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SYNTHESIS OF FURFURYL DERIVATIVES OF 4-ALLYL-1-(4-HYDROXY-3-NITROBENZYL)-3-[2-(4-ALKOXYPHENYL)QUINOLIN-4-YL]-4,5-DIHYDRO-1*H*-1,2,4-TRIAZOL-5-THIONS AND THEIR CYTOTOXIC ACTION ON HUMAN CANCER CELLS

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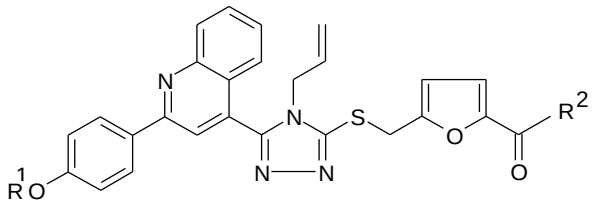
Furfuryl derivatives of 4-allyl-1-(4-hydroxy-3-nitrobenzyl)-3-[2-(4-alkoxyphenyl)quinolin-4- yl]-4,5-dihydro-1H-1,2,4-triazol-5-thiones were synthesized in this study. The docking analysis revealed the affinity of compounds for Epidermal Growth Factor Receptor (EGFR). New compounds exhibit low cytotoxicity in non-small cell lung cancer and breast cancer cell lines.

In the series of furancarboxylic acids, it has been revealed that the cytotoxic effect is enhanced with an increase of the alkoxy radical, while in the series of esters, an increase in the alkoxy radical leads to a loss of activity. The data suggest that chemical invasion of compounds leads to protein degradation in cancer cells.

Fig. 1, tables 2, references 16.

Targeting cancer with small chemical molecules is of great importance giving new possibilities of modulating not only the catalytic activity of proteins, but also functions leading to protein degradation. We have recently shown that furfuryl derivatives of 4-allyl-5-[2-(4-alkoxyphenyl)- quinolin-4-yl]-4H-1,2,4-triazole-3-thiols (compounds **1**, **2**) have a high affinity for the receptor tyro-sine kinase EGFR, and appear to induce degradation of the receptor in cancer cells [1]. It has been established that the pentyloxy derivative in the series of furancarboxylic acids (compound **2**, $R^1 = C_5H_{11}$) is the most potent modulator of EGFR activity and downstream signaling pathways in cancer cell lines. In addition, an antitumor activity detected in murine 180 sarcoma treated with compounds **1** and **2** seems to

correlate with the decrease in the level of DNA methylation in tumor tissue [2]. Structure of active anti-cancer compounds is given below.



1,2

$R^1 = CH_3 – C_5H_{11}$(**1**, **2**); $R^2 = OCH_3$ (**1**), R^2= OH (**2**).

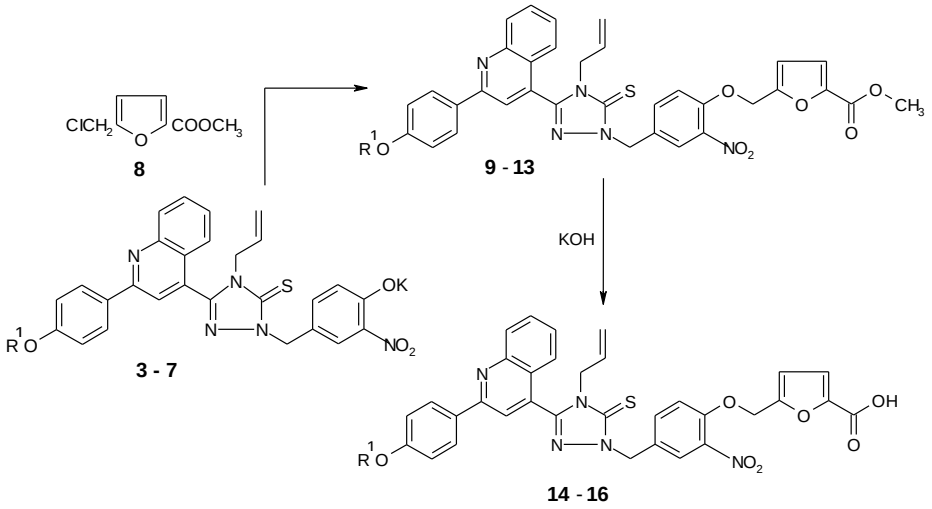
Continuing structure-designed strategy in this direction, new furfuryl derivatives of 4-allyl-1-(4-hydroxy-3-nitrobenzyl)-3-[2-(4-alkoxyphenyl)quinolin-4-yl]-4,5-dihydro-1H-1,2,4-triazol-5-thiones have been synthesized and their toxicity studied in four cancer cell lines: non-small cell lung cancer (strains A549, NSCLC-L6), breast cancer (MDA MB468) and tumor cells of the NCTC 2544 line, representing human transformed keratinocytes.

Docking of compounds **9-16** was first carried out with the catalytic domain of EGFR using the high-resolution structure of 3W32 [5]. The analysis showed that the binding energy for compounds **9-16** was rather high and comparable to known antitumor drugs cabozantinib [6], linsitinib [7] and zarnestra [8] (Table 1). These drugs are blockers of tyrosine kinase receptors, contain quinoline in their structure and have a high energy of interaction with the EGFR receptor. The results of the docking analyses suggest that the catalytic domain of EGFR is a putative target for compounds **9-16**.

