Nontuberculous Pulmonary Mycobacteriosis in Denmark
Incidence and Prognostic Factors

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Rationale: **Few population-based data are available regarding nontuberculous mycobacteria (NTM) pulmonary disease epidemiology and prognosis.**
Objectives: **To examine NTM pulmonary colonization incidence, disease incidence, and prognostic factors.**
Methods: **All adults in Denmark with at least one NTM-positive pulmonary specimen during 1997 to 2008 were identified using national medical databases and were categorized as having possible or definite NTM disease or colonization.**
Measurements and Main Results: **We calculated annual age-standardized NTM incidence rates and adjusted hazard ratios (HR) of death associated with patient age, sex, comorbidity, NTM species, and NTM disease status. Of 1,282 adults with 2,666 NTM-positive pulmonary specimens, 335 (26%) had definite NTM disease, 238 (19%) possible disease, and 709 (55%) colonization only. NTM incidence rates decreased until 2002, followed by an increase from 2003 to 2008 (mean annual rate per 100,000 person-years: NTM colonization, 1.36; NTM disease, 1.08). Five-year mortality after definite NTM disease was 40.1%. After controlling for potential confounders, 5-year mortality for definite NTM disease was slightly higher than for NTM colonization (adjusted hazard ratio [HR], 1.15; 95% confidence interval [CI], 0.90–1.51). *Mycobacterium xenopi* was associated with worse prognosis (adjusted HR, 1.51; 95% CI, 0.99–2.33) than the reference *Mycobacterium avium* complex. High comorbidity level (HR, 2.97), age greater than or equal to 65 years (HR, 9.17), and male sex (female sex HR, 0.73) were predictors of death.**
Conclusions: **NTM disease incidence has remained unchanged in Denmark over the past 12 years. Patients with NTM colonization and disease have similarly poor prognosis. Negative prognostic factors include high levels of comorbidity, advanced age, male sex, and *M. xenopi*.**

Keywords: epidemiological study; atypical mycobacteria; incidence; mortality

Nontuberculous mycobacteria (NTM) are ubiquitous organisms in the environment. Some NTM are associated with pulmonary diseases, including *Mycobacterium avium* complex (MAC), *Mycobacterium kansasii*, *Mycobacterium xenopi*, *Mycobacterium abscessus*, and *Mycobacterium malmoense* (1). There are few population-based data that describe the epidemiology of NTM pulmonary disease. The reported incidence rate, based on voluntary reporting, varies from 0.7 to 1.8 cases annually per 100,000 persons worldwide (1). A French survey, based on 32 sentinel sites, reported an annual incidence rate of 0.73 cases of NTM pulmonary disease per 100,000 person-years in 2002 (2). In contrast, O'Brien and colleagues reported 1.8 cases per 100,000 person-years in a 1982 to 1983 survey of 33 U.S. states (3). Although America and Europe may differ in terms of NTM incidence and pathogen types (4–8), most studies suggest an increasing incidence of NTM isolation over the last decades, possibly due in part to the increasing prevalence of patients with immunosuppression disorders or therapy (9–11). However, the clinical significance of the increases in NTM isolation remains unclear (9, 11).

Mortality after NTM pulmonary disease appears to be high. In a recent cohort study, the 3-year mortality of patients with *M. xenopi* pulmonary disease was 69% (12). Mortality may be influenced by NTM disease severity, by the particular NTM species (8, 13), and by patient age and underlying comorbidities (12, 14). However, very limited data are available regarding prognostic factors. We therefore used Denmark's national population-based healthcare databases to conduct a nationwide epidemiological study on NTM incidence, survival, and prognostic factors. Some of the results of this study were reported previously at the 2009 European Respiratory Society Congress (15, 16).

AT A GLANCE COMMENTARY

Scientific Knowledge on the Subject

Nontuberculous mycobacterial (NTM) pulmonary disease is associated with high mortality, but few population-based data are available concerning its incidence and prognostic factors.

What This Study Adds to the Field

This nationwide population-based study provides an overview of NTM pulmonary disease incidence and patient prognosis in a Northern European country over the past 12 years. The long-term prognosis of patients with NTM colonization or disease is similarly poor. Negative prognostic factors include advanced age, a high level of comorbidity, male sex, and *Mycobacterium xenopi* disease.

METHODS

The Danish healthcare system provides tax-supported healthcare services to all residents, guaranteeing free access to hospitals and

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primary medical care. The civil registration number, a unique identifier assigned to every Danish citizen, allowed for exact linkage among all databases used in this study.

Patients with NTM Colonization and Disease

A nationwide registry at Statens Serum Institut in Copenhagen contains microbiological data from all patients in Denmark with NTM-positive specimens (17). Our study included all Danish residents aged 15 years or older with at least one NTM-positive pulmonary bacteriological specimen between January 1, 1997 and December 31, 2008. Because we did not have access to complete data for clinical and radiological features, we used modified American Thoracic Society (ATS) and Infectious Disease Society of America (IDSA) 2007 criteria to classify patients into three categories on the basis of microbiological data: definite NTM disease, possible NTM disease, and NTM colonization. For additional details and validation of these definitions by medical record review, see the online supplement.

