Zusammenfassung. Intakte Exemplare von *Sepia officinalis* zeigen nach Drehung um alle drei Hauptachsen Augen- und Kopfbewegungen vom Typus des «Nach-Nystagmus». Diese postrotatorischen Reflexe können nicht optisch ausgelöst sein; sie beweisen vielmehr das Vorhandensein eines echten Rotationssinnes. Es wird angenommen, dass dieser auch hier seinen Sitz in den Statocysten mit ihren hochdifferenzierten Cristae hat.

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Phenobarbital Markedly Increases Liver Weight in Walker Carcinosarcoma Bearing Rats

The present work is founded on some recent observations by CONNEY et al.[1]. These authors demonstrated an increase of liver size in weanling rats after a four days treatment with phenobarbital. The increase of liver size was combined with an increased liver protein content and the increased activity of some liver microsomal drug metabolizing enzymes[1,2]. According to our observations, these increases are less intense in adult rats[3,4].

Some observations of ANNAU et al. show that in tumour bearing rats and mice there is an increase of liver weight and of mitotic activity[5]. Similar results were observed in tumour-bearing mice by MALMGREN; the same author could also observe an increase of mitotic activity by simple injection of tumour homogenate[6]. PASCHKIS et al. observed that the liver regeneration was enhanced in the presence of a growing tumour[7]. It appeared evident that in the tumour-bearing rats there is some growth promoting factor(s). Therefore we supposed that tumour-bearing rats might be more sensitive to the phenobarbital effect on the liver enlargement, like growing rats.

In fact, in the present work evidence is given that adult rats bearing Walker carcinosarcoma 256 have a remarkable sensitivity toward liver enlargement induced by phenobarbital.

Male rats of the Sprague-Dawley strain weighing about 200 g were used. The animals were inoculated subcutaneously with Walker carcinosarcoma 256 in the lateral abdominal region. After 18 days, rats bearing well developed tumours were selected and a group of tumour-bearing rats and of controls were treated with phenobarbital (90 mg/kg, i.p.). 48 h later all the rats were killed and the whole liver was weighed. The determination of liver protein, DNA and RNA was carried out, using respectively the methods of LOWRY et al. and SCHNEIDER[8,9].

The results are given in Tables I and II.

Phenobarbital produces only a 5% increase of the liver weight in normal rats, while in the tumour-bearing rats the increase is 28% ($P < 0.001$). As regards the ratio of liver weight for 100 g of body weight, there is a 26% increase in the tumour-bearing rats ($P < 0.001$) against a 6% only in normal rats. Protein, DNA and RNA con-

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Tab. I. Effect of phenobarbital on liver weight of normal and Walker carcinosarcoma 256 bearing rats

	Number of rats	Phenobarbital treatments	Body weight (g)	Tumour weight (g)	Liver weight (g)	Liver weight × 100 / Body weight
(1) Normal rats	14	—	289 ± 3.8		13.0 ± 0.43	4.52 ± 0.13
(2) Normal rats	14	+	284 ± 4.2		13.6 ± 0.41	4.79 ± 0.16
(3) Tumour bearing rats	13	—	286 ± 4.8	17.5 ± 2.4	13.6 ± 0.46[a]	4.76 ± 0.19[a]
(4) Tumour bearing rats	13	+	292 ± 4.5	19.4 ± 2.5	17.4 ± 0.58[a]	5.98 ± 0.24[a]

[a] Significance (4–3) $P < 0.001$

Tab. II. Effect of phenobarbital on protein, DNA and RNA concentration in livers of normal and Walker carcinosarcoma 256 bearing rats

	Phenobarbital treatment	Number of rats	Protein content (mg/g liver)	DNA content (mg/g liver)	RNA content (mg/g liver)
(1) Normal rats	—	9	167 ± 2.5	2.02 ± 0.08	9.80 ± 0.36
(2) Normal rats	+	9	174 ± 3.2	1.91 ± 0.10	11.28 ± 0.58
(3) Tumour bearing rats	—	8	163 ± 3.0	2.15 ± 0.12	10.12 ± 0.60
(4) Tumour bearing rats	+	8	165 ± 3.8	1.98 ± 0.10	10.77 ± 0.51

All differences are statistically not significant.

tents per g of liver of the phenobarbital-treated rats, the tumour-bearing rats or the tumour-bearing phenobarbital-treated rats is not significantly changed.

