

Aminophylline Potentiates Sodium Nitroprusside-induced Hypotension in the Dog

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The biochemical mechanisms by which nitroso-vasodilators cause smooth muscle relaxation remain controversial. One theory states that the effects of nitroso-vasodilators are mediated by increased intracellular levels of cyclic GMP due to activation of guanylate cyclase. To test this hypothesis, the authors examined the effects of sodium nitroprusside (SNP) in anesthetized dogs with and without pretreatment with the phosphodiesterase inhibitor aminophylline. Aminophylline pretreatment resulted in a 2.8-fold potentiation of the hypotensive effects of a continuous infusion of SNP. Potentiation also was seen for the effects of SNP on stroke volume, heart rate, and plasma cyclic GMP levels. These results support the hypothesis that nitroso-vasodilators exert their effects via guanylate cyclase activation. The authors advise caution when vasodilator therapy with agents such as SNP, nitroglycerin, or hydralazine is instituted in patients receiving aminophylline and when aminophylline is either instituted or discontinued in patients on vasodilator therapy. (Key words: Anesthetic techniques: hypotensive; nitroprusside. Blood pressure: hypotension. Pharmacology: aminophylline; cyclic GMP; nitroprusside.)

THE ROLE of cyclic nucleotides as modulators of smooth muscle tone has been investigated intensively during the past decade. It has been proposed that the vasodilator effects of sodium nitroprusside (SNP), nitroglycerin, hydralazine, and related compounds are mediated by activation of the enzyme guanylate cyclase, resulting in an increase in intracellular levels of cyclic GMP (3',5'-guanosine monophosphate).¹⁻⁵ The major evidence for this hypothesis is the correlation between agent-induced relaxation of smooth muscle and changes in levels of cyclic nucleotides in isolated preparations such as aorta, ductus deferens, mesenteric artery, and tracheal smooth muscle. If the actions of nitroso-vasodilators are mediated by increases in intracellular levels of cyclic GMP, then the effects of these vasodilators should be potentiated by cyclic nucleotide phosphodiesterase inhibitors such as aminophylline. Because phosphodiesterase inhibitors

and nitroso-vasodilators frequently are administered to the same patients, clinically significant drug interactions may occur. We chose an animal model to look for such interactions and report that aminophylline results in a 2.8-fold potentiation of the hypotensive effects of a continuous infusion of SNP in the dog.

Methods

Eight adult mongrel dogs, weighing 12-29 kg, were each studied on two occasions, once with and once without aminophylline pretreatment. After an overnight fast, animals were anesthetized with 8 mg/kg ketamine, im, followed by 15 mg/kg sodium pentobarbital, iv. Animals were intubated with a cuffed endotracheal tube and mechanically ventilated with 100% oxygen at a tidal volume of 15 ml/kg and a rate adjusted to maintain arterial blood carbon dioxide tension at 35-40 mmHg. Anesthesia was maintained by continuous pentobarbital infusion at a rate of 7.5 mg · kg⁻¹ · h⁻¹. Systemic artery and quadruple-lumen thermistor-tipped pulmonary artery (Swan-Ganz® VIP catheter; American Edwards) catheters were inserted by cutdown procedures on the femoral artery and vein. A central venous catheter was introduced via the right external jugular vein.

Following baseline hemodynamic measurements (see below), dogs received either 7 mg/kg aminophylline (theophylline ethylenediamine), iv, or an equivalent volume (50 ml) of normal saline over 20 min. Hemodynamic measurements were repeated 10 min later. SNP (Nitropride®; Roche) then was administered at an initial rate of 1 µg · kg⁻¹ · min⁻¹. Fifteen minutes later, hemodynamic measurements were repeated. The infusion rate was increased successively to 2, 4, 8, and 16 µg · kg⁻¹ · min⁻¹, with hemodynamic measurements being obtained after 15 min at each dose. The pentobarbital infusion then was discontinued, catheters were removed, and the dogs were allowed to recover. One week later, the procedure was repeated with the other pretreatment (aminophylline or saline). Total fluid administration during each half of the study was approximately 350 ml.

