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# Primary ciliary dyskinesia in Israel: Prevalence, clinical features, current diagnosis and management practices

Revital Abitbul [a, 1], Israel Amirav [a, b, 1], Hannah Blau [c, d], Soliman Alkrinawi [e], Micha Aviram [e], David Shoseyov [f], Lea Bentur [g], Avraham Avital [f], Chaim Springer [f], Moran Lavie [h], Dario Prais [c, d], Husein Dabbah [i], Nael Elias [j], Arnon Elizur [k], Shmuel Goldberg [l], Avigdor Hevroni [f], Eitan Kerem [f], Anthony Luder [a], Yehudah Roth [m], Malena Cohen-Cymberknoh [f], Marta Ben Ami [a], Avigdor Mandelberg [m], Galit Livnat [n], Elie Picard [l], Joseph Rivlin [n], Moshe Rotschild [g], Ruth Soferman [o], Niki T. Loges [p], Heike Olbrich [p], Claudius Werner [p], Alexander Wolter [p], Martina Herting [p], Julia Wallmeier [p], Johanna Raidt [p], Heymut Omran [p], Huda Mussaffi [c, d, *]

[a] Department of Pediatrics, Ziv Medical Center and Faculty of Medicine, Bar IIan University, Safed, Israel
[b] Department of Pediatrics, University of Alberta T6G 2R3, Edmonton, Canada
[c] Pediatric Pulmonary Institute, Schneider Children's Medical Center of Israel, Petach Tikva, Israel
[d] Sackler Faculty of Medicine, Tel Aviv University, Israel
[e] Soroka Medical Center, Beer Sheva, Israel
[f] Institute of Pulmonology, Hadassah-Hebrew University Medical Centers, Jerusalem, Israel
[g] Rambam Medical Center, Haifa, Israel
[h] Edmond & Lili Safra Children's Hospital, Chaim Sheba Medical Center, Israel
[i] Western Galilee Hospital, Nahariya and Faculty of Medicine, Bar IIan University, Safed, Israel
[j] Saint Vincent De-Paul Hospital, Nazareth, Israel
[k] Assaf Harofeh Medical Center, Zerifin, Israel
[l] Shaare Zedek Medical Center, Jerusalem, Israel
[m] The Edith Wolfson Medical Center, Holon and Sackler Faculty of Medicine, Tel Aviv University, Israel
[n] Carmel Medical Center, Haifa, Israel
[o] Dana Children's Hospital, Tel-Aviv Sourasky Medical Center, Israel
[p] Department of General Pediatrics, University Children's Hospital Muenster, Muenster, Germany

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ABSTRACT

*Background:* Primary Ciliary Dyskinesia (PCD) is rare and its features in Israel have not been described.
*Aims:* to assess prevalence utilizing state-of-the-art diagnostic techniques, and describe clinical features, diagnostic and management practices in Israel.
*Methods:* A national multicenter study from 2012 to 2013 recruited patients diagnosed or suspected of having PCD. Diagnosis was verified using: nasal Nitric Oxide (nNO); High-speed Video Microscope Analysis (HVMA); Transmission Electron Microscopy (TEM) of cilia; Immuno-fluorescence staining (IF) for ciliary proteins, and genetic analysis.
*Results:* Of the 203 patients recruited from 14 pediatric centers, 150 had a PCD diagnosis verified. Median age was 15.05y, with range 0.15–60.5y.
PCD prevalence was 1:54,000 for the general population and 1:25,000 in children (5-14 y). For the non-Jewish (mainly Druze and Arab Moslem) compared to Jewish populations, prevalence was 1:16,500 and 1:139,000 respectively ($p < 0.0001$) and parental consanguinity was 85.4% and 21.9% respectively ($p < 0.0001$).

*Abbreviations:* PCD, primary ciliary dyskinesia; nNO, nasal nitric oxide; TEM, transmission electron microscopy; HVMA, high-speed video-microscopy analysis; IF, immunofluorescence.

* Corresponding author. Pediatric Pulmonary Institute, Schneider Children's Medical Center of Israel, Petach Tikva, 49202, Israel.
*E-mail address:* hmussaffi@clalit.org.il (H. Mussaffi).

[1] Both authors contributed equally to this work.

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Clinical features included bronchiectasis (88%), rhinitis (81%), recurrent pneumonia (78%), recurrent otitis (62%), neonatal pneumonia (60%) and situs inversus (42%). Prior diagnostic practices varied widely between centers with TEM assessed in 55% and abnormal in 61% of these. Management included antibiotics and airway clearance.

Diagnostic verification revealed for 150 PCD patients: 81% nNO<233 ppb, 62% abnormal HVMA, 51% diagnostic TEM, 58% diagnostic IF and, 57% genetic diagnosis.

*Conclusions:* PCD in Israel is rare, with comprehensive diagnostic tests showing prevalence in children similar to Europe. Prevalence was higher in non-Jews, associated with parental consanguinity. Diagnostic and management practices vary. Referral centers providing comprehensive diagnostic and care capabilities should be established.



