Mutations in the *MEN I* gene in sporadic neuroendocrine tumours of gastroenteropancreatic system

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Neuroendocrine tumours of the gastroenteropancreatic (GEP) system occur in sporadic as well as hereditary forms. About one tenth of these tumours are associated with a familial cancer syndrome—multiple endocrine neoplasia (MEN) type I (MEN I). These tumours appear to originate embryonically from organs attributable to the foregut.[1] The *MEN I* gene has been identified by positional cloning. This gene is localised on chromosome 11q13 and encodes a polypeptide (menin) of 610 aminoacid residues. The function of this putative tumour suppressor gene is as yet unknown.[2] With microsatellite markers around the menin-gene region, loss of heterozygosity (LOH) was observed in sporadic neuroendocrine GEP tumours. However, by contrast with existing clinical data[1] LOH was not only found in foregut, but also in midgut and hindgut tumours.[3]

We analysed tumour tissue DNA from 43 patients with sporadic tumours. Mutation screening by the single strand conformation polymorphism (SSC) technique.[4] The open reading frame of 1830 bp was covered by 15, partly overlapping PCR fragments. Primer sequences and PCR conditions are available at http://www.mpimg-berlin-dahlem-mpg.de/abt_rop/moleculargen/men1.html. Several bandshifts were detected in different exons and direct sequencing of the respective PCR products revealed six mutations in sporadic tumours (table). Patients had histologically proven neuroendocrine GEP tumour disease.[5] Patients with sporadic tumours were stratified in four groups, according to the location of the primary tumour: 1 foregut (n=23; stomach 3, pancreas 18, duodenum 2); 2 midgut (n=13; jejunum 1, ileum 8, caecum 3, appendix 1); 3 hindgut (n=3); 4 unknown primary (n=4). Tumour tissues of 18 patients with GEP adenocarcinomas (colorectum, 14; pancreas, 3; stomach 1) served as negative controls.

In six out of 43 patients with sporadic tumours of the foregut, midgut, and hindgut, mutations of the menin gene were identified only in foregut tumours. Affected patients had (age in years tumour size in cm at first diagnosis): solitary benign insulinoma (40/1), metastatic, pancreatic VIPoma (31/6); metastatic (mainly liver) non-functional, pancreatic neuroendocrine tumours (43/6, 51/3, and 76/2); and non-metastatic, non-functional hepatic neuroendocrine tumour (32/14).

Our findings show that in a subset of sporadic neuroendocrine tumours, mutations of the MEN I gene can be found. This suggests that these tumours are also subject to somatic mutation. Consistent with our clinical findings, mutations in the MEN I gene appear to be restricted to neuroendocrine GEP tumours of the foregut (6/23), because no mutations were found in midgut and hindgut neuroendocrine tumours nor tumours with unknown primary (0/20) nor GEP adenocarcinomas (0/18). Possible mechanisms of tumorigenesis include an undetected mutation that inactivates the second MEN I allele or its expression, a dominant-negative effect of the mutation, or the involvement of additional, so far, unidentified altered genes.

1 Scherübl H, Bredeek U, Janson ET, et al. Neuroendocrine gastroenteropancreatic tumors: are they hereditary? *Gastroenterol* 1997; 112 (suppl): A651.

2 Chandrasekharappa SC, Guru SC, Manickam P, et al. Positional cloning of the gene for multiple endocrine neoplasia-type 1. *Science* 1997; 276: 404–07.

3 Jakobovitz O, Nass D, DeMarco L, et al. Carcinoid tumors frequently display genetic abnormalities involving chromosome 11. *J Clin Endocr Metab* 1996; 81: 3164–67.

4 Berger W, van de Pol D, Warburg M, et al. Mutations in the candidate gene for Norrie disease. *Hum Mol Genet* 1992; 1: 461–65.

5 Zimmer T, Stölzel U, Bäder M, et al. Darrhea as the only symptom and normal serum gastrin concentrations in a duodenal gastrinoma. *N Engl J Med* 1995; 333: 634–36.

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Patient	SSCP pattern		Exon	Type of mutation	Sequence alteration in tumour DNA	Menin defect
	Blood	Tumours				
1	+/–	–	5/6	transition, splice acceptor site	A5296G	
2	na	+/–	5	1 bp deletion, frameshift	5187delG[1]	W265...X
3	+	+/–	8	7 bp deletion, 5 bp from intron 7, 2 bp from exon 8	6618del7[1]	
4	na	+/–	2	transition missense	G2411A[1]	G42S
5	na	–	2	deletion 3 bp in frame	2641del3	K119del
6	na	+/–	10	transition missense	G7848A[1]	A541T

NA=not available. *Numbers refer to sequence accession number U93237; +: normal allele; –: mutant allele; [1]: in these cases, the mutant as well as the normal allele were identified by DNA sequencing.

MEN I gene mutations in six of 43 sporadic neuroendocrine gastroenteropancreatic tumours

Pseudolymphoma at site of clonidine patch

Walter B Shelley, E Dorinda Shelley

An 83-year-old woman consulted us for evaluation of a 1·0 cm nodule on her right anterior shoulder (figure). It was symptomless and had arisen at the site of clonidine patches applied during the past year. No lesions were seen on her legs where the patches are now being applied. Her past history included left hemiplegia, aphasia following a right cerebrovascular accident 5 years ago, and bullous pemphigoid. Her blood count was normal.

A skin biopsy specimen showed no changes in the

Nodule on right shoulder