Hemodynamic measurements consisted of heart rate (HR), systemic mean arterial pressure (MAP), central venous pressure (CVP), mean pulmonary artery pressure (MPAP), mean pulmonary artery wedge pressure (PAWP), and cardiac output (CO). Cardiac output was

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TABLE 1. Hemodynamic Variables and Cyclic GMP Levels

Variable	Control	Saline or Aminophylline	Sodium Nitroprusside Dose ($\mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$)				
			1	2	4	8	16
MAP (mmHg)	136 ± 5 132 ± 5	140 ± 6 132 ± 3	136 ± 6 119 ± 4†‡	127 ± 6 108 ± 5*†	117 ± 6* 94 ± 7*†	101 ± 7* 83 ± 6*†	87 ± 2* 69 ± 4*†
CI ($\text{l} \cdot \text{min}^{-1} \cdot \text{m}^{-2}$)	3.8 ± 0.3 3.7 ± 0.3	4.0 ± 0.3 3.9 ± 0.3	4.0 ± 0.2 3.7 ± 0.2	4.0 ± 0.2 3.5 ± 0.2†	4.0 ± 0.2 3.4 ± 0.2§	4.0 ± 0.2 3.3 ± 0.2†	4.0 ± 0.2 3.4 ± 0.1
HR (beats/min)	132 ± 12 127 ± 7	142 ± 13 151 ± 5‡	153 ± 11 173 ± 7*	165 ± 9‡ 178 ± 6*	173 ± 11* 176 ± 7*	177 ± 13* 171 ± 7*	175 ± 14* 168 ± 8*
SI ($\text{ml} \cdot \text{beat}^{-1} \cdot \text{m}^{-2}$)	29 ± 2 30 ± 3	28 ± 1 26 ± 2	27 ± 2 21 ± 2*	25 ± 2* 20 ± 1*†	23 ± 2* 19 ± 1*	23 ± 2* 20 ± 1*	24 ± 2* 21 ± 2*
CVP (mmHg)	1.4 ± 0.7 1.4 ± 0.9	1.6 ± 0.7 1.1 ± 0.8	1.9 ± 1.0 1.2 ± 0.7	1.6 ± 1.1 1.4 ± 0.8	1.2 ± 1.1 0.9 ± 0.9	1.4 ± 1.1 0.5 ± 1.1	1.6 ± 1.3 0.5 ± 1.1
MPAP (mmHg)	10.2 ± 1.0 10.8 ± 1.5	10.4 ± 0.8 9.8 ± 1.3	9.1 ± 0.9 8.9 ± 1.4*	8.4 ± 1.0* 8.5 ± 1.4*	8.0 ± 1.0* 7.6 ± 1.3*	7.2 ± 1.0* 7.5 ± 1.3*	7.2 ± 1.0* 7.5 ± 1.3*
PAWP (mmHg)	3.8 ± 0.5 4.0 ± 1.0	3.6 ± 0.8 2.6 ± 0.8‡	2.0 ± 0.8* 1.9 ± 0.9*	1.1 ± 0.9* 1.5 ± 0.9*	0.9 ± 0.9* 1.1 ± 0.8*	0.6 ± 0.8* 0.9 ± 0.8*	0.5 ± 0.8* 1.1 ± 0.9*
SVRI ($\text{dyn} \cdot \text{s} \cdot \text{cm}^{-5} \cdot \text{m}^2$)	3,014 ± 313 2,911 ± 182	2,897 ± 251 2,773 ± 205	2,724 ± 222 2,622 ± 146	2,577 ± 201* 2,447 ± 120*	2,363 ± 146* 2,255 ± 173*	2,033 ± 160* 2,000 ± 164*	1,740 ± 164* 1,639 ± 108*
PVRI ($\text{dyn} \cdot \text{s} \cdot \text{cm}^{-5} \cdot \text{m}^2$)	139 ± 11 147 ± 8	135 ± 17 146 ± 8	142 ± 11 153 ± 9	144 ± 12 154 ± 7	144 ± 12 154 ± 7	135 ± 10 156 ± 8	137 ± 9 149 ± 12
cGMP (pmol/ml)	21 ± 3 18 ± 2	20 ± 3 20 ± 2	20 ± 3 18 ± 2	19 ± 2 18 ± 2	19 ± 3 20 ± 2	20 ± 2 23 ± 2‡	23 ± 3 30 ± 3*†

Values are means ± SEM of eight subjects. The upper row of values for each variable corresponds to the saline-pretreated group; the lower row corresponds to the aminophylline-pretreated group.

Abbreviations: MAP = systemic mean arterial pressure; CI = cardiac index; HR = heart rate; SI = stroke index; CVP = central venous pressure; MPAP = mean pulmonary artery pressure; PAWP = pulmonary artery wedge pressure; SVRI = systemic vascular resistance index; PVRI = pulmonary vascular resistance index; and cGMP = cyclic

GMP.

* $P < 0.01$ compared with control value (prior to saline or aminophylline pretreatment).

