Summary

Surfactants in general and cationic surfactants in particular have gained lot of importance in the recent years. These surfactants have ever increasing applications ranging from their use as additive in cosmetics to their application as vehicles for the delivery of drugs and genetic materials. In view of the above we have carried out the present work to synthesize some new pyridinium and imidazolium surfactants and to evaluate them for their biological and surface properties.

The work embodied in this thesis has been divided into five chapters.

We have attempted to introduce the work in chapter one and the literature related to the work has also been briefly reviewed in this chapter.

The second chapter deals with the synthesis of thioether spacer containing gemini pyridinium surfactants and effect of increasing hydrophobic alkyl chain length on their surface activity, DNA binding and cytotoxicity.

Synthesis of thioether spacer containing gemini imidazolium surfactants and effect of increasing spacer length and hydrophobic alkyl chain length on their self aggregation and DNA interaction properties has been described as chapter three.

Synthesis and comparative studies of phenoxy ring containing gemini pyridinium and imidazolium surfactants has been detailed in chapter four while chapter five describes the work done for the synthesis of hydroxy group containing pyridinium surfactants and evaluation of their properties.

Chapter 1:
Introduction and Review of Literature

Section 1.1 is the brief introduction of the work embodied in this thesis.

In Section 1.2 the literature related to the synthesis and properties of pyridinium surfactants has been reviewed briefly.

While, the literature related to the synthesis and properties of imidazolium surfactants has been reviewed in Section 1.3.
Chapter 2:
Synthesis of Thioether Spacer Containing Gemini Pyridinium Surfactants and Effect of Increasing Hydrophobic Alkyl Chain Length on Their Surface Activity, DNA Binding and Cytotoxicity.

Section 2.1: Synthesis and characterization of thioether spacer containing gemini pyridinium surfactants.

New gemini pyridinium surfactants containing thioether spacer have been synthesized by regioselective electrophilic cobromination of α-olefins (Scheme 2.1). Ethane-1,2-dithiol \( \text{[1]} \) and \( N \)-bromosuccinimide \( \text{[6]} \) on reaction with α-olefins (dodecene \( \text{[2]} \), tetradecene \( \text{[3]} \), hexadecene \( \text{[4]} \) and octadecene \( \text{[5]} \) ) gave the respective 1,2-bis(2-bromoalkylthio)ethane \( \text{[7-10]} \). The bromoalkylthio ethers \( \text{[7-10]} \) thus obtained on reaction with pyridine \( \text{[11]} \) gave the respective gemini bispyridinium dibromides \( \text{[12-15]} \). The structures of all these new gemini pyridinium surfactants were confirmed by elemental analysis, IR, NMR and Mass spectroscopy.

\[
\begin{align*}
\text{[1]} & \quad \text{[2-5]} & \quad \text{[6]} & \quad 5^\circ C \\
\text{[7-10]} & \quad \text{[11]} & \quad 80^\circ C \\
\end{align*}
\]

\( n = 9 \) (Intermediate 7 & Surfactant 12)  
\( n = 11 \) (Intermediate 8 & Surfactant 13)  
\( n = 13 \) (Intermediate 9 & Surfactant 14)  
\( n = 15 \) (Intermediate 10 & Surfactant 15)

Scheme - 2.1
Section 2.2: Evaluation of surface properties of gemini pyridinium surfactants.
The gemini pyridinium surfactants obtained as above (Scheme - 2.1) were evaluated for their surface properties. The critical micelle concentration (cmc) of these gemini pyridinium surfactant were determined by surface tension and conductivity data. It has been found that cmc of these gemini pyridinium surfactants was lower than previously reported gemini pyridinium surfactants in literature and the cmc value of these surfactants decreased with increase in alkyl chain length.

Section 2.3: Evaluation of DNA binding properties of gemini pyridinium surfactants.
These gemini pyridinium surfactants have also been evaluated for their DNA binding properties. The preliminary studies conducted by agarose gel electrophoresis indicated chain length dependent DNA binding abilities of these surfactants. These results further have been supported by ethidium bromide exclusion experiments and transmission electron microscopy (TEM).

Section 2.4: Evaluation of cytotoxicity of gemini pyridinium surfactants.
These bispyridinium surfactants (12-15) have been found to be less cytotoxic on C6 glioma cells (cancerous brain cell line) compared to quaternary ammonium gemini 1,6-hexanediyl bis-(dimethyldecylammonium)bromide by MTT assay.

