Primer

Human olfactory psychophysics

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Of all the senses, smell is the least understood. Despite centuries of investigation, science can still offer no satisfying theory for why a particular substance smells the way it does. Nor do we understand in any detail how we are able to distinguish the smell of a peach from that of an apricot, or how a particular smell can trigger long-forgotten memories of a distant time or place. Human olfactory psychophysics, the study of how humans perceive odors, is possible because humans have acquired language. Human subjects can report directly if something smells, characterize the smell, or decide if two smells are distinguishable. Answers to these simple questions have the potential to provide insight into important questions: What (if any) is the relationship between the chemical structure of an odor and its perceived smell? What types of olfactory stimuli can be discriminated, and how is this accomplished in the nose and the brain? How does experience modulate our perception of odorants? There are of course many things that cannot be done in humans, for instance genetic manipulation and electrophysiology, but these types of approaches are successfully used in animal models.

The molecular biology of smell

The olfactory system of humans consists of several million olfactory sensory neurons arrayed in a sensory epithelium located inside the nasal cavity. Each of these sensory neurons expresses one of approximately 350 odorant receptor genes, which confers upon that neuron a specific sensitivity to the set of odor molecules that will bind and activate the respective odorant receptor. It is widely believed that only a small region of the odor molecule is recognized by a given odorant receptor. Therefore, unlike hearing or seeing, olfaction is not a spectral sense, but rather consists of a large number of sensors with different specificities and affinities. Any given odor may activate only a single receptor or many different receptors. On the other hand an odorant receptor can be very specific and only be activated by very few odor molecules or be more promiscuous and recognize a variety of odor molecules. We are far from a complete understanding of which odors activate which odorant receptors; however, the available data support the notion that the combinatorial activation of olfactory neurons has the potential to account for the extremely large number of different odors that can be detected. How the activation of populations of olfactory sensory neurons is translated in the brain into a discretely perceived odor quality is still completely mysterious, despite vigorous investigation in model systems as disparate as nematodes, fruit flies, fish, mice, and humans. No clear models have emerged to account for the various psychophysical observations surrounding smell and we do not yet know how different odors are represented in higher brain regions. Olfactory perception depends on peripheral detection and central cortical processing. Very little is known about the mechanisms that guide central olfactory processing and shape the odor percept, which remains one of the most important problems in the field. The fact that odor perception is highly influenced by memory, experience, and input from other sensory modalities makes the problem even more fascinating.

The importance of smell for humans

Humans are commonly thought to have an impoverished sense of smell relative to our rodent and canine cousins. Comparative genomic analysis supports this idea. While humans have approximately 1000 odorant receptor genes, the majority have been mutated into non-coding pseudogenes. This leaves humans with about 350 functional odorant receptor genes, many fewer than the approximately 1000 functional odorant receptor genes in the mouse. Interestingly, there is an enormous diversity in the repertoire of functional odorant receptor genes among different people. Given the marked differences in olfactory preference between people, it is tempting to suggest that some of these differences can be traced directly to genetic diversity in the repertoire of odorant receptors.

Despite the very large number of pseudogenes among human olfactory receptor genes, there is no compelling psychophysical evidence that humans have a substantially worse sense of smell than monkeys, rats, or even dogs. Of course, there are procedural problems with comparing behavioral results collected from humans with those from species that cannot speak, because animals need to be trained to perform olfactory discrimination tasks, whereas humans are merely asked to report differences between odors. Nevertheless, it remains to be seen what specific advantages in odor detection the substantially larger repertoire of odorant receptor genes might confer to mice over humans. Smell is certainly economically important to our species, with sales of scented products constituting an annual market of over $25 billion dollars in the United States alone.

Measuring odor quantity and quality

Olfactory psychophysics relies on simple tools (Figure 1): informed and consenting human subjects, odor stimuli, and a set of questions formulated to obtain clear and reliable answers from the subjects. Psychophysical studies have investigated the effects of age, pregnancy, neurodegenerative diseases, and environmental exposure on the sense of smell, as well as basic questions of odor detection and perception.

