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Original Article

Analysis of the clinical features of Japanese patients with primary ciliary dyskinesia

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ABSTRACT

Objective: Primary ciliary dyskinesia (PCD) is a rare hereditary disease. Most reports of PCD in Japan are case reports, and clinical analysis has not been performed. Differences in the causative genes might affect the clinical features in different ethnic groups. The purpose of this study was to clarify the clinical features of Japanese patients with PCD.

Methods: We performed a retrospective chart review of PCD patients seen at Mie University Hospital and patients whose blood samples were sent to us for genetic analysis from 2011 to 2020. Data on the following items were collected and analyzed: age at first visit to the hospital, age at diagnosis of PCD, process of referral to our facility, chief complaint, situs status, PrImary CiliARy DyskinesiA Rule (PICADAR) score, nasal nitric oxide concentration, otoscopic findings, rhinoscopic findings, and paranasal computed tomography scan findings.

Results: Sixty-seven patients (24 male, 43 female) were diagnosed with PCD during the study period. Age at diagnosis ranged from 2 months to 69 years (median, 17 years). Respiratory symptoms (77%) were the most common complaint, followed by nasal (15%) and aural (8%) symptoms. *Situs inversus* was present in 17 (25%) cases. Only 2 cases had congenital cardiac anomalies. The mean PICADAR score was 7.3 (range, 3–14) points. Approximately 50% of tympanic membranes showed retraction, suggesting otitis media with effusion. The mean Lund-Mackay score was 12.8 (range, 7–17) points, suggesting that the radiographic findings were not always severe. There was no significant difference in the total Lund-Mackay score between patients with and without situs inversus (12.7 vs. 12.6, respectively).

Conclusion: Situs inversus was present in 25% of Japanese PCD patients, which is much lower than observed in other countries. This is a result of differences in the major disease-causing

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genes. The general rule that "situs inversus is observed in approximately 50% of PCD patients" cannot be applied, at least, in Japanese PCD patients.

1. Introduction

Primary ciliary dyskinesia (PCD) is a congenital disease caused by genetic variants related to cilia. PCD occurs in approximately 1 in 20,000 people [1] and is inherited in an autosomal recessive or X-linked manner [2,3]. More than 40 causative genes have been identified to date [4]. The diagnosis of PCD requires the presence of characteristic clinical features such as chronic rhinosinusitis, otitis media with effusion, and bronchiectasis. The paucity of reports on PCD in Japan [5–8] may be due to low awareness of the disease among Japanese physicians; thus, it is probable that PCD is underdiagnosed in Japan.

Disease-causing variants differ markedly among white European (52% of families carry *DNAH5* or *DNAH11* variants), Arab (42% of families carry *CCDC39* or *CCDC40* variants), and South Asian (36% of families carry single *LRRC6* or *CCDC103* variants) patients with PCD [9]. Recently, it has become apparent that *DRC1* is the major cause of PCD in Japanese patients [10–12]. Differences in the causative genes might affect the clinical features in different ethnic groups.

Since most of the reports on PCD in Japan have been case reports [13], little is known about the precise clinical characteristics of the disease, including the prevalence of situs inversus and the severity and features of chronic rhinosinusitis. In this report, we examined clinical signs, symptoms, and imaging findings in order to characterize the clinical features of PCD in the Japanese population.

2. Methods

We retrospectively examined PCD patients seen at Mie University Hospital and PCD patients whose peripheral blood samples and medical histories had been sent to us for analysis, in the period from 2011 to 2020. The patients were diagnosed with PCD according to clinical symptoms, genetic variants, and/or apparent abnormalities on electron microscopy [14].

The diagnosis of PCD requires the presence of characteristic clinical features such as chronic rhinosinusitis, otitis media with effusion, bronchiectasis, and either (1) specific ciliary ultrastructural defects [14] identified by transmission electron microscopy in biopsy samples of the respiratory epithelium and/or (2) biallelic variations of one of the genes known to be associated with PCD [4]. We diagnosed PCD according to these criteria. Situs status was examined either by chest X-ray or chest computed tomography (CT) scan.

For genetic analysis, genomic DNA was extracted from peripheral blood samples taken from the forearm of each participant and we utilized our next-generation sequencing panel of 32 PCD genes [8]. The variants identified by the above panel were validated via Sanger sequencing with a 3500 Series Genetic Analyzer (Thermo Fisher Scientific, Inc., Waltham, MA), as described previously [8]. Genetic and clinical analyses were approved by the Ethics Committee of Mie University (nos. 1363 and 2285, respectively), and written informed consent was obtained from each patient and/or their parent.

Nasal nitric oxide (NO) levels were measured at the National Hospital Organization Mie National Hospital using an ANALYZER CLD 88® (ECO MEDICS AG, Dürnten, Germany) according to the recommendations of the American Thoracic Society/European Respiratory Society [15]. In principle, patients are referred to our hospital with an NO concentration of ≤250 ppb (≤82.5 nL/min) [16].

For electron microscopy studies, a small amount of nasal mucosa was taken from the nasal cavity using a cytology brush. According to the methodology of Rubin [17], >20 cilia from each patient were examined by electron microscopy (JEM–1011; JEOL, Tokyo, Japan).

Information on the following items was collected and analyzed:

- Age at first visit to our hospital and age at diagnosis of PCD
- Process of referral to our facility
- Chief complaint
- Presence or absence of situs inversus
- PrImary CiliARy DyskinesiA Rule (PICADAR) score [18]
- Nasal NO concentration
- Otoscopic findings
- Rhinoscopic and paranasal CT scan findings

Univariate analysis with the Mann–Whitney *U* test was performed to determine significant differences between two groups. Analyses were performed with SPSS statistical software version 21 (IBM, Chicago, IL). A p-value < 0.05 was considered statistically significant.

3. Results

Sixty-seven patients (24 male, 43 female) were diagnosed with PCD during the study period. Fig. 1 shows the distribution of age at first visit to our hospital and age at PCD diagnosis. Age at first visit ranged from 1 month to 66 years (median, 17 years), with 75% of the patients visiting when they were ≤25 years old. Age at diagnosis ranged from 2 months to 69 years (median, 17 years). Age at first visit and age at diagnosis were not significantly different between patients with and without situs inversus.

