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Patient Report

Antenatally diagnosed, intermittently worsened hydronephrosis caused by a ureteral polyp

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Key words fibroepithelial polyp, hyronephrosis, ureteral polyp.

Congenital hydronephrosis is commonly caused by a ureteropelvic junction obstruction (UPJO), which is caused by a developmental anomaly of the ureter in fetal life. This results in stenosis, kinking or vessel compression in the upper part of the ureter.

A ureteral polyp is a very rare cause of UPJO and was first reported in 1958.[1] In 2007 two case reports were published,[2,3] bringing the total now to approximately 40 reported pediatric cases of ureteral polyps in the UPJ.

Here we report a new case of ureteral polyp that caused hydronephrosis. This case was antenatally diagnosed as a left hydronephrosis and several episodes of intermittent worsening of hydronephrosis were experienced since infancy.

Case report

An 8-year-old boy was admitted to University of Tokyo Hospital because of left flank pain and macroscopic hematuria. He had been followed in the outpatient clinic because of bilateral hydronephrosis (UPJO) that was diagnosed at fetal stage. Hydronephrosis was I° in the right side and II° in the left side. In infancy the left-sided hydronephrosis showed periodic worsening on chance ultrasonography (Fig. 1a,b), but there was no symptom. He first experienced left flank pain and macroscopic hematuria at age 5 as his left hydronephrosis worsened at the same time and was confirmed on ultrasonography. Since the first episode he experienced four repetitions of similar flank pain and hematuria. Operative treatment was recommended each time but the parents did not consent to it. Given that renal function did not worsen after the episodes, as ascertained on renal scintigraphy, we decided to observe the natural course. At the last episode, when the patient was 8 years old, his parents agreed to left-sided pyeloplasty. Preoperative magnetic resonance imaging (MRI) indicated irregularity of the left UPJ but the existence of the polyp was not detected (Fig. 1c).

Anterolateral skin incision was made and a retroperitoneal approach was selected for the Anderson–Hynes pyeloplasty. Macroscopically, the left upper ureter was not stenotic at the UPJ and an aberrant vessel did not exist. Instead, the ureter around the UPJ was mildly dilated (Fig. 2a; arrow). When the pelvic incision was made, a thin and long polypoid lesion protruded from the left ureter into the pelvis (Fig. 2a; arrowhead). The lesion originated at approximately 2 cm distal to the UPJ, and was confirmed after the longitudinal opening of the left upper ureter (Fig. 2b). The ureter was resected from the UPJ to the neck of the polypoid lesion and the Anderson–Hynes pyeloplasty was completed. The postoperative course was uneventful and the patient was discharged 9 days after the surgery.

Pathology indicated that the polypoid lesion was benign, and urothelial epithelium covered the fibrotic stroma with some bleeding and inflammatory cell invasion (Fig. 3). Histopathological diagnosis was a fibroepithelial polyp with mild inflammation.

Discussion

Recently hydronephrosis at the UPJ has been commonly diagnosed antenatally owing to the widespread use of fetal ultrasonography. Hydronephrosis is usually caused by ureteral stenosis, kinking, and the compression of the aberrant vessel. Another albeit rare cause of UPJO, however, has been reported: that is, ureteral polyp. According to the large study reported by Adey *et al.*, the incidence of ureteral polyp is 0.5% among total cases of UPJO.[4] Patients were predominantly male (89%) and the polyps were commonly found on the left side (78%). In 2007 two reports were published,[2,3] which added to a total of nine recently described cases. Thus far, approximately 40 pediatric cases have been reported in the English-language literature.

Most common symptoms are hematuria and flank pain on the affected side.[2–4] Intermittent hydronephrosis is another name for the disease. Preoperative diagnosis of the polyp is not easy, as evidenced by the diagnosis of only 22% on i.v. pyelography in one series.[4] Indeed, in the present case MRI showed the irregular shape of the upper ureter but the polyp was not diagnosed preoperatively. It is recommended that retrograde pyeloureterography should be done before the operation to confirm the etiology of the stenosis.

The Anderson–Hynes dismembered pyeloplasty is recommended in most cases, but Ruiz-Lopez *et al.* described a case in which a ureteral polyp was resected first instead of undergoing a

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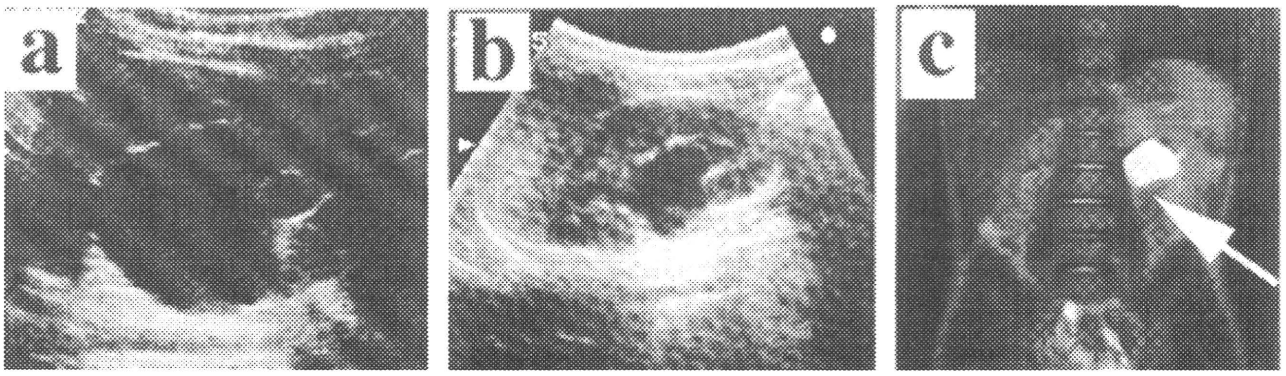

Fig. 1 (a) Ultrasonogram taken at 7 months of age. Pelvic dilatation was prominent and hydronephrosis had apparently worsened. (b) Ultrasonogram taken at 9 months of age. Pelvic dilatation improved compared to that at 7 months of age. (c) Magnetic resonance imaging taken before dismembered pyeloplasty: At the ureteropelvic junction there was some irregular texture detected (arrow).

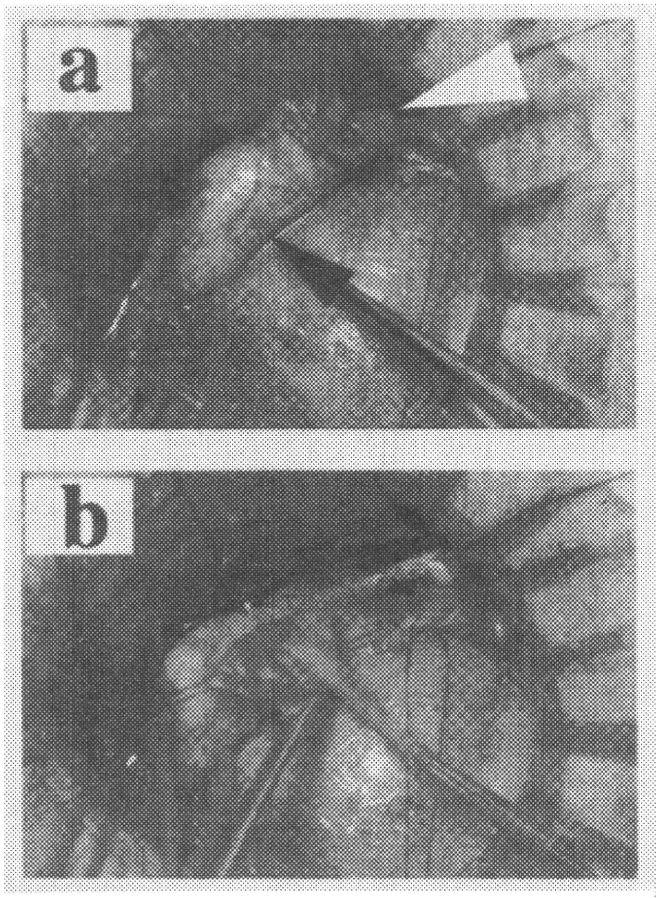

Fig. 2 (a) Macroscopic appearance of the left upper ureter: No narrowing was detected and the diameter of the ureter approximately 2 cm below the ureteropelvic junction was slightly widened (arrow). A thin, long tissue protruded from the cut edge of the pelvis (arrowhead). (b) Macroscopic appearance of the ureteral polyp. It was based at 2 cm distal to the ureteropelvic junction and bled easily when handled with forceps.

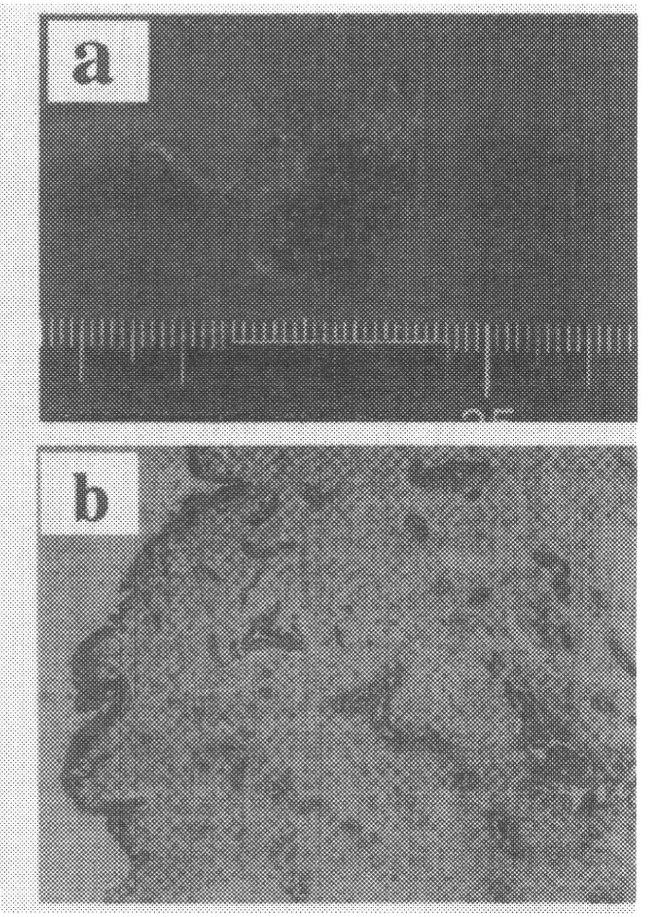

Fig. 3 (a) Macroscopic appearance of the polyp. It was slender and cylindrical in shape and approximately 2 cm long with a fragile mucosal surface. (b) Microscopy features of the ureteral polyp. Urothelial epithelium covered the surface of the polyp and there were loose fibrous tissues with some dilated vessels in the stroma of the polyp.

dismembered pyeloplasty.[5] This resulted in a recurrence of the polyp and necessitated a repeat resection. Thus, a local resection of the polyp is not regarded as the recommended procedure. Others, however, have suggested polypectomy under ureteroscopy in adult and elderly patients.[6] In pediatric patients, however, ureteroscopy is not easy to perform and complete resection of the polyp may be a technically difficult procedure. Thus, dismembered pyeloplasty is a stable and established technique and is recommended for children.

It should be noted that the polyps may be multiple,[6] and have been discovered on both sides in some cases.[4] Careful follow up is needed after the operative procedure.

Pathologically the polyp is usually defined as a fibroepithelial polyp and is not malignant in children. This report now represents the second case of a congenitally derived ureteral polyp causing an obstruction at the UPJ leading to hydronephrosis.

