Head-Down Tilt Posture Attenuates Anaphylactic Hypotension in Mice and Rats

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Abstract: A head-down tilt posture, the Trendelenburg position, which could facilitate venous return from the splanchnic organs and lower extremities, is recommended for the treatment of anaphylactic shock. However, few data of animal studies support its effectiveness. We examined the effects of a head-down tilt maneuver on anaphylactic hypotension in BALB/c mice and Sprague-Dawley rats. We measured systemic arterial pressure (Sap) and portal venous pressure (Pvp) in spontaneously breathing anesthetized animals sensitized with ovalbumin. At either supine (control) or a 30-degree head-down tilt position, anaphylactic hypotension was induced by an intravenous injection of antigen. In the control rats, an increase in Sap by 66 mmHg and a decrease in Pvp by 11.5 cmH2O were observed at 2.5 and 6 min, respectively, after antigen. In contrast, in control mice injected with antigen, Sap decreased similarly, but Pvp increased by only 4 cmH2O. A head-down tilt maneuver in mice substantially attenuated the antigen-induced decrease in Sap throughout the 60 min measurements, though it aggravated slightly, but significantly, only at the late phase of after 25 min in rats. We conclude that a head-down tilt maneuver attenuates anaphylactic hypotension in anesthetized mice and rats. These beneficial effects were smaller in rats than in mice probably because of substantial portal hypertension, which might prevent the head-down tilt-induced increase in venous return from the splanchnic vascular bed.

Key words: anaphylactic shock, hepatic circulation, splanchnic congestion, Trendelenburg, head-down tilt.

Systemic anaphylaxis is a severe immediate-type hypersensitivity reaction characterized by life-threatening hypotension [1]. Although epinephrine injection and intravenous fluid administration are recognized as the first choice against anaphylactic shock, severe cardiovascular collapse is often resistant to this treatment [1, 2]. On the other hand, a cornerstone of initial management is putting the patient in the supine position or the Trendelenburg position, a head-down tilt posture [1, 2]. On the contrary, other investigators reported that the Trendelenburg position is essentially useless for vascular resuscitation [3]. An increase in resistance to venous return is important in the pathogenesis of circulatory collapse in canine anaphylactic shock [4]. We have recently reported that the antigen-induced hepatic vasoconstriction, which may be consistent with increased venous resistance, plays an important role in anaphylactic hypotension in rats [5]. Theoretically, the beneficial effect of the Trendelenburg position could be derived from a facilitation of venous return from the splanchnic or lower extremities. In this respect, blood mobilization from splanchnic organs depends on the hepatic vascular tone, i.e., portal venous pressure [6]. However, few animal studies support the effectiveness of this position in experimental models of anaphylactic shock. Thus we examined the effect of a head-down tilt posture on the systemic arterial pressure and portal venous pressure in anaphylactic shock models of rats and mice.

METHODS

Animals. Twenty-three male BALB/c mice weighing 29.1 ± 2.7 g and 23 male Sprague-Dawley rats weighing 307 ± 24 g (Japan SLC, Japan) were used. These animals were maintained at a room temperature of 23°C and under pathogen-free conditions on a 12:12-h dark/light cycle, with food and water ad libitum. This experiment was approved by the Animal Research Committee of Kanazawa Medical University. We followed the principles of laboratory animal care (NIH publication No. 86-23, revised 1985).

Sensitization. The mice were sensitized by the subcutaneous injection of an emulsion made by mixing aluminum potassium sulfate adjuvant (2 mg) with 0.01 mg ovalbumin (grade V, Sigma) dissolved in saline (0.2 ml). The antigen was injected two times with a 1-week interval [7]. For rat sensitization, an emulsion made of complete Fre-
und’s adjuvant (0.5 ml) and 1 mg ovalbumin dissolved in saline (0.5 ml) was injected subcutaneously only once [5]. Nonsensitized animals were injected with adjuvant and ovalbumin-free saline. Two weeks after the initial injection, the animals were used for the following experiments.

Protocol. Mice or rats were anesthetized with 90 or 70 mg·kg⁻¹ pentobarbital sodium ip, respectively, and placed on a heating pad. The adequacy of anesthesia was monitored by the stability of the systemic arterial pressure (Sap) and respiration during a pinch of the hind paw. Supplemental anesthetic was given intraperitoneally as necessary. The Sap was measured via the right femoral artery and the right common carotid artery in mice and rats, respectively. For the injection of antigen and the measurement of central venous pressure (Cvp), the right external jugular vein was catheterized. Heart rate (HR) was measured by triggering the R wave of the electrocardiogram. The reference level for pressure measurement was the level of the right atrium. The reference level for pressure measurement was set at the level of the catheter tip. These vascular pressures were measured with pressure transducers (TP-400T, Nihon-Kohden, Japan), which were fixed anytime at the level of the right atrium. The reference level for pressure measurement was set at the level of the catheter tip. These vascular pressures were measured with pressure transducers (TP-400T, Nihon-Kohden, Japan), which were fixed anytime at the level of the right atrium.

