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ANTI-TUMOR POTENTIAL OF SUBSTITUTED 6-OXO(THIOXO)-6,7-DIHYDRO-2H-(1,2,4)TRIAZINO(2,3-C)QUINAZOLIN-2-ONES

Quinazoline moiety may be found in structure of drugs that reveal wide spectrum of biological activity[1], including anti-tumor effect[2]. Some of the quinazoline derivatives, namely 4-R-phenylaminoquinazolines («Gefitinib», «Erlotinib», «Vandetanib», «Lapatinib» etc) are inhibitors of CDK2 and p38 kinases, epidermal growth factor receptor, vascular endothelial growth factor and currently used in clinical oncology[3]. It should be mentioned, that antitumor activity of 4-R-phenylaminoquinazolines caused is determined by as nature of basic heterocyclic fragment, so by structure of aniline fragment in position 4. The presence of halogen, hydroxy-group or cyano-group in aniline moiety

1 http://www.drugbank.ca; Dan Wang, Feng Gao (2013). Quinazoline derivatives: synthesis and bioactivities. Chemistry Central Journal. 7 (95): 1–15; Asif, M. (2014). Chemical Characteristics, Synthetic Methods, and Biological Potential of Quinazoline and Quinazolinone Derivatives. International Journal of Medicinal Chemistry, 1–27; Ugale, V. G., & Bari, S. B. (2014). Quinazolines: New horizons in anticonvulsant therapy. European Journal of Medicinal Chemistry, 80, 447–501; Sukriti Srivastava, Sujiti Srivastava. (2015). Biological activity of Quinazoline: A Review. International Journal of Pharma Sciences and Research (IJPSR). 6(9): 1206–1213; Kumar Tiwary, B., Pradhan, K., Kumar Nanda, A., & Chakraborty, R. (2016). Implication of Quinazoline-4(3H)-ones in Medicinal Chemistry: A Brief Review. Journal of Chemical Biology & Therapeutics, 01(02); Gupta, T., Rohilla, A., Pathak, A., Akhtar, M. J., Haider, M. R., & Yar, M. S. (2018). Current perspectives on quinazolines with potent biological activities: A review. Synthetic Communications, 48(10): 1099–1127; Auti, P. S., George, G., & Paul, A. T. (2020). Recent advances in the pharmacological diversification of quinazoline/quinazolinone hybrids. RSC Advances, 10(68), 41353–41392.

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3 Jin, H., Dan, H.-G., & Rao, G.-W. (2018). Research progress in quinazoline derivatives as multi-target tyrosine kinase inhibitors. Heterocyclic Communications, 24(1): 1–10; Bhatia, P., Sharma, V., Alam, O., Manaithiya, A., Alam, P., Kahksha, Imran, M. (2020). Novel quinazoline-based EGFR kinase inhibitors: A review focussing on SAR and molecular docking studies (2015–2019). European Journal of Medicinal Chemistry, 112640.

is essential for the presence of anticancer activity as well. The introduction of additional functional groups to position 6 and 7 is also reasonable for improvement of pharmacokinetic properties, namely bioavailability and lipophilicity. Thus, introduction of vinyl, but-2-ynamide fragments to the molecule increase the metabolic and excretion rate and consequently decrease cumulation of compounds. At the same time presence of alkoxy-groups and saturated nitrogen-containing moities and oxygen containing heterocyclic fragments is essential for hydrophobic interaction with target enzyme[4]. Moreover, recently, it was shown that annulation of heterocyclic fragment to pyrimidine ring (bioisosteric substitution) resulted the extension of anti-cancer activity spectrum, increasing of anti-cancer activity and decreasing of the agent's toxicity[5].

The appearance of novel high effective quinazoline-containing anti-tumor drugs motivated scientist to intensify the studies in the field of substituted quinazolines synthesis and evaluation of their anti-cancer activity. At the same time condensed quinazolines during long period of time were out of the focus and currently anti-cancer potential of abovementioned compounds insufficiently known. [1,2,4]triazino[2,3-c]quinazolines are one of the most promising objects for searching of novel anti-cancer agents due to their synthetic availability, wide possibility for chemical modification and structural similarity to known anti-cancer agents. Considering abovementioned facts the synthesis of thio(oxo)-containing [1,2,4]triazino[2,3-c]quinazolines, their derivatization and study of their anti-cancer activity are interesting in scope of modern medicinal chemistry.

1. Substituted 3-R1-6-oxo(thioxo)-6,7-dihydro-2H-[1,2,4]triazino[2,3-c]quinazolin-2-one: methods of synthesis and chemical modification

The approaches used for synthesis of [1,2,4]triazino[c]quinazolines are based on the sequential formation of pyrimidine and triazine fragments, annulation of triazine cycle to quinazoline carcass or pyrimidine cycle to triazine moiety[6].

One of the common approaches used for annulation of additional heterocycle to quinazoline system based on (5+1)-condensation. Aldehydes, ketones, acetals, acyl halides, anhydrides, carbon disulfide, carbonyldiimidazole etc. were used as equivalent of 1,1-bielectrophilic synthon in reactions with 1,5-binucleophiles (-NCCCN-, -NCCNN-binucleophiles)[7].

4 Bhatia, P., Sharma, V., Alam, O., Manaithiya, A., Alam, P., Kahksha, … Imran, M. (2020). Novel quinazoline-based EGFR kinase inhibitors: A review focussing on SAR and molecular docking studies (2015–2019). European Journal of Medicinal Chemistry, 112640.

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6 Воскобойнік О.Ю., Коваленко С.І., Карпенко О.В., Скорина Д.Ю., Берест Г.Г., Носуле-нко І.С., Кривошей О.В. (2012). Методи синтезу триазинохіназолінів. Журнал органіч-ної та фармацевтичної хімії, 10(1): С. 3–18.

7 Karpenko V. O., Kovalenko S. I., Chekotylo O.O., Shishkina, S.V. (2007). A New One-Step Synthesis of 1,2,4-Triazino[2,3-c]quinazolines. HETEROCYCLES, 71(3), 619; Kovalenko S.I., Karpenko A.V., Krivoshey O.V., Shishkina S.V., Shishkin O.V. (2007). Novel Method for the Synthesis of [1,2,4]Triazino[4,3-c]quinazoline System. Synthetic Communications, 37: 3719–3727; Karpenko A.V., Kovalenko S.I., Shishkin O.V. (2009). Synthesis of spiro-fused (C5)-pyrazolino-(C6)-triazinones, a new heterocyclic system. Tetrahedron, 65(31): 5964–5972; Voskoboynik O.Yu., Karpenko O.V., Kovalenko S.I., Berest G.G., Ivchuk V.V., Shvets V.M. (2014). The structure optimization of [(2-R-quinazolin-4-ylidene)¬hydrazono]carboxylic acids and esters – intention of search a new class of substances with anticancer activity. Journal of Organic and Pharmaceutical Chemistry. 12(4): 3–20; Voskoboynik O.Yu., Skorina D.Yu., Shishkina S.V., Shishkin O.V., Kovalenko S.I., Ivchuk V.V. (2015). Features of interaction between 3-(2-aminophenyl)-6-R-1,2,4-triazin-5(2H)-ones and cyclic anhydrides of non-symmetric dicarboxylic. Journal of Organic and Pharmaceutical Chemistry, 13(1): 25–31; Voskoboynik

Substituted 3-(2-aminophenyl)-6-R_1-1,2,4-triazin-5(2*H*)-ones (1) were used as -NC-CCN-binucleophiles. Abovementioned compounds were obtained by nucleophilic degradation of substituted 3-R_1-2*H*-[1,2,4]triazino[2,3-*c*]quinazolin-2-ones or interaction of 2-aryl-[(3*H*-quinazolin-4-yliden)hydrazono]acetic acids esters with hydrazine hydrate[8].

It was shown that interaction of 3-(2-aminophenyl)-6-R_1-1,2,4-triazin-5(2*H*)-ones (1) with *N,N*-carbonyldiimidazole in anhydrous dioxane resulted corresponding 3-R_1-2*H*-[1,2,4] triazino[2,3-*c*]quinazolin-2,6(7*H*)-diones (2)[35]. Compounds 2 also may be obtained by interaction of compounds 1 chloroethylformiate in acetic acid (Scheme 1).

Unfortunately compounds 2 cannot be used as initial compounds for synthesis of corresponding thiones (3)[9]. Thus, interaction of compounds 2 with phosphorus pentasulfide in dioxane, xylene or DMF in the most case yielded the mixture of the products. The attempts to use *Lawesson's* reagent for obtaining of thio-derivatives 3 were not success as well. Considering the abovementioned facts 3-R_1-6-thioxo-6,7-dihydro-2H-[1,2,4]triazino[2,3-c]quinazolin-2-ones (3) were prepared *via* interaction of anilines 1 with carbon disulfide. The application of potassium/sodium xanthate as reagent allowed to obtain corresponding substituted potassium/sodium 3-R_1-2-oxo-2*H*-[1,2,4]triazino[2,3-*c*] quinazolin-6-thiolates (4). It should be noted, that abovementioned method has such advantages as: effectiveness, safeness, high yields and purity of the products.

