

# Synthesis and Characterization of O-(carboxyl) Chitosan Schiff Base Derivatives and Study Antibacterial Activity

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## ABSTRACT

2-mercaptobenzothiazol [1] was reacted with anhydrous carbonate sodium and  $\text{ClCH}_2\text{COOH}$  in distilled water to produce a compound [2]. Compound [3] was synthesized by reaction compound [2] with  $\text{SOCl}_2$  in benzene. O,N-chitosan derivative [4] had synthesized by reacted chitosan with compound [3] in trichloromethane and pyridine. O-chitosan derivative synthesized according to the method of Fischer and Speier from the reaction of chitosan with compound [2] in distilled water in acidic media to produce compound [5]. O-(carboxyl) chitosan Schiff base derivatives [6-9] had synthesized by reaction O-(carboxyl) chitosan [5] with aromatic aldehydes (syringaldehyde, 3-methoxy,4-hydroxybenzaldehyde, 4-nitrobenzaldehyde, 4-bromobenzaldehyde).

The structure of produced compounds has been characterized through their spectral and analytical data, as in fourier transform infrared (FTIR) spectra,  $^1\text{H-NMR}$ , UV-vis spectroscopy, elemental analysis (CHNS), scanning electron microscope (SEM), differential scanning calorimetry (DSC), and thermo gravimetry analysis (TGA). Finally, study the antibacterial activity of some prepared polymers.

**Keywords:** 2-mercapto benzothiazole, Chitosan derivatives, Schiff base.

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**Conflict of interest:** None

## INTRODUCTION

Benzothiazole derivatives take of great interest in chemistry and pharmacology due to its assorted biological activity.<sup>1,2</sup> The benzothiazole has to get the attention of medicinal chemists because of their broad range of biological activities, which comprise anti-inflammatory,<sup>3</sup> antitumor,<sup>4</sup> vasodilators,<sup>5</sup> antitubercular, antifungal,<sup>6</sup> antimicrobial,<sup>7</sup> anticancer,<sup>8</sup> antidiabetic,<sup>9</sup> and antibacterial activities.<sup>10</sup>

Chitosan has excellent properties: low toxicity, biodegradability, biocompatibility, and bioactivity for its excellent properties chitosan have received interest in many fields, including antimicrobials, biomedical materials, cosmetics, food additives, and agricultural material.<sup>11,12</sup> Chitosan can be modified into many derivatives through physical and chemical processes. In the characterized and also, it acts as a very good chelating agent. Even though it has the advantages mentioned, it also has serious demerits, which limit its use to basic conditions. They are insoluble in an aqueous medium, and it decomposes in acidic.<sup>13</sup>

## MATERIALS AND METHODS

### Materials

The chemicals were supplied from BDH, CDH, and SCR.

### Instrumentation

Spectra of FTIR were registered on a Shimadzu FTIR-8400s, in range 400 to 4,000  $\text{cm}^{-1}$ , utilizing potassium bromide disk.  $^1\text{H-NMR}$  spectra have been performed by the company: Ultra Shield 300 MHz, Bruker, University of Tehran, Iran. TMS was used as an internal standard with DMSO as a solvent.

### Preparation of Compounds

#### *Synthesis of 2-(benzo[d]thiazol-2-ylthio)acetic acid [2]<sup>14</sup>*

An aliquot (0.001 mol) of hot compound [1] mixed with 0.002 mol  $\text{Na}_2\text{CO}_3$  in 15 mL of solvent (distilled water), then added 0.001 mol of  $\text{ClCH}_2\text{COOH}$  and refluxed the solution for 6 hours. After cooling, used concentrated HCl to acidify the solution (pH = 2). The product was filtered, washed with  $\text{H}_2\text{O}$ , and recrystallized by absolute EtOH. MP = 80 to 82, yield 90%. Elemental analysis of compound [2], calculated: C% = 48; H% = 3.11; N% = 6.22; S% = 28.44. Found: C% = 39.87; H% = 3.23; N% = 6.34; S% = 28.28.

#### *Synthesis of 2-(benzo[d]thiazol-2-ylthio)acetyl chloride [3]<sup>15</sup>*

0.01 mol of compound [2] dissolved in dry benzene 15 mL with  $\text{SOCl}_2$  (0.01 mol), then, the solution was refluxed for 8 hours. The surplus of thionyl chloride and benzene were taken off under vacuum after cooling.