## 1. Introduction

Primary Ciliary Dyskinesia (PCD) is a rare autosomal recessive Mendelian disorder. The genetic fault is translated into abnormalities in structure and function of ciliary proteins throughout the body. The main function of the cilia in the upper and lower respiratory tract is to clear mucus secretions from the airways. Impaired ciliary function therefore results in the commonest manifestations of PCD: recurrent respiratory infections and recurrent ear, nasal and sinus disorders [1,2]. Repeated episodes contribute to the development of chronic pathologies within the respiratory system.

The cilia and its constituent proteins play a role in other processes and body systems. Among these is the determination of laterality during embryonic development; when impaired, it often leads to situs inversus [3].

PCD research in the past few years has expanded continuously with particular focus on mechanistic and genetic understanding, although available clinical and epidemiological data is limited. Current estimates of PCD prevalence in the general population vary from 1 in 2200–40,000 [4,5]. The diagnosis is often delayed, and even missed altogether. Various factors may contribute to this: (a) PCD may not be familiar to some primary care physicians (b) phenotypic heterogeneity and the lack of specificity of the clinical phenotype, especially among children, complicate the recognition of PCD, and (c) lack of a single, highly sensitive and specific diagnostic test for PCD [6,7]. In order to achieve a better understanding of PCD, efforts are being made to create a national PCD consortium [8]. In 2009, the European Respiratory Society (ERS) task force on PCD published a consensus statement paper addressing the issue of PCD patients' management based on the existing literature and accumulated clinical experience [5]. Following this publication, a large European group conducted a multi-center survey of 194 centers across Europe, including Israel [3,9]. Eighty seven patients with PCD from Israel were identified at that survey; yet, no details about their clinical features, diagnostic tests performed or management were described.

Therefore, in 2012–2013 we conducted a multicenter national study utilizing comprehensive state of the art diagnostic tests and confirming PCD in many more patients. The study was partially described in a previous publication [10]. During this study we also collected information about previous diagnosis and management practices that were routinely used in Israel.

The aims of this paper is to describe the prevalence of PCD in Israel as assessed by a state-of-the-art battery of diagnostic tests; the demographic and clinical features of these PCD patients and the routine diagnostic and management practices in these patients at the time of the study.

## 2. Methods

This multicenter prospective study was conducted from 2012 to 2013 in 14 pediatric pulmonary centers throughout Israel.

### 2.1. Study population

Patients with a provisional diagnosis or high clinical suspicion of PCD were invited to undergo a battery of diagnostic tests which collectively aid in the diagnosis of PCD. Patients receiving care in pediatric pulmonology centers in Israel were recruited to participate. Some of these centers care for adult PCD patients as well as children. Inclusion criteria for clinical phenotype [7,11] included at least two of the following clinical PCD characteristics: situs inversus or ambiguous; chronic, persistent lower respiratory symptoms (e.g., pneumonias, wet cough); chronic, persistent upper respiratory symptoms (e.g., rhinitis, sinusitis and/or ear infections); neonatal respiratory distress in full term neonates; bronchiectasis of unknown etiology (cystic fibrosis, and hypo-gamma globulinemia ruled out); fertility problems associated with chronic respiratory symptoms; positive family history (sibling) of PCD. Exclusion criteria included acute respiratory infection preceding the study within 4 weeks and a diagnosis other than PCD (e.g. immune deficiency or CF, both of which were excluded in all participants by appropriate tests).

All tested patients or parents/legal guardians signed informed consent forms prior to participation in the national study and each center obtained approval from its ethics committee. The study was registered with Clinical Trial Registration (NCT01070914).

### 2.2. Study questionnaire and clinical evaluation

All pediatric pulmonologists in Israel were invited by mail and in person to participate in the study. They were asked to fill a detailed questionnaire in order to collect clinical and demographic data from their patients. The initial draft of the questionnaire was developed by a national expert panel consisting of three pediatric pulmonologists, one adult pulmonologist and one ENT surgeon with expertise in PCD who reviewed existing questionnaires [2,12,13] and selected items. Based on feedback of the panel, modifications to the questionnaires were made after each iteration. The final questionnaire was divided into 6 sections: demographic information (A), family history (B), medical history (C), clinical evaluation (D), previous PCD investigations (E) and management (F). A list of the items is included in the on-line supplement. The questionnaire included queries regarding clinical features of PCD such as upper and lower respiratory system involvement, laterality abnormalities and fertility disorders. Where questionnaires were not filled for consenting patients, a member of this group (RA) visited the respective center and assisted the physician in obtaining

the data from the patient's medical files.

### 2.3. Procedures

Participants were invited by their primary pulmonary physician to attend the clinic for a 1–2 h visit. During this visit, physicians and patients completed the questionnaire. The patients then were tested for nNO. Regardless of the screening result, blood was drawn for genetic analysis and nasal epithelial cells were collected using a brush swab. Each brush swab sample was tested for PCD using:

a. High-Speed Video-Microscopy Analysis (HVMA).
b. Transmission Electron Microscopy (TEM).
c. Immunofluorescence staining for 4 different constituting proteins of ciliary structure: DNAH5, RSPH9, GAS8 and CCDC39.

