COMMENT

The challenge of diagnosing primary ciliary dyskinesia: a commentary on various causative genes and their pathogenic variants

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Primary ciliary dyskinesia (PCD) is a rare genetic disorder that causes abnormalities in mucociliary clearance in the respiratory epithelium (OMIM: 244400), leading to recurrent infections of the upper and lower airways [1]. Pathogenic variants of nearly 50 disease-causing genes, most of which have been determined to follow a pattern of autosomal recessive inheritance, have been identified to date (Table 1) [1, 2]. Depending on the causative genes, nodal ciliary movement is also hampered during the early embryonic stage, and laterality defects such as situs inversus and heterotaxy can occur, which has been known as Kartagener's syndrome. When sperm flagellar motility is similarly impaired, it often results in male infertility. Female infertility and hydrocephalus caused by dysfunction of motile cilia lining respective tissues have also been reported. Otitis media and ear-related problems appear to improve with age, but upper- and lower-respiratory disease persists and usually gets more severe in adulthood. The incidence of the disease has generally been estimated at 1 in 10,000 to 20,000 live births. However, it is thought to be underestimated because the number of cases with a confirmed diagnosis is still limited, especially in Asia and Africa; moreover, the diagnostic guidelines used in the USA and Europe have not been globally integrated or standardized.

In a recent article, Xu Y., et al. summarized their previous findings and presented the results of PCD-related gene analyses in 66 Japanese families unrelated to each other [3]. Their findings make us focus on the difficulties in diagnosing this disease at an early stage. First, clinicians frequently assume that recurrent sinopulmonary infections are not related to a congenital disorder, particularly when the majority of patients do not display laterality defects; particularly in Asia, a disease characterized by similar symptoms is ambiguously referred to as "sinobronchial syndrome" or often clinically diagnosed as "diffuse panbronchiolitis" [4–6]. Usually, in such cases, no further efforts in seeking a definite diagnosis have been made. Second, all non-genetics-based tests used to aid the diagnosis of PCD, such as nasal nitric oxide (NO) measurements, immunofluorescence staining of key proteins, high-speed video microscopy to evaluate ciliary movement, and transmission electron microscopy-based analysis of ciliary ultrastructure, are not standardized for use with medical equipment, not widely accessible to patients, and not covered by health insurance in many countries. Third, although recent advances in next-generation sequencing techniques have enabled the simultaneous analysis of all exons of multiple PCD-associated genes, variants of unknown significance in a variety of such genes, and pathogenic variants identified in only one allele, make their interpretation quite difficult. Therein, we should carefully and clearly describe variants that contribute to a confirmed diagnosis separately from variants that are not genetically diagnostically certain. It is important to mention the limitations in variant characterization and accurate specification of variant zygosity [3]. Until a reasonable interpretation of the initially identified variants is achieved, these considerations should be taken into account. This includes highlighting the need for new molecular findings, such as the identification of splicing variants at loci distant from canonical exon locations and structural variants that disrupt gene function in the other allele [7]. Fourth, it has recently been reported that the distribution of disease-causing genes and pathogenic variants varies significantly based on ethnicity, and the clinical manifestations and severity of the disease are deeply influenced by the genetic heterogeneity [8]. Although *DNAH5*, *DNAH11*, *DNAI1*, *CCDC39* and *CCDC40* are the most common genes among genetically determined PCD cases in North America and Europe [1, 2], Morimoto et al. recently identified a biallelic 27,748 bp deletion spanning exons 1–4 of *DRC1* in two patients of Japanese and Korean descent, but not in non-Asian populations [5]; subsequent studies have revealed that this large deletion appears to be the most common cause of PCD in Japanese and Korean populations, presumably as a founder mutation [3, 6, 9]. In PCD patients homozygous for this *DRC1* variant, the production of nasal NO was generally low, no situs inversus was observed, and no striking abnormalities in ciliary ultrastructure could be detected by electron microscopy [7]. Therefore, in such a clinical setting, measurement of nasal NO levels can be strongly recommended as an initial screening method, followed by genetic analysis using a simple PCR-based detection system [5] and more advanced methods [7] to reach an accurate diagnosis in patients who often experience non-specific recurrent airway infections, that are common manifestations of chronic rhinosinusitis and bronchiectasis.

It might be considered that clinical relevance of genetic diagnosis is poor in PCD, because no specific gene therapy for

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Table 1. A list of genes causing primary ciliary dyskinesia

