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Genetic susceptibility factors for multiple chemical sensitivity revisited

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ABSTRACT

Multiple chemical sensitivity (MCS) is characterised by adverse effects due to exposure to low levels of chemical substances. Various genes, especially genes of importance to the metabolism of xenobiotic compounds, have been associated with MCS, but findings are inconsistent. The purpose of this study was to investigate genetic susceptibility factors for MCS and self-reported chemical sensitivity in a population sample. Ninety six MCS patients and 1,207 controls from a general population divided into four severity groups of chemical sensitivity were genotyped for variants in the genes encoding cytochrome P450 2D6, arylamine N-acetyltransferase 2, paraoxonase 1, methylene tetrahydrofolate reductase, and the cholecystokinin 2 receptor. No hypotheses were consistently confirmed. An apparent association between number of active cytochrome P450 2D6 alleles and MCS status was not statistically significant (OR=1.2, p=0.28). Fast arylamine N-acetyltransferase 2 metaboliser status was associated with severity of chemical sensitivity only in the most severely affected group in the population sample (OR=3.1, p=0.04). The cholecystokinin 2 receptor allele with 21 CT repeats was associated with MCS when compared in *post hoc* analyses with all individuals from the population sample (p=0.02). Genetic variants in paraoxonase 1 and methylene tetrahydrofolate reductase were not associated with MSC or with self-reported chemical sensitivity in the population sample. Our results suggest that variants in the genes examined are of less importance to MCS than previously reported or that gene-environment interactions or significant degrees of genetic heterogeneity in MCS underlie inconsistent findings in the literature.

Introduction

Individuals experiencing multiple chemical sensitivity (MCS) report symptoms from various organs related to inhalation of multiple unrelated airborne chemicals in concentrations below what is normally associated with toxicological responses (1999 Consensus on Multiple Chemical Sensitivity, 1999; Cullen, 1987; Lacour et al., 2005). A number of hypotheses concerning the etiology and pathophysiology of MCS have been proposed (Winder, 2002), including impaired ability to metabolise toxic chemicals (Cullen, 1987; Winder, 2002) and psychological mechanisms (Labarge and McCaffrey, 2000).

The existence of several well known polymorphisms affecting the activity of enzymes metabolising xenobiotica have prompted research in whether these polymorphisms are associated with MCS and with chemical sensitivity in general populations. In a Canadian study, differences between MCS patients and healthy controls were found for polymorphisms in the genes encoding the enzymes cytochrome P450 2D6 (CYP2D6), arylamine N-acetyltransferase 2 (NAT2), and paraoxonase 1 (PON1) (McKeown-Eyssen et al., 2004). A polymorphism in the gene encoding methylene tetrahydrofolate reductase (MTHFR) was also examined, since vitamin B12 deficiency might be related to neurological symptoms (Carmel, 2000), a key feature of MCS (Lacour et al., 2005) and self-reported chemical sensitivity (Berg et al., 2009). Further, decreased levels of homocysteine have been found in MCS patients (Baines et al., 2004). Recently, however, opposite or no associations have been described between 'chemical-related

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sensitivity' and self-reported MCS and polymorphisms in NAT2 and PON1 (Schnakenberg et al., 2007; Wiesmuller et al., 2008).

Anxiety is often reported together with MCS (Papo et al., 2006). Therefore, a dinucleotide repeat in the promoter region of the gene encoding the cholecystokinin 2 receptor (CCK2R), formerly identified as the CCK-B receptor (Dufresne et al., 2006), associated with panic disorder (Kennedy et al., 1999) was investigated in relation to MCS, and a positive association between MCS and the allele with 22 CT repeats also associated with panic disorder was found (Binkley et al., 2001).

The discrepancies in the results from polymorphisms in *NAT2* and *PON1* described above have been argued to be a result, at least in part, of different study designs and case definitions (Schnakenberg et al., 2007). Furthermore, in general, positive findings from genetic association studies seem to be difficult to replicate (Lohmueller et al., 2003) and possibly inflated by chance (Ioannidis et al., 2001). Therefore, we aimed to investigate the effects of gene variants in *CYP2D6*, *NAT2*, *PON1*, *MTHFR*, and *CCK2R* on the risk of MCS in a sample of MCS patients and a sample of individuals from a general population, and expected to find an excess of individuals with CYP2D6 extensive and NAT2 fast metaboliser genotypes, and heterozygous in the two examined *PON1* polymorphisms in the patient sample, as well as a positive association between slow NAT2 metaboliser status and severity of chemical sensitivity in the population sample. Furthermore, we also expected to find associations between MCS and the C allele in the C677T polymorphism in *MTHFR*, since the T allele has been shown to be associated with increased levels of homocysteine (Kang et al., 1991), and also with the alleles with 21 and 22 CT repeats in *CCK2R*.