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### Synthesis of Polymer [4]<sup>16</sup>

0.01 mol of chitosan was marinated in 50 mL pyridine and trichloromethane (1:1) for 20 hours. To the system was added 0.01 mol of compound [3] under a water-ice bath. The mixture stirred at 100°C for 14 hours, then cooled to room temperature and spilled into a beaker containing 50 mL methanol. After that, cooled at 4°C, then filtered. The precipitate was washed with CH<sub>3</sub>OH and dried at 50°C.

### Synthesis of Polymer [5]<sup>17</sup>

Compound [5] synthesized 0.01 mol of chitosan was hung in 25 mL of H<sub>2</sub>SO<sub>4</sub> (2M), added 0.01 mol of compound [2] to this solution, then was refluxed for 8 hours and cooled to room temperature (r.t.). The pH was settled to 7 by neutralizing with sodium bicarbonate, then precipitated in acetone, filtered, cleaning with acetone, and dried at 60°C in oven.

### Synthesis of Polymers [6-9]<sup>18</sup>

Compound [5] was dissolved in a mixed solution of ethanol with glacial acetic acid and stirred for 30 minutes at r.t. Then, a variety of aromatic aldehydes added to the mixture (4-hydroxy-3,5-dimethoxybenzaldehyde, vaniline, 4-nitrobenzaldehyde,

4-bromobenzaldehyde). The mixture was magnetically stirred and heated at 60°C for 24 hours, then cooled. The crude product was washed with ethanol. The product was dried at r.t. for 24 hours.

## RESULTS AND DISCUSSION

Compound [2] was synthesized through the reaction of 2-mercapto benzothiazole [1] with anhydrous carbonate sodium and ClCH<sub>2</sub>COOH in distilled water. FTIR of compound [2], Figure 1 displayed the appearance bands at 3,400 to 2,400, and 1,695 cm<sup>-1</sup>, which referred to O-H and carbonyl of carboxylic acid, respectively, and disappearance of the SH group at 2,335 cm<sup>-1</sup>. Compound [3] Figure 2 has been synthesized by the reaction of the compound [2] with SOCl<sub>2</sub> in dry benzene. FTIR of compound [3] showed the vanishing of band at 3,400 to 2,400 cm<sup>-1</sup>, due to the OH group of carboxylic acid, and appearance of band at 1,779 cm<sup>-1</sup> connected to acyl chloride.

O,N-chitosan derivative [4] was prepared through the reaction of the compound [3] with chitosan in trichloromethane, and pyridine as a solvent. FTIR of compound [4] Figure 3 demonstrated a big peak at 3,438 cm<sup>-1</sup>, attributed to the stretch

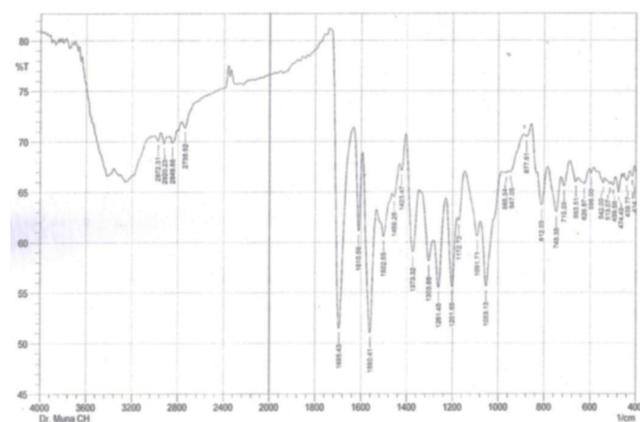


Figure 1: FTIR for compound [2]

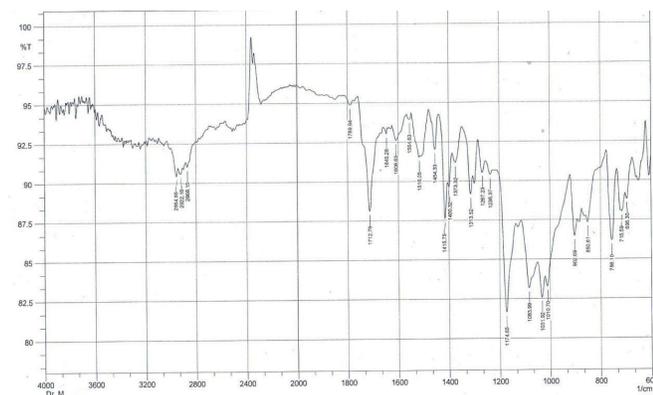


Figure 2: FTIR for compound [3]

