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作成者（著者）	Sato, Kanako / Kawamura, Gaku / Yamada, Yoshitsugu / Kitamura, Takayuki
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Original Article

Differences between the Effects of Sevoflurane and Propofol Anesthesia on Insulin Sensitivity in Fasted Rats Undergoing Descending Colostomy

Kanako Sato[1)]* Gaku Kawamura[2)] Yoshitsugu Yamada[2)] and Takayuki Kitamura[1)]

[1)]Department of Anesthesiology, Toho University Sakura Medical Center, Sakura, Japan
[2)]Department of Anesthesiology, Faculty of Medicine, The University of Tokyo, Tokyo, Japan

ABSTRACT

Introduction: Glucose metabolism is modified perioperatively, resulting in hyperglycemia. While surgical stress is the predominant factor for hyperglycemic responses, anesthesia also modifies glucose metabolism. We examined the effects of anesthetics on intraoperative insulin sensitivity in fasted rats.

Methods: Two experiments were carried out in this study. In experiment 1, fasted rats underwent descending colostomy under sevoflurane (n = 8) and propofol (n = 8) anesthesia without exogenous glucose administration. The surgery took approximately 30 min. Blood glucose, plasma insulin, tumor necrosis factor-α, and high-molecular-weight adiponectin levels were measured. Before and after surgery, insulin sensitivity was evaluated using the quantitative insulin sensitivity check index (QUICKI) in each rat. In experiment 2, fasted rats underwent descending colostomy under sevoflurane (n = 8) and propofol (n = 8) anesthesia with exogenous glucose administration. Subsequently, these rats underwent the insulin tolerance test (ITT); we measured blood glucose levels 30 min after insulin administration.

Results: In experiment 1, rats under propofol anesthesia showed significantly lower blood glucose levels and significantly higher plasma insulin levels than rats under sevoflurane anesthesia before and after surgery. Rats under propofol anesthesia had a significantly lower QUICKI than rats under sevoflurane anesthesia. In experiment 2, rats under propofol anesthesia had less decreases in blood glucose levels during ITT than rats under sevoflurane anesthesia. Additionally, plasma tumor necrosis factor-α (TNF-α) levels under propofol anesthesia were significantly higher than those under sevoflurane anesthesia.

Conclusions: Propofol anesthesia impairs insulin sensitivity during surgery in fasted rats, compared with sevoflurane anesthesia; TNF-α might be involved in insulin resistance.

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KEYWORDS: surgical stress, intraoperative glycemic control, glucose utilization, insulin, adipocytokine

*Corresponding Author: Kanako Sato, 564-1 Shimoshizu, Sakura, Chiba 285-8741, Japan, tel: +81–43–462–8811
e-mail: satok-ane@med.u-tokyo.ac.jp
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Introduction

Glucose metabolism in the perioperative period is modified by many factors and involves complex mechanisms. Hyperglycemia is a common adverse effect of surgery under general anesthesia. Surgical stress is considered as the predominant factor modifying glucose metabolism in the perioperative period, with impaired glucose utilization and enhanced glucose production resulting from the endocrine-metabolic responses to surgical stress.[1, 2] General anesthesia is considered another perioperative factor altering glucose metabolism.[3–7] In clinical settings, several kinds of anesthetics are administered to patients undergoing surgery under general anesthesia. There are two major categories of anesthetics for the maintenance of general anesthesia: volatile and intravenous anesthetics. Sevoflurane is one of the most popular volatile anesthetics, whereas propofol is one of the most popular intravenous anesthetics. Glucose utilization is regulated by both insulin secretion and insulin sensitivity. Previously, we reported the effects of general anesthesia on glucose utilization in fasted rats without surgical stress, showing that sevoflurane anesthesia impairs glucose-induced insulin secretion without affecting basic insulin secretion and insulin sensitivity, whereas propofol anesthesia enhances insulin secretion and induces insulin resistance.[8, 9] In this study, we compared insulin sensitivity in fasted rats undergoing descending colostomy under sevoflurane anesthesia with that under propofol anesthesia for further elucidation of intraoperative glucose metabolism. Several studies reported that adipocytokines, such as TNF-α and high-molecular-weight adiponectin (HMW-adiponectin), affect insulin sensitivity. Thus, we measured plasma levels of TNF-α and HMW-adiponectin.[10–14]

Methods

All experimental protocols were approved by the animal care committee of The University of Tokyo (protocol number: H13-047). Two experiments were carried out in this study. In experiment 1, fasted rats underwent descending colostomy under general anesthesia without exogenous glucose administration, and insulin sensitivity was evaluated using the quantitative insulin sensitivity check index (QUICKI) [15] in each rat (Fig. 1a). In experiment 2, fasted rats underwent descending colostomy under general anesthesia with exogenous glucose administration. Subsequently, these rats underwent the insulin tolerance test (ITT) [9, 16–19] for evaluation of insulin sensitivity (Fig. 1b).

