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SYNTHESIS OF SOME 6-METHYLIDENE, 9-PURINE ACYCLIC NUCLEOSIEDS WITH EXPECTED ANTI-HIV ACTIVITY

Salah El - Kousy

Chemistry Department, Faculty of Science,

Menoufiya University, Shebin El - Kom, EGYPT

ABSTRACT

Reaction of 6 chloro, 9- (2- acetoxyethoxymthyl) pruine 1 with ethyl cyanoacetat, malononitrile and diethyl malonate in presence of NaH and DMF gave the corresponding 6- methylidene derivatives 2a-c, which were deprotected by trearment with methanolic ammonai to give 3a-c. During the reaction of 1 with cyanoacetamide deacetylation took place spontaneously to give the deprotected acyclic nucleoside 4. Treatment of 2a with methyl iodide in presence of NaH and DMF yielded the N- methyl derivative 5 . NMR and mass spectra of the synthesized compounds were discussed.

INTRODUCTION

There is a need for compounds that may be effective in the therapy of acquired immunodificiency syndrome (AIDS). Some compounds have been identified as having an inhibitory effect against retroviruses particularly HIV. It is important to find compounds

which are not expensive to prepare and with less prominent side effects. Therefore, I thought that acyclic nucleosides could realize these characters.

It has been recently reported[1] that some acyclic nucleosides showed a significant anti-HIV activty.

Although 6-alkylated purine nucleosieds have attracted much attention with respect to their physiological activity [2-4], very few reports have appeared on the direct introduction of an alkyl group into a perfomed purine nucoeoside [5-7]. Therefore, I aimed to prepare some 6-methylidene -9-purine acyclic nucleosides through direct alkylation method. I had also the interest to prepare the N-methyl derivative not only to compare its spectra with the other synthesized compounds but also because of the expected antiviral activity similar to N-mthylated copound reported by Chu et al [8] and Finalaneder et al [9].

RESULTS AND DISCUSSION

Few numbers of 6-methylidene-9-purine nucleosides were roported [7,10] and they were prepared by the nucleophilic substitution of 6-chloropuine ribonucleosides with the sodium salts of active methylene compounds. I have applied the same route to prepare the 6-mthylidene-9-purine acyclic nucleosides 2a-c and 4. Ethyl cyanoacetate, malononitrile, α-cyanoacetamide and diethl malonate were used as active methylene compounds. The first step of the re-

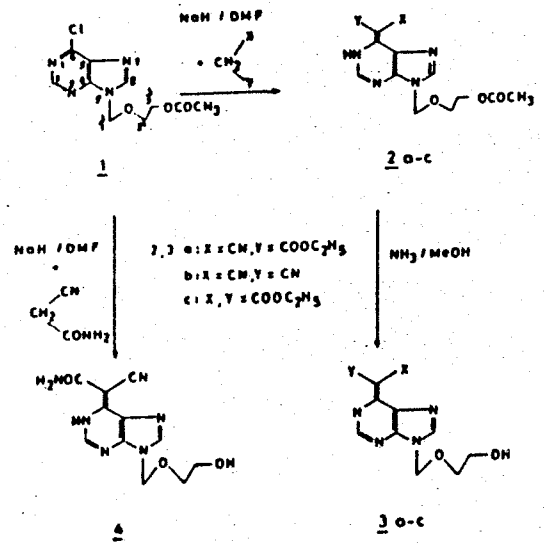


chart 1

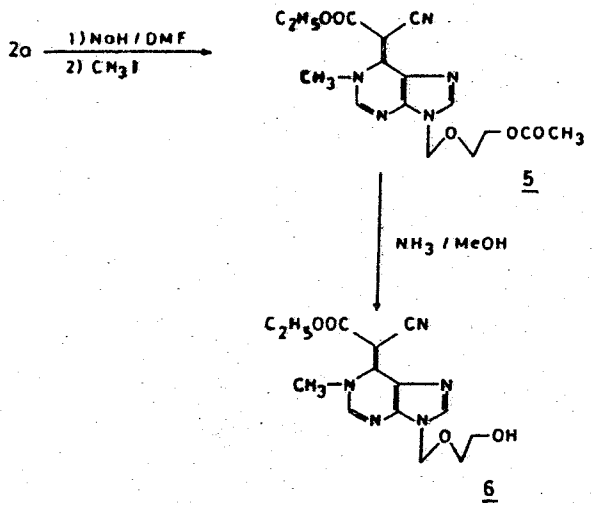
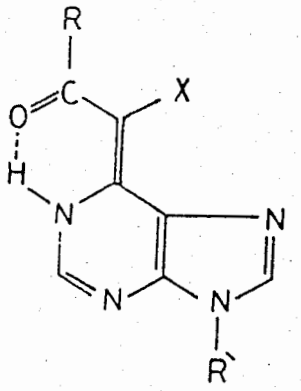


