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A Redefinition of Odor Mixture Quality

Leslie M. Kay, Tanja Crk, and Jennifer Thorngate
University of Chicago

Odor mixtures are perceived as different from (*configural*) or the same as (*elemental*) their components. Recent studies (L. M. Kay, C. A. Lowry, & H. A. Jacobs, 2003; C. Wiltrout, S. Dogra, & C. Linster, 2003) propose that component structural or perceptual similarities predict configural properties of binary mixtures. The authors evaluated this in rats using 4 binary mixtures with varying structural similarity (eucalyptol–benzaldehyde, eugenol–benzaldehyde, octanol–octanal, and [+/−]-limonene). The range of tested ratios for each mixture was determined by the components' vapor pressures. Three results are presented: (a) No mixture maintains purely elemental or configural properties for all concentration ratios, (b) structural similarity or dissimilarity does not predict configural or elemental perception, and (c) overshadowing is significant in responses to all odor sets. The authors offer more precise definitions of elemental and configural properties and overshadowing as they relate to odor mixture perception.

Keywords: binary mixture, olfactory, configural, elemental, overshadowing

A glass of wine contains tens, if not hundreds, of different volatile compounds. Its odor can be described with many adjectives, which refer to the presence of individual chemicals or arise synthetically from the combination of two or more compounds. The chemical and physiological properties of odor mixtures such as these and how they give rise to perceptual properties is one of the most difficult questions in olfactory research.

Each component of an odor mixture has specific physical properties, which affect its volatility, the speed with which it diffuses through air and mucous, and the manner in which it may bind to single or multiple olfactory receptors. Mixtures introduce the possibility of competitive and other types of interactions between component odors at olfactory receptor binding sites. Within the olfactory bulb (OB), neural circuits likely contribute to mixture processing in the representation of odors by mitral cells (Giraudet, Berthommier, & Chaput, 2002; Tabor, Yaksi, Weislogel, & Friedrich, 2004). Downstream areas also play a significant role through internal processing and through feedback to the OB (Kay & Freeman, 1998; Kay & Laurent, 1999; Shipley & Adamek, 1984; Wilson, 2003).

Analysis of binary mixture perception is the first step in understanding how these properties affect the perceptual qualities of odor mixtures. Several studies have suggested that binary mixtures of structurally similar molecules or odors that smell alike produce *configural* or synthetic perceptual qualities in which the mixture does not smell like the components. On the other hand, binary mixtures of very dissimilar molecules or odors that do not smell alike have been shown to produce more *elemental* perceptual qualities in which the mixture smells like the sum of the two odor components (Laing & Francis, 1989; Laing, Panhuber, & Slotnick, 1989; Linster & Cleland, 2004; Wiltrout, Dogra, & Linster, 2003). Our laboratory has recently shown that binary mixtures of monomolecular odors known to interact with olfactory receptor neurons expressing the same receptor type (rat I7 receptor) produce predictable perceptual properties in rats based on overlap at that receptor (Kay, Lowry, & Jacobs, 2003). These results together with the other studies suggest that binary mixture perception may be partly explained by receptor and OB glomerular level interactions.

In this study, we further test the hypothesis that perceptual qualities are driven by receptor biophysics and chemical similarity. We tested four odor pairs of varying levels of structural similarity: one with very dissimilar chemical structures, one with very gross structural similarity, one in which the components differ only at the functional group, and a mixture of two enantiomers. We found, contrary to expectations, that the less similar mixtures have concentration ratio ranges with configural properties and that mixtures of similar molecules can be perceived as elemental (even though the components overlap at several receptor types). In all mixtures, we found evidence of overshadowing of one component by another, depending on the concentration ratio. We argue that classification of a mixture as configural or elemental may not be informative enough to describe the full range of perceptual qualities even in binary mixtures. We therefore propose a method of multiple binary ratios to assist in odor mixture analysis and description based on these studies.

One problem with interpreting studies from different laboratories is the inconsistent use of the terms elemental and configural in cases where overshadowing of one component by another might more appropriately describe the responses. Therefore, in the *Discussion*, we address this issue and offer more precise definitions of these terms as they apply to odor mixture perception.

Leslie M. Kay, Department of Psychology and The College, Institute for Mind & Biology, The University of Chicago; Tanja Crk and Jennifer Thorngate, The College, Institute for Mind & Biology, The University of Chicago.

