Fluorinated Pyrimidines. Part 4. Synthesis, Properties and Stereochemical Conversion of the *cis* and *trans* Isomers of 6-Alkoxy-5-fluoro-5,6-dihydrouracils

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The reaction of 6-acetoxy-5-fluoro-5,6-dihydrouracil with ROH (R = Me, Et, Pr, Pr^i, Bu, Bu^t) under acidic conditions has been investigated using ^{18}F as a tracer. The quantitative replacement of the OAc-group proceeded predominantly with *cisoid* (*gauche*) stereochemistry, but upon prolonged heating the amount of the *trans* compound increased. This isomerization did not originate from epimerization at C^5. The *trans* compounds—apart from being more stable towards substitution and elimination than the corresponding *cis* compounds—gave invariably substitution products with inversion of configuration, while the *cis* compounds mainly retained their configuration. Within the concept of the unifying ion-pair mechanism it is proposed that the *trans* compound reacts *via* an intimate protonated intermediate and the *cis* compound *via* a solvent-separated protonated intermediate. As a result, it was not the *gauche* attraction between the fluorine atom and the incoming nucleophile, but the poor π-donor ability of the C–F bond that was the determining factor in the *cisoid* stereochemistry of 5-fluoro-5,6-dihydrouracil adducts.

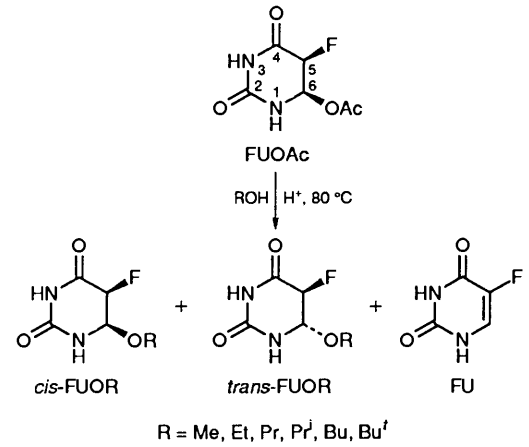
5-Fluorouracil (FU) has been used for several decades for the chemotherapeutic treatment of a variety of human neoplasms, although its efficacy is only about 30%.[1,2] The catabolic enzyme dihydropyrimidine dehydrogenase (DPD) is responsible for the *in vivo* inactivation of up to 90% of an FU dose before it can reach the tumour.[3] Several attempts to increase the therapeutic index of FU have been made by means of N^1-substituted FU precursors or prodrugs that are not directly a target for DPD.[4]

In 1986 we reported on the formation of both *cis* and *trans* isomers of 6-acetoxy-5-fluoro-5,6-dihydro-(FUOAc) and 5-fluoro-6-hydroxy-5,6-dihydro-uracil (FUOH) upon reaction of gaseous acetyl hypofluorite (AcOF) with uracil in AcOH and H_2O respectively.[5] In theory these adducts are not directly a target for DPD owing to the absence of the 5,6-double bond, while the substituent at the 6-position might be versatile enough to be substituted *in vivo* by the SH group of thymidylate synthase (TS) leading to TS-inhibition. In contrast with the N^1-substituted acetoxy adducts,[6–9] the acetoxy group in FUOAc was found to be too labile. By virtue of the hydrogen at N^1, acetic acid elimination took place upon removal of the solvent, leading to an acylimine.[5] On the other hand, FUOH did not lead to acylimine formation, but was found to exert excessive toxic side-effects to have *in vivo* significance. Therefore we decided to investigate whether the 6-alkoxy-5-fluoro-5,6-dihydro-(FUOR) derivatives would be interesting versatile compounds within our DPD-circumventing approach, with the additional advantage that their lipophilicity can be changed by varying the chain length.

In this paper the synthesis, stereochemistry and the acid-catalysed conversion of the *cis* and the *trans* FUOR adducts are described. Differences in substitution rate and product ratio will be correlated to the difference in the geometric position of the fluorine atom, enabling us to propose a refinement of the nature of the preferred *cisoid* stereochemistry. Because of the limited solubility of the uracil derivatives and in order to simplify the detection of the several intermediaries, most of the experiments were carried out using ^{18}F as a tracer.