Subjects

We purchased 8-week-old male Wistar rats (Nippon Bio-Supp. Center, Tokyo, Japan). Rats were housed in a regulated environment for 2 weeks at a room temperature of 25℃ and a 12-h light-dark cycle (07:00 to 19:00). All rats were allowed free access to a standard diet and water. Then, each rat was made to fast for 15 h prior to the experiment, although water was provided until commencement of the experiment. All experiments were performed between 9 a.m. and 3 p.m. A heat lamp and a heating pad were used to prevent hypothermia during the experiments. We monitored the rectal temperature of each rat throughout the experimental period.

Preparations

Anesthesia for perioperative preparations was provided with 5% sevoflurane (Maruishi Pharmaceutical Co., Ltd., Osaka, Japan) in 1 L/min oxygen via a tightly fitting face mask. All rats underwent tracheotomy. After tracheal intubation, sevoflurane (2.5% in 1 L/min oxygen) was administered *via* a tracheal tube, and the lungs were mechanically ventilated; the tidal volume was set at 2.5 mL and the respiratory rate at 60 breaths/min. A catheter was inserted into the right jugular vein, and another catheter was inserted into the right carotid artery. The preparation took approximately 30 min for each rat. All rats were administered 100 IU of heparin intravenously to maintain the patency of the catheters. The rats were subsequently divided into group S and group P, according to whether they received maintenance anesthesia with sevoflurane or propofol, respectively. In group S (n = 16), sevoflurane administration (2.5% in 1 L/min oxygen) was continued. The rats in group S were divided into two subgroups based on the intravenous fluids they received. In group S/saline (n = 8), a bolus dose of 4 mL/kg physiological saline was administered intravenously, followed by its continuous infusion at a rate of 10 mL/kg/h. In group S/glucose (n = 8), a bolus dose of 4 mL/kg physiological saline, followed by its continuous infusion at a rate of 9 mL/kg/h together with a continuous infusion of 50% glucose at a rate of 1 mL/kg/h, was intravenously administered. In group P rats (n = 16), sevoflurane administration was replaced by intravenous administration of a lipid-based formulation of propofol at a concentration of 10 mg/mL (AstraZeneca K. K., Osaka, Japan), the anesthetic being administered at a bolus dose of 4 mL/kg followed by continuous infusion at a rate of 4 mL/

