Thio Analogs of Pyrimidine Bases: Syntheses and EIMS Study of New *ortho*-(*meta*- and *para*-)Bromobenzyl S-Mono and S-N-1-Disubstituted 5-Morpholinomethyl(5-piperidinomethyl)-2-thiouracils.

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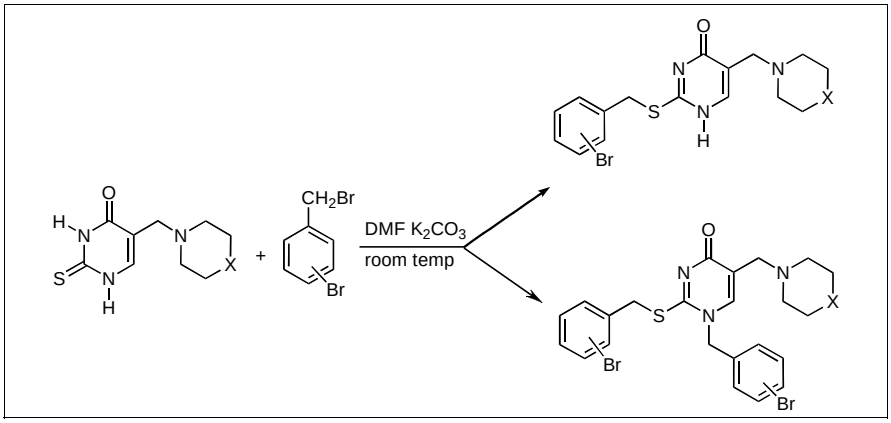

Eleven new *ortho*-(*meta*- and *para*-)bromobenzyl S-mono and S-N-1-disubstituted derivatives of 5-morpholinomethyl-2-thiouracil (MMTU) and 5-piperidinomethyl-2-thiouracil (PMTU) have been prepared and their EI induced mass spectral fragmentation has been investigated. It has been shown that the data derived from EIMS spectra can be used to differentiate the isomers.

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INTRODUCTION.

Thio derivatives of pyrimidine bases have remarkably contributed to biological and medicinal chemistry. Chemical modifications of these compounds have led to a large number of mono- and di-S and N substituted analogs showing therapeutic properties, especially antiviral, antithyroid and antitumor activities [1-7].

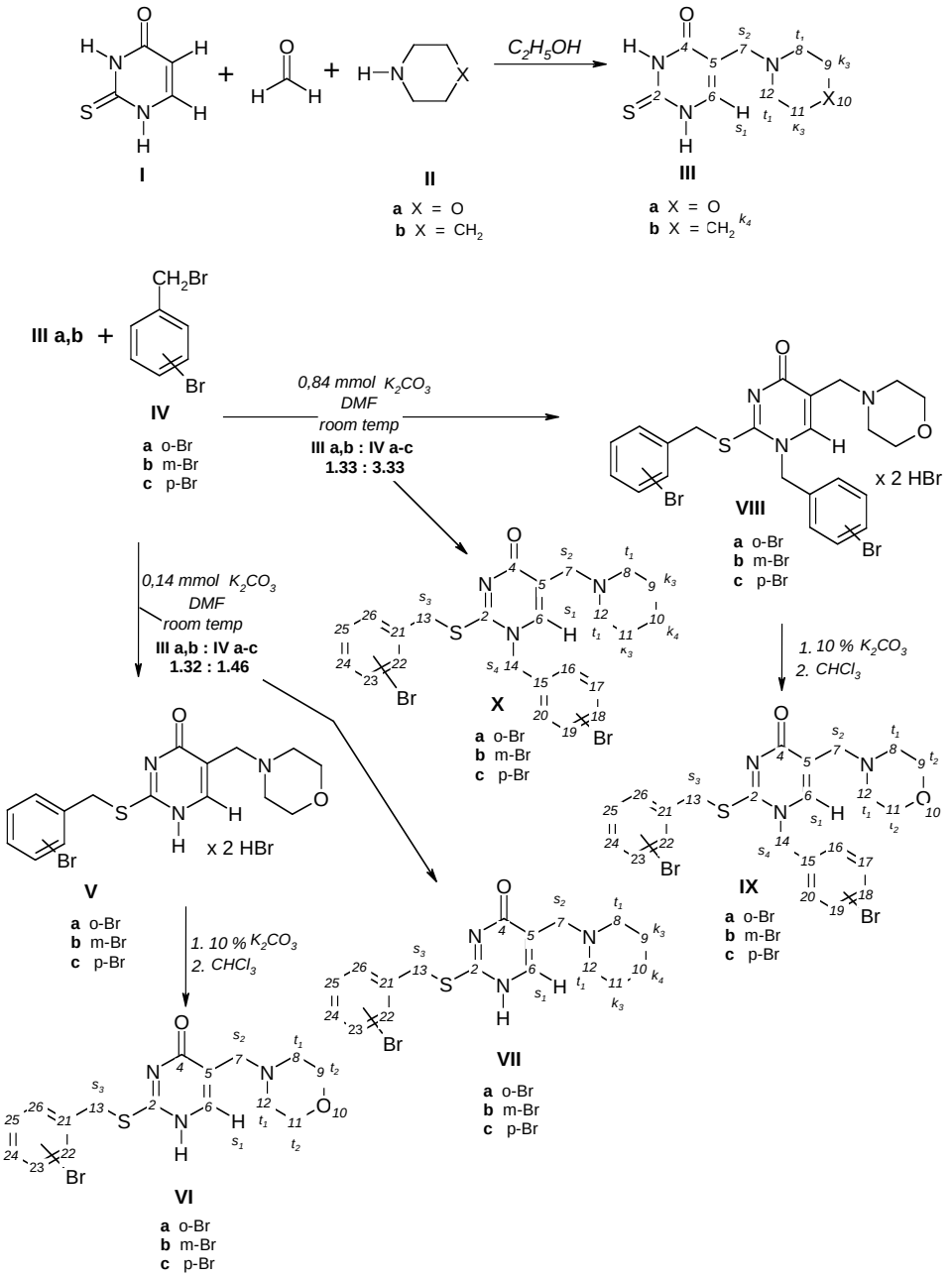
A new antimetabolite of thymine, 5-morpholinomethyl-2-thiouracil (MMTU) has been synthesized *via* the Mannich reaction of 2-thiouracil, formaldehyde and morpholine by Kamalakannan *et al* [8]. However, to the best of our knowledge, no work has been published on the chemoselective and regioselective synthesis as well as on physicochemical properties of the mono- and dibenzyl substituted derivatives of MMTU. This fact has stimulated us to investigate the chemoselectivity and regioselectivity of the reaction of benzylation of MMTU, as well as unknown in literature 5-piperidinomethyl-2-thouracils (PMTU). PMTU has been synthesized by us *via* the Mannich reaction of 2-thiouracil, formaldehyde and piperidine.

