RESEARCH ARTICLE

An Efficient Synthesis of Biologically Active Tetrachloroquinazolin-2,4-dione

Chemical Sciences Journal, Vol. 2012: CSJ-87

An Efficient Synthesis of Biologically Active Tetrachloroquinazolin-2,4-dione

MA Hassan[1], AMM Younes[2], MM Taha[2], WM Salem[3], AH Abdel-Monsef[2*]
[1]Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Sinai University, Arish, Egypt.
[2]Department of Chemistry, Faculty of Science, South Valley University, Qena, Egypt.
[3]Department of Botany, Faculty of Science, South Valley University, Qena, Egypt.

***Correspondence:** bakooos2004@yahoo.com

Accepted: Dec 25, 2012; Published: Dec 31, 2012

Abstract
Pyrimidine is a prominent member of the diazine family of heterocyclics. Pyrimidine compounds have been explored for use as histamine and adenosine receptor antagonists as well as among several other biological receptors and modulators. The aim of the current research work was to synthesize a new set of tetrachloroquinazolin-2,4-dione derivatives by treatment of *N*-phenylsulphonyloxytetrachlorophthalimide with some primary aliphatic and aromatic amines via Lossen Rearrangement. The structures of the synthesized compounds were characterized using physical and spectral data such as IR, ^{1}H NMR and mass spectral studies. The newly synthesized compounds were evaluated for their preliminary in vitro antibacterial activity towards *Salmonella typhi, Staphylococcus aureus* and *Bacillus cereus*. This study leads us to conclude that quinazolinediones have interesting biological and pharmacological properties towards bacteria.

Keywords: 3-Alkyl(1H,3H)tetrachloroquinazolin-2,4-diones; 3-benzyl(1H,3H)tetrachloroquinazolin-2,4-dione; p-anisidine; pyrimidine ring; antibacterial.

1. Introduction

The quinazolinedione template occurs in a large number of bioactive molecules including serotonergic, dopaminergic and adrenergic receptor ligands and inhibitors of aldose reductase, lipoxygenase, cyclooxygenase, collagenase and carbonic anhydrase [1]. Furthermore, quinazolinediones have demonstrated utility in a diverse range of medical and biological applications. Pyrimidine and condensed pyrimidines have received much attention over the years because of their interesting biological and pharmacological properties as sedatives [2], antibacterials [3–9], antimalarial [2], analgesic [2, 4, 8], anti-inflammatory [2, 3, 8], anticonvulsant [9], antipyretic [5], antiparasitic [5, 7], antifungal [6, 10, 11], antitoxic [12], antiviral [9, 12, 13], anticancers [14–17] ad DNA-binding activities [18]. In addition, pyrimidine and quinazolinediones have been found as a key component of many biologically active and pharmaceutical compounds. For this, our strategy was to prepare the targeted 3-substituted tetrachloroquinazoline from *N*-phenylsulphonyloxytetrachlorophthalimide.

2. Methods

2.1. Instrumentation

The main experimental challenge appears to be in fine tuning of experimental parameters (solvent, ratio of reagents, catalysis, etc.) so that pure crystalline material can be isolated. ^{1}H NMR (200 MHz) spectra were recorded on a Varian EM 390 spectrometer. Chemical shift values were recorded in δ units (ppm) relative to tetramethylsilane (TMS) as internal standard. Melting points were determined using an electric melting point apparatus (Kofler). Infrared spectra (IR) were recorded using KBr pellets on a Shimadzu 408 spectrometer. Electron impact mass spectra were obtained at 70 eV using a GCMS sp.1000 Shimadzu. Thin layer chromatography (TLC) was performed on silica gel 60 PF254 plates or aluminum oxide plates obtained from Merck. Elemental analyses were carried out at a Microanalysis Unit at Cairo University, Egypt.

2.2. Chemical procedure

2.2.1. General procedure for the preparation of substituted (alkyl, cyclohexyl and benzyl) tetrachloroquinayplin-2,4-dione (2a–f)

A mixture of *N*-phenylsulphonyloxytetrachlorophthalimide (**1**) (0.44 gm, 1 mmol) and aliphatic amines, namely, methylamine, *n*-propylamine, isobutylamine and *n*-butylamine, cyclohexylamine and benzylamine (2 mmol) in the presence of anhydrous sodium acetate as a basic catalyst (0.12 gm, 1.5 mmol) in glacial acetic acid (20 ml) was refluxed for 6–12 h. After cooling, the reaction mixture was poured on ice water, where a white solid product was formed which filtered off and crystallized from appropriate solvent to give 3-alkyl(1H,3H)tetrachloroquinazolin-2,4-diones (**2a–f**) as white crystals (Scheme 2).

2.2.2. General procedure for the preparation of substituted (aryl)tetrachloroquinayplin-2,4-dione (3a–h)

To a solution of *N*-phenylsulphonyloxytetrachlorophthalimide (**1**) (0.44 gm, 1 mmol), anhydrous sodium acetate (0.12 gm, 1.5 mmol) and glacial acetic acid (20 ml), aromatic amines (1.2 mmol), namely, aniline, p-toulidine, p-aminophenol, p-anisidine, p-chloroaniline, p-aminoacetophenone, p-aminobenzoic acid and p-nitroaniline were added; the reaction mixture was refluxed for 8 h. The reaction mixture was poured into water, and then the mixture was allowed to stand at room temperature overnight. The collected solid was filtered off and crystallized from the appropriate solvent to give 3-aryl(1H,3H)tetrachloroquinazolin-2,4-diones (**3a–h**).

