ORIGINAL

Feasibility of a new hollow fiber silicone membrane oxygenator for long-term ECMO application

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Abstract : Currently in United States, there are no clinically-applicable hollow fiber extracorporeal membrane oxygenation (ECMO) oxygenators available. Therefore, our laboratory is in the process of developing a silicone hollow fiber membrane oxygenator for long-term ECMO usage. This oxygenator incorporates an ultrathin silicone hollow fiber. At this time, a specially-modified blood flow distributor (one chamber distributor) is centered in the module to prevent blood stagnation. An *ex vivo* long-term durability test for ECMO was performed using a healthy miniature calf for 2 weeks. Venous blood was drained from the left jugular vein of a calf, passed through the oxygenator and infused into the left carotid artery using a Gyro C1E3 centrifugal blood pump. A successful 2-week *ex vivo* experiment was performed. The O_2 and CO_2 gas transfer rates were maintained at the same value of 40 ml/min at a blood flow rate of 1 L/min flow and V/Q=3 (V=gas flow rate ; Q=blood flow rate). The plasma free hemoglobin was maintained around 5 mg/dl. After the experiment, no blood clot formation was observed in the module and no abnormal necropsy findings were found. These data suggest that the performance of this newly-improved oxygenator was stable, reliable, and acceptable for long-term ECMO. J. Med. Invest. 49 : 156-162, 2002

Keywords **:** *hollow fiber, silicone membrane, oxygenator, extracorporeal membrane oxygenation, long-term ex vivo study*

INTRODUCTION

An effective microporous hollow fiber oxygenator has been developed for cardiopulmonary bypass procedures ; however, since 1970, no improvements have been made for an effective extracorporeal membrane oxygenation (ECMO) system (1). The Kolobow spiral coil membrane oxygenator (Medtronic Inc., Anaheim, CA, U.S.A.) was developed in 1972 (2) and is the only practical available ECMO system in the United States. This oxygenator is an old-fashioned device that is inefficient and difficult to handle.

To solve this problem, our laboratory is developing a new oxygenator using ultrathin silicone rubber hollow fibers (3-7). These devices have demonstrated better biocompatibility and higher gas transfer performances over and above the already existing ECMO oxygenator (6, 7). For further high performance and antithrombogenicity, an oxygenator with an improved blood flow distributor was fabricated and evaluated with *in vitro* (8) and short term *ex vivo* (9) testing. The purpose of this study was to evaluate its clinical long-term feasibility for ECMO using an animal model.

Received for publication June 27, 2002 ; accepted July 31, 2002.

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METHODS

Animal preparation

The animal involved in this study received humane care in compliance with the *"Guiding Principles in the Case and Use of Animals"* approved by the Council of the American Physiological Society (revised 1980) and the *"Guide for the Care and Use of Laboratory Animals"* published by the National Institutes of Health (NIH Publication No. 85-23, revised 1985). A healthy miniature female calf (Dexter strain) weighing 90 kg and 11 months old was the subject of this study. A calf of this age was essential for this study because an animal with normal physiological adult circulation was needed, without abnormal anatomy due to the fetal circulation (i.e. atrial septal defect). Complete blood cell count (CBC), blood biochemistry, and plasma free hemoglobin were examined before surgery.

Surgical procedure

Anesthesia was induced using 4% halothane with 50% nitrous oxide through a special mask. When the animal was anesthetized, endotracheal intubation was performed and anesthesia was maintained by 1-2% halothane with 100% oxygen. A 15 cm longitudinal incision was made along the jugular vein on the left side of the neck, and the left carotid artery and jugular vein were dissected. The arterial cannula was inserted through a small arteriotomy and threaded proximally into the artery ; the venous cannula was inserted through the jugular vein in the proximal direction in the same manner. After placing the arterial and venous cannulae, both were externalized and connected to the extracorporeal circuit, and the pump circuit was activated. The Gyro C1E3® (Kyocera Corporation, Kyoto, Japan) was used for the centrifugal pump. Mean arterial pressures were maintained at more than 80 mmHg. Activated clotting time (ACT) was maintained at approximately 400 sec throughout the experiment using heparin. When the calf completely recovered from the anesthesia, the ECMO study was started. At the end of experiment, euthanasia and necropsy were performed.

