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Differences in prevalence and treatment of *Pseudomonas aeruginosa* in cystic fibrosis centres in Denmark, Norway and Sweden☆

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Abstract

Background: Chronic *Pseudomonas aeruginosa* (PA) infection causes increased morbidity and mortality in cystic fibrosis (CF). This study aimed to answer the following questions: Does the prevalence of chronic infection with PA differ between the CF centres in Scandinavia? Which differences exist concerning segregation and treatment of PA?
Methods: 989 patients (86%) from all eight CF-centres in Scandinavia were included. Demographic and clinical data, including PA colonisation status based on cultures and serology, were recorded at inclusion. The patients were followed prospectively for 1 year, recording number of days with anti-PA antibiotic treatment.
Results: In all pancreatic insufficient (PI) patients ($n=890$) the prevalence of chronic PA infection at each centre ranged from 25.8% to 48.9%, but were not significantly different. In PI patients <19 years the prevalence was 14.5% in Copenhagen compared to 30.9% in the Swedish centres pooled ($p=0.001$). In intermittently colonised PI patients <19 years the median number of days per year on anti-PA antibiotics was almost 6 times higher in Copenhagen (mean 86 (110), median 61 days) compared to the Swedish centres pooled (mean 27 (52), median 11 days) ($p=0.037$). The pulmonary function was similar.

Abbreviations: SCFSC, Scandinavian CF Study Consortium; PA, *Pseudomonas aeruginosa*; CF, cystic fibrosis; PI, pancreatic insufficient; CIE, crossed immunoelectrophoresis; FEV_1, forced expiratory volume in one second; CFTR, Cystic fibrosis transmembrane conductance regulator.

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Conclusions: It is possible to maintain a very low prevalence of chronic PA infection in CF patients <19 years. We speculate that this was most likely due to a very intensive treatment of intermittently colonised patients with inhaled anti-PA antibiotics over prolonged periods of time in some centres. Since lung function was similar in centres with less intensive use of inhaled antibiotics, studies comparing different treatment modalities and other parts of CF care are needed to define the best clinical practice, including how to use antibiotics in the most rational way.

Keywords: Cystic fibrosis; *Pseudomonas aeruginosa*; Prevalence; Antibiotic therapy

1. Introduction

The most common chronic bacterial pathogen in the cystic fibrosis (CF) lung is *Pseudomonas aeruginosa* (PA) [1,2]. Chronic infection with PA is associated with a decline in lung function [3,4] and increased morbidity and mortality [5,6]. Several studies have shown that early and aggressive antibiotic treatment of PA can postpone and may prevent chronic infection [7–10]. Different antibiotic treatment policies are used in different CF centres, but randomised controlled studies comparing different antibiotic treatment regimens have not been published [11].

Pulmonary infection with PA causes an immune response producing detectable antibodies against specific bacterial antigens. The response is even more pronounced in chronic infection with mucoid PA strains [3,12]. In Danish studies the level of crossed immunoelectrophoresis (CIE) precipitating antibodies (precipitins) against PA showed high sensitivity and specificity for detecting patients with chronic infection [13,14].

Evidence for cross infection with PA between CF patients with epidemic strains [15–17] and after prolonged close social contact (summer/winter camps, physical training courses etc) have been published [18,19]. Other publications report low grade of cross infection [20,21]. Consequently, the question whether patients harbouring PA should be segregated from patients not having the bacteria, is debated [22].

Studies focusing on the impact of differences in treatment policies between CF centres in Scandinavia on the prevalence of chronic PA infection have not been performed. This study aimed to answer the following questions: Does the prevalence of chronic infection with PA differ between the CF centres in Scandinavia? Which differences exist between centres concerning antibiotic treatment strategies, but also segregation- and bacteriological surveillance strategies for PA? Does the prevalence of other chronic Gram-negative infections differ between the centres?

2. Methods

2.1. Patients

The study was initiated by the Scandinavian CF Study Consortium (SCFSC), and all eight CF-centres in Scandinavia participated in the study: Denmark: *Aarhus*, *Copenhagen*; Norway: *Bergen*, *Oslo*; Sweden: *Gothenburg*, *Lund*, *Stockholm* and *Uppsala*. Patients were included consecutively from August 2001 to June 2003, when in clinical stable condition. Exclusion criterion was lung transplant. Patients using pancreatic enzymes were classified as pancreatic insufficient (PI).

