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## Synthesis and Anti-Microbial Activity of 2-(Coumarinyl-4-Oxy)-4, 6-Dichloro-1,3,5-Triazine Tethered Pyrazoline Derivatives.

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### ABSTRACT:

Pyrazolines are a significant class of heterocyclic compounds with vital biological activities. The literature has proven that compounds with an s-triazine core containing other heterocyclic compounds frequently exhibit significant biological activities. s-triazine and pyrazoline both are major structural core systems found in many pharmacologically active Compounds. In the present study, Pyrazoline derivatives are coupled with 4-hydroxy Coumarin comprising s-triazine molecule which contributes to enhance pharmacological activities of Pyrazoline. On the basis of literature survey new molecules were designed and successfully synthesized in the laboratory using convenient synthetic protocols and structures were confirmed by <sup>1</sup>HNMR, IR and Mass Spectroscopy. All Synthesised compounds (**9a-j**) were screened for Anti-Bacterial and Anti-fungal activities.

**Keywords:** 1, 3, 5-Triazine, Chalcones, 4-Hydroxy Coumarin, Pyrazoline, Disubstituted Triazine Derivatives, Anti-bacterial and Anti-fungal Activities.

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## 1. Introduction

Nitrogen comprising heterocycles has received considerable attention from last few decades due to their biological activities, 1, 3, 5-triazine is among them. It has a general formula  $C_3H_3N_3$  and is six membered heterocyclic compounds having three nitrogen atoms and three chlorine atoms which are easily substituted by many nucleophiles. s-triazines and substituted s-triazines are among the many nitrogen-containing heterocyclic compounds that are particularly useful in drug discovery due to their wide range of possible biological actions<sup>1</sup>. Aside from its usage as pharmacophores in medicinal chemistry, 1,3,5-triazine is widely researched due to its numerous applications in biological systems as an anticancer, antibacterial, antiviral, and antifungal agent<sup>2</sup>. The 1,3,5-triazine ring offers a notable framework for the creation of biologically active compounds with a wide range of therapeutic applications and anti-protozoa properties<sup>3</sup>. s-triazine hold a special place in the field of pharmaceutical chemistry. It functions as a protective group in organic chemistry. It's a group that are reactive and adaptable to many synthetic transformations<sup>4</sup>. s-triazine is one of the oldest known organic molecules, some of their derivatives have been known for at least 150 years<sup>5</sup>. Decades of research have revealed a wide range of properties of s-triazine derivatives<sup>6</sup>. Due to the fact that s-triazine is present in numerous medications; its chemistry has been thoroughly investigated<sup>7</sup>. Because of the broad variety of biological activity exhibited by the s-triazine based Chalcones and their derivatives, they have generally been the subject of substantial research<sup>8</sup>. A notable structural core system seen in many pharmacologically active chemicals is shared by an intriguing class of heterocyclic compounds called Chalcones and triazines<sup>9</sup>.

Coumarin belongs to the lactone family and is categorized as an oxygen-heterocyclic molecule. It is also known as benzo-fused lactone. Its skeletal structure is similar to that of benzopyrones<sup>10</sup>. Because of their numerous biological applications, coumarin derivatives have been essential to medicinal chemistry among all the Coumarin derivatives, 4-hydroxy coumarins shown promise for use in anticancer, antimalarial, antifungal, antiviral, and anticoagulant medicinal applications. As antibiotics (Novobiocin), anti-AIDS medications, and anticancer medications (Gelparvarin), they have had significant outcomes. After extensive research, some of these medications have been made from 4-hydroxycoumarin, which has promising biological action<sup>11</sup>.

Pyrazolines are heterocyclic compounds with two nitrogen atoms positioned close to three carbon atoms in a five-membered unsaturated ring structure. Numerous compounds of pyrazolines have significant pharmacological properties, rendering them valuable resources for drug discovery<sup>12</sup>. Due to their numerous biological effects, pyrazoline have drawn the interest of chemists and pharmacologists<sup>13</sup>. It was widely known that pyrazoline-linked heterocyclic compounds had a wide range of pharmacological activities. Strong biological activity with reduced toxicity has been demonstrated by a modified pyrazoline ring connected to other heterocyclic moieties<sup>14</sup>. The biological properties of Chalcones and their pyrazoline derivatives, which include antibacterial, antiviral, antifungal, anti-inflammatory, antihypertensive, and anticancer properties, make them highly significant molecules in pharmaceutical chemistry<sup>15-17</sup>. Many chemical and biological substances contain pyrazolines, which are highly useful heterocyclic molecules that contain nitrogen and improve their actions<sup>18</sup>. Pyrazolines are among the most active classes of compounds in medicinal chemistry and chemical biology and they have very wide range of pharmacological properties<sup>19</sup>. In continuation of our previous work on the pure synthesis and antimicrobial evaluation of s-triazine comprising Chalcones derivatives, herein we have targeted to synthesized new series of s-triazine derivatives with more potent antibacterial and antifungal activities.

