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Synthesis and Antimicrobial Activity of some Phthalazin-1(2H)-One Derivatives

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ABSTRACT:

The phthalazine was allowed to react with 2-aminoethanol, allyl bromide, benzoyl chloride, 2-chloroethanol, phosphorous pentasulfide, ethyl cyanoacetate, phenyl isothiocyanate, piperazine and ethyl piperazine-1-carboxylate. The products were allowed to react with phenyl isothiocyanate, acetic anhydride, 4-aminobenzoic acid, phenylene diamine and piperonaldehyde afforded the more interesting derivatives of phthalazinone. The phthalazinethione reacted with hydrazine hydrate followed by anisaldehyde to give the corresponding arylidene. The phthalazinon-2-ylethylalcohol derivative reacted with phenol and catechol to give phenolic compounds 14 that be acetylated to afford the diacetyl derivatives. The new compounds were synthesized with the objective of studying their antifungal and antimicrobial activity. Some of them gave positive results. The newly synthesized compounds were characterized on the basis of their spectral (1H-NMR, Mass spectrum, IR and Elementary analysis

KEY WORDS: phthalazin-1(2H)one, allyl-phthalazinone, epichlorohydrin, antimicrobial reagents

1. INTRODUCTION

Phthalazin-1(2H)-ones are of considerable interest due to their antidiabetic ¹, antiallergic ², vasorelaxant ³, PDE4 inhibitors ⁴, VEGF (vascular endothelial group factor) receptor tyrosine kinases for the treatment of cancer ^{5,6}, antiasthmatic agents with dual activities of thromboxane A2 (TXA2) synthetase inhibition and bronchodilation ⁷, herbicidal ⁸. A number of established drug molecules like Hydralazine ^{9,10}, Burdralazine ^{11,12}, Azelastine ^{13,14}, Ponalrest ¹⁵, and Zopolrest ¹⁶ are prepared from the corresponding phthalazinones. Several phthalazine derivatives have been reported to possess antitumor ¹⁷⁻¹⁹, antihypertensive ^{20,21}, anticovulsant ²², antimicrobial ²³, antitrypanosomal ²⁴, and anti-inflammatory activity ²⁵. Most of the current nonsteroidal anti-inflammatory drugs (NSAIDs) show serious side effects including gastrointestinal disorders and kidney damage. These studies for developing safer NSAIDs lacking the gastrointestinal and renal side effects of current used ones have recently been of interest for many researches. Most of the classical NSAIDs exerts their side effects by inhibition of COX-1 enzyme since the COX-1 isoform is the constitutive one that is responsible for regulation of physiological processes, and the COX-2 isoform is discovered to be the enzyme induced by inflammatory stimuli, selective inhibition of COX-2 provides a rationale for developing anti-inflammatory and analgesic agents. Although the diaryl heterocyclic compounds are mainly studied as new class of NSAIDs without gastric side effects, many studies have also focused on a different type of compounds to develop safer NSAIDs ²⁶. Also in terms of this aspect, many studies have been focused on pyridazin-(3H)-ones, which are characterized to possess good analgesic and anti-inflammatory activities. Beside pyridazinones, these studies have indicated that the heterocyclic ring substitutions at the six position,

and the presence of acetamide side chain when linked to the lactam nitrogen of pyridazinone ring at the two position of the pyridazinone ring, improve the analgesic and anti-inflammatory activity along with nil or very low ulcerogenicity²⁷⁻³⁰. In view of the aforementioned facts, it seemed most interesting to synthesize some [4-((3-chloro-4-methylphenyl)-2-substituted phthalazin-1(2H)-one)] derivatives with the aim to obtain more precise information about the course of reactions and biological activities.