Furfuryl derivatives **9-13** were obtained by reaction of potassium salts **3-7** [3] with 5-chloro-methylfuran-2-carboxylic acid methyl ether (**8**)[4] in DMFA at 95-100°C. Ethers **9-13** were then hydrolyzed by potassium hydroxide in an aqueous methanol medium to acids **14-16**.

In ^{1}H NMR spectra of compounds **9-16**, the signal from NCH_2-aryl is manifested in the region of 5.55 ppm, which is characteristic of N-substituted 1,2,4-triazol-5-thione derivatives. In IR spectra **9-16**, the signal from the C=S group is fixed at 1203 cm^{-1}, the signal from the C=O groups – in the range of 1720-1727 cm^{-1} for ethers and 1691-1705 cm^{-1} for acids. The synthetic root of compounds **9-16** is shown in Scheme 1.

Scheme 1



R^1 = CH_3 (**3**,**9**,**14**), C_2H_5 (**4**,**10**,**15**), C_3H_7 (**5**,**11**,**16**), C_4H_9 (**6**,**12**), C_5H_{11} (**7**,**13**).

Discussion of the results of Doking analyses

Models of newly synthesized compounds and antitumor drugs Cabozantinib, Linsitinib and Zarnestra were created in the PDB format using the ChemBioDraw Ultra 12.0 software package (http://software.informer.com/getfree-chembio3d-ultra-12.0/). The MM2 program of the ChemBio-Draw Ultra 12.0 software package was used to perform the minimization of the free energy of chemical compounds. Modeling of the interaction of these compounds with a high-resolution 3D structure 3W32 of the cytoplasmic region of EGFR [5] was carried out using the AutoDock Vina software package (http://vina.scripps.edu/index.html) [9]. The interaction profiles were characterized by AutoDock Tools 1.5.6rc3. For each interaction, the 9 conformations with the highest free energies were calculated using the scoring function of Vina.

To determine the mode of interaction with the receptor, the interaction energies of compounds **9-16** with the catalytic domain of EGFR 3W32 were calculated using the docking analysis. The same calculations were carried out for control antitumor drugs. The results of the docking analysis are shown in Table 1.

Table 1

Binding parameters of compounds 9-16 with the catalytic domain of EGFR (human 3W32) cabozantinib, linsitinib, and zarnestra were used as reference molecules for comparison.

Ligand	ΔG_o, *kcal/mol*	K_D, μM
Cabozantinib (XL-184)	-10.1	0.0395
Linsitinib (OSI-906)	-11.1	0.0073
Zarnestra (R-115777)	-11.1	0.0073
9	-10.3	0.0282
10	-10.8	0.0121
11	-10.6	0.0170
12	-9.6	0.0919
13	-10.4	0.0238
14	-10.1	0.0395
15	-10.5	0.0201
16	-10.7	0.0143

The spatial form of representative interactions of cabozantinib and compounds **11** and **16** is shown in Figure.

From presented data it can be seen that all compounds interact nearly in the same site of studied receptor (left side pictures). However, from precise mode of interactions (right side pictures) it can be seen that compounds **11** and **16** interact with receptor in a somewhat different place when compared to cabozantinib.