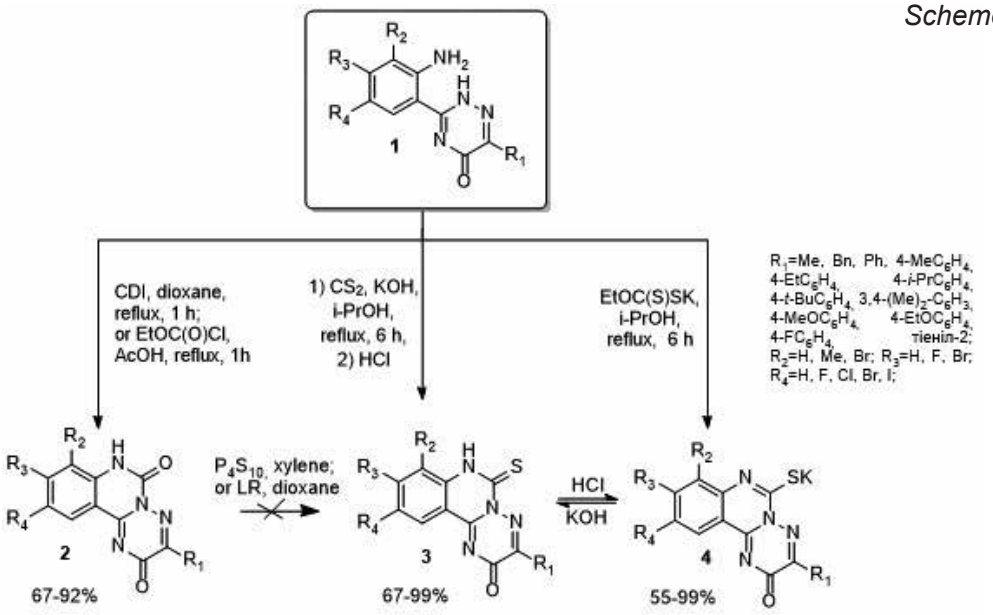
The elaboration of alternative synthetic procedures for thiones 3 that based on interaction of initial compounds (1) with aryl isothiocyanates in propanol-2 resulted mixture of 3-R_1-6-(phenylamino)-2*H*-[1,2,4]triazino[2,3-*c*]quinazolin-2-ones (5) and 3-R_1-6-thioxo-

A.Yu., Scorina D.Yu., Sergeieva T.Yu., Kovalenko S.I., Okovytyy S.I., Omelchenko I.V., Shishkin O.V. (2016). Interaction of 3-(2-Aminophenyl)-6-R1-1,2,4-triazin-5-ones with Acylating Reagents: An Efficient Method for Preparation of 6-Substituted 3-R1-2H-[1,2,4]triazino[2,3-c]quinazolin-2-ones. J. Het. Chem, 53(3): 776–783; Voskoboynik O.Yu., Starosyla S.A., Protopopov M.V., Volynets H.P., Shyshkina S.V., Yarmoliuk S.M., Kovalenko S.I. (2016). Synthesis, anticancer and FGFR1 inhibitory activity of isoindolo[2,1-a][1,2,4]triazino[2,3-c]quinazoline deri¬vatives. Медична та клінічна хімія, 18(1): 5–18; Voskoboynik O.Yu., Kovalenko S.I., Shishkina Sv.V. (2016). 3-R1-8-R2-10-R3-2H-benzo[e][1,2,4]triazino[2,3-c][1,2,3]triazin-2-ones – novel high electro-deficient heterocyclic compounds with promising anticancer activity. Heterocyclic Communications, 22(3): 137–141; Voskoboynik O.Yu., Kolomoets O.S., Kovalenko S.I., Shishkina S.V. (2017). [1,2,4]Triazino[2,3-c]quinazolines 1. Methods for the preparation and spectral characteristics of substituted 3-R1-6-R3-6,7-dihydro-2H-[1,2,4] triazino[2,3-c]-quinazolin-2-ones. Chemistry of Heterocyclic Compounds, 53(8): 892–904; Voskoboynik O.Yu., Kolomoets O.S., Palchikov V.A., Kovalenko S.I., Belenichev I.F., Shishkina S.V. (2017). [1,2,4]Triazino[2,3-c] quinazolines 2*. Synthesis, structure, and anticonvulsant activity of new 3′-R1-spiro[(aza/oxa/thia)cycloalkyl-1(3, 4),6′-[1,2,4]triazino[2,3-c]quinazolin]-2′(7′H)-ones. Chemistry of Heterocyclic Compounds, 53(10): 1134–1147; Voskoboynik O.Yu., Kovalenko S.I., Okovytyy S.I., Shishkina S.V. (2017). Dimethyl acetylenedicarboxylate in reactions with substituted 3-(2-amino¬phenyl)-6-phenyl-1,2,4-triazin-5-(2H)-ones: structure and properties of the products. J. Heterocyclic Chem, 54(3): 2038–2042; Voskoboynik O. Yu., Kolomoets O.S., Antypenko O.M., Zhernova G. O., Nosulenko I.S., Berest G.G., Shvets V.M., Kovalenko S.I. (2017). Synthesis and hypolipidemic activity of new 6,6-disubstituted 3-R-6,7-dihydro-2H-[1,2,4]triazino[2,3-c]quinazolin-2-ones. J. Heterocyclic Chem, 35(1): 318–325; Voskoboynik O.Y., Shishkina S.V., Kovalenko S.I. (2018). [1,2,4]Triazino[2,3-c] quinazolines 3*. Structure and anticancer activity of products obtained from reaction of 3-(2-aminophenyl)-6-R-1,2,4-triazin-5(2H)-ones with aryl iso(thio)cyanates. Chemistry of Heterocyclic Compounds, 54(7), 717–728; Stavytskyi V., Voskoboinik O., Antypenko O., Krasovska N., Shabelnyk K., Konovalova I., Shishkyna S., Kholodniak S., Kovalenko S. (2020). Tandem heterocyclization of 2-(azolyl-(azinyl-))anilines as an efficient method for preparation of substituted pyrrolo[1,2-a]azolo-(azino-)[c]quinazolines.. J. Heterocyclic Chem, 57(3):1249–1260. https://doi.org/10.1002/jhet.3862; Voskoboinik O.Yu., Stavytskyi V.V., Antypenko O.M., Kazunin M.S., Kravtsov D.V., Shishkina S.V., Kovalenko S.I. (2020). Synthesis and structure of pyrido[1,2-a] quinazoline condensed derivatives. Synthetic Communications, 50(13), 2043–2051.

8 Voskoboynik A.Yu., Berest G.G., Skorina D. Yu., Karpenko A.V., Kovalenko S.I. (2011). Synthesis of 6-R-3-(2-aminophenyl)-4H-[1,2,4]thriazin-5-ones: resources and limitations. Chemistry & Chemical Technology. Lviv Polytechnic National University, 5(2): 129–132; Sergeieva T.Yu., Voskoboynik O.Yu., Okovytyy S.I., Kovalenko S.I., Shishkina Sv.V., Shishkin O.V., Leszczynski J. (2014). Hydrazinolysis of 3-R-[1,2,4]Triazino[2,3-c]quinazolin-2-ones. Synthetic and Theoretical Aspects. J. Phys. Chem, A. 118: 1895–1905.

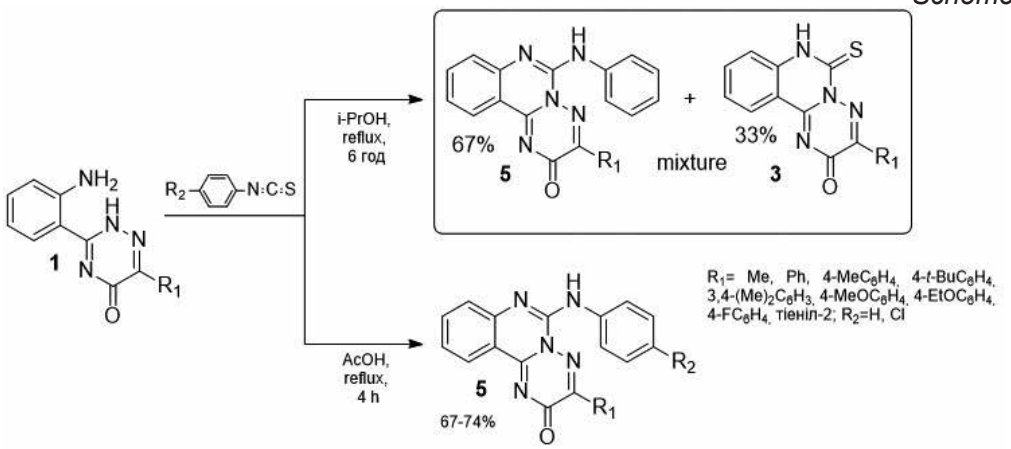
9 Berest G.G., Voskoboynic A.Yu., Kovalenko S.I., Sinyak R.S., Omelchenko I.V., Shishkin O.V., Komarovska-Porokhnyavets E., Novikov V.P. (2010). An Efficient Synthesis of 3-R-6-thio-6,7-dihydro-2H-[1,2,4]triazino[2,3-c]-quinazoline-2-ones and its Derivatives, Antimicrobial and Antifungal Activity. Журнал органічної та фармацевтичної хімії, 8(3): 42–52.

Scheme 1



6,7-dihydro-2*H*-[1,2,4]triazino[2,3-*c*]quinazolin-2-ones (3) in 2:1 ratio (Scheme 2)[10]. The usage of acetic acid as a solvent allowed to obtain the mixtures with less (10%) content of compounds 3. The admixture of thiones 3 easily may be removed by re-crystallization. Thus, abovementioned method may be considered as synthetic approach for of 3-R_1-6-(phenylamino)-2*H*-[1,2,4]triazino[2,3-*c*]quinazolin-2-ones (5).

Scheme 2

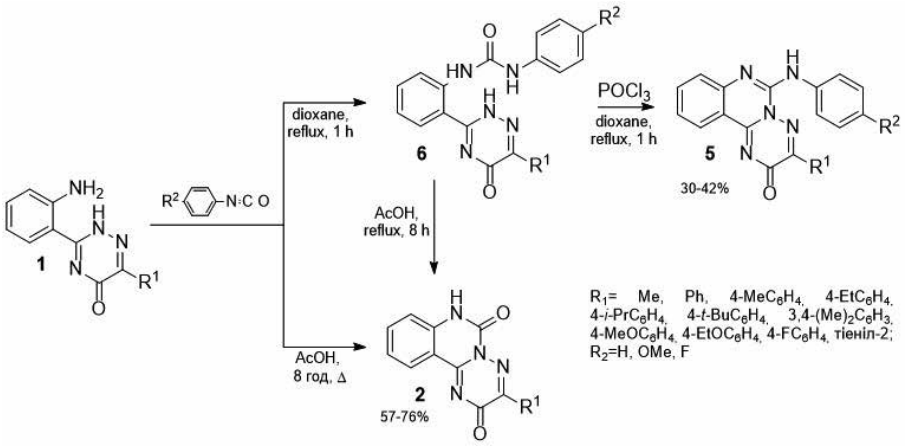


Besides, it was showed, that refluxing of compounds 1 with arylisocyanates in dioxane resulted the formation of corresponding urea 6. The modification of reaction conditions by changing of solvent to acetic acid and prolongation of heating period up to 8 hours resulted the formation of the products of following cyclization, namely - 3-R-2*H*-[1,2,4]triazino[2,3-*c*]quinazolin-2,6(7*H*)-diones (2, Scheme 3)[11].

