

Pentadecapeptide BPC 157 positively affects both non-steroidal anti-inflammatory agent-induced gastrointestinal lesions and adjuvant arthritis in rats

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Summary — Besides a superior protection of the pentadecapeptide BPC 157 (an essential fragment of an organoprotective gastric juice peptide BPC) against different gastrointestinal and liver lesions, an acute anti-inflammatory and analgetic activity was also noted. Consequently, its effect on chronic inflammation lesions, such as adjuvant arthritis, and non-steroidal anti-inflammatory agents (NSAIDs)-induced gastrointestinal lesions was simultaneously studied in rats. In gastrointestinal lesions (indomethacin (30 mg/kg sc), aspirin (400 mg/kg ig) and diclofenac (125 mg/kg ip) studies, BPC 157 (10 µg or 10 ng/kg ip) was regularly given simultaneously and/or 1 h prior to drug application (indomethacin). In the adjuvant arthritis (tail-application of 0.2 mL of Freund's adjuvant) studies (14 days, 30 days, 1 year) BPC 157 (10 µg or 10 ng/kg ip), it was given as a single application (at 1 h either before or following the application of Freund's adjuvant) or in a once daily regimen (0–14th day, 14–30th day, 14th day–1 year). Given with the investigated NSAIDs, BPC 157 consistently reduced the otherwise prominent lesions in the stomach of the control rats, as well as the lesions in the small intestine in the indomethacin groups. In the adjuvant arthritis studies, the lesion's development seems to be considerably reduced after single pentadecapeptide medication, and even more attenuated in rats daily treated with BPC 157. As a therapy of already established adjuvant arthritis, its salutary effect consistently appeared already after 2 weeks of medication and it could be clearly seen also after 1 year of application. Taking together all these results, the data likely point to a special anti-inflammatory and mucosal integrity protective effect.

pentadecapeptide BPC 157 / peptide BPC / gastrointestinal lesions / adjuvant arthritis

Introduction

Because of the likely physiologic significance of gut peptides and as yet not completely explored possible therapeutic applications, the discovery of so far unknown peptides, their original structure and saving actions, have received considerable attention (Thompson *et al*, 1987; Guglietta, 1992). We recently identified a gastric juice protein with mucosal protective properties as well as huge range of organoprotective effects, M_r 40 000 (determined by gel chromatography), code-named BPC (Sikiric *et al*, 1993a, b, c, 1994, 1995, 1996a, b). In line with this, a 15 amino acid fragment (BPC 157), with apparently no sequence homology with known gut peptides, thought to be essential for activity of the entire peptide, was synthesized and characterized (Sikiric *et al*, 1993a, b, c, d, 1994, 1995, 1996a, b). This protective property was independently investigated and confirmed by others (Paré and Klucyznski, 1994; Bosnjak *et al*, 1994; Veljaca *et al*, 1994a, b, 1995a, b; Sandor *et al*, 1996). In toxicology studies, no death or pathologic changes were observed despite the administration of a very high dose (Sikiric *et al*, 1993c). When incubated in human gastric juice or in water, this pentadecapep-

ptide was not subjected to any degradation during 24 h, unlike h-EGF and h-TGF α , stable in water, but degraded in human gastric juice already after 15 min (Veljaca *et al*, 1995a).

Therefore, the recent focus was on the mentioned pentadecapeptide, BPC 157, and its effect on gastrointestinal lesions was successfully evaluated in different experimental models (Paré and Klucyznski, 1992; Sikiric *et al*, 1993a, b, c, d, 1994, 1995, 1996; Bosnjak *et al*, 1994; Veljaca *et al*, 1994a, b, 1995a, b; Sandor *et al*, 1996) in comparison with conventional agents and other gut peptides. The noted positive effects of BPC 157 and other gut peptides (secretin, glucagon and NPY) were in keeping with the suggested peptide's physiologic significance and BPC's importance (Paré and Klucyznski, 1992; Sikiric *et al*, 1993a, b, c, d, 1994, 1995, 1996a, b; Bosnjak *et al*, 1994; Veljaca *et al*, 1994a, b, 1995a, b; Sandor *et al*, 1996). Of note, besides a superior BPC 157 protection against different gastrointestinal and liver lesions, an acute anti-inflammatory and analgetic activity was also noted (Paré and Klucyznski, 1992; Sikiric *et al*, 1993a, b, c, d, 1994, 1995, 1996a, b; Bosnjak *et al*, 1994; Veljaca *et al*, 1994a, b, 1995a, b; Sandor *et al*, 1996). Consequently, the possibility that a similar

beneficial effect could be seen also in chronic inflammation lesions, such as adjuvant arthritis in rats, is discussed here. Because of the significance of gastrointestinal disturbances provoked by anti-rheumatic medication (Shen, 1977, 1984; Caruso and Bianci Porro, 1980; Doherty *et al.*, 1987; Festen, 1988; Sikiric *et al.*, 1988; Rainsford, 1992) the effect on non-steroidal anti-inflammatory agents (NSAIAs)-induced gastrointestinal lesion was simultaneously studied as well.