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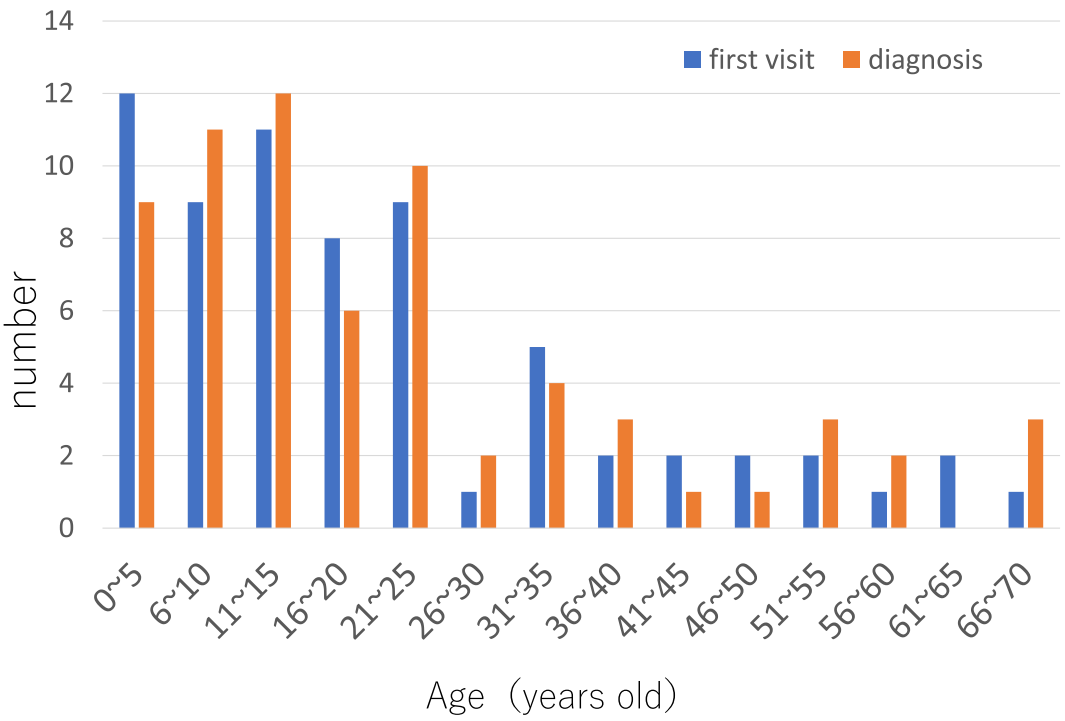

Fig. 1. Distribution of age at first visit to our hospital and age at PCD diagnosis. Age at first visit ranged from 1 month to 66 years (median, 17 years); 75% of patients visited when they were ≤25 years. Age at diagnosis of PCD ranged from 2 months to 69 years (median, 17 years). PCD, primary ciliary dyskinesia.

Forty-five patients visited our hospital and underwent electron microscopy studies and/or genetic tests. Nasal NO concentration was evaluated in 23 of these 45 patients prior to visiting our hospital. For patients who lived far from our hospital (n = 22), blood samples were sent from other hospitals to our facility. On electron microscopy, class 1 or 2 defects [14], which suggest PCD, were found in 26 patients.

Genetic tests revealed biallelic variants in PCD-related genes in 59 cases from 50 families. Twenty-five patients from 20 families had a biallelic deletion of *DRC1*, and 21 patients from 18 families had biallelic *DNAH5* variants. Six patients from 6 families had *DNAH11* variants, and 2 patients in 1 family had *DNAI1* variants. We divided the patients into two groups: those with *DRC1* variants and those with non-*DRC1* variants and examined if there were differences in the clinical features between the two groups. The mean PICADAR score was not significantly different between the *DRC1* and non-*DRC1* groups (6.0 and 8.0, respectively). There was also no significant difference in the Lund-Mackay score between the groups.

The chief complaint was confirmed in 48 cases (Fig. 2). Respiratory symptoms were the most common complaint (37/48, 77%), including chronic wet cough (>3 months) (n = 35), wheezing (n = 1), and sputum (n = 1). Nasal symptoms were the next most common chief complaint (7/48, 15%), including nasal obstruction (n = 5), postnasal drip (n = 1), and rhinorrhea (n = 1). Aural symptoms were the chief complaint in 4 patients (8%), including otorrhea (n = 1), sense of discomfort in the ear (n = 1), ear fullness (n = 1), and recurrent otitis media (n = 1). Eighteen pediatric patients were referred to our hospital because of neonatal respiratory distress; chief complaints were not evaluated in these patients.

Situs inversus was present in 17 of 67 cases (25%). None of the 25 patients with *DRC1* variants had situs inversus. Of the 17 patients with situs inversus, 12 had *DNAH5* variants and 2 had *DNAH11* variants. Genetic analysis was not performed in the remaining 3 patients. Only 2 cases had congenital cardiac anomalies, with corrected transposition of the great arteries in 1 case and complete transposition of the great arteries (type II), double outlet right ventricle, large ventricular septal defect, and patent ductus arteriosus in the other.

PICADAR is a simple diagnostic prediction tool [18] for PCD with good accuracy and validity. The PICADAR score is calculated on the premise that there is wet cough, and the following seven clinical items are scored to predict the possibility of PCD: (1) full-term birth, (2) neonatal respiratory symptoms, (3) neonatal intensive care unit (NICU) admission, (4) misalignment of organs including situs inversus, (5) congenital heart disease, (6) persistent rhinitis, and (7) otitis media effusion, deafness, or perforated eardrum. Two points are assigned to (1), (2), (3), and (5), 4 points to (4), and 1 point to (6) and (7). The PICADAR score could be evaluated in 36 cases and ranged from 3 to 14 (mean, 7.3) points (Fig. 3A). The scores were higher in patients with situs inversus than in those without (mean, 10.2 vs. 5.8, respectively, $p < 0.001$, Fig. 3B). The proportion of cases positive for each of the seven PICADAR score items is shown in Fig. 3C. Most of the patients were born at term and had rhinitis, while only 2 cases had a cardiac anomaly; 75% had otitis media, 66.7% had neonatal respiratory distress, and 50% were admitted to an NICU. Situs inversus was detected in 33.3% of the 36