10 Воскобойнік О.Ю., Шишкіна С.В., Коваленко С.І. (2018). [1,2,4]Триазино[2,3-c]хиназолины 3. Реакция 3-(2-аминофенил)-6-R-1,2,4-триазин-5(2H)-онов с арили-зо(тио)цианатами: строение и противораковая активность продуктов. Chemistry of Heterocyclic Compounds, 54(7): 717–728.

11 Там же.

Scheme 3



The elaboration of alternative cyclization of urea 6 showed, that refluxing of abovementioned compounds with equimolar quantity of phosphorus oxychloride in dioxane yielded 6-(arylamino)-3-R_1-2*H*-[1,2,4]triazino[2,3-*c*]quinazolin-2-ones 5 with low yield (30-42%).

Considering the structural novelty of 6-oxo-(thioxo)-*as*-triazinoquinazolines (2, 3) the method of their alkylation as one of the possible approaches for their chemical modification was elaborated. Alkylation of potassium 3-R-2-oxo-2*H*-[1,2,4]triazino[2,3-c]quinazoline-6-thiolates (4) by halogenalkanes, halogenoalcohols, halogenketones was conducted in propanole-2 – water (1:1) mixture and yielded the corresponding *S*-substituted derivatives 9 (Scheme 4)[12]. Reaction of potassium thiolates (4) with symmetric (1,2-dibromethane) and non-symmetric (1-bromo-2-chloroethane, 1-bromo-3-chloropropane, 1-bromo-4-chlorobutane) in same conditions proceeded without specifics and resulted 6-[(ω-halogenoalkyl) thio]-3-R-2*H*-[1,2,4]triazino[2,3-*c*]quinazoline-2-one (7) with yields (57–88%). It should be mentioned, that formation of bis-derivatives as the admixtures (up to 8%) were observed in conditions of above-mentioned reaction. Thiones 3 also may be alkylated by halogen-containing reagents in ethanol in presence of organic or inorganic base what resulted compounds 8 and 9 lower yields (32–94%).

Synthesis of 6-{[ω-dialkylamino-(heterocyclil-)alkyl]thio}-3-R-2*H*-[1,2,4]triazino[2,3-*c*]quinazolin-2-ones (10, 11) was conducted by reaction of potassium thiolate (4) with hydrochlorides of (2-chloroethyl)-*N*,*N*-dialkylamines or hydrochlorides 1-(2-chloroethyl)heterocyclil in propanol-2 – water (1:1) mixture in presence of organic base (Scheme 5)[13]. The alternative approach for synthesis of compounds 10 and 11 based on the interaction of 6-[ω-halogenalkyl]thio-3-R-2*H*-[1,2,4]triazino[2,3-*c*]quinazoline-2-ones (7) with saturated nitrogen-containing bases (pyrrolidine, piperidine, morpholine) in the presence of potassium iodide in dioxane. It should be mentioned, that yield of compounds 11 in abovementioned conditions were lower and ranged 31–74%.

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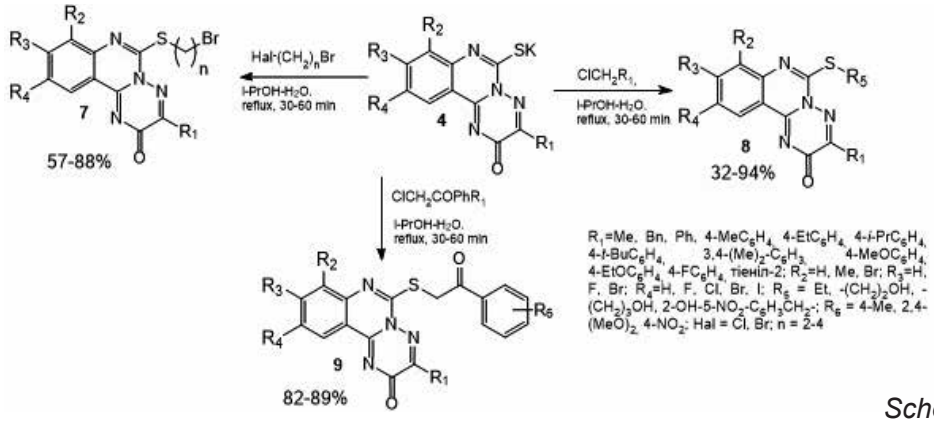
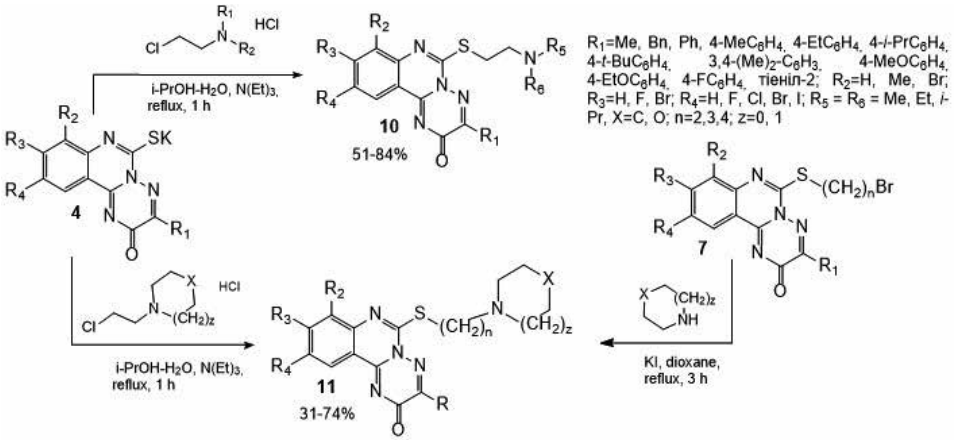
Scheme 4



Scheme 5

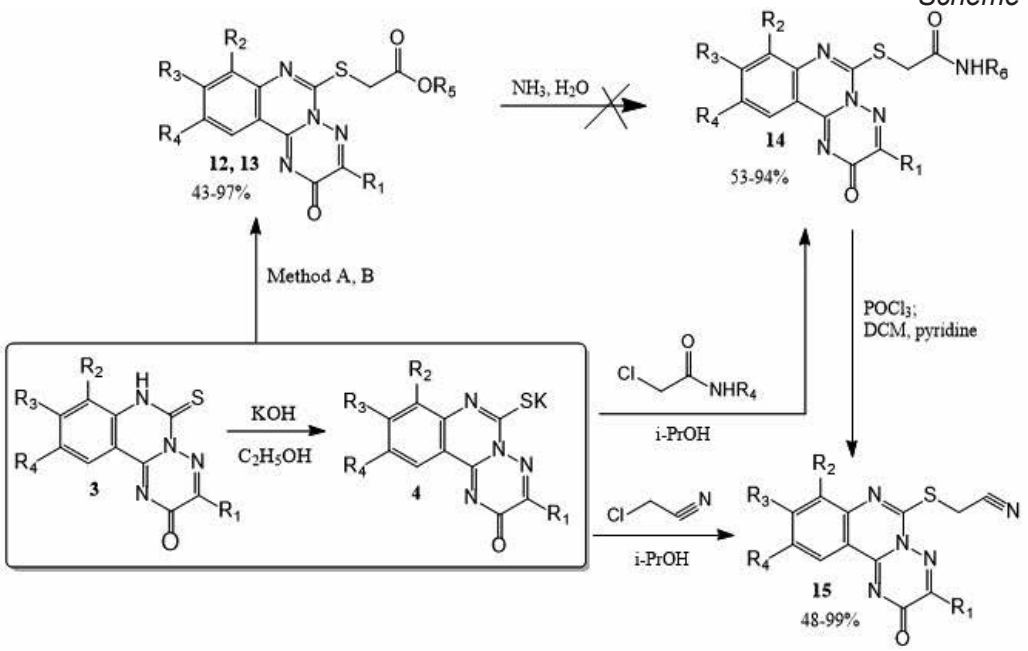


The interaction of thiolates (4) with chloracetic acid in water – alcohol mixture in presence of equimolar quantity of potassium hydroxide resulted formation of 2-((3-R_1-2-oxo-2*H*-[1,2,4]triazino[2,3-*c*]quinazolin-6-yl)thio)acetic acid3 (12) with high yields (method A, Scheme 6)[14]. At the same time target compounds 13-15 were prepared by alkylation of compounds 4 by corresponding functional derivatives of chloroacetic acid (esters, amides, nitriles) in propanol-2 medium (method B). It should be noted that thiones 3 also may be alkylated by chloroacetic acid derivatives in alcohol medium. Addition of potassium hydroxide is essential for conduction of abovementioned. However, the yields of the target products were significantly less and this approach cannot be considered as preparative. Subsequently amides were converted in corresponding nitriles 15 under action of the dehydrating (Scheme 6). It was shown, that esters of substituted 2-[(3-R-2-oxo-2*H*-[1,2,4]triazino[2,3-*c*]quinazoline6-yl)thio]acetic acids (13) can not be used as initial compounds for synthesis of corresponding amides due to their chemical inertness relative to ammonia or primary amines (Scheme 6).

Considering the mentioned above fact, the synthesis of cycloalkylamides (16) was conducted by alkylation of the potassium thiolates (4) *N*-cycloalkyl-(cycloaralkyl-)-2-

14 Voskoboynik O.Yu, Nosulenko I.S., Berest G.G., Kovalenko S.I. (2016). 6-Mono- and 6,6-disubstituted 3-R-6,7-dihydro-2H-[1,2,4]triazino[2,3-c]quinazoline-2-ones – promising class of anticancer agents. Український біофармацевтичний журнал. 2(43): 71–78.