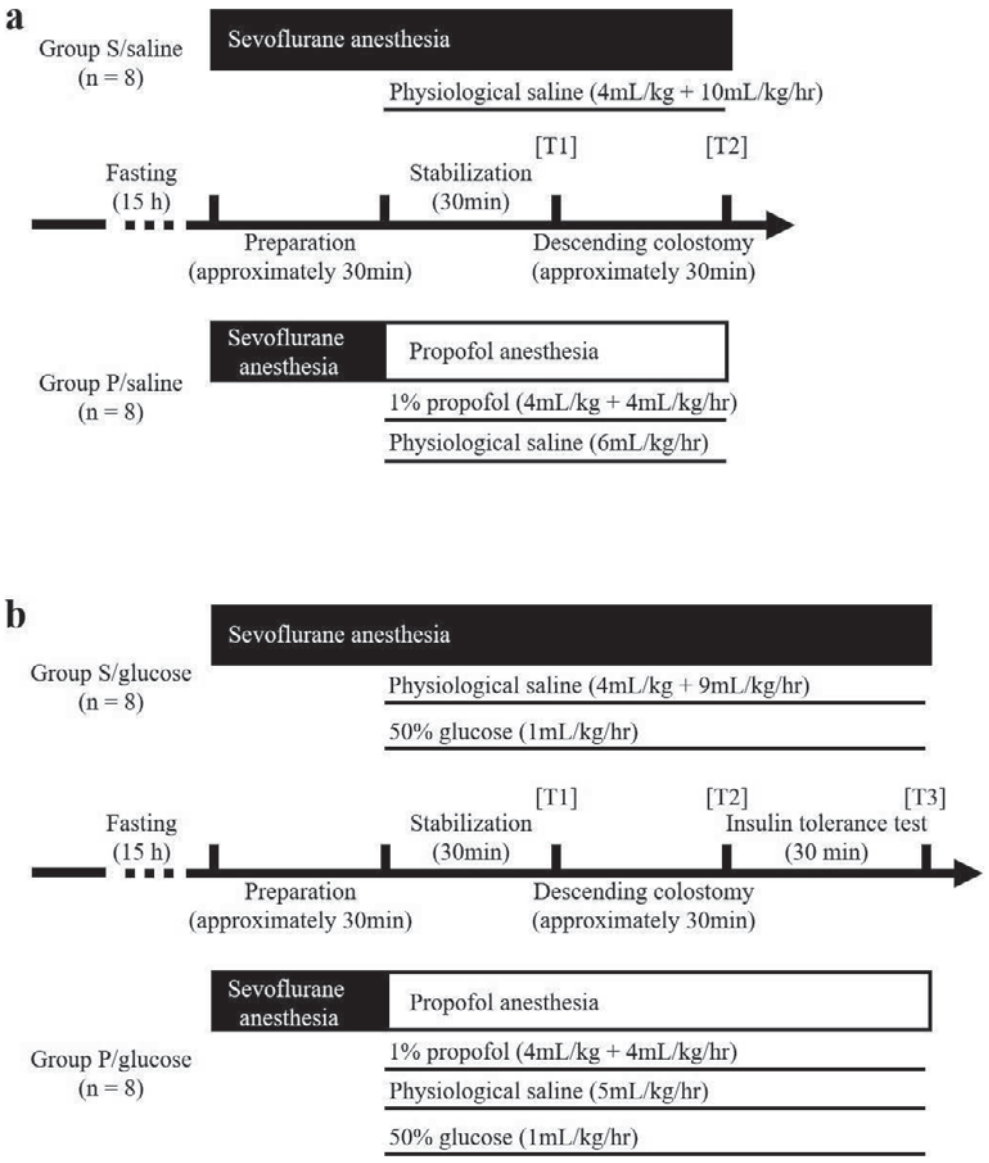

Fig. 1 Experimental protocols. A set of 32 rats were fasted for 15 h prior to the experiment.
a. Protocol of experiment 1. All rats underwent preparations under sevoflurane anesthesia. In group S/saline (n = 8), sevoflurane administration was continued (2.5% in 1 L/min oxygen). In group P/saline (n = 8), sevoflurane administration was discontinued, and instead, propofol solution was administered intravenously. Physiological saline was administered intravenously to rats in groups S/saline and P/saline. After a 30-min stabilization period, each rat underwent descending colostomy.
b. Protocol of experiment 2. All rats underwent preparations under sevoflurane anesthesia. In group S/glucose (n = 8), sevoflurane administration was continued (2.5% in 1 L/min oxygen). In group P/glucose (n = 8), sevoflurane administration was discontinued, and instead, propofol solution was administered intravenously. Physiological saline with glucose was administered intravenously to rats in groups S/glucose and P/glucose. After a 30-min stabilization period, each rat underwent descending colostomy. Subsequently, the rats underwent the insulin tolerance test. T1: just before descending colostomy. T2: just after descending colostomy. T3: 30 min after insulin administration.

kg/h. Rats in group P were also divided into two subgroups based on the intravenous fluids they received. Group P/saline (n = 8) rats received a continuous infusion of physiological saline at a rate of 6 mL/kg/h, whereas those in group P/glucose received a continuous infusion of physiological saline at a rate of 5 mL/kg/h and 50% glu-

cose as a continuous infusion at a rate of 1 mL/kg/h. The doses of sevoflurane and propofol were selected according to the protocols of our previous studies.[8, 9]

Descending colostomy

A 30-min stabilization period was allowed, following which all the rats underwent descending colostomy. Briefly, we made a midline abdominal incision from the xiphoid process to the pubic bone, applied a wound retractor to the abdominal wall, created colostomy in the descending colon, and closed the abdomen. Just before and after descending colostomy (T1 and T2, respectively), 1 mL of arterial blood was sampled.

Insulin tolerance test (ITT)

Immediately after the blood sampling at T2, 10 IU/kg of rapid-acting human insulin analogue (Humulin-R; Eli Lilly Japan K.K., Hyogo, Japan) was intravenously administered to rats in groups S/glucose and P/glucose.[9, 16–19] At 30 min after the insulin administration (T3), 0.2 mL of arterial blood was sampled to measure blood glucose levels.

Measurements and calculations

The arterial catheter was connected to a low-volume pressure transducer for monitoring mean arterial blood pressure (MAP) and heart rate (HR).

Immediately after each blood sampling, blood glucose levels were measured by the glucose oxidase method using a blood glucose meter (Medisafe; Terumo, Tokyo, Japan), and arterial PO_2, PCO_2, and plasma potassium levels were measured using i-STAT 1 Analyzer (Fuso Pharmaceutical Industries, Ltd., Osaka, Japan). Then, each blood sample was centrifuged at 1000 × g for 15 min at 4°C. Each plasma sample was stored at −60°C until analysis. Plasma insulin, TNF-α, and HMW-adiponectin levels were measured by an enzyme-linked immunosorbent assay using AKRIN-010T, AKRTN-010, and AKMAN-011 (Shibayagi Co., Ltd., Gunma, Japan), respectively. Based on the observed absorbance of the test samples, we assumed that the detection limits of TNF-α and HMW-adiponectin are 15.9 pg/mL and 3.13 ng/mL, respectively. Previously, we reported that the lipid-based formulation of propofol at the dose used in this study did not significantly affect any of the assays.[8]

The changes in blood glucose levels from T1 to T2 (Δglucose [surgery]) were calculated in each rat to evaluate the impact of surgical stress (i.e., descending colostomy) on blood glucose levels under general anesthesia: Δglucose [surgery] = [blood glucose level at T2 (mg/dL)] − [blood glucose level at T1 (mg/dL)].

For comparisons of insulin sensitivity between S/saline and P/saline groups, we calculated the QUICKI using blood glucose and plasma insulin levels in each rat as QUICKI = 1 / (log [blood glucose level (mg/dL)] + log [plasma insulin level (μIU/mL)]).[15] For comparisons of insulin sensitivity between groups S/glucose and P/glucose, we calculated the changes in blood glucose levels during ITT (Δglucose [ITT]) in each rat as Δglucose [ITT] = [blood glucose level at T3 (mg/dL)] − [blood glucose level at T2 (mg/dL)].[9]

Statistical analysis

Statistical analyses were performed using JMP 7.0.2 software (SAS Institute, Cary, NC).