2.3. Characterization data

2.3.1. 3-Methyl(1H,3H)tetrachloroquinazolin-2,4-dione (2a)

White crystal (0.28 gm, 72%), m.p. 280–282°C. FT-IR (KBr, cm^{-1}): 3200 (ν NH), 1740, 1660 (ν C=O's). ^{1}H NMR spectrum: (300 MHz, DMSO-d6): 2.5 (s, 3H, CH_3); 11.1 (s, 1H, NH). MS (*m*/*z*, %): 312 (38.05%) (M^+) in addition to the characteristic peaks for compounds containing four chlorine atoms [19] at *m*/*z* = 314 (49.18%) (M+2), 316 (23.29%) (M+4) and 318 (4.88%) (M+6). The main fragment ions for compound **2a** gives an excellent confirmation for the proposed structures, as shown in Scheme 1. Anal. calcd. for $C_9H_4Cl_4N_2O_2$: C, 34.43%; H, 1.28%; N, 8.92%. Found: C, 34.61%; H, 1.27%; N, 9.14%.

2.3.2. 3-Propyl(1H,3H)tetrachloroquinazolin-2,4-dione (2b)

White crystal (0.35 gm, 67%), m.p. 250–252°C. FT-IR (KBr, cm^{-1}): 3200 (ν NH), 1740, 1660 (ν C=O's).^{1}H NMR spectrum: (300 MHz, DMSO-d6): 0.9 (t, 3H, CH_3); 1.6 (m, 2H, CH_2); 3.8 (t, 2H, CH_2); 11.1 (s, 1H, NH). MS (*m*/*z*, %): 340 (22.06%) (M^+) in addition to the characteristic peaks for compounds containing four chlorine atoms at (M+2), (M+4) and (M+6) (Scheme 4). Anal. calcd. for $C_{11}H_8Cl_4N_2O_2$: C, 38.64%; H, 2.36%; N, 8.19%. Found: C, 38.92%; H, 2.36%; N, 8.20%.

2.3.3. 3-Isobutyl(1H,3H)tetrachloroquinazolin-2,4-dione (2c)

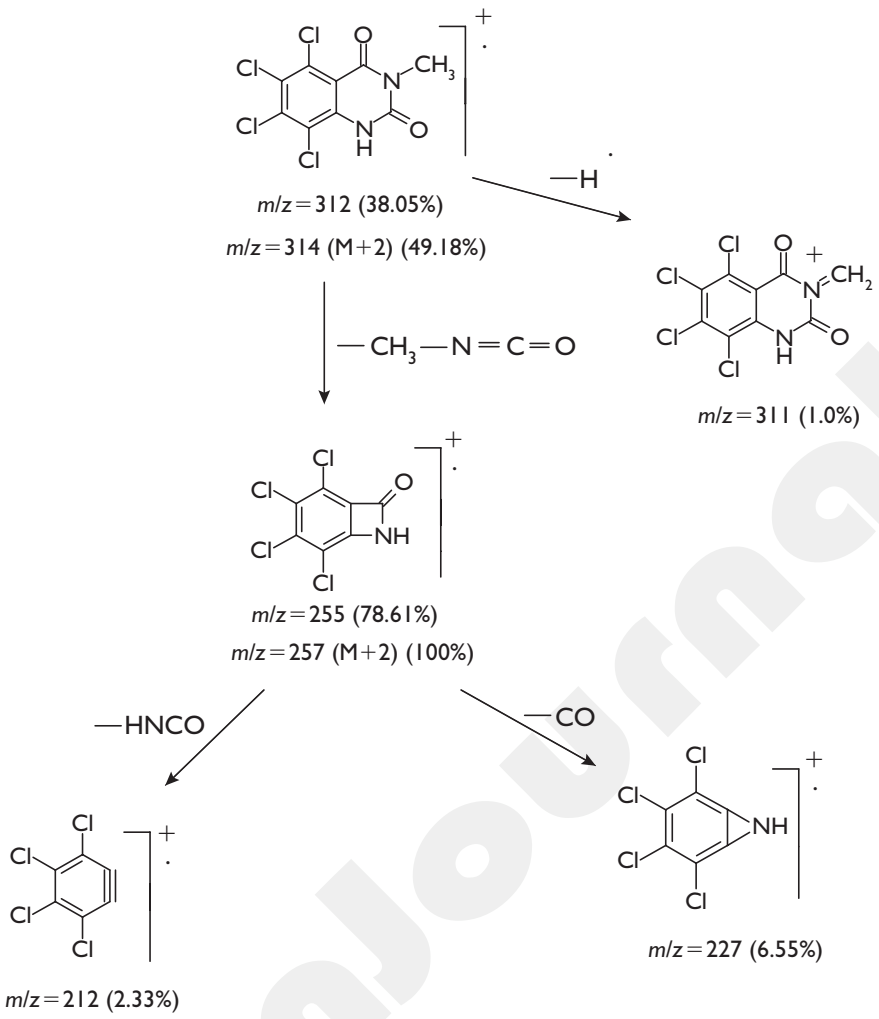
White crystal (0.41 gm, 83%), m.p. 280–282°C. FT-IR (KBr, cm^{-1}): 3206 (ν NH), 1726, 1669 (ν C=O's). ^{1}H NMR spectrum: (300 MHz, DMSO-d6): 0.9 (d, 6H, $2CH_3$); 2.0 (m, 1H, CH); 3.7 (d, 2H, CH_2); 11.1 (s, 1H, NH). Anal. calcd. for $C_{12}H_{10}Cl_4N_2O_2$: C, 40.49%; H, 2.83%; N, 7.87%. Found: C, 40.55%; H, 2.72%; N, 7.89%.