Comorbidity Data

By linkage with the Danish National Registry of Patients, we identified all diagnoses coded during previous hospital contacts for each study subject. We then computed a Charlson Comorbidity Index (CCI) score for each individual based on the complete hospital contact history (18). The CCI includes 19 major disease categories, including risk factors and potential prognostic factors for NTM disease, such as chronic lung disease, solid and hematological malignancy, and HIV infection (18–21). Three comorbidity index levels were defined: low (score of 0), medium (1–2), and high (≥3).

For the primary survival analysis, the date of each patient's first positive sample was used to ascertain comorbidity history. This allowed us to avoid comorbidity ascertainment bias introduced by differential survival times. A second survival analysis included comorbidities recorded either before or after the NTM episode.

Statistical Analysis

Incidence. The annual incidence rates of NTM colonization and possible/definite disease from 1997 through 2008 were computed for the whole series, as well as within age- and sex-specific strata, as the number of patients with an NTM episode divided by the number of citizens in Denmark in the middle of each year (http://www.statistikbanken.dk/statbank5a/default.asp?w=1920). Incidence rates were standardized to the 2002 Danish population using 1-year age groups. Because some researchers regard all *M. gordonae* isolates as colonization episodes, regardless of fulfillment of ATS/IDSA 2007 criteria, we also calculated incidence rates that considered all *M. gordonae* episodes to be NTM colonization (1, 21–23).

Mortality. We ascertained mortality from the Danish Civil Registration System, which is updated daily. Follow-up time was computed from the index date that defined an NTM disease episode or from the date of the first sample for colonized patients until the date of death, migration, or 5 years after the index date, or April 1, 2008, whichever came first. For the prognosis analyses, we included only patients with at least one NTM-positive specimen before April 1, 2008. We used Kaplan-Meier analysis with log-rank testing to calculate and compare crude cumulative survival according to study variables. We used Cox's regression analysis to compute 3-year and 5-year mortality hazard ratios (HR) with 95% confidence intervals (CIs) associated with the following: NTM disease status (colonization [reference], possible disease, definite disease); NTM species (MAC [reference], *M. gordonae*, *M. xenopi*, *M. malmoense*, other non–rapid-growth mycobacteria [NRGM], and rapid-growth mycobacteria [RGM]), sex, age (15–39 [reference], 40–64, ≥65 yr), and level of comorbidity (low [reference], medium, and high), while adjusting for all others factors. HRs for NTM species were not adjusted for NTM disease status, as the NTM species may affect mortality by affecting disease severity. We performed two additional analyses: one that considered the presence of *M. gordonae* to be colonization and one that included comorbidities recorded either before or after NTM-positive samples. The proportional hazards assumption was assessed graphically and found to be appropriate. All Cox proportional hazards models fit the data Akaike's information criterion (AIC). Statistical analyses were performed using SAS version 9.1.

RESULTS

NTM Patients

The mean annual number of samples tested nationwide for NTM between 1997 and 2008 at the Statens Serum Institut, Copenhagen, was 18,888 samples. The total number of samples tested decreased during the first half of our study period and then remained stable. Specifically, the total decreased from approximately 30,000 samples annually in 1993 to 1994 (preceding our study period), to 24,631 in 1997, 16,660 in 2003, and 17,990 in 2008 (data from the Statens Serum Institut). Between January 1, 1997 and December 31, 2008, 2,666 cultures from pulmonary specimens (including gastric aspirations) tested positive for NTM. We identified 1,282 unique patients greater than 15 years old, 3 (0.2%) of whom had two NTM episodes associated with different NTM species several years apart and 4 (0.3%) of whom had two NTM episodes with the same species more than 3 years apart. We excluded 25 children (18 colonized, 4 possibly diseased, and 3 definitely diseased). Of the 1,282 included patients, 335 patients (26.1%) had definite NTM disease according to our criteria, 238 patients (18.6%) had possible NTM disease, and 709 patients (55.3%) showed NTM colonization. We reviewed the medical records of 30 patients (9% of all NTM patients) categorized with definite NTM disease in one of Denmark's 14 counties, North Jutland County, during the study period. We found that all 30 (100%) fulfilled ATS/IDSA 2007 criteria for definite disease and that 85% had been treated with appropriate antibiotics. Among the 21 patients (9% of all nationwide) categorized with possible NTM disease by us in the same county, we found that 19 (90%) fulfilled ATS/IDSA 2007 criteria for definite NTM disease.

Characteristics of the patients and NTM samples according to the three NTM disease status groups are presented in Table 1. The median patient age was 60.2 ± 17 years [15.1–95.9 yr] and 54.7% were male. The most prevalent comorbidity was chronic pulmonary disease (492 patients, 32.9%), of whom 349 (27.2%) were diagnosed with chronic obstructive pulmonary diseases, 133 (10.4%) with asthma, 73 (5.7%) with emphysema, 41 (3.2%) with bronchiectasis, 22 (1.7%) with cystic fibrosis, and 69 (5.4%) with other chronic lung diseases (note that some patients had more than one diagnosis recorded). Other important comorbidities were solid organ cancer (n = 117, 9.2%) and congestive heart failure (n = 80, 6.2%), whereas HIV infection was rare (n = 39, 3.0%) (Table 1). Chronic pulmonary disease before the index date was found in 61.5% of all *M. xenopi* patients and 42.6% of patients with MAC (data not shown). The prevalence of comorbidities was substantially greater when including diagnoses after the NTM index date (Table 1).