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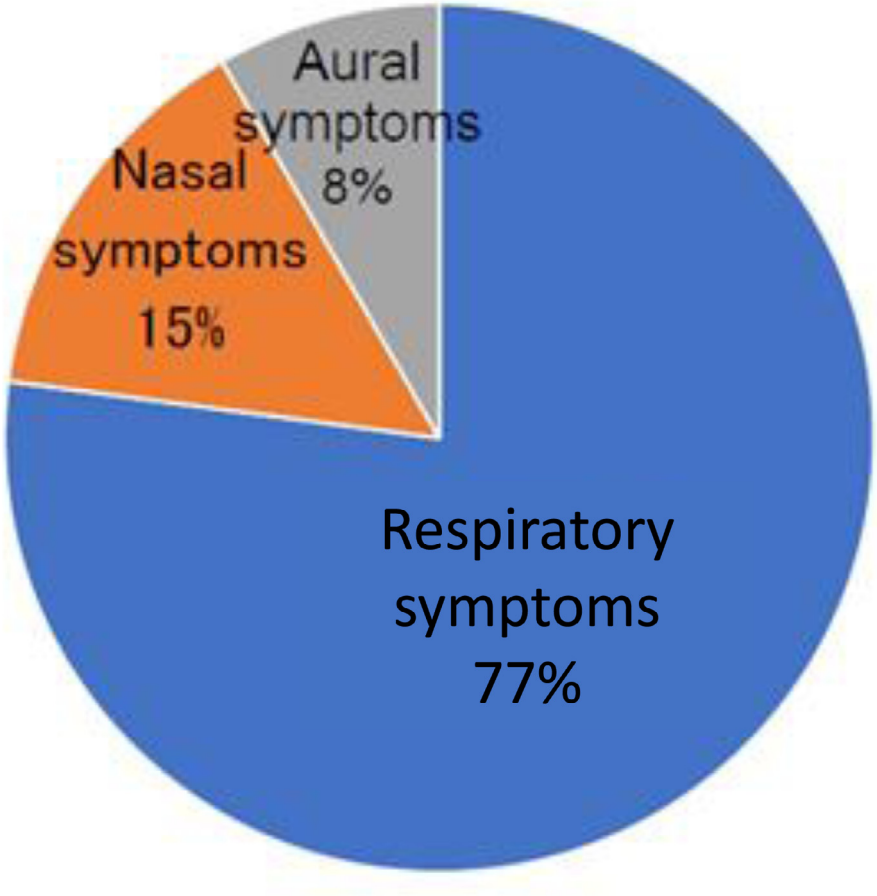

Fig. 2. The chief complaint was confirmed in 48 cases. The most common complaints were related to respiratory systems (77%; 35 cases of wet cough and 1 case each of wheezing and sputum). Nasal symptoms accounted for 15% of the most common complaint (5 cases of nasal obstruction and 1 case each of postnasal drip and rhinorrhea, and aural symptoms accounted for 8% (1 case each of otorrhea, sense of discomfort of the ear, ear fullness, and repeated otitis media).

patients, which was compatible with the percentage of situs inversus (25%) in all patients. A breakdown of the PICADAR scores is shown in Table 1 for each case, presented from the highest to the lowest score. Only 7 patients (19%) were born at term and had rhinitis and/or otitis media.

Nasal NO concentration was measured in 23 patients. Nasal NO concentration ranged from 0.2 ppb (0.066 nL/min) to 650.9 ppb (214.8 nL/min), with a mean of 85.5 ppb (28.3 nL/min). Nasal NO concentration was not significantly different between patients with and without situs inversus.

Otoscopic findings were obtained in 33 cases (Fig. 4). Fifty percent of the tympanic membranes showed retraction, suggesting otitis media with effusion (Fig. 4A). Perforations and ventilation tubes can be regarded as sequelae of otitis media with effusion (Fig. 4A). Eardrums were normal in 33% of patients (Fig. 4A). Various otoscopic findings were observed, including a normal membrane (Fig. 4B), retraction (Fig. 4C), perforation and calcification (Fig. 4D), and ventilation tube insertion (Fig. 4E).

Nasal endoscopic examinations were performed in 34 cases (Fig. 5). Nasal polyps were observed in 12% of patients (Fig. 5A). The rhinoscopic findings of 3 patients with (Fig. 5B, C, D, E) and without (Fig. 5F, G) nasal polyps are presented. Nasal polyps were observed only in adult patients aged $\geq$36 years.

Paranasal sinus CT findings were evaluated in 16 cases using the Lund-Mackay score [19]. This scoring system evaluates opacification of the maxillary, anterior ethmoidal, posterior ethmoidal, sphenoid, and frontal sinuses on a 3-point scale of 0 to 2. In addition, the ostiomeatal complex obstruction is evaluated on a scale of 0 or 2 points, and these scores are totaled separately for right and left to evaluate sinus shadow on a scale of 0 to 12 points. The scores for the ostiomeatal complex were higher than those of the individual sinuses (Fig. 6A). The mean bilateral total score was 12.8 points (Fig. 6B), and the mean score was 6.4 points on both the left and right (Fig. 6C). There was no significant difference in the total Lund-Mackay score between patients with and without situs inversus (12.7 vs. 12.6, respectively).

The frontal and sphenoid sinuses are reported to be hypoplastic in PCD [20]. In the present study, however, we found lower proportions of patients with hypoplasia in these sinuses than previously reported [20]: in the 16 cases evaluated with CT, the frontal sinuses were hypoplastic on the right and left in 31% and 19% of patients, respectively, and the sphenoid sinuses were hypoplastic in only 13% of pa-

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Table 1. Breakdown of the PICADAR scores of the 36 patients.

