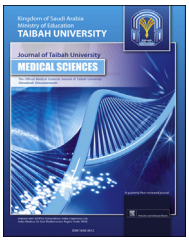
Journal of Taibah University Medical Sciences (2015) 10(3), 333–339

Taibah University

Journal of Taibah University Medical Sciences

www.sciencedirect.com



Original Article

Synthesis and screening of some new fluorinated quinazolinone–sulphonamide hybrids as anticancer agents

Mohamed F. Zayed, PhD [a,d,*], Hany E.A. Ahmed, PhD [a,e], Saleh Ihmaid, PhD [a,f], Abdel-Sattar M. Omar, PhD [b,d] and Adel S. Abdelrahim, PhD [c,d]

[a] *Pharmaceutical Chemistry Department, College of Pharmacy, Taibah University, Almadinah Almunawwarah, KSA*
[b] *Pharmaceutical Chemistry Department, Faculty of Pharmacy, King AbdulAziz University, Jeddah, KSA*
[c] *Pharmaceutical Chemistry Department, Faculty of Pharmacy, Jazan University, Jazan, KSA*
[d] *Pharmaceutical Chemistry Department, Faculty of Pharmacy, Al-Azhar University, Cairo, Egypt*
[e] *Organic Chemistry Department, Faculty of Pharmacy, Al-Azhar University, Cairo, Egypt*
[f] *La Trobe University, Melbourne, Bendigo, Australia*

Received 20 August 2014; revised 5 February 2015; accepted 6 February 2015; Available online 12 March 2015

الملخص

هدف البحث: يهدف هذا البحث إلى تحضير بعض مشتقات مركبات الكوينازولين المفلورة الجديدة المرتبطة بالسلفوناميد، وتقييم نشاطها السام ضد الخلايا في المختبر.

طرق البحث: تم تحضير ثمانية مركبات، وتقييم نشاطها المضاد للسرطان باستخدام ٣ مجموعات من الخلايا تتضمن خلايا سرطان الرئة من المعهد الوطني للسرطان، وخلايا سرطان الثدي من مؤسسة ميتشغان للسرطان، وخلايا طبيعية من كلى الجنين البشرية -٢٩٣.

النتائج: أظهر أحد المركبات نشاطا ذا أهمية مضادة للسرطان مع سمية منخفضة عند مقارنته مع ميثوتركسات كدواء مرجعي. كما أظهر الفحص البيولوجي نشاطا مضادا للسرطان جيدا إلى معتدل للمركبات في القائمة بالمقارنة بالأدوية المرجعية. المركبات الحديثة لها سمية أقل على الخلايا الطبيعية بالمقارنة بالميثوتركسات.

الاستنتاجات: يمكن أن توفر المركبات المحضرة حديثا قالبا ذا قيمة للتحضير الأمثل مستقبلا لإنتاج نظائر أكثر نشاطا من مضادات للسرطان.

الكلمات المفتاحية: كوينازولين؛ سلفوناميد؛ مضاد للسرطان؛ سميات الخلايا؛ تركيب

Abstract

Objectives: The aim of the present research was to synthesise several novel fluorinated quinazoline–sulphonamide derivatives and to evaluate their *in vitro* cytotoxic activity.

Methods: Eight compounds were synthesised. The compounds' anticancer activities were determined through the [3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyltetrazolium bromide] (MTT) assay using a three-cell-line panel consisting of National Cancer Institute (NCI) lung cancer cells, Michigan Cancer Foundation-7 (MCF-7) breast cancer cells, and Human Embryonic Kidney-293 (HEK-293) normal kidney cell. The values of C log P correlations were determined to interpret the results.

Results: One compound exhibited significant anticancer activity with low toxicity compared with the methotrexate as the reference drug. The biological screening showed good to moderate anticancer activity for the title compounds compared with the reference drug. The reference drug exhibited an IC_{50} value of 2.4 μM, whereas compound 9, which was identified as the most active compound, exhibited an IC_{50} value of 2.51 μM on the NCI cell line. The other compounds showed IC_{50} values that ranged from 2.89 to 46.34 μM on the three cell lines. The newly synthesized compounds had lower toxicity on the normal cell line than did methotrexate.

Conclusions: The newly synthesized compounds may provide a valuable template for future design and

* Correspondence address: Department of Pharmaceutical Chemistry, College of Pharmacy, Taibah University, P.O. Box 344, Almadinah Almunawwarah, KSA.
E-mail: mfzayed25@yahoo.com (M.F. Zayed)

Peer review under responsibility of Taibah University.

Production and hosting by Elsevier

 http://dx.doi.org/10.1016/j.jtumed.2015.02.007

optimization to produce analogues that act as more active anticancer agents.