NTM Species

The four main NTM species in this Danish population were *M. gordonae* (485 patients), MAC (425 patients), *M. xenopi* (52 patients), and *M. malmoense* (46 patients). NRGM were found in 110 patients and RGM in 164 patients. For patients with definite disease, the primary NTM was MAC, followed by *M. malmoense* and *M. xenopi* (Table 1). Figure 1 shows the number of patients diagnosed with each of these NTM species per year during 1997 to 2008.

Incidence

The mean annual age-standardized incidence rate of patients with at least one NTM-positive specimen was 2.44 per 100,000 person-years between 1997 and 2008, with an incidence rate per 100,000 person-years of 1.36 for NTM colonization and 1.08 for NTM disease (0.45 for possible and 0.63 for definite NTM

TABLE 1. CHARACTERISTICS OF PATIENTS WITH NONTUBERCULOUS MYCOBACTERIA COLONIZATION, POSSIBLE NONTUBERCULOUS MYCOBACTERIA DISEASE, AND DEFINITE NONTUBERCULOUS MYCOBACTERIA DISEASE

	Colonized Patients (*n = 709*)	Possible NTM Disease (*n = 238*)	Definite NTM Disease (*n = 335*)
Age, years, mean ± SD	58.9 ± 18,4	62.7 ± 16	61.2 ± 16,5
Males	378 (53.3)	127 (53.4)	197 (58.8)
Comorbidity diagnosed before the NTM index date*			
Myocardial infarction	30 (4.2)	15 (6.3)	22 (6.5)
Congestive heart failure	42 (5.9)	12 (5.0)	26 (7.8)
Cerebrovascular disease	38 (5.4)	18 (7.6)	20 (5.9)
Chronic pulmonary disease†	188 (26.5)	80 (33.6)	154 (46)
COPD	154 (21.7)	70 (29.0)	125 (37.3)
Asthma	61 (8.6)	26 (10.9)	46 (13.7)
Bronchiectasis	16 (2.2)	8 (3.3)	17 (5.1)
Cystic fibrosis	4 (0.5)	1 (0.4)	17 (5.1)
Connective tissue disease	44 (6.2)	12 (5)	17 (5.1)
Diabetes (I and II)	36 (5.1)	8 (3.4)	13 (3.9)
Moderate to severe renal disease	13 (1.8)	8 (3.4)	8 (3.4)
Solid cancer	61 (8.6)	26 (10.9)	30 (9)
HIV infection	26 (3.7)	5 (2.1)	8 (2.4)
Low CCI score	356 (50.2)	104 (43.7)	107 (31.9)
Medium CCI score	234 (33.0)	93 (39.1)	172 (51.4)
High CCI score	119 (16.8)	41 (17.3)	56 (16.7)
Comorbidity diagnosed before or after the NTM index date*‡			
Low CCI score	194 (27.4)	55 (23.1)	50 (14.9)
Medium CCI score	253 (35.7)	94 (39.5)	165 (49.2)
High CCI score	262 (36.9)	89 (37.4)	120 (35.8)
Type of specimens for microbiological examination			
Positive smear, (%)	7.2	8.8	75.0
Positive specimens, mean ± SD; median (quartiles)	1 ± 0.07; 1 (1, 1)	1.4 ± 0.5; 1 (1, 2)	4.8 ± 4.4; 3 (3–5)
Gastric aspiration	178 (25.0)	39 (16.7)§	33 (9.9)‖
Pleural effusion	0	1 (0.4)	9 (2.7)
Lung biopsy	0	2 (0.9)	13 (3.9)
Bronchial aspiration	53 (7.5)	8 (3.5)	43 (12.3)
Bronchial wash	0	134 (58)	87 (26.0)
Sputum	474 (66.7)	68 (28.5)	278 (83.4)
NTM species			
Mycobacterium gordonae (n = 485)	392 (55.3)	75 (31.5)	18 (5.3)
Mycobacterium avium complex (n = 425)	137 (19.3)	97 (40.7)	191 (57.0)
Mycobacterium xenopi (n = 52)	14 (1.9)	12 (5.0)	26 (7.8)
Mycobacterium malmoense (n = 46)	12 (1.7)	7 (2.9)	27 (8.1)
Others NRGM (n = 110)	46 (6.5)	25 (10.5)	39 (11.6)
Mycobacterium celatum (n = 25)	11 (1.6)	2 (0.8)	12 (3.6)
Mycobacterium szulgai (n = 12)	0	5 (2.2)	7 (2.1)
Others RGM (n = 164)	108 (15.2)	22 (9.2)	34 (10.1)
Mycobacterium abscessus (n = 58)	28 (3.9)	7 (2.9)	23 (6.9)
Mycobacterium fortuitum (n = 42)	34 (4.8)	4 (1.2)	3 (1.3)

Definition of abbreviations: CCI = Charlson Comorbidity Index; COPD = chronic obstructive pulmonary disease; NRGM = non–rapid-growth mycobacteria; NTM = nontuberculous mycobacteria; RGM = rapid-growth mycobacteria.