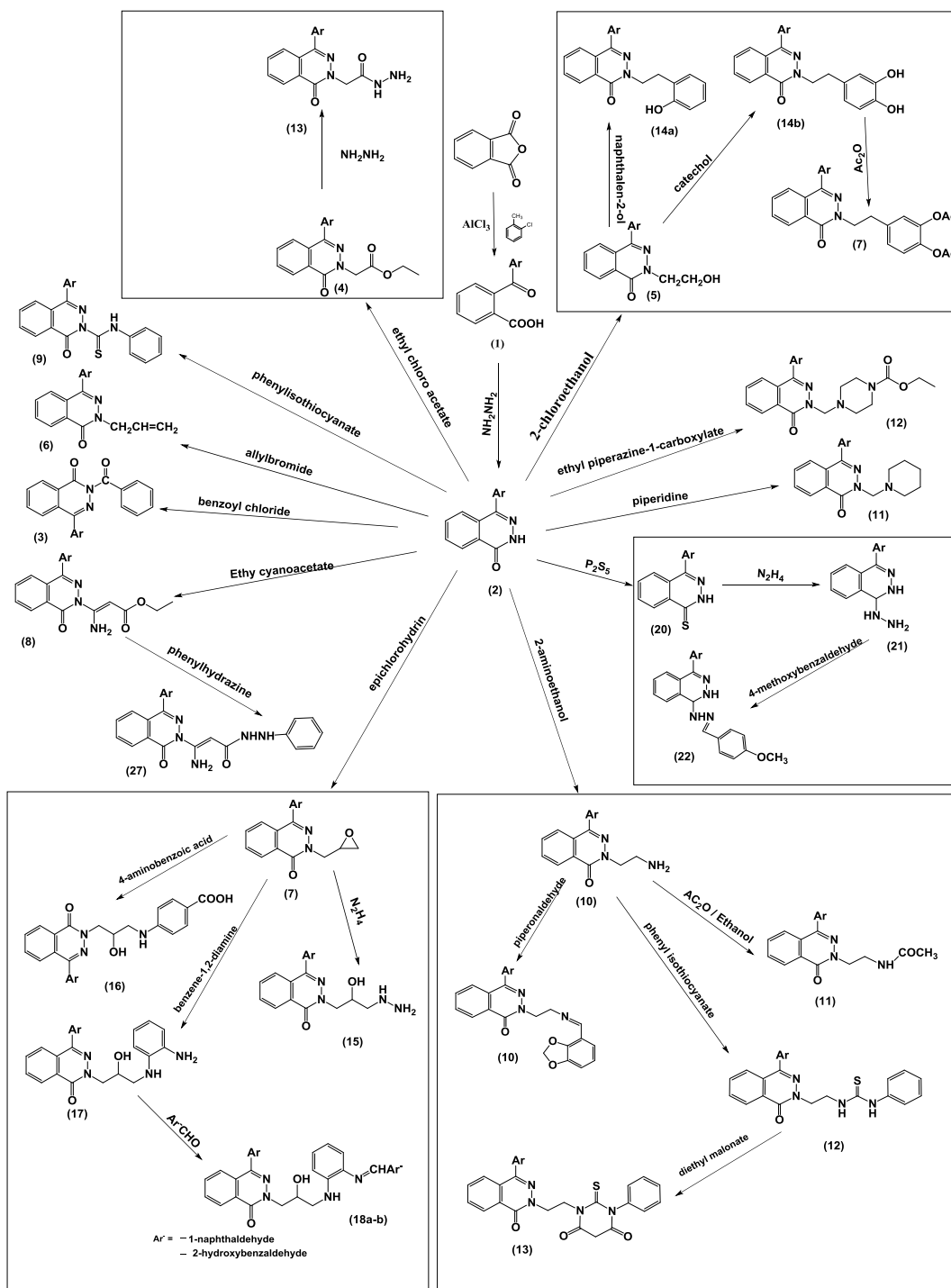
2. RESULTS AND DISCUSSION

2.1 Chemistry

Aroylation of an aromatic system by reaction with phthalic anhydride under Freidel Craft's conditions¹⁴, herein the reaction of 2-chlorotoluene with phthalic anhydride in the presence of anhydrous aluminum chloride was carried out to produce 2-(aryl benzoyl) benzoic acid. Merchant et al prepared phthalazin-1-ones via the condensation of the aroyl benzoic acid with hydrazine hydrate in boiling ethanol¹⁵. Accordingly, our phthalazinone **2** can be synthesized and its IR spectrum showed a characteristic absorption bands at ν 329, 1665 cm^{-1} corresponding to (NH and C=O) respectively. The mass spectrum shows the correct ion peak at m/z 270. Thus, extending the previous work³¹⁻³³, when the compound **2** can be investigated with different carbon electrophiles e.g. benzoyl chloride, 2-chloroethanol, ethylchloroacetate, allyl bromide, epichlorohydrin, ethylcyanoacetate phenylisothiocyanate, ethylenediamine and formaldehyde *via* mannich reaction with different 2ry amine piperidine and ethylpiperazine-1-carboxylate to afford the corresponding compounds **3**, **4**, **5**, **6**, **7**, **8**, **9**, **10**, **11** and **12**. It was treated with ethylchloroacetate and 2-chloroethanol in the presence of anhydrous K_2CO_3 and dry acetone (less polar solvent) to afford 2-alkyl phthalazin-1(2H)-one derivatives **4** and **5** respectively *via* SN^2 N-alkylation (Scheme 1). When compound **2** was allowed to react with allyl bromide, and epichlorohydrin *via* SN^1 the hetero ring opening of highly strained oxirane nucleus followed by ring closure, and not normal SN^2 on the alkyl halide moiety in dry acetone yielded 4-(3-chloro-4-methylphenyl)-2-(2-allyl/ oxiran-2-yl)phthalazin-1(2H)-one **6** and **7** respectively. On the other hand, when compound **2** was allowed to react with ethylcyanoacetate in boiling ethanol afforded 4-(3-chloro-4-methylphenyl)-2-(ethyl-3-aminopropenoat-3-yl) phthalazin-1(2H)-one **8** *via* N-alkylation corresponding by IR spectra that indicate the reaction takes place attacking of cyano group followed by isomerization to give the compound **8** (Scheme 1). Reaction of the compound **2** with phenylisothiocyanate afforded the adduct **9**. Fortunately, reaction of the phthalazinone **2** with ethylene diamine afforded 2-(2-aminoethyl)phthalazine-1(2H)-one **10** *via* removable of ammonia gas. Moreover, when compound **2** was allowed to react with formaldehyde and 2ry amine namely piperidine and ethylpiperazine-1-carboxylate afforded 2-N-(piperidinyl/ethyl-1-piperazinylcarboxylate) methylphthalazin-1(2H)-one derivatives **11** and **12** respectively. When the phthalazinone ester **4** was allowed to react with hydrazine hydrate in boiling ethanol afforded the phthalazinone hydrazide **13**. On the other hand treatment of the 2-oxarinylmethylphthalazinone **7** with different nitrogen nucleophiles e.g. hydrazine hydrate, 4-aminobenzoic acid and *o*-phenylenediamine afforded the corresponding adducts *via* ring opening to relief the strained oxirane ring. Reaction of the 2-(phthalazinon-2-yl)ethanol with active hydrogen of the phenolic compounds namely phenol and catechol in the presence of drops HCl and boiling ethanol afforded the compounds **14a** and **14b** (Scheme 1). Reaction of the 2-oxarinylmethylphthalazinone **7** with nitrogen nucleophiles namely hydrazine hydrate, 4-aminobenzoic acid and *o*-phenylenediamine afforded the corresponding adducts **15**, **16** and **17**. Reaction of the 2-aminoethylphthalazinone with different carbon electrophiles namely acetic anhydride, piperonaldehyde and phenylisothiocyanate afforded the compounds **11**, **12** and **19**. Reaction of the phthalazinthione **20** with hydrazine hydrate afforded the corresponding hydrazine that be condensed with 4-anisaldehyde afforded the corresponding arylidene derivative **22**.

