

# Bio Studies of Novel Macrocyclic Sulfazan and Linear Sulfazan (Creation, Analysis and Anticancer Assessment)

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## Abstract

Sulfazan (Macrocyclic Sulfazan and Linear Sulfazan)- Compounds were created for the first time as a class of organic chemistry in 2019 globally for the first time by Prof. Dr. Nagham Aljamali, then she developed via several studies by using various types conditions and different basic medium like (Pipridine, Triethyl amine, Pyridine, 5 % Sodium hydroxide, etc.) while same researcher prepared chain of these innovated compounds and studied by many applications created and invented Sulfazan compounds as a pharmaceutical studies, anti-bio compounds, and since the references are few on Sulfazan, so we have prepared a series of them and studied their applications in various kinds fields to enrich this topic with references. Several spectroscopic studies have been used to prove their chemical structures which gave clear evidence of its chemical structures via many technical instruments like (FT IR-Spectra, <sup>1</sup>H.NMR-Spectra, <sup>13</sup>C.NMR-Spectra), other studies as anticancer-cancer breast.

**Keywords:** Sulfazan, Macrocyclic Sulfazan, Azo, Coupling, Cancer.

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## INTRODUCTION

**Sulfazan compounds:** In 2019, Sulfazan compounds were invented and prepared for the first time by the researcher Dr. Nagham Aljamali in the first researches<sup>(1-3)</sup>. Sulfazan are organic sulfur compounds with the chemical formula (Ar-N = N-S-R)<sup>(5-8)</sup> which act (Linear Sulfazan). It was created by several steps involving a coupling reaction with Azo with various types conditions (Pyridine, Piperidine, triethylamine, etc..) <sup>(3)</sup>.The researcher Dr. Nagham carried out many researches <sup>(9-15)</sup> in this field through several studies to provide a lot of references on preparation methods and the applications of these newly innovative compounds, and it has been proven that <sup>(5-8)</sup> for these compounds various types applications<sup>(16-25)</sup> work, including microstructure and nano-composites as drug carriers<sup>(26-35)</sup>, antifungal, antibacterial<sup>(36-40)</sup>, antitumor<sup>(2, 41-49)</sup>.

**Macrocyclic Sulfazan Compounds:** Newly invented compounds by researcher Dr. Nagham Aljamali in April 2019 for the first time <sup>(1-3)</sup>. She invited essential and primary methods for preparing these compounds and the conditions <sup>(5-7)</sup> for their interaction and the reaction factors that used in their preparation. It has been considered among the organic compounds for its importance in organic chemistry because it contains two highly reactive groups in many areas of chemistry because it contains pairs<sup>(50-54)</sup> of the free electrons and the donor atoms coordinate with the ions to form

complexes <sup>(4)</sup>. Sulfazan is also included in several medical applications like antibacterial <sup>(1)</sup>, antifungal <sup>(26-35)</sup> and anticancer compounds <sup>(36-42)</sup>, especially breast and laryngeal cancer<sup>(43-45)</sup>, and other applications <sup>(46-49)</sup> such as any other compounds<sup>(50-54)</sup>.

## Instruments and Experimental Part

FT.IR spectra were detailed on Fourier transform infrared spectrometer (FT-IR) in( FT-IR- 3600) infrared spectrometer via employing KBr Pellet technique., <sup>1</sup>H.NMR – spectra and <sup>13</sup>CNMR-Spectra were recorded in DMSO-d<sup>6</sup> as solvent using (TMS) as internal standard and chemical shifts are expressed as (δ ppm).

## EXPERIMENTAL METHODS

### Preparation of 5,5'-(1,4-phenylene)bis(4H-1,2,4-triazole-3-thiol) Compound [1]

Terephthalic acid (0.001 mol, 0.166g) was reacted with thiosemicarbazide (0.002 mol,0.182g) in 30 ml absolute ethanol in basic medium 5% NaOH in rotation and refluxing step for 30 hrs, then cyclization step, the product filtered, dried, recrystallized., by following literatures <sup>(1-3)</sup>.

### Creation of Innovative Linear Sulfazan Compound [2]

Compound [1] was (0.001 mol, 0.276g) dissolved in basic solution of (Piprydine) then reacted with (0.002 mol, 0.286g) of diazo salt of Naphthalen-1-amine via many steps in basic solution to formation invented Sulfazan after (48 hrs), the product filtered, dried, washed by distilled water, recrystallized to yield Innovative Linear Sulfazan by following literatures<sup>(1-3)</sup>.

#### Creation of Innovative Linear Sulfazan Compound [3]

Compound [1] was (0.001 mol, 0.276g) dissolved in basic solution from (Piprydine) then reacted with (0.002 mol, 0.218g) of diazo salt of 3-Aminophenol via many steps in basic solution to formation invented Sulfazan after (48 hrs), the product filtered, dried, washed by distilled water, recrystallized to yield Innovative Linear Sulfazan according to procedures<sup>(1-3)</sup>.

#### Creation of Innovative Linear Sulfazan Compound [4]

Compound [1] was (0.001 mol, 0.276g) dissolved in basic solution from (Piprydine) then reacted with (0.002 mol, 0.290g) of diazo salt of 2-Amino-5-Nitrothiazole via many steps in basic solution to formation invented Sulfazan after (60 hrs), the product filtered, dried, washed by distilled water, recrystallized to yield Innovative Linear Sulfazan according to procedures<sup>(1-3)</sup>.

