

# Synthesis of new unique substances and antibacterial action of Pyrazolo derivatives

Vipin Dubey<sup>1\*</sup>, Daniel Prasad<sup>2</sup>, Rakesh Kumar Gautam<sup>3</sup>, Sachchidanand Tiwari<sup>4</sup>

<sup>1</sup>Department of Chemistry, Govt. Adarsh College, Dindori (M.P.)

<sup>2</sup>Department of Chemistry, Govt. PMCOE Chandrvijay College, Dindori (M.P.)

<sup>3</sup>Department of Chemistry, Govt. Degree College, Baidhan (M.P.)

<sup>4</sup>Department of Chemistry, PMCOE S.G.S Autonomous PG College, Sidhi (M.P.)

## Abstract

Due to their numerous derivatives with various structural alterations, pyrazolo are recognized as adaptable heterocyclic compounds with a variety of biological characteristics. As a result, various novel Pyrazolo quinazoline heterocycles were created from chalcones, and IR, <sup>1</sup>H NMR, <sup>13</sup>C NMR, and mass spectrum data were used to corroborate their structures. The antibacterial activity of each of these synthetic compounds was evaluated in vitro against both gram-positive and gram-negative bacterial strains as well as fungus strains in DMSO. KC-10 had the strongest antibacterial activity among the tested compounds. None of the compound shows antifungal activity. This could be because, in contrast to bacterial cells, fungal cells are more complicated organisms. Additionally, the synthetic chemicals S-CH<sub>3</sub> group may make them inert against the fungus strains under study.

**Keywords:** Antibacterial activity, antifungal activity, pyrazolo derivatives, dimethylsulfoxide and agar-well diffusion method

## Introduction

Pyrazolo derivatives have long piqued interest in organic chemistry because of their numerous pharmacological properties [1], including antibacterial [2-4], anticancer [5-7], anti-inflammatory [8,9], antimicrobial [10-12] and antifungal [13-15] properties. Additionally, the fragment of pyrazolo quinazolines is found in a number of pharmacological compounds as well as several naturally occurring substances that have biological activity [16]. A review of the literature reveals that quinazoline derivatives are crucial for the development of new drugs [17,18]. Owing to these biological characteristics of pyrazolo derivatives, some new fused pyrazolo derivatives are synthesized in this work from (E)-2-benzylidene-3,4-dihydro naphthalen-1(2H)-one, and their characterisation was carried out using mass spectrum data, IR and NMR.

These synthetic compounds were tested in vitro for their antimicrobial activity against a variety of bacterial and fungus species, both gram-positive and gram-negative, in dimethylsulfoxide (DMSO). *Staphylococcus aureus* ATCC 29737 (SA), *Corynebacterium rubrum* ATCC 14898 (CR), *Listeria monocytogenes* ATCC 19112 (LM), *Bacillus cereus* ATCC 11778 (BC), and *Pseudomonas aeruginosa* ATCC 27853 (PA), *Escherichia coli* ATCC 2931 (EC), *Klebsiella pneumoniae* ATCC 2719 (KP), and *Salmonella typhimurium* ATCC 23564 (ST).

## Experimental

### Synthesis

#### *Synthesis of (E)-2-benzylidene-3,4-dihydro naphthalene-1(2H)-one*

In the presence of a catalytic quantity of potassium hydroxide (4%), equimolar solutions (not equal volume) of  $\alpha$ -tetralone and several substituted benzaldehydes in methanol were agitated for 1.5 hours. Different substituted benzaldehydes, including 4-Chloro benzaldehyde, 4-methoxy benzaldehyde, 4-Floro

benzaldehyde, 4-Bromo benzaldehyde, 3,4-dimethoxy benzaldehyde, 4-cyano benzaldehyde, 3-Chloro benzaldehyde, 3-methoxy benzaldehyde, 3-Bromo benzaldehyde, and 4-methyl benzaldehyde. Analytical thin layer chromatography (TLC) (performed on aluminum coated plates Gel 60F254 (E. Merck)) with a 7:3 hexane:ethyl acetate mobile phase was used to confirm the reaction's completion. After the reaction was finished, the solid was filtered, cleaned with water, and vacuum-dried to produce a crude product. The reaction mass was then cooled. By adding an appropriate solvent (diethyl ether) and scraping and swirling the resulting crude product, colored, nonpolar contaminants were eliminated. After allowing the product to settle, the aforementioned solution was decanted. To eliminate contaminants, the process was carried out three to four times (trituration). Gas chromatography revealed that the final product's purity was (99.5%) 0.995 in mole fraction.

### *Synthesis of (5-amino-3-(methylthio)-1H-pyrazole-4 carbonitrile*

For thirty minutes, a combination of dry  $K_2CO_3$  (0.012 mmol) and malanonitrile (0.01 mmol) was agitated in dry DMF at room temperature. After adding 0.02 mole of carbon disulfide dropwise to this reaction mixture, the mixture was agitated for 2.5 hours at room temperature. After that, the solution was cooled from 0-5 °C. After adding 0.02 mole dimethyl sulfate to this cooled solution, it was agitated once more for five to six hours at room temperature. Thin layer chromatography was used to track the reaction's development. It was poured into crushed ice to produce a solid product once the reaction was finished. To obtain the crude product, the resultant solid was filtered, cleaned with cold water, and vacuum-dried. For 30 minutes, an equimolar solution of this crude product and hydrazine hydrate in isopropyl alcohol (IPA) was refluxed. After that, the reaction mixture was added to crushed ice. To create the final product, the solid was filtered, cleaned with water, and vacuum-dried. The resultant product was purified by trituration with hexane to eliminate non-polar impurities, and it was utilized in the subsequent stage without additional purification.

