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A Genome-Wide Screen for Hyposmia Susceptibility Loci

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Abstract

Olfactory dysfunction is an important public health problem in the United States, with approximately 14 million elderly Americans having chronic olfactory impairment. We performed a genome-wide linkage scan for loci influencing susceptibility to hyposmia in the Hutterites, a founder population of European ancestry. Using interviews regarding the olfactory medical history and psychophysical smell testing, we identified 25 individuals with severe hyposmia. Elimination of subjects with confounding conditions yielded 7 hyposmics for analysis. A 52-member pedigree including all affected individuals was constructed from the larger, >1623-member pedigree, and a genome-wide screen for loci influencing the trait of hyposmia using 1123 markers was performed. The most significant evidence for linkage with hyposmia extended over a 45 cM region on chromosome 4q ($P = 0.0013$). Although this signal meets the criteria for suggestive linkage only and will require replication, these results offer the strongest data to date on the effects of genetic variation on olfactory dysfunction.

Key words: genome, hyposmia, linkage, olfaction

Introduction

Olfactory dysfunction is an important public health problem in the United States, with the reported prevalence of olfactory impairment ranging from 0.8% to 24.5% (Wysocki and Gilbert 1989; Hoffman et al. 1998; Murphy et al. 2002). This wide variation probably reflects differences in study design and in the populations examined and known discrepancies between self-reported and tested (albeit subjective depending on cooperation) measures of olfactory function (Nordin et al. 1995). In a recent study using testing, it was estimated that approximately 14 million Americans over the age of 55 years have chronic olfactory impairment (Murphy et al. 2002). Because olfactory function declines with age (Doty et al. 1984), the clinical impact of olfactory dysfunction is likely to increase as our population ages.

Olfactory disorders have been classified as conductive (peripheral transport problems), sensorineural (damage to neuroepithelium), and central impairment (central nervous system derangement). Olfactory dysfunction can arise from multiple etiologies, including certain medications, endocrine or metabolic disorders, congenital defects, industrial exposures, infections, nasal obstruction, neoplasms, neurologic or psychiatric disease, nutritional abnormalities, and pulmonary disease (Murphy et al. 2003). The most common causes of hyposmia or anosmia include prior upper respiratory tract infections, head trauma, and sinonasal disease; these etiologies account for up to two-thirds of patients seen in clinics who have complaints related to their sense of smell (Murphy et al. 2003). Additionally, in up to 18% of patients with

hyposmia, there is no clear cause (Temmel et al. 2002). Although evidence of damage to the olfactory neuroepithelium (including scar formation, neurodegeneration, and inflammation) is observed for some of the many etiologies, common pathways that cause this damage or hinder its repair have not been elucidated. Currently, our ability to diagnose hyposmia or anosmia in patients precisely remains rudimentary, leaving most diagnoses to clinical suspicion.

Treatment options for olfactory impairment of either conductive or sensorineural origin are severely limited by our current poor understanding of the pathophysiology of impaired olfaction. Although medical and surgical treatments are available in cases with conductive causes, they are limited in scope and success (Holbrook and Leopold 2003). Understanding the factors that predispose patients to olfactory loss would provide important contributions to both our basic understanding of olfaction in human beings and advances in clinical care, especially regarding genetic factors that influence susceptibility to environmental insults. Despite the large number of etiologies, the precise pathology involved in these disorders is incompletely understood. Studies of olfactory physiology in man have been limited by access to tissue, influences of environmental factors, and the complexity of the neurosensory physiology. Therefore, novel approaches to investigating olfaction in human subjects are necessary for advances in our understanding of this area.

Better to understand the genetics of olfactory dysfunction, we performed a genome-wide screen for loci that influence hyposmia in the Hutterites, a founder population of European ancestry that lives on communal farms across the western United States and Canada. This population has several important advantages for genetic mapping studies. First, the Hutterites living in South Dakota (the subjects of our studies) are derived from only 62 ancestors (maximum number of independent chromosomes = 124) who were born in the early 1700s to the early 1800s. Thus, reduced genetic heterogeneity and fewer susceptibility alleles present at a ''disease'' locus are expected.

Second, their relatively recent origin results in relatively few meioses that have occurred since the population's founding and, therefore, relatively larger chromosomal regions flanking disease genes that have remained intact than in the general population. As a result, linkage disequilibrium extends over a greater distance than in outbred populations, making the Hutterites a population ideally suited for mapping of genes that influence common diseases, by means of both association tests and linkage studies (Wright et al. 1999; Abney et al. 2000).

Third, the Hutterites practice a communal lifestyle, thus reducing confounding from environmental variables. All Hutterites are exposed to similar environments, which are measurable and consistent. For example, smoking is prohibited and rare in the population, thereby eliminating an environmental exposure that may affect olfaction. The uniform environment also minimizes the effects of nongenetic factors and should enhance the relative effects of genetics on variation in disease expression.

Fourth, prior studies in the Hutterites have shown that nearly all the common alleles that are present in the outbred European population are also present in the Hutterites at the same relative frequencies and show similar associations with common diseases (Bourgain et al. 2003; Newman et al. 2004). Thus, the loci that influence olfactory function in the Hutterites should be the same loci that influence hyposmia in the outbred populations, as our studies of other phenotypes such as asthma indicate so far (Abney et al. 2000; Donfack et al. 2000; Ober et al. 2000; Weiss et al. 2005; Kurz et al. 2006).

Lastly, the Hutterites of South Dakota have been the subjects of complex trait mapping studies for more than 10 years. As part of these studies, about 750 individuals have been genotyped at more than 1000 loci, and association-based mapping statistics have been developed that harness the wealth of information present in this large, extended pedigree (Abney et al. 2000, 2002; Bourgain et al. 2003). This data set has already been used for mapping of genes for asthma and atopy (Ober et al. 1999; Kurz et al. 2006), triglycerides (Newman et al. 2003), morning serum cortisol (Kurina et al. 2005), whole-blood serotonin levels (Weiss et al. 2004), stuttering (Wittke-Thompson et al. 2006), and more than 15 additional quantitative traits (Weiss et al. 2006).

Here, we present the results of a genome-wide screen for loci influencing the trait of hyposmia in this population.

Materials and methods

Subject recruitment

In March 2001, we conducted a population-based study of Hutterites who were the subject of prior studies of the genetics of complex traits in our laboratory (Ober et al. 1999, 2000, 2001; Weiss et al. 2006). We used the following inclusion criteria: age above 13 years, ability to comply with the questionnaire and smell test, and presence in the colony (communal farm) on the day of the study visit. The Institutional Review Board of The University of Chicago approved this study, and informed consent was obtained.