Materials and methods

Preparation of BPC 157

Pentadecapeptide BPC 157 (Gly-Glu-Pro-Pro-Gly-Lys-Pro-Ala-Asp-Asp-Ala-Gly-Leu-Val, M_r 1419) is a partial sequence of human gastric juice protein BPC, freely soluble in water at pH 7.0 and in saline, prepared as described before (Sikiric *et al.*, 1993a, b, c, 1994, 1995, 1996a, b). Peptide with 99% (HPLC) purity (1-des-Gly peptide as impurity), dissolved in saline, was used in all experiments (Sikiric *et al.*, 1993a, b, c, d, 1994, 1995, 1996a, b).

Experimental procedures

Animals

Wistar male (NSAIA-studies) or female (adjuvant arthritis-studies) albino rats randomly assigned, 10–16 rats per group, weighing 200–220 g, were used in all experiments. For ulcerogenic procedures, food and water were withdrawn 24 or 2 h before ulcerogenic procedure.

Ulcer models

Aspirin lesions. Aspirin (Aspirin, Pliva, Zagreb, Croatia) suspended in 1% carboxymethyl cellulose in water (20 mg/mL), was applied in a dose of the 400.0 mg/kg ig. BPC 157 (10.0 µg or 10.0 ng/kg ip) or an equivolume of saline (5 mL/kg bw ip) was simultaneously administered with aspirin.

Diclofenac lesions. Diclofenac (Voltaren, Pliva, Zagreb, Croatia) dissolved in saline was applied in a dose of the 125 mg/kg bw ip. BPC 157 (10.0 µg or 10.0 ng/kg ip) or an equivolume of saline (5 mL/kg bw ip) was simultaneously applied with diclofenac.

Indomethacin lesions. Indomethacin (Indocid, Lek, Slovenia) suspended in 1% carboxymethyl cellulose solution, was given in a dose of 30 mg/kg bw sc. Relative to indomethacin, saline (controls) or BPC 157 (10 µg or 10 ng/kg bw ip) was applied as pre- (1 h before) or co- (simultaneously) treatment.

Assessment of mucosal injury. Immediately after killing, the stomach and small intestine (in the case of indomethacin medication) were removed and the lesions (means ± SEM, mm) were assessed by independent observers as described before (Sikiric *et al.*, 1988, 1993c, 1994, 1995, 1996a, b). Representative sections of the stomach and intestine were processed for further histologic analysis. The ani-

mals were killed 2 h or 4 h after aspirin; following indomethacin or diclofenac, they were killed 24 h later.

Adjuvant arthritis

All the rats received a tail-application of 0.2 mL of Freund's adjuvant (Freund's complete adjuvant, Sigma, USA). BPC (10 µg or 10 ng/kg bw ip) was applied using different protocols: i) 1 h before Freund's adjuvant application; ii) 1 h after Freund's adjuvant application; iii) 1 h after Freund's adjuvant application and subsequently once daily for 2 weeks; and iv) once daily for 2 weeks or 1 year starting from the 14th day after Freund's adjuvant application. Control animals simultaneously received an equivolume of 0.9% NaCl (5.0 mL/kg bw). The animals were killed either after 2 or 4 weeks following Freund's adjuvant application or 1 year starting from the 14th day after Freund's adjuvant application.

Assessment of adjuvant arthritis. Assessment of paw inflammation was done by an independent observer. A macroscopical score of 0 to 3 was used in the following manner. In all paws an assessment including paw thickness and presence of macroscopic arthritic changes (nodes and stiffness) was carried out (once daily till the end of the first month, and once weekly during the rest of the experiment). Healthy animals were used as a control and were given a basal value of 0 (no change); 1, mild; 2, moderate; 3, severe changes. Metatarsal and tarsal joint were fixed in formaline and after decalcination embedded in paraffin and stained according to the H&E method. Pathohistologically, several characteristics were scored (synovitis, arthritis and peri-arthritis, mononuclear cells infiltrations and periostitis with formation of the periosteal new bone) as follows: 0, no change; 1, mild; 2, moderate; 3, severe. Mean sum of the scores was statistically evaluated, using a modification of a previously described system (Glenn and Grey, 1965).

Statistical analysis

Kolmogorov-Smirnov test was performed for estimation of the normality of the data distribution. Further statistical analyses were performed by means of analysis of variance (ANOVA) and/or Kruskal-Wallis, Mann-Whitney tests, Student-Newman-Keuls, Dunn's and Dunnett's tests. Differences of 0.05 or less were considered to be statistically significant.

Results

The effects of pentadecapeptide on NSAIAs induced mucosal lesions

Indomethacin lesions

Indomethacin given in a relatively high dose consistently produced strong lesions in the stomach and small intestine in all of the controls (figs 1, 2). Pentadecapeptide BPC 157 applied simultaneously with indomethacin strongly reduced otherwise severe lesions in both the stomach and small intestine, an effect clearly seen in either µg or ng/kg range (figs 1, 2). Like-

wise, a similar positive effect was obtained when BPC 157 was given 1 h before indomethacin administration.

Aspirin lesions

Significant gastric lesions were noted in the control rats following 2 or 4 h after aspirin administration. BPC 157 applied simultaneously with aspirin markedly reduced the lesions appearance. Of note, a positive effect was noted after both doses, 10 μ g or 10 ng/kg bw, in both tested intervals (fig 1).

