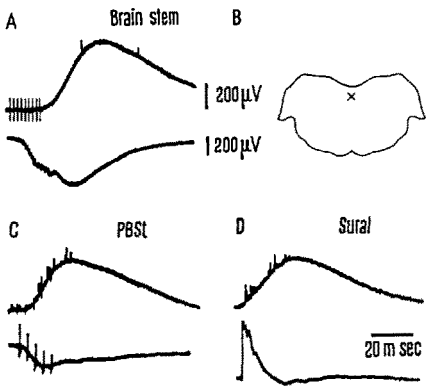

Fig. 1. The upper traces were recorded from a dorsal root filament in lower L_6. The filament was cut 15 mm from the dorsal root entry zone and placed on two electrodes one close to the entry zone and the other on the cut end. Upwards deflection denotes negativity of the central electrode. The lower traces were recorded from the dorsal root entry zone in lower L_7 against an indifferent electrode in the muscle. Record A shows the effect of stimulation of the brain stem at the site shown in B. Record C shows the effect of group I volleys in the nerve to p. biceps-semitendinosus (PBSt) and record D of a single volley in the sural nerve.

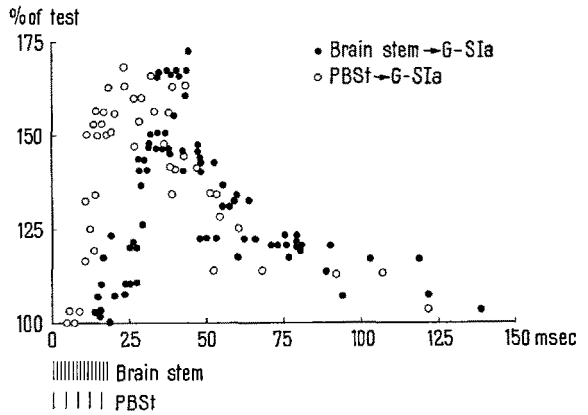

Fig. 2. Effect of conditioning volleys from the brain stem and from the PBSt nerve on the excitability of presynaptic terminals of Ia fibres from gastrocnemius-soleus (G-S). 100% on the ordinate represents the unconditioned test discharge recorded in the nerve to G-S. The testing stimulus was delivered through a micro-electrode inserted into the motor nucleus of G-S at the site where the maximal Ia focal potential could be recorded. The PBSt nerve was stimulated at a strength just maximal for group I afferents. The conditioning stimuli are indicated below the abscissa.

Figure 1 A shows the dorsal root potential evoked by a train of weak stimuli through an electrode inserted into the medulla as shown in B. The dorsal root potentials evoked by repetitive stimulation of group I fibres from p. biceps-semitendinosus (PBSt) and of the sural nerve are shown for comparison in C and D. Transverse exploration revealed a very localized optimum in the brain stem. When the electrode was moved 1 mm from the focus shown the stimulus strength had to be considerably increased to evoke action.

The effect was evoked from the medulla and from the caudal pons but not from more rostral pontine levels. Intraspinal preterminal threshold measurements from various categories of primary afferents revealed that on medullary stimulation there was correspondingly an increased excitability in Ia, Ib and in cutaneous afferents. The curves in Figure 2 show the effect of stimulation of the brain stem and of the PBSt nerve on the excitability of Ia fibres from gastrocnemius-soleus measured with a stimulating electrode in the motor nucleus of this muscle. The actions are evoked through ventral spinal pathways: the effect was abolished after transection of the ventral quadrants but remained when only the dorsal quadrants were transected. The site in Figure 1 B corresponds to the location of the medial longitudinal fasciculus, but further investigations are required to decide if this tract is responsible.

Depolarizing dorsal root potentials can also be evoked from other more widespread predominantly ventral regions of the brain stem. Intraspinal threshold measurements revealed depolarizations in Ib afferents and the FRA after a longer latency than the effect in Figure 1 A. The descending pathways are located in the dorsal quadrants. This system may be of interest in relation to the tonic inhibition exerted from the medial brain stem (through descending pathways in the dorsal part of the lateral funicle) on reflex arcs and transmission to certain ascending pathways. It is, however, not known if this tonic inhibition is caused by presynaptic depolarization or, as originally assumed, by inhibition at an interneuronal level[3]).

