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# Acute effects of sarpogrelate, a 5-$HT_{2A}$ receptor antagonist on cytokine production in endotoxin shock model of rats

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ABSTRACT

Serotonin (5-HT)$_{2A}$ receptors are involved in cytokine production in infection or sepsis. Therefore, 5-$HT_{2A}$ receptor antagonist might be useful to treat sepsis. The present study investigates the effects of a 5-$HT_{2A}$ receptor antagonist, sarpogrelate on endotoxin shock. Catheters were inserted into the femoral artery and vein of Sprague–Dawley rats. First, sarpogrelate 0 (control), 3, or 10 mg/kg dissolved in 0.5 ml of distilled water has been given, followed by endotoxin 10 mg/kg in saline 0.5 ml 5 min later. Blood pressure, pulse rate and survival rate were monitored in 20 rats per dose. Blood gas and plasma cytokine concentrations were measured in 8 rats per dose. In four rats each of sarpogrelate 0, 3, or 10 mg/kg, and sham operation, the lung histology was examined. Zero, 15, and 12 rats survived for 8 h in the control, 3 mg/kg, and 10 mg/kg groups, respectively. The control group had the lowest blood pressure, pulse rate, pH and arterial oxygen tension, and the highest arterial carbon dioxide tension and plasma IL-1β concentration. The increase of TNF-α was significantly lower in 3 mg/kg group than in the control group. Pathological changes of the lung were inhibited in 3 and 10 mg/kg groups. In conclusion, sarpogrelate might be effective to decrease production of pro-inflammatory cytokines, to keep hemodynamics, to inhibit lung damage, and to decrease mortality in endotoxin shock.



## 1. Introduction

Gram-negative bacterial infection triggers multiple host defense mechanisms that result in the activation of monocytes, macrophages, neutrophils, and lymphocytes, which induces production of cytokines. Inflammatory cytokines promote further release of cytotoxic substances (Graziano et al., 1999; Berkow and Dodson, 1988). Therefore, there are many studies investigating the therapy of sepsis, targeting inhibition of cytokine production (Pallua and Heimburg, 2003; Macarthur et al., 2003).

Endotoxic lipopolysaccharides released by bacteria may activate serotonin (5-HT) containing platelets (Timmons et al., 1986). Increased platelet aggregation contributes to microvascular failure and plays a role in the development of organ dysfunction (Levi, 2005). It has been reported that 5-$HT_{2A}$ receptor antagonist, sarpogrelate inhibits platelet aggregation (Uchiyama et al., 2007) and vasoconstriction (Van Nueten et al., 1984). In addition, 5-HT has immunomodulatory effects on monocytes and macrophages (Sternberg et al., 1986). 5-$HT_{2A}$ receptors are reported to be involved in cytokine production of mononuclear cells in infection (Cloez-Tayarani et al., 2003). Therefore, 5-$HT_{2A}$ antagonists may influence cytokine production as well as platelet function during endotoxin shock. Yet only a few studies evaluated the effects of 5-HT on cytokine production (Foon et al., 1976; Cloez-Tayarani et al., 2003).

This study focused on cytokine production and hypothesized that 5-$HT_{2A}$ receptor antagonists might modulate cytokine production and the subsequent mortality in endotoxin shock. The present study investigated the effects of a clinically available 5-$HT_{2A}$ receptor antagonist, sarpogrelate on cytokine production, respiration, hemodynamics, histological changes of the lung, and mortality in endotoxin shock model of rats.

## 2. Material and methods

### 2.1. Animal preparation

After institutional approval, male Sprague–Dawley rats (300–350 g) were anesthetized with halothane 2% in oxygen 100%. Catheters (5 French Atom indwelling feeding tube, Atom Medical International, Tokyo, Japan) were inserted into the femoral artery and the femoral vein (16 rats for histological study were inserted with only the venous catheter.). Both catheters were subcutaneously tunneled under the skin of the head, connected to three-way stop cocks, and locked with heparinized saline. The rats were put in the cage with free access to water. One hour after the catheter insertion, recovery from surgery and anesthesia was checked with locomotor behavior, placing or stepping reflex and righting reflex. None of the rats in this study showed abnormal behavior in these tests.