## Experimental

### Experimental section:

All the Melting points were determined in open capillaries. <sup>1</sup>HNMR spectral data was recorded at 500 MHz (Bruker Avance) Cryo-magnet Spectrometer in CDCl<sub>3</sub> or DMSO Solvent using TMS as an internal standard. IR spectral data were recorded on a FT Infra-Red Spectrophotometer Model RZX Perkin Elmer. The synthesised products were confirmed by the comparison of their Mass, IR, <sup>1</sup>HNMR spectral data. TLC was carried out on Silica gel G (Merk) plates with n-Hexane/Ethyl Acetate (8:2) system.

## 2. Chemicals and Materials

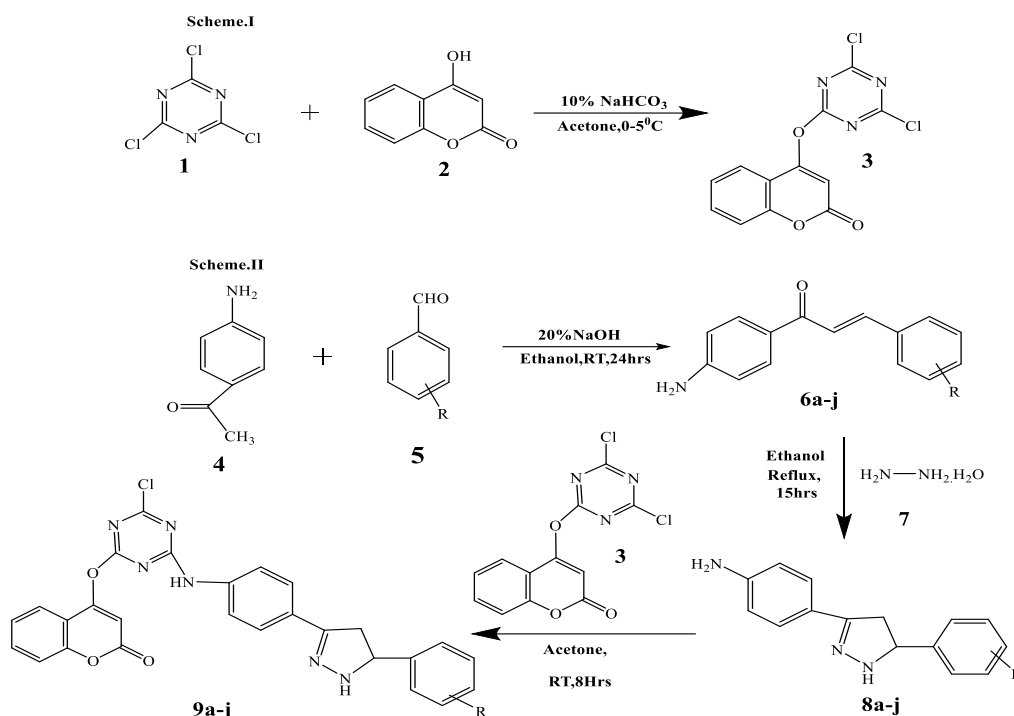
### Synthesis of 2-(Coumarinyl-4-oxy)-4,6-dichloro-s-triazine (3)

To a stirred solution of s-triazine (0.05 mol) in acetone (60 ml) at 0-5°C, the solution of 4-hydroxy Coumarin (0.05 mol) in 10% NaHCO<sub>3</sub> (50 ml) was added drop wise with stirring for 2 hours. The progress of the reaction was being monitored by TLC using n-Hexane/Ethyl Acetate (8:2) as Eluent. After reaction completion, the stirring of reaction mixture was stopped and the reaction mixture was poured in to crushed ice. The obtained product was filtered and dried. The crude product was purified and recrystallized from acetone to give the compound (3); yield 90%, M.P. 207-209°C. The physical data is recorded and correlated with reference<sup>11, 20</sup>.

**General Procedure for the Preparation of 1-(4-aminophenyl)-3-phenylprop-2-en-1-ones (6a-j):** - Equimolar quantity (0.01mol) of 4-Amino Acetophenone and respective aryl aldehyde were mix and dissolved in 30 ml of alcohol. To this add aqueous potassium hydroxide (KOH 20%) solution then it was continuously stirred for 24 hours at room temperature and the reactions progress was monitored on Thin Layer Chromatography (n-Hexane/Ethyl Acetate, 8:2), after completion of reaction, it was poured on crushed Ice and neutralized with dil. HCL and the obtained product was filtered, dried and recrystallized from alcohol. The physical data is recorded and correlated with reference<sup>21-23</sup>.

**General Procedure for the Preparation of 4-(5-phenyl-4,5-dihydro-1H-pyrazol-3-yl) aniline compound (8a-j).** A Mixture of Chalcones (0.01mol) and Hydrazine hydrate (0.02mol) was taken in Ethanol (30ml) and reflux the reaction mixture for 15 hrs. the reactions progress was monitored on Thin Layer Chromatography (n-Hexane/Ethyl Acetate, 8:2), after completion of reaction, it was poured on crushed Ice filtered, dried and recrystallized from alcohol. The physical data is recorded and correlated with reference<sup>8</sup>.