To summarize: there is mounting evidence of complex supraspinal actions on primary afferents. The system depolarizing the presynaptic terminals of Ia afferents is of particular interest since it permits supraspinal regulation of monosynaptic Ia actions.

Zusammenfassung. Es wird gezeigt, dass eine elektrische Medulla oblongata- oder Pons(kaudal)-Stimulation eine Depolarisation (präsynaptische Inhibition) von Ia-, Ib- und Flexor-Reflex-Afferenz im Lumbalmark verursachen kann.

D. Carpenter, I. Engberg, and A. Lundberg

Department of Physiology, University of Göteborg (Sweden), June 21, 1962.

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Blood Glucose and Plasma Unesterified Fatty Acid Changes Induced by the Stress of an Emergency Situation

The discharge of glycogen stores and the rise in the level of blood glucose following the release of adrenaline in an emergency situation was thought traditionally to provide an animal with increased oxidizable substrate for use in impending flight. With the realization of the metabolic importance of the plasma unesterified fatty acids (UFA)[1] and the effect of adrenaline in causing their release from adipose tissue[2,3] a reappraisal was necessary of the relative

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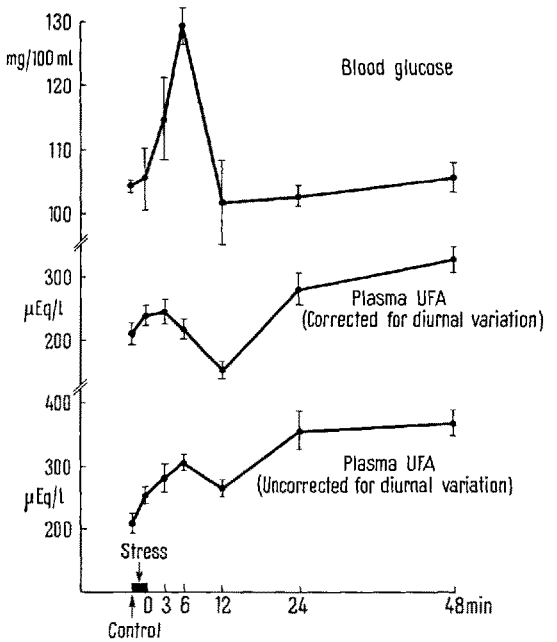
importance of glucose and UFA as fuels of the tissues under emergency situations. SHAFRIR et al. have studied the time course of plasma UFA and glucose responses to the administration of short acting adrenaline in dogs[4] and of long acting adrenaline in dogs[4] and rats[5,6]. Plasma UFA became elevated before the blood glucose and the authors concluded that under the sympathetic discharge of an emergency condition, it is the adipose tissue, and not the glycogen stores, that most rapidly satisfies the demand for oxidizable substrate. However, it has frequently been observed in this laboratory, that untrained rats give erratic and elevated results in the analysis of blood glucose but on the other hand, provided there is no undue delay in sampling, analyses of plasma UFA give low and uniform results. It was decided therefore to follow the changes in blood glucose and plasma UFA of rats submitted to a standard stress which mimiced as closely as possible one that might occur naturally.

Untrained, female, albino rats (200 g) were permitted to escape from their cages and after several mock attempts at recapture they were finally caught and returned to their cages after exactly 2 min at large. At this time (0 min) and subsequently thereafter at 3, 6, 12, 24, and 48 min, batches of four rats were decapitated and blood collected for the estimation of glucose[7] and plasma UFA[8]. The experiments were completed between 10 a.m. and 12.30 p.m. on one day. Control values were obtained from rats that had previously been handled. These did not undergo stress and were killed at 10 a.m. The data were submitted to statistical tests using the Analysis of Variance.

