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# Amino Acid-Based Surfactants as Drug Carriers for the Nonsteroidal Anti-Inflammatory Drug (NSAID) Ibuprofen

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**Abstract:** The development of drug delivery and drug targeting systems for nonsteroidal anti-inflammatory drugs (NSAID) has been an active area of research for several years. The interaction of ibuprofen with $C_{12}$, $C_{14}$ quaternary ammonium surfactants (QUATs) derived from phenylalanine and proline, a proline-derived anionic carboxylate surfactant, as well as cetyltrimethylammonium bromide (CTAB) and sodium dodecyl sulfate (SDS), were carried out by fluorescence spectroscopy. The interactions of CTAB and surfactants **2** and **3** with ibuprofen were mainly hydrophobic, while surfactant **1** interacted via van der Waals forces and hydrogen bonding. Surfactants **2** and **3** showed optimum binding ability with ibuprofen. The antagonistic mixed system of **2** and **3** containing the surfactant **2** at a mole fraction of 0.4 displayed the highest binding ability. The mixture showed the highest membrane permeability at the same mole fraction, showing that this mixture is a promising ingredient for formulating a transdermal drug delivery system.

**Keywords:** Quaternary ammonium surfactants; phenylalanine; proline; mixed surfactant system; ibuprofen; fluorescence spectroscopy; membrane permeability.



## 1. Introduction

Nonsteroidal anti-inflammatory drugs (NSAIDs) are the most widely used painkillers in the world [1-3]. They are known for their anti-inflammatory, analgesic, and antipyretic properties and are used to soothe sprains, arthritis, and other daily discomforts [4,5]. Ibuprofen, chemically called 2-(4-isobutylphenyl) propanoicacidone, is one of the oldest orally administered NSAIDs used extensively for treating fever, pains, and other inflammatory conditions [6,7].

Apart from their benefits, NSAIDs have some major shortcomings [8-10]. These include low drug solubility and permeability and gastrointestinal issues such as vomiting, diarrhea, constipation, gastric ulcers, and even stomach bleeding. They are also nephrotoxic, leading to kidney injury and damage. For these reasons, several researchers have tried to reduce the undesirable side effects of NSAIDs by developing proper drug delivery systems, thus ultimately improving patient compliance in pharmacotherapy [11,12]. Drug delivery systems like microspheres, nanoparticles, dendrimers, and liposomes have been successful in overcoming the unwanted side effects of NSAIDS.

Surfactants are a class of compounds that are effective drug carriers because of their ability to form micellar aggregates in aqueous solutions. They can also act as solubilizers,

wetting agents, emulsifiers, and permeability enhancers. Cetyltrimethylammonium bromide (CTAB) and sodium dodecyl sulfate have been employed to increase the permeation rates of numerous drugs via transdermal absorption [13,14]. Surfactants can alter the permeability characteristics of the stratum corneum, that is, the outer layer of the skin, acting like penetration enhancers that interact with components of the skin to promote drug flux, hence aiding in transdermal drug delivery [15,16]. Recently, Akram *et al.* (2018) reported the interaction of oxy-diester functionalized Gemini surfactants containing betaine-type ethylene oxide units as spacers with ibuprofen, and they found that the surfactants interacted with ibuprofen via van der Waals forces, and hydrogen bonding [17].

Surfactants derived from amino acids are alternatives to conventional surfactants due to their biodegradability, lower toxicity, and biocompatibility [18-24]. They have been reported to show interesting biological properties, such as antibacterial activities. However, there have been limited studies on the interaction of amino acid-based surfactants with NSAIDs for their application as drug carriers.

Recently, our group reported the synthesis, physicochemical, and biological activities of quaternary ammonium surfactants (QUAT) derived from phenylalanine and proline (**1** and **2**) (Figure 1) [25]. These QUATs were found to show interesting physicochemical properties as a single and mixed system with sodium dodecyl sulfate (SDS) and the proline-derived anionic carboxylate surfactant (**3**). The QUATs were also found to possess relatively low cytotoxicity and showed good membrane permeability, rendering them suitable for use in skin products.

In the present work, the interaction of ibuprofen with **1**, **2,** and **3,** as well as the conventional surfactants SDS and CTAB were carried out by fluorescence spectroscopy. The interaction of mixed surfactant systems with ibuprofen was also carried out. The modes of interactions were determined using thermodynamic measurements. The effect of the different surfactants on the membrane permeability of ibuprofen was also studied to investigate their potential as penetration enhancers in drug delivery systems.