2.2 Antimicrobial studies

all the synthesized compounds were done using the agar diffusion assay. This screening was performed against the Gram-positive bacteria, Gram-negative bacteria, staphylococcus aureus atcc 06538, Escherechia coli Atcc 10536, pathogenic fungi candida albicans Atcc 1023 and Aspergills Flavus. A moderate activity was observed with compounds which proved to possess marked activity against E. coli, S. aureus and C. albicans. The strong activity was observed with the most compounds of N-alkylated phthalazinone. The inhibitory concentration was determined for each of the active compounds along with Ampicillin, Streptomycin and Nystatin as positive control.



scheme (1)

Results are shown in the following **Table 1** that outlined the antimicrobial activity of compounds **4** to **14**. The authors can be reported that the N-alkylated phthalazinone derivatives (high HOMO)³⁴ converted to another configuration during the attacked of the bacteria and fungi, these are the one of explanation that why these compounds are weak or moderate effective as antibacterial and antifungal.

3. EXPERIMENTAL

All melting points are corrected and determined on a Stuart electric melting point apparatus (Microanalytical centre, ain shams university, Cairo, Egypt). Elemental analyses were carried out by Elementar Viro El-Microanalysis at the Micro-analytical Center, National Research Center, Egypt. IR spectra (KBr) were recorded on

Table 1: Antimicrobial activity of the synthesized compounds

Synthesized Compounds	Staph. aureus	Escherichia coli	Pseud. aeruginosa	Candida albicans
4a	19±0.21	22±0.12	21±0.24	16±0.22
4b	19±0.24	21±0.32	21±0.26	16±0.15
4c	19±0.11	22±0.15	22±0.02	16±0.07
5a	20±0.13	20±0.10	21±0.11	16±0.21
5b	20±0.11	21±0.23	20±0.23	21±0.14
5c	21±0.32	22±0.21	21±0.23	19±0.26
6a	18±0.11	16±0.12	18±0.21	15±0.13
6b	17±0.14	19±0.02	17±0.0	14±0.01
7a	16±0.08	13±0.04	15±0.03	14±0.09
8	20±0.13	17±0.14	18±0.17	16±0.12
9	15±0.21	17±0.12	21±0.04	12±0.14
Ampicillin	0.0	22±0.03	0.0	0.0
Streptomycin	20±0.02	21±0.01	0.0	0.0
Nystatin	0.0	0.0	0.0	22±0.02

Zone of Inhibition measured in mm. No activity (0.0), inhibition zone (< 7 mm), weak activity (7-10), moderate activity (11-15 mm), strong activity (> 15 mm), solvent CDCl₃ (6 mm).

infrared spectrometer FT-IR 400D (New York, NY, USA) using OMNIC program and are reported frequency of absorption in terms of cm⁻¹ and ¹H-NMR spectra recorded on a Bruker spectrophotometer (Rheinstetten, Germany) at 400 MHz using TMS as internal standard and with residual signals of the deuterated solvent δ = 7.26 ppm for CDCl₃ and δ 2.51 ppm for DMSO-*d*₆. ¹³C-NMR spectra were recorded on the same spectrometer (Rheinstetten, Germany) at 100 MHz and referenced to solvent signals δ = 77 ppm for CDCl₃ and δ 39.50 ppm for DMSO-*d*₆. DEPT 135 NMR spectroscopy were used where appropriate to aid the assignment of signals in the ¹H- and ¹³C-NMR spectra. The mass spectra were recorded on Shimadzu GCMS-QP-1000 EX mass spectrometer (Kyoto, Japan) used the electron ionization technique at 70 e.v. Homogeneity of all synthesized compounds was checked by TLC

4-(3-chloro-4-methylphenyl)phthalazin-1(2H)-one (2).

