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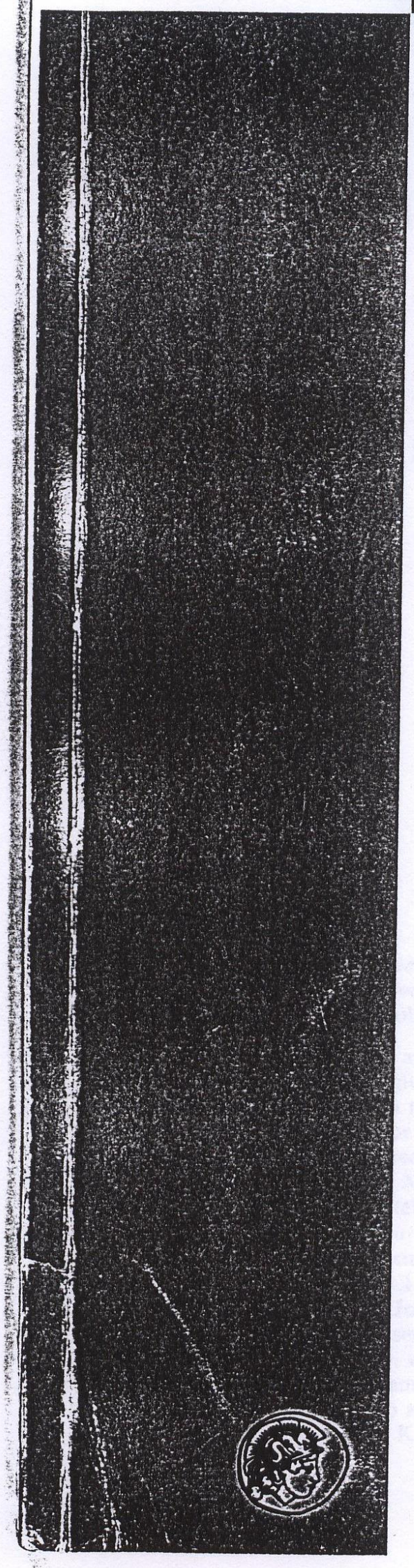

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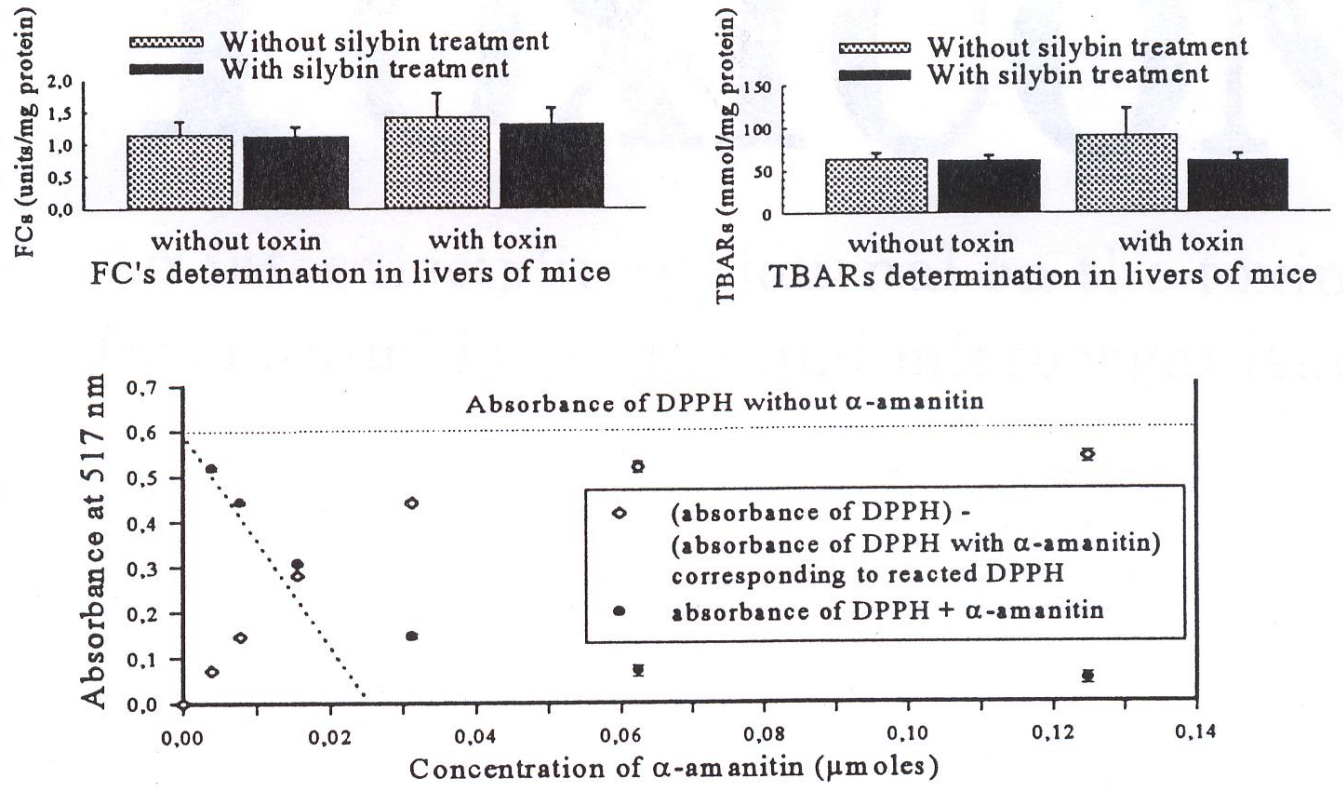
Mode of action of amatoxins: involvement of a peroxidative process in the hepatotoxicity of α-amanitin. Preliminary results. D. Michelot,[1] A. M. Zheleva[2] and Z. D. Zhelev[3] ([1]Laboratoire de Chimie du Muséum National d'Histoire Naturelle, Interactions Fongiques et Microbiennes, URA 401 CNRS, 63, Rue Buffon, F-75005 Paris, France; and [2]Department of Chemistry and Biochemistry, and [3]Department of Molecular Biology and Immunology, 6000 Stara Zagora, Bulgaria).

