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Synthesis and biological screening of new 4-substituted-2-(3,4,5-trimethoxyphenyl)quinazolines as potential anticancer agents

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ABSTRACT

*Cancer is an umbrella term including a diversity of diseases. The need for new anticancer agents is a necessity. On the other hand, quinazolines are biologically interesting scaffold. 4-substituedamino-2-(3,4,5-trimethoxy)quinazolines were synthesized using anthranilamide as a starting material. Thirteen novel 2-(3,4,5-trimethoxyphenyl)quinazolines **4a-d** and **5a-h** were synthesized. The NCI - USA has chosen seven compounds namely **5a** and **5c-h** of the new quinazoline derivatives for the 60-cell lines screening. Compound **5c** showed null growth percent towards the melanoma MDA-MB-435 cell line. The IC_{50} for **5c** was also investigated for both non-small cell lung cancer cell line A549 and colon cancer cell line HCT116. The results were 8.57 and 10.10 μM/ml respectively.*

Keywords: Anticancer agents; 2-(3,4,5-trimethoxyphenyl)quinazolines; Synthesis.

INTRODUCTION

Quinazoline nucleus is an essential building block. This nucleus can be found in a huge number of natural products as well as synthetic derivatives [1]. From the chemistry point of view, quinazoline can be considered as a benzo[*d*]pyrimidine heterocyclic fused ring.

This wide occurrence and prevalence attracted many scientists sight to its diverse biological activities. In fact, quinazoline-skeletoned products were found to possess antimicrobial [2–5], anti-inflammatory [6–9], anticonvulsant [10–12], antiallergic [13], antileishmaniasis [14], in treatment of diabetes and obesity [15]. Moreover, quinazolines have already proven to possess significant anticancer activity [16–20]. Cancer is an infinite term affecting the global health. The ease of diagnosis, severity and curability rates of each type are widely variable. Accordingly, malignant tumors and their treatment are a major contributing factor affecting the health in both developing as well as developed countries [21]. Nevertheless, huge efforts are continuing to discover new safe, effective and selective chemotherapeutic agents. The 4-substitutedaminoquinazoline-based antitumor agents have already proven potential activity in both research [17,22–24] as well as clinical applications. Gefitinib, (Iressa™), erlotinib (Tarceva™), lapatinib (Tykerb®) are FDA approved chemotherapeutic agents in cancer treatment [25] (Figure 1).

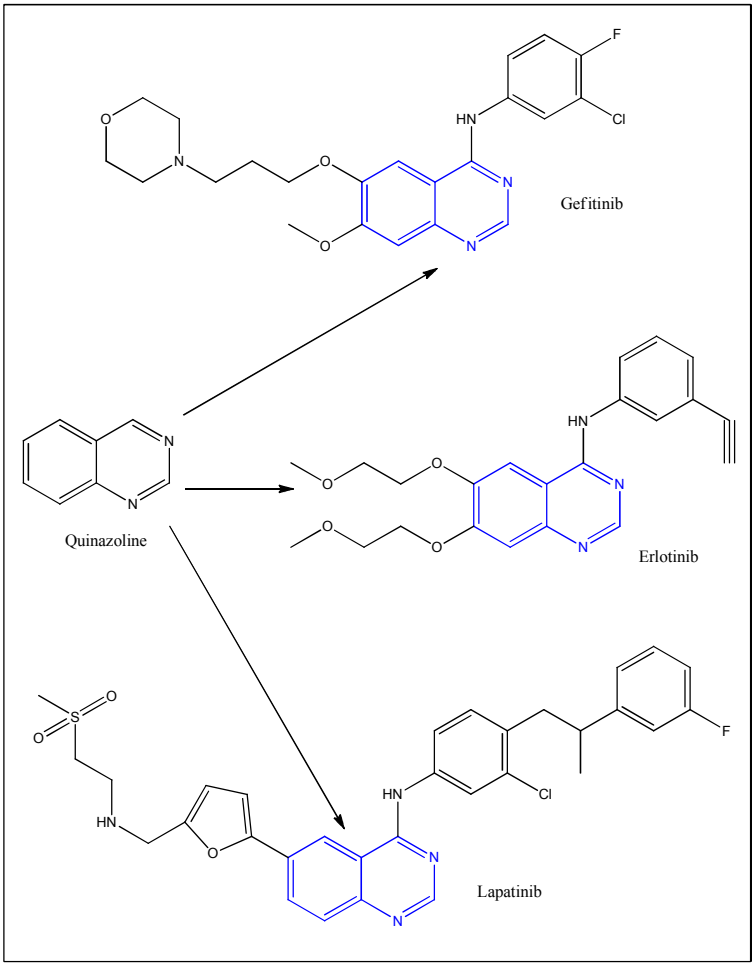


Figure 1. FDA-approved quinazoline-based medications

Accordingly, the 4-substitutedaminoquinazolines have possessed anticancer activity as inhibitors of epidermal growth factor receptor (EGFR) [26–29]. Moreover, the 3,4,5-trimethoxyphenyl moiety is already a biologically active one [30]. Guided by all the above findings and interested in quinazoline nucleus diverse activities, we aimed to the synthesis of this scaffold which bears both the 3,4,5- trimethoxyphenyl moiety and the 4-substitutedaminoquinazolines. we designed and synthesized thirteen new 4-substitutedamino-2-(3,4,5-trimethoxyphenyl)quinolines with expected antitumor cytotoxic activity. (Figure 2)