2.3.4. 3-Butyl(1H,3H)tetrachloroquinazolin-2,4-dione (2d)

White crystal (0.34 gm, 78%), m.p. 280–282°C. FT-IR (KBr, cm^{-1}): 3201 (ν NH), 2960 (ν CH aliph.), 1726, 1660 (ν C=O's). ^{1}H NMR spectrum: (300 MHz, DMSO-d6): 0.9 (t, 3H, CH_3); 1.2 (m, 2H, CH_2); 1.5 (m, 2H, CH_2); 3.8 (t, 2H, CH_2); 11.1 (s, 1H, NH). MS (*m*/*z*, %): 354 (M^+) in addition to the characteristic peaks for compounds containing four chlorine atoms at (M+2), (M+4) and (M+6). Anal. calcd. for $C_{12}H_{10}Cl_4N_2O_2$: C, 40.49%; H, 2.83%; N, 7.87%. Found: C, 40.22%; H, 2.91%; N, 7.69%.

2.3.5. 3-Cyclohexyl(1H,3H)tetrachloroquinazolin-2,4-dione (2e)

White crystal (0.16 gm, 53%), m.p. 306–308°C. FT-IR (KBr, cm^{-1}): 3201 (NH), 2924, 2852 (CH aliph.), 1740, 1659 (C=O's). ^{1}H NMR spectrum: (200 MHz, DMSO-d6): 1.0–2.5 (m, 10H, $5CH_2$); 4.61 (m, 1H, N−CH); 11.1 (s, 1H, NH). MS (*m*/*z*, %): 380 (1.26%) (M^+) in addition to the characteristic peaks for compounds containing four chlorine atoms at (M+2), (M+4) and (M+6). Anal. calcd. for $C_{14}H_{12}Cl_4N_2O_2$: C, 44.04%; H, 3.16%; N, 7.33%. Found: C, 44.20%; H, 3.17%; N, 7.41%.



Scheme 1: The main fragmentations for compound **2a**.

2.3.6. 3-Benzyl(1H,3H)tetrachloroquinazolin-2,4-dione (2f)
White crystal (0.24 gm, 76%), m.p. 262–263°C. FT-IR (KBr, cm^{-1}): 3211 (NH), 3032 (CH arom.), 1721, 1658 (C=O's).^{1}H NMR spectrum: (200 MHz, DMSO-d6): 7.2–7.35 (m, 5H, arom.); 5.1 (s, 2H, CH_2); 11.1 (s, 1H, NH). MS (*m/z*, %): 388 (9.43%) (M^+) in addition to the characteristic peaks for compounds containing four chlorine atoms at (M+2), (M+4) and (M+6).Anal. calcd. for $C_{15}H_8Cl_4N_2O_2$: C, 46.20%; H, 2.06%; N, 7.18%. Found: C, 46.32%; H, 2.07%; N, 7.26%.

2.3.7. 3-Phenyl(1H,3H)tetrachloroquinazolin-2,4-dione (3a)
White crystal (0.22 gm, 68%), m.p. 328–329°C. ^{1}H NMR spectrum: (300 MHz, DMSO-d6): 7.3–7.5 (m, 5H, arom.); 11.3 (s, 1H, NH). MS (*m/z*, %): 374 (M^+) in addition to the characteristic peaks for compounds containing four chlorine atoms at (M+2), (M+4) and (M+6). Anal. calcd. for $C_{14}H_6Cl_4N_2O_2$: C, 44.73%; H, 1.60%; N, 7.45%. Found: C, 44.94%; H, 1.61%; N, 7.56%.

2.3.8. 3-Tolyl(1H,3H)tetrachloroquinazolin-2,4-dione (3b)
White crystal (0.33 gm, 69%), m.p. 325–327°C. FT-IR (KBr, cm^{-1}): 3200 (ν NH), 1740, 1660 (ν C=O's).^{1}H NMR spectrum: (300 MHz, DMSO-d6): 2.2 (s, 3H, CH_3); 7.1–7.3 (2d, 4H,A_2B_2 arom.); coupling constant for aromatic protons = 0.0005 Hz,11.2 (s, 1H, NH). MS (*m/z*, %): 388 (M^+) in addition to the characteristic peaks for compounds containing four chlorine atoms at (M+2), (M+4) and (M+6). Anal. calcd. for $C_{15}H_8Cl_4N_2O_2$: C, 46.20%; H, 2.06%; N, 7.18%. Found: C, 46.41%; H, 2.07%; N, 7.21%.

2.3.9. 3-Hydroxyphenyl(1H,3H)tetrachloroquinazolin-2,4-dione (3c)
Grey crystal (0.45 gm, 81%), m.p. 308–309°C. FT-IR (KBr, cm^{-1}): 3472 (ν OH), 3200 (ν NH), 1726, 1664 (ν C=O's). ^{1}H NMR spectrum: (300 MHz, DMSO-d6): 6.8–7.1 (2d, 4H, A_2B_2 arom.); 9.65 (s, 1H, OH); coupling constant for aromatic protons = 0.0008 Hz, 11.2 (s, 1H, NH). MS (*m*/*z*, %): 390 (M^+) in addition to the characteristic peaks for compounds containing four chlorine atoms at (M+2), (M+4) and (M+6). Anal. calcd. for $C_{14}H_6Cl_4N_2O_3$: C, 42.90%; H, 1.54%; N, 7.14%. Found: C, 43.02%; H, 1.55%; N, 7.17%.

2.3.10. 3-Anisidyl(1H,3 H)tetrachloroquinazolin-2,4-dione (3d)
Buff crystal (0.49 gm, 83%), m.p. 312–314°C. FT-IR (KBr, cm^{-1}): 3196 (ν NH), 3001 (ν CH arom.), 2227 (ν CH aliph.), 1746, 1669 (ν C=O's). ^{1}H NMR spectrum: (300 MHz, DMSO-d6): 3.8 (s, 3H, OCH_3); 7.0–7.24 (2d, 4H, A_2B_2 arom.); coupling constant for aromatic protons = 0.00077 Hz, 11.2 (s, 1H, NH). MS (*m*/*z*, %): 404 (M^+) in addition to the characteristic peaks for compounds containing four chlorine atoms at (M+2), (M+4) and (M+6). Anal. calcd. for $C_{15}H_8Cl_4N_2O_3$: C, 44.37%; H, 1.99%; N, 6.90%. Found: C, 44.62%; H, 2.01%; N, 7.00%.

