Aus den IR-Spektren ermittelte nichtebene NH-Deformationsschwingungen $\gamma_{NH}$ und $NH_2$-Kippschwingungen $\omega_{NH_2}$

| Substanz | Lage des Absorptionsmaximums (in $cm^{-1}$) | «Kälteverschiebung» $\Delta\nu_K$ (in $cm^{-1}$) | Zuordnung |
|---|---|---|---|
| Benzimidazol | 881 | +27 | $\gamma_{NH}$ |
| | 1736 | +15 | $2\gamma_{NH}$ |
| Benzimidazol-[$D_1$] | 1288 | +17 | $2\gamma_{NH}$ |
| Purin | 864 | +22 | $\gamma_{NH}$ |
| Adenin | etwa 665 [b] | | $\omega_{NH_2}$ |
| | 870 [b] | | $\gamma_{NH}$ |
| 7-Methyladenin | 580 | +15 | $\omega_{NH_2}$ |
| | 1150 | + 9 | $2\omega_{NH_2}$ |
| 8-Azaadenin | 880 | +12 | $\gamma_{NH}$ |
| 9-Methyladenin | 690 [c] | +17 | $\omega_{NH_2}$ |
| 1-Methylthymin | 845 | +15 | $\gamma_{NH}$ |
| | 882 [c] | | |
| A-T-Modell | 615 [c] | | $\omega_{NH_2}$ (Adenin) |
| | 917 [c] | | $\gamma_{NH}$ (Thymin) |
| 1-Methylcytosin | 683 | + 5 | $\omega_{NH_2}$ |
| 9-Äthylguanin | 860 | | $\gamma_{NH}$ |
| | 655 | +18 | $\omega_{NH_2}$ |
| G-C-Modell | 890 | +12 | $\gamma_{NH}$ (Guanin) |
| | 700 | | $\omega_{NH_2}$ (Cytosin) |

Präparation: Nujol-Emulsion oder KBr-Tablette; verwendete Geräte: Perkin-Elmer Modell 521 [a], Carl Zeiss Jena UR 10 bezw. UR 20. [a] Die Messungen an diesem Gerät wurden bei einem Studienaufenthalt im Chemischen Institut «Boris Kidrič» (Ljubljana) durchgeführt; für die Ermöglichung dieser Arbeiten danke ich Herrn Prof. D. HADŽI. [b] Messungen von KANASKOVA et al.[2]. [c] Messungen von KYOGOKU et al.[5].

Neben der üblicherweise verwendeten N-Deuterierung können demzufolge zur Zuordnung der NH-Deformationsschwingungen auch die beim Abkühlen der festen Proben auftretenden beträchtlichen Verschiebungen der Absorptionsmaxima zu grösseren Wellenzahlen benutzt werden. Eine Änderung in der Stärke der H-Brücken, wie sie bei Ausbildung der komplementären Basenpaarung in den Nucleinsäuren auftritt, führt oft zu weiteren Verschiebungen dieser IR-Banden und kann die getroffenen Zuordnungen erhärten. Offensichtlich lassen sich schon relativ geringe Änderungen der Stärke der H-Brücken in diesen Systemen aus Verschiebungen der $\gamma_{NH}$ oder $\omega_{NH_2}$ nachweisen. Dadurch wird die Möglichkeit eröffnet, Konformationsunterschiede in den Nukleinsäuren – soweit sie die Stärke der H-Brücken zwischen komplementären Basen beeinflussen – mit Hilfe dieser Schwingungsfrequenzen zu erfassen.

*Summary.* The NH out-of-plane deformation and the $NH_2$ wagging vibrations of base-pairing models of DNA and related model substances in the solid state are assigned by deuteration shifts, cooling shifts, and shifts caused by the complementary base-pairing. The sensitivity of the frequencies of these vibrations to variations of the hydrogen bond strength may be useful to follow conformational changes of nucleic acids.

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## Pteridine Content of Some Methane- and Methanol-Oxidizing Bacteria

The alcohol dehydrogenase of *Pseudomonas sp.* M 27 is thought to contain a pteridine cofactor[1]. Similar ammonium-ion-activated, phenazine-methosulfate-linked alcohol dehydrogenases have been found in a number of methane- and methanol-oxidizing bacteria[2]. These findings prompted us to measure the amounts of pteridines in some of these organisms in an attempt to correlate pteridine content and the presence of a methanol/alcohol dehydrogenase, and as a first step in the actual isolation and elucidation of the structures of individual compounds.

Pteridine content was measured by submitting lyophilized samples of bacteria to alkaline permanganate oxidation as described previously[3]. Measurement of fluorescence (almost completely due to 2-amino-4-hydroxy-6-carboxypteridine) after oxidation provides a simple, accurate value of the pteridine content for comparison among different species. The results are given in the Table.

