

ORIGINAL ARTICLE

## Effects of an intravenous infusion of Ringer's solution on eruption rates of incisor teeth in anesthetized rats

AKEMI SHIMADA, KOICHIRO KOMATSU, TATSUYA SHIBATA & MOTOTSUGU CHIBA

*Department of Pharmacology, School of Dental Medicine, Tsurumi University, Tsurumi-ku, Yokohama, Japan*

### Abstract

**Objective.** The vasculature within the socket is reportedly involved in determining the position of continuously erupting teeth. Thus, loss of body fluid in anesthetized rats, which would affect the vascular physiology, should influence tooth movement. We investigated the effects of an infusion of Ringer's solution on the systemic arterial blood pressure, regional blood flow at the base of the incisor, and axial tooth movement in anesthetized rats to determine the cause of tooth displacement. **Material and methods.** In the experimental group, the animals received intravenous infusions of Ringer's solution at 27  $\mu\text{l}/\text{min}$  for 13 h. In the control group, the animals did not receive the infusion. **Results.** The infusion of Ringer's solution suppressed an increase of the mean arterial blood pressure from 86 to 80 mmHg and a decrease of the regional blood flow from 170 to 217 mV, and increased the eruption rate from 267 to 361  $\mu\text{m}/13$  h during the experimental period. There was a positive correlation between the eruption rate and regional blood flow, and a negative correlation between the blood pressure and regional blood flow. **Conclusions.** These results suggest that an infusion of Ringer's solution can cause an increase in the regional blood flow, resulting in increased fluid volume, elevated intra-socket pressure, and increased eruptive movement. It is possible that the regional vascular volume and/or pressure within the socket play an important role in determining the position of the incisor.

**Key Words:** *Blood flow, infusion, rat, tooth eruption*

### Introduction

There is no consensus concerning the eruptive force involved in tooth eruption, although a variety of theories have been put forward to identify its mechanisms [1–7]. The vasculature within the socket is reportedly involved in determining the position of continuously erupting teeth [2–4,6]. Previous studies have suggested that the position of continuously erupting incisors may be influenced by arterial blood pressure, probably due to changes in the vasculature of the tooth pulp and periodontal ligament [3,8–10].

We previously recorded axial movements of rat mandibular incisors while the animals were immobilized under artificial respiration with halothane anesthesia [11]. Under the experimental conditions, the animals were unable to ingest food or water, and the loss of body fluid by urination and evaporation

increased their risk of dehydration or hypovolemia. We observed a decrease of bodyweight under these conditions [11]. It is possible that such experimental conditions can affect the regulation of body fluid and vascular physiology, which in turn influences tooth movement.

To maintain the fluid volume and salt balance, physiological saline formulations such as Ringer's solution have been exogenously administered during maintenance fluid therapy [12]. It could be expected that maintained tooth movement be observed by maintained fluid volume and salt balance. The main purpose of the present study was to investigate the effect of an infusion of Ringer's solution on systemic arterial blood pressure, regional blood flow, and tooth movement in anesthetized rats to elucidate the factors affecting tooth displacement.

## Material and methods

### Animal preparations

Twenty male Wistar rats, weighing  $371 \pm 33$  (SD) g, were divided into two groups of 10 experimental and 10 control animals. The rats were placed in the supine position and were anesthetized via inhalation of halothane in air (1.0%) through a tracheal tube connected to an artificial respirator. The lower margin of the jawbone was surgically exposed and held with a hemostat, which was fixed to the metal bar of a magnetic stand. The procedures for treating the experimental animals have been described elsewhere [11].

### Infusion of Ringer's solution

In the experimental group, the animals received an intravenous infusion of Ringer's solution (Otsuka Pharmaceutical, Tokyo, Japan) for 13 h through a polyethylene tube inserted in the femoral vein using a pediatric infusion set (TK-A400LK; Terumo, Tokyo, Japan) at a room temperature of about 25°C. The rate of infusion, estimated from the decrease in Ringer's solution at the end of the experimental period, was  $27 \pm 8$  (SD)  $\mu\text{l}/\text{min}$ . The control animals did not receive an infusion.