Diclofenac lesions

The application of diclofenac produced also significant stomach lesions in the control animals. BPC

157 given simultaneously with diclofenac significantly reduced the lesion's development. This effect appears to be dose-dependent (fig 1).

Microscopy investigations

Generally, microscopical data were in full agreement with macroscopic observations. In the groups treated with indomethacin, aspirin or diclofenac, the depth of the lesions varied from mild superficial epithelial damage to the extensive necrosis to all parts of the mucosa. The edges of the stomach lesions were sharp, and the bases of the lesions were infiltrated with granulocytes. The base of the lesions was saturated with blood while granulocytes permeated the rest of the intestinal wall up to the

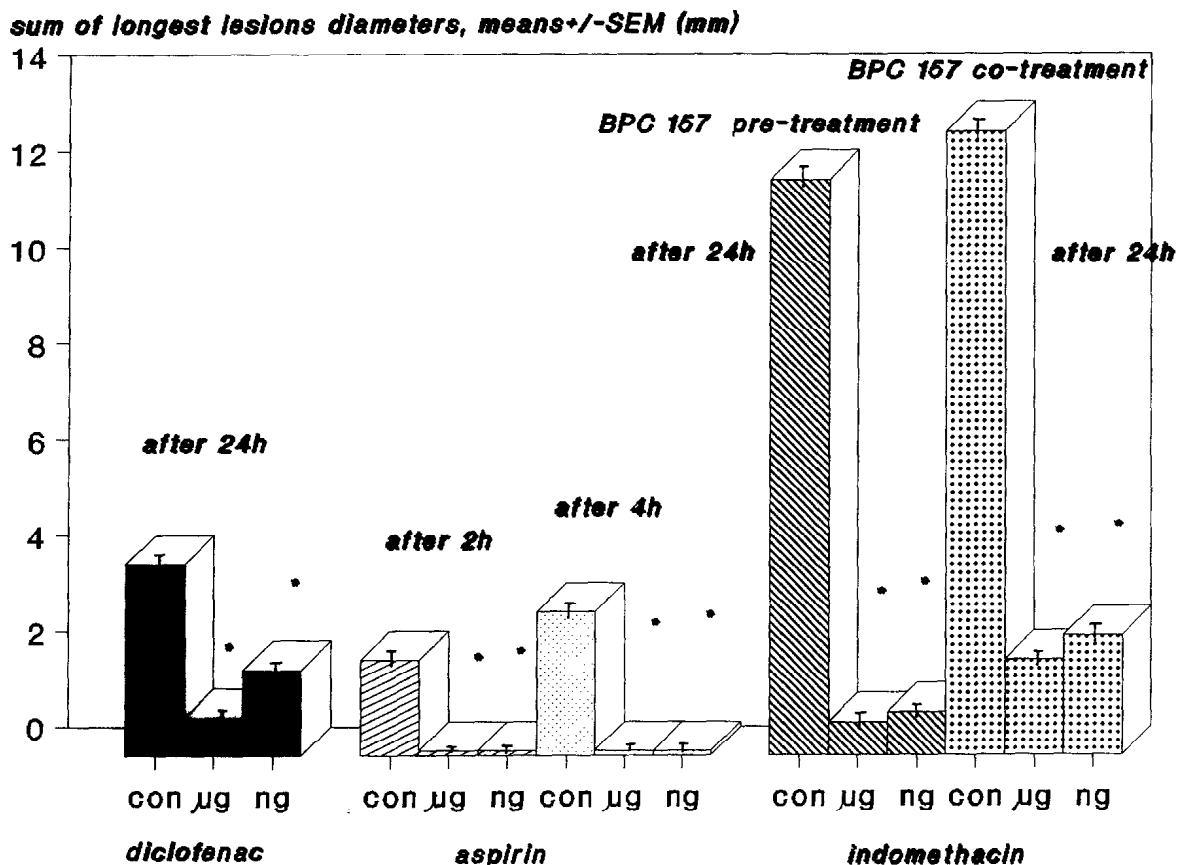


Fig 1. The effect of BPC 157 administration on the development of gastric lesions induced by application of aspirin (400 mg/kg ip), diclofenac (100 mg/kg ip) and indomethacin (30 mg/kg sc). BPC 157 (10 μ g or 10 ng/kg ip) or saline (5 mL/kg) (control) were applied simultaneously (or also 1 h before indomethacin) with noxious agent application. Rats were killed after either 2 or 4 h (aspirin) or 24 h (diclofenac and indomethacin). * $P < 0.05$, at least, vs corresponding control. $n = 6-10$ rats per experimental group.

sum of longest lesions diameters, means \pm SEM, (mm)