Based on the data in our previous study,[8] a standard deviation in QUICKI was assumed to be 0.02. The required sample size to detect a difference of 0.04 in the mean QUICKI between the two groups using a two-tailed t-test with 95% power and a significance level of 0.05 was 16 rats (i.e., 8 rats per group).

Continuous data are shown as mean ± SD. Homogeneity of variance was examined using Bartlett's test; statistical significance was set at $P < 0.05$. The unpaired t-test was used for comparison of data with homogeneity of variance between the two groups at each time point; statistical significance was set at $P < 0.05$. Welch's t-test was used for comparison of data without homogeneity of variance between the two groups at each time point. Plasma TNF-α levels are shown as median [25th, 75th percentiles]. The Wilcoxon test was used for comparison of TNF-α levels between the two groups. Statistical significance was set at $P < 0.05$.

Results

Experiment 1: Insulin sensitivity in groups S/saline and P/saline

Rats in groups S/saline and P/saline weighed 307 ± 10 and 307 ± 8 g, respectively, indicating no significant difference between the two groups ($P = 1.000$, unpaired t-test). The time required for descending colostomy was 28.5 ± 0.6 and 29.3 ± 0.6 min in groups S/saline and P/saline, respectively, which were not significantly different ($P = 0.9713$, unpaired t-test). All rats in groups S/saline and P/saline survived throughout the experimental period.

Table 1 shows rectal temperature, MAP, HR, arterial PO_2, arterial PCO_2, and plasma potassium levels in groups S/saline and P/saline. There were no significant differences in rectal temperature at T1 and T2 between the two

Table 1 Vital signs, blood gas, and plasma potassium levels in groups S/saline and P/saline in experiment 1

		Group S/saline	Group P/saline
Rectal temperature (°C)			
Time point	T1	37.8 ± 0.1	37.2 ± 0.3
	T2	37.2 ± 0.4	37.2 ± 0.1
Mean arterial blood pressure (mmHg)			
Time point	T1	97 ± 10	121 ± 9 *
	T2	83 ± 15	73 ± 13
Heart rate (beats/min)			
Time point	T1	409 ± 29	401 ± 37
	T2	378 ± 38	378 ± 35
Arterial PO_2 (mmHg)			
Time point	T1	465 ± 51	506 ± 31
	T2	488 ± 43	489 ± 68
Arterial PCO_2 (mmHg)			
Time point	T1	34.9 ± 2.7	34.4 ± 2.5
	T2	33.9 ± 2.9	29.8 ± 2.8 *
Plasma potassium levels (mmol/L)			
Time point	T1	4.4 ± 0.4	4.2 ± 0.3
	T2	4.4 ± 0.8	4.4 ± 0.6

Data are shown as mean ± SD. T1: just before descending colostomy. T2: just after descending colostomy.
* $P < 0.05$ versus group S/saline at each time point, unpaired t-test.

groups ($P = 0.1067$ and 0.9746, respectively, Welch's t-test). Rats in group P/saline had significantly higher MAPs than those in group S/saline at T1 ($P = 0.002$, unpaired t-test); there were, however, no significant differences in MAP at T2 between the two groups ($P = 0.1958$, unpaired t-test). There were also no significant differences in HR at T1 and T2 ($P = 0.6176$ and 0.9947, respectively, unpaired t-test), arterial PO_2 at T1 and T2 ($P = 0.0758$ and 0.9519, respectively, unpaired t-test), and arterial PCO_2 at T1 between the two groups ($P = 0.6992$, unpaired t-test); however, group P/saline showed significantly lower PCO_2 than group S/saline at T2 ($P = 0.0123$, unpaired t-test). There were no significant differences in plasma potassium levels at T1 and T2 between the two groups ($P = 0.1301$ and 0.8899, respectively, unpaired t-test). Arterial blood gas analyses were not performed at T3.

Table 2 shows blood glucose levels, plasma insulin levels, QUICKI, plasma TNF-α levels, and plasma HMW-adiponectin levels in groups S/saline and P/saline. Group P/saline showed significantly lower blood glucose levels than group S/saline at T1 and T2 ($P < 0.0001$ and $P < 0.0001$, respectively, Welch's t-test). In group S/saline, blood glucose levels ranged from 98 to 121 mg/dL and from 114 to 151 mg/dL at T1 and T2, respectively. In group P/saline, blood glucose levels ranged from 35 to 89 mg/dL and from 47 to 110 mg/dL at T1 and T2, respectively; blood glucose levels below 50 mg/dL were observed in one of the eight rats at T1 and in one of the eight rats at T2. There were no significant differences in Δglucose [surgery] between the two groups ($P = 0.9351$, unpaired t-test); Δglucose [surgery] was 19 ± 13 and 18 ± 22 mg/dL in groups S/saline and P/saline, respectively. Group P/saline showed significantly higher plasma insulin levels than group S/saline at T1 ($P = 0.0021$, Welch's t-test) and T2 ($P < 0.0001$, unpaired t-test). Group P/saline showed significantly lower QUICKI than group S/saline at T1 ($P < 0.0021$, unpaired t-test) and T2 ($P < 0.0001$, Welch's test). TNF-α was not detected in any of the rats in group S/saline at T1 and T2; however, TNF-α was detected in seven rats at T1 and three rats at T2 in group P/saline. There were no significant differences in plasma HMW-adiponectin levels at T1 and T2 between the two groups ($P = 0.2793$ and 0.7990, respectively, unpaired t-test).