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Correspondence concerning this article should be addressed to Leslie M. Kay, College Institute for Mind & Biology, 940 East 57th Street, Room 331, Chicago, IL 60637. E-mail: lkay@uchicago.edu

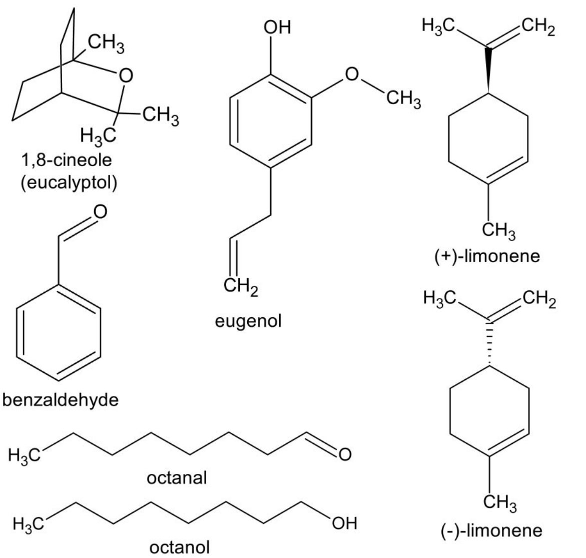

Figure 1. Odorant molecular structures. Benzaldehyde and eucalyptol have no structural similarity, and benzaldehyde and eugenol both have a benzene structure but are otherwise unrelated. Octanol and octanal differ only in the functional group and have identical hydrocarbon chains. The enantiomers of limonene differ very little and smell alike.

Method

Subjects

Twelve adult male Sprague–Dawley rats (420–480 g; obtained from Harlan Sprague Dawley, Indianapolis, IN) were housed singly and maintained on a 14:10-hr light–dark schedule (lights on at 8 a.m.). They reduced to 85% of their ad libitum weight by restricting only food. All testing was done in dim light in the mid to late afternoon. All procedures were performed with approval and oversight by the University of Chicago Institutional Animal Care and Use Committee and conformed to Association for Assessment and Accreditation of Laboratory Animal Care standards.

Odors

Odors were purchased from Sigma-Aldrich (St. Louis, MO) or Fisher Scientific (Atlanta, GA), and all were at ≥96% purity, with most at ≥98% (Fisher Scientific, 99% octanol, 99% eugenol, 98+% benzaldehyde, 98% amyl acetate; Sigma-Aldrich, 99% octanal, 99% eucalyptol, 97% (+)-limonene, 96% (−)-limonene). Chemical structures are shown in Figure 1. Mixtures tested were eucalyptol (1,8-cineole) and benzaldehyde (dissimilar molecules), eugenol and benzaldehyde (grossly similar molecules), octanol and octanal (similar molecules), and (+/−)-limonene (enantiomers). All training odors were binary mixtures diluted in liquid phase to ratios as described in the text, with concentrations above threshold for identification in rats (i.e., they could be trained to identify the individual odors in separate identification tests using the same behavioral methods as in this report). Each mixture set spans the ratio at which the binary mixture ratio is inversely proportional to the ratio of the theoretical vapor pressures of the two chemicals at 25 °C. The absolute concentrations of odors in each mixture are listed in Tables 1–4. Theoretical vapor pressures were estimated using the ACD/I-Lab Web service, ACD/Vapor Pressure 5.0 (Advanced Chemistry Development, Toronto, Canada).

Behavior

The rats were trained to dig in a glass dish (70 mm × 50 mm) of bedding material for a reward of sweet cereal (approximately one fourth of a Kellogg's Froot Loop). After training to the digging task, they were further trained to choose between two dishes, one scented with a drop of an odor diluted in mineral oil on the surface (1% methyl salicylate baited with the reward on the bottom) and the other containing only a drop of mineral oil. After training (approximately 2 weeks), the animals performed odor tests 1–3 times per week.

Each rat was trained and tested on a particular mixture ratio on a single day. After 10 training trials with the reward present (except for probe trials), we tested the mixture, each of the components, and the control odor in random order (balanced across individuals within each odor set). Probe trials were performed during the training trials without the reward to confirm that the rat learned the odor mixture and not the odor of the reward. All rats showed recognition of the training odor. During testing, the experimenters were blind to the test odors. Digging times were measured with a stopwatch and recorded at the end of each extinction trial.

Each mixture ratio was tested on a different day for each rat, with the order within a set balanced across rats. Tests with different odor sets were interleaved so that successive sessions did not use the same odor set. The eucalyptol–benzaldehyde and eugenol–benzaldehyde tests were separated by at least 4 weeks for all rats.