Score	Term birth	Respiratory symptoms	NICU admission	Situs inversus	Cardiac anomaly	Rhinitis	Otitis media
14	2	2	2	4	2	1	1
14	2	2	2	4	2	1	1
12	2	2	2	4	0	1	1
12	2	2	2	4	0	1	1
11	2	2	2	4	0	0	1
11	2	2	2	4	0	1	0
10	0	2	2	4	0	1	1
10	2	2	0	4	0	1	1
8	2	2	2	0	0	1	1
8	2	2	2	0	0	1	1
8	2	2	2	0	0	1	1
8	2	0	0	4	0	1	1
8	2	2	0	4	0	0	0
8	2	2	2	0	0	1	1
8	2	2	2	0	0	1	1
8	2	2	2	0	0	1	1
8	2	2	2	0	0	1	1
7	2	2	2	0	0	1	0
7	2	2	2	0	0	1	0
6	2	2	0	0	0	1	1
6	2	2	0	0	0	1	1
6	2	2	0	0	0	1	1
6	2	2	0	0	0	1	1
6	2	2	0	0	0	1	1
6	2	0	2	0	0	1	1
6	2	2	0	0	0	1	1
6	0	0	0	4	0	1	1
6	2	0	0	4	0	0	0
4	2	0	0	0	0	1	1
4	2	0	0	0	0	1	1
4	2	0	0	0	0	1	1
4	0	0	2	0	0	1	1
3	2	0	0	0	0	1	0
3	2	0	0	0	0	1	0
3	2	0	0	0	0	1	0
3	2	0	0	0	0	1	0

Note: the items of each case are presented from the highest to the lowest score. Positive items are highlighted.

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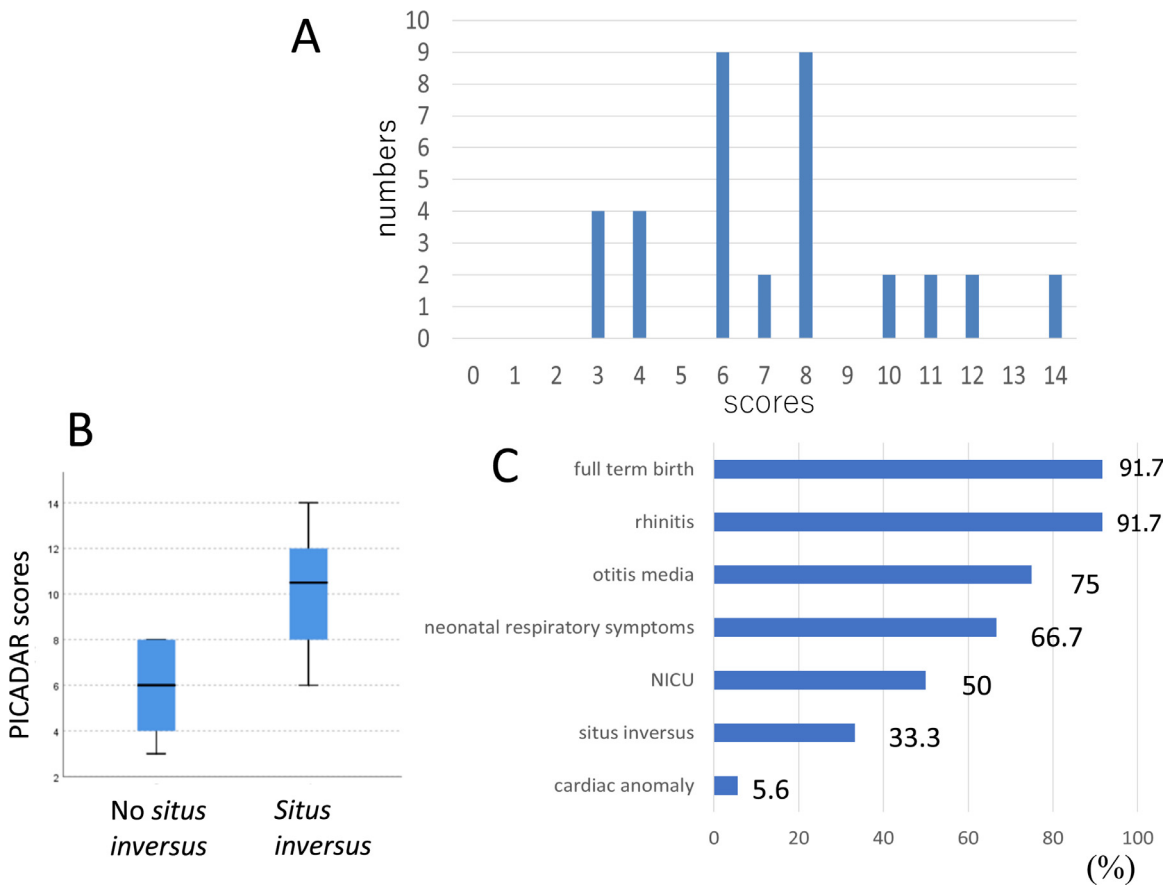

Fig. 3. The PICADAR score was evaluated in 36 cases and the distribution of the total PICADAR score is shown in (A). PICADAR scores ranged from 3 to 14 points (mean, 7.3), with significantly higher scores in patients with situs inversus than in those without (B). The percentage of positive cases for each of the seven PICADAR scores items is shown in (C). Most cases were born at term and had rhinitis, while only a few cases had a cardiac anomaly. PICADAR, PrImary CiliARy DyskinesiA Rule.

tients. Since the number of cases of sinus hypoplasia that could be evaluated in this study was small and defining hypoplasia quantitatively can be difficult, three otolaryngologists examined the CT scans and the presence of hypoplasia was reached by consensus.

4. Discussion

The median age at diagnosis of PCD was 17 years in the present study, which is comparable to the median age (18 years) of an international PCD cohort of 3,013 patients from 18 countries [21]. However, when we focus on patients younger than 20 years of age, the mean age in our series was 9.6 years, which is much older than in European countries [22]. The age at diagnosis in pediatric PCD (<20 years) is 5.0 years in Western Europe, 4.8 years in the British Isles, 5.5 years in Northern Europe, 6.8 years in Eastern Europe, and 6.5 years in Southern Europe, which strongly correlates with general government health expenditure [22]. Thus, PCD patients younger than 20 years of age in Japan are diagnosed much later than those in Europe.