Membrane peroxidation in hepatocytes produced by the mushroom toxin α-amanitin was investigated *in vivo*, together with its reaction with 1,1-diphenyl-2-picrylhydrazyle (DPPH), a stable free radical. Assays characteristic of the products, TBARs (thiobarbituric acid reactive compounds) and FCs (fluorescent compounds), demonstrate that a peroxidative process occurs in the livers of poisoned mice. Previous administration of silybin, an efficient hepatoprotectant known to react with DPPH, protects from the noxious effects of α-amanitin. By means of the latter substance, the toxin probably acts via a radical-type reaction. The investigation of the reactivity of DPPH toward the toxin demonstrates that three molecules react with one toxin molecule. The reactive site is most likely close to the tryptathionin group. Taking into account the literature, the inhibition of RNA polymerase type II, a mode of action commonly admitted, should not be the sole mechanism implicated, and additional peroxidative processes should take place during the severe hepatolysis.



Absorbance of DPPH at 517 nm with α-amanitin.

Bidirectional modulation of evoked-ACh release from Torpedo *synaptosomes by the spider toxins argiotoxin* 636 *and Joro.* Y. Morot Gaudry-Talarmain (Departement de Neurochimie, Laboratoire de Neurobiologie Cellulaire et Moléculaire, CNRS, 91198 Gif-sur-Yvette, France).

Vyas and Bradford (1987) and Israël *et al.* (1993) showed that *Torpedo* synaptosomes co-release acetylcholine and glutamate under some conditions of stimulation. These results raise the possibility that glutamate release might modulate acetylcholine release through a feedback mechanism using glutamate presynaptic autoreceptors and/or related protein targets. In the present study, we addressed this question using spider toxins as pharmacological tools. Arylalkylamine toxins such as argiotoxin 636 from *Argiope lobata* and Joro spider toxin from *Nephila clavata* are low mol. wt toxins, considered to be uncompetitive ligands of glutamate receptors in invertebrates and mammalian central synapses (review in Jackson and Usherwood, 1988). ACh release was measured by the chemiluminescent choline oxidase assay of Israël and Lesbats (1981). The study of the effect of toxins at various concentrations (1–20 μM, 7–10 min in the release medium) showed an increasing stimulation of KCl-evoked ACh release such that an increase to 200% was obtained at 4 μM argiotoxin 636 and 165% 4 μM with Joro toxin. Both toxins were inhibitory for higher concentrations (10 μM and higher). ACh release induced by inserting in the presynaptic membrane the calcium ionophore A23187 (4 μM), which leads to passive calcium entry, was moderately stimulated by both of the toxins at 4–6 μM. Inhibition of the release developed at higher concentrations. From the present results, it is suggested that argiotoxin 636 and Joro spider toxin are able: (1) to activate and then inhibit the coupling between voltage-sensitive calcium entry and ACh release observed after KCl depolarization; and (2) to stimulate in the μM range the latest phase of ACh release which concerns