

**MATERIALS &
EXPERIMENTAL
TECHNIQUES**

2. EXPERIMENTAL

2.1. 1. Materials.

The chemicals, which used in this study supplied from international companies and used without more purification, are shown in Table (1)

Table (1): List of the used chemicals

No.	Material	Source
1	Decyl amine	Merck
2	Dodecyl amine	Merck
3	Hexadecyl amine	Merck
4	Octadecyl amine	Merck
5	Triethyl amine	El-Nasr Co.
6	Propylene oxide	Merck
7	Benzyl chlorid	Merck
8	Sulphoric acid	El-Nasr Co

2.1.2. Synthesis of Tertiary Fatty Amines.

Synthesis of tertiary fatty amine was produced by reaction of one mole of different carbon chain length fatty amines mixed with the desired amount of triethyl amine as catalyst (0.01%) and placed in the reactor. Afterward two moles propylene oxide was dosed. The reaction was carried out at 90 °C in the presence of alkaline catalyst. The reaction mixture was then kept for 30 min at

the reaction temperature, after cooling. The obtained product was discharged and weighed.

2.1.3. Synthesis of Quaternary Ammonium Salts.

A homologous series of cationic surfactants, Benzyl-decyl-bis-(2hydrxy-propyl)-ammonium chloride, compound (I), Benzyl-dodecyl-bis-(2hydrxy-propyl)-ammonium chloride, compound (II), Benzyl-hexadecyl-bis-(2hydrxy-propyl)-ammonium chloride, compound (III), Benzyl-octadecyl-bis-(2hydrxy-propyl)-ammonium chloride, compound(IV), were prepared by the reflux of one mole of prepared tertiary amine with one mole of benzyl chloride in acetone as solvent at temperature of 60°C for 20 hours. The products purified by recrystallization three times in acetone, and then washed with diethyl ether. The products kept in desiccators up to use.

2.2. Experimental Techniques

2.2.1. Structural Confirmation of the Prepared Compounds.

The chemical structure of the prepared compounds was confirmed by:

1. Elemental analysis using Analyzer Vario El III, made in Germany. (Micro Analytical Center, Cairo University).
2. FTIR spectra using ATI Mattsonm Infinity seriesTM, Bench top 961 controlled by Win FirstTM V2.01 software. (Micro Analytical Center, Cairo University).

- ¹H-NMR was measured in DMSO-d₆ by Spect Varian, GEMINI 200 (¹H 200 MHz). (Micro Analytical Center, Cairo University).

2.2. 2. Surface and Interfacial Tension Detremenation:

Surface and interfacial tension of aqueous solutions of prepared compounds were measured using Du-Nouy tensiometer (Kruss type K6). Surface tension was measured at different concentrations at 25° C. The interfacial tension measured between 0.1% surfactant solution and light paraffin oil at 25°C.

2.2.3 Evaluation of Prepared Compounds as Corrosion Inhibitors.

2.2.3.1. Weight loss Techniques.

The carbon steel coupons of 2.5 cm 2.0 cm 0.6 cm were used . The chemical composition of steel sample was recorded in Table (2). The carbon steel coupons abraded with a series of emery papers (grade 320-500-1000-1200) and then washed with distilled water and acetone. After weighing accurately, the specimens were immersed in 300 ml bottle, which contained 250 ml sulfuric acid 1 molar with and without addition of different concentrations of prepared compound.. After 4 h, the specimens were taken out, washed, dried, and weighed accurately. The experiments were carried out in triplicate. The average weight loss of three parallel carbon steel coupons could be obtained. Then the tests were repeated at different temperatures (298, 308 and 318).

Table (2): The chemical composition of steel sample

Element	Fe	C	Mn	Si	P	S
(%)	98.798	0.26	0.637	0.255	0.031	0.019

2.2.3.2. Polarization Techniques.

Polarization experiments were carried out in potentiostat Volta Lap 40 PGZ 301 with conventional three electrodes, a platinum counter electrode, saturated calomel electrode and working electrode.

Before measurement, the electrode was immersed in test solution at natural potential for one hour until a steady state was reached.

The potential was increased with scan rate of 1mVs^{-1} and started from potential of -800 to -350 mV.

2.2.4.3. Electrochemical Impedance Technique.

The Electrochemical Impedance Spectroscopy (EIS) measurements were carried out using alternating current (AC) signals of amplitude 10 mV peak to peak at the open circuit potential (OCP) in the frequency range of 100kHz-30mHz, with the same potentiostat used in polarization technique.

2.2.5. Antimicrobial Activity.

The antimicrobial activity of the synthesized products was measured against a wide range of test organisms comprising: (bacteria and fungi)

Source of microorganisms.

The different species of the tested organisms obtained from the unit of Micro Analytical Center, Cairo University, Cairo, Egypt.

Microorganisms.

Bacteria

Gram-Positive: Staphylococcus albus

Gram-Positive: Streptococcus faecalis

Gram-Positive: Bacillus subtilis

Gram-negative: Escherichia coli

Fungi

Candida albicans

Asperigllus flavus

An assay was made to determine the ability of the prepared compounds to kill or inhibit the growth of living microorganisms. The main technique used for these tests are show below.

Filter-paper disc-agar diffusion (Kirby-Bauer) [109-112].

1. Inoculate flask of melted agar medium with the organism to be tested.
2. Pour this inoculated medium into a Petri dish.
3. After the agar has solidified, a multilobed disc that impregnated with different antibiotics laid on top of the agar.

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4. The antibiotic in each lobe of the disc diffuses into medium and if the organism is sensitive to a particular antibiotic, no growth occur in a large zone surrounding that lobe (clear zone).
5. The diameters of inhibition zones were measured after 24-48 hours at 35-37 °C (for bacteria) and 3-4 days at 25-27 °C (for yeast and fungi) of incubation at 28 °C
6. Measure each clear zone and compare between them to determine the antibiotic which is more efficient inhibitor.