Keywords: Anticancer; Cytotoxic; Quinazolinone; Sulphonamides; Synthesis

Introduction

Quinazolinones have various biological activities, including anticonvulsant,[1,2] antihistaminic,[3] anti-inflammatory, antibacterial,[4] antidiabetic,[5] antifungal,[6] anticancer,[7] anthelmintics[8] and antiviral activities.[9] Quinazolinone derivatives produce their anticancer activity through potent inhibition of various enzymes, such as epidermal growth factor receptor tyrosine kinase, dihydrofolate reductase, folate thymidylate synthase, tyrosine kinase, aldose reductase, cyclic GMP phosphodiesterase and DNA repairing enzymes.[7] Quinazolinone derivatives have therefore been widely used in the production of anticancer drugs.[7] In contrast, sulphonamides have various biological activities, and some of them are widely used in therapy as substantial anticancer agents.[10,11] Sulphonamides act as anticancer agents through a variety of mechanisms, such as cell cycle perturbation in the G1 phase, disruption of microtubule assembly, functional suppression of the transcriptional activator NF-Y[12,13] and inhibition of the carbonic anhydrase (CA) enzyme.[14] In our previous studies,[15] we reported that some derivatives of 6,8-diiodo-2-phenyl-3-substituted-quinazolin-4(3H)-ones, such as compound (A), exhibit good cytotoxic activity (Figure 1). These derivatives are hybrid molecules that included 6,8-diiodo quinazolinone as a fixed moiety with different L-amino acids. Iodine atoms at positions 6 and 8 of the quinazolinone moiety have larger atomic size, atomic radius, atomic covalent bond and van der Waals radius than other halogens, such as fluorine.[18] The large size of iodine atoms may negatively affect the drug receptor binding process by changing the desolvation energy, which plays an important role in ligand−receptor interactions.[19,20] Furthermore, fluorine improved the lipophilicity, absorption and bioavailability of many well-established anticancer drugs[16] such as lapatinib, gefitinib and caneratinib.[17,21] We therefore decided to synthesise some novel fluorinated quinazolinones with the same structure as our previously reported compounds but with an iodine atom instead of a fluorine atom at position 6 of the quinazolinone moiety. Figure 1 shows the structural similarities between the previously reported anticancer quinazolinones (A, B), Thymitaq and the newly synthesized derivatives.[22,23] The present study is thus a continuation of our attempt to identify novel, safe and effective anticancer agents.

Materials and Methods

Synthesis

The strategy used to synthesise the compounds is shown in Scheme 1. It comprises two simple reactions, namely acetylation followed by ring closure of 2-amino-5-flurobenzoic acid (1). This compound was refluxed with acetic anhydride for 1 h to afford a quantitative yield of 6-fluoro-2-methyl-4H-benzo[d][1,3]oxazin-4-one (2). The second reaction is the nucleophilic displacement of the oxygen of benzoxazinone with the nitrogen of the amino group upon treatment with sulphonamides, which was achieved by refluxing compound (2) with the appropriate sulphonamide under dry conditions for 6 h and gave sulphonamide derivatives of 6-fluoro-2-methyl-quinazolinone (4−11) in variable yields ranging from 60 to 79%.

Chemistry

The compounds were analysed at the Analytical Centre, College of Science, Cairo University, Egypt. The melting points were measured using a Griffin melting point apparatus (Griffin) and are uncorrected. The Infrared spectra were recorded as KBr discs on a Nicolet IR 200 (Thermo Fisher Scientific). The ^{1}HNMR spectra were run using TMS as the internal standard (Sigma−Aldrich) on a Varian Mercury VXr-300 NMR instrument (Varian). The mass spectra were measured on a JEOL-SX-102 instrument through electron impact ionization. Elemental analyses (C, H, and N) were performed using a Perkin-Elmer 240C analyser (Perkin-Elmer). All of the values were within ±0.4% of the theoretical values. All of the chemicals were purchased from Sigma−Aldrich.

Experimental

6-Fluoro-2-methyl-4H-benzo[d][1,3]oxazin-4-one (2)

Compound 2 was prepared by refluxing 2-amino-5-fluorobenzoic acid (1, 1.55 g, 0.01 mol) with an appropriate amount of acetic anhydride for 1 h. The residue obtained was evaporated to complete dryness, allowed to cool, washed several times with petroleum ether, collected, filtered and dried without moisture.