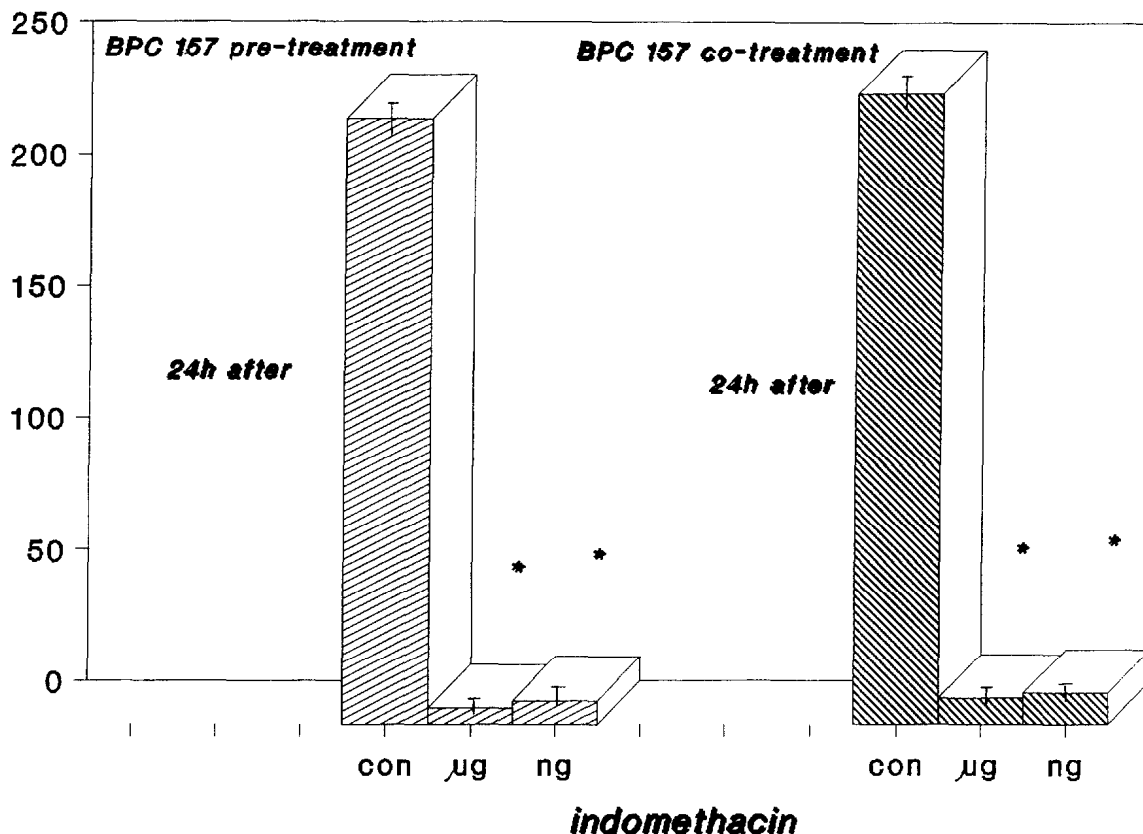


Fig 2. The effect of BPC 157 administration on the development of small intestine lesions induced by application of indomethacin (30 mg/kg sc). BPC 157 (10 µg or 10 ng/kg ip) or saline (5 mL/kg) (control) were applied simultaneously (or 1 h before) with noxious agent application. Rats were killed 24 h after indomethacin application. * $P < 0.05$, at least, vs corresponding control. $n = 6-10$ rats per experimental group.

serosa. Of note, these changes were much less pronounced in BPC 157 treated animals than in corresponding controls.

The effects of BPC 157 on adjuvant arthritis lesions
In the adjuvant arthritis studies, it seems that a favorable effect could be observed throughout the entire experiment.

Development of adjuvant arthritis
Concerning the first phase, the development of adjuvant arthritis after Freund's adjuvant application was clearly shown in controls, during the initial 2 weeks, macroscopically by a prominent paw swelling, nodes and stiffness, as well microscopically by the inflammatory periosteal reactions with accumulations of mononuclear cell infiltrate. It seems interesting that BPC 157 seems to be already effective

when applied prophylactically as only one single application. This effect could be seen when BPC 157 had been given at 1 h either before or following Freund's adjuvant application. This initially noted activity could be likely increased. Supportingly, if BPC 157 was continuously applied as a once daily medication during this phase, the lower dose of the 10 ng/kg also became significantly effective when assessed both macro- and microscopically, unlike single application regimen (effect evident only in macroscopical assessment) (fig 3).

Interestingly, besides the effect on adjuvant arthritic lesion development, an additional effect seems to be evident on general animal conditions as well. Namely, an obvious decrease of body weight from the initiation of the experiment was noted (data not shown). Animals treated with ng-regimens kept the approximately the same weight as before, whereas