It is obvious that no significant correlation exists between pteridine content and the presence of methanol/alcohol dehydrogenases. Values cover a wide range and *Escherischia coli*, for example, has a pteridine content intermediate between the lowest and highest value. The amount in *Anacystis nidulans* (a blue-green alga) is very

[1] C. ANTHONY and L. J. ZATMAN, Biochem. J. *104*, 960 (1967).
[2] R. N. PATEL and D. S. HOARE, personal communication.
[3] F. I. MACLEAN, H. S. FORREST and D. S. HOARE, Arch. Biochem. Biophys. *117*, 54 (1966).

| Organism | Ref. | Methanol/alcohol dehydrogenase | Pteridine content $\mu M$/g Cell (dry weight) | $\mu M$/g Protein |
|---|---|---|---|---|
| *Pseudomonas* M27 | 1 | + | 0.29 | 0.24 |
| *Pseudomonas methanica* | 6 | + | 0.14 | |
| *Methylococcus capsulatus* (Bath) | 7 | + | 0.10 | 0.33 |
| *Methylococcus capsulatus* (Texas) | 8 | + | 0.76 | 0.37 |
| *Hyphomicrobium sp.* | 9 | + | 0.46 | |
| *Methylosinus trichosporium* (strain PG) | 7 | + | 0.25 | 0.075 |
| *Methylosinus sporium* (strain 5) | 7 | + | 0.37 | |
| *Methylobacter sp.* | 7 | + | 0.30 | |
| Pseudomonads | | | | |
| Yellow organism | 10 | + | 0.07 | |
| White organism | 10 | + | 0.10 | |
| Pink organism (grown on methanol) | 10 | + | 0.24 | |
| Pink organism (grown on succinate) | | + | 0.15 | |
| Pink yeast | 10 | − | 0.09 | |
| *Arthrobacter sp.* | 10 | − | 0.27 | 0.108 |
| *Escherischia coli* | 3 | not determined | 0.5 | |
| *Zyotobacter vinelandii* | 3 | not determined | 0.020 | |
| *Anacystis nidulans* | 3 | not determined | 6.8 | |
| *Chromatium* D | 3 | not determined | 0.20 | |

substantially higher than is found in these organisms. A few values were calculated in terms of the protein content of the cells but, although this resolved the apparent contradiction between the two strains of *Methylococcus capsulatus* (the 'Bath' strain must contain a large amount of non-proteinaceous material), it did not show any other significant trends.

The pteridines in the *Pseudomonas sp.* (pink organism) have been identified using methods described previously. The 4 compounds isolated from this organism are neopterin cyclic phosphate[4], 2-amino-4-hydroxy-6-carboxypteridine, neopterin, and 2-amino-4-hydroxy-6-methylpteridine. They were isolated in the ratio 3.5:2:1:4.7, respectively. These have also been isolated from *M. capsulatus*[4], and, apart from the cyclic phosphate, from *Methylosinus sporium* strain 5 and *Methylosinus trichosporium* strain PG[5]. There thus appears to be considerable similarity in the compounds occuring in these bacteria[11].

[4] T. URUSHIBARA, H. S. FORREST, D. S. HOARE and R. PATEL, Biochem. J., *25*, 141 (1971).
[5] T. URUSHIBARA and H. S. FORREST, Biochem. Biophys. Res. Commun. *40*, 1189 (1970).
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[10] R. J. MEHTA and D. S. HOARE, personal communication.
[11] Acknowledgment. This work was supported by the Robert A. Welch Foundation, Houston, Texas, USA, by Grant No. GM 12323 from the National Institutes of Health, U.S. Public Health Service, and by Grant No. GB-8173 from the National Science Foundation.
[12] Deceased, May, 1971.

*Zusammenfassung.* Der Pteridin-Gehalt einiger Methan-oder Methanol-oxydierender Bakterien scheint nicht mit der Anwesenheit einer spezifischen Methanoldehydrogenase zusammenzuhängen. Pteridine einer *Pseudomonas* Sp. (pink organism) wurden identifiziert.

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## An Urinary Metabolite of Bromazepam

Bromazepam[1], 7-bromo-1, 3-dihydro-5(2-pyridyl)-2H-1, 4-benzodiazepin-2-one, is a member of the 1, 4-benzodiazepines such as chlordiazepoxide and diazepam. It is now under clinical evaluation as a psychotropic drug in the control of ambulatory schizophrenics. The present investigation was undertaken in order to isolate and to characterize urinary metabolites of bromazepam administered to dogs as well as to rabbits. The drug was administered orally in a single dose of 200 mg/kg to rabbits and 50 mg/kg to dogs. Urine was collected during the following 48 h and hydrolysis of glucuronides was carried out by the usual method employing $\beta$-glucuronidase. The hydroly-

[1] Bromazepam (Ro 5-3350) is synthesized by R. IAN FRYER, R. A. SCHMIDT and L. H. STERNBACH, Department of Chemical Research, Hoffmann-La Roche, Inc., Nutley, N. J., USA (J. Pharm. Sci., *53*, 264, 1964).