Cancer Chemother Pharmacol (1989) 24: 238–242

Cancer Chemotherapy and Pharmacology

The effect of structural modifications of 5-fluorouracil derivatives on their transport and biodegradation by isolated rat jejunum

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Summary. The continuous-perfusion technique was used in an isolated segment of everted rat jejunum to study transport and biotransformation processes in a series of cancerostatic derivatives of 5-fluorouracil. Metabolic alterations during penetration of the intestinal wall were assessed by high-performance liquid chromatography (HPLC). Octanol-buffer partition coefficients were measured, and the lipophilicity of the study compounds and fragmental constants for their sugar moieties were assessed. In the present series of 5-fluorouracil derivatives, there was no correlation between lipophilicity and metabolic cleavage to 5-fluorouracil, but a correlation was found between lipophilicity and the transport rate. Remarkable stability of the nucleoside bond and high biotransport were observed with 5′-chloro-5-fluorouridine, suggesting a different mode of activation for this derivative.

Introduction

5-Fluorouracil (I)[1] [13, 19], one of the most prominent nucleoside antimetabolites, is used in the treatment of advanced solid tumors, particularly breast, gastrointestinal and gynecological cancers [9, 18]. Until recently, the major growth-inhibitory effect of 5-fluorouracil and its derivatives 5-fluorouridine (II), 5-fluorodeoxyuridine (III), and 1-(tetrahydro-2-furanyl)-5-fluorouracil [Ftorafur (VI)] [15] has been associated with the formation of 5-fluorodeoxyuridine 5′-monophosphate (FdUMP), a potent inhibitor of thymidylate synthetase and thus of DNA synthesis [10, 12, 17].

However, a number of recent reports [16, 23, 24, 32] have demonstrated that 5-fluorouracil and its derivatives are converted to 5-fluorouridine 5′-triphosphate (FUTP), which is incorporated into RNA, thereby disrupting its synthesis and function. The exact mechanism of action of these antimetabolites remains unclear, despite their widespread clinical use. Nevertheless, the conversion of a 5-fluorouracil analog to its nucleotide derivative (FdUMP, FUTP) is considered to be a prerequisite for its antimetabolic and antineoplastic activity. Since 1978, when a series of deoxy derivatives of 5-fluorouridine was described [20, 21] 5′-deoxy-5-fluorouridine (V) has been extensively studied [1, 11, 29] due to its high therapeutic potential, lower toxicity, and broader biological activity compared with 5-fluorouracil (I).

Our recent in vitro screening results [3–5, 22] of 5-fluorouracil derivatives [6–8, 20, 21] have also demonstrated that they differ in their biological activity (Table 1). Briefly summarized, 5-fluorouracil (I) was found to be the strongest inhibitor of *Escherichia coli* growth, 5-fluorouridine (II) was the most potent and specific inhibitor of RNA synthesis, and 5-fluoro-2′-deoxyuridine (III) proved to be the only derivative that inhibited DNA synthesis, with a simultaneous, strong inhibition of RNA synthesis, and expressed virostatic activity. We therefore suggested [4, 5, 21] that the biological activity of 5-fluorouracil, 5-fluorouridine, and/or 5-fluoro-2′-deoxyuridine should at least partly proceed by different mechanisms. Thus, we were interested in determining whether 5-fluorouracil derivatives also differ in their transport and biotransformation processes, which should be important for the development of potent peroral 5-fluorouracil drugs. In addition, the partition coefficients of the study compounds were assessed and used for the calculation of fragmental constants [28] that indicate the lipophilicity of the sugar moiety of different molecules, which is an important characteristic for their biotransport.

