Hemodynamic consequences of chronic parasympathetic blockade with a peripheral muscarinic antagonist

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**Short title:** Blood pressure and chronic parasympathetic blockade

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Abstract

While the sympathetic nervous system has a well established role in blood pressure (BP) regulation, it is not clear whether long-term levels of BP are affected by parasympathetic function or dysfunction. We tested the hypothesis that chronic blockade of the parasympathetic nervous system has sustained effects on BP, heart rate (HR) and BP variability (BPV). Sprague Dawley rats were instrumented for monitoring of BP 22-h per day by telemetry and housed in metabolic cages. After healing from surgery and a baseline control period, scopolamine methyl bromide (SMB), a peripheral muscarinic antagonist, was infused intravenously for 12 days. This was followed by a 10-day recovery period. SMB induced a rapid increase in mean BP from 98±2 mmHg to a peak value of 108±2 mmHg on day 2 of the SMB infusion, and then stabilized at a plateau value of +3±1 mmHg above control (P < 0.05). After cessation of the infusion, the mean BP fell by 6±1 mmHg. There was an immediate elevation in HR that remained significantly above control on the last day of SMB infusion. SMB also induced a decrease in short-term (within 30-min periods) HR variability, and an increase in both short-term and long-term (between 30-min periods) BPV. The data suggest that chronic peripheral muscarinic blockade leads to modest, but sustained increases in BP, HR and BPV, which are known risk factors for cardiovascular morbidity.

Keywords: blood pressure, autonomic nervous system, parasympathetic nervous system, blood pressure variability, muscarinic antagonists
Introduction

The autonomic nervous system maintains cardiovascular homeostasis through the opposing effects of its parasympathetic and sympathetic divisions on cardiac performance, and through a predominantly sympathetic control of the vasculature. While a sympathetic effect on blood pressure (BP) regulation is well established (6), there is also evidence of an involvement of the parasympathetic nervous system in hypertension. This was first suggested by Julius et al. (12) on the basis of acute pharmacological experiments in young individuals with borderline hypertension and a hyperkinetic circulation. Subsequent studies have shown an attenuation of the parasympathetic control of heart rate in hypertension (33) and in a number of conditions predisposing to hypertension such as ageing (27), obesity (1; 31), diabetes mellitus (3), chronic renal failure (2), and physical inactivity (24). However, it is not clear whether parasympathetic dysfunction may affect basal BP levels or exacerbate the hypertensive state. In addition, since vagal dysfunction impairs the baroreflex control of heart rate, parasympathetic dysfunction could promote an increase in BP variability (BPV) (7) which has been shown to be an independent cardiovascular risk factor (9).

The purpose of the present study was to test the hypothesis that sustained blockade of the parasympathetic system would lead to increased BP and BPV. To this end, we investigated the effects of a 12-day administration of scopolamine methyl bromide (SMB) in rats instrumented for monitoring of BP by telemetry. SMB, a quaternary derivative of scopolamine, was chosen because it is a muscarinic antagonist that does not readily cross
the blood-brain barrier (4) and thus avoids potential confounding effects of central muscarinic antagonism, such as increased parasympathetic tone (18).

**Methods**

**Animal Preparation**

Male Sprague-Dawley rats (10-12 weeks old, weighing ~390-400 g) supplied by the R. Janvier Center (France) were used in this study. All protocols were approved by the State Animal Committee. The rats were instrumented (under pentobarbital anesthesia, 60 mg/kg intraperitoneal) with a BP telemeter (model TA11-PA-C40, Data Sciences International) for monitoring of aortic BP as described previously (32). A catheter was also inserted in the left jugular vein and connected to a syringe pump via a single channel swivel as described previously (35), for continuous intravenous infusion of isotonic saline (10 µL/min) starting on the day of surgery and maintained throughout the experimental study. The rats were housed in individual Plexiglas cages equipped for 24h urine collection. The cages were in a quiet air-conditioned room (~21 °C) with a 12h light-dark cycle.