After a midline incision of the abdominal wall, a catheter (ID 0.47 mm, OD 0.67 mm) was inserted into the main portal vein, and the catheter tip was positioned at approximately 0.5 cm from the hepatic hilus for a continuous measurement of the portal venous pressure (Pvp). After closure of the abdomen, the baseline measurements were started.

The Sap, Pvp, and Cvp were continuously measured with pressure transducers (TP-400T, Nihon-Kohden, Japan), which were fixed anytime at the level of the right atrium. The reference level for pressure measurement was set at the level of the catheter tip. These vascular pressures and the HR were continuously displayed on a thermal physiograph and also digitally recorded at 20 Hz (Power-Lab, AD Instruments). The mean values of Psa and Pvp were calculated using the Power-Lab. The values of Cvp were the values measured at expiration. At 20 min after surgery, the position of the animal in the sensitized or nonsensitized head-down group was changed so that the head was lower than the feet at an angle of 30 degrees, but in the other two supine groups, the position remained unchanged. In the head-down groups, Pvp was mathematically corrected after the head-down position change: corrected Pvp (cmH₂O) = measured Pvp (cmH₂O) – 0.5 × the length between the right atrium and liver hilus (cmH₂O). After the baseline measurements, 0.02 mg of the ovalbumin antigen in 100 µl saline was intravenously administered to the mice, and 0.06 mg antigen in 300 µl saline to the rats.

Statistics. All results are expressed as the means ± SD. Statistical analyses were performed with repeated measures analysis of variance, and a P value of less than 0.05 was considered significant. When a significant difference was obtained, post hoc analysis was performed with the Bonferroni posttest method.

RESULTS
1. Effect of head-down tilt in mouse anaphylactic shock
Table 1 shows the basal hemodynamic variables in all groups studied. The basal levels of Sap and Pvp in rats were significantly greater than in mice, and HR in rats was significantly less than in mice. Figure 1A shows the typical recordings in a mouse under supine condition. Figure 2 shows the changes in mean values of mean Sap, mean Pvp, Cvp at expiration, and HR of the mouse. After an antigen injection in the mouse sensitized the supine group, the mean Sap slightly and transiently increased, and it then gradually decreased from the baseline of 91 ± 10 mmHg to the bottom of 54 ± 6 mmHg at 10 min, with recovery to 69 ± 8 mmHg at 60 min.

Table 1. Basal hemodynamic variables in anesthetized mice and rats.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Cvp (mmHg)</th>
<th>Pvp (cmH₂O)</th>
<th>Sap (mmHg)</th>
<th>HR (beats/min)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mouse</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nonsensitized supine</td>
<td>1.1±0.5</td>
<td>6.5±0.5</td>
<td>98±3#</td>
<td>456±22#</td>
</tr>
<tr>
<td>Sensitized supine</td>
<td>1.2±0.8</td>
<td>5.3±0.4#</td>
<td>91±10#</td>
<td>412±47</td>
</tr>
<tr>
<td>Nonsensitized head-down</td>
<td>1.3±0.5</td>
<td>7.0±0.3#</td>
<td>95±5#</td>
<td>476±47#</td>
</tr>
<tr>
<td>Sensitized head-down</td>
<td>1.9±0.9</td>
<td>5.5±0.4#</td>
<td>99±11#</td>
<td>419±49</td>
</tr>
<tr>
<td>Rat</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nonsensitized supine</td>
<td>1.4±0.9</td>
<td>7.8±1.6</td>
<td>113±10</td>
<td>414±26</td>
</tr>
<tr>
<td>Sensitized supine</td>
<td>2.1±1.7</td>
<td>9.0±0.8</td>
<td>130±9</td>
<td>421±27</td>
</tr>
<tr>
<td>Nonsensitized head-down</td>
<td>2.4±0.4</td>
<td>9.9±0.8</td>
<td>115±6</td>
<td>407±15</td>
</tr>
<tr>
<td>Sensitized head-down</td>
<td>2.0±0.9</td>
<td>9.8±1.6</td>
<td>126±18</td>
<td>426±34</td>
</tr>
</tbody>
</table>

Means ± SD; #P < 0.05 vs. rat; Cvp, central venous pressure; Pvp, mean portal venous pressure; Sap, mean systemic arterial pressure; HR, heart rate.
Effect of Head-Down Tilt on Anaphylactic Hypotension

Fig. 1. A representative recording of the response to ovalbumin antigen (0.02 mg) in the mouse sensitized supine group (A) and the mouse sensitized head-down group (B). The pressures were recorded instantaneously.

