



Involvement of mast cells in the priming effect of substance P which allows guinea pig ileum to be contracted by nitric synthase inhibitors

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BY J. FIORAMONTI

**Digestive Disease Week
and the
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Gastroenterological Association**

May 19-22, 1996, San Francisco, California

**Program of the Annual Meeting of the American
Gastroenterological Association, the American
Association for the Study of Liver Diseases, the
Gastroenterology Research Group, the Society for
Surgery of the Alimentary Tract, and the American
Society for Gastrointestinal Endoscopy**

**Abstracts of Papers Submitted to the American
Gastroenterological Association**

**Abstracts of Papers Submitted to the American
Association for the Study of Liver Diseases**

**Abstracts of Papers Submitted to the Society for
Surgery of the Alimentary Tract**

BLOCK OF A COLONIC SMOOTH MUSCLE DELAYED RECTIFIER CHANNEL BY PHENCYCLIDINE. B.W. Frey, F.T. Lynch and A. Carl. University Nevada School of Medicine, Physiology Dept. Reno, NV 89557, USA

Phencyclidine (PCP or "angel dust") is a dissociative anesthetic and potent hallucinogenic drug known to block central nervous system NMDA glutamate receptors and K^+ channels. Recently we found that "delayed rectifier" current in colonic myocytes is composed of two major distinguishable currents: (1) a rapidly activating 4-AP sensitive current, $I_{dK(f)}$ and (2) a slowly activating TEA sensitive current, $I_{dK(s)}$ (J. Physiol. Lond. 484:339-353). In addition, a rapidly activating TEA sensitive current, $I_{dK(n)}$, may contribute to the voltage dependent outward current. The goal of the present experiments was to test whether PCP blocks "delayed rectifier" channels in smooth muscle and whether this drug could be utilized to distinguish between the "delayed rectifier" current components. Outward current was measured with 100 ms step depolarizations to +10 mV from -50 mV holding potential (whole cell voltage clamp). With 10 mM 4-AP in the bath solution, PCP blocked outward current (predominantly $I_{dK(s)}$) at $EC_{50} = 1.1 \mu M$. With 30 mM TEA in the bath solution, PCP (10-4 M) blocked only 20% of control current, probably representing block of residual $I_{dK(s)}$. When 10 mM TEA was included in the pipette solution to isolate $I_{dK(n)}$, PCP (10-4 M) blocked 55% of current. These data suggest that PCP at low concentrations (<10-4 M) might be utilized to investigate the role of $I_{dK(s)}$ in colonic smooth muscles. At higher concentration, PCP also blocks $I_{dK(n)}$ but not $I_{dK(f)}$. These data also show that the two main components of smooth muscle delayed rectifier current can be separated by their pharmacology: (1) a 4-AP sensitive current, $I_{dK(f)}$ and (2) a PCP sensitive current, $I_{dK(n)}$. Supported by the American Heart Association, Nevada Affiliate and NIH-DK 41315.

INVOLVEMENT OF MAST CELLS IN THE PRIMING EFFECT OF SUBSTANCE P WHICH ALLOWS GUINEA-PIG ILEUM TO BE CONTRACTED BY NITRIC OXIDE SYNTHASE INHIBITORS.

R. Garcia-Villar, J. Gay, J. P. Martinolle, J. Fioramonti, & L. Bueno. INRA, Pharmacology & Toxicology Unit, BP.3, 31931, Toulouse, France.

Substance P (SP) immunoreactive nerve fibers have been shown in close apposition with mast cells (MCs) in the intestine. Thus, neuromodulation of the release of mediators from MC has been proposed. We previously showed that *in vitro* SP exerted an as yet not fully understood "priming" effect, which allowed nitric oxide synthase (NOS) inhibitors to contract strips of guinea-pig ileum (Gastroenterology, 108: A603). In this study we addressed the issue of the involvement of MCs in this tissue priming response to SP by investigating the effects of prototypic MC stabilizers and activators.

Methods: Strips of guinea pig ileum (8x2 mm) were placed in the direction of longitudinal fibers in an 8-chamber superfusion system (Krebs, 37°C, 2 mL/min). Drugs, instilled (40 μL /min) in the superfusion flow, were: (i) SP (0.1nM to 10 μM); KCl (60mM); NOS inhibitor, L-NAME (1 μM); all superfused for 3 min; (ii) MC stabilizers ketotifen and doxanzole (1 μM); MC activators, compound 48/80 (10 μg /mL) and concanavalin A (20 μg /mL); all superfused from 20 min before, until 20 min after SP. Contractions were recorded (isometric system) and automatically quantified (g/mm²). Concentration-response curves for both contractile and priming effects of SP were constructed, and EC_{50} s and E_{max} calculated (means [95% CI]; n=12). Contractions were normalized to % of those to KCl (60 mM).

Results: SP elicited concentration-related contractions of strips with an EC_{50} of 0.8nM [0.4-1.6] and E_{max} of 99% [93-106]. L-NAME (1 μM) failed to contract the strips under basal conditions, but contracted them when it was superfused 10 min after SP. This contractile response to L-NAME (1 μM) was dependent on the concentration of SP used for priming the tissue: an EC_{50} of 9.8nM [7.1-14.4] and an E_{max} of 72% [65-79] were calculated for this priming effect of SP. The MC stabilizers doxanzole (1 μM) and ketotifen (1 μM) prevented the contraction to L-NAME (1 μM) after SP (0.1 μM), but did not modify the direct contractile response to SP. Similarly, the MC activators compound 48/80 (10 μg /mL) and concanavalin A (20 μg /mL) did not impair the SP (0.1 μM)-induced contraction, but prevented SP-induced tissue priming, as L-NAME (1 μM) was unable to contract the strips 10 min after SP. However, under the present experimental conditions, these degranulators by themselves were found unable to mimic the priming effect of SP: i.e. L-NAME (1 μM) superfused after either compound 48/80 (10 μg /mL) or concanavalin A (20 μg /mL) did not contract the ileum strips.