Psychophysics can be used to measure detection and identification thresholds, the amount of stimulus needed to

Figure 1. The tools of the psychophysical trade: an informed, consenting human subject and an odor source. Odorants are supplied by a variety of means: squeeze bottles, 'rip-and-sniff' packets, glass vials (as depicted here), or sophisticated computer-controlled olfactometers that deliver odors in a stream of warm and humidified air directly into the nose. Photo by Zach Veilleux, Rockefeller University Communications and Public Affairs.

detect and identify an odor, respectively. The detection threshold is the lowest concentration at which a stimulus can be perceived and is usually measured by comparing the odor intensity of the odor at a very low concentration with the odor intensity of the solvent. The identification threshold is the lowest concentration at which an odor can be identified and is usually considerably higher than the detection threshold. One effective, simple and widely used method is the 'single staircase detection threshold' method, which involves of a number of pairwise presentations of odors (Figure 2). The detection thresholds of different odorants have been shown to vary widely, from 0.00001 to 500,000 parts per billion. Even more interestingly, the detection thresholds vary between people. A lowered sensitivity to one but not all odorants is called 'specific anosmia'. Specific anosmias to musk odors are extremely common and have a genetic basis, although the specific recessive gene defect has not been mapped. In humans and mice, specific anosmias to the rancid smell of isovaleric acid have been documented, and in mice have been mapped to a chromosomal region that contains a number of odorant receptor genes. The extent to which these specific anosmias are a direct cause of polymorphisms or mutations in odorant receptor genes is an interesting avenue for future research.

While determining the odor threshold is relatively straightforward, the problem of assigning a description or quality to a given odor is extremely difficult. Untrained subjects will have no problem reporting that they smell something, but may be incapable of describing the smell using words. Such difficulties are less frequent in psychophysical studies that probe the auditory or visual systems. Despite these inherent difficulties, many experimental designs have been employed in an attempt to determine perceived odor quality. Odors can be profiled, a semantic method in which descriptors from a list are assigned to an odor (Table 1). Odor profiling involves comparing the test odor to mentally stored odor templates, with the list of 146 descriptors serving to jog the memory. The subject has to recall a 'fishy' or 'fruity' odor to assign these descriptors to an odorant. Such profiling has been performed by a large cohort of subjects and when averaged, consensus odor qualities can be extracted. Although it is in widespread use and generally effective, this method has some obvious flaws. Ratings from many subjects must be averaged, which of necessity obscures potentially interesting inter-individual differences in perception. The semantic descriptors, first published by Dravnieks in 1978, are vulnerable to becoming dated as they age and may be incomprehensible to subjects from different cultural backgrounds or non-native English speakers. For instance, while descriptors such as 'sweet' and 'coffee' are likely to remain relevant for the foreseeable future, many contemporary subjects may be baffled by 'kippery' and 'anise, licorice' unless they have direct experience with these smells.

A more direct, non-semantic approach is to compare the odor under investigation to reference odors and use the perceived similarity between odorants to describe the odor. For instance, if an odor is more similar to a floral odor reference than to a musky one, it would be grouped among the floral odors. A serious conceptual problem with this approach is the question of what constitutes a reasonable reference odor. At best, one ends up with a list of floral or woody or musky odors, but the uniqueness of a given odor may be lost for lack of words to describe it. The multidimensionality of the odor identification problem using references would be analogous to asking subjects to group fine art paintings by similarity. Is the Degas more similar to the Vermeer or the Matisse? Is an eight-carbon alcohol more similar to a seven-carbon alcohol or an eight-carbon aldehyde? In both scenarios results can be highly idiosyncratic, arbitrary, and influenced by prior experience. Nevertheless, in the absence of better methods, similarity can be inferred from measures of stimulus discriminability, similarity rating, or grouping of odorants according to similarity. Stimulus discriminability can be measured with a variety of forced choice experiments that, for instance, require subjects to pick the odd odor from a group of three choices. If two odors are reproducibly grouped as being the same, the subjects are scored as not discriminating the odors. All similarity assessments of this type are complicated by the fact that they may measure differences in odor intensity rather than odor quality. Furthermore, odor quality depends on odor concentration: two odorants that smell similar at low concentration may smell different at high concentration.