Results and Discussion

The reaction of gaseous AcOF with uracil in H_2O gives the FUOH adduct as the main product.[5,6] However, when ROH was used as the solvent hardly any FUOR adduct formation was observed, the main reaction of AcOF being the abstraction of a hydrogen from the ROH solvent. Another approach, starting from FUOAc in AcOH, removal of the solvent and adding the desired ROH to the thus formed acylimine gave inconvenient yields, the main product being FU, produced by proton removal by ROH. Obviously under these conditions the acylimine is more stable towards addition than expected, examples of which are known in the literature.[10]



Scheme 1

Quantitative replacement of the OAc group by the OR group was accomplished by the removal of most of the AcOH, followed by the addition of an excess of ROH and a catalytic amount of H_2SO_4, and heating at 80 °C for 10 min (Scheme 1). In this way, besides the *cis* and *trans* alkoxy compounds formed, the reaction mixture contained only the amounts of FUOH and FU which were formed directly during reaction of AcOF with uracil in AcOH (3–10%).[5,6] Under the reaction conditions used no formation of $ROSO_2OH$ or $ROSO_2OR$ was observed, while the HSO_4^- anion, after alkoxonium ion formation, cannot act as a base as long as the stronger base ROH is present in excess. Owing to the presence of the small amount of AcOH during the substitution reaction, a certain amount of the corresponding ROAc ester was formed. This ester is hydrolysed and removed

Table 1 Physical data of the 5-fluoro-5,6-dihydrouracil adducts

Compound	t_R[d]/min 98:2	80:20	δ_{H^6}	δ_{H^5}	$J_{5F,5H}$	$J_{5H,6H}$	$J_{5F,6H}$	δ Alkyl groups
cis FUOMe	5.6	3.8	4.92	5.34	45.8	4.0	2.0	3.44 (OCH_3)
trans FUOMe	10.2	4.9	4.87	4.89	46.8	3.2	7.1	3.44 (OCH_3)
cis FUOEt	10.0	4.7	5.02	5.33	46.0	4.0	2.1	3.78, 3.59 (OCH_{ab});[a] 1.20 (CH_3)
trans FUOEt[c]	22.5	7.1	4.97	4.89	46.5	3.6	7.4	3.80, 3.61 (OCH_{ab});[a] 1.20 (CH_3)
cis FUOPr	29.7	8.6	5.00	5.33	46.0	4.0	2.1	3.67, 3.51 (OCH_{ab});[a] 1.59 (CH_2); 0.90 (CH_3)
trans FUOPr[c]	73.5	16.2	4.96	4.90	46.7	3.4	7.2	3.69, 3.52 (OCH_{ab});[a] 1.59 (CH_2); 0.90 (CH_3)
cis FUOPr[i]	22.6	7.4	5.09	5.30	46.2	4.0	2.3	3.92 (OCH); 1.18 (CH_{3a});[b] 1.17 (CH_{3b})[b]
trans FUOPr[i]	55.2	12.6	5.06	4.85	46.8	3.7	7.2	3.97 (OCH); 1.18 (CH_3)
cis FUOBu	—	19.7	5.00	5.32	46.0	4.0	2.1	3.72, 3.54 (OCH_{ab});[a] 1.56, 1.35 (CH_2); 0.90 (CH_3)
trans FUOBu[c]	—	42.4	4.96	4.89	46.8	3.3	7.1	3.74, 3.55 (OCH_{ab});[a] 1.56, 1.35 (CH_2); 0.90 (CH_3)
cis FUOBu[t]	—	14.3	5.24	5.26	47.1	4.1	2.1	1.25 (CH_3)
trans FUOBu[t]	—	20.2	5.22	4.79	47.3	4.4	7.0	1.28 (CH_3)
cis FUSBu	—	52.5	5.13	5.53	46.8	5.6	2.7	2.72 (SCH_2); 1.56, 1.43 (CH_2); 0.91 (CH_3)
trans FUSBu	—	85.4	4.97	4.99	47.5	5.0	9.5	2.73 (SCH_2); 1.56, 1.43 (CH_2); 0.91 (CH_3)
cis FUSBu[t]	—	25.4	5.10	5.43	47.4	5.5	6.3	1.39 (CH_3)
trans FUSBu[t]	—	40.6	5.05	4.89	47.7	5.9	14.1	1.41 (CH_3)

[a] $J_{Ha,Hb}$ 9.3–9.4 Hz. [b] Through the presence of the F-atom, in *cis* FUOPr[i] the methyl groups show a detectable difference in chemical shift. [c] For these *trans* compounds the two protons and the F-atom form strongly coupled spin systems; the coupling constants and chemical shifts were determined *via* computer simulation and iteration. [d] t_R-value FUOH in 98:2 and 80:20 3.7 min and 3.3 min, respectively; t_R-value FU in 98:2 and 80:20 6.0 min and 3.8 min, respectively; t_R-value uracil in 98:2 and 80:20 5.4 min and 3.6 min, respectively.