According to a systematic review and meta-analysis of PCD in Japan [13], 148 of 316 cases (46.8%) were diagnosed at ≥18 years, and 68 of 316 cases (21.5%) were diagnosed between 0 and 17 years. The authors concluded that delayed diagnosis of this disease with a high frequency of inner dynein arm defects and low frequency of outer dynein arm defects appears to be a historical feature of PCD reported in Japan, when electron microscopy was the main diagnostic tool [13]. At present, inner dynein arm defects are not regarded as diagnostic of PCD [14] and as such, many false-positive cases misdiagnosed as PCD solely due to inner dynein arm defects were probably included in the systematic review. Compared with the review, the present analysis is based on electron microscopic findings supported by international consensus guidelines [14] and/or biallelic pathogenic variants in PCD-causing genes [4].

Patients who do not develop neonatal respiratory distress are usually referred to hospitals for diagnostic purposes later in life. Many patients with intractable chronic rhinosinusitis are referred to our hospital because mucopurulent nasal discharge does not resolve after multiple endonasal sinus surgeries.

In our patients, respiratory symptoms (77%) were the most common complaint, and wet cough was most prevalent. Lucas et al. [23] examined the sensitivity and specificity of each clinical symptom in PCD and reported that chronic (>3 months) wet cough had a sensitivity of 93% and specificity of 15% as an index for PCD. Chronic wet cough, although its specificity is low, is a symptom found in most PCD pa-

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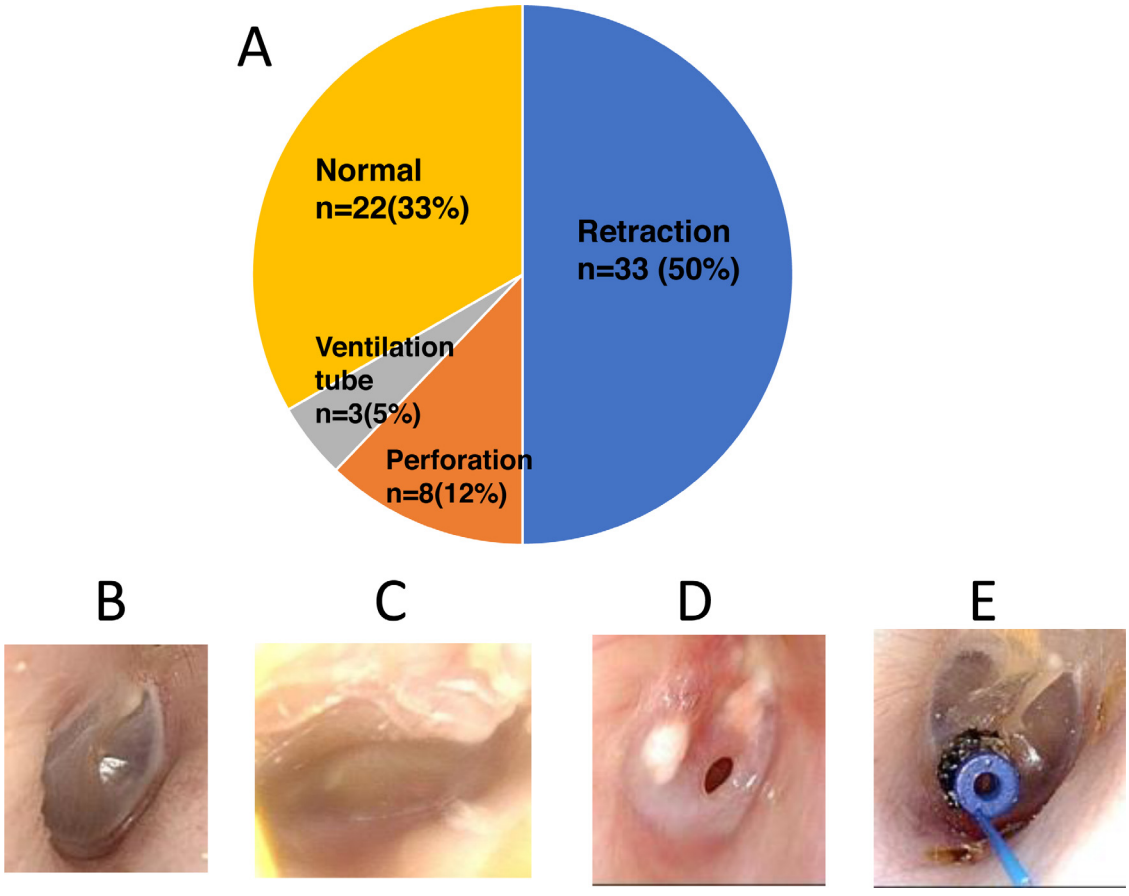

Fig. 4. Otoscopic findings of 66 ears in 33 cases. Fifty percent of tympanic membranes showed retraction, suggesting otitis media with effusion (A). Perforations and ventilation tubes can be regarded as sequelae of otitis media with effusion (A). Eardrums were normal in 33% of patients (A). Various otoscopic findings showing a normal membrane (B), retraction (C), perforation and calcification (D), and ventilation tube insertion (E).

tients and was also the most common complaint in our study. Nasal and aural symptoms were observed in 15% and 8% of patients, respectively. Whether these percentages are higher than in other facilities is not known, because the chief complaint of PCD patients has seldom been reported in the literature.

One of the major findings in this study was the small percentage of Japanese PCD patients with situs inversus (25%). Situs inversus totalis is reportedly observed in 40–50% of individuals with PCD [4]. In a study of 337 PCD patients from the USA (n = 147), Germany (n = 128), Canada (n = 36), and Australia (n = 26), situs solitus was observed in 46.0% of patients, situs inversus totalis in 47.7%, and heterotaxy in 6.3% [24]. The reason for the small percentage of situs inversus in our patients is that a large biallelic deletion in *DRC1* was the major cause of PCD, accounting for 42% of cases (25/59 cases). In the cilia of *DRC1* patients, central microtubules are often lacking. However, the function of primary cilia in the fetal node is preserved even in the absence of central microtubules. Thus, situs inversus does not occur in PCD patients with *DRC1* variants [12]. This deletion in *DRC1* has not been reported in European or American populations. Since this *DRC1* deletion has been suggested to be a founder mutation in Asians [10], the lower percentage of situs inversus may be a characteristic of other Asian populations. The percentage of situs inversus was reported as 63.3% (200/316) in the aforementioned systematic review and meta-analysis conducted in Japan [13]. Since this percentage is remarkably higher than in previous reports [4,24], historically, too much emphasis has been put on the importance of situs inversus in the diagnosis of PCD in Japan.