Evaluation of phenotype

Subjects answered the 12-item Cross-Cultural Smell Identification Test (CC-SIT, now renamed B-SIT: Sensonics, Inc.; Haddon Heights, NJ), a psychophysical test that measures olfactory function. This test, modified from the University of Pennsylvania Smell Identification Test (UPSIT), has been validated in cross-cultural populations (Liu et al. 1995; Doty et al. 1996). We selected this measure because of its ease of administration to large numbers of subjects, its extensive use and validation in other studies, and its ability to provide a score with age and sex norms. The technique for administration of the CC-SIT is described in detail elsewhere (Doty et al.

1996) but involves 12 items of forced preference smell identification in a scratch-and-sniff format. Smell tests were scored with use of age and sex norms (Doty et al. 1996).

Because there are no instruments specifically designed for this purpose, we developed a survey containing closed- and open-ended questions regarding sinonasal and olfactory symptoms and medical history. This questionnaire was modified to meet Hutterite cultural norms and was administered through personal interview overseen by J.M.P. at the South Dakota Hutterite colonies. The questionnaires asked about perception of smell, the morbidities for olfactory dysfunction (current or recent [within the last 3 weeks] common colds, history of nasal trauma, history of rhinosinusitis/polyps/allergies, nasal/paranasal sinus surgery), and nasal symptoms (congestion, postnasal drip, facial pressure, facial pain, headache, and nasal obstruction). Subjects did not know the result of the smell test before answering the smell perception question. Presence of atopy was determined by skin prick testing performed during prior visits (Ober et al. 2000).

The diagnosis of hyposmia was based on the following criteria: 1) a CC-SIT score demonstrating loss of smell according to normative values (≤5th percentile) and 2) absence of confounding medical conditions. Setting our threshold for hyposmia low (≤5th percentile), the phenotype of hyposmia represents a generalized hyposmia. Because of the inherent limitations of the test (12 items, no ability to test threshold) and of phenotyping in the field, and the large number of possible odorants, we cannot accurately further delineate the phenotype into hyposmia to a subset of odorants. It was not possible to repeat testing of the individuals involved. However, the test–rest reliability of the CC-SIT is reported to be relatively high (0.71), and this 12-item test was found to predict accurately the performance on the 40-item UPSIT (Doty et al. 1995).

Genotyping

DNA was extracted from whole blood by standard methods. Genotyping of 658 autosomal microsatellite markers (screening sets 9 and 51) was performed at the Mammalian Genotyping Service in Marshfield, WI (http://research.marshfieldclinic.org/genetics/home/index.asp) by means of a scanning fluorescence detector system developed for high-throughput genotyping. An additional 226 microsatellites and 239 single-nucleotide polymorphisms (SNPs) or insertion/deletions were gentoyped in our laboratory in these individuals in selected regions according to standard methods. The 1123 microsatellites and SNP markers have an average spacing of 2.6 cM. The deCODE genetic map was used for reference in this analysis (www.decode.com) (Kong et al. 2002).

Statistical analysis

Agreement between the perception of smell and the result of the CC-SIT was analyzed by means of kappa statistics. Logistic regression was used for analysis of the associations between the comorbidities and olfactory dysfunction. Such comorbidities included allergic rhinitis, upper respiratory tract infection (cold), and a history of trauma to the nose/paranasal sinuses.

Because the pedigree that included all the hyposmics was too large for programs using algorithms of exact methods, we performed a multipoint nonparametric linkage (NPL) analysis by using Simwalk 2.83 (Sobel and Lange 1996). This program uses a Markov Chain Monte Carlo approach to sample possible allele descent trees according to their likelihood. Two identity by descent (IBD) statistics that measure the degree of allele sharing among affected individuals were calculated and *P* values determined by comparison to a null distribution of these statistics when only the pedigree structure and the affection status of each person was available. NPL_{pairs} is the extent of allele sharing among all affected pairs, and NPL_{all} is the extent of allele sharing among all affected individuals. Both statistics are intended to detect linkage to traits in an additive model.

Results

Demographics

There were 297 subjects who participated in the study. Data were available for 285 subjects for the smell test and for 291 subjects for the sinonasal/olfactory questionnaire. Of the 285 subjects, 114 were male and 171 were female, with ages ranging from 14 to 74 years (mean, 33.4 years). Relevant comorbidities, including the common cold, allergic rhinitis, or a history of trauma to the nose or nasal/paranasal sinus surgery, are shown in Table 1.

The overall prevalence of an abnormal CC-SIT was 8.8% (95% confidence interval [CI]: 5.8–12.7%). This is lower than the 24.5% prevalence reported in a community-based study (Murphy et al. 2002) and may reflect population differences, risk exposures such as pollution or tobacco smoke that are absent in this population, or the severity of our cutoff (≤5th percentile).

The demographic and clinical features of the 25 people with abnormal CC-SIT are shown in Tables 2 and 3. In an unadjusted analysis, female sex (odds ratio [OR] = 1.8), a recent or current cold (OR = 2.88), and past nasal trauma (OR = 1.16) were associated with hyposmia (Table 4). However, in a multivariate analysis, only female sex (adjusted OR = 3.07, 95% CI = 1.01–9.39, $P = 0.05$) and the presence of a common cold (adjusted OR = 2.99, 95% CI = 1.14–7.87, $P = 0.03$) remained significantly associated with olfactory dysfunction in the Hutterites (Table 4). Although a history of trauma to nose and paranasal sinuses was more common in the hyposmics (adjusted OR = 2.24), this was not statistically significant (95% CI = 0.55–9.07, $P = 0.26$). Surprisingly, age was not associated with olfactory dysfunction in this population, and there was a higher percentage of

Table 1 Characteristics and comorbidities of 285 Hutterites

	N (%)	% measured olfactory dysfunction (95% CI)
Sex		
Male	114 (40)	6 (2.5–12.2)
Female	171 (60)	10.5 (6.4–16.1)
Age		
<50 years old	247 (86.7)	9.3 (6.2–13.6)
≥50 years old	38 (13.3)	5.3 (1.4–17)
Allergic rhinitis	88 (30.9)	7.9 (3.3–15.7)
Current or recent common cold	120 (42.1)	13.3 (7.8–20.7)
Nasal trauma, including surgery	31 (10.9)	9.7 (3–25.8)

N, number of subjects in each category. Includes all subjects with both smell test and survey data.

Table 2 Results of olfactory testing by age group

Age (years)	Number evaluated	Number with hyposmia	% with hyposmia (95% CI)
10–14	3	0	0
15–19	49	6	12.3 (5.7–24.2)
20–24	42	8	19 (10.0–33.3)
25–29	30	3	10 (3.5–25.6)
30–34	38	2	5.3 (1.4–17.3)
35–39	44	2	4.5 (1.3–15.1)
40–44	20	1	5 (0.89–23.6)
45–49	19	1	5.3 (0.93–24.6)
50–54	14	1	7.1 (1.3–31.5)
55–59	6	1	16.7 (3.0–56.3)
60–64	8	0	0
65–69	11	0	0
70–74	1	0	0
Total	285	25	8.8 (5.8–12.7)

Includes all subjects with smell test data.

subjects in the younger (less than 50 years) age group with hyposmia. The correlation of hyposmia by CC-SIT and reported normal or abnormal smell perception was 0.08 (95% CI = −0.04–0.2). This is comparable to the results of other studies showing discrepancies between self- reported and tested loss of smell and emphasizes the need for testing (Doty et al. 1986; Nordin et al. 1995; Temmel et al. 2002).

