volved in the development of the stress reaction in the animals with stable cardiovascular functions.

The altered electrolyte levels detected in this study in certain brain nuclei appear to be a reflection of profound metabolic changes in the cellular brain structures responsible for mediating the stress-producing stimulus. The measurements of K^+ and Na^+ concentrations performed in individual brain nuclei support the notion that characteristics of synaptic transmission are the major determinants of the cardiovascular system's resistance to stress.

REFERENCES

1. T. I. Belova and R. Kvetnanskii, *Usp. Fiziol. Nauk*, **12**, No. 2, 67-90 (1981).
2. T. I. Belova, N. V. Petrova, and Yu. Yunson, *Byull. Eksp. Biol. Med.*, **101**, No. 4, 385-387 (1986).
3. A. V. Gorbunova and S. I. Kashtanov, *Fiziol. Zh. SSSR*, **76**, No. 2, 176-183 (1990).
4. V. I. Ruban, E. G. Larskii, and L. S. Orlova, *Biochemical Methods of Investigation in the Clinical Setting* [in Russian], Saratov (1980).
5. E. A. Yumatov, T. I. Belova, L. S. Ulyaninskii, *et al.*, in: *Functional Systems of the Body* [in Russian], Moscow (1987), pp. 245-293.

Antiulcerogenic Effects of Benzylpenicillin in Acutely Stressed Rats

A. V. Kaluev, G. E. Samonina, and I. P. Ashmarin

UDC 615.281:616.33-002.44

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 118, № 8, pp. 131-133, August, 1994
Original article submitted December 6, 1993

The antibiotic benzylpenicillin was found to produce dose-dependent antiulcerogenic effects in rats when administered immediately before their exposure to acute stress (swimming for 1 h) that led to gastric mucosal ulceration. Such effects were not observed in rats given benzylpenicillin 48 h before stress exposure. The results of this study suggest that in acutely stressed animals benzylpenicillin may activate as yet unidentified mechanisms which afford protection to the gastric mucosa and which are not associated with the longer-lasting antimicrobial effects of this antibiotic.

Key Words: *ulcerogenesis; acute stress; benzylpenicillin*

Stress-induced gastric mucosal lesions have customarily been considered to result from an imbalance between protective and aggressive factors and to involve directly a number of neurohumoral mechanisms [7,9]. The properties of various pharmacological agents that influence neurohumoral processes and ulcerogenesis are therefore under intensive study [4,6]. Some benefit in the treatment of gastric ulcer is afforded by certain antibiotics, including benzylpenicillin (BP), which is usually attributed to their antimicrobial properties - their action on microorganisms which have recently been incriminated as possible culprits in the pathogenesis of gastric mucosal lesions [15]. There is also evidence that BP possesses neurotropic properties [1,3,5,8,13]. This calls for studies to find out whether BP and its analogs can exert antiulcerogenic effects via central and/or peripheral mechanisms.

MATERIALS AND METHODS

A total of 240 random-bred white rats of both sexes weighing 150-200 g were used. Before the experiments they had been fasted for 48 h but given free access to water. Immediately (1 min) or 48 h before exposure to acute stress, test rats were injected intraperitoneally with 0.2 ml of BP so-

Chair of Human and Animal Physiology, Department of Biology, Moscow State University

0007-4888/94/0008-0818$12.50

dium in 0.5% Novocain solution, while control rats received the same volume of 0.5% Novocain by the same route. The BP dose of 10,000 U/kg - a dose level close to the daily therapeutic dosage of this drug for humans - was taken as the single standard dose (SSD), and doses equaling 1, 5, 10, and 20 SSD were used. Acute stress was produced in all animals by forcing them to swim in a pool for 1 h at 20°C. Thereafter they were killed with ether, their stomachs were extracted, dissected along the lesser curvature, washed with water, and stretched out on a support, and the state of the gastric mucosas was evaluated under a binocular magnifying glass. The following indicators of mucosal injury were recorded for each stomach: the total area of punctate ulcerous lesions (in mm^2, the area of each lesion being taken as equal to 1 mm^2), the total area of local ulcers (in mm^2), and, in the case of linear ulcers, the ulcer length (in mm), taking the ulcer width to be equal to 1 mm. For the overall evaluation of gastric mucosal lesions, an ulcer index was used, defined as the sum of the areas (in mm^2) of punctate and local ulcers per stomach. The average ulcer index was then calculated for each group of rats. The results were treated statistically by the nonparametric Wilcoxon-Mann-Whitney test [2].