Chapter 3:
Synthesis of Thioether Spacer Containing Gemini Imidazolium Surfactants and Effect of Increasing Spacer Length and Hydrophobic Alkyl Chain Length on Their Self aggregation and DNA Interaction Properties.

Section 3.1: Synthesis and characterization of thioether spacer containing gemini imidazolium surfactants.
Twelve new gemini imidazolium surfactants (22-33) with four different hydrophobic tails i. e. dodecyl/ tetradecyl/ hexadecyl or octadecyl and three different spacers (-S-(CH₂)ₙ-S-, where n is 2, 3 & 4) were synthesized (Scheme – 3. 1). Regioselective cobromination of α-olefins (4-7) using N-bromosuccinimide (8) to get reactive intermediates 1,2-bis((2-bromoalkyl)thio)alkane (9-20). The intermediate were quaternized with N-methylimidazole to get the gemini imidazolium surfactant (22-33). The structure of these new gemini imidazolium surfactants have been established by elemental analysis, IR, NMR and Mass spectroscopy.
Summary

Section 3.2: Evaluation of surface properties of gemini imidazolium surfactants.
The surface properties of these gemini imidazolium surfactants were evaluated by surface tension and conductivity method. Their cmc values decreased with increase in spacer length and hydrophobic alkyl chain length. These surfactants have low cmc values as compared to other category of gemini cationic surfactants and exhibit peculiarities at sufficiently low concentration as they were found to be forming premicellar aggregates in wide range of concentration below their cmc values.

![Reaction Scheme](image)

\[ \text{HS} \left( \text{SH} \right)^{[1-3]} + \text{[4-7]} \rightarrow \text{[9-20]} \]

\[ \text{Br}^{[9-20]} + \text{[21]} \rightarrow \text{[22-33]} \]

n = 2 & m = 9 (Intermediate 9 & Surfactant 22)  
n = 3 & m = 9 (Intermediate 10 & Surfactant 23)  
n = 4 & m = 9 (Intermediate 11 & Surfactant 24)  
n = 2 & m = 11 (Intermediate 12 & Surfactant 25)  
n = 3 & m = 11 (Intermediate 13 & Surfactant 26)  
n = 4 & m = 11 (Intermediate 14 & Surfactant 27)  
n = 2 & m = 13 (Intermediate 15 & Surfactant 28)  
n = 3 & m = 13 (Intermediate 16 & Surfactant 29)  
n = 4 & m = 13 (Intermediate 17 & Surfactant 30)  
n = 2 & m = 15 (Intermediate 18 & Surfactant 31)  
n = 3 & m = 15 (Intermediate 19 & Surfactant 32)  
n = 4 & m = 15 (Intermediate 20 & Surfactant 33)

Scheme - 3.1

Section 3.3: Evaluation of Thermal degradation temperature of imidazolium gemini surfactants by thermogravimetry analysis.
Thermal degradation of these new gemini surfactants was determined by thermogravimetry analysis. The thermal stability of these gemini imidazolium surfactants decreases with increase in hydrophobic alkyl chain length from dodecyl to hexadecyl (for a particular spacer length).
Section 3.4: Evaluation of DNA binding properties of gemini imidazolium surfactants.
The gemini imidazolium surfactants (22-33) were found to exhibit strong DNA binding ability as determined by agarose gel electrophoresis and supported by ethidium bromide exclusion experiments. It has also been observed that due to strong interaction of these surfactants with DNA they were able to protect DNA against enzymatic degradation as determined by DNase sensitivity assay.

Chapter 4:
Synthesis and Comparative Study of Phenoxy Ring Containing Gemini Pyridinium and Imidazolium Surfactants.
Section 4.1: Synthesis and characterization of phenoxy ring containing gemini pyridinium and imidazolium surfactants.
Two series of phenoxy ring containing imidazolium and pyridinium gemini amphiphiles have been synthesized (Scheme 4.1) from allylated hydrogenated cardanol oil (5) having different spacers (-S-(CH$_2$)$_n$-S-, where n is 2, 3, 4 & 6). Dithiols (1-4) were reacted with 1-(allyloxy)-3-pentadecylbenzene (5) and N-bromosuccinimide (6) to give intermediates (7-10) which were further reacted with N-methylimidazole (11) or pyridine (12) at 80 °C for 2 hours to give the corresponding gemini surfactants (13-20). The structure of these new gemini imidazolium and pyridinium surfactants was established by IR, NMR and Mass spectroscopy, while purity was determined by HPLC.
Section 4.2: Comparison of surface properties of gemini pyridinium and imidazolium surfactants.
The surface properties of these gemini imidazolium and pyridinium surfactants could only be evaluated by conductivity method due to limited solubility of these surfactants. These amphiphiles were found to have ultra low cmc values. It has been found that the cmc of these gemini amphiphiles decreases with increase in spacer length. Further, the cmc values of gemini pyridinium amphiphiles were found to be lower as compared to gemini imidazolium congeners.
Section 4.3: Evaluation of DNA binding affinity of gemini pyridinium and imidazolium surfactants.

