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Original Scientific Paper

Synthesis of Potential Anti-Cancer Agents. XVII.* Urea Nitrogen Mustards

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The synthesis of nitrogen mustard ureas from various isocyanates and the free base of *NN*-bis-(2-chloroethylamine) is described. Preliminary pharmacological tests show that some of these compounds have a cytostatic activity.

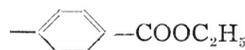
In one of the previous papers of this series compounds with potential cytostatic activity of the urea type were described. Continuing with the preparation of such compounds which would also contain *NN*-bis-(2-chloroethylamino) group the following substances were prepared:



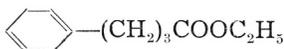
I



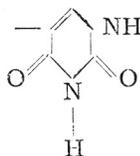
II



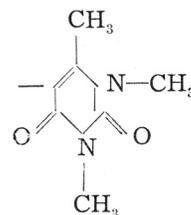
III



IV



V



VI

The isocyanates used for the preparation of IV, V and VI have not been described hitherto.

Preliminary pharmacological tests show that some of these compounds have a cytostatic activity.

EXPERIMENTAL

NN-bis-(2-chloroethyl) hydantoic acid ethyl ester (I)

A solution of 6.5 g. (0.05 mole) of carbethoxymethyl isocyanate in 50 ml. benzene was added to *NN*-bis-(2-chloroethylamine) free base in 50 ml. benzene. The base

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was prepared from 9 g. (0.05 mole) amine hydrochloride* (the temperature was between 15—25°). The reaction mixture was stirred overnight at room temperature. The volatile material was removed under reduced pressure at 40° and the residue was placed in the refrigerator; after several days it crystallized from ethyl acetate-petroleum ether or ethyl acetate in the form of white crystals (8.1 g., 60%), m.p. 64—66°.

Anal. C₉H₁₆Cl₂N₂O₃ (271.148) calc'd.: C 39.86; H 5.95; N 10.33%
found: C 39.75; H 5.99; N 10.45%

2-Benzyl-NN-bis-(2-chloroethyl)hydantoic acid ethyl ester (II)

To a solution of 8.9 g. (0.04 mole) of the isocyanate of phenylalanine ethyl ester¹ in 60 ml. benzene was slowly added *NN*-bis-(2-chloroethylamine) free base (from 8 g. or 0.045 mole of hydrochloride)* in 50 ml. of benzene under ice water cooling at 20—22°. The solution was stirred for 40 hours at room temperature, benzene was removed and the product allowed to crystallize in the refrigerator (16.2 g., 90%), m.p. 84—85° (after recrystallization from 2-propanol). The product is also soluble in ethanol, acetone and hot petroleum ether.

Anal. C₁₆H₂₂Cl₂N₂O₃ (361.266) calc'd.: C 53.19; H 6.14; Cl 19.63; N 7.76%
found: C 53.32; H 6.21; Cl 19.71; N 7.76%

4-Carbethoxyphenylisocyanate

To 150 ml. chlorobenzene was added 75 g. phosgene gas under stirring and cooling in an ice bath (Siefken's method was used³). To this solution at 0° was added slowly 20 g. of finely powdered *p*-aminobenzoic acid ethyl ester. After the reaction mixture was stirred overnight at room temperature, it was slowly heated to 70° and more phosgene was bubbled in. At 90° all had gone into solution. The temperature was raised to 130° for a short time until no more gas escaped. The solvent was removed *in vacuo* and the residue distilled at 0.8 mm., b.p. 118—119° (19.75 g., 78%). The infrared absorption spectrum showed a maximum at 2270 m μ .

NN-bis-(2-chloroethyl)-N'-(4-carbethoxyphenyl) urea (III)

4-Carbethoxyphenylisocyanate (9.6 g., 0.05 mole) was dissolved in 150 ml. of benzene, and *NN*-bis-(2-chloroethylamine) free base was added (from 9 g. or 0.05 mole of hydrochloride*) with stirring and cooling to 15—20°. The reaction mixture was stirred overnight at room temperature. The product precipitated. Benzene was removed, and the residue was recrystallized from absolute ethanol. (13 g., 81%), m.p. 131—32°.

Anal. C₁₄H₁₈Cl₂N₂O₃ (333.214) calc'd.: C 50.46; H 5.44; Cl 21.28; N 8.41%
found: C 50.49; H 5.34; Cl 21.10; N 8.32%

4-(*p*-Aminophenyl) butyric acid ethyl ester hydrochloride

A solution of 5 g. of 4-(*p*-aminophenyl) butyric acid⁴ in 50 ml. of absolute ethanol was stirred and saturated with dry hydrogen chloride without cooling. A solid was obtained. The mixture was evaporated to dryness at 1 mm. To protect the pump from hydrochloric acid a sodium hydroxide absorber was used. Finally the residue was dissolved in a small amount of cold absolute ethanol and the solution added to a large volume of dry ether. White crystals (6 g., 89%) were recrystallized from absolute ethanol to a melting point of 151—53°.