Membrane Oxygenator

A new silicone hollow fiber membrane oxygenator (Preproduction model PPM-04, Fuji Systems Inc., Tokyo, Japan) was fabricated for long-term ECMO application based on the results of a previous model (PPM-03) (8, 9). The major changes in the new improved model (PPM-04) are an increase in the fiber length (from 100 to 150 mm) and the surface area (from 0.8 to 1.0 m^2) to increase the gas transfer rate, a decrease in the packing density (from 45 to 40%) to decrease the pressure drop, and a specially-designed blood flow distributor (from 4 chambers to 1 chamber) incorporated into the center of the module to prevent blood stagnation. Consequently, the new ECMO oxygenator module was 220 mm long and contained in a silicone coated acrylic housing. The priming volume of this module was 200 ml. This oxygenator was of the extracapillary flow type, in which blood flows outside the hollow fibers (Fig. 1).

Measurements

Gas transfer rate : At a blood flow rate of 1 L/min and V/Q=3 (V=gas flow rate ; Q=blood flow rate), the O_2 and CO_2 gas transfer rates were evaluated for 2 weeks (ECMO condition). Blood gas samples, taken from the inlet and outlet sampling ports, were analyzed every day using a System 1306 pH/blood gas analyzer (Instrumentation Laboratories, Lexington, MA, U.S.A.). Three samples were measured for each sampling time. The O_2 content and O_2 transfer rate and the CO_2 content and transfer rates were calculated by the following standard formulas :

$$O_2 \text{ content (Vol\%)} = (Hb \times 1.34 \times \%O_2 \text{ saturation}) / 100 + PO_2 \times 0.003$$

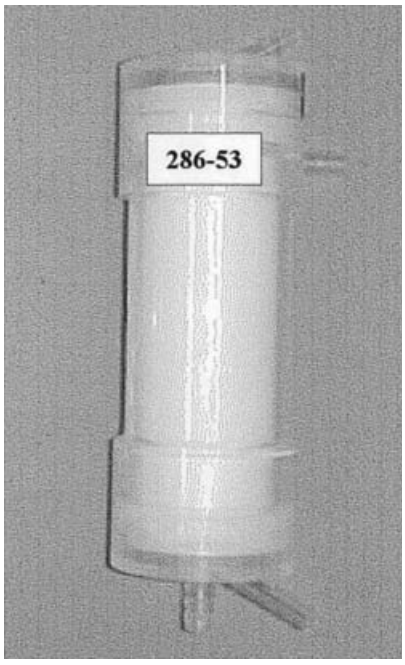

Fig. 1. A photograph shows the new hollow fiber silicone membrane oxygenator.

O_2 transfer rate (ml/min)=(CaO_2 - CvO_2)×blood flow rate

Total CO_2 (mmol/L)=HCO_3^-+0.03×PCO_2

CO_2 transfer rate (ml/min)=22.4×(tCO_2v - tCO_2a)× blood flow rate

where Hb is hemoglobin (g/dl), PO_2 is oxygen partial pressure (mmHg), CaO_2 is arterial oxygen content (Vol%), CvO_2 is venous oxygen content (Vol%), the blood flow rate represents pump flow rate (L/min), HCO_3^- is plasma bicarbonate ion concentration (mmol/L), PCO_2 is CO_2 partial pressure (mmHg), tCO_2v is venous total CO_2 (mmol/L), and tCO_2a is arterial total CO_2 (mmol/L).

Hemolysis test : Plasma free hemoglobin was measured every day. The method of measuring plasma free hemoglobin has been described by Mizuguchi *et al*. (10).

Pressure drop measurement : Pressure differences between the inflow and outflow ports were monitored to assess the pressure drop in the oxygenator throughout the experiment using a pressure monitor (Living Systems Instrumentation, Burlington, VT, U.S.A.).

CBC and biochemistry study : CBC, lactate dehydrogenase (LDH), aspartate aminotransferase (AST), alanine aminotransferase (ALT), blood urea nitrogen (BUN), creatinine, total billirubin (T. Bil), total protein (TP), and electrolytes (Na, K, Cl) were measured every two days postoperatively.

RESULTS

A 2-week *ex vivo* experiment was successfully performed without exchanging the oxygenator. The stable and reliable performance of this new oxygenator was demonstrated for the entire experiment.