This paper deals with the synthesis and physico-chemical properties of new, unknown in literature, dihydrobromides of 2-*o*-(*m*- and *p*-)bromobenzylthio-5-morpholinomethyl- uracils **Va-Vc**, 2-*o*-(*m*- and *p*-)bromo-benzyl-5-morpholinomethyluracils **VIa-VIc**, 2-*o*-(*m*- and *p*-)bromobenzylthio-5-piperidinomethyluracils **VIIa-VIIc**, dihydrobromides of 2-*o*-(*m*- and *p*-)bromobenzylthio-1-*o*-(*m*- and *p*-)bromobenzyl-5-morpholinomethyluracils **VIIIa-VIIIc**, 2-*o*-(*m*- and *p*-)bromobenzylthio-1-*o*-(*m*- and *p*-)bromobenzyl-5-morpholino- methyluracils **IXa-IXc** and 2-*o*-(*m*- and *p*-)bromobenzylthio-1-*o*-(*m*- and *p*-)bromo-benzyl-5-piperidinomethyluracils **Xa-Xc**. The structures of all compounds obtained were determined by examining their UV/VIS, IR, ^{1}H NMR and ^{13}C NMR spectra as well as on the basis of elemental analyses (Tables 1-3). The UV/VIS, IR, ^{1}H NMR, ^{13}C NMR and EIMS spectra of 5-piperidinomethyl-2-thiouracil **IIIb** have also been investi-gated because this compound is unknown in literature. The EIMS spectra of **VIa-VIc**, **VIIa-VIIc**, **IXa-IXc** and **Xa-Xc** have been analysed to check the possibility of differentiation of *o*-(*m*- and *p*-)bromo substituted in benzyl group isomers on the basis of differences in the values of μ_1, μ_2 and μ_3 *i.e.* the ratio of the intensity of selected fragmentation peaks to that of the parent ion peak, and to compare the data with those obtained previously in our laboratory [9-11].

RESULTS AND DISCUSSION.

New 5-piperidinomethyl-2-thiouracil **IIIb** was synthesized *via* the Mannich reaction of 2-thiouracil, formaldehyde and piperidine (Scheme 1). The structure of **IIIb** was determined by examining its UV/VIS, IR, ^{1}H NMR, ^{13}C NMR and EIMS spectra as well as on the basis of elemental analysis (Tables 1, 2, 3, 4). A series of new S-mono-*o*-(*m*- and *p*-) bromobenzyl substituted derivatives of **IIIa** and **IIIb**, as well as S-N-1-di-*o*-(*m*- and *p*-) bromobenzyl substituted derivatives of **IIIa** and **IIIb** was synthesized in the reactions of these compounds with

Scheme 1



the corresponding *o*-(*m*- and *p*-)bromobenzyl bromides **IVa**-**IVc**. Treatment of 1.32 mmole of **IIIa** with 1.46 mmole of the corresponding **IVa-IVc** at room temperature in the solution of DMF in the presence of 0.83 mmole of K_2CO_3 afforded chemoselectively new dihydrobromides of 2-*o*-(*m*- and *p*-)bromobenzylthio-5-morpholinomethyluracils **Va-Vc** (Scheme 1, Table 1). Treatment of 1.32 mmole of **IIIb** with 1.46 mmole of the corresponding **IVa-IVc** at room temperature in the solution of DMF in the presence of 0.83 mmole of K_2CO_3 afforded chemoselectively 2-*o*-(*m*- and *p*-)-bromobenzylthio-5-piperidino- methyluracils **VIIa-VIIc** (Scheme 1, Table 1).

