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Microbial Evaluation of Nanoparticles of Some New Lactosyl 5-Aryl 2,4 Dithiobiurets

Poonam T Agrawal*

Department of Chemistry, Shri R.L.T. College of Science, Akola-444001, Maharashtra, India

***Corresponding author:** Poonam T Agrawal, Department of Chemistry, Shri R.L.T. College of Science, Akola-444001, Maharashtra, India, E-mail: poonamagrawal2575@rediffmail.com

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Abstract

Dithiobiurets shows a wide range of application in medical and industrial field. In view of application of this dithiobiurets and Nanoparticles, we hereby report the microbial evaluation of some new lactosyl 5-Aryl 2,4 dithiobiurets.

Keywords:* *Dithiobiurets; Nanoparticles; Microbial evaluation

Introduction

In view of this application of lactosyl compounds and Nanoparticles in this we have synthesis to investigate the chemistry of this new compound with reference to their application. Properties of many conventional materials changed when form from nanoparticles. This is typically because nanoparticles have a greater surface area per weight than larger particles which causes them to be more reactive to some other molecule. Carbohydrate especially lactosyl compounds have been uses as starting material in the synthesis of nitrogen and sulphur containing open chain and cyclic compound which was already investigated by earlier workers. Nanoparticles exhibit new physical-chemical properties which are not observed either in individual molecules, or in bulk nanoparticles show unique properties that are significantly different from their bulk materials.

Nanostructure materials are attracting a great deal of attention because of their potential for achieving specific processes and selectivity, especially in biological and pharmaceutical applications [1-3]. Recent studies have demonstrated that especially formulated nanoparticles have good antibacterial activity [4,5]

Experimental

UV-visible Spectra is measured using UV Spectrophotomter by using model Single Beam UV-Visible Spectrophotometer with software (BI/CI/SP/SB-S-03) of Bio Era make. IR spectra were recorded on Perkin-Elmer spectrum RXI FTIR spectrophotometer (4000-450 cm^{-1}). ^{1}H NMR was recorded in $CDCl_3$ on Bruker DRX-300 spectrometer operating at 300 MHz.

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a) Synthesis of hepta-O-benzoyl-α-D-lactosyl bromide

The finally powdered lactose octabenzoate (0.03 M, 21.0 g) was added gradually to the brominating agent. After the addition the flask was kept for 2 h at room temperature. Then the reaction mixture with chloroform (130 ml) then the mixture was shaken vigorously for about 15 min. The resultant mixture was poured into ice cold water. The chloroform layer was then separated. It was washed several with aqueous sodium bicarbonate to remove excess of acetic acid followed by aqueous sodium metabisulphite to remove excess of bromine and finally 2-3 times with water. To the chloroform addition of petroleum ether afforded a solid (16.5 g). This solid was expected hepta-O-benzoyl-α-D-lactosyl bromide (yield 77%). It was purified by dissolving it in minimum quantity of chloroform and reprecipitating it with petroleum ether, m.p.168°C.

b) Preparation of lead thiocyanate

Lead thiocyanate was prepared by mixing aqueous solution of lead nitrate and ammonium thiocyanate. The white granular lead thiocyanate was filtered washed with distilled water and dried at 50°C.

c) Preparation of hepta-O-benzoyl-β-D-lactosyl isothiocyanate [6]

To a suspension of hepta-O-benzoyl-α-D-lactosyl bromide (21 g, 0.03 M) in sodium dried xylene (80 ml) was added lead thiocyanate (6 g, 0.03 M). The reaction mixture was then treated for microwave synthesis for about 3 min. This solution was then cooled and liberated lead bromide was removed by filteration. The xylene filterate was then treated with petroleum ether (60-80°C) with stirring, a white solid mass obtained (13 g). This solid was expected hepta-O-benzoyl-β-D-lactosyl isothiocyanate.

It was purified by dissolving it in minimum quantity of chloroform and reprecipitating it with petroleum ether, m.p. 118-120°C. [Found; C; 67.07, H; 4.46, N; 1.22, S; 2.9; $C_{62}H_{49}O_{17}NS$ requires; C; 66.96, H; 4.41, N; 1.26, S; 2.88%].