Gas Transfer Performance : Fig. 2 shows the results of gas exchange performance tests. The O_2 and CO_2 gas transfer rates at a blood flow rate of 1 L/min and V/Q=3 were maintained at 41.72±4.13 (mean±SD) ml/min and 40.97±14.49 ml/min, respectively, for 2 weeks.

Hemolysis test : As shown in Fig. 3, the plasma free hemoglobin was maintained at 5.50±2.20 mg/dl for 2 weeks.

Pressure drop study : The pressure drop in the blood chamber was kept from 20 to 40 mmHg. A high pressure drop was sometimes observed, however, this was not continuously observed (for only 2 or 3 hours) (Fig. 4). The reason of this high pressure drop was unknown.

CBC and biochemistry study : Table 1 shows the CBC and blood biochemistry data throughout the experiment. All the mean values were within normal limits except for anemia due to bleeding towards the end of the experiment. Just after the operation, abnormal levels of WBC, AST and TP were observed, however, they were normalized gradually. Also, the platelet count was maintained above 200,000/μl.

Visual inspection : After the experiment, the visual inspection of the oxygenator revealed no blood clot or thrombus formation (Fig. 5).

Necropsy findings : There were no thromboembolic findings in major organs including liver, kidney, spleen, intestine, etc. in the macroscopic examination. However, a huge hematoma in the chest cavity due to the high ACT level was observed.

DISCUSSION

The microporous membrane oxygenator that was introduced in the early 1960s has been successfully used for cardiopulmonary bypass during cardiac surgery. However, plasma leakage through the pores into the gas phase is one of the limiting factors for long-term use of these microporous membrane oxygenators (11-13). The plasma leakage is thought to be a result of the loss of the hydrophobic characteristics of the micropores by the adsorption of protein, and that is promoted by an increase in the outlet blood pressure (14). Several techniques have been proposed to decrease the plasma leakage, including the use of a heated humiditied gas (11, 12, 15), use of high gas flow rate (15), and development of a more uniform and smaller size porous membrane (16, 17). These techniques require additional complex and expensive devices. In contrast, the nonporous true membrane oxygenator can completely prevent plasma leakage without any other hardware.

ECMO is now a standard treatment for respiratory failure refractory to conventional pulmonary support techniques worldwide, and many patients are treated with ECMO every year (18). The Kolobow spiral coil membrane oxygenator (2), the only ECMO oxygenator approved in the U.S.A., employs a solid silicone membrane. However, because of the mechanically weak characteristics of silicone, manufacturing of hollow fibers from silicone elastomers is difficult and expensive. This author's group developed a novel silicone material with sufficient mechanical strength, and a fine silicone hol-