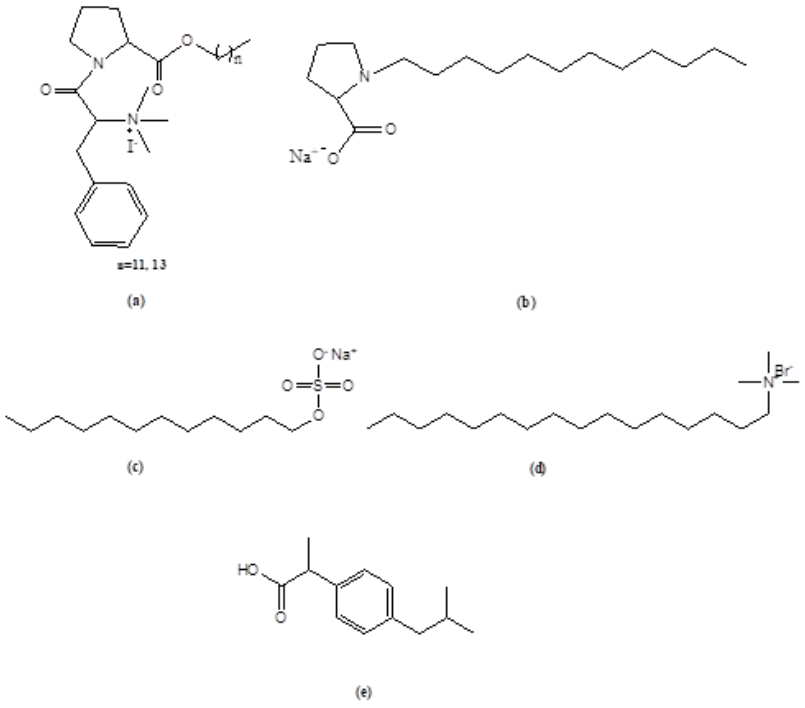


**Figure 1**. Chemical structures of the surfactants **(a)** 1 (n=11), 2 (n=13); **(b)** 3; **(c)** SDS; **(d)** CTAB; **(e)** (*S*)-ibuprofen.

## 2. Materials and Methods

(*S*)-Ibuprofen was purchased from Sigma Aldrich (USA). Cetyl trimethyl ammonium bromide (CTAB) and sodium dodecyl sulfate (SDS) were purchased from BDH Laboratory Supplies (Uganda). The QUATs derived from phenylalanine and proline, **1** and **2,** and proline-derived anionic carboxylate surfactant, **3** were synthesized following the procedure reported earlier [25,26].

UV absorbances were recorded on a Biochrom Libra S22 UV-Vis spectrophotometer. Conductivity measurements were obtained using a Jenway 4320 conductivity meter. Fluorescence spectrophotometry was performed using a Perkin-Elmer LS-55 spectrophotometer.

### *2.1. Binding of surfactants with ibuprofen.*

A stock solution of ibuprofen (100 μM) in phosphate buffer (20 mM, pH 7.4) was titrated with a stock solution of surfactants, and the fluorescence intensity was recorded at 290 nm (excited at 264 nm) at 298 K, 308 K, and 313 K. The excitation and emission slits were set to 10 nm and 5.0 nm, respectively.

### *2.2. Critical micelle concentration.*

The CMCs of the surfactants in phosphate buffer were carried out using conductivity measurements at 25°C.

Mixed surfactant systems **2-3** were prepared by mixing stock solutions of both surfactants in phosphate buffer (pH 7.4), and the solutions were stirred for 1 h. The composition of each solution was expressed as a mole fraction of surfactant **2** (Equation 1).

$$\alpha_2 = \frac{[\mathbf{2}]}{[\mathbf{2}] + [\mathbf{3}]} \qquad \textbf{(1)}$$

where [**2**] and [**3**] are the concentrations of surfactants 2 and 3 in the mixed solutions, respectively.

Mixed systems with varying mole fractions of **2** ($\alpha_2$ = 0, 0.2, 0.4, 0.6, 0.8) were prepared and the CMC of the different mixed systems was determined in phosphate buffer 7.4.

### *2.3. Membrane permeability assay.*

Membrane permeability was measured using a modified Franz cell in which the two cells were joined via an artificial membrane. The receiver cell was filled with phosphate buffer (pH 7.4, 15mL, 20mM), while the donor cell was filled with a solution of ibuprofen in phosphate buffer (pH 7.4, 15mL, 20mM) in the absence and presence of the surfactants (4 × CMC). The two cells were constantly stirred in a water bath at 37°C. The artificial membrane consisted of filter paper submerged in a mixture of 70% silicone oil and 30% isopropyl myristate, which were dried for a few minutes at room temperature and then weighed. The amount of oil absorbed, determined by mass difference, was 0.055 ± 0.002 g, which was kept constant for each experiment. The available diffusion area between cells was 0.7085cm$^2$. At 30 min intervals, 0.5ml samples were withdrawn for analysis from the donor and receiver cells. The concentration of ibuprofen was determined by its absorbance at 222nm on a UV-vis spectrometer.

Fick's first diffusion law was used to calculate the permeability coefficient $K_p$ (cm/h) [27, 28]. The relation between the permeability coefficient ($K_p$) and the steady-state flux is calculated using Equation 2:

$$K_P = \frac{J}{C_i} \quad (2)$$

where $C_i$ (mg/cm$^3$) is the initial permeate concentration in the donor solution, and $J$ (mg/(cm$^2$. h)) is the mass of permeate that passes through a unit area of the membrane in unit time, which is calculated as follows (Equation 3):

$$J = \frac{Q}{At} \quad (3)$$

Q (mg) is the quantity of permeate transported through the membrane in time t (hrs), and A is the exposed membrane area in cm$^2$.

## 3. Results and Discussion

### *3.1. Binding of surfactants with ibuprofen.*

Surfactants have been widely used as drug carriers in the pharmaceutical industry due to their simple preparation, low toxicity, small and narrow size distribution, and long shelf-life. Surfactants can also increase the solubility, bioavailability, stability, and permeability of the drug across physiological barriers [29,30].

Fluorescence spectroscopy has previously been used to measure the stability of drug-surfactant systems in the search for efficient drug carriers. This technique has been used to monitor the conformational changes, dynamics, and intermolecular interactions involved in molecular complexations [17].

The spectroscopic investigations of ibuprofen in different surfactant systems (**1**, **2**, **3**, SDS, and CTAB) were studied to gain an insight into the molecular association phenomena and their mechanism study, with the view of determining their effectiveness as drug carriers. Mixed systems containing cationic and anionic surfactants, known as catanionic mixed systems, have been reported as promising formulations for enhancing drug delivery systems [31]. For this reason, the binding of ibuprofen in catanionic mixed surfactant systems was investigated.