Preparation of 1-hepta O-benzyl-β-D-lactosyl 5 phenyl 2,4-Dithiobiuerts

A suspension of 4 g of Hepta O-benzyl-β-D lactosyl isothiocyante with 20 ml of benzene and 1 g of aniline thiourea was treated for microwave synthesis for about 3 min. This solution was then cooled and the benzene filtrate was then treated with petroleum ether (60-80°C) with stirring, a white solid mass obtained (13 g). This solid was expected 1-hepta-O-β-D lactosyl 5-phenyl 2,4-dithiobiurets.

It was purified by dissolving it in minimum quantity of chloroform and reprecipitating it with petroleum ether, m.p. 145-146°C.

Preparation of Nanoparticles 1-Hepta-O-benzyl-β-D-lactosyl-5-phenyl 2,4-Dithiobiurets

Take about 1 g of 1-Hepta-O-benzyl-β-D-lactosyl-5-phenyl 2,4 Dithiobiurets and dissolve complete 1 Hepta O-benzyl-β-D-lactosyl-5-phenyl 2,4 Dithiobiurets in the 50 ml of solvent in 250 ml beaker. Now put this beaker in sonicator. The highly penetrating acoustic waves are passed through mixture, which create high pressure bubbles in the beaker due to which breakdown of the bulk material is takes place and desired sized nanoparticles are formed. The size determination of nanoparticles is done by the X-ray diffraction studies (TABLE 1).

TABLE 1. **IR spectrum of 1-Hepta-O-benzyl-β-D-lactosyl-5-phenyl 2,4-Dithiobiurets** [7].

Absorption observed (Cm^{-1})	**Assignment**	**Absorption expected (Cm^{-1})**
3068	C-H Ar-stretching	3040-3010
1728	C=O stretching	1750-1735
1176	C-O stretching	1210-1153
1026, 909	Characteristic of lactose	1100-1000 and 910-900

710	Monosubstituted benzene	770-680

NMR Spectral Studies [8,9]

The NMR Spectrum of compound distinctly displayed signals due to N-H Proton at δ 9.05 and d 6.57 ppm, Aromatic Protons at δ 7.47-7.15 ppm, lactosyl protons at d 5.77-3.76 ppm.

Characterisation of Nanoparticles

1. **Charterisation using UV-visible spectrophotometer**

   Characteirisation of nanoparticles was done using visible Spectrophotomter by using model Single Beam UV-Visible Spectrophotometer with software (BI/CI/SP/SB-S-03) of Bio Era make. The UV-Visible Spectrocopy reveals the formation of nanoparticles by showing different absorption those from bulk material.

2. **Size determination of lactose octabenzoate nanoparticle by x-ray diffraction studies**

   From the X-Ray diffraction it comes to know that size of nano octabenzoate is 38 nm.

Antimicrobial activity comparison

All the compounds have been screened for antibacterial activity using cup plate agar diffusion method by measuring the inhibition zone in mm. The compounds were taken at a concentration of 1 mg/ml using dimethyl sulphoxide as solvent. Amikacin (100 mg/ml) was used as a standard for antibacterial activity. The compounds were screened for antibacterial activity against *Escherichia coli*, *Staphylococcus aureus*, *S. typhi*, *P. vulgaris* in nutrient agar medium (TABLE 2).

TABLE 2. **Antimicrobial activity of compounds.**

Antimicrobials	**Bulk***	**Nanoparticles****
E. coli	10 mm	14 mm
S. aureus	10 mm	15 mm
S. typhi	11 mm	16 mm
P. vulgaris	11 mm	15 mm
Amikacin	10 mm	20 mm
Clandamycine	12 mm	14 mm
DMSO	35 mm	28 mm

*including the well diameter of 8 mm. ** zone of inhibition in mm (15 or less) resistance, (16-20 mm) moderate and (more than 20 mm) sensitive.

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