their paper does not contradict the other isomeric structures including those having *o*- and *m*-terphenyl nucleus. The synthetic confirmation of the structure is now in progress.

The toxin B, yellow needles of m.p. 165–170° (decomp.), $C_{23}H_{20}O_2N_2$, also shows cytotoxicity on cultured HeLa cells. The 50% growth inhibitory dose is about 6 µg/ml, and irregularity in cell size and round eosinophilic nucleoli are the morphological changes produced. No remarkable differential supression of ^{3}H-precursors of biopolymers is observed; that is, the incorporation of ^{3}H-thymidine, -uridine and -leucine into the cells treated at 32 µg/ml is below 10% compared to those into the control cells. It also shows an acute toxicity on mice, killing them with doses less than 100 mg/kg body weight by single s.c. injection. It acts rather slowly and caused diffuse hepatotoxic lesion with general jaundice. The chemical investigation on toxin B is also in progress.

Zusammenfassung. Zwei neue Toxine wurden aus den Mycelien und dem Zuchtmedium eines Schimmelpilzes, *Aspergillus candidus*, sowie aus dem experimentell mit demselben Pilz verschimmelten polierten Reis isoliert. Die beiden Substanzen sind sowohl chemisch als auch toxikologisch voneinander völlig verschieden.

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Adsorption to Activated Charcoal and Polarity of Cardenolides and Bufadienolides

For treatment of glycoside intoxication, adsorption of the glycosides to orally applied cholestyramin was recommended[1]. Since cardiac glycosides, except for ouabain, undergo enterohepatric circulation to varying extend, this treatment is suitable not only for oral but also for intravenous glycoside intoxication[1]. HAACKE et al.[2] recently showed adsorption to charcoal of ^{3}H-digitoxin and its metabolites in guinea-pig bile.

The aims of the present study were to establish data on the in vitro adsorption to charcoal of a series of glycosides and derivatives and to solve the question of whether or not the adsorption is influenced by the polarity of these substances.

Material and methods. Digitoxin, lanatoside C and ouabain from E. Merck, Darmstadt; digoxin, β-methyl digoxin and digoxigenin were from Boehringer, Mannheim; proscillaridin, proscillaridin-4′-methyl ether, 3-β-scillarenin and scillarenin-3-β-methyl ether were from Knoll, Ludwigshafen, Germany. Concentrations of the glycosides were determined photometrically (Zeiss PMQ III). For the bufadienolides adsorption in methanol-KOH was measured at 355 nm[3]. With the cardenolides adsorption in H_2SO_4 was measured at 235 nm[4]. Both reactions showed a linear relationship between glycoside concentration and extinction, at least in the tested range of concentrations.

Adsorption of the substances to charcoal. The assays contained 300 µg/ml of the glycoside resp. derivative in 300 µl/ml of ethanol (70%) and 0.025 to 15.0 mg/ml of activated charcoal in aqueous phase. They were incubated for 15 min at 22 °C under continuous shaking. Then the charcoal was sedimented by centrifugation at 4000 *g*. Glycoside concentration in the supernatant was determined and expressed in percent of an assay without charcoal. The results were transferred to semi-logarithmic plots and from this the charcoal concentration was determined which binds 50% of the substance.

Polarity of the substances. 0.8×10^{-5} moles of the substance were shaken mechanically for 30 min in a glass tube containing 2 ml H_2O, 3 ml isopropanol and 5 ml

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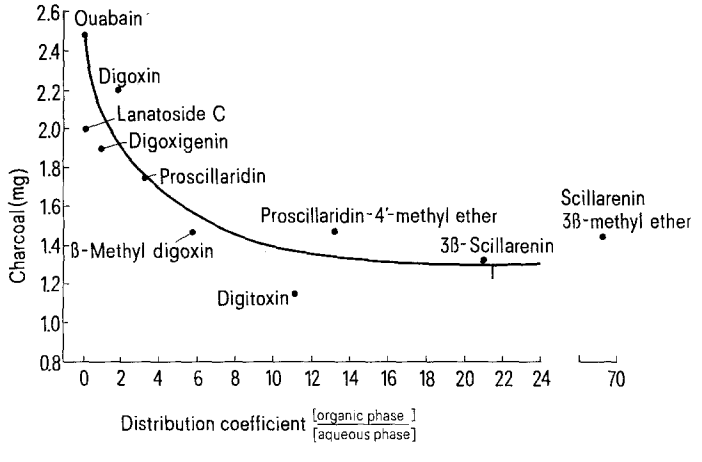

Correlation between polarity of the glycosides/derivatives and charcoal binding. Abscisse: The polarity is given as the quotient of concentrations in organic and aqueous phase of a H_2O, isopropanol, and carbon tetrachloride mixture. Ordinate: mg of activated charcoal needed to bind 50% of 300 µg of the glycosides/derivatives.

carbon tetrachloride[5]. After centrifugation at 2000 *g*, glycoside concentrations were determined in the organic and in the aqueous phases. A quotient Q = organic phase: aqueous phase was calculated.

