Evolutionary Grass Roots for Odor Recognition

Tsviya Olender1 and Doron Lancet1

1Department of Molecular Genetics, The Weizmann Institute of Science, Rehovot 76100, Israel

Correspondence to be sent to: Doron Lancet, Department of Molecular Genetics, The Weizmann Institute of Science, Rehovot 76100, Israel. e-mail: doron.lancet@weizmann.ac.il

Accepted April 15, 2012

Abstract

Considerable evidence supports the idea that odorant recognition depends on specific sequence variations in olfactory receptor (OR) proteins. Much of this emerges from in vitro screens in heterogeneous expression systems. However, the ultimate proof should arise from measurements of odorant thresholds in human individuals harboring different OR genetic variants, a research vein that has so far been only scantily explored. The study of McRae et al., published in this issue of Chemical Senses, shows how the recognition of a grassy odorant depends on specific OR interindividual sequence changes. It provides a clear relevant example for the impact of genetics on olfaction and is an excellent portrayal of the power of human genomics to decipher olfactory perception.

Key Words: genotype, phenotype, haplotype, genetics, SNP, olfactory receptor

Commentary

Different human olfactory receptor (OR) paralogs (members of the receptor repertoire of our species) are widely believed to underlie the capacity to recognize a multitude of odorous chemicals. However, there is another dimension of OR variability that contributes importantly to our understanding of human chemosensory perception: interindividual differences. The pioneering work of John Amoore (1967) has clearly demonstrated that people’s noses are widely different in threshold phenotypes for specific odorants and put forth the conjecture that this phenomenon is attributable to widely different personal arsenals of OR proteins. Further, a natural assumption, supported by evidence (Whissell-Buechy and Amoore 1973; Amoore 1974; Wysocki and Beauchamp 1984; Gross-Isseroff et al. 1992), was that olfactory threshold differences stem from genetics. Based on the huge body of biological data accumulated in the ensuing 60 years, this view is translated to the statement that different humans have different polymorphic versions of OR genes (Young and Trask 2002; Menashe et al. 2003; Hasin-Brumshtein et al. 2009).

In a species vista, each gene locus may harbor an appreciable number of genetic variants (alleles), coding for different protein sequences. An individual receives 2 of these: one from mother and the other from father. Thus, a person’s OR repertoire is composed of pairs of alleles at each of the ~400 gene loci in the active olfactory subgenome. In theory, the allele count could reach ~800, that is, if every locus were heterozygously disposed. In fact, analyses of hundreds of human genomic sequences show that the total number of alleles per person ranges between 500 and 600 (Olender et al., unpublished data). Such a large repository of “personal receptors” is impressive and has profound implications to the variegation human olfaction. Importantly, because ORs are expressed in an allelically excluded mode (only one allele in every sensory cell; Chess et al. 1994), the brain receives a faithful sensory view of such personalized gamut of OR alleles.

An obvious premise is that different allelic protein variants would bind odorants with different affinity, but only one study had thus far provided direct evidence for this. Five years ago, Keller et al. (2007) have shown that the human perception of the steroid odorant androstenone is widely different for 2 allelic variants at the OR7D4 locus. Individuals homozygously carrying a variant with arginine at position 88 and threonine at position 113 (the RT allele) are adequately sensitive to androstenone, whereas individuals with both gene copies encoding tryptophan at position 88 and threonine at position 113 (the WM allele) are much less sensitive to the odorant and found it less unpleasant.

Androstenone is an unusual odorant as pertaining to the human odorant universe. It is a boar pheromone (Claus and Alsing 1976), with allusion of behavior-modifying effects in humans (Araneda and Firestein 2004). This may suggest a highly specialized mammalian receptor, potentially with atypical properties. The article of McRae et al. addresses an odorant much more frequently encountered in human general chemoreception. This is the compound cis-3-hexen-1-ol.
(C3HEX), an unsaturated short-chain alcohol, described as “green grassy,” found in a wide range of foods and beverages and constituting a key flavor of many fruits and vegetables (Jirovetz et al. 2002; Genovese et al. 2004). In an earlier study (Jaegera et al. 2010), a group of authors partly overlapping with the present one reported on a significant genetic association between the ability to detect the C3HEX and a genomic cluster of 25 OR gene on chromosome 6 (clusters 6@29; see Olender et al. (2004)). Such a “low-resolution” genetic signal is typically obtained in genotype–phenotype correlations as exemplified by some other association reports, for example, between isovaleric acid smelling and the receptor OR11H7P exemplified by some other association reports, for example, between isovaleric acid smelling and the receptor OR11H7P between the ability to detect the C3HEX and a genomic association (Jaegera et al. 2010), also clarifies why the genetic factors found were not of the nonsense type, such as stop–gain and insertion–deletion (indels) (Menashe et al. 2003), or deletion copy number variations (Waszak et al. 2010). Instead, all were missense mutations (amino acid replacements), and except R88W of OR7D4 have had Polyphen (Adzhubei et al. 2010) prediction of “benign”, that is, not highly deleterious. Such results suggest caution in passing a-priori judgment on the phenotypic outcome of OR genetic changes. Prudence along similar lines should be exerted when looking at the protein sequence position of observed OR sequence modifications. Among the 4 variations seen (Figure 1), 2 are in intracellular loops (positions 133 and 226), in the broadly defined region of interaction with the olfactory G-protein alpha subunit (Crasto 2009; Kato and Touhara 2009) but not in an obviously conserved or motif-containing region. The other 2 mutations also seem relatively unobstructive: R88W in OR7D4 is in the 1st extracellular loop, a region not hitherto implicated in receptor function, and T113A in OR2J3 is in the upper part of transmembrane helix 3 near to but not overlapping with a predicted odorant binding pocket (Man et al. 2004). Much more insight will become possible when more examples are reported and once a resolved 3-dimensional structure of an OR protein becomes available, as for other G protein–coupled receptors (Mustafi and Palczewski 2009).

