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SYNTHESIS OF SOME NITROSOUREIDO DERIVATIVES OF ACYCLIC NUCLEOSIDES WITH EXRECTED ANTICANCER AND ANTIVIRAL ACTIVITY

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ABSTRACT

6 - CHLORO 9- (2- acetoxyethoxymethyl) Purine 1 reacted with ethylenediamine, p - phenylenediamine or piperazine to give the amino compounds 2 - 4 , which were treated with methylisocyanate to afford the urea derivatives 5 - 7. Reaction of these urea derivatives with sodium nitrite in formic acid led to the formation of the corresponding nitroso urea derivatives 8 - 10.

INTRODUCTION

It Was reported [1] that some nitrosourea derivatives are used as anticancer agents. The nitrosoureido group has an advantage over other reactive substituents in that it can break down in solution into two highly reactive compounds, an azido hydroxide and an isocyanate [1,2]. Either of these products if generated at an enzyme binding site could effectively destory enzyme activity through alky-

lation or carbamoylation of nucleophilic group on the enzyme [3].

It has been found that N^6 - substituted adenosine nuleosides inhibit the growth of certain tumors 4,5.

It is widely held that many cancer may be virally induced. This has suggedted that a suitable antiviral agent may prevent reinduction of a cancer previously controlled by other chemical means[6].

A considerable number of nucleoside analogues have been synthesized and tested for antiviral activity against HSV from which 9- (2 - hydroxy ethoxy methyl) guanine (Acyclovir) [7] is the most active.

It seems desirable to introduce the nitrosourido groups into 9-(2 - hydroxy ethoxy methyl) purine moeity so as to produce analogues which might possess the anticancer properties of nitrosourea and N^6 -substituted adenosine and the antiviral activity of acyclic nucleosides or both.

RESULTS AND DISCUSSION

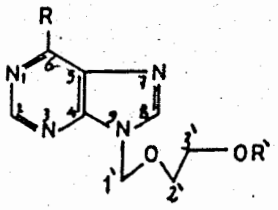
6 - substituted purine ribonucleosides have been synthesized by treatment fo 6-halogenopuine riboncleosides [8-13], 6-methy 1-(or benzyl) thiopurine riboncleosides [14], or 6-trimthylsilyl oxypurine ribonucleosides [15] with appropriate amines.Fleysher et al [11-13], have synthesized several N^6-substituted adenosines by treatment of 6-

chloropuine ribonucleoside with amines in the presence of $CaCo_3$ or Et_3 N, whereas kikugawa et al. [16] used a large excess of various amines in Et OH in absence of an auxiliary acid acceptor to obtain the corresponding N^6 -substituted adenosine dervatives.

In the present syudy ethylenediamine, p-phenylenediamine and piperazine are allowed to react with 6-chloro, 9-(acetoxyethoxy methyl) purine 1 which was prepared by Robins et al. [17] The diamines were used in excess to realize the mono substitution and consequently to obtain the compounds 2-4 which have a free amino group. It has been observed that the reactions are accompanied by elemination of the actyl group to give the deprotected acyclic nucleosides. Ethylenediamine needed one hour to react and the reaction occured in absence of any acid acceptor. The reaction of piperazine durated 48 h.in presence of triethylamine while p-phenylenediamine reacted by using 1,8-diaza bicycio (5, 4, D-) undec-7-ene (DBU) as an acid acceptor. The appearanece of the diamine peaks in NMR spectra beside the peaks of the starting acyclic nucleoside confirmed the stuctures of the compounds 2-4.

The acyclic nucleosides containing free amino group are converted into the corresponding urea derivatives 5-7 rpon treating with methyl isocyanate in absolute ethanol. ^{1}H NMR spectra of 5-7 showed doublets at ~ 2.65 ppm for the methyl group urea aned broad quartets at ~ 6.0 ppm for NH of urea adjcent to methyl group.

A broad triplet was observed at ~ 6.20 ppt in the spectra of 5 indicating the NH of urea adjacent to ethylene group. These results are in agreement withe the results obtained by Elliot et al. [3]. ^{13}C NMR of these compounds showed peaks at ~ 26.1 ppm for CH_3N and at 156 ppm for carbonyl of urea.



