Synthesis of fluoromethyl-containing analogs of antitumor alkaloid luotonin A

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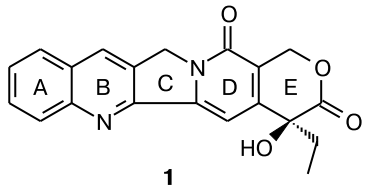
A convenient method for the synthesis of hitherto unknown 3-bromomethyl-2-chloro-4-fluoromethylquinolines has been developed. Coupling of 3-bromomethyl-2-chloro-4-trifluoromethylquinoline with 4(3*H*)-quinazolinone with subsequent intramolecular Heck cyclization leads to 7-trifluoromethylluotonin, an analog of the antitumor alkaloid luotonin A. 7-Trifluoromethylluotonin retains the antitumor activity including apoptosis of cultured tumor cells and inhibiting DNA-topoisomerase I.

Key words: camptothecin, luotonin A, antitumor alkaloid, topoisomerase I, 4-fluoromethyl-2-quinolones, intramolecular Heck cyclization.

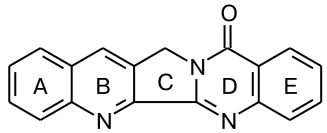
Camptothecin (**1**) is a pentacyclic alkaloid isolated from the tree *Camptotheca acuminata*.[1] Camptothecin remains one of the most promising compounds for development of antitumor drugs.[2] In the 1980s, it was found that camptothecin is an efficient inhibitor of the DNA-dependent enzyme, topoisomerase I in the eukaryotic cells.[3] According to modern concepts, the inhibitory activity of camptothecin is due to the formation of a three-component complex camptothecin—topoisomerase I—DNA,[4] which causes single-strand cleavage of DNA. To date, numerous water-soluble camptothecin analogs have been synthesized; two of them, topotecan (hycampin) and irinotecan (camptosar), are used in clinical practice and about dozen of others undergo clinical trial.[5]

Despite this progress, clinical application of antitumor drugs based on camptothecin faces a number of problems.[6] The major problem is the lability of the lactone ring E at physiological pH values, with the open-chain form of the lactone ring (the carboxyl form) possessing only 10% activity of the lactone form. Another problem of the camptothecin derivatives is their high general resorption toxicity. Chemotherapy causes emergence of multiple drug resistance of tumors because of poor accumulation of the drugs due to their active transport from cells.[7]

Several approaches have been suggested to increase the camptothecin stability, in particular, replacement of the lactone ring by an aromatic one,[8] introduction of a seven-membered β-hydroxylactone ring instead of the metabolically labile six-membered α-hydroxylactone ring,[9] as well as introduction of lipophilic groups into the ring B of camptothecin.[10] It is important to study naturally occurring alkaloid luotonin A (**2**), the structural analog of camptothecin,[11] since its derivatives are suitable for the search for new camptothecin-based antitumor drugs.



1



2

One of the most convenient approaches to the synthesis of luotonin A is formation of the ring C of the pentacycle by fusion of the A/B- and D/E-fragments through the N-alkylation of 4(3*H*)-quinazolinone with 3-bromomethyl-2-haloquinolines and subsequent intramolecular Heck cyclization (Scheme 1).[12] Thus, the substituted 3-bromomethyl-2-haloquinolines are key building blocks in the synthesis of modified luotonins.

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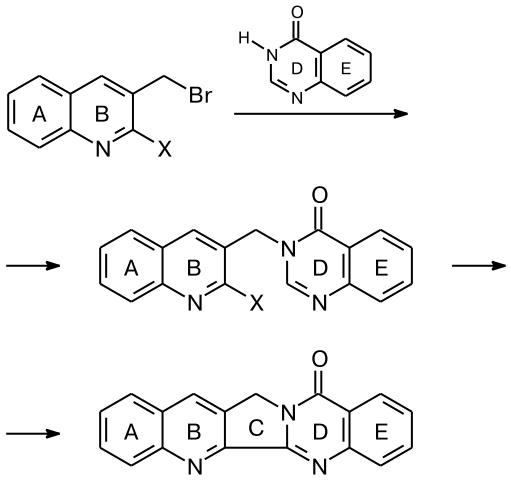
Scheme 1



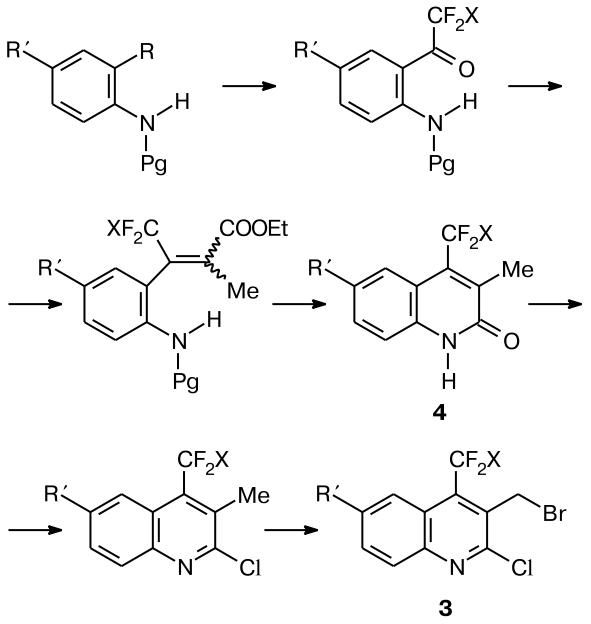
X = Br, Cl

The purpose of this work is the synthesis and assessment of antitumor activity of luotonin A derivatives containing fluoromethyl substituents in the ring B of the pentacycle.[13] Our approach to the synthesis of fluoromethyl analogs of luotonin A is based on the use of new building blocks, *viz.*, 3-bromomethyl-2-chloro-4-fluoromethylquinolines **3** (Scheme 2). In this work, we describe a method for the synthesis of quinolines **3** containing 4-CF_3-, 4-CF_2H-, and 4-CF_2Cl groups. The synthesis of 3-bromomethyl-2-chloro-6-fluoro-4-trifluoromethylquinoline illustrates a possibility of introduction of a substituent (R′ = F, see Scheme 2) into the benzene ring of quinoline **3** in addition to the 4-CF_3-group. This convenient method for the synthesis of quinolines **3** opens a direct route (see Scheme 1) to luotonins containing different fluoromethyl groups in position 7* of the pentacycle essential for the physiological activity: a lipophilic group (CF_3); a group capable of forming hydrogen bonds (CF_2H); a group that can be alkylated with endogenous nitrogen bases (CF_2Cl). More important is that these building blocks can be used for the synthesis of 7-fluoromethyl-substituted camptothecins, homocamptothecins, 14-azocamptothecins, and other compounds of the camptothecin series.

To preparate quinolines **3**, we developed a five-step synthetic scheme. N-Protected anilines served as the starting compounds, synthesis of quinolones **4** being a key step (see Scheme 2).

A scheme for the assembly of 4-trifluoromethyl-substituted 2-quinolone core has been described earlier.[15] This includes *ortho*-trifluoroacetylation of aniline bearing an appropriate N-protecting group, introduction of a C_2-fragment

Scheme 2



R = H, Br; R′ = H, F; X = H, F, Cl; Pg = $COCMe_3$, $COCH_2Me$

by coupling at the keto group, and, finally, the lactam ring closure. In the case of the synthesis of 4-trifluoromethyl-2-quinolones, this scheme is advantageous as compared to the classic Knorr synthesis of 2-quinolones. In the framework of the method developed,[15] 3-methyl-4-trifluoromethyl-2-quinolone **4a** (X = F, R′ = H, see Scheme 2) was synthesized starting from *N*-Boc-aniline using pyrophoric Bu^tLi as a lithiating agent.

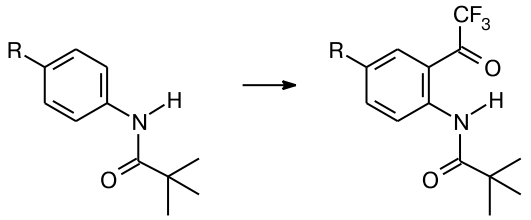
In the synthesis of quinolines **3** based on the Schlosser approach,[15] we modified the stage of *ortho*-fluoroacetylation of anilines. We chose a pivaloyl group (Pg = $COCMe_3$, R = H) and a propionyl group (Pg = $COCH_2Me$, R = Br, see Scheme 2) as protecting and *ortho*-directing groups. Safe BuLi was a lithiating agent and ethyl trifluoroacetate, ethyl difluoroacetate, and ethyl chlorodifluoroacetate were used for the acylation.