† $P < 0.05$ compared with saline-pretreated group.

‡ $P < 0.05$ compared with control value (prior to saline or aminophylline pretreatment).

§ $P < 0.01$ compared with saline-pretreated group.

recorded as the mean of three determinations by thermodilution technique, each using 10 ml iced normal saline; the Edwards Laboratories 9520A Cardiac Output Computer® was used. Cardiac index (CI), stroke index (SI), systemic vascular resistance index (SVRI), and pulmonary vascular resistance index (PVRI) were calculated by standard formulae. Arterial blood for analysis of plasma cyclic GMP levels was obtained with each set of hemodynamic measurements. Arterial blood for analysis of plasma theophylline levels was drawn during the first set of hemodynamic measurements after aminophylline infusion and with the final set of hemodynamic measurements (SNP dose of $16 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$). Blood samples for analysis of plasma cyclic GMP levels were collected, with EDTA added to inhibit phosphodiesterase and immediately cooled to 0–4°C. Samples were centrifuged to separate plasma that was stored at –70°C for later assay. Proteins in plasma were precipitated with cold 18% trichloroacetic acid and centrifuged. Supernatant fractions were extracted with ether, acetylated, and assayed in triplicate for cyclic GMP concentrations by a modification⁶ of the radioimmunoassay procedure of

Steiner *et al.*⁷ Plasma theophylline concentrations were measured by high-pressure liquid chromatography.⁸

STATISTICS

Results are expressed as means ± SEM of the eight dogs. Statistical analysis was by a two-factor repeated measures design analysis of variance and by standard linear regression analysis, with $P < 0.05$ considered significant.⁹

Results

Plasma theophylline levels in the aminophylline-pretreated group were $7.8 \pm 0.9 \mu\text{g}/\text{ml}$ after aminophylline infusion and $5.3 \pm 0.8 \mu\text{g}/\text{ml}$ at the end of the experiment. There were no significant differences between the aminophylline and saline groups in any hemodynamic variable before pretreatment. Within groups the only statistically significant effects of aminophylline pretreatment were a 19% increase in HR and a 1.4 mmHg decrease in PAWP.

SNP resulted in dose-dependent decreases in MAP in both the aminophylline-pretreated and the saline-pre-

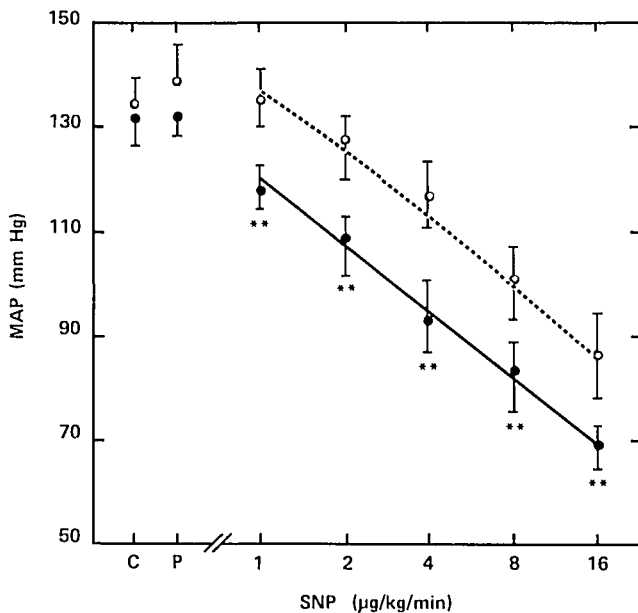


FIG. 1. The effect of sodium nitroprusside (SNP) on mean arterial pressure (MAP) in eight dogs following saline or aminophylline pretreatment. Values are means \pm SEM of eight dogs. C refers to values before pretreatment; P refers to values after saline or aminophylline pretreatment. The open circles represent the saline group; the dotted regression line is $MAP = 138 - 41 \times (\log \text{dose SNP})$; $r = -0.991$; $P < 0.001$. The closed circles represent the aminophylline group; the solid regression line is $MAP = 120 - 42 \times (\log \text{dose SNP})$; $r = -0.999$, $P < 0.001$.

treated groups (table 1). However, the effects of SNP differed between the two groups. The decrease in MAP was significant at all doses of SNP in the aminophylline-pretreated group and at doses of $4 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ and above in the saline-pretreated group. MAP was significantly lower in the aminophylline-pretreated group than in the saline-pretreated group at each dose of SNP. In both groups there was a linear relationship between the log dose of SNP and the MAP (fig. 1); the absolute value of the correlation coefficient r was greater than 0.99 for both groups. The two regression lines were parallel but with different intercepts, indicating a 2.8-fold potentiation by aminophylline of the hypotensive effects of SNP.