RESULTS

Gastric mucosal lesions in the groups of rats injected with BP intraperitoneally immediately before exposure to stress were found to be much less pronounced than those in the control groups, the antiulcerogenic effect of BP being clearly dose-dependent (Table 1 and Fig. 1). Differences between the test and control groups in values of the ulcer index are significant and indicate that BP rapidly afforded protection against gastric mucosal damage in acutely stressed rats. When injected 48 h before exposure to stress, however, BP failed to exhibit antiulcerogenic activity (Fig. 2).

Following parenteral administration, the blood concentration of BP reaches its peak by minutes 20-30, is maintained at a constant level for 3-4 h, and then declines rapidly [4]. In our experiments, therefore, the antiulcerogenic action of BP lasted for not more than 40 min, during which time the drug was distributed to various organs and tissues. It should be noted that there is a time interval during which two oppositely directed events - an increase in the antimicrobial activity of administered BP and a decrease in its short-term antistress activity - are unlikely to overlap. The antiulcerogenic effect of BP we observed indicates that, in addition to its well-known antibiotic properties, this drug also possesses the property of rapidly activating protective mechanisms in stress-induced ulcerogenesis. The time course of this activation suggests that BP probably has neuroregulatory (central or peripheral) antiulcerogenic effects. As a matter of fact, BP is reported to be able to alter the level of gastric secretion [5] and to influence central and peripheral neural mechanisms [3,8]. Of interest in this connection is the evidence implicating gamma-aminobutyric acid

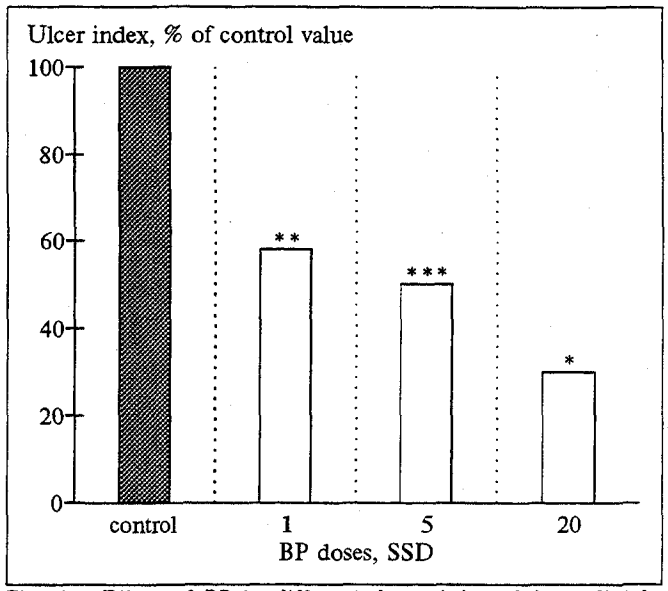

Fig. 1. Effect of BP in different doses injected immediately before stress on the ulcer index. One, two, and three asterisks: significant difference from control rats at $p<0.01$, $p<0.005$, and $p<0.0001$, respectively.