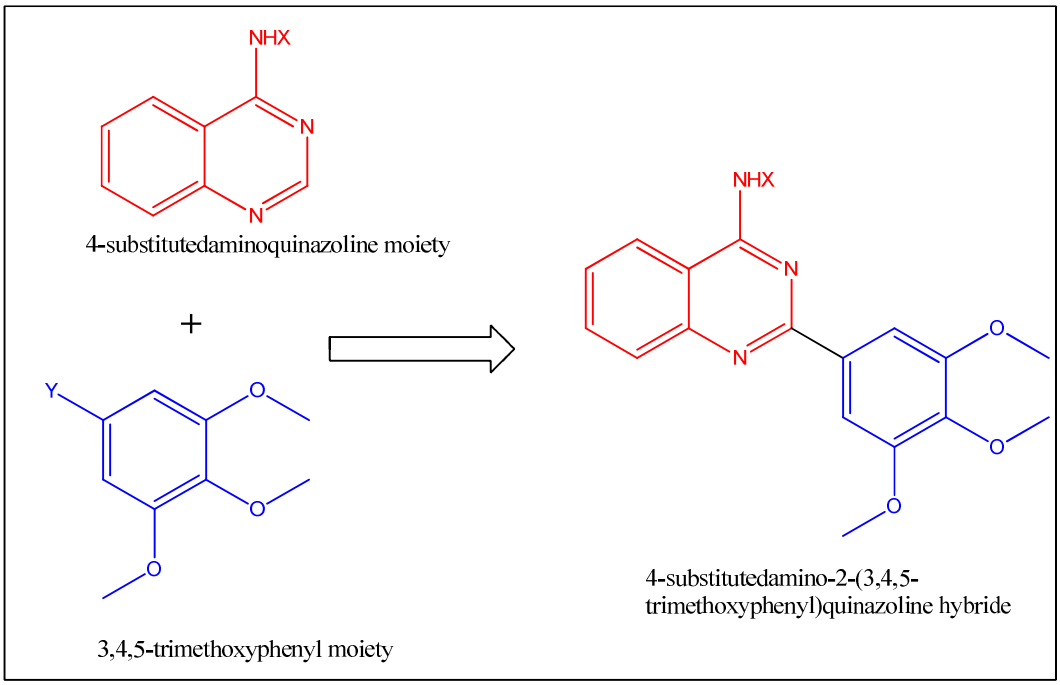


Figure 2. New 4-substitutedamino-2-(3,4,5-trimethoxyphenyl)quinolines scaffold

MATERIALS AND METHODS

Chemistry

General

Melting points were obtained on a Griffin apparatus and were uncorrected. IR spectra were recorded on a Shimadzu 435 spectrometer, using KBr discs. ^{1}H NMR and ^{13}C NMR spectra were performed on JOEL NMR FXQ-300 MHz and JOEL NMR FXQ-400 MHz spectrometers, using TMS as the internal standard. Mass spectra were recorded on a GCM. P.-QP1000 EX Mass spectrometer. Progress of the reactions were monitored by TLC using precoated aluminum sheet silica gel MERCK 60F 254 and was visualized by UV lamp. Element analysis for C, H and N were within 0.4% of the theoretical values and were performed at the regional center for Mycology and Biotechnology, Al-Azhar University. All chemicals were purchased from Sigma-Aldrich company.

4-Oxo-2-(3,4,5-trimethoxyphenyl)quinazoline (1)

Compound **2** was prepared from anthranilamide according to the reported procedure [35] with certain modifications.

4-Chloro-2-(3,4,5-trimethoxyphenyl)quinazoline (2)

A mixture of phosphorus oxychloride (15 ml) and 2-(3,4,5-trimethoxyphenyl)quinazolin-4(3*H*)-one **(2)** (0.003 mol) was refluxed for 1 h. The reaction mixture was concentrated under reduced pressure then poured into ice cold water (100 ml). The precipitated product was filtered, washed with water (2×10 ml), dried and crystallized from ethanol to furnish **3**: Yield 65 %; mp 157-159 ^{o}C; IR cm^{-1}: 3003 (CH aromatic), 2964, 2935 (CH aliphatic), ^{1}HNMR (DMSO-d_6) δ: ^{1}H NMR (DMSO-d_6): δ 3.91 (s, 9H, 3OCH$_3$), 7.56 (s, 2H, ArH), 7.64-7.68 (m, 1H, ArH), 7.95-7.99 (m, 1H, ArH), 8.08-8.08 (d, 1H, ArH) and 8.15-8.17 (d, 1H, ArH); Anal. Calcd. for $C_{17}H_{15}ClN_2O_3$: C, 61.72; H, 4.54; N, 8.47. Found: C, 61.89; H, 4.63; N, 8.64.

