Table II. In vivo incorporation of orotate in rat liver nuclear RNA following an i.p. injection of 30 mg/kg body wt. of CQ.

Animal[a]	Specific activity dpm/100 mg RNA	%
Control (2)	133.300 ± 8.700[b]	100
Treated (2)	129.750 ± 12.250[b]	97

100 μCi of orotate ^3H (17 Ci/mmole) was injected i.v. 4 h after drug. The animals were sacrified 20 min later. RNA was extracted from purified nuclei and the samples were processed for counting according FLOYD, OKAMURA and BUSCH[13]. [a] Number of animals in parentheses. [b] ± Mean deviation.

Table III. Incorporation capacity of liver postmitochondrial fraction of CQ-treated rats after a bilateral adrenalectomy[a]

Dose CQ mg/kg body wt.	Time (h after CQ-treatment)	Incorporation (% relative to adrenalectomized controls)
15 (4)	24	110
30 (2)	4	115

[a] Adrenalectomy was performed 1 week prior to treatment. The incorporating assay was carried out as described in Table I. Number of animals in parenthesis.

This stimulation cannot be ascribed to the interaction of the drug with the protein synthesis components, because CQ, when it is added to a cell-free assay system, produces a definite inhibition.

In order to explore the possibility that CQ mediates the increase of protein synthesis by increasing the rate of mRNA transcription, the labelling of nuclear RNA was determined. As shown in Table II, no difference was observed in the amount of the incorporated uridine between the treated and control animals. This result leads, therefore, to the assumption that the stimulation by CQ of protein biosynthesis in rat liver occurs at the post-transcriptional level.

Several experimental conditions, such as drugs, ionizing radiations, and irritative stimuli, are able to enhance the incorporation of amino acids into rat liver proteins. All these conditions, however, operate in an indirect way, by means of a stimulation of the secretory activity of the adrenal cortex, as demonstrated by the failure of such effects in the adrenalectomized animals.

Drugs, in particular, show a biphasic effect. Initially they produce an inhibition owing to their toxicity, afterwards the liver protein biosynthesis increases because of the higher level of glucocorticoid hormones induced by drugs. With CQ, however, this effect appears within 30 min after the injection and it is not preceded by an inhibition. We also observed that the incorporating ability of the postmitochondrial supernatants from rats subjected to a bilateral adrenalectomy is not depressed within the first 24 h after CQ treatment and a stimulation is still appreciated even at lower dosage of the drug (Table III). On the basis of these results it is conceivable that CQ or its metabolite(s) acts directly on liver hepatocyte rather than promoting an indirect response mediated by the increase of the glucocorticoids.

It has been shown that CQ is selectively concentrated into the liver lysosomes[8,9] and that its presence stabilizes the lysosomal membranes and retards the release of the enzymes from the lysosomes[10]. With regard to the effect of CQ on liver protein biosynthesis, it must be recalled that the lysosome may be involved in the regulation of this process by controlling the release or the activities of its nucleolytic and proteolytic enzymes, condition that is regarded as affecting the rate of mRNA degradation. This action on the permeability of the lysosomal membrane, which has been demonstrated for CQ as well as for the glucocorticoid hormones[11], may therefore play a role in the regulatory mechanism of protein biosynthesis.

Beside this property, both CQ and glucocorticoids display some anti-inflammatory and anti-rheumatic activities and both are able to stimulate the protein biosynthesis in rat liver. While this relationship is conceivably fortuitous, the possibility is suggested that their biological affinities are the expression of a convergent action upon the same target.

Riassunto. L'inoculazione endoperitoneale di Clorochina, a dosi comprese tra 15 e 45 mg/kg di peso corporeo determina una stimolazione precoce delle sintesi proteiche nel fegato di ratto. Questa azione non é correlata ad un aumento della sintesi di RNA, né appare conseguente ad una stimolazione corticosurrenalica. L'effetto osservato é brevemente discusso sulla base delle note interazioni tra lisosomi e Clorochina.

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Fate of Internal Doses of DDT

It has been suggested[1-3] that the insect haemolymph doesn't play a significant role in the transport of topical doses of chlorinated hydrocarbon insecticides to their site of action. The present investigation deals with the role of insect circulation in the transport of injected doses of DDT.