Conclusion: SP exerts a priming effect on the guinea-pig ileum *in vitro* which, only after being stimulated by SP, contracts in response to L-NAME. We suggest that at least part of the mechanism of this intestinal muscle tissue sensitization to NOS inhibitors is mediated through the effects of SP on MCs.

ATTENUATION EFFECT OF PLURONIC L-81 ON SATIATION AFTER A LIPID MEAL IN RATS. K. Fujimoto, Y. Sakata, K. Okamoto, A. Ootani, P. Tso*. Division of Gastroenterology, Department of Internal Medicine, Saga Medical School, Saga, JAPAN, and * Department of Physiology, L.S.U. Medical Center, Shreveport, LA.

Pluronic L-81 (L-81) suppresses lipid transport into mesenteric lymph by preventing the formation of chylomicrons. In this study, we examined the effect of L-81 on satiation after intraduodenal infusion of triglyceride. **Methods:** Under unrestrained and unanesthetized condition, a lipid emulsion containing 13 μ moles of triolein, 2.6 μ moles of phosphatidylcholine and 29 μ moles of sodium taurocholate with or without 0.3 mg L-81 in 1 ml was infused into 24 h fasted rats intraduodenally at 3 ml/h for 15 min, 2 h, 4 h, and 8 h. Following the infusion, food intake during the first 30 min and blood samples were measured. **Results:**

Table. Time course of satiation of the lipid emulsion on 24 fasted rats.

0 min	Duration of infusion of the lipid meal			
	15 min	2 h	4 h	8 h
Without L-81 (Food intake, g/30 min)				
4.8±0.2	3.7±0.3 *	2.4±0.4 *	1.2±0.2 *	1.3±0.2 *
With L-81				
4.9±0.3	3.6±0.4 *	3.5±0.4 **	3.7±0.3 **	3.3±0.5 **

*-P<0.05, compared to 0 min after infusion. **-P<0.05, compared to the lipid without L-81.

Food intake after the 8-h lipid infusion decreased significantly (-75%) compared to rats infused with vehicle only, and the effect of the lipid meal was time-dependent. The satiation effect of lipid feeding was lost after L-81 treatment but food intake was still significantly decreased (-22%). The loss of inhibition of food intake by the feeding of lipid plus L-81 was totally reversed by cessation of L-81 infusion. In rats infused with the lipid emulsion, plasma level of triglyceride and apolipoprotein A-IV increased significantly. This increase was abolished by L-81. **Conclusion:** Judging from the results of L-81, we conclude that post-absorptive factors are important for anorexia observed after a lipid meal in addition to pre-absorptive factors. This study further supports our previous conclusion that apolipoprotein A-IV is an important factor for satiation after a lipid meal.

PNEUMATIC DILATATION (PD) VERSUS INTRASPHERICTIC BOTULINUM TOXIN (BT) IN THE TREATMENT OF ACHALASIA: PRELIMINARY RESULTS OF A CONTROLLED STUDY. M. Gaudin, R. Guimbaud, S. Chaussade, L. Palazzo, G. Quartier, J. Samama, B. Danel, D. Couturier; Service de Gastroenterologie Hôpital Cochin 27 rue du Fbg St Jacques 75014 PARIS. FRANCE.

DP of the gastric cardia represents an effective treatment for patients with achalasia. Good results occur in 70 to 85% of the cases. The perforation risk is 3 to 5%. Recently intrasphincteric injection of BT has shown to be effective without serious adverse effects.

The aim of the present study was to compare results of intrasphincteric BT versus PD

Patients and methods: 16 consecutive patients with achalasia were randomised to treatment with either injection of 80 UI of BT or PD with a 35 mm Rigiflex balloon. Patients were evaluated with a symptom score (0 to 9) and a manometry before treatment and 1 to 6 weeks after treatment.

Results: There was no complication in both groups. 12 patients were seen after treatment.

	symptom score before treatment	symptom score 1 month after treatment	LES pressure fall 1 month after treatment
PD (n=5)	5.6 ± 0.5	0.6 ± 0.9	53.6% (-35/+79)
BT (n=7)	5.7 ± 3.9	2.9 ± 2.1	31.8% (-6/+63)

Group PD: after the first dilatation 3/5 patients (100%) had a symptom score ≤ 2 ; 2/5 patients had a LES pressure fall < 50%, one of them had a second PD (40mm Rigiflex balloon) with a good clinical and manometric result.

Group BT: after the first injection 3/7 patients had a symptom score ≤ 2 ; 5/7 patients had a LES pressure fall < 50%, 4 of them had a second injection and 3 patients had finally to be operated owing to bad results.

Conclusion: These preliminary results concern a few number of patients. They still confirm the efficacy of intrasphincteric BT in the treatment of achalasia. Nevertheless intrasphincteric BT seems to be less effective compared to PD.

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I certify that the material herein has not been submitted to other DDW societies, will not be published as an article before May 19, 1996, and has not been presented at other U.S. national meetings.

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08 DEC. 1995

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I affirm that this abstract meets all of the requirements stated in this booklet, and that the authors named are familiar with the presented data, and that Box C has been completed. On behalf of all of the authors, I hereby transfer copyright in the above abstract to the American Gastroenterological Association.

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