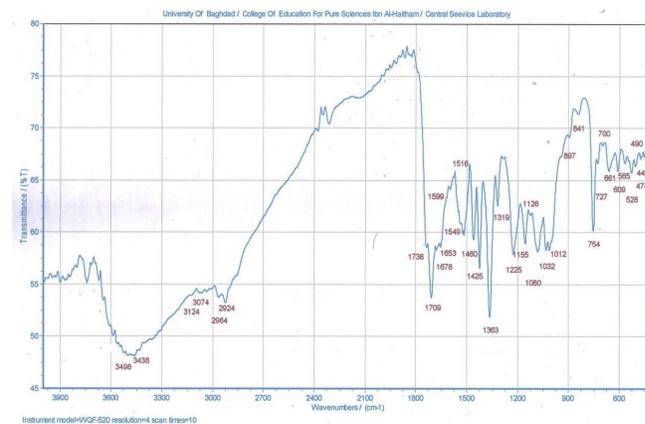


Figure 3: FTIR for compound [4]

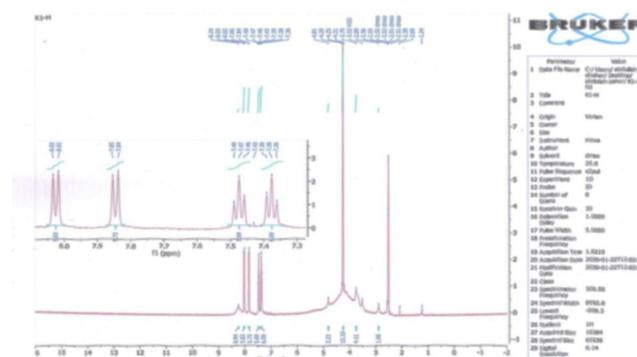


Figure 4: <sup>1</sup>H-NMR for compound [4]

Table 1: FTIR of polymers [6,7,9]

Comp. No.	$\nu$ (O-H) and (N-H)	$\nu$ (C-H) aliph.	$\nu$ (C=O) ester.	$\nu$ (C=N)	$\nu$ (C=C)	$\nu$ (-CH <sub>2</sub> -O-CO)	$\nu$ (C-O-C)
[6]	3,464	2,941–2,850	1,722	1,639	1,577	1,284	1,020
[7]	3,400	2,931–2,872	1,714	1,645	1,590	1,288	1,066
[9]	3,398	2,924–2,854	1,712	1,647	1,585	1,286	1,068

of O-H and N-H from the intra- and inter-molecular hydrogen bonding of chitosan molecules, and showed off new bands at 1,738, 1,678  $\text{cm}^{-1}$ , which refer to the carbonyl group of ester and amide, respectively.<sup>14</sup> While, the  $^1\text{H-NMR}$  of compound [4] Figure 4 shows the signal at 1.24 ppm existed because of the presence of  $\text{CH}_3$  of N-alkylated of glucosamine residue, a signal at 2.09 ppm assigned to H-2, signal at 2.89 ppm due to  $\text{CH}_2\text{O}$ , signals at 3.75 to 4.25 ppm corresponded to the non-anomeric proton (H-3, H-4, H-5, and H-6) of chitosan, singlet signal at  $\delta$  4.29 ppm for proton of  $\text{SCH}_2$  (4H,s), signal at 4.81 ppm refer to H-1 of chitosan, multiple signals at  $\delta$  7.36 to 8.03 for aromatic protons, and singlet signals at  $\delta$  8.24 for  $\text{NHC=O}$  group.<sup>17,19,20</sup>