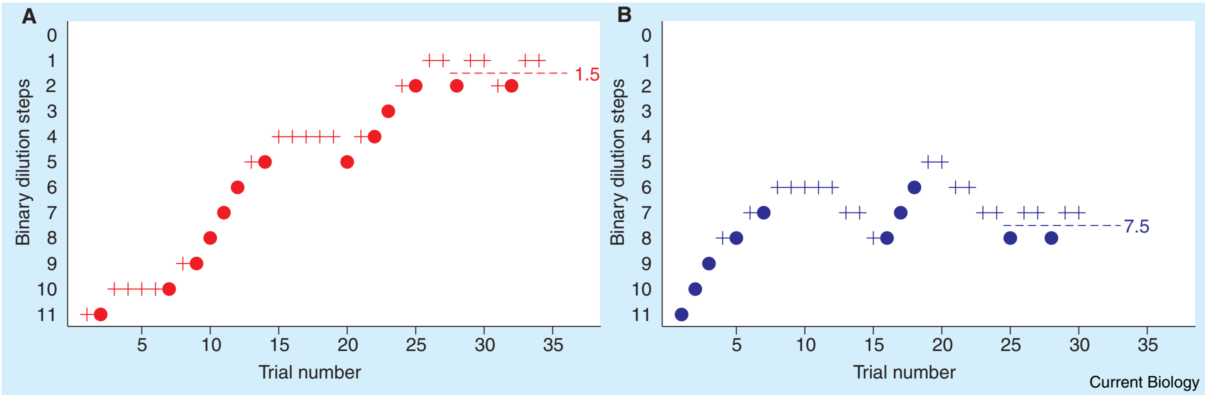

Figure 2. Odor threshold detection using the single-staircase method.
Data for two different human subjects are shown in (A) and (B) for the musk odor, pentadecalactone. Subjects are presented with two vials, one containing a very low concentration of odor and the other containing a solvent control, and are asked to sniff two vials and to identify the one with the stronger odor. A correct answer is scored as '+', while the incorrect answer is indicated by a dot. Odor concentration is increased until five answers are correct at a given concentration, after which the concentration is decreased when two answers are correct and increased when at least one of the two answers is wrong. The threshold value is calculated as the mean of the last four of seven staircase reversals.

The perception of odor mixtures
Naturally occurring smells are virtually always a complex mixture of different odor molecules. The characteristic smell of a rose for example consists of about 260 components. In some sensory modalities, the magnitude of mixed stimuli is perceived as the sum of the magnitudes of the individual stimuli, a characteristic known as additivity. No such rule has been found in olfactory psychophysics. The estimated intensity of the smell of the mixture of two odorants is frequently perceived as being non-additive. This phenomenon is called counteracting. There are three types of counteracting: partial addition, in which the mixture smells more intense than the stronger component; compromise, in which the smell intensity of the mixture is in between the intensities of the components; and compensation, in which the mixture smells less intense than the weaker component. Considerable effort has been expended to understand the mechanisms that underlie these modes of counteracting. Recent *in vitro* experiments that examined odorant receptors expressed in tissue culture cells suggest a model that could account for all of these effects. Antagonists that act on specific odorant receptors have been found and these are thought to block the binding site of odorant receptors without activating the olfactory sensory neuron. Whether complete addition, partial addition, compromise, or compensation occurs in a binary mixture would, therefore, depend on the extent of the antagonistic relationship between the two odorants as they interact with the repertoire of odorant receptors.

Olfactory psychophysical experiments can address whether the sense of smell is analytic or synthetic. An analytic sense of smell would be capable of perceiving the single odorants in a mixture, whereas with a synthetic sense of smell the components of a mixture would form a new odor and the components would not be perceived. Binary odor mixtures produce two different psychophysical results. Mixtures of dissimilar odorants ('poor blenders') in general are not perceived as a blend, but are perceived analytically, whereas binary mixtures of 'good blenders' (similar odors that presumably activate overlapping sets of odorant receptors) may be perceived synthetically as a new odor. It is generally agreed that some odorants lose major characteristic qualities in binary mixtures. Many principles that characterize the perceived quality of the smell of a binary mixture have emerged, but no model has been successful in predicting the odor quality of binary mixtures.

The perceived smell of an odorant at a given concentration changes over time and depends on prior experience. This phenomenon is called adaptation and is caused by repeated or prolonged exposure to an odorant, typically leading to elevated thresholds and reduced responsiveness to suprathreshold stimulation. Adaptation can produce non-reciprocal interactions between odors in mixtures. Sour-smelling propionic acid has little effect on the perception of aromatic-smelling carvone in binary mixtures, although it affects other odorants when mixed with them. Carvone, on the other hand, does influence the perception of propionic acid in the mixture, a finding that can be explained with non-reciprocal cross-adaptation.