A series of 1,2-di-*o*-(*m*- and *p*-)bromobenzyl substituted dihydrobromides of 5-morpholinomethyl-2-thouracils (**VIIIa-VIIIc)** was synthesized regioselectively in the reaction of 1.33 mmole wth 3.33 mmoles of the corresponding **IVa-IVc** in DMF solution in the presence of 1.33 mmole of K_2CO_3 at room temperature. **Va-Vc** and **VIIIa-VIIIc** dissolved 10 % K_2CO_3 water solution were extracted with $CHCl_3$ to give **VIa-VIc** and **IXa-IXc** respectively (Scheme 1, Table 1).

Table 1

Chemical and Physical Data of Compounds **III b, V a-c, VI a-c, VII a-c**,**VIII a-c**, **IX a-c**, **X a-c**.

Comp.	Formula (mol. weight)	M.p. [ºC]	Yield [%]	Elemental analysis calculated (found)			
				C	H	N	S
III b	$C_{10}H_{15}N_3OS$ 225.31	200	91	53.33 (53.42)	6.66 (6.72)	18.66 (18.71)	14.22 (14.32)
V a	$C_{16}H_{18}N_3O_2SBr$ x 2 HBr 558.14	70-2	59	34.40 (34.16)	3.58 (3.54)	7.52 (7.50)	5.73 (5.72)
V b	$C_{16}H_{18}N_3O_2SBr$ x 2 HBr 558.14	67-9	58	(34.75)	(3.56)	(7.55)	(5.59)
V c	$C_{16}H_{18}N_3O_2SBr$ x 2 HBr 558.14	70-3	60	(34.36)	(3.60)	(7.50)	(5.73)
VI a	$C_{16}H_{18}N_3O_2SBr$ x ½ H_2O 405.31	55-7	71	47.40 (47.29)	4.44 (4.48)	10.37 (10.32)	7.90 (7.60)
VI b	$C_{16}H_{18}N_3O_2SBr$ x ½ H_2O 405.31	57-9	72	(47.30)	(4.40)	(10.30)	(7.82)
VI c	$C_{16}H_{18}N_3O_2SBr$ x ½ H_2O 405.31	65-7	69	(47.03)	(4.18)	(10.28)	(6.89)
VII a	$C_{17}H_{20}N_3OSBr$ 394.33	169-170	73	51.77 (51.42)	5.07 (5.15)	10.65 (10.63)	8.12 (8.05)
VII b	$C_{17}H_{20}N_3OSBr$ 394.33	173-5	76	(51.81)	(5.06)	(10.39)	(7.85)
VII c	$C_{17}H_{20}N_3OSBr$ 394.33	185	78	(51.65)	(5.10)	(10.46)	(7.97)
VIII a	$C_{23}H_{23}N_3O_2SBr_2$ x 2 HBr 727.17	139-141	57	37.96 (37.90)	3.16 (3.21)	5.77 (5.93)	4.40 (4.60)
VIII b	$C_{23}H_{23}N_3O_2SBr_2$ x 2 HBr 727.17	55-7	55	(37.65)	(3.18)	(5.62)	(4.20)
VIII c	$C_{23}H_{23}N_3O_2SBr_2$ x 2 HBr 727.17	85-7	59	(37.63)	(3.12)	(5.99)	(4.30)
IX a	$C_{23}H_{23}N_3O_2SBr_2$ x 2 H_2O 601.36	85-7	68	45.92 (45.82)	3.82 (3.51)	6.98 (6.62)	5.32 (5.52)
IX b	$C_{23}H_{23}N_3O_2SBr_2$ x 2 H_2O 601.36	30-2	64	(45.62)	(3.78)	(6.69)	(5.29)
IX c	$C_{23}H_{23}N_3O_2SBr_2$ x 2 H_2O 601.36	65-7	67	(45.67)	(3.87)	(6.68)	(5.04)
X a	$C_{24}H_{25}N_3OSBr_2$ 563.35	130	75	51.15 (51.18)	4.44 (4.49)	7.46 (7.38)	5.68 (5.60)
X b	$C_{24}H_{25}N_3OSBr_2$ 563.35	90-2	82	(51.30)	(4.50)	(7.58)	(5.70)
X c	$C_{24}H_{25}N_3OSBr_2$ 563.35	160	83	(51.30)	(4.39)	(7.42)	(5.78)

Table 2

UV/VIS, IR and ^{1}H NMR Data of Compounds **III b, VI a-c, VII a-c**, **IX a-c**, **X a-c**.