	R	R'
1	Cl	$COCH_3$
2	$HN(CH_2)_2NH_2$	H
3	1,4-$HNC_6H_4NH_2$	H
4	N⟨⟩NH	H
5	$HN(CH_2)_2NHCONHCH_3$	H
6	1,4-$HNC_6H_4NHCONHCH_3$	H
7	N⟨⟩$NCONHCH_3$	H
8	$HN(CH_2)_2NHCON(NO)CH_3$	H
9	1,4-$HNC_6H_4NHCON(NO)CH_3$	H
10	N⟨⟩$NCON(NO)CH_3$	H

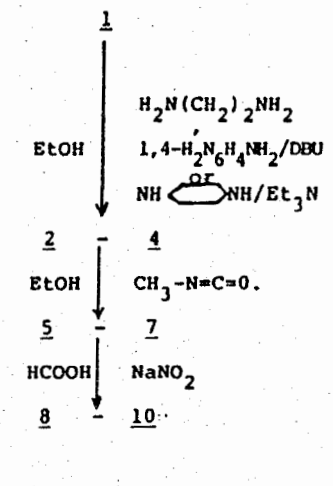


chart 1

Nitrosation of 5-7 with Sodium nitrite in 98 % formic acid gave the nitrosoureido derivatives 8-10. Evidence for the fact that nitrosation has occurred at the urea nitrogen adjacent to the methyl group has gained from the ^{1}H NMR spectroscopy. The nitroso- ureas showed a down field shift of about 0.5 ppm for the N-methyl resonance resulting from anisotropy of the adjacent CONNO group [3,18]. Moreover disappearance of the peak at ~ 6.0 ppm which is consistent with the $NHCH_3$ proton may support the idea of nitrosation at this position.

EXPERIMENTAL

^{13} C NMR and ^{1}H NMR spectra were recorded on a Bruker AC 250 spectromter. Mass spectra were recorded on varian MAT 311A spectrometer. Microanalyses were carried out at the Microanalytical Unit, Cairo University. 6-chlro, 9- (2-actoxyethoxymethyl) purine 1 was prepared using the mthod of Robins et al. [17]

Preparation of N- (2-aminoethyl), 0- (hydroxy ethoxy methyl) adenine 2. :

To an ice- cooled solution of ethylenediamine (3.6g, 60 mmol) in 50 ml abs. EtOH 1(2.7g,10 mmol) was aded. The reaction mixture was stirred and refluxed for 1h. Ethylene diamine hydrochloride was filtered off and the filterate was evaporated under reduced

pressure till dryness. The residue was chromatographed on sillica gel (100 g, 0.04-0.063 mm) with CH_3 OH in CH_2 C_{12} (5-15 %) to give 2, m.p. 157-160 °C, yield 1.0 g (40 %), ^{1}H NMR (CD_3 OD / TMS) δ 2.09 (t,4H, j= 6.0 Hz, NCH_2 CH_2 N), 3.55 (t, 4H, J = 5.9 Hz, 2`, and 3`,-H) , 4.80 (s, 1H, OH), 5.67 (s, 2H, 1,-H), 8.22, 8.30 (2 x s, 2H, 2-and 8-H), 9.37 ppm (s, 1H, NH), ^{13} C NMr (CD_3 OD/ TMS) δ 39.81, 40.15 (NCH_2 CH_2N),59. 94, 70.13 (C-2' and C-3') , 72.28 (C-1'), 118.80 (C-5), 141.90 (C-8), 194.20 (C-4), 152.62 (C-6), 154.53 ppm (C-2), MS: m / 2 =252 (M^+).

$C_{10}H_{16}N_6O_2$ Calcd. C, 47. 6;H, 6.4;N, 33.3 Found C, 47 . 4; H,6.3, N, 33.0 %

prparation of N- (4-aminophenyl), 9-(2 - hydoxy methyl) adenine3.