Fig. 2. The summary of changes in the mean systemic arterial pressure, mean portal venous pressure, central venous pressure, and heart rate after antigen injection in mice. Means ± SD; *significant difference from baseline; #significant difference from the sensitized supine group; open circles, mouse nonsensitized supine group (n = 6); solid circles, mouse sensitized supine group (n = 6); open triangles, mouse nonsensitized head-down group (n = 5); solid triangles, mouse sensitized head-down group (n = 6).

c. HR tended to increase immediately after antigen, and a statistically significant increase was observed after 6 min (Fig. 2).
In the mouse sensitized head-down group (Figs. 1B and 2), basal variables were comparable to those of the sensitized supine group, but the antigen-induced decrease in Sap was significantly attenuated: mean Sap decreased to 80 ± 15 mmHg at 15 min, which was significantly higher than that of the sensitized supine group of 55 ± 5 mmHg.

Fig. 3. A representative recording of the response to ovalbumin antigen (0.06 mg) in the rat sensitized supine group (A) and the rat sensitized head-down group (B). The pressures were recorded instantaneously.

Fig. 4. The summary of changes in the mean systemic arterial pressure, mean portal venous pressure, central venous pressure, and heart rate after antigen injection in rats. Means ± SD; *significant difference from baseline; **significant difference from the sensitized supine group; open circles, rat nonsensitized supine group (n = 5); solid circles, rat sensitized supine group (n = 6); open triangles, rat nonsensitized head-down group (n = 5); solid triangles, rat sensitized head-down group (n = 7).
Effect of Head-Down Tilt on Anaphylactic Hypotension

Actually, significant differences in $Sap$ between these two groups were found from 4.5 min to 60 min. $Pvp$, $Cvp$, and HR changed similarly to that in the sensitized supine group after antigen. In the nonsensitized supine and head-down groups, no significant changes were observed in hemodynamic variables except an increase in HR after 20 min in the nonsensitized supine groups (Fig. 2).

2. Effect of head-down tilt in rat anaphylactic shock

Figures 3 and 4 show typical recordings and the summary data of the hemodynamic variables of the rat, respectively. After antigen in the rat-sensitized supine group (Figs. 3A and 4), mean $Sap$ decreased from the baseline of $130 \pm 9$ mmHg to $64 \pm 9$ mmHg at 6 min, followed by recovery at 60 min. Mean Pvp increased markedly from the baseline of $9.0 \pm 0.8$ cmH$_2$O to the peak of $20.5 \pm 1.9$ cmH$_2$O at 2.5 min after antigen, which was much higher than that in mice, then gradually decreased to the baseline levels at 40 min. After antigen, $Cvp$ tended to decrease, but not significantly. HR did not significantly change after antigen.

In the rat-sensitized head-down group (Figs. 3B and 4), $Sap$ decreased in a manner similar to that in the sensitized supine group until 25 min after antigen. Thereafter $Sap$ of the sensitized head-down group became significantly higher than that of the sensitized supine group. Indeed, mean $Sap$ at 25 min of $119 \pm 13$ mmHg was significantly higher than the corresponding values of the sensitized supine group at $101 \pm 13$ mmHg. The antigen-induced increase in $Pvp$ was similar to that of the sensitized supine group. After antigen, $Cvp$ tended to decrease, but not significantly in the sensitized head-down group. HR did not change significantly after antigen in any rat group studied.

DISCUSSION

We here examined whether the 30-degree head-down tilt attenuates experimental anaphylactic hypotension in anesthetized rats and mice. A major finding was that a head-down tilt maneuver attenuated anaphylactic hypotension substantially in mice, but only slightly in rats. In contrast, anaphylactic portal hypertension was observed considerably in rats, but only slightly in mice. These results suggest that the beneficial effects of a head-down tilt posture in rats, less than in mice, may be due to the presence of a substantial portal hypertension in rats, which might prevent the head-down tilt-induced increase in venous return from the splanchnic vascular bed. This assumption may be supported by the finding that a head-down tilt-induced significant attenuation of anaphylactic hypotension in rats was observed only at the late phase when portal hypertension subsided, as shown in Fig. 4.