4-Hydrazinyl-2-(3,4,5-trimethoxyphenyl)quinazoline (3)

Hydrazine hydrate (99%, 0.62 g, 0.012 mol) was added to a solution of the 4-chloroquinazoline derivative 3 (0.002 mol) in absolute ethanol (20 ml). The reaction mixture was refluxed for 6 h. The precipitate formed upon cooling down was filtered, dried and crystallized from ethanol to give 4: Yield 73 %; m. p.> 300 ^{o}C; IR cm^{-1}: 3307, 3275 (NH, NH$_2$), 1614 (C=N) cm^{-1};^{1}H NMR (DMSO-d_6): δ 3.74 (s, 9H, 3OCH$_3$), 4.99 (br. s, 2H, NH$_2$, D_2O exchangeable), 7.42-7.62 (m, 1H, ArH), 7.74 (s, 2H, ArH), 7.90 (s, 2H, ArH), 8.16-8.18 (m, 1H, ArH) and 9.62 (br.

s, 1H, NH, D_2O exchangeable); ^{13}CNMR (75 MHz, DMSO-d_6): δ: 56.43, 60.59, 105.95, 122.75, 125.62, 133.01, 140.02, 153.12; MS [m/z, %]: 326 [M^+, 100]; Anal. Calcd. for $C_{17}H_{18}N_4O_3$: C, 62.57; H, 5.56; N, 17.17. Found: C, 62.71; H, 5.62; N, 17.29.

***General procedure for the synthesis of compounds 4a-d*:**
To a solution of 4-hydrazinyl-2-(3,4,5-trimethoxyphenyl)quinazoline (**4**) (0.978 g, 0.003 mol) in absolute ethanol (17 ml)) and few drops of glacial acetic acid, the selected aromatic aldehyde (0.003 mol) was added. The reaction mixture was heated under reflux for 6 h. Then cooled and the separated solid was filtered, dried and crystallized from ethanol to afford 5a-d.

4-(2-(4-Fluorobenzylidenehydrazinyl-2-(3,4,5-trimethoxyphenyl)quinazoline (4a)
Yield 68 %; m. p. 219 – 221 ^{o}C; IR cm^{-1}: 3419 (NH), 1627 (C=N); ^{1}H NMR (DMSO-d_6): δ 3.94 (s, 9H, 3OCH_3), 7.31-7.37 (m, 2H, ArH),7.58 (s, 1H, ArH), 7.86-7.93 (m, 7H, ArH), 8.51 (s, 1H, N=CH) and 11.80 (br. s, 1H, NH, D_2O exchangeable); MS [m/z, %]: 432 [M^+, 100]; Anal. Calcd. for $C_{24}H_{21}FN_4O_3$: C, 66.66; H, 4.89; N, 12.96. Found: C, 66.82; H, 4.93; N, 13.04.

4-(2-(4-Chlorobenzylidenehydrazinyl-2-(3,4,5-trimethoxyphenyl)quinazoline (4b): Yield 68 %; m. p. 212 – 214 ^{o}C; IR cm^{-1}: 3380 (NH), 2949, 2831 (CH aliphatic), 1625 (C=N); ^{1}H NMR (DMSO-d_6): δ 3.28 (s, 9H, 3OCH_3), 7.53-7.60 (m, 2H, ArH),7.83-7.92 (m, 8H, ArH), 8.48 (s, 1H, N=CH) and 11.86 (br. s, 1H, NH, D_2O exchangeable); MS [m/z, %]: 448 [M^+, 87.9]; 337 [M- ClC_6H_4, 100]; Anal. Calcd. for $C_{24}H_{21}ClN_4O_3$: C, 64.21; H, 4.72; N, 12.48. Found: C, 64.29; H, 4.69; N, 12.66.

4-(2-(4-Bromobenzylidenehydrazinyl-2-(3,4,5-trimethoxyphenyl)quinazoline (4c): Yield 79 %; m. p. 223 – 225 ^{o}C; IR cm^{-1}: 3371 (NH), 1625 (C=N); ^{1}H NMR (DMSO-d_6): δ 3.22 (s, 9H, 3OCH_3), 7.58-7.92 (m, 10H, ArH), 8.47 (s, 1H, N=CH) and 11.87 (br. s, 1H, NH, D_2O exchangeable); ^{13}CNMR (75 MHz, DMSO-d_6): δ: 56.49, 60.61, 105.95, 113.05 123.36, 126.24, 128.48, 129.08, 132.40, 133.76, 134.00, 134.51, 140.12, 144.61, 153.25, 159.13; MS [m/z, %]: 492, 494 [M^+, M+2, 68, 67]; 337 [M- BrC_6H_4, 100]; Anal. Calcd. for $C_{24}H_{21}BrN_4O_3$: C, 58.43; H, 4.29; N, 11.36. Found: C, 58.58; H, 4.33; N, 11.42.

4-(2-(4-Methoxybenzylidenehydrazinyl-2-(3,4,5-trimethoxyphenyl)quinazoline (4d): Yield 83 %; m. p. 172 – 175 ^{o}C; IR cm^{-1}: 3360 (NH), 2935, 2821 (CH aliphatic), 1627 (C=N); ^{1}H NMR (DMSO-d_6): δ 3.29 (s, 9H, 3OCH_3), 7.04-7.91 (m, 10H, ArH), 8.44 (s, 1H, N=CH) and 11.68 (br. s, 1H, NH, D_2O exchangeable); MS [m/z, %]: 444 [M^+, 100]; Anal. Calcd. for $C_{25}H_{24}N_4O_4$: C, 67.55; H, 5.44; N, 12.60. Found: C, 67.62; H, 5.52; N, 12.81.