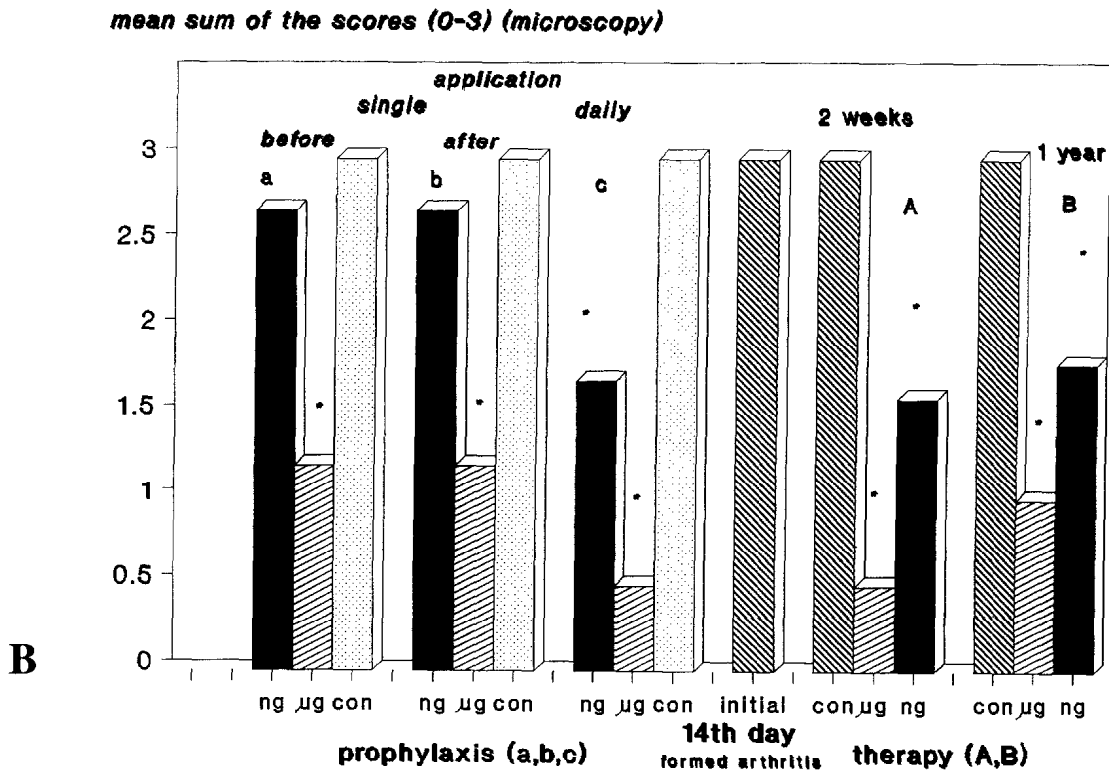
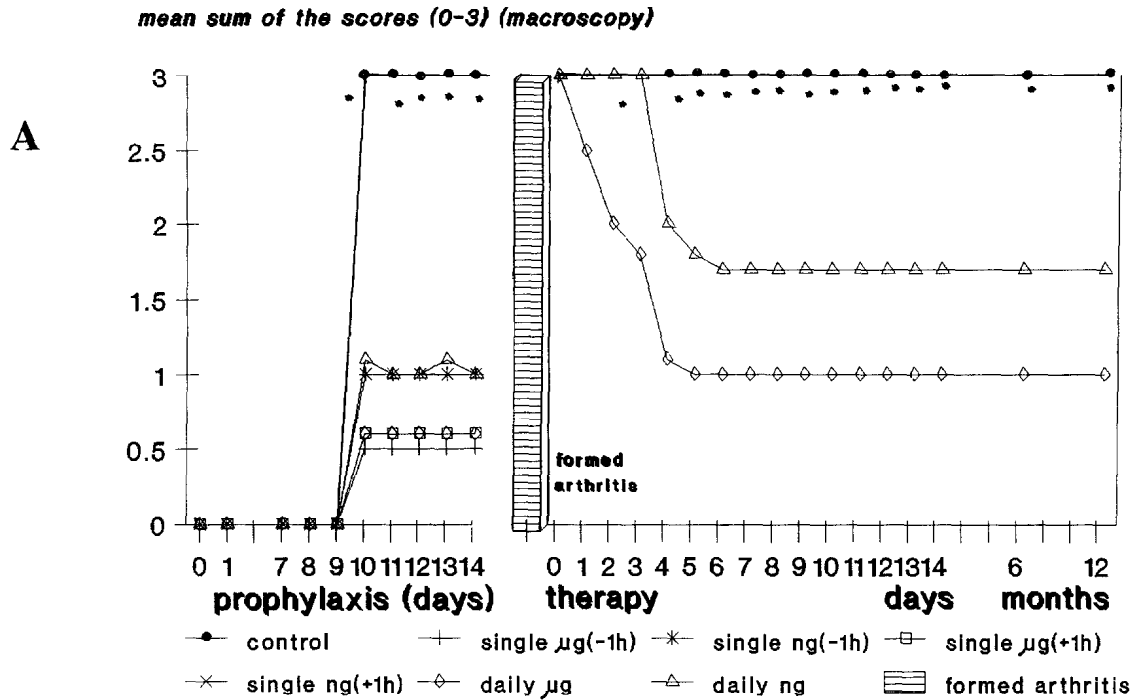


Fig 3. The effect of BPC 157 administration on adjuvant arthritis after Freund's adjuvant application, assessed macroscopically (a, top) and microscopically (b, bottom): on adjuvant arthritis development during the initial 2 weeks (left, prophylaxis, a, b, c) and thereafter (right, therapy, A, B), on already established adjuvant arthritis following 2 weeks or 1 year of medication. BPC 157 (10 μg or 10 ng/kg ip) or saline (5 mL/kg) were given at 1 h either before (a) or after (b) Freund's adjuvant application or it was continuously applied prophylactically as a once daily medication during adjuvant arthritis development phase (c), and later, therapeutically, starting in the conditions of the already established adjuvant arthritis, as a 2-week (A)- or one-year (B) treatment. * $P < 0.05$, at least, vs corresponding control, † $P < 0.05$, at least vs corresponding group. $n = 6-10$ rats per experimental group.

a normal increase in the body weight, similar to the weight gain of the corresponding healthy animals, could be seen in the μg groups.