It should be kept in mind that intermittent worsening of hydronephrosis may suggest an etiology of ureteral polyp and should be examined on MRI or intravenous pyelogram.

References

1 Compere DE, Begley GF, Isaacks HE *et al.* Ureteral polyps. *J. Urol.* 1958; **79**: 209–14.

2 Delia EC, Joseph VT, Sherwood W. Fibroepithelial polyps causing ureteropelvic junction obstruction in children: A case report and review article. *Eur. J. Pediatr. Surg.* 2007; **17**: 142–6.

3 Niu ZB, Yang Y, Hou Y, Chen H *et al.* Ureteral polyps: An etiological factor of hydronephrosis in children that should not be ignored. *Pediatr. Surg. Int.* 2007; **23**: 323–6.

4 Adey GS, Vargas SO, Retik AB *et al.* Fibroepithelial polyps causing ureteropelvic junction obstruction in children. *J. Urol.* 2003; **169**: 1834–6.

5 Ruiz-Lopez MJ, Ramirez-Garrido FR, Nogueras-Ocana M *et al.* Recurrent ureteric fibroepithelial polyp in a child. *Eur. J. Pediatr.* 2004; **163**: 124–5.

6 Zeman L, Dusek M, Lisy J *et al.* Multiple fibroepithelial polyps of the upper ureter in a 17-year-old boy: Case report and review of the literature. *Eur. J. Pediatr. Surg.* 2004; **14**: 358–61.

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Patient Report

Case report: Prenatal intervention for severe anterior urethral valve

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Key words anterior urethral valve, obstructive nephropathy, prenatal intervention, vesico-amniotic shunting.

The presence of an anterior urethral valve (AUV), although uncommon compared with that of a posterior urethral valve (PUV), is one of the causes of obstructive uropathies in children. Congenital obstructive uropathies, when not severe, are rarely diagnosed during infancy, but some are diagnosed prenatally on the basis of findings such as oligohydramnios, hydronephrosis, hydroureter, urinoma and megacystis. Recently, fetal intervention has been performed in some patients with obstructive uropathies,[1–4] although its efficacy is still controversial. We herein present a patient with severe AUV on which the infusion of artificial amniotic fluid and vesico-amniotic shunting were performed. Although the long-term outcome of this patient's renal and urinary bladder functions needs to be followed up carefully, pulmonary maturation was achieved and the exacerbation of the obstructive nephropathy was prevented.

Case Report

A 25-year-old mother was introduced and admitted to The Tokyo University Hospital at 31 weeks of pregnancy because of oligohydramnios. The oligohydramnios was not noted before 30 weeks. Fetal ultrasonography at 31 weeks revealed a left perinephric urinoma, megacystis, bilateral hydronephrosis and hydroureter in the fetus. The fetus was male and had no other congenital anomalies suggesting chromosomal abnormalities. The mother had no other complications.

Lower urinary tract obstruction in the fetus was suspected. Although there was a risk of premature labor, his hydroureter deteriorated in a few days, and pulmonary immaturity was strongly suspected at this gestational age. Therefore, after obtaining informed consent from the parents, the infusion of artificial amniotic fluid followed by vesico-amniotic shunting was performed three times during the 32nd and 33rd weeks of gestation (Fig. 1). After the vesico-amniotic shunting, the hydronephrosis, hydroureter and left urinoma showed no prominent change, and the enlarged urinary bladder decreased in size markedly. The oligohydramnios improved and the volume of amniotic fluid was maintained thereafter. The sodium level of the fetal urine at first was 154 mEq/L; potassium, 0.8 mEq/L; chloride, 144 mEq/L; creatinine, <0.3 mg/dL; N-acetyl-β-D-glucosaminidase (NAG), 0.8 IU/L; and β_2-microglobulin, 0.46 mg/L. A serial chemical analysis of the fetal urine showed a rapid decrease in sodium (to 69 mEq/L) and chloride (to 59 mEq/l) levels.

The amniotic membrane ruptured prematurely and a male infant was vaginally delivered at 33 weeks and 3 days of gestation weighing 2369 g with an Apgar score of 6 at 1 min and presenting with neonatal asphyxia. Because his spontaneous breathing was weak 3 min after the delivery, he was intubated immediately and an artificial surfactant was administered intratracheally. His body temperature was 36.6°C, his heart rate was 151 b.p.m., and his systemic blood pressure was 65/38 mmHg. He had no external malformations or any other abnormal clinical findings. After he was admitted to the neonatal intensive care unit, the basket catheters used for the vesico-amniotic shunting were removed.

Biochemical examination revealed: sodium, 136 mEq/L; potassium, 4.5 mEq/L; chloride, 103 mEq/L; serum creatinine, 0.56 mg/dL; and blood urea nitrogen, 5.6 mg/dL. Urinalysis showed proteinuria (2+), hematuria (occult blood [3+]), 21–50 red blood cells per high-power field and 6–10 white blood cells per high-power field. Urinary examination revealed: osmolarity, 221 mOsm/L; sodium, 89 mEq/L; chloride, 71 mEq/L; NAG, 22.4 IU/L; and α1-microglobulin, 37.2 mg/L. A chest X-ray showed no abnormalities. Ultrasonography revealed left kidney atrophy measuring 3.1 cm × 2.0 cm, left hydronephrosis and a perinephric urinoma. The right kidney was not atrophic, measuring 5.0 cm × 2.7 cm, with hydronephrosis and hydroureter. A thickening of the urinary bladder wall was detected.

He was initially ventilated with high-frequency oscillation and his respiratory condition gradually improved. He was successfully extubated on the 19th postpartum day.

A transurethral catheter (5 Fr) was smoothly inserted after birth and urine was successfully drained. Following tube-feeding with regular formula milk, his serum potassium level increased to 7.31 mEq/L on the 7th postpartum day. His serum creatinine level at that time was 1.68 mg/dL. Glucose-insulin therapy was performed and MM3 milk, a special formula for renal failure

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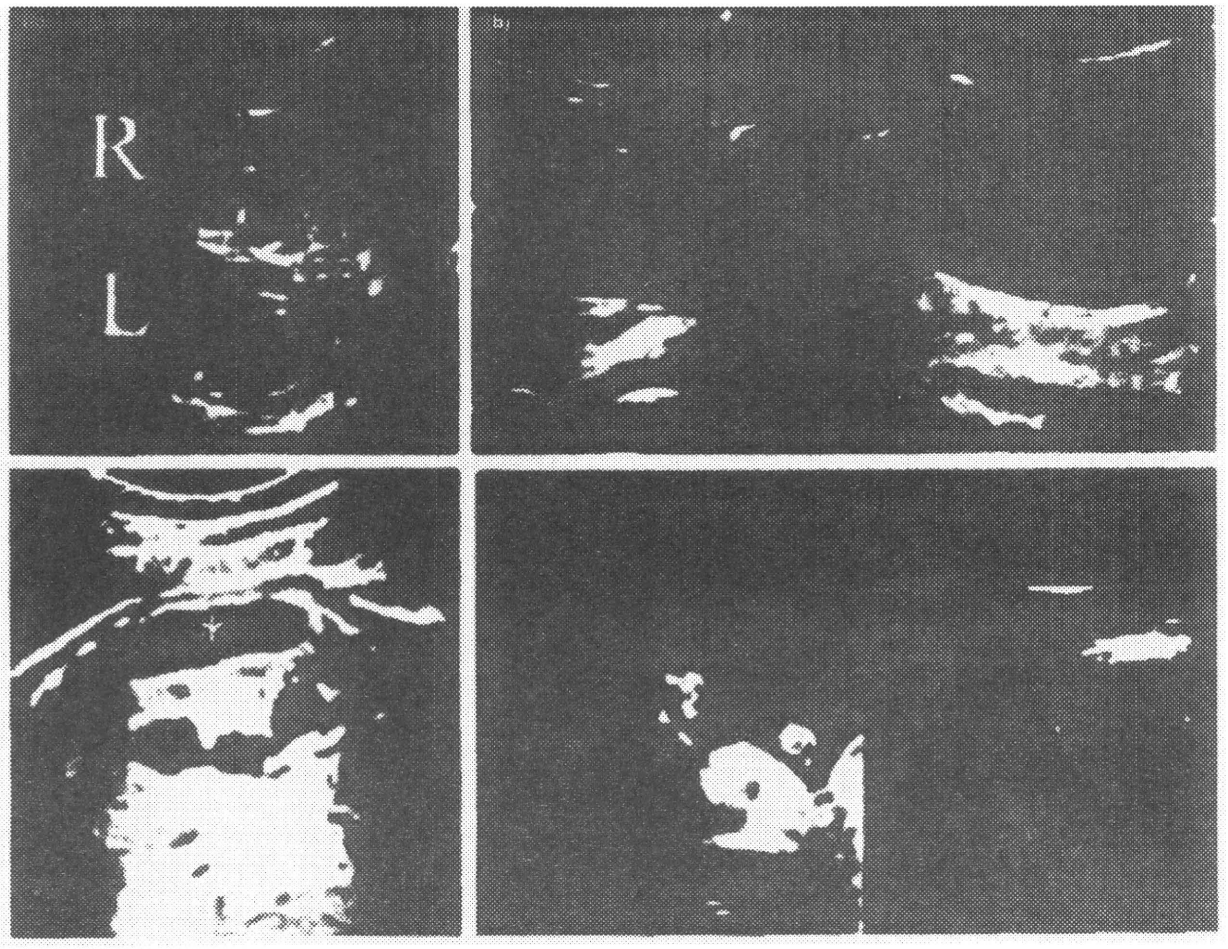

Fig. 1 Fetal ultrasonograms. (a) (Before *in utero* shunting) bilateral hydronephrosis was detected. (b) (Before *in utero* shunting) the urinary bladder was enlarged. (c) (After *in utero* shunting) the left urinoma (*) and hydronephrosis showed no change. (d) (After *in utero* shunting) the size of the urinary bladder was markedly decreased. The urinary bladder wall was markedly thickened.

containing a small amount of potassium (0.17 mEq/dL), was administered. He had an episode of urinary tract infection on the 16th postpartum day. The insertion of a new transurethral catheter was unsuccessful even though the catheter with the smallest diameter (3 Fr) was used, and surgical cystostomy was performed immediately. On the 35th postpartum day, cystostomy was performed again because of the difficulty of replacing the catheter of the initial cystostomy. The time course of his renal function is shown in Figure 2. His creatinine clearance improved up to 50 mL/min/m^2, and the creatinine level decreased to 0.4 mg/dL. When the regular formula milk was restarted, his serum potassium level increased, so he was alternately fed with MM3 and regular formula milk.

When he was 2 months old, voiding cystourethrography (VCUG) (Fig. 3), renal diethylene triamine pentaacetic acid (DTPA) scintigraphy, and magnetic resonance imaging (MRI) were performed. The initial contrast study was postponed to this time because his general condition had been unstable, that is, urinary tract infection had repeated, and his serum potassium level was unstable. VCUG demonstrated left vesicoureteral regurgitation (VUR) with severe dilation of the left ureter but no right VUR. An anterior urethral diverticulum was also identified. Renal DTPA scintigraphy (not shown) revealed a nonfunctional pattern in the left kidney and delayed washout in the right kidney. The glomerular filtration rates (GFR) of the left and right kidneys were 36.4 and 63.7%, respectively. MRI results (not shown) showed bilateral hydronephrosis, hydroureters, and a thickened urinary bladder wall. The anterior urethra showed a water density area 16 mm in diameter. These findings strongly suggested the presence of AUV. Cystourethroscopic examination was not performed at that time because the urethra was too narrow. At 3 months of age, his bodyweight reached 4.4 kg, and he was definitively diagnosed as having AUV by cystourethroscopy. Temporary diversion with cutaneous vesicostomy was conducted, and transurethral incision of diverticulum or open urethral reconstruction will be undergone when his weight increases.