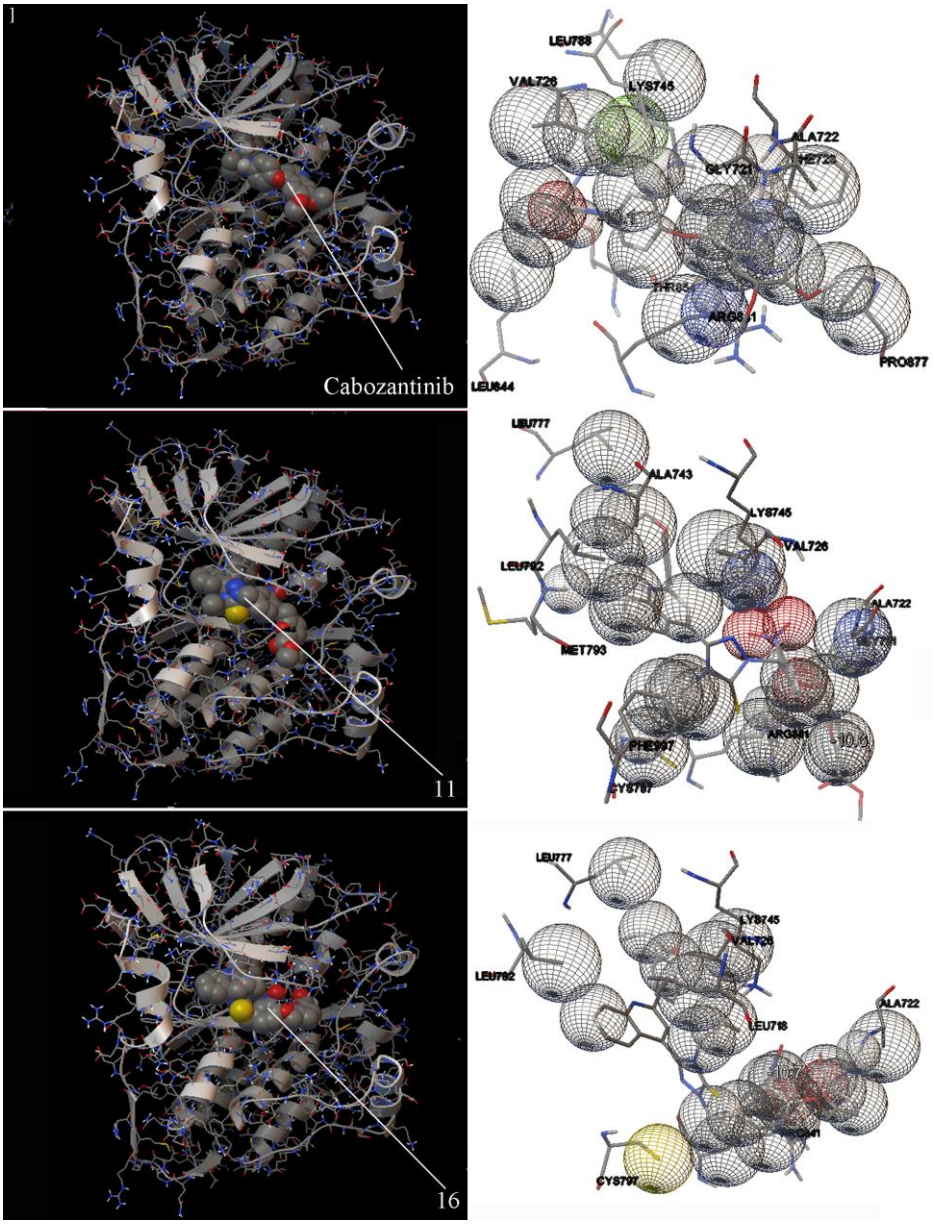


Figure. The binding of compounds **11** and **16** and cabozantinib (for comparison) to the catalytic site of EGFR (3W32).

Discussion of the biological experiments and results

Cell lines. Non-Small-Cell-Lung-Cancer (NSCLC) A549 and NSCLC-L6 cell lines originate from an adenocarcinoma and an epidermoid lung cancer, respectively. NSCLC-L6 is a cell line derived from a NSCLC of a previously untreated patient (moderately differentiated classified as T2N0M0) [10]. A triple negative breast cancer MDA MB468 is characterized by overproduction of epidermal growth factor receptor [11]. The cell line NCTC 2544 represents

transformed human keratinocytes, and A549, NSCLC-L6 and NCTC cell lines were grown in RPMI-1640 supplemented with 2 *mM* glutamine and 5% fetal bovine serum in a humidified atmosphere of 5% CO_2 at 37°C. MDA MB468 cell line was grown in Dulbecco modified eagle medium supplemented with 10% serum.

Cell viability test. Cell lines were seeded at 2×10^4 cells per well in 96-well microtitre plates and the viability of each cell line was assessed by incubation with novel compounds in parallel assays. Each compound was serially diluted and added in cell cultures followed by incubation for 72 h at 37°C. The detection of viable cells was performed with a colorimetric method based on the conversion of tetrazolium dye to blue formazan by live mitochondria [12]. The IC_{50} value of compounds was determined by measuring colour intensity at 570 *nm*. Eight repeats were carried out for each concentration of the compound to be tested.

Next, the evaluation of cell viability revealed that compounds of series **14-16** were more toxic in cell lines than compounds of series **9-13** (Table 2). The level of cytotoxicity of compounds **14-16** was similar in four cell lines tested, with the highest toxicity for compound **16** (IC_{50} 9,9 μM in MDA MB468). Among lower toxic compounds, the most toxic compound **9** had IC_{50} values in the range from 41 μM to 46 μM in cell lines A549, MDA MB468 and NCTC whereas compounds **12** and **13** were inactive at the concentration of 60 μM in all cell lines in the conditions used.

Noteworthy, the range of compound toxicity follows the increasing order of **16** > **15** > **14** in all cell lines whereas the toxicity of less toxic compounds **9-13** appears to have the increasing order of **9** > **10** > **11** > **12** > **13** (see Table 2). Comparison of structures of compounds **1** and **2** suggests that the chemical composition of the side chains R^1 and R^2 can affect the activity of compounds in cancer cells. Indeed, the cytotoxicity of new compounds, which share the same core structure, clearly depends on the length of a hydrophobic chain R^1. The longer hydrophobic R^1 chain gradually increases the cytotoxicity in acids **14-16**. On the contrary, a longer hydrophobic R^1 reduces the cytotoxicity in the series of the ethers **9-13**.

Table 2

Cytotoxicity of novel compounds (IC_{50} in μM) in four cell lines. The viability of cells was determined in parallel assays after incubation with chemical compounds for 72 hours.