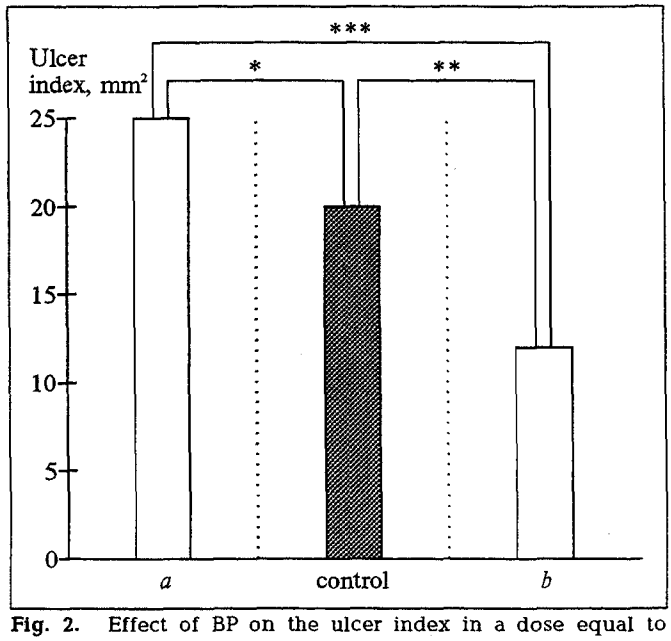

Fig. 2. Effect of BP on the ulcer index in a dose equal to 10 SSD injected 48 h (a) and 1 min (b) before stress. One, two, and three asterisks indicate statistical differences at $p>0.05$, $p<0.05$, and $p<0.0001$, respectively.

TABLE 1. Effect of BP on Gastric Mucosal Lesions in Rats Acutely Stressed by Swimming ($M \pm m$)

Series	No. of rats		Time of BP injection and No. of injected SSD				Ulcer index		p
			48 h before stress		1 min before stress				
	test group	control group	test group	control group	test group	control group	test group	control group	
1	30	30	–	–	1	*	11±2	19±3	<0.005
2	45	45	–	–	5	*	7±1	15±4	<0.0001
3	20	20	*	*	10	*	12±3	20±4	<0.05
	20		10	*	*	*	25±4	20±4	>0.05
4	15	15	–	–	20	*	6±2	21±4	<0.01

** <0.0001

Note. One asterisk: rats given Novocain solution without BP; two asterisks: a significant difference between the two test groups at $p<0.0001$.

(GABA) in the pathogenesis of stress-produced gastric and duodenal mucosal lesions when central and peripheral GABA-ergic mechanisms are activated [10-12,14]. In particular, BP is known to be a specific blocker of GABA-ergic transmission [1,13], which plays a key role in stress. One cannot, however, rule out the possibility that the antiulcerogenic action of BP involves some other mechanisms. It should be emphasized that in our study the antiulcerogenic activity of BP was short-lived and disappeared within 48 h postinjection (Fig. 2).

In acute stress BP may be presumed to activate some rapid protective antiulcerogenic mechanisms not associated with its direct antibiotic effects and, possibly, some central or peripheral neural mechanisms.

REFERENCES

1. I. V. Batueva, N. P. Veselkin, and R. Veskov, *Neirofiziologiya*, **24**, № 2, 151-160 (1992).
2. E. V. Gubler and A. A. Genkin, *Use of Nonparametric Statistical Tests in Biomedical Research* [in Russian], Leningrad (1973).
3. E. L. Dovedova, *Neirokhimiya*, **8**, № 1, 87-94 (1989).
4. M. D. Mashkovskii, *Medicines* [in Russian], Moscow (1987).
5. G. I. Mirziashvili, *On Neural Mechanisms Mediating the Action of Penicillin on the Secretory Activity of Major Digestive Glands (Author's synopsis of dissertation)* [in Russian], Tbilisi (1967).
6. N. A. Natsvlishvili, G. E. Samonina, and I. P. Ashmarin, *Byull. Eksp. Biol. Med.*, **112**, № 7, 9-10 (1991).
7. Ya. S. Tsimmerman, *Klin. Med.*, № 1, 6-11 (1993).
8. A. B. Chernomordik, *Ibid.*, № 2, 102-105 (1980).
9. F. P. Brooks, in: *Peptic Ulcer Disease*, № 3, Edinburgh-London (1985), pp. 45-149.
10. N. Hara, Y. Hara, Y. Natsume, and Y. Goto, *Nippon Yakurigaku Zasshi*, **96**, № 4, 163-168 (1990).
11. N. Hara, Y. Hara, Y. Natsume, and Y. Goto, *Europ. J. Pharmacol.*, **194**, № 1, 77-81 (1991).
12. S. Hara, S. Yanagihara, K. Saton, and S. Morioka, *Jap. J. Pharmacol.*, **47**, № 4, 333-340 (1988).
13. A. Horne, K. Hadingham, A. Macaulay, *et al.*, *Brit. J. Pharmacol.*, **107**, № 3, 732-737 (1992).
14. A. Krantis, *Dig. Dis. Sci.*, **38**, № 8, 1211-1216 (1989).
15. K. K. Wang, *Compr. Ther.*, **17**, № 9, 35-38 (1991).