Values are n (%) unless otherwise noted.

*NTM index date = date of the first positive NTM specimen.

† *See* text for all included pulmonary diseases. Some patients had more than one diagnosis recorded.

‡ For an explanation of the CCI score, *see* the Comorbidity Data subsection of the METHODS.

§ Thirty-five of 39 patients with positive gastric aspiration also had more than one positive pulmonary specimen.

‖ All 33 patients fulfilled criteria for definite NTM disease based on pulmonary specimen alone.

disease). When all *M. gordonae* episodes are considered colonization, the incidence rates for patients with NTM colonization and disease were 1.52 and 0.92 per 100,000 person-years, respectively. There was a peak in colonized patients in 1998 to 1999 (3.3 and 3.8 per 100,000 person-years), particularly among those aged 65 years or older (8.02 and 8.76 per 100,000 person-years, primarily due to *M. gordonae*). The annual incidence rate of possible/definite NTM disease per 100,000 person-years decreased from 1.66 in 1997 to 0.37 in 2002, followed by an increase from 0.58 in 2003 to 1.50 in 2008. The standardized incidence rates of definite and possible disease clearly increased with patient age, increasing from 0.39 in the 15- to 39-year-old age group to 1.08 in the 40- to 64-year-old age group, to 2.78 in the 65 years or older age group (*P* for trend, 0.0001) (Figure 2). In all age groups, the standardized incidence rates for females were lower than for males, with overall NTM 2.25 versus 2.74 and possible/definite disease 0.94 versus 1.25 per 100,000 person-years (*P* for trend, 0.001).

Mortality

Overall cumulative mortality 1, 3, and 5 years after detection of NTM colonization was 15.0, 27.3, and 33.5%, compared with 18.5, 33.6, and 40.1% after detection of definite disease (Figure 3). No major differences in survival were observed within the first year according to disease status (colonized, possible, or definite disease) or NTM species. In terms of crude 3- and 5-year mortality, three different survival groups appeared: patients with *M. gordonae*, other NRGM, or RGM appeared

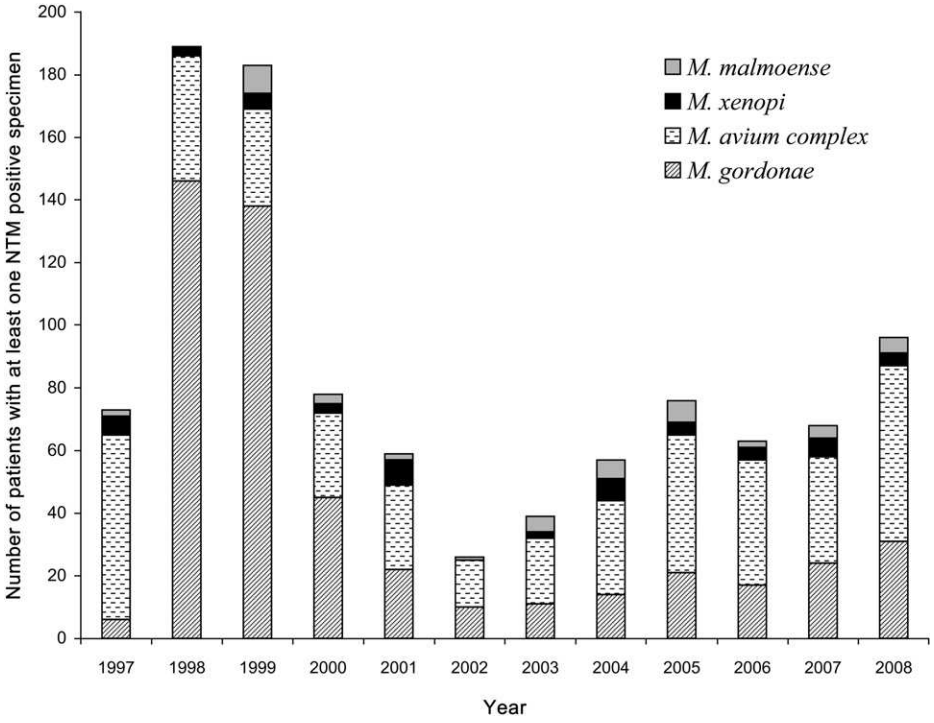

Figure 1. Number of patients per year (1997–2008) in Denmark for each main nontuberculous mycobacteria (NTM) type: *Mycobacterium avium* complex, *Mycobacterium gordonae, Mycobacterium xenopi, Mycobacterium malmoense.*

to have the best prognosis; patients with MAC had an intermediate prognosis; and patients with *M. xenopi* or *M. malmoense* had the worst prognosis (log-rank test, $P = 0.017$ at 3 yr; $P = 0.0003$ at 5 yr) (Figure 4). Three- and 5-year crude mortality also increased for patients with definite disease versus colonization, as shown in the unadjusted survival curves in Figure 4 (log-rank test, $P = 0.05$ at 3 yr and $P = 0.02$ at 5 yr) and in Table 2.