2.2. Collection of data

Demographic and clinical data, including PA colonisation status, were recorded at inclusion and the patients were followed prospectively for 1 year. During the study period, days when antibiotics directed against PA were given, and mode of administration (intravenous (iv), inhaled, oral), were recorded and expressed as number of days per calendar year. When treatment modes were combined, the more "intensive" mode of administration was recorded (iv>inhalation>oral, e.g. in case of combination of iv and inhaled and/or oral antibiotics, only iv administration was recorded). As no centre used oral quinolones as monotherapy against PA, consequently only days with iv or inhaled anti-pseudomonas treatment were recorded. Patients with an inclusion period of less than 275 days, or more than 500 days from the date of inclusion to the date of the end of the study, were excluded from calculations of number of days on antibiotic treatment. Serum samples were obtained a inclusion and at the end of the study for analyses of CIE precipitating antibodies (precipitins) to PA [13]. The precipitin analyses were performed in Copenhagen for all patients.

Lung function test was performed at inclusion (patients older than 6 years) by dynamic spirometry at each centre, according to the local routine. From measured forced expiratory volume in 1 second (FEV_1) in litres, percentage of predicted values were calculated using Solymar [23] and Quanjer [24] reference equations for patients younger and older than/or 19 years, respectively.

Data on policies for surveillance, segregation and treatment of PA were obtained from questionnaires sent to each of the eight participating centres.

2.3. Comparison of the CF centres

Data from all the eight participating centres are presented in tables. Presenting comparisons of all pairs of centres is didactically difficult. It was therefore decided to pool the four Swedish centres in Gothenburg, Lund, Stockholm and Uppsala since they were found to be very similar concerning strategies for antibiotic treatment and segregation policies. The centre in Copenhagen was known to have a different approach to antibiotic treatment and segregation compared to Swedish centres and also differing from the other Danish centre located in Aarhus. It was therefore decided to make comparisons between Copenhagen separately and the four Swedish centres pooled.

2.4. Classification of infection status

2.4.1. Pseudomonas aeruginosa

Chronic infection: Repeated positive bacterial cultures (≥3) during the last six months and/or 2 or more CIE precipitins against PA. Intermittent colonisation: PA cultured at least once during life, but less than three positive cultures during the last six months and precipitins less than 2. Not colonised: PA never cultured and 0–1 precipitins (=normal values).

2.4.2. Other Gram-negative bacteria

Classification of chronic infection with other Gram-negative bacteria was based on each centres own definition, usually three or more positive cultures during a six month period.

2.5. Statistical analysis

Fischer's exact two-sided test was used to compare categorical data (prevalence and incidence of chronic infections) in two groups. Logistic regression with chronic infection as dependent variable and age and centre as covariates was used when comparing centres with different mean age. Independent *t*-test was used to compare normally distributed continuous data (age, FEV_1) in two groups. Linear regression with FEV_1 as dependent variable and age and centre as independent variables was used when comparing centres with different mean age. Mann-Whitney *U*-test was used to compare continuous variables with skewed distribution (number of days per year with antibiotic treatment). The significance level was set to 5%. The statistical analyses were performed by SPSS version 14.0.

2.6. Ethics

The Regional Research Ethics Committees in each country approved the study and informed written consent was obtained from all patients.

3. Results

3.1. Policies for patient segregation, bacteriological surveillance and treatment of PA

The antibiotic treatment policies at each centre are presented in Table 1.