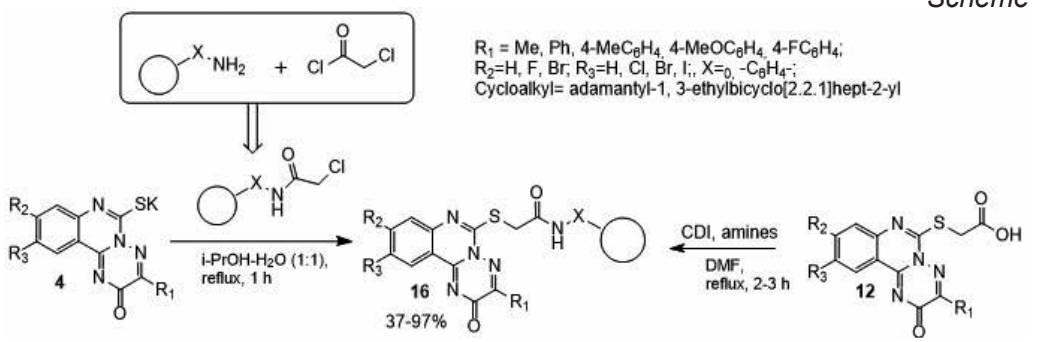
Scheme 6



R_1=Me, Bn, Ph, 4-MeC_6H_4, 4-EtC_6H_4, 4-*i*-PrC_6H_4, 4-*t*-BuC_6H_4, 3,4-$(Me)_2$-C_6H_3, 4-$MeOC_6H_4$, 4-$EtOC_6H_4$, 4-FC_6H_4, тієніл-2; R_2=H, Me, Br; R_3=H, F, Br; R_4=H, F, Cl, Br, I; R_5 = H, Et; R_6 = H, Et

chloracetamides in propanol-2 – water (1:1) mixture (Scheme 7)[15]. Besides, amides 16 were synthesized by aminolysis of generated «in situ»imidazolides of corresponding carboxylic acids 12 in anhydrous solvent.

Scheme 7

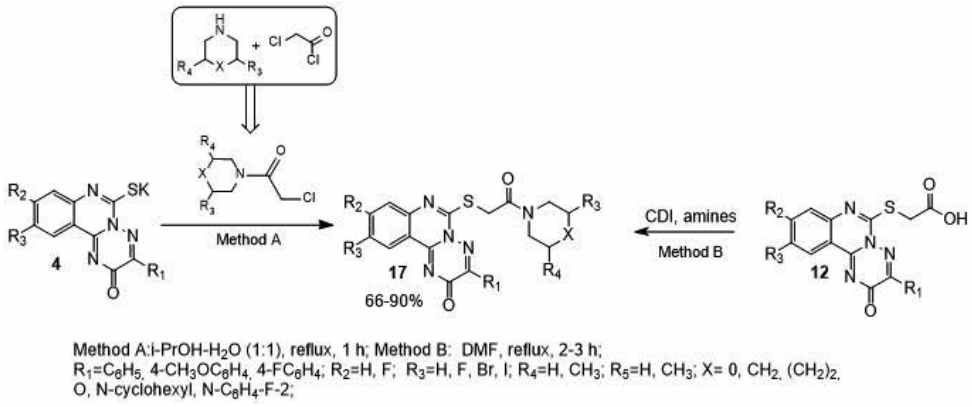


The synthesis of amides 17 was carried out by alkylation of potassium thiolates (4) with *N*-cyclyl-2-chloroacetamides (method A) and aminolysis of activated acids 12 (method B) with pyrrolidine, piperidine, morpholine and their substitutes (Scheme 8)[16]. The conducted experiments showed that these methods of synthesis do not have features, proceed under the conditions discussed above and duration in time.

15 Berest G.G., Voskoboynik A.Yu., Kovalenko S.I., Antypenko A.M., Nosulenko I.S., Katsev A. M., Shandrovskaya A.S. (2011). Synthesis and biological activity of novel N-cycloalkyl-(cycloalkylaryl)-2-[(3-R-2-oxo-2H-[1,2,4]triazino[2,3-c] quinazoline-6-yl)thio]acetamides. European Journal of Medicinal Chemistry. 46(2): 6066–6074; Nosulenko I.S., Voskoboynik O.Yu., Berest G. G., Safronyuk S..L., Kovalenko S. I., Katsev An. V., Sinyak R. S. (2014). Synthesis and antiviral activity [9-R1-10-R2-3-R-2-oxo-2H-[1,2,4]-triazino[2,3-c]quinazolin-6-yl)thio]acetamides derivatives with the fragments of carcase amines. Journal of Organic and Pharmaceutical Chemistry, 12(1): 17–27.

16 Nosulenko I.S., Voskoboynik O.Yu., Berest G.G., Safronyuk S.L., Kovalenko S.I., Kamysh-nyi O.M., Polishchuk N.M., Sinyak R.S., Katsev A.V. (2014). Synthesis 3-R-8-R1-9-R2-10-R3-R-6-thioxo-6,7-dihydro-2H-[1,2,4]

Scheme 8



Method A:i-PrOH-H_2O (1:1), reflux, 1 h; Method B: DMF, reflux, 2-3 h;
R_1=C_6H_5, 4-$CH_3OC_6H_4$, 4-FC_6H_4; R_2=H, F; R_3=H, F, Br, I; R_4=H, CH_3; R_5=H, CH_3; X= 0, CH_2, $(CH_2)_2$, O, N-cyclohexyl, N-C_6H_4-F-2;

The similar approaches, namely aminolysis generated «*in situ*» imidazolides of carboxylic acids 12 and alkylation of potassium thiolates (4) by preliminarily obtained 2-chloro-N-aryl-(benzyl-, phenetyl-)acetamides were used for preparation of *N*-aryl-(benzyl-, phenyl-)-2-[(3-R-9-R_1-10-R_2-2-oxo-2*H*-[1,2,4]triazino[2,3-*c*]quinazoline-6-yl) thio]acetamides (18, Scheme 9)[17]. The alkylation method is express, more flexible, able for scaling, reproduceable and is identified as more optimal for formation of combinatorial libraries despite the satisfying efficiency of both approaches.

The combination of privileged in medicinal chemistry heterocyclic fragments (oxazole, thiazole, thiadiazole) with insignificantly studied triazino[2,3-*c*]quinazoline system by «linker» functional (thioacetamide) group may be considered as systematic approach for searching of drug-like molecules. Considering the abovementioned initial compounds 4 were modified by reaction of *N*-azolyl-2-chloacetamides that were obtained via acylation of aminoazoles by 2-chloroacetyl chloride. Reaction proceeded fast (60-90 min) and was characterized by high yield and purity of target compounds 19-21 (Scheme 10)[18].

Promising anticancer agents 22 that combines in their structures [1,2,4]triazino[2,3-c]quinazoline system, thiadiazole cycle and aniline moiety were prepared by alkylation of thiolates 4 by *N*-{5-[(2-(R_1-anilino)-2-oxoethyl)thio]-1,3,4-thiadiazole-2-yl}-2-chloracetamides in dioxane (scheme 11)[19].

In continuation of our studies aimed to the creation of biologically active compounds with anticancer effect the triazino[2,3-c]quinazoline and pyrazole fragments were joined in single molecule by «linker» group. Especially since pyrazole derivatives are known as promising anti-tumor agents. Synthesis of substituted 23 was conducted by alkylation of potassium thi-

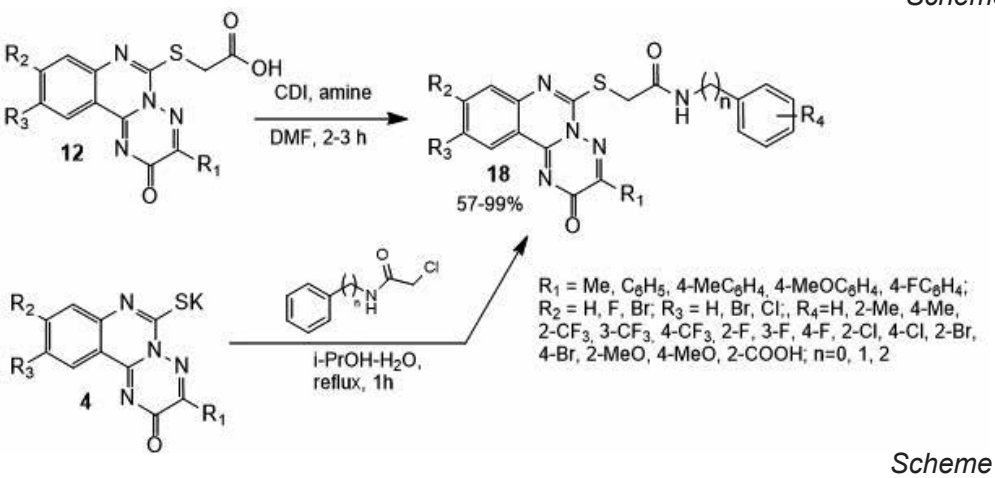
triazino[2,3-c]quinazolin-2-ones, its antibacterial and antifungal activity. Sci Pharm, 82: 483–500.

17 Kovalenko S. I., Nosulenko I. S., Voskoboynik A. Yu., Berest G. G., Antypenko L. M., Antypenko A. N., Katsev A. M. (2013). Novel N-aryl(alkaryl)-2-[(3-R-2-oxo-2H-[1,2,4]-triazino[2,3-c]quinazoline-6-yl)thio]-acetamides: synthesis, cytotoxicity, anticancer activity, compare analysis and docking. Medicinal Chemistry Research, 22(6): 2610–2632.