Gene	Alias	Category	Nasal NO[a]	TEM (cilia)	Laterality defects	Subfertility	Location	RefSeq; MANE Select.	Number of coding exons	Translation length (aa)	Inheritance pattern	OMIM
DNAH5		Outer dynein arm protein	Reduced	ODA defects	Yes	Yes	5p15	NM_001369	79	4624	AR	CILD3 (608644)
DNAH9		Outer dynein arm protein	No abnormalities detected	partial ODA defects	Yes	Yes	17p12	NM_001372	69	4486	AR	CILD40 (618300)
DNAH11		Outer dynein arm protein	Reduced	No defects	Yes	Yes	7p15	NM_001277115	82	4516	AR	CILD7 (611884)
CCDC103		Outer dynein arm docking complex component	Reduced	ODA defects	Yes	Yes	17q21	NM_213607	3	242	AR	CILD17 (614679)
ODAD1	*CCDC114*	Outer dynein arm docking complex component	Reduced	ODA defects	Yes	NR	19q13	NM_001364171	14	707	AR	CILD20 (615067)
ODAD2	*ARMC4*	Outer dynein arm docking complex component	Reduced	ODA defects	Yes	NR	10p12	NM_018076	19	1044	AR	CILD23 (615451)
ODAD3	*CCDC151*	Outer dynein arm docking complex component	Reduced	ODA defects	Yes	NR	19p13	NM_145045	13	595	AR	CILD30 (616037)
ODAD4	*TTC25*	Outer dynein arm docking complex component	Reduced	ODA defects	Yes	NR	17q21	NM_031421	12	672	AR	CILD35 (617092)
DNAI1		Intermediate chain protein	Reduced	ODA defects	Yes	Yes	9p13	NM_012144	20	699	AR	CILD1 (244400)
DNAI2		Intermediate chain protein	Reduced	ODA defects	Yes	Yes	17q25	NM_023036	12	605	AR	CILD9 (612444)
NME8	*TXNDC3*	Intermediate chain protein	Reduced	partial ODA defects	Yes	NR	7p14	NM_016616	15	588	AR	CILD6 (610852)
DNAL1		Dynein light chain	Reduced	ODA defects	Yes	NR	14q24	NM_031427	8	190	AR	CILD16 (614017)
CFAP298	*C21orf59*	Cytoplasmic dynein axonemal assembly factor	Reduced	ODA + IDA defects	Yes	NA	21q22	NM_021254	7	290	AR	CILD26 (615500)
CFAP300	*C11orf70*	Cytoplasmic dynein axonemal assembly factor	Reduced	ODA + IDA defects	Yes	Yes	11q22	NM_032930	7	267	AR	CILD38 (618063)
DNAAF1	*LRRC50*	Cytoplasmic dynein axonemal assembly factor	Reduced	ODA + IDA defects	Yes	Yes	16q24	NM_178452	12	725	AR	CILD13 (613193)
DNAAF2	*KTU*	Cytoplasmic dynein axonemal assembly factor	Reduced	ODA + IDA defects	Yes	Yes	14q21	NM_018139	3	837	AR	CILD10 (612518)
DNAAF3	*C19orf51*	Cytoplasmic dynein axonemal assembly factor	Reduced	ODA + IDA defects	Yes	Yes	19q13	NM_001256715	11	541	AR	CILD2 (606763)
DNAAF4	*DYX1C1*	Cytoplasmic dynein axonemal assembly factor	Reduced	ODA + IDA defects	Yes	Yes	15q21	NM_130810	9	420	AR	CILD25 (615482)
DNAAF5	*HEATR2*	Cytoplasmic dynein axonemal assembly factor	Reduced	ODA + IDA defects	Yes	Yes	7p22	NM_017802	13	855	AR	CILD18 (614874)

Table 1. continued

Gene	Alias	Category	Nasal NO[a]	TEM (cilia)	Laterality defects	Subfertility	Location	RefSeq; MANE Select.	Number of coding exons	Translation length (aa)	Inheritance pattern	OMIM
DNAAF6	*PIH1D3*	Cytoplasmic dynein axonemal assembly factor	Reduced	ODA + IDA defects	Yes	Yes	Xq22	NM_173494	6	214	XR	CILD36 (300991)
DNAAF11	*LRRC6*	Cytoplasmic dynein axonemal assembly factor	Reduced	ODA + IDA defects	Yes	Yes	8q24	NM_012472	12	466	AR	CILD19 (614935)
SPAG1		Cytoplasmic dynein axonemal assembly factor	Reduced	ODA + IDA defects	Yes	Yes	8q22	NM_003114	18	926	AR	CILD28 (615505)
TTC12		Cytoplasmic dynein axonemal assembly factor	Reduced	IDA defects	NR	Yes	11q23	NM_017868	21	705	AR	CILD45 (618801)
ZMYND10		Cytoplasmic dynein axonemal assembly factor	Reduced	ODA + IDA defects	Yes	Yes	3p21	NM_015896	12	440	AR	CILD22 (615444)
CCDC39		Ruler protein	Reduced	IDA defects +MTD	Yes	Yes	3q26	NM_181426	20	941	AR	CILD14 (613807)
CCDC40		Ruler protein	Reduced	IDA defects +MTD	Yes	Yes	17q25	NM_017950	20	1142	AR	CILD15 (613808)
CCDC65	*DRC2*	Nexin dynein regulatory complex component	Reduced	No defects	NR	NR	12q13	NM_033124	8	484	AR	CILD27 (615504)
DRC1	*CCDC164*	Nexin dynein regulatory complex component	Reduced	No defects	NR	NR	2p23	NM_145038	17	740	AR	CILD21 (615294)
GAS8	*DRC4*	Nexin dynein regulatory complex component	No abnormalities detected	No defects	NR	Yes	16q24	NM_001481	11	478	AR	CILD33 (616726)
CFAP74		Central pair component	No abnormalities detected	No defects	NR	NA	1p36	NM_001304360	38	1639	AR	CILD49 (620197)
CFAP221	*PCDP1*	Central pair component	No abnormalities detected	No defects	NR	NR	2q14	NM_001271049	23	840	AR	-
HYDIN		Central pair component	Reduced	No defects	NR	Yes	16q22	NM_001270974	85	5121	AR	CILD5 (608647)
SPEF2		Central pair component	NA	No defects	NR	Yes	5p13	NM_024867	37	1822	AR	-
STK36		Central pair or radial spoke component	No abnormalities detected	CP defects	NR	NR	2q35	NM_015690	26	1315	AR	CILD46 (619436)
DNAJB13		Radial spoke component	No abnormalities detected	CP defects	NR	NR	11q13	NM_153614	8	316	AR	CILD34 (617091)
NME5		Radial spoke component	NA	CP defects	NR	NA	5q31	NM_003551	5	212	AR	CILD48 (620032)
RSPH1		Radial spoke component	No abnormalities detected	CP defects	NR	NR	21q22	NM_080860	9	309	AR	CILD24 (615481)

