In our case only 1 mole of cyanide seems to be bound per mole of protein and this may account for the easy transformation of MeHbCN back to hemoglobin, which is carried out in vivo by means of thiosulfate and represents the final step of cyanide psoioning treatment.

Riassunto. È stata studiata l'interazione tra metemoglobina e cianuro. Il cianuro sembra legarsi ad un solo eme e l'equilibrio non sembra influenzato da interazioni eme-eme.

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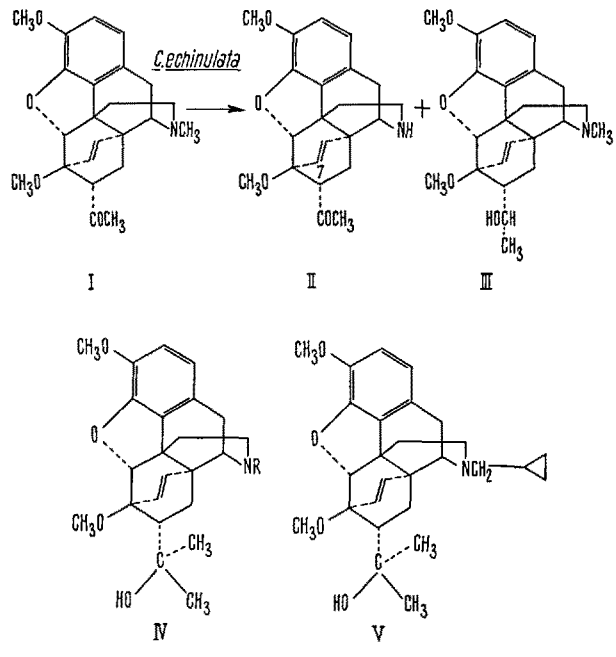
Microbiological Transformation of 6,14-*endo*-Ethenotetrahydrothebaine Alkaloids

The 6,14-*endo*-ethenotetrahydrothebaine alkaloids have attracted considerable scientific attention because they often exhibit extremely high analgesic potency[1]. The results of extensive chemical investigations have been reported recently[2–8]. We have attempted to prepare new members of this class by microbiological means.

In a typical example, a cell suspension of *Cunninghamella echinulata* (NRRL-A-11498) was prepared by shaking the organism for 24 h in a medium composed of 1% glucose, 0.1% yeast extract, 0.1% beef extract, 0.2% bactopeptone and 0.3% corn steep liquor. The substrate (I) was added (100 γ/ml final concentration) and the products were isolated after 6 days fermentation by chloroform extraction. The products were separated and obtained crystalline in 3–10% overall yield after partition chromatography using methanol-saturated hexane on acid washed diatomaceous earth. The products were identified readily from their characteristic micro NMR-spectra[8] which were obtained with a Varian Associates A-60 instrument using a computer of average transients attachment. The formation of secondary alcohol III[3] is the result of a common biological reaction but it is interesting to note that only 1 of the 2 possible stereoisomers was found[3]. The second product (II) clearly lacked the *N*-methyl function, for the characteristic three-proton singlet at 2.33 $\pm$ 0.03 ppm was missing[8]. No other major change in the spectrum was apparent and the spectrum was in fact identical to that of authentic II[4]. This identification was supported by alkylation studies with methyl iodide[2]. A third, minor, product turned out to be the C_7 epimer of II and may have been an artifact of the isolation procedure[1].

Dealkylation through chemical means in this series sometimes leads to difficulties via rearrangements so the microbiological process may be of some utility in difficult cases[4]. Interestingly, dealkylations of this type have not been reported previously through microbiological reactions on alkaloids. These reactions also find an interesting parallel in the proposal that morphine analgesics are dealkylated at the in vivo receptor site whereupon they exert their characteristic biological effects[9,10]. For these various reasons the reaction was studied in greater detail. From the results set out in the Table, it can be seen that the reactions described are fairly general. Furthermore, the nature of the alkyl group on nitrogen is not critical



Microbiological transformation of 6,14-*endo*-ethenotetrahydrothebaine alkaloids

Organism	Substrate	Products
Cunninghamella bertholletiae	I	II, III
C. echinulata	I	II, III
C. bainieri	I	II, III
Xylaria sp.	I	II III
Xylaria sp.	IV (R = CH_3)	IV (R = H)
Xylaria sp.	V	IV (R = H)

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because even a cyclopropylmethyl group was removed. In no case did both dealkylation and reduction take place on the same substrate. Among the alkaloids studied, IV (R = H) was the only substrate that was unattacked by any of the organisms used and it is probably significant that it is the only substrate which did not carry any alkyl function on the piperidino-nitrogen.

