OTITIS MEDIA AND THE IMMOTILE CILIA SYNDROME.*†

ROBERT JAHRSDOERFER, M.D.,
PHILLIP S. FELDMAN, M.D.,
EDWIN W. RUBEL, Ph.D.,
JOHN L. GUERRANT, M.D.,
PEYTON A. EGGLESTON, M.D.,
and
ROBERT F. SELDEN, M.D.,
Charlottesville, Va.

ABSTRACT.

The immotile cilia syndrome appears to be a congenital defect in the ultrastructure of cilia that renders them incapable of movement. Respiratory tract cilia and sperm are predominantly affected. Bronchiectasis, sinusitis and male sterility are the main clinical findings. Situs inversus may be found. To these findings can be added otitis media.

The defect appears to be a complete or partial absence of dynein arms which are believed to be essential for generating movement of cilia or sperm tails. Six patients suspected of having immotile cilia were compared to six patients in a control group. In affected patients, no cilia movement in the middle ear or nasopharynx was observed using the operating microscope. Electron microscopy of cilia from the mucosa of the middle ear and nasopharynx appeared to confirm the ultrastructural defect in two of six patients suspected of having the syndrome.

INTRODUCTION.

Sterile male patients with chronic airway infection have been shown to have an underlying defect in ciliary motility.[1-6] As a ciliated epithelium provides the cover lining of the respiratory tract, it is not surprising that a defect in mucociliary transport would ultimately lead to stasis and chronic infection. Sinusitis and bronchiectasis are common, and may be found in combination with situs inversus (Kartagener's Syndrome). This syndrome may also include chronic bronchitis, rhinitis, nasal polyps and otitis media. Male patients with Kartagener's Syndrome have shown a high rate of sterility.[1,2,4] Analysis has shown that the sperm from these patients is alive and metabolically normal, but immotile. Electron microscopy of the sperm tail has shown absent dynein arms, believed to be essential for normal bending movements.

The knowledge that sperm tails are morphologically similar to cilia led to investigation of ciliated epithelium of the upper and lower airway in affected patients. Cilia from these areas were found to have absent, or defective dynein arms.

While several authors had noted the prevalence of otitis media in Kartagener's Syndrome, as well as the Immotile Cilia Syndrome,[1,2,5-7] none had

*Presented at the Meeting of the Eastern Section of the American Laryngological, Rhinological and Otological Society, Inc., New York, N. Y., January 5, 1979.

†From the Departments of Otolaryngology and Maxillofacial Surgery, Pathology, Internal Medicine and Pediatrics, University of Virginia School of Medicine.

Send Reprint Requests to Robert Jahrsdoerfer, M.D., Department of Otolaryngology and Maxillofacial Surgery, University of Virginia School of Medicine, Box 430, Charlottesville, Va. 22908.

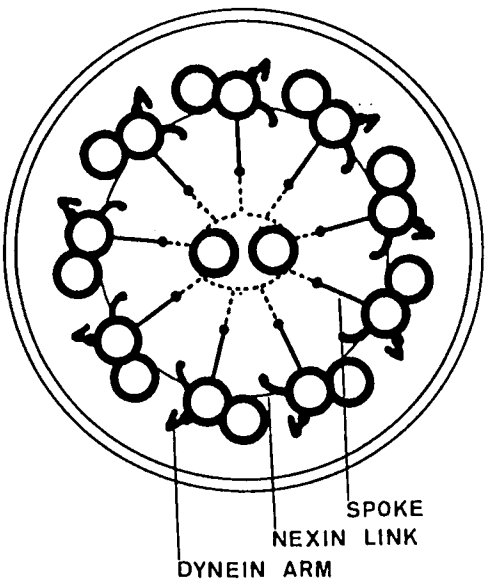

Fig. 1. Diagram of the cross-section of a cilium or of the central portion of a sperm tail. The assembly of the 9 outer microtubular doublets and the 2 central microtubules is held together by 3 kinds of connections: the dynein arms, the nexin links, and the spokes. The dynein arms are generally believed to be responsible for the motility. From Eliasson, *et al.*[1] Reprinted with permission from the New England Journal of Medicine, 297:3, 1977.

stressed the middle ear as a prominent part of the disease. Recently, however, a report specifically focused on the association of otitis media and a middle ear ciliary defect.[8]

ULTRASTRUCTURE OF CILIA.