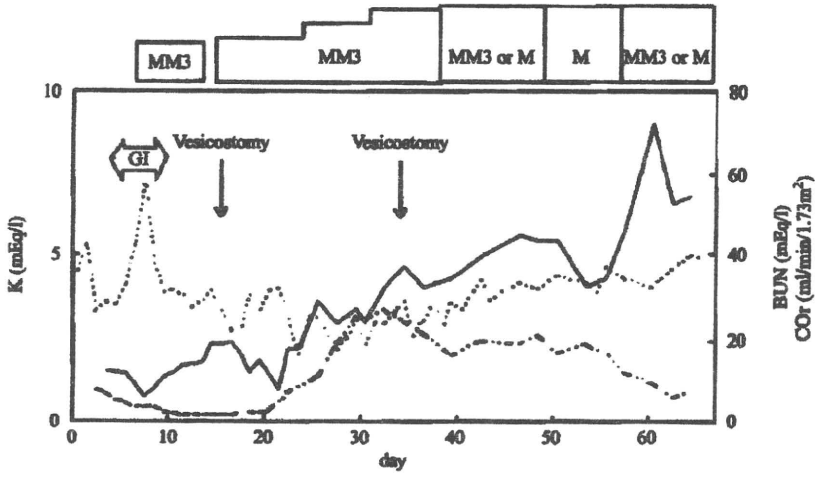

Fig. 2 Time course of renal function and treatment. With a series of treatments including prenatal intervention, postnatal cystostomy, and electrolyte management, the patient's renal function showed improvement during the first two postpartum months. K (dotted line), BUN (dotted and solid line), CCr (solid line). BUN, blood urea nitrogen; CCr, creatinine clearance; GI, glucose-insulin therapy; K, potassium; M, regular formula milk; MM3, special formula milk containing a small amount of potassium.

Discussion

Recently, Merrot *et al.*[5] reported a patient with AUV managed by prenatal urinary ascites evacuation. To our knowledge, this is the first report on a patient with AUV treated by vesico-amniotic shunting. There are several reports on the *in utero* treatment of obstructive uropathies other than AUV, but there are no randomized or large multicenter studies regarding the long-term outcome of *in utero* treatment. Antenatal vesico-amniotic shunting aims at relieving urinary obstruction and providing adequate amniotic fluid to improve renal and urinary bladder functions as well as lung maturation. According to several reports,[1–4] the overall survival rate is 40–91% in the cases treated with *in utero* shunting. Approximately 35% of patients who underwent *in utero* shunting developed renal failure. The complication rate was 10–45%. It is difficult to definitely assess whether prenatal intervention is effective, because the patients who underwent *in utero* shunting might have represented the worst end of the spectrum of obstructive uropathies. Even so, the above rate of renal failure suggests a possible benefit from *in utero* shunting. When the chemical analysis of fetal urine indicates sodium < 100 mEq/L, chloride < 90 mEq/L, osmolarity < 200 mOsm/L, β_2-microglobulin < 6 mg/L, calcium < 8 mg/dL and total protein < 20 mg/dL, the renal function of the fetus is regarded as good.[6]

In this patient, oligohydramnios, hydronephrosis and hydroureter were observed prenatally and lower urinary tract obstruction was suspected. Such urethral obstruction may lead to urinary tract rupture, resulting in ascites or perinephric urinoma.[7] This patient exhibited a left perinephric urinoma suggesting that the urethral obstruction was very severe. When severe urethral obstruction was suspected at 31 weeks of gestation, we had only two possible options: fetal intervention or an emergency cesarean section. Considering the pulmonary immaturity and possible respiratory complications such as respiratory distress syndrome and dry lung syndrome, we performed vesico-amniotic shunting to decompress the urinary tract and maintain an adequate volume of amniotic fluid for lung maturation. Vesico-amniotic shunting is superior to single vesicocentesis for the continuous drainage of fetal urine.

After the vesico-amniotic shunting and infusion of artificial amniotic fluid, the chemical analysis of the fetal urine showed a rapid improvement in sodium and chloride levels. The enlarged urinary bladder decreased in size, and the volume of amniotic fluid was maintained. The patient showed the symptom of acute respiratory distress after birth, but he was successfully extubated and developed no chronic lung disease. The function of the right kidney was maintained. Although it might be partly due to the "pop-off" phenomenon, he does not require dialysis treatment at the moment as a result of a series of treatments including prenatal intervention, postnatal cystostomy, and electrolyte management.

As for an expert and critical treatment for the anterior urethral valve, urethroscopic treatment during the neonatal period should be taken into account. Our patient will undergo transurethral incision of diverticulum or open urethral reconstruction after weight gain.

Because AUV is a very rare obstructive uropathy, there has been no report regarding the prognostic factors of AUV. According to Ylinen *et al.*,[8] who investigated the prognosis of PUV, the initial serum creatinine level, the maximum level of serum creatinine during the first year, the presence of bilateral VUR, and breakthrough urinary tract infections are prognostic factors of PUV. Bajpai *et al.*[9] showed that children with a normal or near normal serum creatinine level (0.8 mg/dL or less) at 1 year of age are able to maintain good renal function at the time of final evaluation (1.0 mg/dL or less). Lopez *et al.*[10] reported that the most significant prognostic factor is GFR at 1 year of age. The onset of proteinuria during the follow-up period is also associated with a poor prognosis. Our patient showed the following poor prognostic factors such as: left unilateral VUR, proteinuria, and an episode of urinary tract infection. The long-term outcome of his renal and urinary bladder functions should be followed up

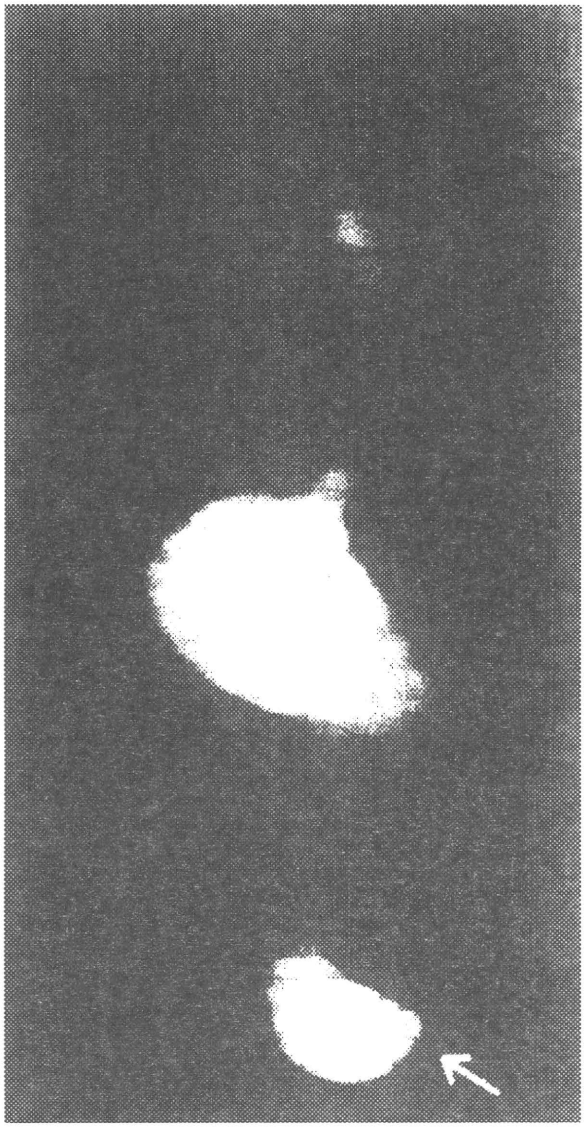

Fig. 3 Voiding cystourethrography demonstrated left vesicoureteral regurgitation with severe dilation of the left ureter. The anterior urethral diverticulum is indicated by an arrow.

paying particular attention to ultrasonographic findings, and serum creatinine level.

In conclusion, we performed prenatal intervention for a patient with severe AUV. Although the efficacy of this intervention on renal function awaits further evaluation, we believe that pulmonary maturation was successfully attained, and the exacerbation of renal function problems was prevented. There are some controversies regarding prenatal intervention and postnatal management; thus, it is important for pediatricians to cooperate with obstetricians and pediatric surgeons to preserve renal function in such cases of severe urinary tract obstruction.

References

1 Clark TJ, Martin WL, Divakaran TG, Whittle MJ, Kilby MD, Khan KS. Prenatal bladder drainage in the management of fetal lower urinary tract obstruction: A systematic review and meta-analysis. *Obstet. Gynecol.* 2003; **102**: 367–82.

2 Coplen DE. Prenatal intervention for hydronephrosis. *J. Urol.* 1997; **157**: 2270–7.

3 Biard J, Johnson MP, Carr MC *et al.* Long-term outcomes in children treated by prenatal vesicoamniotic shunting for lower urinary tract obstruction. *Obstet. Gynecol.* 2005; **106**: 503–8.

4 Freedman AL, Johnson MP, Smith CA, Gonzalez R, Evans MI. Long-term outcome in children after antenatal intervention for obstructive uropathies. *Lancet.* 1999; **354**: 374–7.

5 Merrot T, Chaumoitre K, Shojai R, D'Ercole C, Alessandrini P. Fetal bladder rupture due to anterior urethral valves. *Urology* 2003; **61**: 1259.

6 Johnson MP, Corsi P, Bradfield W *et al.* Sequential urinalysis improves evaluation of renal function in obstructive uropathy. *Am. J. Obstet. Gynecol.* 1995; **173**: 59–65.

7 Obara W, Konda R, Takashi S, Ishikawa K, Kinjo M, Fujioka T. Neonatal abdominal wall urinoma due to rupture of anterior urethral diverticulum. *Int. J. Urol.* 2006; **13**: 395–6.

8 Ylinen E, Ala-Houhala M, Wikstrom S. Prognostic factors of posterior urethral valves and the role of antenatal detection. *Pediatr. Nephrol.* 2004; **19**: 874–9.

9 Bajpai M, Daves S, Gupta DK. Factors affecting outcome in the management of posterior urethral valves. *Pediatr. Surg. Int.* 2001; **17**: 11–5.

10 Lopez PP, Espinosa L, Martinez UMJ, Lobato R, Navarro M, Jaureguizar E. Posterior urethral valves: Prognostic factors. *BJU Int.* 2003; **91**: 687–90.

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Original Article

Early use of probiotics is important therapy in infants with severe congenital anomaly

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Abstract ***Background***: Infants with severe congenital anomaly often need to undergo operation followed by antibiotic therapy. As a result they inevitably acquire abnormal intestinal microbiota, which cause severe infections such as necrotizing enterocolitis. Also, intestinal function deteriorates and their nutritional state is very poor. In order to prevent these situations probiotic therapy is proposed as an effective supporting treatment. Probiotic therapy were therefore applied to infants with severe congenital anomaly as early as possible to ascertain its efficacy.

Methods: As probiotics, two bacteria were used: *Bifidobacterium breve Yakult* and *Lactobacillus casei Shirota*. Probiotic therapy was used in four infants with severe congenital anomaly as early as possible after surgery. Their intestinal microbiota and physical growth were followed through the treatment course.