Table 1
Antibiotic treatment policies against *Pseudomonas aeruginosa* at the eight Scandinavian CF centres

	First PA infection	Intermittent PA colonisation	Chronic PA infection	Azithromycin treatment	Cultures per year from non-sputum producing patients
Aarhus	Oral ciprofloxacin+ inhaled antibiotics*, 3 weeks	Oral ciprofloxacin+inhaled antibiotics*. 3 months if recurrent infection	14 days iv every 3. month / on demand. Aprox. 50% of patients: Inhaled antibiotics*	Not used at the time of the study	10–11 (laryngeal aspiration)
Copenhagen	Oral ciprofloxacin+ inhaled colistin, 3 months	Oral ciprofloxacin+inhaled colistin, 3 months. Iv if recurrent isolate during treatment, or mucoid phenotype, or increasing antibodies	14 days iv every 3. month. Continuous inhaled antibiotics* between iv	Patients with chronic PA infection	12 (laryngeal aspiration)
Bergen	Oral ciprofloxacin+ inhaled antibiotics*, 4 weeks. Small children: 14 days iv	Oral ciprofloxacin+inhaled antibiotics*. 3 months if recurrent infection	12–14 days iv every 3. month. Selected patients: Inhaled antibiotics* between iv	Selected patients with chronic PA infection	4–6 (laryngeal aspiration)
Oslo	Oral ciprofloxacin+ inhaled colistin, 3 weeks. Small children: 14 days iv	Oral ciprofloxacin+inhaled antibiotics*. Duration varies. Sometimes 14 days iv	14 days iv every 3. month. Most patients: Inhaled antibiotics* between iv	No strict policy	9 (laryngeal aspiration)
Gothenburg	10 days iv. Followed by inhaled antibiotics*, 4 weeks since 2000	10 days iv. Followed by inhaled antibiotics*, 4 weeks since 2000	10 days iv on demand	Patients with chronic PA infection	4 (nasopharyngeal swab)
Lund	10 days iv, followed by oral ciprofloxacin and/or inhaled antibiotics*. Information of duration not given	10 days iv, followed by oral ciprofloxacin and/or inhaled antibiotics*. Information of duration not given	10 days iv on demand. Selected patients: Inhaled antibiotics*	Patients with chronic infections or much sputum	0
Stockholm	10 days iv (Repeated within a month if no eradication)	10 days iv, followed by inhaled antibiotics* in selected cases	10 days iv on demand. Selected patients: Inhaled antibiotics*	Selected patients with chronic PA infection	5–6 (nasopharyngeal swab)
Uppsala	10–14 days iv followed by inhaled tobramycin and oral ciprofloxacin and azithromycin, 3 months	10–14 days iv followed by inhaled tobramycin and oral ciprofloxacin and azithromycin, 3 months	10–14 days iv on demand. Selected patients: Inhaled antibiotics* and/or oral ciprofloxacin	Patients with chronic PA infection	4 (nasopharyngeal swab)

*Colistin or tobramycin.
PA, Pseudomonas aeruginosa; iv, intravenous.

In all centres, most PI patients were seen at the outpatient clinic every 4th to 6th week. Patients in Norway and Sweden living long distances from their CF centre, were seen by their local doctor between their yearly (or more frequent) visits at the centre (e.g. shared care with the local hospitals). Bacteriological cultures were obtained from sputum-producing patients at every visit and if the patients noticed symptoms of exacerbation between regular visits. The major differences between centres/countries were:

1. Patient segregation at the outpatient clinic according to PA colonisation status was not practiced in any Swedish centre, in contrast to Danish and Norwegian centres.
2. Bacteriological cultures from non-sputum-producing patients were obtained less often in Sweden compared to Denmark and Norway using laryngeal aspiration at every visit.
3. The main eradication policy of first and intermittent PA infection was 10 days of iv antibiotics in Sweden, but inhaled antibiotics (colistin or tobramycin) in combination with oral ciprofloxacin for several weeks/months in Denmark and Norway.
4. Patients with chronic PA infection were given iv antibiotics on demand in Sweden and in Aarhus, but regularly every 3rd month in Copenhagen and in Norway.
5. Suppression therapy (inhaled colistin or tobramycin) between iv courses was given regularly to patients with chronic PA infection in Copenhagen, to some extent in Aarhus, Bergen and Oslo, but seldom in Swedish centres.