To a solution of 1 (2.7 g, 70 mmol) in 57 ml abs. EtOH p - phenylene diamine (6,5 g, 60mmol) and DBU (6.2 g, 40 mmol) were added. The reation mixture was stirred and refluxed for 48h. The fomed pricipitate was filtered, washed with water then with ethanol to give 3, m.p. 205-207° C, yield 1.5 g (50 %), ^{1}H NMR (DMSO / TMS) 3.34-3.50 (m, 4H, 2'-and 3'-H), 4.86 (s,1H, OH),5.61 (s,2H, 1'-H),7.50-7.60 (m, 4H, Ar-H), 8.31, 8.36 (2xs, 2H, 2-and 8-H), 9.40 (s, 1H, NH), ^{13} C NMR (DMSO / TMS)δ 59.82, 70.72 (C-2'and C-3'), 72.21 (C-1'), 119.0 (C-5), 113.58, 115.23, 132.10, 128.20 (C-AR), 141.21 (C-8), 144.70 (C-4), 149.37 (C-6), 152.44 (C-2). MS : m / z 300 (M^+).

$C_{14}H_{16}N_6O_6$ Calcd. C, 56.O; H, 5.4; N, 28.0.

Found C,55.7; H; 5.8; N, 27.9 %.

Prepartion of 6-Piperaziny1, 9- (2- hydroxyethoxymethy1 purine 4.

To a solution of 1 (2.6 g, 10 mmol) in 60 ml abs. EtoH piperazine (5.2 g,60 mmo1) and triethylamine (4.0 g, 40 mmol) were added. The reaction mixture was stirred and refluxed for 40 h. The piperazine hydrochloride was filtered off and the filterate was evaporated under reduced pressure to dryness. The rdsidue was chromatogrphed on silica gel (100 g, 0.04- 0.063 mm) in whith CH_3 OH (5-10 %) 4, m.p. 120-122 ^oC, yield 1.0 g (36 %); ^1H NMR (DMSO / TMS)δ 3.10-3.25 (m,8H, N $(CH_2)_4N$), 3.40-3.58 (m,4H 2' - and 3'-H), 4.75 (s, 1H, O H), 5.60 (sd, 2H, 1'-H), 8.20, 8.30 ppm (2xs, 2H, 2-and 8-H); ^{13} C NMR (DMSO/ TMS)δ 46.50,. 48.00 (N $(CH_2)_4N$), 59.90, 70.86 (C-2' and C-3), 72.50 (C-1'), 119.02 ppm (C-5); MS: m / z =278 (M^+).

$C_{12}H_{18}N_6O_2$ Calcd. C, 51.8; H, 6.5; N,30.2.

Found C, 52.1; H, 6.4; N, 30.3 %.

preparation of the urea dervatives 5-7 :

To a solution of 1 mmol of the appropriate amino dervative 2-4 in 20 ml abs. EtOH 1.1 mmol (63 mg of methy1 isocyanate was added. The areaction mixture was stirred at room temperature for

30 min. Stirring was continued under reflux for addtional 1 h. The reaction mixture was filtered while hot, and the filterate was evaporated under reduced pressure till dryness. The residue was chromatoqraphed on silica gel (100 g, 0.04-0.063 mm) with CH_3 OH in $CHCl_3$ (0-10 & To give the urea derivatives 5-7 in 42-63 % yield.

N- (2-(3- Methylureido) ethyl), 9- (2- hydroxyethoxymethyl) adenine 5.

M.p.185-187 ^{o}C, yield 150 mg (49 %) ^{1}H NMR (CD_3 / OD/ TMS) δ 2.65 (d,3H, j = 4.1 Hz, CH_3N); 3.49-3.65 (m, 4H, 3, - and 2'-H), 4,65 (broad s, 1H, OH), 5.63 (s, 2H, 1'-H), 5.93 (broad q, 1 H, $NHCH_3$),6.21 (broad t, 1H, $NHCH_2$), 8.37, 8.41 (2 x s, 2H, 2- and 8-H), 9.86 pp, (s,IH, NH) ^{13}C NMR (CD3 OD / TMS) δ 26.41 (CH_3 N),39.78, 40.12 (NCH_2 CH_2N),59.90, 70.88, (C-2' and C'-3), 72.40 (C-1'), 119.24 (C-5), 141.69 (C-8), 149.60 (C-4), 152.0 (C-6), 152.3 (C-2), 155.90 ppm (CO).MS; m/z = 309 (M^+)

$C_{12}H_{19}N_7O_3$ Calcd. C, 46.6; H, 6.2; N,31.8.