Sperm tails, flagella and cilia have basic similarities in their ultrastructure. There is a constant 9 + 2 pattern of outer doublet tubules to a central pair of single tubules. This grouping is enclosed in a membrane in the form of a cylinder and is called an axoneme. The nine outer doublet tubules are interconnected by nexin links, and connected radially to the central pair of single tubules by spokes (Fig. 1). The central tubules run from the basal plate in the cell body to the tip of the cilium where they fuse.[9] The outer doublet filaments stop short of the tip.

Each outer doublet tubule is composed of an "A" fiber and a "B" fiber. Attached to each "A" fiber are two parallel rows of projections called dynein arms, which extend toward the "B" fiber of the adjacent doublet tubule. Motility of the cilium or sperm tail is assumed to occur from the arms holding one doublet in a steady grip while generating an active sliding of any other doublet relative to its neighbor.[10] Thus, a form of temporary cross-over bridging occurs between adjacent microtubules which allows for cilial motility. This "sliding filament" mechanism has been confirmed by other investigators.[9,11,12] The central tubules are devoid of arms.[13]

Dynein is an adenosine triphophatase protein of high molecular weight. Each outer arm is formed from three or four units, suggesting a multiunit polymer of dynein. This probably accounts for the hook-like appearance of the outer arm.[11]

The defect in the Immotile Cilia Syndrome appears to be a complete or partial absence of dynein arms. Other abnormalities of cilia which may

TABLE I.

Clinical Profiles of Six Patients (Test Group) Suspected of Having Immotile Cilia.

Case No.	Sex	Age (yr)	Otitis	Sinusitis	Bronchiectasis	Situs Inversus	Immotile Sperm
1	M	20	+	+	+	−	+
2	F	10	−	+	+	−	
3	F	12	+	+	+	−	
4	M	6	+	+	+	+	?
5	M	9	+	+	+	−	?
6	M	8	+	+	+	−	?

TABLE II.

Biopsy Sites for Cilia.

	Test Group N = 6	Control Group N = 6
Nasopharynx	6	4
Middle ear	5	6
Maxillary sinus	2	0
Bronchus	1	0
Sperm*	1	0

*Sperm tail is a modified cilium.

contribute to immotility are irregular orientation of the central tubules, depleted or extra microtubules, and defective radial spokes.[14]

Normal cilia should be oriented to within 5 to 25 degrees of each other.[1] Lack of orientation is best demonstrated by comparing the paired central single tubules. Additions or deletions to the 9 + 2 cross sectional scheme have been noted in patients with immotile cilia.[15] However, different patterns of tubular arrangement have been noted in other bronchial diseases,[16] as well as other system diseases in man and animals.[6]

MATERIALS AND METHODS.

Test Group.

Six patients suspected of having the Immotile Cilia Syndrome were compared to 6 control patients. In the test group there were 4 males and 2 females, and 5 of these were 12 years of age or younger. Cases 2 and 3 were sisters and Cases 5 and 6 were brothers.

All 6 patients in the test group had both sinusitis and bronchiectasis, and 5 of the 6 had otitis media (Table I). Four of the 5 patients with otitis had perforated tympanic membranes. Two middle ears were actively discharging, 1 was moist, and 1 was dry. Case 4 had bilateral chronic serous otitis media and a past history of recurrent otitis media. Case 5 had had a mastodiectomy at age 6.

A large tympanic membrane perforation in Case 1 afforded good visualization of the hypotympanum and anterior middle ear by use of the operating microscope. No cilial movement was observed. In 3 of 6 patients the nasal turbinates were decongested to allow microscopic rhinoscopy of the posterior wall of the nasopharynx. Cilial movement was not observed. In the control group, cilial activity was readily seen in both the middle ear and nasopharynx.

One patient had situs inversus (Case 4) and had previously been diagnosed as having Kartagener's Syndrome. His young age of 6 years precluded an assessment of his fertility. The only adult in the test group (Case 1) had a total sperm count of 77×10^6 (normal $> 40 \times 10^6$) with 92% immotile sperm. All 6 test patients were carefully evaluated for immune function and cystic fibrosis. Test results were normal. Mucociliary transport studies were not done.

With informed consent, biopsies for cilia were taken from the nasopharynx in 6 patients, middle ear in 5 patients, maxillary sinus in 2 patients and bronchus in 1 patient. All 6 test patients had multiple site biopsies (Table II). All specimens were placed directly in 2% Gluteraldehyde.