Biophysical investigation of these new gemini amphiphiles for their interaction with DNA established greater ability of these amphiphiles to form complex with DNA. This has been established by agarose gel electrophoresis and transmission electron microscopy. Further, ethidium bromide exclusion assay established greater affinity of these gemini amphiphiles
towards DNA as they were able to completely displace ethidium bromide from DNA-EB intercalated complex.

**Section 4.4: Evaluation of cytotoxicity of gemini pyridinium and imidazolium surfactants.**
These gemini cationic surfactants were found to have relatively low cytotoxicity on C6 glioma cells (cancerous brain cell line) by MTT assay. Some of the surfactants were found to display low cytotoxicity even lower than the commercially available transfecting agent dimethyl dioctadecyl ammonium bromide.

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**Chapter 5:**

*Synthesis and Properties of Hydroxy Group Containing Pyridinium Surfactants.*

**Section 5.1: Synthesis and characterization of hydroxy group containing pyridinium surfactants.**

The co-bromination of α-Olefins (1-dodecene (1), 1-tetradecene (2), 1-hexadecene (3), and 1-octadecene (4)), with protected glycerol (solketal (6)) at 60 °C, followed by deprotection with 10% HCl, gave chromatographically inseparable isomeric mixtures of β-bromo monoethers of glycerol: 3-(2-bromoalkyloxy)propane-1,2-diol (11a-14a)/3-(1-bromoalkane-2-yloxy)propane-1,2-diol (11b-14b). These β-Bromo glycerol monoethers, on reaction with pyridine (16), gave hydroxyl group containing pyridinium cationic surfactant (Scheme 5.1), 1-(1-(2,3-dihydroxypropoxy)alkane-2-y1)pyridinium bromide (17a-20a)/1-(2-(2,3-dihydroxypropoxy)alkyl) pyridinium bromide (17b-20b). The structures of all these new gemini pyridinium surfactants were confirmed by elemental analysis, IR, NMR and Mass spectroscopy.

**Section 5.2: Evaluation of surface and biological properties of hydroxy group containing pyridinium surfactants.**

The evaluation of surface and biological properties of hydroxy group containing pyridinium surfactants exhibited that these surfactants have better surface properties, DNA binding abilities, and lower cytotoxicity against C6 glioma cells in comparison to commercially available cationic surfactants i. e. cetyl pyridinium bromide and tetradecyl trimethyl ammonium bromide.
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\text{Summary}
\]

\[\text{RCH=CH}_2 + \text{[5]} \rightarrow 60^\circ \text{C} \]

\[\text{Br} \quad \text{R-CH-CH}_2 \quad \text{[7a-10a]} \quad \text{Br} \quad \text{R-CH-CH}_2 \quad \text{RCH=CH}_2 \quad \text{unreacted} \quad \text{[7b-10b]} \quad 10\% \text{ aq HCl} \quad \text{Room Temperature} \quad 3 \text{ hours} \]

\[\text{Br} \quad \text{R-CH-CH}_2 \quad \text{Br} \quad \text{OH} \quad \text{OH} \quad \text{OH} \quad \text{OH} \quad \text{[15]} \quad \text{[11a-14a]} \quad \text{[11b-14b]} \]

\[\text{100}^\circ \text{C} \quad 10 \text{ hours} \quad \text{[18]} \quad \text{[17a-20a]} \quad \text{[17b-20b]} \]

7, 11 & 17 \quad R = -(\text{CH}_2)_9-\text{CH}_3 \quad 8, 12 & 18 \quad R = -(\text{CH}_2)_{11}-\text{CH}_3 \quad 9, 13 & 19 \quad R = -(\text{CH}_2)_{13}-\text{CH}_3 \quad 10, 14 & 20 \quad R = -(\text{CH}_2)_{15}-\text{CH}_3 \]

\text{Scheme 5.1}