Compound	A549	NSCLC-L6	MDA MB468	NCTC
9	41.4 ± 3.3	17.6 ± 0.3	46.9 ± 3.3	42.6 ± 4.3
10	52.4 ± 9.3	> 60	51.4 ± 4.8	44.6 ± 2.9
11	> 60	> 60	45.3 ± 4.7	> 60
12	> 60	Inactive	Inactive	> 60
13	Inactive	Inactive	Inactive	Inactive
14	19.4 ± 0.8	24.8 ± 1.9	22.3 ± 2.5	21.1 ± 0.6
15	16.3 ± 0.3	17.0 ± 0.2	12.9 ± 0.3	14.8 ± 0.3
16	12.3 ± 0.2	12.0 ± 0.6	9.9 ± 0.1	14.3 ± 0.2

Since recently, the apoptotic death of cancer cells caused by chemical agents that enhance protein degradation has attracted great attention in fighting cancer drug resistance. In particular, EGFR inhibitors can induce degradation of the receptor [13] caused by autophagy and explained as a result of cytoprotective response in cells [14]. Low toxicity of the new compounds described in our study suggests that the binding to EGFR could enhance endocytosis of the receptor leading to autophagic protein degradation. Moreover, the presence of a nitro group in the structure of new compounds suggests the possibility of generating reactive oxygen species that could have an additional impact on the cytotoxicity, as shown by dissection of compounds containing nitrobenzoxydiazole [15,16]. Further studies are required to elucidate the anti-cancer effect of new compounds.

Experimental part

IR spectra were recorded on the spectrophotometer "Nexus" (USA) in vaseline oil. ^{1}H NMR spectra and ^{13}C registered on the device Varian "Mercury-300 VX" in DMSO-d_6 / CCL_4, 1/3, internal standard – TMS. Melting point was defined on the microheating table "Boetius" in ^{o}C. TLC was carried out on "Silufol UV-254" plates for compounds **9-13** in the solvent system benzene-ethyl acetate, 5:1; **14-16** – benzene-ethyl acetate, 1: 1. Revelation of plates was carried out with UV light.

General procedure for the synthesis of methyl 5-{4-allyl-3-[2-(4'-alkoxyphenyl)quinolin-4-yl]-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-ylmethyl-2-nitrophenoxymethyl}-2-furoates (**9-13**). 0.005 *mol* of the corresponding potassium salt **3-7** [3] is dissolved in 10 *ml* of DMF, 0.87 *g* (0.005 *mol*) of methyl ester of 5-chloromethylfuran-2-carboxylic acid (**8**) [4] is added to the solution and heated at 95-100°C for 4-5 *hours*. Then, the greater part of the DMFA is distilled off in vacuo and water is added to the residue. The precipitate formed is filtered off and recrystallized from dimethyl sulfoxide (**9**,**10**,**12**,**13**), from methanol (**11**).

Methyl 5-{4-allyl-3-[2-(4'-methoxyphenyl)quinolin-4-yl]-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-ylmethyl-2-nitrophenoxymethyl}-2-furoate (9). Yield 69%, mp 154-155^{o}C. R_f 0.43. IR spectrum, ν, cm^{-1}: 1723(C=O), 1623, 981, 933(CH=CH_2), 1603, 1499, 833, 768 (CH=CH, aromatic), 1530 (NO_2), 1203 (C=S). ^{1}H NMR, δ, ppm, *Hz*: 3.84 (s, 3H, OCH_3), 3.89 (s, 3H, OCH_3), 4.65 (brd, 2H, J=5,6, $\underline{CH_2}CH=CH_2$), 4.83 (brd, 1H, J=17.3, $CH_2CH=\underline{CH_2}$), 5.02 (brd, 1H, J=10.4, $CH_2CH=\underline{CH_2}$), 5.33 (s, 2H, OCH_2), 5.55 (s, 2H, NCH_2-aryl), 5.77 (ddt,1H, J_1=17.3, J_2=10.4, J_3=5.6, $CH_2\underline{CH}=CH_2$), 6.72 (d, 1H, J=3.5, =CH, fur.), 6.99–7.05 (m, 2H, C_6H_4), 7.16 (d, 1H, J=3.5, =CH, fur.), 7.48 (d,1H, J=8.7, C_6H_3), 7.51–7.58 (m, 1H, C_6H_4), 7.73–7.81 (m, 2H, C_6H_4), 7.80 (dd, 1H, J_1=8.7, J_2=2.2, C_6H_3), 8.02 (d, 1H, J=2.2, C_6H_3), 8.10–8.14 (m, 1H, C_6H_4), 8.21–8.26 (m, 2H, C_6H_4), 8.27 (s, 1H, =CH, pyr.). Found, %: N, 10.48; S, 4.62. $C_{35}H_{29}N_5O_7S$. Calculated, %: N, 10.55; S, 4.83.