**Continuous Hemodynamic Monitoring by Telemetry**

Each cage was equipped with one RMC-1 receiver connected to a calibrated analog adapter (R11CPA) and to an APR-1 barometric pressure reference device. All devices were manufactured by Data Sciences International. The telemetered analog pressure signal was sampled at 500 Hz for 5 sec periods every 30 sec, 24 hours a day, and processed by customized algorithms for beat-to-beat analysis to extract mean arterial pressure (MAP), systolic and diastolic blood pressures (SBP, DBP, respectively), heart
rate (HR), rise time (time to reach peak SBP from the previous DBP point) and maximum rate of change of arterial pressure (max dPa/dt) (17). Telemetry probes were calibrated before implantation and after removal in a sealed pressure chamber at 0 and 200 mmHg, and pressure values were linearly corrected according to calibration values (29). Data were analyzed from 10:00 A.M. to 8:00 A.M. the next morning. The period between 8:00 A.M. and 10:00 A.M. was used for daily maintenance and was omitted from the analysis.

The computer program separated the 22 h data set into a series of 44 half-hour periods. The standard deviation (SD) of BP (for both MAP and SBP) were calculated for each half-hour period. The mean SD of BP was used as a measure of the short-term variability of BP, i.e. the extent of the variation in BP within half-hour periods (19; 30). In addition, we also calculated the SD of the 44 half-hourly mean BP values and used the SD as a measure of the long-term variability of BP, i.e. the extent of the variation in BP between half-hour periods (19; 30). Finally, the SD of all 2,640 daily samples was also computed as an index of overall 22hr-BPV. Similar calculations were performed to calculate the short and long term HR variability. It should be noted that the SD of HR within half-hour periods does not represent beat-to-beat HR variability, but the 30s-to-30s variability of HR. It is thus labeled “short-term” HR variability.

**Experimental Protocol**

In all rats, experimental measurements included measurement of BP and HR 22 hours a day, and monitoring of daily food intake, water and sodium intakes, urine output, and sodium and potassium excretions. Food and water intake were determined by differential weighing (precision at 0.1 g). Urine was collected in a graduated container. Urinary sodium and potassium concentrations were determined by flame photometry (model IL 943). All animals had free access to tap water and were fed with a fixed amount of 20 g
(slightly below the normal ad libitum intake) of standard rat chow per day to ensure a fixed sodium intake (a total of about 4 mmol/day, including 2.2 mmol/day via the infusion).

In seven rats that were successfully instrumented (10.7±0.2 weeks old, weighing 396±14 g), a scopolamine methylbromide (SMB) (Sigma, Switzerland) intravenous infusion was started after at least 12 days of recovery from surgery and an additional control period of 4 days. SMB was infused for 12 days at increasing rates (0.3, 0.6 and 1.2 µg/kg/min i.v., 4 days at each dose, labeled as period 1, 2 and 3). The progressive increase in the SMB-infusion rate was chosen to ensure sustained muscarinic blockade, since pilot studies in which SMB was infused at the single dose of 0.3 µg/kg/min for 10 days showed a higher BP during the first 4 days of infusion (~ +10 mmHg) than during the following days (~ +5mmHg). The SMB-infusion was followed by a ten-day recovery period to test the reversibility of the experimental changes. All solutions (SMB or vehicle saline) were prepared aseptically and infused through a 0.22 µm Millipore filter.

Because BP and HR are known to exhibit a small spontaneous negative drift after surgery, a group of eight rats were submitted to the same surgical procedures (telemetry, intravenous infusion) and housed in identical cages, in order to analyze the changes in BP and HR occurring during vehicle time-control infusion from day 9 after surgery (when the telemeter battery was turned on) up to 28 days after surgery. In two rats, the arterial BP signal could not be used for computation of MAP, due to a large and unstable offset drift, but was adequate for the computation of HR. The rats were studied up to 4 weeks after surgery to quantify the spontaneous drift of the 22-h average of BP and HR.
**Statistical Analysis**

Each rat served as its own control, i.e. the effects of SMB infusion were compared to the average of the 4 control days preceding SMB infusion. Statistical analysis was performed by repeated measures ANOVA with the InStat statistical package (GraphPad Software, Inc.). Experimental and control values were compared using Dunnett’s multiple comparison test. Changes were considered significant if P < 0.05. Values are expressed as mean ± SEM.

**Results**

SMB induced a rapid increase in mean, systolic and diastolic pressures, and heart rate (Figure 1). MAP peaked on day 2 of the SMB infusion at +10.5±0.7 mmHg above a control value of 97.9±1.9 mmHg. The initial increase in MAP was attenuated over the next few days, but MAP remained significantly elevated at a plateau value of +2.9±0.7 mmHg (P < 0.05) during the last 4 days of SMB (period 3), as shown in Table 1. On cessation of SMB infusion, MAP fell by 5.9±0.7 mmHg (day 2 of recovery), and stabilized at 2.6±1.1 mmHg below control value during the last 4 days of the recovery period (days 7 to 10 after SMB cessation).

