Large-Dose Epinephrine Reduces Skeletal Muscle Blood Flow Under General Anesthesia in Rabbits

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The goal of this study was to investigate the effect of an epinephrine continuous infusion on muscle blood flow in rabbits. Sixteen male Japan White rabbits were randomly allocated to 1 of 2 groups: epinephrine continuous infusion at 0.01 $\mu\text{g}/\text{kg}/\text{min}$ (Ep-0.01 group, $n=8$) and at 0.1 $\mu\text{g}/\text{kg}/\text{min}$ (Ep-0.1 group, $n=8$). The observed variables were heart rate, femoral artery blood pressure, common carotid artery blood flow (CCBF), masseter muscle blood flow (MBF), and quadriceps muscle blood flow (QBF). In the Ep-0.01 group, CCBF, MBF, and QBF were increased by 14, 22, and 21% from respective control values. In contrast, in the Ep-0.1 group, CCBF, MBF and QBF were decreased by 10, 30, and 27% from respective control values. There were no differences in the percentage change between MBF and QBF during epinephrine continuous infusion. Positive correlations were observed between CCBF and MBF and between CCBF and QBF. In conclusion, skeletal muscle blood flow was increased during the small-dose epinephrine infusion, whereas it was decreased during large-dose infusion.

Key Words: Epinephrine; Skeletal muscle blood flow; Rabbits.

In dental treatment, especially in oral and maxillofacial surgery, control of bleeding from the surgical field is important in order to achieve smooth procedures. A common method for minimizing bleeding during surgery is an administration of a local anesthetic solution containing epinephrine.^{1–4} Regionally administered epinephrine is immediately absorbed into the regional circulation, and a positive correlation has been observed between basal oral mucosal blood flow and maximum plasma epinephrine concentration.³ Accordingly, though epinephrine is useful because of its strong vasoconstrictive properties, undesirable cardiovascular effects have been reported.^{1,2,4}

When the venous plasma epinephrine concentration is about 200 pg/mL or less, β -adrenergic effects are

dominant. In contrast, when it is above 400 pg/mL, α -adrenergic effects are observed.^{5,6} Because α_1 - and β_2 -adrenergic receptors are distributed in skeletal muscle vessels^{7,8} and β_2 -adrenergic receptors are dominant in these vessels,⁹ it has been reported that skeletal muscle blood flow is increased through vasodilation via β_2 -adrenergic mechanisms.⁹ However, changes in skeletal muscle blood flow during high plasma epinephrine levels remain to be clarified because vasoconstriction may concomitantly occur through α_1 -adrenergic mechanisms. In addition, epinephrine at doses used in routine dental procedures during general anesthesia promotes platelet aggregation through α_2 -adrenergic mechanisms.¹⁰ If large-dose epinephrine causes a decrease in skeletal muscle blood flow along with platelet aggregation, the risk of congestion-related complications such as deep venous thrombosis (DVT) might be increased in clinical settings. It has been reported that DVT has oc-

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curred in patients undergoing oral and maxillofacial surgery.¹¹

In this study, therefore, we observed the changes in skeletal muscle blood flow during small- and large-dose epinephrine infusions in anesthetized rabbits.

METHODS

Sixteen male Japan White rabbits (2.2–2.7 kg), purchased from Sankyo Labo Company, Tokyo, Japan, were utilized. This study was performed according to the Guidelines for the Treatment of Experimental Animals in Tokyo Dental College. All animals were allowed food and water ad libitum until the morning of the experiment. The animals were randomly allocated to 1 of 2 groups: epinephrine continuous infusion at 0.01 $\mu\text{g}/\text{kg}/\text{min}$ (Ep-0.01 group, $n=8$) and epinephrine continuous infusion at 0.1 $\mu\text{g}/\text{kg}/\text{min}$ (Ep-0.1 group, $n=8$).