Selection of cases of hyposmia

To minimize the heterogeneity of the hyposmic phenotype, we focused our studies on subjects who were identified by CC-SIT and we excluded those with comorbidities. Therefore, 120 individuals with a current or recent cold (within the last 3 weeks) as well as 31 individuals with a history of nasal or paranasal surgery or facial trauma were excluded. Subjects with atopy were included in the study because atopy was not associated with hyposmia in this sample. This resulted in a final sample of 7 Hutterites with hyposmia in the absence of these related factors. Review of questionnaires from these subjects did not reveal any other relevant clinical information; specifically, these individuals had no acute or chronic medical conditions, they did not take any medication, and they gave no other medical history. For the genetic studies, we then broke all inbreeding loops in the pedigree and constructed a 52-member pedigree by using PedHunter (Agarwala et al. 1998), which was the smallest pedigree that included the 7 affected individuals (Figure 1).

Table 3 Characteristics and comorbidities of 25 subjects with hyposmia

Age (years)	Sex	Perception of hyposmia	Current or recent cold	Atopy	Nasal trauma/ surgery
15	F	–	–	+	–
16	F	–	+	+	–
17	M	–	+	+	–
18	F	–	+	–	–
19	F	–	–	+	–
19	F	–	–	–	–
21	M	+	+	–	–
21	F	–	+	+	–
21	F	–	–	–	–
22	M	+	+	NA	–
22	F	–	+	–	–
23	F	–	–	–	+
23	F	–	+	+	–
24	F	–	+	–	–
27	F	–	+	NA	–
28	F	+	+	–	–
29	F	–	–	–	–
30	M	–	+	–	–
34	F	–	+	–	–
37	M	NA	NA	NA	NA
40	F	–	–	–	–
42	M	–	+	–	+
48	M	–	+	–	–
53	F	+	+	+	+
59	F	–	–	NA	–

M, male; F, female; NA, data not available; +, presence; –, absence.

Table 4 Association between CC-SIT and comorbidities

	CC-SIT		Crude OR	Adjusted OR	95% CI	*P* value (adjusted OR)
	Abnormal	Normal				
Sex						
Female	18	153	1.8	3.07	1.01–9.39	0.05
Male	7	107				
Age						
<50 years	23	224	0.33	0.34	0.04–2.7	0.31
≥50 years	2	36				
Allergic rhinitis						
Positive	7	81	0.91	0.90	0.34–2.41	0.84
Negative	14	147				
Current/recent cold						
Presence	16	104	2.88	2.99	1.14–7.87	0.03
Absence	8	150				
History of trauma						
Positive	3	28	1.16	2.24	0.55–9.07	0.26
Negative	21	227				

Includes all subjects with both survey and smell data.

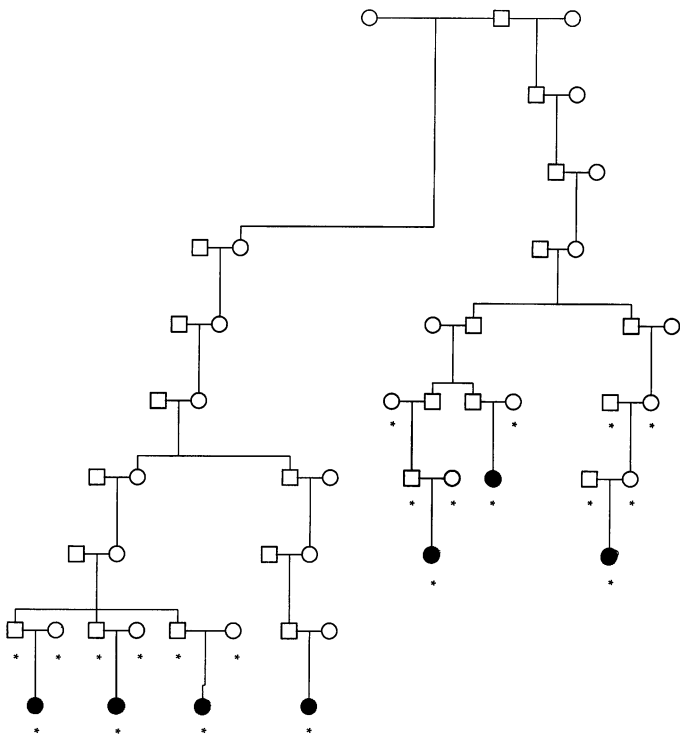

Figure 1 Pedigree of hyposmic subjects and their ancestors. *Individuals who were genotyped.

Genome screen for hyposmia

NPL analysis is a powerful tool for genetic mapping (Kruglyak et al. 1996). This technique is independent of specific models for the inheritance of the phenotype and is based only on IBD measurements at the marker loci. IBD refers to the probabilities that affected individuals share marker alleles identical by descent, that is, inherited from a common ancestor. If a marker is linked to a disease locus, one expects to see a clustering, among the affected individuals, of a few marker alleles descended from the pedigree founders.

The *P* values for the NPL_{all} statistic across the 22 autosomes are plotted in Figure 2. The NPL_{pair} results were virtually indistinguishable from the NPL_{all} results and therefore are not shown. The strongest evidence for linkage to hyposmia in the pedigree was observed on chromosome 4q with marker D4S3333 at 137.4 cM from the p terminus (p ter) ($P = 0.0013$). Additional regions showing nominal evidence for linkage include chromosome 12 ($P = 0.047$ at 52.54 cM from p ter and $P = 0.037$ at 110.24 cM from p ter), chromosome 16 ($P = 0.046$ at 114.00 cM from p ter), and chromosome 17 ($P = 0.048$ at 21.40 cM from p ter and $P = 0.044$ at 56.48 cM from p ter).