Materials and methods

Information on sensitivity related to airborne chemicals was collected with a standardised questionnaire in a sample of individuals diagnosed with MCS and in a population sample.

Participants

The patient sample consisted of individuals consecutively diagnosed with MCS according to Cullen's criteria (Cullen, 1987) (n=136) by an experienced ear nose throat specialist (SV) between January 1st 1990 and January 1st 2007 at Copenhagen University Hospital, Rigshospitalet and at Private Hospital Hamlet, Denmark. Of these MCS patients, five individuals were excluded from the study because of non-Danish ethnicity, leaving 131 potential participants, of whom 97 individuals (74.0%) agreed to participate in the study. DNA was obtained from 96 individuals (73.3%), and a questionnaire was returned by all 96 participating patients (100%).

Individuals from the population sample (n=1,216) had previously participated in a population-based clinical health examination focused on allergic diseases (The Copenhagen Allergy Study) conducted between October 1997 and November 1998. The participation rate for the health examination as well as characteristics of participants and non-participants have been described elsewhere (Linneberg et al., 2000, 2001). All participants from the population sample were Danish citizens born in Denmark. In the period between the health examination and the initiation of the present study, 40 participants had died, 15 had moved outside Denmark, eight had requested not to be contacted again, and three no longer had a known address, leaving 1,150 potential respondents to the questionnaire. The response rate to the questionnaire after one reminder was 83.2% (n=957). Five questionnaires (0.4%) were returned because the address was unknown, eight individuals (0.7%) declined to participate for various reasons, and 180 individuals (15.7%) did not respond. Ten gift vouchers of DKK500, corresponding to €80, were rewarded to randomly selected respondents in an attempt to enhance the response rate.

Table 1
Demographic characteristics of patients with multiple chemical sensitivity and individuals from the population sample.

		Patients		Population		Total	
		n	(%)	n	(%)	n	(%)
Sex	Women	80	(83.3)	550	(57.5)	630	(59.8)
$p<0.001$[a]	Men	16	(16.7)	407	(42.5)	423	(40.2)
	Total	96	(100.0)	957	(100.0)	1,053	(100.0)
Age	≤25	1	(1.0)	21	(1.7)	22	(1.7)
$p<0.001$[b]	25–30	0	(0.0)	122	(10.0)	122	(9.3)
	30–35	4	(4.2)	105	(8.6)	109	(8.3)
	35–40	6	(6.2)	135	(11.1)	141	(10.7)
	40–45	9	(9.4)	170	(14.0)	179	(13.6)
	45–50	11	(11.5)	164	(13.5)	175	(13.3)
	50–55	23	(24.0)	116	(9.5)	139	(10.6)
	55–60	20	(20.8)	86	(7.1)	106	(8.1)
	60–65	16	(16.7)	92	(7.6)	108	(8.2)
	>65	6	(6.2)	205	(16.9)	211	(16.1)
	Total	96	(100.0)	957	(100.0)	1,053	(100.0)

[a] Pearson chi-squared.
[b] T-test for independent samples.

The distributions of sex and age of participants from the two groups are presented in Table 1. Patients were older ($p<0.001$, t-test) and more often women ($p<0.001$, chi-squared (χ^2) test) as compared with individuals from the population sample. Non-participants were also compared with participants within both groups. Participants were older in the patient sample and more often women in the population sample (p=0.006 and $p<0.001$, data not shown).

Questionnaire

The questionnaire included questions on 11 categories of common airborne chemicals, which may elicit symptoms, followed by questions on the character and consequences of the symptoms. The development of the questionnaire as well as internal correlations have been described elsewhere (Berg et al., 2008; Berg et al., 2009). Data from the questionnaire were double entered using SPSS Data Entry Builder (SPSS Data Entry Builder, Rel. 4.0.0. 2003. Chicago: SPSS Inc.).

Definition of variables

Respondents from the population sample were classified in four groups, referred to as severity groups 1-4, in accordance with degree of discomfort elicited by inhalation of airborne chemicals: (1) not bothered by any exposures, (2) reporting symptoms related to exposure but no impact on everyday life, (3) reporting adjustments of personal lifestyle (i.e. by changing products used for personal hygiene, products used for cleaning in the home, or choice of shopping places) due to symptoms, and (4) reporting adjustments of social life or occupational conditions (i.e. adjustments of the use of public transportation, social functions in the private sphere, gatherings in the public sphere, sick leave from work or school, permanently leaving employment or school, or inability to work) due to symptoms.