Anesthesia was induced by inhalation of 4.0% isoflurane (Forane, Abbott Japan, Tokyo, Japan) in oxygen delivered using a mask. Before skin incisions for each of the experimental procedures, appropriate doses of lidocaine (Xylocaine, Astra Zeneca, Osaka, Japan) were injected into the surgical field. A #20 French noncuffed pediatric tracheal tube was inserted into the trachea through a tracheostomy. The left auricular marginal vein and right femoral artery were cannulated with 22- and 20-gauge Teflon indwelling catheters, respectively. After intravenous acetated Ringer's solution was started at 10 mL/kg/h, the animals were paralyzed with rocuronium bromide at 14 $\mu\text{g}/\text{kg}/\text{min}$ (Eslax, Schering-Plough, Tokyo, Japan) and mechanically ventilated to maintain an end-tidal CO_2 at 40 mm Hg. Femoral artery blood pressure was continuously monitored with a pressure transducer (P231D, Gould, Oxnard, Calif). Heart rate (HR) was recorded by a tachograph triggered by the blood pressure wave. Common carotid artery blood flow (CCBF) was measured with an ultrasound flowmeter (T108, Transonic, Ithaca, NY). A flow probe (type 3SB) was applied to the isolated left common carotid artery. Systolic blood pressure, diastolic blood pressure, mean arterial pressure, HR, and CCBF were continuously recorded on a polygraph (Series360, NEC Sanei, Tokyo, Japan).

After the skin incision along the left lower margin of the mandible without local anesthesia, the fascia of the left masseter muscle was detached to expose muscle tissue. A needle probe of the hydrogen clearance tissue blood flowmeter (UHE-100, Unique Medical, Tokyo, Japan) was inserted 3 mm deep into the anterior portion of the left masseter muscle to measure masseter muscle tissue blood flow (MBF). Then, after the skin incision along the left femoral region without local anesthesia, the left quad-

riceps muscle was exposed. A needle probe of the hydrogen clearance tissue blood flowmeter was inserted 5 mm deep into the center of the left quadriceps muscle to measure quadriceps muscle tissue blood flow (QBF).

After completion of experimental preparations, isoflurane inhalation was reduced to a 1.0% end-tidal concentration and maintained at that level for more than 60 minutes to stabilize the hemodynamic and respiratory variables. The isoflurane concentration was continuously monitored with an anesthetic gas monitor (Capnmac, Datex, Helsinki, Finland). Body temperature was continuously monitored with a rectal probe and maintained between 39.0 and 39.5°C with the aid of a heating lamp.

After control measurements, 1 $\mu\text{g}/\text{mL}$ of epinephrine (Bosmin, Daiichi-Sankyo, Tokyo, Japan) was continuously infused at a rate of 0.01 or 0.1 $\mu\text{g}/\text{kg}/\text{min}$, respectively. Data were recorded immediately before epinephrine infusion and 15 minutes after the start of epinephrine continuous infusion. All animals were sacrificed using a fatal dose of thiopental sodium (Ravonal, Mitsubishi-Tanabe, Osaka, Japan) after the experiment.

In this study, data are expressed as mean $\pm$ SD. Student's t test for paired samples was used for within-group comparisons. Student's t test for unpaired samples was used for the comparison of MBF and QBF expressed as a percentage change of the respective control value in Ep-0.1 and Ep-0.01 groups. Pearson's correlation coefficient was used for the regression analysis between CCBF and MBF and between CCBF and QBF. P values less than .05 were considered statistically significant.

RESULTS

In the Ep-0.01 group, blood pressure and HR did not change during epinephrine infusion. CCBF, MBF, and QBF were increased by 14, 22, and 21% from respective control values (Table; Figure 1). In the Ep-0.1 group, HR was slightly decreased, whereas systolic blood pressure was increased by 5% from the control values. CCBF, MBF, and QBF were decreased by 10, 30, and 27% from respective control values (Table; Figure 1). There were no differences between the percentage changes in MBF and QBF during the epinephrine continuous infusion (Figure 1). Positive correlations were observed between CCBF and MBF ($r=0.84$) and between CCBF and QBF ($r=0.79$) (Figures 2 and 3).

