Memoranda/Mémorandums

Therapeutic approaches to cystic fibrosis: Memorandum from a joint WHO/ICF(M)A meeting*

Cystic fibrosis is one of the commonest genetic diseases among Caucasians and represents an important cause of suffering and death among children and adults. In the past two decades marked prolongation of the life of patients with cystic fibrosis has been achieved as the result of improved case-finding and an extensive regimen of therapies. More recently, a variety of new approaches to therapy have been developed or proposed as the result of advances in cell physiology and molecular biology. This article summarizes the presentations and discussions made at a joint WHO/ICF(M)A (International Cystic Fibrosis (Mucoviscidosis) Association) meeting, held in Washington, DC, on 14 October 1992, and reviews the current status of possible therapies for cystic fibrosis and their implications for treatment in various countries of the world.

Introduction

The geographical distribution, control, and screening of cystic fibrosis have been the subject of several joint meetings between WHO and the International Cystic Fibrosis (Mucoviscidosis) Association (ICF(M)A).[a]

In 1989 the cystic fibrosis gene was discovered, which created the possibility of genetically modifying the disease. Subsequently, there has been considerable progress in understanding the pathogenesis of cystic fibrosis.[b]

The life expectancy of those with cystic fibrosis (CF) has increased in many countries probably as the result of increased availability of medication, overall rigorous care, and diagnosis of the milder forms of the disease.[c] There is some uncertainty as to whether there is a difference in survival rate between males and females. The early treatment of newborns screened for CF and well-organized CF centres appear to be important elements in the care of patients in certain countries. Frequent regular visits to such centres lead to more coordinated and careful care and seem to be correlated with better outcomes; as a result, the current median survival of CF patients is about 30 years.

* This Memorandum is based on the report of a joint WHO/International Cystic Fibrosis (Mucoviscidosis) Association (ICF(M)A) Meeting, held in Washington, DC, on 14 October 1992. The participants at the meeting were as follows: F. de Abreu e Silva, Porto Alegre, Brazil; V. Baranov, St. Petersburg, Russian Federation, G.J. Barbero (*Rapporteur*), Columbia, MO, USA; S.L. Brody, Bethesda, MD, USA; G. Döring, Tübingen, Germany; M. Götz (*Chairman*), Vienna, Austria; M.E. Hodson, London, England; N. Hoiby, Copenhagen, Denmark; N. Kapranov, Moscow, Russian Federation; M.R. Knowles, Chapel Hill, NC, USA; L. Lannefors, Lund, Sweden; G. Mastella, Verona, Italy; M. Noirclerc, Marseilles, France; P.M. Quinton, Riverside, CA, USA; D.J. Shale, Cardiff, Wales; R. Shepherd, Herston, Australia; R. Williamson, London, England. *ICF(M)A Secretariat*: M. Weibel. *WHO Secretariat*: V. Boulyjenkov (*Secretary*).

Requests for reprints should be sent to Hereditary Diseases Programme, Division of Noncommunicable Diseases, World Health Organization, 1211 Geneva 27, Switzerland. A French translation of this article will appear in a later issue of the *Bulletin*.

[a] *Report of a Joint WHO/ICF(M)A Meeting on Prevention and Control of Cystic Fibrosis, Oslo, 19 June 1987.* Unpublished WHO document HDP/ICF(M)A/WG/87.3, 1987; Feasibility study of community control programmes for cystic fibrosis: Memorandum from a WHO/ICF(M)A meeting. *Bulletin of the World Health Organization*, 1990, **68**: 709–715; *Report of a Joint WHO/ICF(M)A Task Force on Cystic Fibrosis, Leningrad/Moscow, 26–29 November 1990.* Unpublished document WHO/HDP/ICF(M)A/TF/90.4, 1990.

[b] **Riordan JR et al.** Identification of the cystic fibrosis gene: cloning and characterization of complementary DNA. *Science*, 1989, **245**: 1066–1073.

[c] **Dodge JA et al.** Cystic fibrosis in the United Kingdom 1977–85: an improving picture. *British medical journal*, 1988, **297**: 1599; **Padoan R et al.** Survival in cystic fibrosis: prognostic factors in the Italian population. *Rivista Italiana de pediatria*, 1991, **17**: 669; **Wilmott RW et al.** Cystic fibrosis survival rates. *American journal of diseases of children*, 1985, **139**: 669.

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Bacterial infections[d]

Antibiotic treatment

CF patients frequently have repeated or chronic lung infections, which produce the major morbidity and mortality caused by the disease. Intensive antibacterial therapy has contributed to an improved prognosis.

Patients with CF have a propensity to develop colonization of the respiratory tract with *Staphylococcus aureus*, in the earlier stages, and later with *Pseudomonas aeruginosa*. Careful long-term controlled studies with antibiotics have been difficult to carry out; therefore, many of the antibiotic regimens currently in use have resulted from clinical experience and observations on the longer preservation of lung function, improved clinical status, and extension of life.

The use of antibiotics is related to understanding the course of lung disease in CF. During infancy or beyond it is often quiescent, and children with the disease experience the usual respiratory problems like others in their local community. As such children get older, however, they have increased cough and respiratory signs, and cultures of samples from the upper respiratory tract may show *S. aureus*. At some stage this usually changes to intermittent or chronic colonization with *P. aeruginosa*, which is virtually impossible to eradicate using intensive antibiotic therapy. The microbial population may markedly increase during the acute respiratory exacerbations frequently arising from viral infections. Antibiotics are usually used during these periods of respiratory exacerbation and increased bacterial flora.

A number of key principles for antibiotic therapy are important in the pulmonary infection of CF patients, and differ from the treatment used for other short-term respiratory infections among normal individuals (Table 1); emphasis should be placed on microbial diagnosis, high dosage, and prolonged courses of antibiotics. The pharmacokinetics for some antibiotics is altered, particularly as a result of increased renal excretion. Inhaled antibiotics also have some clinical benefit as an adjunct or replacement for systemic chemotherapy. Table 2 illustrates the programme of antibiotic administration as it relates to sputum bacteriology.

Oral treatment should be used for *S. aureus* infections. They are not always eradicated even with intensive therapy. Some of the early respiratory damage among CF infants is caused by *S. aureus*, which provides a basis for subsequent *P. aeruginosa* colonization. *Haemophilus influenzae* infection may become chronic in some patients and is found moderately frequently during acute exacerbation; a useful treatment is a combination of amoxicillin with beta-lactamase inhibitors, or ciproflaxacin.