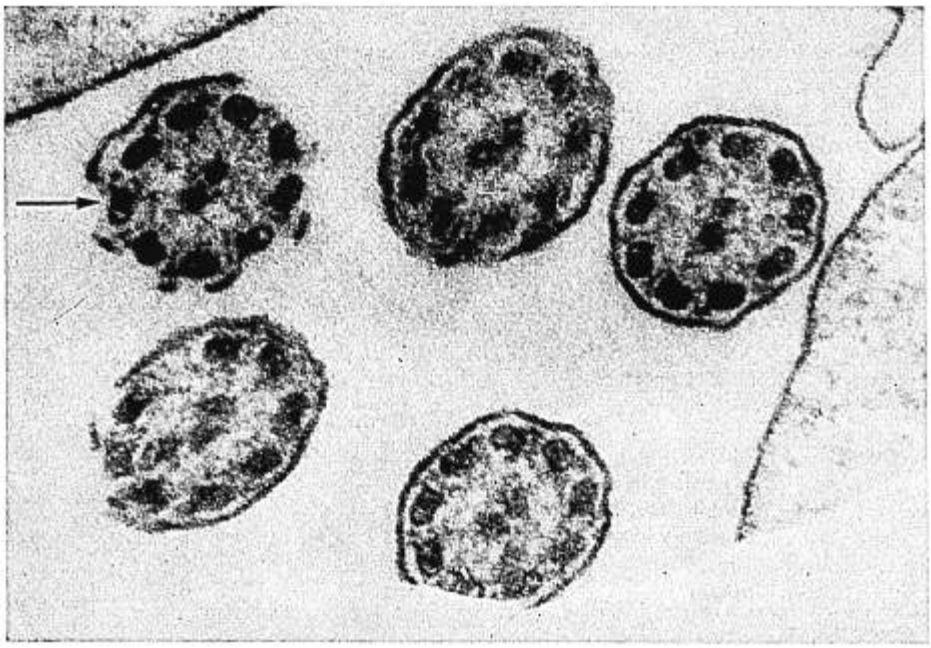

Fig. 2. Cross-section through cilia from the middle ear epithelium of a control group patient. Note doublet (arrow) showing inner and outer dynein arms. Original magnification x 154,000.

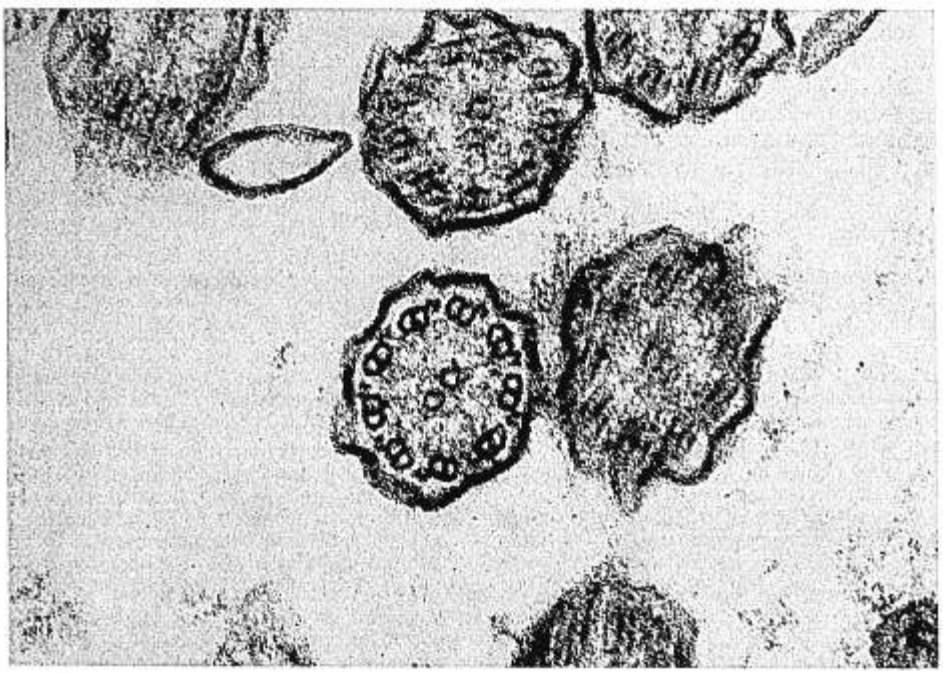

Fig. 3. Cross-section through cilia from the middle ear epithelium of another control group patient. Note normal outer dynein arms with hook-like appearance. Original magnification x 154,000.