Established adjuvant arthritis

Likewise, such a favorable activity seems to be even more prominent when BPC 157 was applied later, in the conditions of the already established adjuvant arthritis. In these animals, besides a prominent paw thickness, and arthritic nodes and stiffness, microscopical investigations showed chronic changes of the tibia, tarsal and metatarsals with chronic synovitis and periostitis characterized by pannus formation and formations of periosteal new bone. Unlike continuous presence of adjuvant arthritis in controls, the beneficial effect following BPC 157 medication could be seen either after a short, or a long lasting period.

Short-time treatment. The salutary effect seems to prominently occur within a few days as could be assumed on the basis of our macroscopical observations. Supportingly, microscopically, it is already fully present in the animals killed following 2 weeks of medication, since a clear (and dose dependent) favorable activity was noted in all rats treated with either μg or ng BPC 157 regimen (fig 3).

Long-lasting treatment. Obviously, the pentadecapeptide BPC 157 activity seems to be long-lasting, clearly seen following an entire year of medication (fig 3). In animals continuously treated with BPC 157 since the establishment of adjuvant arthritis following 2 weeks after Freund's adjuvant application, a prominent salutary (and dose-dependent) effect was obtained in groups receiving either μg or ng BPC 157 regimen (fig 3).

Discussion

These data provide compelling evidence that the lesions induced by either different NSAIA application in the gastrointestinal tract or adjuvant arthritis in rats could be positively influenced by the administration of the pentadecapeptide BPC 157. It is of interest that this agent protects against both these quite different forms of injury. However, the possible mechanisms remain to be further elucidated.

In generally, two distinct mechanisms, inflammation and delayed hypersensitivity, are accepted to be involved in mediation of adjuvant arthritis (Pearson, 1956, 1959; Newbould, 1963; Glenn and Grey, 1965; Brown *et al.*, 1971; Perper *et al.*, 1971; Shen,

1977, 1984). Theoretically, BPC 157 could effect one of them, or both.

Regarding the suggested anti-inflammatory effect, BPC 157 was effective in the models of acute/subacute inflammation and inflammatory/non-inflammatory pain (*eg* turpentine, carrageenin, acetic acid or MgSO_4 writhing (prostaglandin-dependent, prostaglandin-non dependent) tail-pinching tests) (Winter *et al.*, 1962; Damas and Deflandre, 1984; Gyires and Torma, 1984; Willoughby *et al.*, 1986; Sikiric *et al.*, 1993a, b). The used protocols (Gyires and Torma, 1984; Sikiric *et al.*, 1993b, c) were similar to those applied in the present report. Relative to reference standards (*ie* aspirin, indomethacin), BPC 157 was effective in considerable lower doses ($\mu\text{g}/\text{ng}/\text{kg}$ vs mg/kg) (Gyires and Torma, 1984; Sikiric *et al.*, 1993b, c). Along with this was also the noted antipyretic BPC activity (Gyires and Torma, 1984; Sikiric *et al.*, 1993b, c). Thus, it seems likely that BPC 157 has a marked anti-inflammatory capacity.

Concerning the possible BPC 157 effect on delayed hypersensitivity, a positive influence was also evidenced. Recently, we developed a new murine model of inflammatory bowel disease (Brkic *et al.*, 1992a, b; Banic *et al.*, 1993; Sikiric *et al.*, 1993c). Besides an irritative effect, the challenge dose of 2,4-dinitrofluorobenzene applied into the lumen of the colon resulted in specific T-cell mediated injury of the colonic and ileal mucosa (Brkic *et al.*, 1992a, b; Banic *et al.*, 1993; Sikiric *et al.*, 1993c). Like methylprednisolone, a widely used therapeutic drug (Brkic *et al.*, 1992a, 1992b, c; Banic *et al.*, 1993; Sikiric *et al.*, 1993c), BPC 157, applied in the dosage used in the present study, strongly reduced the ulcerations, hemorrhage, edema and particularly mononuclear cell infiltration of the inflamed intestine (Sikiric *et al.*, 1993c). Similarly, in another model of acute and chronic colonic inflammation, application of trinitrobenzene-sulfonic acid (TNBS) in rats (Morris *et al.*, 1984), a prominent lesion reduction was observed in TNBS-rats treated with BPC pentadecapeptide (Veljaca *et al.*, 1994a, b, 1995b). BPC 157 was shown to reduce LTB_4 and TXB_2 serum and inflamed tissue levels (Veljaca *et al.*, 1994a, 1995b). This pentadecapeptide also decreased MPO level in inflamed tissues (Veljaca *et al.*, 1994a). In these studies (Sikiric *et al.*, 1993c; Veljaca *et al.*, 1994a, b, 1995b), an apparently same range of doses as in the present report was used. In analogy, the salutary BPC 157 effects in adjuvant arthritis lesions (*ie* less synovitis, arthritis and peri-arthritis, mononuclear cells infiltrations and periostitis with formation of the periosteal new bone, less paw swelling, nodes and stiffness) could be also interpreted as a likely consequence of a beneficial in-

fluence on delayed type of hypersensitivity reactions.