The PICADAR scoring system is a simple diagnostic clinical prediction tool with good accuracy and validity [17]. In this study, the total PICADAR score could be calculated in 36 cases. Since most of the adult patients did not know whether they had been born at term, their PICADAR scores could not be calculated. The mean PICADAR score obtained in the study (7.3 points) was lower than previously reported (mean ± standard deviation, 7.9 ± 2.8) [17]. The sensitivity and specificity of the PICADAR score are reported to be 90% and 75%, respectively, for a cut-off score of 5 points [17]. In the present study, 8 of 36 (22%) cases had a PICADAR score of ≤5, and this is significantly higher than reported previously by the PCD Diagnostic Center at University Hospital Southampton, UK (6%, 3/50; $p < 0.05$) [18]. Thus, even those with low PICADAR scores can have PCD and patients with chronic wet cough born at term should be considered as potential PCD patients.

The seven PICADAR score items were found to be positive in the following order: term birth (91.7%), rhinitis

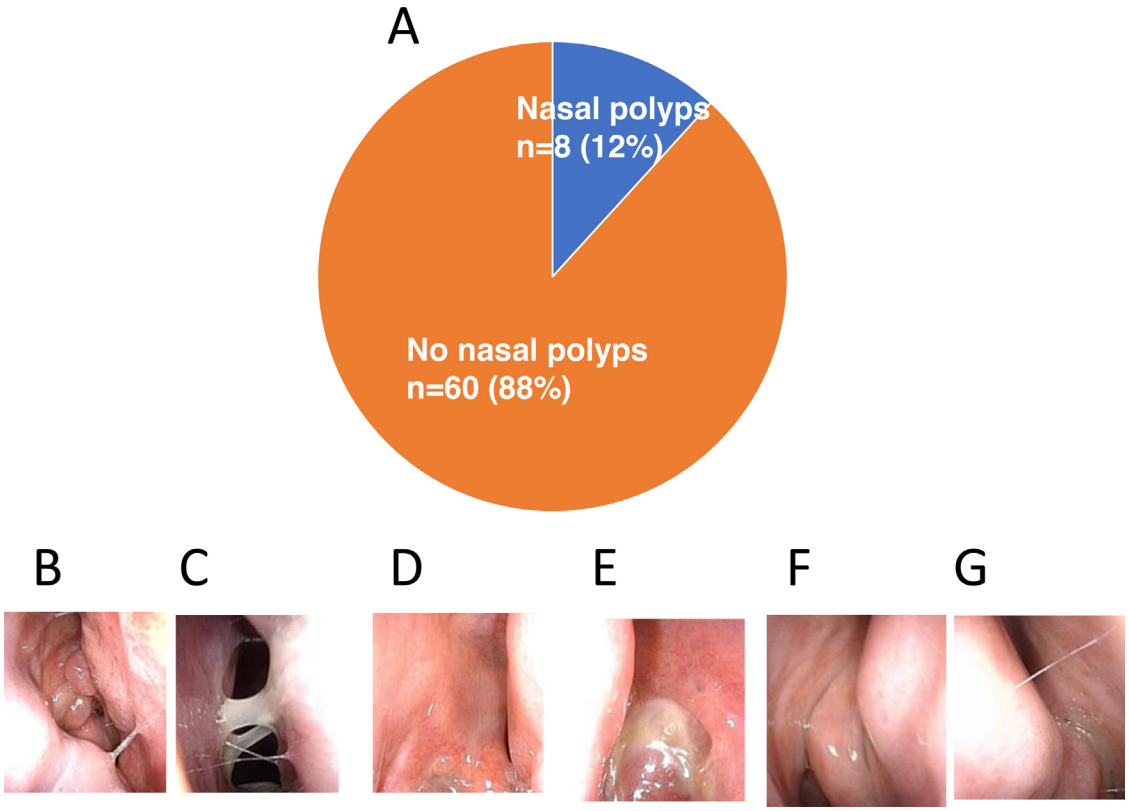

Fig. 5. Rhinoscopic findings of 68 nasal cavities in 34 cases. Nasal polyps were observed in 12% of patients (A). Rhinoscopic findings in 3 patients showing the presence (B, C, D, E,) and absence (F, G) of nasal polyps.