Methyl 5-{4-allyl-3-[2-(4'-ethoxyphenyl)quinolin-4-yl]-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-ylmethyl-2-nitrophenoxymethyl}-2-furoate (10). Yield 70%, mp 134-135°C. R_f 0.47. IR spectrum, ν, cm^{-1}: 1727(C=O), 1620, 982, 936 (CH=CH_2), 1602, 1499, 824, 768 (CH=CH, aromatic), 1530, 1330 (NO_2), 1203 (C=S). ^{1}H NMR, δ, ppm, *Hz*: 1.45 (t, 3H, J=7.0, $CH_2\underline{CH_3}$), 3.84 (s, 3H, OCH_3), 4.13 (q, 2H, J=7.0, $\underline{CH_2}CH_3$), 4.65 (brd, 2H, J=5.5, $\underline{CH_2}CH=CH_2$), 4.82 (brd, 1H, J=17.1, $CH_2CH=\underline{CH_2}$), 5.02 (brd, 1H, J=10.3, $CH_2CH=\underline{CH_2}$), 5.32 (s, 2H, OCH_2), 5.55 (s, 2H, NCH_2-aryl), 5.77 (ddt, 1H, J_1=17.1, J_2=10.3, J_3=5.5, =CH), 6.72 (d, 1H, J=3.5, =CH, fur.), 6.97–7.02 (m, 2H, C_6H_4), 7.15 (d, 1H, J=3.5, =CH, fur.), 7.48 (d, 1H, J=8.7, C_6H_3), 7.51–7.57 (m, 1H, C_6H_4), 7.74–7.82 (m, 3H, C_6H_4), 8.02 (d, 1H, J=2.2, C_6H_3), 8.09–8.14 (m, 1H, C_6H_4), 8.19–8.25 (m, 2H, C_6H_4), 8.25 (s, 1H, =CH, pyr.). Found, %: N, 10.21; S, 4.59. $C_{36}H_{31}N_5O_7S$. Calculated, %: N 10.33; S 4.73.

Methyl 5-{4-allyl-3-[2-(4'-propoxyphenyl)quinolin-4-yl]-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-ylmethyl-2-nitrophenoxymethyl}-2-furoate (11). Yield 58%, mp 108-110°C. R_f 0.54. IR spectrum, ν, cm^{-1}: 1722 (C=O), 1623, 990, 920 (CH=CH_2), 1603, 1502, 837, 769 (CH=CH, aromatic), 1530, 1340 (NO_2), 1203 (C=S). ^{1}H NMR, δ, ppm, *Hz*: 1.09 (t, 3H, J=7.4, CH_3), 1.78–1.90 (m, 2H, $\underline{CH_2}CH_3$), 3.84 (s, 3H, OCH_3), 4.02 (t, 2H, J=6.6, OCH_2), 4.65 (brd, 2H, J=5.6, $\underline{CH_2}CH=CH_2$), 4.82 (brd, 1H, J=17.2, $CH_2CH=\underline{CH_2}$), 5.02 (brd, 1H, J=10.3, $CH_2CH=\underline{CH_2}$), 5.33 (s, 2H, OCH_2), 5.55 (brs, 2H, NCH_2-aryl), 5.76 (ddt, 1H, J_1=17.2, J_2=10.3, J_3=5.6, $CH_2\underline{CH}=CH_2$), 6.72 (d, 1H, J=3.5, =CH, fyr.), 6.97–7.03 (m, 2H, C_6H_4), 7.16 (d, 1H, J=3.5, =CH, fur.), 7.49 (d, 1H, J=8.7, C_6H_3), 7.54 (ddd, 1H, J_1=8.3, J_2=6.8, J_3=1.1, C_6H_4), 7.74–7.82 (m, 3H, C_6H_4), 8.02 (d, 1H, J=2.2, C_6H_3), 8.11 (dd, 1H, J_1=8.7, J_2=1.1, C_6H_4), 8.20–8.25 (m, 2H, C_6H_4), 8.26 (s, 1H, =CH, pyr.). ^{13}C: 10.1(CH_3), 21.9 (CH_3), 47.4(NCH_2), 50.2(NCH_2), 51.1(OCH_3), 63.1(OCH_2), 68.7(OCH_3), 111.9(=CH_2), 114.1(2C, C_6H_4), 115.5(CH), 118.0(CH), 118.1(CH), 119.3(CH), 123.7(C), 124.2 (CH), 124.8(CH), 126.5(CH), 128.3(2C, C_6H_4), 128.4(C), 129.4(CH), 129.7(CH), 129.8(C), 130.6 (C), 131.3(C), 133.9(CH), 139.5(C), 144.0(C), 147.0(C), 147.9(C), 150.2(C), 152.8 (C), 155.2(C), 157.5(C), 160.2(C), 167.1(C). Found, %: N, 10.39; S, 4.49. $C_{37}H_{33}N_5O_7S$. Calculated, %: N, 10.12; S, 4.63.

Methyl 5-{4-allyl-3-[2-(4'-butoxyphenyl)quinolin-4-yl]-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-ylmethyl-2-nitrophenoxymethyl}-2-furoate (12). Yield 64%, mp 138-139°C. R_f 0.58. IR spectrum, ν, cm^{-1}: 1720 (C=O), 1623, 990, 920 (CH=CH_2), 1603, 1501, 835, 772 (CH=CH, aromatic), 1531, 1340 (NO_2), 1205 (C=S). ^{1}H NMR, δ, ppm, *Hz*: 1.02 (t, 3H, J=7.3, $CH_2\underline{CH_3}$), 1.48–1.61 (m, 2H, $\underline{CH_2}CH_3$), 1.75–1.85 (m, 2H, $\underline{CH_2}CH_2CH_3$), 3.84 (s, 3H, OCH_3), 4.05 (t, 2H, J=6.4, OCH_2), 4.65 (dt, 2H, J_1=5.6, J_2=1.4, $\underline{CH_2}CH=CH_2$), 4.83 (dq, 1H, J_1=17.1, J_2=1.4, $CH_2CH=\underline{CH_2}$), 5.02 (dq, 1H, J_1=10.3, J_2=1.4, $CH_2CH=\underline{CH_2}$), 5.33 (s, 2H, OCH_2), 5.55 (s, 2H, NCH_2-aryl), 5.77 (ddt, 1H, J_1=17.1, J_2=10.3, J_3=5.6, $CH_2\underline{CH}=CH_2$), 6.72 (d, 1H, J=3.5, =CH, fur.), 6.97–7.02 (m, 2H, C_6H_4), 7.16 (d, 1H, J=3.5, =CH, fur.), 7.48 (d, 1H, J=8.7, C_6H_3), 7.51–7.57 (m, 1H, C_6H_4), 7.74–7.80 (m, 2H, C_6H_4), 7.80 (dd, 1H, J_1=8.7, J_2=2.2, C_6H_3), 8.02 (d, 1H, J=2.2, C_6H_3), 8.09–8.14 (m, 1H,

