

## Effect of juvenile hormone analogues on the eggs

Treatment	Time of JHA treatment in days after oviposition	No. of eggs treated	Larvae surviving to 1st instar		Larvae surviving to 2nd instar		Larvae surviving to 3rd instar		Larvae surviving to 4th instar	
			No.	%	No.	%	No.	%	No.	%
Ro-84314	0	169	19	11.2	0	0	0	0	0	0
	1	238	66	27.7	0	0	0	0	0	0
	2	197	170	86.3	118	59.9	115	58.4	105	53.3
	3	162	145	89.5	119	73.5	111	68.5	107	66.0
Ro-69550	0	196	0	0	0	0	0	0	0	0
	1	143	120	83.9	102	71.3	90	62.9	86	60.1
	2	220	199	90.5	142	64.6	122	55.4	118	53.6
	3	216	174	80.6	141	65.3	124	57.4	123	56.9
Law's Mimic	0	205	40	19.5	5	2.4	5	2.4	5	2.4
	1	312	88	28.2	60	19.2	47	15.1	43	13.8
	2	249	161	64.7	156	62.6	153	61.4	139	55.8
	3	180	145	80.6	139	77.2	139	77.2	133	73.9
Acetone	0	225	203	90.2	199	88.4	195	86.7	194	86.2
	1	206	199	96.6	192	93.2	183	88.8	179	86.9
	2	202	196	97.0	187	92.6	187	92.6	182	90.0
	3	152	146	96.1	133	87.5	130	85.5	126	82.9

at oviposition or 1 day later. This is consistent with previous work on other insects and suggests that the 1st day of embryonic development in *P. brassicae* is the highly susceptible period of embryogenesis. This is not consistent with previous work on *P. brassicae* in which no mortality effect on the eggs could be found<sup>11</sup>. The small mortality of the acetone controls may be due to an alteration in the structure of the chorion of the eggs which allows excessive water to be lost from them.

Although the majority of the mortality occurred in the eggs, there was also some mortality in the 1st and subsequent ecdyses. This is most likely explained as a delayed mortality effect due to an upset in the programming of the corpora allata, rather than a result of the persistence of hormone residues<sup>5</sup>.

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### Activity alterations of metabolic enzymes in the anterior pituitary of female rats during acute and chronic starvation, as well as after refeeding

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**Summary.** The activity of glycolysis and hexose monophosphate shunt decreases while the activity of some oxydative enzymes and acid phosphatase increases in the anterior pituitary of adult female rats during starvation. The alterations depend on the severity of starvation. The polypeptide hormone production also decreases. A close relationship exists between the metabolic activity of the gland and its endocrine function.

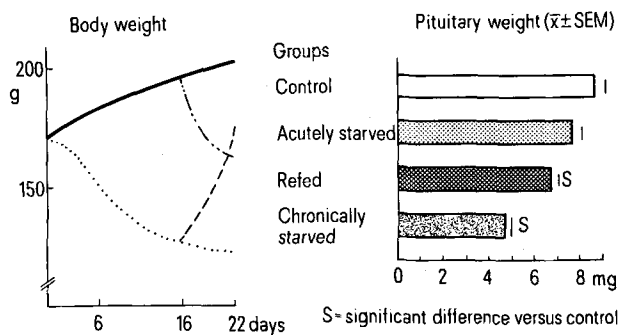
Restriction of food intake involves decreased pituitary secretion and weight reduction of the peripheral endocrine organs<sup>3-5</sup>. A substantial correlation between the decrease in prolactin, growth hormone, FSH, LH and TSH pituitary and blood concentration and/or content and the severity of starvation has been verified in our previous paper<sup>5</sup>. Similar results have been reported by other authors<sup>4,6-11</sup>. The aim of recent experiments was to clarify whether the various starvation conditions did affect anterior pituitary metabolic and lysosomal activity. An answer was sought to: 1. whether starvation-induced biochemical changes in the anterior pituitary were similar to those in other tissues, and 2. whether there was a correlation between pituitary metabolic activity and hormone secretion in starved animals.