Ibuprofen has a strong steady-state fluorescence emission band at $\lambda_{max}$=290nm at an excitation of 264 nm. The gradual addition of the surfactants decreased the fluorescence intensity of the ibuprofen emission band at 290 nm (**1**, **2**, SDS, and CTAB) or caused a red shift (**3**) of the fluorescence emission maxima (Fig.2), suggesting the formation of a non-fluorescent, complex formation between ibuprofen and the surfactant molecules.

Fluorescence quenching occurs through two main mechanisms: dynamic and static quenching, which can be evaluated using the Stern–Volmer equation (Equation 4) [32].

$$F_0/F = K_{SV}[Q] + 1 = k_q\tau_0\,[Q] + 1 \quad (4)$$

where $F_0$ and $F$ denote the fluorescence intensities of ibuprofen in the absence and presence of the surfactant (quencher); $K_{SV}$ and [Q] are the Stern–Volmer quenching constant and the concentration of the quencher respectively; $\tau_0$ is the average lifetime of the biomolecule without the quencher and its value is $10^{-8}$ s; $k_q$ is the apparent bimolecular quenching rate constant [33,34]. The quenching mechanism is dynamic if $k_q$ is less than the maximum scattering collision quenching constant ($2\times 10^{10}$ M$^{-1}$ s$^{-1}$), otherwise, the quenching is considered static.

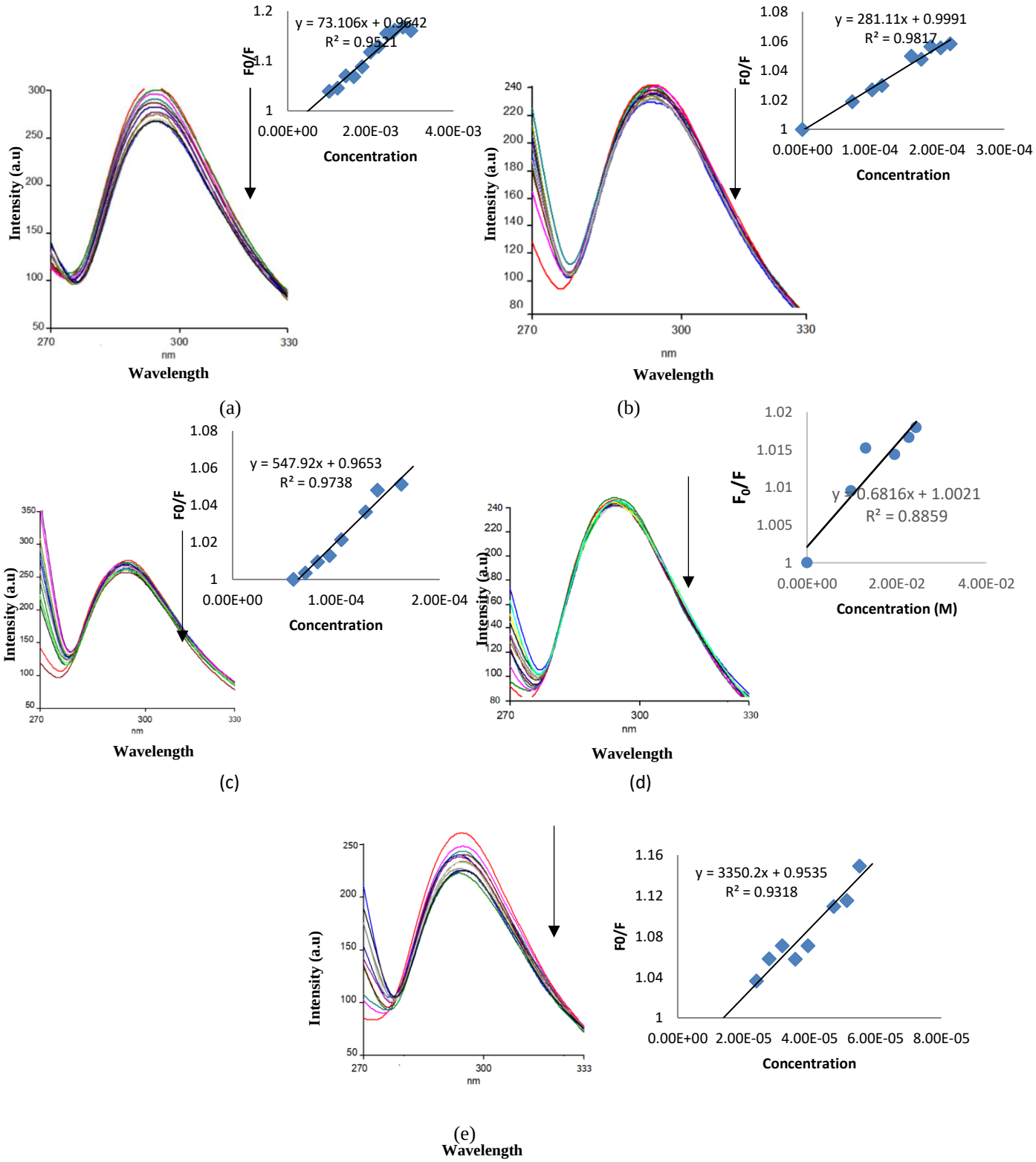


**Figure 2**. Fluorescence spectra of ibuprofen at different concentrations of (a) CTAB, (b) 1 (c) 2 (d) SDS and (e) 3 at 298 K (Inset: Stern–Volmer plots).

The binding of ibuprofen with CTAB and SDS was found to follow dynamic quenching, whereas surfactants **1**, **2,** and **3** followed a static quenching process, most probably due to the formation of ibuprofen-surfactant complexes (Table 1).