Materials and methods

The compounds under investigation were synthesized as cited in Table 2 and illustrated in Fig. 1. They included pyrimidine nucleobases uracil (Ura) and 5-fluorouracil (FUra); pyrimidine ribonucleosides uridine (Urd) and 5-fluorouridine (FUrd); deoxyribonucleosides 2′-deoxyuridine (dUrd), 5-fluoro-2′-deoxyuridine (FdUrd), 5′-chloro-5′-deoxy-5-fluorouridine (5′-ClFUrd), and 5′-deoxy-5-fluorouridine (5′-dFUrd); acetyl derivatives 2′,3′,5′-tri-0-acetyl-5-fluorouridine (Ac_3FUrd), 2′,3′-di-0-acetyl-5′-chloro-5′-deoxy-5-fluorouridine (Ac_2-5′-ClFUrd), and 2′,3′-di-0-acetyl-5′-deoxy-5-fluorouridine (Ac_2-5′-dFUrd); and the drug Ftorafur [1-(tetrahydro-2-furanyl)-5-fluorouracil; Fto]. Uracil, uridine, and 2′-deoxyuridine are commercial products (Pharma-Waldhof GmbH, Mannheim).

The continuous-perfusion technique using an everted rat jejunal preparation as well as the design of permeation

[1] To emphasize the structural difference between 5-fluoro-2′-deoxyuridine and 5′-deoxy-5-fluorouridine, the former is described throughout this paper as the 5-fluoro derivative of the natural nucleoside 2′-deoxyuridine (FdUrd) and the latter, as an unnatural deoxy analog of 5-fluorouridine (5′-dFUrd)

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Table 1. Inhibitory activity of 5-fluorouracil derivatives

Compound	NSC number	DNA[a]	RNA[a]	E. coli[b]		HSV[c]
				Inhibition	Cleavage to FUra	
FUra	19,893	–[d]	55	56 (5)		–
FUrd	146,604	–	0.9	97 (3)	31%	–
FdUrd	27,640	200	58	76 (4)	36%	0.5
5′-dFUrd		–	615	97 (3)	not separated	–
5′-ClFUrd	600,775	–	–	67 (2)	0	–
Ftorafur	148,958	–	–	95 (1)	5%	–

[a] Concentration (in μmol l^{-1}) causing a 50% inhibition of nucleic acid synthesis in cultured lymphoid L1210 cells [5]
[b] Percentage of inhibition of *E. coli* growth [3]; in parentheses: -log of concentration (in mg/l)
[c] Concentration (in μmol l^{-1}) causing >90% inhibition of HSV [22]
[d] Inactive up to 1,000 μmol l^{-1}

Table 2. Transport and biotransformation of 5-fluorouracil derivatives during their penetration of everted rat jejunum

Number	Compound tested	$\bar{v}$ (μmol/30 min)[a] (Total transport)	Percentage of compounds found[b]:				FUra found on serosal side (μmol/30 min)	log P[c]
			Mucosal side		Serosal side			
1.	Uracil	5.68 ± 0.67	Uracil	100	Uracil	100	–	−1.11
2.	Uridine	1.80 ± 0.37	Uridine	100	Uridine	100	–	−1.98
3.	dUrd	3.89 ± 0.11 (3.23[d])	dUrd Ura	99 1	dUrd Ura	83 17	0.66[e]	−1.62
4.	FUra	4.86 ± 0.52	FUra	100	FUra	100	–	−0.93
5.	FUrd [6]	2.49 ± 0.05 (1.79[d])	FUrd FUra	94 6	FUrd FUra	72 28	0.69	−1.70
6.	5′-dFUrd [20]	3.46 ± 0.52 (2.46[d])	5′-dFUrd FUra	98 2	5′-dFUrd FUra	71 29	1.00	−1.16
7.	FdUrd [8]	4.02 ± 0.18 (3.01[d])	FdUrd FUra	95 5	FdUrd FUra	75 25	1.01	−1.21
8.	5′-ClFUrd [20]	3.42 ± 0.98	5′-ClFUrd FUra	99 1	5′-ClFUrd	100	0	−0.54
9.	Ftorafur [15]	6.28 ± 0.47 (4.52[d])	Ftorafur FUra	97 3	Ftorafur FUra	72 28	1.7	−0.37
10.	Ac_3FUrd [6]	1.79 ± 0.15 (0.97[d])	Ac_3FUrd FUrd FUra	68 29 3	 FUrd FUra	 54 46	 0.82	−0.25
11.	Ac_2-5′-dFUrd [7]	2.99 ± 0.11 (2.45[d])	Ac_2-5′-dFUrd 5′-dFUrd FUra	74 25 1	 5′-dFUrd FUra	 82 18	 0.53	−0.02
12.	Ac_2-5′-ClFUrd [7]	4.30 ± 0.83	Ac_2-5′-ClFUrd 5′-ClFUrd	28 72	 5′-ClFUrd	 100	 0	+0.38