HR underwent an immediate increase with SMB infusion, peaking on the first day at +52±5 beats per minute (bpm) above the control value of 345±8 bpm. HR then tended to decrease progressively but remained significantly above control at +22±5 bpm by the last day of SMB infusion (day 12). After cessation of the SMB infusion, HR fell by 36±4 bpm (day 2 of recovery). During the final 4 days of the recovery period, HR reached a value of 318±7 bpm, i.e. 27±6 bpm below the control level.
Rats infused with vehicle infusion showed a small decrease in MAP by 30±0.9 mmHg from 97.7±1.1 mmHg (average of days 9-12 post-surgery) to 94.7±1.0 mmHg (average of days 25-28), with most of the decrease (2.5±1.1 mmHg) occurring during the first 20 days after surgery. HR showed a larger decrease of 31±5 bpm from a control value of 361±9 bpm (average of days 9-12 post-surgery), with most of the decrease (22±5 bpm) occurring during the first 20 days after surgery. Subtracting the average daily negative drift of BP and HR observed in time-control rats (using post-surgery days 13-16 as the control period) from individual values of experimental rats, we estimate the actual effects of SMB on MAP and HR in experimental rats to amount to 10.2±0.4, 5.2±0.7 and 5.5±0.7 mmHg for periods 1, 2 and 3, respectively, and to 60±5, 53±4 and 47±4 bpm for the same periods.

Daily values of pulse pressure (PP), maximum dPa/dt and rise time in SMB-infused rats are shown in Figure 2. Changes from control values are summarized in Table 1. PP and maximum dPa/dt were significantly higher only during the first few days of infusion (on day 3, PP peaked at +3.5±0.7 mmHg above the control value of 22.8±1.4 mmHg and maximum dPa/dt peaked at +840±216 mmHg·s⁻¹ above a control value of 5062±306 mmHg·s⁻¹). Rise time was significantly shorter during days 1-4 (period 1) and 5-8 (period 2), and tended to remain shorter during the last 4 days of SMB infusion (period 3).

Daily values of short-term and long-term MAP and HR variabilities are shown in Figure 3. SMB induced a progressive increase in both short-term (+16% last 4 days of SMB infusion) and long-term (+36%) MAP variabilities, which reverted progressively to control values on cessation of the SMB infusion. Interestingly, the changes in long-term variability were slower to appear and also to disappear. The increase in BP variability was significant for both MAP and SBP, and for all three indices of variabilities (within-30 min SD , between-30 min SD, and overall 22h-SD), as shown in Table 2. SMB also lead to a
marked (-43%) and sustained decrease in short-term HR variability (HRV). In contrast, the decrease in long-term HRV was only transient (-25% on day 2).

Total water intake and urinary output tended to increase progressively during SMB infusion, as shown on Figure 4, and were significantly higher during the last 4 days of infusion (water intake: + 7.2 ml from a control value of 37.8±1.1 ml/d; urine output: +8.6 ml from control value of 24.7±0.7 ml/d). Both variables returned slowly towards control during the recovery period. Changes occurred in parallel and, as a consequence, daily water balance remained unaltered. In 3 rats out of 7, food intake (fixed at a maximum of 20 g/day) decreased slightly on day 2 of SMB infusion, but was back to normal thereafter. This explains the transient decrease in sodium and potassium intakes during day 2. In these three rats the non-ingested sodium of day 2 was compensated on the next day by supplementing food with an estimated equivalent of sodium chloride. Sodium and potassium balances remained stable over the following days.

**Discussion**

While numerous studies have reported the cardiovascular effects of acute parasympathetic inhibition, we are not aware of any published study of the effects of more sustained parasympathetic blockade on hemodynamic function. The main finding of our study is that chronic peripheral muscarinic blockade leads to significant hemodynamic alterations including sustained elevations in blood pressure, heart rate and blood pressure variability.

The effect on BP was most pronounced during the first three days of muscarinic inhibition, with a peak increase in SBP, MAP and DBP of more than 10% during day 2 of SMB. Although BP tended to return towards control level over the following few days, BP remained significantly elevated during the last 4 days of SMB infusion as shown in
Table 1, a conclusion also supported by the off transient in BP when the infusion was halted. The chronic SMB-induced effect on MAP amounted to a 3–6% increase, depending whether its effects are compared to the initial control period (3%) or the final recovery period (6%).