### *Synthesis of pyrazolo[5,1-b] quinazolines*

For 4-5 hours, an equimolar mixture of (E)-2-benzylidene-3,4 dihydronaphthalen-1(2H)-one and (5-amino-3 (methylthio)-1H-pyrazole-4-carbonitrile) was refluxed in n-butanol (Fig. 1). Using (6:4-Hexane:Ethyl acetate) as a mobile phase, Thin Layer Chromatography verified the reaction's completion. After allowing the reaction mixture to cool, the resultant solid was filtered and cleaned of impurities using diethyl ether. To rid the product of contaminants, the process was carried out three or four times.

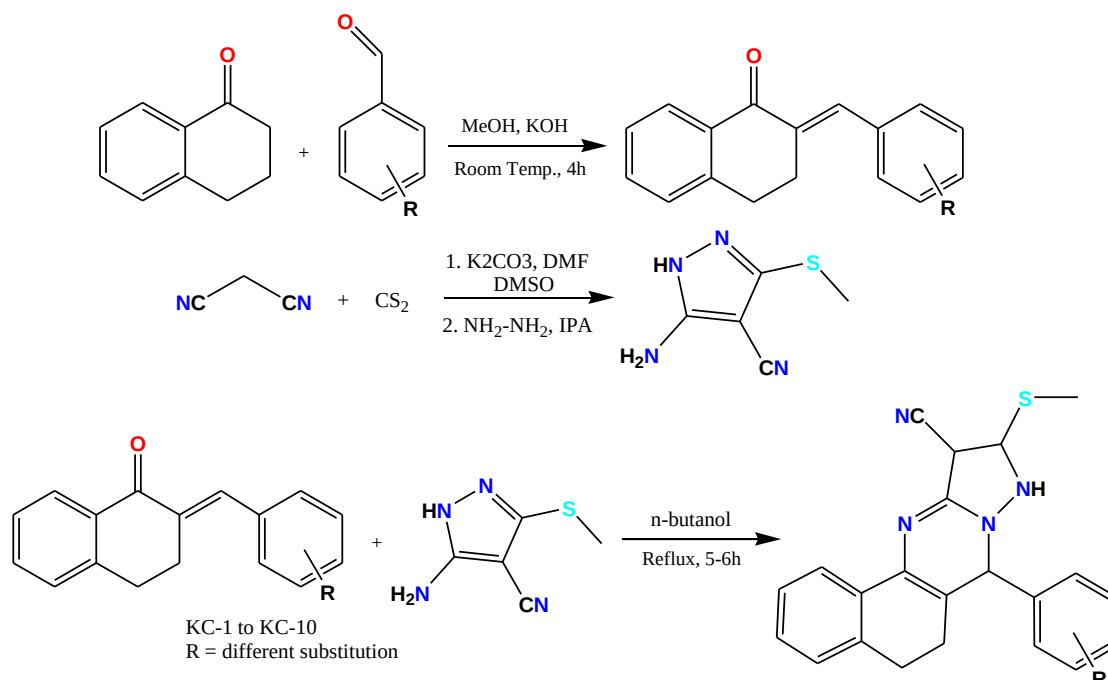


**Fig. 1** Reaction scheme, where R is KC-1 = 4-Cl benzaldehyde, KC-2 = 4-OCH<sub>3</sub> benzaldehyde, KC-3 = 4-F benzaldehyde, KC-4 = 4-Br benzaldehyde, KC-5 = 3,4-diOCH<sub>3</sub> benzaldehyde, KC-6 = 4-CN benzaldehyde, KC-7 = 3-Cl benzaldehyde, KC-8 = 3-OCH<sub>3</sub> benzaldehyde, KC-9 = 3-Br benzaldehyde, KC-10 = 4-CH<sub>3</sub> benzaldehyde.

### Spectroscopic study

IR, mass, <sup>1</sup>H NMR and <sup>13</sup>C NMR spectrum data were used to characterize each of these substances. The Shimadzu FT-IR-8400 instrument was used to record the IR spectra using the KBr pellet method. The direct intake probe method was used to record the mass spectra using the Shimadzu GC-MS-QP-2010 model. Compound KC-2 <sup>1</sup>H NMR was measured in DMSO solution using a Bruker Ac 400 MHz spectrometer and is displayed in Fig. 2. Similarly, compound KC-2 <sup>13</sup>C NMR was measured in DMSO solution using a Bruker Ac 400 MHz spectrometer using TMS as a standard, and it is displayed in Fig. 3. Before being used, these synthetic chemicals were recrystallized. Prior to usage, the solvent DMSO was also purified using conventional techniques [19].