[a] Transport rate: mean of at least three experiments ± SD
[b] Parent compounds and metabolites on mucosal and serosal sides after 30 min
[c] Logarithm of partition coefficient in octanol buffer
[d] Net nucleoside transport calculated from total transport
[e] Uracil

studies have previously been described [14, 26]. Briefly, a 10-cm length of fasted, everted rat jejunum was cannulated from both ends with a tygon cannula and put in an organ bath connected to the outflow from a peristaltic pump on one side and a UV-concentration monitor on the other; the outflowing solution from the concentration monitor was returned to a reservoir for recirculation. The organ bath contained the compound under study at a concentration of 8.2 m*M* in 30 ml Krebs-Ringer's bicarbonate (mucosal fluid). The circulating solution was 100 ml Krebs-Ringer's bicarbonate containing no drug at the beginning of the experiment (serosal fluid). The entire system was kept oxygenated (carbogen) at a constant temperature of 37° C.

The cumulative mucosal-to-serosal transport of compounds was recorded, usually for up to 30 min, after which 10-ml samples of both mucosal and serosal fluids were used for high-performance liquid chromatographic (HPLC) measurements [26] of the added compounds and their metabolites. The concentration measurements necessary for the evaluation of transport parameters and the determination of the partition coefficients were carried out

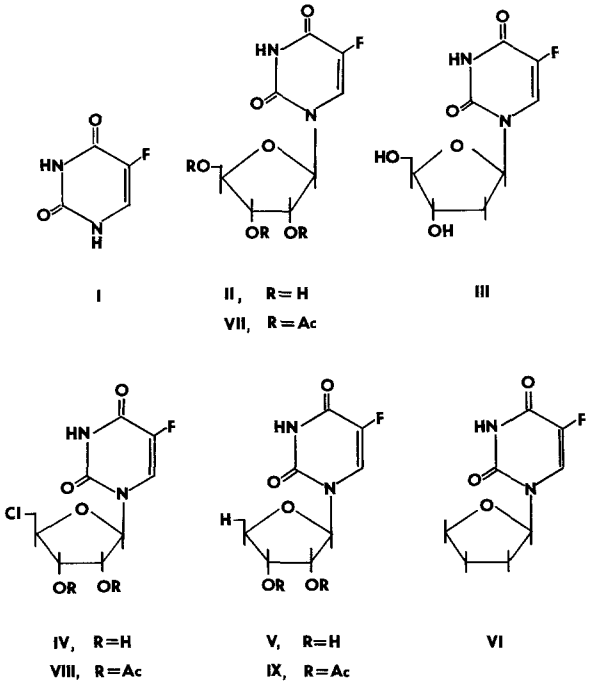

Fig. 1. Structures of 5-fluorouracil derivatives studied

Table 3. Values of the capacity (retention) factors (k) for 5-fluorouracil derivatives on octadecyl silica with aqueous/methanolic mobile phases

