4[9,10], cause two effects. Firstly, capillary damage with extravasation of plasma proteins, as seen in inflammation[11], will lead to concommittant extravasation of drug bound to these proteins and thus cause elevated concentrations of drug in inflamed tissue. Secondly, weak acids show increased lipophilicity in acidic compartments, which may result in increased membrane-concentration[9], and biological activity. Acidic compartments are known to exist in the stomach[9], the kidney[9] and inflamed tissue[11]. Whether these explanations together are sufficient to explain the accumulation and pharmacological activity of acidic NSAID, however, remains a matter of speculation at present[12].

Nevertheless it can be concluded that our observations add a new aspect to the understanding of the mode of action of acidic NSAID. It appears that only acidic drugs which are highly bound to plasma proteins accumulate specifically in inflamed tissue and thus inhibit PG-synthesis in vivo at the site of inflammation. On the other hand, the same physico-chemical characteristics may lead to high membrane concentrations and/or biological activity of these drugs in small intestine, kidney and liver causing the known unwanted side effects in these organs[5]. As a consequence of these observations, it appears almost impossible to dissociate the desired anti-inflammatory action of acidic NSAID from their harmful side effects. Also, since the degree of accumulation in the inflamed tissue may vary from species to species (depending e.g. on the degree of protein binding of therapeutic doses), search for optimal pharmacokinetic behaviour in humans may serve as a useful additional guide line for the development of better anti-inflammatory drugs[13–16].

Zusammenfassung. Es wird gezeigt, dass saure Antiphlogistika im entzündeten Gewebe angereichert werden. Alkalische Strukturverwandte zeigen diesen Effekt nicht. Die Bedeutung dieses Befundes für das Verständnis der Wirkung bekannter und die Entwicklung neuer nicht steroidaler Antiphlogistika wird diskutiert.

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Microsomal Metabolism as a Determinant of Aflatoxin Toxicity

Aflatoxins are a group of toxins elaborated by the fungus *Aspergillus flavus* which are natural contaminants in foods and feeds. They are known to produce acute necrosis, cirrhosis and carcinoma of the liver in a variety of animal species including non-human primates[1–3]. However, there is a marked variation in the response to the toxin between various species of animals[4]. It has not been possible to correlate these differences with differences in the status of the drug metabolizing system[5]. Qualitative changes in the metabolism of the toxin rather than quantitative differences in a single enzyme system may be one of the explanations for the species variations. Examples of such species-specific metabolism have, in fact, been documented[6]. Such a possibility also explains the variation with respect to the zones affected in the livers of different animal species. Aflatoxin liver damage is periportal in the rat, duckling, and monkey, midzonal in the rabbit and centrilobular in the pig, guinea-pig and dog[4]. These observations, therefore, indicate that aflatoxin is metabolically handled differently by different species of animals.

In spite of intensive and extensive studies, there is, as yet, no complete understanding regarding the site and nature of aflatoxin metabolism. It has been suggested that aflatoxin requires to be activated for its acute toxicity[5]. It has also been suggested that its toxicity is dependent upon its being inactivated[7]. Similarly, divergent speculations have been made with respect to its carcinogenic potential. On the basis of reported data from controlled studies in rats subjected to various experimental procedures, an attempt has been made here to explain the metabolic basis for aflatoxin toxicity.

Microsomal drug metabolism is relatively ineffective in young animals[8] and in protein depleted animals[9,10]. In both these situations, susceptibility to acute aflatoxin toxicity is increased[11,12]. Administration of DDT is associated with increased rate of drug metabolism[13] and such treatment results in protection from aflatoxin-induced liver injury[14]. The behaviour of carbon-tetrachloride in these conditions is exactly opposite to that of aflatoxin[7,15]. Such a reciprocal behaviour of these two toxins is also observed from the zones affected in the rat liver during their acute toxicity. In acute aflatoxin toxicity, the lesion is periportal, while in carbon tetrachloride, it is centrilobular. It is usual for the toxins requiring

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activation to result in centrilobular lesion while those detoxified directly produce periportal damage. Those toxins which result in detoxification through intermediates which are more toxic than the parent compound and its ultimate metabolites, produce midzonal necrosis[16].