**General Procedure for the Preparation 4-((4-chloro-6-((4-(5-phenyl-4,5-dihydro-1H-pyrazol-3-yl)phenyl)amino)-1,3,5-triazin-2-yl)oxy)-2H-chromen-2-one Derivatives(9a-j):** To the stirred solution (0.001mol) of compound 3 in 20 ml acetone add Compound 7a-j, (0.001mol) ,and stirred the reaction mixture for 8 hrs, maintaining the temp 40°C the pH was kept neutral by the appropriate addition of 10% NaHCO<sub>3</sub> Solution. The temperature was steadily raised to 45°C during 3 hours and further maintained for 6 hrs. the reactions progress was monitored on Thin Layer Chromatography (n-Hexane/Ethyl Acetate, 8:2), after completion of reaction, it was poured on crushed Ice. The solid obtained product was filtered and dried. The crude was purified and recrystallized from Acetone.



**Scheme II. 4-((4-chloro-6-((4-(5-phenyl-4,5-dihydro-1H-pyrazol-3-yl) phenyl) amino)-1,3,5-triazin-2-yl) oxy)-2H-chromen-2-one derivatives.**

#### Spectral Analysis and Physical Data of Synthesized Compound (9a-j):

**4-((4-chloro-6-((4-(5-phenyl-4,5-dihydro-1H-pyrazol-3-yl)phenyl)amino)-1,3,5-triazin-2-yl)oxy)-2H-chromen-2-one (9a):** Yield 68%, m.p.160<sup>0</sup>C. IR (KBr,  $\nu_{\max}$ ,  $\text{cm}^{-1}$ ): 3405, 3260 (-NH- stretching), 2961(-C-H-stretch in Ar-H), 1715(-C=O stretch in Coumarin), 1605(C=N stretch), 1215(-C-O-C- stretch in Coumarin).

<sup>1</sup>H NMR (DMSO,500 MHz,  $\delta$  ppm): 9.82 (s,1H,-NH-), 9.40 (s,1H,-NH-,exchangeable with D<sub>2</sub>O),7.72(d,2H),7.35(d,2H),7.30(d,2H),7.26(m,1H, ),7.25(d,2H),7.35-7.70(m,4H),

5.70(s,1H), 3.80(dd,1H,H<sub>a</sub>), 3.60(dd,1H,H<sub>b</sub>), 4.02(m,1Hx). Mass (m/z):510.94[m+1], Anal. Calcd. For C<sub>27</sub>H<sub>19</sub>ClN<sub>6</sub>O<sub>3</sub>, C: 63.47, H: 3.75, N: 16.45, Found C: 63.42, H: 3.71, N: 16.41.

#### **4-((4-chloro-6-((4-(5-(2-chlorophenyl)-4,5-dihydro-1H-pyrazol-3-yl)phenyl)amino)-1,3,5-triazin-2-yl)oxy)-2H-chromen-2-one (9b):**

Yield 84%, m.p.190<sup>0</sup>C. IR (KBr,  $\nu_{\max}$ ,  $\text{cm}^{-1}$ ): 3415,3220 (-NH- stretch), 2980(-C-H- stretch in Ar), 1720 (-C=O stretch in Coumarin), 1612 (C=N), 1221 (-C-O-C- stretch in Coumarin), 750 (C-Cl stretch).<sup>1</sup>HNMR (DMSO,500 MHz,  $\delta$  ppm): 9.85 (s,1H,-NH-), 9.43 (s, 1H, -NH-, exchangeable with D<sub>2</sub>O), 7.74 (d, 2H ), 7.66 (d,1H), 7.20-7.28 (m,3H), 7.25 (d,2H, ), 7.35-7.70(m,4H), 5.70(s,1H), 3.90(dd,1H,H<sub>a</sub>), 3.62(dd,1H,H<sub>b</sub>), 4.06(m,1Hx). Mass (m/z): 545.38 [m+1], Anal. Calcd. For C<sub>27</sub>H<sub>18</sub>Cl<sub>2</sub>N<sub>6</sub>O<sub>3</sub>, C:59.46, H: 3.33, N:15.41, Found C:59.41, H: 3.28, N:15.38.