Compound	k					
	Methanol content, volume %					
	30	25	20	15	10	5
Uracil	–	0.12	0.17	0.24	0.36	0.61
FUra	–	0.15	0.20	0.28	0.42	0.67
Uridine	–	0.08	0.16	0.29	0.52	1.09
FUrd	–	0.17	0.27	0.45	0.80	1.59
2′-Deoxyuridine	–	0.16	0.29	0.48	0.91	1.90
FdUrd	–	0.25	0.40	0.66	1.30	2.58
5′-dFUrd	–	0.61	0.99	1.56	2.99	5.87
5′-ClFUrd	–	1.47	2.29	3.69	7.06	13.8
Ac[2]-5′-ClFUrd	6.30	10.4	20.1	–	–	–
Ac[2]-5′-dFUrd	3.60	6.56	13.1	–	–	–
Ac[3]FUrd	4.14	7.44	15.9	–	–	–
Ftorafur	0.86	1.32	2.06	3.33	5.89	12.2

by reversed-phase HPLC [26, 30]. The HPLC system consisted of an SP 8700 solvent delivery unit, an SP 8440 variable-wavelength UV detector operated at 254 nm, and an SP 4200 computing integrator (Spectra-Physics; San Jose, Calif, USA). Separations were carried out on a 250 × 4.2 mm (inside diameter) stainless steel column packed with 10-μm Separon SI C 18 octadecylsilica (Laboratorní přístroje; Prague, Czechoslovakia) with aqueous/methanolic mobile phases. Injections were done with a 10-μl Rheodyne 7125 sampling valve (Rheodyne; Cotati, Calif, USA).

Table 3 shows the dependence of the capacity (retention) factors (k) on the methanol content of the mobile phase for the study compounds. The chromatographic conditions were chosen according to the lipophilicity of the compound to obtain reasonable retention times and baseline resolution of the metabolites. The determination of the partition coefficient (P) was carried out as previously described [14, 28]; fragmental constants of the sugar moiety, which characterize the lipophilicity of the studied part of the molecule, were calculated according to Nys and Rekker [27].

Results and discussion

The above-described continuous-perfusion technique was used in an isolated segment of everted rat jejunum to study both transport and biotransformation processes in a series of cancerostatic derivatives of 5-fluorouracil. Table 2 summarizes the transport rates ($\bar{v}$), expressed as micromoles of the study compounds per 30 min; the percentages of the measured compounds occuring on both mucosal and serosal sides whether in intact form or as a degradation product; and the corresponding log of octanol/water partition coefficient. From Table 2 it is clear that the pyrimidine bases pass the intestinal wall with no metabolic alteration and exhibit higher transport rates than the corresponding nucleoside derivatives, in agreement with previous reports [14, 26]. This is to be expected, since the transport of such substances has been attributed to both active and passive mechanisms [31]. The slight decrease in the transport rate of 5-fluorouracil (I) (4.7 μmol/30 min) compared with that of uracil (5.7 μmol/30 min) might have been due to an alteration in the acidity of both molecules (pK_a = 9.45 and 8.15 for uracil and 5-fluorouracil, respectively), although the stability of the nucleobases was not influenced by the electronegative substituent.

Whereas in the uracil and cytosine series the nucleoside bond of the ribo-, arabino-, and cyclonucleoside derivatives was found to be completely stable during biotransport through rat intestine [14, 26] and far more stable than that of the corresponding 2′-deoxyribo nucleosides, quite opposite results were obtained with the nucleoside derivatives of 5-fluorouracil. The substitution by the fluoro atom dramatically reduced the stability of the ribonucleoside bond, even to below that of 2′-deoxyribonucleoside. The cleavage of the ribonucleoside bond was increased from 0% in the uracil series (uridine) to 28% in the 5-fluorouracil series [5-fluorouridine (II)]; the cleavage of the deoxyribonucleoside bond was only very slightly enhanced, from 17% in the uracil series (2′-deoxyuridine) to 25% in the 5-fluorouracil series [5-fluoro-2′-deoxyuridine (III)]. The level of cleavage of the nucleoside bond remained very similar (about 28%) in most of the nucleosides studied. The only cleavage product was released 5-fluorouracil, and its quantity was independent of the lipophilicity of the study compound. With the change in partition coefficient (Table 2) from −0.37 to −1.70, the amount of 5-fluorouracil released was changed from 25% to only 28%, being identical for 5-fluorouridine (II), 5′-deoxy-5-fluorouridine (V), and even Ftorafur (VI), whose structure differs from that of the other nucleosides.