O-chitosan derivative [5] was synthesized through the reaction compound [2] with chitosan in distilled water in acidic media. FTIR of compound [5], Figure 5 showed a big peak at 3,411  $\text{cm}^{-1}$ , which refers to the stretch of N-H and O-H from the intra-and extra-molecular hydrogen bonding of chitosan molecules, and showed off a new band at 1,707  $\text{cm}^{-1}$ , which refers to C=O group of ester.<sup>15</sup> Compounds [6-9] had synthesized by the reaction of O-(carboxyl) chitosan [5] with aromatic aldehydes. FTIR of compound [8] Figure 6 shows a presence of a big peak at 3,448  $\text{cm}^{-1}$ , which refers to the stretch of O-H and N-H from the intra-and extra-molecular hydrogen bonding of chitosan molecules, and showed a new band at 1,710  $\text{cm}^{-1}$ , due to C=O group of ester, and

appeared 1,641, corresponding to the C=N characteristic vibration of imines.<sup>20</sup> All the spectral data for these polymers in Table 1. While, the  $^1\text{H-NMR}$  of compound [8] Figure 7 shows the signal at 1.13 ppm existed because of the presence of  $\text{CH}_3$  of N-alkylated of glucosamine residue, a signal at 2.21 ppm assigned to H-2, signals at 3.34 to 4.25 ppm, corresponded to the non-anomeric proton (H-1, H-3, H-4, H-5, and H-6) of chitosan, singlet signal at  $\delta$  4.26 ppm for proton of  $\text{SCH}_2$  (2H,s), multiple signals at  $\delta$  8.01 to 8.43 for aromatic protons, and singlet signals at  $\delta$  10.13 for H-C=N group.<sup>19,20</sup>

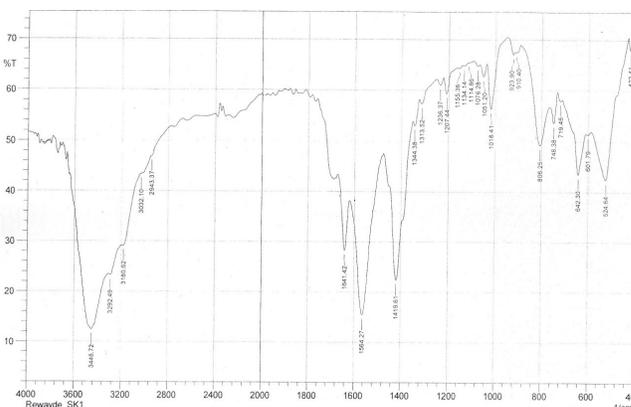


Figure 6: FTIR of compound [8]

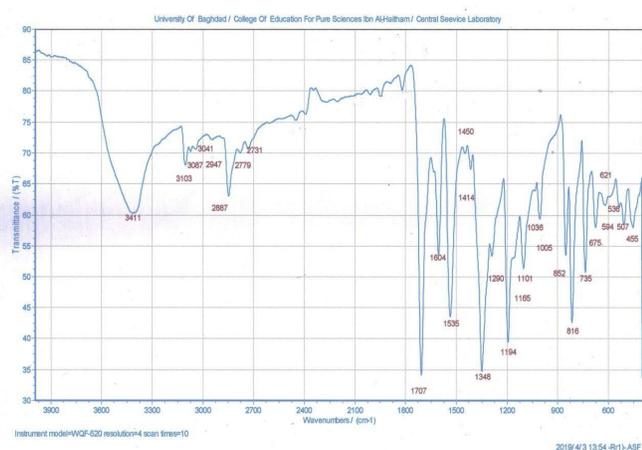
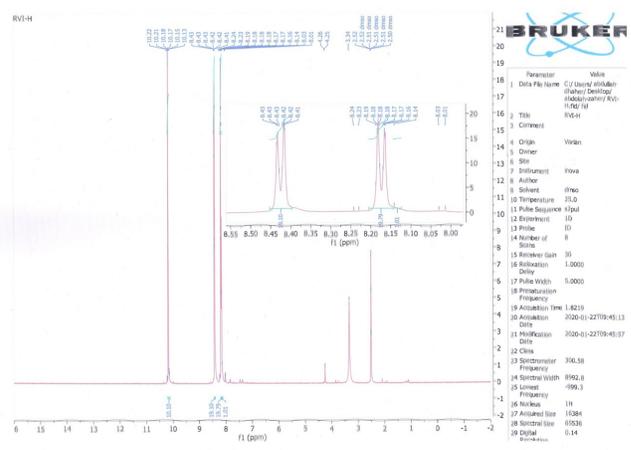


Figure 5: FTIR of compound [5]