Another possible reason why the head-down tilt posture was more effective against anaphylactic hypotension in mice than in rats may be related to the difference in blood flow distribution to the lower extremities. It is reported that the blood flow to muscles of the lower extremities is greater in mice (30% of cardiac output) than in rats (~20%) [8]. Thus the head-down tilt position, which facilitates the venous return from the lower extremities, could mobilize more blood from the lower extremities in mice than in rats. In contrast, there seemed to be no difference in portal blood flow from splanchnic organs between rats and mice [8, 9].

Species differences in the target organs affected by anaphylaxis occur with right heart failure taking place in the rabbit [10] and hepatic congestion occurring in the dog [10, 11]. In the guinea pig, heart is the target organ of systemic anaphylaxis [12]. In the rat and mouse, it is not well known which organs are vulnerable to systemic anaphylaxis. However, we and others [13] are proposing that the liver and splanchnic vascular beds are anaphylactic target organs in the rat. On the other hand, although anaphylactic hepatic venoconstriction was observed in mice, as shown in the present study, it was very weak in comparison with the other animals. This species difference may account for the difference in the effect of the head-down tilt posture on anaphylactic hypotension, as described above.

We previously reported that anaphylactic hepatic venoconstriction is involved in anaphylactic hypotension in rats, based on the finding that the total hepatectomy combined with ligation of the celiac and mesenteric arteries attenuated antigen-induced systemic hypotension during the period of portal hypertension [5]. The present study suggests that the anaphylactic hepatic venoconstriction could also serve as an impedance against venous return from splanchnic organs during a head-down maneuver. However, the extrapolation of the present findings in the patients with anaphylactic shock should be awaited, because it is still uncertain whether anaphylactic shock in humans is accompanied by portal hypertension. Clinical observation of the portal pressure in patients with anaphylactic shock is required.

After antigen, HR significantly increased in sensitized mice under either a supine or head-down condition. This antigen-induced tachycardia in mice may be caused by the baroreceptor reflex in response to hypotension. Furthermore, the possibility exists of a direct positive chronotropic effect of anaphylactic mediators such as histamine on the sinus nodes, as seen in sensitized guinea pig hearts [14]. In this study, no significant differences in the HR response were found between the sensitized supine and head-down mice. This finding was unexpected because a head-down tilt posture could activate cardiopulmonary baroreceptors resulting in the attenuation of the baroreflex function [15, 16]. A direct positive chronotropic effect of anaphylaxis might have surpassed a possible sympathoinhibitory action of the head-down tilt maneuver. In this respect, HR in nonsensitized supine mice significantly in-
creased at the end of the experiment, but HR did not significantly change in the nonsensitized head-down mice (Fig. 2). In contrast to the tachycardiac response of mice to antigen, the sensitized rats under either supine or head-down condition showed no significant changes in HR when Sap considerably decreased after antigen. The absence of tachycardia in response to anaphylactic hypoten-
sion in anesthetized rats was consistent with our previous studies [5]. Koyama et al. [17] demonstrated that systemic baroreceptor reflex control of heart rate and renal sympathetic nerve activity is reduced during anaphylactic hypoten-
sion in pentobarbital-anesthetized dogs. A similar impairment of arterial baroreceptor reflex might occur during anaphylactic shock in the rat, but not in the mouse. The present study was carried in pentobarbital-anesthetized rats and mice. Anesthetic agents have substantial ef-
fects on the results of the present study. The vagolytic property of pentobarbital sodium [18] may account for the high levels of HR at baseline (approximately 400 beats/ min) as compared with the unanesthetized rats (approximately 350 beats/min) [19]. In the present study, when Sap decreased after the antigen, HR did not significantly increase. This finding contrasts with the tachycardia re-
sponse to the antigen of the unanesthetized SD rats sensi-
tized with ovalbumin in the same manner as our study (un-
published observation).

CONCLUSIONS

We here clearly showed that a head-down tilt maneuver attenuates anaphylactic hypotension in anesthetized mice and rats. These beneficial effects were smaller in rats than in mice probably because of substantial portal hyperten-
sion, which might prevent the head-down tilt-induced in-
crease in venous return from the splanchnic vascular bed. This study was supported by a Grant-in-Aid for Scientific Research (18591730) from the Ministry for Education, Culture, Science and Technology of Japan, Dr. Zhanheng Zhao and Dr. Wei Zhang were supported by postdoctoral fellowships for foreign researchers of Kanazawa Medical University in 2006 and 2007, respectively.

REFERENCES