Extracts from dried skins (149 skins weighing 430 g) were fractionated as described by DEULOFEU and DUPRAT[4b]. After extraction of the bufotenine, the barium ions were precipitated as sulfate, the remaining solution brought to pH 6 and concentrated to 50 ml. This concentrate on paper chromatography[7], gave spots corresponding to bufotenine, dehydrobufotenine and 5-hydroxytryptamine. It was then saturated with ammonium sulfate and extracted with *n*-butanol. The 5-hydroxytryptamine in the butanol phase was precipitated by addition of 5-nitrobarbituric acid, and crystalline 5-hydroxytryptamine was prepared following the usual procedure[8]. Material recrystallized from acetone-water had m.p. 207–210° (Kofler), undepressed when mixed with an authentic sample, Rf 0.12 (*n*-butanol saturated with *N* hydrochloric acid) and 0.43 (*n*-butanol : acetic acid : water; 4:1:5).

The presence of 5-hydroxytryptamine was also detected by paper chromatography in a sample of the dried secretion of the paratoid glands of the same species of toad, kindly given to us by Prof. J. A. IZQUIERDO.

*Zusammenfassung.* 5-Hydroxytryptamin wurde aus der Haut der in Argentinien häufigsten Krötenart *B. arenarum* Hensel isoliert und in trockenen Paratoidsekreten chromatographisch nachgewiesen.

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## The Uptake of Pyrithiamine by Cerebral Tissue

The only *in vitro* effect of pyrithiamine (PT) is the inhibition of thiamine (T) phosphorylation[1–4] by thiaminokinase, which is considered to explain the antivitamin mechanism of PT[5].

*In vivo* a particular action of PT was shown by DE CARO et al. in rats[6–8] and mice[9,10]. The oral or intraperitoneal administration of the antivitamin, at doses ranging from 0.2 to 6 mg, alone or together with small amounts of T, as a single dose or repeated for 4–6 days, always caused an earlier and deeper decrease of T (total and cocarboxylase) content in brain than in other organs.

This must be considered a distinctive feature of PT athiaminosis as compared with nutritional athiaminosis, where the brain is known to preserve its T content for a longer time than other organs[11–13].

As a consequence of this action of PT on cerebral tissue, most animals show, in 10–15 days, the typical neurological signs of beri-beri, which are difficult to produce in rodents, even in extreme nutritional T deficiency. These results have been confirmed by KOEDAM in pigeons[5] and mice[14]. It is clear that knowledge of the PT tissue levels after PT administration could clarify its apparent specificity in lowering the total T (esterified T) content of the brain. In fact, the hypothesis can be made that PT accumulates preferably in the cerebral tissue, displacing T and thus causing the early onset of the beri-beri symptoms.

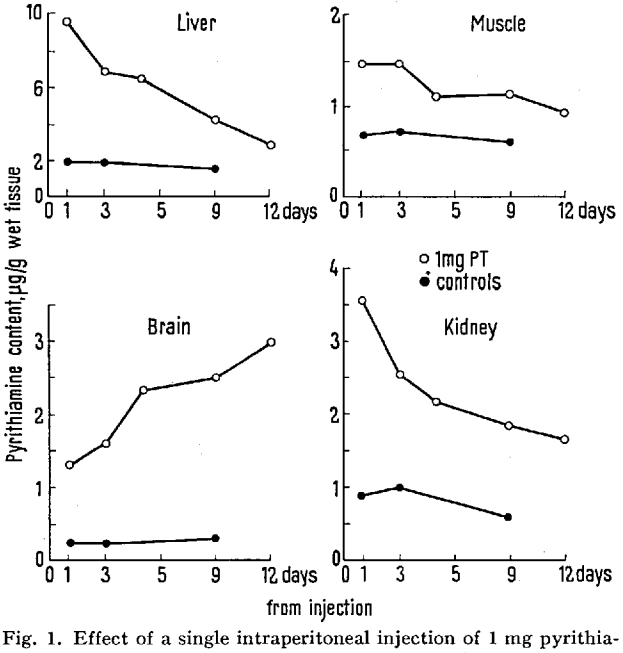


Fig. 1. Effect of a single intraperitoneal injection of 1 mg pyrithiamine (PT) on the PT-content in various organs. Rats reared on a thiamine-deficient diet. Each point is the average of 7 determinations.

The specific microfluorimetric method of RINDI and PERRI[15,16] for the determination of PT in presence of T enabled us to verify this hypothesis. The experimental conditions chosen were quite similar to those in which we found the earliest and the most striking decrease of the cerebral T content[8]. A single dose of 1 mg of PT (Calif. Biochem. Corp., Los Angeles), dissolved in 0.2 ml of saline, was injected intraperitoneally in male albino rats (body weight 80–90 g). The animals, reared on standard T-deficient diet[8], were sacrificed at different time intervals after injection. In brain, liver, muscle, and kidney of treated and control (untreated) rats the PT content was determined. The results are summarized in Figure 1. As is shown, the PT content found in all organs except brain 24 h after injection, was approximately that of T usually present. Successively, the PT content decreased in liver,