For quality control all of the above were reviewed at least twice.

nNO measurements were obtained according to established American Thoracic Society guidelines [14]. A single manufacture unit (CLD 88 SP, Eco Physics Duernten, Switzerland) was employed in all centers. Most of the nNO tests were performed by a single team of investigators using the same unit. Some of the nNO tests were obtained by experienced technicians at local centers using identical standardized operating procedures (single breath with breath hold). An average of three measurements was taken for statistical analysis. In young children (usually less than 4 years) the tidal breath method was employed.

Clinical evaluation, questionnaire filling, nNO and HVMA tests were performed in Israel [10], whereas IF, TEM and genetic analysis were performed in the laboratory of Professor Omran in Münster, Germany as described previously [15].

### 2.4. Diagnostic criteria

Confirmation of PCD in these high-risk subjects was based on at least one of the following criteria:

1. Hallmark ultrastructural defects detected by TEM, including: Outer Dynein Arm (ODA) defects – absence or short ODA; Inner Dynein Arm (IDA) defect – absence; Central Pair (CP) defect/organization defect of the 9 + 2 cilia structure [16].
2. Low nNO levels ($\leq$233 ppb) [11] as well as dyskinetic function of the cilia, as evident by HVMA, including: immotile cilia, dyskinetic motility, stiff movement, low amplitude motility, no ciliary recovery stroke, hyperkinetic movement, rotatory movement and epithelial cells with reduced number of cilia organelles [17,18].
3. Absent protein on IF of at least one of the following four structural proteins of the cilia: DNAH5, RSPH9, CCDC39 and GAS8 [15].
4. In cases where clinical suspicion was very high and diagnostic tests were indeterminate or unavailable (e.g., lack of adequate nasal sample), diagnosis of PCD was further verified by presence of bi-allelic mutation of known PCD causing genes (e.g. *DNAH5, LRRC6, CCDC39, RSPH9, DNALI1, DNAH11, CCDC65, DNAAF3, MCIDAS, RSPH4A, CCDC103, ZMYND10, DYX1C1*). In cases where a previous genetic diagnosis of 2 mutations had been made, this was not repeated.

### 2.5. Statistical analysis

The data was analyzed using BMDP Statistical Software, Inc. (Statistical Solutions Ltd. Statistical Solutions, Unit 1A, South Ring Business Park, Stonehill Corporate Center, Suite 104Kinsale Road, Cork, Ireland, 999 Broadway, and Saugus, MA 01906, USA). Prevalence was calculated for each relevant cohort. For example, patients under the age of 18 years were not included in the statistical calculation of fertility disorders. Discrete values were compared using Pearson's Chi squared test.

## 3. Results

Of 51 pediatric pulmonologists in Israel, 30 reported treating patients with PCD. Of these, 28 pediatric pulmonologists from 14 centers agreed to participate in this study.

Two hundred and forty four patients known to the physicians with a provisional or suspected diagnosis of PCD were invited to participate and of these, 223 agreed to participate. Six withdrew consent, and 14 no longer fulfilled inclusion criteria for suspected PCD, so that 203 patients were enrolled in the study (Fig. 1).

The response rate for questionnaires was 178/203 (88%) of consenting, enrolled patients. Data relating to the other 25 patients was collected directly from the consenting patient's files as described.

Prior to the study, 6 patients had abnormal IF staining (identified at Prof. Omran's laboratory) and 19 patients were found to have 2 PCD mutations. These were not repeated during the study.

By implementing all the diagnostic techniques 150 patients were finally diagnosed with PCD. The diagnostic yield of tests performed during this study were: 146 completed nasal NO analysis and 122/146 (84%) had a nasal NO < 233 ppb. One hundred forty nine of the 150 PCD patients had a sample for TEM analysis, of those 119 samples were adequate for analysis with 77/119 (65%) were diagnostic for PCD. Samples from HVMA analysis were obtained from 147 of 150 PCD patients, of them only 111 samples were analyzable with 93/111 (84%) had abnormal ciliary beat consistent with PCD. One hundred and nineteen of the PCD patients had a technically acceptable sample for IF, and 79/119 (68%) of these had at least one abnormal IF staining for PCD proteins. Genetic studies were done for 85 patients and 68/85 (80%) were found to have 2 mutations. It should be noted that prior to the study, 6 additional patients had abnormal IF staining (identified at Prof. Omran's laboratory) and 19 additional patients were found to have 2 PCD mutations. These were not repeated during the study and were incorporated in our results. Thus, the total positive tests in the 150 patients with a verified diagnosis of PCD were: nNO 122/150 (81%), HVMA 93/150 (62%), TEM 77/150 (51%), IF staining 85/150 (58%) and genetic analysis 87/150 (57%).

Twenty two patients had 4 positive criteria for diagnosis of PCD and 95 patients had at least two positive criteria. Fifty five had only one positive criterion for diagnosis; 13 with low nNO as well as pathologic HVMA (considered a single criterion). Two had pathologic EM, both with low nNO. Eleven with positive IF staining (7 with a low nNO level) and 29 had two functional PCD mutations (11 with a low nNO level).

### 3.1. Prevalence of PCD in Israel

PCD diagnosis was established in 150 patients. Based upon published national population figures [19], the prevalence of PCD in the general population was 1:54,000. The prevalence in the non-Jewish and Jewish population was 1:16,500 and 1:139,000 respectively (Table 1).