Lithiation of *N*-pivaloylaniline (**5a**) and *N*-pivaloyl-*p*-fluoroaniline (**5b**) was performed in THF with a threefold excess of BuLi in the temperature range from −10 to 0 °C for 3 h (see Ref. 16). The acylating agent, ethyl trifluoroacetate (2 equiv.), was added at −70 °C and, after standard work-up, products **6a** and **6b** were obtained in preparative yields (~50%) (Scheme 3). However, in the case of ethyl difluoroacetate and ethyl chlorodifluoroacetate, the yields of the corresponding ketones were unsatisfactory.

Ketones **6c** and **6d** were successfully obtained starting from *N*-propionyl-2-bromoaniline **5c** (Scheme 4). They were isolated in >50% yields. Lithiation with BuLi was performed in this case at −75 °C in THF.[17] It should be noted that the lithiation of 2-bromoaniline **5c** is accompanied by formation of *N*-propionyl-2-*n*-butylaniline **5d**

* Two general ways for the luotonin A numbering were used. The first is based on the camptothecin numeration, see Refs 12c and 14a. The second uses the IUPAC nomenclature, see Refs 13b,c, and 14b.

Scheme 3

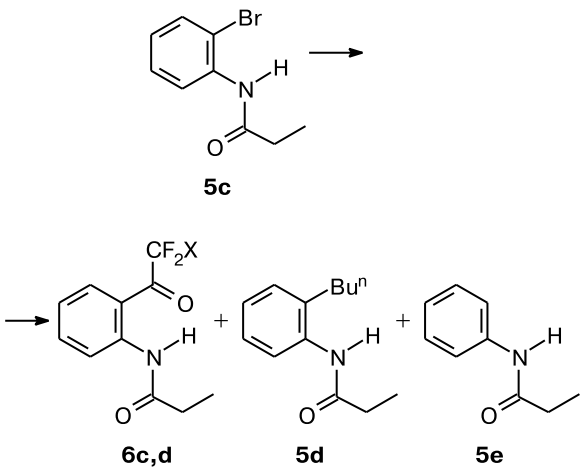


5a,b **6a,b**

R = H (**5a**, **6a**), F (**5b**, **6b**)

Reactants and conditions: 1) Bu^nLi, –10—0 °C; 2) CF_3CO_2Et, –75 °C.

Scheme 4



5c **6c,d** **5d** **5e**

X = H (**6c**), Cl (**6d**)

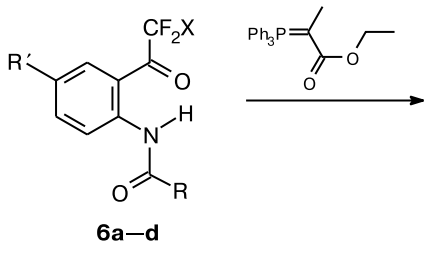
Reactants and conditions: 1) Bu^nLi, –75 °C; 2) CF_2XCO_2Et, –75 °C.

(see Ref. 18) the yield of which reached 10%. The lithiation of 2-bromoaniline **5c** results also in the formation of noticeable amounts (up to 10%) of debromination product, *N*-propionylaniline **5e**.

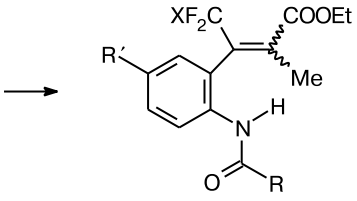
The formation of the carbon skeleton of 4-fluoromethyl-2-quinolones involved introduction of the C_2-fragment by coupling ketones **6a—d** with 1-(ethoxycarbonylethylidene)triphenylphosphorane[15] (Scheme 5). The Wittig reaction products **7a—d** were isolated by column chromatography as mixtures of *E*,*Z*-isomers, which were characterized by ^1H and ^{19}F NMR spectra and used in the subsequent steps of the lactam ring formation as such (Scheme 6). Compounds **7a,b** underwent cyclization to form 2-quinolones **4a,b** upon refluxing in a mixture of 6*M* HCl and acetic acid. However, the crucial factor for the preparation of 2-quinolones **4c,d** was performing the cyclization of compounds **7c,d** under less drastic conditions, *viz.*, upon refluxing in 2*M* HCl.

2-Quinolones **4a—d** upon refluxing in an excess of $POCl_3$ furnish 2-chloroquinolines **8a—d** (Scheme 7). The

Scheme 5



6a—d

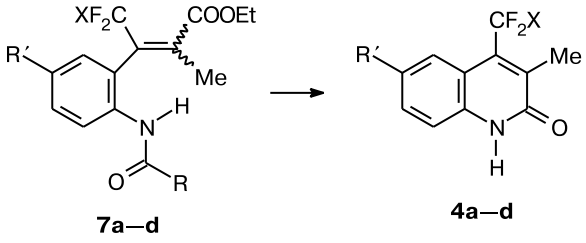


7a—d

	R	R′	X
7a	CMe_3	H	F
7b	CMe_3	F	F
7c	CH_2Me	H	H
7d	CH_2Me	H	Cl

Conditions: toluene, 100 °C.

Scheme 6



7a—d **4a—d**

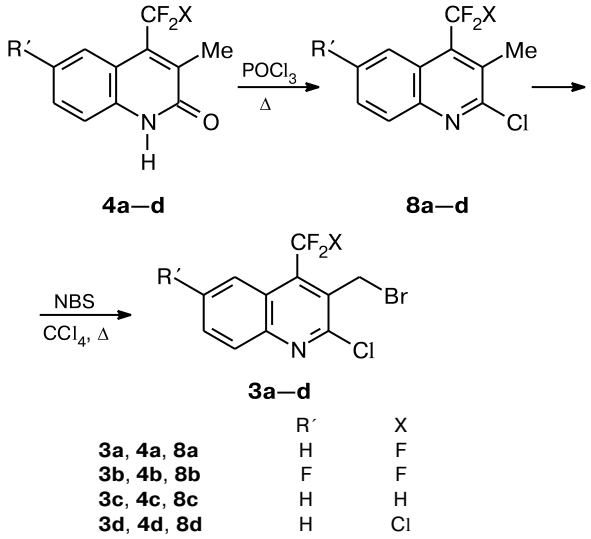
	R′	X
4a	H	F
4b	F	F
4c	H	H
4d	H	Cl

Reactants and conditions: 6 *M* HCl or 2 *M* HCl.

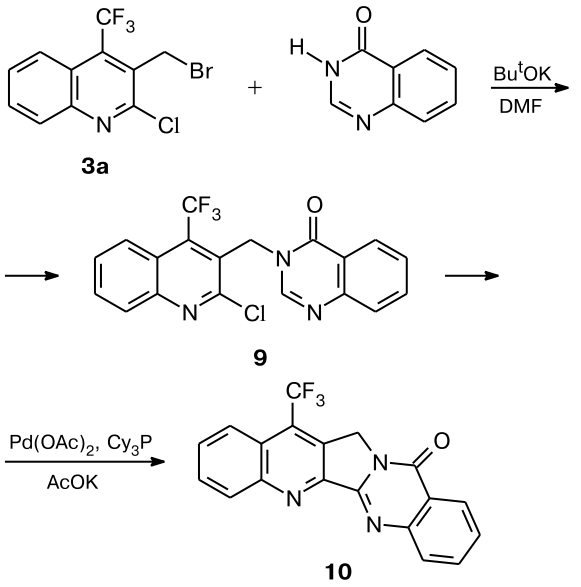
reaction requires short time and proceeds in high yields. Upon bromination with NBS in boiling CCl_4 in the presence of a radical initiator,[19] quinolines **8a—d** give the target quinolines **3a—d** (see Scheme 7).

