Table 1 Preoperative clinical condition and laboratory data

	Group A ($n = 10$)	Group B ($n = 14$)	p value
Age at appearance (days)	2 (0–56)	10.5 (0–46)	0.10
Complaints at appearance			
Non-bile vomiting	6 (60 %)	6 (43 %)	0.68
Bile vomiting	1 (10 %)	4 (29 %)	0.36
Abdominal distention	8 (80 %)	6 (43 %)	0.10
Poor oral intake	1 (10 %)	6 (43 %)	0.17
Complaints on the admission			
Vomiting (not bile like)	1 (10 %)	3 (21 %)	0.61
Vomiting (bile like)	5 (50 %)	6 (43 %)	1.0
Abdominal distention	10 (100 %)	14 (100 %)	1.0
Blood test on the first visit			
White blood cell (/μl)	11,700 (6,000–22,600)	12,600 (5,200–18,100)	0.60
Eosinophil (/μl)	150 (0–1,064) ($n = 8$)	252 (86–1,724) ($n = 10$)	0.29
Eosinophil (%)	1 (0–7) ($n = 8$)	3 (1–12) ($n = 10$)	0.19
C-reactive protein (mg/dl)	0.2 (0.05–14.6) ($n = 9$)	0.1 (0.03–1.06) ($n = 9$)	0.11
SI for cow's milk by LST	152 (100–236) ($n = 3$)	583 (397–2,069) ($n = 10$)	0.01
% Body weight before surgery (%)	101 (79–112) ($n = 8$)	101.6 (60.9–117.7) ($n = 14$)	0.73
Enterocolitis	0 (0 %)	4 (29 %)	0.11
Number of times of enterocolitis, median (range)	0	0 (0–1)	0.07
Preoperative period	63 (23–171)	50 (6–773)	0.73
Pediatric ED feeding (days)	56 (19–115)	36 (6–148)	0.47
Intestinal dilatation on X-ray	3/10 (30 %)	9/14 (64 %)	0.21
Age of rectal biopsy (days)	6 (3–59)	18 (5–719)	0.07
Age of operation (days)	66 (25–171)	77 (19–793)	0.52

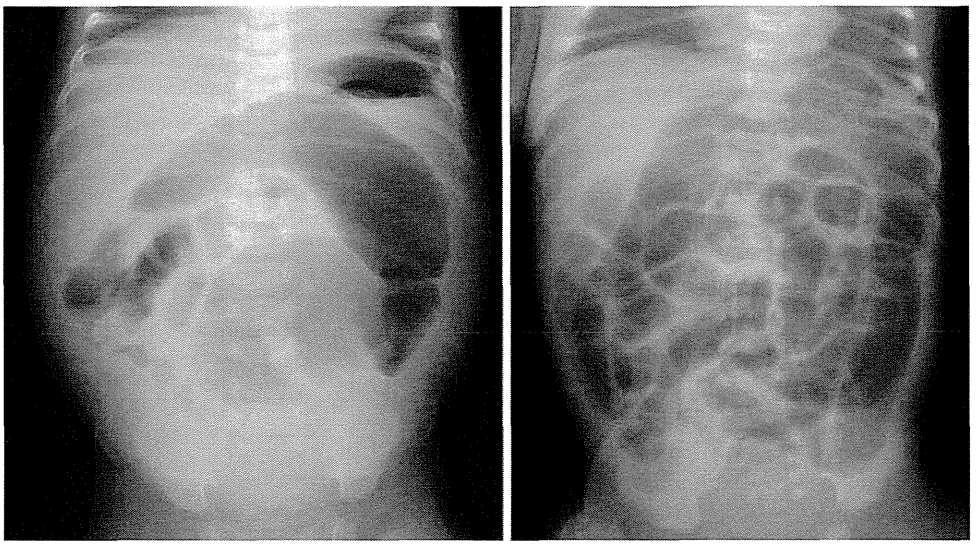

Fig. 1 Plain abdominal X-ray of a Group A patient, showing a dilated intestinal gas shadow mainly in the colon (*left*), and that of a Group B patient, showing a dilated intestinal gas shadow in both the small intestine and colon (*right*)

Postoperative clinical conditions and laboratory data (Table 2)

Enterocolitis did not occur in Group A patients, but occurred in 5 (36 %) Group B patients ($p = 0.05$). Enterocolitis was more frequent in Group B than in Group A ($p = 0.04$). The period of elemental formula feeding was significantly longer in Group B than in Group A ($p = 0.02$). There was no significant difference in medication at 6 months after operation between Groups A and B.

Table 2 Postoperative clinical condition and laboratory data

	Group A	Group B	*p* value
Enterocolitis	0 (0 %)	5 (36 %)	0.05
Number of times of enterocolitis, median (range)	0	0 (0–1)	0.04
Period of elemental formula feeding (days)	$n = 9$ 118 (8–1,287)	$n = 14$ 156 (10–1,868)	0.02
Medication at 6 months after operation	$n = 9$	$n = 14$	
No medication	3 (33 %)	2 (14 %)	0.34
Glycerin enema	4 (44 %)	7 (50 %)	1.0
Laxative	5 (56 %)	9 (64 %)	1.0

Discussion

CMA patients often present with various gastrointestinal symptoms, and it is sometimes difficult to discriminate CMA from HD [5]. The incidence of CMA is reported to be 0.21 to 3 % [6–9]. It was reported that the incidence of CMA in infants with birth weight <1,000 g was significantly higher than that in infants with birth weight of 1,500–2,500 g [8]. The diagnosis of CMA was correctly made if gastrointestinal symptoms were relieved by cow's milk elimination and appeared on challenge test. Because non-IgE-mediated delayed type allergic reactions are thought to play a predominant role in the majority of CMA, cow's milk-specific IgE antibody level has a limited role as an indicator of CMA. Therefore, LST has been proposed as an alternative diagnostic test for CMA [10, 11]. Ikeda et al. [4] proposed that LST is interpreted as positive when SI is greater than 300 %, based on the receiver operating characteristic curve obtained from the data of LST values of CM in 94 infants with and without CMA (sensitivity 95 %, specificity 69 %). Most patients classified as having possible CMA in this study showed SI of LST much higher than 300 %.

Comparing clinical and laboratory data between non-CMA and possible CMA patients, the frequency of postoperative enterocolitis and the period of elemental formula feeding were significantly greater in possible CMA patients. Allergic characteristics, e.g. a positive result of LST, presence of eosinophilic infiltration in the colon resected at surgery or rectal suction biopsy specimen, are considered to be risk factors for enterocolitis in HD patients. Our finding that non-CMA patients did not have a history of enterocolitis before or after the Soave procedure needs to be validated in a larger number of HD patients without CMA.

The incidence of preoperative enterocolitis in HD patients was reported to be 15 to 50 %, and postoperative enterocolitis occurs in 2 to 33 % of patients [12]. Many risk factors, including increased length of the aganglionic segment, female sex, trisomy 21, and the presence of other associated congenital anomalies, have been identified and help support the current model for the pathophysiology of enterocolitis associated with HD [13–16], whereas, to our knowledge, CMA has not been proposed as a risk factor for enterocolitis in HD patients.

It was not clarified in this study whether the HD patients had allergy for food antigens other than cow's milk, because LST for other foods was not conducted in the HD patients. Some patients were examined with radioallergosorbent test (RAST) for IgE-mediated food allergy. One patient of Group A showed a positive result of RAST for egg white at 99 days of age. Six patients of Group B showed positive results of RAST for egg white, and two of the six showed positive results of RAST for egg yolk at the median age of 511 days (234–1,385 days). The correct incidence of co-existing other food allergy remains unknown, because systematic follow-up study was not conducted in the patients.

In conclusion, examination of the association of CMA is worthwhile, and it is a possible risk factor for enterocolitis in HD patients. Large, prospective cohort studies are needed to clarify the clinical impact of CMA on HD patients.

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Ghrelin and glucagon-like peptide-2 increase immediately following massive small bowel resection

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ABSTRACT

Children with short bowel syndrome face life-threatening complications. Therefore, there is an urgent need for a new therapy to induce effective adaptation of the remnant intestine. Adaptation occurs only during feeding. We focused on preprandial acyl ghrelin and des-acyl ghrelin, and postprandial glucagon-like peptide-2 (GLP-2), which are known to have active orexigenic and trophic actions. This study aims to clarify the secretion trends of these hormones after massive small bowel resection and to obtain basic data for developing a new treatment. Sixty-three growing male rats were used: 3 were designated as controls receiving no operation and 60 were randomized into the 80% small bowel resection (80% SBR) group and the transection and re-anastomosis group. Changes in body weight, food intake, and remnant intestine morphology were also assessed for 15 days after the operation. Acyl ghrelin and des-acyl ghrelin levels increased immediately, equivalently in both operation groups ($P = 0.09$ and 0.70). Interestingly, in 80% SBR animals, des-acyl ghrelin peaked on day 1 and acyl ghrelin peaked on day 4 ($P = 0.0007$ and $P = 0.049$ vs controls). GLP-2 secretion was obvious in 80% SBR animals ($P = 2.25 \times 10^{-6}$), which increased immediately and peaked on day 4 ($P = 0.009$ vs. controls). Body weight and food intake in 80% SBR animals recovered to preoperative levels on day 4. Morphological adaptations were evident after day 4. Our results may suggest a management strategy to reinforce these physiological hormone secretion patterns in developing a new therapy for short bowel syndrome.

1. Introduction

The loss of a large part of the small bowel in infants, owing to surgical removal or a congenital defect, leads to a condition called short bowel syndrome. The 3 most common causes of short bowel syndrome in children are necrotizing enterocolitis, intestinal atresia, and midgut volvulus [22]. When a large part of the small intestine is lost, the functional ability of the remaining intestine is often inadequate to support growth and hydration, and prolonged parenteral nutritional support is required. Children with a short bowel are at risk for many life-threatening complications such as sepsis due to catheter-related blood stream infection and parenteral nutrition-associated liver disease even when these children are under total parenteral nutrition. In the clinic, decisions about the optimal management of pediatric short bowel syndrome are often based on repeated trial-and-error treatments, depending on the condition of a specific patient. Therefore, there is an urgent need for a new therapy to compensate for the lost functionality of the small intestine.