Comp.	UV/VIS	IR	^{1}H NMR						
	λ max (lg ε)	νC4=O νC5=C6	s_1	s_2	s_3	s_4	t_1 t_2	k_3 k_4	Ph-H M
III b	213.0 (4.08)	1640	7.24	3.12	-	-	2.50	1.47	-
	273.0 (4.14)	1569					-	1.37	
VI a	210.0 (4.29)	1673	7.60	3.22	4.46	-	2.37	-	7.20-7.95
	285.0 (3.86)	1534					3.59	-	
VI b	211.0 (4.30)	1652	7.63	3.20	4.24	-	2.35	-	7.25-7.80
	287.5 (3.90)	1558					3.56	-	
VI c	209.5 (4.21)	1662	7.61	3.14	4.24	-	2.33	-	7.36-7.48
	287.0 (3.94)	1528					3.54	-	
VII a	220.5 (4.34) 248.5 (4.01) 286.0 (4.01)	1675 1473	7.78	3.54	4.41	-	2.50 -	1.58 1.44	7.18 - 7.64
VII b	220.5 (4.33)	1675	7.72	3.34	4.27	-	2.50	1.53	7.22–7.62
	248.5 (3.98)	1473					-	1.40	
	285.5 (3.99)								
VII c	223.5 (4.36)	1675	7.76	3.50	4.30	-	2.50	1.58	7.18-7.37
	249.0 (4.03)	1484					-	1.41	
	286.0 (4.00)								
IX a	218.0 (4.37)	1644	8.32	3.34	4.43	5.15	2.37	-	7.19 –7.81
	282.0 (4.05)	1568					3.55	-	
IX b	215.0 (4.43)	1646	8.32	3.31	4.33	5.20	2.36	-	7.15 –7.95
	282.0 (4.10)	1569					3.57	-	
IX c	218.0 (4.43)	1642	8.35	3.30	4.35	5.10	2.33	-	7.35-7.48
	282.5 (4.05)	1486					3.55	-	
X a	215.0 (4.31)	1670	8.58	3.35	4.53	5.54	2.50	1.62	7.21 –7.72
	281.0 (3.94)	1589					-	1.41	
X b	216.5 (4.34)	1669	8.51	3.35	4.53	5.42	2.50	1.60	7.24 –7.81
	279.5 (3.87)	1582					-	1.41	
X c	226.5 (5.37)	1669	8.50	3.39	4.50	5.43	2.50	1.62	7.36-7.48
	282.5 (4.89)	1583					-	1.44	

A series of new 1,2-di-*o*-(*m*- and *p*-)bromobenzyl substituted derivatives of 5-piperidinomethyl-2-thouracils (**Xa-Xc**) was synthesized regioselectively in the reaction of 1.33 mmole of PMTU (**IIIb**) with 3.33 mmoles of corresponding **IVa-IVc** in DMF solution in the presence of 1.33 mmole of K_2CO_3 at room temperature (Scheme 1, Table 1). Noteworthy is the fact that the presence of the piperidinomethyl substituent at the C-5 atom of the uracil ring of the **IIIb** molecule changes the reactivity of this compound in the reaction of bromobenzylation relative to that of **IIIa**. In particular, the reactions of S-bromobenzylation and S-N-1-dibromobenzylation of **IIIa** afforded dihydrobromides of mono bromobenzyl substituted derivatives of MMTU (**Va-Vc**), and dihydrobromides of dibromobenzyl substituted derivatives of MMTU (**VIIIa-VIIIc**). In the reactions of benzylation of **IIIb** in the same conditions only S-bromobenzylated and S-N-1-dibromobenzylated derivatives of PMTU (**VIIa-VIIc, Xa-Xc**) were obtained. So, it is clear that these reactions are largely governed by the structure of MMTU (or PMTU) and by the nature of the benzylation agent.

Assignments of the ^{1}H NMR and ^{13}C NMR resonances of **IIIb**, **VIa-VIc**, **VIIa-VIIc**, **IXa-IXc** and **Xa-Xc** were deduced on the bases of signal multiplicities, and by the concerted application of two-dimensional NMR technique HETCOR.

Table 3
^{13}C NMR Shifts of **III b, VI a-c, VII a-c, IX a-c** and **X a-c** δ (ppm).

Comp.	Carbon 2	4	5	6	7	13	14	8, 12	9, 11	10
IIIb	174.92	161.39	113.84	139.31	53.04	-	-	53.57	25.50	23.76
VIa	163.11	160.08	117.60	161.75	52.96	30.30	-	53.50	66.05	-
VIb	163.00	160.02	117.60	151.70	52.95	31.00	-	53.52	66.03	-
VIc	163.18	160.00	117.61	151.87	52.95	32.60	-	53.54	66.04	-
VIIa	163.20	160.00	117.60	153.03	52.60	34.29	-	54.46	24.10	22.73
VIIb	163.10	160.04	117.66	154.43	53.92	32.76	-	55.05	24.74	23.44
VIIc	163.68	160.12	117.60	152.95	52.66	32.77	-	53.47	24.18	22.79
IXa	169.39	160.49	117.26	151.96	55.98	34.16	66.10	53.17	61.45	-
IXb	166.35	161.22	117.13	151.83	55.97	33.90	66.10	53.15	61.45	-
IXc	166.30	161.20	117.77	151.80	55.53	33.90	66.16	53.07	61.40	-
Xa	166.42	161.19	117.90	153.20	56.20	35.13	68.19	52.66	22.63	20.90
Xb	166.82	162.63	117.80	151.58	56.00	33.92	68.07	53.46	22.60	19.36
Xc	166.78	162.70	117.84	154.70	55.56	33.64	67.99	52.11	22.61	20.96