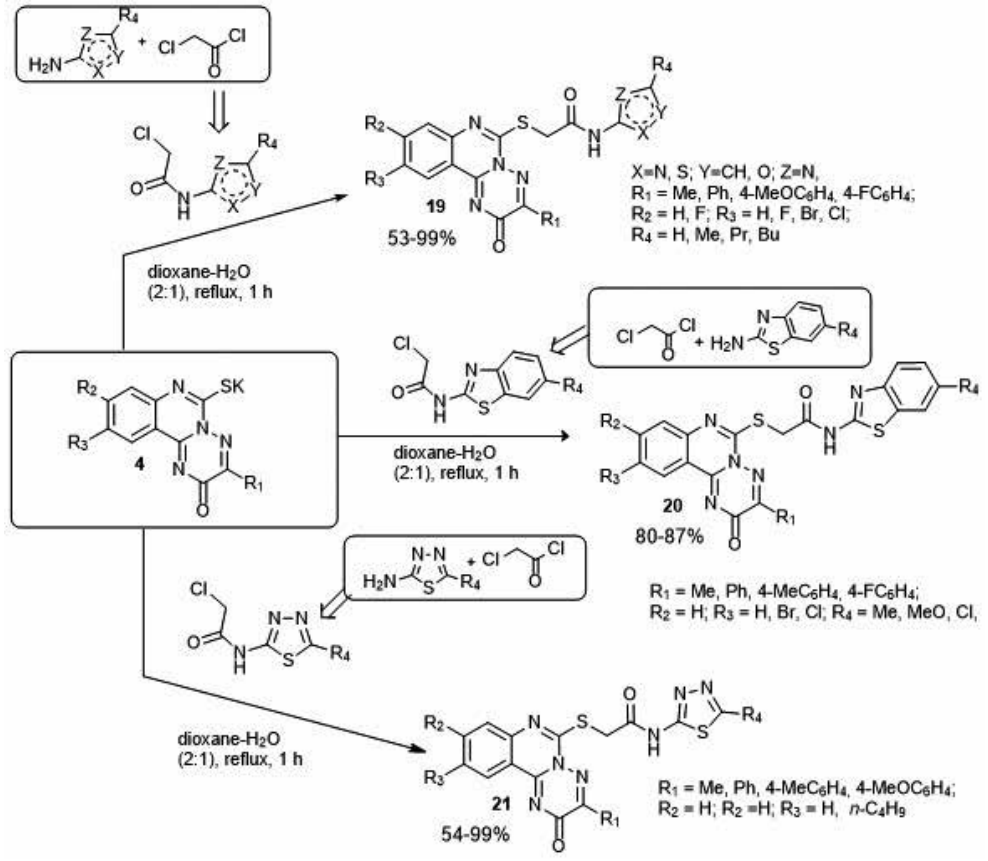
18 Kovalenko S.I. , Nosulenko S.S., Voskoboynik A.Yu., Berest G.G., Antypenko L.M., Antypenko A.N., Katsev A.M. (2012). N-R-2-[(3-R-2-oxo-2H-[1,2,4]triazino[2,3-c]quinazoline-6-yl)thio]acetamides with thiazole and thiadiazole fragments in a molecules. Synthesis, physico-chemical properties, cytotoxicity research by bioluminescence inhibition, anticancer activity. Sci. Pharm. 80: 837-865.

19 Kovalenko S.I. , Nosulenko S.S., Voskoboynik A.Yu., Berest G.G., Antypenko L.M., Antypenko A.N., Katsev A.M. (2012). N-R-2-[(3-R-2-oxo-2H-[1,2,4]triazino[2,3-c]quinazoline-6-yl)thio]acetamides with thiazole and thiadiazole fragments in a molecules. Synthesis, physico-chemical properties, cytotoxicity research by bioluminescence inhibition, anticancer activity. Sci. Pharm. 80: 837-865.

Scheme 9



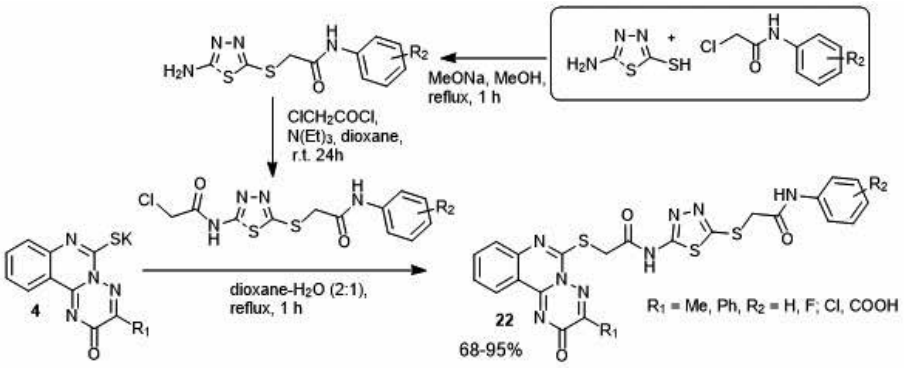
Scheme 10



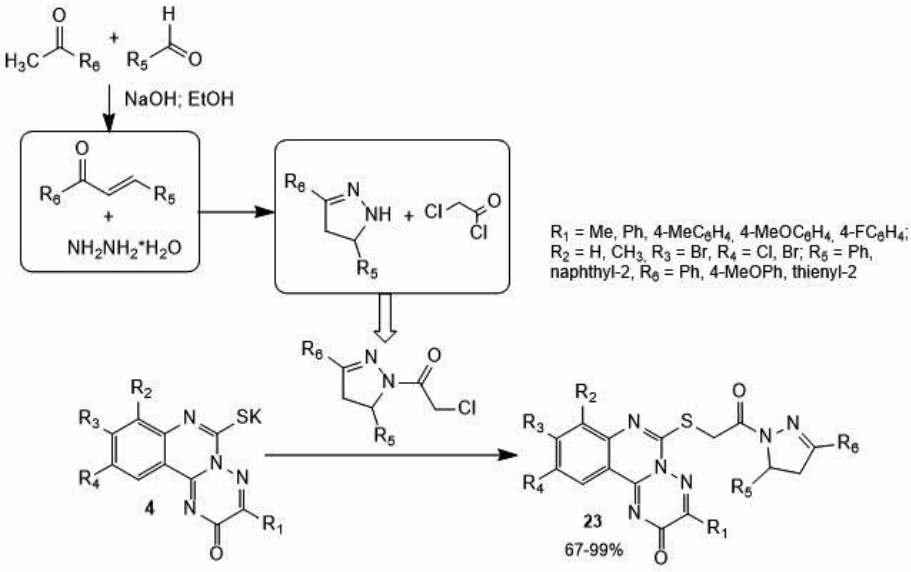
olates (4) by corresponding 1-(chloracetyl)-3-aryl-(hetaryl-)-5-aryl-4,5-dihydro-1*H*-pyrazoles (Scheme 12)[20]. The last ones were obtained by series of chemical transformations including synthesis of chalcones via condensation of methylaryl-(heteryl-)ketones with aromatic aldehydes, their transformation to 3-aryl(heteryl)-5-aryl-4,5-dihydro-1*H*-pyrazoles and acylation of abovementioned compounds by 2-chloroacetyl chloride.

20 Патент України на корисну модель №94614 МПК C07D 277/08 (2006.01).

Scheme 11



Scheme 12



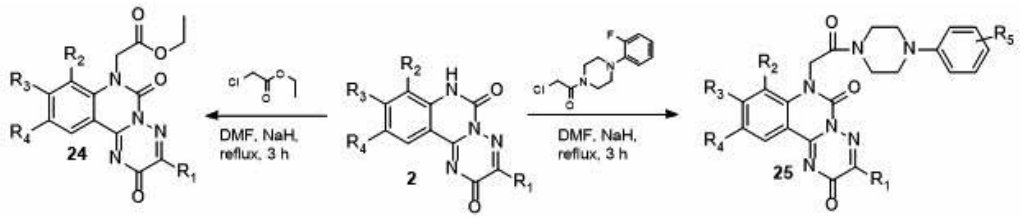
The features of 3-R_1-2*H*-[1,2,4]triazino[2,3-*c*]quinazoline-2,6(7*H*)-diones alkylation were studied as logical development of anticancer agents design strategy. It was shown that refluxing of abovementioned heterocyclic derivatives with ethyl 2-chloroacetate or 2-chloro-1-(4-(2-fluorophenyl)piperazin-1-yl)ethan-1-one in DMF with presence of sodium hydride allowed to obtain esters 24 and amides 25 (Scheme 13)[21]. At the same time reaction of compounds 2 with 2-chloro-*N*-aryl-(benzyl-)acetamides at the same conditions no reaction occurred what may be explained by presence of NH-acidic center in the molecules of acylating agents.

The introduction of amine or hydrazine group to the position 6 of the [1,2,4]triazino[2,3-c]quinazoline system is one more promising route of cytostatic drug development. The in detail study of the reactivity of S-alkylation products relative to *N*-nucleophiles showed, that long-term refluxing of compounds 8 in morpholine yielded corresponding 6-morpholino-derivatives (26)[22]. Besides, the refluxing of compounds 8 and 12

21 Voskoboynik O.Yu. (2015). Synthesis, physicochemical properties and anticancer activity of 6-(heterocyclyl-N-ylmethyl)-3-R1-9-R2-2H-[1,2,4]triazino[2,3-c]quinazolin-2-ones. Вопросы химии и химической технологии. 1 (99): 9–12.

22 Voskoboynik O.Yu. (2015). Synthesis, physicochemical and biological properties of 6-S- and 6-N-substituted 3-R-2H-[1,2,4]triazino[2,3-c]quinazolin-2-ones. Вопросы химии и химичес-кой технологии. 4(102): 9–16.