Fundamentally, when a large section of the small intestine is lost, the reduction in nutritional absorption is compensated gradually by an increase in the mucosal surface area of the remaining bowel, accompanied by increases in the villus height and crypt cell proliferation rates. This process is known as adaptation [32]. The regulation and augmentation of the function of the remaining intestine is induced through a complex interaction of many different factors, including luminal nutrients and gastrointestinal hormones [22,28]. Physiologically, bowel adaptation is supposed to occur only in response to oral feeding [32]. In this study, we investigated the levels of 3 gastrointestinal hormones, acyl ghrelin, des-acyl ghrelin, and glucagon-like peptide-2 (GLP-2).

Ghrelin is secreted by the X/A-like cells of the stomach and the proximal small intestine. Two major molecular forms of ghrelin exist, of which acyl ghrelin with n-octanoylated modification appears to serve multiple functions [7,15,27], including exerting positive effects on food intake, growth hormone

Abbreviations: GLP-2, glucagon-like peptide-2; 80% SBR, 80% small bowel resection.

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secretory action, glucose and lipid metabolism, gastrointestinal motility, cell proliferation, and hemodynamics, all of which may contribute to intestinal adaptation after massive small bowel resection. On the other hand, non-acylated des-acyl ghrelin induces a negative energy balance by decreasing food intake and delaying gastric emptying [2]. Furthermore, des-acyl ghrelin suppresses acyl ghrelin-induced food intake [10]; a continuous infusion of des-acyl ghrelin is reported to reduce weight gain [21].

GLP-2 is secreted by the intestinal L-cells of the distal ileum and proximal colon in response to both direct stimulation of luminal nutrients and vagally mediated pathways, which are activated by the presence of nutrients in the proximal bowel [12]. GLP-2 is best known for its beneficial role in intestinal adaptation and has become a focus of studies on short bowel syndrome [32]. A randomized placebo-controlled study of teduglutide, a GLP-2 analog, showed a potential reduction in the dependency on parenteral support of adult patients with short bowel syndrome [11]. However, this treatment has not been applied clinically in children.

The purpose of this study was to clarify the trends in the secretion of endogenous acyl ghrelin, des-acyl ghrelin, and GLP-2 following massive small bowel resection in order to obtain basic data for the future investigation of a new treatment that may induce efficient intestinal adaptation in patients with short bowel syndrome.

2. Materials and methods

2.1. Animals

Sixty-three 7-week-old male Sprague-Dawley rats weighing 200–240 g (purchased from Kyudo Co., Ltd., Saga, Japan) were used in this experiment. The animals were individually housed in cages with free access to standard rat chow and water, and maintained under standardized temperature (23 °C ± 1 °C), humidity (50% ± 10%), and 12-h light-dark cycles (lights on at 7:00 a.m.).

All experimental procedures were approved by the Laboratory Animal Committees of Kagoshima University Graduate School and were performed in accordance with the "Guidelines for the Care and Use of Laboratory Animals."

2.2. Study design

Sixty animals were randomized to either the 80% small bowel resection (SBR) group or the transection and re-anastomosis operation (sham) group and allowed to acclimatize to their environment for 6 days before experimentation. Changes in body weight, food intake, water intake, amount of stool, and amount of urine were measured from 7:00 to 8:00 a.m. throughout the experimental period. Preprandial plasma acyl ghrelin and des-acyl ghrelin levels, postprandial plasma GLP-2 levels, and intestinal morphology were assessed at days 1, 4, 7, 11, and 15 after the operation (6 animals per day for the 2 operation groups). As a control, the same measurements described above were assessed in 3 animals at day 0. Plasma acyl and des-acyl ghrelin levels are known to fluctuate according to psychological or physical stresses [16,34]; therefore, the environments of the experimental animals were noted to be uniform. The 80% SBR animal model has been well established [18,37]. Adaptive response in an 80% SBR rat model was reported to be most pronounced in the first week [18], and the morphological changes reached a plateau (equivalent to 30-postoperative-day levels) within 12 days in a 70% SBR rat model [9]. Thus, we set 15 days as the experimental period for this study. The promotion of functional alterations in response to morphological adaptations following massive small bowel resection has been previously reported [19,26,37].

2.3. Surgical methods

The animals were fasted overnight, anesthetized with isoflurane (1.5% inhalation by mask), and explored through a midline laparotomy under sterile conditions. Intestinal length was measured in a standardized fashion, and 80% SBR was performed, leaving 15 cm of the ileum above the ileocecal valve anastomosed to the jejunum 5 cm below the ligament of Treitz [13]. Bowel anastomoses were completed with the aid of an operating microscope, using interrupted 6-0 silk sutures (Alfresa Pharma Corporation, Tokyo, Japan), and the abdominal incision was closed with 3-0 polyglycolic sutures (Johnson & Johnson K.K., Tokyo, Japan). Sham-operated rats were transected at 15 cm above the ileocecal valve and re-anastomosed [13].

All animals received cefazolin (50 mg/kg per dose subcutaneously; Otsuka Pharmaceutical Factory, Inc., Tokushima, Japan) to prevent postoperative infection and buprenorphine (0.01 mg/kg per dose subcutaneously; Otsuka Pharmaceutical Co., Ltd., Tokyo, Japan) for analgesia [13]. Additionally, a subcutaneous injection of 10 mL isotonic saline was given to prevent postoperative dehydration. The animals were allowed free access to water immediately after surgery and standard rat chow ad libitum at the dark cycle of the first postoperative day.

2.4. Measurement of plasma acyl ghrelin, des-acyl ghrelin, and GLP-2 levels

After an overnight fast, rats were anesthetized by isoflurane inhalation. Blood was obtained from the tail between 10:00 and 12:00 a.m. then immediately centrifuged at 1500 × g for 15 min at 4 °C. All plasma samples were stored at −80 °C until assayed.

2.4.1. Preprandial acyl ghrelin and des-acyl ghrelin

For the acyl ghrelin and des-acyl ghrelin assay, blood samples were drawn into chilled polypropylene tubes containing 0.2 M ethylenediaminetetraacetic acid, disodium salt (EDTA·2Na) (20 μL/1 mL blood sample) and aprotinin (0.3–0.8 trypsin inhibitor unit/1 mL of blood sample) and then centrifuged. Aliquots of plasma were acidified with 1 N hydrogen chloride and then stored. Plasma acyl ghrelin and des-acyl ghrelin levels were measured using an enzyme-linked immunosorbent assay kit (Mitsubishi Chemical Medicine Corporation, Tokyo, Japan).

2.4.2. Postprandial GLP-2

One hour after gavage of the animals with 2 mL of a liquid meal (ENSURE H; Abbott Japan Co., Ltd., Tokyo, Japan), blood samples were drawn into chilled polypropylene tubes containing 0.2 M EDTA·2Na (20 μL/1 mL blood sample) and centrifuged. Aliquots of plasma were stored. Plasma GLP-2 levels were measured by an enzyme immunoassay kit (Yanaihara Institute Inc., Shizuoka, Japan).

2.5. Gross intestinal morphology and histology

After the collection of blood for the GLP-2 assay, the animals were euthanized by exsanguination. The total small intestine was harvested for gross and microscopic morphological analysis.

The mesentery was removed, and the total length of the small bowel was measured from the ligament of Treitz to the ileocecal valve along the antimesenteric border. The harvested small intestine was quickly opened along the mesenteric border, rinsed in cold saline, and then weighed. Bowel width was measured at the middle point of the opened jejunum and ileum. Samples for microscopic analysis were harvested from the jejunum (2.5 cm below the ligament of Treitz), the proximal ileum (12.5 cm above the ileocecal valve, i.e., 2.5 cm below the anastomotic line), and

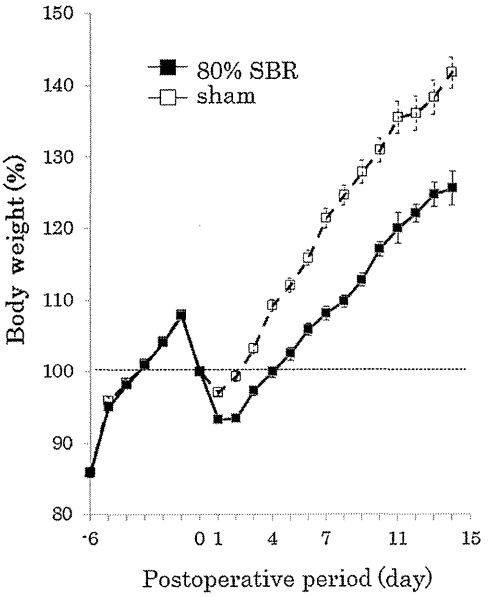

Fig. 1. Changes in body weight. Body weight was set at 100% at the time of the operation. Data are reported as the means ± standard error. A significant difference ($P < 0.01$) in body weight was found between the 80% SBR group and the sham operation group throughout the postoperative period, as analyzed by Student's *t*-test. 80% SBR, 80% small bowel resection.

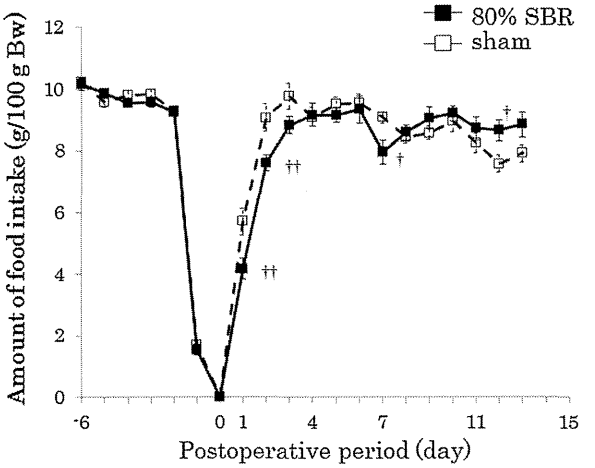

Fig. 2. Changes in the amount of food intake. Data are shown as the mean value of intake per 100 g of body weight ± standard error. The statistical differences between the groups were analyzed by Student's *t*-test. ††$P < 0.01$ and †$P < 0.05$ versus the sham operation group at similar time points. 80% SBR, 80% small bowel resection.

the distal ileum (2.5 cm above the ileocecal valve) and fixed in a 10% formaldehyde neutral buffer solution for 24 h. Mucosal scrapings from the residual jejunum and ileum were weighed. Paraffin sections of formalin-fixed tissue were cut at 3-μm thickness and stained with hematoxylin and eosin. For each sample slide, microscopic measurements of the villus height, villus width, crypt depth, and number of villi per 1 mm were recorded from 5 well-oriented villi/crypt units. The quantification was performed with the help of an expert pathologist. The absorptive mucosal surface area per 1 cm^2 of the intestine was calculated using methods discussed previously. In brief, the mucosal surface area was calculated by first considering the intestine as a cylinder and then multiplying the added mucosal surface area contributed by the villi, considering each villus as a cone [19,23].