The apparent equilibrium constant ($K_a$) and the number of binding sites ($n$) can be calculated using the double logarithmic plots (Fig. 3) [35].

$$Log\left[\frac{F_0 - F}{F}\right] = \log K_a + n\log[Q] \qquad (5)$$

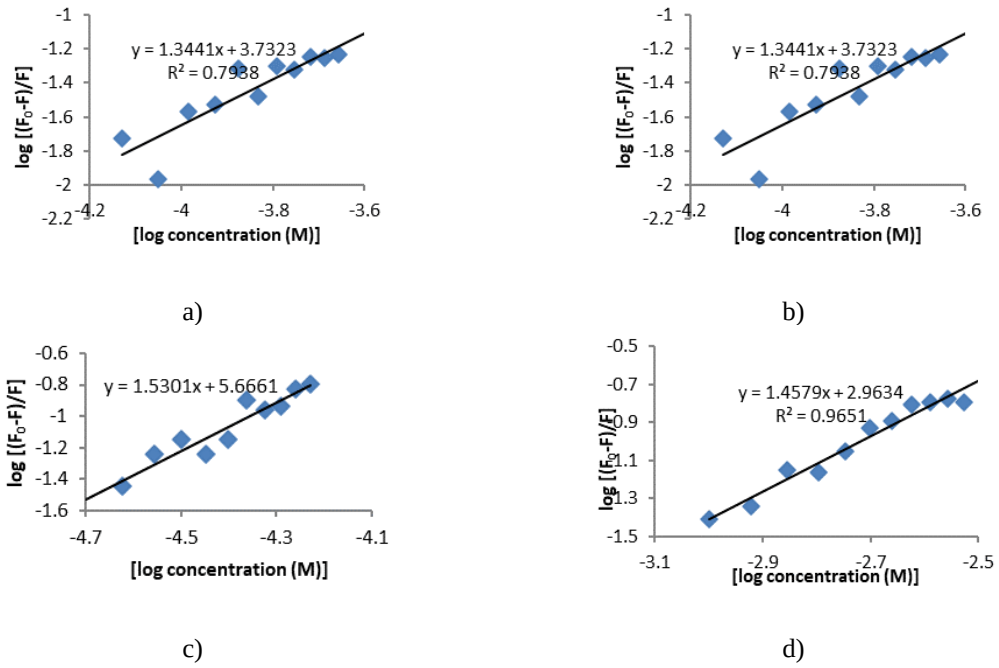


**Figure 3**. Double logarithmic plots for the binding of **(a)** 1; **(b)** 2; **(c)** 3; **(d)** CTAB with ibuprofen at 298 K.

**Table 1**. Stern–Volmer quenching constant ($K_{SV}$), bimolecular quenching constant ($k_q$), binding constant ($K_a$), and binding sites ($n$) for the binding of the surfactants with ibuprofen

| Surfactant | $K_{sv}$ ($M^{-1}$) | $k_q$ ($M^{-1}$ $s^{-1}$) | $K_a \times 10^2$($M^{-1}$) | .*n* |
|---|---|---|---|---|
| **1** | 281.1 | 2.81 X $10^{10}$ | 5.40 × $10^1$ | 1.00 |
| **2** | 547.9 | 5.48 X $10^{10}$ | 8.74 × $10^6$ | 0.85 |
| **3** | 2242 | 2.24 X $10^{11}$ | 4.64 × $10^3$ | 0.77 |
| **SDS** | 0.6816 | 6.81 X $10^7$ | 9.19 × $10^0$ | 0.67 |
| **CTAB** | 73.11 | 7.31 X $10^9$ | 0.68 × $10^{-4}$ | 0.76 |

Comparing the binding abilities of the surfactants, the binding was found to decrease in the following order: **2** > **3** > **1** > CTAB > SDS (Table 1). The higher binding affinity of **2** to ibuprofen was probably due to the higher hydrophobicity compared to **1** and CTAB, which enables **2** to form micellar aggregates and encapsulate the ibuprofen drug molecules.

It has been reported that catanionic mixed surfactant systems containing anionic amino acid-based carboxylate surfactants form unilamellar vesicles with drug entrapment ability [36]. The anionic carboxylate surfactant **3** and surfactant **2** showed the highest $K_a$ values, indicating their higher affinity for ibuprofen, and therefore, both of these surfactants are better drug carriers. Consequently, the binding of ibuprofen in a catanionic mixed surfactant system containing varying mole fractions of **2** ($\alpha_2$) and **3** was investigated (Table 2).

**Table 2.** Stern–Volmer quenching constant ($K_{SV}$), bimolecular quenching constant ($k_q$), binding constant ($K_a$), and binding sites ($n$) for the binding of the 2-3 mixed surfactant with ibuprofen.

| Mole fraction of 2 ($\alpha_2$) | Mole fraction of 3 | $K_{sv}$ ($M^{-1}$) | $k_q$ ($M^{-1}$ $s^{-1}$) | $K_a$ ($M^{-1}$) | *n* |
|---|---|---|---|---|---|
| **0** | 1.0 | 2242 | 2.24 x $10^{11}$ | 4.64 x $10^5$ | 0.77 |
| **0.2** | 0.8 | 2577 | 2.58 x $10^{11}$ | 9.10 x $10^6$ | 1.90 |
| **0.4** | 0.6 | 2875 | 2.88 x $10^{11}$ | 3.16 x $10^{14}$ | 2.11 |
| **0.6** | 0.4 | 2066 | 2.07 x $10^{11}$ | 1.74 x $10^8$ | 3.77 |
| **0.8** | 0.2 | 1749 | 1.75 x $10^{11}$ | 2.90 x $10^2$ | 0.80 |
| **1.0** | 0 | 547 | 5.48 x $10^{10}$ | 8.74 x $10^8$ | 0.85 |

In all the mole fractions studied, ibuprofen binding to the **2- 3** mixed surfactant system was found to be static rather than dynamic, as observed by the calculated $k_q$ values.