#### Creation of Innovative Linear Sulfazan Compound [5]

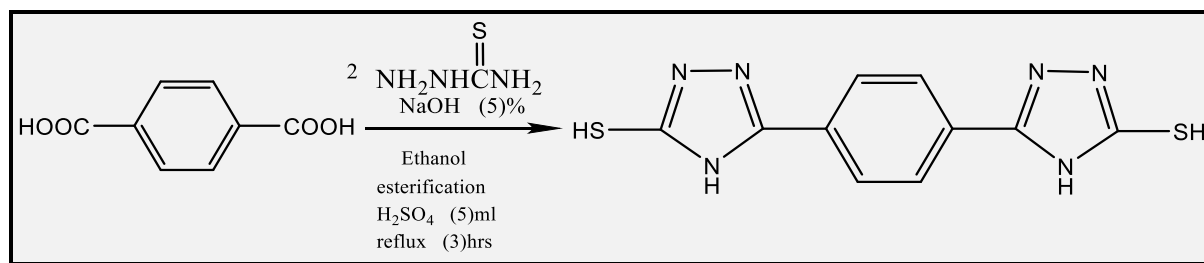
Compound [1] was (0.001 mol, 0.276g) dissolved in basic solution from (Piprydine) then reacted with (0.002 mol, 0.228g) of diazo salt of 2-Amino-5-methylthiazole via many steps in basic solution to formation invented Sulfazan after (55 hrs), the product filtered, dried, washed by distilled water, recrystallized to yield Innovative Linear Sulfazan according to procedures<sup>(1-3)</sup>.

#### Creation of Inventive Macrocylic Sulfazan Compound [6]

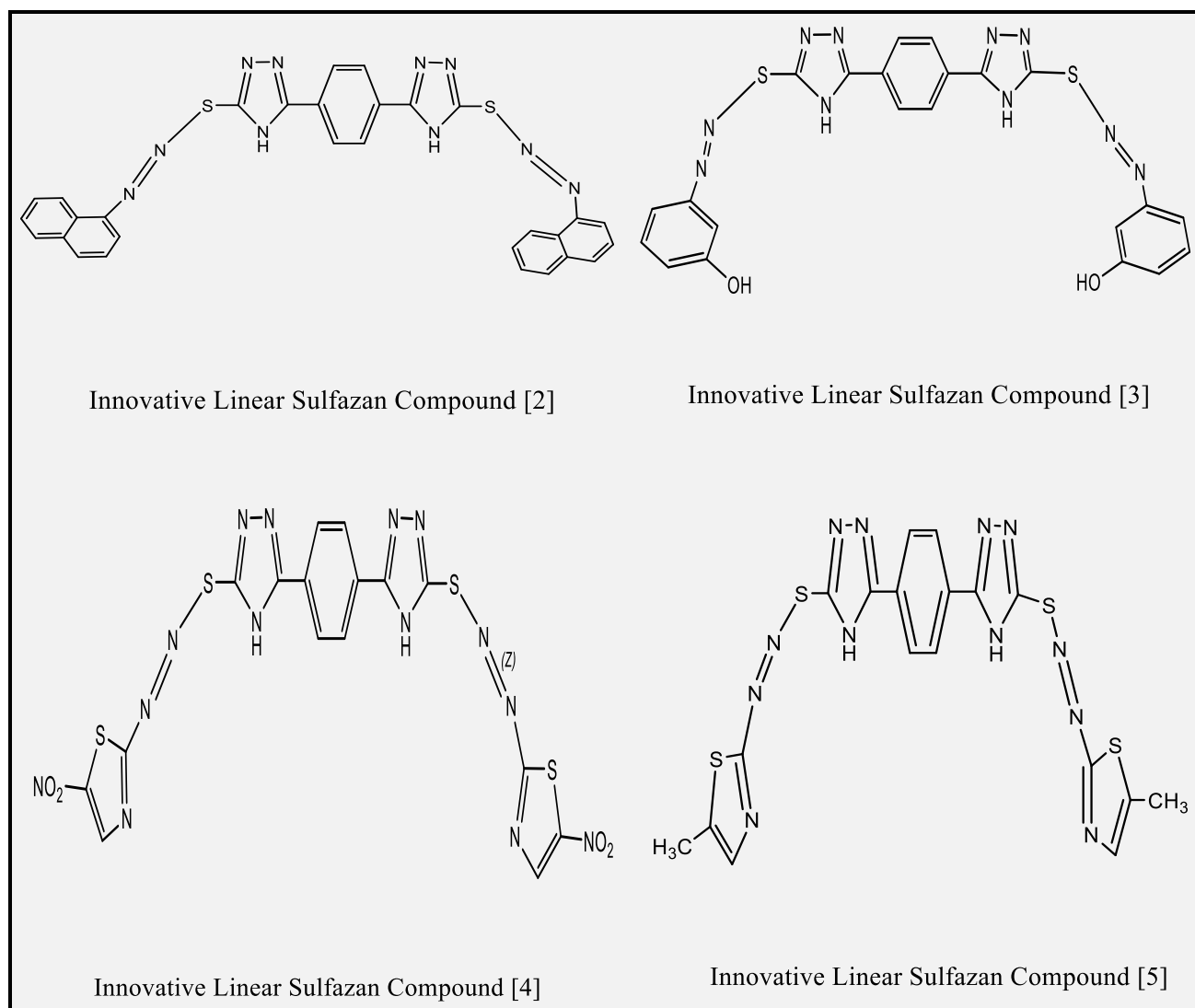
Compound [1] was (0.001 mol, 0.276g) dissolved in basic solution from (Piprydine) then reacted with (0.001 mol, 0.290g) of diazo salt of Trimethoprim via many steps in basic solution to formation invented Macrocylic Sulfazan after (60 hrs), the product filtered, dried, washed by distilled water, recrystallized to yield Invented Macrocylic Sulfazan by following literatures<sup>(1-3)</sup>.

