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LETTER TO EDITOR

Mannan-binding lectin-associated serine protease-2 (MASP-2) deficiency in two patients with pulmonary tuberculosis and one healthy control

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Hereditary complement deficiencies are usually associated with increased susceptibility to infections and/or autoimmune diseases. Generally, they are rare: the majority have been detected in no more than a few dozen individuals. However, due to lack of population-based studies, such cases are usually detected by chance, e.g., in severely ill persons and their family members. An exception is mannan-binding lectin (MBL) deficiency, affecting perhaps 5%–10% of the population. MBL, like collectin-10, -11 and the ficolins (M-, L-, H-), is a pattern-recognition molecule, cooperating with MBL-associated serine proteases (MASPs) in the initiation of complement activation *via* the lectin pathway. In contrast, deficiency of other factors of the lectin pathway seems to be very rare.

Within the MASP family, three proteases (MASP-1, MASP-2, MASP-3) and two related, non-enzymatic proteins, MAp19 (sMAP) and MAp44 (MAP-1), have been identified. MASP-1, MASP-3 and MAp44 are the products of alternative splicing of the *MASP1/3* gene while MASP-2 and MAp19 are synthesized under the control of the *MASP2* gene. MASP-2 is responsible for activation of complement factors C4 and C2. MASP-2 is composed of six domains: CUB1, EGF, CUB2, CCP1, CCP2 and the serine protease domain.[1]

The afore-mentioned *MASP2* gene, comprising 12 exons, is localized to chromosome 1p.36.2-3. Among its single-nucleotide polymorphisms (SNPs), three: p.R99Q (c.296G>A), p.D120G (c.359A>G) and p.P126L (c.377C>T), affect the structure of the CUB1 domain. Another SNP, p.H155R (c.464A>G) and a duplication p.156_159dupCHNH (c.466_477TGCCACAACCAC) are located in the fourth (d) exon, encoding the EFG-domain. The p.V377A (c.1103T>C) SNP affects the structure of the CCP2 domain.[1] As MASP-2 shares its first two domains with MAp19, almost all (with an exception of p.V377A) mentioned polymorphisms, influence the structure of the latter.

Three (p. R99Q, p.D120G, p.V377A) polymorphisms have been found among Caucasians. The p.D120G SNP, in a homozygous state, is the primary cause of MASP-2 deficiency.[1] It leads to an exchange of aspartic acid for glycine at residue 120 in the CUB1 domain,[2] resulting in a reduced serum level of MASP-2 and MAp19. The main consequence of the G/G genotype is, however, in preventing formation of complexes with lectins[2] (as the collagen-binding sites are located in the CUB1–EGF–CUB2 domains). Thus, G/G homozygotes are practically unable to activate the lectin pathway of complement. Although perhaps >10% individuals within Caucasian populations may be heterozygous, the frequency of total MASP-2 deficiency has been estimated as 6/10 000.[1] The cases of MASP-2 deficiency first described suggested clear disease associations;[2,3] however, several later reports revealed G/G genotype carriers among healthy controls as well.[4–7] Totally, homozygosity for the p.D120G mutation has been found in eight patients suffering from various diseases and six healthy controls (Table 1B).

We report three other instances of MASP-2 deficiency—two in persons with pulmonary tuberculosis (among 440 tested) and one in a perfectly healthy individual (Table 1A). One of the patients was a 72-year-old male, diagnosed also with chronic obstructive pulmonary disease and angina pectoris. He was infected with a strain of *Mycobacterium tuberculosis* resistant to streptomycin, isoniazid and ethambutol. The other tuberculosis patient was a 36-year-old woman without any comorbidities. No case of tuberculosis was found among their closest relatives. Moreover, among

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Table 1 Characterization of MASP-2-deficient subjects identified in this work (A). Reported previously cases of MASP-2 inherited deficiency, having or not clinical associations (B)

A

Age	*Sex*	*MASP-2 concentration*[a,b] *(ng/ml)*	MBL2 *genotype*	*MBL concentration*[a] *(ng/ml)*	*MBL–MASP-2 activity*[a,b] *(mU/ml)*	*MBL–MASP-1 activity*[a] *(mU/ml)*
Pulmonary tuberculosis patients						
72	M	41	HYA/LYB	472	85	455
36	F	169	HYA/LXA	1121	64	249
Healthy individual						
29	M	44	LXA/LXA	719	68	307