In contrast to the metabolic relationships, the transport rate of the 5-fluorouracil nucleosides was not influenced by the electronegative substituent (fluoro atom), although it was directly related to the lipophilicity of the derivatives (Table 4, Fig. 2). The total transport value was thus confounded by the cleavage of the nucleoside bond and the subsequent, fast transport of the nucleobase released. The net transport of 5-fluorouridine (II) (1.8 μmol/30 min) was identical to that of uridine. The value for 5-fluoro-2′-deoxy-

Table 4. Lipophilicity (log P)-transport rate and/or lipophilicity-net transport rate correlation of 5-fluorouracil derivatives[a]

Group	Compound	Number[b]	A	B	*r*	Line in Fig. 2
A	Uracil	1	10.69	4.396	0.990	a
	Uridine	2	(10.61	4.483	0.998)[c]	
	dUrd	3				
B	FUra	4	7.736	3.082	1.000	b
	FUrd	5	(8.107	3.818	0.963)[c]	
	FdUrd	7				
C	group B		7.311	2.883	0.938	–
	5′-dFUrd	6	(7.367	3.469	0.852)[c]	
D	group A		8.382	3.222	0.899	c
	group B		(8.169	3.350	0.848)[c]	
E	Ac^3FUrd	10	2.883	3.905	0.992	d
	Ac^2-5′-dFUrd	11	(2.382	5.212	0.995)[c]	
	Ac^2-5′-ClFUrd	12				

[a] The relationship is expressed by the linear equation $y = A + B \cdot \log P$

[b] For compound numbers, refer to Table 2

[c] Values for net transport rate correlation

Values of points 8 and 9 were not used for calculation of the transport rate-lipophilicity correlation due to the different chemical structures of 5′-Cl-5-FUrd (8) and Ftorafur (9)

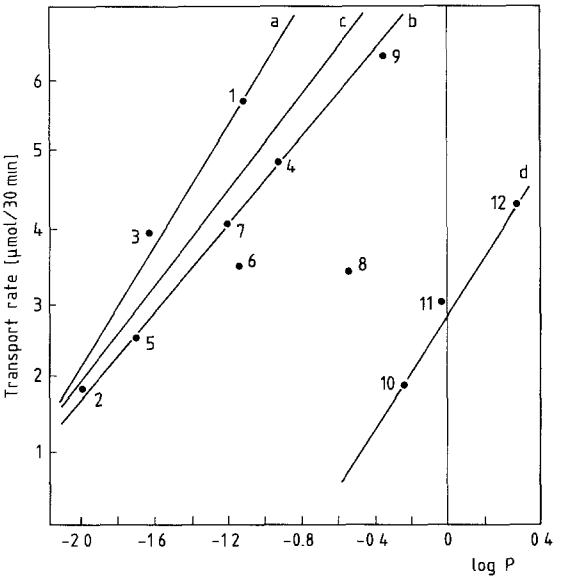

Fig. 2. Lipophilicity (log P)-transport rate correlation of 5-fluorouracil derivatives (compound numbers as indicated in Table 2)

uridine (III) (3.0 µmol/30 min) was similar to that for deoxyuridine (3.2 µmol/30 min), and that for 5-fluorouracil (I) (4.9 µmol/30 min) was similar to that for uracil (5.7 µmol/30 min).