#### Creation of Inventive Macrocylic Sulfazan Compound [7]

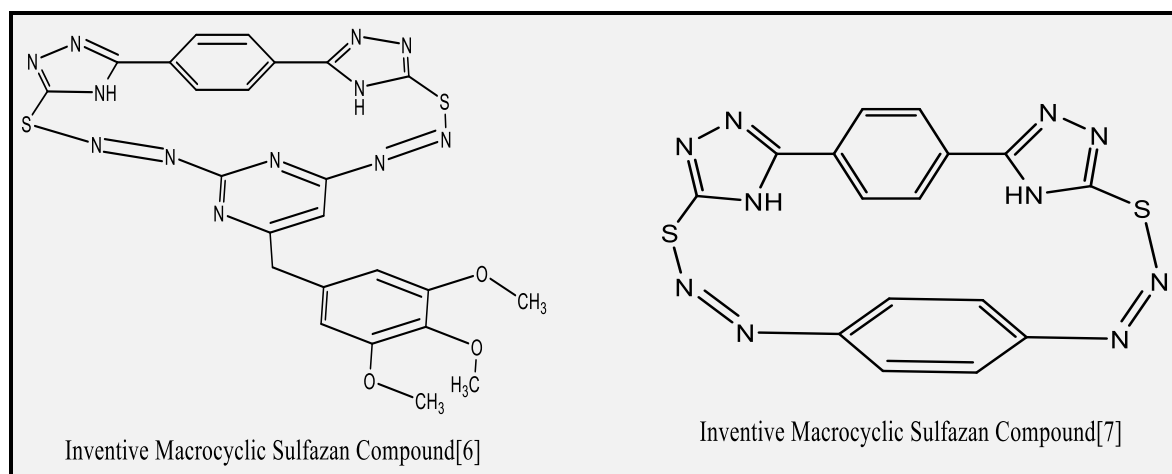
Compound [1] was (0.001 mol, 0.276g) dissolved in basic solution from (Piprydine) then reacted with (0.001 mol, 0.108g) of diazo salt of Benzene-1,4-diamine via many steps in basic solution to formation invented Macrocylic Sulfazan after (60 hrs), the product filtered, dried, washed by distilled water, recrystallized to yield Invented Macrocylic Sulfazan according to procedures<sup>(1-3)</sup>.



Scheme 1: Preparation of Compound [1]



Scheme 2: Creation of Invented Linear Sulfazan Compounds [2-5]



Scheme 3: Creation of Invented Macrocylic Sulfazan Compounds [6, 7]

## RESULTS AND DISCUSSION

Recently study, various of Invented Macrocylic Sulfazan

and linear Sulfazan have been created in same procedure that followed and invented<sup>(1-5)</sup> by Dr. Nagham in year 2019 that got her patent to invention of Linear Sulfazan and

Macrocylic Sulfazan compounds <sup>(19,22)</sup>, then several studies were carried out to improve these innovative compounds by the using of spectral identification like : <sup>1</sup>H.NMR spectra, FT.IR- Spectra, <sup>13</sup>CNMR-Spectra, other studies represented by anticancer –breast evaluation.

**Spectral Indications**

**FT.IR- Spectral Indication of Liner Sulfazan and Macrocylic Sulfazan compounds:** It indicated to formation our Invented Sulfazan compounds by appearance of band at :(2400) cm<sup>-1</sup> for (SH) thiol group in compound

[1] that disappeared in Sulfazan compounds [2, 3, 4, 5, 6, 7] and formation new band instead of it represented by (-S-N=N-) Sulfazan group at : {(1400, 1454 ), (1440, 1501),(1400, 1452), (1400, 1453), (1440, 1508), (1432, 1482)} cm<sup>-1</sup> respectively,also appearance band (-S-N) in the prepared Sulfazan compounds [2,3, 4, 5, 6, 7] at :{(1257) (1247), (1246), (1244), (1249),(1257) } cm<sup>-1</sup> respectively, all frequencies clarified according to reference<sup>(28)</sup>, and other functional groups in figures (1-7):

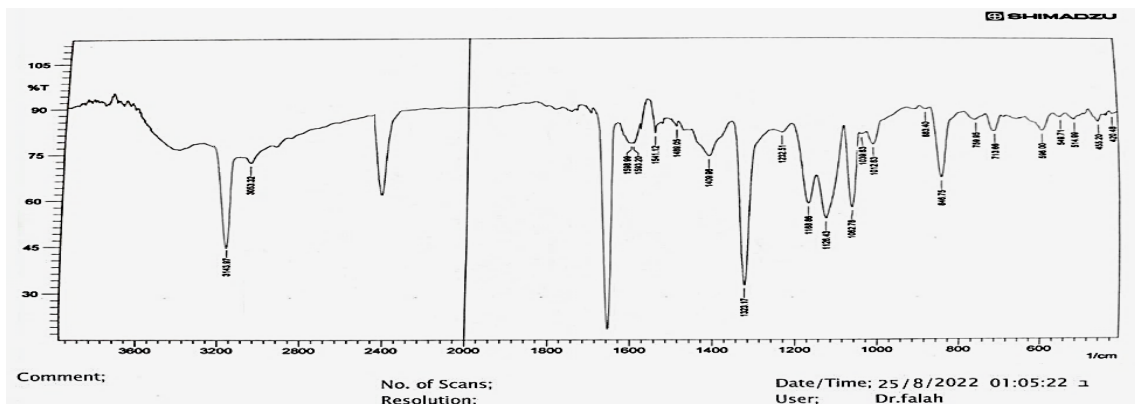


Fig (1): I.R of Compound [1]