Analysis

Data are digging times, in seconds, normalized to *z* scores (zero mean and unit standard deviation) for each rat for each test day, as reported in earlier studies (Kay et al., 2003; Nusser, Kay, Laurent, Homanics, & Mody, 2001). We normalized in this fashion because digging times vary considerably across time and animals, and this made for easy application of post hoc comparisons. Rats that did not dig in any of the four test odors for a given data set were excluded from the analysis for that set. Normalized data were analyzed with a one-way analysis of variance for each mixture ratio followed by Newman–Keuls post hoc tests to assess pairwise differences among members of the odor set.

Table 1
Eucalyptol–Benzaldehyde Concentrations

	Liquid ratio				
Odorant	20:1 (51:2.5 Pa)	10:1 (25.6:2.5 Pa)	1:1 (2.6:2.5 Pa)	1:10 (2.6:25 Pa)	1:20 (2.6:50 Pa)
Mixture	10%:0.5%	5%:0.5%	0.5%:0.5%	0.5%:5%	0.5%:10%
Eucalyptol	10%	5%	0.5%	0.5%	0.5%
Benzaldehyde	0.5%	0.5%	0.5%	5%	10%
Amyl acetate	0.25%	0.25%	0.25%	0.25%	0.25%

Note. Pa = Pascals.

Table 2
Eugenol–Benzaldehyde Concentrations

Odorant	Liquid ratio					
	300:1 (2:1 Pa)	150:1 (1:1 Pa)	97.5:1 (0.66:1 Pa)	30:1 (0.2:1 Pa)	1:1 (0.02:2.5 Pa)	1:10 (0.02:12.5 Pa)
Mixture	30%:0.1%	30%:0.2%	19.5%:0.2%	5%:0.2%	0.5%:0.5%	0.5%:2.5%
Eugenol	30%	30%	19.5%	5.0%	0.5%	0.5%
Benzaldehyde	0.1%	0.2%	0.2%	0.2%	0.5%	2.5%
Amyl acetate	0.25%	0.25%	0.25%	0.25%	0.25%	0.25%

Note. Pa = Pascals.

Results

None of the mixtures tested shows the same perceptual results for all ratios (see Figures 2 and 3). Each graph has the mixture ratios arrayed along the horizontal axis and the normalized (*z* score) digging times on the vertical axis (absolute concentrations of individual component odors in liquid phase are in Tables 1–4, and chemical structures are in Figure 1). Each odor set spanned the ratio at which the relative concentrations are close to the inverse ratio of the theoretical vapor pressures.

The odor sets in Figure 2 were chosen to examine the perceptual qualities of mixtures of dissimilar and only grossly similar chemicals. The vapor pressures for eucalyptol and benzaldehyde were approximately 1.7 to 1 (0.22 to 0.13 kPa), so that the 1:1 ratio was the closest to the inverse ratio of the vapor pressures (IVP). For this mixture (see Figure 2A), training ratios of 1:1 (IVP) and 1:10 (eucalyptol to benzaldehyde) showed a marked configural representation, with the rats overwhelmingly identifying only the trained mixture significantly over the control odor (amyl acetate). This includes an order of magnitude increase in the absolute concentration of benzaldehyde in the mixture from the 1:1 (0.5% eucalyptol and 0.5% benzaldehyde) to the 1:10 (0.5% eucalyptol and 5% benzaldehyde) ratio. On either side of the configural range, the animals generalized only to the component of higher concentration.

Figure 2B shows the results for eugenol–benzaldehyde mixtures. There was a much larger difference in vapor pressures in this case, as the ratio for eugenol to benzaldehyde was 1 to 97.5. Again, there was a range of configural representation in which only the trained odor mixture is recognized. However, digging in response to benzaldehyde for the 25:1 test was nearly significant ($p = .08$ in a paired one-tailed t test of benzaldehyde over amyl acetate). In the 1:1 test, both eugenol and benzaldehyde responses were significant in paired one-tailed t tests over amyl acetate ($p = .03$ and .048, respectively), but this significance did not survive the post hoc comparison. As in the eucalyptol–benzaldehyde tests, on either side of this range, the rats generalized only to the component of highest concentration.