Moreover, in the transport-lipophilicity correlation, an exact dependence of the transport rate of 5-fluorouracil nucleosides on their partition coefficient was observed, beginning with 5-fluorouridine (II), followed by 5′-deoxy-5-fluorouridine (V), 5-fluoro-2′-deoxyuridine (III), 5′-chloro-5-fluorouridine (IV), and Ftorafur (VI) (1.8, 2.5, 3.0, 3.4, and 4.5 µmol/30 min, respectively). An identical transport-lipophilicity relationship was also observed in the series of acetyl derivatives of 5-fluorouracil nucleosides, beginning with triacetyl-5-fluorouridine (VII), followed by diacetyl-5′-deoxy-5-fluorouridine (IX) and diacetyl-5′-chloro-5-fluorouridine (VIII) (1.0, 2.5, and 4.3 µmol/30 min, respectively). The clinically used compound Ftorafur (VI) was tested for comparative purposes. Its transport was the highest of all compounds tested, and its higher lipophilicity might also suggest a passive transport component [25].

In our previous work in the arabinosylcytosine series [14, 26], both the transport rate and biotransformation were found to be affected by the acetylation of arabinosylcytosine, with no cleavage of the nucleoside bond in any of the acetyl derivatives. The results in the 5-fluorouracil series differed from those obtained with the acetyl derivatives of arabinosylcytosine. The single types of 5-fluorouracil analogs differend in their behavior: whereas in 5-fluorouridine both processes were influenced in a negative way by its acetylation (VII) (the transport rate was decreased and the cleavage of the nucleoside bond was substantially increased), in diacetyl-5′-deoxy-5-fluorouridine (IX) the transport rate remained unchanged and the stability of the nucleoside bond was slightly increased, and in diacetyl-5′-chloro-5-fluorouridine (VIII) the transport rate was increased and the stability of the nucleoside bond was not attacked at all. In contrast to the arabinosylcytosine series, all acetyl derivatives of the 5-fluorouracil series were completely deacetylated during biotransport.

A fundamental exception to the other 5-fluorouracil derivatives can be seen in the effect of substitution of the sugar moiety in the 5′-position by the chloro atom. High stability was observed with 5′-chloro-5-fluorouridine as well as its diacetyl derivatives. The 5′-chloro compounds did not release any 5-fluorouracil under our experimental conditions, although 5′-chloro-5-fluorouridine (IV) has expressed a significant antitumor activity in experimental animals [1]. In addition, both 5′-chloro-5-fluorouridine and its diacetyl derivative (VIII) were transported through the intestinal wall at the highest rate in the study series, the transport rate of the diacetyl derivative (VIII) approaching that of Ftorafur (VI). However, we did not test the relationship between the rate of transport and the concentration of different uracil nucleosides to find out the type of transport kinetics involved.

Although our model for transport and metabolism study is not intended for the investigation of drug metabolism but rather for the determination of biodegradation during transport, nevertheless it shows that 5′-chloro-5-fluorouridine, being an active cancerostatic [2], did not produce 5-fluorouracil in our experiments. The significant stability of the nucleoside bond and the simultaneous, high transport rate of 5′-chloro-5-fluorouridine and its diacetyl derivative, together with their promising biological activity [1], encourage their further investigation, particularly for oral administration. We suppose that the 5′-chloro derivatives would pass through the cell or intestinal wall after oral application better than any other 5-fluorouracil analog, with no cleavage of the nucleoside bond, resulting in a higher concentration of the nucleoside inside the cell.

From the measured estimates of partition coefficient in Table 2, we also calculated the fragmental constant of the sugar moiety, as previously described [26]. This constant characterizes the lipophilicity of the sugar moiety of the molecule and may be used for the prediction of the partition coefficient of new antimetabolite preparations. The

Table 5. Estimates of lipophilicity of sugar moieties (fragmental constant)

Sugar moiety	Fragmental constant
1-β-D-Ribofuranosyl	−0.58
2-Deoxy-1-β-D-erythropentofuranosyl	−0.09
5-Deoxy-1-β-D-ribofuranosyl	−0.04
5-Chloro-5-deoxy-1-β-D-ribofuranosyl	+0.58
2,3,5-Tri-O-acetyl-1-β-D-ribofuranosyl	+0.87
2,3-Di-O-acetyl-5-deoxy-1-β-D-ribofuranosyl	+1.10
2,3-Di-O-acetyl-5-chloro-5-deoxy-1-β-D-ribofuranosyl	+1.50
Tetrahydro-2-furanyl	+0.75

calculated values are recorded in Table 5 and the results are in agreement with those of our previous studies [26].