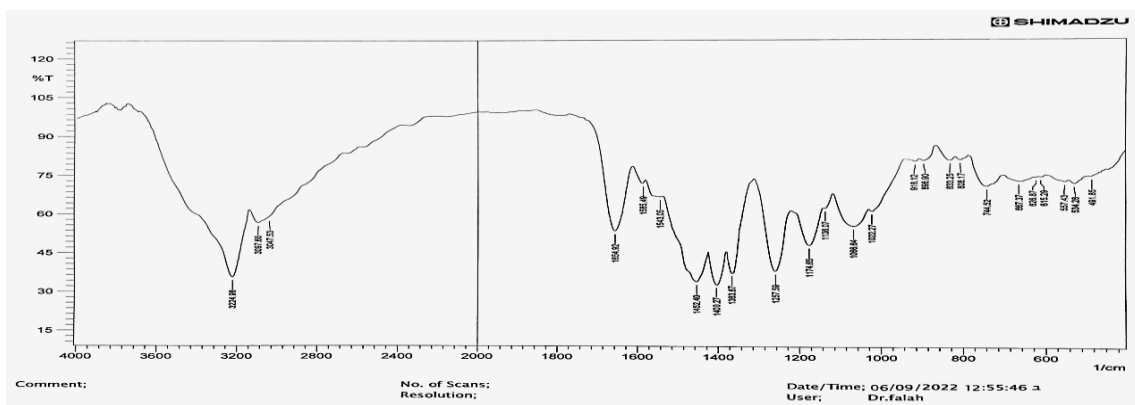


Fig (2): I.R of Compound [2]

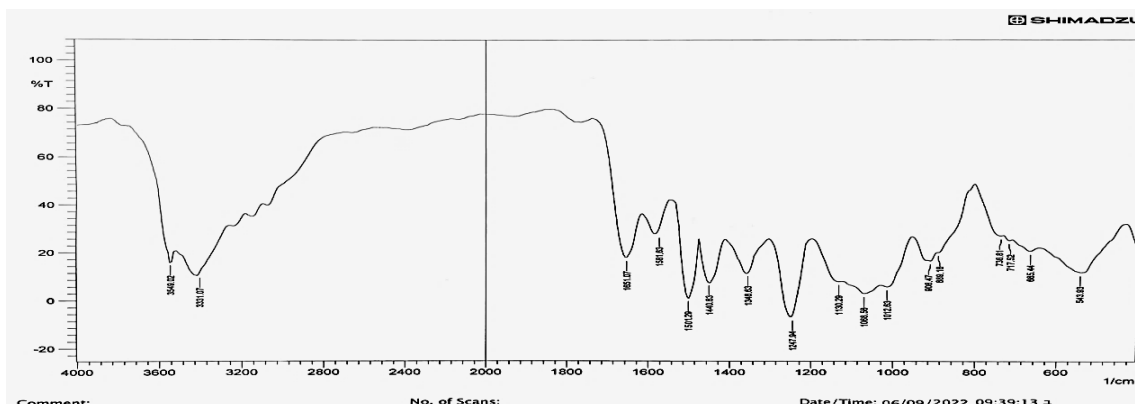


Fig (3): I.R of Compound [3]

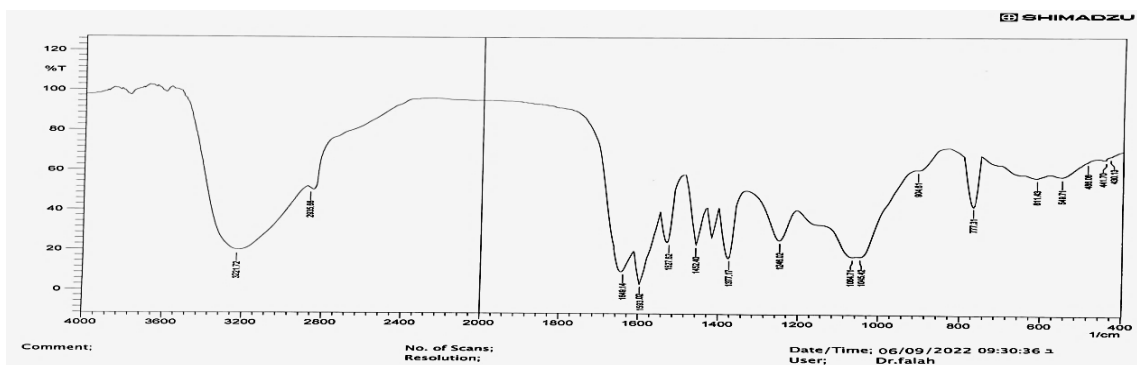


Fig (4): I.R of Compound [4]

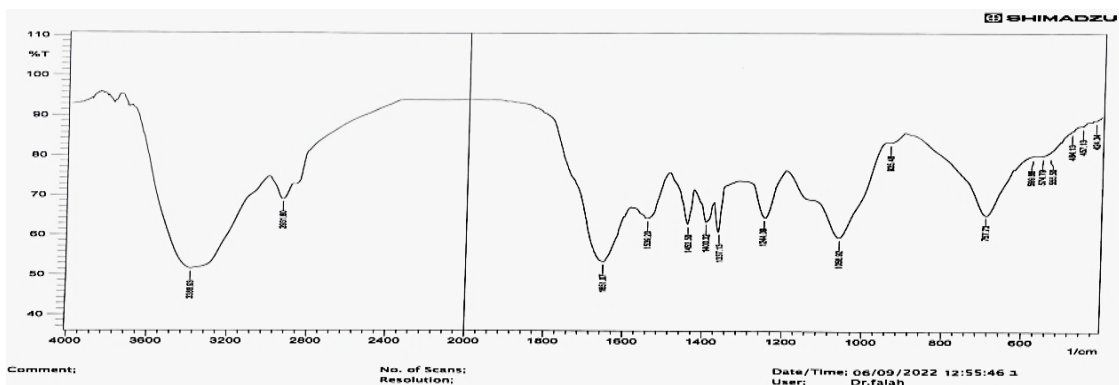


Fig (5): I.R of Compound [5]