chart 2

action was the formation of the sodium salt of the active methylene compounds by using sodium hydride. Cimethylsulfoxide, N,N-dimethyl formamide and tetrahydrofuran were tested as solvents, but I found that dimethlformamide is the best one. It could be easly removed and had a good dissolution strength for the sodium salt of active methylene compounds. Treatment of 6- chloro-9- (2-acetoxythoxymethyl) purine (11 1 with the sodium salts of active methyline compounds at room temperature led to the formation of 2 and 4. Elelvated temperatures (50 and 75 % C) have also been tested but they diminished the yields. During the reaction of 1 with α-cyanoacetamid, the acetyl group was removed and the deprotected nucleoside 4 was obtained. This could be indicated by absence of the absorption of the ester carbonyl group at 1734 cm^{-1} in IR spectrum, absence of the singlet of $COCH_3$ at ~ 1.9 ppm in ^{1} H NMR spectrum and absence of the peaks of ^{13} C NMR at δ - 20.3 and 168 ppm of CH_3 and CO of the acetyl group, respectively.

The acetyl group in compounds 2a-c could be easly removed by treatment with methanolic ammonia solution. Compounds 2-4 were assigned the 1H purinylidene structure due to the presence of singlets at 12.8-15 for NH in their ^{1}H NMR spectra. All these compounds except the malononitrile derivatives 2_b and 3_b showed shifts at 13.5-15 ppm. These last peaks occur in the region of strongly hydrogen bonded protons and this confirmed the following stucture:



These results are in agreement with the reported rsesults for 2(1H) quinolylidene compound [12] indicated by UV IR and ^{1}H NMR spectra.

In my trials to introduce an alkyl group at N^1 of 6-methylidene purine derivatives, I have succeeded to prpare the N^1 methyl dervative 5 by using methyl iodide in presence of sodium hydide. The ^{1}H NMR spectrum of 5 is similar to that of the other dervatives except the abscence of the peak of NH.

Acetyl group was splited off from 5 again by treatment with methanolic ammonia to give 6.

Mass spectra of compounds 2a-c showed similar fragmentations of the acyclic parts of the compounds . Splitting of the frag-

ments COCH , CH_2CH_2O $COCH_3$, $OCH_2CH_2OCOCH_3$ and CH_2-$COCH_2CH_2OCOCH_3$ were recoded in each case.

EXPERIMENTAL

^{13} C NMR and ^{1}H NMR spectra were recorded on a Brucker AC 250 spectrometer. Mass spectra were recorded or varian MAT 211 A Spectrometer. IR spectra were recorded on perkin- Elmer 1720 FTIR spectrophotometer. Microanalyses were carried out by microanalytical center at cairo university.

6-Chlro, 9-(2-acetoxyethoxy methtyl) purine 1 was prepared according to the methods of Robins et al[11]

Preparation of the 6-methylidene -9-puine acyclic nucleosides 2a-c,4.

General procedure :

To an ice-cold solution of the appropriate active mthylene compound (25 mmol) in 15 ml of N,N-dimthyl formamide were added portionwise 60 % oil-immersed sodium hydride (0.64g, 16 mmol). The resulting mixture was stirred at room temperature for 1h. 6-Chlro-9- (2-acetoxyethoxy methyl) purine 1[11](1.08g, 5 mmol)was added and stirring was continued for 40 h at room temperatur. The solvent was evaporated under reduced pressure at 1 Torr. The residue was mixed with 100 ml of water. In case of matononitrile dervative, it was necessary to adjust the pH of the aqueous work-up to 7

with concentrated hydrochloric acid to effect precipiation. The crude products were crystallized from dioxane to give $\underline{2}_{a-c}$ and $\underline{4}$ in 40-60 % yield.

6- (Carboethoxy, cyano methylidene), 1H, 9- (2-acetoxyethoxymethy1) purine 2a.