Treatment of pulmonary *P. aeruginosa* infections of CF patients is intensive, usually involving a combination of two intravenous antibiotics, most commonly tobramycin with ticarcillin or piperacillin, in high dosages to achieve blood levels sufficient to penetrate the intraluminal respiratory tract of the lungs. In Denmark an intensive preventive approach to chronic *Pseudomonas* persistence has been pioneered by treating patients every three months. This pattern of management has markedly changed the process of pulmonary progression among Danish CF patients. During episodes of severe chronic illness aerosol tobramycin or colistin can help to stabilize the clinical state and decrease the frequency of exacerbations requiring hospitalization.

Although some studies have shown little difference between groups treated with antibiotics or placebos, their findings have not been convincing enough to alter the rigorous therapeutic intervention against *P. aeruginosa* in most centres. The value of antibiotics is reinforced by the improved mortality seen in those centres that use more aggressive anti-pseudomonal treatment.

In recent years *P. cepacia* infections have occurred in some centres and have been accompanied by considerably increased deterioration in lung function

Table 1: **Principles of chemotherapy for lung infections in cystic fibrosis patients**

- Carry out microbial diagnosis based on secretions from the lower respiratory tract before initiating chemotherapy.
- Administer high doses of bactericidal antibiotics for 14 days.
- Preferably use antibiotics when resistant variants are rarely encountered or use combinations of antibiotics.
- Avoid prophylactic chemotherapy.
- Be aware of the cumulative side-effects resulting from frequent use of antibiotics.
- Be aware of the altered pharmacokinetics of some antibiotics when administered to CF patients, especially increased renal excretion.
- Remember that inhalation of antibiotics may be useful as a support or replacement for systemic chemotherapy.

[d] **Hoiby N.** Prevention and treatment of infections in cystic fibrosis. *International journal of antimicrobial agents*, 1992, **1**: 229–238. **Jensen T. et al.** Use of antibiotics in cystic fibrosis: the Danish approach. In: Hoiby N et al., eds. Pseudomonas aeruginosa *infection*. Basel, Karger, 1989: 237–246 (Antibiotics and chemotherapy, vol. 42). **Marks MI.** Antibiotic therapy for bronchopulmonary infections in cystic fibrosis: the American approach. In: Hoiby N et al., eds. Pseudomonas aeruginosa *infection*. Basel, Karger, 1989: 229–236 (Antibiotics and chemotherapy, vol. 42).

Table 2: **Antibiotics used to treat lung infections in cystic fibrosis patients**

Infecting organism	
Staphylococcus aureus	Oral dicloxacillin (25 mg per kg per 24 hours) + fusidic acid (50 mg per kg per 24 hours) The following alternative drugs can replace one of the above: flucloxacillin, rifampicin and clindamycin.
Haemophilus influenzae	Oral pivampicillin (35 mg per kg per 24 hours) or amoxicillin (25–50 mg per kg per 24 hours) Alternative drugs: amoxicillin + clavulanate or rifampicin in combination with erythromycin.
Pseudomonas aeruginosa[a]	Oral ciprofloxacin (20–30 per mg per kg per 24 hours) + aerosolized colistin ($2–3 \times 10^6$ units per 24 hours).
P. aeruginosa[b]	Intravenous (or aerosolized) tobramycin (10–20 (30) mg per kg per 24 hours) + piperacillin (300 mg per kg per 24 hours) or + cefsulodin (100–150 mg per kg per 24 hours) or + ceftazidime (150–250 mg per kg per 24 hours) or + aztreonam (150–250 mg per kg per 24 hours) or + thienamycin 50–75 mg per kg per 24 hours) and + aerosolized colistin ($2–4 \times 10^6$ units and/or + ciprofloxacin (20–40 mg per kg per 24 hours)
Probenecid is given orally to all patients receiving betalactam antibiotics eliminated by tubular excretion	

[a] Intermittently colonized.
[b] Chronically colonized.

and patient status. *P. cepacia* develops antibiotic resistance more readily than *P. aeruginosa*: chronic suppression with high doses of doxycycline may diminish some symptoms. The approach to *P. cepacia* in some centres has been to separate those patients with this condition from regular contact with other patients who have *P. aeruginosa* infection. This step has been found to increase recovery from *P. cepacia* and suggests that cross-infection occurs. In an attempt to minimize cross-infection the Danish centre has separated its patient population into the following categories: those without *Pseudomonas* infections; those with *P. aeruginosa*; those with multiple-resistant *P. aeruginosa*; and those with *P. cepacia*. Some preliminary data suggest that such measures may help to prevent the development of more severe *Pseudomonas* infection. Certainly direct skin contact or kissing should be avoided among patients in these groups.

Other organisms such as atypical *Mycobacteria* spp., *Klebsiella* spp., and *Proteus* spp. may be isolated from CF patients and warrant treatment when present. *Aspergillus fumigatus* is not uncommonly cultivated from the sputum of CF patients and allergic bronchopulmonary aspergillosis develops in some; this can be treated with prednisone.

The cost of antimicrobial chemotherapy for CF patients can be high and is a major problem where resources are limited, as illustrated in Table 3 for Denmark. Such costs usually exceed normal familial resources and require support from other public or private systems. The high cost of antibiotic programmes as well as the extreme demands on the patients highlight the need for newer approaches to diminish the vulnerability of those with CF to pulmonary bacterial colonization. Early preventive efforts, the use of aerosols or oral agents, and reduction of the costs of drugs by arrangement with drug companies may be areas for development.

Immunology[e]

The immune system of CF patients responds to chronic bacterial lung infections by producing specific antibodies against many bacterial antigens, forming immune complexes, rapidly recruiting neutrophils from the bloodstream, and producing cytokines. During this type-III hypersensitivity reaction, lysosomal enzymes are released that are held responsible for tissue damage. Neutrophil elastase (NE) is by far the best-studied of these enzymes and may reach concentrations of >100 µg/ml of sputum. About 90% of the endogenous inhibitor for NE, α_1-proteinase inhibitor, is locally inactivated by NE and probably also by oxidative attack. Thus, a high imbalance between proteinases and proteinase inhibitors is present in the inflamed lungs of these patients. The pathological effects of free NE, demonstrated in a number of *in vitro* and *in vivo* investigations, include cleavage of fibronectin, lung elastin, immunoglobulins and immune complexes, complement, complement receptors on neutrophils, and other

[e] **Döring G, Knight R, Bellon G.** Immunology of cystic fibrosis, In: Hodson M, Geddes D, eds. *Textbook of cystic fibrosis.* London, Chapman & Hall, 1993.