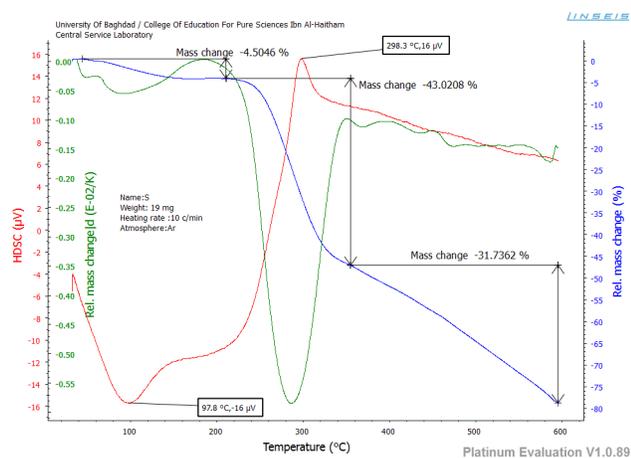


Figure 8: Thermal analysis of pure CS

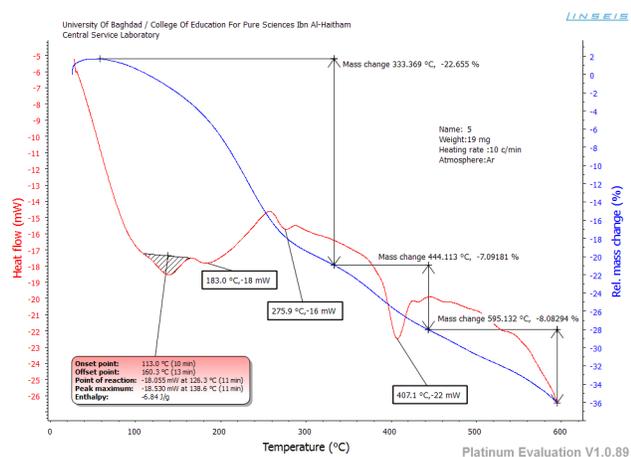


Figure 9: Thermal analysis of polymer [4]

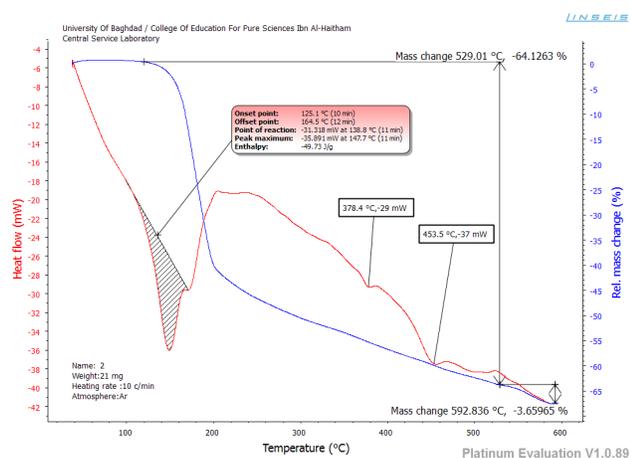


Figure 10: Thermal analysis of polymer [7]

The UV-vis spectrum of compounds [4] and [6] showed the absorption at 320 to 402, 335 to 422, and 332 due to ( $\pi-\pi^*$ ) and ( $n-\pi^*$ ), respectively.

### Thermogravimetric Analysis<sup>21</sup>

Figures 8-10 show the curves (DSC/TG) of pure chitosan and prepared polymers were acquired at the rate of heat  $10^\circ\text{C}\cdot\text{min}^{-1}$