There is now considerable evidence to show that carbon tetrachloride needs to be activated by microsomal enzyme system in the liver before becoming toxic[17,18]. Administration of glucocorticoids increases toxicity of carbon tetrachloride[19] indicating an effective stimulation of drug metabolism during cortisone therapy. Such a treatment in acute aflatoxicosis, on the other hand, results in protection against liver damage[20].

Pregnancy depresses metabolism of various foreign compounds[21] and increases susceptibility to acute aflatoxin damage[22]. Similarly, vitamin A deficiency has been shown to depress the metabolism of a variety of drugs[23] and has been demonstrated to increase the susceptibility to acute aflatoxin damage[24]. A more direct and clinching evidence for the role of metabolism of aflatoxin as a factor determining its toxicity, is provided by the studies on hypophysectomized rats. Hypophysectomy reduced the metabolism of aminopyrin and of aflatoxin[25]. There was a decreased formation of the metabolic products of aflatoxin produced by the microsomal enzymes[25]. However, the acute susceptibility to aflatoxin was greatly increased by hypophysectomy[26].

These data therefore strongly suggest that an effective drug metabolizing system protects the animal from the acute effects of aflatoxin. On the other hand, this effective drug metabolizing system would appear to have exactly the opposite effect on the carcinogenic potential of aflatoxin. Protein deficiency and vitamin A deficiency increase the acute susceptibility, but reduce the incidence of hepatomas[27,28]. Again, hypophysectomy markedly reduces the LD_{50}[26], but is not compatible with the production of liver tumours[29]. These results indicate that during the metabolism of aflatoxin in the body, compounds which are less toxic with respect to acute lesions but more effective in hepatocarcinogenesis are formed. This possibility is supported by the observation that administration of a single large dose of 500 µg of the toxin results in the development of hepatomas in 50% of rats[30], while a smaller dose of less than 100 µg when administered in multiple divided doses over a long period results in liver cancer in 100% of animals[31].

The observation that simultaneous administration of phenobarbitone and aflatoxin reduces the incidence of aflatoxin-induced hepatic tumours, however, suggests that aflatoxin is detoxified with respect to its carcinogenic potential also[32]. This could have resulted from the competitive inhibition of the metabolism of the carcinogen administered while the drug-metabolizing system is handling the phenobartitone.

It appears, therefore, that the microsomal drug metabolizing enzymes are rate limiting in the degradation of aflatoxin in the rat liver. It is likely that these 'detoxified' products are more carcinogenic to the liver. Further studies are necessary to substantiate this hypothesis.

Summary. The evidences to incriminate the possible role of microsomal drug metabolizing system in aflatoxin toxicity in rat have been reviewed. It is suggested that an effective drug metabolism results in decreased acute toxicity and the products of metabolism are more carcinogenic than the native toxin to the rat liver.

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Benzodiazepines: a Comparison of their Effects in Mice on the Magnitude of the Palmar Skin Conductivity Response and on Pentylenetetrazole-Induced Seizures

We have previously demonstrated[1] that a wide variety of CNS depressants inhibit the palmar skin conductivity response in mice (PSCR-test). This test might thus be used to study non-specific sedative activity. On the other hand, the blocking of pentylenetetrazole-induced seizures (anti PIS = APIS-test) at very low doses is characteristic of benzodiazepines and might be related to the anti-epileptic and anxiolytic actions of these drugs. Consequently, a systematic comparison between the two methods deserves investigation.

Methods. Swiss Or1 male mice (body weight 18 to 25 g), randomized into batches of 10, were used for both methods. PSCR- and APIS-tests were carried out following[1] and[2] respectively. The PSCR-test is based on the increase of the palmar skin conductivity level in response to a photostimulus; 13 benzodiazepines and 2 other drugs (cf. Table) were injected i.p. 20 min before the i.p. administration of 125 mg/kg of pentylenetetrazole in the APIS-test or before the 2nd photostimulus in the PSCR-test. Each batch of mice was dosed in such a way as to achieve a logarithmic increase of dosage over the whole experiment. For each drug tested the following values were calculated: the ED 50 (following[3]) preventing tonic convulsions and

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