General procedure for the synthesis of compounds 5a-h:
The appropriate secondary amine / *N*-(substituted)phenylpiperazine was added to a mixture of the 4-hydrazinoquinazoline derivative **3** (0.326 g, 0.001 mol) and triethylamine (0.36 ml) in absolute ethanol (12 ml). The reaction mixture was heated under reflux for 15 h. The reaction mixture was cooled and the separated solid was filtered, dried and crystallized from ethanol to afford series 6a-h.

4-(2-(3,4,5-trimethoxyphenyl)quinazolin-4-yl)morpholine (5a): Yield: 76 %; m. p. 142 – 144 ^{o}C IR cm^{-1}: 3040 (CH aromatic), 2995, 2994 (CH aliphatic), 1610 (N=C); ^{1}H NMR (DMSO-d_6): δ 3.75 (m, 4H, 2NCH_2), 3.82 (m, 4H, 2OCH_2), 3.89 (s, 9H, 3OCH_3), 7.48-7.53 (m, 1H, ArH), 7.79-7.91 (m, 4H, ArH), 8.02-8.04 (m, 1H, ArH); ^{13}CNMR (75 MHz, DMSO-d_6): δ 50.28, 56.59, 60.61, 66.49, 105.75, 115.01, 125.73, 125.81, 128.82, 133.42, 133.89, 134.00, 140.21, 152.50, 153.28, 158.16, 164.53; MS: m/z (% abundance) 381 (M^+, 100), 323 ($C_5H_{10}N$, 68); Anal. Calcd. for $C_{21}H_{23}N_3O_4$: C, 66.13; H, 6.08; N, 11.02. Found: C, 66.41; H, 6.13; N, 11.14.

4-(piperidin-1-yl)-2-(3,4,5-trimethoxyphenyl)quinazoline (5b): Yield: 68 %; m. p. 132 – 134 ^{o}C IR cm^{-1}: 3103, 3066, 3035 (CH aromatic), 2995, 2939, 2841 (CH aliphatic); ^{1}H NMR (DMSO-d_6): δ 1.13-1.23 (m, 6H, 3CH_2), 3.66-3.93 (m, 13H, 3OCH_3 + CH_2NCH_2), 7.45-7.95 (m, 6H, ArH); MS: m/z (% abundance) 379 (M^+, 0.29), 324 ($C_5H_{11}N$, 100); (Anal. Calcd. for $C_{22}H_{25}N_3O_3$: C, 69.64; H, 6.64; N, 11.07. Found: C, 69.73; H, 6.73; N, 11.13.

4-(4-methylpiperazin-1-yl)-2-(3,4,5-trimethoxyphenyl)quinazoline (5c): Yield: 78 %; m. p. 98 – 100 ^{o}C; IR cm^{-1}: 3103, 3066, 3035 (CH aromatic), 2995, 2939, 2841 (CH aliphatic).; ^{1}H NMR (DMSO-d_6): δ 3.75 (s, 8H, 4NCH_2), 3.29 (s, 12H, 3OCH_3 + NCH_3), 7.20-7.56 (m, 2H, ArH), 7.82-7.90 (m, 4H, ArH); MS: m/z (% abundance) 394 (M^+,

2.55), 324 ($C_5H_{11}N$, 100); Anal. Calcd. for $C_{22}H_{26}N_4O_3$: C, 66.99; H, 6.64; N, 14.20. Found: C, 67.13; H, 6.98; N, 14.37.

4-(4-ethylpiperazin-1-yl)-2-(3,4,5-trimethoxyphenyl)quinazoline (5d): Yield: 85 %; m. p. 124 – 126 ^oC; IR cm^{-1}: 3040 (CH aromatic), 2972, 2935 (CH aliphatic), 1612 (N=C); ^1H NMR (DMSO-d_6): δ 1.22-1.31 (t, 3H, CH_3), 3.75 (s, 3H, OCH_3), 3.92 (s, 6H, $2OCH_3$), 7.54-7.58 (m, 1H, ArH), 7.83 (m, 4H, ArH), 7.98-8.08 (m, 1H, ArH); MS: m/z (% abundance) 408 (M^+, 2.82); 324 ($C_5H_{11}N$, 100); Anal. Calcd. for $C_{23}H_{28}N_4O_3$: C, 67.63; H, 6.91; N, 13.72. Found: C, 67.80; H, 6.98; N, 13.81.

4-(4-phenylpiperazin-1-yl)-2-(3,4,5-trimethoxyphenyl)quinazoline (5e): Yield: 72 %; m. p. 222 – 226 ^oC; IR cm^{-1}: 3005, 3043, 3053, 3074 (CH aromatic), 2980, 2962, 2935, 2891 (CH aliphatic), 1598 (N=C); ^1H NMR (DMSO-d_6): δ 3.42, 3.97 (br. s, 8H, $4NCH_2$ piperidinyl), 3.75 (s, 3H, OCH_3), 3.91 (s, 6H, $2OCH_3$), 6.79-6.83 (m, 1H, ArH), 6.98-7.01 (m, 2H, ArH), 7.22-7.28 (m, 2H, ArH), 7.50-7.55 (m, 1H, ArH), 7.79-7.92 (m, 4H, ArH), 8.05-8.08 (m, 1H, ArH); ^{13}CNMR (75 MHz, DMSO-d_6) δ: 31.16, 48.48, 49.52, 56.38, 60.62, 105.78, 115.09, 115.88, 119.53, 125.81, 128.82, 129.51, 133.44, 133.93, 140.22, 151.28, 153.29, 158.18, 164.42; MS: m/z (% abundance) 456 (M^+, 6.82); 324 ($C_5H_{11}N$, 100); Anal. Calcd. for $C_{27}H_{28}N_4O_3$: C, 71.03; H, 6.18; N, 12.27. Found: C, 71.27; H, 6.22; N, 12.42.

