SYNTHESIS OF 6-/N,N-1' ,6'-HEXYLENEFORMAMIDINE-^{14}C/ -PENICILLANIC ACID

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SUMMARY

Synthesis of 6-/N,N-1',6'-hexyleneformamidine-^{14}C/-penicillanic acid /VII/ was carried out. N-formyl-^{14}C-hexamethyleneimine /II/ was obtained from formic-^{14}C acid and hexamethyleneimine /I/. The product was chlorinated with oxalyl chloride. The obtained N,N-1,6-hexylenechloroformimino-^{14}C chloride /III/ was condensed with trimethylsilyl ester of N-trimethylsilyl-6-amino penicillanic acid /V/. The final product /VII/ was separated with a radiochemical yield of 46.5%.

INTRODUCTION

6-/N,N-1',6'-hexyleneformamidine/-penicillanic acid /VII/ is a new derivative of 6-aminopenicillanic acid[1].
It exhibits strong bacteriostatic action against a number of Gram-negative strains of bacteria particularly against E.coli species[2, 3].

The mode of antibiotic action of this compound differs

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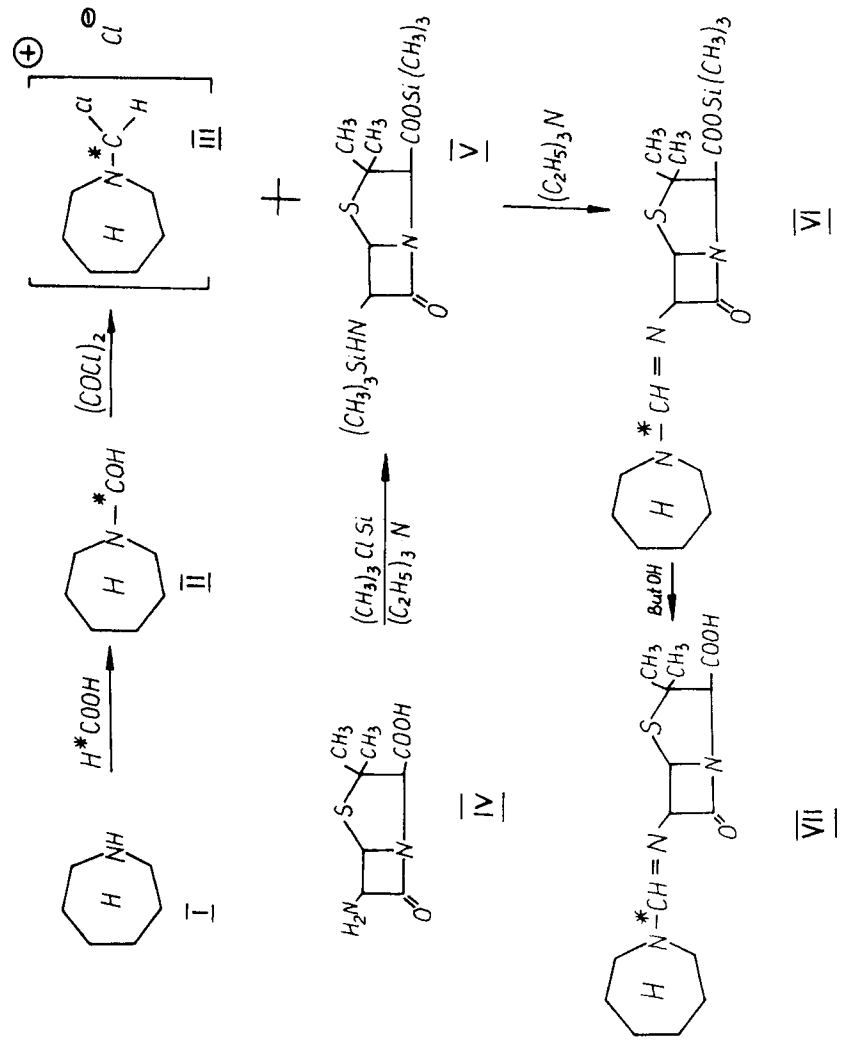

significantly from that of natural and semi-synthetic penicillins[4].