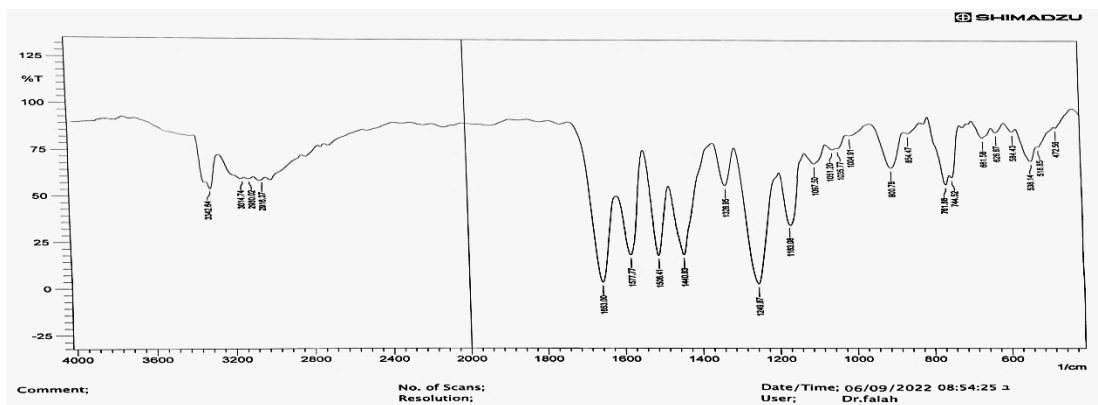


Fig (6): I.R of Compound [6]

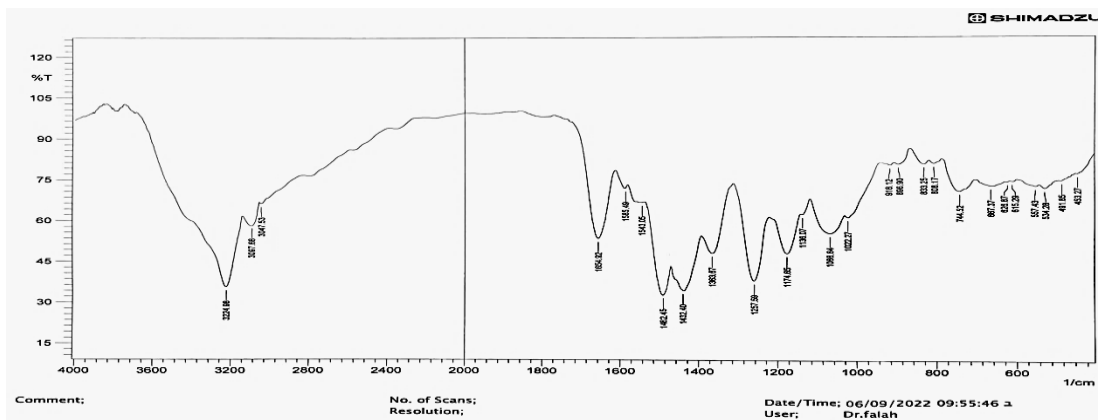


Fig (7): I.R of Compound [7]





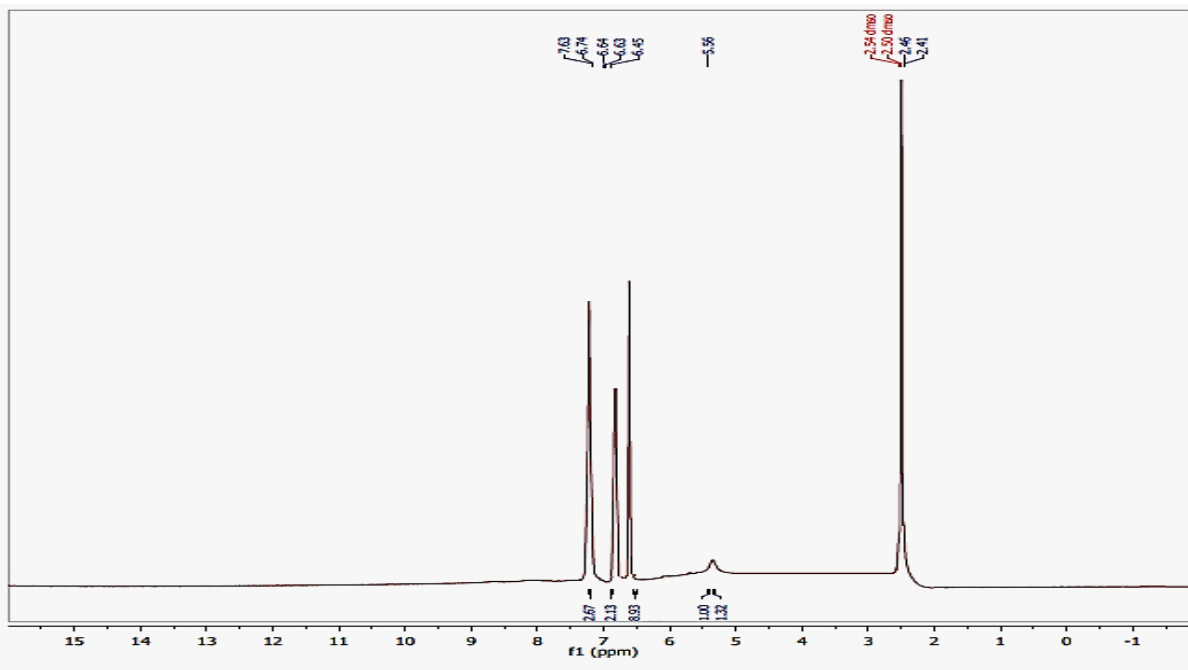


Fig (14): H.NMR of Compound [7]