2.3.11. 3-Chlorophenyl(1H,3H)tetrachloroquinazolin-2,4-dione (3e)
White crystal (0.55 gm, 67%), m.p. 320–322°C. ^{1}H NMR spectrum: (300 MHz, DMSO-d6): 7.3–7.6 (2d, 4H, A_2B_2 arom.); coupling constant for aromatic protons = 0.00088 Hz, 11.3 (s, 1H, NH). MS (*m*/*z*, %): 408 (M^+) in addition to the characteristic peaks for compounds containing four chlorine atoms at (M+2), (M+4) and (M+6). Anal. calcd. for $C_{14}H_5Cl_5N_2O_2$: C, 40.98%; H, 1.23%; N, 6.82%. Found: C, 41.16%; H, 1.24%; N, 6.95%.

2.3.12. 3-Acetophenyl(1H,3H)tetrachloroquinazolin-2,4-dione (3f)
Yellow crystal (0.3 gm, 65%), m.p. 210–212°C. FT-IR (KBr, cm^{-1}): 3231 (ν NH), 3057 (ν CH arom.), 2919 (ν CH aliph.), 1740, 1660 (ν C=O's). ^{1}H NMR spectrum: (200 MHz, DMSO-d6): 2.6 (s, 3H, CH_3); 7.5–8.1 (2d, 4H, A_2B_2 arom.); coupling constant for aromatic protons = 0.0031 Hz, 11.4 (s, 1H, NH). MS (*m*/*z*, %): 416 (M^+) in addition to the characteristic peaks for compounds containing four chlorine atoms at (M+2), (M+4) and (M+6). Anal. calcd. for $C_{16}H_8Cl_4N_2O_3$: C, 45.98%; H, 1.92%; N, 6.70%. Found: C, 46.35%; H, 1.94%; N, 6.92%.

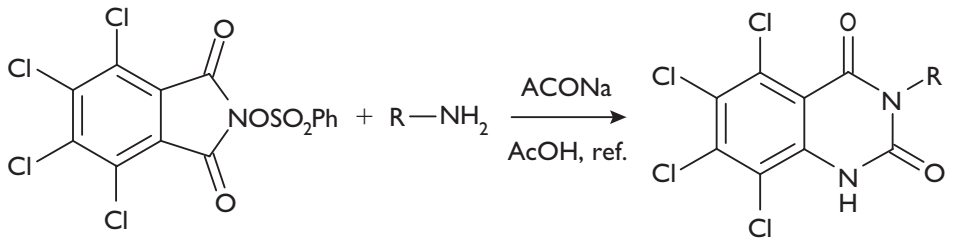
2.3.13. 3-Carboxyphenyl(1H,3H)tetrachloroquinazolin-2,4-dione (3g)
White crystal (0.29 gm, 72%), m.p. 318–319°C. FT-IR (KBr, cm^{-1}): 3395 (ν NH), highly bonded OH extending from 3267–2724, 1740, 1660 (ν C=O's). ^{1}H NMR spectrum: (200 MHz, DMSO-d6): 7.4–8.1 (2d, 4H, A_2B_2 arom.); coupling constant for aromatic protons = 0.0032 Hz, 11.4 (s, 1H, NH); 12.8 (broad s, 1H, COOH). MS (*m*/*z*, %): 418 (M^+) in addition to the characteristic peaks for compounds containing four chlorine atoms at (M+2), (M+4) and (M+6). Anal. calcd. for $C_{15}H_6Cl_4N_2O_4$: C, 42.89%; H, 1.44%; N, 6.67%. Found: C, 43.28%; H, 1.45%; N, 6.98%.

2.3.14. 3-Nitrophenyl(1H,3H)tetrachloroquinazolin-2,4-dione (3h)
Yellow crystal (0.34 gm, 77%), m.p. 304–306°C. ^{1}H NMR spectrum: (300 MHz, DMSO-d6): 7.6–8.4 (2d, 4H, A_2B_2 arom.); coupling constant for aromatic protons = 0.0024 Hz, 11.4 (s, 1H, NH). Anal. calcd. for $C_{14}H_5Cl_4N_3O_4$: C, 39.95%; H, 1.19%; N, 9.91%. Found: C, 40.03%; H, 1.18%; N, 10.0%.

2.3.15. Antibacterial studies
Antibacterial activity was determined against the above-mentioned bacteria using the paper disk assay method [20] with Whatman No. 1 filter paper disk of diameter 6 mm, which was sterilized by autoclaving for 15 min at 121°C. The sterile disks were impregnated with different tested compounds (50 mg/ml). Agar plates were surface inoculated uniformly from the broth culture of the tested microorganisms. In all cases, the concentration was approximately 1.2×10^8 CFU/ml [21]. The impregnated disks were placed on the Muller Hinton medium suitably spaced apart, and the plates were incubated at 37°C for 24 h. Dimethylformamide (DMF) was used as negative control, while commercial antibiotic disks (tetracycline, 30 mg/disk) were used as a positive control. The diameter of the growth inhibition halos caused by different compounds tested was measured by a ruler and expressed in millimeter. All the assays were carried out in triplicate.

3. Results and Discussion

3.1. Synthesis
The synthesis of the compounds **2a–f** and **3a–h** resulted from three steps [22], sequence starting from tetrachlorophthalic anhydride followed by *N*-hydroxytetrachlorophthalimide, then *N*-phenylsulphonyloxytetrachlorophthalimide (**1**).