Table 2
Demographic and clinical data on patients from the eight Scandinavian CF centres and from the Swedish centres pooled

	Denmark		Norway		Sweden					
Centres	A	C	B	O	G	L	S	U	G+L+S+U	*p*-value*
All patients (n=989)										
Patients, *n* (F)	119 (56)	240 (121)	42 (20)	159 (73)	118 (51)	109 (53)	158 (73)	44 (23)	429 (200)	
Genotypes, *n* (%)										
dF508/dF508	87 (73.1)	171 (71.3)	14 (33.3)	67 (42.1)	51 (43.2)	58 (53.2)	69 (43.7)	17 (38.6)	195 (45.5)	$p < 0.001$
Class I/I, I/II, II/II[#]	106 (89.1)	201 (83.8)	15 (35.7)	90 (56.6)	76 (64.4)	81 (74.3)	120 (75.9)	35 (79.5)	312 (72.7)	$p = 0.002$
PS patients, *n* (%)	6 (5.0)	1 (0.4)	11 (26.2)	23 (14.5)	26 (22.0)	15 (13.8)	15 (9.5)	2 (4.5)	58 (13.5)	$p < 0.001$
PI patients (n=890)										
Patients, *n* (F)	113 (50)	239 (121)	31 (14)	136 (58)	92 (41)	94 (45)	143 (66)	42 (21)	371 (173)	
Mean (SD) age (y)	14.5 (9.3)	19.5 (11.3)	15.2 (13.0)	19.6 (12.5)	18.3 (10.1)	18.5 (11.1)	16.9 (11.5)	17.7 (11.5)	17.7 (11.1)	$p = 0.058$
Genotypes, *n* (%)										
dF508/dF508	86 (76.1)	171 (71.5)	14 (45.2)	66 (48.5)	51 (55.4)	58 (61.7)	69 (48.3)	17 (40.5)	195 (52.6)	$p < 0.001$
Class I/I, I/II, II/II[#]	103 (91.2)	201 (84.1)	15 (48.4)	89 (65.4)	75 (81.5)	80 (85.1)	119 (83.2)	35 (83.3)	309 (83.3)	$p = 0.88$
Unknown[†]	3 (2.7)	6 (2.5)	14 (45.2)	27 (19.9)	2 (2.2)	7 (7.4)	11 (7.7)	4 (9.5)	24 (6.5)	$p = 0.044$
Chronic PA infection										
All PI patients, *n* (%)	37 (32.7)	116 (48.5)	8 (25.8)	55 (40.4)	42 (45.7)	46 (48.9)	67 (46.9)	19 (45.2)	174 (46.9)	$p = 0.33$[¶]
PI patients < 19 y, n/n_{Total} (%)	13/80 (16.3)	18/124 (14.5)	3/23 (13.0)	12/67 (17.9)	15/53 (28.3)	15/49 (30.6)	35/95 (36.8)	3/23 (13.0)	68/220 (30.9)	$p = 0.001$
Chronic BC infection										
All PI patients, *n* (%)	5 (4.4)	16 (6.7)	0	4(2.9)	2 (2.2)	0	3 (2.1)	2 (4.8)	7 (1.9)	$p = 0.004$
PI patients < 19 y, n/n_{Total} (%)	1/80 (1.3)	4/124 (3.2)	0/23	1/67 (1.5)	0/53	0/49	1/95 (1.1)	0/23	1/220 (0.5)	$p = 0.059$
Chronic SM infection										
All PI patients, *n* (%)	1 (0.9)	9 (3.8)	0	18 (13.2)	6 (6.5)	3 (3.2)	5 (3.5)	0	14 (3.8)	$p = 1.0$
PI patients < 19 y, n/n_{Total} (%)	0/80	4/124 (3.2)	0/23	4/67 (6.0)	1/53 (1.9)	0/49	2/95 (2.1)	0/23	3/220 (1.4)	$p = 0.26$
Chronic AX infection										
All PI patients, *n* (%)	3 (2.7)	14 (5.9)	0	0	0	0	0	0	0	$p < 0.001$
PI patients < 19 y, n/n_{Total} (%)	2/80 (2.5)	9/124 (7.3)	0/23	0/67	0/53	0/49	0/95	0/23	0/220	$p < 0.001$
FEV_1 (% pred.), mean (SD)										
All PI patients	66 (25)	76 (23)	73 (22)	68 (25)	85 (26)	76 (25)	80 (23)	72 (26)	79 (25)	$p = 0.28$[§]
PI patients < 19 y	75 (22)	88 (16)	79 (18)	80 (22)	95 (18)	85 (22)	88 (19)	76 (23)	88 (20)	$p = 0.99$
PI patients with chronic PA	57 (27)	64 (23)	51 (19)	56 (22)	75 (28)	65 (25)	74 (24)	62 (25)	71 (26)	$p = 0.16$[§]