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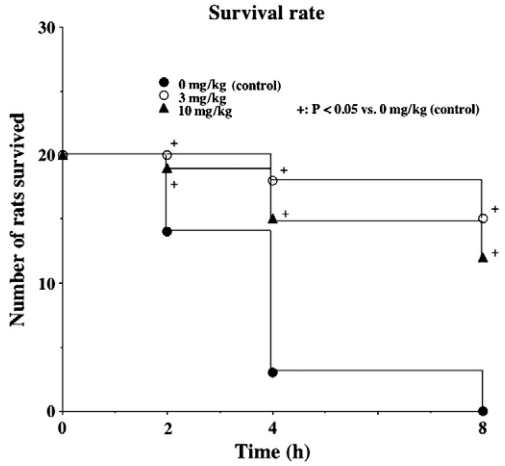


**Fig. 1.** Mortality. The Kaplan–Meier plot is shown. Fourteen, 20, and 19 rats at 2 h, 3, 18, and 15 rats at 4 h, and 0, 15, and 12 rats at 8 h survived in the control, 3 mg/kg, and 10 mg/kg groups, respectively.

### 2.2. Drug administration

Sarpogrelate (Mitsubishi Pharma Co. Ltd., Osaka, Japan) 0 (control), 3, or 10 mg/kg was dissolved in 0.5 ml of distilled water as recommended by the manufacturer and was administered intravenously. Five minutes later, endotoxin (*E. coli* 055:B5, Sigma, St. Louis, MO) 10 mg/kg dissolved in saline 0.5 ml was administered intravenously. Each administration was followed by injection of saline 2 ml to flush the catheter.

### 2.3. Mortality and hemodynamic study

Arterial blood pressure, pulse rate by arterial blood pressure wave, and survival rate were monitored for 8 h after endotoxin administration. Blood pressure and pulse rate were measured by connecting the catheters to the transducer when the rats were freely moving. Twenty rats were tested in each dose (60 rats in total). Only the data in blood pressure and pulse rate before death were included. All rats were sacrificed after 8 h observation by intraperitoneal injection of pentobarbital 50 mg/kg because no rats survived at 8 h in the control group.

### 2.4. Cytokine study

Twenty-four rats were prepared, as shown in the mortality study, after final exclusion of dead rats during the study (32, 1, and 2 rats in the control, 3 mg/kg, and 10 mg/kg groups, respectively). Drug administration was also the same (finally 8 rats in each group). Arterial blood 1.5 ml was drawn from femoral artery before sarpogrelate administration, and 2 and 4 h after endotoxin administration. After each drawing, saline 3 ml was injected to replace blood loss. Blood gas analysis was performed with ABL 625™ (Radiometer, Copenhagen, Denmark). The rest of the blood was centrifuged for 10 min with 3000 *g* and plasma was stored at −40 °C until the measurements. Plasma concentrations of IL-1β, 6, 8, and 10 and TNF-α were measured with enzyme-linked immunosorbent assay (Immunoassay Kit for Rat, BioSource International. Co. Ltd., Camarillo, CA, USA) at BCL Laboratory (Tokyo, Japan). The detection limit was 3 pg/ml for IL-1β, 8 pg/ml for IL-6, 5 pg/ml for IL-8 and IL-10, and 4 pg/ml for TNF-α.

### 2.5. Histological study

For rats that were inserted with only a femoral vein catheter, drug administration was the same as with the other tests. No drugs were administered to the sham operation group. Four rats in each group were studied after exclusion of dead rats (0, 13, 1, and 2 rats in sham, control, 3 mg/kg, and 10 mg/kg groups, respectively). Four h later, immediately after the animals have been sacrificed with intraperitoneal injection of pentobarbital 50 mg/kg, 10% formalin was infused into the lung through the trachea. Then the lung was excised with trachea and fixed with 10% formalin. Histology of the lung was examined using hematoxylin-eosin staining and scored as 0 (no changes), 1 (very slight changes), 2 (slight changes), 3 (moderate changes), or 4 (severe changes) by an animal pathologist who was blind to the treatment. Four sections were examined from each lung.

### 2.6. Data analysis

The data are shown as mean ± standard deviation or the number of rats. The Kaplan–Meier method was used to analyze mortality. Statistical analysis was performed with two-way factorial analysis of variance (ANOVA) followed by Bonferroni/Dunn test for blood pressure, pulse rate, blood gas analysis, and cytokine concentrations. Kruskal–Wallis test and the Mann–Whitney *U* test were used for analysis of the histological scores. A *P* value less than 0.05 was considered to be statistically significant.

