Bei Poly (rA + rA) (Figur 1) bleibt D in den Grenzen der Messgenauigkeit (± 2%) bei der Dehydratation konstant. Änderungen der Basenorientierungen relativ zur Helixachse treten also nicht auf, wie auch aus Röntgenbeugungsuntersuchungen an Poly (rA + rA) hervorgeht[11]. Dieses Verhalten wird auf die Bindungsverhältnisse zwischen den Adeninbasen und dem Zucker-Phosphatgerüst zurückgeführt (Figur 3).

Die Adeninbasen sind durch 2 Wasserstoffbrücken miteinander verbunden. Zusätzlich bildet jede Adeninbase mit einem Sauerstoffatom einer Phosphatgruppe des gegenüberliegenden Strangs eine dritte Wasserstoffbrücke (N_{10}–H...O_6 in Figur 3). Ausserdem sind die N_1-Atome der Adeninbasen protoniert. Durch die elektrostatische Wechselwirkung der Protonen mit den negativ geladenen Phosphatgruppen der Gerüststränge wird der Molekülkomplex in hohem Masse stabilisiert. Daher bleibt die geordnete Sekundärstruktur von Poly (rA + rA) auch bei vollständiger Dehydratation der Proben erhalten.

Summary. LD measurements on poly (rA + rU) show a hysteresis of conformational changes within the dehydration-hydration cycle. The different conformational changes of poly (rA + rU) and DNA during hydration are discussed. With poly (rA + rA) no conformational changes could be detected during dehydration.

D. ZIRWER und M. BECKER

Deutsche Akademie der Wissenschaften zu Berlin, Institut für Biophysik, Berlin-Buch (DDR-1115), 10. Juni 1970.

Ethanol Increases Liver Uridine-Diphosphate-Glucuronyltransferase

The chronic administration of ethanol to humans and animals has been shown to induce proliferation of the smooth endoplasmic reticulum of the liver parenchymal cell[1–4], and to enhance some liver microsomal drug-metabolizing enzymes[5–10]. WALTMAN et al.[11] have lately reported a statistically significant reduction of serum bilirubin levels in new-born babies whose mothers had been given ethanol before delivery.

A possible explanation is that ethanol induces liver uridine-diphosphate-glucuronyltransferase (UDPG-transferase, E.C. 2.4.1.17), which is a microsomal enzyme necessary for the conjugation of free bilirubin. In order to test this hypothesis, the behaviour of UDPG-transferase was studied in the liver of rats chronically treated with ethanol. Aniline hydroxylase (E.C. 1.14.1.1) and nitroreductase (E.C. 1.6.6.2), microsomal enzymes, which are well known to be induced by ethanol[7], were also studied to provide a comparison with UDPG-transferase variations.

Moreover, the effect of ethanol administration on bilirubinaemia and liver UDPG-transferase values was studied in a young man with congenital Gilbert's type jaundice, which was found to be associated with low levels of liver UDPG-transferase activity[12,13].

Materials and methods. 20 male Wistar rats, weighing about 250 g, and fed with standard laboratory diet, were studied. 10 rats were treated daily for 15 days with ethanol (3 g/kg of body wt.). Ethanol was diluted 40% v/v with physiological saline and administered by gastric intubation. Each rat fed ethanol was matched with a control rat that received an isocaloric amount of glucose. After the treatment, the animals were fasting for 12 h before being killed by exsanguination. Portions of the livers were homogenized in 0.25 *M* sucrose + 1 m*M* EDTA, 20% w/v. Microsomes were isolated according to STRITTMATTER[14]. Enzyme assays were carried out on whole homogenates as well as on washed microsomes.

Aniline hydroxylase was assayed according to IMAI et al.[15]. Nitroreductase was determined as described by FOUTS and BRODIE[16]. UDPG-transferase was assayed according to VAN ROY and HEIRWEGH[17] and HEIRWEGH and MEUWISSEN[18], using unconjugated bilirubin as substrate. Total protein in whole homogenates and in washed microsomes was measured according to LOWRY et al.[19].