2.6. Statistical analysis

Data are presented as the mean values ± standard error (SE). Statistical analyse between groups and time courses were performed by 2-factor factorial analysis of variance (ANOVA) followed by Tukey's multiple-comparison posttest. Comparisons with controls were performed by Dunnett's test. Comparisons between the experimental groups at similar time points were performed by Student's *t*-test. Statistical analysis was completed using Ekuseru-Toukei 2010 (Social Survey Research Information Co., Ltd., Tokyo). All results were considered statistically significant when *P* values were <0.05.

3. Results

3.1. Changes in daily assessment data

The body weight of animals in the 80% SBR group returned to preoperative levels within 4 days, and continued to increase steadily (Fig. 1). Food intake in the 80% SBR animals recovered to preoperative levels on postoperative day 4. After day 4, roughly equivalent intake was maintained between the 2 operative groups, the 80% SBR animals and the sham-operated animals (Fig. 2). The sham-operated animals showed higher water intake, amount of stool, and amount of urine than the 80% SBR animals until postoperative day 4. Subsequently, these measures remained almost the same in both groups (data not shown). Loose stools were observed in the 80% SBR animals several days after the operation; however, watery or muddy stools were not observed.

3.2. Changes in intestinal morphology

Gross morphological changes in bowel weight and mucosal weight were evident in the 80% SBR animals after postoperative day 4 compared with controls. Moreover, these changes were significantly different in comparisons with the sham-operated animals at similar time points (Table 1). The increase in villus height of the 80% SBR animals was more evident than that of the sham-operated animals after postoperative day 4 (Fig. 3). In the microscopic quantification, villus height and crypt depth were significantly increased after day 4 in the 80% SBR animals (Table 2). There were no significant differences in the changes of villus width or the number of villi per 1 mm (data not shown). The increase in absorptive mucosal surface per unit area was based on the growth of the villus height. The expansion of absorptive mucosal surface was observed starting on day 4 (Table 2).

3.3. Changes in gastrointestinal hormone levels

The levels of all 3 gastrointestinal hormones immediately increased following massive small bowel resection compared with the controls.

In 80% SBR animals, preprandial plasma acyl ghrelin peaked on day 4, with a significant difference versus controls (means ± standard error, 104.7 ± 14.1 fmol/mL, $P = 0.049$). The time when acyl ghrelin reached its peak level accorded with the time when the body weight and food intake recovered to the preoperative levels. It also matched the timing when the morphology of the remaining intestine began to change significantly. Interestingly, the peak of preprandial plasma des-acyl ghrelin was observed on day 1 (1021.6 ± 93.1 fmol/mL, $P = 0.0007$ vs. control). Concerning preprandial acyl ghrelin and des-acyl ghrelin, equivalent plasma levels were maintained in 80% SBR animals, i.e., under short bowel conditions, and in sham-operated animals, i.e., under native small bowel length conditions ($P = 0.09$ and $P = 0.70$).

The postprandial plasma GLP-2 concentration in 80% SBR animals peaked on day 4, with a significant difference versus controls

Table 1
Changes in gross intestinal morphology.

			Jejunum				Ileum			
		n	Bowel length (cm)	Bowel width (cm)	Bowel weight (mg/cm)	Mucosal weight (mg/cm)	Bowel length (cm)	Bowel width (cm)	Bowel weight (mg/cm)	Mucosal weight (mg/cm)
Control		3	5.0 ± 0.0	1.0 ± 0.0	101.3 ± 6.4	38.5 ± 4.3	15.0 ± 0.0	1.0 ± 0.0	105.6 ± 7.2	44.5 ± 4.1
80%SBR	Day 1	6	5.1 ± 0.1	1.1 ± 0.1	95.8 ± 2.3	41.5 ± 4.0	15.2 ± 0.4	1.2 ± 0.1	104.9 ± 6.3	43.7 ± 2.7 †
	Day 4	6	5.2 ± 0.2	1.2 ± 0.1	142.9 ± 10.9 ††	60.1 ± 6.0 ††	15.8 ± 0.5	1.4 ± 0.0 **,††	131.5 ± 2.7 ††	57.5 ± 2.7 ††
	Day 7	6	5.2 ± 0.1	1.4 ± 0.1 **,††	166.0 ± 13.8 *,††	58.4 ± 4.2 ††	16.6 ± 0.8	1.5 ± 0.1 **,††	177.5 ± 9.4 **,††	73.7 ± 4.0 **,††
	Day 11	6	5.1 ± 0.2	1.6 ± 0.0 **,††	261.5 ± 25.4 **,††	98.9 ± 12.1 **,††	16.4 ± 0.2	1.8 ± 0.0 **,††	208.0 ± 10.8 **,††	78.9 ± 4.8 **,††
	Day 15	6	5.3 ± 0.3	1.7 ± 0.1 **,††	267.3 ± 21.2 **,††	93.2 ± 11.3 **,††	15.7 ± 0.3	1.7 ± 0.0 **,††	223.9 ± 8.9 **,††	89.2 ± 4.8 **,††
Sham	Day 1	6	5.0 ± 0.0	1.0 ± 0.0	86.0 ± 10.1	30.4 ± 5.2	14.9 ± 0.1	1.1 ± 0.0	90.3 ± 3.1	34.2 ± 2.5
	Day 4	6	5.0 ± 0.0	1.1 ± 0.0	87.0 ± 3.4	32.5 ± 2.0	15.3 ± 0.5	1.1 ± 0.0	90.5 ± 3.6	36.4 ± 3.2
	Day 7	6	5.0 ± 0.0	1.0 ± 0.0	93.4 ± 3.6	34.9 ± 1.8	16.3 ± 0.5	1.1 ± 0.0	110.2 ± 7.5	40.5 ± 4.3
	Day 11	6	5.0 ± 0.0	1.0 ± 0.0	100.3 ± 3.7	37.4 ± 1.8	15.7 ± 0.2	1.1 ± 0.0	133.7 ± 7.8	46.3 ± 3.4
	Day 15	6	5.0 ± 0.0	1.1 ± 0.1	106.3 ± 5.0	40.8 ± 3.2	15.7 ± 0.4	1.2 ± 0.1	133.4 ± 8.2	43.2 ± 2.6

Data are expressed as the means ± SE. Comparisons with controls were performed by Dunnett's test. Comparisons among the groups at similar time points were analyzed by Student's *t*-test.
* $P<0.05$ versus controls.
** $P<0.01$ versus controls.
† $P<0.05$ versus the sham operation subjects.
†† $P<0.01$ versus the sham operation subjects.
80% SBR, 80% small bowel resection.

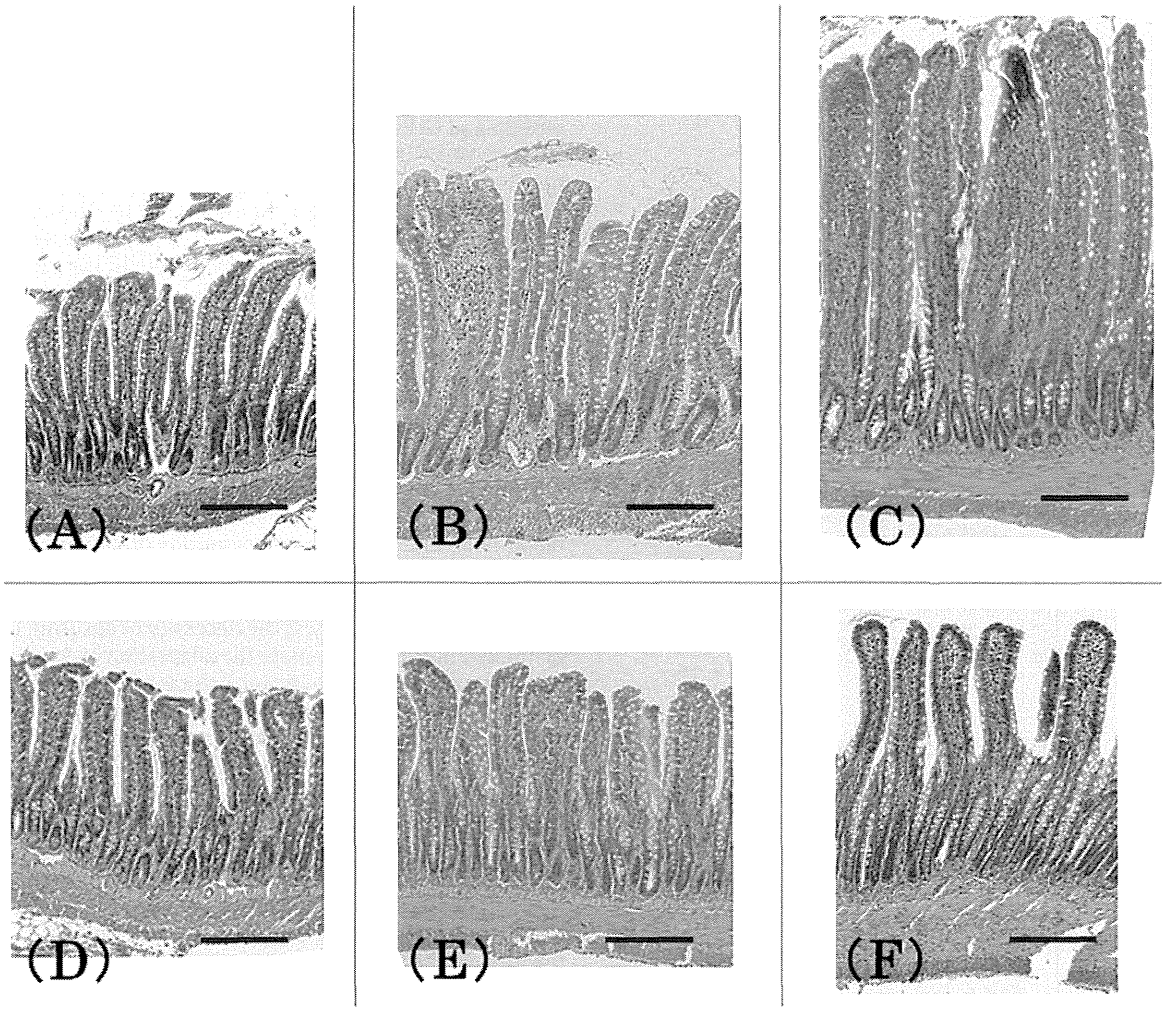

Fig. 3. Changes in intestinal morphology in the proximal ileum. Morphological changes were evident after day 4 in the 80% SBR animals but not in sham-operated animals. This tendency was seen in both the remaining jejunum and ileum. The changes in microscopic morphology in the proximal ileum (12.5 cm above the ileocecal valve) are shown. (A) Post 80% SBR on day 1; (B) post 80% SBR on day 4; (C) post 80% SBR on day 15; (D) post sham operation on day 1; (E) post sham operation on day 4; (F) post sham operation on day 15. Bar, 200 μm. 80% SBR, 80% small bowel resection.