Table 2 Yields (%) of the fluoro compounds upon incubation of *cis* and *trans* FUOEt in acidified EtOH ($EtOH-H^+$ 100:1) at room temperature

Substrate	Δt/day	Yield (%) *cis* FUOEt	*trans* FUOEt	FU
cis FUOEt	1	94	5	0.5
	2	90	7	1
	5	83	13	3
	13	75	19	4
trans FUOEt	1	8	90	—
	2	15	84	—
	5	28	70	0.5
	13	53	45	1

Fig. 1 Conformation of the *cis* and *trans* FUOR adducts

upon rotary evaporation during work-up, while the H_2O formed during this side-esterification process did not lead to the formation of significant amounts of FUOH as a side product ($\leqslant 3\%$). Under the given reaction conditions, a 1:1 molar mixture of primary alcohols like EtOH–PrOH gave a 1:1 FUOEt–FUOPr product ratio.

From the $J_{5H,6H}$ and $J_{5F,6H}$ coupling constants (Table 1) it could be calculated (CAGPLUS), that with respect to their conformation in both the *cis* and the *trans* compounds—irrespective of the nature of the alkyl group—the anomeric effect (substituent at C^6 axial to N^1) was the predominant factor (Fig. 1). This strong conformational preference was also observed for the corresponding FUOAc and FUOH adducts.[5–7]

As representatives of the sulfide series, we took BuSH and Bu^{t}SH. In this case the OAc group was also replaced with the same ease. According to expectation the anomeric effect becomes smaller on replacing an oxygen atom with a sulfur atom (Table 1). From the $J_{5H,6H}$ and $J_{5F,6H}$ coupling constants of the *trans* compounds it appears that the pyrimidine ring is almost non-puckered, while the position of the C^6 substituent is between an axial and equatorial.

After heating for 10 min at 80 °C the *cis* isomer was the main product (*c*:*t* ratio 10–20:1), which implies that the replacement of the OAc-group proceeds through an S_N1-like reaction with an energetically favourable *cisoid* (*gauche*) stereochemistry.[11] Upon prolonged heating, the FUSBu and FUSBu^t adducts were slowly converted into FU. However, under the same conditions the alkoxy compounds underwent an interesting reaction. The amount of *trans* compound was found to increase with a concomitant slight increase in FU formation (Fig. 2). The rate of isomerization was only slightly dependent on the H^+ concentration (studied in the range 2000:1–12:1), but for ROH/H^+ ratios below 25, elimination—giving FU—became a significant side reaction. The *cis*–*trans* isomerization—starting from the pure alkoxy isomers—also took place at room temperature, albeit at a lower rate (Table 2). However, an overwhelming kinetic and thermodynamic preference for the *cis* product remained. The latter observation seems to be in accordance with well-known geometric arguments. In the *cis* compound there is stabilization by the anomeric effect, by the *gauche* effect (between the F atom and the OR group), and by the synperiplanar relationship of the F atom with the carbonyl function at C^4 (α-halo ketone effect). In the *trans* compound only the anomeric effect remains, since there now exists no *gauche* interaction between the F atom and OR while the F atom is in an antiperiplanar relationship with the carbonyl functions at C^4.

In order to determine whether the observed interesting isomerization was due to an enolization process at C^5 (Fig. 3), an exchange at C^6 or both, we studied the conversion of equimolar amounts of each pure FUOR isomer in an excess of R'OH, to which no acid, AcOH or H_2SO_4 was added. For these cross experiments, the majority were carried out with *cis* and *trans* FUOEt and *cis* and *trans* FUOPr. These four compounds and the elimination product FU have distinctly different t_R values upon HPLC analysis, while the corresponding solvents (EtOH, PrOH) exhibited no serious difference in protonation rate, as could be verified by the mixed solvent experiments. After *cis* FUOEt had been heated in PrOH at 80 °C, about 40% FU was formed, after 4 h about 70%; for the *trans* FUOEt