Comp.	Carbon 21 (15)	22 (16)	23 (17)	24 (18)	25 (19)	26 (20)
VIa	135.80	123.80	132.70	127.97	129.57	131.55
VIb	140.80	131.60	121.30	129.74	130.37	127.88
VIc	139.45	138.82	130.82	120.29	130.82	138.82
VIIa	137.01	124.04	132.60	127.86	129.33	132.41
VIIb	142.20	131.43	121.37	129.54	130.43	127.93
VIIc	137.87	131.13	131.22	120.06	131.22	131.13
IXa	135.22	128.25	132.57	127.75	129.69	131.56
	(135.11)	(127.96)	(132.34)	(127.65)	(129.27)	(131.25)
IXb	140.80	131.60	121.36	127.88	129.76	131.25
	(139.59)	(131.40)	(121.27)	(127.79)	(129.69)	(131.40)
IXc	139.40	138.80	130.80	119.20	130.86	138.80
	(138.96)	(138.78)	(130.52)	(119.11)	(130.52)	(138.78)
Xa	136.14	123.81	132.64	127.88	130.35	131.00
	(135.52)	(122.60)	(132.55)	(127.80)	(129.41)	(130.41)
Xb	140.66	131.51	122.10	129.59	130.73	127.92
	(138.12)	(131.09)	(121.97)	(128.31)	(130.19)	(127.32)
Xc	137.45	135.62	131.48	120.85	131.48	135.62
	(137.26)	(135.26)	(131.36)	(120.31)	(131.36)	(135.26)

On the basis of the low- and high-resolution electron impact as well as B/E linked scan mass spectra (Table 4) the principal mass spectral fragmentation routes of compound **IIIb** are proposed. The characteristic features of the mass spectral fragmentation of the molecular ion **IIIb** are the cleavages of the three bonds of the piperidine ring. By these cleavages the fragment ions [M-$CH_3^{\cdot}$] **f**, [M-$^{\cdot}CH_2CH_3$] **g**, [M-$^{\cdot}C_3H_7$] **h,** [M-C_4H_8] **j** are derived. The cleavages of the two Csp^3-Csp^3 bonds of this ring lead to the ejection of C_4H_8 neutral molecules to give odd-

electron fragment ions **j**. By the simple inductive cleavage of Csp^3-N bond in the piperidinomethyl substituent the complementary even-electron fragment ions $[M-^{\cdot}C_5H_{10}N]^+$ **l** and $[M-^{\cdot}C_5H_5N_2SO]^+$ **h** (100 % rel. int.) are derived. The cleavage of the Csp^3-N bonds of the piperidinomethyl substituent also involves H rearrangement and yields odd-electron fragment ion **k**. Simple inductive cleavage of Csp^2-Csp^3 bond between the uracil ring and the piperidinomethyl substituent *i.e.* the cleavage of the β bond to annular nitrogen atom of the piperidine ring has also been observed (ion **m**). The common features of the mass spectral fragmentations of the molecular ions of **VIa-VIc** and **VIIa-VIIc** (Table 4) are the cleavages of Csp^3-S bonds in the bromobenzylthio substituent (ions **i**) and of Csp^3-N bond in the 5-piperidinomethyl (or 5-morpholinomethyl) substituent (ions **e**). In the fragmentation of the molecular ions of **VIa-VIc** and **VIIa-VIIc**, simple radical-site initiated α-cleavages of the Csp^3-Csp^2 bonds of the 5-piperidinomethyl (or 5-morpholinomethyl) substituents have also been observed (ion **m**). The inductive

Table 4

Elemental composition and Relative Intensities of the Ion Peaks in the Spectra of **IIIb**,**VIa - VIc** and **VIIa - VIIc** according to high resolution Data.

Ion	m/z	Elemental composition	% Relative Intensity						
			IIIb	VIa	VIb	VIc	VIIa	VIIb	VIIc
$M^{+\cdot}$**a**	225	$C_{10}H_{15}N_3OS$	17	-	-	-	-	-	-
	395/397	$C_{16}H_{18}N_3O_2SBr$	-	12/11	7/6	3/2	-	-	-
	393/395	$C_{17}H_{20}N_3OSBr$	-	-	-	-	7/6	9/8	12/11
b	337/339	$C_{13}H_{12}N_3OSBr$	-	8/9	4/3	3/2	-	-	-
c	308/310	$C_{12}H_9N_2OSBr$	-	19/20	17/16	10/9	9/8	12/11	21/20
d	229	$C_{12}H_9N_2OS$	-	34	4	2	27	3	5
e	226	$C_9H_{12}N_3O_2S$	-	39	28	13	-	-	-
	224	$C_{10}H_{14}N_3OS$	-	-	-	-	11	7	13
f	210	$C_9H_{12}N_3OS$	1	-	-	-	-	-	-
g	196	$C_8H_{10}N_3OS$	3	-	-	-	-	-	-
h	182	$C_7H_8N_3OS$	14	-	-	-	-	-	-
l	169/171	C_7H_6Br	-	100/99	100/99	26/25	27/26	16/15	49/48
j	169	$C_6H_7N_3OS$	5	-	-	-	-	-	-
k	142	$C_6H_6N_2OS$	5	-	-	-	-	-	-
l	141	$C_5H_5N_2OS$	6	21	19	8	8	6	7
m	100	$C_5H_{10}NO$	-	38	82	100	-	-	-
	98	$C_6H_{12}N$	3	-	-	-	10	12	17
n	86	C_4H_8NO	-	67	90	49	-	-	-
	84	$C_5H_{10}N$	100	-	-	-	100	100	100
o	82	C_4H_4NO	22	-	-	-	-	-	-
p	70	C_4H_8N	5	-	-	-	-	-	-
r	57	C_3H_5O	-	50	95	39	-	-	-
	55	C_3H_5N	19	-	-	-	13	13	12