Quinoline **3a** was successfully used in the synthesis of 7-trifluoromethylluotonin A. The reaction of **3a** with 4(3*H*)-quinazolinone in DMF in the presence of potassium *tert*-butoxide[20] occurs at 0 °C to form compound **9** (Scheme 8). Subsequent intramolecular Heck cyclization in the presence of $Pd(OAc)_2$, tri(cyclohexyl)phosphine, and potassium acetate in DMF at 160 °C (see Ref. 20) afforded 7-trifluoromethylluotonin **10** in 30% yield (Scheme 8).

Scheme 7



Scheme 8



Structures of all the compounds obtained were confirmed by elemental analysis data (Table 1) and their spectral characteristics (Table 2).

Biological

Cytotoxicity. Compound **10** caused apoptosis of cultured human colon adenocarcinoma and leukemia cells in relatively high concentrations: 50% growth inhibitory concentrations after cell exposure for 72 h were 17±3 and 21±4 μmol L^{-1}, respectively. These values are lower than those found for the most antitumor compounds (micromolar and submicromolar ranges).

Table 1. Yields and some physicochemical characteristics of compounds **3a—d**, **4b—d**, **5d**, **6a—d**, **8a—d**, **9**, **10**

Compound	Yield (%)	M.p. /°C	Found / Calculated (%)			Molecular formula
			C	H	N	
3a	74	60—61 (LP)	40.87 / 40.71	1.86 / 1.86	4.24 / 4.32	$C_{11}H_6BrClF_3N$
3b	72	95—96 (LP)	38.59 / 38.57	1.54 / 1.47	4.05 / 4.09	$C_{11}H_5BrClF_4N$
3c	37	67—68 (LP)	42.95 / 43.10	2.15 / 2.30	4.34 / 4.57	$C_{11}H_7BrClF_2N$
3d	47	65—66 (LP)	38.91 / 38.75	1.75 / 1.77	4.07 / 4.11	$C_{11}H_6BrCl_2F_2N$
4b	65	213—214 (ethanol)	53.76 / 53.89	2.79 / 2.88	5.61 / 5.71	$C_{11}H_7F_4NO$
4c	30	229—230 (ethanol)	62.91 / 63.16	4.30 / 4.34	6.78 / 6.70	$C_{11}H_9F_2NO$
4d	32	215—216 (ethanol)	54.11 / 54.23	3.35 / 3.31	5.82 / 5.75	$C_{11}H_8ClF_2NO$
5d	—	79—80 (LP)	76.07 / 76.06	9.41 / 9.33	6.65 / 6.82	$C_{13}H_{19}NO$
6a	47	yellow oil	57.04 / 57.14	5.11 / 5.16	4.98 / 5.13	$C_{13}H_{14}F_3NO_2$
6b	48	47—48 (LP)	53.49 / 53.61	4.48 / 4.50	4.80 / 4.81	$C_{13}H_{13}F_4NO_2$
6c	60	94—95 (EA—LP)	58.09 / 58.15	4.90 / 4.88	6.14 / 6.16	$C_{11}H_{11}F_2NO_2$
6d	51	yellow oil	50.51 / 50.49	3.94 / 3.85	5.29 / 5.35	$C_{11}H_{10}ClF_2NO_2$
8a	91	57—58 (LP)	53.94 / 53.79	2.91 / 2.87	5.69 / 5.70	$C_{11}H_7ClF_3N$
8b	91	64—65 (LP)	50.09 / 50.12	2.21 / 2.29	5.11 / 5.31	$C_{11}H_6ClF_4N$
8c	72	119—120 (LP)	58.09 / 58.04	3.60 / 3.54	6.17 / 6.15	$C_{11}H_8ClF_2N$
8d	78	66—67 (LP)	50.32 / 50.41	2.67 / 2.69	5.24 / 5.34	$C_{11}H_7Cl_2F_2N$
9	60	190—191 (EA—LP)	58.61 / 58.55	2.75 / 2.84	10.82 / 10.78	$C_{19}H_{11}ClF_3N_3O$
10	30	256—258 (CH_2Cl_2—LP)	64.61 / 64.59	2.79 / 2.85	11.78 / 11.89	$C_{19}H_{10}F_3N_3O$

Activity of topoisomerase I. Compound **10** caused inhibition of topoisomerase I activity at relatively high concentration, 40 μmol L^{-1} (Fig. 1). Even at this concentration, only insignificant amount of the rapidly migrating plasmid was found. The lower concentrations had no effect of retarding untwisting of DNA. Like for cytotoxicity, the range of concentrations inhibiting topoisomerase I demonstrates low activity of **10** with respect to topoisomerase I. Hence, a conclusion can be drawn that the luotonin derivative synthesized suppresses the topoisomerase I activity only in relatively high concentrations.

Thus, we developed an efficient and reliable scheme for the synthesis of hitherto unknown 3-bromomethyl-2-chlo-

Table 2. Spectral characteristics of compounds **3a—d**, **4b—d**, **5d**, **6a—d**, **8a—d**, **9**, **10**