An increase in $\alpha_2$ from 0 to 0.4 caused an increase in the $K_a$ value, and a further increase in $\alpha_2$ up to 0.8 caused a decrease in the binding constant. This implies that the catanionic mixture of **2-3** acts as a more efficient drug encapsulator at a composition of $\alpha_2$ = 0.4. The decrease in binding constant with higher $\alpha_2$ might be due to a change in the shape and composition of the mixed micellar systems, which rendered them unfavorable for the binding and encapsulation of ibuprofen.

*3.2. Thermodynamics of surfactant binding.*

The effect of temperature on the binding ability of the surfactants **1**, **2**, **3**, CTAB, and SDS, as well as the **2- 3** mixed surfactant system, were investigated, and the thermodynamic parameters were determined at temperatures 298, 308, and 313 K (Table 3, Table 4).

Thermodynamic parameters were determined using Van't Hoff formula (Equation 6).

$$\ln K_a = \frac{-\Delta H^\circ}{RT} + \frac{\Delta S^\circ}{R} \qquad (6)$$

where $K_a$ is the binding constant, R is the gas constant, and T is the absolute temperature, respectively. The free energy change ($\Delta G^0$) was calculated according to Equation 7:

$$\Delta G^\circ = -RT \ln K_a \qquad (7)$$

The interaction of ibuprofen and the surfactants can be explained using the enthalpy and entropy of binding obtained from the Van't Hoff plots (Fig.4)

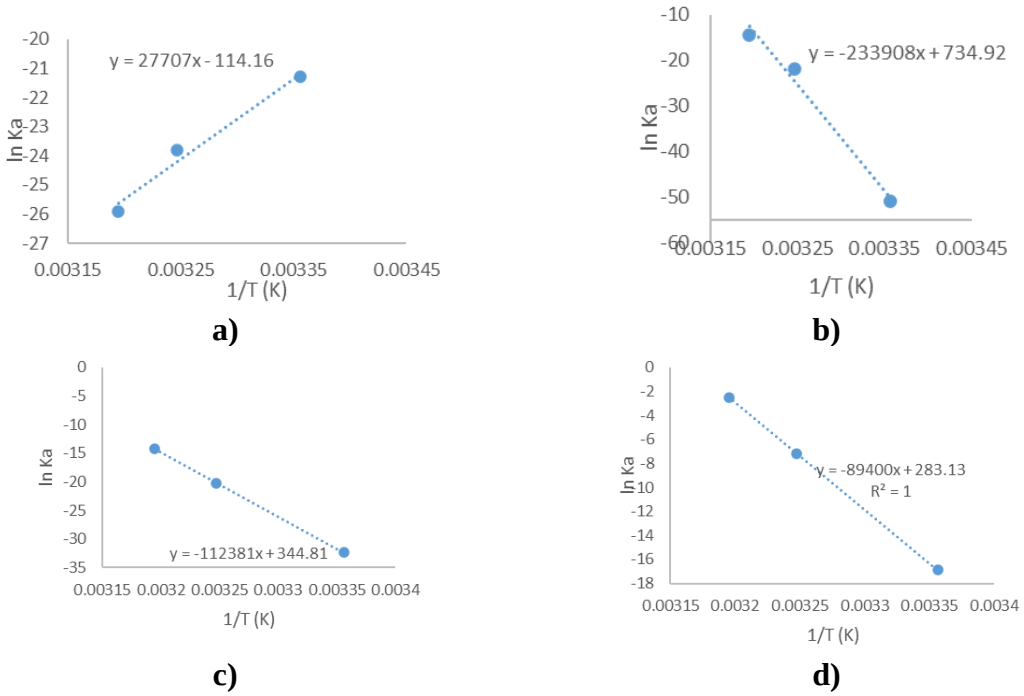


**Figure 4.** Van't Hoff plots of the binding of ibuprofen to **(a)** 1; **(b)** 2; **(c)** 3; **(d)** CTAB.

**Table 3**. Thermodynamic parameters of ibuprofen–surfactant interaction in phosphate buffer.

| Temperature (K) | *n* | $K_a$ ($M^{-1}$) | ln $K_a$ | 1/T ($K^{-1}$) | ΔG (kJ $mol^{-1}$) | ΔH (kJ $mol^{-1}$) | ΔS ($JK^{-1}$ $mol^{-1}$) |
|---|---|---|---|---|---|---|---|
| **SDS** | | | | | | | |
| 298 | 0.76 | 0.6816 | | 0.003356 | | | |
| 308 | 0.98 | | | 0.003247 | | | |
| 313 | 0.50 | | | 0.003195 | | | |
| **CTAB** | | | | | | | |
| 298 | 0.67 | $9.19 \times 10^2$ | 6.82 | 0.003356 | -16.89 | | |
| 308 | 0.97 | $1.64 \times 10^1$ | 2.80 | 0.003247 | -7.17 | 742.9 | 2352 |