The responses to the octanol–octanal mixtures were different from the previous two sets, and the chemical structures share an identical hydrocarbon chain only differing at the functional group (see Figures 1 and 3A). The vapor pressure ratio of octanol to octanal was approximately 1 to 18 (0.0152 kPa and 0.276 kPa, respectively), so that the ratio closest to the IVP was 20:1 in the figure. At every ratio in this set, at least one component was recognized, and at the IVP ratio, the animals generalized to both components equally with the mixture in elemental fashion. It is notable that for the 50:1 octanol (10%) to octanal (0.2%) mixture, the recognition of octanal was completely absent, whereas for the 20:1 ratio, with the concentration of octanal held constant and octanol decreased to 4%, there was a dramatic change in the response to octanal. The response to octanol disappeared when the concentration was dropped from 4% to 0.5%, even though this is well within the range in which rats can identify this odor (data not presented).

Figure 3B shows the responses to the mixtures of the enantiomers (+)-limonene and (−)-limonene. To human observers, these chemicals smell alike, and in the 1:1 ratio, subjects responded to neither the mixture nor the components significantly above the control odor. At ratios above and below the 1:1 ratio, the component of highest concentration was equated with the mixture.

Table 3
Octanol–Octanal Concentrations

Odorant	Liquid ratio				
	200:1 (15:1.3 Pa)	50:1 (3.75:1.3 Pa)	20:1 (1.5:1.3 Pa)	1:1 (1.5:3.3 Pa)	1:20 (1.5:66 Pa)
Mixture	40%:0.2%	10%:0.2%	4%:0.2%	0.5%:0.5%	0.5%:10%
Octanol	40%	10%	4.0%	0.5%	0.5%
Octanal	0.20%	0.20%	0.20%	0.5%	10%
Amyl acetate	0.25%	0.25%	0.25%	0.25%	0.25%

Note. Pa = Pascals.

Table 4
(+)/(−)Limonene Concentrations

	Liquid ratio				
Odorant	100:1 (100:1 Pa)	10:1 (10:1 Pa)	1:1 (1:1 Pa)	1:10 (1:10 Pa)	1:100 (1:100 Pa)
Mixture	20%:0.2%	2%:0.2%	0.2%:0.2%	0.2%:2%	0.2%:20%
(+)Limonene	20.0%	2.0%	0.20%	0.20%	0.20%
(−)Limonene	0.20%	0.20%	0.20%	2.0%	20.0%
Amyl acetate	0.25%	0.25%	0.25%	0.25%	0.25%

Note. Pa = Pascals.

Discussion

Summary

The data presented here show that over large ranges of concentration ratios in binary mixtures various answers may be obtained regarding component perception. Configural representation is sometimes maintained over extended ranges in concentration ratios. Both mixtures containing benzaldehyde show a configural range in which only the trained mixture is identified over the control odorant (see Figure 2). Our laboratory showed previously that a mixture of citronellal and octanal, both agonists of the I7 receptor, produced an extended configural range, displayed in