Results and discussion. The substances tested show a range of Q from 0.004 to 70, indicating great differences in the polarity (Figure). The charcoal concentration needed to obtain half maximal adsorption of the glycosides and derivatives varies from 1.15 to 2.49 mg/ml. It is evident that a correlation exists between polarity and charcoal binding. The higher the polarity, the more charcoal is needed to obtain the same adsorption. With Q increasing > 10 no further increase of the charcoal-glycoside affinity is observed.

The results raise the question of in vivo adsorption of glycosides to charcoal. In the case of glycoside intoxication, charcoal would be a cheep and non-dangerous antidote. On the other hand, a reduction of bioavailability of the cardiac glycosides may occur when a patient on digitalis therapy receives charcoal from other indications.

The stoechiometric relations between glycoside binding and charcoal concentration should not be transferred without criticism to in vivo conditions, as an interference of the glycosides with other substances absorbed to charcoal is most probable.

Zusammenfassung. Die Adsorption von 10 Herzglykosiden und Geninen an Aktivkohle wurde untersucht. Es zeigte sich, dass mit zunehmender Polarität der Substanzen die zur Adsorption benötigten Kohlemengen anwuchsen.

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Prevention by Cystamine of the Rat Liver Polysomal Disaggregation Induced by Carbon Tetrachloride (CCl_4)

Cystamine prevents some biochemical and morphological alterations induced by CCl_4 in the rat liver[1,2]. As suggested by Castro et al.[2], the protective effect of cystamine against CCl_4 intoxication can be explained in 3 ways: 1. cystamine inhibits CCl_4-activation leading to free radicals; 2. cystamine acts as free radical 'trapping agent'; 3. cystamine interacts with the target structures (the membranes of endoplasmic reticulum), in such a way that it shields the unsaturated lipids against the action of free radical.

Since the formation of free radicals from the homolytic scission of CCl_4 is considered the first step in the chain of reactions leading to liver damage[3], it seemed important to study whether cystamine prevents the polysomal damage (disaggregation) produced by CCl_4.

Methods. Male rats of Wistar strain weighing 180–230 g were fasted 12 h before treatment, water was given ad libitum. Cystamine dihydrochloride (Fluka) dissolved in saline, was given per os. Pure CCl_4 was given i.p. in the amount of 250 µl/100 g body wt. Controls were given equal volumes of saline, cystamine and CCl_4 respectively. Rats were sacrificed 30 min after CCl_4 administration. Polyribosomes sedimentation patterns were studied as described by Borghetti et al.[4]; the amount of post-mitochondrial supernatant stratified on a gradient was exactly determined, as suggested by Fleck and Munro[5], by the RNA content.

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Table I. Effect of different doses of cystamine on the polysomal disaggregation caused by CCl_4

Treatment		Polysomes[c]
		Total ribosomes
Control[a]	(5)	0.30 ± 0.02
Cystamine[b] (mg/100 g body wt.)		
5	(5)	0.35 ± 0.01
10	(5)	0.47 ± 0.01
20	(5)	0.57 ± 0.008
40	(5)	0.57 ± 0.03
100	(5)	0.60 ± 0.008

[a]-Control receiving i.p. 250 µl/100 g b.w. of CCl_4 alone. [b]-Cystamine was given po 120 min before CCl_4. Animals were sacrificed 30 min after CCl_4. [c]-Calculated by areas of the polysomal patterns. Mean ± SE. In parentheses number of experiments.

Table II. Protection of cystamine administered at different times prior CCl_4, on CCl_4-polysomal disaggregation.

Cystamine pretreatment[a]		Polysomes[b]
time (h)		Total ribosomes
2	(5)	0.60 ± 0.008
12	(5)	0.57 ± 0.01
16	(5)	0.48 ± 0.007
24	(5)	0.40 ± 0.04
Control	(5)[c]	0.30 ± 0.02

[a]-Cystamine was given po at the dose of 60 mg/100 g body wt. Pretreated animals were sacrificed 30 min after CCl_4 administration. [b]-Calculated by areas of the polysomal patterns. Mean ± SE. [c]-Controls receiving i.p. 250 µl/100 g body wt. of CCl_4. In parentheses number of experiments.