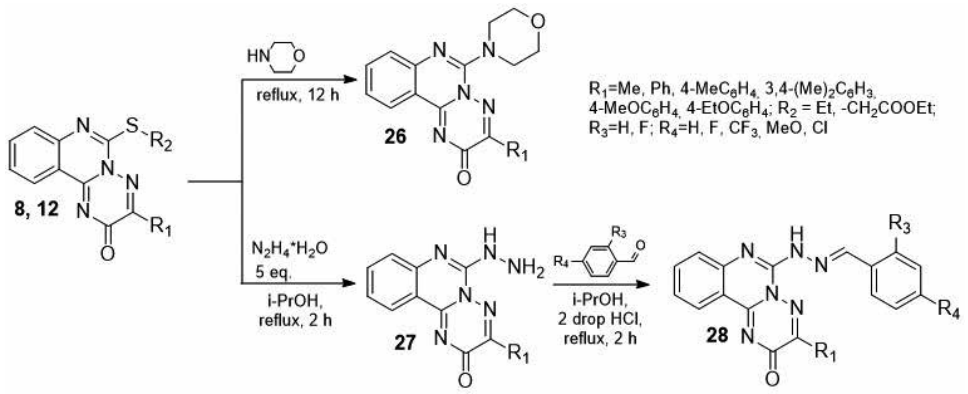
Scheme 13



R_1=Me, Ph, 4-$MeOC_6H_4$, 4-FC_6H_4; R_2=H, Me, Br; R_3=H, F, R_4=H, F, Cl; R_5 = 2-F, 4-F

with hydrazine hydrate in propanol-2 resulted formation of 6-hydrazino-3-R_1-2*H*-[1,2,4] triazino[2,3-*c*]quinazoline-2-ones (27, Scheme 14). Corresponding hydrazones 28 were obtained by reaction of compounds 27, шляхом with aromatic aldehydes.

Scheme 14



2. Spectral characteristics of 3-R_1-6-oxo(thioxo)-6,7-dihydro-2H-[1,2,4]triazino[2,3-c] quinazoline-2-ones their functional derivatives.

The IR spectra of compounds 2 and 3 were characterized by a band of valence vibrations of NH groups at 3494-3079 cm^{-1} and a band at 1743-1600 cm^{-1}, which correspond to the valence vibrations of νC=S(O)-groups. It should be mentioned that in IR-spectra of compounds 4 signals associated with the vibrations of the νC=S-group have a significant batochromic shift, which confirms the nature of the ionic bond. The absorption band at 1767–1700 cm^{-1} which correspond to the vibrations of νC = S-group were not observed in IR spectra of compounds 2, 5 and 6, what significantly distinguishes them from the initial compounds. However, in the spectra of some of the members of compounds 7 characteristic low-intensive absorbtion bands caused by stretching vibrations of the νS-R bond were observed at 2783–2432 cm^{-1}. Moreover, the IR-spectra of synthesized compounds 5-7 were characterized by stretching vibrations of νC = O-bond at 1695–1646 cm^{-1}. Usually, the above mentioned signals in IR-spectra of carboxylic acids and their derivatives have a significant hypsochromic shift. The characteristic bands associated with stretching symmetric and asymmetric vibrations of CH_2 groups at the range 2998-2800 cm^{-1}, deformation vibrations at the 1496–1470 cm^{-1} were observed in the IR spectra of compounds 8 and 9. IR-spectra of derivatives 8 and 9 that contain long aliphatic moieties were additionally characterized by absorption bands associated with rocking vibration at the 769-760 cm^{-1}. IR-spectra of halogen-containing com-

pounds 8 were characterized by the absorption band vC-Br at the 615–607 cm^{-1} or vC-Cl at the 750–700 cm^{-1}. In the spectra of alkylamines 9 the bands 1395-1001 cm^{-1} (stretching vibrations of the C-N bond) at 3070-3055 cm^{-1} and 1662-1608 cm^{-1} (stretching and deformation vibrations of the N-H bonds) were observed. Additionally, the following absorption band were observed in IR-spectra of studied compounds: vC = O- (1759-1658 cm^{-1}), vC-C (1520cm^{-1} and 1449 cm^{-1}), vCS (713-604 cm^{-1}), vCS and vCN (1593-710 cm^{-1}) and δCH (902–649 cm^{-1}). Abovementioned signals are associated with triazinoquinazoline system.

^{1}H NMR spectra of compounds 3 were characterized by the signal of thioamide group that were observed as broad singlet at 13.92–13.83 ppm. Signals of protons of the triazinoquinazoline cycle with appropriate chemical shifts and multiplicity (H-11 (d.), H-9 (t.), H-10 (t.) and H-8 (d.) were registered in spectra of compounds 3 as well. It should be mentioned that signals of H-8 and H-10 protons in spectra of the most of compounds have a similar chemical shift and observed as a multiplets. In ^{1}H NMR-spectra of compounds 4-23 the signals of the -S-CH_2 group protons were observed as singlet, triplet, quintet or multiplet at the 4.92–3.20 ppm. The multiplicity of abovementioned signals was caused by the proton environment and their chemical shifts by the donor-acceptor properties of the substituent.

In ^{13}C NMR-spectra of compounds 3 characteristic signals of the deshilded Carbon atoms at the positions 6 and 2 were observed at the 171.05–168.79 ppm and 158.68–160.25 ppm correspondingly and confirmed the formation of a new heterocyclic system. Additionally, signals of *sp*^{3}-hybridized Carbon atoms were registered in high field of compounds 3 ^{13}C NMR-spectra. According to chemical shifts the abovementioned signals correspond to the following fragments: CH_3- (20.04 ppm), C_6H_5-CH_2- (36.93 ppm), 4-$CH_3C_6H_4$ - (21.50 ppm), 4-$CH_3OC_6H_4$ - (55.90 ppm), 3,4-$(CH_3)_2C_6H_3$- (20.00 and 20.15 ppm, respectively). As for ^{13}C NMR-spectra *S*-substituted 5-23 characteristic signal of *sp*^{3}-hybridized Carbon atoms associated with the -S-CH_2 groups (34.21–38.94 ppm) were observed in high field. The signals of Carbons at the positions 6 and 2 were characteristic in the ^{13}C NMR spectra of compounds 9 and were observed 159–155 ppm and 160 ppm respectively. The structure of obtained compounds and *S*-regioselectivity of alkylation process were additionally confirmed by the signal -SCH2 group Carbon atom at the 32.31–28.70 ppm.

The mass-spectra (EI) of thions 3 has features and significantly differs from the spectra of described heterylsulfides[23]. The fragmentation of the molecular ion under electron impact occurs on C(2)–C (3) and N(4) –N(5) bonds and resulted the elimination methyl-(aryl-)nitrile radicals and the formation fragment ion with high intensive signal (I_{rel} 100-96%). Therefore, the triazine cycle is subject to destruction. Subsequently, the formed fragment ion eliminates the particles of S, SH, CNS, CHNS and CNO with the formation of ions with appropriate m/z value. The behavior of molecular ion of acids 12 under electron impact was similar to thions 3. The features in fragmentation of molecular ions of compounds 12 are associated with elimination of CO_2, COOH and SCH_2COOH. Abovementioned processes resulted the formation of following fragmentary ions: *m/z* 217 (I_{rel} = 100–75%), *m/z* 216 (I_{rel} = 88–20%), *m/z* 171 (I_{rel} = 100–48%).

23 Воскобойнік О.Ю., Карпенко О.В., Берест Г.Г., Скорина Д.Ю., Носуленко І.С., Ковале-нко С.І., Коломоєць О.С. (2013). Мас-спектральне дослідження поведінки [1,2,4]триазино[2.3-с]хіназолінів під дією електронного удару. Ученые записки ТНУ се-рия «Биология, химия». 26(65), 4: 229–241.

The X-ray study [(3-metthyl-2- oxo-2H-[1,2,4]triazino[2,3-c]quinazoline-6-yl)thio] acetic acid was conducted for unquestionable proof of it is chemical structure[24]. The final atomic coordinates, and crystallographic data for abovementioned molecule have been deposited to with the Cambridge Crystallographic Data Centre, and are available on request quoting the deposition numbers CCDC 766559).

3. In vitro screening of anticancer activity on 60 cancer cell lenes.

As a result of virtual prescreening of the synthesized compounds, 126 promising substances were selected for Phase I studies and tested at the US National Cancer Institute (DTP program, www.dtp.nci.nih.gov) on 60 human cancer cell lines at a concentration of 10.00 μM (the lines cover major human cancers, including: leukemia (CCRF-CEM, HL-60 (TB), K-562, MOLT-4, RPMI-8226, SR), non-small cell lung cancer (A549 / ATCC, EKVX, HOP) -62, HOP-92, NCI-H226, NCI-H23, NCI-H322M, NCI-H460, NCI-H522), colon cancer (COLO 205, HCC-2998, HCT-116, HCT-15, HT29, KM12 , SW-620), CNS cancer (SF-268, SF-295, SF-539, SNB-19, SNB-75, U251), melanoma (LOX IMVI, MALME-3M, M14, MDA-MB-435, SK -MEL-2, SK-MEL-5, SK-MEL-28, UACC-62, UACC-257), ovarian cancer (IGROV-1, OVCAR-3, OVCAR-4, OVCAR-5, OVCAR-8, NCI / ADR-RES, SK-OV-3), kidney cancer (786/0, A498, ACHN, CAKI-1, RXF 393, SN12C, UO-31), prostate cancer (PC-3, DU-145) and breast cancer (MCF-7, MDA-MB-231 / ATCC, HS 578T, BT-549, T-47D, MDA-MB-468). The experiment was performed by estimating of the quantity of cells by fluorescent method (dye - sulforodamine B, standards - 5-fluorouracil and adriamycin)[25]. The results were presented as the percentage of cell growth of each line and the average value for all lines.

Preliminary studies proved the antitumor activity of S-substituted 3-R_1-6-thio-6,7-dihydro-2*H*-[1,2,4]triazino[2,3-*c*]quinazoline-2-ones and allowed to select 22 high active compounds for the next stage of research (Fig. 1).

In phase II selected compounds were studied in five concentrations at 10-fold dilution (100μM, 10μM, 1.0μM, 0.1μM, 0.01μM) on 57–59 cell lines of 9 types of cancer. According to the result of the experiment, three dose-dependent parameters were calculated: GI_{50}, TGI, LC_{50}.The mean values of the experiment (mean graph midpoints, MG_MID) were calculated for each of the parameters.