## 3. Results

### 3.1. Mortality and hemodynamic study

Mortality was the highest in the control group (Fig. 1). Some rats died during the study, therefore, only 14, 20, and 19 rats at 2 h, 3, 18,

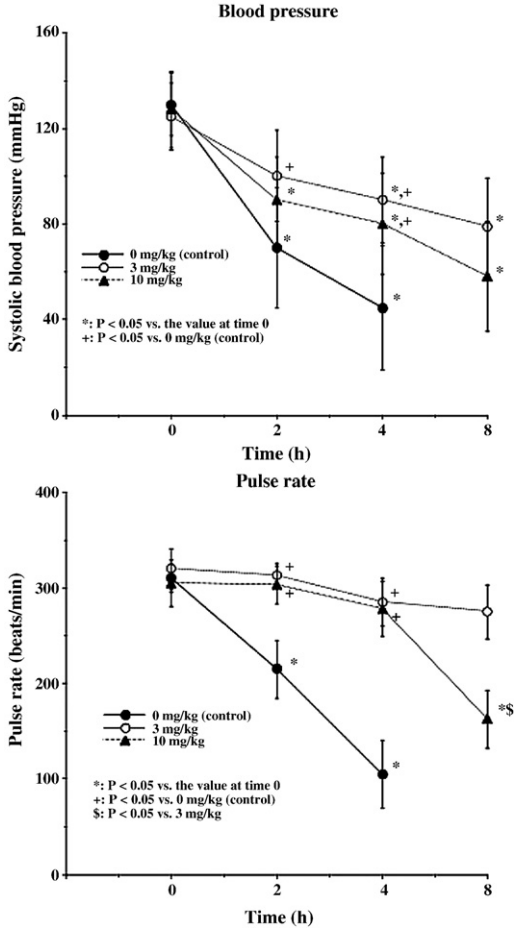


**Fig. 2.** Hemodynamics. Blood pressure (upper) and heart rate (lower). Mean ± standard deviation $N = 20$ at time 0 in all doses, 14, 20, and 19 at 2 h, 3, 18, and 15 at 4 h, and 0, 15, and 12 at 8 h, in the control, 3 mg/kg, and 10 mg/kg groups, respectively. Therefore, no data of the control group are shown at 8 h.

**Table 1**
Arterial blood gas analysis.

| Sarpogrelate dose | Time (h) | pH | $PaO_2$ (mmHg) | $PaCO_2$ (mmHg) |
|---|---|---|---|---|
| 0 mg/kg (control) | 0 (40) | 7.35 ± 0.06 | 124.5 ± 15.4 | 40.5 ± 2.7 |
| | 2 (31) | 7.19 ± 0.25 | 82.3 ± 25.5[a] | 60.7 ± 9.5[a] |
| | 4 (8) | 7.06 ± 0.18[a] | 55.7 ± 25.6[a] | 78.3 ± 12.6[a] |
| 3 mg/kg | 0 (9) | 7.34 ± 0.07 | 125.7 ± 17.9 | 40.1 ± 2.8 |
| | 2 (9) | 7.31 ± 0.11 | 115.0 ± 22.2[b] | 47.4 ± 3.8[a,b] |
| | 4 (8) | 7.30 ± 0.13[b] | 102.1 ± 19.5[a,b] | 50.2 ± 7.9[a,b] |
| 10 mg/kg | 0 (10) | 7.35 ± 0.09 | 120.5 ± 20.3 | 41.6 ± 1.8 |
| | 2 (9) | 7.30 ± 0.11 | 103.8 ± 23.1 | 48.2 ± 4.3[a,b] |
| | 4 (8) | 7.27 ± 0.17 | 98.5 ± 20.9[a,b] | 52.7 ± 6.8[a,b] |

Time 0, before sarpogrelate administration; Time 2 and 4, 2 and 4 h after endotoxin administration, respectively.
$N = 8$ for each dose group. In the parenthesis in the Time, number of rats tested is shown.
[a] $P<0.05$ vs. the value at time 0.
[b] $P<0.05$ vs. 0 mg/kg (control).

and 15 rats at 4 h, and 0, 15, and 12 rats at 8 h in the control, 3 mg/kg, and 10 mg/kg groups, respectively were included in analysis of hemodynamic data.