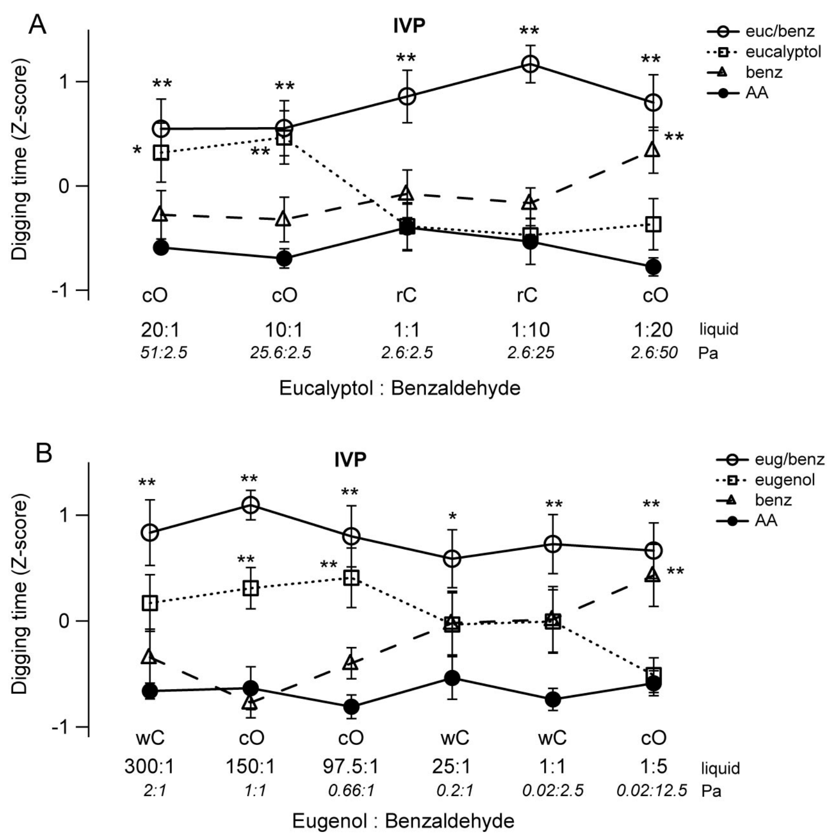

Figure 2. Mixtures with little structural similarity. Each plot shows mean response (z score of digging time with standard error) on the vertical axis and the training mixture component ratio on the horizontal axis. The line labeled “liquid” shows the ratio of volume/volume dilutions used (absolute liquid concentrations in Tables 1–2). The line labeled “Pa” shows the theoretical vapor pressures (in Pascals) of the components as ratios. Note that at the inverse ratio of the vapor pressures (IVP), the ratios are close to 1:1 Pa. A: Eucalyptol (1,8-cineole; euc) and benzaldehyde (benz) with a robustly configural (rC) range at 1:1 and 1:10 flanked by regions of complete overshadowing (cO) (see Discussion). B: Eugenol (eug) and benzaldehyde with a weakly configural (wC) range at 25:1 and 1:1, flanked by cO. The wC response at 300:1 is similar to the cO response at 150:1 (see Discussion). Asterisks indicate significance of response over control odor response (* $p < .05$, ** $p < .01$). AA = n-amyl acetate.

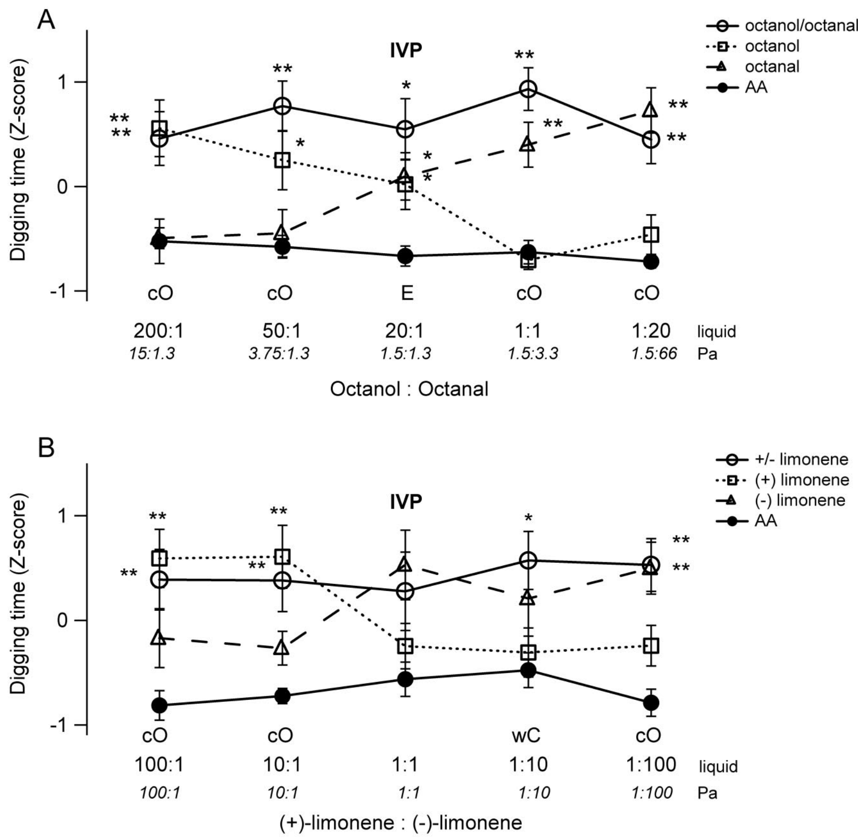

Figure 3. Mixtures with great structural similarity. Each plot shows mean response (*z* score of digging time with standard error) on the vertical axis and the training mixture component ratio on the horizontal axis. The line labeled "liquid" shows the ratio of vol/vol dilutions used (absolute liquid concentrations in Tables 3–4). The line labeled "Pa" shows the theoretical vapor pressures (in Pascals) of the components as ratios. A: Responses to mixtures of octanol and octanal. Note the point of elemental response (E) at the inverse ratio of the vapor pressures (IVP). On either side, one component overshadows the other. B: Responses to (+/−)-limonene mixtures. At each point except at the 1:1 ratio, the component of greater concentration is equated with the mixture, with most of the responses designated at complete overshadowing. At the 1:1 ratio, none of the responses to test odors is significant. AA = n-amylacetate; cO = complete overshadowing; wC = weakly configural. $^{*}p < .05$; $^{**}p < .01$.