For studied compounds mean value of growth inhibition rate (MG_MID) was at the range $lgGI_{50}$ -4.50 – -5.67. Among 6-{[ω-dialkylaminialkyl]thio}-3-R-2*H*-[1,2,4triazino[2,3-*c*]quinazolin-2-ones (Ia, IIa, IIb) the highest lg GI_{50} values were detected for compound Ia against cell line SNB-75 CNS cancer (lg GI_{50}=-6.07), CAKI-1 kidney cancer (lg GI_{50}=-5.94), IIa – against cell line A498 kidney cancer (lg GI_{50}=-6.29), IIb – against cell line HOP-92 non-small cell lung cancer (lg GI_{50}=-6.20), HCT-116 (lg GI_{50}=-5.93) and HT29 (lg GI_{50}=-5.96) colon cancer[26].

Among tested *N*-alkyl-(aryl-, benzyl-)-2-[(3-R-2-oxo-2H-[1,2,4]triazino[2,3-c] quinazolin-6-yl)thio]acetamides (VIIa-VIIj) the highest values of lg GI_{50} were detected

24 Berest G.G., Voskoboynic A.Yu., Kovalenko S.I., Sinyak R.S., Omelchenko I.V., Shishkin O.V., Komarovska-Porokhnyavets E., Novikov V.P. (2010). An Efficient Synthesis of 3-R-6-thio-6,7-dihydro-2H-[1,2,4]triazino[2,3-c]-quinazoline-2-ones and its Derivatives, Antimicrobial and Antifungal Activity. Журнал органічної та фармацевтичної хімії, 8(3): 42–52.

25 Boyd M. R. (1997). The NCI in vitro anticancer drug discovery screen. Concept, implemen-tation and operation. Humana Press. 2: 23–43.

26 Berest G.G., Voskoboynik O.Yu., Kovalenko S.I., Nosulenko I.S., Antypenko L.M., Antypenko O.M., Shvets V.M., Katsev A.M. (2012). Synthesis of new 6-{[—(dialkylamino-(heterocyclyl)alkyl]thio}-3-R-2H-[1,2,4]triazino[2,3-c]quinazoline-2-ones and evaluation of their anticancer and antimicrobial activities. Scientia Pharmaceutica. 80(1): 37–65.

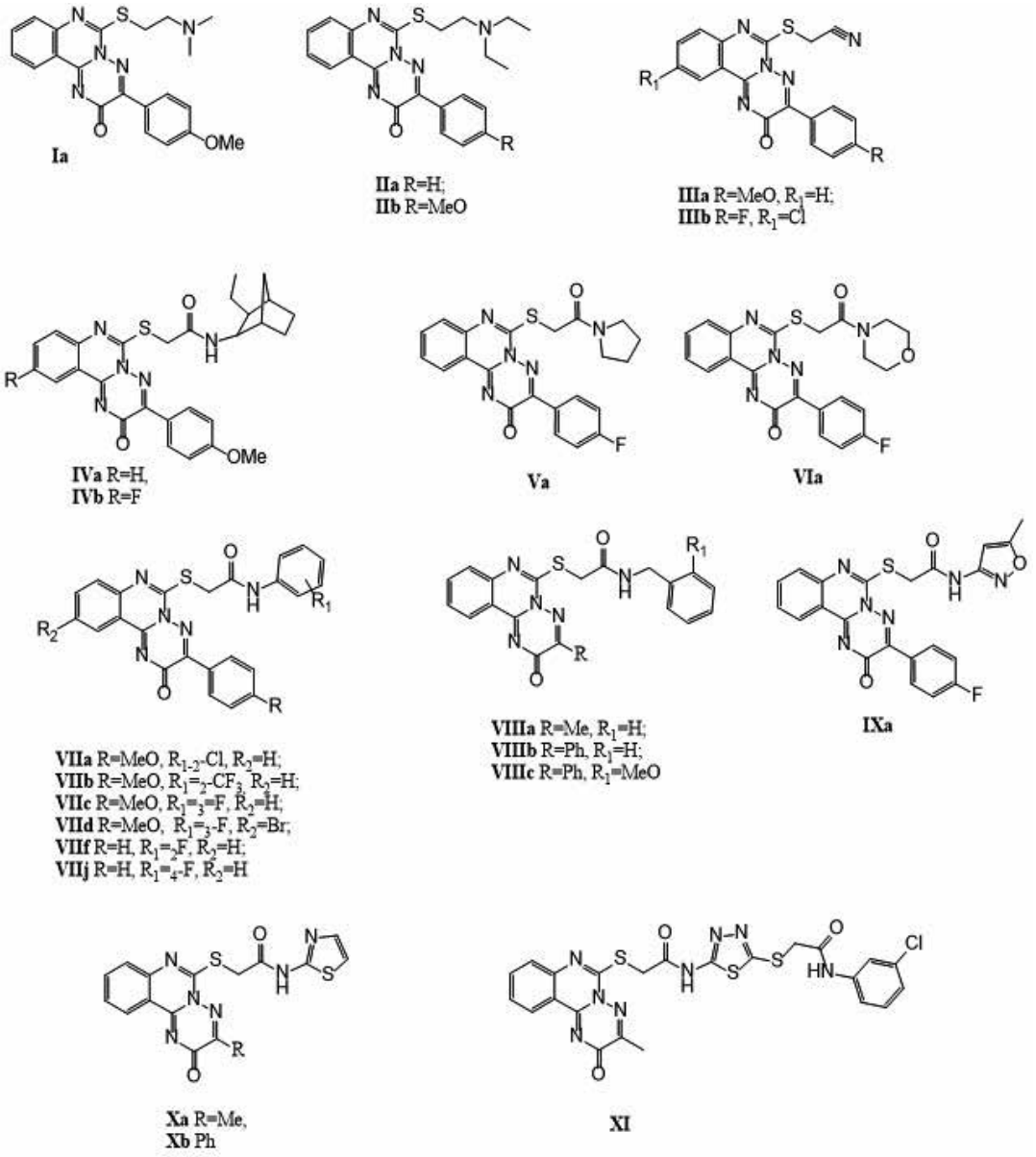


Figure 1. «Lead-compounds» that were studied on cancer cell line at the range of concentrations

for compounds : VIIa – against cell line HOP-92 of non-small lung cancer (lg GI_{50}=-6.01), VIIc – against cell line A498 of kidney cancer (lg GI_{50}=-7,57), VIIf – against cell line A498 of kidney cancer (lg GI_{50}=-5.93)[27].

N-(5-methylisoxazolyl-3-), (4-R_1-thiazolyl-2-), (5-R_1-1,3,4-thiadiazolyl-2-)-2-[(3-R-2-oxo-2H-[1,2,4]triazino[2,3-*c*]quinazolin-6-yl)thio]acetamides (IXa, Xa, Xb, XIa) were also

27 Kovalenko S. I., Nosulenko I. S., Voskoboynik A. Yu., Berest G. G., Antypenko L. M., Antypenko A. N., Katsev A. M. (2013). Novel N-aryl(alkaryl)-2-[(3-R-2-oxo-2H-[1,2,4]-triazino[2,3-c]quinazoline-6-yl)thio]-acetamides: synthesis, cytotoxicity, anticancer activity, compare analysis and docking. Medicinal Chemistry Research, 22(6): 2610–632.

active against proper cancer cell lines[28]. Thus compound Xa inhibited growth of following cell lines: K-562 (lg GI_{50}=-6.47), SR (lg GI_{50}=-6.42) of leukemia, NCI-H522 (lg GI_{50}=-6.65) of non-small cell line cancer, COLO 205 (lg GI_{50}=-6.30), HCT-116 (lg GI_{50}=-6.35), HT29 (lg GI_{50}=-6.39), KM12 (lg GI_{50}=-6.31) of colon cancer, MALME-3M (lg GI_{50}=-6.28), SK-MEL-5 (lg GI_{50}=-6.32) melanoma, OVCAR-3 (lg GI_{50}=-6.59) of ovarian cancer, A498 (lg GI_{50}=-6.17) of kidney cancer, MCF7 (lg GI_{50}=-6.32), MDA-MB-468 (lg GI_{50}=-6.51) of breast cancer. It should be mentioned that compound Xa reveal the broadest spectrum of anticancer activity (MG_MID lg GI_{50}=-5.67). Thus abovementioned compound effectively inhibited growth of leukemia cells (lg GI_{50}=-5.67), lung cancer cells (lg GI_{50}=-5.55), colon cancer cells (lg GI_{50}=-6.01), melanoma cells (lg GI_{50}=-5.65), ovarian cancer cells (lg GI_{50}=-5.87), prostate cancer cells (lg GI_{50}=-5,58) and breast cancer cells (lg GI_{50}=-5.61).

Considering the abovementioned facts antitumor potential of the studied compounds and the possible prospects of profound pharmacological studies, we calculated quantitative indicators of selectivity of growth inhibitor effects against different types of tumors based on experimental results of high-throughput screening. The results of these calculations show that only some of compounds are characterized by a moderate and high level of selectivity against non-small cell lung cancer (13.6, SI=4.52), CNS cancer (12.20, SI=3.25; 13.2, SI=6.75; 13.6, SI=5.34; 13.4, SI=4.63), melanoma (13.6, SI=3.43), kidney cancer (13.6, SI=8.56), prostate cancer (13.6, SI=4.98) breast cancer (13.6, SI=4.72).