In conclusion, the correlation between the lipophilicity of the investigated compounds and their biotransformation in the everted-rat-intestine model is not evident. However, there is a correlation between the lipophilicity of the compounds and their rate of transport from the mucosal to the serosal side of rat jejunum. The cleavage of the nucleoside bond and the release of 5-fluorouracil is common to all of the compounds except 5′-chloro-5-fluorouridine and its diacetyl derivative, being closely similar in most of the unsubstituted nucleosides as well as Ftorafur. Finally, the effect of the substitution of 5′-hydroxyl by chlorine is interesting: 5′-chloro-5-fluorouridine is transported across the intestinal wall at a higher rate, exhibiting no biodegradation to 5-fluorouracil. The fact that this compound possesses a high biological activity without biodegradation suggests a different mode of activation.

Acknowledgements. The authors wish to express their sincere thanks to Dr. M. Ryba for this efficient cooperation in carrying out HPLC measurements.

References

1. Ajmera S, Danenberg PV (1982) Synthesis and biological activity of 5′-substituted 5-fluoropyrimidine nucleosides. J Med Chem 25: 999
2. Amstrong R, Diasio R (1980) Metabolism and biological activity of 5′-deoxy-5-fluorouridine, a novel fluoropyrimidine. Cancer Res 40: 3333
3. Bártová M, Ryba M, Jedličková Z, Novotný L, Hřebabecký H, Beránek J (1983) Growth inhibition of *Escherichia coli* B by nucleoside analogs. Collect Czech Chem Commun 48: 2088
4. Beránek J (1986) A study on structure-activity relationships of nucleoside analogues. Drugs Exp Clin Res 12: 355
5. Beránek J, Acton EM (1984) Inhibition of nucleic acid synthesis in L1210 cells by nucleoside analogs. Collect Czech Chem Commun 49: 2551
6. Beránek J, Hřebabecký H (1976) Acetylation and cleavage of purine nucleosides. Synthesis of 6-azauridine, 5-fluorouridine and 5-methyluridine. Nucleic Acids Res 3: 1387
7. Beránek J, Hřebabecký H, Novotný L, Cech D, König J (1984) Fluorination of 5′-deoxyuridine and 5′-chloro-5′-deoxyuridine. Nucleic Acids Symp Ser 14: 241
8. Brokeš J, Hřebabecký H, Beránek J (1979) Preparation of 2′-deoxyribonucleosides and their 5-halogeno derivatives. Collect Czech Chem Commun 44: 439
9. Carter SK, Bakowski MT, Hellmann K (1977) Chemotherapy of cancer. John Wiley & Sons, New York, p 56
10. Cohen SS, Flaks JG, Barner HD, Loeb MR, Lichtenstein J (1958) The mode of action of 5-fluorouracil and its derivatives. Proc Natl Acad Sci USA 44: 1004
11. Cook AF, Holman MJ, Kramer MJ, Trown PW (1979) Synthesis and antitumor activity of a series of 5′-deoxy-5-fluoropyrimidine nucleosides. J Med Chem 22: 1330
12. Danenberg PV (1977) Thymidylate synthetase – a target enzyme in cancer chemotherapy. Biochem Biophys Acta 473: 73
13. Duschinsky R, Pleven E, Heidelberger C (1957) The synthesis of 5-fluoropyrimidines. J Am Chem Soc 79: 4559
14. Farghali H, Novotný L, Ryba M, Beránek J, Janků I (1984) Kinetics of transport and metabolism of 1-β-D-arabinofuranosylcytosine and structural analogs by everted perfused rat jejunum. Biochem Pharmacol 33: 655