**Material and methods.** The experiments have been carried out on adult CFY female rats. The animals were kept in individual cages. The daily intake of the granulated food was measured regularly. To prevent coprophagia a wide-meshed wire net was applied to the bottom of cages. The animals were grouped as follows: control group, fed ad libitum; acutely starved group, fed ad libitum to day 16, thereafter 6 days total food deprivation; chronically starved group, 6 days total food deprivation, thereafter  $\frac{1}{4}$  of the original food intake; the so-called refeed group, 6 days total food deprivation, then 10 days  $\frac{1}{4}$  of the original food intake, thereafter ad libitum feeding. At the end of the experimental period, the animals were decapitated in light ether narcosis. The anterior pituitary has been homoge-

Enzyme activities (mU/mg wet tissue) and protein concentration ( $\mu\text{g}/\text{mg}$  wet tissue) of rat anterior pituitary

Groups	LDH	G6P-DH	ICDH	MDH	Acid phosphatase	Protein
1 Control (10)	86.70 $\pm$ 2.33	3.59 $\pm$ 0.21	2.15 $\pm$ 0.07	256.4 $\pm$ 8.3	1.75 $\pm$ 0.05	111.6 $\pm$ 2.7
2 Acutely starved (9)	74.10 $\pm$ 2.58	3.19 $\pm$ 0.17	2.27 $\pm$ 0.06	249.5 $\pm$ 6.7	1.98 $\pm$ 0.06	113.3 $\pm$ 4.5
3 Chronically starved (10)	54.22 $\pm$ 2.46	2.22 $\pm$ 0.10	2.59 $\pm$ 0.09	300.6 $\pm$ 4.5	2.10 $\pm$ 0.05	116.0 $\pm$ 2.5
4 Refed (10)	73.75 $\pm$ 3.44	3.10 $\pm$ 0.20	2.37 $\pm$ 0.08	290.0 $\pm$ 6.6	1.89 $\pm$ 0.04	109.0 $\pm$ 2.6
p values						
1-2	<0.002	NS	NS	NS	<0.01	NS
1-3	$\leq$ 0.0001	<0.0001	<0.002	<0.0001	<0.0001	NS
1-4	<0.006	NS	NS	<0.03	<0.05	NS

Enzyme activities and protein concentration are expressed as mean  $\pm$  SE of mean. NS not significant ( $p > 0.05$ ). Number of animals in parentheses.



nized in cold 0.05 M, pH 7.2 triethanolamine-HCl buffer. After centrifugation at 4°C by 3000 rpm for 15 min, the protein concentration in the supernatant was determined with Ponceau S<sup>12</sup>. Standardized kinetic methods were used to determine enzyme activities<sup>13</sup>. LDH activity (L-lactate: NAD oxidoreductase EC 1.1.1.27) was used to characterize glycolysis, G6P-DH (D-glucose-6-phosphate: NADP oxidoreductase EC 1.1.1.49) for the hexose monophosphate shunt, MDH (L-malate: NAD oxidoreductase EC 1.1.1.37) and ICDH (threo-D<sub>2</sub>-isocitrate: NADP oxidoreductase EC 1.1.1.42) for the citrate cycle, as well as acid phosphatase (orthophosphate-monoester phosphohydrolase EC 3.1.3.1) for the lysosomal activity.

**Results.** Dynamics of b.wt alterations were in accordance with the diet (figure). There has been a constant weight increase in the control group, and weight loss was observed in the chronically underfed group. As compared to the substantial weight loss after total deprivation of food, refeeding resulted in weight gain. Pituitary weight after chronic starvation was only half of that in controls (figure). There was a minor weight decrease after acute starvation that rose essentially after refeeding, remaining on average, however, below the control value. The results of the biochemical analyses are summarized in the table. Pituitary LDH activity showed a definite decrease after chronic starvation, and was moderately but statistically significantly reduced after acute deprivation of food and refeeding. The decrease of G6P-DH activity was highest in the chronically starved group and was found moderate in the acutely starved and refed animals. Enzyme activities of the citrate cycle differed from the above. MDH activity remained unchanged after acute starvation, and showed a moderate but significant elevation in the chronically starved animals and after refeeding. The ICDH activity showed no significant increase in the acutely starved and refed groups; activity increase of about 20% was found significant, however, in chronic starvation. Pituitary acid phosphatase activity significantly increased in all of the experimental groups. Activity increase was highest in the chronically starved group. It has been found most remarkable that the concentration of water-soluble proteins in the pituitary had not been affected by starvation inspite of the well defined

quantitative and activity changes, respectively of bioactive proteins, polypeptide hormones and the enzymes.