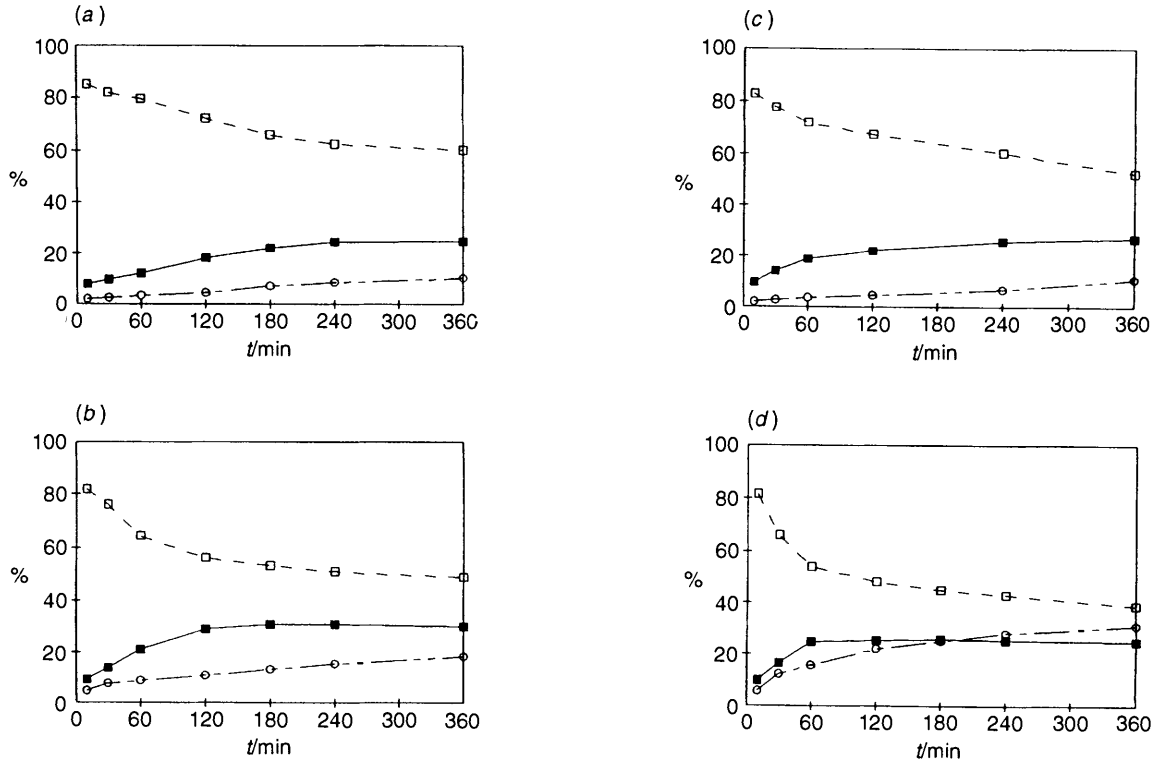


Fig. 2 Isomerization profile of several of the FUOR adducts upon prolonged heating, at 80 °C (ROH–H^+ 100:1): R = (*a*) Me; (*b*) Pr; (*c*) Et; (*d*) Pr^i. □, *cis*; ■, *trans*; ○, FU.

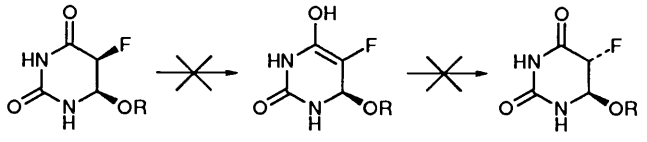


Fig. 3 Hypothetical pathway for the isomerization of the FUOR adducts

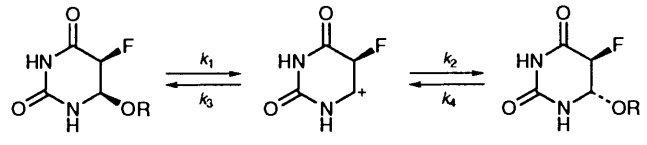


Fig. 4 Possible pathway for the formation of FUOR isomers

compound in PrOH this was 10% and 20%, respectively. The same results were obtained for the *cis* and *trans* FUOPr isomers in EtOH at 80 °C. In all four cases no isomerization or substitution was observed. The difference in elimination rate is in accordance with the geometry of the compounds and with the known preference of weak bases for *anti*-elimination: the *cis* adduct possesses the perfect *anti*-arrangement (the proton and the leaving group is axially disposed); for the *trans* compound only *syn* elimination is possible, this being a less favourable elimination route. In the presence of a stronger base such as OH^- (aqueous alkaline pH = 8 solution) both *cis* and *trans* compounds gave FU quantitatively with no difference in elimination rate.

In the presence of AcOH (ROH–AcOH 10:1 v/v; 80 °C, 4 h), for *cis* and *trans* FUOEt in PrOH or *cis* and *trans* FUOPr in EtOH, no substitution and hardly any elimination or isomerization was observed; minor amounts of FU (1–2%) were formed, while only the *trans* compound underwent isomerization. However, this was less than 1%. Heating the separate isomers in pure AcOH at 80 °C for 4 h did not lead to detectable isomerization; only minor amounts of FU (1–2%) were formed. This latter result is consistent with the fact that for our alkoxy adducts no detectable exchange of H^5 for D was observed in CD_3CO_2D even after several days. This strongly indicates that an enolization as depicted in Fig. 3 does *not* occur, although such an enolization has been proposed for the $5F,6SO_3^-$ adduct of deoxyuridine[12] and has been observed to occur for 5F,6OH cytosine adducts[6] (rapid H^5 for D exchange in CD_3CO_2D during NMR measurements).