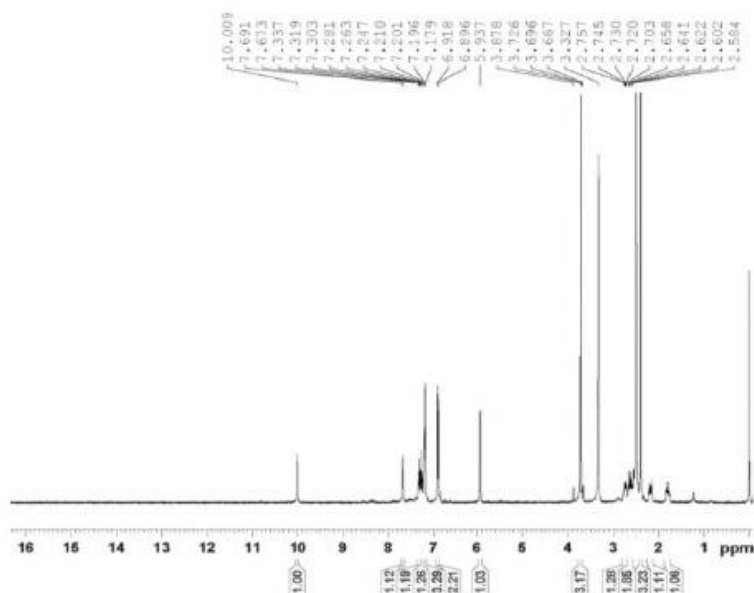


**Fig. 2** <sup>1</sup>H NMR spectrum of KC-02

**Fig. 3**  $^{13}\text{C}$  NMR spectrum of KC-02

### Microorganisms tested

The National Chemical Laboratory (NCL) in Pune, India, provided the microorganisms under study. The temperature of the microorganisms was kept at 4°C. *Staphylococcus aureus* was one of the Gram-positive microorganisms under investigation. The gram-negative bacteria were *Pseudomonas aeruginosa* ATCC27853 (PA), *Escherichia coli* NCIM2931 (EC), *Klebsiella pneumoniae* NCIM2719 (KP) and *Salmonella typhimurium* ATCC23564 (ST); the fungi were *Candida albicans* ATCC2091 (CA), *Cryptococcus neoformans* NCIM3542 (CN), *Candida glabrata* NCIM3448 (CG) and *Candida cereus* ATCC11778 (BC). Before being used, the organisms were subcultured and kept at 4°C on nutritional agar for bacteria and MGY medium (Hi Media, India) for fungi. A number of diseases and food deterioration are caused by the clinically significant microorganisms under study. The gram-positive and gram-negative bacteria that were chosen are common pathogenic bacteria, and their study is clinically significant. They cause a variety of infections, food-borne illnesses, spoilages and skin diseases and it is crucial to combat them with some effective therapeutic medicines. To investigate a wide range of synthetic compound activity, both gram-positive and gram-negative were chosen.

### Preparation of compounds solutions

DMSO was used to check for antibacterial activity in each chemical. For every chemical, a 20 mg/ml concentration solution was made.

### Diffusion method in an agar well

The Agar well diffusion method was used to investigate the various Pyrazoloquinazolines' in vitro antibacterial efficacy against harmful microbiological strains. Mueller Hinton No. 2/Sabouraud dextrose agar (Hi-media) was utilized for the antibacterial and antifungal susceptibility tests, respectively [20,21]. After melting and cooling the Mueller Hinton agar and Sabouraud dextrose agar to 48–50°C, a standardized inoculum ( $1.5 \times 10^8$  CFU/ml, 0.5 McFarland) was added aseptically to the molten agar and placed into sterile Petri dishes; wells (8.5 mm) were formed in the seeded agar plates. The well was filled with 100 µl of the test chemical. For bacteria and fungi, the plates were incubated for 24 and 48 hours, respectively, at 37°C and 28°C. The negative control was DMSO. The microbial growth was assessed by measuring the diameter of the zone of inhibition and the mean values are used.

## Results and discussion

Ten compounds in all (KC-1 to KC-10) were created. Table 1 lists all of the produced compounds physical constants. Their chemical structure was verified by the IR, NMR and mass spectral data.

**Table 1** Physical constants of compounds

| Compound | R                       | Molecular Formula                                               | Mol. weight | Yield (%) |
|----------|-------------------------|-----------------------------------------------------------------|-------------|-----------|
| KC-1     | -4-Cl                   | C <sub>22</sub> H <sub>17</sub> ClN <sub>4</sub> S              | 404.09      | 78        |
| KC-2     | -4-OCH <sub>3</sub>     | C <sub>23</sub> H <sub>20</sub> N <sub>4</sub> OS               | 400.14      | 79        |
| KC-3     | -4-F                    | C <sub>22</sub> H <sub>17</sub> FN <sub>4</sub> S               | 388.46      | 78        |
| KC-4     | -4-Br                   | C <sub>22</sub> H <sub>17</sub> BrN <sub>4</sub> S              | 449.37      | 76        |
| KC-5     | -3,4-diOCH <sub>3</sub> | C <sub>24</sub> H <sub>22</sub> N <sub>4</sub> O <sub>2</sub> S | 430.52      | 71        |
| KC-6     | -4-CN                   | C <sub>23</sub> H <sub>17</sub> N <sub>5</sub> S                | 395.48      | 60        |
| KC-7     | -3-Cl                   | C <sub>22</sub> H <sub>17</sub> ClN <sub>4</sub> S              | 404.92      | 77        |
| KC-8     | -3-OCH <sub>3</sub>     | C <sub>23</sub> H <sub>20</sub> N <sub>4</sub> OS               | 400.14      | 76        |
| KC-9     | -3-Br                   | C <sub>22</sub> H <sub>17</sub> BrN <sub>4</sub> S              | 449.37      | 71        |
| KC-10    | -4-CH <sub>3</sub>      | C <sub>23</sub> H <sub>20</sub> N <sub>4</sub> S                | 384.50      | 64        |