### 3.2. Demographic and clinical characterization of the diagnosed patients

The demographic characteristics of the confirmed PCD group (n = 150) are shown in Table 2. Upper and lower respiratory tract

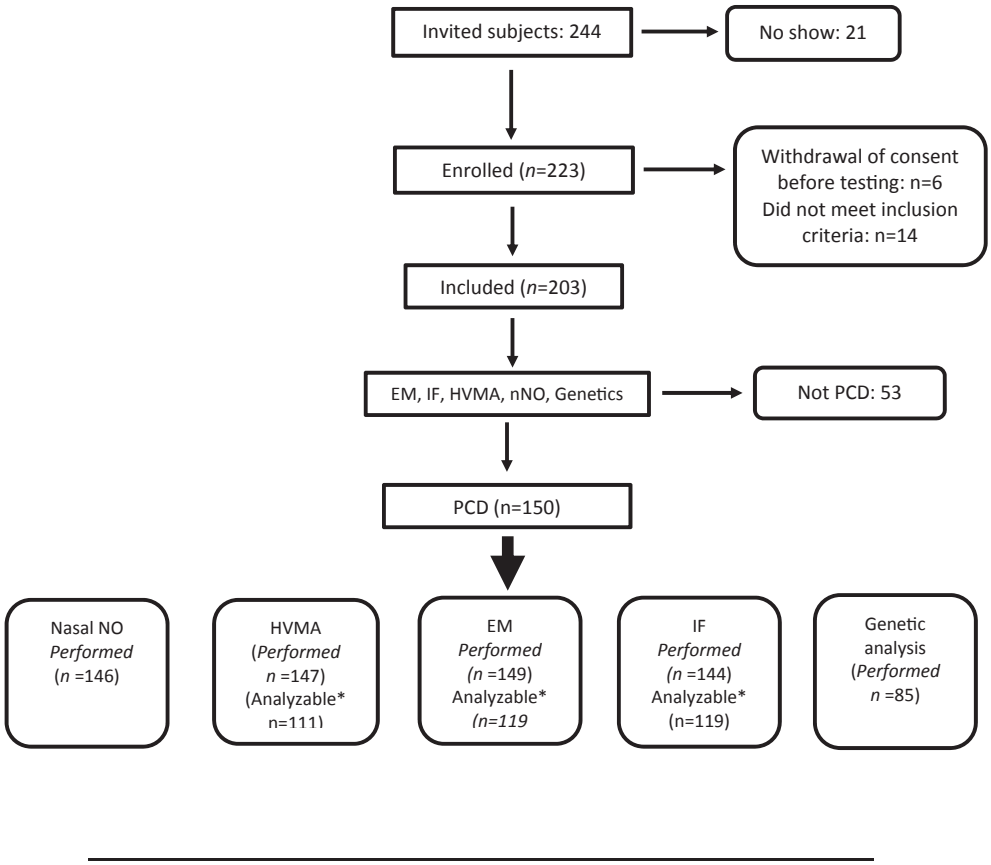



**Fig. 1.** Flow chart of patient's enrollment and the diagnostic tests performed. PCD, primary ciliary Dyskinesia; HVMA, high-speed video-microscopy analysis; TEM, transmission electron microscopy; IF, immunofluorescence. *Analyzable: technically adequate respiratory epithelial specimen for HVMA, EM and IF.

**Table 1**
Prevalence of PCD in Israel.

| Age group (years) | Population in 2013 (1000's) | | | Number of patients reported | | | Prevalence | | |
|---|---|---|---|---|---|---|---|---|---|
| | Total | Jewish | Non- Jewish[a] | Total | Jewish | Non- Jewish[a] | Total | Jewish | Non- Jewish[a] |
| 5–14 | 1453.9 | 1012.3 | 400.3 | 57 | 9 | 47[b] | 1:25,000 | 1:112,500 | 1:8500 |
| 0–19 | 2926.6 | 2061.5 | 781.6 | 98 | 25 | 71[b] | 1:30,000 | 1:82,500 | 1:11,000 |
| ≥19 | 5207.9 | 4043 | 1164,9 | 52 | 19 | 33 | 1:100,000 | 1:213,000 | 1:28,000 |
| Total population | 8134.5 | 6104.5 | 1714.6 | 150 | 44 | 104[b] | 1:54,000 | 1:139,000 | 1:16,500 |

[a] Non-Jewish population includes Muslims, Christians and Druze and does not include population not classified by religion (313,600).
[b] Two adopted patients <19 years old in Jewish families were excluded from the prevalence calculation for the non-Jewish population.

morbidities: respiratory distress or pneumonia in the first month of life were reported in 71 patients (60%, out of available responders n = 119), and 82% of the patients were born at term. Chronic cough reported in119 patients (88%; n = 136), starting before the age of one year in more than 48% of the cases. Recurrent pneumonia was reported in 110 patients (78%; n = 141), most of them commencing before five years of age (69%). Prolonged lung atelectasis was reported in 35 patients (27%; n = 135). Bronchiectasis evident on chest CT was reported in 98 patients (88%; n = 111). A variety of ENT disorders were reported: 110 patients had chronic rhinitis (81%, n = 136) and 86 patients (62%, n = 138) had recurrent otitis media. Chronic rhinitis started early with 41% and 61% before the ages of one and five years, respectively (Fig. 2). Situs abnormalities were reported in 62 patients (42%, n = 146). Out of 30 adults (≥18 years) who provided an answer about fertility, 15 subjects (11 males, 4 females) reported fertility problems and six males and one female reported no fertility problems.