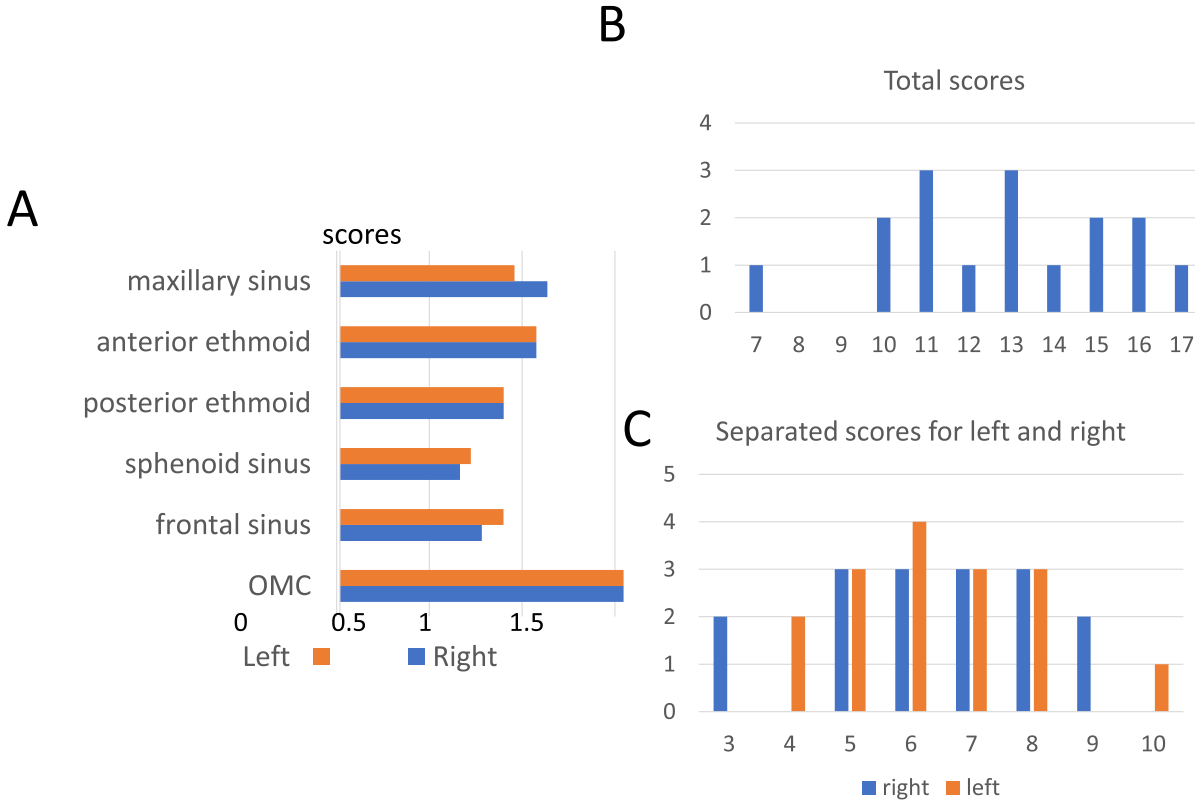

Fig. 6. Lund-Mackay scores were evaluated in 16 patients who had paranasal CT scans available. Although scores were high for the OMC, scores for individual sinuses were relatively low (A). The mean total score (left and right) was 12.8 points (B), and the mean score on both the left and right was 6.4 points (C). CT, computed tomography; OMC, ostiomeatal complex.

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(91.7%), otitis media (75%), neonatal respiratory symptoms (66.7%), NICU admission (50%), situs inversus (33.3%), and cardiac anomaly (5.6%). According to the original report on the PICADAR score, the order was as follows: term birth (90.7%), rhinitis (81.3%), neonatal respiratory symptoms (74.6%), NICU admission (61.3%), otitis media (57.3%), situs inversus (44%), and cardiac anomaly (8.0%) [18]. The higher percentage of otitis media cases in the present study may be because we are specialists in otology and can detect subtle abnormalities of the tympanic membranes. The lower percentage of situs inversus cases can be explained by differences in the causative genes, as stated earlier. When we compared consultation age, diagnosis age, and Lund-Mackay score between different PICADAR score groups with a cut-off score of 6 or 7, no significant differences were found between the groups with either cut-off score.

In most cases with PCD, certain abnormalities are found on otoscopic examination. Since perforation of the eardrums and ventilation tubes can be regarded as sequelae of otitis media with effusion, the basic aural abnormality is otitis media with effusion [25]. It is noteworthy that tympanic membrane findings were normal in approximately 30% of our cases, because all of the 30 eardrums examined in a previous study showed some abnormality [25]. This suggests that the absence of otitis media cannot rule out the possibility of PCD.

The mean Lund-Mackay score was 12.8 (range, 7–17) points, which is comparable to the score of 10.6 (range, 6–16) reported in a PCD cohort [26] and to those reported in children with cystic fibrosis (17.3 for patients undergoing surgery and 11.5 for those treated medically) [27]. It has been reported that the Lund-Mackay score is not a predictor for endoscopic sinus surgery or hospitalization in PCD patients [28]. The wide range of Lund-Mackay scores in PCD patients suggests that the radiographic severity of paranasal sinusitis varies between patients and that this scoring system is not useful for the diagnosis of PCD.

The limitations of the present study are as follows. We could evaluate PICADAR scores in only 54% of patients. The PICADAR score is a relatively new diagnostic tool (first reported in 2016) and we did not obtain sufficient information in the cases that were recruited prior to this. It was also difficult for adult patients to provide their term status and neonatal respiratory status. Otoscopic and rhinoscopic findings were obtained in only 49% and 51% of patients, respectively, because 45 of 67 (67%) patients visited our hospital, where we could perform these examinations. Moreover, most of the patients were examined in otoscopy and rhinoscopy only once when they visited our hospital. Currently, there is no obvious definition of sinus hypoplasia. Thus, three otolaryngologists examined the CT scan and the presence of hypoplasia was reached by consensus. Further studies should be conducted to address these limitations.

Conclusions

The age at which PCD was diagnosed in Japan was higher than in other countries. In the future, it will be important to raise awareness so that PCD can be diagnosed at an early stage.

Situs inversus was present in 25% of Japanese patients with PCD, which is much lower than observed in other countries. This resulted in the rather low PICADAR scores in our PCD patients. Knowledge of the clinical features of PCD may lead to its early detection.

The PICADAR score is a useful tool for diagnosing PCD. However, since *DRC1* is the causative gene, which is not usually associated with situs inversus in Japan, it should be noted that there were a number of cases with low PICADAR scores. In addition, the PICADAR score is a relatively new diagnostic tool, and we were only able to evaluate the total score in approximately 50% of cases. Further research is expected to overcome the difficulties with confirming the evaluation items such as respiratory symptoms in the neonatal period and further evaluate the usefulness of the PICADAR score.

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Declaration of Competing Interest

The authors have no conflicts of interest to declare.

References

[1] Afzelius BA, Mossberg B. Immotile-cilia syndrome (primary ciliary dyskinesia), including Kartagener syndrome. In: Scriver CR, Beaudet AL, Sly WS, Valle D, editors. The metabolic and molecular bases of inherited disease. New York: McGraw-Hill, Inc.; 1995. p. 3943–54.

[2] Paff T, Loges NT, Aprea I, Wu K, Bakey Z, Haarman EG, et al. Mutations in *PIH1D3* cause X-linked primary ciliary dyskinesia with outer and inner dynein arm defects. Am J Hum Genet 2017;100:160–8.

[3] Olcese C, Patel MP, Shoemark A, Kiviluoto S, Legendre M, Williams HJ, et al. X-linked primary ciliary dyskinesia due to mutations in the cytoplasmic axonemal dynein assembly factor PIH1D3. Nat Commun 2017;8:14279.