Yield 79%, mp: 87−89 °C; ^{1}H NMR (DMSO-d_6): δ 1.41 (s, 3H, CH_3), 7.31−8.24 (m, 3H, Ar−H). ^{13}C NMR (DMSO-d_6): δ 24.23, 82, 90.5, 118.5, 138.2, 149.6, 150, 154.1, 158. Anal. Calcd. For $C_9H_6FNO_2$ (179.04): C, 60.34; H, 3.38; N, 7.82. Found C, 60.12; H, 3.61; N, 7.64. MS (EI) m/z 180.04 [M + 1].

General method for preparation of test compounds (4−11)

Compounds 4−11 were prepared by mixing 1.79 g (0.01 mol) of compound (2) with 0.01 mol of sulphonamide derivatives in 100 ml of dry pyridine, refluxing for 6 h, cooling, treating with a small amount of 10% hydrochloric acid and pouring onto crushed ice. The crystals obtained were collected by filtration and re-crystallised from ethanol or glacial acetic acid.

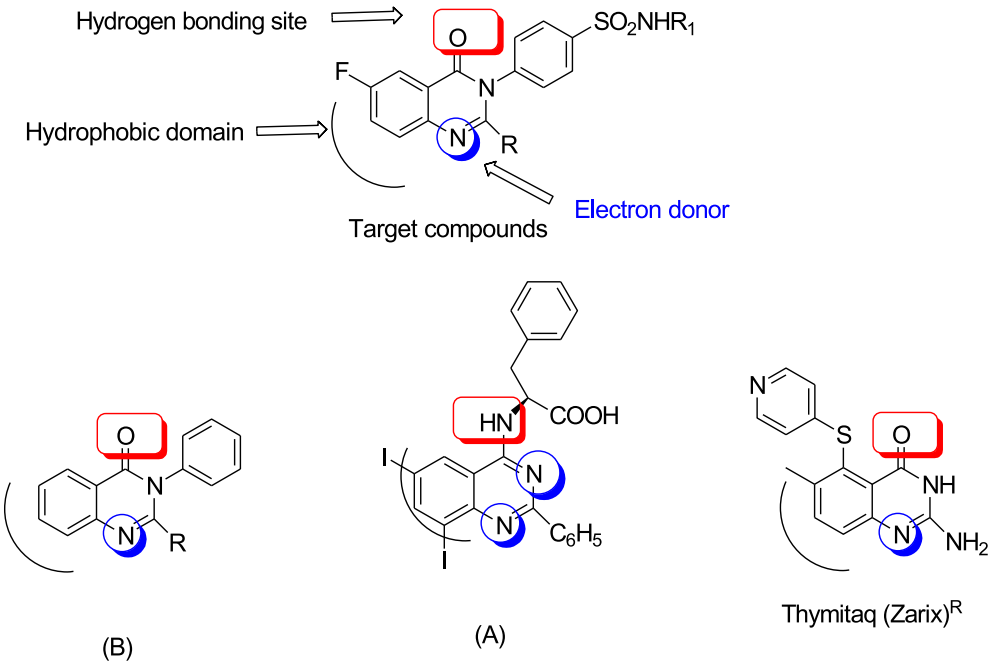


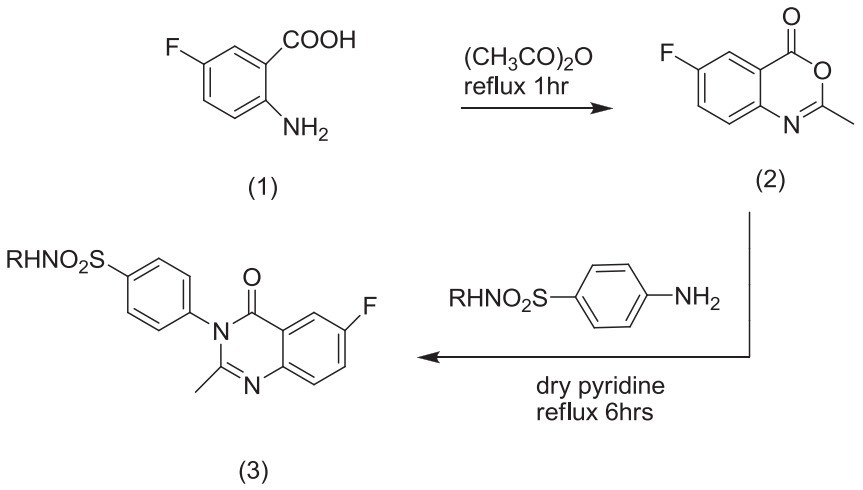
Figure 1: Structural similarities and pharmacophoric features of the reported and designed quinazolinones **(4–14)** as cytotoxic agents.