Results: Two patients suffered from meconium peritonitis with ileal atresia. One patient was born with complex anomalies (omphalocele, bladder exstrophy, myelomeningocele). The fourth patient suffered from complete urorectal septum malformation. The intestinal microbiota of these four patients was first induced to be probiotic dominant and finally changed to commensal anaerobe dominant that was similar to normal intestinal microbiota. Pathogenic bacteria were seldom detected. The patients' physical growth was excellent despite short bowel and pulmonary hypoplasia.

Conclusion: Probiotic therapy was effective in inducing probiotic dominant intestinal microbiota and normal intestinal microbiota in infants with severe congenital anomalies. As a result their intestinal absorptive functions were activated and severe infections were completely prevented. All of the infants grew well despite their physical disadvantages.

Key words congenital anomaly, prebiotics, probiotics.

Recent advances in the field of perinatal care have led to improved survival outcomes for some critically ill neonates. These advances include fetal intensive care (including fetal intervention), neonatal respiratory care, and pre- and post-surgical care. Critically ill neonates, however, need this intensive care for a considerable time after birth; that is, repeated use of antibiotics, prolonged respiratory care and restriction of enteral feeding. These prolonged periods of intensive care can often affect the colonization of the gut by normal intestinal microbiota in neonatal period, and instead can encourage pathogenic microorganisms to become established in the intestine. Such abnormal intestinal microbiota can often cause severe infections and malnutrition, which will again require the affected infants to return to, or prolong their time in intensive care.

We have reported previously the effects of synbiotic therapy (combined use of probiotics and prebiotics)[1] in such critically ill pediatric surgical patients since 2001.[2–5] Synbiotic therapy induced a probiotic-dominant intestinal microbiota and prevented the development of severe infections as well as improving the nutritional state of these severely ill patients.

Here we discuss the preliminary findings of four patients with very severe congenital anomalies who were treated soon after birth using our probiotic therapy. Their intestinal microbiota were well controlled as probiotics became dominant during early infancy and their physical growth was excellent despite abnormally short bowels and prolonged periods of intensive care.

Methods

Patients

Ten pediatric surgical patients were selected, who had been admitted to hospital immediately after birth and who had remained there for more than 1 year. Their fecal microbiota were analyzed before therapy in order to ascertain if long-term intensive care had resulted in abnormal intestinal microbiota.

Four other patients, who had been diagnosed antenatally with congenital anomalies, were treated with our probiotic therapy as soon as possible after birth. Two patients suffered from giant cystic-type meconium peritonitis with intestinal atresia, one had

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the omphalocele, exstrophy of bladder, imperforated anus, spinal defects (OEIS) complex, and the fourth had complete urorectal septum malformation, which is usually considered to be fatal.

Protocol of our probiotic and synbiotic therapy

For probiotic therapy we used two bacteria, *Bifidobacterium breve Yakult* (Yakult Central Institute, Tokyo, Japan) and *Lactobacillus casei Shirota* (Biolactis powder;, Yakult Honsya, Tokyo, Japan). These products include more than 10^9–10^{10} bacteria per 1.0 g pack For prebiotic therapy we used galacto-oligosaccharide (Oligomate; Yakult Honsya, Tokyo, Japan). Probiotics were introduced via a nasogastric tube as soon as intestinal feeding became possible. As a first step, two probiotics were administered at a dose of 0.12 g per day each, in four equal doses. When the amount of milk intake increased, two probiotics were administered at a dose of 3.0 g per day, in three equal doses, and 3.0 g of galacto-oligosaccharides were added in three equal doses.

Quantitative analysis of intestinal microbiota in feces

Fresh fecal samples were harvested in transport tubes and stored at 4°C under anaerobic conditions until bacterial analysis, which was performed at the Yakult Central Institute. Initial processing, subsequent weighing and dilution of the fresh feces were also all carried out under strict anaerobic conditions.[4] VLG (Yakult Central Institute, Tokyo, Japan), MPN (Yakult Central Institute), LBS agar with Laboratory Lemco powder (Yakult Central Institute), DHL agar (Nissui Pharmaceutical, Tokyo, Japan), Sabouraud agar, OPA *Staphylococcus* agar (Japan Becton Dickinson, Tokyo, Japan), Colistin-Oxolinic Acid-Blood agar, and NAC agar (Eiken Chemical, Tokyo, Japan) were used for the selective isolation of *Bacteroides, Bifidobacterium, Lactobacillus, Enterobacteriaceae, Candida,* methicillin-resistant *Staphylococcus aureus* (MRSA), *Enterococcus* and *Streptococcus*, and *Pseudomonas* species, respectively. The details of culture techniques and other media have been described previously.[6,7] The numbers of viable *L. casei Shirota* and *B. breve Yakult* bacteria in the fecal specimens were estimated from the number of colonies formed on the selective media, LLV agar and T-CBPC agar (Yakult Central Institute), respectively,[8,9] and the identities of the bacteria were confirmed on enzyme-linked immunoassay using specific monoclonal antibodies against each probiotic.

Phase classification of intestinal microbiota in pediatric patients

We classified the intestinal microbiota into four colonization phases, based on our previous analyses of the intestinal microbiota of many pediatric surgical patients as well as those of normal healthy infants (Table 1).[10]

Phase I-a is characterized by a microbiota dominated by commensal aerobes. The microbiota of normal infants in the few days after birth show this pattern. During this unstable early phase there is a low risk of intestinal infection, but the microbiota in healthy infants will move on to phase III within 1 month.

Phase I-b is characterized by a microbiota dominated by pathogenic bacteria. Typically, many pediatric surgical patients are in this phase. It is considered to be of high risk to survival because severe infections can develop and there is concomitant deterioration in the absorptive functions of the intestine.

Table 1 Phase classification of intestinal microbiota†

Phase	Dominant biota	Risk
I-a	Commensal aerobes	Low
I-b	Pathogenic aerobes	High
II	Probiotics	Controlled
III	Commensal anaerobes	Safe

†Obtained from analyses of microbiota in pediatric surgical patients and normal children.

Phase II is characterized by a microbiota dominated by probiotics. This is a controlled phase that is maintained by extrinsic probiotics, and one that can be well maintained as long as the synbiotic therapy is continued in the patient while in intensive care.

Phase III is characterized by a microbiota dominated by commensal anaerobes. Most normal healthy infants will acquire this microbiota within 1 month of birth and in this phase the child's gut develops a strong mucosal barrier against attack from pathogenic bacteria. The colons of normal healthy adults typically have a thousand-fold more anaerobes than aerobes.

The clinical courses of the individual patients were analyzed from their clinical records. This study was authorized by the ethics committee of Tokyo University Hospital, and the legal guardians of all patients gave written informed consent.

Results

Intestinal microbiota in 10 pediatric surgical patients without any control treatment of intestinal bacteria

Ten patients were admitted to the hospital immediately after birth, and remained there for >1 year. *Bacteroides* was detected in the feces of two patients (20%) and *Bifidobacterium* in one patient (10%; Table 2). These two bacteria are the most dominant in normal healthy infants (*Bacteroides* was detected in 72.2% and *Bifidobacterium* in 66.7% of 18 normal infants at 1 month of age; data not shown). Conversely *Pseudomonas* was detected in four patients (40%) and *Candida* in seven (70%; *Pseudomonas* and *Candida* were never detected in 18 normal infants at 1 month of age; data not shown). Based on the pre-defined phases (Table 1), five patients were in phase I-a, and the other five were in phase I-b. From this analysis it can be seen that almost all of the 10 present pediatric surgical patients acquired abnormal intestinal microbiota if they were treated intensively in the long term, without therapy to control intestinal bacterial colonization.

Intestinal microbiota and physical growth in four patients who were treated with synbiotic therapy as early as possible

Case 1

This male infant was born in University of Tokyo Hospital at 40 weeks 3 days of gestation with a bodyweight of 2920 g. Fetal ultrasonography indicated that he had giant cystic-type meconium peritonitis. First, we drained the cystic fluid via needle

Table 2 Abnormal intestinal microbiota in 10 critically ill pediatric surgical patients (log10/1.0 g wet feces)

Diagnosis	*Bacteroides*	*Bifidobacterium*	*Lactobacillus*	*Enterobacteriacea*	*Enterococcus*	*Staphylococcus*	*Pseudomonas*	*Candida*
VACTERL association	0	0	5.62	9.78	9.38	4.30	3.34	0
Short bowel syndrome	0	0	0	9.20	7.68	6.05	0	3.08
Giant omphalocele	0	0	0	9.17	0	6.38	0	3.66
Tracheal stenosis	10.04	0	0	9.41	4.08	3.26	7.15	0
Refractory enterocolitis	0	0	5.78	9.76	6.75	3.66	0	0
Short bowel syndrome	0	0	0	0	0	0	0	9.41
Trachoesophageal fistula	0	0	6.70	9.78	9.70	4.88	5.76	7.95
Hirschsprung disease	0	7.66	0	8.24	8.38	4.04	0	4.03
Laryngotracheo esophageal cleft	0	0	0	8.73	10.07	0	0	7.08
Diaphragmatic eventration	10.49	0	0	9.09	9.32	3.62	7.14	6.44

VACTERL, vertebral anomaly, anal anomaly, cardiac disease, tracheo-esophageal fistula, renal anomaly, and limb defects.

puncture, which was retained in the cyst for 25 days after birth. The patient underwent his first operation at 25 days old. A strangulated and severely dilated ileum, which had formed a pseudocyst, was resected and an end-to-end anastomosis of the ileum was performed. MRSA peritonitis developed after surgery and peritoneal drainage was performed; the patient underwent a second operation at 67 days old. Re-anastomosis of the ileum was completed and oral feeding started when he was 76 days old. The length of the small intestine was calculated at the time of the second operation to be approximately 70 cm. Synbiotic therapy was started with milk feeding when he was 76 days old.

Before the initiation of synbiotic therapy, the intestinal bacterial microbiota were very similar to that of an infant aged 1–2 days (phase I-a); no anaerobic bacteria were detected (Table 3). The probiotics became well established in the intestine, however, and his intestinal microbiota was well maintained in a probiotic bacteria-dominant pattern (phase II; Table 3). During the course of treatment, pathogenic organisms such as *Candida*, *Pseudomonas*, and MRSA were detected only rarely (Table 3). The patient's growth was excellent after he started oral feeding and concomitant synbiotic therapy. His bodyweight gain was 23.4 g/day in the first 3 months following the start of synbiotics (Fig. 1a); enteritis did not occur.

Case 2

The mother was transferred to University of Tokyo Hospital because her fetus, at 35 weeks' gestation, suffered from massive ascites and hydrops fetalis. A female infant was delivered by cesarean section. Massive ascites was aspirated and drainage tubes were inserted into the peritoneal space. At the age of 5 days the patient underwent laparotomy, which confirmed ileal atresia with a large pseudocyst. The necrotic section of the intestine was resected and an end-to-end anastomosis of the ileum performed. The length of the small intestine was approximately 75 cm. Postoperative recovery was satisfactory with intensive care. At 12 days old, milk feeding and synbiotic therapy were started.

Analysis of fecal microbiota, just before starting the probiotic treatment, indicated phase I-a; no significant bacteria were detected (Table 4). A probiotic-dominant microbiota (phase II) was maintained for approximately 2 months and then intrinsic *Bifidobacteria* and *Bacteroides* became dominant (phase III; Table 4).

The patient's growth rate was reasonable, with a gain in bodyweight of 32 g/day for the first 3 months (Fig. 1b). Enterocolitis was not experienced, and the patient was discharged at 39 days of age.