DISCUSSION

This is the first study to clarify the decrease in muscle blood flow during a large-dose epinephrine infusion. In

Table Haemodynamics and Tissue Blood Flow†

	Ep-0.01 Group		Ep-0.1 Group	
	Control	Epinephrine	Control	Epinephrine
HR, bpm	297.5 ± 19.1	296.3 ± 16	302.5 ± 22.5	298.8 ± 25.2*
SBP, mm Hg	131.3 ± 10.9	130.0 ± 10.4	127.0 ± 19.8	133.1 ± 18.7*
DBP, mm Hg	66.9 ± 5.9	66.3 ± 6.4	63.8 ± 23.7	65.0 ± 24.6
MAP, mm Hg	81.3 ± 11.3	80.6 ± 10.8	78.1 ± 9.2	78.1 ± 10.3
CCBF, mL/min	31.3 ± 6.6	35.5 ± 7.1*	28.6 ± 5.2	25.6 ± 4.7*
MBF, mL/min/100 g	43.5 ± 7.4	52.5 ± 6.3*	44.0 ± 8.2	30.9 ± 7.7*
QBF, mL/min/100 g	64.8 ± 8.3	77.9 ± 7.4*	66.2 ± 8.7	49.1 ± 12.7*

* $P < .05$ vs control.† HR indicates heart rate; SBP, systolic blood pressure; DBP, diastolic blood pressure; MAP, mean arterial pressure; CCBF, common carotid artery blood flow; MBF, masseter muscle tissue blood flow; and QBF, quadriceps muscle tissue blood flow. Data are expressed as mean ± SD ($n=8$).

this study, epinephrine was infused at 0.1 and 0.01 $\mu\text{g}/\text{kg}/\text{min}$. The area under the curve of epinephrine after 0.05 mg intravenous injection has been reported to be 236 ± 37 ng/mL/min in rabbits.¹² Because total body clearance is calculated as the injected dose divided by area under the curve, total body clearance of epinephrine in rabbits is estimated to be 212 mL/min. Because steady state plasma concentration is calculated as the infusion rate divided by total body clearance, steady state plasma concentration during a 0.1 $\mu\text{g}/\text{kg}/\text{min}$ infusion was estimated as 1179 pg/mL. Likewise, steady state plasma concentration during 0.01 $\mu\text{g}/\text{kg}/\text{min}$ infusion was estimated as 118 pg/mL in this study. In human experiments, when the venous plasma epinephrine concentration is about 200 pg/mL or less, β -adrenergic effects are dominant. In contrast, when it is above 400 pg/mL, α -adrenergic effects were observed.^{5,6} In addition,

Figure 1. Changes in muscle blood flow in Ep-0.01 and Ep-0.1 groups. Data are expressed as the percentage change of respective control values. MBF indicates masseter muscle tissue blood flow; QBF, quadriceps muscle tissue blood flow mean ± SD ($n=8$).

tion, α - and β -adrenergic receptors are stimulated during large-dose (0.04 $\mu\text{g}/\text{kg}/\text{min}$ or more) epinephrine infusion, whereas β -adrenergic receptors are stimulated during small-dose (0.02 $\mu\text{g}/\text{kg}/\text{min}$ or less) infusion.^{13,14} It is reported that α -¹⁵ and β -adrenergic receptors¹⁶ are distributed in rabbit skeletal muscle vessels. However, there is no report on the plasma threshold of epinephrine on α - and β -adrenergic effects in rabbits. Accordingly, if the plasma threshold of epinephrine on these effects in rabbits was similar with those in humans, both α - and β -adrenergic effects should be observed during 0.1 $\mu\text{g}/\text{kg}/\text{min}$ infusion and β -adrenergic effects should be observed during 0.01 $\mu\text{g}/\text{kg}/\text{min}$ infusion, respectively. It is therefore suggested that the decrease in muscle blood flow in the Ep-0.1 group was caused by vasoconstriction through α -adrenergic mechanisms whereas the increase in the Ep-0.01 group was caused by vasodilatation through β_2 -adrenergic mechanisms.