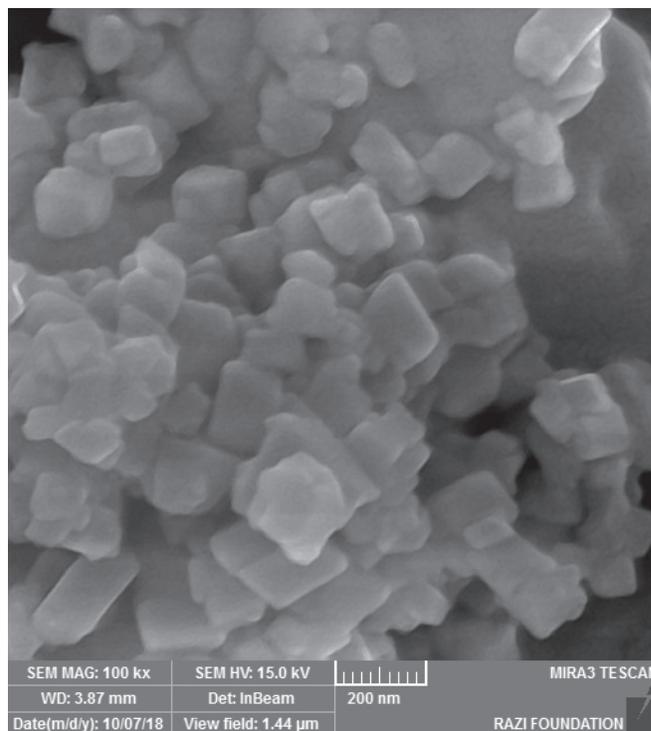


Figure 11: SEM of polymer [4]

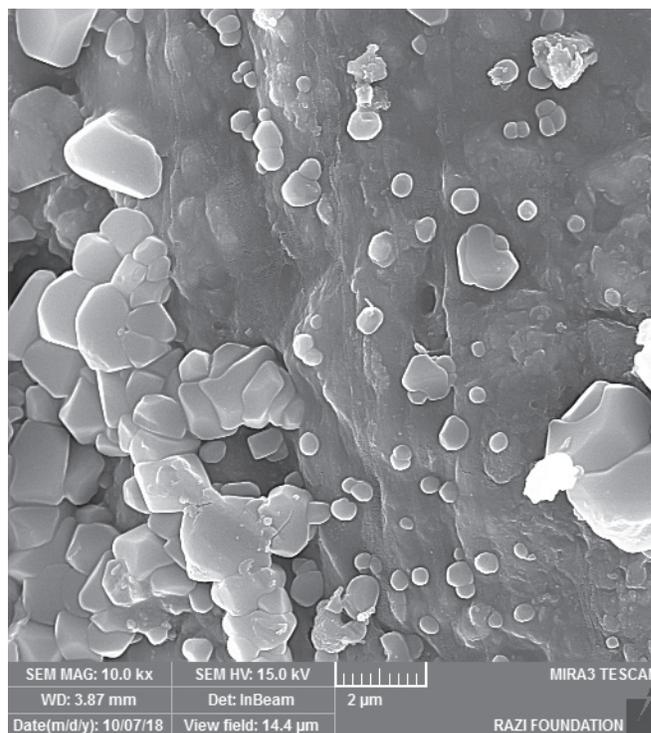


Figure 12: SEM of polymer [8]

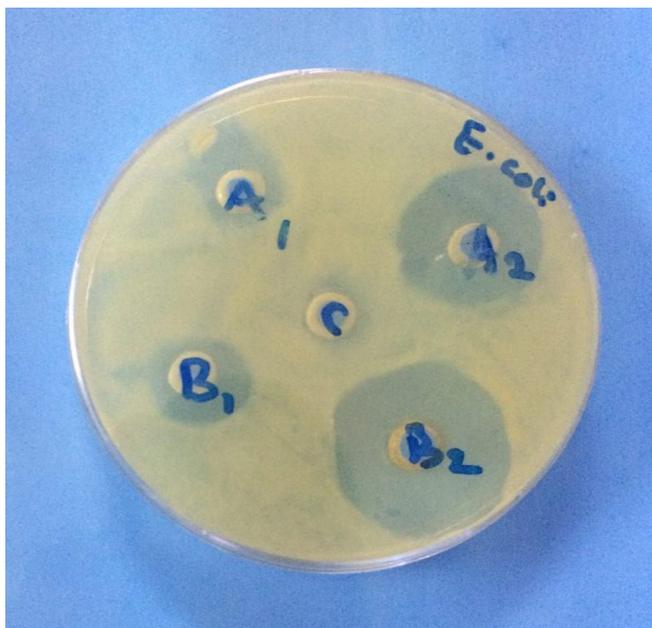
under argon and measured in temperature 0 to  $600^\circ\text{C}$ . Table 2 shows DSC and TGA for chitosan and some of prepared polymers.