### Measurements of systemic arterial blood pressure, regional blood flow, and tooth movement

The mean arterial blood pressure was measured using a pressure transducer (MP-15; Micron Instruments, Calif., USA) via a polyurethane tube inserted in the right femoral artery.

Regional blood flow at the base of the incisor was measured using a laser Doppler flowmeter (BRL-100; Bio Research Center, Nagoya, Japan). To measure the blood flow, the bone surface at the base of the incisor was surgically exposed, the buccinator muscles were excluded by a polyethylene ring, and then the tip of the needle-type probe (BN-0.5; Bio Research Center) of the flowmeter was placed near the exposed bone surface [10]. The blood flow was estimated as the relative value of mV.

Axial movement of the incisor was measured with a displacement detector (503-FD; EMIC, Tokyo, Japan), i.e. a device in which the movement of a metal plate attached to the tooth surface is recorded continuously [11]. The data were corrected for the curvature of the incisor and the radial distance between the tooth surface and the metal plate [11].

The data for the arterial blood pressure, regional blood flow, and tooth movement were simultaneously loaded onto a computer (Macintosh PowerBook G3; Apple Japan Inc., Tokyo, Japan) using an analog-digital converter (MacLab/400; ADInstruments Japan Inc., Nagoya, Japan) at intervals of 0.5 s during the experimental period.

### Statistical analyses

The difference between the mean values was examined using Student's *t*-test. The mean values of 10 rats estimated for a period of 1 h were used for the regression analysis. Correlation coefficients between the regional blood flow and eruption rate, and between the regional blood flow and arterial blood pressure, were examined. The differences between the regression coefficients and between the correlation coefficient (*r*) and zero were also examined using Student's *t*-test.

## Results

### Systemic arterial blood pressure

Figure 1a shows changes in the systemic arterial blood pressure during a 13 h period. In the control group, the mean blood pressures slightly decreased in the initial 2 h, and then slightly increased during the rest of the experimental period. In the experimental group, the mean blood pressures slightly decreased in the initial 2 h, and then slightly increased until about 6 h and leveled off during the rest of the experimental period. The mean blood pressures were kept at a lower level in the

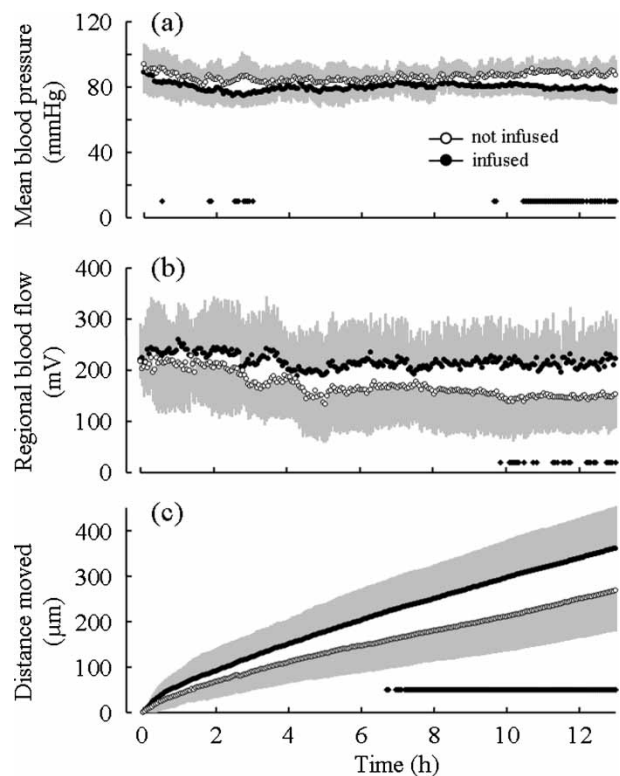


Figure 1. Mean arterial blood pressure (mmHg) (a), regional blood flow (mV) at the base of the incisor (b), and tooth movement ( $\mu\text{m}$ ) (c) in control (not infused) and experimental (infused with Ringer's solution for 13 h) rats. Each point and vertical bar represents the mean  $\pm 1$  SD of 10 rats measured at intervals of 3 min. Significant differences between the two groups ( $\blacklozenge$   $p < 0.05 \sim 0.001$ ).