WHO susceptible strain of houseflies was used. LC_{50} and LC_{90} for males averaging 16 mg in weight were 1 and 2.5 μg of technical DDT, respectively. 2.5 μg of a mixture of C^{14} and carrier DDT (activity 700 ± 100 cps) in 0.1 μl acetone were injected deep into the thorax between the

scutellum and postnotum. 0.3 mm × 3/4″ hypodermic needles with a short sharp point were used. And, although injection was made with great care, activity measurements made immediately after injection suggested that not more than 20% of the activity was recovered from the interior of the thorax while the remaining 80% was recovered from the thoracic cuticle. This is due to the spontaneous uptake of insecticide by the cuticle from the tip of the needle at the start of injection. This difficulty appears to have been overcome in the technique of GEROLT[3] in which insecticide crystals were introduced into the haemocoele by means of a metal loop through an incision made in the ventral abdomen. The following technique however, is thought to be more satisfactory: The DDT solution from a 100 µl Hamilton Syringe mounted on a gear-controlled micrometer head is brought to the tip of the needle. It is then withdrawn by a volume of 0.1 µl. The tip of the needle is wiped clean of insecticide with tissue paper wetted with acetone. The needle is then inserted and injection of the 0.1 µl of air followed by the required dose is made. In this method, varying amounts of insecticide not exceeding 10% of the internal dose can nevertheless be recovered from the thoracic cuticle immediately after injection. It is certain that the latter resulted from contact of the cuticle with the tip of needle when the latter was withdrawn. Symptoms of poisoning appeared 15 min after injection and, no doubt, toxicity was caused by the internal rather than the topical dose.

Fate of Intra-thoracic DDT. Flies were killed instantly by placing on Cardic at 0 and 6 h after injection. They were separated into heads, thoraces and abdomens and these were homogenized and assayed for activity.

DDT poisoning leads to a considerable loss of body fluid through vomiting and anal excretion. It was therefore difficult to collect blood samples exceeding 0.05 µl from anyone fly. A total of 0.7 µl of blood collected from a large number of flies at various stages of poisoning showed no traces of radio-activity. Frozen sections of thoraces 2 h after injection were autoradiographed. These show that the injected DDT was largely localised at the site of injection from which gradual uptake by the surrounding thoracic muscles takes place. Uniform distribution indicating the exhaustion of the reservoir of insecticide was not yet reached in 12-h-old material. In all sections studied there was a concentration gradient of insecticide across the thorax.

Thoraces 9 h after injection were cut with scissors and homogenized in carrier-saturated 0.35 *M* sucrose. Cuticle and debris were separated by straining through a gauze pad. The remainder was fractionated in an MSC Model 18 ultracentrifuge into myofibrils (150–200 rpm), nuclei (1,000 rpm), mitochondria (6,000 rpm), microsomes (18,000) and supernatant. These fractions showed the following percentage activities, respectively: 90, 2, 3, 3 and 2%.

The use of ion exchange resin in the determination of blood volume in houseflies[4] showed that exchange equilibrium was reached within seconds, indicating that a whole circulation of the haemolymph is completed in a very short time. One would expect that the injected DDT will form a stable suspension and that activity in all parts of the body will be detected shortly after injection. Our results, however, indicate that an intrathoracic dose of DDT of the order of 2.5 µg becomes localised and that it diffuses slowly into the neighbouring thoracic muscles and nerves setting up a concentration gradiant across the thorax. This results in the increase in the concentration of DDT in the thoracic cuticle of older material as indicated by the results in the experiment, in which the DDT in the thoracic cuticle rose from 10 to 30% in a few hours. The concentration gradient will no doubt extend from the thoracic to both cephalic and abdominal cuticle accounting for the rise in their DDT content. It appears, therefore, that the housefly haemolymph doesn't play a significant role in the transport of internal doses of DDT, perhaps as a result of a response mechanism whereby substances foreign to the circulation are confined locally. The results of the fractionation experiment suggest that DDT toxicity can be exaggerated by the remarkable affinity of the insecticide to myofibrils. Its absence from supernatant and microsomes is expected since this strain lacks both aldrin epoxidase and DDT-ase.

Résumé. Des doses de C^{14}DDT de l'ordre de 2.5 γ injectées dans le thorax de mouches domestiques – témoins restèrent localisées. Des auto-radiographes du thorax montrent que la courbe de concentration de l'insecticide décroît vers la périphérie. On peut donc présumer que chez l'insecte la circulation joue un rôle mineur dans le transport du DDT vers son site d'action.

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Time (h)	Activity (%)				
	Thoracic cuticle	Thoracic muscles	Abdominal cuticle	Internal viscera	Head
0	10	87	2	0	1
6	30	60	7	7	1.5

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Influence of Temperature and Seasons on H^3-Norepinephrine Uptake by Isolated Strip Ventricle of Frog

The order of potency of a group of sympathomimetic amines in the frog heart has been found to be isoproterenol > epinephrine > norepinephrine (NICKERSON and NOMAGUCHI[1]; LAND and HOWARD[2]). ERLIJ et al.[3] reported that isoproterenol is 10,000 times more potent than epinephrine. The results of a study (SANCHEZ-GARCIA et al.) on isolated ventricle of the frog indicated that isoproterenol was approximately 10 times more potent than epinephrine. The discrepancy in these results might be attributed to changes on the uptake system of catechol-