Experiment 2: Insulin sensitivity in groups S/glucose and P/glucose

Rats in groups S/glucose and P/glucose weighed 309 ± 19 and 310 ± 15 g, respectively, indicating no significant difference between the two groups ($P = 0.8836$, unpaired t-test). The time required for descending colostomy was not significantly different between the two groups, at 29.3 ± 1.8 and 29.0 ± 0.8 min in groups S/glucose and P/glucose, re-

Table 2 Assessment of parameters of glucose metabolism in groups S/saline and P/saline in experiment 1

		Group S/saline	Group P/saline
Blood glucose levels (mg/dL)			
Time point	T1	111 ± 7	61 ± 18 * *
	T2	129 ± 12	78 ± 21 * *
Plasma insulin levels (μIU/mL)			
Time point	T1	38 ± 18	910 ± 522 * *
	T2	42 ± 24	552 ± 36 *
Quantitative insulin sensitivity index			
Time point	T1	0.280 ± 0.014	0.214 ± 0.013 *
	T2	0.272 ± 0.016	0.217 ± 0.006 * *
Plasma tumor necrosis factor-α levels (pg/mL)			
Time point	T1	＜15.9 [＜15.9, ＜15.9]	1161.9 [802.3, 1897]
	T2	＜15.9 [＜15.9, ＜15.9]	＜15.9 [＜15.9, 238.2]
Plasma high-molecular-weight adiponectin levels (ng/mL)			
Time point	T1	610 ± 297	774 ± 285
	T2	585 ± 378	636 ± 409

Continuous data are shown as mean ± SD. Plasma tumor necrosis factor-α levels are shown as median [25th, 75th percentiles]. Plasma tumor necrosis factor-α levels in all rats in group S/saline were below the detection limit of our assay.
T1: just before descending colostomy. T2: just after descending colostomy. * P＜0.05 versus group S/saline at each time point, unpaired t-test. * * P＜0.05 versus group S/saline at each time point, Welch's t-test.

Table 3 Vital signs, blood gas, and plasma potassium levels in groups S/saline and P/saline in experiment 2

		Group S/glucose	Group P/glucose
Rectal temperature (℃)			
Time point	T1	37.6 ± 0.6	38.0 ± 0.7
	T2	36.9 ± 1.2	37.1 ± 1.4
	T3	37.5 ± 0.4	37.2 ± 0.6
Mean arterial blood pressure (mmHg)			
Time point	T1	89 ± 17	108 ± 9 *
	T2	74 ± 10	70 ± 10
	T3	71 ± 18	41 ± 11 *
Heart rate (beats/min)			
Time point	T1	387 ± 36	414 ± 50
	T2	339 ± 35	365 ± 19
	T3	413 ± 46	369 ± 28 *
Arterial PO_2 (mmHg)			
Time point	T1	461 ± 25	453 ± 73
	T2	493 ± 36	480 ± 30
Arterial PCO_2 (mmHg)			
Time point	T1	34.6 ± 2.8	35.2 ± 3.7
	T2	30.5 ± 2.9	30.6 ± 1.8
Plasma potassium levels (mmol/L)			
Time point	T1	4.0 ± 0.2	4.1 ± 0.6
	T2	4.1 ± 0.6	4.0 ± 0.4

Data are shown as mean ± SD. T1: just before descending colostomy. T2: just after descending colostomy. T3: 30 min after insulin administration. In all rats, arterial blood gas analysis was not performed at T3. * P＜0.05 versus group S/glucose at each time point, unpaired t-test.

spectively (P = 0.7292, Welch's t-test). All rats in groups S/glucose and P/glucose survived throughout the experimental period.

