuir 23:1033–1040

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