Blood pressure decreased in all doses with significantly lower values at 2 and 4 h in the control group (Fig. 2). Pulse rate decreased in the control and 10 mg/kg groups. It was significantly lower in the control group at 2 and 4 h (Fig. 2). At 8 h, pulse rate in the 10 mg/kg group was lower than that in the 3 mg/kg group.

### 3.2. Cytokine study

The study was repeated until 8 rats in each group lived for 4 h, therefore, 40, 9, and 10 rats for the control value, 31, 9, and 9 rats at 2 h, and 19, 8, and 8 rats at 4 h in the control, 3 mg/kg, and 10 mg/kg groups, respectively were included. The pH and arterial oxygen tension ($PaO_2$) decreased and arterial carbon dioxide tension ($PaCO_2$) increased significantly in all groups, and these changes were smaller in the 3 mg/kg and 10 mg/kg groups than in the control group (Table 1).

Plasma concentration of IL-1β increased in all groups but showed significantly higher levels at 2 and 4 h in the control group (Fig. 3). Plasma concentration of IL-8 increased in all groups with significantly higher values at 2 h in the control group (Fig. 3). Plasma TNFα levels were the highest at 2 h with significantly higher values in the control group (Fig. 3). Interleukin-6 increased in all groups without any inter group differences (Fig. 3). Plasma IL-10 concentrations increased with significantly higher levels at 2 h in the 3 mg/kg group (Fig. 3).

### 3.3. Histological study

Samples from 4 rats in each group were examined. Inflammatory changes such as peri-vascular, peri-bronchial, and alveolar granulocyte and lymphocyte infiltration, and increases of alveolar macrophages were severe in the endotoxin treated groups in comparison with the sham group, and these inflammatory changes significantly decreased in the 3 mg/kg group, while no changes were observed in the 10 mg/kg group (Table 2). Typical example of histology in each group was shown in the Fig. 4.

## 4. Discussion

Pretreatment with sarpogrelate inhibited the decrease in blood pressure, pulse rate, pH and $PaO_2$; decreased inflammatory changes of the lung and mortality; inhibited the increase of $PaCO_2$, IL-1β, IL-8 and TNF-α; increased IL-10 in endotoxin shock model of rats. However, these effects were not dose dependent.

Sarpogrelate was administered before the onset of endotoxin. Therefore, the results are not applicable to the clinical practice where the endotoxin effect is already present before drug administration. We

