having a pheromone effect are emitted by the males of this insect.

For Trypetidae Diptera sex pheromone emission has up to now been attributed to the males and, with the one exception of *Ceratitis capitata* Wied.[8] no chemical characterization of these substances has been accomplished. However for this insect also field experiments[10] did not ascertain the sexual specificity of the substances previously reported as sex pheromones.

For *D. oleae* we can affirm that the females emit chemical signals both attracting and aphrodisiac; however, their direction in sexual terms has not so far been ascertained.

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Inhibitory effect of fasting on the glucagon-induced increase of liver phosphorylase A activity in rats[1]

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Summary. The increase of liver phosphorylase A activity observed 1, 2 and 3 min after i.v. administration of 0.1 μg kg^{-1} glucagon to fed rats was found to be completely absent in 24 h fasted animals, although there is even an exaggerated liver cAMP response to glucagon after fasting.

In a previous paper from our laboratory[2], maximal increases in the activity of the active (A) form of liver glycogen phosphorylase (EC 2.4.1.1) in rats subjected to low 'stress-producing' doses (400 revolutions during 6 min and 40 sec) of the Noble-Collip[3] drum procedure were described. An important stimulator of phosphorylase A activity is glucagon, which increases the level of cAMP and activates the whole glycogenolytic cascade (for review see Van de Werve[4]). Under the conditions of the above-mentioned procedure, we have also demonstrated the release of glucagon[5] and an increased level of hepatic cAMP[2]. However, fasting for 24 h substantially reduced both glucagon release (unpublished results) and enzyme response[6] during the stress. Yet the possibility that decreased tissue sensitivity to glucagon might also be involved after fasting cannot be ruled out on the basis of the results available.

In this paper, the phosphorylase response of fed and 24 h fasted rats to physiological doses of exogenous glucagon is compared. As glucagon is able to increase enzyme activity within seconds[7], and the response to the drum procedure also appears very rapidly (after 2 min), phosphorylase activity was studied during the 1st 6 min after i.v. glucagon. Our results have shown the absence of phosphorylase response to 0.1 μg kg^{-1} of glucagon after fasting.

Materials and methods. Adult, male SPF rats of the Wistar strain with an average weight of 330 g supplied by VELAZ (Prague) were used. They were kept at a constant light (from 06.00 h to 18.00 h) - dark schedule and on a standard laboratory diet (caloric percent protein 25, carbohydrate 53, lipid 22, minerals and vitamins added) and tap water. The animals were allowed to adapt to the conditions of our animal house for 3 weeks. Before glucagon administration, which took place during the morning hours, 50% of the animals were fasted for 24 h. Both fed and fasted rats were anaesthetized with Pentobarbital (SPOFA, Prague, 50 mg kg^{-1} i.p.) and injected via a tail vein with 1.0 ml kg^{-1} of saline (controls) or 1.0 ml kg^{-1} of a glucagon solution (Eli Lilly and Comp., USA) in saline. In a pilot study in which only phosphorylase activity was determined the hormone concentration was 0.2 μg ml^{-1} and the animals were studied

Effect of 24 h fasting in rats on the response to an i.v. bolus of 0.1 μg kg^{-1} of glucagon. Means of 6 values per group ± SEM

	Fasting	Control	Min after glucagon 1	2	3	6
cAMP nmole g^{-1}	+	5.10±0.95	19.06±3.40[c,e]	7.94±1.68	6.20±0.37	-
(protein)	−	5.72±0.59	9.08±1.39[a]	5.74±1.31	5.45±0.65	3.07±0.22[c]
Phosphorylase	+	15.3±1.6	18.8±2.2[f]	15.8±1.7[h]	13.5±1.6[h]	-
Form A	−	18.9±2.4	26.1±1.2[a]	34.8±2.4[d]	34.2±2.4[c]	13.2±1.1
Total	+	48.3±1.9	45.1±2.6	43.4±6.4	42.2±2.5	-
	−	56.2±3.9	50.7±2.3	50.9±2.2	46.2±3.1	44.4±1.5[b]
Percent A	+	31.9±3.9	43.8±8.0	37.8±3.8[g]	32.7±4.3[h]	-
	−	33.2±2.9	51.5±1.9[d]	69.0±5.8[d]	75.0±4.9[d]	29.9±3.1

[a] Against control significant at $p<0.05$; [b] at $p<0.02$; [c] at $p<0.01$; [d] at $p<0.001$; [e] Against fed significant at $p<0.05$; [f] at $p<0.02$; [g] at $p<0.01$; [h] at $p<0.001$.