Table 5

Elemental composition and Relative Intensities of the Ion Peaks in the Spectra of **IXa - IXc** and **Xa - Xc** according to high resolution Data.

Ion	m/z	Elemental compositon	% Relative intensity					
			IXa	IXb	IXc	Xa	Xb	Xc
$M^{+\cdot}$**a**	565	$C_{23}H_{23}N_3O_2SBr$	15	25	7	-	-	-
	563	$C_{24}H_{25}N_3OSBr$	-	-	-	6	3	2
b	394/396	$C_{16}H_{17}N_3O_2SBr$	18/17	33/32	15/14	-	-	-
	392/394	$C_{17}H_{19}N_3OSBr$	-	-	-	5/4	5/4	2/1
c	309/311	$C_{12}H_{10}N_2OSBr$	17/16	24/23	15/14	6/5	3/2	2/1
d	293/295	$C_{11}H_6N_2OSBr$	15/14	22/21	30/29	2/1	2/1	2/1
e	250/252	$C_{11}H_9NOBr$	13/12	11/10	5/4	13/12	25/24	14/13
f	229	$C_{12}H_9N_2OS$	22	4	4	14	2	1
g	226	$C_9H_{12}N_3O_2S$	14	14	10	-	-	-
	224	$C_{10}H_{14}N_3OS$	-	-	-	6	4	2
h	169/271	C_7H_6Br	100/99	100/99	100/99	100/99	100/99	48/47
i	100	$C_5H_{10}NO$	39	56	19	-	-	-
	98	$C_6H_{12}N$	-	-	-	52	64	43
j	86	C_4H_8NO	29	88	45	-	-	-
	84	$C_5H_{10}N$	-	-	-	60	94	100
k	57	C_3H_5O	23	35	6	-	-	-
	55	C_3H_5N	-	-	-	22	16	10

cleavages of Csp^3-N bonds of 5-piperidinomethyl and 5-morpholinomethyl substituents of **VIa-VIc** and **VIIa-VIIb** give the even-electron fragment ions **n**. The cleavage of the same bonds with a simultaneous transfer of γ-hydrogen to the nitrogen atom of the piperidine (or morpholine) ring leads to the odd-electron fragment ions **c**.

As shown in Table 5, the principal fragmentation pathways of the molecular ions of **IXa-IXc** and **Xa-Xc** are similar to those of **VIa-VIc** as well as **VIIa-VIIc**. Inductive cleavages of the Csp^3-S bonds of the bromobenzylthio substituent proceed with the retention of charge, or the migration of charge and elimination of bromobenzyl (or thiouracils radicals) to form the complementary even-electron fragment ions **b** and **h**, respectively.

The simultaneous cleavages of Csp^3-S bonds in bromobenzylthio substituents as well as Csp^3-N bonds in morpholinomethyl (piperidinomethyl) substituent lead to the even-electron fragment ions **c**. The inductive cleavage of Csp^3-N bonds of morpholinomethyl (piperidinomethyl) substituent proceeds also with the retention of charge to form even-electron fragment ions **j**. The inductive cleavage of Csp^3Csp^2 bonds of these substituents leads to even-electron fragment ions **i**.

The differences in the fragmentations within each of the four sets of isomeric compounds within the series of **VIa-VIc**; **VIIa-VIIc**; **IXa-IXc** and **Xa-Xc** are expressed quantitatively by comparing the calculated values of the coefficients μ, *i.e.* the abundances of selected even-electron fragment ions relative to the abundances of the corresponding molecular ions. Table 6 presents the values of μ_1, μ_2 and μ_3. As shown in this table, the differences in the values of μ_1, μ_2 and μ_3 are sufficient to differentiate among the isomeric sets **VIa-VIc**, **VIIa-VIIc**, **IXa-IXc** and **Xa-Xc**.

Conclusions.

The Mannich reaction of 2-thiouracil, formaldehyde and piperidine leads to 5-piperidinomethyl-2-tiouracil. The reaction of 1.32 mmole of PMTU with 1.46 (or 3.33) mmole of **IVa-IVc** in DMF in the presence of 0.83 (or 1.33) mmole of K_2CO_3 at room temperature leads chemoselectively to **VIIa-VIIc** or regioselectively to **Xa-Xc**. The reaction of 1.32 mmole of MMTU with 1.46 (or 3.33) mmole of **IVa-IVc** in DMF in the presence of 0.83 (or 1.33) mmole of K_2CO_3 at room temperature leads chemoselectively to **Va-Vc** or regioselectively to **VIIIa-VIIIc**. Treatment of **Va-Vc** and **VIIIa-VIIIc** with 10 % K_2CO_3 water solution affords the corresponding free bases **VIa-VIc** and **IXa-IXc**.