Found C' 46.2; H' 6.5; N, 31.5 %.

N- (2- (3- Methylureido) phenyl,) 9- (2- hydroxyethoxy methyl) adenine 6.

M.p. 200-202 ^{o}C, yield 240 mg (67 %) , ^{1}H NMR (DMSO / TMS) δ 2.63 (d,3H, j = 4.1 Hz, CH_3N, 3.50 - 3.52 (m, 4H, 3' - and 2'-H), 4.70 (t, 1H, J = 4.1 Hz, OH) , 5.64 (s, 2H, 1'-H), 5.95 (broad

s, ^{1}H, $NHCH_3$), 7.34 (d, 2H, j = 8.5 Hz, Ar - H), 7.73 (d, 2H, J = 8.4 Hz, Ar-H)8.37, 8.41- (2xs, 2H, 2- and 8-H), 9.86ppm (s, ^{1}H, NH) ^{13} C NMR (DMSO / TMS) δ 26.12 (CH_3 N), 59.81, 70.76 (C-2' and C-3') , 72.27 (C-I') 119.32 (C-5),117.84, 121.65; 132.94, 135.86 (Ar-H), 141.62 (C-8), 149.80 (C-4), 152.06 (C-6), 152.33 (C-2), 155.86 ppm (CO). MS: m/ z = 357 (M^+).

$C_{16}H_{19}N_7O_3$ Calcd. C, 53.8; H, 5.4; N, 27.4

Found C, 53.4, H, 5.3; N, 27.1 % .

6- (N- (N.Methylcarboxamido) piperazino), 9- (2-hydroxyethoxy methyl purine 7.

m.p. 170-173 °C,yield 140 mg (42 %) ; ^{1}H NMR (DMSO / TMS) δ 2.65 (d, 3H, J = 4.0 Hz, CH_3N) , 3.20 (m, 8H $N(CH_2)_4N$),3.50 3.53 (m, 4H, 3'-and 2'-H), 4.66 (s, IH, OH), 5.65 (s,2H, I'H)m 5.99 ppm (broad q, ^{1}H, NH CH_3), ^{13} C NMR (DMSO / TMS) δ 26.15 (CH_3 N) , 48.8, 49.1 (N $(CH_2)_4N$), 59.90, 70.45 (C-2' and C-3') , 72.40 (C-I') , II9.24 (C-5), 141.96 (C-8) , 149.60 (C-4), 152.00 (C-6) , 152.30 (C-2), 155.92 (CO). MS : m / z = 335 (M^+).

$C_{14}H_{21}N_7O_3$ Calcd. C, 50.I; H, 6.3; N, 29.2

Found C, 50.4, H, 6.I; N, 29.0 %

preparation of the nitrosourea derivatives 8-10.

To a cold (ice- MeoH bath) solution of each of the urea derva-

tives 5-7 (0.4 mmol) in 98 % formic acid (18 ml) sodium nitrite (138 mg, 2 mmol) was added. The resulting solution was, stirred on cold Ih, and then evaporated to dryness in vaccu. The residue was chromatogaphed on sillica gel (50 g, 0.04-0.63 mm) with CH_3 OH in $CHCI_3$ (0-2 %) to give the nitroso urea dervatives 8-10 in 58-80 % yield.

N- (2- (3- Methyl- 3- nitrosouredo ethyl), 9- (2-hydoxy ethoxymethyl) adenine 8.

M.P. 120-123 ^{o}C dec. Yield 80 mg (58%), ^{1}H NMR (DMSO/ TMS) δ 3.18 (s, 3H, CH_3N), 3.20-3.25 (m, 4H, NCH_2 CH_2N), 3.49-3.55 (m, 4H,3'- and 2'-H), 4.65 (s, 1H, OH), 5.64 (s, 2H, 1'-H), 7.90 (broad s, 1H, $HNCH_2$), 8.38, 8.45 (2xs, 2H, 2-and 8-H), 10.59 ppm (s,1H,NH). ^{13}C NMR (DMSO/TMS) δ 26.97 (CH_3N), (40.74, 40.95 (NCH_2CH_2N), (40.74, 40.95 (NCH_2 CH_2N),59.80, 70.78 (C-2' and C-3'), 72.31 (C-1'), 119.39 (C-5), 141, 91 (C-8), 150.90 (C-4), 152.01 (C-6), 142.25 (C-2), 157.00 ppm (Co). MS: M/z 338 (M^+).