4-(6-Fluoro-2-methyl-4-oxoquinazolin-3(4H)-yl) benzensulphonamide (4)

Yield 68%, mp: 152–154 °C; ^{1}H NMR (DMSO-d_6): δ 2.46 (s, 3H, CH_3), 5.2 (s, 2H, NH_2), 7.02–7.94 (m, 7H, Ar–H). Anal. Calcd. for $C_{15}H_{12}FN_3O_3S$ (333.06): C, 54.05; H, 3.63; N, 12.61. Found C, 53.91; H, 3.82; N, 12.49. MS (EI) m/z 334.06 [M + 1].

4-(6-Fluoro-2-methyl-4-oxoquinazolin-3(4H)-yl)-N-(pyridin-2-yl) benzensulphonamide (5)

Yield 65%, mp: 161–163 °C; ^{1}H NMR (DMSO-d_6): δ 2.12 (s, 3H, CH_3), 7.02–8.25 (m, 11H, Ar–H), 8.41 (s, 1H, NH). Anal. Calcd. For $C_{20}H_{15}FN_4O_3S$ (410.08): C, 58.53; H, 3.68; N, 13.65. Found C, 58.62; H, 3.71; N, 13.83. MS (EI) m/z 411.08 [M + 1].



4) R = Hydrogen
5) R = Pyridin-2-yl
6) R = 3,4-dimethylisoxazol-5-yl
7) R = Thiazole-2-yl
8) R = 5-methylisoxazol-3-yl
9) R = 4,6-dimethlpyrimidin-2-yl
10) R = 4-methylpyrimidin-2-yl
11) R = Diaminomethylene

Scheme 1: Synthesis of the target compounds.

4-(6-Fluoro-2-methyl-4-oxoquinazolin-3(4H)-yl)-N-(3,4-dimethylisoxazol-5-yl)benzensulphonamide (6)

Yield 62%, mp: 156–158 °C; ^{1}H NMR (DMSO-d_6): δ 2.27 (s, 6H, $2CH_3$), 2.51 (s, 3H, CH_3), 7.13–8.27 (m, 7H, Ar–H), 8.41 (s, 1H, NH). Anal. Calcd. For $C_{20}H_{17}FN_4O_4S$ (428.1): C, 56.07; H, 4.00; N, 13.08. Found C, 56.31; H, 3.87; N, 12.97. MS (EI) m/z 429.1 [M + 1].

4-(6-Fluoro-2-methyl-4-oxoquinazolin-3(4H)-yl)-N-(thiazole-2-yl)benzensulphonamide (7)

Yield 60%, mp: 174–176 °C; ^{1}H NMR (DMSO-d_6): δ 2.45 (s, 3H, CH_3), 7.12–8.43 (m, 9H, Ar–H), 8.61 (s, 1H, NH). Anal. Calcd. For $C_{18}H_{13}FN_4O_3S_2$ (416.04): C, 51.91; H, 3.15; N, 13.45. Found C, 51.81; H, 3.42; N, 13.25. MS (EI) m/z 417.04 [M + 1].

4-(6-Fluoro-2-methyl-4-oxoquinazolin-3(4H)-yl)-N-(5-methylisoxazol-3-yl)benzensulphonamide (8)

Yield 62%, mp: 164–166 °C; ^{1}H NMR (DMSO-d_6): δ 2.42 (s, 3H, CH_3), 2.76 (s, 3H, CH_3), 7.11–8.24 (m, 8H, Ar–H), 8.41 (s, 1H, NH). Anal. Calcd. For $C_{19}H_{15}FN_4O_4S$ (414.08): C, 55.07; H, 3.65; N, 13.52. Found C, 55.19; H, 3.91; N, 13.44. MS (EI) m/z 415.08 [M + 1].

4-(6-Fluoror-2-methyl-4-oxoquinazolin-3(4H)-yl)-N-(4,6-dimethlpyrimidin-2-yl)benzensulphonamide (9)

Yield 65%, mp: 168–170 °C; ^{1}H NMR (DMSO-d_6): δ 2.31 (s, 3H, CH_3), 2.46 (s, 6H, $2CH_3$), 6.98–8.08 (m, 8H, Ar–H), 8.31 (s, 1H, NH). Anal. Calcd. For $C_{21}H_{18}FN_5O_3S$ (439.11): C, 57.39; H, 4.13; N, 15.94. Found C, 57.24; H, 4.51; N, 15.78. MS (EI) m/z 440.11 [M + 1].

4-(6-Fluoro-2-methyl-4-oxoquinazolin-3(4H)-yl)-N-(4-methylpyrimidin-2-yl)benzensulphonamide (10)

Yield 60%, mp: 163–165 °C; ^{1}H NMR (DMSO-d_6): δ 2.25 (s, 3H, CH_3), 2.64 (s, 3H, CH_3), 7.02–8.33 (m, 9H, Ar–H), 8.51 (s, 1H, NH). Anal. Calcd. For $C_{20}H_{16}FN_5O_3S$ (425.1): C, 56.46; H, 3.79; N, 16.46. Found C, 56.31; H, 3.62; N, 16.37. MS (EI) m/z 426.1 [M + 1].