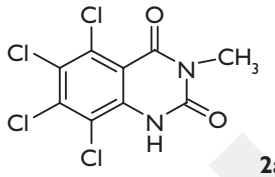
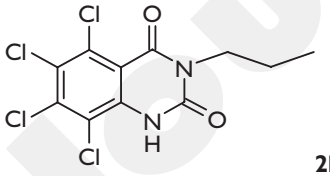
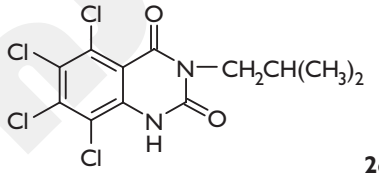
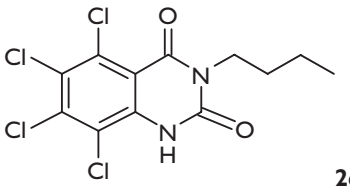
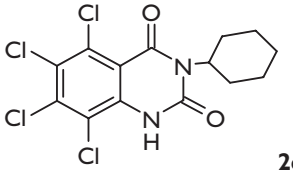
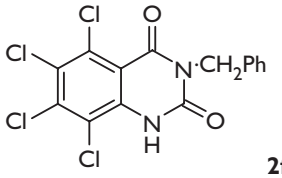


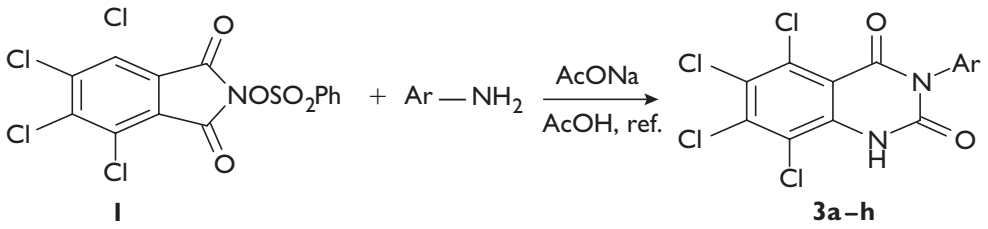
I **2a–f**

N-phenylsulphonyloxytetrachlorophthalimide 3-alkyl(1H,3H)tetrachloroquinazolin-2,4-diones

Scheme 2

Table 1: Preparation of substituted (alkyl, cyclohexyl and aralkyl)tetrachloroquinazolin-2,4-dione (**2a–f**).

Entry	Alkyl group	Product	Time (h)	Yield (%)
A	CH_3	 **2a**	6	72
B	$CH_3CH_2CH_2$	 **2b**	8	67
C	$(CH_3)_2CHCH_2$	 **2c**	9	83
D	$CH_3CH_2CH_2CH_2$	 **2d**	12	78
E	Cyclohexyl	 **2e**	8	53
F	$PhCH_2$	 **2f**	6	76



I — *N*-phenylsulphonyloxytetrachlorophthalimide

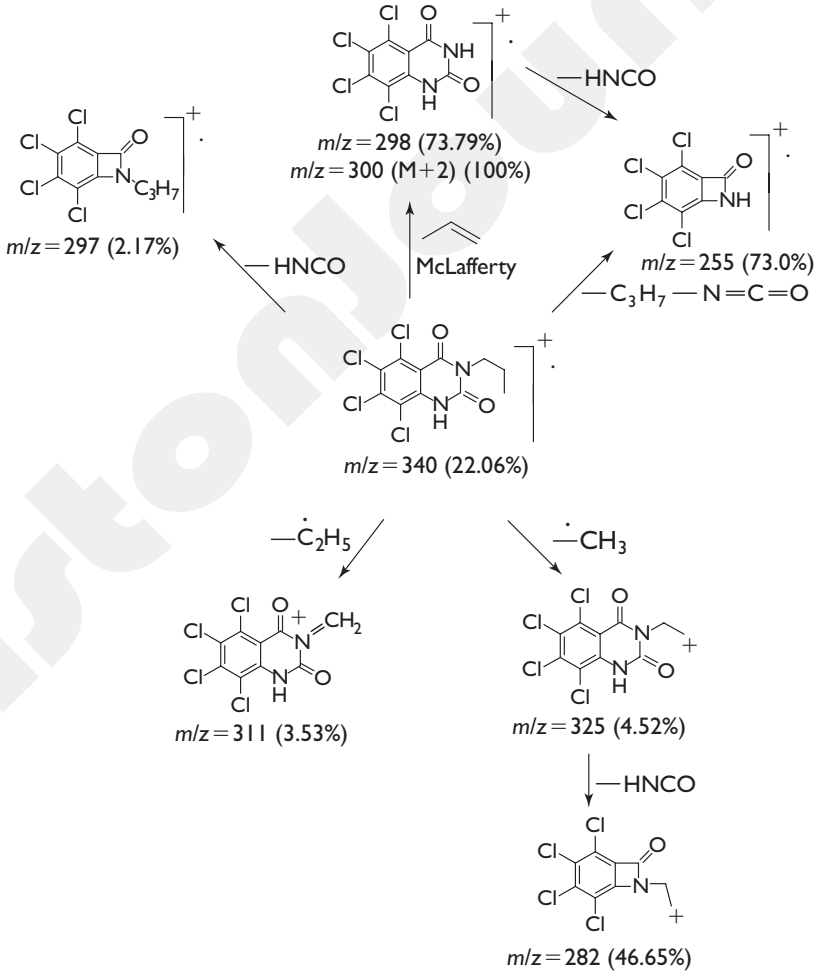
3a–h — 3-aryl(1H,3H)tetrachloroquinazolin-2,4-diones

Scheme 3

Scheme 2 outlines the synthetic pathway used to obtain compounds **2a–f**. The starting material *N*-phenylsulphonyloxytetrachlorophthalimide was prepared by allowing *N*-hydroxytetrachlorophthalimide to react with benzenesulfonyl chloride. Upon mixing of compound **I** with primary aliphatic amines, cyclohexylamine and benzylamine (Table 1) in acetic acid, 3-alkyl(1H,3H)tetrachloroquinazolin-2,4-diones **2a–f** were obtained in relatively good yields (Scheme 2).