Table 2
Changes in microscopic intestinal morphology.

		n	Jejunum			Proximal ileum			Distal ileum		
			Villus height (μm)	Crypt depth (μm)	Absorptive mucosal surface per unit area ($cm^2/1 cm^2$)	Villus height (μm)	Crypt depth (μm)	Absorptive mucosal surface per unit area ($cm^2/1 cm^2$)	Villus height (μm)	Crypt depth (μm)	Absorptive mucosal surface per unit area ($cm^2/1 cm^2$)
Control		3	468.7 ± 35.0	167.3 ± 17.4	12.9 ± 1.9	338.2 ± 29.9	150.5 ± 12.0	9.2 ± 1.5	365.4 ± 29.9	158.2 ± 9.2	9.8 ± 1.6
80%SBR	Day 1	6	452.5 ± 18.7 †	180.2 ± 9.6 *	12.5 ± 0.8	382.8 ± 25.3 †	163.9 ± 6.3	10.8 ± 0.9	367.4 ± 18.2 ††	168.5 ± 9.5	9.9 ± 1.0
	Day 4	6	591.7 ± 39.6 *,†	220.2 ± 6.1 *,††	17.7 ± 1.2	463.4 ± 24.1 **,††	174.3 ± 6.9	14.0 ± 1.5 *	452.3 ± 24.7 **,††	176.5 ± 8.1	13.4 ± 0.8 *,†
	Day 7	6	553.6 ± 13.3 ††	220.8 ± 15.8 *,††	16.1 ± 0.9	513.6 ± 22.8 **,††	207.4 ± 13.2 **	14.6 ± 0.8 *,†	476.5 ± 28.1 **,††	191.5 ± 9.9 †	14.6 ± 1.1 **,††
	Day 11	6	639.7 ± 41.3 **,††	247.4 ± 18.4 **,††	21.9 ± 1.9 **,††	538.0 ± 25.1 **,††	212.3 ± 9.1 **	16.4 ± 1.0 **,†	519.4 ± 23.9 **,††	210.1 ± 17.6 *	15.9 ± 0.9 **,††
	Day 15	6	708.9 ± 24.1 **,††	250.7 ± 14.0 **,††	23.8 ± 1.1 **,††	615.7 ± 31.1 **,††	230.3 ± 8.4 **,†	19.0 ± 1.1 **,††	580.3 ± 17.3 **,††	246.0 ± 18.8 **	20.2 ± 0.7 **,††
Sham	Day 1	6	390.6 ± 10.1	152.5 ± 7.0	11.8 ± 0.6	305.6 ± 21.3	148.5 ± 7.4	8.0 ± 1.0	291.4 ± 9.5	146.3 ± 5.0	7.9 ± 0.3
	Day 4	6	444.0 ± 31.6	160.0 ± 8.2	14.0 ± 1.9	337.5 ± 8.2	168.8 ± 9.7	11.1 ± 1.4	328.6 ± 11.2	152.9 ± 12.1	9.7 ± 1.1
	Day 7	6	445.9 ± 14.9	160.8 ± 9.9	13.1 ± 1.0	355.2 ± 16.9	173.7 ± 8.3	10.4 ± 1.1	329.6 ± 14.0	152.1 ± 8.7	9.5 ± 0.7
	Day 11	6	479.2 ± 13.7	176.8 ± 8.6	14.1 ± 0.5	423.1 ± 14.6	201.7 ± 11.3 **	12.6 ± 0.8	386.8 ± 8.3	184.1 ± 7.0	11.4 ± 0.3
	Day 15	6	503.8 ± 16.0	198.4 ± 5.5	15.8 ± 0.9	425.2 ± 14.4	200.4 ± 5.4 **	13.6 ± 1.0	413.4 ± 12.3	208.2 ± 8.6 *	13.9 ± 0.8

Data are expressed as the means ± SE. Comparisons with controls were performed by Dunnett's test. Comparisons among the groups at similar time points were analyzed by Student's *t*-test.
* $P < 0.05$ versus controls.
** $P < 0.01$ versus controls.
† $P < 0.05$ versus the sham operation subjects.
†† $P < 0.01$ versus the sham operation subjects.
80% SBR, 80% small bowel resection.

(4.3 ± 0.8 ng/mL, $P = 0.009$). GLP-2 levels were maintained at significantly higher levels under short bowel conditions than in native small bowel length conditions ($P = 2.25 \times 10^{-6}$, Table 3).

4. Discussion

Ideally, the progression of intestinal adaptation in infants with short bowel syndrome would occur gradually over 1 to 2 years [32]. In the first 1 to 2 weeks after resection, ileus occurs in the remaining bowel. The next 1 to 6 months are characterized by hypersecretion. Fluid and electrolytes are lost owing to a large amount of watery stool excretion. Subsequently, morphological and functional adaptations occur, such as an increase in the absorptive mucosal surface area. Adaptation is known to result from an increase in villus height and in the rate of crypt cell proliferation [32]. After 1 to 2 years, the compensated absorptive mucosa could perform adequate nutrition and fluid absorption. However, we have encountered some cases in which ideal adaptation cannot be obtained. The clinical challenge is to induce intestinal adaptation early and effectively among children with short bowel syndrome. Postresection intestinal adaptation is supposed to occur only in response to feeding [32]. It was reported that adaptation was impaired in the absence of luminal nutrients in case of total parenteral nutrition [17]. Moreover, only 25% reduction in oral intake caused significantly lower enterocyte production in the crypts [6]. Here, we considered a management strategy to achieve both an orexigenic effect, which promotes food intake, and a trophic effect, which increases the absorptive mucosal surface. Among several gastrointestinal hormones, we focused on ghrelin and GLP-2, which are known to actively bring about these 2 effects [2,12,18,24]. They may provide a clue about how to solve the clinical problem of short bowel syndrome.

In short bowel environments, acyl ghrelin was maintained at an equivalent level compared with the native bowel length conditions. Acyl ghrelin levels increased immediately in the early postoperative period (Table 3). The timie when acyl ghrelin reached its peak on day 4 accorded with the time when the body weight and food intake recovered to the preoperative levels (Figs. 1 and 2). It also matched the start of the morphological adaptation of the remaining intestine. Among the multiple functions of acyl ghrelin [15], here we want to focus on the regulation of food intake. Numerous peptides secreted from the gastrointestinal tract decrease food intake; only one, acyl ghrelin has a promoting effect [35]. Luminal nutrients ingested orally can stimulate intestinal peristalsis, mucosal blood flow, and endogenous secretions of various hormones and growth factors [15,26,28]. The significant increase in endogenous acyl ghrelin levels in our 80% SBR animals within the first 4 postoperative days may suggest the necessity of ensuring the presence of luminal nutrients to initiate the adaptation of the residual intestine. Interestingly, a significant increase in endogenous des-acyl ghrelin level was seen on postoperative day 1, before a peak in the acyl ghrelin levels was reached on day 4. It was reported that the metabolic machinery that induces intestinal adaptation had already been turned on within the first 24 h in the 80% SBR model rats [37]; however, intestinal permeability, which may be detrimental to the organism, was also increasing at the same time [37]. Des-acyl ghrelin induces an anorexigenic effect by decreasing food intake and delaying gastric emptying [2,10]. Therfore, des-acyl ghrelin secretion may suppress the intake of luminal nutrients until the intestinal condition becomes well regulated. It is thus reasonable to say that the control of intake is an indispensable factor for intestinal compensation.

As was found in previous reports [18,19], significantly higher levels of GLP-2 were secreted under short bowel conditions compared with native bowel length conditions. GLP-2 levels increased immediately after the operation (Table 3). The peak GLP-2 level

Table 3
The secretion trends of endogenous acyl ghrelin, des-acyl ghrelin, and glucagon-like peptide-2 (GLP-2) following massive small bowel resection.

		n	Pre-prandial plasma acyl ghrelin (fmol/ml)	Pre-prandial plasma des-acyl ghrelin (fmol/ml)	Postprandial plasma glucagon-like peptide-2 (ng/ml)
Control		3	53.6 ± 9.2	583.6 ± 79.1	2.0 ± 0.2
80%SBR	Day 1	6	93.3 ± 22.7	1021.6 ± 93.1 **	3.6 ± 0.4
	Day 4	6	104.7 ± 14.1 *	709.1 ± 62.6	4.3 ± 0.8 **,†
	Day 7	6	65.4 ± 6.7	490.1 ± 41.8	3.1 ± 0.2 ††
	Day 11	6	72.1 ± 10.0	508.2 ± 54.7	3.3 ± 0.3 ††
	Day 15	6	73.4 ± 6.1	481.7 ± 46.0	2.8 ± 0.4
Sham	Day 1	6	93.7 ± 11.6	917.2 ± 108.2 *	2.9 ± 0.4
	Day 4	6	110.7 ± 24.1	694.8 ± 22.1	2.0 ± 0.2
	Day 7	6	105.4 ± 19.5	584.5 ± 83.9	2.1 ± 0.2
	Day 11	6	91.2 ± 11.8	565.0 ± 72.5	1.9 ± 0.1
	Day 15	6	95.2 ± 21.6	538.7 ± 102.9	2.2 ± 0.2

Data are expressed as means ± SE. The differences between the groups and the time courses were evaluated by a 2-factor factorial analysis of variance (ANOVA) followed by Tukey's multiple-comparison posttest. Comparisons with controls were performed by Dunnett's test, and comparisons between groups at similar time points were performed by Student's *t*-test. There were no significant differences between groups ($F = 2.94$, $P = 0.09$) and time courses ($F = 0.86$, $P = 0.49$) in the levels of acyl ghrelin. The time course difference was significant ($F = 13.73$, $P < 0.01$); however, there was no difference between groups ($F = 0.15$, $P = 0.70$) in the levels of des-acyl ghrelin. There were significant differences between groups ($F = 28.55$, $P < 0.01$) but not between the time course changes ($F = 1.97$, $P = 0.11$) in the levels of GLP-2.