The strongest linkage signal (D4S3333) meets the criteria for suggestive linkage ($P < 1.7 \times 10^{-3}$) following Lander and Kruglyak (1995). Whereas it is not computationally feasible to conduct the simulations required for determining if our

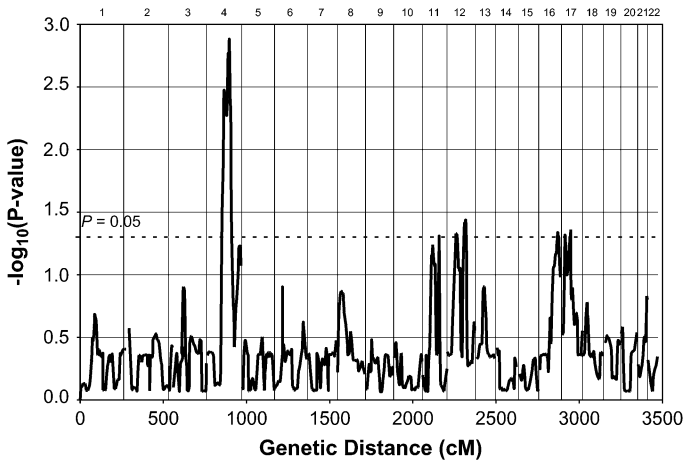

Figure 2 Genome-wide linkage scan for susceptibility to hyposmia. Position in cM according to the deCODE genetic map is shown on the *x* axis and –log10 (*P* value) of the NPL_{all} statistic on the *y* axis. Chromosome number is indicated along the top.

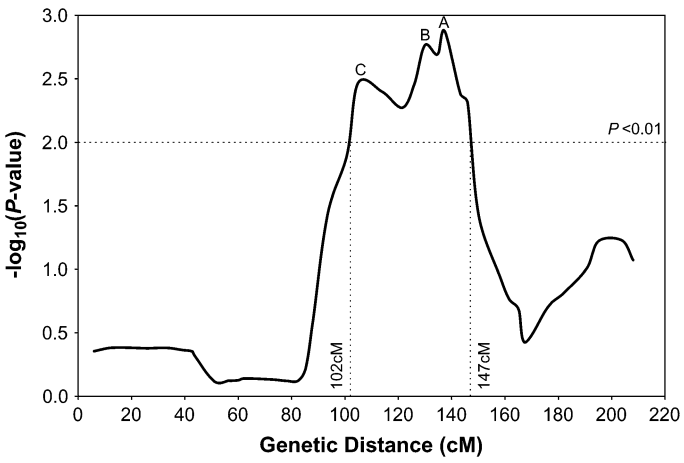

Figure 3 Linkage for susceptibility to hyposmia for chromosome 4. The highest linkage peak, A ($P = 0.0013$), occurs at 137.4 cM (D4S3333). The critical region, significant at the $P < 0.01$ threshold, is indicated by vertical dashed lines at 102 and 147 cM. Two additional linkage peaks occur within this region: B ($P = 0.0034$) at 104.9 cM (D4S1647) and C ($P = 0.0017$) at 129.2 cM (D4S2394). Axes are labeled as in Figure 2.

results meet genome-wide significance, it is important to note that estimates of the empirical *P* value using Simwalk2 were found to be conservative of the true *P* value when compared with exact results for large simulated pedigrees where the exact results were known and for pedigrees small enough so that exact *P* values could be calculated (Sobel and Lange 1996). Therefore, we are likely to be underestimating the true significance of our findings in these analyses. We are much less confident of the robustness of the weaker linkage signals, which marginally meet nominal significance at best. These could easily represent false positive results. Hence, we focused subsequent investigations on our most significant linkage peak only.

To define a critical region for further investigation, we set a $P < 0.01$ significance threshold boundary of 102–147 cM (Figure 3), corresponding to a 53-Mb physical distance mapping to the ~95–148 Mb position on chromosome 4 (4q22.3–4q31.22) (NCBI Build 36.2, September 2006). Within this critical region, 2 subpeaks are evident in addition to the most significant peak at 137.4 cM; they correspond to D4S1647 ($P = 0.0034$ at 104.9 cM; peak C in Figure 3) and D4S2394 ($P = 0.0017$ at 129.2 cM; peak B in Figure 3). This 52-Mb critical region contains 288 genes (183 known RefSeq genes, 105 hypothetical genes), none of which are olfactory receptor genes.

Discussion

Demographics of olfactory dysfunction in the Hutterites

The prevalence of olfactory dysfunction among the Hutterites, as defined by the performance on the CC-SIT, was 8.8% (95% CI = 5.8–12.7%). This prevalence was higher than in some previous community-based studies (Wysocki and Gilbert 1989; Hoffman et al. 1998) but lower than others (Murphy et al. 2002) that may reflect differences in study design (e.g., tested vs. self-reported olfactory determination, self vs. monitored administration). The agreement between the awareness of olfactory loss and the CC-SIT results was quite poor in this study. That people usually are unaware of their smell loss despite having olfactory dysfunction implies a lack of perception, minimal loss of smell, or misinterpretation by the subject of the test. Likewise, people believing their olfactory sense was abnormal had, in fact, normal olfaction by CC-SIT, implying that the test is not capturing the perceived abnormality. Therefore, self-reported smell function may provide misleading information, and olfactory testing is necessary for assessment of olfactory function. Doty et al. (1988) found that, in the case of Parkinson's disease, approximately 90% of patients had a demonstrable olfactory deficit, even though only 28% of such patients were aware of their deficit. Studies in other clinical settings noted similar observations (Doty et al. 1986; Nordin et al. 1995; Temmel et al. 2002), including the use of threshold testing (Philpott et al. 2006). Murphy et al. (2002) also showed that self-report in older adults significantly underestimated the prevalence rates obtained by olfactory testing.

The common cold and female sex were found to be 2 factors contributing to olfactory dysfunction in the Hutterites, with the adjusted ORs of 2.99 for presence of the common cold (95% CI = 1.14–7.87) and 3.07 for female sex (95% CI = 1.01–9.39), respectively. Age above 50 years, allergic rhinitis, and a history of nasal/facial trauma including nasal/paranasal sinus surgery, after adjusting for other effects, did not show any statistically significant effect on olfactory dysfunction in this study. Although there was a trend toward increased olfactory dysfunction with a history of trauma to the nose/paranasal sinuses (adjusted OR = 2.24 with a wide 95% CI of 0.55–9.07), this was not statistically significant, perhaps as a result of the lack of severe head trauma in this

rural population (there were no motor vehicle accidents or assaults).

The common cold has been postulated to be one of the leading causes of olfactory dysfunction (Temmel et al. 2002). This could occur as a result of the conduction problems caused by nasal congestion or inflammatory effects on the olfactory pathway (Cowart et al. 1993). Rydzewski et al. (2000) demonstrated that the prevalence of hyposmia in allergic rhinitis was 21.4%, higher than the prevalence in our study of 7.9%, and may reflect the stringency of the definition of hyposmia in our study. Simola and Malmberg (1998) compared olfactory thresholds in 105 rhinitis patients and 104 healthy controls to analyze possible relationships between the sense of smell and rhinitis, age, sex, smoking, prick-test results, nasal resistance, and history of nasal or paranasal surgery. They found that age and rhinitis were the only variables with a significant effect on the olfactory threshold. Neither smoking habits nor a history of nasal or paranasal surgery were related to olfactory thresholds in that study. The differences between our results and those of other studies might be due to the differences in population (urban vs. rural), study design (hospital based vs. population based), different measurement of olfactory function (threshold vs. identification vs. subjective report), or the different status of allergic rhinitis at the time of test administration (all testing in our study was done in the winter months, outside of certain allergen seasons). In particular, the lifelong absence of exposure to environmental tobacco smoke (which is known to impair olfactory function [Frye et al. 1990; Murphy et al. 2002]) in the Hutterites may contribute both to the overall lower prevalence of hyposmia and the lack of an association with age.