Table 3: **Cost of antimicrobial chemotherapy in Denmark**[a]

Regimen	Cost per patient per year (US$)
Antipseudomonas treatment, intravenous (4 courses per annum)	7 400
Antipseudomonas, aerosol (colistin, 365 days per annum)	2 500
Antistaphylococcal treatment (oral)	640
Antihaemophilus treatment (oral)	60
Probenecid (oral)	10
Total	10 610

[a] Data provided by the Danish Cystic Fibrosis Centre.

receptors on T-cells and B-cells. Furthermore, NE inhibits ciliary beating and stimulates mucus production from goblet cells and facilitates *P. aeruginosa* adherence. Finally, by cleaving receptors for interleukin (IL)-1 and IL-2 or the T-cell antigen receptor, it may hypothetically inhibit message transmission and immune recognition. Thus, besides destruction, chronic infection may lead to acquired immune suppression. These topics clearly should be investigated in greater detail in order to design more specific treatment strategies to protect the lung tissues of CF patients.

Prevention[f]

Prevention of bacterial colonization of the lungs is another strategy for early treatment of the CF host. Active immunization against *P. aeruginosa* with an exotoxin A–polysaccharide conjugate is currently being studied in non-infected CF children at an early age. Additionally, two passive immunization studies were recently carried out using intravenous gammaglobulin preparations. In both studies transient improvement in lung function was noticed in the treatment group, and larger studies should be carried out to validate these data.

Bacterial transmission routes, in general, and especially within the hospital between patients, or between patients and healthy carriers (e.g., hospital personnel), and patients and environmental sources have been investigated using reliable and highly discriminatory typing methods for *P. aeruginosa* and other bacteria. Normal hand washing without appropriate disinfection thereafter may lead in some cases to contamination with microorganisms, particularly *P. aeruginosa*. Therefore, hygienic measures to decontaminate wash-basins and toilets have been recommended as well as improved hygienic measures for hand disinfection. Such approaches may include the installation of heating devices in hospital sink drains. An improved heating device has recently been developed and is currently under clinical investigation. However, improvements in the methods of dispensing and the use of disinfectants, isolation of infected patients, and improvements in aseptic techniques, notably the use of gloves for many nursing procedures, are successful in reducing *P. aeruginosa* infections in hospitals.

Respiratory diseases—novel therapies[g]

The manifestations of CF in the lung are complex and change during the evolution of the disease. The primary problem involves airway epithelial electrolyte and fluid balance, which reduces clearance of airway secretions and leads to abnormalities in the airway surface microenvironment. This is probably most important early in life, while later the epithelium is damaged, perhaps by viral infection, leading to inflammation which results in predisposition to a vicious cycle of bacterial infection and continuing host defence-based injury. The current policies of antibiotic treatment for the three main bacteria causing injury (*S. aureus, H. influenzae* and *P. aeruginosa*) have only a small impact on halting the progression of lung destruction.

A range of treatments has been proposed to address the two important aspects that contribute to lung destruction in CF:

— agents able to improve clearance of secretions from the lung; and

— anti-inflammatory or anti-injury mediator therapy.

There are a variety of therapeutic agents available that are designed to normalize defective electrolyte transport. However, in patients with established airway infections one of the major factors leading to decreased secretion clearance is the presence of host-derived DNA. Although the thickened secretions consist of those normally produced by patients with CF and host-derived products of plasma exudation in response to infection, a major contribution is DNA derived almost entirely from disintegrated inflamma-

[f] **Döring G et al.** Generation of *Pseudomas aeruginosa* aerosols during hand washing from contaminated sink drains, transmission to hands of hospital personnel, and its prevention by use of a new heating device. *Zentralblatt für die gesamte Hygiene*, 1991, **191**: 494–505.

[g] **Hubbard RC et al.** A preliminary study of aerosolized recombinant human deoxyribonuclease I in the treatment of cystic fibrosis. *New England journal of medicine*, 1992, **326**: 812–815; **Shak S et al.** Recombinant human DNase I reduces the viscosity of cystic fibrosis sputum. *Proceedings of the National Academy of Sciences of the USA*, 1990, **87**: 9188–9192.

tory cells, e.g., neutrophils. DNA inherently forms thick viscous gels, and interaction with mucus already present in the airways leads to extremely adherent secretions. DNA may reach high concentrations (up to 15 g/l) in sputum. Studies with aerosolized, recombinant human DNase (rhDNase) have suggested that cleaving DNA in purulent lung secretions may facilitate CF patients to improve their airways clearance. The patients taking part in these studies reported improvements in their breathing ability while receiving rhDNase and showed objective improvements in both forced vital capacity (FVC) and forced expiratory volume in one second (FEV_1), compared with baseline values. The *in vivo* biological activity of the aerosolized DNase product was demonstrated by detection of cleaved DNA in the sputum.

A variety of other drugs such as ibuprofen (a broad spectrum anti-inflammatory agent) and corticosteroids are of interest in suppressing the host inflammatory response in CF patients. Recently, more specific compounds capable of antagonizing proteases, such as α_1-antitrypsin, serum leukocyte protease inhibitor (SLPI), and ICI 200 800 have been studied in patients with CF. Of further interest would be the use of agents such as specific anticytokines, e.g., antitumour necrosis factor (TNFα) or anti-IL2 or IL8, which may have more profound effects on the inflammatory process. These substances can be specifically designed as receptor-blocking agents, or possibly humanized monoclonals capable of stopping the action of cytokines, or other agents, e.g., pentoxifylline.

The value of corticosteroids remains to be proved despite an earlier report that they preserved lung function and reduced immunoglobulin levels in children. Recent studies with a higher dose have had to be stopped, however, because of unacceptable side-effects, while a lower-dose study continues. Pentoxifylline is promising as an anticytokine capable *in vivo* of inhibiting the transcription of the TNFα gene. Hence, this may go some way towards reducing the inflammatory process within the lungs, but more research needs to be carried out. A side-effect of such an agent may be better control of the nutritional state of CF patients since TNFα may contribute to some of the cachexia associated with infection.