Table 1. continued

Gene	Alias	Category	Nasal NO[a]	TEM (cilia)	Laterality defects	Subfertility	Location	RefSeq; MANE Select.	Number of coding exons	Translation length (aa)	Inheritance pattern	OMIM
RSPH3		Radial spoke component	Reduced	CP defects	NR	NR	6q25	NM_031924	8	418	AR	CILD32 (616481)
RSPH4A		Radial spoke component	Reduced	CP defects	NR	NR	6q22	NM_001010892	6	716	AR	CILD11 (612649)
RSPH9		Radial spoke component	Reduced	CP defects	NR	NR	6p21	NM_152732	5	276	AR	CILD12 (612650)
GAS2L2		Ciliary base structure	No abnormalities detected	No defects	Yes	No	17q12	NM_139285	6	880	AR	CILD41 (618449)
OFD1		Ciliary base structure	NA	No defects	Yes	NA	Xp22	NM_003611	23	1012	XR	-
LRRC56		IFT associated protein	Reduced	No defects	Yes	NA	11p15	NM_198075	11	542	AR	CILD39 (618254)
CCNO		Proteins involved in multiciliogenesis	Reduced	Oligocilia	NR	Yes	5q11	NM_021147	3	350	AR	CILD29 (615872)
FOXJ1		Proteins involved in multiciliogenesis	No abnormalities detected	Oligocilia	Yes	Yes	17q25	NM_001454	2	421	AD	CILD43 (618699)
MCIDAS		Proteins involved in multiciliogenesis	Reduced	Oligocilia	NR	Yes	5q11	NM_001190787	7	385	AR	CILD42 (618695)
NEK10		Proteins involved in multiciliogenesis	No abnormalities detected	Short cilia	NR	NA	3p24	NM_001394966	35	1115	AR	CILD44 (618781)
TP73		Proteins involved in multiciliogenesis	NA	Oligocilia	NR	NA	1p36	NM_005427	13	636	AR	CILD47 (619466)

NO nitric oxide, *TEM* transmission electron microscopy, *MANE* matched annotation from NCBI and EMBL-EBI, *aa* amino acids, *NA* not available, *NR* not reported, *ODA* outer dynein arm, *IDA* inner dynein arm, *MTD* microtubular disorganization, *CP* central pair, *IFT* intraflagellar transport, *XR* X- linked recessive, *AD* autosomal dominant, *AR* autosomal recessive

[a]This gene list was created by referencing sources #1 and #2, as well as OMIM accessed in April 2023. Genes associated with primarily non-respiratory symptoms were excluded. The list will be updated with new relevant information as it becomes available in the future

improving impaired cilia motion is yet available; however, if a confirmed diagnosis is made, an intensive patient care protocol can be initiated; chest physiotherapy to promote sputum clearance, vaccination to prevent respiratory infections, appropriate use of antimicrobials to reduce exacerbations, smoking cessation, and exercise-based therapy are all important components of the management. It is recommended that authorities establish a system for providing lifelong medical care subsidies. Genetic counseling can be beneficial for patients and their families, particularly for those considering having children. Global collaborations among patient associations would be needed to avoid isolation, to share updates on the disease, and to ensure timely participation of patients if and when an international clinical trial is conducted. The first steps toward evidence-based treatment of PCD have already been taken; a recent multicenter phase-3 trial demonstrated that azithromycin reduced the incidence rate of exacerbations by half, and analysis of the already available in vitro data on the restoration of ciliary function is a crucial preliminary step to pave the way for developing new treatment strategies for PCD in the future [10].

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COMPETING INTERESTS

The authors declare no competing interests.

ADDITIONAL INFORMATION

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