

# Incidence and prevalence of multiple endocrine neoplasia 2B in Denmark: a nationwide study

## Dear Editor,

Multiple endocrine neoplasia 2B (MEN2B) is an autosomal dominant inherited cancer syndrome associating medullary thyroid carcinoma (MTC), pheochromocytoma (PHEO), ganglioneuromatosis of the aerodigestive tract and facial, ophthalmologic and skeletal abnormalities. MEN2B is caused by the M918T and A883F mutation of the *REarranged during Transfection (RET)* proto-oncogene in approximately 95% and <5% of cases, respectively. Only very few other mutations have been reported to cause MEN2B. In approximately 75% of MEN2B patients, mutations occur as *de novo* (Wells et al. 2015).

The epidemiology of MEN2B is poorly defined. A nationwide study from Northern Ireland reported of three MEN2B patients and 1,824,000 inhabitants alive at April 21, 2012, yielding a point prevalence of 1.65 per million (Znaczko et al. 2014). However, it is unclear if this is representative of larger populations. A German study reported an MEN2B incidence (M918T carriers only) of 1.4 per million live births per year from 1991 to 2000 and estimated that at least half of all German *RET* carriers were captured (Machens et al. 2013). Meanwhile, the incidence of MEN2B and M918T carriers in a complete population is undisclosed. We conducted a nationwide study of the incidence and prevalence of MEN2B in Denmark from 1941 to 2014.

This retrospective cohort study included 12 unique MEN2B patients identified through the following sources:

1. A nationwide *RET* cohort containing all patients ( $n=1583$ ) *RET* tested in Denmark from 1994 to 2014 (Mathiesen et al. 2017b). Eight MEN2B patients were identified from this source.
2. An MTC cohort comprising 476 unique MTC patients identified from three nationwide registries: the Danish Thyroid Cancer database (DATHYRCA), the Danish Cancer Registry and the Danish Pathology Register between January 1960 and December 2014.

Inclusion criteria were histologically diagnosed MTC or cytologically diagnosed MTC verified by positive staining of calcitonin.

The DATHYRCA database has prospectively registered all patients with thyroid carcinomas in Denmark since January 1996 (Londero et al. 2014). During 1996 the database was retrospectively supplemented with 57 MTC patients diagnosed from 1960 to 1995 and mainly treated at the Department of Oncology, Copenhagen University Hospital. Based on the histology variable (HIST=5) 206 MTC patients were identified from January 1996 to December 2014.

The Danish Cancer Registry has recorded new cancer patients in Denmark since 1943. Recording became mandatory in 1987 (Gjerstorff 2011). Based on the combination of codes (ICD7 topography 1940; 4940; 5940; 6940; 8940; 9940, TOPO1 topography 1939 and MORFO1 morphology 83463; 85101; 85103; 85113; 85123 for the period from 1943 to 1977 and ICD-10 topography DC739 and MORFO3 morphology 83453; 83463; 83473; 85101; 85103; 85123 for the period from 1978 to 2014) 459 MTC patients were recorded from 1943 to December 2014. No MTC patients appeared before 1960.

The Danish Pathology Register has registered diagnosis of pathological specimens in Denmark since September 1968 (www.patobank.dk). Registration became mandatory in 1997. Based on the combination of topographical (T96000; T96010; T96050; T96100; T96200; T96300; T96400; T96500) and morphological SNOMED codes (M85103; M85104; M85106; M85107) 379 MTC patients were identified.

Thus, 263, 459 and 379 MTC patients were found in the DATHYRCA database, the Danish Cancer Registry and the Danish Pathology Register, respectively. Removal of 145 duplicates and 217 triplicates yielded 522 unique MTC patients. Subtraction of those not fulfilling inclusion criteria resulted in 476 patients (474

diagnosed by histology and 2 by cytology). Medical records were available for 407 patients. A review revealed 11 MEN2B patients.

- A PHEO cohort accounting for 478 unique patients with histologically proven PHEO identified from the Danish Pathology Register between September 1968 and December 2014. Topographical (T93000; T93010; T93020; T93100; T93110; T93120; T93130; T93200; T93300) and morphological SNOMED codes (M87000; M87001; M87003; M87004; M87006; M87007; M87009) were used. Among the 24 patients present in both the PHEO and MTC cohort 20 had MEN2A and four had MEN2B.
- A nationwide collaboration of endocrinologists from all relevant university hospitals (Copenhagen, Aarhus, Odense and Aalborg). Upon inquiry, each endocrinologist has contributed with all MEN2B patients registered locally. This provided eight MEN2B patients.
- A systematic literature search performed on October 21, 2016 in the following databases: Cochrane, Embase, PubMed, Scopus and Web of Science with the search term 'medullary thyroid carcinoma Denmark' OR 'medullary thyroid carcinoma Danish OR multiple endocrine neoplasia 2 Denmark OR multiple endocrine neoplasia 2 Danish'. No filters were employed.