The use of antielastases is of special interest in preventing destruction of lung tissue. Nebulized human α_1-antitrypsin has been delivered to the airways of CF patients in an attempt to correct the imbalance of the protease-anti-protease components in epithelial lining fluid. A preliminary study demonstrated that α_1-antitrypsin could be safely delivered to the airways and produce biologically active antielastase function in the airways. A side-effect noted in the initial study was that neutralizing neutrophil elastase in the environment around neutrophils enhanced their ability to kill *P. aeruginosa in vitro*.

Therapy with oxygen radical scavengers is another potential way to reduce inflammation. A combination of superoxide dismutase and catalase is needed to reduce completely the superoxide radical anion. Markers for oxidative lung damage have to be developed.

At an early phase, it would seem worthwhile to treat CF patients who are already infected and undergoing continuous lung injury, both from the point of view of improving clearance and of dealing with the consequences of the host inflammatory response, in addition to traditional antibacterial therapy and physiotherapy. It may thus be possible to enhance protection of the lung and possibly affect long-term survival. Larger and longer studies need to be carried out in order to define inhibitor deposition after aerosolization, the degree of proteinase inhibition, aerosolization time, and other parameters. In particular, markers for lung tissue damage by neutrophil elastase or other proteinases have to be developed.

Physiotherapy[h]

Pulmonary hypersecretion that blocks airways and paves the way for chronic bacterial colonization with frequent exacerbations is one of the main problems caused by CF, since it results in airway obstruction and destruction of the lung parenchyma. Eventually, breathing muscles become overexerted because of gradually increased pulmonary obstruction. Malnutrition, inactivity, frequent infections and the heavy work load of breathing lead to muscle wasting and poor endurance. Without an optimal treatment, including efficient chest physiotherapy, each infection results in further physical impairment.

The objective of regular chest physiotherapy is to make leading a normal life possible for CF patients, by counteracting exacerbations and optimizing the body's physical function. The frequent pulmonary exacerbations are counteracted by improving airway clearance with ensuing maintenance or improvement of lung function and diminution of airway destruction due to removal of secretions containing proteolytic enzymes. The physical function of the body is optimized by physical exercises of different kinds. Chest physiotherapy is considered to be an integral part of the treatment.

[h] **Lannefors L, Wollmer P.** Mucus clearance with three chest physiotherapy regimens in cystic fibrosis: a comparison between postural drainage, PEP and physical exercise. *European respiratory journal*, 1992, **5**: 748–753; **Oberwaldner B, Evans JC, Zach MS.** Forced expirations against a variable resistance: a new chest physiotherapy method in cystic fibrosis. *Pediatric pulmonology*, 1986, **2**: 358–367.

Different techniques to improve airway clearance have been developed throughout the world. The results of short- and long-term studies of mucus clearance techniques have been somewhat controversial. Current techniques used to loosen, transport, and evacuate pulmonary secretions are listed below.

- Active cycle-of-breathing techniques. A cycle consists of postural drainage, thoracic expansion exercises, forced expirations and breathing controls; it can be carried out with or without percussion.
- Autogenic drainage; different modified ways exist.
- Positive expiratory pressure (PEP) used in different ways:
  - PEP combined with forced expirations and breathing controls;
  - High-pressure PEP; and
  - oscillating PEP.
- Physical exercise interspersed or combined with forced expirations and breathing controls.

Evaluation of different physiotherapy methods shows that a highly individualized approach is necessary and that there is no one universally accepted optimal method. Combined use of the various methods is the rule. All the different techniques can be used either exclusively, combined, or mixed. Factors such as age, condition of the lungs, mucosal swelling, airway reactivity, airway stability, treatment motivation, culture, individual interest, surroundings, time of the day, etc., affect the content of each chest physiotherapy programme. Over time the most suitable techniques usually change depending on, e.g., disease stage. Only by jointly involving parents, partners, patients, and therapists can chest physiotherapy be established as a set routine of daily life and thereby of the treatment. The programme tailored for each individual must be designed in such a way that it is likely to be carried out. All youngsters should be educated to perform the daily treatment independently in an efficient way. Independence is to be viewed as a possibility where the programme performed and techniques used are checked at regular intervals and changed whenever needed.

Use of nebulizing therapy with bronchodilators and/or mucolytics should precede or intersperse each chest physiotherapy session, if prescribed, and is of benefit to the patient. If nebulized corticosteroids or antibiotics are prescribed they should be taken after chest physiotherapy; the choice of nebulizer and training for the inhalation technique are essential to obtain the optimum result. Most chest physiotherapy techniques and physical exercises can be performed without special devices. However, a device or a system made for the patient can be of value; there are a number of such products available commercially but most can be custom-made. In a few cases it is difficult to obtain the results expected without using the specific device developed for the technique concerned.

The success of physiotherapy should be monitored using regular pulmonary function tests along with oxygen measurements. Untoward side-effects caused by chest physiotherapy are infrequent. All techniques and devices used are continually being evaluated and developed and new ones are occasionally introduced; however, designing and performing controlled studies is difficult. Better objective methods to evaluate current treatment are needed. Comparative data are scarce and many questions remain to be answered; for example, the following:

- Is there a logical chronological order for introduction to various physiotherapy techniques according to patients' ages?
- What are the comparative short- and long-term benefits of physiotherapy, according to patients' characteristics?
- What type of physiotherapy suits best under special circumstances (malnutrition, cor pulmonale, pneumothorax, etc.)?
- What are the optimal criteria for successful physiotherapy?
- How can acceptance of and compliance with physiotherapy be improved?