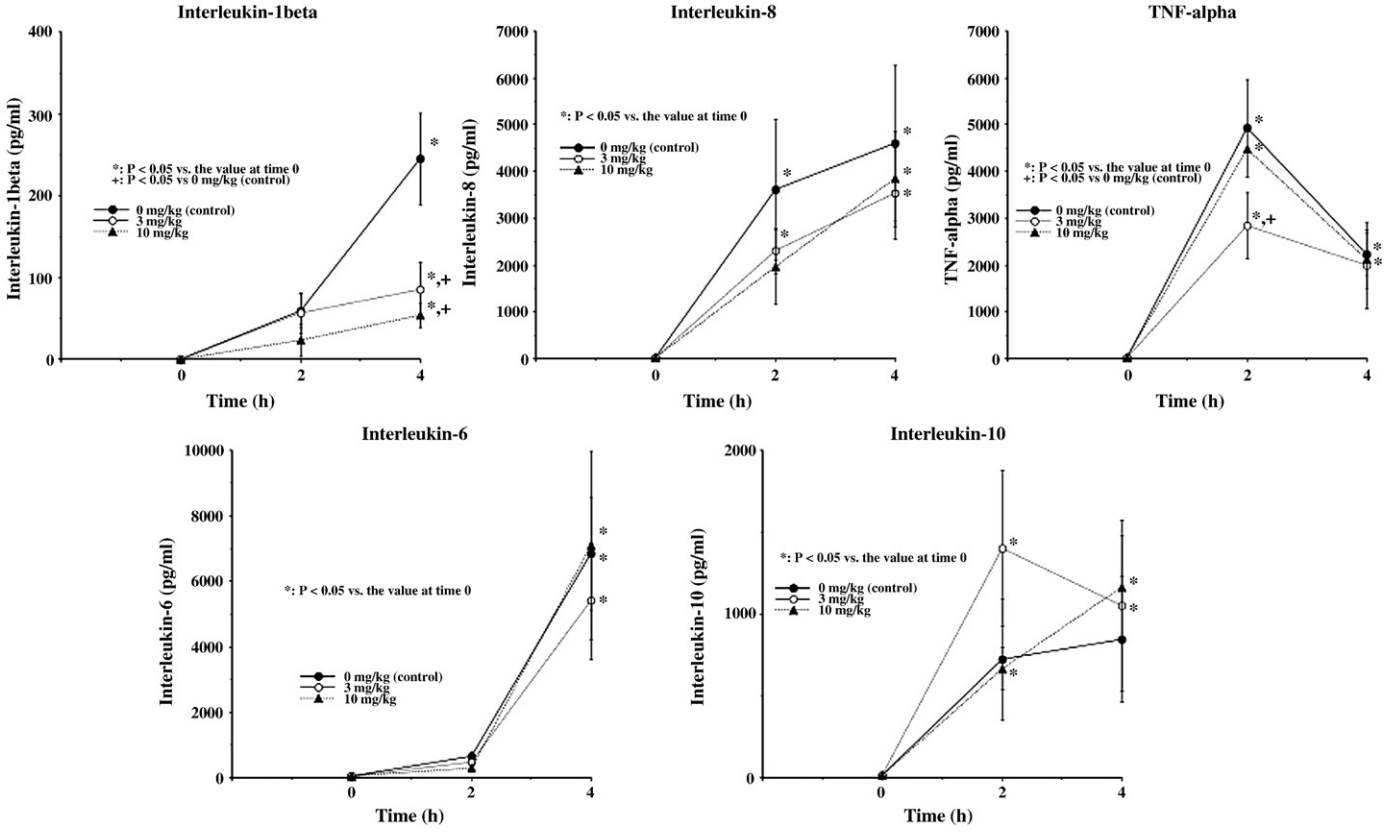


**Fig. 3.** Cytokines. Interleukin-1β, interleukin-6, interleukin-8, interleukin-10, and tumor necrosis factorα. Mean ± standard deviation. $N = 40$, 31, and 8 at time 0, 2, and 4 in the control (0 mg/kg) group, 9, 9, and 8 at time 0, 2, and 4 in the 3 mg/kg group, and 10, 9, and 8 at time 0, 2, and 4 in the 10 mg/kg group, respectively.

**Table 2**
Score of the histological changes.

| | Sham | Control 0 mg/kg | 3 mg/kg | 10 mg/kg |
|---|---|---|---|---|
| Accumulation of mucus in the bronchus | 0,0,0,0 | 1,1,3,0 | 0,0,1,0 | 1,1,1,0,1 |
| | 0 | 1 | 0 | 1 |
| Hyperplasia of the bronchial epithelium | 2,5,0,1 | 3,2,5,4 | 0,3,3,0 | 4,4,2,0 |
| | 1 | 1 | 0 | 1 |
| Increase of goblet cells in the bronchial epithelium | 2,2,3,1 | 6,3,8,6[a,b] | 0,1,3,0 | 5,7,3,2 |
| | 1 | 2 | 0 | 1 |
| Formation of lymphatic follicles in peri-bronchus and vessels | 2,4,5,3 | 9,8,7,10[b] | 4,9,7,7 | 8,6,11,6[b] |
| | 1 | 2 | 1 | 2 |
| Proliferation and degeneration of the glandular epithelium | 0,2,0,0 | 0,0,3,0 | 0,0,2,0 | 0,0,0,0 |
| | 0 | 0 | 0 | 0 |
| Peri-bronchial granulocyte infiltration | 2,2,2,3 | 5,5,6,6[a,b] | 2,1,3,1 | 4,3,6,4[a,b] |
| | 1 | 1 | 0 | 2 |
| Peri-bronchial lymphocyte infiltration | 2,3,3,4 | 9,8,7,10[b] | 6,8,6,5[b] | 7,8,7,8[b] |
| | 1 | 2 | 2 | 2 |
| Peri-bronchial fibrosis | 2,2,4,2 | 2,4,4,4 | 0,3,2,3 | 3,0,3,2 |
| | 1 | 1 | 1 | 0 |
| Peri-vascular granulocyte infiltration | 2,2,4,0 | 2,2,4,4 | 0,3,1,4 | 2,1,1,2 |
| | 1 | 1 | 0 | 0 |
| Peri-vascular lymphocyte infiltration | 3,6,8,4 | 5,4,5,7 | 3,6,2,8 | 4,4,5,6 |
| | 2 | 2 | 2 | 1 |
| Increase of alveolar macrophage | 2,5,3,1 | 3,3,4,4 | 4,4,7,1 | 0,0,5,2 |
| | 1 | 1 | 1 | 0 |
| Alveolar granulocyte infiltration | 3,3,1,0 | 4,1,3,3 | 5,3,5,0 | 1,2,4,4 |
| | 0 | 1 | 1 | 1 |