N-(diaminomethylene)-4-(7-fluoro-2-methyl-4-oxoquinazolin-3(4H)-yl)benzensulphonamide (11)

Yield 60%, mp: 145–147 °C; ^{1}H NMR (DMSO-d_6): δ 2.31 (s, 3H, CH_3), 6.91–8.14 (m, 7H, Ar–H), 8.29–8.65 (m, 4H, $2NH_2$). Anal. Calcd. For $C_{16}H_{14}FN_5O_3S$ (375.08): C, 51.19; H, 3.76; N, 18.66. Found C, 51.03; H, 3.58; N, 18.81. MS (EI) m/z 376.05 [M + 1].

Biological screening

The newly synthesized compounds were screened for their cytotoxic activities using NCI (lung cancer cell line), MCF 7 (breast cancer cell line) and HEK293 (normal kidney cell line) cells by the MTT (3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyltetrazolium bromide) assay.[24] The screening experiments were conducted at the National Cancer Institute (NCI, Cairo, Egypt) and undertaken with approval from Taibah University, Al-madinah Al-munawarah, Saudi Arabia (approval #3006/434).

The cultures were inspected using an inverted microscope to determine the degree of viability. The absence of fungal and bacterial contaminants was confirmed. The cell monolayer was washed with phosphate buffer saline (PBS) without calcium and magnesium ions using a volume equivalent to half the volume of culture medium. A Trypsin/EDTA mixture was added to the washed cell monolayer using 1 ml per 25 cm^2 of surface area. The flask containing this mixture was rotated to cover the monolayer with trypsin and then incubated for 2–4 min. All of the cells were examined using an inverted microscope to confirm that they were detached and floated. The cells were suspended in a small volume of fresh serum containing HEK-293 medium, and 100–200 µl was removed for cell counting. The required number of cells were moved to a new labelled flask containing warmed HEK-293 medium and incubated as appropriate for the cell line. All of the cytotoxicity screening experiments were performed in 96-well plates. Methotrexate was used as a reference standard for measuring the cytotoxic activity. The test compounds were prepared using 2% DMSO solution. The IC_{50} values, which are the drug concentrations causing a 50% inhibition of cell proliferation, were calculated.

Cytotoxic assay

Cytotoxic assays were performed according to the reported method.[24] First, the cells were incubated at a concentration of 1×10^6 cells/ml in culture medium for 3 h at 37 °C and 6.5% CO_2. After incubation, the cells were seeded at a concentration of 5×10^4 cells/well in 100 µl of culture medium with different concentrations (0.005–100 µM/ml) of the reference drug methotrexate and the synthesized compounds, which were dissolved in 2% DMSO (dimethylsulphoxide) solution into microplates (tissue culture grade, 96 wells, flat bottom) and incubated for 24 h at 37 °C and 6.5% CO_2. The cell proliferation process is based on the ability of the mitochondrial succinate tetrazolium reductase system to convert 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) to a blue-coloured formazan. The test indicates the survival of the cells after toxic exposure. Then, 10 µl of the MTT labelling mixture was added, and the mixture was incubated for 4 h at the same conditions. Each experiment was performed three times. Then, 100 µl of the solubilization solution was added into each well, and the mixtures were incubated overnight. The absorbance of the samples was measured by spectrophotometry using a microplate (ELISA) reader. The formazan wavelength is between 550 and 600 nm according to the filters used for the ELISA reader. The reference wavelength should be more than 650 nm. The value of IC_{50} was then calculated as the drug concentration that caused a 50% inhibition of cell proliferation. The HEK 293 cell line was used to determine the cytotoxic effects of the synthesised compound on the non-cancerous cells.

Statistical analysis

The data obtained are expressed as the means ± SE. The statistical significance of the data was evaluated by one-way analysis of variance (ANOVA) followed by Tukey's test as a post ANOVA multiple-comparison test of the IC_{50} of the all of the compounds against methotrexate (Sigma Stat, version 3; SPSS Inc.). A P value <0.05 was considered statistically significant.

Results

Compounds 4–11 were synthesized according to the reaction sequence illustrated in Scheme 1. The structures of the compounds were deduced by their ^{1}H NMR, IR and mass spectra, and their composition was determined through elemental analyses. The chemical shift and multiplicity patterns correlated well with the proposed structures, and the elemental analysis showed good agreement between the experimentally determined and the theoretically calculated values.