* $P < 0.05$ versus controls.
** $P < 0.01$ versus controls.
† $P < 0.05$ versus the sham-operated subjects.
†† $P < 0.01$ versus the sham-operated subjects.
80% SBR, 80% small bowel resection.

in the 80% SBR animals was reached at the same time as the start of the expansion of the absorptive mucosal surface on day 4. Subsequent steady gain in body weight implied the production of functionally mature enterocytes [29]. Both in the preclinical and clinical models, GLP-2 is a well-studied intestinotrophic factor that enhances nutrient and fluid absorption in the context of massive small bowel resection [11,22,28]. Exogenous GLP-2 stimulates crypt cell proliferation and results in a significant increase in the absorptive mucosal area due to villus lengthening [8,20,31]. Additionally, it promotes nutrient transporter expression, intestinal blood flow [20,31], and intestinal barrier function [5]. The significant increase in endogenous GLP-2 in our 80% SBR animals within the first 4 postoperative days may suggest the necessity of ensuring enough stimulation to initiate the expansion of the absorptive mucosal surface. GLP-2 secretion is stimulated by luminal nutrients [12,19,25]. It was interesting that both acyl ghrelin and GLP-2 increased immediately at the same time and peaked at the time when morphological adaptation became evident.

From our laboratory findings, we could imagine a more effective method of inducing residual intestinal adaptation after massive small bowel resection. In a previous study, GLP-2 receptor expression was shown to have significantly increased by postoperative day 3 in 90% SBR model rats [13]. Moreover, continued intravenous administration of GLP-2 during the first postoperative week was required to maximize the adaptation of the remaining bowel in rats [12,13]. These findings suggest that early treatment with a continuous infusion of GLP-2 would be clinically beneficial for short bowel syndrome patients. However, it has been reported that exogenous GLP-2 suppresses ghrelin secretion by nearly 10% in humans [4]. In addition, GLP-2 administration led to a significant elevation of glucagon level [33]. Glucagon is a powerful inhibitor of ghrelin in humans [1]. In rats, intravenous glucagon has been reported to upregulate the synthesis and release of des-acyl ghrelin [14]. The regulation of food intake with exogenous GLP-2 is controversial. Both peripheral [3] and central [36] administration of GLP-2 inhibited food intake in rodents. In healthy humans, the physiologic [33] and pharmacologic [30] GLP-2 doses given intravenously for 3 to 4.5 h had no effect. Yet, the effects of the sustained administration of GLP-2 on food intake in humans are still unknown. These reports imply that a combined administration of acyl ghrelin and GLP-2 would be more useful. As an example of a possible clinical application of a combination that reinforces physiological hormone secretion patterns, the therapeutic schema may be as follows. Under total parenteral nutritional support, a continuous intravenous administration of GLP-2 should be initiated to induce augmentation of the absorptive mucosal area. Additionally, oral food intake should be started as early as possible, and GLP-2 supplementation should be continued in the postprandial period with administration of acyl ghrelin in the preprandial period to stimulate adequate compensation in the remaining small intestine.

Further studies on the relation between acyl ghrelin, des-acyl ghrelin and GLP-2 after massive small bowel resection are necessary to gain concrete information that may aid in developing a more effective method to induce remnant intestinal adaptation.

5. Conclusion

This is the first report to show the trends of endogenous preprandial plasma acyl ghrelin, des-acyl ghrelin and postprandial plasma GLP-2 in the context of massive small bowel loss. The expansion of the absorptive mucosal surface area became evident after postoperative day 4. All the 3 gastrointestinal hormones studied were elevated immediately after resection. The acyl ghrelin and GLP-2 levels were peaked at the same time as when body weight and food intake recovered to the preoperative levels and as when the remnant intestinal adaptation started. A management strategy that could achieve active orexigenic and trophic effects at the same time may provide a clue as to the development of a new therapy for inducing intestinal compensation in short bowel syndrome patients.

Conflicts of interest

The authors declare that they have no conflicts of interest.

Acknowledgments

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●第39回日本小児栄養消化器肝臓学会　シンポジウム
小児の在宅栄養支援の問題点と今後の展開

小児腸管不全症例に対する在宅静脈栄養の現状と問題点

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Key Words：在宅静脈栄養，腸管不全，ヒルシュスプルング病類縁疾患，短腸症候群

要　旨

腸管不全患児において在宅静脈栄養（HPN）は社会・家庭復帰のために重要な役割を果たす。当科におけるHPNの現状と問題点について検討した。対象は1983年から2012年まで当科にてHPNを施行した40例。施行期間は8カ月～27年，疾患は腸管運動障害19例，難治性下痢10例，短腸症候群6例，その他5例であった。カテーテル（CVC）はカフ付きもしくは埋め込み型リザーバーを用いた。HPN施行においては院外薬局による無菌調剤とし，輸液ラインはクローズドシステムを用いた。微量元素，ビタミンは年齢に応じて一日必要量を毎日投与し，必要に応じてSeを投与した。転帰は離脱による終了が11例，原疾患による死亡が8例，転医が3例で，1例で小腸移植を行った。17例が現在も継続中である。1,000日あたりのCVC感染頻度は0.26であった。カテーテル関連合併症はカフ付きCVCで少なかった。適切なHPNの施行により合併症も少なく，多くの患児で成長発達を維持し，QOLの向上が可能となった。

緒　言

栄養療法においては消化管が機能している場合には経腸栄養が優先されるのが大原則である。しかし短腸症候群やヒルシュスプルング病類縁疾患をはじめとする腸管不全症例に対しては中心静脈栄養（Total Parenteral Nutirition：TPN）が長期にわたり必須となる。

在宅（中心）静脈栄養法（Home Parenteral Nutrition：HPN）とはTPNによるサポートを必要とする患児に家庭でTPNを行うことである。HPNにより患児は入院生活から解放され，成長していく上でかけがえのない大切な場所である家庭や学校への復帰を果たすことができるようになる。患児・家族ともquality of life（QOL）の向上がもたらされるだけでなく，その後の成長や発達に重要な機会を得ることができる。すなわち成長発達過程の小児においては，このHPNの成否が腸管不全の患児の予後とQOLを大きく左右するといっても過言ではない。

一方でHPNにも合併症が起こり得るのも事実である。カテーテル関連血流感染（Catheter Related Blood Stream Infection：CRBSI）をはじめとする感染性合併症，カテーテルの閉塞，破損などの機械的合併症，肝機能障害などの代謝性合併症が起こる危険があり，家庭においても入院時と同様に正しい知識によ

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表1 小児の在宅静脈栄養施行症例

基礎疾患	
腸管運動障害	19
難治性下痢	10
短腸症候群	6
その他	5
合計	40
性別	男児21 女児19
開始年齢	6カ月～15歳(平均3.2歳)
施行期間	244日～25.7年 (平均8.7年)

1983～2012年

る厳密な管理を要する。

当科では30年にわたり小児の腸管不全症例に対してHPNを行ってきた。これらの現状と問題点について後方視的に検討したので報告する。

対象・方法

1983年から2012年まで当科にてHPNを施行してきた小児40例を対象とした。男児21例，女児19例であり，対象となった疾患はHirschsprung病類縁疾患などの腸管運動障害19例，短腸症候群6例，難治性下痢10例，その他5例であった（表1）。

当科においてHPNは以下の手順で導入・維持した。症状が落ち着き家庭復帰を目指すにあたり，長期留置用のダクロンカフ付きカテーテル（Broviacカテーテル®）もしくは，皮下埋め込み型リザーバーを留置選択した（図1）。夜間を中心に間欠的TPNを行い，家族が十分にTPNの手技を習熟した段階でHPNに移行した。輸液は原則として院外薬局による無菌調剤とし，輸液ラインはクローズドシステムを用いた（図2）。微量元素，ビタミンは年齢に応じて一日必要量を毎日投与した。必要に応じて院内製剤によりSeを投与した。カテーテル刺入部の管理では刺入部の消毒はクロルヘキシジンまたはポビドンヨードを用い，患者の皮膚にあわせてフィルム型もしくはパッド型のドレッシングを選択した。

外来受診は安定期には月に1回程度とし，身長，体重を成長曲線にプロットして静的動的に成長を評価した。

カテーテルの種類による合併症の頻度を生存曲線にて評価した。またHPNの転帰，その他の合併症について後方視的に検討した。

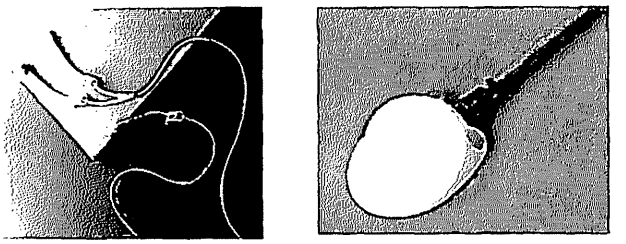

図1 HPNに用いるカテーテル

右：Broviacカテーテルと Hickmanカテーテル。ダクロンカフが皮下に癒着することにより長期間使用可能。シリコン製。破損すれば部位によっては修復可能である。
左：完全皮下埋込型ポート。これを皮下に留置する。中央の膜を特殊な針（Huber針）で穿刺して使用する。輸液をしていないときには体の外のデバイスがない状態を維持できる。