15. Giller SA, Zhuk RA, Lidak MYu (1967) N_1-(α-Furanidilnye) proizvodnye prirodnych osnovanii i ikh antimetabolitov. Dokl Akad Nauk SSSR 176: 332
16. Glazer RI, Hartman KD (1980) The effect of 5-fluorouracil on the synthesis and methylation of low molecular weight nuclear RNA in L1210 cells. Mol Pharmacol 17: 245
17. Heidelberger C (1972) The nucleotides of fluorinated pyrimidines and their biological activities. In: Carbon-fluorine compounds. A Ciba Foundation symposium. Elsevier/Excerpta Medica/North-Holland, Amsterdam, p 125
18. Heidelberger C (1975) Fluorinated pyrimidines and their nucleosides. In: Sartorelli A, Johns D (eds) Antineoplastic and immunosuppressive agents, part II. Springer-Verlag, Berlin, p 193
19. Heidelberger C, Chaudhuri N, Danenberg P, Mooren D, Griesbach L, Duschinsky R, Schnizer R, Pleven E (1957) Fluorinated pyrimidines, a new class of tumor-inhibitory compounds. Nature 179: 663
20. Hřebabecký H, Beránek J (1978) 5′-Halogeno-2′,3′-cyclic sulphite isomers in the preparation of 5′-halogeno nucleosides. Synthesis of 5′-deoxyuridine and 5′-deoxy-5-fluorouridine. Nucleic Acids Res 5: 1029
21. Hřebabecký H, Beránek J (1978) 5′-Halogeno-2′,3′-sulphites in the synthesis of 2′,5′-dideoxy-5-fluorouridine and related analogues. Collect Czech Chem Commun 43: 3268
22. Reference deleted
23. Kufe DW, Egan EM (1981) Enhancement of 5-fluorouracil incorporation into human lymphoblast ribonucleic acid. Biochem Pharmacol 30: 129
24. Kufe DW, Major PP (1981) 5-Fluorouracil incorporation into human breast carcinoma RNA correlates with cytotoxicity. J Biol Chem 256: 9802
25. Moon AY, Poland DC, Sherage HA (1965) Thermodynamic data from fluorescence spectra: I. The system phenol-acetate. J Phys Chem 69: 2960
26. Novotný L, Farghali H, Ryba M, Janků I, Beránek J (1984) Structure-intestinal transport and structure-metabolism correlations of some potential cancerostatic pyrimidine nucleosides in isolated rat jejunum. Cancer Chemother Pharmacol 13: 195
27. Nys GG, Rekker RF (1974) The concept of hydrophobic fragmental constants (f values): II. Extension of its applicability to the calculation of lipophilicities of aromatic and heteroaromatic structures. Eur J Med Chem 9: 361
28. Rekker RF (1977) The hydrophobic fragmental constant. Elsevier, Amsterdam
29. Rosowsky A, Kim S-H, Trites D, Wick M (1982) Synthesis and in vivo antitumor activity of potential 5-fluorouracil prodrugs. J Med Chem 25: 1034
30. Ryba M, Beránek J (1981) Liquid chromatographic separation of purines, pyrimidines and their nucleosides on silica gel columns. J Chromatogr 211: 337
31. Shanker LS, Tocco DJ (1960) Active transport of some pyrimidines across the rat intestinal epithelium. J Pharmacol Exp Ther 128: 115
32. Wilkinson DW, Tlsty TD, Hanas RJ (1975) The inhibition of ribosomal RNA synthesis and maturation in Novikoff hepatome cells by 5-fluorouridine. Cancer Res 35: 3014

Received 7 July 1987/Accepted 17 January 1989