The same kind of cross experiment, but in the presence of a small amount of H_2SO_4 (ROH–H^+ 100:1) instead of AcOH, gave the results as shown in Table 3 (at 60 °C and at room temperature). It can be concluded that, in the presence of this stronger acid *cis–trans* isomerization also does not occur *via* epimerization at C^5 (otherwise, *e.g.*, *cis* FUOEt would have given *trans* FUOEt). Instead, the isomerization is the result of a new attack at C^6, whereby the *cis* compound gives mainly a *cis* product, while the *trans* compound is predominantly converted into a *cis* product, but at a distinctly lower rate than its corresponding *cis* compound. So in the non-cross experiments of Table 2, the *cis* compound is only apparently more stable.

As expected, the addition of 400 mm^3 of AcOH—and its concomitant formation of a small amount of water—during the cross experiment with H_2SO_4 did not affect the substitution yield or the *cis*:*trans* FUOR product ratio. This is due to the fact that, when formed, FUOH is subject to the same fast substitution reactions as the corresponding FUOR adducts, as was checked by starting from pure FUOH.

At first glance it seems that these results and consequently those from Fig. 2 and Tables 2 and 3 are explained by the two equilibria given in Fig. 4. However, in general, S_N1 substitution gives a nearly completely racemized mixture and is accompanied by much elimination. So when postulating the open carbocation equilibria, in which one needs a remarkably powerful product-directing *gauche* interaction between the F atom and the incoming nucleophile ($k_3 \gg k_2$), one has to explain why $k_4 < k_2$ (more of the *trans* being generated than consumed) and why such a relatively low incidence of elimination (leading to FU) occurs.

Table 3 Yields (%) of the fluoro compounds[a] upon incubation of *cis* and *trans* FUOEt in acidified PrOH (PrOH–H^+ 100:1) at 60 °C and at room temperature (r.t.)

Substrate	Δt	Yield (%) *cis* FUOEt	*trans* FUOEt	*cis* FUOPr^n	*trans* FUOPr^n
cis FUOEt (60 °C)	10 min	70	—	24	2.4
	20 min	46	—	44	6.2
	30 min	28	—	59	8.4
trans FUOEt (60 °C)	10 min	—	91	4	0.1
	20 min	—	85	10	0.6
	30 min	—	80	14	1.2
cis FUOEt (r.t.)	1 day	51	—	42	3.1
	2 days	26	—	62	7.0
	3 days	13	—	74	9.1
	10 days	1	—	74	20.0
trans FUOEt (r.t.)	1 day	—	85	10	0.5
	2 days	—	77	18	0.8
	3 days	—	70	23	2.0
	10 days	—	41	45	9.0

[a] FU formation in the order of 1–4%.

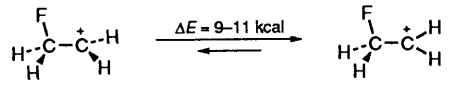


Fig. 5 Most unfavourable (C–F bond parallel) and most stable (C–F bond orthogonal) conformer of the β-fluoroethyl carbocation

Fig. 6 Visually represented difference in π-donor abilities in the *cis* and *trans* FUOR adduct

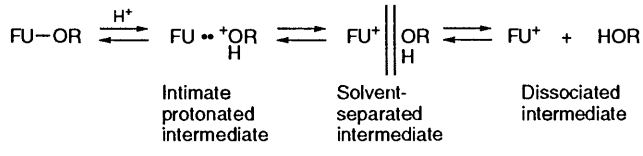
The *gauche* effect has been estimated to be 0.2–1.2 kcal mol^{-1},[13–14] the α-halo ketone effect 0.2–0.4 kcal mol^{-1},[15] and the anomeric effect 1.5 kcal mol^{-1}.[16] These values correspond to the fact that in the *trans* compound the anomeric effect surpasses the *gauche* effect, so it is inconceivable that a stereoelectronic effect like the *gauche* effect would be such a powerful product director.

S_N2 and S_N1 reactions are the extremes of a mechanistic continuum. Winstein,[17,18] Shiner[19] and Sneen,[20] have shown the existence of at least two distinct ion-pair intermediates before coming to an open carbocation, an intimate ion pair and a solvent-separated ion pair.

$$RX \rightleftharpoons R^+X^- \rightleftharpoons R^+ \,||\, X^- \rightleftharpoons R^+ + X^-$$

In this mechanistic continuum attack on the intimate ion pair corresponds with the 'old' S_N2 reaction, resulting in inversion of configuration, while the solvent-separated intermediate gives a high retention of configuration through immediate front-side solvent collapse. Of course the open carbocation gives a racemized mixture of products. Thermodynamic proof for the existence of the two intermediates has been reported.[21]