As far as the experiment in man is concerned, ethanol (1.5 g/kg body wt./day) was diluted with 900 ml of physiological saline, and administered i.v. at a rate of 2 ml/min. The ethanol administration was repeated daily for a week. Liver needle biopsies were taken before and after the treatment, and the specimens were used for UDPG-transferase assays and light microscopy study. The specimens were homogenized 8% weight per volume, as described for rat livers. Enzyme assays were carried out on the homogenates by the same method used for rat livers. Bilirubinaemia was assayed every other day.

Results and discussion. The mean aniline hydroxylase activity was strikingly increased in rat livers after ethanol administration; that of nitroreductase was significantly increased in washed microsomes only (Table). These data are in agreement with those of RUBIN et al.[7].

Liver UDPG-transferase activity was markedly enhanced in rats given ethanol: significant increases were

Mean hepatic activity (± S.E.) of aniline hydroxylase, nitroreductase and UDPG-transferase in control rats and in rats given 3 g/kg of ethanol/day for 15 days

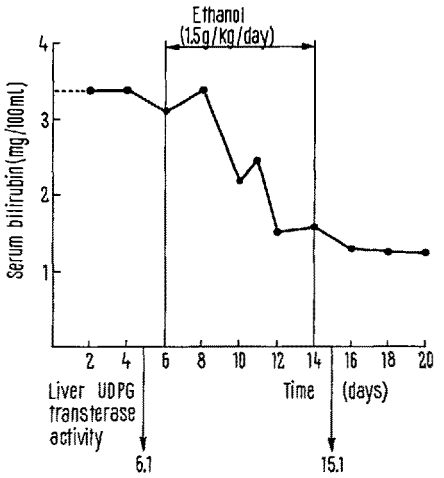
Samples	Rats	Aniline hydroxylase		Nitroreductase		UDPG transferase	
Whole homogenate	Controls	57.3 ± 5.3	$P<0.001$	176 ± 25	$P>0.05$	246 ± 16	$P<0.001$
	Treated	123.3 ± 11.9		244 ± 31		439 ± 41	
Washed microsomes	Controls	219.6 ± 21	$P<0.005$	46 ± 7	$P<0.005$	590 ± 69	$P<0.02$
	Treated	495.9 ± 83		160 ± 33		1337 ± 230	

Enzyme activities expressed as nmoles of *p*-aminophenol, *p*-aminobenzoic acid and conjugated bilirubin respectively formed per g of liver protein or microsomal protein per min at 37 °C.

observed in whole homogenates as well as in washed microsomes (Table).

A striking decrease in bilirubinaemia was observed in man during the treatment with ethanol (Figure); bilirubin level was still low 90 days after the end of the treatment. Liver UDPG-transferase activity was 6.1 nmoles of bilirubin conjugated/g of wet liver/min before the treatment, and 15.1 nmoles of bilirubin conjugated/g of wet liver/min at the end of the treatment (Figure).

These data suggest an inductive action of ethanol on liver UDPG-transferase. However, the mechanism of this induction is still uncertain. These observations confirm our preliminary communication[20], and might provide an explanation for the decreased bilirubin levels observed by WALTMAN et al.[11] in new-born babies whose mothers had been given ethanol before delivery.



Behaviour of serum total bilirubin and of liver bilirubin UDPG-transferase in a young man with congenital Gilbert's type jaundice, before and after the i.v. administration of ethanol for 7 days. Conjugated bilirubin always ranged from 0.20 to 0.25 mg/100 ml. Bilirubin UDPG-transferase activity expressed as nmoles of bilirubin conjugated/g of wet liver/min. In our laboratory, normal liver bilirubin UDPG-transferase values range from 14.5 to 44.

Riassunto. La bilirubina UDPG-transferasi epatica si è incrementata nel ratto ed in un paziente affetto da ittero di Gilbert dopo somministrazione di etanolo. Nell'uomo la bilirubinemia si è nettamente ridotta durante il trattamento ed è ancora su livelli normali a tre mesi di distanza dalla fine di questo. Questi dati potrebbero fornire una spiegazione ai bassi livelli bilirubinemici riscontrati in neonati da madre trattata con etanolo poco tempo prima del parto[11].

G. IDÉO, R. DE FRANCHIS,
E. DEL NINNO and N. DIOGUARDI

Istituto di Patologia Medica II dell'Università di Milano, 'Antonio Migliavacca Centre' for Liver Diseases, I-20122 Milano (Italy), 6 July 1970.