How to combine these findings with the known effectiveness of the drugs therapeutically used in rheumatic disorders (Pearson, 1956, 1959; Newbould, 1963; Glenn and Grey, 1965; Brown *et al.*, 1971; Perper *et al.*, 1971; Shen, 1977, 1984)? From this point of view, the noted BPC 157 effectiveness in adjuvant arthritis (prophylactic/therapeutic effect, non-established/established adjuvant arthritis) seems to be not observed with the present therapeutic agents. Glucocorticoids are effective in adjuvant arthritis prevention when daily applied, but not in short treatment, immunosuppressants (in high dosage) and non-steroidal analgetics only in pre-treatment or post-treatment respectively (Pearson, 1956, 1959; Newbould, 1963; Glenn and Grey, 1965; Brown *et al.*, 1971; Perper *et al.*, 1971; Shen, 1977, 1984). Thus, it could be speculated that a particular combination of their mechanisms (regardless of the not fully defined) is involved in the BPC beneficial activity in adjuvant arthritis lesions.

In this, whether and how BPC 157 would affect the initial immune response, remains to be seen. However, a beneficial effect on adjuvant arthritis development is noted already after a single BPC 157 application, given 1 h before or after Freund's adjuvant application. Consequently, an inhibition or interference with the initial immune phenomena involved in this delayed hypersensitivity reaction, most probably lymphocytes, known to be of considerable importance respectively (Pearson, 1956, 1959; Newbould, 1963; Brown *et al.*, 1971; Perper *et al.*, 1971) for inductive phase of immunologic response, could be likely suggested. This initial effectiveness in adjuvant arthritis prevention could be even enhanced with its daily application. Higher (10 µg/kg) dosage was effective either after single or daily regimen, whereas lower (10 ng/kg) dose, previously ineffective given as a single application, becomes efficacious applied as a daily regimen. This finding in conjecture with a therapeutic effect of BPC 157 on fully established arthritis, favors its further beneficial role in the entire course of adjuvant arthritis.

However, despite the presented evidence, the precise mechanism of the BPC 157 efficacy in adjuvant arthritis remains still elusive. Theoretically, despite general limitations (Pearson, 1956, 1959; Winter *et al.*, 1962; Newbould, 1963; Glenn and Grey, 1965; Brown *et al.*, 1971; Perper *et al.*, 1971; Shen, 1977, 1984; Damas and Deflandre, 1984; Gyires and Torma, 1984; Willoughby *et al.*, 1986), a possible solution could be further searched within generally known complexity of processes involved in the inflammation and different systems' interactions in the

various models of inflammation (Pearson, 1956, 1959; Winter *et al.*, 1962; Newbould, 1963; Glenn and Grey, 1965; Brown *et al.*, 1971; Perper *et al.*, 1971; Shen, 1977, 1984; Damas and Deflandre, 1984; Gyires and Torma, 1984; Willoughby *et al.*, 1986). For instance, the histamine, 5-HT, bradykinin and prostaglandin systems are sequentially involved in the carrageenin edema formation (Damas and Deflandre, 1984), but the prostaglandins are widely accepted to be very important (Shen, 1977, 1984). Concerning their involvement in rheumatic processes and prostaglandin-synthesis inhibitors, besides cyclo-oxygenase inhibition, an inhibition of other products of arachidonic acid seems to be even more therapeutically important (Shen, 1977, 1984). Thus, the mentioned inhibitory effect of BPC 157 on inflammatory mediators, MPO, LTB₄ and TXB₂ (Veljaca *et al.*, 1994a, b, 1995b) could provide an explanation for the observed efficacy. For instance, pentadecapeptide BPC is also able to dose-dependently reduce the release of LTB₄ in human blood stimulated by a calcium ionophore A23187 (Veljaca *et al.*, 1995b). Besides this, there is a likely possibility that some of the beneficial effects of BPC 157 in adjuvant arthritis could be (directly and/or indirectly) mediated by glucocorticoids. BPC peptide beneficial activity (*eg* gastrointestinal tract protection) would disappear after adrenalectomy in rats (Sikiric *et al.*, 1993b). Applied in stress conditions, besides mucosal integrity of gastrointestinal tract, pentadecapeptide BPC 157 would protect the adrenal gland as well (Paré and Klucyznski, 1994; Sikiric *et al.*, 1993b).

Of note, these findings have to be viewed in the light of the salutary effects noted in gastrointestinal tract lesions, particularly those induced by NSAIA applications. An overproduction of leucotriens, as a consequence of cyclo-oxygenase inhibition by NSAIA, was convincingly implicated in pathogenesis of NSAIA-GI tract ulcers. As mentioned, BPC 157 induced modulation (and/or inhibition) of a damaging overactivity of leucotriens and other inflammatory mediators (*ie* MPO, TXB₂) was noted in inflamed animal tissues and human blood (Veljaca *et al.*, 1994a, b, 1995b). Whether this could provide a common explanation for this pentadecapeptide salutary effect on NSAIA-gastrointestinal tract lesions as well as adjuvant arthritis remains to be seen. In any case, a BPC 157-beneficial effect on NSAIA-gastrointestinal tract lesions could be clearly expected. Besides mentioned salutary effect in colitis induced by DNFB or TNBS (Sikiric *et al.*, 1993c; Veljaca *et al.*, 1994a, b, 1995b) a strong protective effect was demonstrated on ethanol and restraint stress gastric lesions (Paré and Klucyznski, 1994; Sikiric *et al.*, 1993c, 1994, 1996a, b; Sandor *et al.*,