C_6H_4), 8.19–8.25 (m, 2H, C_6H_4), 8.26 (s, 1H, =CH, pyr.). ^{13}C: 13.3 (CH_3), 18.6(CH_2), 30.6(CH_2), 47.4 (NCH_2), 50.3(OCH_3), 51.0(NCH_2), 63.1(OCH_2), 66.9(OCH_2), 111.7(CH_2), 111.8(CH), 114.1(2C, C_6H_4), 115.5 (CH), 118.0(2CH), 119.3(C), 123.7(C), 124.2 (CH), 124.9(CH), 126.5(CH), 128.3(2C, C_6H_4), 128.3(C), 129.4(CH), 129.6(CH), 129.8(C), 130.6 (CH), 131.2(C), 133.9(CH), 139.5(C), 144.0(C), 147.0(C), 147.9(C), 150.1(C), 152.8(C), 154.9 (C), 160.7(C), 167.1(C). Found, %: N, 9.83; S, 4.39. $C_{38}H_{35}N_5O_7S$. Calculated, %: N, 9.92; S, 4.54.

Methyl 5-{4-allyl-3-[2-(4'-pentyloxyphenyl)quinolin-4-yl]-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-ylmethyl-2-nitrophenoxymethyl}-2-furoate (13). Yield 61%, mp 129-130°C. R_f 0.64. IR spectrum, ν, *cm*$^{-1}$: 1726 (C=O), 1622, 985, 936 (CH=CH_2), 1602, 1499, 830, 768 (CH=CH, aromatic), 1530, 1340 (NO_2), 1203 (C=S). ^{1}H NMR, δ, ppm, *Hz*: 0.97 (t, 3H, J=7.0, CH_3), 1.36–1.55 (m, 4H, $\underline{CH_2}CH_2CH_3$), 1.76–1.87 (m, 2H, CH_2),3.84 (s, 3H, OCH_3), 4.04 (t, 2H, J=6.4, OCH_2), 4.66 (brd, 2H, J=5.4, $\underline{CH_2}CH=CH_2$), 4.83 (brd, 1H, J=17.1, $CH_2CH=\underline{CH_2}$), 5.02 (brd, 1H, J=10.3, $CH_2CH=\underline{CH_2}$), 5.33 (s, 2H, OCH_2), 5.55 (s, 2H, NCH_2-aryl), 5.77 (ddt, 1H, J_1=17.1, J_2=10.3, J_3=5.4, $CH_2\underline{CH}=CH_2$), 6.72 (d, 1H, J=3.5, =CH, fur.), 6.96–7.02 (m, 2H, C_6H_4O), 7.16 (d, 1H, J=3.5, =CH, fur), 7.48 (d, 1H, J=8.7, C_6H_3), 7.51–7.57 (m, 1H, C_6H_4), 7.74–7.82 (m, 3H, C_6H_4 and C_6H_3), 8.02 (d, 1H, J=2.1, C_6H_3), 8.09–8.14 (m, 1H, C_6H_4), 8.18–8.25 (m, 2H, C_6H_4O), 8.26 (s, 1H, =CH, pyr). Found, %: N, 9.70; S, 4.61. $C_{39}H_{37}N_5O_7S$. Calculated, %: N, 9.73; S, 4.45.

General procedure for the synthesis of 5-{4-allyl-3-[2-(4'-alkoxyphenyl) quinolin-4-yl]-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-ylmethyl-2-nitrophenoxy}-2-furoate (**14-16**). A mixture of 0.001 *mol* of the corresponding ester **9-11**, 0.11 *g* (0.002 *mol*) of potassium hydroxide and 16 *ml* of 50% methanol is boiled for 3-4 *h*, the solution is acidified with acetic acid, the precipitate is filtered off and recrystallized from ethanol.