Compound	NMR ($CDCl_3$), δ (*J*/Hz)			MS (EI, 70 eV), *m/z* (I_{rel} (%))
	^1H	^{13}C	^{19}F	
3a	4.96 (s, 2 H, CH_2Br); 7.76 (t, 1 H, Ar, *J* = 7.8); 7.89 (t, 1 H, Ar, *J* = 7.5); 8.15 (d, 1 H, Ar, *J* = 8.5); 8.28 (dm, 1 H, Ar, *J* = 8.5)	25.55 (q, $J_{C,F}$ = 6.1), 123.07, 123.72 (q, $J_{C,F}$ = 278.3), 125.00 (q, $J_{C,F}$ = 5.1), 128.42, 128.96, 129.49, 131.56, 134.81 (q, $J_{C,F}$ = 30.2), 147.51, 151.65	24.73 (s, CF_3)	327 $[M + H]^+$ (4), 325 $[M + H]^+$ (11), 245 (39), 244 (17), 243 (100), 209 (17), 208 (31), 204 (75), 176 (44), 140 (14)
3b	5.01 (s, 2 H, CH_2Br); 7.67 (td, 1 H, Ar, *J* = 7.7, *J* = 2.5); 7.91 (dm, 1 H, Ar, *J* = 9.7); 8.15 (dd, 1 H, Ar, *J* = 9.3, *J* = 5.5)	25.28 (q, $J_{C,F}$ = 5.5), 109.45 (qd, $J_{C,F}$ = 26.2, $J_{C,F}$ = 5.2), 121.99 (d, $J_{C,F}$ = 26.2), 123.51 (q, $J_{C,F}$ = 278.7), 124.00 (d, $J_{C,F}$ = 10.6), 129.52, 132.00 (d, $J_{C,F}$ = 9.5), 134.08 (qd, $J_{C,F}$ = 31.0, $J_{C,F}$ = 5.8), 144.63, 151.01, 161.63 (d, $J_{C,F}$ = 251.7)	−31.54 (s, F, 1 F); 22.02 (s, CF_3, 3 F)	345 $[M + H]^+$ (18), 343 $[M + H]^+$ (31), 263 (40), 262 (17), 261 (100), 226 (26), 225 (40), 207 (59), 176 (57), 158 (44)
3c	4.95 (s, 2 H, CH_2Br); 7.47 (t, 1 H, CF_2H, *J* = 53.0); 7.74 (td, 1 H, Ar, *J* = 7.4, *J* = 1.4); 7.88 (td, 1 H, Ar, *J* = 7.4, *J* = 0.9); 8.14 (dd, 1 H, Ar, *J* = 8.4, *J* = 0.9); 8.36 (dm, 1 H, Ar, *J* = 8.4)	24.50, 112.10 (t, $J_{C,F}$ = 241.3), 123.41, 124.68 (t, $J_{C,F}$ = 3.4), 128.18 (t, $J_{C,F}$ = 5.2), 128.53, 129.41, 131.57, 138.31 (t, $J_{C,F}$ = 20.7), 147.72, 150.59	−32.66 (s, CF_2H)	309 $[M + H]^+$ (15), 307 $[M + H]^+$ (20), 228 (40), 226 (100), 190 (32), 140 (45)
3d	5.00 (s, 2 H, CH_2Br); 7.75 (td, 1 H, Ar, *J* = 7.1, *J* = 1.4); 7.88 (td, 1 H, Ar, *J* = 8.4, *J* = 1.1); 8.14 (dd, 1 H, Ar, *J* = 8.4, *J* = 0.9); 8.36 (dm, 1 H, Ar, *J* = 8.4)	26.20 (t, $J_{C,F}$ = 8.2), 122.07, 125.07 (t, $J_{C,F}$ = 300.1), 125.30 (t, $J_{C,F}$ = 7.2), 126.44, 128.70, 129.57, 131.48, 140.21 (t, $J_{C,F}$ = 23.5), 147.67, 151.84	34.34 (d, AB system, J_{AB} = 169.7), 35.43 (d, AB system, J_{AB} = 169.7)	343 $[M + H]^+$ (19), 341 $[M + H]^+$ (31), 305 (13), 261 (60), 259 (83), 224 (85), 190 (100), 189 (89), 140 (75)
4a*	2.32 (q, 3 H, Me, *J* = 3.7); 7.24 (t, 1 H, Ar, *J* = 7.6); 7.38 (d, 1 H, Ar, *J* = 8.3); 7.53 (t, 1 H, Ar, *J* = 7.6); 7.70 (d, 1 H, Ar, *J* = 7.1); 12.33 (br.s, 1 H, NH)	13.27 (q, $J_{C,F}$ = 4.4), 114.66, 116.34, 122.97, 124.62 (q, $J_{C,F}$ = 278.6), 124.67 (q, $J_{C,F}$ = 4.4), 130.63, 131.85 (q, $J_{C,F}$ = 28.8), 132.82, 137.82, 161.31	23.60 (s, CF_3)	228 $[M + H]^+$ (30), 227 $[M]^+$ (100), 199 (82), 198 (40), 158 (27), 130 (93)
4b*	2.38 (q, 3 H, Me, *J* = 3.9); 7.36—7.42 (m, 2 H, Ar); 7.54 (td, 1 H, Ar, *J* = 8.5, *J* = 2.4); 12.39 (br.s, 1 H, NH)	13.43 (q, $J_{C,F}$ = 4.4), 109.96 (qd, $J_{C,F}$ = 26.5, $J_{C,F}$ = 5.5), 115.12 (d, $J_{C,F}$ = 7.7), 118.18 (d, $J_{C,F}$ = 8.9), 118.90 (d, $J_{C,F}$ = 24.3), 124.35 (q, $J_{C,F}$ = 278.6), 131.30 (qd, $J_{C,F}$ = 29.9, $J_{C,F}$ = 3.3), 134.54, 134.64, 157.53 (d, $J_{C,F}$ = 237.7), 160.96	−40.89 (s, F, 1 F), 23.60 (s, CF_3, 3 F)	246 $[M + H]^+$ (30), 245 $[M]^+$ (100), 217 (43), 216 (86), 207 (22), 176 (22), 148 (80)
4c*	2.28 (t, 3 H, Me, *J* = 1.8); 7.23 (dd, 1 H, Ar, *J* = 7.7, *J* = 1.0); 7.37 (d, 1 H, Ar, *J* = 7.7); 7.51 (t, 1 H, Ar, *J* = 7.7); 7.58 (t, 1 H, CF_2H, *J* = 52.7); 7.93 (d, 1 H, Ar, *J* = 8.2); 12.13 (br.s, 1 H, NH)	12.21, 114.38 (t, $J_{C,F}$ = 237), 115.42, 116.18, 122.52, 125.00, 130.22, 132.09 (t, $J_{C,F}$ = 8), 135.56 (t, $J_{C,F}$ = 22), 138.03, 161.70	−35.37 (s, CF_2H)	210 $[M + H]^+$ (30), 209 $[M]^+$ (100), 181 (22), 180 (36), 162 (17), 161 (20), 130 (53)
4d**	2.57 (t, 3 H, Me, *J* = 4.8); 7.35 (dd, 1 H, Ar, *J* = 7.6, *J* = 8.2); 7.41 (d, 1 H, Ar, *J* = 8.2); 7.59 (dd, 1 H, Ar, *J* = 8.2, *J* = 7.6); 8.06 (d, 1 H, Ar, *J* = 8.2); 11.40 (br.s, 1 H, NH)	14.00 (t, $J_{C,F}$ = 6.9), 114.09, 116.49, 122.81, 125.09 (t, $J_{C,F}$ = 6.6), 126.07 (t, $J_{C,F}$ = 293.7), 130.57, 130.60, 137.41 (t, $J_{C,F}$ = 23.3), 137.99, 161.38	33.98 (s, CF_2Cl)	245 $[M]^+$ (8), 243 $[M]^+$ (21), 209 (18), 208 (100), 207 (12), 180 (76), 179 (23), 130 (14)

(*to be continued*)

Table 2 (*continued*)

Compound	NMR ($CDCl_3$), δ (*J*/Hz)			MS (EI, 70 eV), *m*/*z* (I_{rel} (%))
	^1H	^{13}C	^{19}F	
5d	1.00 (t, 3 H, $(CH_2)_3\underline{Me}$, *J* = 7.3); 1.34 (t, 3 H, $CH_2\underline{Me}$, *J* = 7.3), 1.44 (m, 2 H, $(CH_2)_2C\underline{H}_2Me$); 1.63 (m, 2 H, $CH_2C\underline{H}_2CH_2Me$); 2.49 (q, 2 H, $C\underline{H}_2Me$, *J* = 7.3); 2.63 (t, 2 H, $C\underline{H}_2(CH_2)_2Me$, *J* = 7.3); 7.07 (br.s, 1 H, NH); 7.13—7.29 (m, 3 H, Ar); 7.87 (d, 1 H, Ar, *J* = 7.5)	9.96, 13.98, 22.60, 30.55, 31.13, 32.00, 124.23, 125.37, 126.59, 129.47, 134.16, 135.11, 172.31	—	205 $[M]^+$ (11), 176 (72), 148 (17), 120 (14), 107 (18), 106 (100)
6a	1.35 (s, 9 H, 3 Me); 7.17 (ddd, 1 H, Ar, *J* = 8.3, *J* = 7.3, *J* = 1.1); 7.68 (ddd, 1 H, Ar, *J* = 8.7, *J* = 7.3, *J* = 1.6); 7.96 (dm, 1 H, Ar, *J* = 8.3); 8.88 (dd, 1 H, Ar, *J* = 8.7, *J* = 1.0); 11.24 (br.s, 1 H, NH)	27.51, 40.61, 115.35, 116.62 (q, $J_{C,F}$ = 291.2), 121.21, 122.43, 131.85 (q, $J_{C,F}$ = 4.4), 137.76, 144.06, 178.38, 182.70 (q, $J_{C,F}$ = 33.7)	8.49 (s, CF_3)	273 $[M]^+$ (4), 230 (9), 205 (15), 204 (100), 207 (24), 189 (14), 146 (15), 121 (7), 120 (56)
6b	1.42 (s, 9 H, 3 Me); 7.51 (ddd, 1 H, Ar, *J* = 9.4, *J* = 7.3, *J* = 3.0); 7.70 (dm, 1 H, Ar, *J* = 9.4); 8.98 (dd, 1 H, Ar, *J* = 9.4, *J* = 5.0); 11.14 (br.s, 1 H, NH)	27.48, 40.55, 115.85 (d, $J_{C,F}$ = 6.3), 116.31 (q, $J_{C,F}$ = 291.4); 117.28 (d.q, $J_{C,F}$ = 24.4, $J_{C,F}$ = 4.3); 123.25 (d, $J_{C,F}$ = 6.9), 125.24 (d, $J_{C,F}$ = 21.8); 140.51, 156.71 (d, $J_{C,F}$ = 244.8); 178.26, 182.30 (q, $J_{C,F}$ = 35.1)	−40.01 (s, F, 1 F); 8.00 (s, CF_3, 3 F)	292 $[M + H]^+$ (7), 291 $[M]^+$ (13), 248 (12), 223 (14), 222 (100), 207 (24), 164 (19), 139 (19), 138 (52)
6c	1.35 (t, 3 H, Me, *J* = 7.5); 2.56 (q, 2 H, CH_2, *J* = 7.5); 6.45 (t, 1 H, CF_2H, *J* = 53.4); 7.22 (td, 1 H, Ar, *J* = 8.2, *J* = 0.8); 7.73 (td, 1 H, Ar, *J* = 8.6, *J* = 1.2); 8.04 (dd, 1 H, Ar, *J* = 8.2,*J* = 1.5); 8.90 (d, 1 H, Ar, *J* = 8.6); 11.22 (br.s, 1 H, NH)	9.41, 31.70, 110.52 (t, $J_{C,F}$ = 253.4); 116.77, 121.03, 122.40, 131.36 (t, $J_{C,F}$ = 4.3); 137.06, 142.95, 173.25, 190.10 (t, $J_{C,F}$ = 24.1)	−43.03 (s, CF_2H)	227 $[M]^+$ (5), 177 (14), 176 (100), 171 (15), 149 (14), 120 (95)
6d	1.34 (t, 3 H, Me, *J* = 7.5); 2.56 (q, 2 H, CH_2, *J* = 7.5); 7.21 (ddd, 1 H, Ar, *J* = 8.5, *J* = 7.3, *J* = 1.3); 7.72 (ddd, 1 H, Ar, *J* = 8.5, *J* = 7.3, *J* = 1.3); 8.15 (dm, 1 H, Ar, *J* = 8.5, *J* = 1.0); 8.89 (dd, 1 H, Ar, *J* = 8.5, *J* = 1.0); 11.00 (br.s, 1 H, NH)	9.41, 31.70, 114.46, 119.85 (t, $J_{C,F}$ = 304.9); 121.40, 122.26, 132.00 (t, $J_{C,F}$ = 4.9); 137.41, 143.74, 173.16, 183.85 (t, $J_{C,F}$ = 28.4)	18.87 (s, CF_2Cl)	263 $[M]^+$ (13), 261 $[M]^+$ (42), 206 (75), 176 (43), 121 (33), 120 (100)
8a	2.82 (q, 3 H, Me, *J* = 2.8); 7.70 (dd, 1 H, Ar, *J* = 8.2, *J* = 7.3); 7.81 (dd, 1 H, Ar, *J* = 8.2, *J* = 7.3); 8.11 (d,1 H, Ar, *J* = 8.6); 8.26 (d,1 H, Ar, *J* = 8.6)	17.85 (q, $J_{C,F}$ = 4.6); 123.27, 124.18 (q, $J_{C,F}$ = 5.1); 124.28 (q, $J_{C,F}$ = 278.2); 128.40, 129.25, 129.87 (q, $J_{C,F}$ = 2.0); 130.11, 134.20 (q, $J_{C,F}$ = 30.2); 146.37, 152.94	24.71 (s, CF_3)	247 $[M]^+$ (30), 245 $[M]^+$ (100), 210 (13), 209 (25), 190 (35), 189 (73), 140 (20)
8b	2.76 (q, 3 H, Me, *J* = 2.9); 7.54 (td, 1 H, Ar, *J* = 8.3, *J* = 2.4); 7.84 (d, 1 H, Ar, *J* = 11.0); 8.05 (dd, 1 H, Ar, *J* = 9.2, *J* = 5.7)	17.88 (q, $J_{C,F}$ = 4.4); 108.68 (qd, $J_{C,F}$ = 26.5, $J_{C,F}$ = 5.5); 120.38 (d, $J_{C,F}$ = 25.4); 124.07 (d, $J_{C,F}$ = 10.0); 124.09 (q, $J_{C,F}$ = 278.6); 131.05, 131.75 (d, $J_{C,F}$ = 10.0); 133.71 (qd, $J_{C,F}$ = 30.5, $J_{C,F}$ = 6.6); 143.54, 152.32 (d, $J_{C,F}$ = 3.3); 161.40 (d, $J_{C,F}$ = 249.9)	−30.93 (s, F, 1 F); 23.99 (s, CF_3, 3 F)	265 $[M]^+$ (30), 263 $[M]^+$ (60), 228 (42), 209 (43), 208 (100), 158 (81)