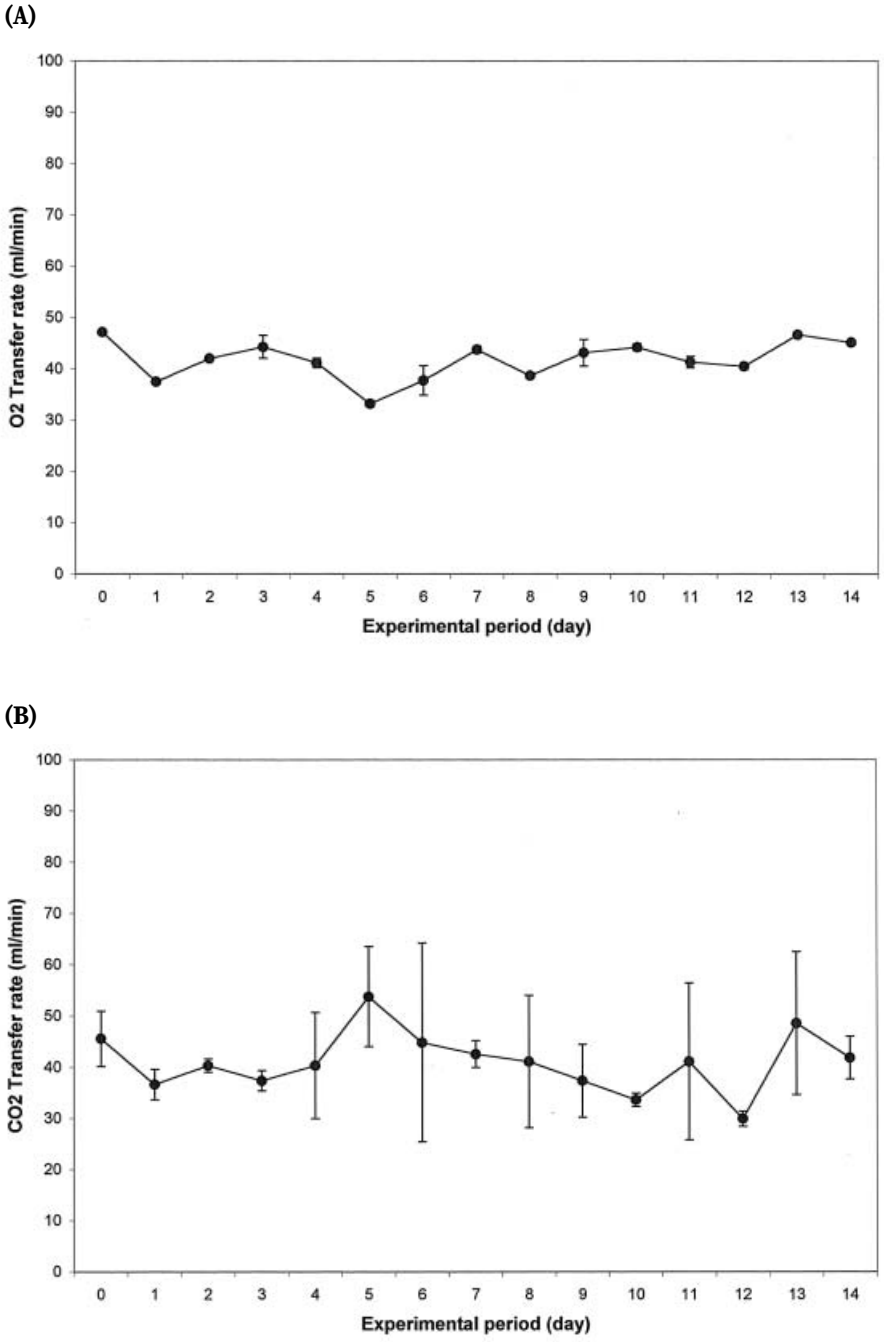

Fig. 2. Gas transfer performance changes of O_2 transfer rate (A) and CO_2 transfer rate (B) for a 2-week ECMO study. Stable and sufficient gas transfer levels were revealed. Data are expressed as mean ±SD.

low fiber with a diameter of 300 μm and wall thickness of 50 μm (3). Utilizing this hollow fiber, an oxygenator with an improved blood flow distributor was fabricated. After satisfactory *in vitro* (8) and short-term *ex vivo* (9) performances were achieved, a 2-week *ex vivo* experiment was successfully performed. Other groups are also developing high-performance ECMO oxygenators, for example, Takewa *et al.* (19) reported long-term ECMO experiments using a heparin bonded oxygenator. On the other hand, our oxygenator was made of silicone rubber with excellent biocompatibility. This is the first all silicone rubber hollow fiber ECMO oxygenator in the world.

One of the limitations of conventional silicone hollow fiber compared with microporous membrane oxygenators is poor gas permeability. We solved the problem to some extent by using a novel fine ultrathin silicone fiber. In this *ex vivo* experiment, the O_2 and CO_2 gas transfer rates were maintained at the same value of 40 ml/min at a blood flow rate of 1 L/min flow and V/Q=3 for 2 weeks. Stable and reliable gas transfer performance under the ECMO condition is advantageous for long-term usage. However, O_2 and CO_2 transfer rates are still limited comparing the results of *in vitro* experiments (8).