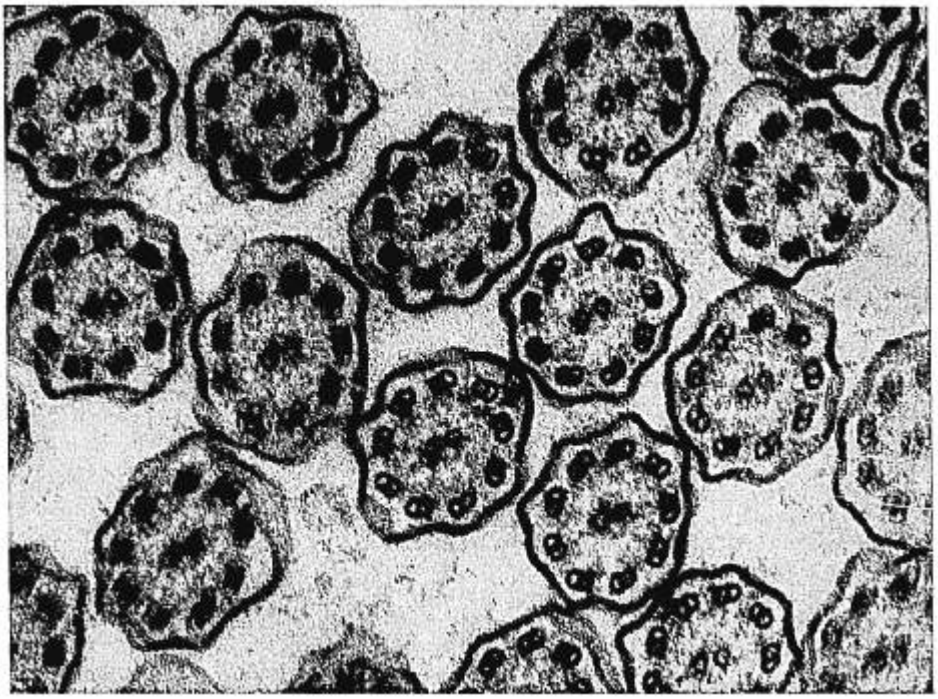

Fig. 4. Cross-section through cilia from the nasopharynx of patient with Kartagener's Syndrome (Case 4). Note absence of dynein arms. An occasional outer arm stub may be seen. Original magnification x 126,000.

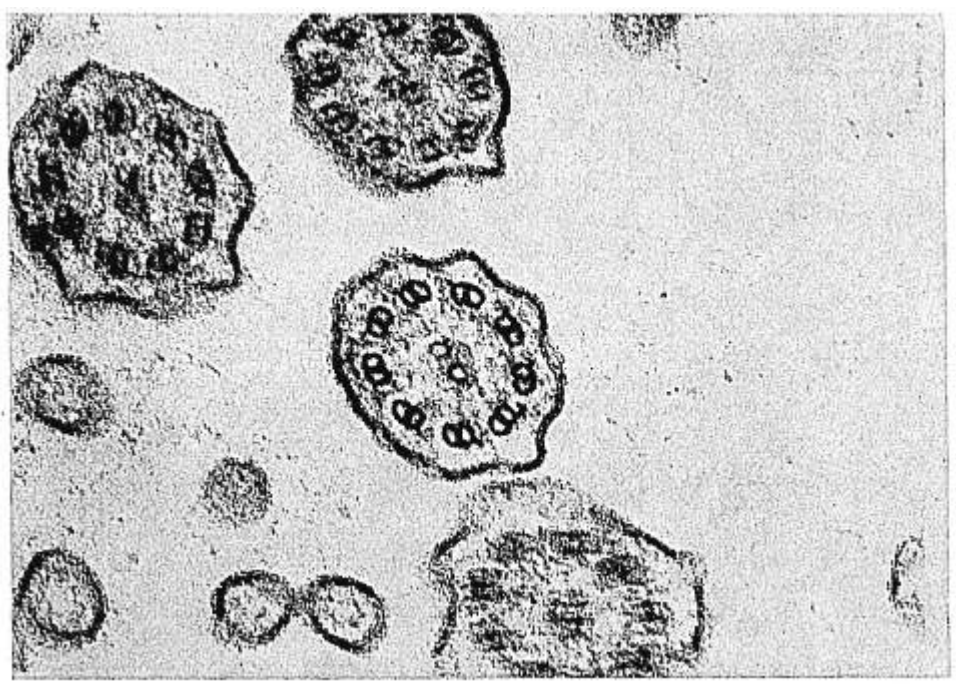

Fig. 5. Cross-section through cilia from the middle ear epithelium of patient with defective dynein arms (Case 1). Note short outer arm segment. Original magnification x 154.000.