Hydrazine hydrate (0.015 mol) was added to a solution of 3-chloro-4-methylphenyl benzoic acid (0.01 mol) in absolute ethanol and the reaction mixture was heated under reflux for 3h. The solid that separated after cooling was filtered off and recrystallized from ethanol to

give the phthalazinone **2**, 80% yield as colorless crystals, m.p. 220-222°C; The ¹H-NMR spectrum showed signal at 2.35 (methyl group), 7.3 – 7.7 (m, 7H, ArH), 10.5 (s, 1H, NH, exchangeable with D₂O). IR (KBr) ν : 3296 (NH), 1665 (C=O), 1605 (C=N) cm⁻¹. EIMS (70 eV) *m/z* (%): 276 (M⁺, 100), 248 (43), 220 (15), 131 (25), 105 (15). Anal calcd for C₁₅H₁₁N₂OCl: C, 66.66; H, 4.07; N 10.37; found C, 66.45; H 4.02; N 10.

2-Benzoyl-4-(3-chloro-4-methylphenyl)phthalazin-1(2H)-one (3).

A solution of phthalazone derivative **2** (0.01 mol) benzoyl chloride (0.015 mol), and potassium carbonate anhydrous (3 mol) in dry acetone (50 ml) was heated under reflux for 24 h. The excess solvent was removed by distillation, and the reaction mixture was diluted with water. The solid product was obtained filtered off and crystallized from the ethanol to give **3**, 60 % yield, m.p. 110°C. IR(KBr) ν : 1668, 2852 and, 2920 cm⁻¹ attributable to C=O and CH. Anal calcd for C₂₂H₁₅N₂O₂Cl: C, 70.59; H, 4.01; N 7.49; found C, 70.43; H, 3.95; N 7.11.

2-Allyl-4-(3-chloro-4-methylphenyl)phthalazin-1(2H)-one (4).

A solution of **2** (0.01 mol), allyl bromide (0.03 mol), and potassium carbonate anhydrous (0.03 mol) in 50 ml dry acetone was heated under reflux for 24 h. The excess solvent was removed by distillation, and the reaction mixture was diluted with water. The solid product was obtained filtered off and crystallized from the ethanol to give **4**, 30 % yield, m.p. 110°C. EIMS (70 eV) *m/z* (%): M⁺, 310 (1), 268 (42). IR (KBr) ν : 1660, 2853 and 2923 cm⁻¹ attributable to C=O and CH. Anal calcd for C₁₈H₁₅N₂OCl: C, 69.45; H, 5.14; N, 9.03; found C, 68.31; H, 5.08; N, 8.92.

4-(3-Chloro-4-methylphenyl)-2-(2-hydroxyethyl)phthalazin-1(2H)-one (5).

A mixture of the phthalazinone **2** (0.01 mol), 2-chloroethanol (0.03 mol), and potassium carbonate anhydrous (0.03 mol) in 50 ml dry acetone was heated under reflux for 24 h. The excess solvent was removed by distillation and the reaction mixture was diluted with water filtered off and crystallized from the ethanol to give **5**, 50 % yield, m.p. 130°C. ¹H-NMR δ : 2.35 (Methyl group), 3.8 (t, CH₂ OH), 7.2 - 7.9 (2m, 7H, Ar-H), 8.37 (s, 1H, OH, exchangeable with D₂O). EIMS (70 eV) *m/z* (%): M⁺, 314 (2), 302 (2.2) 290 (35), 276 (100), 248 (40). IR

(KBr) ν : 1632 , 2852 , 2921 , and 3436 cm^{-1} attributable to C=O , CH and OH . Anal calcd for $\text{C}_{17}\text{H}_{15}\text{N}_2\text{O}_2\text{Cl}$: C, 64.96 ; H, 4.77 ; N, 8.91 ; found C , 64.52 ; H , 4.77 ; N , 8.67.

2-Aralkyl-4-(3-chloro-4-methylphenyl)phthalazin-1(2H)-one (6a-b).