#### **4-((4-chloro-6-((4-(5-(4-chlorophenyl)-4,5-dihydro-1H-pyrazol-3-yl)phenyl)amino)-1,3,5-triazin-2-yl)oxy)-2H-chromen-2-one (9c):**

Yield 85%, m.p.160<sup>0</sup>C. IR (KBr,  $\nu_{\max}$ ,  $\text{cm}^{-1}$ ): IR (KBr,  $\nu_{\max}$ ,  $\text{cm}^{-1}$ ): 3484,3126 (-NH- stretch), 2881(-C-H- stretch in Ar), 1721 (-C=O stretch in Coumarin), 1616 (C=N), 1218 (-C-O-C- stretch in Coumarin), 761 (C-Cl stretch).<sup>1</sup>H NMR (DMSO,500 MHz,  $\delta$  ppm): 9.91 (s,1H,-NH-), 9.46 (s, 1H, -NH-, exchangeable with D<sub>2</sub>O), 7.76 (d, 2H ), 7.47 (d,2H), 7.46 (d,2H), 7.27 (d,2H, ), 7.35-7.70(m,4H), 5.72(s,1H), 3.92(dd,1H,H<sub>a</sub>), 3.63(dd,1H,H<sub>b</sub>), 4.09(m,1Hx). Mass (m/z): 545.38 [m+1], Anal. Calcd. For C<sub>27</sub>H<sub>18</sub>Cl<sub>2</sub>N<sub>6</sub>O<sub>3</sub>, C:59.46, H: 3.33, N:15.41, Found C:59.41, H: 3.28, N:15.38.

**4-((4-chloro-6-((4-(5-(2,4-dichlorophenyl)-4,5-dihydro-1H-pyrazol-3-yl)phenyl)amino)-1,3,5-triazin-2-yl)oxy)-2H-chromen-2-one (9d):** Yield 76%, m.p.200<sup>0</sup>C. IR (KBr,  $\nu_{\max}$ ,  $\text{cm}^{-1}$ ): 3486,3222 (-NH- stretch), 2980(-C-H- stretch in Ar), 1722 (-C=O stretch in Coumarin), 1619 (C=N), 1220 (-C-O-C- stretch in Coumarin), 763 (C-Cl stretch).<sup>1</sup>H NMR (DMSO,500 MHz,  $\delta$  ppm): 9.85 (s,1H,-NH-), 9.44 (s, 1H, -NH-, exchangeable with D<sub>2</sub>O), 7.75 (d, 2H ), 7.71 (s,1H), 7.40 (d,1H), 7.26 (d,2H, ),7.01(d,1H), 7.35-7.70(m,4H), 5.69(s,1H), 3.93(dd,1H,H<sub>a</sub>), 3.62(dd,1H,H<sub>b</sub>), 4.07(m,1Hx). Mass (m/z): 579.82 [m+1], Anal. Calcd. For C<sub>27</sub>H<sub>17</sub>Cl<sub>3</sub>N<sub>6</sub>O<sub>3</sub>, C:55.93, H: 2.96, N:14.49, Found C:55.89, H: 2.92, N:14.46

**4-((4-chloro-6-((4-(5-(2-fluorophenyl)-4,5-dihydro-1H-pyrazol-3-yl)phenyl)amino)-1,3,5-triazin-2-yl)oxy)-2H-chromen-2-one (9e):** Yield 67%, m.p.180<sup>0</sup>C. IR (KBr,  $\nu_{\max}$ ,  $\text{cm}^{-1}$ ): 3410,3210 (-NH- stretch), 2985(-C-H- stretch in Ar), 1722 (-C=O stretch in Coumarin), 1600 (C=N), 1219 (-C-O-C- stretch in Coumarin), 978 (C-F stretch).<sup>1</sup>H NMR (DMSO,500 MHz,  $\delta$  ppm): 9.85 (s,1H,-NH-), 9.38 (s, 1H, -NH-, exchangeable with D<sub>2</sub>O), 7.73 (d, 2H ), 7.70 (m,1H), 7.20-7.60 (m,3H), 7.24 (d,2H, ), 7.35-7.70(m,4H), 5.65(s,1H), 3.89(dd,1H,H<sub>a</sub>), 3.60(dd,1H,H<sub>b</sub>), 4.03(m,1Hx). Mass (m/z): 528.93 [m+1], Anal. Calcd. For C<sub>27</sub>H<sub>18</sub>ClFN<sub>6</sub>O<sub>3</sub>, C:61.31, H: 3.43, N:15.89,Found C:61.25, H: 3.38, N:15.84.

**4-((4-chloro-6-((4-(5-(4-fluorophenyl)-4,5-dihydro-1H-pyrazol-3-yl)phenyl)amino)-1,3,5-triazin-2-yl)oxy)-2H-chromen-2-one (9f):** Yield 73%, m.p.130<sup>0</sup>C. IR (KBr,  $\nu_{\max}$ ,  $\text{cm}^{-1}$ ): 3475,3220 (-NH- stretch), 2886(-C-H- stretch in Ar), 1723 (-C=O stretch in Coumarin), 1605 (C=N), 1212 (-C-O-C- stretch in Coumarin), 993 (C-F stretch).<sup>1</sup>H NMR (DMSO,500 MHz,  $\delta$  ppm): 9.89 (s,1H,-NH-), 9.47 (s, 1H, -NH-, exchangeable with D<sub>2</sub>O), 7.72 (d, 2H ), 7.24 (d,2H), 7.20 (d,2H), 7.21 (d,2H, ), 7.35-7.70(m,4H), 5.69(s,1H), 3.89(dd,1H,H<sub>a</sub>), 3.59(dd,1H,H<sub>b</sub>), 4.04(m,1Hx). Mass (m/z): 528.93 [m+1], Anal. Calcd. For C<sub>27</sub>H<sub>18</sub>ClFN<sub>6</sub>O<sub>3</sub>, C:61.31, H: 3.43, N:15.89,Found C:61.25, H: 3.38, N:15.84.