These results indicate that a treatment with phenobarbital produces a marked liver weight increase in only 48 h. This enlargement is a real hypertrophy, since at the same time there is an increase of total proteins, DNA and RNA.

The phenobarbital activity on tumour-bearing rats can be compared with that observed in weanling rats. It is probable that some growth-promoting factor(s) from the tumour is responsible for the greater sensitivity toward the phenobarbital action on the liver. Our previous results show that there is an increased sensitivity toward phenobarbital in partial hepatectomized or in pregnant rats, and in both cases the presence of growth-promoting factor(s) has been reported [10–12].

Riassunto. Gli autori hanno dimostrato che nei ratti portatori di tumore di Walker (carcinosarcoma 256), un trattamento eseguito 48 h prima con phenobarbital produce un aumento del rapporto peso fegato/peso corporeo pari al 26%, mentre esso è soltanto del 6% dei ratti normali. Tale fatto si accompagna ad aumento di contenuto totale di proteine, di RNA e di DNA.

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Decreased Activities of Liver Microsomal Drug-Metabolizing Enzymes in the Rats Bearing Walker Carcinosarcoma

There are many reports concerning systemic effects of malignant tumours on host animals[1]. NAKAHARA and FUKUOKA, NAKAGAWA et al. and KAMPSCHMIDT and McCOY reported a marked decrease of liver catalase activity in the tumour-bearing animals, and also that injection of the tumour extracts produced a depression of liver catalase activity[2–4]. On the other hand, GREENE and HAVEN have shown an increase in cytochrome oxidase activity in liver homogenates or mitochondria from livers of rats bearing Walker carcinosarcoma 256[5].

In recent years, the importance of oxidative processes in liver microsomes for drug metabolism have been widely recognized. The microsomal enzymes have a common requirement of reduced triphosphopyridinenucleotide (TPNH) and oxygen. The oxidation of TPNH in the microsomes by TPNH oxidase and oxygen may involve a formation of organic peroxide, but the mechanism of action is not yet clear[6].

In the present work, evidence is given that rats bearing Walker carcinosarcoma 256 show a depressed activity of the liver microsomal enzymes responsible for metabolism of hexobarbital, strychnine and meprobamate.

Male rats of the Sprague-Dawley strain, weighing about 200 g were used. Walker carcinosarcoma 256 were inoculated subcutaneously in the lateral abdominal region. The animals were killed 22 days after the inoculation.

The enzyme activities were determined by measuring the metabolized hexobarbital, strychnine and meprobamate by a liver microsomal preparation during incubation for 1 h. The liver was immediately removed and homogenized in 3 parts of isotonic KCl (1.15%) with a Potter-Elvehjem type homogenizer. The nuclei and mitochondria were sedimented by centrifugation of the homogenate at 8500 *g* for 15 min. The incubation mixture (5.0 ml) contained 3 ml of the microsome-containing supernatant, 0.1 ml of 20 μmole glucose-6-phosphate, 0.4 μmole TPN, 50 μmole nicotinamide and 75 μmole $MgCl_2$ and 2 *M* KCl and more 1.3 ml of 0.2 *M* sodium phosphate buffer pH 7.4 (pH 8.2 for strychnine) and 0.2 ml of the substrate. Final concentrations of hexobarbital, strychnine and meprobamate were 6×10^{-4}, 2×10^{-4} and $3 \times 10^{-4} M$ respectively.

The determinations of hexobarbital, strychnine and meprobamate were carried out according to the methods of AXELROD et al., KATO et al., and HOFFMAN and LUDWIG respectively with little modification[7–9]. The results are shown in the Table.

The metabolisms of hexobarbital, strychnine and meprobamate were markedly decreased in liver of Walker carcinosarcoma-bearing rats. Hexobarbital metabolism was decreased 53%. In preliminary experiments, we could not demonstrate the presence of an inhibitor in the liver of the tumour-bearing rats. Similarly, decreased enzyme activity was also observed by us in rats bearing Flexner-Jobling carcinosarcoma even of less intense degree.

The mechanism which produced the decreased activities of the microsomal drug metabolizing enzymes is not yet clear. It is probable that as toxohormone produces a decrease in the activity of liver catalase, some factors elaborated by the tumour cell may act directly or indirectly on the liver microsomes to produce a decrease of the oxidative processes. We also observed a decreased activity of microsomal TPNH oxidase in the tumour-bearing rats, but according to GILLETTE et al.[6], the production of H_2O_2 and its decomposition by catalase are not connected with the process of drug metabolisms in liver

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