(*to be continued*)

Table 2 (*continued*)

Compound	NMR ($CDCl_3$), δ (*J*/Hz)			MS (EI, 70 eV), *m/z* (I_{rel} (%))
	^1H	^{13}C	^{19}F	
8c	2.76 (t, 3 H, Me, *J* = 1.6); 7.42 (t, 1 H, CF_2H, *J* = 53.5); 7.66 (ddd, 1 H, Ar, *J* = 8.4, *J* = 7.3, *J* = 1.1); 7.80 (dd, 1 H, Ar, *J* = 8.4, *J* = 7.3); 8.11 (d, 1 H, Ar, *J* = 8.4); 8.31 (dd, 1 H, Ar, *J* = 8.4, *J* = 1.1)	16.08, 112.79 (t, $J_{C,F}$ = 230.1); 123.67, 123.72 (t, $J_{C,F}$ = 1.9); 127.97, 129.11 (t, $J_{C,F}$ = 5.5); 129.22, 130.04, 136.89 (t, $J_{C,F}$ = 22.1); 146.52, 152.38	−33.31 (s, CF_2H)	229 $[M]^+$ (37), 227 $[M]^+$ (100), 192 (5), 172 (12), 171 (28)
8d	2.79 (t, 3 H, Me, *J* = 4.0); 7.70 (ddd, 1 H, Ar, *J* = 8.6, *J* = 7.0, *J* = 1.4); 7.80 (ddd, 1 H, Ar, *J* = 8.4, *J* = 7.0, *J* = 1.0); 8.11 (dd, 1 H, Ar, *J* = 8.4, *J* = 1.0); 8.31 (dm, 1 H, Ar, *J* = 8.6)	18.42 (t, $J_{C,F}$ = 6.6); 122.26, 124.45 (t, $J_{C,F}$ = 7.7); 125.59 (t, $J_{C,F}$ = 294.1); 128.11, 128.16, 129.31, 130.00, 139.68 (t, $J_{C,F}$ = 24.3); 146.54, 152.98	35.56 (s, CF_2Cl)	263 $[M]^+$ (20), 261 $[M]^+$ (34), 228 (32), 227 (12), 226 (100), 191 (10), 190 (33), 188 (20)
9	5.65 (q, 2 H, CH_2, *J* = 1.0); 7.51 (ddd, 1 H, Ar, *J* = 8.2, *J* = 7.0, *J* = 1.4); 7.67—7.80 (m, 3 H, Ar); 7.85 (s, 1 H, H(2));7.87 (ddd, 1 H, Ar, *J* = 8.2, *J* = 7.0, *J* = 1.4); 8.12 (dd, 1 H, Ar, *J* = 8.4, *J* = 0.9); 8.20—8.31 (m, 2 H, Ar)	45.44 (q, $J_{C,F}$ = 4.4); 121.78, 122.93, 123.63 (q, $J_{C,F}$ = 278.6); 124.63, 124.94 (q, $J_{C,F}$ = 5.5); 126.75, 127.55, 127.55, 129.24, 129.54, 132.00, 134.57, 137.60 (q, $J_{C,F}$ = 29.8); 144.57, 147.63, 147.99, 151.82, 161.04	24.80 (s, CF_3)	355 $[M + H - Cl]^+$ (22), 341 $[M - Cl]^+$ (100), 227 (11), 226 (59)
10	5.62 (q, 2 H, CH_2, *J* = 2.4); 7.68 (t, 1 H, Ar, *J* = 7.0); 7.89—7.98 (m, 2 H, Ar); 8.03 (dd, 1 H, Ar, *J* = 7.8, *J* = 7.1); 8.20 (d, 1 H, Ar, *J* = 8.1); 8.37 (d, 1 H, Ar, *J* = 8.1); 8.52 (d, 1 H, Ar, *J* = 7.8); 8.66 (d, 1 H, Ar, *J* = 8.6)	47.93 (q, $J_{C,F}$ = 5.5); 121.22, 123.60, 123.74 (q, $J_{C,F}$ = 278.6); 124.07 (q, $J_{C,F}$ = 2.2), 126.40, 127.16 (q, $J_{C,F}$ = 2.2), 127.76, 128.70, 130.20 (q,$J_{C,F}$ = 33.2), 130.26, 131.27, 131.52, 134.70, 148.92, 150.06, 151.11, 151.32, 160.04	20.73 (s, CF_3)	354 $[M + H]^+$ (9), 353 $[M]^+$ (42), 325 (13), 306 (23), 292 (26), 291 (38), 277 (39), 235 (26), 219 (26), 203 (31), 189 (71), 147 (100)

* ^1H, ^{13}C, and ^{19}F NMR spectra were recorded in DMSO-d_6.
** ^{13}C NMR spectrum was obtained in DMSO-d_6.

ro-4-fluoromethylquinolines **3**, which are promising building blocks for the synthesis of 7-fluoromethyl-containing alkaloids of the camptothecin series, in particular, 7-trifluoromethylluotonin **10**. It was found that 7-trifluoromethylluotonin **10** retains some antitumor activity, causing death of tumor cells and inhibiting topoisomerase I. Using the method developed for the synthesis of 7-fluoromethyl-substituted luotonins, we hope to optimize the structure of topoisomerase I inhibitors of the camptothecin series, the prototypes of the target-specific antitumor drugs.