SNP increased heart rate in both groups; the maximum heart rate of approximately 175 beats/min was reached at a SNP dose of $1 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ in the aminophylline-pretreated group and $4 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ in the saline-pretreated group. The increase in heart rate was significant at all doses of SNP in the aminophylline-pretreated group and at doses of $2 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ and above in the saline-pretreated group.

SNP decreased stroke index in both the aminophylline-pretreated group and the saline-pretreated group. The decrease in stroke index was significant at all doses of

SNP in the aminophylline-pretreated group and at doses of $2 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ and above in the saline-pretreated group. Although stroke index was lower at each dose of SNP in the aminophylline-pretreated group than in the saline-pretreated group, this difference was statistically significant only at the $2 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ dose of SNP. Cardiac index was lower at each dose of SNP in the aminophylline-pretreated group than in the saline-pretreated group; this difference was statistically significant at SNP doses of 2, 4, and $8 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$.

SNP resulted in dose-dependent decreases in SVRI, MPAP, and PAWP in both the aminophylline-pretreated and the saline-pretreated groups; however, there were no significant differences between the two groups. There were no significant changes in PVRI or CVP.

Plasma cyclic GMP levels were not affected by aminophylline or saline pretreatment or by low doses of SNP. Cyclic GMP levels were increased significantly at the 8 and $16 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ doses of SNP in the aminophylline-pretreated group. The increase in cyclic GMP levels at the $16 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ dose of SNP in the saline-pretreated group did not reach statistical significance. Cyclic GMP levels were significantly higher at the $16 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ dose of SNP in the aminophylline-pretreated group than in the saline-pretreated group.

Discussion

Aminophylline pretreatment resulted in a 2.8-fold potentiation of the hypotensive effects of SNP. Potentiation (increased effect or effect at a lower dose of SNP) also was seen in SI, HR, and plasma cyclic GMP levels. The increase in plasma cyclic GMP levels only at the two largest doses of SNP in the aminophylline-pretreated group may reflect the existence of an intracellular threshold level below which cyclic GMP does not exit from smooth muscle cells into blood. The absence of potentiation in all hemodynamic variables may be due to direct cardiovascular effects of aminophylline (such as positive chronotropy), which are unrelated to the interaction with SNP. In order to limit the problem of interpreting such direct effects, we intentionally chose an aminophylline dose calculated to produce low therapeutic levels.¹⁰ If we had used an aminophylline dose that produced high therapeutic levels, *i.e.*, 15–20 $\mu\text{g}/\text{ml}$, an even greater potentiation of the effects of SNP might have been seen.

The hemodynamic mechanisms for the increased hypotensive effects of SNP following aminophylline pretreatment are not clear. The major effect appears to be related to a decrease in CI rather than to a further decrease in SVRI. It is possible that either aminophylline or SNP produce a negative inotropic effect; however, there is little evidence in the literature to support such

a hypothesis. A possible explanation for the decrease in CI is that aminophylline pretreatment potentiates the venodilating effects of SNP. Our failure to demonstrate an interaction between aminophylline pretreatment and SNP on CVP or PAWP may have been due to difficulties in accurately measuring small differences in filling pressures that were already low—venodilation in the presence of low filling pressures may have marked hemodynamic effects with little absolute change in the filling pressures.

Since aminophylline is a potent inhibitor of cyclic nucleotide phosphodiesterase, the enzyme that degrades cyclic GMP, our results are consistent with the hypothesis that the effects of SNP are mediated by activation of guanylate cyclase, resulting in increased intracellular levels of cyclic GMP. There are two possible clinical implications to our findings. The first is that patients who are receiving aminophylline or related compounds may be prone to develop severe hypotension when treated with standard doses of SNP. The second possible implication relates to the hypothesis that the other nitroso-vasodilators such as nitroglycerin and hydralazine also work through activation of guanylate cyclase. Some patients receiving aminophylline therefore may be more sensitive to the effects of these vasodilators, such as the antianginal effects of the nitrates and the afterload-reducing effects of hydralazine. Instituting aminophylline treatment in some patients on vasodilators therefore could result in hypotension; discontinuing aminophylline in patients on nitrates or hydralazine could result in cardiac ischemia, pulmonary edema, and low-output congestive heart failure. Future studies are needed to determine whether such clinical interactions occur.

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