Anal. C₁₂H₁₇NO₂ · HCl (243.729) calc'd.: C 59.14; H 7.45%
found: C 59.36; H 7.59%

* 9 g. (0.05 mole) of *NN*-bis-(2-chloroethylamine hydrochloride) according to Mann² was suspended in cold benzene and 50 ml. of 5% NaOH was added.

NN-bis-(2-chloroethyl)-N'-(4-carbethoxypropylphenyl)-urea (IV)

4-(*p*-Aminophenyl) butyric acid ethyl ester hydrochloride (1 g., 0.0041 mole) was added to a cold solution (10°) of phosgene in 100 ml. of chlorobenzene and was stirred at room temperature overnight. Then the solution was concentrated to 50 ml. The isocyanate was distilled at 1 mm., b.p. 133°. IR 2269 $\text{m}\mu$. To this solution was added with cooling a benzene solution of dichloroethylamine free base from 0.9 g. of hydrochloride in 100 ml. of benzene*. After stirring overnight and boiling for one hour the solution was washed successively with a small amount of 10% HCl and water. The dry solution was concentrated to 10 ml. and petroleum ether added. The crystals were recrystallized from petroleum ether, (0.4 g., 19.8%) to a melting point of 54–55°.

Anal. $\text{C}_{17}\text{H}_{24}\text{Cl}_2\text{N}_2\text{O}_3$ (375.292) calc'd.: C 54.40; H 6.45; Cl 18.89; N 7.47%
found: C 54.55; H 6.60; Cl 19.02; N 7.35%

NN-bis-(2-chloroethyl)-N'-uracil-(5)-urea (V)

6 g. of 5-aminouracil and 600 ml. of benzene were stirred and boiled while phosgene was bubbled in for four hours. Subsequently the reaction mixture was stirred at room temperature and nitrogen was blown in overnight. The solvent was removed. The crude product gave a sharp maximum in infrared at 2270 $\text{m}\mu$. It was dissolved in 150 ml. of dimethylformamide at room temperature (at high temperatures the solution of the isocyanate polymerized) and with cooling to 10°, dichloroethylamine free base (from 7 g. of hydrochloride*) in benzene was added.

After stirring overnight at room temperature, all dimethylformamide was removed by repeated distillation with benzene under reduced pressure. The crude product, (6.9 g., 49.0%) was extracted nine times with hot ethyl acetate. After cooling, only 5 g. of a product, melting between 180–85°, precipitated. After 3 recrystallizations from ethyl acetate, a product with a melting point of 189–90° was obtained.

Anal. $\text{C}_9\text{H}_{12}\text{Cl}_2\text{N}_4\text{O}_3$ (295.132) calc'd.: C 36.62; H 4.10; Cl 24.03; N 18.98%
found: C 36.62; H 3.92; Cl 24.03; N 18.88%

NN-bis-(2-chloroethyl)-N'-(1,3,4-trimethyluracil)-(5)-urea (VI)

1.7 g. of 5-amino-1,3,4-trimethyluracil and 100 ml. of benzene were stirred and boiled while phosgene was bubbled in for four hours. The excess of phosgene was removed with nitrogen. The benzene solution was nearly clear; half of the benzene volume was removed. The product gave a sharp peak in IR at 2270 $\text{m}\mu$. The solution after cooling to 10° was treated with NN-bis-(2-chloroethylamine) free base (from 1.8 g. of hydrochloride*) in benzene.

After boiling for half an hour and stirring 15 hours at room temperature, the benzene solution was washed with a small amount of water, dried over magnesium sulfate, and concentrated to a volume of 10 ml. A small amount of petroleum ether was added. Some oil precipitated. The clear benzene-petroleum ether solution was separated and the product crystallized after one day in the refrigerator (1.5 g. 47%), m.p. 138–40°. Recrystallized from benzene, m.p. 142–43°.

Anal. $\text{C}_{12}\text{H}_{18}\text{Cl}_2\text{N}_4\text{O}_3$ (337.22) calc'd.: C 42.74; H 5.38; Cl 21.03; N 16.62%
found: C 42.95; H 5.17; Cl 21.07; N 16.80%

REFERENCES

1. S. Goldschmidt and M. Wick, *Liebigs Ann.* **575** (1952) 217.
2. F. G. Maan, *J. Chem. Soc.* **1934**, 461.
3. W. Siefken, *Liebigs Ann.* **562** (1949) 107.
4. W. Reppe, *Liebigs Ann.* **596** (1955) 21.

* See page 34.

IZVOD**Sinteza potencijalnih antikancerogenih supstancija. XVII. Uree diklordietilamina***R. C. Elderfield i D. Kolbah*

Pripravljen je niz derivata uree, koji sadrže diklordietilaminsku skupinu, na taj način da se slobodna baza *NN*-bis-(2-kloretilamina) kondenzirala s raznim izocianatima. Pretpostvlja se da će ovi spojevi imati citostatsko djelovanje.

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