Case 3

This patient was diagnosed antenatally with omphalocele and myelomeningocele. At 33 weeks 5 days' gestation she was delivered by cesarean section. There was also lower celosomia and exstrophy of the bladder; the meningocele was covered by normal skin. On the day of her birth the patient underwent a laparotomy. Terminal ileum connected to the exstrophied bladder

Table 3 Intestinal microbiota in patient 1 (log10/1.0 g wet feces)

Species	Age (months)					
	2	3	5	8	16	23
Bacteroides	ND	ND	ND	ND	10.18	9.93
Bifidobacterium sp.	ND	8.90	9.78	7.70	10.08	9.95
Bifidobacterium breve(probiotics)	ND	8.90	9.78	7.26	9.38	8.78
Lactobacillus sp.	ND	8.78	8.24	7.91	9.38	9.45
Lactobacillus casei (probiotics)	ND	8.78	7.64	7.91	7.73	7.60
Enterobacteriaceae	8.56	7.88	8.07	8.90	7.76	6.90
Enterococcus	9.20	9.86	8.78	9.78	7.83	8.16
MRSA	ND	ND	ND	ND	ND	ND
Pseudomonas	ND	ND	3.53	ND	ND	ND
Candida	ND	ND	ND	ND	ND	ND
Phase	I-a		II		III	

MRSA, methicillin-resistant *Staphylococcus aureus*; ND, not detected.

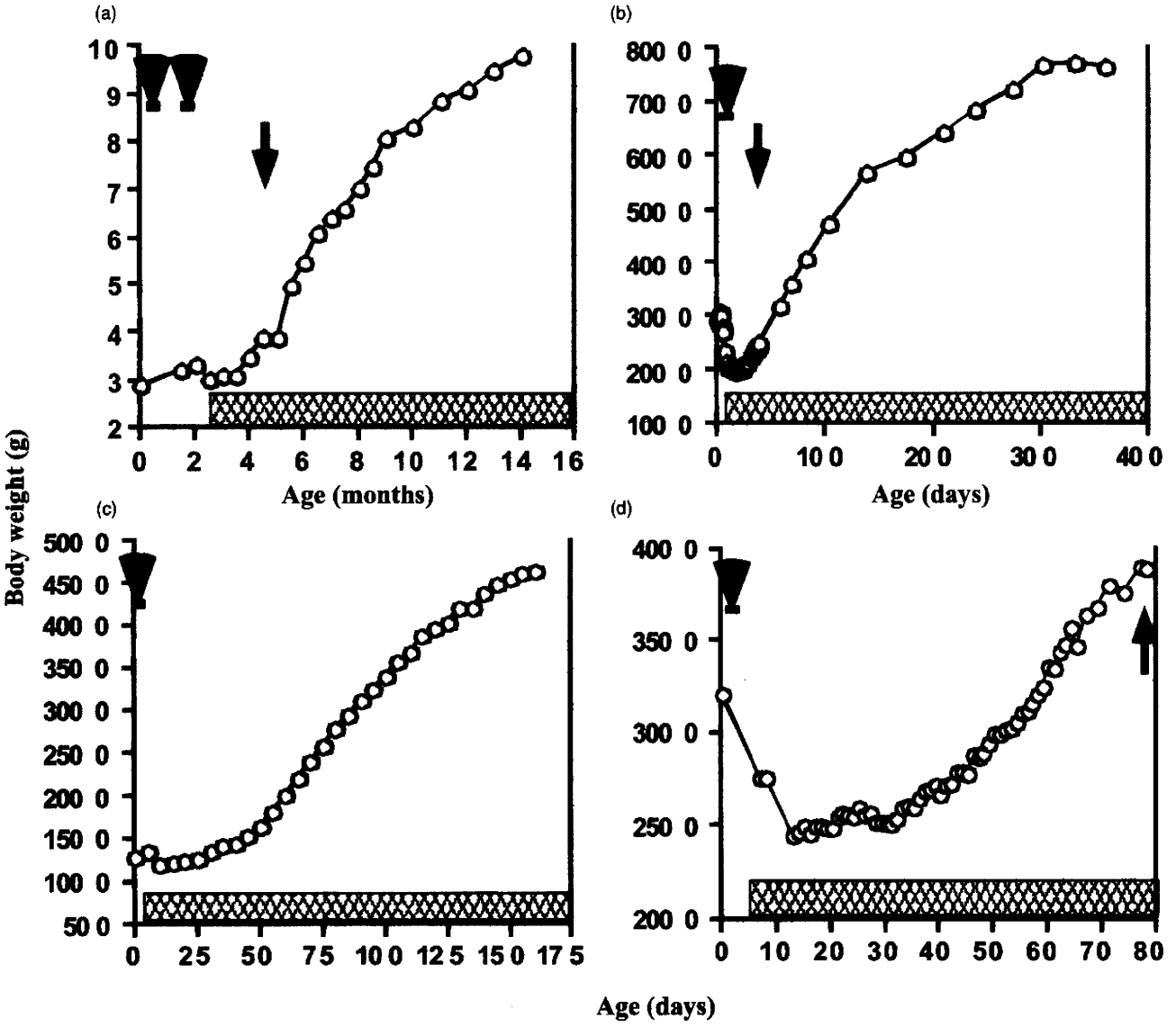

Fig. 1 Physical growth (bodyweight) in four patients with severe congenital anomalies. Arrowheads, surgical treatment; arrows, discharge from hospital. Cross-hatched bars, period of probiotic therapy. (a) The patient underwent surgery twice in the 2 months after birth and probiotic therapy was started at 76 days of age. Bodyweight gain was good after the treatment (23.4 g/day). (b) The patient underwent surgery at 5 days of age and started probiotic therapy at 12 days of age. The physical growth of the patient was good (bodyweight gain: 32.0 g/day) after the treatment. (c) This patient underwent surgery on the day of birth and probiotic therapy was started at 4 days of age. Patient growth was good despite ileostomy (bodyweight gain: 25.6 g/day). (d) This patient underwent surgery at 2 days of age and probiotic therapy was started at 5 days of age. The growth of the patient was good despite short bowel (bodyweight gain: 27.0 g/day).

and short colon was detected caudally. An ileostomy was constructed and the abdominal wall was closed. The patient also had a bilateral hydroureter and hydronephrosis. The postoperative course was uneventful and probiotic therapy was started at 4 days of age. The patient experienced several urinary tract infections, which were treated with antibiotics.

The intestinal microbiota, analyzed just after starting probiotics, were in phase I-a (Table 5). Only probiotic *L. casei* was detected in low numbers (Table 5). During approximately 2 months the probiotic-dominant intestinal microbiota was maintained (phase II) and thereafter intrinsic *Bifidobacteria* and *Bacteroides* became dominant (phase III; Table 5).

The patient's growth was excellent despite the ileostomy, with a bodyweight gain of 25.6 g/day in 3 months (Fig. 1c).

Case 4

This female patient[11] suffered from a complete urorectal septum malformation sequence, which is usually considered to be fatal. She had no urethral opening and suffered from anal atresia without any fistula. While still in the womb this patient under-

Table 4 Intestinal microbiota in patient 2 (log10/1.0 g wet feces)

Species	Age (months)						
	0.5	1.0	2.0	3.0	4.0	6.0	9.0
Bacteroides	ND	ND	ND	3.60	ND	7.85	8.16
Bifidobacterium sp.	ND	9.51	10.42	10.34	10.04	10.52	10.49
Bifidobacterium breve(probiotics)	ND	9.51	10.32	ND	ND	ND	ND
Lactobacillus sp.	ND	7.08	7.86	7.71	6.37	3.52	7.19
Lactobacillus casei (probiotics)	ND	7.08	7.86	7.71	6.37	3.52	7.19
Enterobacteriaceae	ND	8.90	7.92	9.30	7.84	9.23	7.97
Enterococcus	ND	8.18	10.06	9.38	7.10	9.62	9.12
MRSA	ND	ND	ND	ND	ND	ND	ND
Pseudomonas	ND	ND	ND	3.51	ND	ND	ND
Candida	ND	ND	ND	ND	ND	4.03	ND
Phase	I-a	II			III		

MRSA, methicillin-resistant *Staphylococcus aureus*; ND, not detected.

went vesico-amniotic shunting, which successfully saved her renal function, and she was delivered by cesarean section at 35 weeks 2 days. At 2 days of age she underwent a laparotomy, and a colostomy and cutaneo-vesicostomy were constructed. The length of the small intestine was only 60 cm. She suffered occasionally from urinary tract infections after the operation and was treated by antibiotics.

Probiotic therapy was started when the patient was 5 days old. Her intestinal microbiota at the age of 0.5 months showed *Bifidobacteria* including a probiotic strain, and the probiotic-dominant phase II was maintained for approximately 2 months (Table 6). At 4.5 months of age her intestinal microbiota shifted to phase III with intrinsic *Bifidobacteria* and *Bacteroides* (Table 6).

The patient's growth was excellent despite congenital short bowel and there was a gain in bodyweight of 27 g/day in her first 2 months (Fig. 1d).

Discussion

In the present report we have shown for the first time that the intestinal microbiota of severely ill pediatric surgical patients becomes abnormal if no control measures are introduced; pathogenic bacteria become dominant and anaerobes are seldom detected. This abnormal microbiota leads to intestinal dysfunction and results in severe infection and malnutrition.

Recently, progress has been made in fetal and neonatal care and this has meant that the lives of some patients with very severe congenital anomalies can now be saved. Four patients discussed in the present report were typical of such severe anomalies. They often had a short bowel, pulmonary hypoplasia, and renal dysfunction and needed long-term intensive care after birth. As suggested in Table 2, they would inevitably acquire abnormal intestinal microbiota if their intestinal microbiota remained uncontrolled through medical intervention.

The probiotic and synbiotic therapy produced excellent results in encouraging and maintaining a normal intestinal microbiota, with probiotics dominant early in life, and with a commensal anaerobe-dominant microbiota taking over thereafter. With the early application of the probiotic and synbiotic therapy regimen, the bodyweight gain in these four patients was reasonably good and was equivalent to that of normal infants in spite of their congenital short bowels. None of the four patients experienced

Table 5 Intestinal microbiota in patient 3 (log10/1.0 g wet feces)

Species	Age (months)				
	0.5	1.5	2.5	3.5	4.5
Bacteroides	ND	ND	ND	8.19	ND
Bifidobacterium sp.	ND	9.83	9.39	10.41	9.25
Bifidobacterium breve(probiotics)	ND	9.83	4.41	ND	7.03
Lactobacillus sp.	6.31	7.92	8.26	7.81	7.13
Lactobacillus casei (probiotics)	6.31	7.92	8.26	7.81	7.13
Enterobacteriaceae	ND	7.90	6.95	6.64	9.28
Enterococcus	ND	9.45	9.00	8.76	8.44
MRSA	ND	ND	ND	ND	ND
Pseudomonas	ND	ND	ND	ND	ND
Candida	ND	ND	ND	ND	ND
Phase	I-a	II		III	

MRSA, methicillin-resistant *Staphylococcus aureus*; ND, not detected.

Table 6 Intestinal microbiota in patient 4 (log10/1.0 g wet feces)

Species	Age (months)			
	0.5	2.0	4.5	7.0
Bacteroides	ND	ND	7.9	ND
Bifidobacterium sp.	8.66	9.88	9.5	9.2
Bifidobacterium breve(probiotics)	5.61	9.88	ND	7.8
Lactobacillus sp.	4.02	8.92	8.3	8.3
Lactobacillus casei (probiotics)	4.02	5.61	ND	ND
Enterobacteriaceae	ND	8.27	7.70	7.6
Enterococcus	7.11	3.87	8.10	8.4
MRSA	ND	ND	ND	ND
Pseudomonas	ND	ND	ND	ND
Candida	ND	ND	7.3	ND
Phase	II		III	

MRSA, methicillin-resistant *Staphylococcus aureus*; ND, not detected.

enterocolitis and their clinical course was also reasonably good. By keeping the intestinal microbiota normal in early life it was possible to maintain good intestinal absorptive functions in the small intestine, and this was the main contributory factors to the good physical growth of these patients who had been born with severe congenital anomalies. Recently it was demonstrated that commensal anaerobes have the potential to promote lipogenesis in the liver and fat storage in the peripheral fat tissues.[12] We hypothesized that the present probiotics also have a similar metabolic potential, and that such a potential of probiotics and commensals was another contributory factor to the additional good physical growth seen in the present patients at a very young age despite their congenital short bowels.