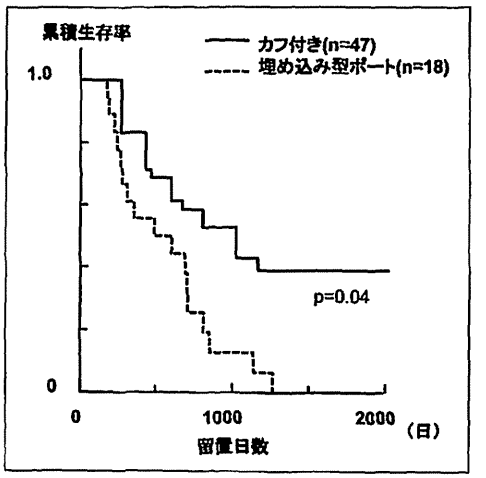

図2 カテーテルサバイバル分析

小児においてはカフ付きカテーテル（Broviacカテーテル）が合併症も少なく長持ちする傾向にあった。($p<0.05$)

結　果

開始時の年齢は6カ月～15歳（平均5.2歳），施行期間は継続中を含め，8カ月から27年（平均8.7年）であった。終了症例は離脱による終了が11例，死亡が8例，転医が3例，1例で長期の静脈栄養に伴う肝障害により，小腸移植を行った（表2）。死亡例のうち腸管不全およびHPNに関連する死亡は2例で腸炎による敗血症1例，肝不全1例であった。その他の4例は併存する消化管以外の疾患による死亡であった。

カテーテル1本当たりのCVCの使用期間は6カ月～7年（758±147日）で，1,000日あたりのCVC感染

表2　小児在宅静脈栄養の疾患別転帰

疾患名	症例数	継続	離脱	死亡	小腸移植	転医
腸管運動障害	19	10	2	5		2
難治性下痢	10	5	3	1	1	
短腸症候群	6	1	3	1		1
その他	5	1	3	1		
合計	40	17	11	8	1	3

表3　小児の在宅静脈栄養の合併症

合併症	症例数
成長障害	5例
肝障害	12例
血管閉塞	2例
その他	5例

※カテーテル関連の合併症をのぞく

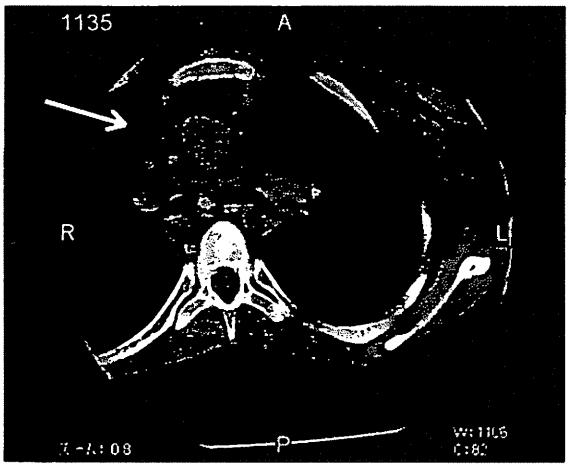

図3　長期HPNの合併症—SVC閉塞—

22歳女性。20年間HPNを継続していたが，上大静脈が完全閉塞した。現在大伏在静脈よりカテーテルを先端が下大静脈になるように留置している。カテーテルは腹部から体の外に出している。

頻度は0.26であった。カテーテル関連の合併症によるカテーテル使用期間は完全皮下埋め込み型ポートの方がカフ付きカテーテルよりも有意に高かった（図2）。

カテーテル関連以外の合併症では腸管運動障害の3例，難治性下痢の2例に成長障害を認め，GH補充療法を必要とした（表3）。肝障害は12例に認めた。その他に短腸症候群に伴うD-lactic acidosisを2例，性腺発育遅延2例，腎障害を1例に認めた。10年以上HPNを行った症例のうち，2例でSVCの完全閉塞を認めた（図3）。

考　察

在宅静脈栄養とは入院中に行っている栄養療法を自宅にて行い，家庭生活への復帰や，就学などを目指すものである[1)~4)]。在宅栄養療法の導入によって患児は入院生活から解放され，家庭復帰，就学・社会復帰を果たすことができるようになりQOLの向上がもたらされる。

適応となる疾患は静脈栄養を要する疾患の全てである。主に小児では消化吸収障害もしくは運動機能障害を来す腸管不全症例が適応になる。消化吸収障害を来す疾患としては壊死性腸炎・小腸閉鎖・中腸軸捻転などの腸管大量切除後の短腸症候群，難治性下痢があげられる。腸管運動機能障害は小児領域ではヒルシュスプルング病類縁疾患と分類され，hypoganglionosis，慢性偽性腸閉塞症（CIPO：Chronic idiopathic intestinal pseudo-obstruction），MMIHS（Megacystis Microcolon Intestinal Hypoperistalsis Syndrome）がある。その他にも重症のクローン病などの経腸栄養を行うことが病態の増悪につながるような疾患や，ある程度の消化管機能は保たれているものの経口摂取および経腸栄養において十分な栄養を投与することができない病態で，原疾患の管理に入院を必要としない患者全てがHPNの適応となる[5)~11)]。

HPN施行の条件は，病状に急激な変化がない慢性期であること，継続的な栄養療法が必要であること，そして栄養療法により確実な治療効果が得られることである。特に小児においては自己による管理を期待することはほとんどできず，家族などの保護者が在宅静脈栄養療法における介護の中心的役割を果たすことになるので，介護者でもある家族がHPNに積極的であることや，HPNとその合併症対策について十分な理解があることが不可欠である[1)~4),12)]。施行する施設においても，HPNの専門的知識と技術を有する医師，薬剤師，看護師，管理栄養士により，入院中から退院

後も継続して家族に対する教育，サポートを行うことが必要である[12]。さらに，さまざまなトラブル，緊急事態に対応が可能であるというのが条件である。

小児の在宅中心静脈栄養では，特に成人に比べ，破損や感染などのカテーテルに関する合併症が多い[13)~15)]。したがって長期に使用できる，長期留置用カテーテルが優先される。特にダクロンカフ付きのBroviacカテーテル®は長期を目的として作成されたもの[16]で，HPNに適している。成人でよく用いられる完全皮下埋め込み式ポートは輸液をしていない時間は体外部分がないという利点がある[15),17)]。しかし輸液の度に皮膚を穿刺するため，小児ではその痛みに耐え難いこともしばしばみられる。自験例でも学童期には通学の利便性から埋め込み型ポートを選択する患児もみられたが，穿刺の痛みに耐えられず，多くの患児が次の入れ替え時にはBroviacカテーテルを選択する傾向があった。また，破損や感染などの合併症は小児ではポートよりもBroviacタイプの方で少ない傾向がみられた。いずれにせよ，それぞれのカテーテルの特徴を熟知し，疾患，年齢，投与方法および患児のライフスタイルに合わせて選択すべきである。

HPNにおける最も重要な注意点の一つは感染の予防である。HPNにおいてもCRBSI（カテーテル関連血流感染症）は重篤な合併症である。刺入部・輸液ラインの観察は怠ってはならないのはいうまでもなく，薬剤師による無菌調剤，クローズドシステムによる輸液ラインの管理，無菌的カテーテル管理が必要である[13]。当科ではTPN導入時期よりクローズドシステムによる管理の重要性を報告してきている[18]。これらの手技をHPNにおいても徹底することにより，低い感染率を保つことが可能となっている。近年，長期の在宅静脈栄養時のCRBSI予防のためのエタノールロックが注目されている[19]。その他にも抗生剤ロック，ウロキナーゼロック，NaOHロックなど，カテーテルの入れ替えがそう簡単ではない小児において，カテーテルを長持ちさせるための様々な工夫がされている。ただし，いたずらにカテーテルロックを繰り返してはならない。重篤な感染を来した場合，全身状態がよくない場合，保存的治療に抵抗する場合には躊躇なくカテーテルを抜去することも必要である。

その他，カテーテルに関連する合併症に血管閉塞がある。当科でも10年以上HPNを施行した2症例においてSVCの閉塞を来す結果となった。幸い，両症例ともTPNの依存度が低く，現時点では小腸移植の適応とはなっていないが，長期にわたるHPNを行う場合には十分注意を要する合併症の一つである。

HPNは消化管が使えない患児においては，QOLを保ちながら栄養状態を維持する有効な手段であるが，長期のTPNにおいては感染のみならず代謝上の合併症や成長障害を引き起こすため，可能な限り早くに経腸栄養，経口摂取に移行し，離脱を図るべきである。特に短腸症候群では多くの症例で離脱可能と考えられる[1),20)~22)]。ただし，これらの症例でも無理な離脱は受験などのストレスにより急速な栄養障害に陥る危険がありその場合は速やかに静脈栄養を再開すべきである。

一方で，とくにヒルシュスプルング病類縁疾患の患児においてはTPNの依存度が高く[9]，多くの場合で離脱の見込みも少ないと考えられる。長期に及ぶHPNにより，肝障害や成長障害を来す症例がしばしばみられる。また，腸炎を繰り返すことにより，敗血症や重篤な肝障害に陥り，命を落とすこともある。こういった症例には今後，小腸移植を視野に入れた検討を行うべきである。

このような長期静脈栄養を行う腸管不全患児に見られる胆汁鬱滞性肝障害（Intestinal Failure-Associated Liver Disease：IFALD）は，難治性であり，ときに肝不全に進行する場合もある[23]。原因として長期の静脈栄養による血漿アミノ酸パターンの変化や，細菌叢の増殖，腸炎，感染症などが考えられる。過栄養を回避し，経腸栄養を早期開始し，敗血症を予防することが，胆汁鬱滞の発生を抑制することにつながると考えられている[24)~26)]。また最近では，このIFALDに対しω-3系の多価不飽和脂肪酸を含んだ脂肪乳剤が，減黄効果と肝酵素の上昇を抑えることが報告されている[16]。本邦では薬事認可を受けていないが，一部の施設で試験的に使用され，その有効性が報告されており[27),28)]，その効果が期待されている。

このように非常に長期にわたるHPN症例ではSe欠乏が問題となる[29]。Seは現在市販されている静脈栄養剤にはSeは含まれていない。Seは微量元素のひとつであるが，欠乏により心筋症[30]を起こすことが言われている。当科ではSeの院内製剤を静脈内投与し，Seの血中濃度を維持している[31]。

結　論

成長・発達の過程にある小児にとって，毎日が貴重でかけがえのない日である。症状が安定していれば漫然と入院を長引かせることなく，一日も早い家庭・社会復帰を目指すべきであり，消化管に障害のある患児にとって在宅静脈栄養は家庭復帰のための強力なツー

ルとなる。適切なHPNの施行により合併症も少なく，多くの患児で成長発達を維持し，QOLの向上が可能であった。

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Long-term out come of pediatric patients receiving home parenteral nutrition

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Some pediatric patients who either cannot be fed enterally or are unable to tolerate sufficient enteral calories to provide their nutrition requirements, particularly those with short bowel syndrome or intestinal failure, will eventually require home parenteral nutrition (HPN). During the past 30 years, we have managed 40 patients on a HPN for periods ranging from 244 days to 27 years. Silastic Broviac catheters or catheters with subcutaneous implantable reservoirs were inserted into the superior vena cava. Solutions were infused using a closed line system and volumetric pump. All patients improved their nutritional status. Following HPN, bowel adaptation and initiation of full oral alimentation become possible in 14 patients. Administration of HPN is a safe, successful technique for maintaining an optimal nutritional status in children with severe digestive disorders, and permits resumption of a more normal daily lifestyle.