## Spectral Data

### KC-1: 7-(4-chlorophenyl)-10-(methylthio)-5,6,7,9 tetra hydrobenzo[h]pyrazolo[5,1-b] quinazoline-11 carbonitrile

IR(cm<sup>-1</sup>, KBr): 3475.85 (-NH (sec.) str.), 3049.56 (Ar H str.), 2924.18(-CH<sub>2</sub> sym. str.), 2227.86 (-CN str.), 1664.62(C=C str. α, β unsaturated 6-member ring), 1604.83(-NH bending vib. Secondary amine), 1381.08 (-CH bending.), 1315.50 (C-N (sec) bending.), 1242 1010 (C-H in plane bending, phenyl ring), 767.69 (C-H str. 5-adjacent c atoms), 767.69(C-Cl str.), <sup>1</sup>H NMR (DMSO-d<sub>6</sub>) δ(ppm): 2.40 (3H, singlet, CH<sub>3</sub>), 1.79-2.75 (4H, multiplet, C-H), 6.07 (1H, singlet, C-H), 7.22-7.70 (8H, multiplet C-H), 10.14 (1H, singlet, -NH). Elemental analysis: %C = 65.29 (65.34), %H = 4.21 (4.20), %N = 13.88 (13.86), %S = 7.92 (7.92). Mass: (m/z) =404.09.

### KC-2: 7-(4-methoxyphenyl)-10-(methylthio)-5,6,7,9 tetrahydrobenzo[h]pyrazolo[5,1-b] quinazoline-11 carbonitrile

IR(cm<sup>-1</sup>, KBr): 3077.11 (Ar-H asym. str.), 1658.38 (C=N str), 2945.40-2843.17 (CH<sub>2</sub> str. of cyclohexanone ring), 1597.11(C=C str.), 1090.95 (C-O-C sym. str. 954.80 (ring str. in cyclohexanone), 1210.45 (C-O-C asym. Str.) 2814.11(C-H str. Alkane) <sup>1</sup>H NMR (DMSO-d<sub>6</sub>) δ(ppm): 2.44 (3H, singlet,-CH<sub>3</sub>), 3.70 (3H, singlet-OCH<sub>3</sub>), 1.80 2.76 (4H, multiplet, C-H), 5.94 (1H, singlet, C-H), 6.90 7.70 (8H, multiplet C-H), 10.01 (1H, singlet, N-H). Elemental analysis: %C = 68.92 (69.00), %H = 5.06 (5.00), %N = 13.97 (14.00), %S = 8.03 (8.00), %O = 4.02 (4.00). Mass: (m/z) =400.50.

### KC-3: 7-(4-fluorophenyl)-10-(methylthio)-5,6,7,9 tetrahydrobenzo[h]pyrazolo[5,1-b] quinazoline-11 carbonitrile

IR(cm<sup>-1</sup>, KBr): 3479.70 (-NH (sec.) str.), 3037.99 (Ar H str.), 2918.40(-CH<sub>2</sub> sym. str.), 2227.86 (-CN str.), 1666.55 (C=C str. α, β unsaturated 6-member ring), 1599.04 (-NH bending vib. Secondary amine), 1381.08 (-CH bending.), 1319.08 (C-N (sec) bending.), 1242 1010(C-H in plane bending, phenyl ring), 1093.67 (C-F str.), 725.26 (C-H str. 5-adjacent c atoms), <sup>1</sup>H NMR (DMSO-d<sub>6</sub>) δ(ppm): 2.42 (3H, singlet, CH<sub>3</sub>), 1.80-2.77 (4H, multiplet, C-H), 5.99 (1H, singlet, C-H), 7.15-7.69 (8H, multiplet C-H), 10.25 (1H, singlet, -NH). Elemental analysis: %C = 68.07 (68.04), %H = 4.42 (4.38), %N = 14.41 (14.43), %S = 8.25 (8.24). Mass: (m/z) =388.46.

**KC-4: 7-(4-bromophenyl)-10-(methylthio)-5,6,7,9 tetrahydrobenzo[h]pyrazolo[5,1-b]quinazoline-11 carbonitrile**

IR( $\text{cm}^{-1}$ , KBr): 3257.88 (-NH (sec.) str.), 3047.63 (Ar H str.), 2929.97 (-CH<sub>2</sub> sym. str.), 2227.86 (-CN str.), 1653.05 (C=C str.  $\alpha$ ,  $\beta$  unsaturated 6-member ring), 1604.83 (-NH bending vib. Secondary amine), 1383.01 (-CH bending.), 1315.50 (C-N (sec) bending.), 1242 1010(C-H in plane bending, phenyl ring), 723.33 (C-H str. 5-adjacent c atoms), 582.52 (C-Br str.), <sup>1</sup>H NMR (DMSO-d<sub>6</sub>)  $\delta$ (ppm): 2.42 (3H, singlet, CH<sub>3</sub>), 1.80-2.79 (4H, multiplet, C-H), 5.99 (1H, singlet, C-H), 7.21-7.82 (8H, multiplet C-H), 10.25 (1H, singlet, -NH). Elemental analysis: %C = 58.87 (58.93), %H = 3.79 (3.80), %N = 12.45 (12.50), %S = 7.07 (7.14). Mass: (m/z) = 448.37.