[1] O. A. ISERI, C. S. LIEBER and L. S. GOTTLIEB, Am. J. Path. *48*, 535 (1966).
[2] B. P. LANE and C. S. LIEBER, Am. J. Path. *49*, 593 (1966).
[3] E. RUBIN and C. S. LIEBER, Fedn Proc. *26*, 1458 (1967).
[4] E. RUBIN and C. S. LIEBER, New Engl. J. Med. *278*, 869 (1968).
[5] C. S. LIEBER and L. M. DE CARLI, J. clin. Invest. *47*, 62a (1968).
[6] C. S. LIEBER and E. RUBIN, Gastroenterology *54*, 642 (1968).
[7] E. RUBIN, F. HUTTERER and C. S. LIEBER, Science *159*, 1469 (1968).
[8] E. RUBIN and C. S. LIEBER, Science *162*, 690 (1968).
[9] C. S. LIEBER and L. M. DE CARLI, Science *162*, 917 (1968).
[10] C. S. LIEBER and E. RUBIN, New Engl. J. Med. *280*, 705 (1969).
[11] R. WALTMAN, F. BONURA, G. NIGRIN and C. PIPAT, Lancet *ii*, 1265 (1969).
[12] M. BLACK and B. H. BILLING, New Engl. J. Med. *280*, 1266 (1969).
[13] I. M. ARIAS, L. M. GARTNER, M. COHEN, J. BEN EZZER and A. J. LEVI, Am. J. Med. *47*, 395 (1969).
[14] C. F. STRITTMATTER and F. T. HUMBERGER, Biochim. biophys. Acta *180*, 18 (1969).
[15] Y. IMAI, A. ITO and R. SATO, J. Biochem., Tokyo *60*, 417 (1966).
[16] J. R. FOUTS and B. B. BRODIE, J. Pharm. exp. Ther. *119*, 197 (1957).
[17] F. P. VAN ROY and K. P. M. HEIRWEGH, Biochem. J. *107*, 507 (1968).
[18] K. P. M. HEIRWEGH and J. A. T. P. MEUWISSEN, Biochem. J. *110*, 31P (1968).
[19] O. H. LOWRY, N. J. ROSEBROUGH, A. FARR and R. J. RANDALL, J. biol. Chem. *193*, 265 (1951).
[20] N. DIOGUARDI, G. IDÉO, E. DEL NINNO and R. DE FRANCHIS, Lancet *i*, 1063 (1970).

Actinomycin D Inhibition of Intestinal Transport of L-Histidine and of Vitamin D Action

Renal aminoaciduria and reduced intestinal histidine transport were observed in rabbits with experimental vitamin D deficiency rickets[1], while administration of high doses of vitamin D to intact animals resulted in increased intestinal absorption of histidine[1]. Since absorption may involve a protein carrier in the intestinal mucosa, the present study was carried out to determine the effect of actinomycin D, an inhibitor of protein synthesis[2], on histidine absorption.

White rabbits weighing 700–1000 g, fasted for 12 h, were used. Under light thiobarbital anesthesia the small intestine was rapidly excised, washed in normal saline and everted, using the technique of WILSON and WISEMAN[3]. 3 or 4 sacs, each of about 3.5 cm long, were obtained from each animal. These sacs were each filled with 1.5 ml of L-histidine solution and placed in a flask containing 25 ml of the same solution. The solution used was Krebs-Henseleit bicarbonate saline, containing 0.3% (w/v) glucose in which L-histidine (Wako Pure chemical Industory; chemically pure grade) was dissolved. At the end of incubation the sacs were removed from the flask and the volume of the sac content was measured. Samples of the initial amino acid solution and of the sac fluid were analyzed with an automatic amino acid analyzer (Model KLA3, Hitachi Co.). The weight of each sac was determined after drying for 2 h at 110 °C. The rate of accumulation of histidine in serosal fluid during the incubation was calculated in mmoles/g dry wt. of sac per 90 min.

[1] M. SUGAI and I. MATSUDA, Biochim. biophys. Acta *170*, 474 (1968).
[2] E. REICH, Science *143*, 684 (1964).
[3] T. H. WILSON and G. WISEMAN, J. Physiol., Lond. *124*, 414 (1954).