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ORIGINAL ARTICLE

Impact of pediatric intestinal transplantation on intestinal failure in Japan: findings based on the Japanese intestinal transplant registry

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Abstract
Introduction We assessed the impact of intestinal transplantation on Japanese pediatric patients with intestinal failure with data from the Japanese intestinal transplant registry.
Methods Standardized forms were sent to all known intestinal transplantation programs, requesting information on transplants performed between 1996 and June 30, 2012. Patients younger than 18 years were analyzed. Patient and graft survival estimates were obtained using the Kaplan–Meier method.
Results Of the 14 intestinal transplants, 4 were deceased and 10 were living donor transplants. The primary indications were: short gut syndrome ($n = 7$), intestinal functional disorder ($n = 6$), and re-transplantation ($n = 1$). The overall 1- and 5-year patient survival rates were 77 and 57 %, respectively. In transplants performed after 2006 ($n = 6$), the one-year patient survival rate was 83 %, and the 5-year survival rate was 83 %. Graft one- and 5-year survival rates were 83 and 83 %, respectively. The living-related transplant survival rate was 80 % at 1 year and 68 % at 2 years, compared to 67 and 67 % for cadaveric transplant recipients. There were no statistically significant differences in patient ($p = 0.88$) and graft ($p = 0.76$) survival rates between living donor and cadaveric transplant recipients. All current survivors discontinued PN.
Conclusion Intestinal transplantation has become an effective therapy for patients with intestinal failure who cannot tolerate PN.

Keywords Intestinal transplant · Pediatric transplant · Japanese registry

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Introduction

Intestinal failure is caused by a critical reduction of functional gut mass to below the minimal amount necessary for adequate digestion and absorption to satisfy nutrient and fluid requirements for maintenance in adults and growth in children [1]. The most common type of intestinal failure is short bowel syndrome with an estimated incidence of 3–5 cases per 100 000 births per year

[2]. Advances in neonatal intensive care, anesthesia, nutritional support, and surgical techniques have improved the survival of children, so the prevalence of common causes of short bowel syndrome, including gastroschisis, necrotizing enterocolitis, and intestinal atresia has likely increased in recent years [3]. Some survivors, however, develop irreversible intestinal failure. The prognosis for intestinal failure related to short gut syndrome and intestinal motility disorders has improved dramatically owing to the development of parenteral nutrition (PN). Some children achieve long-term survival with PN at home with a relatively good quality of life, but others develop serious side effects that can eventually lead to death. However, PN-related complications, such as loss of venous access and intestinal failure-associated liver disease (IFALD), are still major problems for patients with intestinal failure [4]. Intestinal transplantation can significantly improve their prognosis and quality of life. Early efforts to transplant the small bowel have failed due to refractory graft rejection and sepsis. Outcomes improved during the early 1990s, but survival rates were still inferior to those for other organ transplants. Over the past 5 years, individual centers have reported improved outcomes with better long-term intestinal engraftment.

The first intestinal transplant in Japan was performed in 1996. The total number of intestinal transplants in Japan has increased to 24 as of June 2011. We assessed the impact of intestinal transplantation on Japanese pediatric patients with intestinal failure based on data from the Japanese intestinal transplant registry.

Methods

Standardized forms were sent to all known intestinal transplantation programs, requesting information on intestinal transplants performed between 1996 and June 30, 2012. The data included age, sex, date of birth, date of transplant, type of donor (deceased or living), pre-transplant status (home or hospital), underlying disease, procedure, ABO blood type, immunosuppression regimen (induction and maintenance therapy), and post-transplant status (PN requirement, intravenous (IV) fluid requirement, and daily life restrictions). Patients under 18 years of age were analyzed. The data were entered into a Microsoft Excel spreadsheet and analyzed with JMP version 10.0 (SAS Institute Inc, USA). Patient and graft survival estimates were obtained using the Kaplan–Meier method. For survival analysis, failure was defined as occurring on the date of graft removal or death. A p value <0.05 was considered statistically significant. This study was approved by the institutional review board.

Results

Four programs provided data on 14 grafts in 13 patients who were received transplants between 1 April 1996, and 30 June 2012 in Japan. The participation rate was 100 %. All intestinal transplants performed in Japan are captured in the registry database. All patients were followed, unless the patient has passed way. Ten grafts were obtained from living donors, and four cases involved deceased donors. The annual number of intestinal transplants, according to organ donation type, is shown in Fig. 1. Prior to 2005, 25 % of patients who underwent transplantation were called in from home, as compared with 66 % in the last 5 years (Fig. 2).

There were nine male and five female recipients. The age distribution of the recipients is shown in Fig. 3. Two-thirds of the patients were over 6 years old. The youngest recipient was 8 months. The causes of intestinal failure requiring intestinal transplantation are shown in Fig. 4. Approximately half of the patients had conditions that result in short gut syndrome.

Most patients ($n = 13$) received isolated intestinal transplants. There was only one case of simultaneous liver-intestinal transplantation from two living-related donors. Twelve patients received grafts from donors with an identical ABO blood type. Two patients received grafts from ABO compatible donors. There were no transplants involving ABO incompatibility. All patients were on tacrolimus maintenance therapy. The types of induction therapy used are shown in Fig. 5. Antibody-based induction therapy and tacrolimus-based maintenance immunosuppression were used even if the medication was not commercially available in Japan.

Graft and patient overall survival as of June 2011 are shown in Kaplan–Meier plots (Fig. 6a, b, respectively). The one-year and 5-year patient survival rates were 77 and 57 %, respectively, comparable with rates from the international intestinal transplant registry. Five recipients died.

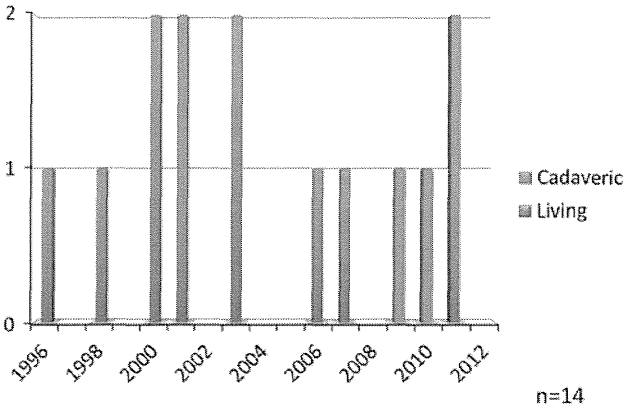

Fig. 1 Number of intestinal transplants by year

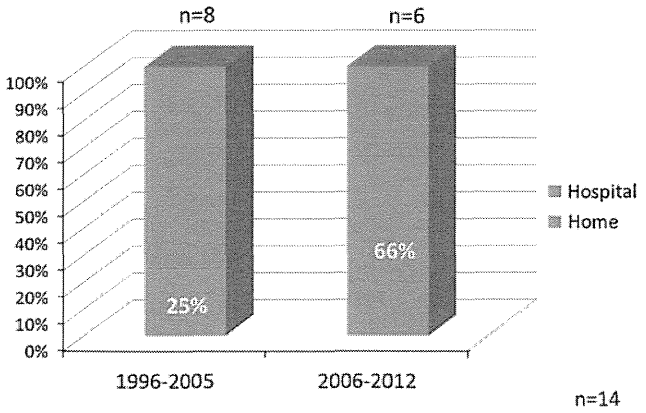

Fig. 2 Pre-transplant patient status

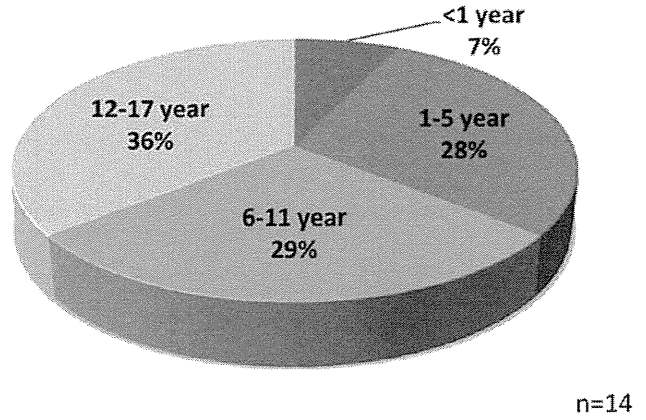

Fig. 3 Recipient age at transplant

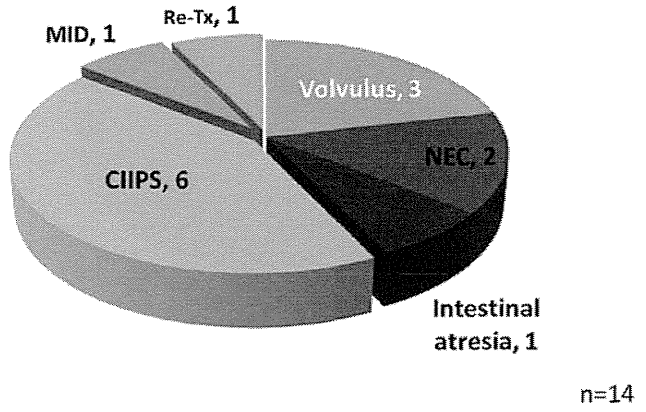

Fig. 4 Cause of intestinal failure *NEC* necrotizing enterocolitis, *CIIPS* chronic idiopathic intestinal pseudo-obstruction syndrome, *MID* microvillus inclusion disease, *Re-Tx* Re-transplant

The causes of death included sepsis ($n = 3$), post-transplant lymphoma ($n = 1$) and intra cranial hemorrhage ($n = 1$).