**C.NMR - Spectral Indication of Liner Sulfazan and Macrocylic Sulfazan compounds :** It indicated to formation our Invented Sulfazan compounds by appearance of peaks at :  $\delta$  (106.0-141.9) for signals of Aromatic carbon atoms in Sulfazan compounds [1, 2, 3, 4, 5, 6, 7], also peak at :  $\delta$  (60.4) for signal of carbon atom of methoxy group (-

OCH<sub>3</sub>) in Sulfazan compound [6], other peak at : $\delta$  (12.50) for signal of carbon atom of methylene group (-CH<sub>2</sub>) in Sulfazan compound [6], all other signals clarified according to reference<sup>(28)</sup>, and other functional groups in figures (15-18):

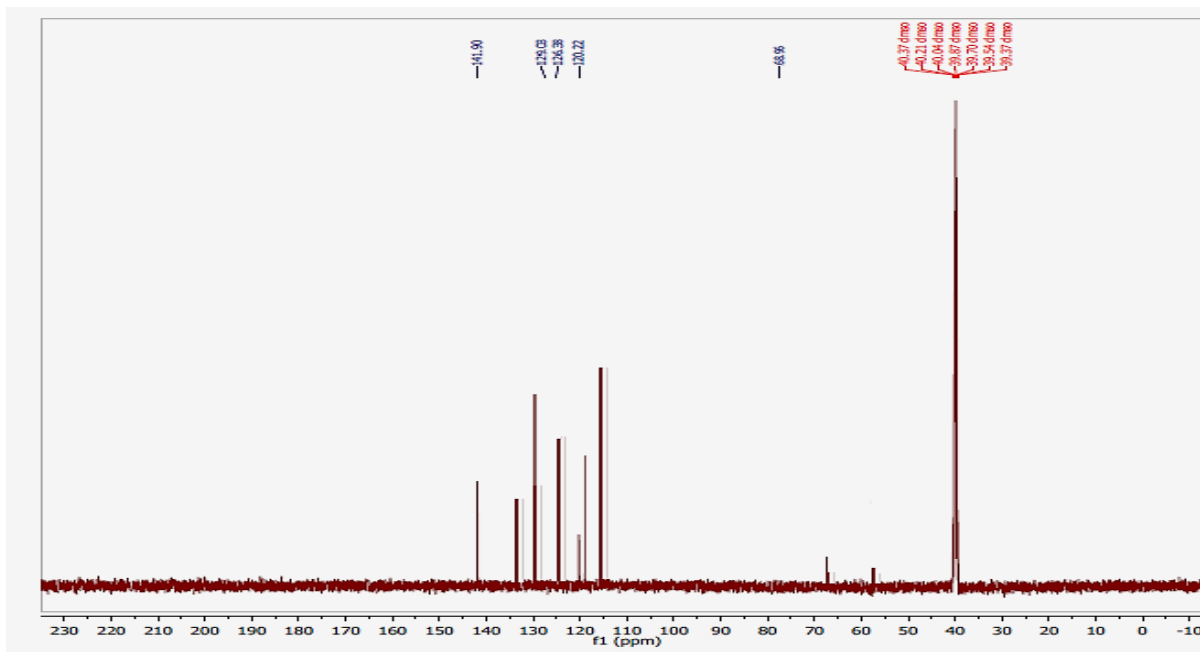


Fig (15): C.NMR of Compound [2]

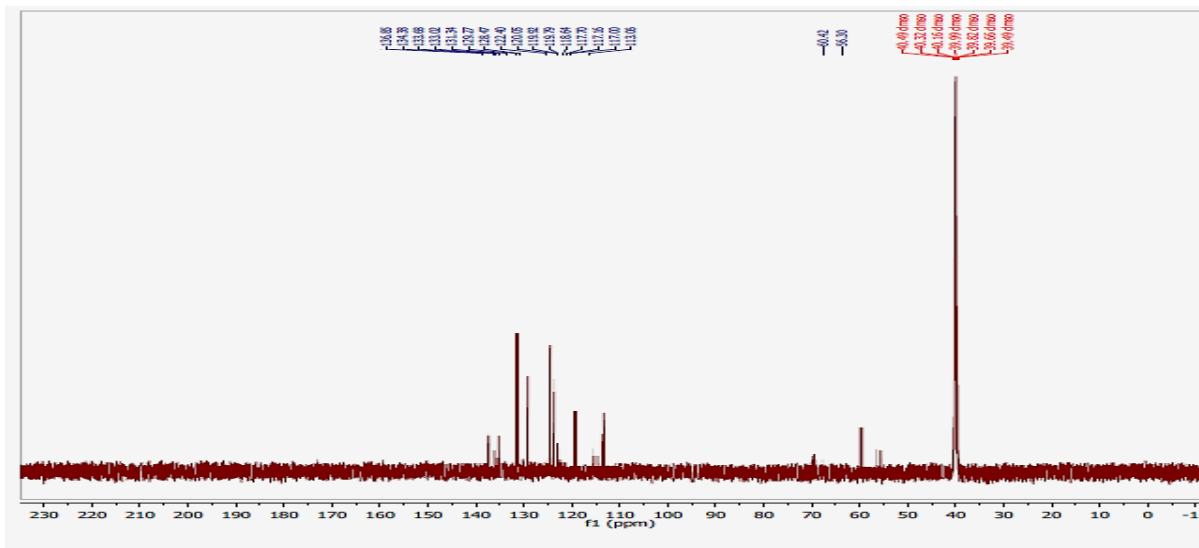


Fig (16): C.NMR of Compound [3]