5-{4-Allyl-3-[2-(4'-methoxyphenyl)quinolin-4-yl]-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1- ylmethyl-2-nitrophenoxymethyl}-2-furoate (14). Yield 62%, mp 128-130°C. R_f 0.44. IR spectrum, ν, *cm*$^{-1}$: 3100-2500 (OH), 1691 (C=O), 1620, (CH=CH_2), 1603, 1500, 834, 765 (CH=CH, aromatic), 1529 (NO_2), 1203 (C=S). ^{1}H NMR, δ, ppm, *Hz*: 3.89 (s, 3H, OCH_3), 4.65 (dt, 2H, J_1=5.6, J_2=1.7, $\underline{CH_2}CH=CH_2$), 4.82 (dq, 1H, J_1=17.2, J_2=1.7, $CH_2CH=\underline{CH_2}$), 5.02 (dq, 1H, J_1= 10.3, J_2=1.7, $CH_2CH=\underline{CH_2}$), 5.31 (s, 2H, OCH_2), 5.55 (s, 2H, NCH_2-aryl), 5.76 (ddt, 1H, J_1=17.2, J_2=10.3, J_3=5.6, $CH_2\underline{CH}=CH_2$), 6.68 (d, 1H, J=3.4, =CH, fur.), 7.00–7.05 (m, 2H, C_6H_4O), 7.06 (d, 1H, J=3.4, =CH, fur.), 7.50 (d, 1H, J=8.7, C_6H_3), 7.54 (ddd, 1H, J_1=8.3, J_2=6.8, J_3=1.2, C_6H_4), 7.74–7.82 (m, 3H, C_6H_4 and C_6H_3), 8.01 (d, 1H, J=2.2, C_6H_3), 8.10–8.14 (m, 1H, C_6H_4), 8.21–8.26 (m, 2H, C_6H_4O), 8.28 (s, 1H, =CH, pyr.). Found, %: N, 10.62; S, 4.80. $C_{34}H_{27}N_5O_7S$. Calculated, %: N, 10.78; S, 4.93.

5-{4-Allyl-3-[2-(4'-ethoxyphenyl)quinolin-4-yl]-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-ylmethyl-2-nitrophenoxymethyl}-2-furoate (15). Yield 64%, mp 134-136°C. R_f 0.46. IR spectrum, ν, *cm*$^{-1}$: 3100-2500 (OH), 1705 (C=O), 1625

($CH=CH_2$), 1604, 1503, 762 (CH=CH, aromatic), 1529 (NO_2), 1203 (C=S). ^{1}H NMR, δ, ppm, *Hz*: 1.45 (t, 3H, J=6.9, CH_3), 4.13 (q, 2H, J=6.9, $\underline{CH_2}CH_3$), 4.65 (brd, 2H, J=5.1, $\underline{CH_2}CH=CH_2$), 4.83 (d, 1H, J=17.3, $CH_2CH=\underline{CH_2}$), 5.02 (d, 1H, J=10.3, $CH_2CH=\underline{CH_2}$), 5.31 (s, 2H, CH_2), 5.55 (s, 2H, NCH_2-aryl), 5.77 (ddt, 1H, J_1=17.3, J_2=10.3, J_3=5.1, $CH_2=\underline{CH}CH_2$), 6.68 (d, 1H, J=3.4, =CH, fur.), 6.96–7.03 (m, 2H, C_6H_4O), 7.08 (d, 1H, J=3.4, =CH, fur.), 7,49 (d, 1H, J=8.7, C_6H_3), 7.50–7.57 (m, 1H, C_6H_4), 7.71–7.82 (m, 3H, C_6H_4), 8.02 (d, 1H, J=2.1, C_6H_3), 8.12 (brd, 1H, J=8.5, C_6H_4), 8.19–8.25 (m, 2H, C_6H_4O), 8.26 (s, 1H, =CH, pyr.), 12.06 (br, 1H, COOH). Found, %: N, 10.47; S, 4.76. $C_{35}H_{29}N_5O_7S$. Calculated, %: N, 10.55; S, 4.83.

5-{4-Allyl-3-[2-(4'-propoxyphenyl)quinolin-4-yl]-5-thioxo-4,5-dihydro-1H-1,2,4-triazol-1-ylmethyl-2-nitrophenoxymethyl}-2-furoate (16). Yield 71%, mp 144-145°C. R_f 0.47. IR spectrum, ν, cm^{-1}: 3100-2500 (OH), 1717 (C=O), 1620 ($CH=CH_2$), 1602, 1500, 835, 768 (CH=CH, aromatic), 1529 (NO_2), 1203 (C=S). ^{1}H NMR, δ, ppm, *Hz*: 1.09 (t, 3H, J=7.4, CH_3), 1.78–1.91 (m, 2H, $\underline{CH_2}CH_3$), 4.02 (t, 2H, J=6.4, OCH_2), 4.65 (brd, 2H, J=5.3, $\underline{CH_2}CH=CH_2$), 4.82 (brd, 1H, J=17.2, $CH_2CH=\underline{CH_2}$), 5.02 (brd, 1H, J=10.3, $CH_2CH=\underline{CH_2}$), 5.32 (s, 2H, OCH_2), 5.55 (s, 2H, NCH_2-aryl), 5.76 (ddt, 1H, J_1=17.2, J_2=10.3, J_3=5.3, $CH_2\underline{CH}=CH_2$), 6.68 (d, 1H, J=3.4, =CH, fur.), 6.97–7.03 (m, 2H, C_6H_3), 7.08 (d, 1H, J=3.4, =CH, fur.), 7.50 (d, 1H, J=8.8, C_6H_3), 7.52–7.58 (m, 1H, C_6H_4), 7.74–7.82 (m, 3H, C_6H_4 and C_6H_3), 8.02 (d, 1H, J=2.1, C_6H_3), 8.09–8.14 (m, 1H, C_6H_4), 8.19–8.25 (m, 2H, C_6H_4O), 8.27 (s, 1H, =CH, pyr.), 12.80 (br, 1H, COOH). Found, %: N, 10.48; S, 4.59. $C_{36}H_{31}N_5O_7S$. Calculated, %: N, 10.33; S, 4.73.