M.p. 215-217 ^{o}C,yield 0.8 g(60 %), ^{1}H NMR (DMSO / TMS) α 1.92 (t, 3H J = 7.1 Hz, CH_3), 1.95 (s, 3H, $COCH_3$),3.74 5 Hz, 3'-H),.(t,2H,J=4.5 Hz, 2` ,-H), 4.09 (t,2H, J = 4,5 Hz, 3` - H) 4.24 (q, 2H, J = 7.1 Hz, CH_2),5.65 (s, 2H, 1'-H), 8.53 (s,2H,2-H, 8-H), 13.95 (s, 1H, NH). ^{13} C NMR (DMSO / TMS) δ 14.20 (CH_3), 20.35 ($COCH_3$), 95.92 (CH_2), 61.95 (= C<), 62.54, 66,97 (C-2', C-3`), 72.24 (C-1'), 117.33 (CN), 122.25 (C-5), 142.64 (C-8), 144,77 (C-2), 146.29 (C-4), 194.22 (C-6), 168.06 ($COCH_3$), 170.03 (COO). Ms (EI): m / z (%) = 347 (M+, 11.5) , 304 (0.77), 287 (1),260 (1.8), 244 (3.8),231 (6.9). IR (cm^{-1}), 1738, 2206

$C_{15}H_{17}N_5O_5$ Calcd. C,51.9, H, 4.9; N, 20.2

Found C,51.5; H, 5.1; N, 20.0 %.

6- (Dicyanomethylidene), 1H, 9-(2-acetoxy ethoxymethyl) purine 2b.

M.p. 220-221 ^{o}C, Yield 0.7 g (58%) ^{1}H NMR a 1.96 (s, 2H $COCH_3$), 3.50-3.74 (m,4H, 2'H,3'-H, 5.63 (s, 2H, 1'-H), 8.28, 8.51 (2 x s, 2H, 2-, 8-H), 12.88 (s, 1H, NH). ^{13}C NMR (DMSO/ TMS) δ

20.37 (COCH3), 59.69 (=C), 62.52, 66.96 (C'-2', C'-3), 72.51(C-1'), 116.3 (CN), 122.25 (C-25), 142.89 (C-8), 145.52 (C-2), 145.88 (C-4), 150.34 (C-6) 168.90 (CO CH_3). MS (EI): m/z (%) = 300 (M^+, 15.4), 240 (1) , 213 (7.7), 197 (23), 184 (32.3) . IR (Cm^{-1}), 1734, 2212.

$C_{13}H_{12}N_6O_3$ Calcd. c, 52.0; H,4.0; N, 28.0

Found C,51.7; H, 4.2; N,27.8%

.6- (Dicarboethoxyt methylidene) 1H, 9- (2 acetoxythoxymethyl) purine 2c.

M.P. 108-111 ^oC yield 0.6g (40%) ^1H NMR (DMSO/ TMS) δ 1.29 (t,6H, J= 7.0 Hz, 2 x CH_3),1.91 (s, 3H, $COCH_3$), 3.75 (t, 2H, J, = 4.5 H2, 2'H), 4.09 (t, 2H, J= 4.5, 3`-H), 4.16-4.29 (m,4H, 2 x CH2), 5.73 (s, 2H, 1'-H), 8.73, 8.92(2 x s 2H, 2,-8-H). 14.80 (s,1H, NH). ^{13}CNMR (DMSO/ TMS) α 13.66 (CH_3), 20.34 (OCN_3), 61.49 (CH_2), 62.85 (C-2'), 67.19 (C.3'), 72.39 (C-1'), 81.84 (=C), 130.94 (C-5), 146.85 (C-8), 151.02 (C-2), 151.99 (C-4), 156.20 (C-6), 167.99 ($COCH_3$), 170,50 (COO). MS (EI) m/z (%) =394 (M^+,1,2),322 (11) 307 (305), 291 (1.5) 277 (5). IR (Cm^{-1}), 1734.

$C_{17}H_{22}N_4O_7$ Calcd C,51.8; H, 5.6; N, 14.2.

Found C, 51.7, H, 5.6; N, 14.1 % .

6- (Carboxamido, cyanomethylidene) IH, 9- (2-hydroxyethoxymethyl) purine 4.

M.p 225-227° Cyield 0.62g (60 %), ^{1}H NMR (DMSO / TMS) δ 3.48-3.53 (m, 4H, 2'-H,3'-H), 4.65 (s, 1H, OH), 5.61 (s, 2H, 1'-H), 7.03 (broad s, 2H, NH_2), 8.43, 8.84 (2xs, 2H, 2-H,8-H) , 14.97 (s, 1H, NH), ^{13}C NMR (CMSO / TMS) δ 59.75 70.84 (C-2', C-3'), 62.26 (=C <) 72.56 (C-1'), 119.06 (CN), 121.96 (C-5), 141.95 (C-8), 144.35 (C-2), 145.66 (C-2), 149.22 (C-6), 170.40 (CO), MS: m/z = 276 (M^+), IR (cm^{-1}), 1631, 2196, 3250-3350.