**KC-5: 7-(3,4-dimethoxyphenyl)-10-(methylthio)-5,6, 7,9-tetrahydrobenzo[h]pyrazolo [5,1-b]quinazoline 11-carbonitrile**

IR( $\text{cm}^{-1}$ , KBr): 3236.66 (-NH (sec.) str.), 3007.12 (Ar H str.), 2929.97 (-CH<sub>2</sub> sym. str.), 2224.40 (-CN str.), 1666.55(C=C str.  $\alpha$ ,  $\beta$  unsaturated 6-member ring), 1604.83(-NH bending vib. Secondary amine), 1383.09 (-CH bending.), 1334.78 (C-N (sec) bending.), 1242 1010(C-H in plane bending, phenyl ring), 702.11 (C-H str. 5-adjacent c atoms), 731.05(C-H in plane bending), <sup>1</sup>H NMR (DMSO-d<sub>6</sub>)  $\delta$ (ppm): 2.45 (3H, singlet,-CH<sub>3</sub>), 3.71 (3H, singlet-OCH<sub>3</sub>), 4.02 (3H, singlet -OCH<sub>3</sub>), 1.80-2.76 (4H, multiplet, C-H), 5.98 (1H, singlet, C-H), 7.00-7.76 (8H, multiplet C-H), 10.09 (1H, singlet, -NH). Elemental analysis: %C = 66.91 (66.97), %H = 5.21 (5.12), %N = 13.08 (13.02), %S = 7.49 (7.44), %O = 7.31 (7.44). Mass: (m/z) = 430.52.

**KC-6: 7-(4-cyanophenyl)-10-(methylthio)-5,6,7,9 tetrahydrobenzo[h]pyrazolo[5,1-b] quinazoline-11 carbonitrile**

IR( $\text{cm}^{-1}$ , KBr): 3310.18 (-NH (sec.) str.), 2990.17 (Ar H str.), 2896.57 (-CH<sub>2</sub> sym. str.), 2227.12 (-CN str.), 1666.55 (C=C str.  $\alpha$ ,  $\beta$  unsaturated 6-member ring), 1597.64 (-NH bending vib. Secondary amine), 1381.13 (-CH bending.), 1289.74 (C-N (sec) bending.), 1242 1010(C-H in plane bending, phenyl ring), 712.87 (C-H str. 5-adjacent c atoms), 729.67(C-H in plane bending), <sup>1</sup>H NMR (DMSO-d<sub>6</sub>)  $\delta$ (ppm): 2.43 (3H, singlet,-CH<sub>3</sub>), 1.71-2.72 (4H, multiplet, C-H), 5.60 (1H, singlet, C-H), 7.22-7.95 (8H, multiplet C-H), 10.14 (1H, singlet, -NH). Elemental analysis: %C = 69.81 (69.87), %H = 4.39 (4.30), %N = 17.68 (17.72), %S = 8.12 (8.10). Mass: (m/z) = 395.48.

**KC-7: 7-(3-chlorophenyl)-10-(methylthio)-5,6,7,9 tetrahydrobenzo[h]pyrazolo[5,1-b] quinazoline-11 carbonitrile**

IR( $\text{cm}^{-1}$ , KBr): 3387.87 (-NH (sec.) str.), 2967.46 (Ar H str.), 2894.57 (-CH<sub>2</sub> sym. str.), 2224.40 (-CN str.), 1667.99(C=C str.  $\alpha$ ,  $\beta$  unsaturated 6-member ring), 1590.12(-NH bending vib. Secondary amine), 1357.23 (-CH bending.), 1309.47 (C-N (sec) bending.), 1242 1010(C-H in plane bending, phenyl ring), 699.24 (C-H str. 5-adjacent c atoms), 724.51(C-H in plane bending), 767.69(C-Cl str.), <sup>1</sup>H NMR (DMSO-d<sub>6</sub>)  $\delta$ (ppm): 2.40 (3H, singlet,-CH<sub>3</sub>), 1.79-2.75 (4H, multiplet, C-H), 6.07 (1H, singlet, C-H), 7.22-7.70 (8H, multiplet C-H), 10.14 (1H, singlet, -NH). Elemental analysis: %C = 65.22 (65.34), %H = 4.21 (4.21), %N = 13.78 (13.86), %S = 8.00 (7.92). Mass: (m/z) = 404.92.