Another important factor in controlling intestinal microbiota is to establish colonization resistance and prevent enteritis.[13,14] Commensal anaerobes (including probiotics) cover the intestinal mucosa as biofilms and provide an effective barrier that prevents the invasion of pathogenic bacteria (bacterial translocation), by immunological surveillance of the intestinal mucosa.[15] A recent review concluded that probiotics might reduce the risk of necrotizing enterocolitis in preterm neonates born under 33 weeks' gestation.[16] Indeed none of the present four patients suffered from enteritis and tolerated enteral feeding very well, despite their short bowels.

The present results are still preliminary; one limitation of the study was the absence of any control cases. Based on our past experiences, however (Table 2), we propose that our probiotic and synbiotic therapy is a very strong modulator of intestinal microbiota and can induce an anaerobe-dominant environment in the intestine. Therefore, we recommend the use of probiotics in severely ill pediatric surgical patients as early as possible to maintain an anaerobe-dominant intestinal microbiota. We believe that this treatment will lessen the mortality and morbidity of severely ill neonates born with severe congenital anomalies.

References

1 Gibson R, Roberfroid MB. Dietary modulation of the human colonic microbiota: Introducing the concept of prebiotics. *J. Nutr.* 1995; **125**: 1401–12.

2 Kanamori Y, Hashizume K, Sugiyama M *et al.* Combination therapy with Bifidobacterium breve, Lactobacillus casei, and galactooligosaccharides dramatically improved the intestinal function in a girl with short bowel syndrome: A novel synbiotics therapy for intestinal failure. *Dig. Dis. Sci.* 2001; **46**: 2010–16.

3 Kanamori Y, Hashizume K, Sugiyama M *et al.* A novel synbiotic therapy dramatically improved the intestinal function of a pediatric patient with laryngotracheo-esophageal cleft (LTEC) in the intensive care unit. *Clin. Nutr.* 2002; **21**: 527–30.

4 Kanamori Y, Sugiyama M, Hashizume K *et al.* Experience of long-term synbiotic therapy in seven short bowel patients with refractory enterocolitis. *J. Pediatr. Surg.* 2004; **39**: 1686–92.

5 Kanamori Y, Sugiyama M, Komura M *et al.* Synbiotic therapy: An important supportive therapy for pediatric patients with severe respiratory distress. *Int. J. Probiot. Prebiot.* 2006; **1**: 161–8.

6 Holdeman LV, Cato EP, Moore WEC. *Anaerobe Laboratory Manual*, 4th edn. Virginia Polytechnic Institute and State University, Blackburg, 1977.

7 Kanazawa H, Nagino M, Kamiya S *et al.* Synbiotics reduce postoperative infectious complications: A randomized controlled trial in biliary cancer patients undergoing hepatectomy. *Langenbecks Arch. Surg.* 2005; **390**: 104–13.

8 Kado Y, Yuki N, Kushiro A *et al.* Survival of a probiotic, *Bifidobacterium breve* strain Yakult, in the human gastrointestinal tract: Selective isolation from feces and identification using randomly amplified polymorphic DNA polymerase chain reaction technique. *J. Intest. Microbiol.* 2001; **15**: 9–14.

9 Yuki N, Watanabe K, Mike A *et al.* Survival of a probiotic, *Lactobacillus casei* strain Shirota, in the gastrointestinal tract: Selective isolation from faeces and identification using monoclonal antibodies. *Int. J. Food Microbiol.* 1999; **48**: 51–7.

10 Kanamori Y, Sugiyama M, Goishi K *et al.* Abnormal intestinal microbiota in pediatric surgical patients and the effects of a newly designed synbiotic therapy. *Int. J. Probiot. Prebiot.* 2006; **1**: 149–60.

11 Kanamori Y, Iwanaka T, Nakahara S *et al.* Survival in a neonate with complete urorectal malformation sequence after fetal vesicoamniotic shunting for a prominently dilated cloaca. *Fetal Diagn. Ther.* 2008; **24**: 458–61.

12 Backhed F, Ding H, Wang T *et al.* The gut microbiota as an environmental factor that regulates fat storage. *Proc. Natl Acad. Sci. USA* 2004; **101**: 15 718–23.

13 Van der Waaij D, Berghuis-De Vries JM, Lekkerkerk-Van der W. Colonization resistance of the digestive tract in conventional and antibiotic-treated mice. *J. Hyg.* 1971; **69**: 405–11.

14 Madson K, Cornish A, Soper P *et al.* Probiotic bacteria enhance murine and human intestinal epithelial barrier function. *Gastroenterology* 2001; **121**: 580–91.

15 Macpherson AJ, Uhr T. Induction of protective IgA by intestinal dendritic cells carrying commensal bacteria. *Science* 2004; **303**: 1662–5.

16 Deshpande G, Rao S, Patole S. Probiotics for prevention of necrotising enterocolitis in preterm neonates with very low birthweight: a systematic review of randomised controlled trials. *Lancet* 2007; **369**: 1614–20.

Bifidobacterium Septicemia Associated with Postoperative Probiotic Therapy in a Neonate with Omphalocele

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We report the one case of sepsis caused by *Bifidobacterium breve* administered as probiotic therapy. Probiotics can be a potential cause of an invasive disease and should be used with care in vulnerable patients. *(J Pediatr 2010;156:679-81)*

Probiotic therapy has been associated with reduced risk of necrotizing enetrocolitis[1-4] and methicillin-resistant *Staphylococcus aureus* colonization in the intestine of premature infants.[5] This approach has been adopted in neonatal intensive care units in many countries. At our center, we have been administering *Bifidobacterium breve* BBG-01 to all neonates admitted to neonatal intensive care units because of surgical problems or prematurity. The present case is the first in which we have encountered sepsis secondary to *Bifidobacterium breve* BBG-01 probiotic therapy.

Case Report

A fetus was diagnosed with omphalocele at 13 weeks' gestation. A female infant was delivered at 37 weeks and 2 days of gestation by scheduled cesarean delivery. The birth weight was 2060 g; Apgar scores at 1 and 5 minutes were 6 and 7, respectively. The liver and intestine were prolapsed, but the hernia sac was intact. There were no external malformations other than omphalocele and polydactyly of the right hand. The other anatomic examinations and laboratory tests on admission showed no abnormalities.

Four hours after birth, surgery for omphalocele was performed (**Figure 1**). On day 2, administration of *Bifidobacterium breve* BBG-01 was begun. On day 8, a peripheral arterial catheter was removed. On day 10, gastric fluid became bilious, and the laboratory findings were as follows: C-reactive protein, 1.2 mg/dL(normal range: <0.2 mg/dL); white blood cell count 3500/mm^3, with 18% bands and 26% neutrophils. A peripherally inserted central catheter was replaced. Peripheral aerobic and anaerobic blood cultures were obtained; ampicillin/sulbactam and amikacin were initiated empirically and enteral feedings were discontinued. Cerebrospinal fluid cultures were not obtained because the patient was not in critical condition and had no abnormal neurogical signs and symptoms. On day 12, C-reactive protein and white blood cell count increased to 8.2 mg/dL and 9520 /mm^3 (16% bands and 42% neutrophils), respectively. Antibiotics were changed from ampicillin/sulbactam to meropenem. On day 14, the blood cultures obtained on day 10 grew *Bifidobacterium* spp; oral *Bifidobacterium breve* BBG-01 therapy was discontinued. The patient continued to improve, and repeated blood cultures were negative. She recovered without any sequelae or complications. Isolated *Bifidobacterium* spp was susceptible in vitro to penicillin and ampicillin sulbactam (MICs 1 μg/mL) but not to meropenem (MIC >8 μg/mL) or amikacin (MIC ≥32 μg/mL).

The strain detected from the patient was therefore genetically identical to the probiotic *Bifidobacterium breve* BBG-01. Polymerase chain reaction analysis of *Bifidobacterium* spp isolated from the blood cultures showed positive results for *Bifidobacterium*, *Bifidobacterium breve*, and more specifically, *Bifidobacterium breve* BBG-01. Strain-specific identification by a randomly amplified polymorphic DNA analysis using strain-specific primers confirmed *Bifidobacterium breve* BBG-01 (**Figure 2**). The isolates were positive for a monoclonal antibody against *Bifidobacterium breve* BBG-01.

Discussion

"Probiotics" are defined by the Food and Agriculture Organization of the United Nations and the World Health Organization as "live microorganisms, which when administered in adequate amounts, confer a health benefit on the host."[6]

In our center, we use *Bifidobacterium breve* BBG-01 as the probiotic agent. *Bifidobacterium breve* BBG-01 is supplied as a freeze-dried powder in corn starch, containing about 10^9 CFU/g (Yakult Honsya Co Ltd, Tokyo, Japan). Before administering to infants, the nurses add 1 g of the powder in 1.5 mL of sterile water and obtain 0.5 mL of the supernatant after centrifugation in a sterile environment. The supernatant is given 2 times daily. This case report describes invasive disease attributed to *Bifidobacterium* used in a neonate. Although it is uncertain, we suspect that the systemic edema

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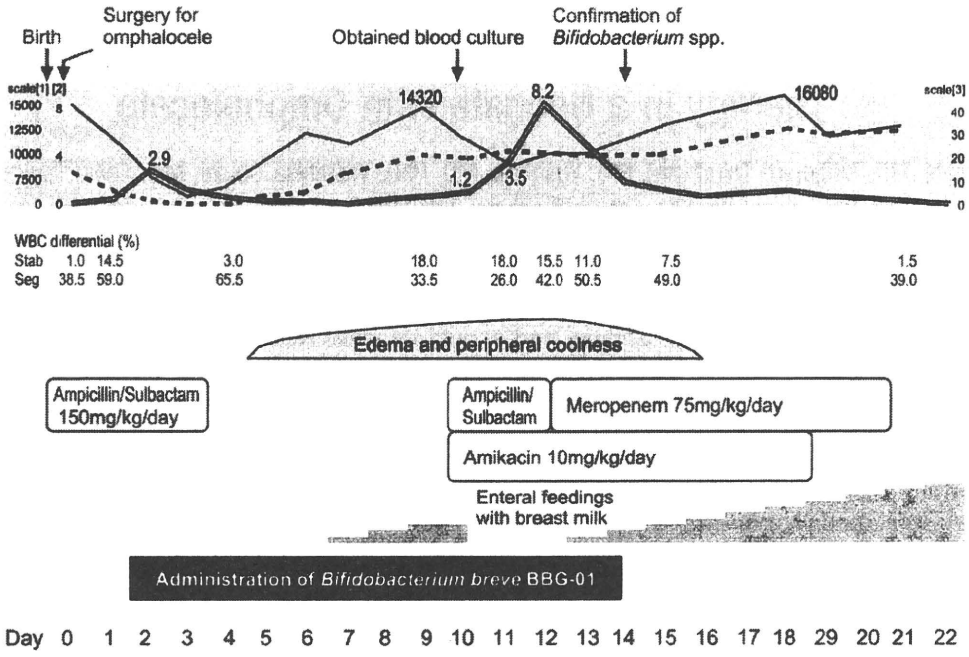

Figure 1. Clinical course in this case of *Bifidobacterium breve* septicemia.

and elevated inflammatory markers resulted from the *Bifidobacterium breve* BBG-01 sepsis for the following reasons: (1) the bacteria were detected from the blood when the increase in inflammatory markers was observed; (2) the intestinal inflammation from the surgical repair could have led to the translocation of *Bifidobacterium breve* BBG-01; and (3) no organisms were detected in other cultures except the blood cultures.