Alcohols are relatively weak nucleophiles (compared with RO^-) and weak bases, while in our system C^6 is a secondary carbon-centre. Not only have secondary carbon-centres minimal reactivity both for S_N2 and S_N1 reactions when compared with a corresponding primary or tertiary carbon-centre, but also elimination occurs *via* mechanisms that are intermediate between the extreme mechanistic E_1 and E_2 types (the variable E_2 transition-state theory).[22] Besides this, theoretical calculations for the β-fluoroethyl carbocation (Fig. 5) in the gas phase have shown that the energy difference between the C–F bond being parallel with the empty π-orbital and orthogonal to it is 9–11 kcal mol^{-1}.[23,24] Although we are in solution now and not in the gas phase, the principle that the C–H bond is a better π-donor than the C–F bond cannot be neglected when compared with anomeric, *gauche*, α-halo ketone and temperature effects. In the β-fluoroethyl carbocation free rotation around the C–C bond is possible; however, in our *cis*/*trans* compounds the situation is more or less fixed. In the *trans* compound we have a C–F bond parallel and in the *cis* compound a C–F bond orthogonal to the developing carbocation-like intermediate (Figs. 1 and 6). As a result, instead of the occurrence of the two equilibria in Fig. 4 we propose the mechanism outlined in Scheme 2.



Scheme 2

We postulate that after protonation, the parallel C–F bond in the *trans* compound simply blocks the development of any positive charge at the neighbouring C atom, *i.e.*, locks the compound in the intimate protonated intermediate state. Consequently only a displacement process is possible with inversion of configuration. In the case of the *cis* compound the C–H bond—being a better π-donor than the C–F bond—allows the formation of a solvent-separated intermediate, whereafter immediate front-side solvent collapse results in the remarkable high retention of configuration observed. Within this hypothesis, the results given in Table 3 for the *trans* compound are now interpreted as an exclusive attack on the locked intimate protonated intermediate, wherafter the small amount of *trans* compound formed during reaction is the result from attack on the just-formed *cis* compound. For the *cis* compound the substituent at C^6 is relatively rapidly (re)substituted.

This leaves us with the question of where, in the case of the attack on the *cis* compound, the *trans* compound and the elimination product come from. The *trans* compound might

Table 4 Yields (%) of the fluoro compounds[a] upon incubation of FUOAc in acidified ROH (ROH–H^+ 100:1) at 80 °C for 1 h

Substrate	Yield (%)			
	MeOH	EtOH	PrOH	BuOH
cis FUOR	80	72	65	59
trans FUOR	12	19	22	24
FU	3	4	9	12

[a] The amount of FU formed directly during the reaction of AcOF with uracil has been subtracted; the amount of FUOH has been omitted.

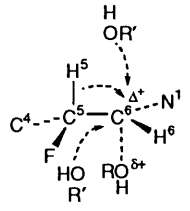


Fig. 7 *cis/trans* formation and elimination during conversion of the *cis* FUOR adduct

come from attack on the *cis* compound during its intimate protonated intermediate state or the *trans* compound and FU might be formed from a certain amount of open carbocation. This question can be answered by comparing the results with variation of the group R. Table 4 shows us the results under similar reaction conditions and molar ratios in the series R = Me, Et, Pr, Bu. From MeOH to BuOH the ionizing power of the solution decreases and it is inconceivable that somehow in the less polar BuOH solution more of an open carbocation would be formed than in the more polar MeOH. Pursuing our reasoning, in the less polar solvent BuOH, the formation of a solvent-separated protonated intermediate is slightly more difficult (there now exists a slightly higher energy barrier between the intimate and solvent-separated intermediate corresponding to the necessity of creating a void between the ion and the leaving group that must grow to molecular size before a solvent can occupy it). From the concomitant increased amount of FU, it can implicitly be concluded that this product is not an E_1 but an E_2-like elimination product.

The situation is depicted in Fig. 7. We propose that the E_2-like elimination takes place just before the solvent-separated intermediate is formed (followed by a front-side solvent collapse yielding the *cis* compound), while the S_N2-like substitution occurs just before the elimination. At this point the departing group has not left the carbocation completely and backside attack from the BuOH molecule can occur. The more positive charge that develops at C^6, the more steric hindrance there will be from the π-donating C^5–H^5 bond approaching C^6 (at this point we have a situation analogous to hydronium-ion formation). The approaching backside R'OH can now only be of assistance by taking up the H^5 proton resulting in the E_2 elimination product FU. For the most polar solvent in this series, MeOH, the formation of the solvent-separated intermediate is easiest, thus giving the S_N2-like substitution or E_2 elimination less chance. Proof of steric hindrance during these processes, if present, could, unfortunately, not be demonstrated experimentally. Using a mixture of alcohols, *e.g.*, a 1:1 mixture of Pr^{i}OH and PrOH, more of the FUOPr adduct was found to be formed (ratio 1:2.5). However this does not necessarily imply the influence of steric hindrance owing to the fact that in such a mixture Pr^{i}OH is the more easily protonated of the two alcohols, resulting in a relatively higher incidence of free PrOH which is consequently incorporated.