4-ԱԼԻԼ-1-(4-ՀԻԴՐՕՔՍԻ-3-ՆԻՏՐՈԲԵՆԶԻԼ)-3-[2-(4-ԱԼԿՕՔՍԻՖԵՆԻԼ)ԽԻՆՈԼԻՆ-4-ԻԼ]-4,5-ԴԻՀԻԴՐՈ-1Հ-1,2,4-ՏՐԻԱԶՈԼ-5-ԹԻՈՆՆԵՐԻ ՖՈՒՐՖՈՒՐԻԼ ԱԾԱՆՑՅԱԼՆԵՐԻ ՍԻՆԹԵԶԸ ԵՎ ՄԱՐԴՈՒ ՔԱՂՑԿԵՂԱՅԻՆ ԲՋԻՋՆԵՐԻ ՎՐԱ ԴՐԱՆՑ ՑԻՏՈՏՈՔՍԻԿ ԱԶԴԵՑՈՒԹՅՈՒՆԸ

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Ներկայացված աշխատանքում բերված է 4-ալիլ-1-(4-հիդրօքսի-3-նիտրոբենզիլ)-3-[2-(4-ալկօքսիֆենիլ)խինոլին-4-իլ]-4,5-դիհիդրո-1Հ-1,2,4-տրիազոլ-5-թիոնների ֆուրֆուրիլ ածանցյալների սինթեզը և կառուցվածքային անալիզը: Դոքինգ անալիզի միջոցով հնարավոր սպիտակուցային թիրախների հետ մոլեկուլային փոխազդեցության ուսումնասիրությունը ցույց տվեց, որ EGFR-ի կատալիտիկ դոմենի հետ նոր նյութերի կապման էներգիան բարձր է և մակարդակով մոտ է համեմատության համար օգտագործված հակաքաղցկեղային պրեպարատների (կաբոզանտինիբ, լինսիտինիբ և զոնեստրա) փոխազդեցության էներգիաներին: Ուսումնասիրվել է նյութերի ցիտոտոքսիկ ազդեցությունը հետևյալ բջջային գծերի վրա. կրծքագեղձի քաղցկեղ՝ MDA MB468, թոքերի ոչ մանր բջջային քաղցկեղ՝ NSCLC A549 և NSCLC-L16, մարդու տրանսֆորմացված կերատինոցիտներ՝ NCTC 2544: Ցույց է տրվել, որ ֆուրանկարբո-նաֆթիունների շարքում ցիտոտոքսիկ ակտիվությունը մեծանում է ալկօքսի ռադիկալի մեծացման հետ, իսկ էսթերների դեպքում ալկօքսի ռադիկալի մեծացումը բերում է ակտիվության անկման:

Նյութերի ցածր տոքսիկությունը ենթադրում է, որ քիմիական ինվազիան բերում է EGFR-ի էնդոցիտոզի և ռեցեպտորի և քաղցկեղային բջջում դրա հետ ասոցված սպիտակուցների հետագա դեգրադացման:

СИНТЕЗ ФУРФУРИЛЬНЫХ ПРОИЗВОДНЫХ 4-АЛЛИЛ-1-(4-ГИДРОКСИ-3-НИТРОБЕНЗИЛ)-3-[2-(4-АЛКОКСИФЕНИЛ)ХИНОЛИН-4-ИЛ]-4,5-ДИГИДРО-1*H*-1,2,4-ТРИАЗОЛ-5-ТИОНОВ И ИХ ЦИТОТОКСИЧЕСКОЕ ДЕЙСТВИЕ НА РАКОВЫЕ КЛЕТКИ ЧЕЛОВЕКА

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В представленной работе проведен синтез и структурный анализ фурфурильных произ- водных 4-аллил-1-(4-гидрокси-3-нитробензил)]-3-[2-(4-алкоксифенил)хинолин-4-ил]-4,5-дигидро-1*H*-1,2,4-триазол-5-тионов. Изучение молекулярных взаимодействий с возможными белковыми мишенями методом докинга показал, что энергия связывания новых соединений с каталитическим доменом EGFR высокая и находится на уровне, близком для противоопухолевых препаратов кабозантиниба, линситиниба и зарнестры, использованных для сравнения. Исследовано цитотоксическое действие соединений на клеточной линии рака молочной железы MDA MB468, линиях немелкоклеточного рака легких NSCLC A549 и NSCLC-L16, а также на линии трансформированных кератиноцитов человека NCTC 2544. Выявлено, что в ряду фуранкарбоновых кислот цитотоксическое действие усиливается с повышением алкоксильного радикала, а в ряду эфиров повышение алкоксильного радикала приводит к потере активности. Низкая токсичность соединений предполагает, что химическая инвазия приводит к повышенному эндоцитозу EGFR и последующей деградации рецептора и ассоциированных с ним белков в раковых клетках.

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