The partial recovery of blood pressure during SMB infusion occurred despite evidence of the continued effectiveness of muscarinic blockade by SMB. Heart rate, which initially increased by as much as 15% above control value, remained elevated by 11% on the last day of SMB infusion (based on the size of the off transient, day 2 of recovery). The attenuation of the “short-term” HR variability was even more dramatic, responding promptly at the onset and end of SMB infusion, and not wavering throughout the 12 day infusion period. Combined with the observation that doubling the SMB dosage on days 5 – 8 (period 2) and again on days 9 – 12 (period 3) failed to cause further alterations in heart rate, “short-term” HR variability, or any other measured parameter, our data suggest that the SMB infusion provided an effective and sustained blockade of cardiac muscarinic receptors.

The values of BP and HR during days 7-10 of the recovery period (-2.6±1.1 mmHg and -27±6 bpm below control values for MAP and HR, respectively) are consistent with a moderate spontaneous decrease in hemodynamic values often observed in chronic experiments after a surgical procedure. Indeed, in a group of vehicle-infused rats instrumented in a similar way, there was an average decrease in MAP and HR of 3% and 8%, respectively, from day 9-12 after surgery to day 25-28. This slow decrease in vehicle-infused animals, possibly related to the gradual disappearance of expected post-surgery inflammatory reactions, strengthens our findings in SMB-infused rats that the significant increases in MAP and HR associated with long-term SMB infusion are not due to a slow positive baseline drift but represent a real phenomenon. Indeed, after correcting for the
negative drift occurring in time-control animals, the estimated chronic effects of SMB amounted to a 6% increase in MAP and a 13% increase in HR (period 3).

Although our experimental approach (recording throughout the day, with animals left undisturbed as much as possible) did not allow a more precise determination of the mechanisms underlying the SMB-induced increase in BP, at least three factors could be involved: (a) an increase in cardiac output (CO) due to a primary increase in HR and/or stroke volume; (b) an increase in peripheral vascular resistance; and (c) an altered ability of the kidney to excrete salt and water.

(a) While an increase in HR is expected initially to increase CO, stroke volume tends to fall with time and changes in HR per se may not lead to sustained increases in CO (8). However, a combination of increased HR and contractility could potentially cause a larger increase in CO. Tachycardia alone is expected to decrease PP and maximum dPa/dt as shown in heart-pacing experiments (26). In our experiments, the observed increases (and the lack of decreases) in PP and maximum dPa/dt, and the decrease in rise time, are consistent with an increase in cardiac contractility during the SMB infusion although we cannot exclude an effect of SMB to decrease arterial distensibility.

(b) Muscarinic blockade could promote vasoconstriction. There is, however, little vascular parasympathetic innervation to be blocked except for sparse specialized circulations (33), but there is evidence for a local release of acetylcholine (ACh) by endothelial cells themselves. The presence of choline acetyltransferase, the enzyme responsible for the synthesis of ACh, has been shown in cerebral microvascular endothelial cells of rats (20) and pigs (11), as well as in bovine carotid artery endothelial
cells (13). Milner et al. (16) could demonstrate a local release of ACh in isolated human umbilical vein endothelial cells, which is triggered by shear stress. Release of ACh by cultured endothelial cells prepared from bovine carotid artery was verified by radioimmunoassay (13). Taken together, these results show that endothelial cells can synthesize and release ACh, which may act as an autacoid and reinforce stretch-induced NO release. Inhibition of endothelial muscarinic receptors by SMB may thus impair stretch-induced NO release. Interestingly, the hypertensive effect of NO-synthase inhibition is potentiated by acute cholinergic blockade (14), suggesting an interaction between cholinergic and nitrergic vasodilatation.

Finally, the kidneys may also be involved in the SMB-increase in BP. Normally, a rapid increase in BP by vasoconstrictors would be expected to result in a pressure-induced natriuresis (10). Yet, no initial natriuresis was observed in our experiments despite a rise in BP of more than 10 mmHg, suggesting sodium-retaining forces that could offset pressure-induced natriuresis. ACh has a natriuretic effect on the kidney, which is inhibited by atropine (34). Although there is no clear parasympathetic innervation of the kidneys (5), the antagonism of endothelial mechanisms, as described above, could possibly promote renal vasoconstriction and sodium retention.