Electron Microscopy.

Specimens were washed in sodium cacodylate and post fixed in osmic acid 2%. They were imbedded in Epon and thick sections were cut. Ultrathin sections were then cut and stained with uranyl acetate and Reynold's lead citrate. The sections were examined and photographed on Zeiss 9 S-2 and Philips 300 electron microscopes.

Interestingly, in Case 6, biopsies from three different sites — bronchus, nasopharynx and middle ear — failed to reveal cilia on electron microscopy. The patient's brother (Case 5) had his nasopharynx and middle ear biopsied at the same time using similar techniques of fixation and preparation. Cilia were easily demonstrated. It would seem unusual to have three separate biopsies fail to reveal cilia while microvilli and other epithelial ultrastructures were present. One may speculate that perhaps cilia were indeed absent. Unfortunately, light microscopy was not done and any inference as to the presence or absence of cilia would be premature. The child will be restudied.

Control Group.

Six patients not suspected of having immotile cilia gave informed consent for middle ear and nasopharynx biopsies while under general anesthesia for other, unrelated conditions. The middle ear mucosa was biopsied for cilia in all 6 patients and the nasopharynx was biopsied in 4 of 6 patients.

RESULTS.

Electron micrographs of cilia cross sections in the control group satisfactorily demonstrated both inner and outer dynein arms as well as a "hook" on the outer arm (Fig. 2 and 3). Spokes could be adequately seen and orientation of the paired central tubules was usually good, but not uniformly so. For each patient in either control or test group, the cilia were identical regardless of the site of biopsy. For example, if a defect was seen in cilia taken from the nasopharynx, the same defect was seen in cilia from the middle ear.

Dynein Arms.

In Case 4, the dynein arms were absent (Fig. 4). This patient had previously been diagnosed as having Kartagener's Syndrome. In Case 1, the outer dynein arms were defective, being represented by a short projection rather than the typical hook-like appearance seen in normal cilia (Fig. 5). The inner arms were difficult to see and may have been absent. In general, regardless of the patient group, inner dynein arms were more difficult to demonstrate than outer arms. Dynein arms in Cases 2, 3 and 5 were present.

Orientation of Central Tubules.

Electron micrographs of cilia from Cases 1 and 5 showed poor orientation of the central tubules (Fig. 6). The angle between central tubules of adjacent cilia was 90°. Case 4 showed only fair orientation. Of interest is the finding that the central tubules in cilia from one of the control patients was also poorly oriented.

Cilia in Case 5 were unusual in that they exhibited multiple central microtubules. In place of the usual 9 + 2 array, there were cilia with 8 + 4, 9 + 3, 9 + 5, and 9 + 7 combinations (Fig. 7). While this finding may be considered a deviation from normal, there is disagreement in the literature as to whether or not this should be classified as abnormal.[6,16] The results are summarized in Table III.

DISCUSSION.

This study was encouraged by the paucity of information concerning the middle ear in patients with immotile cilia. In man, cilia are commonly