$C_{11}H_{12}N_6O_3$ Calcd. C,47.8, H, 4.4; N, 30.4

Found C,47.4; H, 4.3; N,30.2 % .

preparation of 6- (Carboethoxy, cyano methylidene) 1-methyl, 9- (2-acetoxyethoxy methyl) purine 5.

To a a stirred ice-cold sloution of 2a (1.24 g, 5 mmol) in 20 ml N,N-dimethlyformamide were added portionwise 60 % oil immrersed sodium hydride (0.2 g, 5 mmol). The reaction mixture was then stirred for lh at room temperature. Methyl iodide (0.7g, 5 mmol was added to the reaction mixture and stirring was continued for overnight at the same temperature. The solvent was evaporatedunder reduced pressure at 1 Torr. The residue was chromatographed on sillica gel (50 g 0.04 - 0.063 mm) with CH_3OH / CH_2Cl_2 (1-3 %) to give 5, m.p 155-158 °C. yield 0.2g (17 %) ^{1}H NMR ($CDCl_3$ /

TMS) & 1.30 (t, 3H, J = 7.0 Hz, CH_3), 1.97 (s, 3H, $COCH_3$), 3.65-3.81 (m, 4H, 2'-H, 3'-H), 3.91 (s, 3H, NCH_3), 4.20 (q, 2H, J = 7.2 Hz, CH_2) 5.65 (s, 2H, 1' -H), 8.48, 8.59 (2xs, 2H, 2-H, 8-H). ^{13}C NMR ($CDCl_3$ / TMS) & 13.90 (CH_3), 20.37 ($COCH_3$), 43.40 (NCH_3), 59.90 (CH_2), 61.93 (= C), 62.54, 66.77 (C-2', C-3') 72.69 (C-1'), 116.99 (CN), 121.93 (C-5), 142.61 (C-8), 144.50 (C-2), 146.99(C-4), 149.35 (C-6), 176.92 ($CO CH_3$), 170.21 (COO) MS: m/z = 361 (M^+).

$C_{16}H_{19}N_5O_5$ Calcd, C, 53.2; H; 5.3; N, 19.4

Found C, 53.6; H, 5.0; N, 19.7 % .

Deprotection of the compounds 2 a-c and 5 to give 3 a-c and 6.

In 20 ml of sarurated solution of ammonia gas in methanol were suspended 1 mmol of the protected compound 2 a-c or 5. The reaction mixture was stirred for overnight. The solvent was evaported unedr reduced pressure and the residue was chromatogrphed on silica gel (50 g 0.04 - 0.063 mm) with CH_3OH / $CHCl_3$ (10-15 %) to give teh deprotected compounds 2 a-c and 6 in 54-75 % yield.

6- (Carboethoxy, cyano methylidene) 1H, 9- (2-hydroxy ethoxy methyl) purine 3 a.

M.p. 240-242 ^{o}C; yield 230 mg (80 %) ^{1}H NMR (DMSO/ TMS)δ 1.29 (t, 3H, J = 6.9 Hz, CH_3), 3.74-3.95 (m, 4H 2'-H,3'-H),

4.24 (q, 2H, J = 7.0 Hz , CH2), 4.56 (s, 1H , OH) , 5.65 (s,2H , 1` -H), 8.50-8.62 (2 x s, 2H, 2-H, 8-H), 13.92 (s, 1H, NH), $^{13}$C NMR (DMSO/TMS) & 14.15 ($CH_3$ ) 59.92 ($CH_3$ ), 61.80 (= C ), 62.50, 70.98 (C-2', C-3`), 72.68 (C-l') , 117.02 (CN), 121.93 (C-5), 142.46 (C-8) 144.71 (C-2,C-3), 146.92 (C-4), 149.35 (C-6), 170.13 (COO).

$C_{13}H_{15}N_5O_4$ Calcd. C, 5l. l; H; 5.0; N, 23.0.

Found C, 50.7; H; 5.3; N; 22.8 %.

6- (Dicyanomthylidene) lH, 9- (2-hydroxy ethoxymthyl) purine 3 b.