Lung transplantation[i]

The first successful heart–lung transplant performed on a CF patient was carried out in the United Kingdom in 1984. Since then, approximately 180 CF patients have received such transplants worldwide, with encouraging results. During this time other options for lung transplantation have become available. Single-lung transplantation is unsuitable since the native lung would be a source of infection. One-block, double-lung transplantation with revascularization requires further evaluation. The 1-year actuarial survival for 79 patients after heart-lung transplantation at Harefield Hospital in England was 69%. The Papworth English series of 34 patients,

[i] **Frist WH et al.** Cystic fibrosis treated with heart–lung transplantation: North American results. *Transplantation proceedings*, 1991, **23**: 1205–1206; **Madden BP et al.** Intermediate term results of heart-lung transplantation for cystic fibrosis. *Lancet*, 1992, **339**: 1583–1587; **R de Leval M. et al.** Heart and lung transplantation for terminal cystic fibrosis: a 4 1/2-year experience. *Journal of thoracic and cardiovascular surgery*, 1991, **101**: 633–642; **Shennib H et al. and the Cystic Fibrosis Transplant Study Group.** Double-lung transplantation for cystic fibrosis. *Annals of thoracic surgery*, 1992, **54**: 27–32.

who were more selected, had an actuarial survival of 79% at 1 year and 66% at 2 years. A North American series had an actuarial survival of 42% at 1 year. In Toronto the 1-year survival of 17 patients after bilateral, single-lung transplantation was 58%. The University of North Carolina has performed bilateral lung transplants on 27 CF patient (100% operative survival) resulting in a 1-year survival rate of 90%.

The demand for transplantation is far greater than the availability of organs. It is therefore essential that patients are properly assessed so that no one receives a transplant without a proper trial of maximum medical treatment. Indications for transplantation are deteriorating chronic respiratory failure, despite maximum medical treatment, a severely impaired quality of life, and a life expectancy of less than 18 months. The patients must also want to have a transplant. Among the contraindications are high-dose corticosteroids, psychosocial instability, infection with mycobacteria or aspergillus, other end-organ failure, and gross malnutrition. Factors known to increase the risks are previous thoracic surgery, pleurodesis, ventilation, and severe liver insufficiency. The major matching criteria are donor size, patient size, blood group, and cytomegaloviral status. Patients require a very detailed medical and psychological assessment before transplant surgery. The routine immunosuppressants are cyclosporin and azathioprine, with steroids being used for acute episodes of rejection. Major postoperative problems are haemorrhage, multiorgan failure, infection, rejection, and obliterative bronchiolitis; the reported incidence of obliterative bronchiolitis is about 30%, increasing with the time elapsed after surgery. There are specific challenges when a CF patient receives a transplant, which the surgical team does not encounter with other patients, and all transplant teams treating CF patients should include a physician experienced in the medical treatment of the condition.

Lung transplantation is now an accepted technique for the management of patients with end-stage CF. The problems that should be addressed in the future are outlined below.

- At present there is no adequate way of predicting how ill patients are and when they should receive a transplant. An individual parameter, e.g., FEV_1 less than 30% of the predicted value, cannot be relied upon solely to determine the candidacy of a patient for transplantation. Currently, evidence of progressive decline in status, despite maximum medical and nutritional support, is a commonly used criterion. Since the long-term results of lung transplantation are not satisfactory, it is preferable to delay performing a transplant. The logistics of organ allocation in each country and region must also be considered in deciding when a patient should be put on the transplantation list.
- For certain groups of patients it is not clear whether lung transplantation is contraindicated or not, e.g., those with *P. cepacia* or mycobacterial infections. While some programmes would accept patients with mycobacterial as well as fungal infections, there are concerns about accepting those with *P. cepacia*.
- The incidence of brochiolitis obliterans is quite alarming and, on long-term follow-up, results in the demise or dysfunction of a significant number of CF patients who receive transplants. The emphasis should be on identifying strategies for the prevention and treatment of bronchiolitis obliterans.

These issues cannot be answered through individual centre approaches. In France, for example, the Cystic Fibrosis Transplant Study Group has been developed with the specific purpose of organizing clinical research to address these problems. A transplant registry with detailed patient information has been set up, and this could be expanded for the purpose of retrospective analysis as well as for the design of prospective clinical trials.

CF transplants should be carried out only in a limited number of centres where special expertise is available. It may be more appropriate that poorer countries concentrate their resources on providing good nutrition, enzyme replacement therapy, physiotherapy, and antibiotics to CF patients. Also, medical treatment should be improved so that fewer CF patients need transplants. Malnutrition, upper respiratory tract infection, malabsorption of cyclosporin, diabetes mellitus, salt loss, bowel obstruction, and liver disease can all cause problems in CF patients.

Cell physiology[i]

Possible approaches to pharmacological treatment of CF

Over the past few years, understanding about the basic abnormality in CF patients at the cellular level

[i] **Anderson MP** Nucleoside triphosphates are required to open the CFTR chloride channel. *Cell*, 1991, **67**: 775–784; **Anderson MP et al.** Demonstration that CFTR is a chloride channel by alteration of its anion selectivity. *Science*, 1991, **253**: 202–204; **Cheng SH et al.** Defective intracellular transport and processing of CFTR is the molecular basis of most cystic fibrosis. *Cell*, 1990, **63**: 827–834; **Knowles MR et al.** A pilot study of aerosolized amiloride for the treatment of cystic fibrosis lung disease. *New England journal of medicine*, 1990, **322**: 1189–1194; **Knowles MR, Clarke LL, Boucher RC.** Extracellular nucleotides activate chloride secretion in cystic fibrosis airway epithelia. *New England journal of medicine*, 1991, **325**: 533–538; **Quinton PM, Reddy MM.** Control of CFTR-C1-conductance by energy levels and non-hydrolytic ATP-binding. *Nature*, 1992, **360**: 79–81.

has advanced very rapidly. Nevertheless, it is not yet possible to advocate specific therapies for direct intervention in the disease process. Several cellular defects that may become principal targets for drug therapy are, however, now clearly recognized. Current views are that CF is associated with a protein (cystic fibrosis transmembrane conductance regulator (CFTR)), which forms a channel for the passive, conductive movement of chloride ions through the cell membrane. The normal opening or activation of this chloride-specific channel is dependent upon phosphorylation of CFTR by protein kinase A, whose enzymatic activity is governed by cellular concentrations of AMP. In numerous fluid-secreting systems the concentration of cyclic AMP, and hence the opening of the chloride channel, depends on stimulating the cell with a β-adrenergic agonist. In CF-affected tissues, β-adrenergic stimulation fails to open chloride channels. Although there is no direct link between this failure and the pathology manifested in patients with CF, the most prevalent assumption is that correcting the defect in chloride permeability will markedly ameliorate the disease process.

