crease in Na absorption. The high final serosal fluid glucose concentrations seen in the insulin treated rats indicate a marked increase in the sugar concentrating activity of the mucosal cells, a process which has also been taken to be a measure of sugar transport[13]. Since water accompanies Na movement from mucosal fluid to serosal fluid, the small intestine does not concentrate Na, and absorption is generally taken as the measure of Na transport[13]. Segments from alloxan-diabetic rats transported more glucose than non-diabetics, confirming the results of previous studies. In addition, Na transport was approximately doubled in the diabetic animals. The final series of experiments was concerned with the effect of insulin in alloxan-diabetes. Given first at the same dose level employed in normal rats (0.75 U/rat) the hormone depressed Na absorption and increased serosal fluid glucose concentration as was observed in normal animals, but failed to change glucose absorption. The same results were observed in a second group in which insulin was given in larger amounts (2.5 U/rat). The failure of insulin to increase sugar absorption in alloxan-diabetes may be related to decreased fluid absorption, an effect of the hormone seen in diabetic but not in normal rats.

The increases in sugar transport seen in both alloxan-diabetic and insulin treated normal rats appears contradictory. However, the increased glucose and Na transport in alloxan diabetes may have been the result of mucosal cell metabolic changes compensatory to the renal losses of glucose and Na characteristic of the diabetic state.

SCHULTZ has recently proposed a model system suggesting linked entry of Na and actively transported sugars into the mucosal cell in which Na and the sugar share a common carrier[10]. The complex dissociates inside the cell and Na is removed by a 'pump' located at the serosal surface of the cell. Sodium removal favors dissociation of the common carrier and the sugar probably moves out of the cell down its concentration gradient. Changes in alloxan-diabetes appear to fit this hypothesis of linked Na and glucose entry, since absorption of both is increased. However, it appears that insulin increased glucose transport by some other pathway, since Na absorption is decreased by the hormone. The decreased Na absorption produced by insulin may be accountable to depression of dissociation of the Na-carrier-glucose complex by high intracellular levels of sugar accumulated under the influence of the hormone, reducing the amount of Na available to the serosal 'pump'[14].

Zusammenfassung. Die Natrium- und Glucoseabsorption in Dünndarmsegmenten alloxandiabetischer Ratten war erhöht. Durch Insulininjektionen wurde bei diesen Tieren die Natriumabsorption herabgesetzt, während bei normalen Ratten Insulin die Glucoseabsorption erhöhte und die Natriumabsorption erniedrigte.

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Department of Physiology, University Medical Center, Little Rock (Arkansas USA), December 23, 1964.

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Potentiation of Bradykinin and Eledoisin by BPF (Bradykinin Potentiating Factor) from *Bothrops jararaca* Venom

As shown in previous papers from this laboratory[1-3], a factor called BPF (bradykinin potentiating factor), extracted from the venom of *Bothrops jararaca*, was capable of increasing some of the pharmacological activities of bradykinin on the guinea-pig ileum, rat uterus and rabbit duodenum, as well as the hypotensive effects of the polypeptides in cats and dogs. BPF has no effect upon contractions elicited by histamine and acetylcholine, and little effect on the actions of angiotensin and oxitocin on the smooth muscles of the guinea-pig ileum and the rat uterus respectively.

In this paper an analysis of the effects of BPF was extended to the polypeptide eledoisin, since it has been shown that this substance is more resistant to inactivation by plasma enzymes and considerably more active than bradykinin on the guinea-pig ileum and blood pressure of the cat and the dog[4-7].

(a) *Experiments on the guinea-pig ileum.* The deductions as to the potentiation of the agonists by BPF were taken after plotting the reciprocals of the effects (1/y) against the reciprocals of the doses (1/x) according to the technique described previously[8,9]. BPF produced a definite potentiating effect upon bradykinin (BRS 640, Sandoz) and kallidin (KL 698, Sandoz) responses without any apparent action on eledoisin (ELD 950, Sandoz) contractions. Figure 1 gives a view of the experimental data recorded from two typical experiments. We can see that bradykinin and kallidin lines tend to displace towards eledoisin lines during the treatment of the guinea-pig ileum with BPF. It must be noticed that, when the concentration of BPF reached 5 μg/ml, the sensitivity of the isolated preparation to bradykinin and eledoisin was almost the same. Concentrations of BPF up to 10 μg/ml did not consistently change the pattern of responses obtained with 5 μg/ml.

(b) *Arterial blood pressure experiments.* These experiments were performed on the arterial blood pressure of the dog and cat recorded from a common carotid artery by means of a mercury manometer. BPF potentiates the

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hypotensive action of bradykinin in the cat as well as in the dog. The intensity of this potentiation could be appreciated by measuring comparatively the initial fall in arterial blood pressure or, even better, by the hypotensive area recorded after bradykinin administration. This hypotensive area was measured in a graph in which the mean arterial blood pressure variations from the original base line were plotted against time.

Considering the long duration of the bradykinin effects after BPF administration, histamine, acetylcholine and eledoisin were administered before bradykinin. All agents were injected intravenously.