In order to pursue studies on the mode of the action and on the metabolism of compound VII in the bodies of animals and

humans the synthesis of preparation VII labelled with C-14 was carried out. The formamidine group was chosen as labelling site. This choice was guided on one hand by the possibility of carrying out the synthesis of a compound labelled with ^{14}C, on the other hand by the usefulness of examination of the stability of the $\rangle C = N$ bond in a side chain.

Experimental

N-formyl-^{14}C-hexamethyleneimine /II/

0.738 g /16 mM/ of ^{14}C-formic acid of a concentration of approx. 99% and an activity of 34.5 mCi was mixed with 1.104 g /24 mM/, of inactive formic acid. A preparation of an activity of 0.860 mCi/mM was obtained. 3.9 ml /34.5 mM/ of hexamethyleneimine was added to the acid and the mixture was heated at a temperature of 80 - 85° C for 6 hours. The water and the excess of ^{14}C-formic acid was separated by fractional vacuum distillation, and the principal fraction was collected at a temperature 90 - 95° C and at a pressure of 0.1 mm Hg.
4.094 g /32.2 mM/ of N-formyl-^{14}C-hexamethyleneimine of $n_D^{23} = 1.4864$ was obtained. The radiochemical yield was 80.5%.

/N,N-1,6-hexylene/-chloroformimino-^{14}C chloride /III/

4.094 g /32.2 mM/ of N-formyl-^{14}C-hexamethyleneimine was dissolved in 30 ml of water-free chloroform and cooled down to -20° C. A solution containing 2 - 5 ml /29 mM/ of oxalyl chloride in 25 ml of chloroform was added dropwise during 15 minutes upon continuous stirring. Stirring was continued for 20 minutes and

the temperature was mantained at -20° C.

Trimethylsilyl ester of 6-/N-trimethylsilyl/amino-penicillanic acid /VIII/

6.27 g /29 mM/ of 6-aminopenicillanic acid freshly crystallized and dried over phosphorous pentoxide was suspended in 120 ml of water-free chloroform. The mixture was cooled to -5° C and 7.6 ml /approx. 60 mM/ of trimethylchlorosilane was added dropwise upon vigorous stirring. After 10 minutes 8.1 ml /60 mM/ of triethylamine was added slowly.
The reaction was carried out for 1.5 hour and the temperature was mantained in the range 0° - 5°.

6-/N,N-1',6'-hexyleneformamidine-^{14}C/-penicillanic acid /VII/

Trimethylsilyl ester of 6-/N-trimethylsilyl/-aminopenicillanic acid was cooled to -70°. A solution of /N,N-1,6-hexylene/-chloroformimino-^{14}C chloride cooled to -20° was added dropwise upon vigorous stirring. Then 8.1 ml /60 mM/ of triethylamine was added dropwise. The reaction was carried out for 1.5 h, the temperature being raised slowly up to -10°. At this temperature the solution was concentrated almost to dryness. The yellowish syrup was extracted with 70 ml portions of water-free ether four times. The extract was filtered and kept at a temperature 0° C for 20 hours. Then 3 ml of water-free n-butanol was added dropwise and the mixture was stirred for half an hour. The mixture was kept for 2 hours at a temperature of 0° C.
The white precipitate was filtered off, washed quickly with ether and dried under vacuum over phosphorus pentoxide.

6.623 g /21 mM/ of 6-/N,N-1',6'-hexyleneformamidine-^{14}C/-penicillanic acid of a melting point of 150 - 152° C and of a total activity of 16.64 mCi and a specific activity of 0.76 mCi/mM. The chemical yield was 52.7%, the radiochemical yield, with respect to ^{14}C-formic acid was 46.5%.

The radiochemical purity was determined by thin-layer chromatography and autoradiography.

Silicagel G /Merck's product/ was used. The developing solvent consisted of n-butanol : water : acetic acid /40 : 40 : 1/. The R_f value for the obtained preparation was 0.53 and the radiochemical purity was 93.7%.

An IR-spectrum was also measured using KBr and Unicam SP-200 spectrophotometer.

Characteristic bands appeared at 1760 cm^{-1} /C = O -lactam/, 1680 cm^{-1} /C = N/ and 1605 cm^{-1} /COOH/.

Literature

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