After adjustment for possible confounders, definite NTM disease showed a nonsignificant increase in mortality at 3 years (adjusted HR, 1.10; 95% CI, 0.82–1.48) and 5 years (adjusted HR, 1.15; 95% CI, 0.90–1.51), compared with NTM colonization (Table 2). *M. xenopi* tended to be associated with a worse prognosis (adjusted HR, 1.51; 95% CI, 0.99–2.33) compared with MAC. A CCI greater than 2 (adjusted HR, 2.97; 95% CI, 2.20–4.01) and age greater than 65 years compared with age less than 40 years (adjusted HR, 9.17; 95% CI, 4.98–16.86) were strong predictors of death at 5 years. Women had a better prognosis then men (adjusted HR, 0.73; 95% CI, 0.60–0.91).

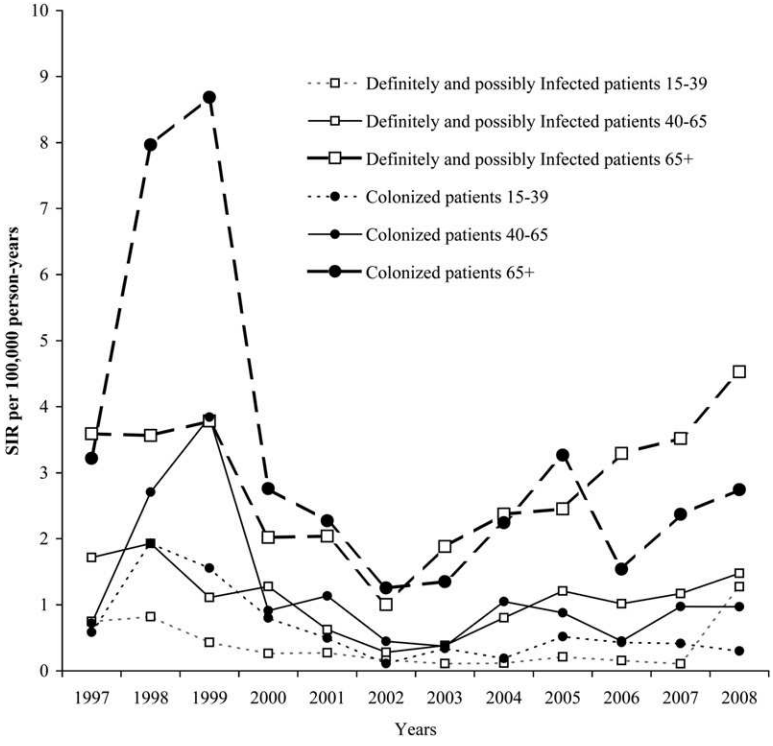

Figure 2. Standardized incidence rates (SIR) of possible or definite nontuberculous mycobacteria (NTM) disease and NTM colonization in Denmark, 1997–2008, stratified by age group. Rates were age-standardized for the Danish population in 2002 using 1-year age groups.