Finally, acetylcholine is known to inhibit the release of norepinephrine from sympathetic nerve terminals via stimulation of prejunctional muscarinic receptors (36). Thus, an additional possibility is that blockade of these receptors by SMB could effectively increased sympathetic tone at the level of the heart and vasculature.
Although a chronic increase in 24h BP of 3–6% is physiologically relatively modest, human clinical trials indicate that sustained similar increases in BP would have substantial effects on cardiovascular risk. Furthermore, it is possible that the tonic influence of the parasympathetic nervous system on BP may play a larger role in humans or in other circumstances. Indeed, rats have a modest parasympathetic tone compared to larger animals such as dogs and humans, perhaps in part because they are usually studied at laboratory temperatures (~20-22 °C) which are well below the range of ambient temperatures that requires minimal thermoregulatory effort to maintain body temperature (thermoneutrality zone). The thermal neutral zone of healthy rats of common strains in experimental setups similar to ours (single cage housing, no bedding) is around 30 °C (23). Warming ambient temperature into the zone of thermoneutrality has been shown to reduce BP and HR, and to increase HRV in both rats and mice (28). The parasympathetic system may thus play a potential larger role in BP regulation in humans than in rats and mice maintained at standard laboratory temperatures.

Chronic muscarinic blockade also led to significant increases in short-term (within-30 min SD), long-term (between-30 min SD) and overall (22hr-SD) BPV. Vagal dysfunction impairs the baroreflex control of HR, which in turn could lead to greater oscillations in BP and hence to an increased short-term BPV (7). Indeed, muscarinic blockade in our study led to a marked and sustained decrease in “short-term” HRV and an increase in short-term BPV. The slow increase in long-term BPV observed during SMB infusion is intriguing, but the underlying mechanism remains unclear.

Increases in BPV could contribute to hypertensive target organ damage (TOD). Indeed, several clinical investigations have found TOD to be more advanced in patients with increased BPV (19; 21). These findings have been reinforced by a prospective
investigation (9) in which patients that had taken part in one of the original studies (19) were followed-up approximately 7 years later. The role of systolic BPV in promoting TOD has been confirmed in large clinical studies, such as the PAMELA study (25) and the Syst-Eur trial (22).

**Perspectives**

Parasympathetic dysfunction is not a rare event. In addition to hypertension itself, reductions in cardiac parasympathetic tone or reactivity have been described in obesity (1; 31), diabetes mellitus (3), chronic renal failure (2), physical inactivity (24), and ageing (27). Other factors may promote parasympathetic dysfunction via direct muscarinic receptor (MR) inhibition. Autoantibodies against MR, whose occurrence increases with age, have been found in healthy subjects (15). Moreover, some major drugs, such as neuroleptics, antiarrhythmics and antidepressants, may have anticholinergic effects.

While a contribution of parasympathetic dysfunction to cardiovascular morbidity has been suggested by many authors, the hemodynamic effects of chronic muscarinic blockade have not been reported. Our study shows that chronic MR-inhibition favors a small increase in BP and leads to a significant increase in both short-term and long-term BPV. Our data suggest that chronic peripheral muscarinic blockade could contribute to the development or aggravation of hypertension and TOD.
Acknowledgements

The authors would like to thank Aldo Tempini for expert technical assistance.
References


Table 1

<table>
<thead>
<tr>
<th>Variable</th>
<th>Period 1</th>
<th>Period 2</th>
<th>Period 3</th>
<th>Recovery</th>
</tr>
</thead>
<tbody>
<tr>
<td>SBP (mmHg)</td>
<td>9.3±0.5 **</td>
<td>3.5±0.9 *</td>
<td>3.3±1.1 *</td>
<td>-2.8±1.6</td>
</tr>
<tr>
<td>MAP (mmHg)</td>
<td>8.2±0.4 **</td>
<td>3.3±0.7 **</td>
<td>2.9±0.7 *</td>
<td>-2.6±1.1 *</td>
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<tr>
<td>DBP (mmHg)</td>
<td>6.7±0.5 **</td>
<td>2.9±0.7 *</td>
<td>2.5±0.5 *</td>
<td>-2±0.9</td>
</tr>
<tr>
<td>HR (bpm)</td>
<td>47±5 **</td>
<td>35±4 **</td>
<td>24±4 **</td>
<td>-27±6 **</td>
</tr>
<tr>
<td>PP (mmHg)</td>
<td>2.6±0.7 *</td>
<td>0.7±0.8</td>
<td>0.8±1</td>
<td>-0.8±1.5</td>
</tr>
<tr>
<td>Max dPa/dt (mmHg/s)</td>
<td>579±184 *</td>
<td>185±163</td>
<td>66±264</td>
<td>-138±260</td>
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<tr>
<td>Rise time (ms)</td>
<td>-1.7±0.2 *</td>
<td>-1.3±0.4 *</td>
<td>-1.0±0.4</td>
<td>0.2±1</td>
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</tbody>
</table>