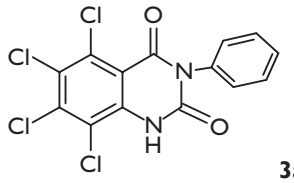
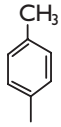
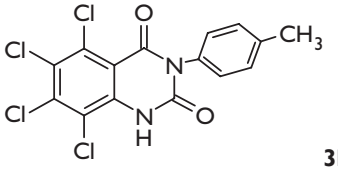
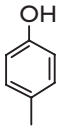
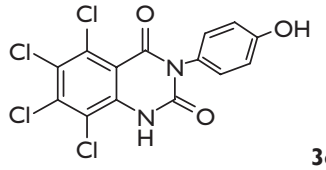
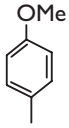
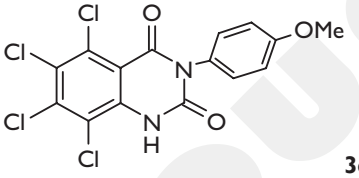
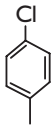
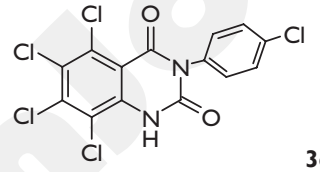
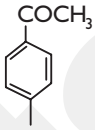
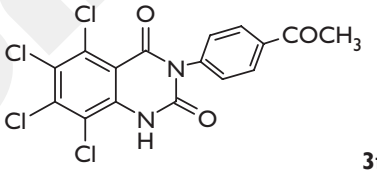
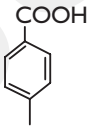
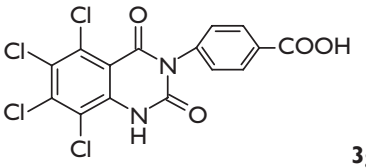
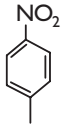
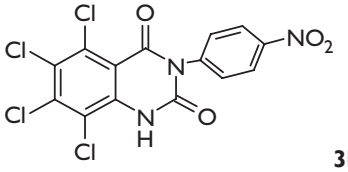
In conjugation with our current research with the action of amines on compound **I**, we study the action of primary aromatic amines, which have been found to be less basic than alkyl- and aralkylamines.

Scheme 3 outlines the synthetic pathway used to obtain compounds **3a–h**, which is prepared by the treatment of compound **I** with different aromatic amines, namely, aniline, p-toulidine, p-aminophenol, p-anisidine,



Scheme 4: The main fragmentations for compound **2b**.

Table 2: Preparation of substituted (aryl)tetrachloroquinazolin-2,4-dione (**3a–h**).

Entry	Aryl group	Product	Time (h)	Yield (%)
A		 **3a**	6	68
B		 **3b**	8	69
C		 **3c**	9	81
D		 **3d**	7	83
E		 **3e**	10	67
F		 **3f**	8	65
G		 **3g**	7	72
H		 **3h**	9	77

p-chloroaniline, p-aminoaceto-phenone, p-aminobenzoic acid and p-nitroaniline (Table 2) in the presence of anhydrous sodium acetate as a base catalyst (0.12 gm, 1.5 mmol) in glacial acetic acid (20 ml) and was refluxed for 6–10 h to (Scheme 3).

4. Biological Activity

Bacterial infection causes high rate of mortality in human population and aquaculture organisms [23]. For example, *B. cereus* is responsible for causing foodborne diseases [24]. *S. aureus* causes diseases such as mastitis, abortion and upper respiratory complications, while *Salmonella* sp. causes diarrhea and typhoid fever [25]. The revolutionized therapy of infectious diseases by the use of antimicrobial drugs has certain limitations because of changing patterns of resistance in pathogens and side effects they produced. These limitations demand for improved pharmacokinetic properties, which necessitate continued research for new antimicrobial compounds for the development of drugs [26]. So accordingly, pharmaceutical industries are giving importance to the compounds derived from quinazolinedione sources.

4.1. Bacterial source and culture conditions

The bacteria used in this study were *S. aureus*, *S. typhi* and *B. cereus* (obtained from the Pathology Department, Faculty of Veterinary Medicine, South Valley University). These bacterial strains were maintained on suitable medium at 4°C and subcultured on Mueller Hinton Broth at 37°C for 18 h before testing.

4.2. Antibacterial assays

Most of the tested compounds exhibited antibacterial activity against all the tested bacterial species. The gram-negative *S. typhi* was the most sensitive to most of the compounds tested (**2a–d** and **3f**). The higher antibacterial activity (indicated as zone of inhibition) was recorded for compound **3f** followed by **2d** and **2f** (11, 10 and 10 mm), respectively (Figure 1). Hence, the susceptibility of the gram-positive *B. cereus* to compounds **2d**, **2f**, **3a**, **3e–g** was more pronounced when compared to the other tested compounds. The most observed antibacterial activity was recorded for compound **2d** for both gram-positive and negative bacteria; this may be due to the four chlorine atoms [27] and pyrimidine ring [28–30]. On the other hand, *S. aureus* and *B. cereus* were resistant to compounds **2a–c**, while they were sensitive to the other compounds. It is worthy to mention that the clear zone caused by compounds **2a**, **b** and **d** with *S. typhi* was nearly closely to the inhibition zone caused by tetracycline disk; the sensitivity of these bacteria toward our compounds may be due to the presence of four chlorine atoms [27] and pyrimidine ring [28–30] (Figure 1). The efficiency of compounds **2a–d**, and **3f** as antibacterial products recorded the highest inhibition with the gram-negative bacteria; while **2d**, **2f**, **3a**, **3e–g** were more efficient for gram-positive bacteria. Thus,