Histological changes of the lung were scored as 0 (no changes), 1 (very slight changes), 2 (slight changes), 3 (moderate changes), or 4 (severe changes). The numbers in the upper row show the total score of each 4 sections of the lung from one rat. Four rats in each group, therefore, 4 numbers are shown in one group. In the lower row median score of the group was shown.
[a] $P<0.05$ vs. 3 mg/kg.
[b] $P<0.05$ vs. Sham.

need further studies of post administration of sarpogrelate in order to confirm its "curative" clinical usefulness, while the present study with pretreatment is still useful as a first experiment.

In the histological study, we did not assess the histology of dead rats within 4 h. Therefore, when we consider the real effects of sarpogrelate, 3 mg/kg might have induced larger improvement than presented in this study because the dead rats might have more deleterious histological changes.

We used a rat model of intravenous administration of endotoxin (*E. coli*) 10 mg/kg. Symptoms of gram-negative bacterial sepsis can be reproduced experimentally by treating animals with endotoxin (Zivot et al., 1995). In rats administered endotoxin 15 mg/kg intravenously, blood pressure significantly decreased in 3 h, heart rate decreased in 4 h, serum TNF-α increased in 2 h, and IL-6 increased in 4 h (Taniguchi et al., 2003). Our model showed almost consistent results with their study. This model is quite severe, therefore, we could obtain the data only for 8 h. In addition, all rats did not receive fluid infusion, respiratory support, or other treatment, which is different from the clinical situation. However, sarpogrelate had significant effects in this model, therefore, sarpogrelate is expected to be useful in improving endotoxin shock, which should be further studied in the future using other animal models such as cecal ligation and puncture model or pneumonia, etc.

We separated the mortality and hemodynamic study, cytokine study, and histological study because drawing a large amount of blood samples might have caused changes in hemodynamics, mortality and histology. We did not study the sham group for mortality, hemodynamic and cytokine studies to save the number of the animals, while some effects of catheter insertion on these studies might exist.

There are some 5-$HT_{2A}$ receptor antagonists other than sarpogrelate. Sarpogrelate has higher binding affinity to 5-$HT_{2A}$ receptor in comparison to other 5-$HT_2$ receptor antagonists, but its affinity is slight lower than that of ketanserin (Muntasir et al., 2006). Sarpogrelate shows strong affinity to 5-$HT_2$ receptors, but not to 5-$HT_1$, $\alpha_1$-,$\alpha_2$-, and β-adrenergic receptors and muscarinic receptors (Maruyama et al., 1991). In 5-$HT_2$ receptor subtypes, sarpogrelate has higher binding affinity to 5-$HT_{2A}$ receptor than 5-$HT_{2B}$ and 5-$HT_{2C}$ receptors (Rashid et al., 2003). Ketanserin has higher affinity to the$\alpha_1$-adrenergic