Conclusions

The *cis* and *trans* FUOR adducts appeared to be interesting tools for a study of nucleophilic substitution reactions at β-fluoro carbon-centres with the fluorine atom in two different geometric positions. Our results indicate that the poor π-donor ability of the C–F bond is the predominant factor determining product formation. Consequently, in pyrimidines the observed preferred *cisoid* (*gauche*) stereochemistry for fluorine during 5-halo-5,6-dihydro adduct formation (for I, Br and Cl all adducts are found to exist in the *trans* form) is due to the fact that the π-donor ability follows the sequence C–I > C–Br > C–Cl > C–H > C–F, while *gauche* attraction—if any—between the F-atom and the incoming nucleophile is of minor importance. The latter stereoelectronic effect might only lead to conformational preference after product formation when it is not surpassed by, *e.g.*, an anomeric effect or the polarity of the solvent.

Finally, these results imply that—within our proposed SET mechanism for the reaction of AcOF with uracil derivatives[6]—either immediate recombination of the radical at C^6 with the acetoxyl radical occurs (leading to a *cis* adduct) or the acetoxyl radical carries out a second SET—but only when the C–F bond is in an orthogonal position—after which immediate front-side solvent collapse still results in a *cis* adduct.[6] Consequently, a high amount of *trans* compound is not an indication of the formation of an open carbocation and subsequent random recombination of the two ions,[25] but of the occurrence of a (rapidly) inverting radical and subsequent recombination with the electron-delocalized acetoxyl radical, an expectation that is borne out in the reaction of AcOF with cyclohexene, where no resonance-stabilized carbocation can be formed, but where a *cis/trans* ratio of 1.5 was obtained for 1-acetoxy-2-fluoro-cyclohexane.[26]

Experimental

For most of the experiments ^{18}F was used as a tracer, which permitted a simple determination of the yields of the different fluorinated products using HPLC techniques. Gaseous Ac$O^{18}F$ was produced by passing $^{18}F_2$ through a column of KOAc–HOAc. For reversed-phase HPLC analysis all products were dissolved in H_2O (for R = Me, Et, Pr, Pr^i) or a 40:60 mixture of MeCN–H_2O (for R = Bu, Bu^t, and for OR = SBu, SBu^t). The analysis of the products were performed on a 20 cm CPTM-spher, C18 column (Chrompack), eluent 98:2 or 80:20 v/v 0.01 mol dm^{-3} $NH_4H_2PO_4$–MeOH, flow rate 1 cm^3 min^{-1}. Peaks were detected using a radioactivity monitor and UV detector (210 and 254 nm); fractions of 500 mm^3 were collected and counted for radioactivity. Separation of the F-containing products was performed by reversed-phase column chromatography (Lobar Lichroprep RP-8, 40–63 μm, Merck), eluent: various H_2O–MeOH mixtures (*vide infra*). Solvents were removed by evaporation at 50 °C or by freeze-drying. ^1H NMR spectra were measured on a Bruker WM 250 spectrometer using CD_3CO_2D as the solvent. Chemical shifts are reported in δ relative to $\delta(CD_2HCO_2D) = 2.04$.

General Procedure for the Isolation of the cis *and* trans *Fluorinated Adducts.*—Gaseous AcOF (390 μmol) was bubbled through a solution of 45 mg (402 μmol) uracil in 20 cm^3 of AcOH. Subsequently the solvent was evaporated until about 0.5 cm^3 of AcOH was left, 15 cm^3 of ROH were added (R = Me, Et, Pr, Pr^i), followed by 55–100 mm^3 conc. H_2SO_4 (molar ratio ROH–H^+ 100:1). The mixture was heated at 80 °C until a *cis*:*trans* ratio of about two was obtained (for Me and Et, 3 h; for Pr, 2 h; for Pr^i, 1 h), after which it was cooled in ice, 10 cm^3 of H_2O were added and the solution was brought to pH = 2.5–3 with the aid of a 1.8 mol dm^{-3} K_2CO_3 solution. After

evaporation almost to dryness, the reaction products were dissolved in 2 cm^3 of the column eluent by use of ultrasonic frequencies and applied to two Lichroprep columns in series. The following were eluents: H_2O for Me, H_2O–MeOH 95:5 for Et and Pr^i, H_2O–MeOH 90:10 for Pr. After separation, the *cis* and *trans* fractions were collected, evaporated to a volume of about 2 cm^3 and freeze-dried; the overall efficiency was 70–80%.