### 3.3. Pre-study investigations and diagnosis

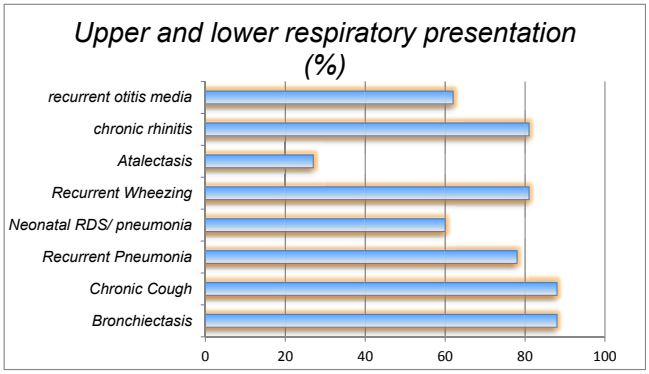
For the 150 patients diagnosed as above, questionnaires revealed pre-study diagnostic investigations with a relatively high degree of variability between centers.

Screening: NO analyzers were available in two centers. A total of 65 patients (43%) had already been screened by nNO testing prior to the current study, 38 were patients from the equipped centers and

**Table 2**
Demographic details (n = 150).

| | |
|---|---|
| Gender | Male 79 (53%)<br>Female 71 (47%) |
| Origin | Jewish 44 (29%)<br>Arab 91 (61%)<br>Druze 13 (9%)<br>unknown 2 (1%) |
| Age in years | Mean: 17.08; SD ± 11.96<br>Median: 15.05<br>Range: 0.15–60.47 |
| Adults, above 19years old | 52 (34.6%) |
| Consanguinity (n = 137)[a]: 1st, 2nd, and 3rd degree | 91 (66%) |
| Jewish (n = 41) | 9 (21.9%) |
| Non- Jewish (n = 96) | 82 (85.4%) |
| Patients distribution | |
| CF centers with multidisciplinary team | 89 (59%) |
| Other chronic lung disease center | 61 (41%) |

[a] Parental consanguinity is significantly higher in the non-Jewish population. P < 0.0001.




RDS, respiratory distress syndrome.

**Fig. 2.** Upper and lower respiratory presentation. RDS, respiratory distress syndrome.

27 were referred from 6 other centers. Abnormal nNO (<233 ppb) was found in 55/65 (85%) patients.

Diagnosis: The most common pre-study test used for PCD diagnosis was TEM, done in 83 patients (55%) from 10 centers. Five centers tested more than 75% of their patients. Pre-study ciliary function tests by light microscopy or HVMA were performed in 51 (34%) patients and abnormal ciliary function was reported in 41 of them (80%). Abnormal pre-study TEM features were found in 51/83 patients (61%), normal ultrastructure in 19/83 (23%) and no definitive diagnosis in 13/83 (16%). Pre-study IF staining and genetic testing were performed in 31 (21%) and 23 (15%) patients, respectively.

### 3.4. Management practices

The most commonly used treatments were antibiotics and airway clearance therapies, chest physiotherapy to drain the lungs using percussion (clapping), vibration, deep breathing and huffing. Follow up care of PCD patients included regular performance of lung function studies and sputum cultures. In the year before the current study, 101 patients (74%, n = 136; age ≥4 years) underwent lung function tests and the average value of FEV1% of predicted was 71% (range 29–117%). Sputum cultures during the year before enrollment were collected from 84 patients and were positive for: Hemophilus influenza 37 (44%), Pseudomonas aeruginosa 19 (23%), Staphylococcus aureus 10 (12%), Moraxella catarrhalis 4 (5%) and Mycobacteria abscessus 2 (2.5%).

Eighty nine patients with PCD (59%) were treated in six accredited Cystic Fibrosis centers that have a multi-disciplinary team. The remainder, were followed by pediatric pulmonologists in eight other hospitals.

Medical and drug therapies one year before enrollment are listed in Table 3. Four patients underwent lobectomy and 1 patient had lung transplantation.

## 4. Discussion

This multi-center national PCD study in Israel is one of the largest reported national cohorts, confirmed by a combination of state-of-the-art diagnostic tests. It provides a unique opportunity to evaluate the clinical characteristics, prevalence and diagnostic and management measures routinely practiced in Israel at the time of the study.

### 4.1. PCD prevalence in Israel

Based on our initial cohort of patients submitted for evaluation, we were now able to accurately identify many more patients with PCD. Prior to this national study there were 87 patients identified with PCD in Israel [4], whereas the number now significantly increased to 150 patients with PCD. The cases now brought for evaluation demonstrate a heightened awareness of this orphan disease over recent years, with a higher index of suspicion by primary physicians and pulmonologists [20,21]. The unique feature of this national study was the thorough diagnostic evaluation these patients now underwent. Many were subsequently excluded, while others were found to have PCD even though one test, e.g. TEM, was found to be normal.