Quality and quantity of more complex mixtures have not yet been studied as extensively. A remarkable finding with mixtures of more than two odorants is that subjects are only able to identify three or, rarely, four components of a complex mixture. Neither training nor experience increases the number of identifiable components. Mixtures of complex odors, which are themselves mixtures, tend to behave like mixtures of single odors. The number of components in a mixture is invariably underestimated, and the odor of mixtures is not perceived as more complex than the odor of single chemicals.

Table 1. Semantic odor quality descriptors.

Materials	Chemicals	Outdoors	Fruits	Foods	Spices
dry, powdery	sharp, pungent, acid	hay	cherry, berry	buttery, fresh	almond
chalky	sour, acid, vinegar	grainy	strawberry	caramel	cinnamon
cork	ammonia	herbal, cut grass	peach	chocolate	vanilla
cardboard	camphor	crushed weed	pear	molasses	anise, licorice
wet paper	gasoline, solvent	crushed grass	pineapple	honey	clove
wet wool, wet dog	alcohol	woody, resinous	grapefruit	peanut butter	maple syrup
rubbery, new	kerosene	bark, birch	grape juice	soupy	dill
tar	household gas	musty, earthy, moldy	apple	beer	caraway
leather	chemical	cedarwood	cantaloupe	cheesy	minty, peppermint
rope	turpentine, pine oil	oakwood, cognac	orange	eggs, fresh	nut, walnut
metallic	varnish	rose	lemon	raisins	eucalyptus
burnt, smoky	paint	geranium leaves	banana	popcorn	malt
burnt paper	sulphidic	violets	coconut	fried chicken	yeast
burnt candle	soapy	lavender	fruity, citrus	bakery, fresh bread	black pepper
burnt rubber	medicinal	laurel leaves	fruity, other	coffee	tea leaves
burnt milk	disinfectant, carbolic				spicy
creosote	ether, anaesthetic				
sooty	cleaning fluid, carbona				
fresh tobacco smoke	mothballs				
stale tobacco smoke	nail polish remover				

Foul	Common	Common	Meats	Vegetables	Body
fermented, rotten fruit	sweet	sweaty	meat seasoning	fresh vegetables	dirty linen
sickening	fragrant	cool, cooling	animal	garlic, onion	sour milk
rancid	perfumery	light	fish	mushroom	sewer
putrid, foul, decayed	floral	heavy	kippery, smoked fish	raw cucumber	fecal, manure
dead animal	cologne	warm	blood, raw meat	raw potato	urine
mouse-like	aromatic		meat, cooked good	bean	cat urine
	musky		oily, fatty	green pepper	seminal, like sperm
	incense		sauerkraut		
	bitter		celery		
	stale		cooked vegetables		

Semantic odor quality rating chart after Dravnieks. Subjects are presented with an odor and asked to rate the suitability of 146 odor descriptors on a scale from 0 (no resemblance to odor) to 5 (extremely good description of odor). The odor profile of a given chemical obtained in this way has been shown to be stable when averaged across relatively large sample sizes.

Influence of the other senses

The outcome of psychophysical experiments with olfactory stimuli is highly context dependent. Visual, gustatory, perceptual, and cognitive factors strongly modulate the performance in olfactory psychophysical experiments. For example, odors that have been presented with sugar previously will be rated as sweeter and less sour. Color has been shown to affect perceived odor intensity. Most notorious is the study carried out at the University of Bordeaux in which oenology students smelled wines and assigned red wine descriptors to white wine that had been dyed red without their knowledge.

Future directions

The availability of the complete sequence of human genome provides enormous opportunities to relate olfactory phenotype to the underlying genotype of odorant receptor genes. It will be of interest to relate the specific anosmias encountered in various populations with the underlying gene defects. In all of these approaches, human olfactory psychophysics will be a key tool. Without information about the ultimate behavioral relevance of a given stimulus, it will be difficult to evaluate whether any phenomenon recorded along the pathway of perception is important for the brain to crack the odor code.

Further reading

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