**KC-8: 7-(3-methoxyphenyl)-10-(methylthio)-5,6,7,9 tetrahydrobenzo[h]pyrazolo[5,1-b] quinazoline-11 carbonitrile**

IR( $\text{cm}^{-1}$ , KBr): 3337.71 (-NH (sec.) str.), 3015.87 (Ar H str.), 2897.12 (-CH<sub>2</sub> sym. str.), 2227.97 (-CN str.), 1666.55 (C=C str.  $\alpha$ ,  $\beta$  unsaturated 6-member ring), 1615.92 (-NH bending vib. Secondary amine), 1397.54 (-CH bending.), 1337.56 (C-N (sec) bending.), 1242 1010(C-H in plane bending, phenyl ring), 709.57 (C-H str. 5-adjacent c atoms), 724.69(C-H in plane bending), <sup>1</sup>H NMR (DMSO-d<sub>6</sub>)  $\delta$ (ppm): 2.45 (3H, singlet,-CH<sub>3</sub>), 3.70 (3H, singlet-OCH<sub>3</sub>), 1.77-2.81 (4H, multiplet, C H), 6.02 (1H, singlet, C-H), 7.09-

7.83 (8H, multiplet C H), 10.11 (1H, singlet, -NH). Elemental analysis: %C = 68.92 (69.00), %H = 5.01 (5.00), %N = 13.93 (14.00), %S = 8.12 (8.00), %O = 4.02 (4.00). Mass: (m/z) = 400.14.

### KC-9: 7-(3-bromophenyl)-10-(methylthio)-5,6,7,9 tetrahydrobenzo[h]pyrazolo[5,1-b]

#### quinazoline-11 carbonitrile

IR( $\text{cm}^{-1}$ , KBr): 3337.69 (-NH (sec.) str.), 2967.43 (Ar H str.), 2937.65 (-CH<sub>2</sub> sym. str.), 2229.70 (-CN str.), 1666.55(C=C str.  $\alpha$ ,  $\beta$  unsaturated 6-member ring), 1591.25(-NH bending vib. Secondary amine), 1403.17 (-CH bending.), 1312.36 (C-N (sec) bending.), 1242 1010(C-H in plane bending, phenyl ring), 699.14 (C-H str. 5-adjacent c atoms), 737.45(C-H in plane bending), <sup>1</sup>H NMR (DMSO-d<sub>6</sub>)  $\delta$ (ppm): 2.40 (3H, singlet,-CH<sub>3</sub>), 1.70-2.70 (4H, multiplet, C-H), 5.95 (1H, singlet, C-H), 6.99-7.72 (8H, multiplet C-H), 10.12 (1H, singlet, -NH). Elemental analysis: %C = 58.86 (58.93), %H = 3.87 (3.80), %N = 12.45 912.50), %S = 7.11 (7.14). Mass: (m/z) = 448.37.

### KC-10: 10-(methylthio)-7-(p-tolyl)-5,6,7,9 tetrahydro benzo[h]pyrazolo[5,1-b] quinazoline-11-carbonitrile

IR( $\text{cm}^{-1}$ , KBr): 3314.25 (-NH (sec.) str.), 2987.34 (Ar H str.), 2945.67 (-CH<sub>2</sub> sym. str.), 227.64 (-CN str.), 1666.55(C=C str.  $\alpha$ ,  $\beta$  unsaturated 6-member ring), 1593.14(-NH bending vib. Secondary amine), 1374.28 (-CH bending.), 1340.05 (C-N (sec) bending.), 1242 1010(C-H in plane bending, phenyl ring), 698.47 (C-H str. 5-adjacent c atoms), 742.36(C-H in plane bending), <sup>1</sup>H NMR (DMSO-d<sub>6</sub>)  $\delta$ (ppm): 2.43 (3H, singlet,-CH<sub>3</sub>), 3.83 (3H, singlet-CH<sub>3</sub>), 1.80-2.80 (4H, multiplet, C-H), 6.02 (1H, singlet, C-H), 7.11-7.81 (8H, multiplet C-H), 10.07 (1H, singlet, -NH). Elemental analysis: %C = 71.89 (71.88), %H = 5.28 (5.21), %N = 14.52 (14.58), %S = 8.31 (8.33). Mass: (m/z) = 384.50.

### Antimicrobial Activity

The antibacterial efficacy of each synthetic chemical in DMSO against gram-positive bacteria is displayed in Fig. 4. Figure shows that KC-10 inhibited all tested gram-positive bacteria, whereas KC-3 and KC-7 inhibited 75% of them. The highest zone of inhibition against *Bacillus cereus* (BC) was demonstrated by KC-3. KC-4 demonstrated the greatest suppression of *Staphylococcus aureus* (SA), followed by KC-3 and KC-10. KC-2, KC-5 and KC-8 were unable to inhibit any of the gram-positive bacteria, whereas KC-6 and KC-3 shown the greatest inhibition against strains of *Listeria monocytogenes* (LM) and *Corynebacterium rubrum* (CR), respectively. Additionally, all of these Gram-positive bacteria may be inhibited by KC-10. Pyrazolo quinazoline, which has various side chains, is the core component in each of the ten compounds. According to reports, the antibacterial activity differs with the same core moiety and different side chains [22,23].