**4-((4-((4-(5-(4-bromophenyl)-4,5-dihydro-1H-pyrazol-3-yl)phenyl)amino)-6-chloro-1,3,5-triazin-2-yl)oxy)-2H-chromen-2-one (9g):** Yield 83%, m.p.140<sup>0</sup>C. IR (KBr,  $\nu_{\max}$ ,  $\text{cm}^{-1}$ ): 3460,3205 (-NH- stretch), 2880(-C-H- stretch in Ar), 1710 (-C=O stretch in Coumarin), 1600 (C=N), 1205 (-C-O-C- stretch in Coumarin), 810 (C-Br stretch).<sup>1</sup>H NMR (DMSO,500 MHz,  $\delta$  ppm): 9.87 (s,1H,-NH-), 9.38 (s, 1H, -NH-, exchangeable with D<sub>2</sub>O), 7.71 (d, 2H ), 7.69 (d,2H), 7.16 (d,2H), 7.23 (d,2H, ), 7.35-7.70(m,4H), 5.68(s,1H), 3.90(dd,1H,H<sub>a</sub>), 3.60(dd,1H,H<sub>b</sub>), 4.03(m,1Hx).Mass (m/z): 589.83 [m+1], Anal. Calcd. For C<sub>27</sub>H<sub>18</sub>ClBrN<sub>6</sub>O<sub>3</sub>, C:54.98, H: 3.08, N:14.25, Found C:54.93, H: 3.04, N:14.21.

**4-((4-chloro-6-((4-(5-(o-tolyl)-4,5-dihydro-1H-pyrazol-3-yl)phenyl)amino)-1,3,5-triazin-2-yl)oxy)-2H-chromen-2-one (9h):** Yield 78%, m.p.226<sup>0</sup>C. IR (KBr,  $\nu_{\max}$ ,  $\text{cm}^{-1}$ ): 3400,3195 (-NH- stretch), 2885(-C-H- stretch in Ar), 1720 (-C=O stretch in Coumarin), 1590 (C=N), 1205 (-C-O-C- stretch in Coumarin).<sup>1</sup>H NMR (DMSO,500 MHz,  $\delta$  ppm): 9.74 (s,1H,-NH-), 9.25 (s, 1H, -NH-, exchangeable with D<sub>2</sub>O), 7.69 (d, 2H ), 7.30 (m,1H), 6.90-7.20 (m,3H), 7.20 (d,2H), 7.35-7.70(m,4H), 5.50(s,1H), 3.75(dd,1H,H<sub>a</sub>), 3.89(dd,1H,H<sub>b</sub>), 4.01(m,1Hx),2.26(s,3H).Mass (m/z): 524.97 [m+1], Anal. Calcd. For C<sub>28</sub>H<sub>21</sub>ClN<sub>6</sub>O<sub>3</sub>, C:64.06, H: 4.03, N:16.01, Found C:64.01, H: 3.99, N:15.98.

**4-((4-chloro-6-((4-(5-(p-tolyl)-4,5-dihydro-1H-pyrazol-3-yl)phenyl)amino)-1,3,5-triazin-2-yl)oxy)-2H-chromen-2-one (9i) :** Yield 76%, m.p.210<sup>0</sup>C. IR (KBr,  $\nu_{\max}$ ,  $\text{cm}^{-1}$ ): 3395,3190 (-NH- stretch), 2870(-C-H- stretch in Ar), 1715 (-C=O stretch in Coumarin), 1588 (C=N), 1200 (-C-O-C- stretch in Coumarin).<sup>1</sup>H NMR (DMSO,500 MHz,  $\delta$  ppm): 9.69 (s,1H,-NH-), 9.22 (s, 1H, -NH-, exchangeable with D<sub>2</sub>O), 7.68 (d, 2H ), 7.20 (d,2H), 7.05 (d,2H), 7.19 (d,2H), 7.35-

7.70(m,4H), 5.62(s,1H), 3.68(dd,1H,H<sub>a</sub>), 3.79(dd,1H,H<sub>b</sub>), 3.00(m,1H<sub>x</sub>),2.27(s,3H). Mass (m/z): 524.97 [m+1], Anal. Calcd. For C<sub>28</sub>H<sub>21</sub>ClN<sub>6</sub>O<sub>3</sub>, C:64.06, H: 4.03, N:16.01, Found C:64.01, H: 3.99, N:15.98