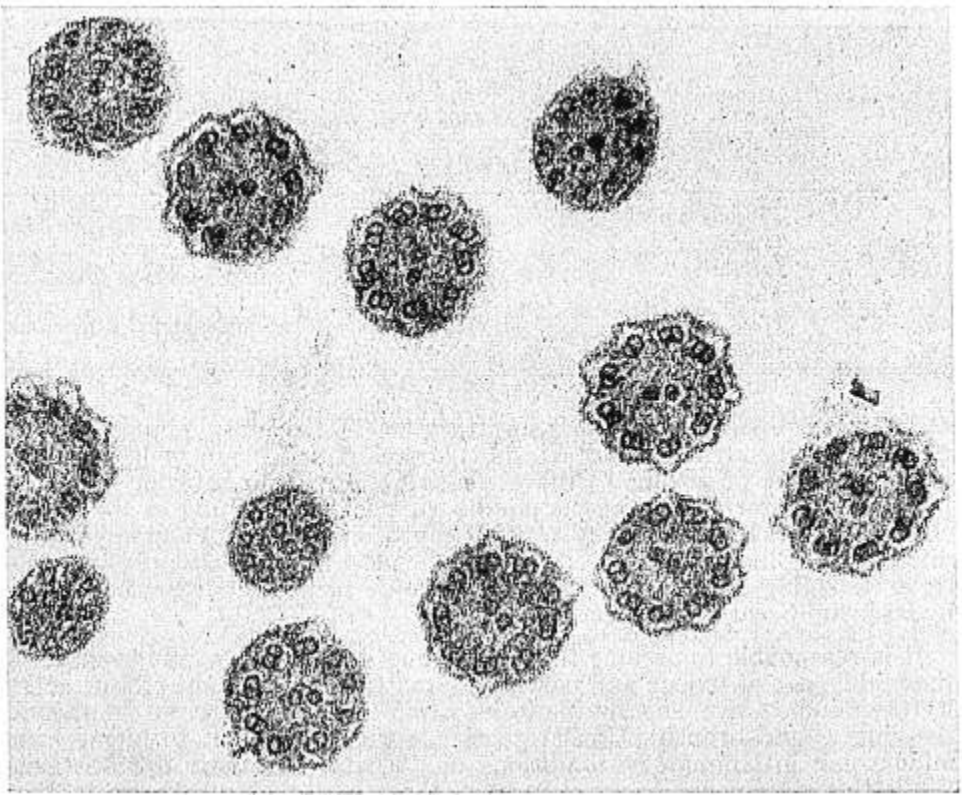

Fig. 6. Cross-section through cilia from the middle ear epithelium of same patient as shown in Fig. 5. Note poor orientation of central microtubules. Original magnification x 126,000.

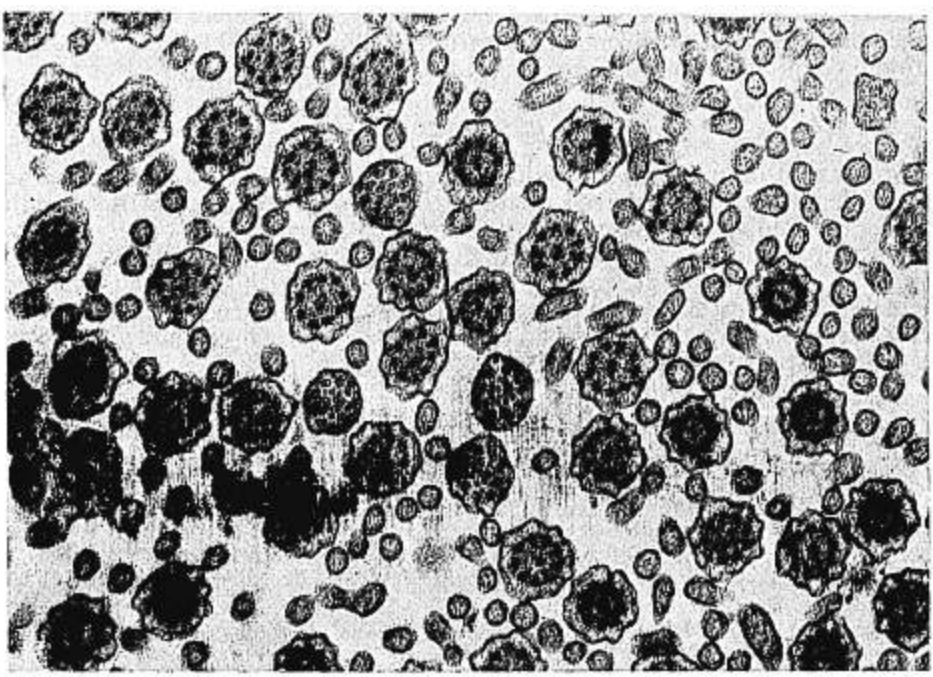

Fig. 7. Cross-section through cilia from the nasopharynx of a test group patient (Case 5). Note multiple central microtubules. Original magnification x 42,700.

TABLE III.
Ultrastructure of Cilia (Test Group).

Case	Dynein Arms	Orientation of Central Microtubules	Other
1	Patrially absent	Poor	
2	Present	Good	
3	Present	Good	
4	Absent	Fair	
5	Present	Poor	Multiple central microtubules
6	Cilia not seen		

found in the upper and lower respiratory tracts, paranasal sinuses, Eustachian tube, ependymal lining of the spinal cord and brain ventricles, and in the oviducts of women.[5] Cilia are also found in the anterior hypotympanum and anterior part of the middle ear near the opening of the Eustachian tube. Middle ear ciliary activity can be observed with the operating microscope. Ciliary motility can be documented by cinematography, video tape, recording of surface light reflections by photoelectric methods[17] and by laser light-scattering spectroscopy.[18]

It is reasonable to assume that ciliary motility is important in clearing the middle ear of mucus and particulate matter, and without ciliary activity the middle ear is susceptible to infection in much the same manner as the sinuses and bronchi. Until recently, it was convenient to blame most middle ear inflammatory conditions on "Eustachian tube dysfunction." While this statement is correct in part, there is probably more to it. It is not enough for the contemporary physician to know that cilia are normally present in the middle ear and Eustachian tube, and that ciliary motility is an excellent indicator of a healthy middle ear. He must be aware that there exists a congenital syndrome of non-functioning cilia which may place the middle ear at risk and dispose the patient to chronic ear infections.