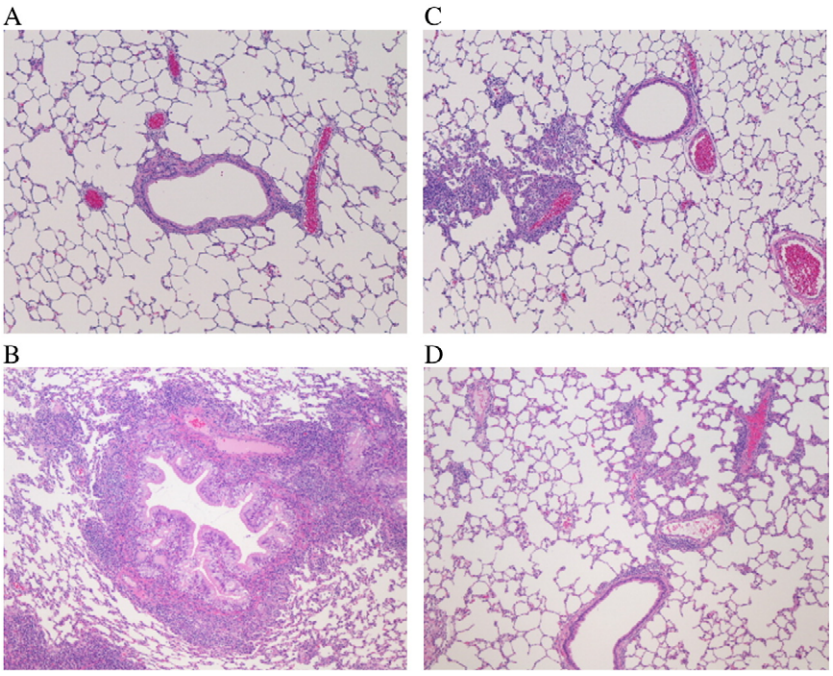


**Fig. 4.** Histology. Typical histology of each group is shown by hematoxylin-eosin staining (×10). Sham group (A); Very slight peri-vascular lymphocyte infiltration and peri-bronchial lymphatic follicles are seen. Control (0 mg/kg) group (B); Moderate hyperplasia of the bronchial epithelium, peri-bronchial and peri-vascular lymphatic follicles, and peri-bronchial lymphocyte infiltration, slight increase of goblet cells in the bronchial epithelium, peri-bronchial granulocyte infiltration, and peri-vascular lymphocyte infiltration are observed. 3 mg/kg group (C); Moderate peri-bronchial and peri-vascular lymphatic follicles, slight increase of goblet cells in the bronchial epithelium, and very slight peri-bronchial and peri-vascular lymphocyte infiltration are seen. 10 mg/kg group (D); Slight peri-bronchial and peri-vascular lymphocyte infiltration, and very slight increase of goblet cells in the bronchial epithelium, and peri-bronchial and peri-vascular granulocyte infiltration are observed.

receptor than sarpogrelate (Israilova et al., 2001). Therefore, we selected sarpogrelate as a 5-$HT_{2A}$ receptor antagonist.

According to the manufacturer, the maximum safe i.v. dose of sarpogrelate in rat was 45 mg/kg, therefore, we did a preliminary study administering 3, 10, and 30 mg/kg sarpogrelate. In that study with the absence of endotoxin, blood pressure decreased dose dependently by sarpogrelate (3, 10, and 30 mg/kg). The 30 mg/kg dose killed some animals, probably due to the vasodilating effects of sarpogrelate (Frishman et al., 1995). Therefore, we did not increase the dose to 30 mg/kg in the present study. The difference in blood pressure between 3 mg/kg and 10 mg/kg was not statistically significant, but at 2 h the blood pressure significantly decreased compared to the control value only with 10 mg/kg. Heart rate did not increase after endotoxin administration in the present study as sometimes shown in the acute phase of septic shock. This might be due to severe endotoxin shock of this model and because the first measurement was done 2 h after the administration when already heart rate might decrease. The decrease in blood pressure and heart rate might have some effects on cytokine production and inflammatory cell infiltration. That might be the reason why no dose dependency was observed. One another possibility for this could be that sarpogrelate (or 5-$HT_{2A}$ receptor) has different actions on cytokine production depending on the dose (or receptor activity).

The cytokines implicated as major mediators of the pathophysiologic changes of sepsis syndrome are TNF-α and IL-1β (Cannon et al., 1990; Casey et al., 1993; Libert et al., 1992; Michie et al., 1988). IL-6 is also considered to play an important role in modulating both acute-phase protein synthesis and immunologic response to infection (Gennari et al., 1994). The balance between cytokines with pro-inflammatory properties (TNFα, IL-1β, IL-6, and IL-8) and those with anti-inflammatory properties (IL-4 and IL-10) is important in the control of the duration and severity of an inflammatory response.