4. SAR- and QSAR-analysis of studied compounds.

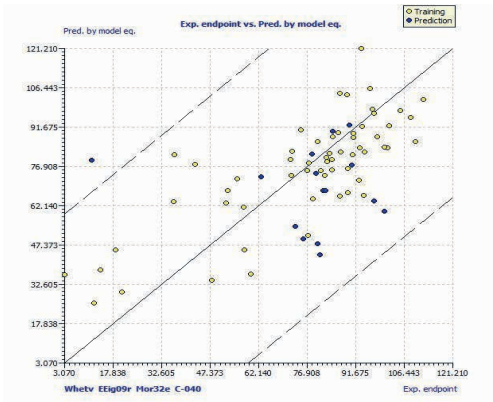
as-Triazino[2,3-*c*]quinazolines may be considered as promising heterocyclic compounds with antitumor activity and are interesting objects of structural optimization and chemical modification. SAR analysis showed that the antitumor activity of the synthesized compounds is determined both by structure of the basic heterocycle (as-triazino[2,3-*c*]quinazoline system) and by nature of substituents in the positions 3 and 6. Thus, one of the optimal approaches to increase the antitumor activity is the functionalization of the substituent in the position 3, namely, the introduction of an aryl fragment with various groups (alkyl-, ethoxy-, trifluoromethyl-, halogen groups, etc.). Besides, it is important to functionalize the substituent in the position 6, by diversification of the combinatorial library of amines used for introduction of amide fragment in molecule, by replacing the thio group by an oxo-, alkyl- (aryl-, heteryl) amino-, hydrazino group, etc and by introduction of carboxyalkyl substituents to position 7 and their subsequent functionalization. The introduction of substituents at positions 8, 9, 10, 11 of the triazinoquinazoline system also may be considered as promising approaches for functionalization of «leader compounds». Similar approaches were used to enhance the level and expand the spectrum of kinase inhibitory activity of 4-anilinoquinazolines[29].

28 Kovalenko S.I. , Nosulenko S.S., Voskoboynik A.Yu., Berest G.G., Antypenko L.M., Antypenko A.N., Katsev A.M. (2012). N-R-2-[(3-R-2-oxo-2H-[1,2,4]triazino[2,3-c]quinazoline-6-yl)thio]acetamides with thiazole and thiadiazole fragments in a molecules. Synthesis, physico-chemical properties, cytotoxicity research by bioluminescence inhibition, anticancer activity. Sci. Pharm. 80: 837–865.

29 Ravez, S., Castillo-Aguilera, O., Depreux, P., & Goossens, L. (2015). Quinazoline derivatives as anticancer drugs: a patent review (2011 – present). Expert Opinion on Therapeutic Patents, 25(7): 789–804; Hameed, A., Al-Rashida, M., Uroos, M., Ali, S. A., Arshia, Ishtiaq, M., Khan, K. M. (2018). Quinazoline and quinazolinone as important medicinal scaffolds: a comparative patent review (2011–2016). Expert Opinion on Therapeutic Patents, 28(4): 281–297.

3-R-6-thioxo-2*H*-[1,2,4] triazino [2,3-c]quinazolin-2-ones and their *S*-substituted with established antitumor activity in vitro were selected for QSAR analysis (126 compounds). The obtained data about inhibition of cancer cells growth at a concentration of 10^{-5} M were used to create QSAR models. Descriptors were calculated using Dragon software (> 1600 descriptors). Definition of all used descriptors and protocols of their calculations are described[30]. The affinity of synthesized compounds to the epidermal growth factor receptor (EGFR) and protein kinase CK2 were calculated by molecular docking method using Autodock software.

Six QSAR-models were obtained for leukemia (MOLT-4 and SR cell lines), non-small cell lung cancer (EKVX and NCI-H522 cell lines), colon cancer (KM12 cell lines), ovarian (SK12 cell lines) -3) and chest (cell lines MDA-MB-468) (Fig. _). The obtained equations contain 7 descriptors. Most of the descriptors used to form the model belong to the 3D type (RDF, 3D-MoRSE, WHIM, GETAWAY, etc.). Thus, the results of the research showed that the antitumor activity of the synthesized compounds is determined not only by the presence of a «pharmacophore», but also by their electronic environment.



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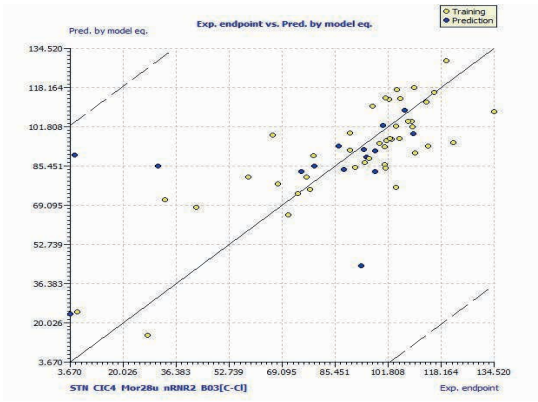

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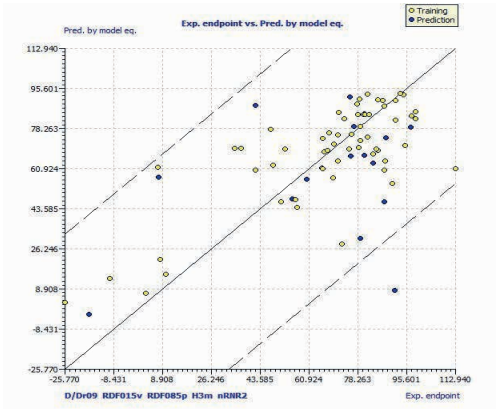

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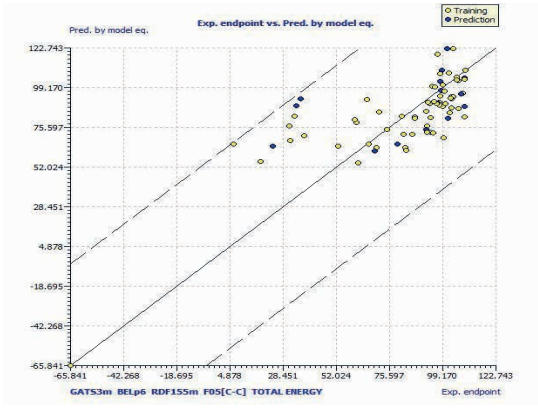

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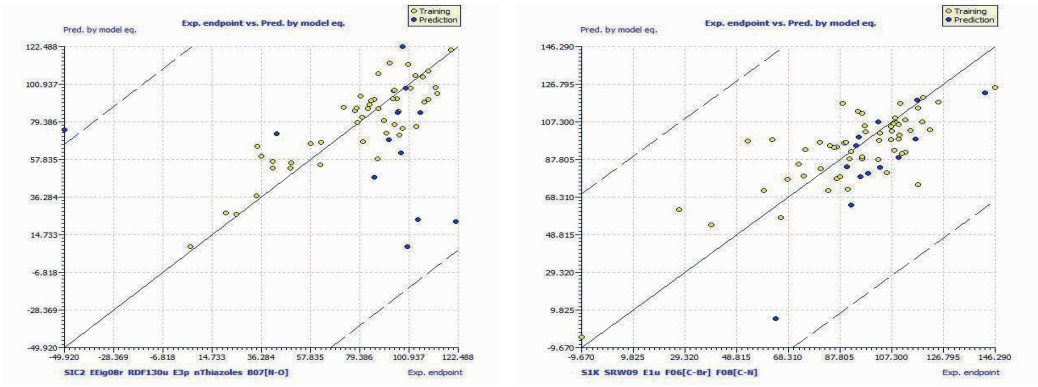
30 Воскобойнік О.Ю., Коваленко С.І. (2018). Спрямований пошук протиракових агентів серед похідних [1,2,4] триазино[2,3-с]хіназоліну як один з напрямків реалізації стратегії по імпортозаміщенню хіміотерапевтичних препаратів. Управління, економіка та забес-печення якості у фармації, 3(55). 4–10; Nosulenko I.S, Voskoboynik O.Y, Antypenko O.M, Berest G.G., Kovalenko S.I. (2015). Methodology for prediction of anticancer action of 2-oxo-2H-[1,2,4] triazino[2,3-c]quinazolin-6-yl)thiones via QSAR and docking studies. Запорізький медичний журнал, 1(88): 99–104.



C **F**

Figure 2. Observed and predicted by QSAR-models values of the growth (in %) of the leukemia (cell line MOLT-4 (A), SR (B)), non-small lung cancer (NCI-H522, C), colon cancer (KM12, D), ovarian cancer (SK-OV-3, E) and breast cancer (MDA-MB-468, F) cells.

5. COMPARE-analysis, molecular docking and study of the cell mechanisms of synthesized compounds anticancer activity.

In silico methods of COMPARE analysis and molecular docking were used to estimate the probable mechanism of action of the most active compounds. The aim of the COMPARE analysis was to evaluate the selectivity of antitumor cytotoxicity and its similarity to known anticancer agents (synthetic and natural compounds) from the NCI database. The evaluation of selectivity is conducted by comparing of the growth inhibition (lg GI_{50}) parameters of differential cancer cell lines. It was found that the studied compounds in most cases have moderate correlation with the specificity of antitumor activity of included in NCI-base of synthesized compounds anticancer agents. Abovementioned fact allowed to suppose the unknown mechanism of synthesized compounds anticancer activity. A number of compounds have a Pearson correlation coefficient (RCC)> 0.6 units and may be considered as possible EGFR inhibitors, topoisomerase I and II and apoptosis inducers.

Receptor-oriented virtual screening was used to analyze the affinity of synthesized compounds to some molecular targets associated with anticancer activity.. Molecular docking was performed at ATP binding sites of protein kinases SK2 (RCSB database code 3NSZ - 1.30Å) and FGFR1 (RCSB database code 3GQI - 2.50Å), using the program Autodock4 (http://autodock.scripps.edu/) , preprocessing and processing of results was performed using the program MGL Tools (http://mgltools.scripps.edu). Based on the obtained data, it was found that among the studied compounds, some of the amides 13 and 14 reveal high affinity to the EGRF and SC-2[31].

Hereinafter ofEGRF and SC-2 inhibitory activity of 2 - [(3-R-8-R^1-9 -R^2-10-R^3-2-oxo-2H-[1,2,4]triazino[2,3-c]quinazolin-6-yl)thio]acetamides was studied in vitro. Compound Xa was identified as high active inhibitor of CK2 (IC_{50} = 9.3 μM).

31 Воскобойнік О.Ю., Коваленко С.І. (2018). Спрямований пошук протиракових агентів серед похідних [1,2,4] триазино[2,3-с]хіназоліну як один з напрямків реалізації стратегії по імпортозаміщенню хіміотерапевтичних препаратів. Управління, економіка та забес-печення якості у фармації, 3(55). 4–10.

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