A mixture of compound **5** (0.01 mol) and compounds containing active hydrogen namely , phenol and catechol , (0.015 mol) and 2 drops from conc. HCl in 50 ml ethanol was heated under reflux for 3h. The solid product was obtained after concentration and cooling was filtered off and crystallized from the ethanol to give **6(a-b)**. (**6b**), 60 % yield, m.p. 210°C. $^1\text{H-NMR}$ δ : 2.35 (methyl group) , 1.7 (t , 2H , CH_2) , 6.64-7.9 (3m , 11H , Ar-H) , 10.28 (s , 2H , OH , exchangeable with D_2O) . IR (KBr) ν : 1665 and 3400, 3430 cm^{-1} attributable to C=O and OH. Anal calcd for $\text{C}_{23}\text{H}_{19}\text{N}_2\text{O}_3\text{Cl}$: C , 67.98; H , 4.67 ; N , 6.89; found C ,67.42 ; H , 4.15; N, 6.56 . (**6a**), 70 % yield, m.p. 240°C. IR (KBr) ν 1665 and 3400 cm^{-1} attributable to C=O and OH. Anal calcd for $\text{C}_{23}\text{H}_{19}\text{N}_2\text{O}_2\text{Cl}$: C 70.76, H 4.87, N 7.17 ; found C 71.02, H 5.01, N 7.52 .

4-(2-(1-Oxo-4-(3-chloro-4-methylphenyl)phthalazin-2(2H)-yl)ethyl)-1,2-phenylene diacetate (7).

A solution of compound **6b** (0.01 mol) in acetic anhydride (15 ml) was heated under reflux for 1 h. The reaction mixture was diluted with water. The solid that obtained was crystallized from ethanol to give **7**, 45 % yield, m.p. 195°C. IR (KBr) ν : 1666 and 1761 cm^{-1} attributable to two C=O. Anal calcd for $\text{C}_{27}\text{H}_{23}\text{N}_2\text{O}_5\text{Cl}$:C ,66.12 ; H , 4.69 ; N , 5.71 ; found C , 65.46 ; H 4.31; N , 5.63 .

4-(3-Chloro-4-methylphenyl)-1-oxo-N-phenylphthalazine-2(1H)-carbothioamide (8).

A mixture of compound **2** (0.01 mol) and phenyl isothiocyanate (0.01 mol) in benzene (20 ml) was refluxed for 6h. after cooling , The solid that separated was filtered off and crystallized from the ethanol to give **8** , 20 % yield , m.p. 95°C. IR (KBr) ν :1156 , 1665 and 3155 cm^{-1} attributable to C=S , C=O and NH. EIMS (70 eV) m/z (%): M^+ , 403, (3), 402 (5) and 400 (2) Anal calcd for $\text{C}_{22}\text{H}_{16}\text{N}_3\text{OSCl}$: C , 65.18 ; H , 3.95 ; N , 10.37 ; found C , 64.66 ; H , 3.54 ; N , 10. .

2-(2-Aminoethyl)-4-(3-chloro-4-methylphenyl)phthalazin-1(2H)-one (9).

A mixture of compound **2** (0.01 mol) and ethanolamine (0.015 mol) was heated in oil bath at 150 °C for 2 h. The reaction mixture was diluted with water and the solid that

separated was filtered off and crystallized from the ethanol to give **9** , 90 % yield , m.p. 210 °C. IR (KBr) ν : 1655 , 3210 and 3478 cm^{-1} attributable to C=O and NH. Anal calcd for $\text{C}_{17}\text{H}_{16}\text{N}_3\text{O}_1\text{Cl}$: C , 64.96; H,5.09 ; N,13.37 ;found C ,64.12 ; H ,5.21; N ,12.98.

2-((Z)-2-((Benzof[d][1,3]dioxol-7-yl)methyleneamino)ethyl)-4-(3-chloro-4-methylphenyl)phthalazin-1(2H)-one (10).

A mixture of compound **9** (0.01 mol) , and piperanal (0.015 mol) in ethanol (30 ml) was heated under reflux for 3h. after cooling the solid that separated was filtered off and crystallized from the ethanol to give **10** , 40 % yield, m.p. 230°C. IR (KBr) ν : 1622 and 1675 cm^{-1} attributable to C=N and C=O. Anal calcd for $\text{C}_{25}\text{H}_{20}\text{N}_3\text{O}_3\text{Cl}$: C, 67.41 ; H, 4.49 ; N, 9.43 ; found C ,67.22 ; H ,4.49 ; N ,9.56

N-(2-(4-(3-Chloro-4-methylphenyl)-1-oxophthalazin-2(1H)-yl)ethyl)acetamide (11).

Acetic anhydride (0.002 mol) was added rapidly to a boiling solution of **9** (0.001 mol) in 20ml ethanol was heated under reflux for 2 hs, cooled and poured on cold water to give a solid which was separated and crystallized from benzene to give **11**, 50 % yield, m.p. 190°C. $^1\text{H-NMR}$ δ : 2.35 (methyl group), 1.3 (s, 3H, CH_3CO), 2.1 (s, 3H, CH_3), 7.2-7.8 (m, 7H, Ar-H) , 10.41 (s, 1H, NH, exchangeable with D_2O). IR (KBr) ν : 1666 and 3155 cm^{-1} attributable to C=O and NH. Anal calcd for $\text{C}_{19}\text{H}_{17}\text{N}_3\text{O}_2\text{Cl}$: C, 64.40; H, 4.80; N, 11.86; found C, 64.22; H, 4.62; N, 10.34.