A rapid BPF injection (2 mg/kg) produced a small and transitory hypotensive effect which did not occur when the administration of BPF was slow. BPF had no detectable effect on histamine or acetylcholine responses (Figure 2). Although BPF produced in the cat a small potentiation of the action of eledoisin on blood pressure, no such effect could be observed in the dog (Figures 3 and 4).

Under normal conditions, eledoisin has been found to be 8 times more active than bradykinin in the cat, and 20–50 times more active in the dog[4,5]. If the animals were treated with equipotent doses of both peptides before and after treatment with BPF, bradykinin became more potent than eledoisin in cats and only half as potent in dogs.

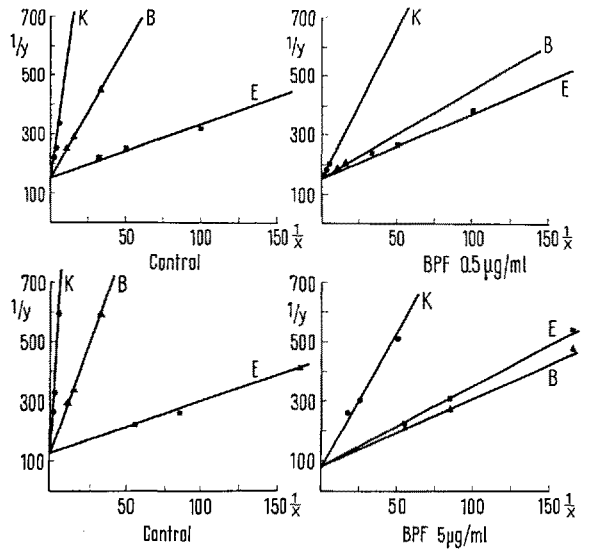

Fig. 1. Two experiments represented by the double reciprocal plots. In ordinate the reciprocal of the responses (1/y); and in abscissae the reciprocal of the final concentration of the agonists. Note that kallidin (K) and bradykinin (B) lines tend to displace towards eledoisin (E) line during the treatment of the guinea-pig ileum with BPF. Tyrode containing atropine ($1 \cdot 10^{-4}$ mg/l) and Benadryl ($5 \cdot 10^{-5}$ mg/l).

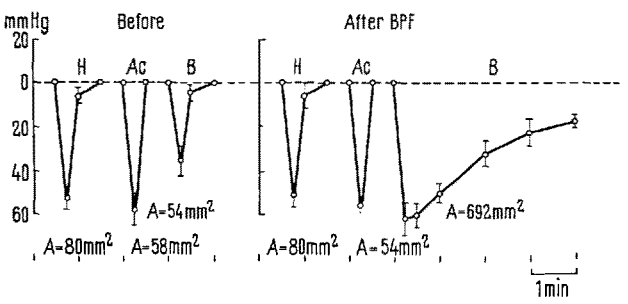

Fig. 2. Action of BPF upon the hypotensive effects of histamine (H = 2 µg/kg), acetylcholine (Ac = 0.1 µg/kg) and bradykinin (B = 2 µg/kg) in the cat. The dose of BPF was 2 mg/kg. The points represent the mean blood pressure variation in three animals. Standard errors given by vertical bars. A = hypotension area. Anaesthetic: Nembutal 30 mg/kg i.v.

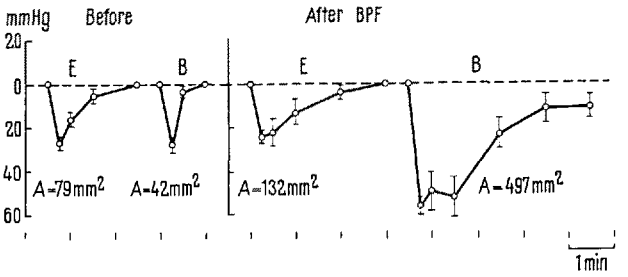

Fig. 3. Action of BPF upon the hypotensive effects of bradykinin (B = 2 µg/kg) and eledoisin (E = 1 µg/kg) in the cat. The dose of BPF was 2 mg/kg. The points represent the mean arterial blood pressure variation in three animals. Standard errors given by vertical bars. A = hypotension area. Anaesthetic: Nembutal 30 mg/kg i.v.

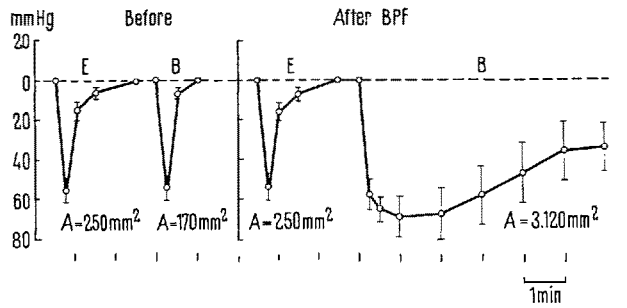

Fig. 4. Action of BPF upon the hypotensive effects of equipotent doses of bradykinin (B = 1 µg/kg) and eledoisin (E = 0.024 to 0.066 µg/kg) in the dog. The dose of BPF was 2 mg/kg). The points represent the mean arterial blood pressure variation in five animals. Standard errors given by vertical bars. A = hypotension area. Anaesthetic: Nembutal 30 mg/kg i.v.