In order to assess the relative physiological importance of the magnitude of changes in blood glucose and plasma UFA it is necessary to plot the data on scales which are physiologically comparable. Other work[9] using fed and fasting rats has shown that the plasma UFA rises in concentration by approximately 100 μEq/l for every 10 mg/100 ml fall in blood glucose. The scales used in the Figure are based on this relationship. Caution must also be exercised in the interpretation of changes in plasma UFA since normal fed rats show a diurnal rise of approximately 0.75 μEq/l/min between 10 a.m. and 12 noon[9]. Presumably this rise would alter the 'baseline value' of the UFA on which the experimental results are superimposed, although at present there is no information available to verify this presumption. The data for plasma UFA are presented both corrected and uncorrected for this variation (Figure). The correction was applied by subtracting from the results 0.75 μEq/l of UFA for every minute exceeding 10.0 a.m. up to the time the animal was killed. When corrected, the values are equivalent to those that would have been obtained had the animals been stressed at an appropriate time before 10.0 a.m. and all been killed at 10.0 a.m. exactly.

Immediately after the period of stress there was a rise in blood glucose which reached a maximum of nearly 130 mg/100 ml after 6 min (P 0.001–0.01). By 12 min the concentration had fallen to within the normal range where it remained up to the termination of the experiment. The plasma level of UFA (corrected) rose by an insignificant amount ($P > 0.2$) for up to 3 min after the stress and then fell significantly (P 0.001–0.01) during the next 6 min, the fall coinciding with the maximum increase in blood glucose. The suppressive effect of a rise in blood glucose on the level of UFA has been seen by previous workers after the injection of adrenaline[4,5]. As soon as the blood glucose had returned to within the normal range, the plasma UFA rose significantly ($P = 0.001$) for the remainder of the experiment. With respect to the results uncorrected for the diurnal variation, there was a small but significant (P 0.001–0.01) initial increase lasting up to 6 min after the stress. The magnitude of this rise was no more however than that which occurs diurnally between 10 a.m. and 12 noon in fed rats[9]. A small but insignificant fall (P 0.05–0.2) occurred between 6 and 12 min, coinciding with the maximum rise in blood sugar.



Changes in blood glucose and plasma UFA concentrations in rats stressed for 2 min. Each point is the mean of four results except for the controls, which are the mean of five results in the case of the blood glucose and six in the case of the UFA. The bars show the S.E.M.

Present results indicate that a sympathetic discharge occurring under natural conditions as a result of an emergency situation, causes an immediate and large rise in blood glucose and at the most only a small rise in plasma UFA. As soon as the glucose rise becomes prominent, any rise in UFA is suppressed and it only rises again significantly when the glucose rise has subsided. The eventual rise in plasma UFA is of a much longer duration than the rise in blood glucose. If these changes in concentration in the blood reflect the availability of these two substrates to the cells, then glucose is clearly the predominant fuel used in the first minutes of an emergency and UFA are mobilized subsequently to supplement or take the place of carbohydrate as this becomes inadequate as a source of energy. These conclusions agree with the known metabolic properties of glucose and fatty acids. The fuel of most use

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to muscle in the first minutes of flight is one that can be oxidized anaerobically such as glucose. Later, a fuel such as the UFA, which must be oxidized aerobically, which is of high calorific value, and which is available in relatively unlimited quantity, comes into its own when the respiratory and circulatory systems are delivering more oxygen to the tissues.

Résumé. Lorsque l'organisme est mis brusquement dans une situation exigeant de promptes réactions, une décharge sympathique cause immédiatement une importante élévation du glucose sanguin alors qu'il n'y a tout au plus qu'une légère augmentation des acides gras non-estérifiés du plasma. Une augmentation plus durable de ces acides gras non-estérifiés suit le retour à la normale du glucose sanguin.

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Inhibition of Pentobarbital and Meprobamate Metabolism by some 'Inducers' of Drug-Metabolizing Enzymes

Recently, a variety of 'prolonging agents', which enhance drug action by virtue of their ability to inhibit drug biotransformation, have been reported.