A, Aarhus; C, Copenhagen; B, Bergen; O, Oslo; G, Gothenburg; L, Lund; S, Stockholm; U, Uppsala.
F, female; PS, pancreatic sufficient; PI, pancreatic insufficient; y, years; PA, *Pseudomonas aeruginosa*; BC, species of the *Burkholderia cepacia* complex; SM, *Stenotrophomonas maltophilia*; AX, *Achromobacter xylosoxidans*; FEV_1, forced expiratory volume in 1 s.
* C vs G+L+S+U.
[#] Patients homozygous or compound heterozygous for class I or class II CFTR mutations.
[†] One or both mutations unknown.
[¶] Logistic regression with chronic PA infection as dependent variable and group (C and G+L+S+U) and age as covariates.
[§] Linear regression with FEV_1 as dependent variable and group (C and G+L+S+U) and age as independent variables.

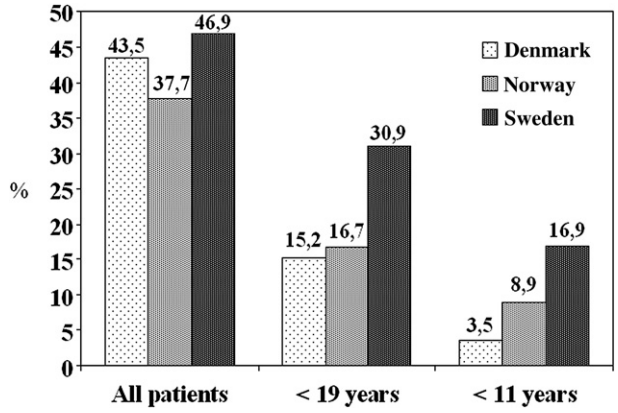

Fig. 1. Prevalence of chronic infection with *Pseudomonas aeruginosa* in 890 pancreatic insufficient CF patients in Denmark, Norway and Sweden.

3.2. Demographics

989 (48% females) cystic fibrosis patients were included in the study, accounting for about 86% of the Scandinavian CF population. Demographic and clinical data from each centre are shown in Table 2. The frequency of PI patients was higher in Denmark, due to differences between the countries in distribution of CFTR mutations (Table 2). Due to this huge difference in prevalence of pancreatic insufficiency, a known risk factor for pseudomonas colonisation and deterioration of lung function, only PI patients ($n=890$ (47% females)) were included in the further analyses, diminishing the differences between the countries related to differences in distribution of CFTR mutations.

3.3. Prevalence of chronic PA infection

The prevalence of chronic PA infection in each country is shown in Fig. 1, and for each centre in Table 2.

When comparing the prevalence in all pairs of centres, after correcting for differences in age, only Oslo and Stockholm were found to be significantly different ($p=0.041$). The prevalence did not differ between Copenhagen and the pooled Swedish centres (Table 2).

In patients < 19 years the prevalence of chronic PA infection was similar in the four centres in Denmark and Norway (p-values not shown), but was significantly lower in Copenhagen compared to each of the three larger Swedish centres: Gothenburg ($p=0.037$), Lund ($p=0.019$), Stockholm ($p<0.001$), and compared to the four Swedish centres pooled ($p=0.001$) (Table 2). However, the prevalence was similar in Copenhagen and at the smallest Swedish centre in Uppsala ($p=1.0$). The prevalence was significantly higher in Stockholm compared to the other three centres in Denmark and Norway: Aarhus ($p=0.004$), Bergen ($p=0.045$) and Oslo ($p=0.013$), and when compared to Uppsala ($p=0.045$).