CONCLUSION.

Specimens of ciliated epithelium from 6 patients suspected of having immotile cilia were compared to cilia from 6 control patients. Biopsy sites were the middle ear, nasopharynx, maxillary antrum, and bronchus. Two of 6 test patients showed absent or defective dynein arms on electron microscopy. The results of this study are in accord with previous investigations and affirm the validity of the Immotile Cilia Syndrome. Patients with sinusitis, bronchiectasis and otitis media, with or without situs inversus, should be suspected of having immotile cilia.

BIBLIOGRAPHY.

1. ELIASSON, R., ET AL.: The Immotile Cilia Syndrome. *N. Engl. J. Med.*, 297:1-6, 1977.

2. MOSSBERG, B. and HANNGREN, A.: Kartagener's Syndrome — A Ciliary Immotility Syndrome. *Mt. Sinai J. Med.*, 44:837-843, 1977.

3. CAMNER, P., MOSSBERG, B. and AFZELIUS, B. A.: Evidence for Congenitally Nonfunctioning Cilia in the Tracheobronchial Tract in Two Subjects. *Am. Rev. Respir. Dis.*, 112:807-809, 1975.

4. PEDERSEN, H. and MYGIND, N.: Absence of Axonemal Arms in Nasal Mucosa Cilia in Kartagener's Syndrome. *Nature*, 262:494-495, 1976.

5. AFZELIUS, B. A.: A Human Syndrome Caused by Immotile Cilia. *Science*, 193:317-319, 1976.

6. LUPIN, A. J. and MISKO, G. J.: Kartagener's Syndrome with Abnormalities of Cilia. *J. Otolaryngol.*, 7:95-102, 1978.

7. HARTLINE, J. V. and ZELKOWITZ, P. S.: Kartagener's Syndrome in Childhood. *Am. J. Dis. Child.*, 121:349-352, 1971.

8. FISCHER, T. J., ET AL.: Middle Ear Ciliary Defect in Kartagener's Syndrome. *Pediatrics*, 62:443-445, 1978.

9. SATIR, P.: Studies on Cilia. III. Further Studies on the Cilium Tip and a "Sliding Filament" Model of Ciliary Motility. *J. Cell. Biol.*, 39:77-94, 1968.

10. AFZELIUS, B. A., ET AL.: Lack of Dynein Arms in Immotile Human Spermatozoa. *J. Cell. Biol.*, 66:225-232, 1975.

11. WARNER, F. D., MITCHELL, D. R. and PERKINS, C. R.: Structural Conformation of the Ciliary ATPase Dynein. *J. Mol. Biol.*, 114:367-384, 1977.

12. SUMMERS, K. E. and GIBBONS, I. R.: Effects of Trypsin Digestion on Flagellar Structures and Their Relationship to Motility. *J. Cell. Biol.*, 58:618-629, 1973.

13. KERJASCHKI, D.: The Central Tubuli in Distal Segments of Olfactory Cilia Lack Dynein Arms. *Experientia.*, 32:1459-1460, 1976.

14. STURGESS, J. M., ET AL.: Cilia with Defective Radial Spokes. A Cause of Human Respiratory Disease. *N. Engl. J. Med.*, 300:53-56, 1979.

15. KATZ, M. S., ET AL.: Kartagener's Syndrome and Abnormal Cilia. *N. Engl. J. Med.*, 297: 1011-1012, 1977. (Correspondence.)

16. AFZELIUS, B. A.: Kartagener's Syndrome and Abnormal Cilia. *N. Engl. J. Med.*, 297:1012, 1977. (Correspondence.)

17. REIMER, A., ET AL.: The Mucociliary Activity of the Upper Respiratory Tract. I. A Method for Use in Experimental Studies on Human Material. *Acta Otolaryngol.*, 83:491-497, 1977.