1-(2-(4-(3-Chloro-4-methylphenyl)-1-oxophthalazin-2(1H)-yl)ethyl)-3-phenylthiourea (12)

A mixture of compound **9** (0.01 mol) and phenyl isothiocyanate(0.015 mol) in dry benzene (30 ml) was refluxed for 5h. After cooling , the solid that separated was filtered off and crystallized from the ethanol to give **12** , 40 % yield, m.p. 230 °C. The $^1\text{H-NMR}$ δ : 2.35 (methyl group) , 1.42 (t, 2H , CH_2) , 7.2-8.5 (3m, 12H, Ar-H) , 10.49 (s, 2H, 2 NH, exchangeable with D_2O). IR (KBr) ν : 1156, 1667 and 3443 cm^{-1} attributable to C=S, C=O and NH. Anal calcd for $\text{C}_{24}\text{H}_{21}\text{N}_4\text{OSCl}$: C, 64.28 ; H ,4.68 ; N ,12.5 ; found C, 63.88 ; H, 4.19 ; N, 11.92 .

3-(2-(4-(3-Chloro-4-methylphenyl)-1-oxophthalazin-2(1H)-yl)ethyl)-dihydro-1-phenyl-2 thioxopyrimidine-4,6(1H,5H)-dione (13).

A mixture of compound **12** (0.01 mol), and diethyl malonate (0.015 mol) in ethanol (30 ml) was heated under reflux for 3h. After cooling the solid that separated was filtered off and crystallized from the ethanol to give **13**, 50 % yield, m.p. 260°C. IR (KBr) ν : 1156 and 1667 cm^{-1} attributable to C=S and C=O. Anal calcd for $\text{C}_{27}\text{H}_{21}\text{N}_4\text{O}_3\text{S}$: C, 62.79; H, 4.06; N, 10.85; found C, 62.31; H, 3.87; N, 10.15.

4-(3-Chloro-4-methylphenyl)-2-(oxiran-2-yl)methylphthalazin-1(2H)-one (14).

A mixture of the phthalazinone **2** (0.01mol), epichlorohydrin (0.03 mol) and potassium carbonate anhydrous (0.03 mol) in dry acetone (50 ml) was heated under reflux for 24 h. The excess solvent was removed by distillation and the solid that separated after dilution with water was filtered off and crystallized from the ethanol to give **14**, 50% yield as colorless crystals mp 130-135 °C. $^1\text{H-NMR}$ (DMSO-d_6 , 300 MHz) δ : 2.35 (methyl group), 7.2-7.8 (m, 7H, Ar-H). IR (KBr) ν : 1653(C=O), 1579 (C=N), 1134 (C-O) cm^{-1} ; EIMS (70 eV) m/z (%): M^+ , 316 (10), 305 (21), 238 (88). Anal calcd for $\text{C}_{18}\text{H}_{15}\text{N}_2\text{O}_2\text{Cl}$: C, 66.25; H, 4.60; N, 8.58; found C, 65.87; H, 4.09; N, 8.17.

4-(3-chloro-4-methylphenyl)-2-(3-hydrazinyl-2-hydroxypropyl)phthalazin-1(2H)-one (15).

A solution of compound **14** (0.01 mol), and hydrazine hydrate (0.015 mol) in ethanol (30 ml) was heated under reflux for 3h. The solid that obtained after cooling was filtered off and crystallized from the ethanol to give **15**, 30% yield as colorless crystals m.p. 85°C. $^1\text{H-NMR}$ (DMSO-d_6 , 300 MHz) δ : 2.35 (methyl group), 7.2-7.8(2m, 8H, Ar-H), 8.5 (s, 2H, NH_2 , exchangeable with D_2O), 10.4(s, 1H, NH, exchangeable with D_2O). IR (KBr) ν : 3376 (OH), 2927, 2857 (NH), 1669,1647(C=O), 1574 (C=N) cm^{-1} . Anal calcd for $\text{C}_{18}\text{H}_{19}\text{N}_4\text{O}_2\text{Cl}$: C, 60.33; H, 5.30; N, 15.64; found C, 60.21; H, 5.04; N, 15.20.

4-(2-hydroxy-3-(3-chloro-4-methylphenyl)phthalazin-2(1H)-yl)propylamino benzoic acid (16). A solution of compound **14** (0.01 mol), and 4-amino benzoic acid (0.015 mol) in ethanol (30 ml) was heated under reflux for 3h. The solid that obtained after cooling was filtered off and crystallized from the ethanol to give **16**, 40% yield as colorless crystals m.p. 85 °C. IR (KBr) ν : 3459 (OH), 3360 (NH), 1670, 1629 (C=O), 15798 (C=N), 1158 (C-O) cm^{-1} EIMS (70 eV) m/z (%): M^+ , 462 Anal calcd for $\text{C}_{25}\text{H}_{22}\text{N}_3\text{O}_4\text{Cl}$: C, 64.79; H, 4.75; N, 9.07; found C, 64.61; H, 4.32; N, 9.11.

2-(3-(2-aminophenylamino)-2-hydroxypropyl)-4-(3-chloro-4-methylphenyl)phthalazin-1(2H)-one (17).