Because of solubility problems, for the compounds with RO = BuO, Bu'O, BuS and Bu'S only 195 μmol AcOF, 200 μmol uracil, and 37–45 mm^3 conc. H_2SO_4 were used. After 1 h at 80 °C and cooling, a solution of MeCN–H_2O 40:60 was added until a homogenous solvent mixture was obtained, which was subsequently brought to pH = 2.5–3. After evaporation almost to dryness, the residue was dissolved in MeCN–H_2O 40:60 using ultrasonic frequencies. The following were column eluents: H_2O–MeOH 80:20 for Bu'O, H_2O–MeOH 70:30 for BuO and Bu'S and H_2O–MeOH 50:50 for BuS. Overall efficiency 60% for BuO, 40% for Bu'O (owing to the fact that during isomerization about 20% of FUOH was formed) and 40% for BuS and Bu'S (owing to the fact that during isomerization about 40% of FU was formed while only a *cis*:*trans* ratio of about 10 was obtained).

Substitution Reactions and Related Experiments.—For a comparison of the isomerization rate and concomitant elimination under similar reaction conditions and molar ratios, 0.5 cm^3 of 160 μmol cm^{-3} stock solution of FUOAc in AcOH was added to 150 mmol ROH (R = Me, 6.1 cm^3; Et, 8.7 cm^3; Pr and Pr^i, 11.2 cm^3; Bu and Bu', 13.7 cm^3) or RSH (BuS and Bu'S: 16.7 cm^3) followed by 40 mm^3 (0.75 mmol) conc. H_2SO_4 (RXH–H^+ 100:1) and heated at 80 °C. Samples were taken at 10 min, 30 min, 1 h, 4 h and 6 h. Each sample was cooled, and after addition of 1 cm^3 of H_2O (for R = Me, Et, Pr and Pr^i) or 3 cm^3 MeCN–H_2O 40:60 (for R = Bu, Bu', and RO = BuS, Bu'S), brought to pH = 2.5–3 with the aid of a 1.8 mol dm^{-3} K_2CO_3 solution. The solvent was removed *in vacuo* almost until dryness, and subsequently the reaction mixture was dissolved in water or MeCN–H_2O, respectively, by use of ultrasonic frequencies and analysed by HPLC. These conversions were also measured using ^{18}F ($t_{\frac{1}{2}}$ = 110 min) as a tracer.

Mixed solvent experiments were carried out in the same way, but instead of 150 mmol of ROH a mixture of 75 mmol ROH + 75 mmol R'OH was used; sets investigated were EtOH–PrOH; EtOH–BuOH; PrOH–Pr^iOH and PrOH–BuOH.

For a check of the influence of the ROH–H^+ ratio, EtOH was used. Conditions were: 0.17 cm^3 of the stock solution and 2.9 cm^3 EtOH, while the chosen EtOH–H^+ molar ratios were achieved by addition of the appropriate amounts of a H_2SO_4–MeCN 1:9 solution. Ratios studied were EtOH–H^+ 2000:1; 1000:1; 500:1; 150:1; 100:1; 75:1; 50:1; 25:1 and 12:1. After 1 h at 80 °C, samples were worked up as described above.

For the substitution reactions on FUOH, 4.5 mg (30 μmol), was dissolved in 0.5 cm^3 HOAc, followed by the addition of either 8.7 cm^3 EtOH, 11.2 cm^3 PrOH or 13.7 cm^3 BuOH and 40 mm^3 conc. H_2SO_4 and heating at 80 °C. Samples were taken at 10 min, 30 min and 1 h and worked up as described above.

Cross- and Related Experiments.—For a study of the nature of the isomerization process, three basic sets of experiments were carried out. (*i*) Either 10 μmol of *cis* FUOEt or *trans* FUOEt were dissolved in 5 cm^3 PrOH or 10 μmol of *cis* FUOPr or *trans* FUOPr were dissolved in 5 cm^3 EtOH. After addition of H_2SO_4 (molar ratio ROH–H 100:1), the reaction mixture was heated at 60 °C or left to stand at room temperature. At appropriate time intervals samples were taken, worked up as described above and analysed by HPLC. (*ii*) The same set-up, but instead of H_2SO_4, 0.5 cm^3 HOAc was added. During heating at 80 °C, samples were taken at 1 h and 4 h; the solvent was removed by evaporation and water was added, followed by HPLC analysis. (*iii*) The same set-up, but no acid was added at all. Sampling and work-up as in (*ii*).

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