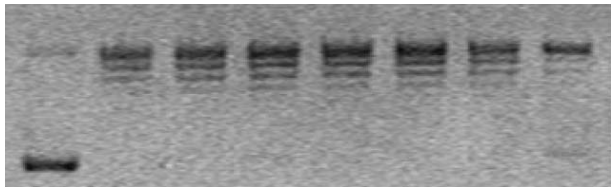


Fig. 1. Influence of compound **10** on the activity of topoisomerases I *in vitro*. *C* is concentration of compound **10**. DNAst is a supertwisted DNA.

Experimental

^1H and $^{13}C\{^1H\}$ NMR spectra were recorded on a Bruker Avance^{TM}600 spectrometer (600.22 MHz and 150.925 MHz, respectively). Chemical shifts for protons and ^{13}C nuclei were determined with respect to the residual signal for chloroform (7.27 ppm) or the signal for $CDCl_3$ (77.0 ppm), respectively and recalculated from the $SiMe_4$ signal. Chemical shifts were determined with accuracy no less than 0.001 ppm and 0.03 ppm, respectively. ^{19}F $\{^1H\}$ NMR spectra were recorded on a Bruker Avance^{TM}300 spectrometer (288.38 MHz). Chemical shifts for ^{19}F nuclei were determined with respect to trifluoro-

acetic acid as an external standard with accuracy no less than 0.01 ppm.

Mass spectra were obtained on a Finnigan Polaris Q instrument (ion trap, 70 eV, the DIP procedure was used for the sample injection) and on a MS-30 Kratos instrument (70 eV, temperature of ionizing chamber was 200 °C, a system for direct injection of the sample was used). Silica gel with the particle size of 0.06—0.20 mm (Merck Kieselgel 60) was used for column chromatography. Monitoring of the reaction course and purity of products obtained was performed using Merck Kieselgel 60 F_{254} TLC plates. *n*-Butyllithium (1.6 *M* solution in hexane) was purchased from Acros. Tetrahydrofuran (Aldrich) was prepared for the lithiation reaction by reflux over sodium metal in the presence of benzophenone until dark blue color of the solution was persistent with subsequent distillation under argon. All the lithiation reactions were performed under argon. Elemental analysis was performed in the Laboratory of Elemental Analysis of the A. N. Nesmeyanov Institute of Organoelement Compounds, Russian Academy of Sciences. Light petroleum (LP) and ethyl acetate (EA) were used as the solvents.

N-Substituted anilines **5a,b** were synthesized using a general procedure.[21] Physicochemical and spectral characteristics of *N*-phenyl-2,2-dimethylpropionamide[22] (**5a**) and *N*-(4-fluorophenyl)-2,2-dimethylpropionamide[16b] (**5b**) completely agreed with the data reported in the literature. *N*-(2-Bromophenyl)-propionamide (**5c**) was synthesized using procedure given in work.[23]

Compounds **3a—d**, **4a—d**, **5d**, **5e**, **8a—d** are colorless crystalline substances.

***N*-[2-(Trifluoroacetyl)phenyl]-2,2-dimethylpropionamide (6a).** *n*-Butyllithium (1.6 *M* solution in hexane, 78.2 mL, 125.1 mmol) was added over 1 h to a solution of *N*-phenyl-2,2-dimethylpropionamide (**5a**) (7.4 g, 41.7 mmol) in anhydrous THF (80 mL) at temperatures from −10 °C to 0 °C. A suspension obtained was stirred for 3 h at −5—0 °C. Then the reaction mixture was cooled to −70 °C, followed by addition of ethyl trifluoroacetate (20.9 g, 147.1 mmol). The reaction mixture was stirred for 1 h at −70 °C, slowly warmed to 0 °C, treated with saturated aqueous NH_4Cl (50 mL) and ethyl acetate (150 mL), the water layer was separated. The organic layer was washed with brine (2×50 mL), dried with $MgSO_4$, and concentrated. The residue was separated by column chromatography eluting with a LP—EA (6 : 1) solvent mixture to obtain compound **6a** (5.4 g) as a yellowish dense liquid.

***N*-[4-Fluoro-2-(trifluoroacetyl)phenyl]-2,2-dimethylpropionamide (6b)** was obtained similarly from *N*-(4-fluorophenyl)-2,2-dimethylpropionamide (**5b**) and ethyl trifluoroacetate. The product as light yellow crystals was isolated by column chromatography on silica gel with LP—CH_2Cl_2 (1 : 1) solvent mixture as the eluent.

***N*-[2-(Difluoroacetyl)phenyl]propionamide (6c).** *n*-Butyllithium (1.6 *M* solution in hexane, 10.5 mL, 33 mmol) was added to a solution of *N*-(2-bromophenyl)propionamide (**5c**) (3.64 g, 16 mmol) in anhydrous THF (50 mL) at −75 °C. The solution obtained was stirred for 1 h at −75 °C followed by addition of ethyl difluoroacetate (3.97 g, 32 mmol). The reaction mixture was stirred for 1 h at −75 °C, slowly warmed to 0 °C, treated with saturated aq. NH_4Cl (10 mL) and ethyl acetate (50 mL), the water layer was separated. The organic layer was washed with brine (2×20 mL), dried with $MgSO_4$, and concentrated. The residue was separated by column chromatography eluting with a LP—EA (3 : 1) solvent mixture. Compound **6c** (2.2 g) was isolated first as light yellow crystals. Further elution of the column gave first *N*-(2-*n*-butylphenyl)propionamide (**5d**) (0.32 g) and then *N*-phenylpropionamide (**5e**) (0.2 g), m.p. 103—104 °C (from LP); see Ref. 24: 103—104 °C. ^{1}H NMR spectrum of compound **5e** completely agreed with that reported in the literature.[24]

***N*-[2-(Chlorodifluoroacetyl)phenyl]propionamide (6d)** was obtained from *N*-(2-bromophenyl)propionamide (**5c**) and ethyl chlorodifluoroacetate similarly the synthesis of compound **6c** as a yellowish dense liquid.

Ethyl 4,4,4-trifluoro-3-[2-(2,2-dimethylpropionylamino)-phenyl]-2-methylbut-2-enoate (7a) (a mixture of *E*,*Z*-isomers in the ratio 3 : 1). A solution of (1-ethoxycarbonylethylidene)-triphenylphosphorane (0.77 g, 2.1 mmol) and compound **6a** (0.58 g, 2.1 mmol) in anhydrous toluene (10 mL) was heated for 6 h at 100 °C. Toluene was evaporated *in vacuo*. The residue was extracted with boiling pentane (3×30 mL). A precipitate formed after cooling pentane was filtered off. The pentane extracts were concentrated. The residue was purified by column chromatography on silica gel using a LP—EA (6 : 1) solvent mixture as an eluent to obtain compound **7a** (0.55 g) as a colorless dense liquid. The yield was 73%. ^{1}H NMR of the predominant isomer in the mixture of isomers ($CDCl_3$), δ: 0.91 (t, 3 H, $CH_2\underline{Me}$, J = 7.1 Hz); 1.35 (s, 9 H, 3 Me); 2.36 (q, 3 H, Me(2), J = 2.2 Hz); 3.96 (q, 2 H, $C\underline{H}_2Me$, J = 7.1 Hz); 7.11—7.14 (m, 2 H, Ar); 7.43 (td, 1 H, Ar, J = 8.4 Hz, J = 4.5 Hz); 7.60 (br.s, 1 H, NH); 8.16 (d, 1 H, Ar, J = 8.2 Hz). ^{19}F NMR of the predominant isomer in the mixture of isomers ($CDCl_3$), δ: 18.96 (s, CF_3). ^{1}H NMR of the minor isomer in the mixture of isomers ($CDCl_3$), δ: 1.35 (s, 9 H, 3 Me); 1.42 (t, 3 H, $CH_2\underline{Me}$, J = 7.1 Hz); 1.83 (q, 3 H, Me(2), J = 2.4 Hz); 4.39 (q, 2 H, $C\underline{H}_2Me$, J = 7.14 Hz); 7.18—7.24 (m, 2 H, Ar); 7.51 (td, 1 H, Ar, J = 8.4 Hz, J = 4.5 Hz); 7.52 (br.s, 1 H, NH); 8.36 (d, 1 H, Ar, J = 8.2 Hz). ^{19}F NMR of the minor isomer in the mixture of isomers ($CDCl_3$), δ: 15.61 (s, CF_3). No analytically pure sample was obtained.