**Discussion.** On the basis of our results, a correlation of pituitary metabolic enzymes activity with starvation has been established. The changes were found most explicit in the group of chronic starvation. According to the literature<sup>14</sup>, the glucose utilization of the various tissues decreases on starvation. In consequence of increase fatty acid metabolism, the NADH/NAD ratio increases and a large amount of acetyl-coenzyme-A accumulates that will be metabolized in the citrate cycle<sup>14</sup>. Activity changes of the studied respiratory enzymes suggest that alterations in pituitary glycolysis and oxidative metabolism during starvation do not differ basically from those observed in other tissues. During starvation, only qualitative differences can be observed between the respiratory activity of the anterior pituitary and other tissues. The essential decrease of G6P-DH activity confirms that glucose breakdown activated by the hexose monophosphate shunt becomes reduced in the anterior pituitary of starved animals. The approx. 50% decrease in LDH activity suggests an essential restriction of glycolysis. Not only glycolysis but also the adaptation of oxidative enzymes has been observed. Although increase of ICDH and MDH activities has been moderate, it was present consistently in the starved groups. In analogy with other organs<sup>14</sup>, it may be assumed that the phenomenon is related to the increased metabolism of the raised acetyl-coenzyme-A quantity, and suggests the predominant role of the citrate cycle in ATP production required for pituitary function and possibly also for the synthesizing processes in the starved animals.

The acid phosphatase indicates lysosomal activity<sup>15</sup>. The activity increase observed in starvation suggests a raised intrapituitary degradation of peptide hormones<sup>15</sup>. The mechanism might be responsible for the essential serum trophic hormone decrease in starved animals. The experimental findings are direct evidence for the close relationship between pituitary metabolic activity and the endocrine function. The parallelism between glycolytic enzyme activity and decrease of anterior pituitary hormone secretion<sup>5</sup> suggests the possibility that glycolytic processes supply an essential part of energy requirements of polypeptide hormone production.

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## TERMINOLOGICA

### Concerning the concept 'organelle'

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**Summary.** Since the essential cytological functions are endergonic processes, the organelle cannot be defined without considering the necessary energy transfer. Therefore, a sensible definition may read: The organelle is a cytoplasmic structure with functional energy consumption.

The vital functions of ultrastructural cell differentiations consist of *endergonic* processes. Therefore, a definition of the organelle must include the capacity of energy transfer of the structure under consideration.

As a consequence, individual ribosomes are not organelles. Their function as synthesists of polypeptides is only possible in cooperation with a ribonucleic acid strand when they are organized as polysomes capable of putting into action free energy furnished by the surrounding groundplasm. So the polysome, the ATP converting system, must be interpreted as an organelle, although it is not enveloped by a biomembrane.

In the same way, an individual microtubule is not an organelle. Only an association of numerous microtubules cooperating with the energy furnishing groundplasm can function as an organelle (nuclear spindle, cortical microtubular system, etc.).

The same is true of individual enzyme molecules. Yet even crowds of such molecules suspended in the groundplasm or the enchylema<sup>1</sup> have not the status of organelles, in spite of their functional possibilities, because the catabolic breakdown which they catalize (digestion, fermentation, hydrolysis) are *exergonic* processes. Relevant experiments *in vitro* proceed without the addition of an energy donor (ATP, UTP, etc.) necessary for the functioning of the polysomal anabolism *in vitro*.

The structure which functions as an organelle in the case of hydrolases is the lysosome<sup>2</sup>. The lysosomal vesicles survive only as long as the surrounding groundplasm furnishes

ATP or another donor of free energy. If this energy transfer is cut off, the enzymes in the vesicle not only hydrolyse its metabolites, but also autolyse the lysosomal membrane, previously maintained in a labile structural equilibrium, by a constant influx of energy.

Similar aspects are valid for the plasmalemma (plasma membrane). It can only resorb ions or molecules against their concentration gradients as long as it is furnished with free energy by the groundplasm.

Also groups of plasmic<sup>3</sup> or muscular fibrils which are involved in the dynamics of motility can only act as organelles if the necessary energy transfer is ensured.

In contrast to such open systems, the organelle status of coated vesicles with endergonic energy input can more easily be demonstrated. This is true for the endoplasmic reticulum (synthesis of protein, intracellular nutritional transfer) or the Golgi apparatus (polymerization of oligosaccharides, secretion and excretion activity).

Thus the organelle can be defined as a cytoplasmic structural system which functions by endergonic energy transformation.

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## PRO EXPERIMENTIS

### Polyvinylchloride (PVC) particles implantation in mouse liver. A technique for experimental study of schistosome eggs-induced liver pathology<sup>1</sup>

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**Summary.** An injection of suspended PVC particles in the caecal vein of mice induces a foreign-body portal granuloma reaction in the liver. Plastic casts of the portal system, after PVC particles implantation, show modifications in the portal bed and are compared with plastic casts obtained in mice infested by *Schistosoma mansoni*. This technique can be useful to study the cellular dynamics of the portal granuloma and can be a model for schistosomal eggs induced liver pathology.