4-(4-(4-chlorophenyl)piperazin-1-yl)-2-(3,4,5-trimethoxyphenyl)quinazoline (5f): Yield: 75 %; m. p. 210 – 212 ^oC; IR cm^{-1}: 3040 (CH aromatic), 2997, 2978, 2864 (CH aliphatic), 1608 (N=C); ^1H NMR (DMSO-d_6) δ: 3.37, 3.97 (br. s, 8H, $4NCH_2$ piperidinyl), 3.75 (s, 3H, OCH_3), 3.91 (s, 6H, $2OCH_3$), 7.00-7.12 (m, 4H, ArH), 7.51-7.56 (m, 1H, ArH), 7.80-7.92 (m, 4H, ArH), 8.06-8.09 (m, 1H, ArH); MS: m/z (% abundance) 324 ($C_5H_{11}N$, 100); Anal. Calcd. for $C_{27}H_{27}ClN_4O_3$: C, 66.05; H, 5.54; N, 11.41. Found: C, 66.17; H, 5.58; N, 11.54.

4-(4-(2-chlorophenyl)piperazin-1-yl)-2-(3,4,5-trimethoxyphenyl)quinazoline (5g): Yield: 82 %; m. p. 233 – 235 ^oC; IR cm^{-1}: 3076, 3064 (CH aromatic), 2997, 2978, 2864 (CH aliphatic), 1608 (N=C); ^1H NMR (DMSO-d_6) δ: 3.24, 4.01 (br. s, 8H, $4NCH_2$ piperidinyl), 3.75 (s, 3H, OCH_3), 3.90 (s, 6H, $2OCH_3$), 7.08-7.10 (m, 1H, ArH), 7.22-7.32 (m, 2H, ArH), 7.44-7.55 (m, 2H, ArH), 7.82-7.92 (m, 4H, ArH), 8.06-8.09 (m, 1H, ArH); MS: m/z (% abundance) 490 (M^+, 4.95); 324 ($C_5H_{11}N$, 100); Anal. Calcd. for $C_{27}H_{27}ClN_4O_3$: C, 66.05; H, 5.54; N, 11.41. Found: C, 66.19; H, 5.60; N, 11.52.

4-(4-(4-methoxyphenyl)piperazin-1-yl)-2-(3,4,5-trimethoxyphenyl)quinazoline (5h): Yield: 73 %; m. p. 112 – 114 ^oC IR cm^{-1}: 3040 (CH aromatic), 2937, 2829 (CH aliphatic), 1610 (N=C); ^1H NMR (DMSO-d_6) δ: 3.24, 4.01 (br. s, 8H, $4NCH_2$ piperidinyl), 3.75 (s, 3H, OCH_3), 3.90 (s, 6H, $2OCH_3$), 7.08-7.10 (m, 1H, ArH), 7.22-7.32 (m, 2H, ArH), 7.44-7.55 (m, 2H, ArH), 7.82-7.92 (m, 4H, ArH), 8.06-8.09 (m, 1H, ArH); MS: m/z (% abundance) 486 (M^+, 11.65); 324 ($C_5H_{11}N$, 100); Anal. Calcd. for $C_{28}H_{30}N_4O_4$: C, 69.12; H, 6.21; N, 11.51. Found: C, 69.23; H, 6.27; N, 11.59.

Anticancer activity screening

Anticancer activity screening of the newly synthesized compounds was measured in vitro utilizing 59 different human tumor cell lines provided by US National Cancer Institute according to previously reported standard procedure [36,37] as follows:

Cells are inoculated into 96 well microtitre plates in 100 µL. After cell inoculation, the microtitre plates are incubated at 37° C, 5 % CO_2, 95 % air and 100 % relative humidity for 24 h prior to addition of experimental drugs. After 24 h, two plates of each cell line are fixed *in situ* with TCA, to represent a measurement of the cell population for each cell line at the time of drug addition (Tz). Experimental drugs are solubilized in dimethyl sulfoxide at 400-fold the desired final maximum test concentration and stored frozen prior to use. At the time of drug addition, an aliquot of frozen concentrate is thawed and diluted to twice the desired final maximum test concentration with complete medium containing 50 µg/ml gentamicin. Additional four, 10-fold or ½ log serial dilutions are made to provide a total of five drug concentrations plus control. Aliquots of 100 µL of these different drug dilutions are added to the appropriate microtitre wells already containing 100 µL of medium, resulting in the required final drug concentrations. Following drug addition, the plates are incubated for an additional 48 h at 37°C, 5 % CO_2, 95 % air, and 100 % relative humidity. For adherent cells, the assay is terminated by the addition of cold TCA. Cells are fixed *in situ* by the gentle addition of 50 µL of cold 50 % (w/v) TCA (final concentration, 10 % TCA) and incubated for 60 minutes at 4°C. The supernatant is discarded, and the plates are washed five times with tap water and air dried.