Genetics of olfaction

A genetic perspective to understanding olfactory dysfunction should provide insight into chemosensory physiology. Studies employing a genetic approach to olfaction in man are limited, although animal studies suggest a genetic basis for a variety of related phenotypes (reviewed in Segal et al. 1995). Limited data from human studies suggests that genetic factors may affect the sense of smell. For example, idiopathic hypogonadotrophic hypogonadism with anosmia, or Kallmann's syndrome (KS), is a rare genetic disorder with variable expressivity (Kallmann et al. 1944). Family members have a variety of phenotypes, including anosmia alone. This condition is genetically heterogeneous, with both X-linked, autosomal dominant, and autosomal recessive transmission reported (Kallmann et al. 1944; Hockaday 1966; White et al. 1983). Mutations in the KS1 locus (*KAL1*) on chromosome Xp22.3 have been identified in the X-linked form (Franco et al. 1991; Legouis et al. 1991; Bick et al. 1992; Hardelin et al. 1992), and mutations in the fibroblast growth factor receptor 1 locus (*KAL2*) on chromosome 8p11.2–12 cause one autosomal dominant form of the disease (Dode et al. 2003). A recent study also reported linkage of KS to chromosome19p13, with loss-of-function mutations in the *GPR54* locus causing KS (de Roux et al. 2003).

Studies of nonsyndromic forms of anosmia or hyposmia also support a genetic basis of olfactory function. Forrai et al. (1981) examined the genetic influence on the ability to smell ketones by using a twin pair study design and observed genetic effects on the detection of acetone. Wysocki and Beauchamp (1984) studied sensitivity to androstenone and pyridine by using twin pairs, showing that all the monozygotic twin pairs, but only 61% of dizygotic pairs, were concordant for sensitivity to androstenone. A genetic influence on androstenone detection was confirmed by Gross-Isserhoff et al. (1992) who also used a twin pair design and showing a genetic effect on sensitivity to isoamyl nitrate. Using the UPSIT, Doty et al. (1984) and Segal et al. (1995) found a higher correlation of scores between monozygotic twins ($r = 0.31$) than between dizygotic twins ($r = 0.15$). Using a different measure of preference ratings of UPSIT odors, Topolski reported a genetic influence on certain items (Segal et al. 1995). Also, in a twin pair study, Finkel et al. (2001) found moderate heritability for several olfactory measures, including odor identification, intensity, detection, and pleasantness.

Knaapila et al. (2007) recently reported the first genome-wide screen of olfactory phenotypes. They did not find any significant results for linkage with the phenotype of identification by CC-SIT, although there were methodologic differences in their analytic strategy (phenotyping all subjects without exclusions; quantitative mapping based on total score on the B-SIT vs. qualitative analysis based on selecting severe hyposmia by age/sex norms in our study) and marker density (350 markers vs. 1123 in our study). Additionally, this population sample of 146 individuals from 26 families was recruited through a study of migraine in Finland. Population, diet, allele frequency, or environmental differences, inclusion of smokers, and/or the presence of migraine may explain the discrepancy between their results and ours. Interestingly, they did show a significant linkage with the phenotype of pleasantness of cinnamon odor (logarithm of the odds = 3.01) on chromosome 4q33.3 at 163.65 cM, just outside our region.

Recently, subjects with Bardet–Biedl syndrome (BBS), which encompasses retinal degeneration, truncal obesity, renal and limb malformations, and developmental delay, have been shown to have partial or complete anosmia (Kulaga et al. 2004). To date, 12 BBS loci have been identified (reviewed in Li and Wong 2001; Mykytyn et al. 2001; Nishimura et al. 2001; Sheffield et al. 2001; Mykytyn et al. 2002; Ansley et al. 2003; Badano et al. 2003; Chiang et al. 2004; Fan et al. 2004; Nishimura et al. 2005; Chiang et al. 2006; Stoetzel et al. 2006, 2007). In fact, the loci for 2 members of this novel group of genes, *BBS7* (Badano et al. 2003) and *BBS12* (Stoetzel et al. 2007), lie within our most significant region on chromosome 4. This finding may indicate that more subtle perturbations in BBS7 and/or BBS12 function may underlie olfaction in nonsyndromic

olfactory dysfunction and make these loci excellent candidates for association with hyposmia.

Although the large number of candidate genes precludes extensive discussion here, other promising candidate genes include *SCYE1* (Shalak et al. 2007), a cytokine generated in apotosis, and *CASP6*, a caspase that is activated in human neurons upon an apoptotic insult. Interestingly, apoptosis has been implicated in olfactory pathology (Kern et al. 2004). Additionally, another candidate in the region, *UNC5C*, a receptor involved in both neural migration and development, has also been reported to be involved in apoptosis (Thiebault et al. 2003).

Our results further support a genetic etiology for hyposmia, and they suggest that genetic variation influences the olfactory system. We selected our phenotype and threshold in order carefully to reduce the issue of confounding environmental and host factors as much as possible to focus on idiopathic olfactory decline. Nevertheless, it is possible that our affected individuals had an environmental exposure influencing their susceptibility to olfactory loss. As viral illness is one of the most common reported causes of the loss of the sense of smell, it is possible that postviral damage is the etiology of the olfactory loss in our affected individuals. In olfactory clinics, some patients note the loss of smell to be temporally associated with an especially severe viral upper respiratory tract infection (URI), but some do not notice the insult. Moreover, injury from a URI that is subtotal or predisposing to further loss by apoptosis or other mechanisms without patient perception is entirely possible. In this case, the phenotype being mapped in our study might be an example of a gene–environment interaction where genetically susceptible individuals who experience the appropriate viral insult are predisposed to loss of smell. Nevertheless, mapping of genes that underlie such a phenomenon would provide information on the mechanisms involved in this process. Hence, our data would still provide useful information on susceptibility to olfactory loss, although perhaps not on nonvirally mediated decline.

Another possibility is that the affected individuals share a common diagnosis of isolated, nonsyndromal congenital hyposmia. We believe that it is unlikely that our subjects had congenital hyposmia, given the lack of a supportive medical history and the absence of behaviors (inability to detect body odor, complaints of flavorless food, inability to notice farm odors, spoiled milk, etc.) that would be associated with this disease. Although a number of our affected subjects were in their teens, we believe that our demographics reflects the age structure of our population, which has a large number of younger individuals because of large family size.