| Temperature (K) | *n* | $K_a$ ($M^{-1}$) | ln $K_a$ | 1/T ($K^{-1}$) | ΔG (kJ $mol^{-1}$) | ΔH (kJ $mol^{-1}$) | ΔS ($JK^{-1}$ $mol^{-1}$) |
|---|---|---|---|---|---|---|---|
| 313 | 1.30 | $2.60 \times 10^0$ | 0.956 | 0.003195 | -2.49 | | |
| **1** | | | | | | | |
| 298 | 1.00 | $5.40 \times 10^3$ | 8.59 | 0.003356 | -21.3 | | |
| 308 | 1.26 | $1.10 \times 10^4$ | 9.31 | 0.003247 | -23.8 | -230.2 | -948.67 |
| 313 | 1.45 | $2.10 \times 10^4$ | 9.95 | 0.003195 | -25.9 | | |
| **2** | | | | | | | |
| 298 | 0.85 | $8.74 \times 10^8$ | 20.59 | 0.003356 | -50.99 | | |
| 308 | 1.25 | $4.90 \times 10^3$ | 8.50 | 0.003247 | -21.76 | 1943 | 6107 |
| 313 | 0.89 | $2.46 \times 10^2$ | 5.51 | 0.003195 | -14.33 | | |
| **3** | | | | | | | |
| 298 | 0.77 | $4.65 \times 10^5$ | 13.05 | 0.003356 | -32.32 | | |
| 308 | 0.65 | $2.65\times 10^3$ | 7.88 | 0.003247 | -20.17 | 933.89 | 2865 |
| 313 | 0.88 | $2.34 \times 10^2$ | 5.46 | 0.003195 | -14.20 | | |

**Table 4**. Thermodynamic parameters of the binding of 2-3 mixed system with ibuprofen in phosphate buffer.

| Temperature (K) | *n* | $K_a$ ($M^{-1}$) | ln $K_a$ | 1/T ($K^{-1}$) | ΔG (kJ $mol^{-1}$) | ΔH (kJ $mol^{-1}$) | ΔS ($JK^{-1}$ $mol^{-1}$) |
|---|---|---|---|---|---|---|---|
| $\alpha_2$ = **0** | | | | | | | |
| 298 | 0.77 | $4.65 \times 10^5$ | 13.05 | 0.003356 | -32.32 | | |
| 308 | 0.65 | $2.65\times 10^3$ | 7.88 | 0.003247 | -20.17 | 933.89 | 2865 |
| 313 | 0.88 | $2.34 \times 10^2$ | 5.46 | 0.003195 | -14.20 | | |
| $\alpha_2$ **= 0.2** | | | | | | | |
| 298 | 1.9 | $9.10 \times 10^6$ | 16.02 | 0.003356 | -39.67 | | |
| 308 | 2.13 | $8.13 \times 10^7$ | 18.21 | 0.003247 | -46.61 | -893.50 | -3318 |
| 313 | 2.50 | $5.37 \times 10^9$ | 22.40 | 0.003195 | -58.26 | | |
| $\alpha_2$**=0.4** | | | | | | | |
| 298 | 3.77 | $3.16 \times 10^{14}$ | 33.39 | 0.003356 | -82.69 | | |
| 308 | 2.90 | $7.45 \times 10^{11}$ | 27.34 | 0.003247 | -69.98 | 1369.5 | 3898 |
| 313 | 2.40 | $1.38 \times 10^9$ | 21.05 | 0.003195 | -54.75 | | |
| $\alpha_2$**=0.6** | | | | | | | |
| 298 | 2.11 | $1.74 \times 10^8$ | 18.97 | 0.003356 | -46.98 | | |
| 308 | 1.4 | $1.34 \times 10^5$ | 11.81 | 0.003247 | -30.23 | 1222.02 | 3732 |
| 313 | 1.07 | $7.98 \times 10^3$ | 8.98 | 0.003195 | -23.36 | | |
| $\alpha_2$ **=0.8** | | | | | | | |
| 298 | 0.8 | $2.90 \times 10^2$ | 5.67 | 0.003356 | -14.04 | | |
| 308 | 1.75 | $2.34 \times 10^6$ | 14.67 | 0.003247 | -37.55 | -2010 | -6858.5 |
| 313 | 1.80 | $9.46 \times 10^6$ | 20.67 | 0.003195 | -53.76 | | |
| $\alpha_2$ **=1** | | | | | | | |
| 298 | 0.85 | $8.74 \times 10^8$ | 20.59 | 0.003356 | -50.99 | | |
| 308 | 1.25 | $4.90 \times 10^3$ | 8.50 | 0.003247 | -21.76 | 1943 | 6107 |
| 313 | 0.89 | $2.46 \times 10^2$ | 5.51 | 0.003195 | -14.33 | | |

The four binding modes through which molecules can bind to each other are hydrogen bonds, hydrophobic forces, electrostatic forces, and van der Waals' interactions. These interactions can be described by thermodynamic parameters such as enthalpy and entropy energies. In the case of CTAB **2** and **3**, positive enthalpy and entropy values were obtained, implying that their interactions with ibuprofen were mostly hydrophobic. The negative enthalpy and entropy values observed for binding ibuprofen to **1** showed that the interactions involved are mainly van der Waals forces and hydrogen bonding [37].

The binding of ibuprofen with a mixture of 2 and 3 was found to be mostly due to hydrophobic interactions, except in the case of $\alpha_2$ =0.2 and 0.8, where the binding was mainly due to van der Waals forces and hydrogen bonding. The lower binding constant observed for $\alpha_2$ = 0.2 and 0.8 can be accounted for by a change in the micellar shape and composition, leading to different interactions between the surfactants and the drug molecule, which are less favorable for their binding efficiency.

*3.3. Critical micelle concentration.*

The ability of surfactants to form colloidal-sized clusters called micelles has particular importance in pharmacy due to their ability to enhance the solubility of partially soluble substances in aqueous medium [38]. In an endeavor to increase the bioavailability of drugs, several drug delivery systems have been reported in an attempt to minimize drug degradation and also to prevent harmful side effects [39]. In this view, applying micelles as drug carriers is advantageous compared to other alternatives, such as soluble polymers and liposomes. Micellar assemblies can increase the solubility and bioavailability of poorly soluble drugs through solvent-free drug delivery systems such as topical ointments [40].