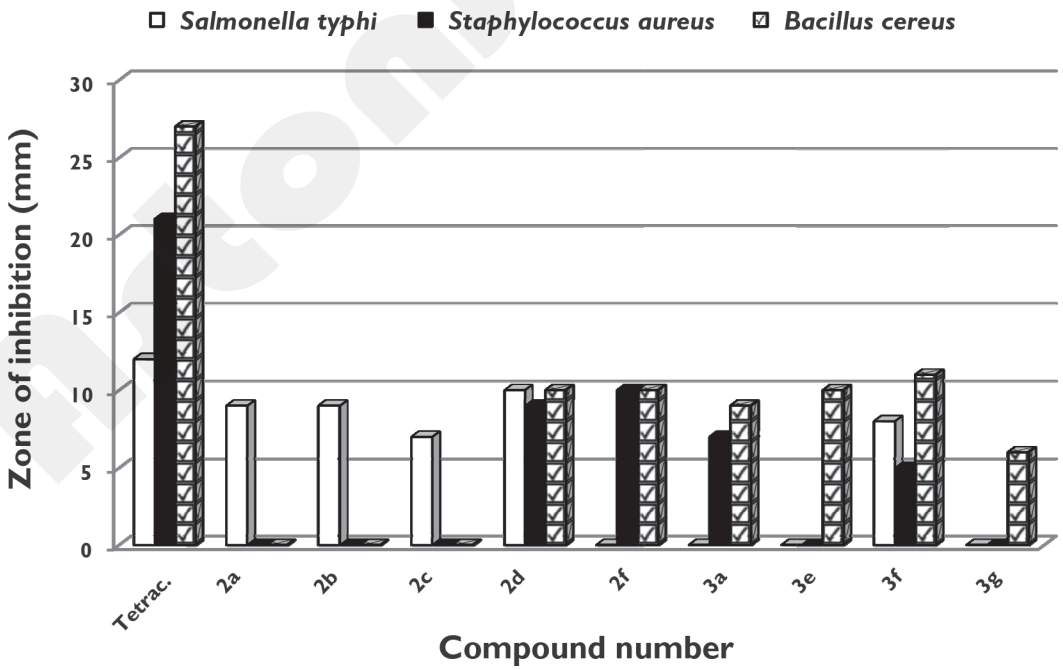


Figure 1: Antibacterial activities of quinazoline dione compounds against some gram-positive and negative bacteria.

the susceptibility of gram-positive bacteria to quinazolinediones was more than those of gram-negative bacteria. Many authors recorded similar observations [31]. The greater susceptibility of gram-positive bacteria to quinazoline dione compounds was because of the differences in their cell wall structure and their composition [32]. In gram-negative bacteria, the outer membrane acts as a barrier to many environmental substances, including antibiotics [33]. The presence of thick murine layer in the cell wall also prevents the entry of the inhibitors [34]. The above results confirm the broad antibacterial effect of quinazolinedione compounds.

Competing Interests
None declared.

Authors' Contributions
All authors contributed equally to this work.

Acknowledgement
The authors are grateful to the spirit of Prof. Dr. Yousef H. Ebeed, Faculty of Science, South Valley University for his guidance and encouragement.

References

[1] American Chemical Society, 1989. UNT Digital Library, Division of Chemical Information. Chemical Information Bulletin, 41(3). New York: Philadelphia, Pennsylvania.
[2] Bakhite EA, Radwan SM, El-deen AMK, 2000. Synthesis of novel pyridothienopyrimidines, pyridothienopyrimidothiazines, pyridothienopyrimidobenzthiazoles and triazolopyridothienopyrimidines. Journal of Chinese Chemical Society, 47(5): 1105.
[3] Younes MI, Abbas HH, Metwally SAM, 1999. Synthesis of ethyl-5-amino-1-(5-ethyl-5H-1,2,4-triazino [5,6-b]indol-3-yl)-1H-pyrazole-4-carboxylate and pyrazolo[3,4-d]pyrimidine derivatives. Pharmazine, 46(2): 98.
[4] Rideout JL, Krenitesky TA, Chao EY, Elion GB, Williams RB, Latter VS, 1983. Pyrazolo[3,4-d]pyrimidine ribonucleosides as anticoccidials. 3. Synthesis and activity of some nucleosides of 4-[(arylalkenyl)thio]-pyrazolo[3,4-d]pyrimidines. Journal of Medicinal Chemistry, 26(10): 1489.
[5] Rideout JL, Krenitesky TA, Koszalka GW, Coln NK, Chao EY, Elion GB, *et al.*, 1982. Pyrazolo[3,4-d]pyrimidine ribonucleosides as anticoccidials. 2. Synthesis and activity of some nucleosides of 4-(alkylamino)-1H-pyrazolo[3,4-d]pyrimidines. Journal of Medicinal Chemistry, 25 (9): 1040.
[6] Marie MG, Aly DM, Mishrikey MM, 1992. A new synthesis of pyrazolo[1,5-c]pyrimidines from acetylenic β-diketones. Bulletin of the Chemical Society of Japan, 65(12): 3419.
[7] Krenitesky TA, Rideout JL, Koszalka GW, Inmmon RB, Chao EY, Elion GB, 1982. Pyrazolo[3,4-d]pyrimidine ribonucleosides as anticoccidials. 2. Synthesis and activity of some nucleosides of 4-(alkylamino)-1H-pyrazolo[3,4-d]pyrimidines. Journal of Medicinal Chemistry, 25(1): 32.
[8] Gatta F, Perotti F, Gradoni L, Gramiccia M, Orsini S, Palazzo G, *et al.*, 1990. Synthesis of some 1-(dihydroxypropyl)pyrazolo [3,4-d]-pyrimidines and in vivo evaluation of their antileishmanial and antitrypanosomal activity. European Journal of Medicinal Chemistry, 30: 419.
[9] Ugarkar BG, Cottam HB, Mekernan PA, Robins RK, Revankar GR, 1984. Synthesis and antiviral/antitumor activities of certain pyrazolo[3,4-d]pyrimidine-4(5H)-selone nucleosides and related compounds. Journal of Medicinal Chemistry, 27(8): 1026.
[10] Makara GM, Ewing W, Winter E, 2001. Synthesis of bicyclic pyrimidine derivatives as ATP analogues. Journal of Organic Chemistry, 66(17): 5783.
[11] Mishra B, Muddin N, 1989. A convenient and facile synthesis of 1-aroyl-4-oxo-5-substituted-phenylpyrazolo[3,4-d]pyrimidine-6-thiones. Indian Journal of Chemistry, 28B: 346.
[12] Miller DJ, Shen H, Such JK, Kerwin SM, Robertass JD, 2002. Structure-based design and characterization of novel platforms for ricin and shiga toxin inhibition. Journal of Medicinal Chemistry, 45(1): 90.
[13] El-Baendary ER, Badria FA, 2000. Synthesis, DNA-binding, and antiviral activity of certain pyrazolo[3,4-d]pyrimidine derivatives. Archives of Pharmacy, 99: 333.
[14] Balzarini J, Mc Guingan C, 2002. Bicyclic pyrimidine nucleoside analogues (BCNAs) as highly selective and potent inhibitors of varicella-zoster virus replication. Journal of Antimicrobial Chemotherapy, 50: 5.