Lastly, isolated congenital hyposmia is extremely rare and has been mapped in one small family study to a different region of the genome (chromosome 18p) (Ghadami et al. 2004). We believe that a number of factors influence the complex trait of hyposmia, both environmental and genetic, and teasing out this interaction remains a challenge. Although the injuries might be heterogeneous, there may be a common genetic susceptibility to a variety of insults that lead to a common outcome of olfactory decline. Our studies will eventually require replication in other population samples for analysis of these issues.

The ability of the nervous system to distinguish among many odors and the variability in olfactory function among individuals are under intense scrutiny. To understand how genetic factors may explain the variability in human olfaction between individuals, Menashe et al. (2002, 2003) examined the prevalence of segregating pseudogenes in the olfactory receptor gene family and proposed that the combination of these polymorphisms could underlie variability in olfaction between individuals and populations. Indeed, Keller et al. (2007) recently reported evidence that supports this concept, finding that variation in an olfactory receptor gene, *OR7D4*, affects odorant perception in human subjects and is associated with differential binding to its odorant in vitro. Additionally, 2 of the common deletion polymorphisms in the human genome are in olfactory receptor genes (McCarroll et al. 2006). These are intriguing ideas that attempt to explain how olfactory function varies among individuals. However, although specific olfactory receptor genes have been implicated in specific anosmia in mouse models (Griff and Reed 1995; Zhang and Firestein 2002), our phenotype of generalized hyposmia is perhaps less likely to be caused by variation in olfactory receptor genes. Indeed, there are no olfactory receptor genes in the linked region on chromosome 4q, indicating that variations in nonolfactory receptor genes also influence smell perception.

This study was the first step in identifying novel genes that contribute to variation in olfaction in man. Subsequent studies to identify the specific gene within our linked region are currently under way.

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References

Abney M, McPeek MS, Ober C. 2000. Estimation of variance components of quantitative traits in inbred populations. Am J Hum Genet. 66(2):629–650.

Abney M, Ober C, McPeek MS. 2002. Quantitative-trait homozygosity and association mapping and empirical genomewide significance in large, complex pedigrees: fasting serum-insulin level in the Hutterites. Am J Hum Genet. 70(4):920–934.

Agarwala R, Biesecker LG, Hopkins KA, Francomano CA, Schaffer AA. 1998. Software for constructing and verifying pedigrees within large genealogies and an application to the Old Order Amish of Lancaster County. Genome Res. 8(3):211–221.

Ansley SJ, Badano JL, Blacque OE, Hill J, Hoskins BE, Leitch CC, Kim JC, Ross AJ, Eichers ER, Teslovich TM, et al. 2003. Basal body dysfunction is a likely cause of pleiotropic Bardet-Biedl syndrome. Nature. 425(6958):628–633.

Badano JL, Ansley SJ, Leitch CC, Lewis RA, Lupski JR, Katsanis N. 2003. Identification of a novel Bardet-Biedl syndrome protein, BBS7, that shares structural features with BBS1 and BBS2. Am J Hum Genet. 72(3):650–658.

Bick D, Franco B, Sherins RJ, Heye B, Pike L, Crawford J, Maddalena A, Incerti B, Pragliola A, Meitinger T, et al. 1992. Brief report: intragenic deletion of the KALIG-1 gene in Kallmann's syndrome. N Engl J Med. 326(26):1752–1755.

Bourgain C, Hoffjan S, Nicolae R, Newman D, Steiner L, Walker K, Reynolds R, Ober C, McPeek MS. 2003. Novel case-control test in a founder population identifies P-selectin as an atopy-susceptibility locus. Am J Hum Genet. 73(3):612–626.

Chiang AP, Beck JS, Yen HJ, Tayeh MK, Scheetz TE, Swiderski RE, Nishimura DY, Braun TA, Kim KY, Huang J, et al. 2006. Homozygosity mapping with SNP arrays identifies TRIM32, an E3 ubiquitin ligase, as a Bardet-Biedl syndrome gene (BBS11). Proc Natl Acad Sci USA. 103(16):6287–6292.

Chiang AP, Nishimura D, Searby C, Elbedour K, Carmi R, Ferguson AL, Secrist J, Braun T, Casavant T, Stone EM, et al. 2004. Comparative genomic analysis identifies an ADP-ribosylation factor-like gene as the cause of Bardet-Biedl syndrome (BBS3). Am J Hum Genet. 75(3):475–484.

Cowart BJ, Flynn-Rodden K, McGeady SJ, Lowry LD. 1993. Hyposmia in allergic rhinitis. J Allergy Clin Immunol. 91(3):747–751.

de Roux N, Genin E, Carel JC, Matsuda F, Chaussain JL, Milgrom E. 2003. Hypogonadotropic hypogonadism due to loss of function of the KiSS1-derived peptide receptor GPR54. Proc Natl Acad Sci USA. 100(19): 10972–10976.

Dodé C, Levilliers J, Dupont JM, De Paepe A, Le Dû N, Soussi-Yanicostas N, Coimbra RS, Delmaghani S, Compain-Nouaille S, Baverel F, et al. 2003. Loss-of-function mutations in FGFR1 cause autosomal dominant Kallmann syndrome. Nat Genet. 33(4):463–465.

Donfack J, Tsalenko A, Hoki DM, Parry R, Solway J, Lester LA, Ober C. 2000. HLA-DRB1*01 alleles are associated with sensitization to cockroach allergens. J Allergy Clin Immunol. 105(5):960–966.

Doty RL, Deems DA, Frye RE, Pelberg R, Shapiro A. 1988. Olfactory dysfunction in parkinsonism: a general deficit unrelated to neurologic signs, disease stage, or disease duration. Neurology. 38(8):1237–1244.

Doty RL, Gregor T, Monroe C. 1986. Quantitative assessment of olfactory function in an industrial setting. J Occup Med. 28(6):457–460.

Doty RL, Marcus A, Lee WW. 1996. Development of the 12-item Cross-Cultural Smell Identification Test (CC-SIT). Laryngoscope. 106(3 Pt 1): 353–356.

Doty RL, McKeown DA, Lee WW, Shaman P. 1995. A study of the test-retest reliability of ten olfactory tests. Chem Senses. 20(6):645–656.

Doty RL, Shaman P, Applebaum SL, Giberson R, Siksorski L, Rosenberg L. 1984. Smell identification ability: changes with age. Science. 226(4681): 1441–1443.

Fan Y, Rahman P, Peddle L, Hefferton D, Gladney N, Moore SJ, Green JS, Parfrey PS, Davidson WS. 2004. Bardet-Biedl syndrome 1 genotype and obesity in the Newfoundland population. Int J Obes Relat Metab Disord. 28(5):680–684.

Finkel D, Pedersen NL, Larsson M. 2001. Olfactory functioning and cognitive abilities: a twin study. J Gerontol B Psychol Sci Soc Sci. 56(4): P226–P333.