3.4. Incidence of chronic PA infection

500 of the PI patients were not chronically infected with PA at inclusion. PA colonisation status at the end of the study was available from 456 of these patients with mean age 12.7 (9.4) years. The mean number of days from the start to the end of the study was 374 (35). During the observation period 33 patients (7.2%) became chronically infected with PA. The incidence was 6.3% in Denmark, 5.2% in Norway and 9.4% in Sweden. The incidence in Sweden was more than twice the incidence in Copenhagen: 5/120 (4.2%), but this difference did not reach statistical significance ($p=0.11$).

3.5. Prevalence of other infections

The prevalence of other chronic infections at each centre are shown in Table 2. Chronic infection with *Achromobacter xylosoxidans* was only found in Danish patients, and the prevalence of both *A. xylosoxidans* and species of the *Burkholderia cepacia* complex was significantly higher in Copenhagen compared to the Swedish centres pooled.

3.6. Use of antibiotics directed against PA

3.6.1. Patients chronically infected with PA

The use of antibiotics differed between centres, also within the same country, shown in Table 3. When comparing Copenhagen and the pooled Swedish centres, the number of days

Table 3
Mean (SD), median days per year on antibiotics against PA in patients with pancreatic insufficiency at each centre and the Swedish centres pooled

	Denmark		Norway		Sweden					
Centres	A	C	B	O	G	L	S	U	G+L+S+U	p-value*
Chronic PA, *n*	32	107	8	42	30	42	59	15	146	
Inhaled	131 (157), 19	283 (80), 310	14 (31), 0	119 (116), 122	16 (38), 0	47 (111), 0	13 (31), 0	80 (80), 32	30 (73), 0	$p<0.001$
Iv	17 (24), 0	50 (31), 49	40 (27), 47	42 (19), 49	25 (20), 20	29 (23), 21	40 (24), 36	36 (36), 28	34 (25), 30	$p<0.001$
Inhaled and/or iv	149 (164), 71	333 (84), 365	54 (46), 50	161 (126), 172	41 (42), 32	76 (112), 35	53 (41), 40	117 (96), 81	64 (77), 40	$p<0.001$
Intermittent PA# <19 years, *n*	21	65	5	21	14	15	22	2	53	
Inhaled	18 (36), 0	83 (106), 61	4 (10), 0	63 (118), 0	6 (16), 0	12 (47), 0	12 (44), 0	43 (20), 43	12 (39), 0	$p<0.001$
Iv	1 (5), 0	2 (7), 0	3 (6), 0	8 (13), 0	7 (8), 5	8 (10), 0	25 (28), 15	21 (29), 21	15 (21), 10	$p<0.001$
Inhaled and/or iv	19 (36), 0	86 (110), 61	7 (10), 0	71 (121), 0	13 (21), 5	20 (46), 11	36 (68), 15	64 (49), 64	27 (52), 11	$p=0.037$

*C vs G+L+S+U.
#No other chronic Gram-negative infection.
PA, Pseudomonas aeruginosa; A, Aarhus; C, Copenhagen; B, Bergen; O, Oslo; G, Gothenburg; L, Lund; S, Stockholm; U, Uppsala; Iv, intravenous.

Table 4
Prevalence of pancreatic insufficient patients younger than 19 years getting other medication at each centre

	Denmark		Norway		Sweden			
Centres	A	C	B	O	G	L	S	U
Number of patients	80	124	23	67	53	49	95	23
Dornase alpha (%)	70	99	35	45	30	59	26	0
Inhaled N-acetylcysteine (%)	0	0	96	91	66	10	90	39
Oral N-acetylcysteine (%)	1	14	91	49	9	20	77	39
Oral bromhexin (%)	0	0	0	2	98	25	98	100
Inhaled steroids (%)	15	27	13	9	13	18	13	44
Oral steroids (%)	3	10	0	2	0	0	0	4
Continuous macrolides (%)	0	23	9	5	40	39	14	17

A, Aarhus; C, Copenhagen; B, Bergen; O, Oslo; G, Gothenburg; L, Lund; S, Stockholm; U, Uppsala.

per year with inhaled antibiotics was higher in Copenhagen, reflecting the difference in treatment policy. Also the number of days per year with intravenous antibiotics was significantly higher in Copenhagen, using regular intravenous courses, compared to the Swedish centres, using intravenous antibiotics on demand.