**4-((4-chloro-6-((4-(5-(4-methoxyphenyl)-4,5-dihydro-1H-pyrazol-3-yl)phenyl)amino)-1,3,5-triazin-2-yl)oxy)-2H-chromen-2-one (9j):** Yield 76%, m.p.155<sup>0</sup>C. IR (KBr, ν<sub>max</sub>, cm<sup>-1</sup>): 3390,3185 (-NH- stretch), 2860(-C-H- stretch in Ar), 1705 (-C=O stretch in Coumarin), 1582 (C=N), 1196 (-C-O-C- stretch in coumarin), 1020(-C-O-C-). <sup>1</sup>HNMR (DMSO,500 MHz, δ ppm): 9.66 (s,1H,-NH-), 9.20 (s, 1H, -NH-, exchangeable with D<sub>2</sub>O), 7.63 (d, 2H ), 7.19 (d,2H), 6.86 (d,2H), 7.16 (d,2H, ),7.35-7.70(m,4H), 5.69(s,1H), 3.65(dd,1H,H<sub>a</sub>), 3.77(dd,1H,H<sub>b</sub>), 4.00(m,1H<sub>x</sub>),3.78(s,3H). Mass (m/z): 540.96 [m+1], Anal. Calcd. For C<sub>28</sub>H<sub>21</sub>ClN<sub>6</sub>O<sub>4</sub>, C:62.17, H: 3.91, N:15.54, Found C:62.12, H: 3.87, N:15.50.

### Biological Activity:

#### Anti-bacteria and Anti-fungal Activities.

All the novel synthesized compounds from the series (9a-j) were screened for antibacterial activity against two Gram-Positive Bacteria viz. *B. licheniformis* and *B. subtilis*, and Gram-Negative Bacteria viz. *E.coli* by disk diffusion assay<sup>24</sup>. Using Chloramphenicol (100 μs/disk) the reference standard for comparing the results. The Anti-bacterial activity was screened by using nutrient agar obtained from Hi-media. Composition (g/L<sup>-1</sup>). Sodium chloride-5 : Beef extract-3: Penton 5.0 (P<sup>H</sup> 7.2).The novel synthesized series of compounds (9a-j) were also screened for Anti-fungal activity against *A.niger* and *C. albicans* by agar diffusion assay<sup>25</sup>, Using Amphotericin B (100 units /disk) as the reference standard. The Antifungal activity is screened by using Sabouraud Agar Media and DMSO as control Solvent. The diameter of the Zone is measured by Vernier Calliper. The Anti-Bacterial and Anti-Fungal Activity of the 4-((4-chloro-6-((4-(5-phenyl-4,5-dihydro-1H-pyrazol-3-yl)phenyl)amino)-1,3,5-triazin-2-yl)oxy)-2H-chromen-2-one compound derivatives (9a-j) is shown in **Table No.1**.

TABLE NO 1. Anti-bacterial and Anti-fungal Screening of Compounds (9a-j)

Compound		Anti-bacterial Activity			Anti-Fungal Activity	
		<i>B. licheniformis</i>	<i>B. subtilis</i>	<i>E. Coli</i>	<i>P. aeruginosa</i>	<i>C. albicans</i>
9a	H	7.9	-	-	-	8.7
9b	2-Cl	8.1	-	-	-	8.9
9c	4-Cl	7.9	8.1	-	-	9.0
9d	2,4-Cl	-	9.4	-	-	9.0
9e	2-F	-	-	-	-	13.2
9f	4-F	-	8.3	-	8.4	16.2
9g	4-Br	-	-	-	-	11.2
9h	2-Me	-	-	-	8.3	17.1
9i	4-Me	8.6	-	-	-	12.6
9j	4-OCH <sub>3</sub>	-	-	-	9.0	13.3
Chloramphenicol		17	19	27.1	16.00	NA
Amphotericin B		NA	NA	NA	NA	10.11

### 3. Result And Discussion

Literature survey reveals that there are no reports of 4-((4-chloro-6-((4-(5-phenyl-4,5-dihydro-1H-pyrazol-3-yl) phenyl) amino)-1,3,5-triazin-2-yl) oxy)-2H-chromen-2-ones, hence it was