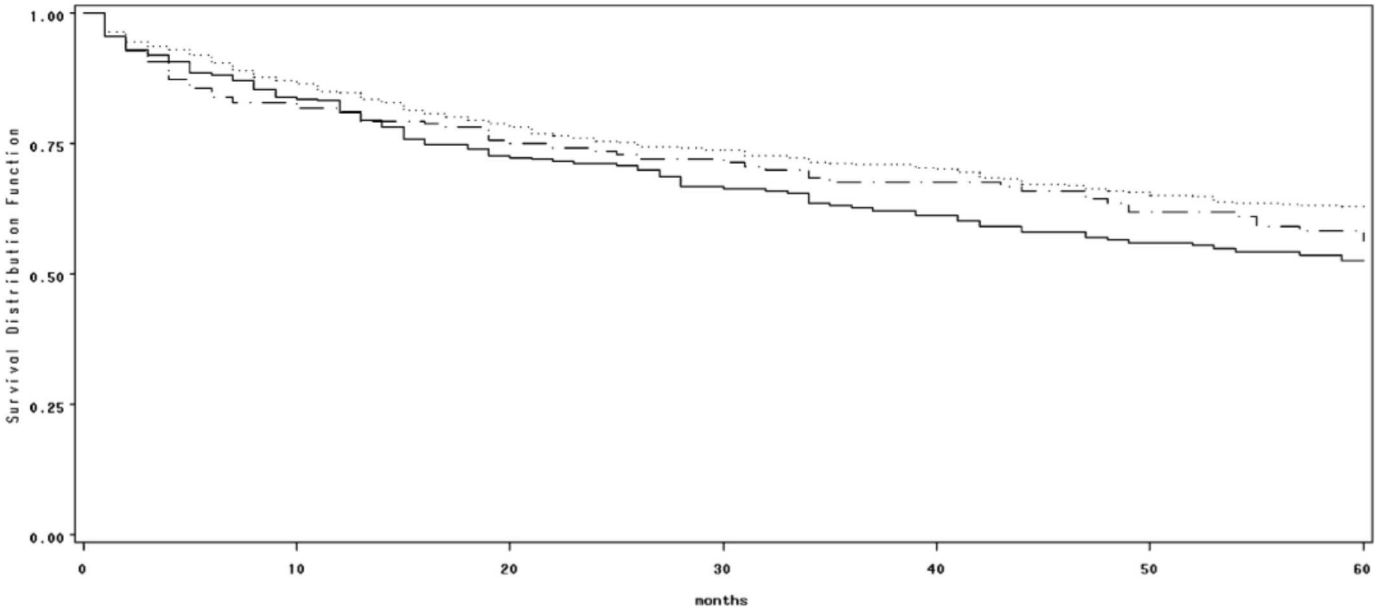

Figure 3. Five-year cumulative survival according to nontuberculous mycobacteria (NTM) disease status: colonization, possible disease, and definite disease. *Dotted line* indicates NTM colonization; *dot-and-dash* line indicates possible disease; *solid line* indicates definite disease. For definitions of the three categories of NTM disease status, please refer to the online supplement. The log-rank test of a difference between these three survival curves was not significant ($P = 0.07$).

When counting all *M. gordonae* patients as NTM-colonized patients, the HRs associated with NTM disease status, sex, age, and comorbidity were virtually unchanged.

When all comorbidities diagnosed either before or after the NTM index date were included in the model, *M. xenopi* was associated with an even worse prognosis compared with MAC (adjusted HR, 1.60; 95% CI, 1.04–2.45). For other variables, the adjusted HRs tended to be slightly lower than those derived from the primary model using only pre-NTM comorbidities.

In supplementary *post hoc* analyses, we found indications that patients with a positive smear had poorer 5-year survival than patients with a negative smear (crude HR, 1.40; 95% CI, 1.12–1.74; adjusted HR, 1.23; 95% CI, 0.96–1.58). Positive pulmonary specimens were also associated with a worse 5-year prognosis than were positive gastric aspirations (crude HR, 1.98; 95% CI, 1.48–2.65; adjusted HR, 1.20; 95% CI, 0.87–1.64).

DISCUSSION

This study provides an overview of NTM colonization and disease incidence and prognosis in a Northern European country over the last decade. To our knowledge, it is the largest

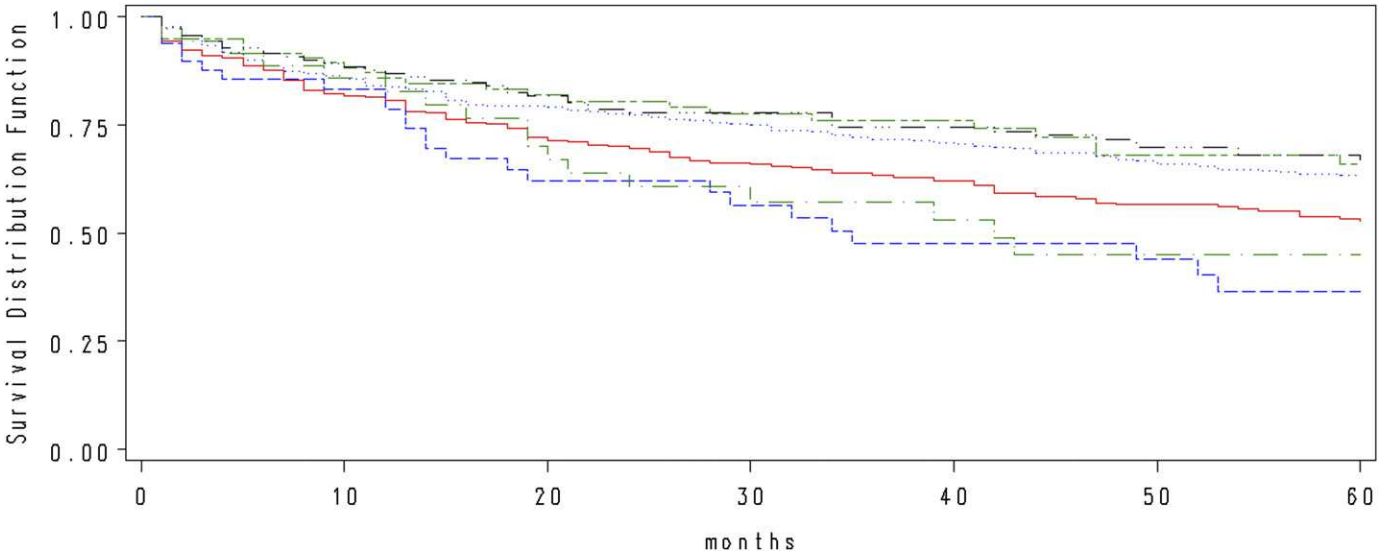

Figure 4. Five-year cumulative survival according to NTM species. *Black dot-and-dash line* indicates rapid-growth mycobacteria; *green dashed line* indicates non–rapid-growth mycobacteria; *blue dotted line* indicates *M. gordonae*; *red solid line* indicates *M. avium* complex; *green dot-and-dash line* indicates *M. malmoense*; *blue dashed line* indicates *M. xenopi*. The log-rank test of a difference between these six survival curves was significant ($P = 0.0003$).