Forrai G, Bánkövi G, Szabados T, Papp ES. 1981. Ketone compound smelling ability: study in Hungarian twins. Acta Med Acad Sci Hung. 38(2): 153–158.

Franco B, Guioli S, Pragliola A, Incerti B, Bardoni B, Tonlorenzi R, Carrozzo R, Maestrini E, Pieretti M, Taillon-Miller P, et al. 1991. A gene deleted in Kallmann's syndrome shares homology with neural cell adhesion and axonal path-finding molecules. Nature. 353(6344): 529–536.

Frye RE, Schwartz BS, Doty RL. 1990. Dose-related effects of cigarette smoking on olfactory function. JAMA. 263(9):1233–1236.

Ghadami M, Morovvati S, Majidzadeh-AK, Damavandi E, Nishimura G, Kinoshita A, Pasalar P, Komatsu K, Najafi MT, Niikawa N, et al. 2004. Isolated congenital anosmia locus maps to 18p11.23-q12.2. J Med Genet. 41(4):299–303.

Griff IC, Reed RR. 1995. The genetic basis for specific anosmia to isovaleric acid in the mouse. Cell. 83(3):407–414.

Gross-Isseroff R, Ophir D, Bartana A, Voet H, Lancet D. 1992. Evidence for genetic determination in human twins of olfactory thresholds for a standard odorant. Neurosci Lett. 141(1):115–118.

Hardelin JP, Levilliers J, del Castillo I, Cohen-Salmon M, Legouis R, Blanchard S, Compain S, Bouloux P, Kirk J, Moraine C. et al. 1992. X chromosome-linked Kallmann syndrome: stop mutations validate the candidate gene. Proc Natl Acad Sci USA. 89(17):8190–8194.

Hockaday TD. 1966. Hypogonadism and life-long anosmia. Postgrad Med J. 42(491):572–574.

Hoffman HJ, Ishii EK, MacTurk RH. 1998. Age-related changes in the prevalence of smell/taste problems among the United States adult population. Results of the 1994 disability supplement to the National Health Interview Survey (NHIS). Ann N Y Acad Sci. 855:716–722.

Holbrook EH, Leopold DA. 2003. Anosmia: diagnosis and management. Curr Opin Otolaryngol Head Neck Surg. 11(1):54–60.

Kallmann FJ, Schönfeld WA, Barrera SE. 1944. The genetic aspects of primary eunuchoidism. Am J Ment Defic. 48:203–236.

Keller A, Zhuang H, Chi Q, Vosshall LB, Matsunami H. 2007. Genetic variation in a human odorant receptor alters odour perception. Nature. 449(7161):468–472.

Kern RC, Conley DB, Haines GK 3rd, Robinson AM. 2004. Pathology of the olfactory mucosa: implications for the treatment of olfactory dysfunction. Laryngoscope. 114(2):279–285.

Knaapila A, Keskitalo K, Kallela M, Wessman M, Sammalisto S, Hiekkalinna T, Palotie A, Peltonen L, Tuorila H, Perola M. 2007. Genetic component of identification, intensity and pleasantness of odours: a Finnish family study. Eur J Hum Genet. 15(5):596–602.

Kong A, Gudbjartsson DF, Sainz J, Jonsdottir GM, Gudjonsson SA, Richardsson B, Sigurdardottir S, Barnard J, Hallbeck B, Masson G, et al. 2002. A high-resolution recombination map of the human genome. Nat Genet. 31(3):241–247.

Kruglyak L, Daly MJ, Reeve-Daly MP, Lander ES. 1996. Parametric and nonparametric linkage analysis: a unified multipoint approach. Am J Hum Genet. 58(6):1347–1363.

Kulaga HM, Leitch CC, Eichers ER, Badano JL, Lesemann A, Hoskins BE, Lupski JR, Beales PL, Reed RR, Katsanis N. 2004. Loss of BBS proteins causes anosmia in humans and defects in olfactory cilia structure and function in the mouse. Nat Genet. 36(9):994–998.

Kurina LM, Weiss LA, Graves SW, Parry R, Williams GH, Abney M, Ober C. 2005. Sex differences in the genetic basis of morning serum cortisol levels: genome-wide screen identifies two novel loci specific to women. J Clin Endocrinol Metab. 90(8):4747–4752.

Kurz T, Hoffjan S, Hayes MG, Schneider D, Nicolae R, Heinzmann A, Jerkic SP, Parry R, Cox NJ, Deichmann KA, et al. 2006. Fine mapping and positional candidate studies on chromosome 5p13 identify multiple asthma susceptibility loci. J Allergy Clin Immunol. 118(2):396–402.

Lander E, Kruglyak L. 1995. Genetic dissection of complex traits: guidelines for interpreting and reporting linkage results. Nat Genet. 11(3):241–247.

Legouis R, Hardelin JP, Levilliers J, Claverie JM, Compain S, Wunderle V, Millasseau P, Le Paslier D, Cohen D, Caterina D, et al. 1991. The candidate gene for the X-linked Kallmann syndrome encodes a protein related to adhesion molecules. Cell. 67(2):423–435.

Li C, Wong WH. 2001. Model-based analysis of oligonucleotide arrays: expression index computation and outlier detection. Proc Natl Acad Sci USA. 98(1):31–36.

Liu HC, Wang SJ, Lin KP, Lin KN, Fuh JL, Teng EL. 1995. Performance on a smell screening test (the MODSIT): a study of 510 predominantly illiterate Chinese subjects. Physiol Behav. 58(6):1251–1255.

McCarroll SA, Hadnott TN, Perry GH, Sabeti PC, Zody MC, Barrett JC, Dallaire S, Gabriel SB, Lee C, Daly MJ, et al. 2006. Common deletion polymorphisms in the human genome. Nat Genet. 38(1):86–92.

Menashe I, Man O, Lancet D, Gilad Y. 2002. Population differences in haplotype structure within a human olfactory receptor gene cluster. Hum Mol Genet. 11(12):1381–1390.

Menashe I, Man O, Lancet D, Gilad Y. 2003. Different noses for different people. Nat Genet. 34(2):143–144.

Murphy C, Doty R, Duncan, HJ. 2003. Clinical disorders of olfaction. In: Doty R, editor. Handbook of olfaction and gustation. New York: Marcel Dekker.

Murphy C, Schubert CR, Cruickshanks KJ, Klein BE, Klein R, Nondahl DM. 2002. Prevalence of olfactory impairment in older adults. JAMA. 288(18):2307–2312.

Mykytyn K, Braun T, Carmi R, Haider NB, Searby CC, Shastri M, Beck G, Wright AF, Iannaccone A, Elbedour K, et al. 2001. Identification of the gene that, when mutated, causes the human obesity syndrome BBS4. Nat Genet. 28(2):188–191.