B

Number of cases	*Ethnicity*	*Description*	*Reference*
1	Danish	Adult with autoimmune diseases and severe infections (including pneumonias and sepsis)	Stengaard-Pedersen *et al.*, 2003[2]
1	Polish	Pediatric patient with recurrent pneumonias	Cedzynski *et al.*, 2004[3]
1	Danish	Pediatric patient with cystic fibrosis	Olesen *et al.*, 2006[9]
2	Danish	Adult patients of lung clinics	Thiel *et al.*, 2007[1]
1	Italian	Adult with hepatocellular carcinoma	Segat *et al.*, 2008[10]
1	Polish	Pediatric patient with recurrent upper respiratory infections and skin abscesses	Cedzynski *et al.*, 2009[8]
1	Danish	Adult with colorectal cancer	Ytting *et al.*, 2011[11]
1	British		Stover *et al.*, 2005[6]
2	Spanish		Garcia-Laorden *et al.*, 2006[4]
1	Italian	All healthy adults	Segat *et al.*, 2008[10]
2[c]	Spanish		Garcia-Laorden *et al.*, 2008[5]
1	Polish		Olszowski *et al.*, 2013[7]

Abbreviations: MASP, mannan-binding lectin-associated serine protease; MBL, mannan-binding lectin.
[a] Average (median) values among Polish healthy adults: 375 ng/ml (MASP-2 concentration; $n=164$); 877 ng/ml (MBL concentration; $n=394$); 383 mU/ml (MBL–MASP-2 activity; $n=265$); 354 mU/ml (MBL–MASP-1 activity; $n=260$) (unpublished).
[b] Values corresponding to the fifth percentile of Polish healthy adults: 171 ng/ml (MASP-2 concentration); 62 mU/ml (MBL–MASP-2 activity).
[c] One case identified for the first time.

276 healthy controls, we found a 29-year-old man, carrying the p.D120G mutation on both alleles. Chickenpox during childhood was the most serious disease reported by him. Patients came from the Voivodeship Hospital of Lung Diseases in Jaroszowiec and the Masovian Center of Lung Diseases and Tuberculosis Treatment in Otwock, Poland. The serum and DNA from healthy volunteers was obtained from APC Medical Analyses Laboratory, Lodz, Poland. Approval of the local ethical committee was obtained, as was the written informed consent of patients.

All three subjects had low MASP-2 concentrations and low MBL–MASP-2 complex activities (Table 1A). Additionally, one of the tuberculosis patients had a *MBL2* gene mutation, at codon 54 of the exon 1 (B variant allele), affecting both MBL serum concentration and activity (Table 1A). Two cases of MASP-2 deficiency previously reported by us were a 12-year-old boy with recurrent pneumonias and a 4-year-old girl with recurrence of upper respiratory infections and skin abscesses.[3,8] Detailed diagnostics (both were patients of the Unit of Immunodisorders) showed no other immune abnormalities. They had LYA/LXA and HYA/LXA *MBL2* genotypes with serum MBL concentrations of 1.4 μg/ml and 1.8 μg/ml, respectively, while MBL–MASP-2 complex activity was undetectable.[3,8] Taking into account our experiences as well as data published by others (Table 1), it might be suspected that MASP-2 deficiency is mainly associated with respiratory disease and/or certain types of cancer. However, as seven examples have been found in healthy controls (compared with 10 patients with serious diseases), it equally might be disease modifier, potentially important when accompanied by other factors, as suggested by Thiel *et al.*[1] and Olszowski *et al.*[7] Thus, the clinical impact of MASP-2 deficiency remains uncertain.

Another issue is the frequency of the G/G genotype. It was originally estimated as six cases per 10 000 individuals.[1] However, more recent data from healthy controls: two cases per 868 (frequency: 0.0023),[4] 2/1447 (0.0014),[5] 1/596 (0.0017),[6] 1/164 (0.0061)[9] or 1/276 (0.0036, this investigation) suggest that MASP-2 deficiency may be commoner than previously thought.

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