A solution of compound **14** (0.01 mol), and o-phenylene diamine (0.015 mol) in ethanol (30 ml) was heated under reflux for 3h. The solid that obtained after cooling was filtered off and crystallized from the ethanol to give **17**, 30% yield as colorless crystals m.p 78°C. $^1\text{H-NMR}$ (DMSO-d_6 , 300 MHz) δ : 2.35 (methyl group), 3.27 (s, 2H, NH_2 exchangeable with D_2O), 4.29 (s, methine proton), 6.3-7.26 (m, 11H, Ar-H), 7.45(s, 1H,NH), 7.9 (s, 1H, OH, exchangeable with D_2O). IR (KBr) ν : 3384, 3362 (NH_2), 3287 (OH), 3280 (NH), 1633 (C=O), 1589 (C=N) cm^{-1} . EIMS (70 eV) m/z (%): M^+ , 433 (13), 295 (16), 106 (21). Anal calcd for $\text{C}_{24}\text{H}_{23}\text{N}_4\text{O}_2\text{Cl}$: C, 66.35; H, 5.29; N, 12.90; found C, 66.51; H, 5.12; N, 12.30.

2[3-(2-arylidione amine phenylamino)-2-hydroxy]n-propyl-4-(3-chloro-4-methylphenyl)phthalazin-1(2H)-one (18a-b).

A solution of compound **18** (0.01 mol), and appropriate aldehyde namely, 2-hydroxybenzaldehyde and 1-naphthaldehyde in ethanol (30 ml) was heated under reflux for 3h. The solid that obtained after cooling was filtered off and crystallized from the ethanol to give **18(a-b)**. IR spectrum of compound **18b** showed strong absorption bands at 1650 ν C=N, 1687 ν C=O, 3052 ν NH, and 3400 ν OH

4-(3-chloro-4-methylphenyl)-2-(piperidin-1-yl)methylphthalazin-1(2H)-one (19).

A solution of compound **2** (0.01mol), piperidine (0.02 mol) formaldehyde (1ml), and conc.HCl (4 drops) in ethanol (30 ml) was heated under reflux for 3h. The solid that obtained after cooling was filtered off and crystallized from the ethanol to give **19**, 75 % yield, m.p.205 °C. EIMS (70 eV) m/z (%): M^+ , 366 (13), 270 (73) and 242 (20), 224 (17), 155 (100). IR (KBr) ν : 1654, 1583, 2929 cm^{-1} attributable to C=O, C=N and CH. Anal calcd for $\text{C}_{21}\text{H}_{22}\text{N}_3\text{OCl}$: C, 68.66; H, 5.99; N 11.44; found C, 67.17; H, 5.20; N, 11.11.

4-(3-chloro-4-methylphenyl)phthalazine-1(2H)-thione (20).

A solution of compound **2** (0.01 mol), and P_2S_5 (0.03 mol) in dry xylene (30ml), was heated under reflux for 1h. The solution was filtered with hot and the solid that obtained after cooling was filtered off and crystallized from the

ethanol to give the thione **20**, 40 % yield, m.p. 170°C. EIMS (70 eV) m/z (%): M^+ , M^++1 and M^++2 , 285 (100), 283 (20) and 280 (6) corresponding to M^+ . IR (KBr) ν : 1205, 1428, 2855, 2932 and 3147 cm^{-1} attributable to C=S, CH and NH. Anal calcd for $\text{C}_{15}\text{H}_{11}\text{N}_2\text{S}$: C, 62.93; H, 3.84; N, 9.79; found C, 61.56; H, 3.01; N, 9.11.

1-hydrazinyl-4-(3-chloro-4-methylphenyl)-1,2-dihydrophthalazine (21).

A solution of thione **20** (0.01 mol), and hydrazine hydrate (0.015 mol) in ethanol (30 ml) was heated under reflux for 3h. The solid that obtained after cooling was filtered off and crystallized from the ethanol to give **21**, 30 % yield, m.p. 210°C. Anal calcd for $\text{C}_{15}\text{H}_{14}\text{N}_4\text{Cl}$: C, 63.15; H, 4.91; N, 19.64; found C, 61.04; H, 5.12; N, 18.31.

(19E)-2-(4-methoxybenzylidene)-1-(4-(3-chloro-4-methylphenyl)-1,2-dihydrophthalazin-1-yl)hydrazine (22)

:

A solution of **21** (0.01 mol), and 4-methoxy benzaldehyde (0.015 mol) in ethanol (30 ml) was heated under reflux for 3h. The solid that obtained after cooling was filtered off and crystallized from the ethanol to give **22**, 30 % yield, m.p. 140°C. IR (KBr) ν : 1601, 1508 cm^{-1} , attributed to C=N. Anal calcd for $\text{C}_{23}\text{H}_{20}\text{N}_4\text{OCl}$: C, 68.48; H, 4.96; N, 13.89; found C, 66.92; H, 4.22; N, 13.01.

Ethyl-4-((1-oxo-4-(3-chloro-4-methylphenyl)phthalazine-2(1H)-yl)methyl)piperazine-1-carboxylate (23).