Anti-inflammatory cytokines such as IL-4, and IL-10 inhibit the formation of TNFα and IL-1β (Aderka et al., 1989; Cassatella et al., 1993; Selzman et al., 1998; Tilg et al., 1994; Vannier et al., 1992). Our results showed that sarpogrelate might inhibit the production of pro-inflammatory cytokines, TNFα, IL-8, and IL-1β. In addition, the rats treated with sarpogrelate had a higher level of IL-10, an anti-inflammatory cytokine than the control group. The mechanism of this cannot be discussed from the present results. However, this increase might have some effects on decreasing pro-inflammatory cytokines. The effect of 5-HT on IL-6 production was significantly inhibited by sarpogrelate in the study by Ito et al. (2000). It is also reported that 5-HT inhibited the production of TNF-α, but it did not significantly alter the production of other cytokines such as IL-6, and IL-10 in human peripheral blood mononuclear cells (Cloez-Tayarani et al., 2003). The inhibition of TNFα production by 5-HT involves the participation of the 5-$HT_{2A}$ receptor subtypes in human blood mononuclear cells (Cloez-Tayarani et al., 2003). However, our study showed that sarpogrelate, a 5-$HT_{2A}$ receptor antagonist inhibited the increase of TNFα. The quoted study (Cloez-Tayarani et al., 2003) was performed *in vitro* using human mononuclear cells while our study was done *in vivo* in rats. Therefore, many factors other than pure receptor function on a single cell line, i.e. blood pressure, respiratory factor, humoral factor, or other mediators could have affected our results. Especially, inhibition of the increase of IL-1βand increase of IL-10 might have inhibited the increase of TNFα in the present study.

The decrease in $PaO_2$ and increase in $PaCO_2$ were inhibited by sarpogrelate in the present study. In endotoxin shock, pulmonary edema, followed by acute lung injury with pulmonary hypertension occurs, in which serotonin had some roles (Sibbald et al., 1980). Walther et al. (2001) reported that leukocyte independent vascular permeability induced by endotoxin was inhibited by methysergide, a 5-$HT_1$ and 5-$HT_2$ receptor antagonist with a stronger affinity to the 5-$HT_2$ receptor. Ketanserin, one of the 5-$HT_{2A}$ receptor antagonists was effective in maintaining vascular wall shear rate, reducing leukocyte–endothelial interaction, and reducing macromolecular efflux during endotoxemia (Walther et al., 2007). Therefore, sarpogrelate might have inhibited vascular permeability in the lung in the present study, although we only studied blood gas and histological changes, along with inflammatory changes. In addition, sarpogrelate has its effects on restoring microcirculation. Further study to measure pulmonary arterial pressure, lung water volume, and microcirculation would be necessary to confirm this assumption.

Platelet activation and coagulation disorder are also important in pathophysiology of sepsis (Faust et al., 2001; Levi, 2005). A 5-$HT_{2A}$ receptor is involved in activating platelet aggregation and sarpogrelate inhibits it dose dependently (Uchiyama et al., 2007). In addition, sarpogrelate inhibited vasoconstriction by thrombus formation (Nishihara et al., 2006). Therefore, we assume that these effects of sarpogrelate must have contributed to the decrease of mortality and hemodynamic depression in the present study, although we did not measure anything in platelet function and coagulation factors. In addition, sarpogrelate inhibited vasoconstriction by thrombus formation (Nishihara et al., 2006). However, to know which is the major mechanism of the effect of sarpogrelate on sepsis, further studies on platelet function and coagulation are necessary using the same model.

To inhibit platelet aggregation by sarpogrelate carries the risk of bleeding. We did not perform necropsy, therefore, we did not know exactly the reason of their death. Although no apparent bleeding was observed, we can't deny the bleeding as a causative factor of their death and blood pressure decrease.

5-HT receptors exist in platelets, mast cells, endothelial cells of the vessels, brain, etc. We administered sarpogrelate intravenously. It is reported that sarpogrelate does not go into the brain from systemic circulation blocked at the blood brain barrier (Komatsu et al., 1991). Therefore, the effects of sarpogrelate in the present study were not mediated by the 5-HT receptors in the brain.

In conclusion, sarpogrelate might be effective on endotoxin shock as it decreases pro-inflammatory cytokines, increases anti-inflammatory cytokines, maintains hemodynamics, decreases inflammation of the lung, and decreases mortality.

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