1 min after saline or hormone. Our main results, however, were obtained using a glucagon solution of 0.1 μg ml^{-1}, and 1, 2, 3 or 6 min after glucagon or 1 min after saline, liver samples were obtained by laparotomy and immediately deep frozen between 2 blocks of dry ice to be analyzed for active (A) and total phosphorylase activity[8], and cAMP[9] and glycogen content (using an anthrone method). The protein level of the 10% liver homogenate used for enzyme analysis was also determined[10]. After obtaining liver samples the animals were decapitated for collecting mixed blood for serum glucose determinations using an orthotoluidine method (test sets prepared by LACHEMA, Prague).

Results. In the screening experiment with 0.2 μg kg^{-1} glucagon no influence of fasting on the liver phosphorylase response was observed. The percentage of the total enzyme activity represented by form A in fed and fasted rats, respectively, was as follows; without hormone administration 33.6 ± 1.4 vs 23.6 ± 1.2 $p < 0.01$; 1 min after glucagon 79.1 ± 7.4 vs 80.4 ± 4.6/(means ± SEM).

In our experiment with the lower glucagon dose (table), no significant differences were found in either A or total phosphorylase activity between fed and fasted control animals. However, 0.1 μg kg^{-1} of the hormone failed to affect phosphorylase A activity in fasted animals, whereas the same dose increased it at 1, 2 and 3 min in fed rats. Substantial increases of cAMP were observed 1 min after glucagon (table), the response in the fasted group being even more pronounced than in the fed animals.

At 6 min a return of phosphorylase A vlaues to control activities and a decreased cAMP concentration was noted in fed animals. No effect of glucagon on liver glycogen content or serum glucose level of either fed or fasted animals was observed during our observation period (glucose values of control animals in mmole l^{-1}: fasted 6.6 ± 0.2, fed 8.9 ± 0.8, NS), the glycogen levels of fasted animals being extremely low as a consequence of food withdrawal (in fasted controls in mg g^{-1} 2.2 ± 0.3 in comparison with 87.5 ± 5.8 in the corresponding fed group).

Discussion. A successive lowering of rat liver phosphorylase A during fasting for several days with a decrease by about 36% after the 1st 24 h of fasting has been described[11] and a lowering of A and total enzyme activity after fasting for 24 h was reported more recently[12]. On the other hand, no effect of fasting for 19–21 h on percent phosphorylase A and total enzyme activity was found by a further group of authors[13]. In our pilot study the phosphorylase A activity of the fasted control animals was significantly lower ($p < 0.05$) than that of the fed group. The inhibitory effect of fasting was also obvious after administration of 0.1 μg kg^{-1} of glucagon. This dose was supposed to mimic portal glucagon concentrations (about 500 pg ml^{-1}) of rats subjected to the Noble-Collip drum procedure.

After the greater, probably supraphysiological glucagon dose no difference in enzyme response between fed and fasted rats was noted, therefore there seems to be an increased threshold for the glucagon effect rather than total insensitivity.

According to published reports, fasting for 18 h elevates basal and glucagon-stimulated cAMP level of isolated liver cells[14] and increases the basal cAMP level in vivo[15]. On the other hand, a decreased glucagon binding and glucagon stimulated cAMP accumulation in isolated liver cells of rats fasted for 48 h was reported[16]. In our experiments the cAMP level in the liver of control animals was uninfluenced by the food withdrawal and after glucagon it increased in fasted rats more than in fed animals. So the target of inhibition of phosphorylase by fasting should be sought somewhere on the glycogenolytic cascade between cAMP and phosphorylase. In rats fasted for 24 h a changed distribution of liver type I and type II protein kinase in the sense of predominance of type II enzyme (in contrast to the prevalence of type I in fed animals) was observed; the physiological significance of this phenomenon remains obscure[17].

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Ketoconazole, an inhibitor of calcium transport in skeletal muscle sarcoplasmic reticulum

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Summary. Ketoconazole, an antimycotic agent, inhibits calcium binding and accumulation, and induces calcium release in sarcoplasmic reticulum. The Mg^{2+}-ATPase and the $(Ca^{2+} + Mg^{2+})$-ATPase activities are stimulated at low but inhibited at high concentrations of ketoconazole.

Ketoconazole, cis-1-acetyl-4- (4((2-(2,4-dichloro-phenyl)-2-(1H-imidazole-1-yl-methyl)-1,3-dioxalan-4-yl)methoxy)-phenyl) piperazine, an antimycotic agent, inhibits the growth of yeast by inhibiting ergosterol synthesis; this is accompanied by changes in cell membrane permeability[2]. Miconazole, an antifungal imidazole chemically related to ketoconazole, inhibits the membrane-bound plasma membrane ATPase and mitochondrial ATPase of yeast[3], and affects the exchange[4] of intracellular K^+ for extracellular H^+. It has been suggested that miconazole affects mem-