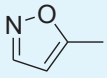
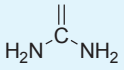
The results of the biological screening and statistical analysis (Table 1) showed that compounds 9 ($IC_{50} = 2.51 \pm 0.48$ μM), 5 ($IC_{50} = 3.27 \pm 0.89$ μM), and 7 ($IC_{50} = 4.57 \pm 0.64$ μM) had high activity on the NCI (lung cancer) cell line. Compound 9, which was the most potent compound, exhibited comparable activity to the standard drug methotrexate ($IC_{50} = 2.43 \pm 0.23$ μM). Compound 11 had the lowest activity ($IC_{50} = 25.48 \pm 0.72$). The order of the activities of the target compounds on the NCI cell line was the following: 9 < 5 < 7 < 8 < 10 < 6 < 4 < 11. The screening analyses on MCF-7 cells (breast cancer cell line) showed that compound 5 had the highest cytotoxic activity and compound 11 had the lowest activity. The order of the activities of the target compounds on the MCF-7 cell line was 5 < 9 < 7 < 8 < 10 < 4 < 6 < 11. The comparison of the compounds (5, 7, 8 and 9) showed comparable activity to methotrexate ($IC_{50} = 2.59 \pm 0.12$ μM). The assay on the HEK-293 normal cell line showed that compounds 9 ($IC_{50} = 6.81 \pm 0.61$ μM), 5 ($IC_{50} = 5.82 \pm 0.61$), and 7 ($IC_{50} = 13 \pm 0.95$ μM) killed less normal cells compared with methotrexate ($IC_{50} = 1.89 \pm 0.55$ μM). The C log P data for the tested compounds (Table 2) ranged from 3.10376 to 0.250538.

Table 1: Cytotoxic activity of the tested compounds.

Compound No.	Mean IC_{50} (μM) ± SE		
	NCI	MCF-7	HEK-293
4	16.91 ± 0.74***	14.97 ± 0.57***	12.31 ± 0.42***
5	3.27 ± 0.89	2.87 ± 0.91	5.82 ± 0.61***
6	12.32 ± 0.73***	16 ± 0.54***	14.82 ± 0.22***
7	4.57 ± 0.64	6.97 ± 0.11***	13 ± 0.59***
8	6.14 ± 0.81*	7.41 ± 0.72***	6.75 ± 0.66***
9	2.51 ± 0.48	3.42 ± 0.48	6.81 ± 0.61***
10	9.72 ± 0.82***	8.39 ± 0.57***	10.12 ± 0.41***
11	25.48 ± 0.72***	31 ± 0.41***	46.34 ± 0.23***
Methotrexate	2.43 ± 0.23	2.59 ± 0.12	1.89 ± 0.55***

* indicates significant difference at $P < 0.05$ and *** indicates significant difference at $P < 0.001$ compared with the reference methotrexate, as determined using ANOVA and Tukey's post-test.

Table 2: C log P values of the target compounds.

Compound No.	R	C log P value
4	H	1.85604
5		2.98086
6		2.30449
7		2.8276
8		2.64549
9		3.10376
10		2.60476
11		0.250538

Discussion

Cancer is the leading cause of death in economically developed countries and the second leading cause of death in developing countries.[14] The major challenges associated with the currently available anticancer agents include their selectivity and toxicity, resistance and the development of a secondary malignancy.[21] These drawbacks have motivated the search for newer, more efficacious, and better-tolerated antitumour drugs. Many techniques have been discovered and described for the synthesis of anticancer drugs, which include hybrid molecules and joined moieties. In a previous study, we described the synthesis of some hybrid molecules composed of a quinazolinone nucleus and amino acid derivatives.[15] These compounds were screened for their anticancer activities, and the results revealed good activities against different types of cell lines. In addition, these findings encouraged us to modify the structure of these compounds as a trial to produce more efficacious anticancer agents. The newly synthesised compounds were designed by joining two different moieties, namely quinazolinone and sulphonamide. The structure of these derivatives can be divided into two parts: the fixed part of the 6-fluoroquinazolinone moiety and the changed part of the sulphonamide moiety at the third position of the quinazolinone ring. The optimisation of these molecules was performed at the third position of quinazolinone using different sulphonamide derivatives, namely, sulphanilamide in compound (4), sulphapyridine in compound (5), sulphafurazole in compound (6), sulphathiazole in compound (7), sulphamethoxazole in compound (8), sulphamethazine in compound (9), sulphamerazine in compound (10) and sulphaguanidine in compound (11). These compounds were subjected to *in vitro* cytotoxic activity assays against two types of cancer cell lines: NCI (lung cancer cell line) and MCF-7 (breast cancer cell line) cells to measure their anticancer activity and one type of normal cell line HEK-293 (normal epidermal kidney cell line) to measure their cytotoxic activity