[4] Zariwala MA, Knowles MR, Leigh MW. Primary ciliary dyskinesia. SourceGeneReviews [Internet]. Pagon RA, Bird TD, Dolan CR, Stephens K, editors, Seattle (WA): University of Washington, Seattle; 1993. 2007 Jan 24 [updated December 5, 2019].

[5] Kano G, Tsujii H, Takeuchi K, Nakatani K, Ikejiri M, Ogawa S, et al. Whole-exome sequencing identification of novel DNAH5 mutations in a young patient with primary ciliary dyskinesia. Mol Med Rep 2016;14:5077–83.

[6] Orimo M, Kondo M, Takeyama K, Abe K, Miyoshi A, Honda N, et al. A Japanese case of primary ciliary dyskinesia with DNAH5 mutations. Intern Med 2019;58:2383–6.

[7] Orimo K, Kondo M, Arimura K, Takeyama K, Takeuchi K, Tamaoki J. Primary ciliary dyskinesia with complex abnormalities including cleavage of B-subfibers. Respirol Case Rep 2016;4:e00150.

[8] Takeuchi K, Kitano M, Kiyotoshi H, Ikegami K, Ogawa S, Ikejiri M, et al. A targeted next-generation sequencing panel reveals novel mutations in Japanese patients with primary ciliary dyskinesia. Auris Nasus Larynx 2018;45:585–91.

[9] Fassad MR, Patel MP, Shoemark A, Cullup T, Hayward J, Dixon M, et al. Clinical utility of NGS diagnosis and disease stratification in a multiethnic primary ciliary dyskinesia cohort. J Med Genet 2020;57 322–0.
[10] Morimoto K, Hijikata M, Zariwala MA, Nykamp K, Inaba A, Guo TC, et al. Recurring large deletion in DRC1 (CCDC164) identified as causing primary ciliary dyskinesia in two Asian patients. Mol Genet Genomic Med 2019;7:e838.
[11] Keicho N, Hijikata M, Morimoto K, Homma S, Taguchi Y, Azuma A, et al. Primary ciliary dyskinesia caused by a large homozygous deletion including exons 1-4 of DRC1 in Japanese patients with recurrent sinopulmonary infection. Mol Genet Genomic Med 2020;8:e1033.
[12] Takeuchi K, Xu Y, Kitano M, Chiyonobu K, Abo M, Ikegami K, et al. Copy number variation in DRC1 is the major cause of primary ciliary dyskinesia in the Japanese population. Mol Genet Genomic Med 2020;8:e1137.
[13] Inaba A, Furuhata M, Morimoto K, Rahman M, Takahashi O, Hijikata M, et al. Primary ciliary dyskinesia in Japan: systematic review and meta-analysis. BMC Pulm Med 2019;19:13.
[14] Shoemark A, Boon M, Brochhausen C, Bukowy-Bieryllo Z, De Santi MM, Goggin P, et al. International consensus guideline for reporting transmission electron microscopy results in the diagnosis of primary ciliary dyskinesia (BEAT PCD TEM Criteria). Eur Respir J 2020;55:1900725.
[15] American Thoracic Society; European Respiratory SocietyATS/ERS recommendations for standardized procedures for the online and offline measurement of exhaled lower respiratory nitric oxide and nasal nitric oxide, 2005. Am J Respir Crit Care Med 2005;171:912–30.
[16] Karadag B, James AJ, Gültekin E, Wilson NM, Bush A. Nasal and lower airway level of nitric oxide in children with primary ciliary dyskinesia. Eur Respir J 1999;13:1402–5.
[17] Rubin BK. Immotile cilia syndrome (primary ciliary dyskinesia) and inflammatory lung disease. Clin Chest Med 1988;9:657–68.
[18] Behan L, Dimitrov BD, Kuehni CE, Hogg C, Carroll M, Evans HJ, et al. PICADAR: a diagnostic predictive tool for primary ciliary dyskinesia. Eur Respir J 2016;47:1103–12.
[19] Lund VJ, Mackay IS. Staging in rhinosinusitis. Rhinology 1993;31:183–4.
[20] Pifferi M, Bush A, Caramella D, Di Cicco M, Zangani M, Chinellato I, et al. Agenesis of paranasal sinuses and nasal nitric oxide in primary ciliary dyskinesia. Eur Respir J 2011;37:566–71.
[21] Goutaki M, Maurer E, Halbeisen FS, Amirav I, Barbato A, Behan L, et al. The international primary ciliary dyskinesia cohort (iPCD Cohort): methods and first results. Eur Respir J 2017;49:1601181.
[22] Kuehni CE, Frischer T, Strippoli MP, Maurer E, Bush A, Nielsen KG, et al. Factors influencing age at diagnosis of primary ciliary dyskinesia in European children. Eur Respir J 2010;36:1248–58.
[23] Lucas JS, Barbato A, Collins SA, Goutaki M, Behan L, Caudri D, et al. European Respiratory Society guidelines for the diagnosis of primary ciliary dyskinesia. Eur Respir J 2017;49:1601090.
[24] Kennedy MP, Omran H, Leigh MW, Dell S, Morgan L, Molina PL, et al. Congenital heart disease and other heterotaxic defects in a large cohort of patients with primary ciliary dyskinesia. Circulation 2007;115:2814–21.
[25] Takeuchi K, Kitano M, Sakaida H, Usui S, Masuda S, Ogawa S, et al. Analysis of otologic features of patients with primary ciliary dyskinesia. Otol Neurotol 2017;38:e451–6.
[26] Pappa AK, Sullivan KM, Lopez EM, Adams KN, Zanation AM, Ebert CS Jr, et al. Sinus development and pneumatization in a primary ciliary dyskinesia cohort. Am J Rhinol Allergy 2021;35:72–6.
[27] Do BA, Lands LC, Mascarella MA, Fanous A, Saint-Martin C, Manoukian JJ, et al. Lund-Mackay and modified Lund-Mackay score for sinus surgery in children with cystic fibrosis. Int J Pediatr Otorhinolaryngol 2015;79:1341–5.
[28] Bhatt JM, Muhonen EG, Meier M, Sagel SD, Chan KH. Rhinosinusitis in pediatric primary ciliary dyskinesia: impact of disease. Otolaryngol Head Neck Surg 2019;161:877–80.

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