In Israeli children, PCD prevalence was similar to that reported in Norway and not much less than Switzerland and Denmark (about 1:20,000). These countries have the highest prevalence in Europe, except for Cyprus where PCD diagnosis was reported at 1:10,000 children [4].

The prevalence of PCD in the non-Jewish patients was significantly higher than in the Jewish population most probably due to high rates of parental consanguinity found in this study and described for the Israeli Arab community in general [22]. Similarly, a higher prevalence of PCD has been demonstrated in other populations with high rates of consanguinity such as the north African population in Belgium and a British Asian population reported previously [6,23]. Previous published cohorts reported parental consanguinity to be 13% in the US cohort and 19.6% in the Belgian cohort [6,24].

**Table 3**
Treatment one year prior to enrollment in the study (n = 150).

| Treatment | N (%) |
|---|---|
| Antibiotics | 122 (81%) |
| Continuous treatment | 42 (34%) |
| During exacerbations only or alternately | 72 (59%) |
| Pattern was not reported | 8 (7%) |
| Bronchodilators | 94 (63%) |
| Inhaled corticosteroids | 83 (55%) |
| Nasal steroids | 33 (22%) |
| Hypertonic saline | 47 (31%) |
| Airway clearance therapy[a] | 94 (63%) |
| Physical activity n = 125 (age> 6 years) | 38 (30%) |

[a] Chest physiotherapy to drain the lungs using percussion (clapping), vibration, deep breathing and huffing.

### 4.2. Clinical features of the Israeli PCD population

As expected, the clinical spectrum of PCD patients in Israel is similar to that previously described, with predominant upper and lower respiratory tract morbidities and a relatively high prevalence of neonatal pneumonia (60%), which is, however, lower than previously reported [24]. In our cohort, situs inversus was reported in 42% of subjects which is similar to the proportion reported in Europe [4] and less than the expected 50%. This could be attributed to the presence of specific mutations not associated with situs inversus, such as mutations causing reduced generation of cilia [25].

### 4.3. Pre-study investigations and diagnosis

Similar to other European centers [9], there was great variability in diagnostic practices used among different Israeli centers prior to and at the time of this national study. Three centers based PCD diagnosis only on clinical features. Some patients only underwent screening with nNO and others underwent specific diagnostic tests (TEM or HVMA).

TEM was the most commonly used pre-study test whereas functional tests were used less commonly (using Light microscopy or HVMA). We attribute this to the low availability of HVMA technology in Israel. At the time of the study, there were only two centers performing HVMA for PCD patients. As part of the current project, a portable outreach HVMA system for improved diagnosis of PCD in remote regions was developed and described. Sensitivity of the outreach HVMA system was no different than sensitivity calculated using on-site HVMA [10]. Similarly, nasal NO evaluation was not readily available in all centers.

Pre-study genetic testing was performed mostly in families with more than one case of PCD. Currently, genetic testing for PCD in Israel is not funded routinely. As this study had a high yield of positive genetic diagnoses, many patients without definitive results of TEM and functional studies could undoubtedly benefit from subsidization of PCD genetic testing in Israel.

### 4.4. Management practices of PCD in Israel

Airway Clearance Therapy (ACT) was prescribed for the majority of patients. Although, as for cystic fibrosis, there is only a moderate level of evidence supporting ACT [5] the clinical experience of the treating physicians and the patient's satisfaction are very convincing. Airway clearance therapy was recently (2015) approved for government funding for PCD patients in Israel.

Antibiotics were also broadly used. Most physicians prescribed antibiotics intermittently, during pulmonary exacerbations. Continuous prophylactic antibiotic therapy was more commonly prescribed for patients followed in hospitals with large cystic fibrosis populations. There is no evidence supporting this approach, although it is favored by some large centers in Europe and might be considered in children requiring frequent intermittent courses of antibiotics [5,7]. Clearly there is a need for multicenter studies to evaluate this and other therapeutic interventions for patients with PCD. The use of other drugs like bronchodilators, steroids and hypertonic saline has been reported variably.

At the time of this study, PCD care in Israel was not centralized and there were no particular requirements for a pulmonary clinic managing these patients. All the patients included in the study were cared for by pediatric pulmonology centers, six of whom are accredited CF centers with a multidisciplinary team. As recommended by the ERS [5] and the UK experience, PCD patients receiving specialist care in centers with multidisciplinary team may have improved healthcare outcomes [26]. Due to the close follow up a PCD patient requires, it is recommended that PCD patients visit referral centers regularly. Establishing such centers in Israel is highly recommended. Such centers could contribute to a national PCD database and hopefully become part of an international program including the development of a PCD registry as was recently published by Werner and al [27].

### 4.5. Limitations

Adults with PCD are often followed in pediatric centers. However, those managed in adult pulmonary clinics were not included in this study. This could lead to an underestimation of PCD prevalence in Israeli adults.