on normal cells. The anticancer activity was assayed using the MTT (3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyltetrazolium bromide) assay. The IC_{50} values for the test and standard compounds were calculated. In this study, compound 9 was found to be significantly active against the NCI (lung cancer) cell line, whereas compounds 5 and 7 were also significantly active but less active than compound 9. The other compounds exerted less activity on this cell line. Compound 5 was significantly active against MCF-7 (breast cancer cell line) while compounds 5 and 7 were also significantly active but less than compound 5. The other compounds exerted less activity on this cell line. Compound 9 had the lowest cytotoxic activity on the normal cell line (kidney HEK-293 cells), whereas the other compounds had higher cytotoxic activity on this cell line. Based on the findings of the present study, the authors concluded that compound 9 is the most active compound with the lowest cytotoxic activity on normal cells. This compound can be subjected to further optimisation and *in vivo* studies to understand its action. The relationship between the lipophilicity of the newly synthesised compounds and their biological activity was measured through the correlation of anticancer activity with the C log P values for all of the compounds. The C log P value expresses the degree of lipophilicity of the chemical compound. An increase in this value indicates an increase in the lipophilic character of the tested compound. This value was calculated using computerised models (Cambridge software). It is worthwhile to note that the C log P values for compounds 9, 5, 7 and 8 were higher than those of the other four compounds, namely 10, 6, 4 and 11. These values may explain the variation in their biological activity compared with their lipophilicity. Interestingly, the C log P values for the tested compounds agreed with their potency levels. Compound 9, the most active compound, had the highest C log P value, whereas compound 11, the least active compound, had the lowest C log P value. Based on these results, we noted a correlation between the anticancer activities of the target compounds and their lipophilic characters.

Based on the results of this study, the strategy for the synthesis of anticancer agents comprising the joining of two pharmacophoric groups of sulphonamides and fluorinated quinazolinone may be considered a promising way to produce significantly active compounds; in addition, increasing the lipophilicity of these compounds using lipophilic substituents increases the anticancer activity of the resulting compounds. Molecular optimisation, enzymatic assay and *in vivo* studies of compound 9 may be considered a future plan for producing an effective anticancer agent.

Conclusions

Eight novel quinazolinone–sulphonamide derivatives were synthesised and tested for their *in vitro* cytotoxic activity. All of the newly synthesized compounds had significant activity. Compound **9** showed promising activity and was found to be safety because it was able to kill cancerous cells more effectively than non-cancerous cells. Therefore, this compound can be optimised and evaluated through enzymatic assays and *in vivo* animal models for its future development into a prototype molecule of a new class of anticancer agents.

Contributions

The design of the study, the syntheses of the compounds, and all of the correspondence and editing were performed by M. Zayed. The purification of the compounds and the data analysis and interpretation were performed by H. Ahmed and S. Ihmaid. The biological screening and the revising of the manuscript were performed by A. Omar and A. Abdelrahim.

Conflict of interest

The authors have no conflict of interest to declare.

Acknowledgement

The authors would like to express their appreciation to the Deanship of Scientific Research at Taibah University for funding the work through Research Group Project No. 3006/434.

References

1. Bayomi AH, El-Helby AA, Abdel Rahman AA, et al. Synthesis and anticonvulsant activity of some new 2-styryl-4(3H)quinazolinone derivatives. **Egypt J Biomed Sci 2005**; 19: 216–225.
2. Zayed MF, Ahmed EA, Omar AM, et al. Design, synthesis, and biological evaluation studies of novel quinazolinone derivatives as anticonvulsant agents. **Med Chem Res 2013**; 22(12): 5823–5831.
3. Lemura R, Manabe H, Todayaki S. Bioisosteric transformation of H1-antihistaminic benzimidazole derivatives. **Chem Pharm Bull 1989**; 37: 2723–2726.
4. Zayed MF, Hassan MH. Synthesis and biological evaluation studies of novel quinazolinone derivatives as antibacterial and anti-inflammatory agents. **Saudi Pharm J 2014**; 22: 157–162.
5. Ram VJ, Farhanullah BK, Srivastava AK. Synthesis and antihyperglycemic activity of suitably functionalized 3H-quinazolin-4-ones. **Bioorg Med Chem 2003**; 1: 2439–2444.
6. Liu J, Wilson JC, Ye P, et al. Privileged structure based quinazolinone natural product-templated libraries: identification of novel tubulin polymerization inhibitors. **Bioorg Med Chem Lett 2006**; 16: 686–690.
7. Shang LC, Yu PF, Run TL, et al. Synthesis and in vitro antitumor activity of 4(3H)-quinazolinone derivatives with dithiocarbamate side chains. **Bioorg Med Chem Lett 2005**; 15: 1915–1917.
8. Connolly JD, Cusack D, Sullivan PT, et al. Synthesis of quinazolinones and quinazolines. **Tetrahedron 2005**; 61: 10153–10202.
9. Dinakaran M, Selvam P, Declercq E, et al. Synthesis, antiviral and cytotoxic activity of 6-bromo-2,3-disubstituted-4(3H)-quinazolinones. **Biol Pharm Bull 2003**; 26: 1278–1282.
10. Bano S, Javed K, Ahmad S, et al. Synthesis and biological evaluation of some new 2-pyrazolines bearing benzene sulfonamide moiety as potential anti-inflammatory and anti-cancer agents. **Eur J Med Chem 2011**; 46(12): 5763–5768.
11. Mert S, Yalýolu SA, Demirtas I, et al. Synthesis and antiproliferative activities of some pyrazole-sulfonamide derivatives. **Med Chem Res 2014**; 23(3): 1278–1289.
12. Stawinski J, Szafranski K, Vullo D, et al. Carbonic anhydrase inhibitors. Synthesis of heterocyclic 4-substituted pyridine-3-sulfonamide derivatives and their inhibition of the human cytosolic isozymes I and II and transmembrane tumor-