Figure 4 (Araneda, Kini, & Firestein, 2000; Kay et al., 2003). On the other hand, we show here that elemental responses can occur in a mixture of compounds (octanol and octanal) that are known to activate one or more overlapping receptor types (Araneda, Peterlin, Zhang, Chesler, & Firestein, 2004). In the octanol–octanal tests, we show that rats recognize the element of highest concentration at every point but the IVP ratio, and they produce an elemental response at the IVP ratio (see Figure 3A). Our previous experiments with mixtures of citral and octanal (putative antagonist and agonist, respectively, of the I7 receptor) showed that an elemental range may extend over several ratios (Kay et al., 2003). We now know that these two structurally related compounds are both agonists of other receptors (Araneda et al., 2004).

Mixtures of enantiomers give us a window on the extremes of structural and perceptual similarity. When presented with (+/−)-limonene mixtures, the rats appeared to perceive the component of higher concentration in each of the mixtures flanking the IVP ratio (1:1; see Figure 3B), whereas at the IVP ratio, neither the mixture nor the components produced significant responses over the control odor. Because the enantiomers smell alike, in the tests that were not 1:1, the rats may simply have responded to the component with intensity near that of the trained mixture as has been seen in human mixture psychophysics (Laing, Panhuber, Willcox, & Pittman, 1984). It has been shown that when these enantiomers are differentially reinforced, it is possible for rats to distinguish them, even though in a habituation paradigm they are indistinguishable (Linster, Johnson, Morse, Yue, & Leon, 2002; Linster et al., 2001). However, at these levels of similarity, the animals may cue on impurities present in the two solutions rather than discriminating the enantiomers themselves. Our task represents a spontaneous perception of the mixture components, as the rats were not trained to detect them, and the results at the 1:1 ratio differ from those published previously using a similar task in which a configural response was seen (Wiltrout et al., 2003). The differences in results between the two studies may be due to the