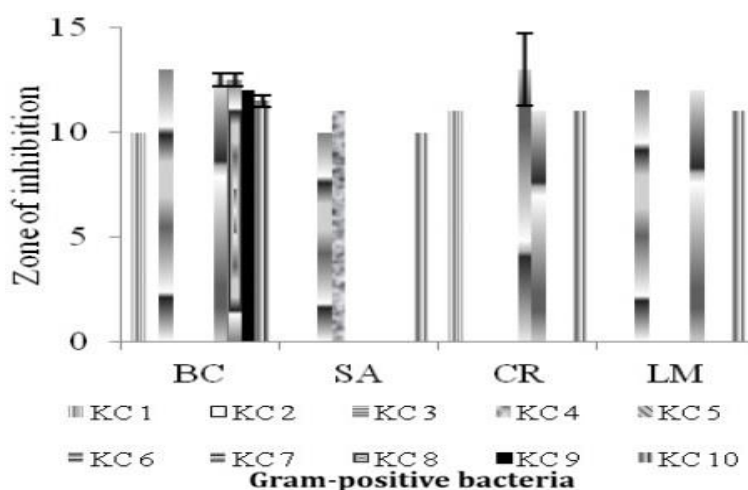


Fig. 4 Antimicrobial activity against Gram positive bacteria

Therefore, halogen and methyl-containing groups work best to suppress these particular gram-positive bacteria. According to Rai *et al.*, [24], the presence of methyl and halogen groups causes greatest inhibition. Other researchers have also reported similar findings [25,26]. Additionally, these investigators have noted that the presence of halogen groups might sometimes augment the inhibition. According to Ibrahim *et al.*, [27], there is good biological activity when the  $-CH_3$  group is present as a side chain. The most vulnerable gram-positive bacteria were *Bacillus cereus* (BC) while the most resistant were *Staphylococcus aureus* (SA).

Compounds antibacterial efficacy against gram-negative bacterial strains is depicted in Fig. 5. *Escherichia coli* (EC) was the most resistant of the four gram-negative bacteria that were chosen; it was not inhibited by any of the substances that were tested. KC-9 showed the greatest inhibition of the other three microorganisms. KC-4 exhibits the least amount of inhibition against *Salmonella typhimurium* (ST) and *Klebsiella pneumoniae* (KP). Additionally, whereas KC-2 might inhibit *Pseudomonas aeruginosa* (PA), KC-5 demonstrated inhibition against *Klebsiella pneumoniae* (KP) and *Salmonella typhimurium* (ST).

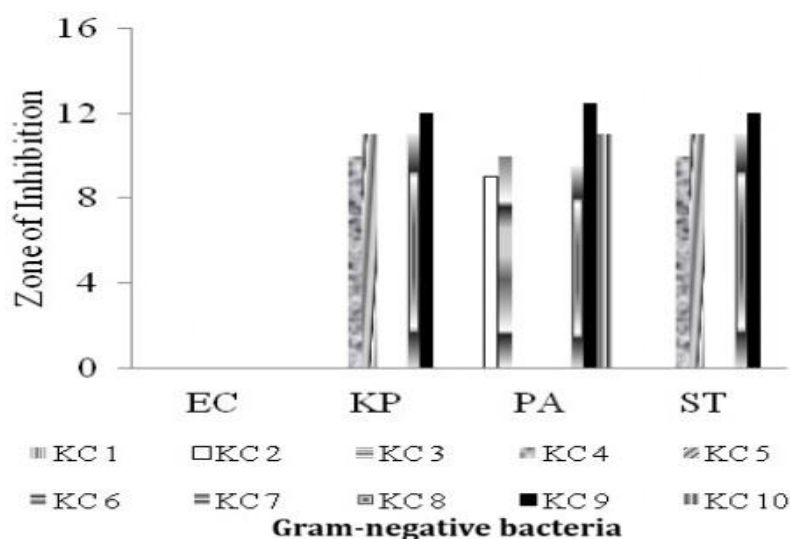


**Fig. 5** Antimicrobial activity against Gram negative bacteria

Once more, none of these gram-negative bacterial strains were inhibited by KC-8. While methoxy groups are present in all three compounds, KC-2, KC-5 and KC-8, their locations differ. It is in para position in KC-2 and meta position in KC-8. Two methoxy groups are present in KC-5 at the meta and para locations. Therefore, the inhibition is also influenced by group positions. A comparison of KC-9 and KC-4 inhibition provides additional evidence for this. Although the bromine group is present in both of these molecules, its locations differ. It is in the meta position in KC-9 and the para position in KC-4. Therefore, the group's position plays a significant role in the suppression of microorganisms.

None of the compounds exhibited antifungal efficacy against any of the chosen fungal strains. The  $-S-CH_3$  group is found in the moiety of the compounds under study. This could be the cause of these substances' inability to exhibit fungal strains. The majority of Pyrazolo quinazoline derivatives with amide or other groups (but not the  $-S-CH_3$  group) demonstrated notable antifungal inhibition, according to a review of the literature [28-30]. The effective prediction of a lead molecule and drug-like qualities at the beginning of drug design will pay dividends later in drug development, which is why screening different organic compounds and identifying the active agents is crucial.

## Conclusion

The degree of activity against gram-positive and gram-negative bacteria differed across the ten compounds. In general, the chosen gram-positive and gram-negative bacteria are more effectively inhibited by chemicals with methyl and halogen groups. *B. Cereus* and *P. aeruginosa* are the most sensitive and *S.*

*aureus* and *E. coli* are the most resistant bacteria. None of the ten compounds were shown to be effective against the chosen fungal strains.