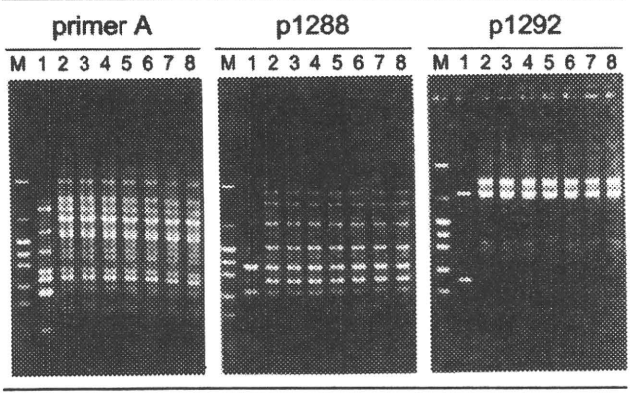

Figure 2. Strain-specific identification of *Bifidobacterium breve* BBG-01 by randomly amplified polymorphic DNA analysis. Three strain-specific primers were used: primer A (-CCGCAGCCAA-), p1288 (-GGGGTTGACC-), and P1292 (-CCCGTCAGCA-). Lane M, pHY DNA size marker; lane 1, other strain of *Bifidobacterium breve*; lanes 2 through 7, *Bifidobacterium* spp detected from the patient; lane 8, *Bifidobacterium breve* BBG-01.

The benefits of probiotics therapy for preterm infants are indicated in many studies; it is shown to reduce the incidence of necrotizing enterocolitis[1-4] and to prevent infections and other diseases.[7] Moreover, contribution of *Bifidobacterium breve* to the intestinal function of premature infants and their prognosis has been suggested.[8] Recently, Kanamori et al[9] reported the beneficial effects of *Bifidobacterium breve* for patients with short bowel syndrome.

Use of probiotic therapy has the fundamental dilemma that patients with the greatest potential benefit from therapy, such as neonates who have immature immune systems or require surgery for gastrointestinal abnormalities, are those who are prone to sepsis from common microorganisms. On the other hand, invasive diseases caused by well-characterized, less pathogenic organisms might not be life-threatening and might be eradicated easily or even be self-limiting.

Some authors have questioned the safety of probiotic therapy. Guarner et al[10] reported that the risk of infection, unrestricted stimulation of the immune system, and gain of antibiotic resistance to virulent microorganisms should be evaluated while administering antibiotic therapy. Some reports demonstrate *Lactobacillus* infection during probiotic therapy.[11] It is also possible that sepsis related to *Bifidobacterium* is under-diagnosed because the anaerobic microorganism is undetected under regular aerobic conditions. In this case, the culture became positive within 40 hours of incubation. Subculture onto several agar plates yielded only a few colonies on an ABHK agar plate (Nissui Pharmatheuticals, Tokyo, Japan) incubated under anaerobic conditions.

Although the clinical course of this case was not life-threatening, one should be aware that the introduction of a new

living microorganism as a therapeutic agent can be harmful, especially in patients who are predisposed to invasive disease. Further studies should aim for better understanding of the appropriate method, strains or combination of strains administered, dosage, and period of administration. Moreover, it is necessary to demonstrate whether the method is cost-effective and safe when performed in neonates.

This report should serve as a reminder that probiotic agents have potential benefits and risks of invasive disease under certain physiological conditions. ■

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References

1. Lin HC, Hsu CH, Chen HL, Chung MY, Hsu JF, Lien RI. Oral probiotics prevent necrotizing enterocolitis in very low birth weight preterm infants: a multicenter, randomized, controlled trial. Pediatrics 2008;122: 693-700.
2. Bin-Nun A, Bromiker R, Wilschanski M, Kaplan M, Rudensky B, Caplan M, et al. Oral probiotics prevent necrotizing enterocolitis in very low birth weight neonates. J Pediatr 2005;147:192-6.
3. Deshpande G, Rao S, Patole S. Probiotics for prevention of necrotising enterocolitis in preterm neonates with very low birthweight: a systematic review of randomised controlled trials. Lancet 2007;369:1614-20.
4. Alfaleh K, Bassler D. Probiotics for prevention of necrotizing enterocolitis in preterm infants. Obstet Gynecol 2008;111:1202-4.
5. Kanamori Y, Hashizume K, Kitano Y, Tanaka Y, Morotomi M, Yuki N, et al. Anaerobic dominant flora was reconstructed by synbiotics in an infant with MRSA enteritis. Pediatr Int 2003;45:359-62.
6. Joint FAO/WHO Working Group meetings. Guidelines for the evaluation of probiotics in food. London, Ontario, Canada, 2002.
7. Kopp-Hoolihan L. Prophylactic and therapeutic uses of probiotics: a review. J Am Dietetic Assoc 2001;101:229-41.
8. Kitajima H, Sumida Y, Tanaka R. Norikatsu Yuki, Hiroo Takayama, Masanori Fujimura. Early administration of *Bifidobacterium breve* to preterm infants: randomized controlled trial. Arch Dis Child 1997;76:F101-7.
9. Kanamori Y, Hashizue K, Sugiyama M, Morotomi M, Yuki N. Combination therapy with *Bifidobacterium breve, Lactobacillus casei*, and galactooligosaccharides dramatically improved the intestinal function in a girl with short bowel syndrome: a novel synbiotics therapy for intestinal failure. Dig Dis Sci 2001;46:2010-6.
10. Guarner F, Schaafsma GJ. Probiotics. Int J Food Microbiol 1998;39:237-8.
11. Kunz AN, Noel JM, Fairchok MP. Two cases of *Lactobacillus* bacteremia during probiotic treatment of short gut syndrome. J Pediatr Gastroenterol Nutr 2004;38:457-8.

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ORIGINAL ARTICLE

CT and MR imaging for pediatric cochlear implantation: emphasis on the relationship between the cochlear nerve canal and the cochlear nerve

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Abstract
Background Cochlear implantation has become an accepted treatment for deafness. As the frequency of cochlear implantation has increased, requests for images have also increased in the work-up for candidates. An absent cochlear nerve (CN) is a contraindication to cochlear implantation. Therefore, MRI is performed to evaluate the CN in patients with sensorineural hearing loss. Recently, some authors have reported the relationship between cochlear nerve canal (CNC) stenosis and CN hypoplasia.
Objective To review the relationship between CNC and CN.
Materials and methods During a period of 78 months, 21 children (42 ears) with unilateral or bilateral sensorineural hearing loss underwent both HRCT and MRI of the cochlear nerve. We retrospectively reviewed two factors: the evaluation of inner ear malformations and the relationship between CNC stenosis and CN hypoplasia.
Results Inner ear malformations were recognized in ten ears. The mean CNC diameter was approximately 2 mm (ranging from 0.6 to 2.7 mm). CN hypoplasia was seen in eight of the 42 ears; all eight were associated with CNC stenosis (≤1.5 mm). Of the 34 ears with normal CN, 32 had CNC >1.5 mm in diameter and the remaining two ears, with incomplete partition type I, had CNC stenosis.
Conclusion Children with CNC stenosis had a high incidence of CN hypoplasia. CNC stenosis (≤1.5 mm) suggests CN hypoplasia. On the other hand, CN hypoplasia was not seen in children with CNC diameter >1.5 mm. Therefore, we conclude that children with CNC stenosis or malformations on HRCT should receive MR imaging of the CN.

Keywords Cochlear implantation · Cochlear nerve · Cochlear nerve canal · Children

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Introduction

Hearing loss is one of the most common birth defects, affecting 3–4 of every 1,000 newborns [1]. A variety of pathologic conditions cause hearing loss in children [2]. With the advent of universal newborn screening, deafness can be diagnosed earlier in life [3].

Cochlear implantation has become an accepted treatment for deafness. As the frequency of cochlear implantation has increased, requests for imaging have also increased in the work-up of cochlear implant candidates. Radiologists must be familiar with imaging findings that contraindicate implantation (cochlear aplasia and cochlear nerve aplasia) [4]. High-resolution CT (HRCT) of the temporal bone has been used to evaluate mastoid air cell aeration, facial nerve position, and inner ear malformations. On the other hand, MRI is better to visualize the cochlear nerve and intracranial structure directly . Therefore, high-resolution MRI has been used to detect CN aplasia or hypoplasia in children with sensorineural hearing loss (SNHL) [1, 3–6]. Recently, some authors have reported a relationship between cochlear nerve canal stenosis and CN hypoplasia [7–9]. Those findings have important implications for clinicians who evaluate children with SNHL. CNC stenosis might be a key

finding to select children with SNHL who should undergo further evaluation with MRI.

The purposes of this study were to evaluate a variety of pathological conditions for children with SNHL and to review the relationship between CNC and CN.

Materials and methods

This study was reviewed and approved by the ethics committee of our hospital.

During a period of 78 months, 21 children (42 ears) with unilateral or bilateral SNHL underwent both high-resolution CT of the temporal bone and MRI of the cochlear nerve (Table 1). These 21 children included eight boys and 13 girls, with a mean age of 7 years (ranging from 1 to 13 years) at the time of the MRI. Of these, 11 children had unilateral and ten children had bilateral SNHL (31 ears with SNHL and 11 without). Cochlear implantation was performed in three children who had bilateral SNHL and a normal CN.