Sulforhodamine B (SRB) solution (100 µL) at 0.4 % (w/v) in 1 % acetic acid is added to each well, and plates are incubated for 10 minutes at room temperature. After staining, unbound dye is removed by washing five times with 1 % acetic acid and the plates are air dried. Bound stain is subsequently solubilized with 10 mM trizma base, and the absorbance is read on an automated plate reader at a wavelength of 515 nm. For suspension cells, the methodology is the same except that the assay is terminated by fixing settled cells at the bottom of the wells by gently adding 50 µL of 80 % TCA (final concentration, 16 % TCA). Using the seven absorbance measurements [time zero, (Tz), control growth, (C), and test growth in the presence of drug at the five concentration levels (Ti)], the percentage growth is calculated at each of the drug concentrations levels. Percentage growth inhibition is calculated as:
[(Ti-Tz)/(C-Tz)] x 100 for concentrations for which Ti>/=Tz
[(Ti-Tz)/Tz] x 100 for concentrations for which Ti<Tz.

RESULTS AND DISCUSSION

Chemistry

This piece of work was sketched out in one scheme. Compound **1** is considered as the building block for this scheme. It is worth mentioning that **1** was previously prepared starting from either anthanilamide [31,32] or isatoic acid [33,34]. In this paper, compound **1** was prepared starting with anthranilamide. The reaction of anthranilamide with 3,4,5-trimethoxybenzaldehyde in dimethylformamide with drops of conc. hydrochloric acid afforded the 2-substitutedquinazolin-4-one derivative **1**. Furthermore, refluxing of **1** with phosphorous oxychloride for 1 hour yielded the corresponding chloroquinazoline derivative **2** in a good yield. It is worth mentioning that **2** was reacted with nitrogenous compounds to afford the final products. Accordingly, the reaction of **2** with hydrazine hydrate yielded the corresponding 4-hydrazinoquinazoline derivative **3**. The existence of compound **3** was confirmed through the appearance of different important key spectroscopic clues. In ^{1}HNMR, the appearance of broad peak at δ 4.99 ppm equivalent to two protons and its disappearance by D_2O. Furthermore, the appearance of two IR. peaks in the range of 3307 and 3275 cm^{-1} corresponding to the (NH) and (NH_2) groups. The preparation of Schiff's bases **4a-d** were achieved through the reaction of the 4-hydazinoquinazoline derivative **3** with different aldehydes in the presence of a catalytic amount of glacial acetic acid. Once more the spectroscopic elucidation of the various spectral data confirmed such formation. Series **5** was prepared with the aim of preparing the 4-substituedminoquinazoline derivatives. Refluxing different secondary amines with the 4-chloro-2-(3,4,5-trimethoxyphenyl)quinazoline (**2**) in absolute ethanol and in the presence of triethylamine as a catalyst afforded **5a-h** in quite good yields. Interestingly, series **5** showed a unique mass fragmentation pattern. A very common fragment peak m/z 324 was appearing in this series members. A plausible mechanism explaining this fragment formation is sketched out in figure 3.

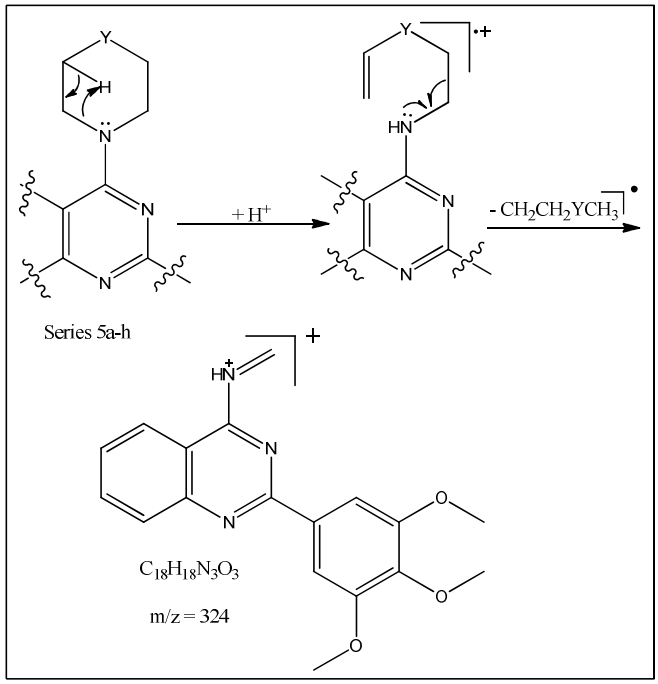
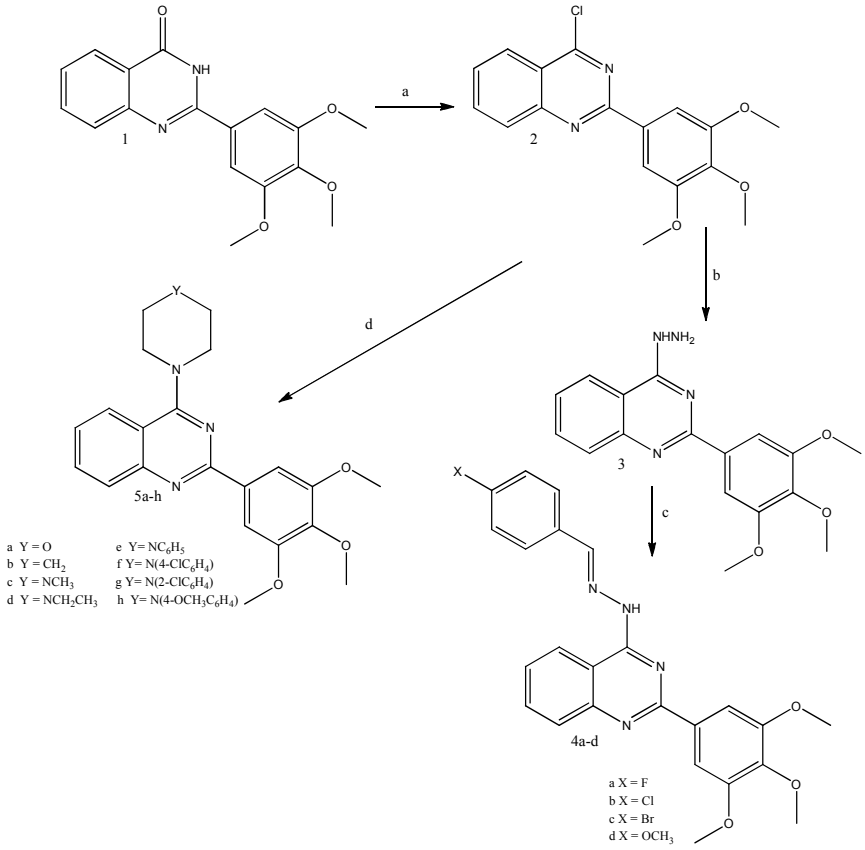