### SEM Studies

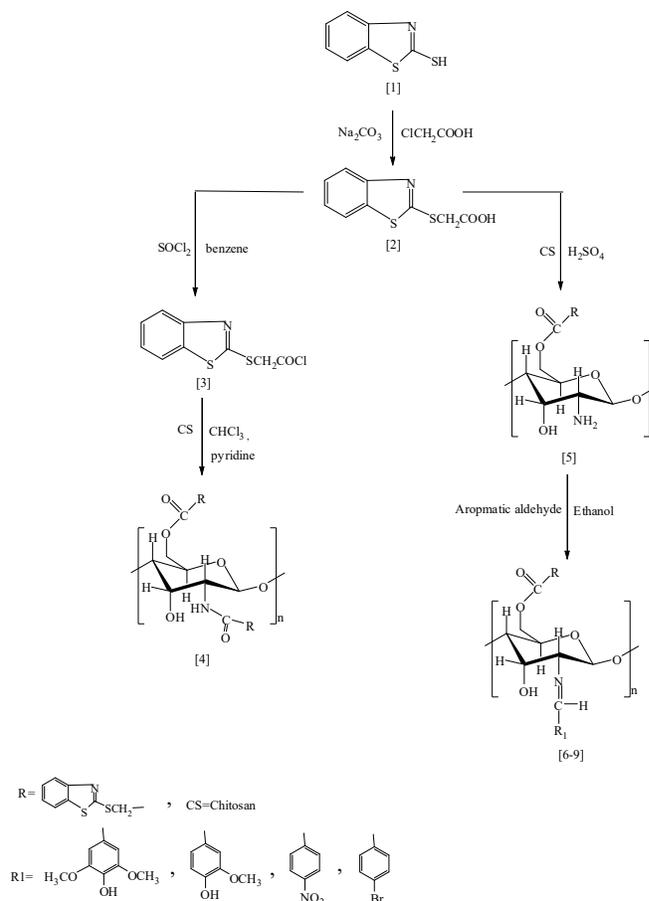
The SEM micrographs were used to study the changes in the

**Table 3:** Anti-bacterial activity of some prepared polymers

Comp.	<i>E. coli</i>	<i>Bacillus cereus</i>
Penicillin	17	23
[4]	15	16
[5]	15	16
[8]	24	20
[9]	27	22


**Figure 13:** Antibacterial activities of polymers

surface morphology for the prepared polymer. Figures 11, 12 illustrate the surface morphology of the modified chitosan. In the SEM photos, we can notice an increase in the pore average size compared to the pore size in chitosan. The changes in surface morphology are due to the new bonds in the prepared polymer.


**Scheme 1:** The synthetic route for target derivatives [1-9]

### Anti-bacteria Activity<sup>22</sup>

The antibacterial activity of some synthesized compounds has been examined (*in vitro*) against *Bacillus subtilisa* (G<sup>+</sup>) and *E. coli* (G<sup>-</sup>), based on the approach of agar diffusion.<sup>22</sup> A standard drug used to compare with the synthesized polymers is penicillin (50 µg/mL) all compounds and their antibacterial activities listed in Table 3 (Figure 13 and Scheme 1).

### CONCLUSION

The present study aimed to achieving synthesis, characterization and study biological activity of some new synthesized polymers. The results showed that all polymers had higher growth inhibition zone diameter, compound [9] showed very good inhibition against *Bacillus subtilisa* and *E. coli*.

### REFERENCES

- El-Sayed R. Synthesis, antibacterial and surface activity of 1,2,4-triazole derivatives. *Grasas Y Aceites*. 2006;57(2): 180-188.
- Vedavathi M, Somashekar B, Sreenivasa GM, Jayachandran E. Synthesis, Characterization and Anti-microbial activity of fluoro benzothiazole incorporated with 1,3,4-Thiadiazole. *J. Pharm. Sct. & Res.* 2010;2(1):53-63.
- Gurupadaya BM, Gopal M, Padmashali B, Vaidya VP. Synthesis and biological activities of fluoro benzothiazoles. *Int J Heterocyclic Chem.* 2005;15:169-172.