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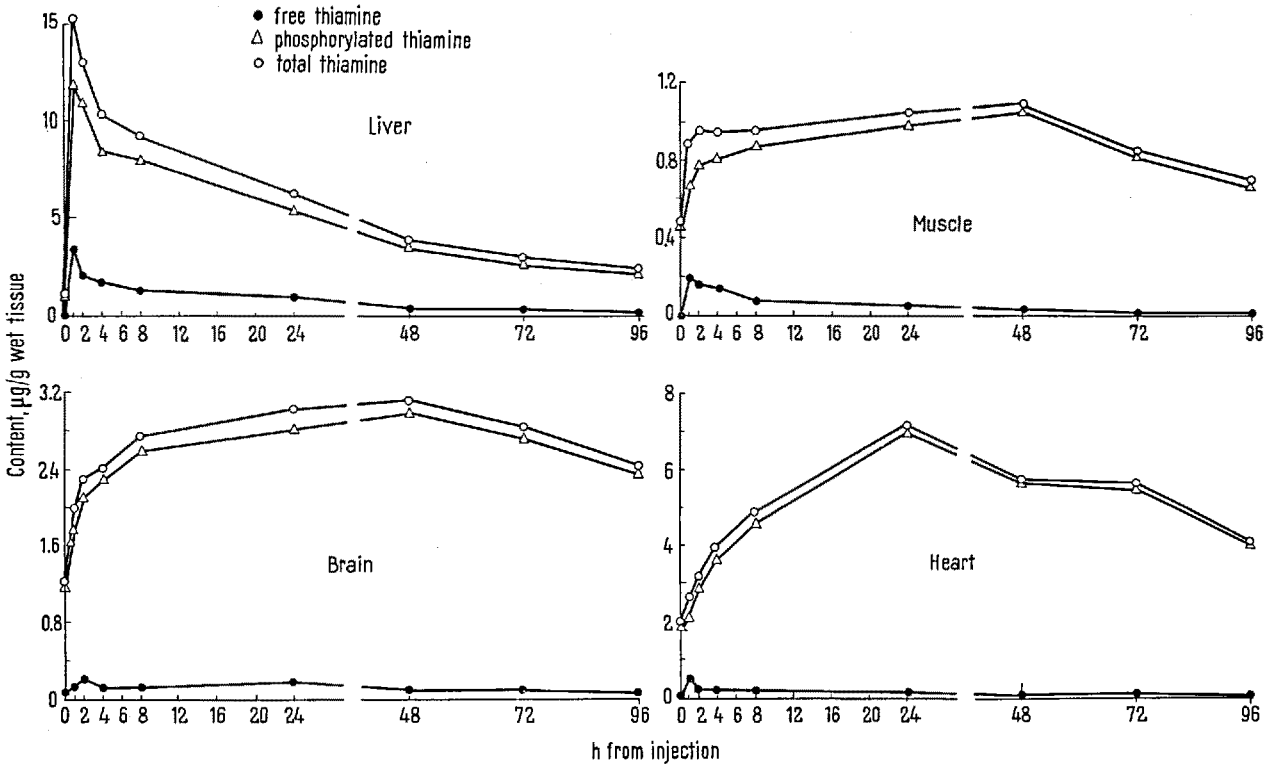


Fig. 2. Thiamine content in organs of thiamine-deficient rats (15 days), injected intraperitoneally with 1 mg of thiamine. Each point is the average of 7 determinations.

muscle and kidney, but it gradually increased in brain, reaching a level very close to that of total T usually present. Of course, the uptake of PT by the tissues caused a shift of T, of which the urinary excretion was greatly increased, as previously shown[7,8].

Thus the distinctive feature of brain towards PT, as compared with other tissues, lies in the accumulation of the antivitamin.

In this connection it is suggestive that a single i.p. injection of T (1 mg) in T deficient rats (15 days) caused in the brain a steady increase of vitamin (determined by Thiochrome method[17]), probably at the expense of liver stores, which rapidly depleted after a quick initial rise (Figure 2). Heart and skeletal muscle behaved similarly to brain.

Therefore, the selective increase of PT in cerebral tissue may be interpreted in terms of structural analogy. The specific accumulation of PT in brain results in a shift of total T/PT ratio towards a greater efficacy of the antivitamin, causing a sort of organ athiaminosis with consequent profound neurological disturbances.

The present results strongly support the validity of the hypothesis of a preferential deposition of PT in cerebral tissue[18].

*Riassunto.* Il livello di PT che si instaura nel fegato, rene e muscolo di ratti iniettati con 1 mg di PT in peritoneo e tenuti a dieta carente di T diminuisce continuamente nel giro di 12 giorni, mentre aumenta gradatamente nel cervello. Ciò può spiegare la particolare efficacia della PT nell'abbassare selettivamente il contenuto in T totale nel tessuto cerebrale.

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[18] We are deeply grateful to Prof. R. R. Williams of the Research Corporation, New York, for the generous gift of the Farraud Fluorimeter used in this research.

## Neoplasms in *Lumbricus terrestris* L.

Few examples of induced tumors in invertebrates have been described in the literature. An excellent review of the subject is that of Scharrer and Lochhead[1]. Many of these 'tumors' have been questioned as to their manifest equivalence with mammalian neoplasms, and the lesions in this note present no exception. Huxley[2] has commented on the unresolved and perplexing problem involving the phylogenetic comparisons of tumors.

During the course of work with Lumbricus, using a variety of chemical carcinogens and X-irradiation, several neoplastic appearing lesions were observed. One of the most commonly found changes resembled that of a granular cell myoblastoma. One such lesion is shown in Figure 5, which occurred on the prostomium of a worm

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