Micellar systems containing mixed surfactants are known to have altered physical properties compared to individual surfactants, and they are used in several applications [41, 42]. Mixed systems consisting of cationic and anionic surfactants are known to have enhanced surface activities due to electrostatic interactions between the individual surfactants.

The critical micelle concentration (CMC) of the surfactants **1**, **2**, **3**, CTAB, and SDS (Table 5), as well as that of the **2-3** mixed surfactant system (Table 6), were determined in phosphate buffer pH 7.4.

**Table 5**. CMC of the surfactants in phosphate buffer pH 7.4.

| Surfactants | Critical micelle concentration (mM) |
|---|---|
| SDS | 4700 |
| CTAB | 720 |
| **1** | 61 |
| **2** | 46 |
| **3** | 20 |

The Clint equation (Equation 8) is used to predict the ideal CMC (cmc*) of the binary mixtures [43].

$$\frac{1}{cmc*} = \frac{\alpha_2}{cmc_2} + \frac{(1-\alpha_2)}{cmc_3} \quad (8)$$

where $\alpha_2$ is the mole fraction of **2** in the mixture, $cmc_2$ and $cmc_3$ are the critical micelle concentrations of pure **2** and **3,** respectively, and cmc* is the value under ideal mixing.

**Table 6.** CMC of 2- 3 mixed surfactant systems.

| Mole fraction | | | |
|---|---|---|---|
| **2** | **3** | **Theoretical CMC (μM)*** | **Experimental CMC (μM)** |
| 0 | 1 | 20.00 | 20.00 |
| 0.2 | 0.8 | 22.52 | 19.20 |
| 0.4 | 0.6 | 25.78 | 28.30 |
| 0.6 | 0.4 | 30.13 | 37.00 |
| 0.8 | 0.2 | 36.25 | 41.20 |
| 1 | 0 | 45.50 | 45.50 |

The micellar molar fraction of **2** in the ideal state, $\chi_2^{id}$ was calculated according to Equation 9, assuming a binary ideal mixture.

$$\chi_2^{id} = \frac{\alpha_2 cmc_3}{\alpha_2 cmc_3 + (1-\alpha_2) cmc_3} \quad (9)$$

The CMC values obtained experimentally were higher than those predicted by the Clint equation (Equation 5), suggesting antagonism within the mixed surfactant systems. The antagonistic behavior of the mixed **2-3** was presumed to be due to the precipitation of the

neutral catanionic complex formed between anionic surfactant **3** and the cationic QUAT **2** at an equimolar ratio.

The regular solution theory (RST) was used to estimate the degree of non-ideality of the surfactant interactions. This includes characterizing the interactions between the two surfactants in the mixed micelle using the interaction parameter $\beta_{12}$. $\beta_{12}$ is related to the activity coefficients, *f*, of the surfactants within the micelle, according to equations (10-12):

$$f_1 = \exp \beta_{12} (1- \chi_2)^2 \quad \textbf{(10)}$$

$$f_2 = \exp \beta_{12} \chi_2 ^2 \quad \textbf{(11)}$$

Where $\chi_2$, the molar fraction of **2** in the mixed micelle, can be obtained by solving the following equation iteratively:

$$\frac{\chi_2^2 \ln(\alpha_2 cmc/\chi_2 cmc_2)}{(1-\chi_2)^2 \ln[(1-\alpha_2)cmc/(1-\chi_2)cmc_3} = 1 \quad \textbf{(12)}$$

$\beta_{12}$ can then be calculated from

$$\beta_{12} = \frac{\ln(\alpha_2 cmc/\chi_2 cmc_2)}{(1-\chi_2)^2} \quad \textbf{(13)}$$

From Equation 13, the molar fraction of **2** in the mixed micelle, $\chi_2$ was lower than the mole fractions of 2 in bulk ($\boldsymbol{\alpha}_2$), suggesting that **2** had a lower tendency to micelles in the surfactant **3** mixed surfactant system. Positive $\beta_{12}$ values were obtained for **2**-**3** mixed systems for the different mole ratios studied, confirming antagonistic behavior within the mixed micellar system [44].

*3.4. Franz cell diffusion of ibuprofen in the presence of surfactants.*

Transdermal drug delivery has become one of the most successful and innovative focuses for research in drug delivery, with around 40% of the drug candidates being under clinical evaluation related to transdermal or dermal systems [45]. This drug delivery method involves applying drug formulations onto healthy and intact skin ,followed by the diffusion of the drug through the skin to the site of action.

To evaluate the effectiveness of surfactants **1**, **2**, **3**, CTAB, and SDS in promoting the permeability of ibuprofen for transdermal applications, the permeation rate of the drug in the absence and presence of surfactants at a concentration above their respective CMC values (x 4) was evaluated using a Franz cell, simulating specific conditions of human skin.