planned to synthesize these compounds. In the present study step-I, 1,3,5-triazine(1) is reacted with 4-Hydroxy Coumarin (2) in the presence of aq.NaHCO<sub>3</sub> in acetone to the yield 2-(Coumarinyl-4- oxy)-4,6-dichloro-s-triazine (3) in good yield. In the step-II, 4-Amino acetophenone (4) is reacted with the substituted aryl aldehydes (5) in the presence of NaOH followed by condensation reaction to yield 1-(4-aminophenyl)-3-phenylprop-2-en-1-one compound derivatives (6a-6j) with good yield. Further synthesis, Compounds (6a-6j) reacted with the compound (7) which yield compound (8a-j), in the final step compound (8a-j) reacted with compound (3) in the presence of aq.NaHCO<sub>3</sub> and Acetone as solvent to Yield (9a-9j) with excellent yield. The IR spectra of (9c) show strong absorption band at 3484 cm<sup>-1</sup> and 3126 cm<sup>-1</sup> indicated the Stretching frequency of -NH- functional group. 1721cm<sup>-1</sup> is stretching of (-C=O) in Coumarin ring, 1616cm<sup>-1</sup> is the value for (-C=N) stretching in pyrazoline confirm the synthesis of coupling of pyrazoline derivatives with s-triazine. <sup>1</sup>H NMR Spectrum of (9c) show that, Singlet at δ 9.30 ppm for (-NH-) gives confirmation of secondary amine, again singlet at δ 9.90 ppm confirm the (-NH-) of pyrazoline whereas δ 5.85 ppm for (-C-O-C-) confirm that the coupling of s-triazine and 4-hydroxy Coumarin. The Mass Spectra shows the molecular Ion peak at 545.38 [m+1]. All these Spectral analyses show that the Confirmation of synthesis of (9a-9j) compounds.

Screening of the biological activities of synthesized compounds revealed that, compound 9a,9b,9c,9i show good Anti-bacterial activity against Gram Positive bacteria i.e. *B. licheniformis*. Compounds 9c, 9d, 9f having electron withdrawing chlorine and fluorine show good Anti-bacterial activity against Gram-Positive bacteria i.e. *B.subtilis*. Compounds No any compound show antibacterial activity against Gram-Negative bacteria i.e. *E.coli*. and *P. aeruginosa*. The Investigation of Anti-fungal activity data revealed that, the compound which has chlorine substituent shows less zone of inhibition as compare to other substituent against *C. albicans*. Fungal strain compared with the standard Amphotericin B drug.

#### 4. Conclusion

In the present research, we have reported the synthesis of new series of 4-((4-chloro-6-((4-(5-phenyl-4,5-dihydro-1H-pyrazol-3-yl)phenyl)amino)-1,3,5-triazin-2-yl)oxy)-2H-chromen-2-one derivatives (9a-j). The Compound with electron withdrawing group shows promising Anti-bacterial activities as compared to the standard Chloramphenicol drug. All synthesized compound shows potent Antifungal activities against standard Amphotericin B. drug. But chlorine substituent has less antifungal activity as compare to another substituent. All the synthesized series of compound shows excellent to moderate activity against the Pathogens and are very promising core molecule as potent Antifungal agents, further investigation is needed

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**Conflict of Interest:** The authors affirm that their publication of these articles does not present a conflict of interest.

## 5. References

1. Gui-Feng Kang and Gang Zhang Beilstein J. Org. Chem. 16, 2020,1447-1455.doi:10.3762/bjoc.16.120
2. Sharma, A.; Sheyi, R.;de la Torre, B.G.; El-Faham, A.;Albericio, F. s-Triazine: A Privileged Structure for Drug Discovery and Bioconjugation. *Molecules*, 26, 2021,864.
3. A. Majeed Ganai, T. Khan Pathan, G. A. Hampannavar, C. Pawar, V. A. Obakachi, B. Kushwaha, N. Deshwar Kushwaha, R. Karpoormath, Recent Advances on the s-Triazine Scaffold with Emphasis on Synthesis, Structure-Activity and Pharmacological Aspects: A Concise Review *Chemistry Select*, 6, 2021,1616–1660.
4. Rajeev Kumar, Neeraj Kumar, Ram Kumar Roy and Anita Singh, Triazines – A comprehensive review of their synthesis and diverse biological importance, *Curr Med Drug Res*, 1, 2017, (1), Article ID 173.
5. Dávila Cerón, V.; Illicachi, L.A.; Insuasty, B. Triazine: An Important Building Block of Organic Materials for Solar Cell Application. *Molecules*, 28, 2023,257.
6. Maliszewski, D. Drozdowska, D. Recent Advances in the Biological Activity of s-Triazine Core Compounds. *Pharmaceuticals*, 15, 2022,221.
7. S. G. Kansara, R. D. Pandit and V. G. Bhawe, synthesis of some new ibuprofen derivatives containing chief heterocyclic moiety like s-triazine and evaluated for their analgesic activity, (*RASAYAN. J. Chem*, Vol.2, No.3, 2009, 699-705.
8. Anjani Solankee, Kishor Kapadia, Ana Ciric, Marina Sokovic, Irini Doytchinova, Athina Geronikaki, Synthesis of some new S-triazine based chalcones and their Derivatives as potent antimicrobial agents, *European Journal of Medicinal Chemistry* 45 2010,510–518.
9. G. V. Pavan Kumar, D. Srinivasa Rao, B. Pooja, G. Harika & Y. Anil Kumar, Design, Synthesis, Spectral Characterization of Some New Fully Unsaturated 2-Substituted-4,6 Dichloro Symmetric Triazine based Chalcones Hybrids, *Global Journal of Medical Research: B Pharma, Drug Discovery, Toxicology & Medicine*, Volume 16 Issue 1 Version 1.0 Year 2016.
10. Abhimanyu Pawara, Kishor Naktodea, Kishore Purib and Santosh Gaikwad. Synthesis of Coumarin-coupled pyrazole and isoxazole compounds, *Heterocyclic Letters* Vol. 14| No.1, 2024,73-81.
11. Archana Y Cholera, Kartik D Ladva, A Convenient Synthesis of Trisubstituted 1,3,5-triazine Derivatives and their Antimicrobial Screening, *Der Pharma Chemica*, 10(4): 2018,57-61.
12. Balasubramanian, S., Irfan, N., Umamaheswari, A., & Puratchikody, A. (2018). Design and virtual screening of novel fluoroquinolone analogs as effective mutant DNA Gyrase inhibitors against urinary tract infection-causing fluoroquinolone resistant *Escherichia coli*. *RSC advances*, 8(42), 23629-23647.
13. Navabshan, I., Sakthivel, B., Pandiyan, R., Antoniraj, M. G., Dharmaraj, S., Ashokkumar, V., ... & Show, P. L. (2021). Computational lock and key and dynamic trajectory analysis of natural biophors against COVID-19 spike protein to identify effective lead molecules. *Molecular biotechnology*, 63(10), 898-908.
14. Sugumaran, A., Pandiyan, R., Kandasamy, P., Antoniraj, M. G., Navabshan, I., Sakthivel, B., ... & Ngamcharussrivichai, C. (2022). Marine biome-derived secondary metabolites, a class of promising antineoplastic agents: A systematic review on their classification, mechanism of action and future perspectives. *Science of the Total Environment*, 836, 155445.
15. Balasubramanian, S., Irfan, N., Senthilkumar, C., Umamaheswari, A., & Puratchikody, A. (2020). The synthesis and biological evaluation of virtually designed fluoroquinolone