4. Geoffrey W, Tracey DB, Patrizia D, Angela S, Dong-Fang S, Andrew D, Westwell MF, Stevens G, Antitumour Benzothiazoles. Part 10: The Synthesis and Antitumour Activity of Benzothiazole Substituted Quinol Derivatives. *Bioorg. & Med. Chem. Lett.* 2000;10:513-515.
5. Kochichiro Y, Katsumi G, Kazuya Y, Tominori M, Goro T. Synthesis and evaluation of novel benzothiazole derivatives against human cervical cancer cell lines. *Int. J. Pharm. Sci.* 2007;69(1):46-50.
6. Ahmad MR, Ahmad AH. Synthesis and characterization of some New Derivatives from 2- Mercaptobenzothiazole. *Iraqi Journal of Science.* 2014; 55, (2A):319-333
7. Rajeeva B, Srinivasulu N, Shantakumar S. Synthesis and Antimicrobial activity of some new 2-substituted benzothiazole derivatives. *E-Journal of Chemistry.* 2009;6(3):775-779.
8. Stanton HLK, Gambari R, Chung HC, Johny COT, Filly C, Albert SCC, Synthesis and anti-cancer activity of benzothiazole containing phthalimide on human carcinoma cell lines. *Bioorg Med Chem.*, 2008; 16:3626-3631.
9. Pattan S, Suresh C, Pujar V, Reddy V, Rasal V, Koti B., Synthesis and antidiabetic activity of 2-amino[5''(4-sulphonylbenzylidene)-2,4-thiazolidinenone]-7-chloro-6-fluorobenzothiazole. *Ind J Chem.*, 2005; 44B:2404-2408.
10. Kaur H., S. Kumar, I. Singh, K. K. Saxena, A. Kumar. Synthesis, Characterization and Biological activity of various substituted benzothiazole derivatives. *Digest J. Nanomaterials and Biostructures*, 2010 ; 5(1): 67-76.
11. Al-Issa MA, Ayat AA, Fadhel SM. Synthesis and characterization of schiff base derived from chitosan and its complexes with (Co<sup>+2</sup>, Ni<sup>+2</sup> and Cu<sup>+2</sup>). *Ibn Al-Haitham Jour. for Pure & Appl. Sci.* 2016;29(2):115-129.
12. Younus MA, Shihab HA. Synthesis & Characterization of Chitosan Schiff Base Hydrogel for Controlled Drug Release. *Journal of Global Pharma Technology.* 2019;11(02 Suppl.):392-398.
13. Supriya Prasad, Thandapani Gomathi and P.N. Sudha, Synthesis and Characterization Chitosan Schiff Base (CSB) and its Polyethylene Glycol (PEG) Blend. *Novel Trends In Pharmaceutical Sciences.* 2017;7 (2): 2277 - 2782
14. Ruwaidah S. Saeed, Fadhel S. Matty and Ali H. Samir, Chemical modification of PVA with four, five and seven heterocyclic compounds and study anticancer activity. *J. Pharm. Sci. & Res.* 2019;11(3):733-740
15. Ali H. Samir, Ruwaidah S. Saeed and Fadhel S. Matty, Synthesis and Study of Modified Polyvinyl Alcohol Containing Amino Acid Moieties as Anticancer Agent. *Orient. J. Chem.* 2018; 34(1):286-294
16. Zhimei Z, Bayaer A, Hui X, Shuang Z. Structure and antimicrobial activities of benzoyl phenyl-thiosemicarbazone-chitosans. *International Journal of Biological Macromolecules.* 2012;50(4):1169-1174
17. Badawy MEI, Rabea EI. Preparation and antimicrobial activity of O-(benzoyl) chitosan derivatives against some plant pathogens, *African Journal of Microbiology Research.* 2013;7(20):2259-2268.
18. Emad Soliman, Salah M. El-Kousy, Hamed M. Abd-Elbary and Ahmed R. Abou-zeid, Low Molecular Weight Chitosan-based Schiff Bases: Synthesis, Characterization and Antibacterial Activity. *American Journal of Food Technology.* 2013;8(1): 17-30
19. Jinping C, Qifeng D, Chengsheng L, Teng W, Bing F, Jingqun Y, Yanyan X., Preparation, characterization and antibacterial activity of O-acetyl-chitosan-N-2-hydroxypropyl trimethyl ammonium chloride. *International journal of Biological Macromolecules.* 2015;80:8-15
20. Wang J. and Wang H., Preparation of soluble p-aminobenzoyl chitosan ester by Schiff's base and antibacterial activity of the derivatives *International journal of Biological Macromolecules.* 2011;48:523-529.
21. Saral Sarojini K, Indumathi MP and Rajarajeswari GR, Mahua oil-based polyurethane/chitosan/nano ZnO composite films for biodegradable food packaging applications. *Int J Biol Macromol.* 2019;124:163-174.
22. Barry AL. *The Anti microbial Susceptibility Test: principle and Practices*, (Len and Febiger, Philadelphia, USA). 1977; 180, *Biol Abstr.* 64, 25183