For a definitive diagnosis of PCD, nasal brushing for HVMA and TEM should optimally be repeated, due to the possibility of a poor sample or secondary changes in the epithelium. Our study involved a single patient visit with only one nasal brushing. Therefore, we were more stringent in requirements for quality control of the nasal brushing, reviewing HVMA, TEM and IF at least twice. In addition, we required both nNO and HVMA to be abnormal in the case of a normal TEM.

## 5. Conclusion

We present the first detailed report on the demographics, clinical features and diagnosis and management practices for PCD in Israel. Prevalence was more accurately defined using a comprehensive battery of diagnostic tests. We found that PCD is rare in Israel as in Europe, with a higher prevalence in the non-Jewish, mainly Moslem and Druze Arabs, probably due to higher rates of consanguinity in this population. Current diagnostic and management routines in Israel are variable. We recommend that PCD referral centers be established based on consensus guidelines, to optimize identification and best care for patients with this rare disease.

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All authors have indicated they have no financial relationships to disclose relevant to this article.

## Conflict of interest

The authors have no conflicts of interest to disclose relevant disclose relevant to this article.

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## Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.rmed.2016.08.015.

## References

[1] P.H. Hosie, D.A. Fitzgerald, A. Jaffe, C.S. Birman, J. Rutland, L.C. Morgan,

Presentation of primary ciliary dyskinesia in children: 30 years' experience, J. Paediatr. Child. Health 51 (7) (2015 Jul) 722–726.
[2] P.G. Noone, M.W. Leigh, A. Sannuti, S.L. Minnix, J.L. Carson, M. Hazucha, M.A. Zariwala, M.R. Knowles, Primary ciliary dyskinesia: diagnostic and phenotypic features, Am. J. RespirCrit Care Med. 169 (4) (2004 Feb 15) 459–467.
[3] S. Nonaka, Y. Tanaka, Y. Okada, S. Takeda, A. Harada, Y. Kanai, M. Kido, N. Hirokawa, Randomization of left-right asymmetry due to loss of nodal cilia generating leftward flow of extraembryonic fluid in mice lacking KIF3B motor protein, Cell 95 (6) (1998 Dec 11) 829–837.
[4] C.E. Kuehni, T. Frischer, M.P. Strippoli, E. Maurer, A. Bush, K.G. Nielsen, A. Escribano, J.S. Lucas, P. Yiallouros, H. Omran, E. Eber, C. O'Callaghan, D. Snijders, A. Barbato, ERS Task Force on Primary Ciliary Dyskinesia in Children. Factors influencing age at diagnosis of primary ciliary dyskinesia in European children, EurRespir J. 36 (6) (2010 Dec) 1248–1258.
[5] A. Barbato, T. Frischer, C.E. Kuehni, D. Snijders, I. Azevedo, G. Baktai, L. Bartoloni, E. Eber, A. Escribano, E. Haarman, B. Hesselmar, C. Hogg, M. Jorissen, J. Lucas, K.G. Nielsen, C. O'Callaghan, H. Omran, P. Pohunek, M.P. Strippoli, A. Bush, Primary ciliary dyskinesia: a consensus statement on diagnostic and treatment approaches in children, EurRespir J. 34 (6) (2009 Dec) 1264–1276.
[6] M. Boon, A. Smits, H. Cuppens, M. Jaspers, M. Proesmans, L.J. Dupont, F.L. Vermeulen, S. Van Daele, A. Malfroot, V. Godding, M. Jorissen, K. De Boeck, Primary ciliary dyskinesia: critical evaluation of clinical symptoms and diagnosis in patients with normal and abnormal ultrastructure, Orphanet J. Rare Dis. 9 (2014 Jan 22) 11.
[7] C. Werner, J.G. Onnebrink, H. Omran, Diagnosis and management of primary ciliary dyskinesia, Cilia 4 (1) (2015 Jan 22) 2.
[8] http://www.bestcilia.eu/.
[9] M.P. Strippoli, T. Frischer, A. Barbato, D. Snijders, E. Maurer, J.S. Lucas, E. Eber, B. Karadag, P. Pohunek, Z. Zivkovic, A. Escribano, C. O'Callaghan, A. Bush, C.E. Kuehni, ERS Task Force on Primary Ciliary Dyskinesia in Children. Management of primary ciliary dyskinesia in European children: recommendations and clinical practice, EurRespir J. 39 (6) (2012 Jun) 1482–1491.
[10] I. Amirav, H. Mussaffi, Y. Roth, M. Schmidts, H. Omran, C. Werner, Israeli PCD Consortium Investigators. A reach-out system for video microscopy analysis of ciliary motions aiding PCD diagnosis, BMC Res. Notes 8 (1) (2015 Dec) 999.