Mykytyn K, Nishimura DY, Searby CC, Shastri M, Yen HJ, Beck JS, Braun T, Streb LM, Cornier AS, Cox GF, et al. 2002. Identification of the gene (BBS1) most commonly involved in Bardet-Biedl syndrome, a complex human obesity syndrome. Nat Genet. 31(4):435–438.

Newman DL, Abney M, Dytch H, Parry R, McPeek MS, Ober C. 2003. Major loci influencing serum triglyceride levels on 2q14 and 9p21 localized by homozygosity-by-descent mapping in a large Hutterite pedigree. Hum Mol Genet. 12(2):137–144.

Newman DL, Hoffjan S, Bourgain C, Abney M, Nicolae RI, Profits ET, Grow MA, Walker K, Steiner L, Parry R, et al. 2004. Are common disease susceptibility alleles the same in outbred and founder populations? Eur J Hum Genet. 12(7):584–590.

Nishimura DY, Searby CC, Carmi R, Elbedour K, Van Maldergem L, Fulton AB, Lam BL, Powell BR, Swiderski RE, Bugge KE, et al. 2001. Positional cloning of a novel gene on chromosome 16q causing Bardet-Biedl syndrome (BBS2). Hum Mol Genet. 10(8):865–874.

Nishimura DY, Swiderski RE, Searby CC, Berg EM, Ferguson AL, Hennekam R, Merin S, Weleber RG, Biesecker LG, Stone EM et al. 2005. Comparative genomics and gene expression analysis identifies BBS9, a new Bardet-Biedl syndrome gene. Am J Hum Genet. 77(6):1021–1033.

Nordin S, Monsch AU, Murphy C. 1995. Unawareness of smell loss in normal aging and Alzheimer's disease: discrepancy between self-reported and diagnosed smell sensitivity. J Gerontol B Psychol Sci Soc Sci. 50(4):P187–P192.

Ober C, Abney M, McPeek MS. 2001. The genetic dissection of complex traits in a founder population. Am J Hum Genet. 69(5):1068–1079.

Ober C, Tsalenko A, Willadsen S, Newman D, Daniel R, Wu X, Andal J, Hoki D, Schneider D, True K. 1999. Genome-wide screen for atopy susceptibility alleles in the Hutterites. Clin Exp Allergy. 29(Suppl 4):11–15.

Ober C, Tsalenko A, Parry R, Cox NJ. 2000. A second-generation genomewide screen for asthma-susceptibility alleles in a founder population. Am J Hum Genet. 67(5):1154–1162.

Philpott CM, Wolstenholme CR, Goodenough PC, Clark A, Murty GE. 2006. Comparison of subjective perception with objective measurement of olfaction. Otolaryngol Head Neck Surg. 134(3):488–490.

Rydzewski B, Pruszewicz A, Sulkowski WJ. 2000. Assessment of smell and taste in patients with allergic rhinitis. Acta Otolaryngol. 120(2):323–326.

Segal NL, Topolski TD, Wilson SM, Brown KW, Araki L. 1995. Twin analysis of odor identification and perception. Physiol Behav. 57(3):605–609.

Shalak V, Guigou L, Kaminska M, Wautier MP, Wautier JL, Mirande M. 2007. Characterization of p43(ARF), a derivative of the p43 component of multi-aminoacyl-tRNA synthetase complex released during apoptosis. J Biol Chem. 282(15):10935–10943.

Sheffield VC, Nishimura D, Stone EM. 2001. The molecular genetics of Bardet-Biedl syndrome. Curr Opin Genet Dev. 11(3):317–321.

Simola M, Malmberg H. 1998. Sense of smell in allergic and nonallergic rhinitis. Allergy. 53(2):190–194.

Sobel E, Lange K. 1996. Descent graphs in pedigree analysis: applications to haplotyping, location scores, and marker-sharing statistics. Am J Hum Genet. 58(6):1323–1337.

Stoetzel C, Laurier V, Davis EE, Muller J, Rix S, Badano JL, Leitch CC, Salem N, Chouery E, Corbani S, et al. 2006. BBS10 encodes a vertebrate-specific chaperonin-like protein and is a major BBS locus. Nat Genet. 38(5):521–524.

Stoetzel C, Muller J, Laurier V, Davis EE, Zaghloul NA, Vicaire S, Jacquelin C, Plewniak F, Leitch CC, Sarda P, et al. 2007. Identification of a novel BBS gene (BBS12) highlights the major role of a vertebrate-specific branch of chaperonin-related proteins in Bardet-Biedl syndrome. Am J Hum Genet. 80(1):1–11.

Temmel AF, Quint C, Schickinger-Fischer B, Klimek L, Stoller E, Hummel T. 2002. Characteristics of olfactory disorders in relation to major causes of olfactory loss. Arch Otolaryngol Head Neck Surg. 128(6):635–641.

Thiebault K, Mazelin L, Pays L, Llambi F, Joly MO, Scoazec JY, Saurin JC, Romeo G, Mehlen P. 2003. The netrin-1 receptors UNC5H are putative tumor suppressors controlling cell death commitment. Proc Natl Acad Sci USA. 100(7):4173–4178.

Weiss LA, Lester LA, Gern JE, Wolf RL, Parry R, Lemanske RF, Solway J, Ober C. 2005. Variation in ITGB3 is associated with asthma and sensitization to mold allergen in four populations. Am J Respir Crit Care Med. 172(1):67–73.

Weiss LA, Pan L, Abney M, Ober C. 2006. The sex-specific genetic architecture of quantitative traits in humans. Nat Genet. 38(2):218–222.

Weiss LA, Veenstra-Vanderweele J, Newman DL, Kim SJ, Dytch H, McPeek MS, Cheng S, Ober C, Cook EH Jr, Abney M. 2004. Genome-wide association study identifies ITGB3 as a QTL for whole blood serotonin. Eur J Hum Genet. 12(11):949–954.

White BJ, Rogol AD, Brown KS, Lieblich JM, Rosen SW. 1983. The syndrome of anosmia with hypogonadotropic hypogonadism: a genetic study of 18 new families and a review. Am J Med Genet. 15(3):417–435.

Wittke-Thompson JK, Ambrose N, Yairi E, Roe C, Cook EH, Ober C, Cox NJ. 2006. Genetic studies of stuttering in a founder population. J Fluency Disord. 32(1):33–50.

Wright AF, Carothers AD, Pirastu M. 1999. Population choice in mapping genes for complex diseases. Nat Genet. 23(4):397–404.

Wysocki CJ, Beauchamp GK. 1984. Ability to smell androstenone is genetically determined. Proc Natl Acad Sci USA. 81(15):4899–4902.

Wysocki CJ, Gilbert AN. 1989. National Geographic smell survey. Effects of age are heterogenous. Ann N Y Acad Sci. 561:12–28.

Zhang X, Firestein S. 2002. The olfactory receptor gene superfamily of the mouse. Nat Neurosci. 5(2):124–133.

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