## References

- [1] Dangi, R., N. Hussain, A. Joshi, G. Pemawat and G. Talesara, Ind. J. Chem., 50(B), 1165-1172 (2011).
- [2] Bedi, P.M.S., V. Kumar and M.P. Mahajan, Bioorg. Med. Chem. Lett., 14, 5211-5213 (2004).
- [3] Khalil, A.M., M.A. Berghot and M.A. Gouda, Eur. J. Med. Chem., 44, 4434-4440 (2009).
- [4] Yassin, F.A., Chem. Het. Compd., 45, 35-41 (2009).
- [5] Ali, M.A., M. Shaharyar and A.A. Siddiqui, Eur. J. Med. Chem., 42, 268-275 (2007).
- [6] Joseph, A., A. Pai, K.K. Srinivasan, T. Kedar, A.T. Thomas, E. M. Jessy and R.K. Singala, Orbital Elect. J. Chem., 2, 158-167 (2010).
- [7] Zhang, Y., Y. Huang, H. Xiang, P. Wang, D. Hu, W. Xue, B. Song and S. Yang, Eur. J. Med. Chem., 78, 23-34 (2014).
- [8] Chandrika, P.M., A.R. Rao, B. Narsaiah and M.B. Raju, Int. J. Chem. Sci., 6, 1119-1146 (2008).
- [9] Kumar, P., N. Chandak, P. Kaushik, C. Sharma, D. Kaushik, K.R. Aneja and P.K. Sharma, Med. Chem. Res., 21, 3396-3405 (2012).
- [10] Patel, N.B. and G.G. Bharat, J. Saudi Chem. Soc., 14, 157-164 (2010).
- [11] Patil, S.S., S.D. Jadhav and M.B. Deshmukh, J. Chem. Sci., 124, 1043-1048 (2012).
- [12] Trivedi, P.B., N.K. Undavia, A.M. Dave, K.N. Bhatt and N.C. Desai, Ind. J. Chem., 32, 497-500 (1993).
- [13] Pandey, V.K., L.P. Pathak and S.K. Mishra, Indian. J. Chem. B, 44, 1940-1944 (2005).
- [14] Pattanaik, J. M., M. Pattanaik and D. Bhatta, Ind. J. Pharm. Sci., 53, 229-232 (1991).
- [15] Shivananda, M.K. and B.H. Shivarama, J. Chem. Pharm. Res., 3, 83-86 (2011).
- [16] Kumari, K., D.S. Raghuvanshi, V. Jouikov and K.N. Singh, Tetrahedron Lett., 53, 1130-1133 (2012).
- [17] Eweas A.F., A.O.H. El-Nezhawy, A.R. Baiuomy and M.M. Awad, Med. Chem. Res., 22, 1011-1020 (2013).
- [18] Helali, A.Y.H., M.T.M. Sarg, M.M.S. Koraa and M.S.F. El-zoghbi, Open J. Med. Chem., 4, 12-37 (2014).
- [19] Riddick, J.A., W.B. Bunger and T. Sakano, Organic solvents-physical properties and methods of purification, Fourth Edition., Techniques of Chemistry II, John Wiley, New York (1986).
- [20] Perez, C., M. Paul and P. Bazerque, Acta Biol. Med. Exp., 15, 113-115 (1990).

- [21] Parekh, J., P. Inamdhar, R. Nair, S. Baluja and S. Chanda, J. Serb. Chem. Soc., 70, 1155-1158 (2005).
- [22] Baluja, S., N. Kacchadia and S. Chanda, Pharm. Chem. J., 46, 117-121 (2012).
- [23] Vaghasiya, Y., R. Nair, M. Soni, S. Baluja and S. Chanda, J. Serb. Chem. Soc., 69, 991-998 (2004).
- [24] Rai, N.S., B. Kalluraya, B. Lingappa, S. Shenoy and V.G. Puranic, Eur. J. Med. Chem., 43, 1715-1720 (2008).
- [25] Jantova, S., G. Greif, K. Spirkova, S. Stankovsky and M. Oravcova, Folia Microbiol., 45, 133-137 (2000).
- [26] Zaranappa, H.M. Vagdevi, N.D. Jayanna and K.P. Latha, Der. Pharm. Chem., 4, 1754-1758 (2012).
- [27] Ibrahim, S.S., A.M. Abdel-Halim, Y. Gabr, S. El Edfawy and R.M. Abdel-Rahaman, J. Chem. Res.(S), 5, 154-155 (1997).
- [28] Gouda, M.A., M.A. Berghot, G.A. Ghani and A.M. Khalil, Eur. J. Med. Chem., 45, 1338-1345 (2010).
- [29] Antipenko, L., A. Karpenko, S. Kovalenko, A. Katsev, E. Porokhnyavets, V. Novikov and A. Chekotilo, Chem. Pharm. Bull., 57, 580-585 (2009).
- [30] Abdel-Aal, M.T., H. Abdel-Aleem, L.I. Ibahim and A.L. Zein, Arch. Pharm. Res., 33, 1891-1900 (2010).

**Copyright & License:**

© Authors retain the copyright of this article. This work is published under the Creative Commons Attribution 4.0 International License (CC BY 4.0), permitting unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.