Figure 3. The mechanism explaining the formation of the common mass fragment



Reagents and conditions: a) $POCl_3$, reflux 2h; **b)**$NH_2NH_2.H_2O$, absolute ethanol, reflux 6h; c) aromatic aldehyde, drops of glacial acetic acid, absolute ethanol, reflux 6 h; d) appropriate secondery amine, absolute ethanol, TEA, reflux 15 h.

Scheme 1. The synthetic pathways and reagents for the preparation of series 4a-d and 5a-h

Anticancer activity

The National Cancer Institute (NCI) USA, has chosen seven compounds for anticancer evaluation. The denoted compounds were **5a**, and **5c-h**. The chosen compounds were evaluated at a single dose (10^{-5} M) utilizing 60 different human tumor cell lines representing different types of cancers. The tumor growth inhibition properties of the seven compounds **5a** and **5c-h** with the NCI codes NSC D-784287/1, D-784290/1, D-784291/1, D-784292/1, D-784293/1, D-784294/1, D-784295/1 selected among **3** - **5a-h** by the NCI at the NIH, Bethesda, Maryland, USA, under the drug discovery program of the NCI. The results obtained from the single dose test of the selected compounds are shown in Table 1. The chosen compounds showed mild activity against certain tumor cell lines. Compound **5a** showed growth inhibition percentages 63.24, 61.24, 63.74 and 71.9 towards leukemia SR cancer cell line, non-small cell lung cancer HOP-92 cell line, renal cancer 786-0 and breast cancer T-47D cell line respectively. Interestingly, compound **5c** showed pronounced growth inhibition percentages. In fact, **5c** possessed 100% growth inhibition towards melanoma MDA-MB-35 cancer cell line, 96.82% towards ovarian cancer cell line OVCAR-3, 91.42% towards non-small cell lung cancer NCI-H522 and 70.46% towards MCF7 breast cancer cell line. Compound **5d** showed growth inhibition percentages 42.96 and 42.88 towards CNS cancer cell line U251 and breast cancer MDA-MB-231 respectively. Finally, both **5g** and **5h** exhibited growth inhibition percentages 51.64 and 58.68 respectively against PC-3 prostate cancer cell line. Although none of the selected compounds was selected for further 5-log dose molar range we were interested in checking the IC50 for compound **5c**. Two cancer cell lines, namely, non-small cell lung cancer A549-1 and HCT-E colon cancer cell line were used to check the IC50 of compound **5c**. This 5-log dose molar range in *vitro* assay was done in the NCI, Egypt. Interestingly, the results of compound **5c** were 8.57 and 10.10 μM/ml respectively.

Table 1. The Sixty human tumor cell line anticancer screening data at a single dose assay (10^{-5} M) as a percent cell growth for compounds 6a and 6c-6h