Three potential approaches to drug therapy are protein synthesis and processing; chloride channel activation; and collateral compensation. Shortly after the gene for CFTR was expressed *in vitro*, it was reported that in CF cells CFTR failed to undergo complete glycosylation. Further immunocytochemical localizations with antibodies to the protein demonstrated that in CF the protein appeared to accumulate in the cytoplasm, in contrast to normal cells where it is primarily associated with the apical membrane. However, it is doubtful that failure to process the protein is the only, and absolute, defect in CF patients. Insect and amphibian cells induced to synthesize mutant CFTR proteins from corresponding messenger RNA templates clearly show that mutant CFTR is processed to the cell membrane even though it exhibits different kinetics with respect to stimulation and function after activation. Very recent studies have demonstrated that this failure to process CFTR is temperature sensitive, i.e., recombinant mammalian cells grown a few degrees below body temperature appear to process mutant CFTR proteins much more efficiently than those grown at normal temperatures. These observations suggest that drugs that affect the manner in which proteins are folded and processed in order to reach the cell membrane could have an important role in therapeutic interventions. Since at least some chloride conductance can be elicited from mutant chloride channel proteins by certain drugs, the latter may have an effect on protein processing. The action of these drugs should be studied further.

As far as activation of the chloride channel is concerned, the observation that extremely high doses of phosphorylating agonists seem to restore at least some function to mutant CFTR chloride channels suggests that drugs which enhance phosphorylation or inhibit dephosphorylation could potentially be used to "hyperphosphorylate" mutant proteins. The success of this approach will depend on the complete processing of at least some of the mutant CFTR to the membrane and on the mutant having at least some functional activity once it is in place in the appropriate cell membrane.

The commonest mutation in CF, delta F508, occurs in a region of the CFTR protein that should bind nucleotides. Recently, it has been suggested that binding of ATP at this site is an additional requirement for opening the chloride channel. Mutations in this region may very well interfere with normal ATP binding, phosphorylation, or function. Consequently, analogues of ATP that bind more readily to this region and potentiate the activation of mutant CFTR chloride channels could be a potential class of drugs.

The third approach would be one that does not target defects directly involved with the mutant CFTR protein, but which treats the expression of the defect in affected systems. The object is to compensate for abnormal or deleterious functions caused by the mutant channel. For example, it is now well established that other types of chloride channels are present in many cell membranes. Drugs that could affect the function of these channels in such a way that they compensate for the loss of the CFTR chloride channel could lessen the impact of a defective physiological expression. Furthermore, CFTR may have functions other than chloride conductance or be closely associated with other cellular functions that require chloride conductance, e.g., the CFTR protein may conduct chloride when stimulated and carry out additional functions that are as yet unknown. Such functions probably involve hydrogen ions, and hence pH level, because the normal movement of bicarbonate in the pancreatic duct is thought to be highly dependent upon an open chloride channel. Failure of this system is almost certain to alter the pH of secreted fluid and may be closely tied to the pathology of CF. Drugs that inhibit or stimulate hydrogen ion or bicarbonate transport might therefore conceivably be used to compensate for, and at least partially correct, a defect in physiological expression precipitated by loss of CFTR chloride channel function.

The above considerations should not be taken to be all inclusive, since numerous other avenues will probably be discovered that may provide much better approaches to controlling CF. It is more than encour-

aging to note, however, that it has recently become possible, for the first time in the history of the disease, to discuss potential drug therapy in terms of observations and fundamental understanding of the biochemical and physiological processes involved. Thus, research in CF has apparently entered a new era that has every possibility of being characterized by the discovery and application of therapeutic drugs.

Aerosolized amiloride and triphosphate nucleotides

Normal airway epithelial physiology. Conducting airway epithelia absorb surface liquid as it is moved by the mucociliary escalator from distal regions to proximal conducting airways. Active Na^+ absorption is the dominant basal ion flow, and provides the driving force for reabsorption of airway surface liquid. The absorption of Na^+ occurs in two steps; Na^+ enters the cell down an electrochemical gradient through an amiloride-sensitive Na^+ channel on the apical membrane and is pumped from the cell by the basolateral membrane Na^+/K^+/ATPase. The accompanying movement of chloride ion occurs via cellular and/or paracellular paths.

There is no net liquid (i.e., Cl^-) secretion under resting conditions, because electrochemical driving forces do not favour movement of Cl^- across the apical membrane. However, airway epithelia have the ability to secrete liquid across the apical membrane into the airway lumen by active Cl^- secretion across several types of Cl^- channels. For example, Cl^- secretion can occur via AMP-dependent (CAMP) or calcium-mediated pathways. Thus, airway epithelia have the capability for either salt and water absorption (driven by active Na^+ transport) or liquid secretion (driven by active Cl^- transport).

Abnormal ion transport in CF airway epithelia. CF airway epithelia exhibit two defects in ion transport that contribute to abnormal airway secretions. The predominant transport dysfunction is excessive absorption of Na^+ (and liquid); CF airway epithelia also have limited ability to secrete Cl^- via the CFTR protein (Cl^- channel) in response to CAMP-mediated stimulation. Taken together, excessive Na^+ absorption and limited Cl^- secretion contribute to dehydration of airway surface liquid and impaired mucociliary clearance.

Treatment of excessive Na^+ (and liquid) absorption in CF airways. Amiloride, a Na^+ channel blocker, inhibits Na^+ absorption across normal and CF airway epithelia. Thus, amiloride delivered to the surface of CF airways might limit excessive Na^+ absorption and prevent dehydration of airway surface liquid. Studies of the short-term use of amiloride aerosol indicate improved mucociliary clearance compared to the vehicle. Also, a double-blind crossover study of chronic amiloride aerosol in 14 adult CF patients suggested clinical benefit. This study tested the effect of amiloride aerosol on pulmonary function over a 6-month period after parenteral administration of antibiotic to normalize lung function and provide for optimal aerosol delivery. The decrease in FVC produced by the amiloride aerosol was only approximately 40% of that produced by the control vehicle. The safety and efficacy of amiloride aerosol in patients with CF is being investigated in a multicentre placebo-controlled trial involving patients aged 12 years and above. An important aspect of the use of amiloride aerosol in CF patients relates to its "protective effect" from mucus impaction, airway damage, and prevention of chronic bacterial infections. Under optimal circumstances, aerosolized amiloride instituted in early childhood might retard or prevent airways disease.

