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CARTAS AL EDITOR

Effect of Somatostatin on Leucineaminopeptidase Activity in the Small Intestine

Somatostatin (SRIF) is a small peptide hormone which is widely distributed in the organism and has great activity in the gastrointestinal mucosa. The fact that plasma somatostatin levels rise after ingestion of fat, glucose or protein suggest that this peptide probably controls the transfer of nutrients to the circulation (1) and in spite of there existing few indications of how this is done, it is thought that SRIF regulates intestinal absorption by modifying the activities of enzymes directly involved in the process. The present article describes the effect of somatostatin on the activity of leucineaminopeptides (LAP), an enterocyte hidrolase which cleaves N-terminal L-peptide residues with a free amino group.

The animals used were male and female hamsters weighing 80-100 g, male and female rats weighing 120-140 g and 4-6 week old chicks. A dose of SRIF of 6 μg/100 g body weight was administered i.p. in saline solution 4 or 14 h before the animals were killed. Control animals were injected with saline solution alone. Brush border membrane vesicles of chicks and rats were prepared using the method of KESSLER *et al.* (6). The brush border membrane from hamsters were obtained according to the method of EICHHOLZ and CRANE (4). LAP activity was assayed as per GOLDBARG and RUTENBURG (5) and expressed as a specific activity as mU per mg of protein. Protein was determined by the LOWRY *et al.* (7) method.

Since the preparation of the brush border membranes of hamsters required the use of EDTA, which inhibits LAP, the enzyme was activated before assay by 10 min incubation at 40°C with 0.002 M $MnCl_2$ buffered with 0.04 M Tris (pH 8). The substrate solution used in the assays contained L-leucyl-beta-naphthylamide hydrochloride and an appropriate amount of $MnCl_2$ buffered with Tris at pH 8.6 (10, 11).

Student's t test for paired data were used to compare sample means.

The LAP activity of homogenates were not modified by hormone. In isolated brush border membranes of chicks, LAP activity fell slightly in the duodenum and jejunum and significantly in the ileum when SRIF was injected 4 h before decapitation, but when it was 14 h before, LAP activity rose in the duodenum and

Table I. *Somastostatin effect on LAP activity in brush border of chick.* Results are expressed as the mean + SEM. Number of animals = 7.

Time h	Duodenum		Jejunum		Ileum	
	Control	Treated	Control	Treated	Control	Treated
4	546.3 ± 92.6	485.7 ± 35.5	570.7 ± 62.8	521.8 ± 58.0	1299 ± 316	945 ± 68.3*
14	386.2 ± 60.8	597.4 ± 60.5*	561.7 ± 40.7	702.0 ± 47.0*	1573 ± 215*	1926 ± 264.2

* Statistical significance $P < 0.01$.

Table II. *Somatostatin effect on LAP activity in brush border of rat and hamster.* Results are expresed as the mean + SEM. Number of animals = 10. Statistical significance.

	Time h	Rats		Hamsters	
		Males	Females	Males	Females
Control	4	477.4 ± 32.6	396.6 ± 48.7	1288.3 ± 91.4	1798.4 ± 353.3
Treated	4	346.4 ± 37.3*	366.0 ± 64.1	1330.0 ± 144.5	1867.7 ± 392.8
Control	14	510.3 ± 38.8	436.2 ± 17.8	1520.1 46 67.2	1791.3 ± 212.5
Treated	14	297.4 ± 16.0**	402.4 ± 41.8	1791.3 ± 212.5	2843.4 ± 299.5*

* $P < 0.01$ and ** $P < 0.005$.

jejunum (table I). These results may be explained by the existence of two kinds of somatostatin receptors, with either low or high affinity (2), for the inhibitory effect observed within a few hours after injecting the hormone mediated by high-affinity binding sites on the enterocyte membrane, this effect reversing when slower low-affinity bindign sites later trigger stimulation of LAP activity.

In male rats, SRIF produces a decrease in enzymatic brush border activity, while LAP activity increased in brush border of female hamsters injected 14 h earlier (table II). The inhibitory action of the hormone on brush border enzymatic activity might be mediated by a decrease in cyclic AMP concentration. In rats, SRIF inhibits cyclic AMP or increases cyclic GMP and it is an activator of phosphoprotein phosphates in the pancreas, liver and enterocytes (2, 3). The finding that no effect was exerted by somatostatin in male hamsters is in keeping with those reported by SENEGAS-BALAS *et al.* (9). The increase in LAP activity in the female hamsters 14 h after hormone administration, may be due either to the hormone indirectly affecting pancreatic function, to changes associated with the administration of SRIF or to a fall in enzymatic degradation rates (8). In view o the above considerations, the effect of somatostatin may be said to depend on both sex and species, due perphaps to the existence of different kinds of hormone receptors.

Key words: Somatostatin, Leucine amino peptidase, Brush border.

Palabras clave: Somatostatina, Leucina amino peptidasa, Borde en cepillo.

References

1. Burhol, P. G., Lygren, I., Jenssen, T. G., Elorholmen, J. and Jorde, R.: *Acta Physiol., Scand.*, 121, 223-228, 1984.
2. Catalán, R. E., Miguel, B. G., Godoy, J. E., Robles, A., Aragonés, M. D. and Martínez, A. M.: *Biochem. Med.*, 31, 36-41, 1984.
3. Catalán, R. E., Martínez, A. M. and Arago-

nés, M. D.: *Regul. Pep.*, 8, 147-159, 1984.
4. Eichholz, A. and Crane, R. K.: In «Methods in Enzymology» (S. Fleischer and L. Packer, eds.), Academic Press, New York, 1974, vol. 31, pp. 123-125.
5. Goldbarg, J. A. and Rutenburg, A. M.: *Cancer*, 11, 283-291, 1958.
6. Kessler, M., Acuto, O., Storelli, C., Murer, H., Muller, M. and Semenza, G.: *Biochim. Biophys. Acta*, 506, 136-154, 1978.
7. Lowry, O. M., Rosebrough, N. J., Farr, A. L. and Randall, N. J.: *J. Biol. Chem.*, 193, 265-275, 1951.
8. Ramaswamy, K. and Flint, P. W.: *Am. J. Physiol.*, 238, G114-G118, 1980.
9. Senegas-Balas, F., Balas, D., Pradayrol, L., Laval, J., Bertrand, C. and Ribet, A.: *Acta Anat.*, 121, 124-132, 1985.
10. Smith, E. L. and Spackman, D. M.: *J. Biol. Chem.*, 212, 255-269, 1955.
11. Spackman, D. M., Smith, E. L. and Brown, D. M.: *J. Biol. Chem.*, 212, 271-299, 1955.

M. C. TABOADA*, J. MARTÍNEZ-SAPIÑA, M. D. ANDRÉS, E. REBOLLEDO and M. P. FERNÁNDEZ-OTERO**

Departamento de Fisiología
Facultad de Farmacia
15706 Santiago de Compostela (Spain)

(Received on July 15, 1986)

* To whom all correspondence should be addressed.

** Address: Departamento de Fisiología. Facultad de Farmacia. Universidad de Navarra. 31080 Pamplona (Spain).