- analogs against fluoroquinolone-resistant *Escherichia coli* intended for UTI treatment. *New Journal of Chemistry*, 44(31), 13308-13318.
16. Puratchikody, A., Irfan, N., & Balasubramanian, S. (2019). Conceptual design of hybrid PCSK9 lead inhibitors against coronary artery disease. *Biocatalysis and Agricultural Biotechnology*, 17, 427-440.
  17. Santhaseelan, H., Dinakaran, V. T., Sakthivel, B., Somasundaram, M., Thanamegam, K., Devendiran, V., ... & Rathinam, A. J. (2022). Bioactive efficacy of novel carboxylic acid from halophilic *Pseudomonas aeruginosa* against methicillin-resistant *Staphylococcus aureus*. *Metabolites*, 12(11), 1094.
  18. Irfan, N., Balasubramanian, S., Ali, D. M., & Puratchikody, A. (2023). Bioisosteric replacements of tyrosine kinases inhibitors to make potent and safe chemotherapy against malignant cells. *Journal of Biomolecular Structure and Dynamics*, 41(19), 9437-9447.
  19. Nagendran, S., Balasubramanian, S., & Irfan, N. (2023). Virtually screened novel sulfathiazole derivatives as a potential drug candidate for methicillin-resistant *Staphylococcus aureus* and multidrug-resistant tuberculosis. *Journal of Biomolecular Structure and Dynamics*, 41(11), 5086-5095.
  20. Venkatesh, G., Vennila, P., & Balasubramanian, S. (2024). Solvent effects, chemical reactivity, docking and antimicrobial activity of silver and gold nanocages glimepiride: Experimental and theoretical calculations. *Chemical Physics Impact*, 8, 100498.
  21. Kotakonda, M., Marappan, M., Dharmar, P., Sakthivel, B., & Sunnapu, P. (2023). Isolation and Identification of Bioactive Compounds with Antimicrobial Activity from Marine Facultative Anaerobe, *Bacillus subtilis*. *Current Pharmaceutical Biotechnology*, 24(5), 698-707.
  22. Faisal, M., Saeed, A., Hussain, S., Dar, P., Ali larik, F.,. Recent developments in synthetic chemistry and biological activities of pyrazole derivatives. *J. Chem. Sci.* ,131, 2019,70.
  23. S. Sathiya, A. Keerthika, B. S. Krishnamoorthy, S. Nandhabala, S. Aravind, N. Hari and R. Ravikumar, synthesis of novel pyrazolines and their antimicrobial activity, *Rasayan J. Chem.*, 13(1) Tok F, Koçyiğit-Kaymakçioğlu B, Sağlık BN, Levent S, Özkay Y, Kaplançıklı ZA.
  24. Synthesis and biological evaluation of new pyrazolone Schiff bases as monoamine oxidase and cholinesterase inhibitors. *Bioorg Chem.* 84: 2019,41-50., 2020,676-683.
  25. Kaplançıklı ZA, Turan-Zitouni G, Özdemir A, Can ÖD, Chevallet P. Synthesis and anti-nociceptive activities of some pyrazoline derivatives. *Eur J Med Chem.* 44(6): 2009,2606-2610.