Table 3 shows rectal temperature, MAP, HR, arterial

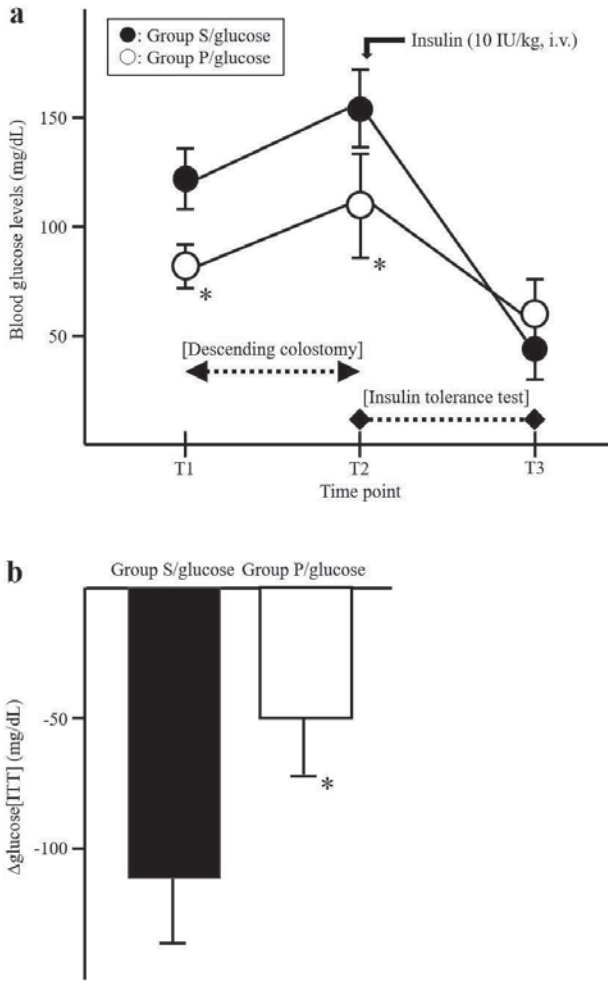

Fig. 2 Time courses of blood glucose levels and changes in blood glucose levels during the insulin tolerance test in groups S/glucose and P/glucose. a Time courses of blood glucose levels during descending colostomy and the insulin tolerance test. Group P/glucose showed significantly lower blood glucose levels than group S/glucose at T1 and T2 ($P<0.05$, unpaired t-test); however, there were no significant differences in blood glucose levels between the two groups at T3 ($P>0.05$, unpaired t-test). b Changes in blood glucose levels during the insulin tolerance test (Δglucose [ITT]). There was a significant difference in Δglucose [ITT] between the two groups ($P<0.05$, unpaired t-test). T1: just before descending colostomy. T2: just after descending colostomy. T3: 30 min after insulin administration.

PO_2, arterial PCO_2, and plasma potassium levels in groups S/glucose and P/glucose. There were no significant differences in rectal temperature at T1, T2, and T3 between the two groups (P = 0.1633, 0.6970 and 0.2009, respectively, unpaired t-test). When compared with group S/glucose, group P/glucose showed significantly higher MAP at T1 (P = 0.0137, unpaired t-test) and significantly lower MAP at T3 (P = 0.0011, unpaired t-test); however, no significant difference between the two groups was detected at T2 (P = 0.4166, unpaired t-test). Although there were no significant differences in HR at T1 and T2 between the two groups (P = 0.2408 and 0.0845, respectively, unpaired t-test), group P/glucose showed significantly lower HR at T 3 (P = 0.0344, unpaired t-test). There were no significant differences in arterial PO_2 at T1 and T2 (P = 0.7703 and 0.4705, respectively, unpaired t-test), arterial PCO_2 at T1 and T2 (P = 0.6993 and 0.9596, respectively, unpaired t-test), and plasma potassium levels at T1 and T2 between the two groups (P = 0.1311 and 0.8437, respectively, unpaired t-test). Arterial blood gas analyses were not performed at T3.

The time courses of blood glucose levels and Δglucose [ITT] in groups S/glucose and P/glucose are shown in Fig. 2. Group P/glucose showed significantly lower blood glucose levels than group S/glucose at T1 and T2 ($P < 0.0001$ and = 0.0009, respectively, unpaired t-test); however, there were no significant differences in blood glucose levels at T 3 between the two groups (P = 0.0624, unpaired t-test). In group S/glucose, blood glucose levels ranged from 107 to 147 mg/dL and from 125 to 178 mg/dL at T1 and T2, respectively. In group P/glucose, blood glucose levels ranged from 68 to 98 mg/dL and from 76 to 143 mg/dL at T1 and T2, respectively. There were no significant differences in Δglucose [surgery] between the two groups (P = 0.6571, unpaired t-test); Δglucose [surgery] was 33 ± 21 and 28 ± 24 mg/dL in groups S/glucose and P/glucose, respectively. There was a significant difference in Δglucose [ITT] between the two groups ($P < 0.0001$, unpaired t-test); Δglucose [ITT] was −111 ± 24 and −50 ± 21 mg/dL in groups S/glucose and P/glucose, respectively.

Table 4 shows plasma levels of insulin, TNF-α, and HMW-adiponectin in groups S/glucose and P/glucose. Group P/glucose showed significantly higher plasma insulin levels than group S/glucose at T1 and T2 (P = 0.0016 and 0.0019, respectively, Welch's t-test). TNF-α was detected in all rats in both groups at T1 and T2. Group P/glucose showed significantly higher plasma TNF-α levels than group S/glucose at both T1 and T2 (P = 0.0011 and = 0.0023, respectively, Wilcoxon test). There were no significant differences in plasma HMW-adiponectin levels at T1 and T2 between the two groups (P = 0.1947 and 0.1452, respectively, unpaired t-test).