A total of 211 citations were found. Removal of seven duplicates and two triplets yielded 200 unique citations, of which 61 had Danish affiliations. Full-text was retrieved for all citations. Among the 61 citations nine reported of

Danish MEN2B patients. The reference list of each citation was scrutinized to uncover patients published more than once. If needed an author of the concerned publication was contacted for clarification. Five of the nine citations mentioned a patient reported elsewhere. Two of the remaining four citations reported of patients already identified through the Danish *RET* cohort. The final two citations described a total of five patients (Rasmussen 1980, Emmertsen 1984). The author of each citation was contacted and four of the five MEN2B patients were identified.

- A four-generation pedigree was created for all 10 MEN2B index patients using the Civil Registration System ([www.cpr.dk](http://www.cpr.dk)) and the Danish National Archives ([www.sa.dk/en/](http://www.sa.dk/en/)). Two additional MEN2B patients were ascertained.

Data were collected from medical records and when insufficient supplemented by previous publications of the patients.

Point prevalence was calculated as the number MEN2B patients alive at January 1 divided by the number of inhabitants alive at the same date. Incidence was calculated as the number of MEN2B patients born in each decade divided by the number of live births in Denmark for the respective decade. Patients born between 1941 and 1970 all died before *RET* testing was available and were solely diagnosed by the MEN2B phenotype. Patients born between 1971 and 2014 were diagnosed both by phenotype and by a verified *RET* germline mutation (M918T or A883F). Thus, to estimate the incidence of

**Table 1** Demographic, clinical and follow-up data of 12 Danish MEN2B patients.

Patient no.	Sex	RET mutation	Mucosal neuroma	Marfanoid habitus	At histological adrenal diagnosis		At histological thyroid diagnosis			At last follow-up	
					Age (years)	PHEO	Age (years)	MTCTNM <sup>a</sup>	Procedure	Age (years)	Status
1 <sup>b</sup>	F	M918T	Yes	Yes	27.1	Unilateral	17.6	T4aN1bM0	TTX+LND	27.7	Alive
2	F	M918T	No	No		None		TxNxMx	None	0.5	Dead
3	F	M918T	Yes	No		None	0.8	T1aN0M0	TTX	4.2	Alive
4	M	M918T	Yes	Yes		None	9.1	T1aN0M0	TTX+LND	14.7	Alive
5	F	M918T	Yes	No	20.4	Bilateral	15.0	T3N1aM0	TTX+LND	23.4	Alive
6	F	M918T	Yes	No	33.8	Unilateral	17.5	T3N0M0	TTX	35.9	Alive
7	F	M918T	Yes	Yes		None <sup>c</sup>	25.7	T4aN1bM0	TTX+LND <sup>d</sup>	29.2	Dead
8	M	A883F	Yes	Yes	11.9	Bilateral	10.9	T1aN1bM0	TTX+LND	19.1	Alive
9	M	NA	Yes	Yes		None	16.7	T4aN1bM0	ST+LND	39.2	Dead
10	M	NA	Yes	Yes	36.9	Bilateral	17.3	T2N0M0	HT	36.9	Dead
11	M	NA	Yes	Yes		None <sup>e</sup>	13.6	T1aN0M0	TTX	21.6	Dead
12	F	NA	Yes	NA	33.4	Bilateral	33.5	T1bN1aM0	TTX+LND	45.5	Dead

<sup>a</sup>According to the 7th Edition of the American Joint Committee on Cancer; <sup>b</sup>the mother of patient no. 2 and 3; <sup>c</sup>PHEO diagnosed at age 26.2 years by biochemistry and imaging exclusively; <sup>d</sup>included laryngectomy; <sup>e</sup>PHEO diagnosed at age 13.6 years by biochemistry exclusively.

F, female; HT, hemithyroidectomy; LND, lymph node dissection; M, male; MEN, multiple endocrine neoplasia; MTC, medullary thyroid carcinoma; NA, not available; PHEO, pheochromocytoma; *RET*, *REarranged during Transfection*; ST, subtotal thyroidectomy; TNM, tumor, node, metastases; TTX, total thyroidectomy.