The OR2J3 haplotypes reveal an intriguing evolutionary narrative (Table 1). The reference haplotype repetition time

![Figure 1](https://academic.oup.com/chemse/article-abstract/37/7/581/300903)
Table 1  Evolutionary portrayal of OR positions 113 and 226 (p113 and p226), shown to affect C3HEX binding to OR2J3, as appearing in other OR homologs both in human and in other organisms documented in the HORDE database (http://genome.weizmann.ac.il/horde/).

<table>
<thead>
<tr>
<th>OR gene</th>
<th>Species</th>
<th>Ivt react</th>
<th>%id</th>
<th>p113</th>
<th>p226</th>
<th>Relation</th>
</tr>
</thead>
<tbody>
<tr>
<td>OR2J3 HAP1*</td>
<td>human</td>
<td>278</td>
<td>T</td>
<td>R</td>
<td>var</td>
<td></td>
</tr>
<tr>
<td>OR2J3 HAP3</td>
<td>human</td>
<td>156</td>
<td>99.5</td>
<td>T</td>
<td>Q</td>
<td>var</td>
</tr>
<tr>
<td>OR2J3 HAP6</td>
<td>human</td>
<td>100</td>
<td>99.5</td>
<td>A</td>
<td>R</td>
<td>var</td>
</tr>
<tr>
<td>OR2J3 HAP2</td>
<td>human</td>
<td>0</td>
<td>99</td>
<td>A</td>
<td>Q</td>
<td>var</td>
</tr>
<tr>
<td>OR2J4P</td>
<td>human</td>
<td>89</td>
<td>T</td>
<td>Q</td>
<td></td>
<td>paralog</td>
</tr>
<tr>
<td>OR2J2</td>
<td>human</td>
<td>88</td>
<td>A</td>
<td>R</td>
<td></td>
<td>paralog</td>
</tr>
<tr>
<td>OR2J1P</td>
<td>human</td>
<td>89</td>
<td>A</td>
<td>R</td>
<td></td>
<td>paralog</td>
</tr>
<tr>
<td>C696.71-2984</td>
<td>chimp</td>
<td>98</td>
<td>T</td>
<td>R</td>
<td></td>
<td>ortholog</td>
</tr>
<tr>
<td>C696.65-251</td>
<td>chimp</td>
<td>95</td>
<td>T</td>
<td>R</td>
<td></td>
<td>ortholog</td>
</tr>
<tr>
<td>C696.63-9755</td>
<td>chimp</td>
<td>89</td>
<td>T</td>
<td>Q</td>
<td></td>
<td>homolog</td>
</tr>
<tr>
<td>C696.74-3848</td>
<td>chimp</td>
<td>90</td>
<td>A</td>
<td>R</td>
<td></td>
<td>homolog</td>
</tr>
<tr>
<td>NM_001000266</td>
<td>rat</td>
<td>83</td>
<td>T</td>
<td>W</td>
<td></td>
<td>ortholog</td>
</tr>
<tr>
<td>MOR256-18</td>
<td>mouse</td>
<td>82</td>
<td>T</td>
<td>W</td>
<td></td>
<td>ortholog</td>
</tr>
<tr>
<td>cOR2B2</td>
<td>dog</td>
<td>62</td>
<td>T</td>
<td>Q</td>
<td></td>
<td>homolog</td>
</tr>
<tr>
<td>Modo-OR28B8P</td>
<td>opossum</td>
<td>61</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>aa-OR2G22</td>
<td>platypus</td>
<td>60</td>
<td>T</td>
<td>R</td>
<td></td>
<td>homolog</td>
</tr>
</tbody>
</table>

%id is the percent amino acid sequence identity. The top 4 sequences represent the different human variants (var) whose analyses are reported by McRae et al., along with the nomenclature used by them for the haplotypes (HAPI) and their reported in vitro activity toward C3HEX (Ivt react). The human reference genome variant is marked with “*”. Sequences marked as “paralogs” are the 3 closest OR2J3 homologs in human. Sequences marked as “orthologs” are OR2J3 homologs in other species showing mutual best homology relationship. Sequences marked as “homologs” are the closest OR2J3 homologs in the other species, but without mutual best homology. For chimpanzee identifiers, “C” represents contig.

(TR) is also present in 2 very near chimpanzee orthologs, suggesting short-term conservation of the “smeller” version. However, in the presumed rodent orthologs, a completely different amino acid combination appears (TW), casting some doubt on the assumption that orthologous ORs have similar odorant binding underpinnings (Man et al. 2004). On the other hand, none of the species examined have the “null” haplotype AQ. Curiously, both human and chimpan have other OR repertoire members (paralogs) with the intermediate haplotypic combinations TQ and AR, and the nearest OR2J3 dog sequence also has a TQ version. Finally, in the evolutionarily remote platypus, the most similar sequence carries the human reference combination TR. This type of evolutionary “lego” may further attest to the functional significance of these 2 sequence positions.

The human OR repertoire is currently defined mostly based on the reference human genome (Olender et al. 2004). The present study highlights the importance of establishing a complete compendium human OR alleles, expected soon in the HORDE database (Olender et al. 2004), together with extensive screening and psychophysics. This will likely generate a framework for additional genotype–phenotype association studies and prepare the arena for comprehensive future assessment of functional OR key residues both computationally (Crasto 2009) and experimentally. Such progress would hopefully lead to a “pharmacological” mapping of the entire OR repertoire, providing invaluable tools for the fragrance and flavor industries.

Funding
The writing of this commentary was funded by the Crown Human Genome Center at the Weizmann Institute of Science.

References