Ethyl 3-[5-fluorophenyl-2-(2,2-dimethylpropionylamino)]-4,4,4-trifluoro-2-methylbut-2-enoate (7b) (a mixture of *E*,*Z*-isomers in the ratio 4 : 1) was obtained from compound **6b** similarly to the preparation and isolation of compound **7a**. Colorless dense liquid. The yield was 70%. ^{1}H NMR of the predominant isomer in the mixture of isomers ($CDCl_3$), δ: 0.97 (t, 3 H, $CH_2\underline{Me}$, J = 7.0 Hz); 1.33 (s, 9 H, 3 Me); 2.35 (q, 3 H, Me(2), J = 2.0 Hz); 4.01 (q, 2 H, $C\underline{H}_2Me$, J = 7.0 Hz); 6.89 (dd, 1 H, Ar, J = 8.3 Hz, J = 2.8 Hz); 7.13 (td, 1 H, Ar, J = 8.4 Hz, J = 2.5 Hz); 7.53 (br.s, 1 H, NH); 8.05 (dd, 1 H, Ar, J = 8.4 Hz, J = 5.3 Hz). ^{19}F NMR of the predominant isomer in the mixture of isomers ($CDCl_3$), δ: −40.01 (s, 1 F); 19.05 (s, 3 F). ^{1}H NMR of the minor isomer in the mixture of isomers ($CDCl_3$), δ: 1.34 (s, 9 H, 3 Me); 1.41 (t, 3 H, $CH_2\underline{Me}$, J = 7.0 Hz); 1.83 (q, 3 H, Me(2), J = 2.0 Hz); 4.39 (q, 2 H, $C\underline{H}_2Me$, J = 7.3 Hz); 6.99 (dd, 1 H, Ar, J = 8.3 Hz, J = 3.0 Hz); 7.20 (td, 1 H, Ar, J = 8.4 Hz, J = 2.7 Hz); 7.42 (br.s, 1 H, NH); 8.27 (dd, 1 H, Ar, J = 8.4 Hz, J = 5.0 Hz). ^{19}F NMR of the minor isomer in the mixture of isomers ($CDCl_3$), δ: −39.34 (s, 1 F); 15.61 (s, 3 F). No analytically pure sample was obtained.

Ethyl 4,4-difluoro-2-methyl-3-(2-propionylaminophenyl)but-2-enoate (7c) (a mixture of *E*,*Z*-isomers in the ratio 1 : 2) was obtained from compound **6c** similarly to the synthesis of compound **7a**. A predominant isomer with higher R_f was isolated by column chromatography on silica gel as colorless crystals eluting with a LP—EA (2 : 1) solvent mixture. M.p. 68—69 °C. ^{1}H NMR of the predominant isomer ($CDCl_3$), δ: 1.27 (t, 3 H, $CH_2\underline{Me}$, J = 7.5 Hz); 1.43 (t, 3 H, $OCH_2\underline{Me}$, J = 7.2 Hz); 1.79 (t, 3 H,

Me(2), J = 3.4 Hz); 2.42 (q, 2 H, C$\underline{H}_2$Me, J = 7.5 Hz); 4.40 (q, 2 H, OC$\underline{H}_2$Me, J = 7.2 Hz); 7.07 (t, 1 H, CF_2H, J = 55.0 Hz); 7.07—7.27 (m, 3 H, Ar, NH); 7.49 (t, 1 H, Ar, J = 7.0 Hz); 8.22 (d, 1 H, Ar, J = 7.0 Hz). ^{19}F NMR of the predominant isomer ($CDCl_3$), δ: −36.99 (d, AB-system, J_{AB} = 319.1 Hz); −34.09 (d, AB-system, J_{AB} = 319.1 Hz). ^{13}C NMR of the predominant isomer ($CDCl_3$), δ: 9.64, 14.08, 17.35, 31.92, 61.98, 111.91 (t, $J_{C,F}$ = 234.5 Hz), 122.75, 123.64, 124.51, 129.61, 129.90, 135.54, 135.74 (t, $J_{C,F}$ = 24.4 Hz), 138.48 (t, $J_{C,F}$ = 8.3 Hz), 166.97, 172.04. MS (EI, 70 eV), m/z (I_{rel} (%)): 311 $[M]^+$ (6), 255 (19), 254 (23), 210 (59), 182 (89), 181 (26), 162 (100). Found (%): C, 61.79; H, 6.27; N, 4.44. $C_{16}H_{19}F_2NO_3$. Calculated (%): C, 61.73; H, 6.15; N, 4.50. The minor isomer was isolated by further elution in a mixture with the predominant isomer. ^{1}H NMR of the minor isomer in the mixture of isomers ($CDCl_3$), δ: 0.88 (t, 3 H, OCH$_2$$\underline{Me}$, J = 7.0 Hz); 1.27 (t, 3 H, CH$_2$$\underline{Me}$, J = 7.5 Hz); 2.29 (t, 3 H, Me(2), J = 1.0 Hz); 2.43 (q, 2 H, C$\underline{H}_2$Me, J = 7.5 Hz); 3.94 (q, 2 H, OC$\underline{H}_2$Me, J = 7.0 Hz); 6.59 (t, 1 H, CF_2H, J = 56.0 Hz); 7.07—7.27 (m, 3 H, Ar, NH); 7.40 (t, 1 H, Ar, J = 7.0 Hz); 8.11 (d, 1 H, Ar, J = 7.3 Hz). ^{19}F NMR of the minor isomer in the mixture of isomers ($CDCl_3$), δ: −37.13 (d, AB-system, J_{AB} = 313.6 Hz); −34.65 (d, AB-system, J_{AB} = = 313.6 Hz). The overall yield of two isomers was 63%.

Ethyl 4-chloro-4,4-difluoro-2-methyl-3-(2-propionylaminophenyl)but-2-enoate (7d) (a mixture of *E*,*Z*-isomers in the ratio 2 : 3) was obtained from compound **6d** similarly to the preparation and isolation of compound **7a**. Yellowish dense liquid. The yield was 53%. ^{1}H NMR of the predominant isomer ($CDCl_3$), δ: 1.32 (t, 3 H, CH$_2$$\underline{Me}$, J = 7.5 Hz); 1.44 (t, 3 H, OCH$_2$$\underline{Me}$, J = 7.2 Hz); 1.79 (t, 3 H, Me(2), J = 3.4 Hz); 2.47 (q, 2 H, C$\underline{H}_2$Me, J = 7.5 Hz); 4.40 (q, 2 H, OC$\underline{H}_2$Me, J = 7.2 Hz); 7.20—7.37 (m, 3 H, Ar, NH); 7.51 (t, 1 H, Ar, J = 7.5 Hz); 8.31 (d, 1 H, Ar, J = 8.2 Hz). ^{19}F NMR of the predominant isomer ($CDCl_3$), δ: 27.83 (d, AB-system, J_{AB} = 161.3 Hz); 29.33 (d, AB-system, J_{AB} = 161.3 Hz). ^{1}H NMR of the minor isomer in a mixture of isomers ($CDCl_3$), δ: 0.92 (t, 3 H, OCH$_2$$\underline{Me}$, J = 7.0 Hz); 1.27 (t, 3 H, CH$_2$$\underline{Me}$, J = 7.5 Hz); 2.39 (t, 3 H, Me(2), J = 1.0 Hz); 2.43 (q, 2 H, C$\underline{H}_2$Me, J = 7.5 Hz); 3.94 (q, 2 H, OC$\underline{H}_2$Me, J = 7.0 Hz); 7.20—7.37 (m, 3 H, Ar, NH); 7.44 (t, 1 H, Ar, J = 7.0 Hz); 8.13 (d, 1 H, Ar, J = 8.2 Hz). ^{19}F NMR of the minor isomer in a mixture of isomers ($CDCl_3$), δ: 31.55 (d, AB-system, J_{AB} = 161.3 Hz); 32.98 (d, AB-system, J_{AB} = 161.3 Hz). No analytically pure sample was obtained.