[15] Shalaby AM, Fathalla OA, Kassem EMM, Zaki MEA, 2000. Synthesis of new 5-*N*-pyrazolyl amino acids, pyrazolopyrimidines and pyrazolopyridines derivatives. Acta Chimica Slovenica, 47: 187.
[16] El-Afalek EI, Abubshit SA, 2001. Heterocyclic o-aminonitriles: preparation of pyrazolo[3,4-d]-pyrimidines with modification of the substituents at the 1-position. Molecules, 6: 621.
[17] Bhuyan PJ, Borah HN, Lekhok KC, Sandhu JS, 2001. Studies on uracils: a facile one-pot synthesis of pyrazolo[3,4-d]pyrimidines. Journal of Heterocyclic Chemistry, 38: 491.
[18] Finch RA, Revankar GR, Chen PK, 1996. Structural and functional relationships of toyocamycin on NPM-translocation. Anticancer Drug Design, 12: 205.
[19] Silverstein RM, Bassler GC, Morrill TC, 1916. Spectrometric Identification of Organic Compounds. New York: John Wiley, 35.
[20] Seeley HW, VanDemark PJ, 1981. Selected Exercises from Microbes in Action: A Laboratory Manual of Microbiology, Third Edition, USA: W.H. Freeman and Company.
[21] Salem WM, Galal H, Nasr El-deen F, 2011. Screening for antibacterial activities in some marine algae from the red sea (Hurghada, Egypt). African Journal of Microbiology Research, 5(15): 2160–2167.
[22] Hassan MA, Younes AMM, Taha MM, Abdel-Monsef AH, 2012. Action of some heterocyclic amines and difunctional nucleophiles on *N*-phenylsulphonyloxytetrachlorophthalimide. Chemistry Journal, 158–165.
[23] Kandhasamy M, Arunachalam KD, 2008. Evaluation of in vitro antibacterial property of seaweeds of southeast coast of India. African Journal of Biotechnology, 7(12): 1958–1961.
[24] Kotiranta A, Lounatmaa K, Haapasalo M, 2000. Epidemiology and pathogenesis of *B. cereus* infections. Microbes Infection 2(2): 189–198.
[25] Kandhasamy M, Arunachalam KD, 2008. Evaluation of in vitro antibacterial property of seaweeds of southeast coast of India. African Journal of Biotechnology, 7(12): 1958–1961.
[26] Al-Haj NA, Mashan NI, Shamsudin MN, 2009. Antibacterial activity in marine algae *Eucheuma denticulatum* against *S. aureus* and *S. pyogenes*. Research Journal of Biological Sciences, 4(4): 519–524.
[27] Ryan KJ, Ray CG, 2004. Sherris Medical Microbiology, Fourth Edition. McGraw Hill. ISBN 0-8385-8529-9.
[28] Selassie CD, Li R, Poe M, Hansch C, 1991. On the optimizationof hydrophobic and hydrophilic substituent interactions of 2,4-diamino-5-(substituted-benzyl)pyrimidines with dihydrofolate reductase. Journal of Medicinal Chemistry, 34: 46.
[29] Mc Nair-Scott DB, Ulbrich TLV, Rogers ML, Chu E, Rose C, 1959. Effect of substituted pyrimidines on growth and biosynthesis of microorganisms. Cancer Research, 19: 15.
[30] Cheng CC, 1969. 3 some pyrimidines of biological and medicinal interest-I. Progress in Medicinal Chemistry, 6: 67.
[31] Demirel Z, Yilmaz-Koz FF, Karabay-Yavasoglu UN, Ozdemir G, Sukatar A, 2009. Antimicrobial and antioxidant activity of brown algae from the Aegean sea. Journal of Serbian Chemical Society, 74(6): 619–628.
[32] Taskin E, Ozturk M, Taskin E, Kurt O, 2007. Antibacterial activities of some marine algae from the Aegean sea (Turkey). African Journal of Biotechnology, 6(24): 2746–2751.
[33] Tortora GJ, Funke BR, Case CL, 2001. Microbiology: An Introduction. San Francisco: Benjamin Cummings, 88.
[34] Paul John Peter M, Venkatesan M, Yesu Raj J, 2012. The antibacterial activity and phytochemicals of the leaves of *stylosanthes fruticosa*. International Journal of Phytopharmacy, 2(4): 98–106.