3.6.2. Patients <19 years old intermittently colonised with PA

The number of days per year with antibiotics to patients <19 years intermittently colonised with PA and not having chronic infection with any other Gram-negative bacteria, is shown in Table 3. The number of days per year with inhaled antibiotics was significantly higher in Copenhagen compared to the pooled Swedish centres. In contrast, the number of days per year with intravenous antibiotics was higher in the Swedish centres. In total, the use of antibiotics calculated as the number of days per year with inhaled and/or intravenous antibiotics, was significantly higher in Copenhagen.

3.7. Lung function

Lung function for each centre and for the Swedish centres pooled are shown in Table 2. There were no significant differences in FEV_1 between Copenhagen and the Swedish centres pooled, neither for all PI patients nor for subgroups (PI patients <19 years, PI patients with chronic PA infection).

3.8. Other treatment

Data on other medical treatment are shown in Table 4. The treatment differed between centres. One major difference was that Dornase alpha was the most prevalent mucolytic agent used in Denmark in contrast to most Norwegian and Swedish centres using inhaled N-acetylcysteine. Oral bromhexin was used only in Sweden.

4. Discussion

This study explored the policies for treatment and prevention of PA and the prevalence of chronic PA infection in eight CF-centres in three geographically and socially similar countries. The policies for segregation of patients infected with PA differed with strict segregation at the centres in Denmark and Norway, but not in Sweden. Antibiotic treatment of PA infections was significantly different with antibiotics given more days per year to patients mainly in Copenhagen but also in Aarhus and in the Norwegian centres compared to the Swedish centres. The prevalence of chronic PA infection was similar in all centres when all age groups were included, but significantly higher in the pooled Swedish centres compared to Copenhagen in patients <19 years. This was further supported by a trend towards a higher incidence of chronic PA infection in Sweden during the study period.

One major obstacle of comparisons between centres is different definitions regarding criteria for classification of PA infection status. Definitions only relying on cultures depend on number of cultures obtained over a time period, how the cultures are obtained and facilities at the local microbiology laboratory. This was overcome in this multi-centre study by defining chronic PA infection with a combination of repeated positive cultures and/or ≥2 CIE precipitins against PA detected [13]. The precipitin analyses were centralised to Copenhagen. The prevalence of chronic PA infection in each centre is thus based on the same diagnostic test and is directly comparable. Only one of all chronically infected patients had less than two precipitins (but three or more positive cultures during a six month period).

The prevalence of chronic PA infection was low in all centres compared to other published data [2,25], and the prevalence was probably not underestimated. Instead the prevalence was slightly overestimated since there is a known serological cross-reaction with other Gram-negative bacteria like *A. xylosoxidans*, species of the *B. cepacia* complex and *Stenotrophomonas maltophilia* [26,27]. Some patients were therefore classified as chronically infected with PA but actually infected with another Gram-negative bacteria. These patients were few, and it was decided by the SCFSC that it was important to use a definition of chronic PA infection that was robust and with no bias depending on clinical practice.

There are various possible explanations to the difference in prevalence of chronic PA infection in the younger cohort. First, the eradication policy may be less effective in Sweden. Copenhagen introduced in the late 1980s inhalation of colistin and oral ciprofloxacin as standard treatment once PA was detected, increasing the dosage and length of treatment if recurrent infections occurred. The Swedish centres have from the same time adopted a policy with intravenous treatment for 10 days at first infection and repeating this at recurrent infections. Only during the last few years inhaled antibiotics have been added to the eradication regimen in some centres in Sweden. Both the longer duration and the mode of antibiotic therapy in Copenhagen may be of importance for more successful eradication. Combining inhaled and systemic treatment results in high concentrations of antibiotics to both the central bronchi and sputum and to the peripheral bronchioles and alveoli.