Table 4 Plasma levels of insulin, tumor necrosis factor-α, high-molecular-weight adiponectin in groups S/glucose and P/glucose in experiment 2

		Group S/glucose	Group P/glucose
Plasma insulin levels (μIU/mL)			
Time point	T1	50 ± 28	952 ± 513 * *
	T2	59 ± 29	914 ± 502 * *
Plasma tumor necrosis factor-α levels (pg/mL)			
Time point	T1	806 [129.7, 919]	2237.3 [1912.4, 2603]
	T2	724.6 [152.7, 1024.2]	1454.3 [1168, 1785.9]
Plasma high-molecular-weight adiponectin levels (ng/mL)			
Time point	T1	492 ± 554	880 ± 587
	T2	177 ± 286	493 ± 503

Data are shown as mean ± SD. Plasma tumor necrosis factor-α levels are shown as median [25th, 75th percentiles]. T1: just before descending colostomy. T2: just after descending colostomy. * $P<0.05$ versus group S/glucose at each time point, unpaired t-test. * * $P<0.05$ versus group S/glucose at each time point, Welch's t-test.

Discussion

Consistent with our previous findings,[8, 9] the significantly lower QUICKI values at T1 in group P/saline compared with group S/saline suggest insulin resistance in fasted rats without surgical stress under propofol anesthesia. The significantly lower QUICKI values at T2 in group P/saline than group S/saline and the significantly smaller decreases in blood glucose levels during the ITT in group P/glucose than group S/glucose suggest insulin resistance in fasted rats exposed to surgical stress under propofol anesthesia. Blood glucose levels increased during descending colostomy in all four groups, S/saline, P/saline, S/glucose, and P/glucose, although no significant differences in Δglucose [surgery] (i.e., changes in blood glucose levels during descending colostomy) were detected between groups S/saline and P/saline and between groups S/glucose and P/glucose. These results suggest that the suppressive effects of propofol anesthesia on endocrine-metabolic responses to surgical stress associated with descending colostomy were equivalent to those of sevoflurane anesthesia, at the doses tested in this study. Sevoflurane has both analgesic and hypnotic effects. On the other hand, propofol is a hypnotic agent that has no analgesic effects. In clinical settings, surgical patients are administered propofol in combination with different kinds of analgesics. We intended to examine the effects of propofol alone on intraoperative insulin sensitivity. Thus, general anesthesia was maintained using propofol alone during the experiments in groups P/saline and P/glucose. We suppose that the surgical stress applied to each rat during descending colostomy is not small, because the surgical procedures include digestive tract manipulations. Based on these results, it can be assumed that the differences in the effects of sevoflurane and propofol anesthesia on insulin sensitivity seemed to be the major cause of the observed differences in intraoperative insulin sensitivity, and the contributions of surgical stress to the observed differences in intraoperative insulin sensitivity might be insignificant. These findings need to be verified by further investigations in which stronger surgical stress than descending colostomy is applied to test animals. Examination of insulin sensitivity during surgery in test animals anesthetized with propofol in combination with some kind of analgesic, such as opioids, should also be performed in the future.

Adenosine triphosphate-sensitive potassium channels (K_{ATP} channels) in β-islet cells regulate insulin secretion.[20] Our recent findings suggest that sevoflurane anesthesia attenuates glucose-induced insulin secretion by activating K_{ATP} channels in β-islet cells, whereas propofol anesthesia enhances insulin secretion by inhibiting K_{ATP} channels in β-islet cells.[8, 21] Previously, we reported that steep increases in plasma insulin levels were observed 30 min after the changing anesthesia regimens from sevoflurane anesthesia to propofol anesthesia.[8, 9, 21] Consistent with our previous findings,[8, 9, 21] plasma insulin levels in fasted rats under propofol anesthesia were significantly higher than those in fasted rats under sevoflurane anesthesia in this study. Taken together, glucose utilization during surgery under propofol anesthesia is characterized by insulin resistance with hyperinsulinemia. It was previously reported that plasma levels of adipocytokines (i.e., cytokines and hormones derived from adipocytes) affect insulin sensitivity.[10–14] Increases in plasma TNF-α levels[10, 13] and decreases

in plasma HMW-adiponectin levels are associated with insulin resistance.[11–14] In this study, plasma TNF-α levels in fasted rats during surgery under propofol anesthesia were notably higher than those under sevoflurane anesthesia; however, no significant differences in plasma HMW-adiponectin levels were detected. Therefore, we consider that insulin resistance during surgery under propofol anesthesia might be induced by the elevated plasma TNF-α levels. The mechanisms underlying the elevated plasma TNF-α levels could not be elucidated in this study. Generally, a lipid-based formulation of propofol is used for anesthesia because propofol has a hydrophobic nature. Thus, propofol anesthesia is always accompanied by an acute lipid load. Reportedly, insulin resistance can be induced by an acute lipid load.[22–25] In our recent study using fasted rats without surgical stress, we reported that an acute lipid load may induce insulin resistance under propofol anesthesia.[9] In the current study, plasma insulin levels were extraordinarily high during descending colostomy in group P/saline; however, blood glucose levels less than 50 mg/dL were observed in one out of eight rats just before descending colostomy and after descending colostomy. It is possible that insulin resistance plays an important role as a counterreaction to hyperinsulinemia for the prevention of critical hypoglycemia during surgery under propofol anesthesia.