experimental group than in the control group. The most significant differences between the two groups at intervals of 3 min were from 10.4 h following the infusion ( $p < 0.05 \sim 0.001$ ). The mean values of arterial blood pressure during a period of 13 h were 86 and 80 mmHg in the control and experimental groups, respectively, i.e. a decrease of 7% ( $p < 0.05$ ; Table I).

#### Regional blood flow

Figure 1b shows changes in the regional blood flow at the base of the incisor during the 13 h period. In the control group, the regional blood flow decreased gradually during the experimental period. In the experimental group, the regional blood flow decreased from about 2 to about 5 h, and then leveled off. The regional blood flows were kept at a relatively higher level in the experimental group than in the control group. The differences between the two groups at intervals of 3 min were significant from 9.9 h following the infusion ( $p < 0.05 \sim 0.01$ ). The mean values for regional blood flow during the 13-h period was 170 and 217 mV in the control and experimental groups, respectively, i.e. an increase of 28%, although the difference between the two groups was not significant (Table I).

#### Tooth movement

Figure 1c shows the extrusive tooth movement during the 13 h period. While the tooth movement was obviously accelerated by the infusion, a significant difference between the two groups at intervals of 3 min was first detected at 6.7 h following the infusion ( $p < 0.05$ ). The differences were all significant thereafter ( $p < 0.05$ ). The total amount of tooth movement during the 13-h period was  $267 \pm 88$  (SD) and  $361 \pm 92$   $\mu\text{m}$  in the control and experimental groups, respectively, i.e. an increase of 35% ( $p < 0.05$ ). The mean eruption rates were estimated to be  $493 \pm 163$  (SD)  $\mu\text{m}/24$  h in the control group and  $666 \pm 171$   $\mu\text{m}/24$  h in the experimental group (Table I).

Figure 2 shows the eruption rates ( $\mu\text{m}/\text{h}$ ) of the incisor during the 13-h period. The eruption rates were generally greater in the experimental group

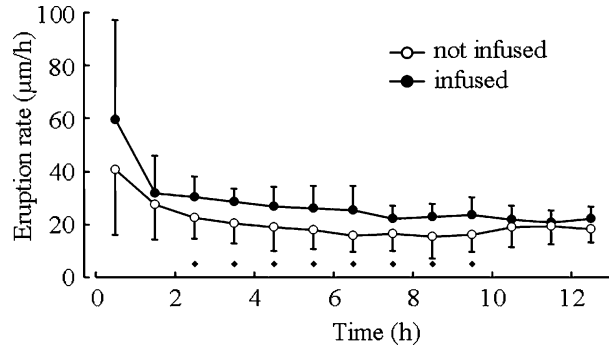


Figure 2. Eruption rate ( $\mu\text{m}/\text{h}$ ) of the mandibular incisor in control (not infused) and experimental (infused with Ringer's solution for 13 h) rats. Each point and vertical bar represents the mean  $\pm 1$  SD of 10 rats. Significant differences between the two groups ( $\blacklozenge p < 0.05 \sim 0.01$ ).

than in the control group. However, significant differences were only observed between 3 and 9 h following the infusion ( $p < 0.05 \sim 0.01$ ). In both groups, the greatest fluctuations, due to large variations among individuals, occurred in the initial few hours, and fluctuations were less pronounced during the rest of the experimental period. Therefore, the values in the first hour in both groups were excluded for the regression analysis.