Approach to treating abnormal Cl^- secretion in CF airways. Recent efforts have targeted the abnormality in Cl^- (liquid) secretion. Triphosphate nucleotides (adenosine triphosphate (ATP) and uridine triphosphate (UTP)) induce Cl^- secretion across human airway epithelia *in vitro* and *in vivo* via interaction with extracellular purinergic (P_2) receptors. *In vivo* superfusion of ATP and UTP onto amiloride-pretreated nasal epithelium induced Cl^- secretion in normal subjects and CF patients. This effect was dose related and the nucleotides were equipotent (approximately $3–5 \times 10^{-6}$ mol/l) in both groups (maximal effective concentration, approximately 10^{-4} mol/l). Interestingly, the efficacy of these compounds was greater in CF patients than in normal subjects.

The mechanism of nucleotide-induced Cl^- secretion was explored *in vitro* and *in vivo*. Primary cultures of airway epithelial cells were studied with double-barrelled Cl^--selective microelectrodes; the tissues were pretreated with amiloride on the luminal membrane to induce a favourable driving force Cl^- secretion. Both ATP and UTP induced greater Cl^- secretion in epithelial cells from CF patients compared with normal subjects. This greater response in CF epithelia reflected a change from a lower basal rate of Cl^- secretion in cells from CF patients to a level of secretion similar to that of normal epithelia after nucleotide application. These observations are consistent with activation of an apical membrane Cl^- conductance and Cl^- secretion.

The relatively prolonged duration of Cl^- secretion in response to these nucleotides *in vivo* suggests that more than one mechanism may be operating. One mechanism clearly involves P_2 receptor activa-

tion, phospholipase C and inositol triphosphate generation, and an increase in cytosolic calcium. This type of activation is usually short lived, and may contribute only to the initial phase of the Cl^- secretory response. The duration of action suggests that a more direct grating effect on the Cl^- channel may be present and that this mode of action may sustain the secretory response.

ATP and UTP also stimulate ciliary beat frequency and goblet cell degranulation of canine and human airway epithelia, also probably via P_2 receptor mechanisms. These effects, together with the Cl^- secretory effect of luminal nucleotides, suggest that a physiological and/or integrative effect of these compounds is involved in the clearance of airway secretions as a defense mechanism. For example, a coordinated response to an inhaled irritant or infectious agent might be amplified by luminal nucleotides to stimulate ciliary beat frequency and induce goblet cell degranulation and liquid secretion in the airway lumen, thereby assisting in mucociliary clearance.

Future developments. In CF, defined abnormalities of ion transport by airway epithelia provide the rationale for therapeutic intervention with aerosolized pharmacological agents that modulate ion transport. In CF patients, aerolized amiloride inhibits excessive Na^+ absorption, improves biorheology and mucociliary clearance of airway secretions, and may retard the loss of lung function in adults. Recent studies suggest that it may also be possible to attack the other limb of abnormal ion transport, i.e., the defect in Cl^- secretion. Aerolized nucleotides hold promise for inducing Cl^- secretion in CF airway epithelia. Pyrimidine compounds, such as UTP, may be preferable to ATP because the metabolic products of UTP may have less effect on airway epithelia than those of ATP.

Gene therapy[k]

New developments may permit the introduction of a normal copy of the CFTR gene into the affected tissue of CF patients. Somatic genetic therapy provides a great hope for the future and raises no ethical problems. It should be noted that there are no proposals currently to carry out germ-line gene therapy, which would raise completely different issues.

Because CF is a recessive disease, a single copy of the normal gene is sufficient for correct physiological function, a hypothesis that has been confirmed using cells from CF patients in culture. There is now an accurate CF mouse model, which should permit more rapid assessment of the efficacy and safety of gene treatment. Vectors based on a disabled adenovirus, which appears to meet stringent safety criteria, can introduce the gene into epithelial cells, such as those lining the lungs. The lung is relatively accessible to genetic constructs; however, it should be noted that it is not certain that correction of the ion defect is equivalent to correction of the lung pathology.

The major questions raised by gene therapy are whether it will differ in a revolutionary way from existing treatments, whether it will be available within a reasonable length of time, and whether it will be sufficiently easy to use and inexpensive enough to permit its possible application to all who could benefit.

The first clinical trials, which started in 1993, will address specific questions about the entry and expression of the CFTR gene in human respiratory epithelium, and also questions of safety, all of which will also be studied in the CF mouse model. If clinical benefit is evident, early approval by regulatory agencies will result in a product entering trials to assess clinical benefit as early as 1995. However, it should not be forgotten that the genetic therapies currently being studied will only treat cells lining the lungs, and that the gut and liver pathologies and infertility associated with CF are not yet being investigated. Not everyone with CF will derive clinical benefit since they will experience other systemic problems in time. Genetic therapy may therefore be an improvement on existing therapy rather than a cure for CF. It should also be borne in mind that there has only been one successful clinical trial of gene therapy, to date, involving a handful of patients with severe combined immunodeficiency.

Genetic therapy will be easy to deliver in any country that has a modern medical system; therefore, there are no reasons why any patient should be deprived of the benefits of such therapy once its efficacy and safety have been established. However, the development costs of gene therapy are considerable, and private companies will expect to recover these costs when treatments are introduced.

Thus, in summary the following points can be made about the use of gene therapy for CF.

- Somatic genetic therapy for CF should be encouraged.
- The time-scale for this development is being reduced by the provision of adequate funding and the availability of a mouse model. Safety and efficacy studies may be well under way over the next 2 years.
- The genetic therapies that are currently under consideration for CF will only improve lung function,

[k] **Crystal RG.** Gene therapy strategies for pulmonary disease. *American journal of medicine*, 1992, **92**(suppl. 6A): 44S–52S; **Drumm ML et al.** Correction of the cystic fibrosis defect *in vitro* by retrovirus-mediated gene transfer. *Cell*, 1990, **62**: 1227–1233.

and not other body functions; therefore it is important to regard gene therapy as improved treatment rather than as a cure.

- WHO/ICF(M)A should maintain contact with the major CF charities, government agencies, and the biotechnology companies involved to ensure that new therapies become available as quickly as possible at reasonable cost, so that treatment is not limited solely by finance.