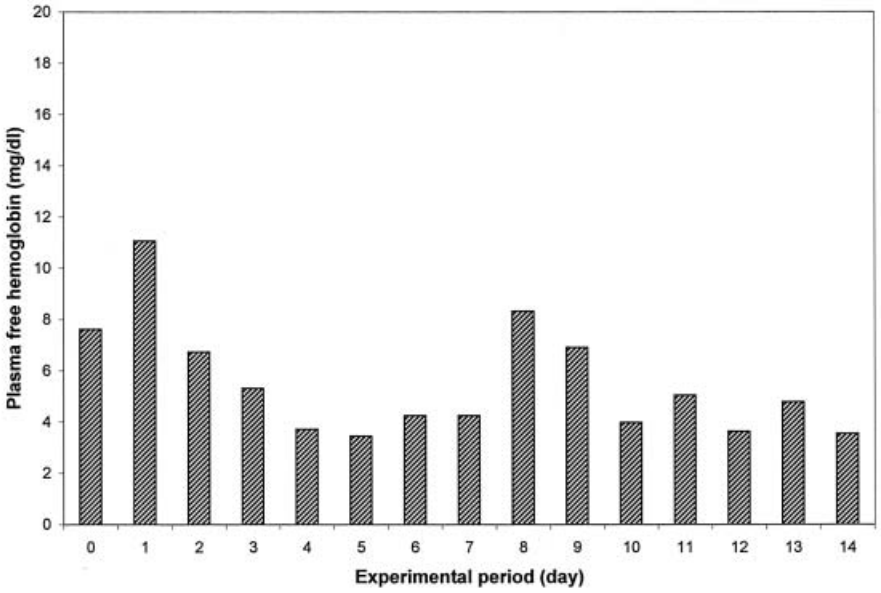

Fig. 3. The change of plasma free hemoglobin during the 2-week ECMO study.

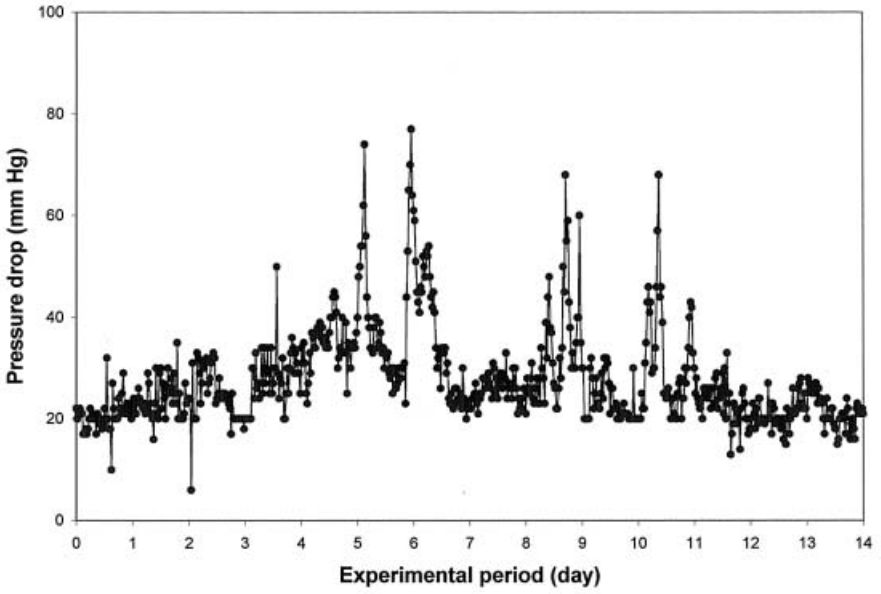

Fig. 4. The pressure drop in the blood chamber during the 2-week ECMO study.

Further improvements (further increase of the fiber length and the surface area, etc.) are necessary. Additionally, the plasma free hemoglobin levels and pressure drops were within acceptable levels, and no abnormalities during the blood examination or necropsy were observed. Excellent biocompatibility of our silicone membrane oxygenator was proven in this *ex vivo* experiment.

The remaining problem is the bleeding complication due to the anti-coagulant therapy. Because the preliminary study cases developed oxygenator occlusion after several days, it was decided that the ACT level be increased from 200-250 sec to around 400 sec. However, in this study, a huge hematoma in the chest cavity due to the high ACT level was observed. During the next ECMO experiment, this group is proposing the ACT level be in the range of 250-300 sec. These results indicate that this oxygenator can be used for prolonged ECMO, although further improvement in thrombo-resistant properties should be achieved.