The protocol of HRCT of the temporal bone was performed with a multidetector-row CT scanner (8-detector,

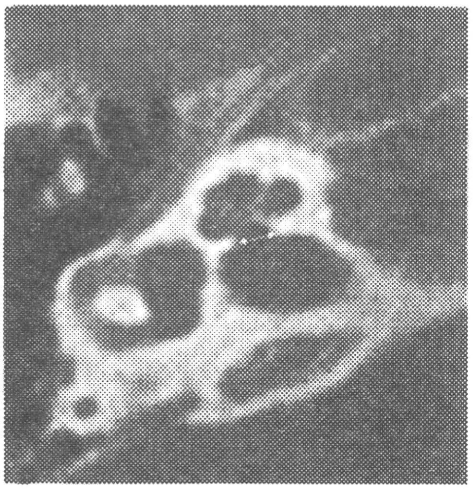

Fig. 1 Measurement of CNC on axial image of temporal HRCT. The CNC is the bony canal from the fundus of the IAC to the base of the modiolus. The diameter of the CNC width (*line*) was measured along the inner margin of its bony walls at its middle portion on an axial image of the base of the modiolus

Table 1 Summary of CT and MRI findings in children with SNHL

Case no.	Age (years) /sex	Side of SNHL	Inner ear malformations		CNC diameter (mm)		CN on MRI	
			Right	Left	Right	Left	Right	Left
1	8/F	Right	Michel deformity	No	–	2	Absent	Normal
2	2/F	Bilateral	Cochlear aplasia	Cochlear aplasia	–	–	Absent	Absent
3	10/F	Left	IP-I	IP-I	1.2	1.0	Normal	Normal
4	11/M	Bilateral	Large vestibular aqueduct syndrome	Large vestibular aqueduct syndrome	2.0	2.0	Normal	Normal
5	13/F	Right	Duplication of IAC	No	0.68	2	Absent	Normal
6	6/F	Bilateral	Large vestibular aqueduct syndrome	Large vestibular aqueduct syndrome	2.1	2	Normal	Normal
7	5/F	Bilateral	No	No	2	1.9	Normal	Normal
8	1/M	Right	No	No	1.5	2	Absent	Normal
9	2/F	Bilateral	No	No	2.2	2.1	Normal	Normal
10	6/F	Right	No	No	2.2	2.1	Normal	Normal
11	8/M	Bilateral	No	No	2.4	2.4	Normal	Normal
12	10/M	Left	No	No	2.4	2.5	Normal	Normal
13	5/F	Bilateral	No	No	2.1	2.1	Normal	Normal
14	3/F	Bilateral	No	No	2.5	2.2	Normal	Normal
15	5/M	Left	No	No	1.9	–	Normal	Hypoplasia
16	7/F	Right	No	No	0.59	1.8	Hypoplasia	Normal
17	7/M	Right	No	No	2.2	1.8	Normal	Normal
18	13/M	Right	No	No	2.5	2.7	Normal	Normal
19	13/F	Bilateral	No	No	2.4	2.6	Normal	Normal
20	7/F	Right	No	No	1.5	2.4	Hypoplasia	Normal
21	3/M	Bilateral	No	No	2.1	2.2	Normal	Normal

SNHL sensorineural hearing loss, *CNC* cochlear nerve canal, *CN* cochlear nerve, *M* male, *F* female, *No* no abnormality, –not evaluated, *IP-I* Incomplete partition type I

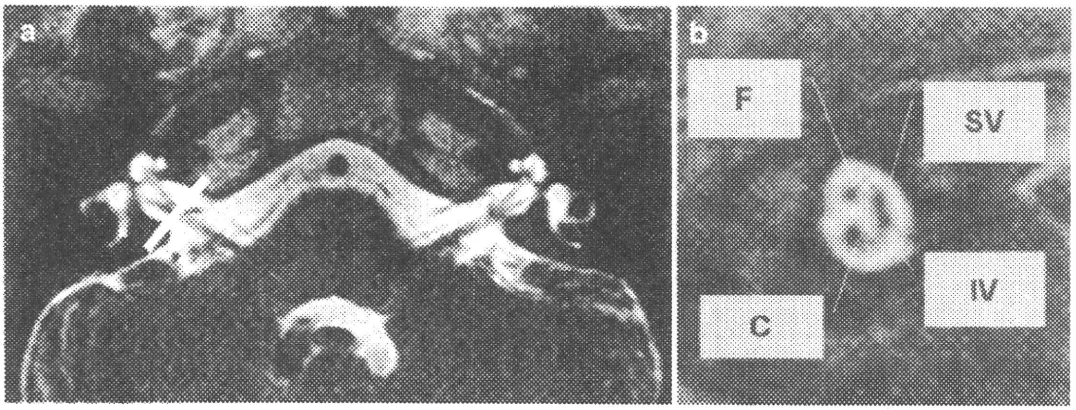

Fig. 2 Axial and oblique sagittal image of normal IAC on DRIVE. **a** Axial image of the cerebellopontine region and IAC shows the normal anatomy. Line illustrates the plane prescribed for oblique plane sagittal images obtained perpendicular to the nerves of the IAC. **b** Oblique sagittal image obtained at the fundus of the IAC, oriented with anterior to the left and the cerebellum to the right. Four nerves were recognized. *F:* Facial nerve, *C:* Cochlear nerve, *SV:* Superior vestibular nerve, *IV:* Inferior vestibular nerve

LightSpeed Ultra, GE, Milwaukee, WI, USA). Images were acquired in the direct axial planes using a 0.625-mm slice thickness, a field of view (FOV) of 96 mm, 140 kV, 150 mA, 1.25 pitch, and a bone algorithm. CT was performed with the minimal radiation dose, according to the ALARA (as low as reasonably achievable) concept. The radiation dose ranged from 35.55 mGy to 44.44 mGy for CT dose index volume ($CTDI_{vol}$). We are now using low kV and mA to lower the $CTDI_{vol}$. Coronal multi-planar reconstruction images were obtained, as well. The CNC is a bony canal extending from the fundus of the inner auditory canal (IAC) to the base of the modiolus. The diameter of the CNC was measured along the inner margin of its bony walls at its middle portion on an axial image of the base of the modiolus (Fig. 1).

All MR images were obtained using a 1.5-T or 1.0-T magnet (Intera 1.5T and 1.0T; Philips, The Netherlands). The children were studied in the supine position with a head coil. The MRI scan sequences included a 3-D T2-weighted fast spin-echo sequence (using DRIVE: driven equilibrium) in axial and oblique sagittal planes of the IAC with a 0.7-mm slice thickness and an FOV of 130 mm. Oblique sagittal images were obtained perpendicular to the course of the nerves through the IAC (Fig. 2). The scan time was approximately 30 min. In oblique sagittal images of the lateral aspect of the IAC, four nerves were recognized (facial, superior vestibular, inferior vestibular and cochlear).

We retrospectively reviewed the following factors: (1) the evaluation of the inner ear malformations on HRCT, (2) the CNC measurements on HRCT, (3) the presence of CN hypoplasia or aplasia on MRI, (4) the relationship between CNC stenosis and CN hypoplasia or aplasia. The cochleovestibular malformations were listed according to the new classification system for inner ear malformations (Table 2), published in 2002 by Sennaroglu and Saatci [10]. As Kim et al. [11] reported, the CN is larger than either the superior or inferior vestibular nerve in 90% of normal ears and is of similar size or larger than the facial nerve in 65%. Therefore, we designated the CN as hypoplastic when it appeared smaller than the facial nerve and as aplastic when it could not be identified on oblique sagittal images.

Results

Evaluation of inner ear malformations on HRCT

Of the 42 ears, cochlear malformations were recognized in ten ears (six children). The details of the inner ear malformations were as follows: Michel deformity in one ear, cochlear aplasia in two ears, incomplete partition type I (IP-I) in two ears (Fig. 3), large vestibular aqueduct syndrome in four ears, and duplication of IAC in one ear (Fig. 4) [12–15].

CNC measurements on HRCT

Of the 42 ears, four ears without a CNC could be evaluated. These four ears had Michel deformity (one ear), cochlear

Table 2 A new classification for inner ear malformations [10]

-Michel deformity
-Cochlear aplasia
-Common cavity deformity
-Cochlear hypoplasia
-Incomplete partition type I (IP-I)
-Cochlear hypoplasia
-Incomplete partition type II (IP-II) = Mondini deformity

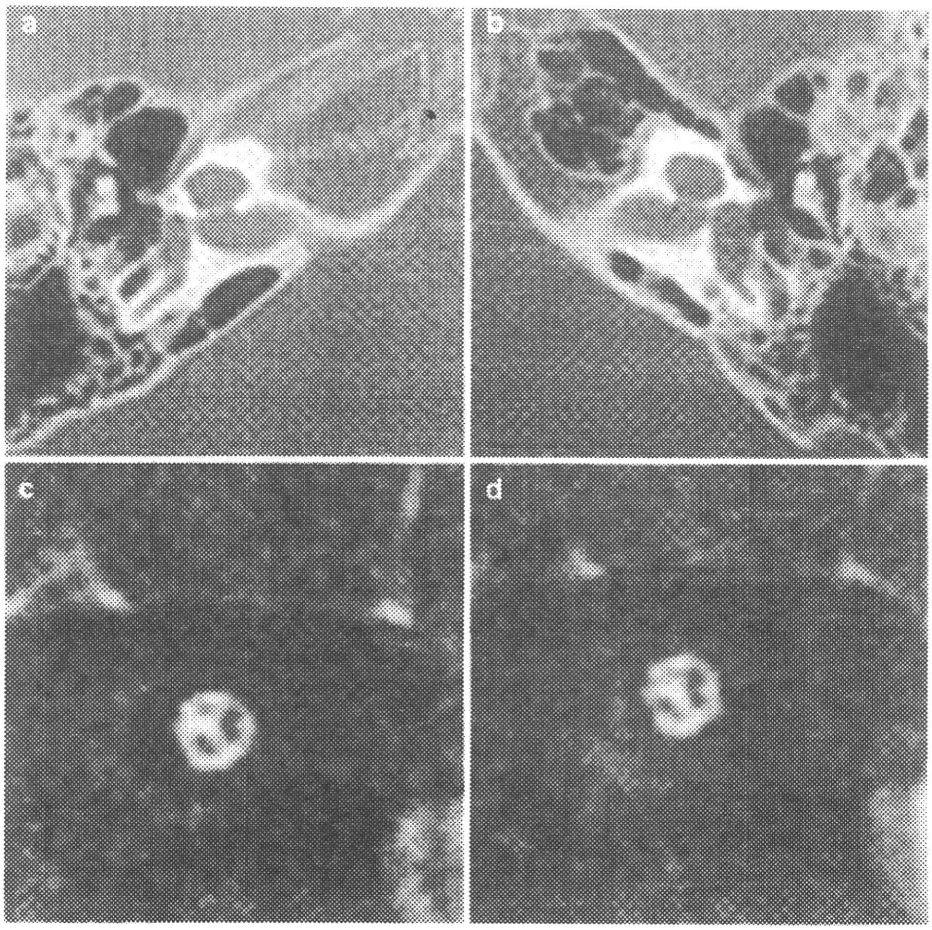

Fig. 3 IP-I in a 10-year-old girl with left SNHL (case 3). **a, b** Axial images show cystic cochlea and the lack of modiolus, which is diagnosed as IP-I. The narrowing of the CNC is also seen. **c, d** Oblique sagittal DRIVE images show normal CN, bilaterally

aplasia (two ears) and a closed CNC without cochlear dysplasia (one ear).

The CNC was measurable in the remaining 38 ears. The CNC diameters ranged from 0.6 mm to 2.7 mm in 27 ears with SNHL and from 1.2 mm to 2.7 mm in 11 ears without SNHL. The mean CNC diameter was 2 mm in each ear, with or without SNHL.

Presence of CN hypoplasia or aplasia on MRI

The CN of all ears could be evaluated on MRI. CN hypoplasia and aplasia were seen in three and five ears, respectively. These corresponded to the affected side. The remaining 23 ears with SNHL and 11 ears without SNHL had a normal CN.

The relationship between CNC stenosis and CN hypoplasia/aplasia (Table 3)

All eight ears with CN hypoplasia or aplasia were associated with CNC stenosis (CNC diameter ≤1.5 mm; mean, 1.1 mm). The remaining 34 ears had a normal CN. Of these, 32 ears had normal CN and CNC diameters >1.5 mm (mean, 2.2 mm). Despite CNC stenosis (<1.5 mm), normal CN was seen in two ears with IP-I. A CNC with CN hypoplasia or aplasia was smaller than those with a normal CN.

Of the eight ears with CN hypoplasia or aplasia, four had inner ear malformations, as follows: one with Michel deformity, two with cochlear aplasia, and one with duplication of the IAC. The remaining four ears with CN hypoplasia or aplasia failed to show inner ear malformation (Fig. 5). On the other hand, four ears with large vestibular aqueduct syndrome and two ears with IP-I did not have CN hypoplasia, and three ears had CN hypoplasia without inner ear malformations. CN aplasia without inner ear malformation was seen in one ear.

Discussion

A variety of pathologic conditions cause SNHL in infants and children [2]. Recent studies suggest that cochlear nerve dysfunction accounts for up to 10% of newly diagnosed