#### Correlations between measurements

Figure 3a shows a regression line between the mean values of regional blood flow ( $x$ , mV) and the eruption rates of the incisors ( $y$ ,  $\mu\text{m}/\text{h}$ ) estimated for a period of 1 h for both control and experimental groups. A positive correlation was found between the eruption rate and the regional blood flow. The correlation coefficient ( $r = 0.842$ ) was significantly different from zero ( $p < 0.001$ ). When the two groups were calculated separately, each regression line was  $y = 0.117x - 0.829$  ( $r = 0.745$ ;  $p < 0.05$ ) in the control group, and  $y = 0.205x - 14.6$  ( $r = 0.529$ ;  $p < 0.05$ ) in the experimental group. The difference of regression coefficients between the two regression lines was not significant.

Figure 3b shows a regression line between the mean values of regional blood flow ( $x$ , mV) and the systemic arterial blood pressure ( $y$ , mmHg) estimated for a period of 1 h for both control and experimental groups. A negative correlation was found between the systemic arterial blood pressure and the regional blood flow. The correlation coefficient ( $r = -0.891$ ) was significantly different from zero ( $p < 0.001$ ). When two groups were calculated separately, each regression line was  $y = -0.0938x + 100$  ( $r = -0.795$ ;  $p < 0.01$ ) in the control group, and  $y = -0.0264x + 84.4$  ( $r = -0.215$ ; NS) in the experimental group. The difference of regression coefficients between the two regression lines was not significant.

Table I. Effects of infusion on systemic arterial blood pressure (BP), regional blood flow (BF), and eruption rate of the incisor (ER)

	Not infused	Infused	$p$
BP (mmHg)	$86 \pm 6$	$80 \pm 6$	$< 0.05$
BF (mV)	$170 \pm 69$	$217 \pm 67$	NS ( $p = 0.14$ )
ER ( $\mu\text{m}/24$ h)	$493 \pm 163$	$666 \pm 171$	$< 0.05$

Mean  $\pm 1$  SD of 10 rats is shown.  $P$  = level of significance; NS = not significant.

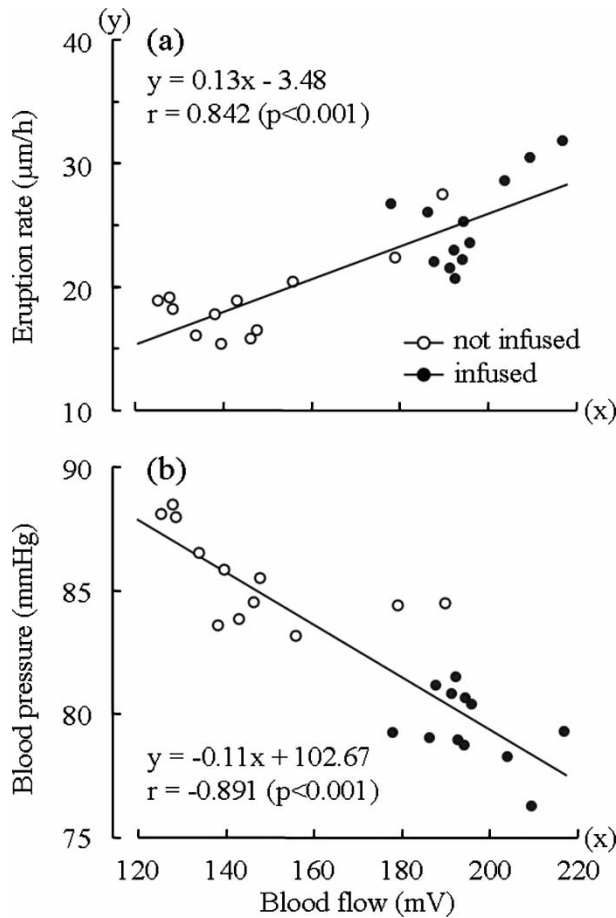


Figure 3. Regression lines between the mean values of regional blood flow ( $x$ , mV) and eruption rate ( $y$ ,  $\mu\text{m/h}$ ) (a), and between the regional blood flow ( $x$ , mV) and arterial blood pressure ( $y$ , mmHg) (b). Each point represents the mean value of 10 rats estimated for a period of 1 h following an infusion of Ringer's solution. The values in both the control (not infused) and experimental (infused with Ringer's solution for 13 h) groups are included. The values in the first hour in both groups are excluded because of the great fluctuations due to large variations among individuals. The correlation coefficients ( $r$ ) were significantly different from zero ( $p < 0.001$ ).