It is generally accepted that the standard method for evaluation of insulin sensitivity is the hyperinsulinemic-normoglycemic clamp.[26] Considering the study design, it was difficult to perform the hyperinsulinemic-normoglycemic clamp in this study. In clinical settings, QUICKI is considered as a useful index of insulin sensitivity.[15] Insulin sensitivity evaluated by QUICKI correlates well with that evaluated by the hyperinsulinemic-normoglycemic clamp. In addition, some studies reported that QUICKI is also useful for the evaluation of insulin sensitivity in animal studies.[27, 28] Fasting blood glucose levels and plasma insulin levels are required for the evaluation of insulin sensitivity using QUICKI. Exogenous glucose administration influences blood glucose levels and plasma insulin levels. Thus, we consider that QUICKI is not appropriate for the evaluation of insulin sensitivity of rats in groups S/glucose and P/glucose. Thus, we applied QUICKI for the evaluation of insulin sensitivity in groups S/saline and P/saline. Due to the insufficient blood glucose levels for the performance of the ITT in group P/saline at T2 (i.e., just after descending colostomy), we performed ITT using rats undergoing descending colostomy with exogenous glucose administration (i.e., rats in groups S/glucose and P/glucose) for verifying the insulin sensitivity evaluated by QUICKI in groups S/saline and P/saline. There were significant differences in plasma insulin levels between groups S/glucose and P/glucose just before insulin administration: plasma insulin levels at T2 were 59 ± 29 and 914 ± 502 μIU/mL in groups S/glucose and P/glucose, respectively. For the ITT, we administered an extraordinarily high dose (i.e., 10 IU/kg) of a rapid-acting human insulin analogue to the rats in this study. It is, therefore, assumable that the differences in plasma insulin levels during ITT between groups S/glucose and P/glucose were negligible. There were also no significant differences in plasma potassium levels at T2 between groups S/glucose and P/glucose. We, thus, suppose that plasma potassium levels were not involved in the observed differences in insulin sensitivity between groups S/glucose and P/glucose.

Perioperative hyperglycemia is considered to be an independent risk factor for adverse events related to surgery.[29–31] Thus, adequate control of blood glucose levels throughout the perioperative period is required for the improvement of surgical prognosis. However, the optimal blood glucose level in the perioperative period is still unknown. Elucidation of the mechanisms underlying the perioperative hyperglycemic responses is required for the establishment of guidelines for glycemic control. Blood glucose levels reflect the balance between glucose production and utilization. Both surgical stress and general anesthesia are considered to be factors affecting glucose utilization and production. It is, therefore, necessary to elucidate the details of both these metabolic processes during surgery under general anesthesia. The results of this study using fasted rats suggest significant differences in the effects of sevoflurane and propofol anesthesia on glucose utilization during surgery. Indeed, we believe that these findings might be useful information for clinical practice, although further elucidation of these effects in clinical studies is still required.

There are several limitations to this study. Anesthesia for preoperative preparations was provided with sevoflurane in all the rats. Although a 30-min stabilization period was allowed in this study, there might have been residual effects of sevoflurane anesthesia on the observed insulin sensitivity in groups P/saline and P/glucose. There were significant differences in hemodynamic parameters between groups S/saline and P/saline and between groups

S/glucose and P/glucose, suggesting possible differences in sympathetic nervous activity, which is generally accepted as a factor affecting glucose metabolism. The fasting period prior to the experiment was set in each rat, because fasting blood samples are required for the evaluation of insulin sensitivity using QUICKI.[15, 32, 33] Glucose metabolism is drastically modified by fasting. In addition, it is reported that postoperative insulin resistance can be ameliorated by intake of a carbohydrate-rich drink during the fasting period.[26, 34, 35] Thus, the effects of a 15-h fasting period on the observed insulin sensitivity in this study cannot be neglected. There is a difference in the method applied for the evaluation of insulin sensitivity between the two groups; we applied QUICKI in groups S/saline and P/saline, while we performed ITT in groups S/glucose and P/glucose. Although acute lipid load might be a causative of insulin resistance under propofol anesthesia, we could not elucidate it in this study. According to the protocol of our previous study, [9] we took a 30 min observation period to evaluate insulin sensitivity during ITT in this study. Applying a longer observational period during ITT, we might observe further differences in Δglucose [ITT] between groups S/glucose and P/glucose. We used normal rats in this study. Further investigations using diabetic rats are required to further elucidate intraoperative insulin sensitivity in diabetic patients.

In summary, unlike sevoflurane anesthesia, propofol anesthesia induces insulin resistance accompanied by hyperinsulinemia in fasted rats undergoing descending colostomy. Increases in plasma TNF-α levels might be involved in the impaired insulin sensitivity under propofol anesthesia.

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Author contributions: KS, GK, and TK designed and conducted the experiments, analyzed the data, and prepared the manuscript. YY designed the experiments and prepared the manuscript. All authors approved the final version of the manuscript.

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