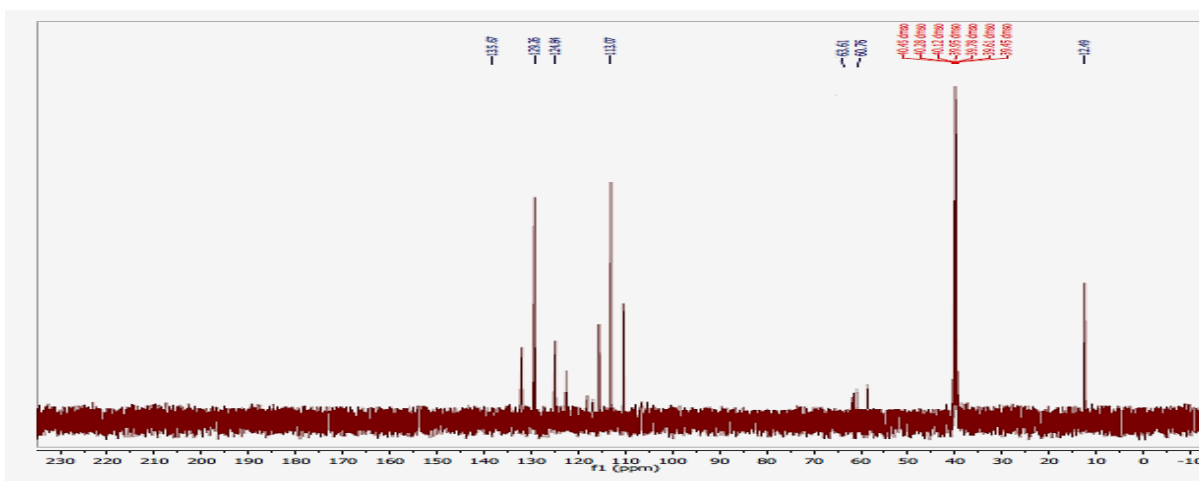


Fig (17): C.NMR of Compound [5]

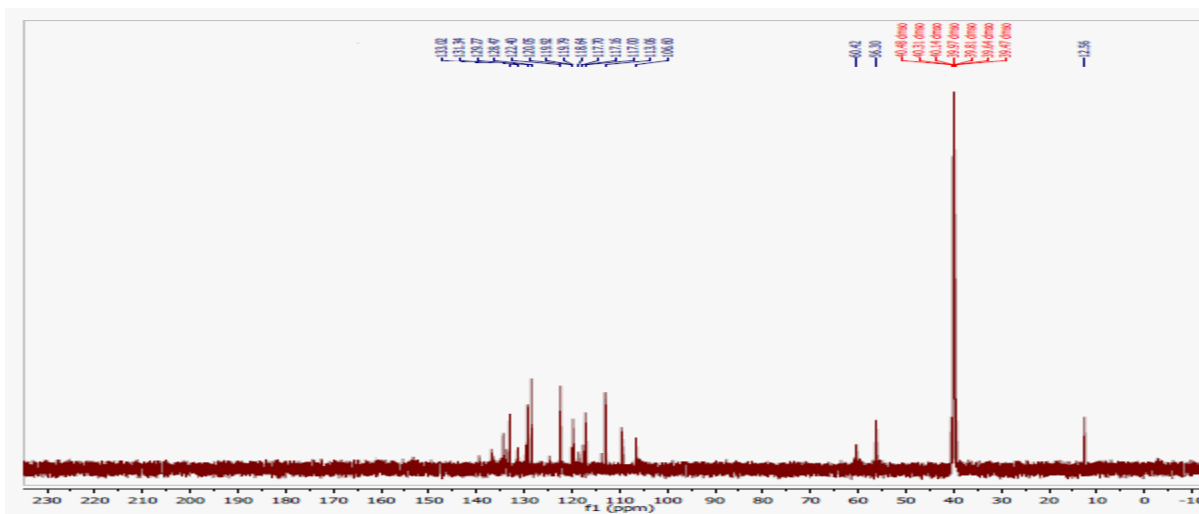


Fig (18): C.NMR of Compound [6]

**Anticancer-Assay<sup>(1-3)</sup>**

Two compounds were selected for anticancer evaluation (breast cancer) represented by Linear Sulfazan Compound [4] and Macrocylic Sulfazan compound [7] by MTT-Test

of two types of Cells (MCF-7) as Cancer cell and (MCF- 10 A)as healthy cell according to studies<sup>(1-3)</sup>. all data in Figures(19, 20), Tables(1,2).

Table. (1): Cytotoxic Activity of Linear Sulfazan Compound{4} on Breast Cancer Cells line (MCF-7) and Healthy Cells (MCF-10A) at the same concentration using 24 hrs., MTT test 37<sup>o</sup>c

Con. (µg.mL <sup>-1</sup> )	Mean Percentage (%) for each cell line			
	MCF-7 / IC <sub>50</sub> = 20.37		MCF-10A / IC <sub>50</sub> = 189.75	
Linear Sulfazan Compound {4}	Cancerous line cells of MCF-7		Normal line cells of MCF-10A	
	Cell Viability	Cell Inhibition	Cell Viability	Cell Inhibition
15. 62	95.26	4.74	94.11	5.89
31. 25	80.61	19.39	91.48	8.52
62. 5	77.0	23.0	92.4	7.60
125.0	75.87	24.13	96.25	3.75
250	68.62	31.38	94.37	5.63
500	47.86	52.14	94.19	5.81
Control	100		95.92	4.08