The differences in the values of the coefficients μ_1, μ_2 and μ_3 calculated from the EIMS spectra of **VIa-VIc**;

Table 6

The values of μ_1-μ_3 calculated from the EI mass spectra of metameric **VIa-VIc; VIIa-VIIc; IXa-IXc; Xa-Xc**

Comp	μ_1	μ_2	μ_3
VIa	3.25	2.91	5.58
VIb	4.50	11.71	12.85
VIc	4.33	33.33	13.00
VIIa	0.63	0.31	14.28
VIIb	0.77	1.22	11.11
VIIc	1.08	4.08	8.33
IXa	6.66	2.60	1.93
IXb	4.00	2.24	3.52
IXc	14.28	2.71	6.42
Xa	16.60	8.66	10.00
Xb	33.33	21.33	31.33
Xc	24.00	21.50	50.00

$$\mu_1 = \frac{\%\ \text{rel. abund. } \mathbf{i}\rceil^+}{\%\ \text{rel. abund. } \mathbf{a}\rceil^{+\cdot}}\ \textbf{VIa-VIc}\ \textbf{VIIa-VIIc} \qquad \mu_1 = \frac{\%\ \text{rel. abund. } \mathbf{h}\rceil^+}{\%\ \text{rel. abund. } \mathbf{a}\rceil^+}\ \textbf{IXa-IXc}\ \textbf{Xa-Xc}$$

$$\mu_2 = \frac{\%\ \text{rel. abund. } \mathbf{m}\rceil^+}{\%\ \text{rel. abund. } \mathbf{a}\rceil^{+\cdot}}\ \textbf{VIa-VIc}\ \textbf{VIIa-VIIc} \qquad \mu_2 = \frac{\%\ \text{rel. abund .i}\rceil^+}{\%\ \text{rel. abund. } \mathbf{a}\rceil^+}\ \textbf{IXa-IXc}\ \textbf{Xa-Xc}$$

$$\mu_3 = \frac{\%\ \text{rel. abund. } \mathbf{n}\rceil^+}{\%\ \text{rel. abund. } \mathbf{a}\rceil^{+\cdot}}\ \textbf{VIa-VIc}\ \textbf{VIIa-VIIc} \qquad \mu_3 = \frac{\%\ \text{rel. abund. } \mathbf{j}\rceil^+}{\%\ \text{rel. abund. } \mathbf{a}\rceil^+}\ \textbf{IXa-IXc}\ \textbf{Xa-Xc}$$

VIIa-VIIc; **IXa-IXc** and **Xa-Xc** (Table 6) allow a differentiation of the isomers.

EXPERIMENTAL

The purity of all compounds described was checked by m.p.'s, TLC and elemental analysis. Melting points (uncorrected) were determined on a Böetius microscope stage UV/VIS spectra were recorded with a Specord UV/VIS spectrophotometer in methanol. IR spectra were recorded with a FT-IR Bruker IFS–113 v spectrophotometer in KBr pellets. The ^{1}H NMR and ^{13}C NMR spectra were determined with a Varian Mercury Spectrometer operating at 300.07 MHz (proton) or 75.40 MHz (carbon). The data were obtained from DMSO-d_6 solution at a concentration between 0.25 and 0.40 *M* at ambient temperature. The chemical shifts were referred to tetramethylsilane. The heteronuclear 2D ^{13}C NMR and ^{1}H NMR chemical shift correlation experiments were carried out using HETCOR spectra. Elemental analyses were performed with a Vector Euro EA 3000 analyzer. Low- and high-resolution mass spectra were recorded on an AMD - Intectra GmbH-Harpstedt D-27243 Model 402 two - sector mass spectrometer (ionizing voltage 70 eV, accelerating voltage 8 kV, resolution 10 000). Samples were introduced by a direct insertion probe at the source temperature of ~150°C. The elemental compositions of the ions were determined by a peak matching method relative to perfluorokerosene and on the same instrument. All masses measured were in agreement with the composition given in column 3 of Tables 4 and 5 to within ± 2 ppm. The B/E linked scan spectra in the first field - free region were measured on the same spectrometer. The values of μ_1, μ_2 and μ_3 were calculated as averages of three measurements. 2-thiouracil and *o*-(*m*- and *p*-)bromobenzyl bromides were available from Merck Company, paraformaldehyde and piperidine from Fluka, morpholine from Aldrich. MMTU was obtained according to the literature [8].

The synthesis of 5-piperidinomethyl-2-thiouracil (PMTU) (IIIb). An equimolar mixture of 2-thiouracil (12.8 g), paraformaldehyde (3 g) and piperidine (9.87 ml) was suspended in 400 ml of ethanol and refluxed for 48 hours. The homogeneous solution obtained was filtered and concentrated on a rotatory evaporator to 200 ml. The reaction mixture was kept at room temperature for 24 hours. The precipitated solid was isolated by filtration, dried at room temperature, and recrystallized from methanol.