Values are means ± SEM (n=7), computed as changes from control levels (last 4 days before starting SMB infusion). Periods 1, 2 and 3 correspond to days 1-4, 5-8 and 9-12, respectively, of SMB infusion. Recovery corresponds to days 7-10 after cessation of SMB infusion. * < 0.05 and ** < 0.01 versus control period (Dunnett's)
Table 2

Table 2. Short-term (within-30 min), long-term (between-30 min) and overall (22hr) standard deviation (SD) of systolic blood pressure (SBP) and mean arterial pressure (MAP)

<table>
<thead>
<tr>
<th>Variable</th>
<th>Control</th>
<th>Period 1</th>
<th>Period 2</th>
<th>Period 3</th>
<th>Recovery</th>
</tr>
</thead>
<tbody>
<tr>
<td>SBP Short Term SD (mmHg)</td>
<td>4.9±0.3</td>
<td>5.6±0.2 *</td>
<td>5.9±0.2 **</td>
<td>5.7±0.2 **</td>
<td>4.8±0.2</td>
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<tr>
<td>SBP Long Term SD (mmHg)</td>
<td>4.5±0.3</td>
<td>5.0±0.3</td>
<td>5.9±0.4 **</td>
<td>6.3±0.6 **</td>
<td>4.9±0.4</td>
</tr>
<tr>
<td>SBP Overall SD (mmHg)</td>
<td>6.6±0.3</td>
<td>7.5±0.2 *</td>
<td>8.3±0.4 **</td>
<td>8.5±0.5 **</td>
<td>6.9±0.4</td>
</tr>
<tr>
<td>MAP Short Term SD (mmHg)</td>
<td>4.6±0.3</td>
<td>5.1±0.2 *</td>
<td>5.4±0.2 **</td>
<td>5.3±0.1 **</td>
<td>4.5±0.2</td>
</tr>
<tr>
<td>MAP Long Term SD (mmHg)</td>
<td>4.1±0.3</td>
<td>4.5±0.2</td>
<td>5.3±0.4 **</td>
<td>5.6±0.5 **</td>
<td>4.5±0.3</td>
</tr>
<tr>
<td>MAP Overall SD (mmHg)</td>
<td>6.1±0.3</td>
<td>6.8±0.2 *</td>
<td>7.5±0.3 **</td>
<td>7.7±0.4 **</td>
<td>6.3±0.3</td>
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</tbody>
</table>

Values are means ± SEM (n=7). Control represents the last 4 days before starting SMB infusion. Periods 1, 2 and 3 correspond to days 1-4, 5-8 and 9-12, respectively, of SMB infusion. Recovery corresponds to days 7-10 after cessation of SMB infusion. * < 0.05 and ** < 0.01 versus control period (Dunnett's)
Figure Legends

Figure 1  Effects of a 12-day SMB infusion on mean arterial pressure (MAP), systolic blood pressure (SBP) and diastolic blood pressure (DBP), and on heart rate.

Figure 2  Effects of a 12-day SMB infusion on pulse pressure, maximum dPa/dt and rise time.

Figure 3  Effects of a 12-day SMB infusion on short-term and long-term MAP and HR variabilities. The time-course of short-term and long-term SBP variabilities was very similar to the time-course of MAP variabilities and is therefore not shown in the Figure.

Figure 4  Effects of a 12-day SMB infusion on the daily balance of water, sodium and potassium. Water intake and sodium intake include the amount of water and sodium, respectively, contained in the intravenous infusion. Balance was computed as intake minus urinary excretion.
Figure 1

SMB infusion (μg/kg/min)

Blood Pressure (mmHg)

Heart Rate (bpm)

Copyright Information
Figure 2

![Graph showing changes in pulse pressure, max dPa/dt, and rise time over time (days) with different SMB infusion rates (0.3, 0.6, 1.2 μg/kg/min).]
Figure 3

Short-term MAP variability

Short-term HR variability

Long-term MAP variability

Long-term HR variability

SMB infusion (µg/kg/min)

0.3 0.6 1.2