TABLE 2. CRUDE AND ADJUSTED 3- AND 5-YEAR MORTALITY IN PATIENTS AFTER DETECTION OF NONTUBERCULOUS MYCOBACTERIAL EPISODES

	3-yr			5-yr		
	Dead (%)	Crude HR (95% CI)	Adjusted HR* (95% CI)	Dead (%)	Crude HR (95% CI)	Adjusted HR* (95% CI)
NTM disease status						
Colonization	27.3	1	1	33.5	1	1
Possible disease	29.3	1.17 (0.86–1.57)	1.10 (0.80–1.51)	36.4	1.21 (0.91–1.59)	1.13 (0.84–1.50)
Definite disease	33.6	1.33 (1.04–1.71)	1.10 (0,82–1.48)	40.1	1.35 (1.08–1.70)	1.15 (0.90–1.51)
NTM species						
MAC	33.2	1	1	39.7	1	1
Mycobacterium gordonae	27.3	0.73 (0.56–0.95)	1.00 (0.77–1.30)	34.1	0.72 (0.57–0.91)	1.01 (0.73–1.18)
Mycobacterium xenopi	44.9	1.46 (0.92–2.30)	1.37 (0.87–2.17)	51.0	1.44 (0.94–2.20)	1.51 (0.99–2.33)
Mycobacterium malmoense	38.9	1.16 (0.66–2.01)	1.12 (0.64–1.97)	47.2	1.21 (0.73–201)	1.37 (0.82–2.28)
NRGM	24.1	0.73 (0.46–1.17)	1.05 (0.69–1.60)	27.6	0.71 (0.46–1.09)	0.94 (0.63–1.38)
RGM	21.8	0.58 (0.39–0.85)	0.75 (0.49–1.15)	28.5	0.59 (0.42–0.84)	0.73 (0.50–1.07)
Age, years						
15–39	4.9	1	1	6.7	1	1
40–64	18.0	4.12 (1.98–8.55)	3.54 (1.70–7.37)	22.5	3.89 (2.08–7.27)	3.44 (1.83–6.45)
⩾65	45.5	12.37 (6.1–25.02)	9.21 (4.52–18.77)	54.9	11.99 (6.57–21.92)	9.17 (4.98–16.86)
Sex						
Male	32.5	1	1	38.9	1	1
Female	25.5	0.76 (0.61–0.95)	0.70 (0.55–0.87)	32.0	0.80 (0.65–0.98)	0.73 (0.60–0.91)
Comorbidity index†						
Low	14.6	1	1	19.5	1	1
Medium	39.0	3.25 (2.46–4.29)	2.21 (1.65–2.94)	48.1	3.21 (2.51–4.09)	2.18 (1.69–2.81)
High	46.6	4.35 (3.16–5.99)	3.16 (2.28–4.38)	50.6	4.04 (3.01–5.40)	2.97 (2.20–4.01)

Definition of abbreviations: HR = hazard ratio; MAC = *Mycobacterium avium* complex; NRGM = non–rapid-growth mycobacteria; NTM = nontuberculous mycobacteria; RGM = rapid-growth mycobacteria.

*NTM disease status adjusted using Cox proportional hazards regression analysis for NTM species, age, sex, and comorbidity; NTM species adjusted for age, sex, and comorbidity; age adjusted for NTM disease status, NTM species, sex, and comorbidity; sex adjusted for NTM disease status, NTM species, age, and comorbidity; comorbidity adjusted for NTM disease status, NTM species, age, and sex.

† Value of Charlson Comorbidity Index score at the index date.

population-based study to date to examine long-term mortality associated with NTM disease after adjustment for individual confounders. We found that the prognosis of patients with NTM colonization and disease is similarly poor. A high level of comorbidity, advanced age, and *M. xenopi* disease were strong negative prognostic factors, with *M. xenopi* disease the strongest prognostic factor.

The comprehensive Danish public healthcare system allowed this study to have a population-based design with inclusion of all NTM-positive specimens in Denmark during a 12-year period; this eliminated some of the selection problems in other studies. Another strength of the study was its comparison of colonized and diseased patients. Earlier studies of NTM incidence seldom differentiated between patients with disease versus colonization or focused only on diseased patients. We were able to adjust for a wide range of important underlying diseases at the individual patient level in the survival analysis, and the large study size provided more precise estimates than earlier studies.

Study limitations included the use of routine hospital discharge data with the possibility of some coding errors. Although preexisting comorbidities could have been discovered after the diagnosis of NTM disease, including these diagnoses did not change the observed associations. Another study limitation was the lack of clinical and radiological data in the databases. However, our medical chart review of all possibly infected NTM patients in a single county found that 90% of these patients met the ATS/IDSA 2007 criteria for definite NTM pulmonary disease. A misclassification of exposure status (i.e., disease versus colonization) would probably have resulted in more conservative mortality estimates and a smaller difference in mortality between NTM colonized and diseased patients. Finally, misclassification of comorbidity data may have led to some residual confounding, and unmeasured or unknown confounders could also have affected our mortality estimates.