General Procedure for the Preparation of Va-Vc and VIIIa-VIIIc. A mixture of 0.83 (or 1.32) mmole of K_2CO_3 and 1.32 mmole of MMTU in 10 ml of dry DMF was stirred at room temperature for 2 hours. Next 1.46 (or 3.33) mmoles of corresponding **IVa-IVc** were added. After stirring at room temperature for 24 hours, 10 ml of distilled water was added. The reaction mixture was kept at room temperature for 24 hours. The oily organic layer was separated, triturated with dry diethyl ether and left for 24 hours in a refrigerator. The product formed was collected by filtration and crystallized from methanol.

General Procedure for the Preparation of VIa-VIc and IXa-IXc. Compounds **Va-Vc** (or **VIIIa-VIIIc**) were dissolved in 10 % K_2CO_3 water solution and stirred for 10 minutes. Then the reaction mixture was extracted with $CHCl_3$. Half of the volume of the solvent was evaporized, and the obtained **VIa-VIc** were collected by filtration, washed with diethyl ether and dry air direct. **IXa-IXc** were separated by silica gel column chromatography (Merck 203-400 mesh) using the following solvent mixtures CH_2Cl_2 – CH_3OH 80:1 (100 ml), CH_2Cl_2 – CH_3OH 60:1 (100 ml), CH_2Cl_2 – CH_3OH 40:1 (100 ml). The fractions of 20 ml were collected. **IXa-IXc** were present in fractions 5-8. On the basis of analytical TLC, large fractions of products desired were obtained by combining 20 ml fractions. They were concentrated on a rotatory evaporator. Compounds **IXa-IXc** were shown to be analytically pure.

General Procedure for the Preparation of VIIa-VIIc and Xa-Xc. A mixture of 0.83 (or 1.32) mmole of K_2CO_3 and 1.33 mmole of PMTU in 10 ml of dry DMF was stirred at room temperature for 2 hours. Next 1.45 (or 3.33) mmoles of the corresponding **IVa-IVc** were added. After stirring at room temperature for 24 hours, 10 ml of distilled water was added. The reaction mixture was kept at room temperature for 24 hours. The oily organic layer was separated, triturated with dry diethyl ether and left for 24 hours in refrigerator. The obtained solid of **VIIa-VIIc** was collected by filtration and crystallized from methanol. The crude solids of **Xa-Xc** were separated by silica gel column chromatography (Merck 203-400 mesh) using the following solvent mixtures CH_2Cl_2 – CH_3OH 80:1 (100 ml), CH_2Cl_2 – CH_3OH 60:1 (100 ml), CH_2Cl_2 – CH_3OH 40:1 (100 ml). The fractions of 20 ml were collected. **Xa-Xc** were present in fractions 6-9. On the basis of analytical TLC large fractions of products desired were obtained by combining 20 ml fractions. They were concentrated on a rotatory evaporator. Compounds **Xa-Xc** were shown to be analytically pure.

REFERENCES AND NOTES

[1] E. De Clercq, J. Balzarini, *Il Farmaco*, **50**, 735-747 (1995).

[2] H. Tanaka, H. Takashima, M. Ubasawa, K. Sekiya, N. Inouye, M. Baba, S. Shigeta, R. T. Walker, E. De Clercq, *J. Med. Chem.*, **38**, 2860-2865 (1995).

[3] J. Crooks, in "*Side Effects of Drugs*." Expertia Medica, Eds. L. Meyler, A. Herzheimer, Elsevier, Amsterdam 1972 p. 573-576.

[4] R. F. Berth, A. H. Soloway, R. G. Fairchild, *Cancer Res.*, **50**, 1061– 070 (1990).

[5] M. F. Hawthorne, *Angew. Chem.*, **105**, 997-1033 (1993).

[6] W. Tjarks, D. Gabel, *J. Med. Chem.*, **34**, 315-319 (1991).

[7] R. A. Nugent, S. T. Schlachter, M. J. Murphy, G. J. Cleek, T. J. Poel, D. G. Wishka, D. R. Graber, Y. Yagi, B. J. Kaiser, R. A. Olmsted, L. A. Kopta, S. M. Swaney, S. M. Poppe, J. Morris, W. G. Tarpley, R. C. Thomas, *J. Med. Chem.*, **41**, 3793-3803 (1998).

[8] P. Kamalakannan, D. Venkappayya, T. Balasubramanian, *J. Chem. Soc. Dalton Trans.*, 3381-3391 (2002).

[9] E. Wyrzykiewicz, T. Pospieszny, A. Barłóg, *Rapid Commun. Mass Spectrom.*, **19**, 580-584 (2005).

[10] E. Wyrzykiewicz, A. Szponar - Krajewicz, *Rapid Commun. Mass Spectrom.*, **17**, 1919-1923 (2003).

[11] E. Wyrzykiewicz, T. Pospieszny, S. Mielcarek, *Phosphorus , Sulfur and Silicon and the Related Elements*, **180**, 2051-2070 (2005).