1996) and cysteamine duodenal ulcers (Sikiric *et al.*, 1994; Sandor *et al.*, 1996). These effects were noted after either parenteral or peroral application, a finding in keeping with a particular stability of this pentadecapeptide in human gastric juice (Paré and Klucyznski, 1994; Sikiric *et al.*, 1993, 1994, 1996a, b; Veljaca *et al.*, 1995a; Sandor *et al.*, 1996). In line with the original isolation of BPC peptide from gastric juice, and its stomach origin (Sikiric *et al.*, 1993a, b, c, d, 1994, 1995, 1996a, b), this pentadecapeptide, incubated in human gastric juice or in water, was not subjected to any degradation during 24 h, unlike h-EGF and h-TGF α , stable in water, but degraded in gastric juice already after 15 min (Veljaca *et al.*, 1995a).

Additionally, the described BPC 157 effects in the present report seem to be at least partly shared also by drugs therapeutically used in gastrointestinal ulcers or rheumatic disorders (Robert, 1979; Hernandez *et al.*, 1984; Kuwayama and Eastwood, 1985; Malone *et al.*, 1991; Marabini *et al.*, 1991; Robert *et al.*, 1991; Tascilar *et al.*, 1993). Besides improving gastrointestinal disturbances in rheumatic patients with NSAIA therapy, several antiulcer drugs would also favorably affect the rheumatic disorders as well (Malone *et al.*, 1991; Marabini *et al.*, 1991; Tascilar *et al.*, 1993). For instance, misoprostol increased the anti-inflammatory effect of acetylsalicylic acid in adjuvant arthritis in rats at some dosage regimens (Tascilar *et al.*, 1993). Cimetidine substantially decreased dermal and synovial mast cell-induced vasopermeability (Malone *et al.*, 1991). Intra-articular somatostatin induced a clinical improvement in rheumatic patients (Marabini *et al.*, 1991). Similarly, there is evidence that the drugs currently used in rheumatic disorders, and thought to have an ulcerogenic effect, could have an opposite salutary activity on gastric mucosa as well. Instead of an increased ulcerogenesis, a high tolerance was regularly produced by a prolonged treatment with NSAIA (Robert, 1991; Rainsford, 1992). Moreover, the concept of cytoprotection, introduced by Robert, is based on the evidence that aspirin would protect gastric mucosa against damaging effect of indomethacin (Robert, 1979). An apparent lack of ulcers in the experimental animals and increased proliferation of the gastric mucosa are noted after 2 weeks of daily treatment with either aspirin (Robert *et al.*, 1991) or indomethacin (Kuwayama and Eastwood, 1985). Interestingly, the resistance to ulcer formation persisted for 10 days following 4 days of treatment with aspirin and cold (Robert *et al.*, 1991). A beneficial effect on gastric stress ulcers was noted for corticosteroids as well (Hernandez *et al.*, 1984).

Could this commonality in the positive effects of the seemingly opposite medications provide a common basis for both gastrointestinal ulcerations and

rheumatic disorders? Supportingly, the incidence of the gastrointestinal ulcers was increased in rheumatic patients even without use of NSAIA (Caruso and Bianchi Porro, 1980; Doherty *et al.*, 1987; Festen, 1988). Previously, it was shown that raised serum immunoreactive gastrin was consistently raised in both rats with adjuvant arthritis and rheumatic patients (Rooney *et al.*, 1973a, b). An important role of foregut hormones was consequently suggested (Rooney *et al.*, 1973a).

As an additional link, capsaicin-sensitive neurons (Holzer, 1991) could be proposed. Besides in maintenance of gastrointestinal mucosa integrity, capsaicin sensitive neurons have been implicated in the both afferent central transmissions of nociceptive information and in the efferent regulation of inflammation and sensitization of joint sensory endings in a chronic pain state, for example arthritis (Holzer, 1991). Recently, in addition to a powerful mucosa protection, a close interaction with capsaicin sensitive neurons was clearly demonstrated for BPC 157 (Sikiric *et al.*, 1993b, 1994). Thus, it has been suggested to be one of the neuropeptides implicated in conduction of nociceptive information (Sikiric *et al.*, 1993b, c). Indeed, in rheumatic disorders, a raised level of neuropeptides in conjecture with an increased number of nerve fibers staining for neuropeptides such as substance P, calcitonin gene related peptide and somatostatin in inflamed tissues, has been consistently noted (Holzer, 1991; Mantyh, 1991).

In summary, the present study demonstrated that BPC 157 pentadecapeptide has a beneficial effect on NSAIA-induced gastrointestinal tract lesions and non-established and established adjuvant arthritis in rats. It is possible that its efficacy appears as a consequence of a specific activity in inflammatory processes (*eg* interference with release of arachidonic acid metabolites, particularly LTB₄) (Veljaca *et al.*, 1994a, b, 1995b). These potentially interesting findings need to be further investigated in clinical trials.

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