For example, SKF 535 A (2-diethylaminoethyl-diphenylpropyl-acetate) Lilly 18947, (N, N-diethyl-N-2-phenyl-4, 6-dichlorophenoxy-ethylamine) iproniazid and N-ethyl-3-piperidyldiphenylacetate, owe their prolonging action to nonspecific inhibition of some microsomal drug-metabolizing enzymes[1,2].

On the other hand, drugs, such as phenobarbital, phenaglycodol, glutethimide, chlorcyclizine, phenylbutazone, aminopyrine, zoxazolamine, meprobamate, chlorpromazine, niketamide, chloretone, if injected 12–24 h in advance, induce an increase activity of some microsomal drug-metabolizing enzymes[3–11].

The hypothesis arises that some of the above mentioned drugs inducing a late increase of activity of drug-metabolizing enzymes, have an immediate blocking action on the same enzyme systems.

Male rats of the Sprague-Dawley strain, weighing about 60 g were used.

The enzyme activities were determined by measuring the amount of metabolized drugs during an incubation of 1 h with liver microsome-containing supernatant as described previously.

Concentrations of pentobarbital and meprobamate were determined according to the methods of BRODIE et al., and HOFFMANN and LUDWIG, respectively[12,13].

In every case, the possibility of an interference in the determinations of pentobarbital and meprobamate concentrations by the inhibitors and their metabolites was excluded.

The concentrations of inhibitors which produce 50% inhibition of pentobarbital and meprobamate metabolism is shown in the Table.

Concentration of SKF 525 A[14] and Lilly 18947[14] producing 50% inhibition were also given, because they can induce the microsomal drug-metabolizing enzymes[15].

Chlorcyclizine had the most potent inhibitory action which is almost of the same potency as that of SKF 525 A and about 2 times more potent than that of Lilly 18947.

Glutethimide was potent not only in the metabolism of pentobarbital, but also in the meprobamate metabolism. These facts suggest that glutethimide inhibits pentobarbital metabolism not only by virtue of the similarity of its chemical structure to pentobarbital.

Phenobarbital which has the most potent inducing action of the microsomal drug-metabolizing enzymes, shows however, a weak inhibitory action on the me-

Inhibition of pentobarbital and meprobamate metabolism by some inducing-drugs of the microsomal drug-metabolizing enzymes

Inhibitors	Concentrations producing 50% inhibition (Mol)	
	Metabolism of pentobarbital	Metabolism of meprobamate
1 Chlorcyclizine	2.0×10^{-5}	1.4×10^{-4}
2 Glutethimide	8.3×10^{-5}	6.5×10^{-5}
3 Phenaglycodol	1.2×10^{-4}	9.3×10^{-5}
4 Phenobarbital	—	4.2×10^{-4}
5 Chlorpromazine	1.6×10^{-4}	2.3×10^{-4}
6 Zoxazolamine	1.8×10^{-4}	2.3×10^{-4}
7 Chloretone	1.9×10^{-4}	2.2×10^{-4}
8 Phenylbutazone	—	3.9×10^{-4}
9 Niketamide	5.2×10^{-4}	7.8×10^{-4}
10 Aminopyrine	9.6×10^{-4}	7.2×10^{-4}
11 SKF 525 A	1.7×10^{-5}	1.5×10^{-5}
12 Lilly 18947	4.4×10^{-5}	5.2×10^{-5}

The incubation mixture (5.0 ml) contained 2 ml of the microsome-containing supernatant (obtained by centrifugation at 8500 *g* for 15 min), 0.1 ml of 20 µmole glucose-6-phosphate, 0.4 µmole TPN 50 µmole nicotinamide and 75 µmole $MgCl_2$ and 1 *M* KCl, and more 2.3 ml of 0.1 *M* sodium phosphate buffer pH 7.4 and 0.2 ml of the substrates (final concentration were 2×10^{-4} Mol ×) and 0.1 ml of inhibitors. Phenobarbital and phenylbutazone interfered with determination of pentobarbital concentration.

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