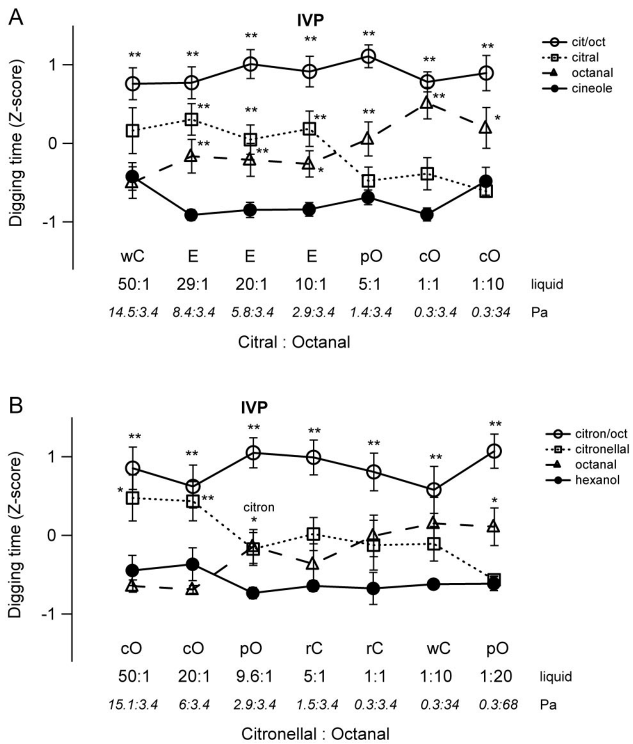

Figure 4. Agonists and antagonists of the I7 receptor. Each plot shows mean response (*z* score of digging time with standard error) on the vertical axis and the training mixture component ratio on the horizontal axis. The line labeled "liquid" shows the ratio of vol/vol dilutions used (absolute liquid concentrations in Tables 1–4). The line labeled "Pa" shows the theoretical vapor pressures (in Pascals) of the components as ratios. This figure is adapted from "Receptor Contributions to Configural and Elemental Odor Mixture Perception," by L. M. Kay, C. A. Lowry, & H. A. Jacobs, 2003, *Behavioral Neuroscience, 117,* p. 1110. Copyright 2003 by the American Psychological Association. A: Responses to citral (putative I7 antagonist) and octanal (I7 agonist) mixtures and components. The elemental (E) range (29:1 to 10:1) is flanked on the right by first partial overshadowing (pO) and then complete overshadowing (cO) responses. The leftmost ratio (50:1) is classified as weakly configural (wC), but if the response to the control odor had been lower, it would easily have been classified as either pO or cO. B: Responses to citronellal (I7 agonist) and octanal mixtures. The configural range extends from 5:1 to 1:10, including robust and weak regimes. At the inverse ratio of the vapor pressures (IVP) the response is classified as pO, but it is nearly identical to the neighboring robustly configural (rC) response. Flanking the configural range are overshadowing responses. $^{*}p < .05$; $^{**}p < .01$.

absolute concentrations of the two odor preparations (our concentrations were approximately one tenth of those used in that study).

A likely influence on mixture perception is the presence of contaminants, which may be of higher vapor pressure than the intended components and thus be more salient (Brockerhoff & Grant, 1999; Fraser, Mechaber, & Hildebrand, 2003). We also expect that different compounds will have different impurities and that some will be more affected than others. For instance, aldehydes are particularly subject to oxidation. One way to begin to approach a solution to this problem, in addition to using odorants of the greatest purity possible, is to use the same compound from many different sources and of many levels of purity in the same combinations with other compounds, so that the differences due to contaminants can be averaged out over many experiments.

Redefinition of Terms

The data presented here suggest a reinterpretation of previous mixture studies and our resultant theories about odor mixture perception. First, in many of the ratios tested there is recognition of only one component in the binary mixture; these mixtures have been called both partially elemental (Kay et al., 2003) and configural (Wiltrout et al., 2003). We propose that the most accurate description of this latter phenomenon is overshadowing (Linster & Smith, 1997; Pelz, Gerber, & Menzel, 1997; Smith, 1996). We use overshadowing to represent the decrement in representation of one component by an increase in concentration of the other, such that at some concentration ratio one component and the trained mixture are recognized, while the other is ignored. In every mixture tested, we found a region of overshadowing by a component of higher concentration. In almost all binary mixtures we tested, these points can be found at both extremes of the ratios, and in some of these mixtures the overshadowing region covers an extended range including the IVP ratio (see Figure 2). It is likely that overshadowing is caused by masking during learning, which can be due to intensity differences or interactions at the receptor or glomerular level. However, we cannot rule out the possibility that higher level cortical processing is involved in overshadowing without masking in some odor pairs.

In most cases of overshadowing, particularly those driven by extreme concentration ratios, the mixture and the stronger component are perceived as equal. We call this *complete overshadowing*. However, overshadowing may not always be complete, as is the case for 150:1 eugenol to benzaldehyde responses. Both the mixture and eugenol receive responses above those to the control odor, but the mixture responses are significantly stronger than those to eugenol ($p < .01$). This type of response we call *partial overshadowing* because a significant difference between perception of the mixture and the dominant element is likely influenced by the "silent" component. A second reinterpretation follows from overshadowing. Depending on the concentration ratio, there may be various interpretations regarding the mixture as configural or elemental. For example, the 1:1 ratio of eucalyptol and benzaldehyde suggests a configural representation, but increasing the amount of eucalyptol changes the apparent representation to complete overshadowing.

The question remains whether it is appropriate to call any mixture elemental or configural. We argue that a truly elemental response is one in which (a) both components are detected above control, and in the most stringent definition, either (b) both component responses are equal to the mixture response (e.g., the octanol–octanal IVP ratio in Figure 3A), or (c) the component responses sum to the mixture response. We also argue that a *robust configural* response is one in which only the trained mixture is recognized, with the response significantly higher than to both components and a control odor (e.g., eucalyptol–benzaldehyde 1:1 and 1:10 ratios in Figure 2A). A *weakly configural* response is then one in which only the trained mixture is recognized, but the response is not significantly larger than each component. The line between weakly configural and partial overshadowing is therefore indistinct, and it is our hope that as we understand the mechanisms for these effects better, the distinction will be clearer. Because configural and elemental responses are normally flanked by overshadowing regimes, it is important when describing any response to include the relative and absolute concentrations of odorants.