18. LEE, W. I. and VERDUGO, P.: Ciliary Activity by Laser Light-Scattering Spectroscopy. *Ann. Biomed. Eng.*, 5:248-259, 1977.

DISCUSSION.

CHARLES D. BLUESTONE, M.D., (Pittsburgh, Pa.): Dr. Jahrsdoerfer and his co-workers should be congratulated for their continuing interest in the ultrastructure of cilia in patients with generalized inflammation of the respiratory tract, which naturally includes the middle ear. The present study confirms the recent report by Fisher, *et al.*,* that abnormal cilia are present in the middle ear of patients with Kartagener's Syndrome, which appears to be a subgroup of the somewhat more prevalent Immotile Cilia Syndrome.

I was particularly interested in the authors' conclusion that this finding may play an important role in the pathogenesis of otitis media rather than "Eustachian tube dysfunction." In an attempt to investigate this assumption, we recently had the opportunity to test the Eustachian tube function in two children (ages 12 and 13) with Kartagener's Syndrome. Both had had myringotomy and tympanostomy tubes inserted for chronic otitis media with effusion. Two weeks after the surgical procedure, the hearing had returned to normal and their middle ears were dry. This latter finding is no doubt related to the change in middle ear-nasopharynx pressure relationships following insertion of tympanostomy tubes (non-intact tympanic membrane) so that the middle ear secretions drain down the Eustachian tube even in the absence of a normal mucociliary transport system. At this visit, the two children had the standard inflation-deflation Eustachian tube function test and also a new function study, the forced response test. This latter test is unique in that it eliminates the "mucous forces" in the lumen of the Eustachian tube that may interfere with the results of the inflation-deflation test when attempting to assess the active opening mechanism, *i.e.*, contraction of the tensor veli palatini muscle, and the compliance of the tube. This is accomplished by forcing the Eustachian tube open with a constant flow of air from the middle ear end of the tube and then assessing tubal resistance before and during active dilation of the tube during swallowing activity. Both children had abnormal inflation-deflation studies, and one had abnormal active tubal opening, as demonstrated by the forced response test. However, the other child had *normal* active dilation of the Eustachian tube during the forced response test.

From these findings and our other studies, it appears that the vast majority of patients with otitis media have abnormal function of the Eustachian tube which is primarily due to functional obstruction secondary to an abnormal opening mechanism or increased compliance or both; or, less commonly, due to intrinsic or extrinsic mechanical obstruction. Abnormal patency of the Eustachian tube also has been a frequent finding. Eustachian tube dysfunction can then

*Middle Ear Ciliary Defect in Kartagener's Syndrome. Pediatrics, 62:443-445, 1978.

result in secondary middle ear mucous membrane disease. A much smaller number of patients may have *primary* mucosal disease of the middle ear, which has been purported to be caused by allergy (controversial and unproven) or, as described in the present paper, due to an abnormality of the cilia. However, as demonstrated by our Eustachian function studies in the two children with Kartagener's Syndrome, primary mucosal disease may also affect Eustachian tube function.

Since the overwhelming number of patients with otitis media do not have generalized respiratory tract mucous membrane disease, the Immotile Cilia Syndrome related to the pathogenesis of otitis media should be considered as a rare entity. However, the condition should be considered when otitis media is associated with sinusitis, bronchitis and bronchiectasis.

Finally, I would like to ask Dr. Jahrsdoerfer if the control subjects had otitis media, or did they have normal middle ears, since studies of the cilia in patients with otitis media in whom the other stigmata of the Immotile Cilia Syndrome are absent would seem to be the next logical step in investigating this interesting finding.

DR. JAHRSDOERFER: I thank Dr. Bluestone for his comments. None of the patients in the control group had active infectious or inflammatory middle ear disease at the time of biopsy. The middle ear mucosa appeared normal in every control group case.

SPANISH AMERICAN MEDICAL SOCIETY.

This 50-year-old organization of Spanish speaking physicians in the metropolitan area, will sponsor The III Interamerican Medical Congress in New York City at the Americana Hotel on October 5-8, 1979, celebrating: "El Dia De La Raza" (Columbus Day.)

The faculty has prominant physicians from the Spanish speaking countries as well as American physicians.

This Congress has 20 hours' credit for C.M.E. and will be held annually: October 10-13, 1980; October 9-12, 1981; October 8-11, 1982; and October 7-10, 1983.

For further information, contact: Dr. Rene F. Rodriguez, Chairman, 37-21 75th Street, Jackson Heights, New York 11372.