On the other hand, the longer periods with inhaled antibiotics used by some of the intermittently colonised Copenhagen patients raise the possibility of keeping patients in the group

defined as intermittently colonised by delaying the increase of precipitins or recurrent positive cultures although the patient may be culture-positive once the treatment stops. This possibility is supported by the fact that some intermittently colonised patients used anti-pseudomonas antibiotics for more days per year in Copenhagen than chronically infected patients in Sweden.

Thirdly, the infection might be detected earlier in Denmark and Norway obtaining cultures from non-sputum producing patients more frequently than in Sweden. Longer time from the acquisition of the bacteria to the start of eradication treatment may increase the risk of eradication failure.

A fourth explanation of higher prevalence might be a higher incidence of cross infections in Swedish CF centres that do not practice segregation at the outpatient clinic or at social events like meetings and summer camps. This seems less likely since pulse field gel electrophoresis analyses do not indicate spread of epidemic strains within these centres until 2004 [28]. After this study was ended, cross-infection between Swedish CF patients during a winter camp has though been reported [29].

A higher prevalence of other Gram-negative chronic infections was found in Copenhagen (*A. xylosoxidans*, species of the *B. cepacia* complex) and Oslo (*S. maltophilia*). Reasons for this can only be speculated on, but prolonged use of broad-spectrum antibiotics might increase the risk of acquisition of resistant pathogens. A multi-centre comparison suggested a higher prevalence of resistant Gram-negative bacteria in centres with more frequent antibiotic treatment [30].

Infection with PA has been linked to a decline in lung function [3,4]. In this study we found at least as good lung function in Swedish patients despite a higher prevalence of chronic PA infection (patients <19 years old). One weakness of the study is that FEV_1 was only measured at inclusion making it impossible to evaluate the annual decline in pulmonary function in the different centres. Similar lung function in chronically infected patients in Copenhagen and the Swedish centres despite a more than 5-fold higher use of antibiotics against PA in Copenhagen, makes it important to define other factors that can affect the progress of CF lung disease. A higher prevalence in Copenhagen of patients chronically infected with *A. xylosoxidans* and species of the *B. cepacia* complex, known to cause a fast decline in lung function in infected CF patients, may be one such factor [31,32].

Some studies have shown a short term beneficial effect of beta-2 agonists on lung function in CF patients, and a long-term effect in CF patients with bronchial hyperreactivity [33]. Data on use of beta-2 agonists were not collected in this study, but the Swedish centres report that practically all their patients are given nebulised beta-2 agonists once or twice daily, in contrast to Danish and Norwegian centres where the treatment policy varies. This may be a factor of importance for the FEV_1 reported, since the spirometry procedures were not standardised in relation to inhalation of beta-2 agonists.

We also speculate that frequent use of N-acetylcysteine in Sweden may have a positive impact on the patients' pulmonary function, although N-acetylcysteine was also prescribed to Norwegian patients having lower lung function parameters. Clinical evidences of beneficial effects of N-acetylcysteine in CF are scarce [34], however results from some published studies suggest a possible positive effect of oral N-acetylcysteine on airway inflammation [35] and on lung function in CF patients [36].

Other factors, such as nutrition, physiotherapy and exercise, may also have an impact on pulmonary function. There are differences in how physiotherapy and physical exercise are performed at the different centres, but this question was not part of the study.

The differences in practical CF care and medication, including use of antibiotics, were huge (Table 3 and 4). These differences were not dependent on economical factors in the three countries but on individual decisions at each centre of how to treat. The divergences of treatment strategies between centres and countries were surprising to the members of the consortium, indicating lack of consensus and clear treatment recommendations. This implies that benchmarking between centres should include not only simple measures like lung function and weight but also treatment policies.

In conclusion, the prevalence of chronic PA infection was low in all Scandinavian CF centres compared to other published data. The prevalence among patients <19 years was significantly lower in Copenhagen compared to Sweden. Copenhagen and the Swedish centres differed both concerning mode and duration of antibiotic treatment against PA and the annual number of cultures obtained. These factors may be of importance for the observed difference in prevalence of chronic PA infection. Despite the differences in PA prevalence and use of antibiotics, the patients attending these centres had similar lung function. More studies comparing different treatment modalities and other parts of CF care are needed to define the best clinical practice, including how to use antibiotics in the most rational way.

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