Table. (2): Cytotoxic Activity of Macrocylic Sulfazan Compound{7} on Breast Cancer Cells line (MCF-7) and Healthy Cells (MCF-10A) at the same concentration using 24 hrs., MTT test 37<sup>o</sup>c

Con. (µg.mL <sup>-1</sup> )	Mean Percentage (%) for each cell line			
	MCF-7 / IC <sub>50</sub> = 28.0		MCF-10A / IC <sub>50</sub> = 184.09	
Macrocylic Sulfazan Compound {7}	Cancerous line cells of MCF-7		Normal line cells of MCF-10A	
	Cell Viability	Cell Inhibition	Cell Viability	Cell Inhibition
15. 62	97.97	2.03	92.92	7.08
31. 25	83.77	16.23	91.82	8.18
62. 5	82.57	17.43	92.66	7.34
125.0	79.25	20.75	94.54	5.46
250	56.13	43.87	93.36	6.64
500	42.04	57.96	93.55	6.50
Control	100		95.92	4.08

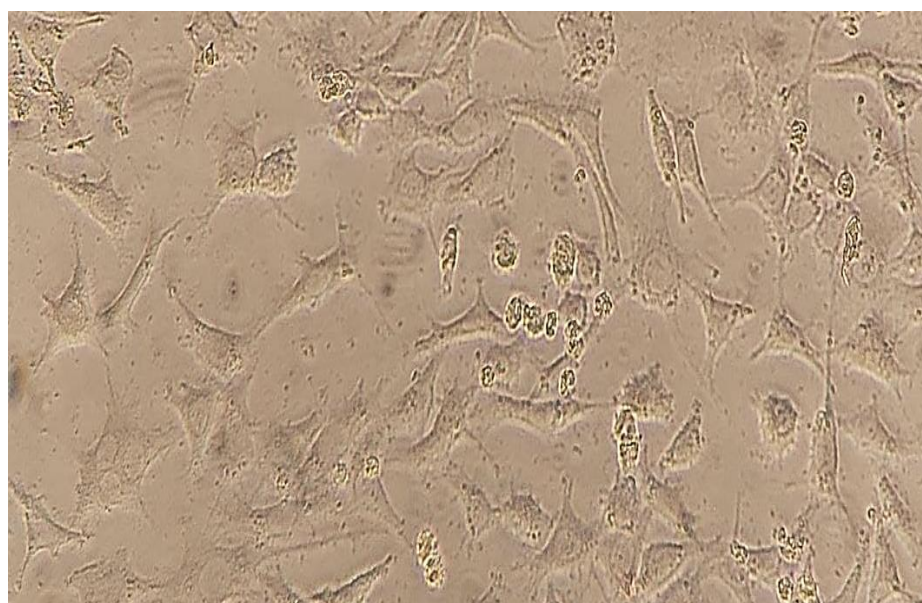


Fig. 19:Anti-cancer activity of Macrocylic Sulfazan Compound{7}on ( MCF-7) at 500µg/ml

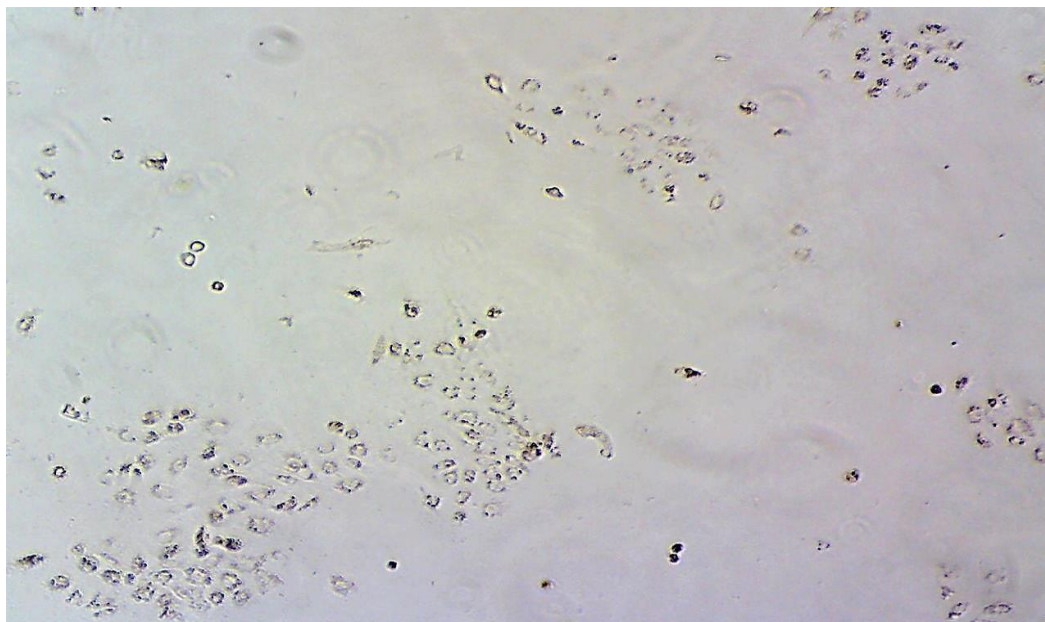


Fig. 20: Anti-cancer activity of Macrocylic Sulfazan Compound (MCF-A 10) at 500 µg/ml

## CONCLUSIONS

From results of anticancer test, we noted that Linear Sulfazan Compound [4] has higher efficiency towards Cancer cells and high inhibition on cancer cells than Macrocylic Sulfazan compound [7] due to presence of double groups of Sulfazan in same compound.

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