Nutritional management[l]

Growth retardation affects a significant number of CF patients and may influence the progression of the disease and the survival of those afflicted. A wide range of nutritional problems are associated with CF. As patients age, their height and weight are displaced to lower percentiles. There are deficiencies in body cell mass and fat, as well as diminished muscle mass, and an increase in extracelluar water. The diminished body cell mass can appear during the first months of life. Specific deficits of essential fatty acids, fat-soluble vitamins, some water-soluble vitamins, and some micronutrients have been demonstrated. Excessive loss of sodium chloride in sweat, particularly in young infants, leads to the significant collapse of some patients. The nutritional problems arise from a combination of factors, including the following: increased nutrient loss in the stools from maldigestion; diminished food intake during acute illnesses; and increased energy utilization to sustain breathing because of diminished lung function. Some evidence supports that a basic defect in patients with CF is a maladaptation to undernutrition, although it is difficult to specify the many factors involved. The major consequences of malnutrition in CF are growth retardation, delayed puberty, pulmonary disorders, malabsorption, poor immune status, and finally the adverse progression of the disease. Improved nutritional status has been reported for patients who have both milder pulmonary involvement and pancreatic sufficiency. The nutritional rehabilitation of malnourished patients improves the course of the pulmonary disease; aggressive nutritional surveillance and intervention are indicated at all stages of the disease.

To achieve adequate protein–energy balance, daily consumption of more than 130% of the recommended dietary intake may be necessary to compensate for increased needs and losses. Fat intakes of 40% of the total energy requirement are well tolerated with adequate pancreatic enzyme support. Dietary supplements as well as vitamin and mineral supplements are also required to treat deficiencies. Furthermore, dietary support and counselling are of central importance for maintaining adequate nutrition. Significant failure to grow or weight loss may necessitate nocturnal intragastric feedings.

Pancreatic enzyme supplements do not fully correct the maldigestion associated with CF but there is considerable individual variation. In some patients significant maldigestion still persists despite ingestion of high levels of pancreatic enzyme. Adjuncts such as H_2-receptor antagonist plus prostaglandin analogues (misoprostol) have sometimes been found to be beneficial. The key to dealing with nutrition in CF is to place its surveillance and management at a high priority in the patient care programme from the time of diagnosis.

Social and emotional adaptation[m]

The psychosocial aspects of CF are related specifically to the nature of the disease, and in general to the problems posed by all chronic diseases. The following are important obstacles that make it difficult to adapt to and accept the illness:

— about 20% of patients experience progressive or perceived disability that limits normal vocational aspirations;

— a rigorous regimen of therapy that requires time, knowledge, and commitment is needed to maintain or attain a functional state of well-being;

— it is not easy to conceal feelings of deficiency and some of the symptoms of the disease, such as cough and activity limitations;

— about 20% of CF patients refuse to accept their condition and avoid the indicated therapeutic interventions, sometimes at some biological cost to themselves;

— for about 40–50% of patients the moderate-to-high economic cost of maintaining the therapeutic programme usually requires significant input from their families and public support systems.

[l] **Dalzell AM et al.** Nutritional rehabilitation in cystic fibrosis: a 5-year follow-up study. *Journal of pediatric gastroenterology and nutrition*, 1992, **15**: 141–145; **Ramsey BW, Farrell P, Pencharz PB.** Nutritional assessment and management in cystic fibrosis: a consensus report. *American journal of clinical nutrition*, 1992, **55**: 108–116.

[m] **Bartholomew LK et al.** Development of a health education programme to promote the self-management of cystic fibrosis. *Health education*, 1991, **18**: 429–443; **Mador JA, Smith DH.** The psychological adaptation of adolescents with cystic fibrosis: a review of the literature. *Journal of adolescent health care*, 1989, **10**: 136–142; **Pearson DA, Pumariega AJ, Seilheimer DK.** The development of psychiatric symptomatology in patients with cystic fibrosis. *Journal of the American Academy of Child and Adolescent Psychiatry*, 1991, **30**: 290–297.

The problems related to the chronicity of the condition are as follows:

— significant potential for and actual acute short- or long-term interruption of activities in at least 25% of CF patients;

— need for a sustained process of chronic care and daily effort to maintain health;

— fear of progressive morbidity or of a shortened life span; and

— problems associated with perception by others and guilt in the family about the CF patient.

In the past, dysfunctional maladaptive emotional states were thought to be very common among CF patients. However, recent studies have shown that they have a remarkably high level of functional adaptation and strength. Similarly, many families have been strengthened by the demands of the disease. In contrast, it does appear that at least 20% of CF patients and their families have borderline or serious maladaptations to life, and some of these individuals might benefit from formal psychological treatment. All CF patients and their families may have periods of psychological maladaptation and readjustment requiring ongoing counselling and advocacy by specialist care providers through the stressful periods of diagnosis, maintenance, acute exacerbations, and the grief surrounding the death of the patient.

Of special importance is compliance with a complicated regimen that requires considerable organizational skill to manage and integrate into each day. Many individuals achieve a compromise as they try to blend their treatment needs with their life priorities. In particular, the care providers need to understand the problems posed by the disease's chronicity and the psychological adaptation required to cope with it, and be skilled in counselling families and patients.

The crises created by family breakdown, divorce, death, and other significant life events can have a marked impact on maintaining the therapeutic programme, the biological course of the disease, and the use of health resources. As with all chronic diseases, a "biopsychosocial" approach provides orientation in helping patients and their families. Although the degree of the disease certainly influences the psychosocial aspects associated with CF, it is of interest to note that some patients with mild CF exhibit marked insecurity and emotional immaturity, which prolongs adolescence into the adult years.

A major requirement of CF patients and their families is dedicated, compassionate carers providing continuity and advocacy. It is also clear that the psychosocial problems arising from CF are as important as its specific biological aspects and may require as much care, time, and commitment.

Conclusions and recommendations

The various therapies now available have increased survival and reduced morbidity among individuals with CF. It is now possible to identify the majority of the genetic distribution of CF in a population and apply current and soon-to-be available treatment in countries whose health systems are sufficiently organized to achieve this goal. New genetic therapies appear ready to be studied and carry promise to alter the pattern of the disease.

WHO and ICF(M)A should continue their collaborative efforts in the following areas:

— issue regular reports to countries about the status of therapeutic developments in cystic fibrosis;

— bring together scientists and medical leaders to plan and share technical knowledge that will be made available to all countries;

— assist in the development of collaborative efforts to improve and facilitate the more rapid development of effective therapeutic modalities;

— help in the development of financial support for such scientific programmes and research;

— plan an educational campaign with countries interested in sharing information on care and research; and

— work on trade agreements with countries and pharmaceutical companies to improve the wider availability, at the lowest cost, of medications for the treatment of CF.