## Discussion

It is clear that arterial pressure is regulated by several interrelated systems, each of which performs a specific function; that is, baroreceptor, chemoreceptor, and sympathetic activity by central nervous system ischemic response react rapidly, within second or minutes, after change in the arterial blood pressure; stress relaxation of vasculatures, renin-angiotensin vasoconstriction, and fluid shift through the tissue capillary walls respond over an intermediate time period, minutes or hours; and aldosterone and the renal-body fluid pressure control system provide long-term arterial pressure regulation, days, months, and years [13]. Among these mechanisms, it is well established that plasma rennin activity and the concentration of arginine vasopressin increase during periods of fluid deprivation, and that the vasoconstrictor actions of angiotensin II [14–16]

and vasopressin [15–19] on peripheral vasculature play critical roles in determining the level of the systemic arterial blood pressure in dehydrated animals. In the control group (not infused rats), the slight increase of systemic arterial blood pressure was observed during the experimental period of 2 to 13 h (Figure 1a); the regional blood flow decreased gradually during the same period (Figure 1b). It is possible that the systemic arterial blood pressure in the control group was maintained by vasoconstriction on the peripheral vasculature, which caused the decrease of regional blood flow at the base of the incisor.

The systemic arterial blood pressure (Figure 1a) and regional blood flow (Figure 1b) were generally more stable in the experimental group than in the control group. It is possible that the infusion of Ringer's solution properly maintained the level of body fluid during the experimental period, and suppressed induction of the pressor systems such as renin-angiotensin and vasopressin. We suggest that the infusion of Ringer's solution reduced the constriction of the peripheral blood vessels and maintained relatively high levels of regional blood flow within the socket in the experimental group.

Following the infusion, the eruptive movement of the incisors was obviously accelerated in association with the higher levels of regional blood flow during the experimental period (Figure 1c, 2, Table I). Indeed, a positive correlation was found between the eruption rate and regional blood flow (Figure 3a). It is conceivable that the infusion primarily reduced the constriction of the peripheral vasculature by the pressor systems, resulting in increased regional blood flow followed by increased fluid volume and elevated intra-socket pressure. The increase of eruptive movement of the incisors could be caused by these increased fluid volume and elevated intra-socket pressure. We propose that the local pressure system within the socket is an important determinant of tooth position [3,9,10], as suggested by other investigators [2,4,6,8,20–22].

In a previous study [10], we found that an infusion of angiotensin II decreased the regional blood flow and tooth eruption, but increased the arterial blood pressure. In the present study, the infusion of Ringer's solution increased the regional blood flow (Figure 1b) and tooth eruption (Figure 1c, 2), but decreased the arterial blood pressure (Figure 1a). In both studies, a positive correlation was found between the regional blood flow and tooth eruption rate, and a negative correlation was found between the regional blood flow and arterial blood pressure. These studies indicate that the eruption rate of the rat incisor is modulated by the regional blood flow, and that regional blood flow and arterial blood pressure are inversely related.

The present study on the effects of an infusion of Ringer's solution on axial movements of the rat

incisor indicates that the regional vascular volume and/or pressure within the socket play an important role in determining the position of the incisor. However, many other factors could regulate the vascular volume and/or pressure within the socket, such as transcapillary fluid exchange based on Starling's hypotheses [8,23], the capacitance vessels in the periodontal ligament [24,25], and a low-compliance environment within the socket [26,27]. Future studies should attempt to clarify the precise mechanisms that determine the tooth position relative to the regional vascular pressure within the socket.

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