In conclusion, a 2-week *ex vivo* experiment with the silicone membrane hollow fiber oxygenator was successfully completed. Stable and reliable performance of this oxygenator was proven. These data demonstrate the feasibility for long-term application of this newly improved silicone hollow fiber oxygenator. However, this manuscript is essentially the results of a single experiment, which confirms the feasibility of our newly-developed oxygenator. It is not a sufficient number of experiments to confirm the advantages of our oxygenator. This oxygenator is still

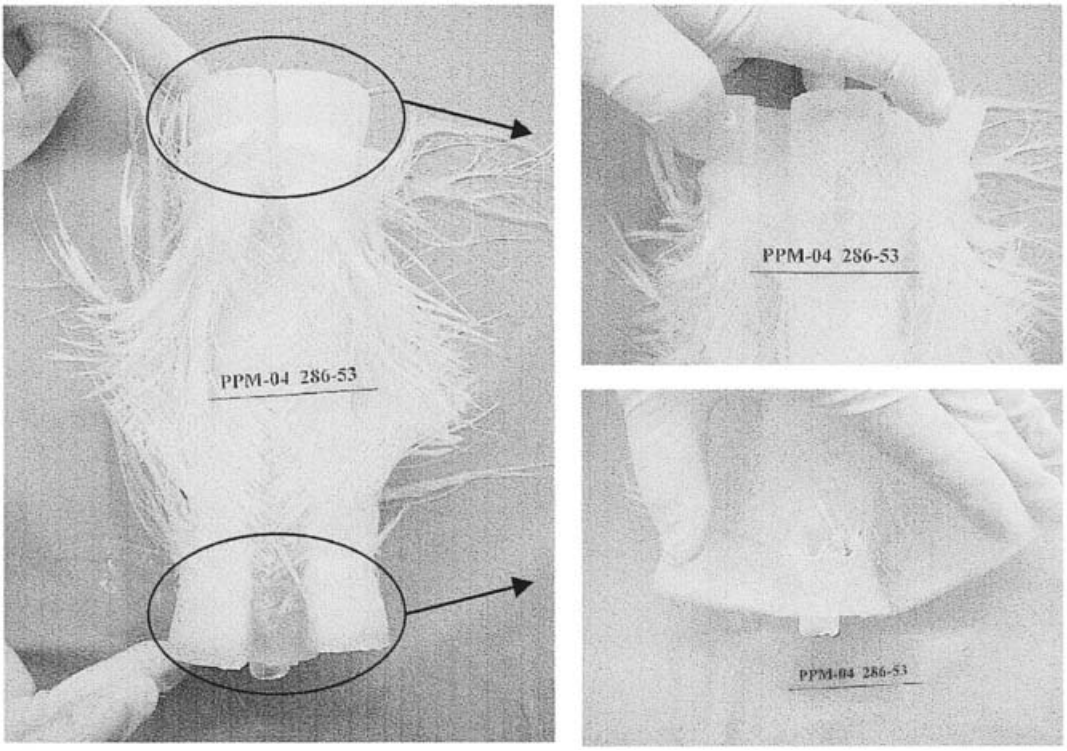

Fig. 5. A photograph taken of the oxygenator after the experiment.

Table 1. Complete blood cell count and biochemistry data

	Mean±SD	Range
WBC (/μl)	11589±6214	4400-22000
RBC (×10⁴/μl)	444±171	210-740
Ht (%)	18.9±6.3	10.0-30.0
Plt (×10⁴/μl)	35.7±6.2	22.3-43.6
LDH (IU/L)	652±202	404-955
AST (IU/L)	64±34	37-126
ALT (IU/L)	16±6	10-27
BUN (mg/dl)	12±4	9-21
Creatinine (mg/dl)	0.7±0.1	0.6-0.9
T. Bil (mg/dl)	0.4±0.3	0.1-1.1
TP(g/dl)	6.0±0.7	4.8-6.7
Na (mEq/L)	136±3	133-140
K (mEq/L)	4.6±0.5	3.4-5.2
Cl (mEq/L)	96±7	85-106

All data are shown as mean±SD.

WBC, white blood cell ; RBC, red blood cell ; Ht, hematocrit ; Plt, platelet ; LDH, lactate dehydrogenase ; AST, aspartate aminotransferase ; ALT, alanine aminotransferase ; BUN, blood urea nitrogen ; T. Bil, total bilirubin ; TP, total protain ; Na, sodium ; K, potassium ; Cl, chroride

in the development stage, and further experiments and improvements will be necessary.

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