3-Methyl-4-trifluoromethylquinolin-2(1*H*)-one (4a). Compound **7a** (0.55 g, 1.5 mmol) was added to a mixture of HCl (6 *M*, 5 mL) and acetic acid (5 mL) and this was refluxed for 6 h Crystals formed after cooling were filtered off and washed with water on the filter until the washings were neutral to obtain compound **4a** (0.22 g, 62%). M.p. 235—236 °C (from ethanol); Ref. 15: 237—239 °C.

6-Fluoro-3-methyl-4-trifluoromethylquinolin-2(1*H*)-one (4b) was obtained from compound **7b** similarly to compound **4a**.

4-Difluoromethyl-3-methylquinolin-2(1*H*)-one (4c). Compound **7c** (2.23 g, 7.2 mmol) was refluxed for 4 h in HCl (2 *M*, 40 mL). Crystals formed after cooling were filtered off and washed with water on the filter until the washings were neutral to obtain compound **4c** (0.63 g).

4-Chlorodifluoromethyl-3-methylquinolin-2(1*H*)-one (4d) was obtained from compound **7d** similarly to compound **4c**.

2-Chloro-3-methyl-4-trifluoromethylquinoline (8a). A solution of **4a** (0.8 g, 3.5 mmol) in phosphorus oxychloride (2.7 g, 17.6 mmol) was refluxed for 1 h. The reaction mixture was cooled. Unreacted phosphorus oxychloride was evaporated *in vacuo* (12 Torr). The residue was poured in ice. Crystals formed were filtered off, washed with saturated aq. $NaHCO_3$ on the filter to pH 7, dried in air to obtain compound **8a** (0.78 g).

2-Chloro-6-fluoro-3-methyl-4-trifluoromethylquinoline (8b), 2-chloro-4-difluoromethyl-3-methylquinoline (8c), and **2-chloro-4-chlorodifluoromethyl-3-methylquinoline (8d)** were obtained similarly from the corresponding 3-methylquinolin-2(1*H*)-ones.

3-Bromomethyl-2-chloro-4-trifluoromethylquinoline (3a). A solution of **8a** (0.25 g, 1 mmol), *N*-bromosuccinimide (0.2 g, 1.1 mmol), and 1,1′-azobis(cycleohexanecarbonitrile) (10 mg) in dry CCl_4 (10 mL) was refluxed for 24 h. The reaction course was monitored by TLC using a LP—EA solvent mixture (25 : 1) as an eluent, R_f of the starting compound was 0.4; R_f of the product was 0.3. The solvent was evaporated, the residue was purified by column chromatography on silica gel with a mixture of LP and EA (15 : 1) as an eluent to obtain compound **3a** (0.24 g).

3-Bromomethyl-2-chloro-6-fluoro-4-trifluoromethylquinoline (3b), 3-bromomethyl-2-chloro-4-difluoromethylquinoline (3c) and **3-bromomethyl-2-chloro-4-chlorodifluoromethylquinoline (3d)** were obtained similarly from corresponding 2-chloro-3-methylquinolines.

3-[(2-Chloro-4-trifluoromethylquinolin-3-yl)methyl]quinazolin-4(3*H*)-one (9). Potassium *tert*-butoxide (0.2 g, 1.8 mmol) was added to a solution of 4(3*H*)-quinazolinone (0.24 g, 1.65 mmol) in dry DMF (20 mL). The reaction mixture was stirred for 1 h at 20 °C and cooled to 0 °C, followed by a dropwise addition of a solution of **3a** (0.52 g, 1.6 mmol) in DMF (10 mL). The reaction mixture was stirred for 1 h at 0 °C, warmed to 20 °C, diluted with 5% aq. Na_2CO_3 to pH 10, and extracted with ethyl acetate (3×30 mL). The organic extracts were combined, washed with brine, dried with $MgSO_4$, and concentrated. The product was purified by column chromatography on silica gel eluting with a LP—EA (1 : 1) solvent mixture to obtain compound **10** (350 mg) as grayish crystals.

7-Trifluoromethylluotonin or 14-trifluoromethylquino[2′,3′:3,4]-pyrrolo[2,1-*b*]quinazolin-11(13*H*)-one (10). Compound **9** (0.27 g, 0.69 mmol), $Pd(OAc)_2$ (16 mg, 0.07 mmol), tri(cyclohexyl)-phosphine (40 mg, 0.14 mmol), and potassium acetate (137 mg, 1.4 mmol) were mixed in a flask equipped with an inlet for an inert gas. Dry DMF (15 mL) was added to the mixture, a suspension obtained was stirred for 30 min under argon at 20 °C, then warmed to 160 °C, and kept for 15 min at this temperature. The reaction mixture was warmed to 20 °C, diluted with 5% aq. Na_2CO_3 to pH 10, and extracted with ethyl acetate (3×30 mL). The organic extract were combined, washed with brine, dried with $MgSO_4$, and concentrated. The product was purified by column chromatography on silica gel eluting with a LP—EA (1 : 1) solvent mixture to obtain compound **10** (73 mg) as grayish crystals.

Biological testing. *Cytotoxicity.* The method[25] is based on the reduction of tetrazolium salt to formazan in living cells. Compound **10** was dissolved in dimethyl sulfoxide to final concentration of 10 mmol L^{-1}. This solution was used for the preparation of the drug dilutions in the medium. The K562 leukemia and HCT116 colon carcinoma cell lines were propagated in Dulbecco modified Eagle's medium supplemented with 5% fetal calf serum, L-glutamine (2 mmol L^{-1}), penicillin (100 unit mL^{-1}), streptomycin (100 μg mL^{-1})). Cells were plated on 96-well plates ($3 \cdot 10^3$ cells in 190 μL of cultural medium). Compound **10** was

added at final concentrations 0.2—50 μmol L^{-1}. Control wells were filled with the medium. Each concentration of **10** was tested in triplicates. The cells were incubated for 72 h at 37 °C, 5% of CO_2, then aqueous solution of tetrazolium bromide (2.5 μg mL) was added. After 2 h, formazan was dissolved in dimethyl sulfoxide (100 μL), and optical density was measured at 540 nm on an LKB spectrophotometer (Sweden). The percentage of dying cells at given concentration of **10** was calculated as average optical density in the wells with the agent divided by average optical density of control wells.

Modulation of topoisomerase I activity. The method is based on the comparison of electrophoretic mobility of DNA depending on its conformation, *i.e.*, degree of relaxation under the action of topoisomerase I. Inhibition of the enzyme prevents the relaxation of the supercoiled plasmid DNA, therefore, the unwound DNA migrates faster in the gel compared to relaxed DNA. The influence of compound **10** on topoisomerase I activity was studied as described.[26] The reaction mixture (20 μL) containing DNA (0.25 μg) (pBR322 plasmid; Fermentas, Lithuania) and compound **10** was incubated for 30 min at 37 °C in the buffer: TRIS-HCl (35 mmol L^{-1}, pH 8.0), KCl (72 mmol L^{-1}), $MgCl_2$ (5 mmol L^{-1}), dithiothreitol (5 mmol L^{-1}), spermidine (2 mmol L^{-1}), bovine serum albumin (100 μg mL^{-1}); all the reagents were from Sigma, USA. The reaction was terminated by addition of sodium dodecyl sulfate (final concentration 1%), proteinase K was added for 40 min at 37 °C. The samples were analyzed by electrophoresis in 1% agarose gel (3 V cm^{-1}) for 4—5 h in the buffer cotaining TRIS-base (40 mmol L^{-1}), EDTA (1 mmol L^{-1}), glacial acetic acid (30 mmol L^{-1}). The gels were stained with ethidium bromide (0.5 μg mL^{-1}) and visualized under the UV light.

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