associated isozymes IX and XII. **Eur J Med Chem 2013**; 69: 701–710.
13. Xu F, Xu H, Wang X, et al. Discovery of N-(3-((7H-purin-6-yl) thio)-4-hydroxynaphthalen-1-yl)-sulfonamide derivatives as novel protein kinase and angiogenesis inhibitors for the treatment of cancer: synthesis and biological evaluation. Part III. **Bioorg Med Chem 2014**; 22(4): 1487–1495.
14. Supuran CT, Briganti F, Tilli S, et al. Carbonic anhydrase inhibitors: sulfonamides as antitumor agents. **Bioorg Med Chem 2001**; 9(3): 703–714.
15. Zayed MF, Hassan MH. Design, synthesis and biological evaluation studies of novel quinazoline derivatives as cytotoxic agents. **Drug Res 2013**; 63: 210–215.
16. Chundawat ST, Nutan SN, Sunita BS. Synthesis and in vitro antimicrobial evaluation of novel fluorine-containing 3-benzofuran-2-yl-5-phenyl-4,5-dihydro-1H-pyrazoles and 3-benzofuran-2-yl-5-phenyl-4,5-dihydro-isoxazoles. **Med Chem Res 2014**; 23(3): 1350–1359.
17. Buzalaf AM, Leite LA, Carvalho TN, et al. Bioavailability of fluoride administered as sodium fluoride or monofluorophosphate to humans. **J Fluor Chem 2008**; 129(8): 691–694.
18. Kirk LK. Fluorine in medicinal chemistry: recent therapeutic applications of fluorinated small molecules. **J Fluor Chem 2006**; 127(8): 1013–1029.
19. Englert L, Biela A, Zayed M, et al. Displacement of disordered water molecules from hydrophobic pocket creates enthalpic signature: binding of phosphonamidate to the S1'-pocket of thermolysin. **Biochim et Biophys Acta 2010**; 1800(11): 1192–1202.
20. Baum B, Mohamed M, Zayed M, Gerlach C, et al. More than a simple lipophilic contact: a detailed thermodynamic analysis of nonbasic residues in the S1 pocket of thrombin. **J Mol Biol 2009**; 390(1): 56–59.
21. Stepan AF, Walker DP, Bauman J, et al. Structural alert/reactive metabolite concept as applied in medicinal chemistry to mitigate the risk of idiosyncratic drug toxicity: a perspective based on the critical examination of trends in the top 200 drugs marketed in the United States. **Chem Res Toxicol 2011**; 24(9): 1345–1410.
22. Abou El Ella AD, Ghorab MM, Heiba IH, et al. Synthesis of some new thiazolopyrane and thiazolopyranopyrimidine derivatives bearing a sulfonamide moiety for evaluation as anticancer and radiosensitizing agents. **Med Chem Res 2012**; 21(9): 2395–2407.
23. Al-Obaid AM, Abdel-Hamide SG, El-Kashef HA, et al. Substituted quinazolines, part 3. Synthesis, in vitro antitumor activity and molecular modeling study of certain 2-thieno-4(3H)-quinazolinone analogs. **Eur J Med Chem 2009**; 44: 2379–2391.
24. Rathi SG, Suthar M, Patel P, et al. In-vitro cytotoxic screening of Glycyrrhiza glabra L. (Fabaceae): a natural anticancer drug. **J Young Pharm 2009**; 1: 239–243.