Previous work on the microbiological transformation of morphine alkaloids has led to a variety of interesting results[11–16], but our findings represent reactions which are apparently novel in this class. Virtually all the positions involved in previous studies are blocked by the ethenobridge in our substrates and the shape of the molecule is rigidly prescribed. It is, therefore, not surprising that our transformations followed another path.

Zusammenfassung. In der vorliegenden Arbeit werden mikrobiologische Umwandlungen von mehreren 6,14-*endo*-Äthenotetrahydrothebain Alkaloiden beschrieben. Mit Hilfe von verschiedenen Mikroorganismen konnten stereospezifische Reduktionen und *N*-Desalkylierungen ausgeführt werden.

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The Variation in Active Tension with Sarcomere Length in Vertebrate Skeletal Muscle and its Relation to Fibre Width

According to the sliding-filament model of muscular contraction[1,2], the tension output is related to the number of active sites formed, and hence to the area of overlap between the 2 sets of interdigitating filaments of the myofibril. As has recently been demonstrated[3,4], the length-tension relation exhibited by the vertebrate skeletal muscle fibre is accordant with this idea. Contraction does not, however, merely involve a longitudinal movement of the myofilaments. Due to the fact that the fibre maintains a virtually constant volume throughout changes in length[5,6], the myofilament packing density, i.e. the centre-to-centre distance between the filaments, may be assumed to vary as an inverse square root function of the sarcomere length. Data obtained in X-ray diffraction studies[7–9] support this view. On this basis it may be assumed that as the number of interacting sites is increased when the fibre shortens, the distance over which the active links have to operate in order to propel the A and I rods relative to each other is steadily increased. For example, by shortening of the sarcomere spacing from 3.6 μ (zero overlap) to 1.4 μ the centre-to-centre distance between the A and I filaments is increased by a factor of 1.6. For an adequate evaluation of the kinetics of the sliding-filament process, it is essential to find out whether the variation in active tension with sarcomere spacing exclusively refers to the length dimension of the contractile system, i.e. the area of overlap between the A and I filaments, or whether the tension output is also dependent on the width of the myofilament lattice. In the present study we have approached this problem by defining the length-tension curve in isolated skeletal muscle fibres that were subjected to various degrees of hydration. Evidence will be presented that changes in width of the fibre do not affect the relation between tetanic output and sarcomere spacing to any substantial degree.

Methods. The recording technique used was similar to that described previously[4,10]. The isolated fibre (dissected from the ventral head of the semitendinosus muscle of *Rana temporaria*) was mounted horizontally in a thermostated Perspex trough (1–2 °C) between an RCA 5734 tension transducer and a light isotonic lever. The resting length of the fibre and the amount of active shortening could be adjusted by means of microscrews in front of and behind the lever. The sarcomere spacing in the middle segment of the fibre was measured at rest for various degrees of stretch of the fibre in the beginning and at the end of the experiment[4]. The sarcomere spacing during activity was derived by cinephotographic recording (64 f/sec, 2.5 msec exposure time) of thin nylon filaments placed on the fibre surface across the fibre axis. Tetanic contractions were produced at 3 min intervals by passing a square pulse train of 1.0–1.5 sec duration (40 c/sec, pulse width 1 msec) through a pair of platinum electrodes mounted in the floor of the Perspex trough. Changes in fibre width produced by altering the osmotic strenght of the Ringer's solution were measured from the cine film (see above) for a given region of the fibre (close to a nylon filament) at 3 different sarcomere spacings (2.5–3.0 μ). The photographic records after enlargement were read to

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