The 1-year overall graft survival rate was 80 % for cadaveric grafts versus 50 % for living donor grafts ($p = 0.76$), as shown in Fig. 7a. The 1-year overall patient survival rate was 80 % for cadaveric grafts versus 67 % for living donor grafts ($p = 0.88$), as shown in Fig. 7b.

Graft survival improved over the last 5 years. The one- and five-year graft survival rates were 83 and 83 % for 2006–2011 versus 63 and 38 % for 1996–2005 ($p = 0.14$), as shown in Fig. 8a. The 1- and 5-year patient survival rates were 83 and 83 % for 2006–2011 versus 71 and 43 % for 1996–2005 ($p = 0.27$), as shown in Fig. 8b.

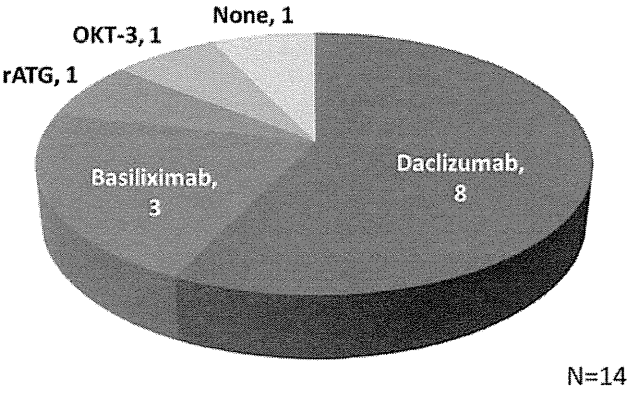

Fig. 5 Induction immunosuppression therapy *rATG* rabbit anti-thymus globulin, *OKT-3* anti-CD3 monoclonal antibody

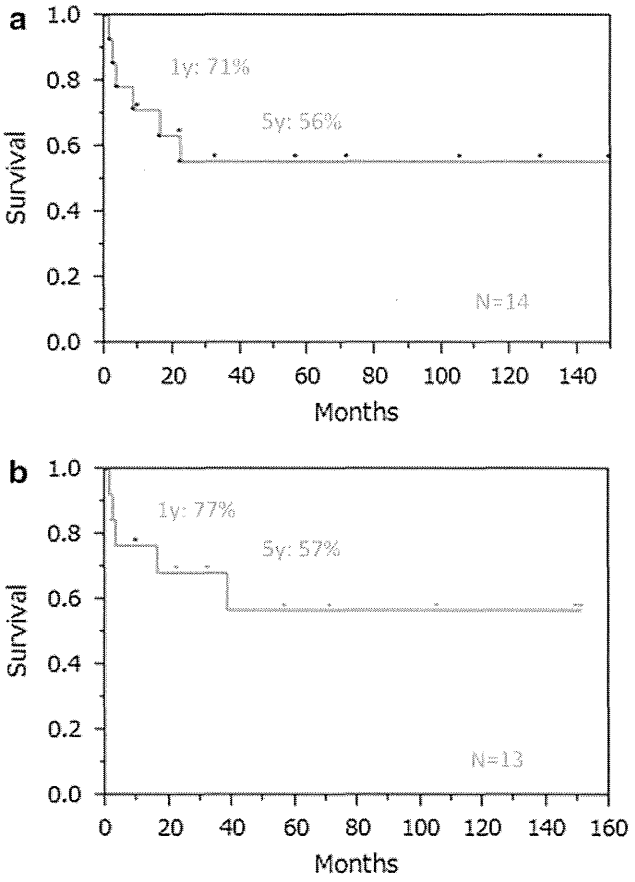

Fig. 6 Overall graft (**a**) and patient (**b**) survival

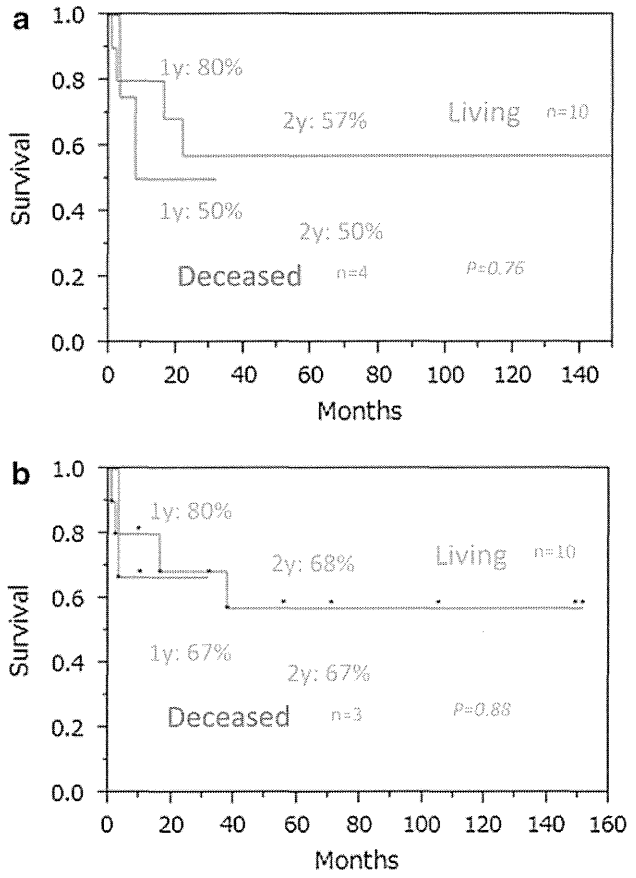

Fig. 7 Graft (**a**) and patient (**b**) survival according to graft type

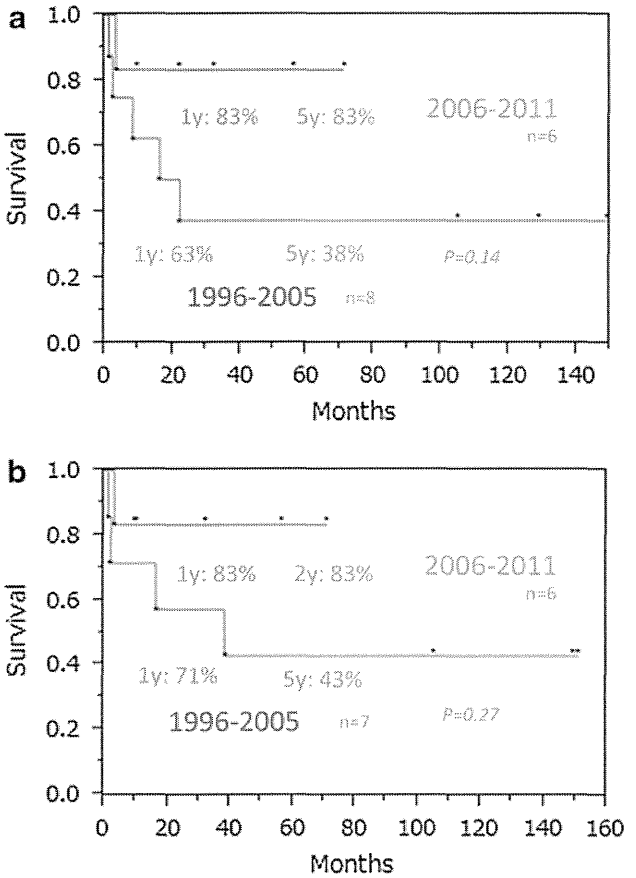

Fig. 8 Graft (**a**) and patient (**b**) survival by era

Graft function in terms of PN dependence was excellent. All patients became PN-free after intestinal transplantation, although two-thirds of patients require continuous or intermittent intravenous fluid support. Of the eight patients who were alive at the time of data collection, all patients were off parenteral nutrition, with three patients requiring intravenous fluids daily, two patients requiring intravenous fluids occasionally (Fig. 9). Most recipients stopped parenteral supplementation, eat, and have resumed normal activities. Of the seven surviving patients 1 year after transplant, six lead a full life.

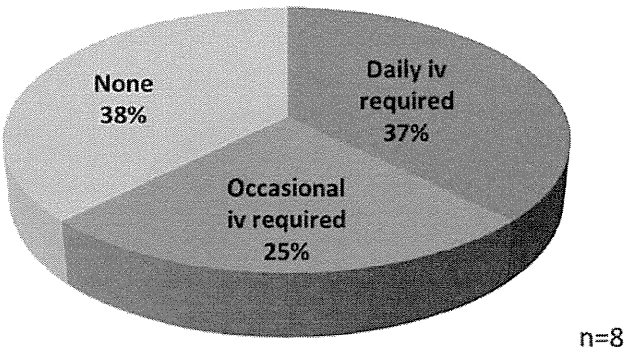

Fig. 9 Intravenous (IV) fluid requirement after intestinal transplantation

Discussion

Children with intestinal failure are at risk for numerous complications, especially PN-related complications. For example, loss of venous access and IFALD are still major problems for patients with intestinal failure because they are potentially life-threatening [4].

Catheter-related bloodstream infections were common in patients with intestinal failure [5]. Survival of children with chronic intestinal failure has increased as result of home PN. Adequate central venous accesses crucial for the successful management of home PN, but venous access can be complicated by episodes of catheter-associated infection, repeated procedures to replace catheters, and catheter-related thrombosis. Management and prevention of catheter-related thrombosis are of vital importance. [6].

IFALD can be a progressive and fatal entity in children with short gut syndrome. Parenteral fish oil-based fat emulsions are safe and may be effective in the treatment of PN-associated liver disease [7]. A lipid reduction protocol may prevent cholestasis [8]. Despite all efforts to prevent