[11] M.W. Leigh, A.J. Shapiro, J.E. Pittman, S.D. Davis, H. Lee, J. Krischer, T.W. Ferkol, J.J. Atkinson, S.D. Sagel, M. Rosenfeld, S.D. Dell, C. Milla, K.N. Olivier, M.R. Knowles, Genetic diseases of mucociliary clearance consortium (GDMCC). Definition of clinical criteria for diagnosis of primary ciliary dyskinesia, Am. J. RespirCrit Care Med. 185 (2012) A2483.
[12] E.M. Noll, C.H. Rieger, E. Hamelmann, T.G. Nüsslein, Questionnaire to preselect patients with a high probability of primary ciliary dyskinesia, Klin. Padiatr 223 (1) (2011 Jan) 22–26.
[13] J.U. Sommer, K. Schäfer, H. Omran, H. Olbrich, J. Wallmeier, A. Blum, K. Hörmann, B.A. Stuck, ENT manifestations in patients with primary ciliary dyskinesia: prevalence and significance of otorhinolaryngologic co-morbidities, Eur. Arch. Otorhinolaryngol. 268 (3) (2011 Mar) 383–388.
[14] American Thoracic Society; European Respiratory Society, ATS/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. 171 (8) (2005 Apr 15) 912–930.
[15] H. Omran, N.T. Loges, Immunofluorescence staining of ciliated respiratory epithelial cells, Methods Cell Biol. 91 (2009) 123–133.
[16] Papon JF, Coste A, Roudot-Thoraval F, Boucherat M, Roger G, Tamalet A, Vojtek AM, Amselem S, Escudier E. A 20-Year Experience of Electron.
[17] W.A. Stannard, M.A. Chilvers, A.R. Rutman, C.D. Williams, C. O'Callaghan, Diagnostic testing of patients suspected of primary ciliary dyskinesia, Am. J. Respir. Crit. care Med. 181 (4) (2010) 307–314.
[18] J. Raidt, J. Wallmeier, R. Hjeij, J.G. Onnebrink, P. Pennekamp, N.T. Loges, H. Olbrich, K. Häffner, G.W. Dougherty, H. Omran, C. Werner, Ciliary beat pattern and frequency in genetic variants of primary ciliary dyskinesia, Eur. Respir. J. 44 (6) (2014 Dec) 1579–1588, http://dx.doi.org/10.1183/09031936.00052014. Epub 2014 Sep. 3.
[19] http://www.cbs.gov.il/reader/cw_usr_view_SHTML?ID=629.
[20] L. Behan, B.D. Dimitrov, C.E. Kuehni, C. Hogg, M. Carroll, H.J. Evans, M. Goutaki, A. Harris, S. Packham, W.T. Walker, J.S. Lucas, PICADAR: a diagnostic Predict. tool Prim. ciliary dyskinesia, Eur Respir J 47 (4) (2016 Apr) 1103–1112.
[21] M.W. Leigh, T.W. Ferkol, S.D. Davis, H.S. Lee, M. Rosenfeld, S.D. Dell, S.D. Sagel, C. Milla, K.N. Olivier, K.M. Sullivan, M.A. Zariwala, J. Pittman, A.J. Shapiro, J.L. Carson, J. Krischer, M.J. Hazucha, M.R. Knowles, Clinical features and associated likelihood of primary ciliary dyskinesia in children and adolescents, Ann. Am. Thorac. Soc. 13 (8) (2016 Aug) 1305–13013.
[22] Jaber L1, M. Shohat, G.J. Halpern, Demographic characteristics of the Israeli Arab community in connection with consanguinity, Isr. J. Med. Sci. 32 (12) (1996 Dec) 1286–1289.
[23] C. O'Callaghan, P. Chetcuti, E. Moya, High prevalence of primary ciliary dyskinesia in a British Asian population, Arch. Dis. Child. 95 (2010) 51–52.
[24] M.R. Knowles, L.A. Daniels, S.D. Davis, M.A. Zariwala, M.W. Leigh, Primary ciliary dyskinesia. Recent advances in diagnostics, genetics, and characterization of clinical disease, Am. J. RespirCrit Care Med. 188 (8) (2013 Oct 15) 913–922.
[25] J. Wallmeier, D.A. Al-Mutairi, C.T. Chen, N.T. Loges, P. Pennekamp, T. Menchen, L. Ma, H.E. Shamseldin, H. Olbrich, G.W. Dougherty, C. Werner, B.H. Alsabah, G. Köhler, M. Jaspers, M. Boon, M. Griese, S. Schmitt-Grohé, T. Zimmermann, C. Koerner-Rettberg, E. Horak, C. Kintner, F.S. Alkuraya, H. Omran, Mutations in CCNO result in congenital mucociliary clearance disorder with reduced generation of multiple motile cilia, Nat. Genet. 46 (6) (2014 Jun) 646–651.
[26] J. Lucas, P.H. Chetcuti, F. Copeland, C. Hogg, T. Kenny, E. Moya, C.H. O'Callaghan, W.T. Walker, Overcoming challenges in the management of primary ciliary dyskinesia: the UK model, Paediatr. Respir. Rev. 15 (2014) 142–145.
[27] C. Werner, M. Lablans, M. Ataian, J. Raidt, J. Wallmeier, J. Große-Onnebrink, C.E. Kuehni, E.G. Haarman, M.W. Leigh, A.L. Quittner, J.S. Lucas, C. Hogg, M. Witt, K.N. Priftis, P. Yiallouros, K.G. Nielsen, F. Santamaria, F. Ückert, H. Omran, An international registry for primary ciliary dyskinesia, Eur. Respir. J. 47 (3) (2016 Mar) 849–859.