A mixture of **2** (0.01 mol), N-ethoxycarbonyl piperazine (0.015 mol), formaldehyde (2ml) and few drops from conc. HCl in absolute ethanol (30 ml) was heated under reflux for 3h. The solid that obtained after cooling was filtered off and crystallized from the ethanol to give **23**, 22 % yield, m.p. 160°C. $^1\text{H-NMR}$ δ : 1.3 (t, 3H, CH_2CH_3), 1.8 (s, 4H, β -methylene protons), 2.9 (s, 4H, α -methylene protons), 4.1 (q, 2H, CH_2CH_3), 4.2 (m, 8H of piperazine moiety), 5.7 (s, 1H, NCH₂N), 7.2-7.8 (m, 8H, Ar-H). IR (KBr) ν : 1653, 1702, 2859 and 2927 cm^{-1} attributable to two C=O, and CH. Anal calcd for $\text{C}_{23}\text{H}_{12}\text{N}_4\text{O}_3\text{Cl}$: C, 74.98; H, 6.29; N, 8.74; found C, 75.48; H, 5.90; N, 8.21.

ethyl 2-(4-(3-chloro-4-methylphenyl)-1-oxophthalazin-2(1H)-yl)acetate (24)

A mixture of **2** (0.01 mol), ethyl chloroacetate (0.03 mol) and (0.03 mol) potassium carbonate in 30 ml dry acetone was refluxed for 24 h. cooled at room temperature and

poured into water the obtained solid was filtered off and crystallized from ethanol to give **(24)**, 50 % yield, m.p. 230°C. $^1\text{H-NMR}$ δ : 1.3 (t, 3H, CH_2CH_3), 1.8 (s, 4H, β -methylene protons), 2.9 (s, 4H, α -methylene protons), 4.1 (q, 2H, CH_2CH_3), 7.2-7.8 (m, 8H, Ar-H). IR (KBr) ν : 1750, 1650 cm^{-1} attributable to two C=O, and CH. Anal calcd for $\text{C}_{19}\text{H}_{17}\text{N}_2\text{O}_3\text{Cl}$: C, 64.04; H, 4.77; N, 7.86; found C, 63.21; H, 3.12; N, 6.17.

2-(4-(3-chloro-4-methylphenyl)-1-oxophthalazin-2(1H)-yl)acetohydrazide(25)

A mixture of **24** (0.01 mol), hydrazine hydrate (0.03 mol) in 30 ml ethanol was refluxed for 3 h. the resultant solid was filtered off and crystallized from ethanol to give **(25)**, 70 % yield, m.p. 210°C. $^1\text{H-NMR}$ 2.35 (methyl group), 7.2-7.8 (2m, 8H, Ar-H), 7.2 (s, 2H, NH₂, exchangeable with D₂O), 10.4 (s, 1H, NH, exchangeable with D₂O), 7.2-7.8 (m, 8H, Ar-H). IR (KBr) ν : 1750, 1650 cm^{-1} attributable to two C=O, and CH. Anal calcd for $\text{C}_{17}\text{H}_{15}\text{N}_2\text{O}_3\text{Cl}$: C, 59.64; H, 4.38; N, 16.37; found C, 61.11; H, 4.89; N, 16.77.

(2E)-ethyl 3-amino-3-(4-(3-chloro-4-methylphenyl)-1-oxophthalazin-2(1H)-yl)acrylate(26)

A mixture of **2** (0.01 mol), ethyl cyanoacetate (0.03 mol) in 30 ml ethanol was refluxed for 3 h. the resultant solid was filtered off and crystallized from ethanol to give **(26)**, 80 % yield, m.p. 225°C. $^1\text{H-NMR}$ 2.35 (methyl group), 3.7 (H, α -methylene protons), 7.2-7.8 (2m, 8H, Ar-H), 7.2 (s, 2H, NH₂, exchangeable with D₂O), 7.2-7.8 (m, 8H, Ar-H). IR (KBr) ν : 1750, 1650 cm^{-1} attributable to two C=O, and CH. Anal calcd for $\text{C}_{20}\text{H}_{17}\text{N}_3\text{O}_3\text{Cl}$: C, 62.82; H, 4.45; N, 10.99; found C, 60.91; H, 3.39; N, 10.01.

(2E)-3-amino-3-(4-(3-chloro-4-methylphenyl)-1-oxophthalazin-2(1H)-yl)-N'-phenylacrylohydrazide (27)

A mixture of **26** (0.01 mol), phenylhydrazine (0.03 mol) in 30 ml ethanol was refluxed for 3 h. the resultant solid was filtered off and crystallized from ethanol to give **(27)**, 80 % yield, m.p. 240°C. IR (KBr) ν : 1750, 1650 cm^{-1} attributable to two C=O, and CH. Anal calcd for $\text{C}_{24}\text{H}_{20}\text{N}_5\text{O}_2\text{Cl}$: C, 64.71; H, 4.49; N, 15.73; found C, 63.52; H, 3.89; N, 14.61.

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