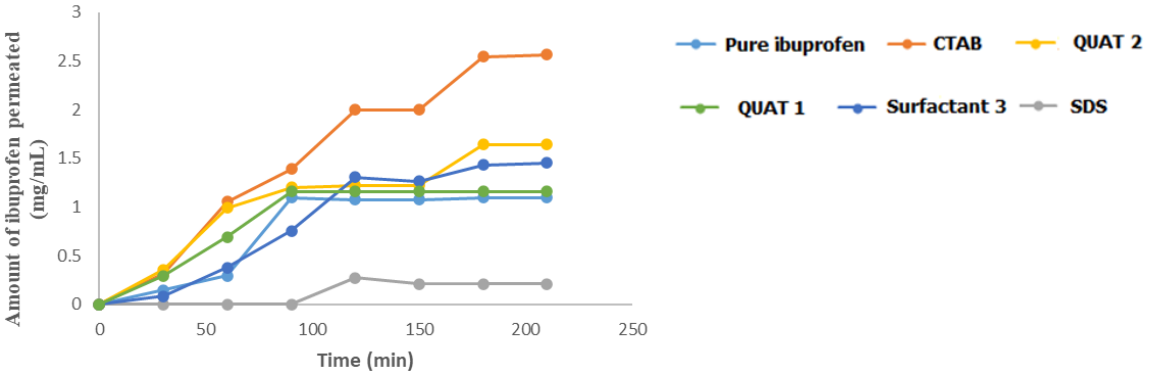


**Figure 5**. Effect of the different surfactant on permeation of ibuprofen.

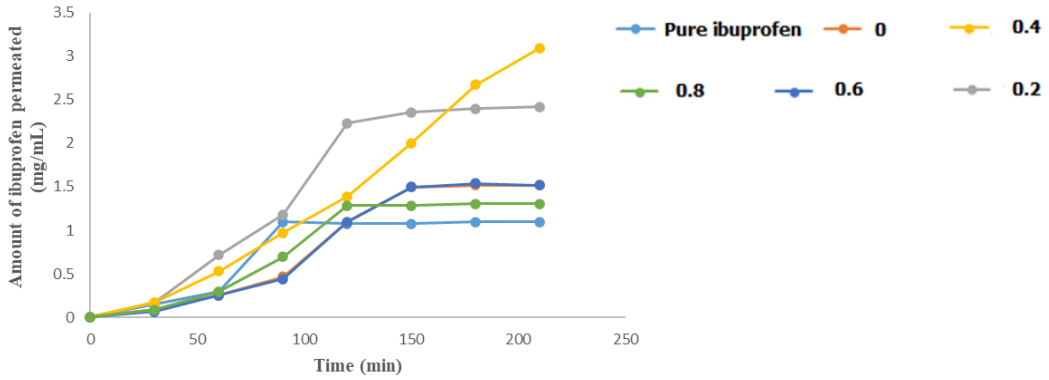


**Figure 6.** Effect of the different surfactants on permeation of ibuprofen.

An increase of the flux (*J*) and permeability coefficient ($K_p$) of ibuprofen was observed in the presence of **1**, **2**, **3,** and CTAB, which indicates that these surfactants were able to promote the membrane permeability of ibuprofen (Table 7). CTAB was found to cause an approximately two-fold increase in the permeability of ibuprofen compared to solutions without surfactants.

The decrease in permeability of ibuprofen in the presence of SDS might account for the unfavorable binding between SDS and ibuprofen, as indicated by a very low binding constant $K_a$.

**Table 7**. Flux (*J*) and permeability coefficient ($K_p$) of ibuprofen through a silicone/isopropyl myristate membrane (70:30) at 25 °C in the presence and absence of surfactants.

| Compound | $J$ (mg/(cm$^2$. h) × 10$^3$ | $K_p$ (cm/h) × 10$^3$ | *Log* $K_p$ (cm/s) |
|---|---|---|---|
| No surfactant | 5.80 | 9.00 | -2.04 |
| SDS | 1.12 | 1.74 | -2.76 |
| CTAB | 10.59 | 16.50 | -1.78 |
| **1** | 7.40 | 11.50 | -1.94 |
| **2** | 8.70 | 13.60 | -1.87 |
| **3** | 6.70 | 10.40 | -1.98 |

The mixed surfactant systems containing **2** and **3** were also evaluated for their ability to influence the membrane permeability of ibuprofen. An increase in mole fraction of **2** was found to increase the flux (*J*) and permeability coefficient ($K_p$) of ibuprofen up to a mole fraction of 0.4, above which the permeability was found to decrease (Table 8). This mirrors the change in $K_a$ with the mole fraction noted earlier.

**Table 8**. Flux (*J*) and permeability coefficient ($K_p$) of ibuprofen through a silicone/isopropyl myristate membrane (70:30) at 25°C in the presence of a mixed surfactant system.

| Mole fraction of QUAT 2 | $J$ (mg/(cm$^2$. h) × 10$^3$ | $K_p$ (cm/h) × 10$^3$ | *Log* $K_p$ (cm/s) |
|---|---|---|---|
| 0 | 6.70 | 10.40 | -1.98 |
| 0.2 | 12.70 | 19.80 | -1.70 |
| 0.4 | 16.40 | 22.60 | -1.65 |
| 0.6 | 8.14 | 12.70 | -1.90 |
| 0.8 | 8.17 | 12.80 | -1.89 |
| 1 | 8.70 | 13.60 | -1.87 |

## 4. Conclusions

The surfactants **1**, **2**, **3,** and CTAB showed promising interaction with ibuprofen, while SDS showed minimal interaction with the drug. The surfactants **2**, **3,** and CTAB were found to interact with ibuprofen via hydrophobic interactions, while surfactant **1** interacted via van der Waals forces and hydrogen bonding. Optimum binding abilities were observed for **2** and **3**. Investigation of the mixed surfactant system containing **2** and **3** showed that the mixture

containing **2** at mole fraction 0.4 displayed the highest binding ability. The mixture also showed the highest membrane permeability at the same mole fraction, suggesting that this mixture could be employed as a promising carrier for the formulation of transdermal drug delivery of ibuprofen.

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## Conflicts of Interest

The authors declare no conflict of interest.

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