**Table 2** Incidence of MEN2B in Denmark according to decade and mutation.

	1941–1950 (853,113 live births)	1951–1960 (761,648 live births)	1961–1970 (792,197 live births)	1971–1980 (672,095 live births)	1981–1990 (557,279 live births)	1991–2000 (673,654 live births)	2001–2010 (643,356 live births)	2011–2014 (229,657 live births)
	Patients, <i>n</i> (per 1 million and year)	Patients, <i>n</i> (per 1 million and year)	Patients, <i>n</i> (per 1 million and year)	Patients, <i>n</i> (per 1 million and year)	Patients, <i>n</i> (per 1 million and year)	Patients, <i>n</i> (per 1 million and year)	Patients, <i>n</i> (per 1 million and year)	Patients, <i>n</i> (per 1 million and year)
A883F	NA	NA	0	0	0	1 (1.5)	0	0
M918T	NA	NA	0	2 (3.0)	1 (1.8)	1 (1.5)	1 (1.6)	2 (8.7)
MEN2B <sup>a</sup>	3 (3.5)	1 (1.3)	0	2 (3.0)	1 (1.8)	2 (3.0)	1 (1.6)	2 (8.7)

<sup>a</sup>Patients born between 1941 and 1970 died before *RET* testing was available and were diagnosed by phenotype. Patients born between 1971 and 2014 were diagnosed by phenotype and genotype.

MEN, multiple endocrine neoplasia.

MEN2B in regards to mutation carriers the period of 1971–2000 was chosen, taking the precaution that some MEN2B born from 2001 to 2014 might still be unrecognized (mean age at diagnosis; 13 years (Brauckhoff et al. 2014)). Danish population data were retrieved from Statistics Denmark (www.statbank.dk). All analyses were done using Stata 14.1 (StataCorp, USA). The investigation was approved by the Danish Health Authority (3-3013-395/2) and the Danish Data Protection Agency (13/19275).

A total of 12 MEN2B patients from 10 unrelated families were included. Demographic, clinical and follow-up data are shown in Table 1. All but patient 2 and 3 were classified as having a *de novo* mutation. The remaining 10 patients were recorded as index patients.

The point prevalence at January 1, 2015, was 1.06 per million (95% CI: 0.39–2.30) (six MEN2B patients and 5,659,715 inhabitants alive). If only including index patients in analysis, the point prevalence at January 1, 2012 and 2015, were 0.90 (95% CI: 0.29–2.10) and 0.88 (95% CI: 0.28–2.06) per million, respectively.

Table 2 depicts the incidence of MEN2B by decade and mutation. The incidence from 1971 to 2000 was 2.6 (95% CI: 0.85–6.13) per million live births per year. If subdivided by mutation, the incidences of M918T and A883F carriers were 2.1 (95% CI: 0.57–5.38) and 0.5 (95% CI: 0.01–2.93) per million live births per year, respectively. In a German study, the incidence of M918T carriers from 1971 to 2000 was 1.0 per million live births per year (Machens et al. 2013). The differences in incidence probably reflect more or less complete ascertainment rather than a genuine difference.

This study shares limitations inherent with retrospective studies of rare diseases. Small sample sizes limiting generalization are often seen when studying rare diseases, as in this study. To increase sample size, we depleted virtually all possibilities to identify MEN2B patients in Denmark.

Also, the MEN2B diagnosis was not verified by *RET* mutations in four (no. 9–12) of the 12 included patients.

However, three (no. 9–11) demonstrated endocrine (MTC), musculoskeletal (marfanoid habitus, femoral epiphysiolysis, pes cavus, scoliosis), intestinal (constipation) and oral (mucosal neuromas) manifestations consistent with MEN2B. Pertinent data for the remaining patient (no. 12) revealed MTC, bilateral PHEO, constipation and neuromas of the tongue, also consistent with MEN2B.

The A883F carrier introduces heterogeneity into the study. However, inclusion of this carrier seems appropriate when calculating the overall MEN2B prevalence and incidence as such carriers have been identified in at least five other countries (Mathiesen et al. 2017a).

The point prevalence of MEN2B index patients in Denmark was 0.90 per million at January 1, 2012. In a nationwide study consisting solely of MEN2B index patients from Northern Ireland, the point prevalence was 1.65 per million at April 21, 2012 (Znaczkowski et al. 2014). This is considerably higher than seen in Denmark. However, no statistical significant difference can be proven. This might be due the small sample sizes.

In conclusion, the incidence and prevalence of MEN2B in this nationwide study does not differ significantly from that reported in Germany and Northern Ireland, respectively.

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#### Declaration of interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

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#### Author contribution statement

J S Mathiesen conceived and coordinated the study, collected data, performed statistical analyses and drafted the manuscript. J P Kroustrup, P Vestergaard, M Madsen, K Stochholm, P L Logstrup, Å K Rasmussen, U Feldt-Rasmussen, S Schytte, H B Pedersen, C H Hahn, J Bentzen, M Gaustadnes, T F Ørntoft, T v O Hansen, F C Nielsen, K Brixen, A L Frederiksen and C Godballe participated in data collection, statistical analyses and drafting of the manuscript.

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