Our mean annual incidence rate of 1.08 per 100,000 person-years for possible/definite NTM disease is comparable to results from New Zealand and France (2, 24). Contrary to some reports suggesting a steady increase in the incidence of NTM diseases in recent decades, we found that 2008 incidence rates in Denmark were close to 1997 levels (4, 6). As in the present study, some earlier reports described a concurrent increase in the incidence of episodes with NTM isolation (i.e., NTM colonization and disease). Over the past few years, diagnostic methods have improved for detecting and for identifying NTM, and there is a constant emergence of new NTM species. The main challenge has been to differentiate colonization from disease. In a study by Van Ingen and colleagues, only 25% of the patient met the ATS/IDSA criteria (25). In a study by Donnabella and colleagues, a dramatic increase in the incidence of *M. xenopi* isolates in a U.S. hospital was related to a more sensitive laboratory isolation technique rather than to a true increase in clinical disease (26). In our study, the decreasing incidence of patients with NTM isolation, and particularly the decrease in NTM colonization between 1997 and 2002, was probably due to a combination of improved national guidelines for the use of sterile water for gastric lavage (because many *M. gordonae* are found in lavage specimens) and a decrease in the total number of specimens tested. In contrast, and in accordance with recent findings by Billinger and colleagues (27), our data suggest that the incidence of NTM disease has increased during the last 5 years, especially in the elderly. Notably, the annual number of specimens tested in this period has been rather stable.

Because we standardized the NTM incidence rates, demographic changes in the study population cannot explain the observed incidence changes, yet we think that possible risk factors for NTM disease, such as comorbidity (5, 6, 28), have become more prevalent in Denmark because of a number of

factors including population aging, lifestyle factors, and a longer disease survival (20).

We found a high 3- and 5-year cumulative mortality of 27.3 and 33.5% after NTM colonization versus 33.6 and 40.1% after definite NTM disease. The apparent differences in survival according to disease versus colonization status were small when controlling for confounding factors. In our review of the medical charts of a subset of our patients, the vast majority were treated appropriately according to consensus guidelines, suggesting that timely and appropriate treatment of NTM disease is associated with a prognosis similar to that of NTM colonization. The small long-term survival differences might suggest that patients die primarily from severe comorbid conditions rather than from NTM disease *per se*. As suggested by ATS/IDSA, disease criteria may differ by NTM species. In our cohort, chronic obstructive pulmonary disease was closely associated with *M. xenopi* disease, possibly explaining the poor outcome of these patients. However, even when controlling for comorbidity, *M. xenopi* remained significantly associated with a worse prognosis as compared with MAC. In the two randomized British Thoracic Society studies, *M. xenopi* was also associated with the highest mortality rate (57% at 5 yr in the first study) (13, 29).

In the present study, a CCI score greater than 2 and an age older than 65 years were strong predictors of death at 5 years; this is similar to findings for other severe infections, such as pneumonia (21). It is notable that survival was higher for females than males. The reasons for this are unclear, but some hypothesize that NTM disease affects males and females differently; this has been described as the "Lady Windermere" syndrome, which is associated with a better prognosis (30).

We conclude that the current incidence of NTM disease in Denmark remains close to 1997 levels. The prognosis of patients colonized with NTM seems to be about as poor as the prognosis of patients who have NTM disease, yet appropriate treatment of the latter may have obscured a considerable difference in prognosis. A high level of comorbidity, advanced age, male sex, and *M. xenopi* disease are negative prognostic factors.

Conflict of Interest Statement: C.A. does not have a financial relationship with a commercial entity that has an interest in the subject of this manuscript; V.Ø.T.'s spouse/life partner is an employee of Nordisk and V.Ø.T. is an employee of Statens Serum Institut (SSI), each receiving more than $100,001 in compensation. V.Ø.T. received up to $1,000 from SSI as a member of the institute board and $10,001–$50,001 from GenMab in institutional grants. V.Ø.T.'s spouse/life partner holds $10,001–$50,000 in shares in Cellestis and ALK-Abello (pension) and V.Ø.T. holds $10,001–$50,000 in shares in Ambu, Roche, Novo Nordisk, and Zymogenetics (pension). I.S.J. does not have a financial relationship with a commercial entity that has an interest in the subject of this manuscript; A.R. does not have a financial relationship with a commercial entity that has an interest in the subject of this manuscript; T.L.B. received $1,001–$5,000 from BMS in advisory board fees and $1,001–$5,000 from BMS in lecture fees. P.D. does not have a financial relationship with a commercial entity that has an interest in the subject of this manuscript. H.T.S.'s Department of Clinical Epidemiology is involved in studies with funding from various companies (Amgen, Pfizer, Glaxo, and Centocor). F-X.L. does not have a financial relationship with a commercial entity that has an interest in the subject of this manuscript. R.W.T.'s Department of Clinical Epidemiology is involved in studies with funding from various companies (Amgen, Pfizer, Glaxo, and Centocor).

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