Ratios of potency of eledoisin/bradykinin on the arterial blood pressure of the dog before and after treatment with BPF

Dog No.	Before BPF			After BPF		
	B	E	Ratio	B	E	Ratio
I	1	0.024	41.6	0.024	0.012	2
II	1	0.066	15.1	0.066	0.033 0.066	2
III	1	0.024	41.6	0.024	0.024	1
IV	1	0.048	20.8	0.048	0.024	2
Range of ratios			15.1–41.6			1.0–2.0

The figures indicate the equivalence of doses, in µg/kg, of bradykinin (B) and eledoisin (E), before and after administration of BPF (2 mg/kg). In dog II, after BPF treatment, the indicated dose of bradykinin produced an effect intermediate between the two doses of eledoisin.

In our experiments, the doses of eledoisin which produced an initial fall in arterial blood pressure equal to that induced by 1 μg/kg of bradykinin varied from 0.024 to 0.066 μg/kg.

The Table indicates the equivalence of doses of bradykinin and eledoisin before and after administration of BPF in 4 different dogs. The ratios of the activity of eledoisin/bradykinin, which varied from 15–41 in the control experiments, were reduced to almost equivalency ranging from 1.0 to 2.0 after treatment with BPF. It is to be noted that the doses of bradykinin (0.024–0.066 μg/kg), which produced noticeable effects after treatment with BPF, were below the threshold doses necessary to produce any effect before the treatment. Kallidin showed a behaviour similar to that of bradykinin, being strongly potentiated by BPF.

In conclusion, the results presented in this communication show that the pharmacological potency of bradykinin on guinea-pig ileum and arterial blood pressure of the cat and the dog becomes similar or stronger than that of eledoisin after BPF treatment.

Our present findings give further support to the suggestion[2,3] that bradykinin potentiation by BPF could be due to the blockade of the peptide inactivation by blood, or by kininases possibly present in the smooth muscles, at sites close to the pharmacological receptors[10].

Zusammenfassung. Nachweis der pharmakologischen Wirkung des Bradykinins am isolierten Meerschweinchenileum und am deutlichen Blutdruckanstieg bei Hund und Katze nach Vorbehandlung mit BPF (Bradykininpotenzierender Faktor aus Jararacagift). Unter diesen Bedingungen werden mit Bradykinin gleich intensive Effekte wie mit Eledoisin erhalten.

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Department of Pharmacology, Faculty of Medicine, University of São Paulo, Ribeirão Prêto, Est. de São Paulo (Brazil), February 1, 1965.

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COGITATIONES

Mitotic Rate in Organs and Tissues in Relation to Metabolic Body Size ($kg^{3/4}$)

On the molecular level of metabolism, the concentration of certain respiratory enzymes, such as cytochrome-*c*, cytochrome oxidase, etc., per unit of metabolically active mass ($kg^{¾}$) is proportional to overall energy exchange per kg – for rats, dogs, men and cows[1,2]. At least in mammalian liver, the amount of mitochondria bears the same quantitative relation to total metabolism as to total body size, and it is likely that the relative amounts of these elements in any given tissue will prove to be the controlling factor in determining the regression of oxygen utilization on total body size of the species[3,4].

In a broad and metabolic sense, cellular life-span is closely correlated with the life-span of certain cellular components or metabolites. Thus, the $\tau_{½}$ – half time – of cytochrome-*c* in liver may be as representative of the turnover time of the liver cell as is the $\tau_{½}$ of hemoglobin of that of the red blood cell[5].

Metabolic processes in the rat, for example, go at rates some 4.6 times faster than those in man. The metabolic rate factor 4.6 reflects the degree of difference between the adult rat and adult man in rate per unit time and per unit mass of such measurable quantities as the basal metabolic rate (i.e. oxygen consumption), the outflow of nitrogen in the urine, or of protein production[5].

The mitochondrial energy transduction in aerobic organisms is based on citric acid cycle oxidations and fatty acid oxidation, coupling electron flow to synthesis of adenosine triphosphate (ATP), that is, oxidative phosphorylation. The components of the electron transport system are currently though of as consisting of a series of physically interconnected lipoproteins with electron acceptors (coenzymes) tightly bound. The terminal catalyst appears to be cytochrome oxidase and may be the *rate-limiting factor.*

Other studies, although indirectly, substantiate the function of cytochrome oxidase as a rate-limiting factor. Chance's work[6] indicates that in the intact mitochondria, the cytochrome oxidase velocity constant is 4 to 23-fold less than that of the other reactions. Another item of evidence is that the absolute values of the total cytochrome oxidase activity in certain large as well as small animals were found to be very nearly the same as the maximal metabolism of the intact animal[7].

With cytochrome oxidase as a rate-limiting factor, increasing body size is accompanied by decreasing respiratory intensity – in analogy, a small clock must have a faster-swinging pendulum than a large one – in vivo and in vitro[8], and the duration of the metabolic process, Life,

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