$C_{12}H_{18}N_8O_4$ Calcd. C, 42.6; H, 5.4; N, 33.1

Found C, 42.2; H, 5.6; N, 32.8%.

N-(4- (3- Methyl -3- nitrosoureido (phenyl), 9-(2-hydroxy-0 methyl) adenine 9.

M.p. 197-200 ^{o}C dec. yield 123 mg (80 %) . ^{1}H NMR (DMSO / TMS) δ 3.19 (s, 3H, (CH_3N), 3.50-3.55(m, 4H, 3'- Hand 2'-H) 4.65

(s, 1H, OH), 5.66 (s, 2H, I'-H) , 7.65 (d, 2H, J = 8.2 Hz, Ar-H), 7.93 (d,2H, J = 8.3 Hz, Ar-H) , 8.36, 8.42 (2xs,2H 2-and 8-H), 9.95 (s, ^{1}H NH), 10.60 ppm (s, ^{I}H, NH), ^{13}C NMR (DMSO / TMS) δ 26.97 (CH_3N), 59.8, 70.87 (C-3' and C-2'), 72.31 (C-1') , 119.39 (C-5),117.50, 120.91, 132.83, 135.98 (Ar-H) , 141.91 (C-8) , 150.90 (C-4), 142.01 (C-6), 152.52 (C-2), 157.03 ppm (CO); MS: m/z = 286 (M^+)

$C_{16}H_{18}N_8O_4$ Calcd. C, 49.7; H, 4.7; N, 29.0

Found C, 49.3; H, 4.9; N, 28.7%.

6-(N-(N-Methyl - N - nitroso carboxamido) piperazino), 9- (2-hydroxyethoxymethyl) , puine 10.

M.p. 130-134 °C dec. Yield 101 mg (70 %), ^{1}H NMR (DMSO /TMS) δ 3.18 (s, 3H, CH_3N), 3.23-3.30 (m, 8H, N $(CH_2)_4$N), 3.49-3.54 (m,4H, 3'-and 2'-H), 4.65 (s, 1H , OH), 5.64 (s, 1H, 1'-H), 8.32-8.42 ppm (2x s, 2H, 2-and 8 - H), ^{13}C NMR (DMSO / TMS) δ 29.92 (CH_3N), 47.21, 47.90 (N $(CH_2)_2$N), 59.75,70.56 (C-2' and C-3') , 72.35 (C-I'), 119.5 (C-5) , 141.00 (C-8), 149.23 (C-4), 152.05 (C-6), 152.90 (C-2), 157.12 PPm (CO), Ms: m / z = 364 (M^+).

$C_{14}H_{20}N_8O_4$ Calcd. C , 46.2.H, 5.5; N, 30.8.

Found C, 46.3, H, 5.3; N, 30.5 %.

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تخليق بعض مشتقات النيتروزويوريدات لنيكليوزيدات غير حلقية لها تأثير متوقع ضد السرطان والڤيروسات

صلاح القوصى

قسم الكيمياء - كلية العلوم - جامعة المنوفية

شبين الكوم - مصر

تم التفاعل بين ٦-كلورو-٩-(٢- اسيتوكس ايثوكس ميثيل) البيورين ١ وكل من ايثيلين ثنائى الامين والبارافينيلين ثنائى الامين والبيبرازين ليعطى المركبات الامينية ٢-٤. تفاعلت هذه المواد مع ايزوسيانات الميثيل فى الكحول المطلق مكونة مشتقات اليوريا لهذه النيكليوزيدات. أما مشتقات النيتروزويوريدات فقد تم الحصول عليها بمعاملة مشتقات اليوريا مع نيتريت الصوديوم فى وسط من حمض الفورميك.