Skeletal muscles consist of slow- and fast-twitch fibers. Fifty to 90% of the total muscle fibers in the anterior and deep portions of the masseter muscle have a slow-twitch property.¹⁷ In contrast, 90% of the total

Figure 2. Relationship between common carotid artery blood flow (CCBF) and masseter muscle tissue blood flow (MBF). Positive correlation was observed between CCBF and MBF ($r=0.84$).

Figure 3. Relationship between common carotid artery blood flow (CCBF) and quadriceps muscle tissue blood flow (QBF). Positive correlation was observed between CCBF and QBF ($r=0.79$).

muscle fibers in the quadriceps muscle have a fast-twitch property.¹⁸ Therefore, both muscles were used in this study because we believed that the masseter and quadriceps muscles would be good indicators of blood flow changes in the slow- and fast-twitch muscles, respectively. Because distributions of α -⁷ and β -adrenergic receptors¹⁹ are greater in slow-twitch muscle vessels than in fast-twitch muscle vessels, it is speculated that the increase in MBF is larger than in QBF in the Ep-0.01 group and the decrease in MBF is larger than in QBF in the Ep-0.1 group. However, there were no differences in the changes in skeletal muscle blood flow between MBF and QBF. Further study will be needed to clarify the involvement of adrenergic receptors in skeletal muscle blood flow regulation.

It is reported that platelet retention rate, which is a good index of platelet aggregating function, was increased by 30–40% after submucosal injection of a relatively large dose of epinephrine with 1% lidocaine hydrochloride.¹⁰ Therefore, there might be a possible increased risk of DVT because intravascular congestion might occur through the decrease in skeletal muscle blood flow and enhanced platelet aggregation caused by epinephrine contained in a local anesthetic solution.¹⁰ Dehydrated, obese, and geriatric patients or patients with a fracture of the mandible have an increased risk for DVT²⁰ and are among the patients commonly treated by oral and maxillofacial surgeons. In fact, it has been reported that DVT has occurred in patients undergoing oral and maxillofacial surgery.¹¹ Accordingly, careful attention should be paid to these high-risk patients for possible DVT.

In our study, anesthesia was maintained with 1.0% isoflurane throughout the experiment. This concentration was equal to 0.5 minimum alveolar concentration (MAC) of isoflurane in the rabbit.^{21,22} Because isoflurane has vasodilating effects, total peripheral resistance has

been reported to decrease by 24% and forearm blood flow to increase by 199% in humans after 1 hour inhalation of 1 MAC isoflurane.²³ In addition, mesenteric blood flow increases by 50% during 0.65 MAC of isoflurane anesthesia.²⁴ In our study, peripheral blood vessels might be dilated through isoflurane inhalation. However, there were no major differences in tissue blood flow between this study and general anesthesia with sevoflurane²⁵ at the blood concentration of propofol for skin incision (CP50_{skinincision}, blood concentration at which 50% of patients do not respond to skin incision). Therefore, effects of vasodilatation induced by isoflurane should be minimal in this study. In a previous study involving an infusion rate of rocuronium bromide for stable muscular relaxation was 14 $\mu\text{g}/\text{kg}/\text{min}$ during inhalation of 0.5 MAC sevoflurane, tissue oxygen tension of the masseter muscle, which was an index of MBF,²⁶ was not changed.²⁷ Therefore, the infusion rate of rocuronium bromide in our study was set at 14 $\mu\text{g}/\text{kg}/\text{min}$ during an inhalation of 0.5 MAC (1%) isoflurane throughout the experiment.

In conclusion, skeletal muscle blood flow was increased during small-dose epinephrine infusion, whereas it was decreased during large-dose infusion.

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