M.p. > 270 °C yield 155 mg (60 %) ^{1}H NMR (DMSO / TMS) & 3.52-3.69 (m, 4H, 2'-H, 3,-H), 4.59 (s, lH, OH), 5,63 (s, 2H, l;-H) , 8.30, 8.61 (2 x s, 2H, 2-H, 8-H), 12.50 (s, lH, NH) ^{13}C NMR (DMSO / TMS) & 59.43 (= C), 62.52, 66.83 (C-2', C-3') , 72.24 (C-1'), 116.50 (CN) 121.90 (C-5), 142.56 (C-8), 145.30 (C-2), 145.85 (C-4), 150.43 (C-6).

$C_{11}H_{10}N_6O_2$ Calcd. C, 51.2; H; 3.9; N; 32.5.

Found C, 50.7; H; 3.8; N; 32.1. %.

6- (Dicarboethoxy methylidene) lH, 9- (2-hydroxy ethoxymethyl) purine 3 c .

M.p. 150-154 °C, yield 158 mg (45 %). ^{1}H NMR (DMSO /

TMS)& 1.72 (t, 6H, J = 6.9 Hz, 2 x CH_3), 3.75-3.95 (m, 4H, 2'-H, 3'-H), 4.05-4.18 (m, 4H, 2 x CH 2) , 4.62 (s, 1H OH), 5.70 (s, 2H, l'-H), 8.70-8.88 (2 x s, 2H; x-2 - H, 8 -H), 14.90 (s, lH, NH). ^{13} C NMR (DMS / TMS) & 14.01 (CH 3) 61.00 (CH_2) , 62.10, 66.98 (C-2' C-3'), 72.28 (C-1'), 81.50 (= C), 129.85 (C-5), 146.20 (C-8), 150.56 (C-2_), 152.00 (C-4), 155.78 (C-6), 170.23 (COO).

$C_{15}H_{20}N_4O_6$ Calcd C,51.1; H; 5.7 N; 15.9.

Found C, 51.4; H; 5.5; N; 15.8 %.

6- (Carbethoxy, cyano mthylidene) 1-methyl, 9- (2-hydroxy ethoxymethyl) purine 6.

M.p. 175 -177 ^{o}C yield, 159 mg (50 %) , ^{1}H NMR ($CDCl_3$ / TMS) δ 1.28 (t, 3H, J = 7.1 Hz, CH_3), 3.61-3.79 (m, 4H, 2'-H, 3'-H), 3.92 (s, 3H, NCH_3) 4.23 (q, 2H; J = 7.0 Hz, CH_2) 4.59 (s, lH, OH), 5.65 (s, 2H, l'H) , 8.45, (8.60 (2 x s, 2H, 2-H, 8-H), ^{13} C NMR ($CDCl_3$ / TMS) α 14.01 (CH_3), 43.35 (NCH_3), 59.95 (CH_2), 61.80 (= C), 62.54, 66.86 (C-2', C-3'), 72.20 (C-1'), 116.54 (CN) , 122.02 (C-5), 142.52 (C-8), 144.95 (C-2), 146.83 (C-4), 149.75 (C-6), 170.73 (COO).

$C_{14}H_{17}N_5O_4$ Calcd C,52.7; H; 5.4; N, 21.9.

Found C, 52.2; H; 5.7; N; 21.7 %.

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تخليق بعض نيكليوزيدات ٦- ميثيليدين -٩- البيورين غير الحلقية والمتوقع لها فعالية ضد فيروسات فقدان المناعة المكتسبة

صلاح القوصى

قسم الكيمياء - كلية العلوم - جامعة المنوفية

شبين الكوم - مصر

تفاعلت المادة ٦- كلورو -٩- (٢- اسيتوكس ايثوكس ميثيل) البيورين ١ مع خلات سيانو الايثيل والمالونونيتريل وثانى مالونات الايثيل بمساعدة هيدريد الصوديوم فى ثنائى ميثيل الفورماميد- وادى ذلك الى تكوين مشتقات المثيليدين المناظرة ٢. تم نزع مجموعة الاسيتيل من هذه المركبات بمفاعلتها مع محلول النشادر فى الميثانول لتتكون النيكيوزيدات غير المحمية ٣. واثناء تفاعل المادة ١ مع سيانو الاسيتاميد تم إنفصال مجموعة الاستيل تلقائياً وتكونت المادة ٤. تفاعلت احدى المواد ٢ مع يوديد الميثيل فى وجود هيدريد الصوديوم ونشأ عن ذلك تكوين المشتق المحتوى على مجموعة الميثيل متصلة بذرة النيتروچين ٥ - وقد تم نزع مجموعة الاستيل منه ليتكون النيكليوزيد غير المحمى ٦ . نوقشت بعض القياسات الطيفية لهذه المركبات.