In light of these new definitions, we reexamined our previous study (Kay et al., 2003). The data are displayed in Figure 4. We previously claimed that mixtures of citral and octanal presented largely elemental responses because of the presumed inhibition of the I7 receptor by citral. Over several concentration ratios (29:1, 20:1, and 10:1), we showed true elemental responses with responses to both components significantly above the control odor. To the right of the elemental range, the responses drop to partial and then to complete overshadowing. On the left of the elemental range, the response is classified as weakly configural, primarily because the response to the control odor was elevated. If this response were more in line with control responses on other tests, it would likely have been classified as partial or complete overshadowing, showing a relationship between overshadowing and weak configural responses. The assessment of mixtures of citronellal and octanal (both I7 agonists) as containing a configural range also still holds, but it is only robustly configural for the ratios of 5:1 and 1:1. At the IVP ratio (9.6:1), the relationship between overshadowing and weak configural responses again becomes apparent. On either end of this configural range is complete overshadowing.

We have not encountered any data sets which do not vary smoothly with changes in ratios, and we have not seen both elemental and robust configural responses, as stringently defined above, for the same mixture set. This suggests that there are some simple interactions either in intensity or in receptor biophysics that vary the degree to which a component is recognized in a mixture. Overshadowing provides a means by which to understand these smooth transitions, as components increase and decrease relative to each other and pass from overshadowing into either elemental or configural regimes.

Theoretical Implications

In natural odor mixtures, components are not usually found in a 1:1 or IVP ratio, so it is important to test a range of concentration ratios before forming rules about mixture perception. Thus, we are in a position to examine two rules of mixture perception which are now gaining favor: (a) mixtures of structurally similar chemicals activate overlapping receptors and glomeruli and produce configural perceptual qualities, and (b) mixtures of structurally different chemicals result in less overlap of activated glomeruli and produce elemental perceptual qualities. (A recent computational model explains how these representations can occur within the OB via lateral inhibition between glomeruli; Linster & Cleland, 2004.) Our data show that in some binary mixtures the opposite is true. Octanol and octanal are structurally similar, differing at the functional group but with the same 8-carbon aliphatic chain, and they activate several overlapping olfactory receptors and presumably overlapping glomeruli (Araneda et al., 2004; Mombaerts et al., 1996), so the first rule predicts that mixtures of these two should show a configural range. However, the mixture shows an elemental response at the IVP ratio with complete overshadowing at ratios larger and smaller than this (see Figure 3A). It is important to note that these two chemicals do not smell alike, in contrast to other structurally similar compounds tested (Wiltrout et al., 2003). On the other hand, the eucalyptol–benzaldehyde mixture shows a robustly configural range, even though the components are not structurally similar and do not smell alike, challenging the validity of the second rule. Eugenol–benzaldehyde responses show a weakly configural range, and the components show only very gross structural similarity arguably not more similar than eucalyptol and benzaldehyde.

We previously proposed that overlap at the receptor level may lead to configural perception (Kay et al., 2003), but the results from the present study suggest that the mechanism is more complex than this. It is unlikely that all receptor types are equally represented in the olfactory epithelium or in their projections to the OB. It is equally unlikely that all odors activate equal distributions of receptors and are equally objective tests of the hypotheses. For example, benzaldehyde has pheromonal effects on some insect species and it is present, along with octanal, in the body odors of some mammals (Natale, Mattiacci, Hern, Pasqualini, & Dorn, 2003; Wood & Weldon, 2002), so these odor representations may have special significance. Many chemicals have trigeminal effects even at relatively modest concentrations, which can contribute to perceptual quality (Cometto-Muniz, Cain, Abraham, & Gola, 2001). Anatomical and physiological studies also show that direct action on OB neurons could be effected by stimulation of trigeminal fibers in the nasal epithelium (Olpe, Heid, Bittiger, & Steinmann, 1987; Schaefer, Bottger, Silver, & Finger, 2002). Absolute concentration of odorants, not just ratios, can also play a significant role in component perception, perhaps separately from trigeminal influences (Cometto-Muniz, Cain, & Abraham, 2005).

Although we are unable at this point to replace the old rules with new ones, we can infer that combined action of most odor pairs at multiple receptors will produce competing effects of agonism, antagonism, and intensity and in the end account for much of the perceptual qualities of odors. Thus, a prudent way to proceed is to use many techniques and many odor sets with known receptor and glomerular activation patterns at multiple ratios and concentrations to describe the multidimensional aspects of odor mixture psychophysics.

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