Panel/ cell line	Compound no.						
	5a	5c	5d	5e	5f	5g	5h
Leukemia							
CCRF-CEM	56.65	99.29	105.53	105.49	138.38	108.27	91.32
HL-60(TB)	56.55	67.02	71.31	68.86	90.50	88.44	44.38
K-562	70.78	44.75	96.00	98.33	126.10	109.37	79.14
RPMI-8226	70.45	71.24	85.53	118.39	101.73	73.17	54.82
SR	36.76	48.92	89.78	90.16	111.80	76.83	42.56
Non-Small Cell Lung Cancer							
A549/ATCC	61.80	72.54	100.07	104.76	101.94	85.37	80.49
EKVX	50.32	67.18	71.78	96.60	96.46	88.74	84.97
HOP-62	49.24	47.35	71.85	96.30	75.51	112.07	83.88
HOP-92	38.44	44.28	58.30	110.61	86.50	79.84	56.63
NCI-H226	57.76	66.22	75.46	92.75	94.64	83.16	78.37
NCI-H23	79.63	74.40	83.95	90.43	91.50	85.64	81.13
NCI-H322M	78.95	84.40	81.93	96.65	95.87	99.79	91.36
NCI-H460	95.60	84.53	102.84	96.10	101.93	89.20	87.01
NCI-H522	44.91	8.58	83.09	87.98	93.54	69.48	55.15
Melanoma							
LOX IMVI	72.20	44.25	87.18	97.94	96.71	89.33	79.35
MALME-3M	99.30	63.80	93.16	102.84	102.23	116.78	108.40
M14	72.32	41.99	81.86	97.76	90.85	90.38	71.29
MDA-MB-435	70.74	0.00	81.87	95.98	96.62	81.78	67.11
SK-MEL-2	93.70	81.20	98.92	104.53	103.81	98.73	93.58
SK-MEL-28	84.29	59.46	98.71	108.38	105.91	86.71	87.10
SK-MEL-5	62.24	47.05	86.29	96.32	93.70	63.84	46.43
UACC-257	91.69	58.19	123.56	120.30	98.10	99.99	102.39
UACC-62	38.81	18.19	80.54	88.38	86.53	72.78	55.98

Panel/ cell line	Compound no.						
	5a	5c	5d	5e	5f	5g	5h
Colon Cancer							
COLO 205	67.30	81.45	74.61	93.47	93.46	76.63	62.87
HCC-2998	76.03	88.37	91.62	86.46	101.95	106.39	95.12
HCT-116	67.45	31.94	77.30	95.66	94.59	64.19	69.99
HCT-15	68.09	56.93	87.99	99.60	99.64	57.90	61.81
HT29	46.61	49.53	79.70	103.82	99.50	68.21	81.50
KM12	76.52	53.42	86.46	90.51	96.18	83.01	69.96
SW-620	87.57	53.30	81.48	85.50	93.90	88.66	85.57
CNS Cancer							
SF-268	76.32	56.75	82.45	90.49	98.64	89.22	87.72
SF-295	76.01	28.56	96.49	107.95	103.50	100.29	95.16
SF-539	71.81	39.27	92.85	104.85	101.47	99.82	96.89
SNB-19	86.92	85.19	92.64	107.82	99.85	100.85	93.45
SNB-75	47.61	15.70	57.04	75.24	87.00	76.10	84.52
U251	80.99	60.21	97.49	100.95	99.09	90.65	96.13

Panel/ cell line	Compound no.						
	5a	5c	5d	5e	5f	5g	5h
Ovarian Cancer							
IGROV1	1 58.13	62.47	90.81	100.53	98.45	105.42	97.05
OVCAR-3	75.51	3.18	88.94	93.40	105.35	95.15	94.62
OVCAR-4	71.80	55.86	92.90	95.13	100.94	86.72	87.99
OVCAR-5	82.96	88.54	90.70	98.12	100.44	95.69	93.50
OVCAR-8	72.30	75.69	103.92	108.73	103.16	90.56	71.11
NCI/ADR-RES	56.63	10.31	84.13	96.85	92.03	81.85	65.26
SK-OV-3	3 64.51	68.42	99.61	103.41	90.40	94.77	91.42
Renal Cancer							
786-0	36.26	47.51	69.80	105.84	104.76	82.40	85.70
A498	51.60	21.63	98.76	94.92	101.02	106.25	79.51
ACHN	74.85	47.50	91.30	103.64	100.26	93.28	90.64
CAKI-1	68.47	50.26	88.56	91.62	97.12	92.00	84.07
RXF 393	76.29	51.38	100.89	103.85	100.21	94.51	92.61

SN12C	73.61	82.57	85.03	107.87	101.53	99.07	90.11
TK-10	45.75	94.13	109.58	138.41	124.18	114.22	113.37
UO-31	40.36	44.97	76.77	94.61	95.14	83.45	69.91
Prostate Cancer							
PC-3	41.39	52.79	67.13	93.44	73.59	48.36	41.32
DU-145	80.87	82.40	95.54	102.16	109.25	93.06	93.46
Breast Cancer							
MCF7	68.42	29.54	83.41	90.70	92.24	91.22	83.33
MDA-MB-231/ATCC	61.47	61.23	57.12	97.70	85.68	81.86	70.96
HS 578T	50.74	41.11	74.09	84.35	88.54	78.49	65.18
BT-549	53.58	84.72	84.56	94.62	90.79	85.25	39.89
T-47D	28.10	65.34	82.78	83.49	84.11	65.22	45.40
MDA-MB-468	67.52	38.44	66.42	98.34	96.54	82.96	76.20

CONCLUSION

In this piece of work, we succeeded in synthesizing a new hybrid 4-substitutedaminoquinazolino-3,4,5-trimethoxyphenyl motif. The newly designed and synthesized series of compounds beard both active pharmacophore moieties at quinazoline C-4 position in combination with trimethoxyphenyl moiety at position 2 in the quinazoline ring. Compounds **6a** and **6c-h** were screened against the antineoplastic activity. Compound **6a** showed the best activity towards melanoma MDA-MB-435 cell lines, non-small cell lung cancer NCI-H522 and ovarian cell cancer OVAR-3 with percent of inhibition 100, 91.42 and 96.82 respectively. The IC50 for **6c** was investigated for both non-small cell lung cancer A549 and colon cancer cell line HCT-E. The results were 8.57 and 10.10 µM/ml respectively.

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