# CSA-SFC Joint Meeting: Understanding Mixtures

## Understanding the ways in which chemicals are perceived both alone and in combination with other materials

Consumers of foods and beverages don't buy chemicals, they buy perceptions, explained Terry Acree (Cornell University) during the recent joint meeting of the Society of Flavor Chemists and Chemical Sources Association at the Valley Regency in Clifton, New Jersey. And so, understanding the relationships between chemicals and perceptions has always been the challenge of flavor science. Aside from a few isolated examples, consumers eat no pure chemicals—everything that is perceived is a mixture of materials.

"What your companies do is sell chemicals," said Acree. "We know that these chemicals produce what the consumers want, which is some kind of a perception." Understanding what happens in between was the subject of the day's talk.

Acree began his discussion by presenting the definition of flavor posited by Richard Hall in 1968, which still holds true to this day: "Flavor is the sensation produced by a material taken in the mouth, perceived primarily by the senses of taste and smell, and also by the general pain, tactile and temperature receptors in the mouth. Flavor also denotes the sum of the characteristics of the material which produce that sensation."

In 2005, Dana Small and John Prescott published a complimentary psychological-neurobiological definition: "Perceptions of the flavors of foods or beverages reflect information derived from multiple sensory afferents, including gustatory, olfactory, and somatosensory fibers. Although flavor perception therefore arises from the central integration of multiple sensory inputs, it is possible to distinguish the different modalities contributing to flavor, especially when attention is drawn to particular sensory characteristics."[1]

That, explained Acree, is a process of analysis: "We can analyze flavors and extract from them the individual components of what those flavors are."

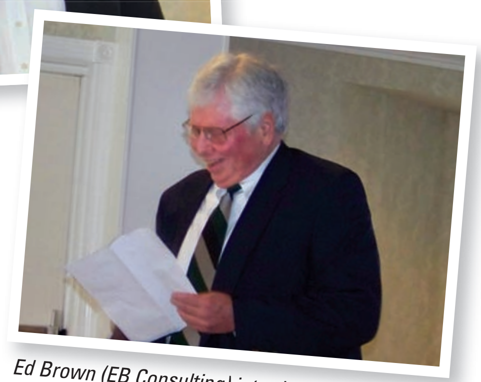
Small and Prescott's definition goes on to note, "Nevertheless, our experiences of the flavor of a food or beverage are also simultaneously of an overall unitary perception."

In other words, when humans eat, they typically don't analyze individual components, but instead "experience it." "That experience is instantaneous, complete, and requires no analysis," said Acree. "The neat thing is that humans can't avoid analyzing everything. So we do that too."

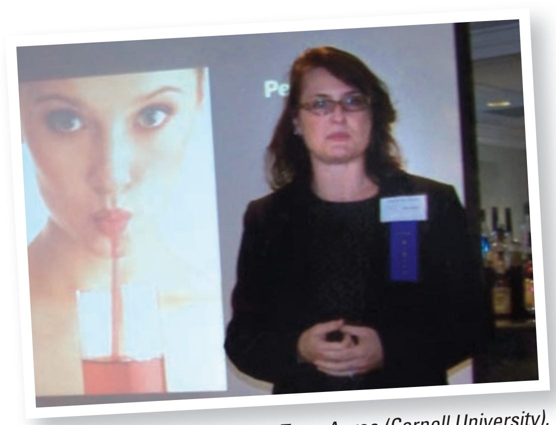
Ligands, which are chemicals or parts of chemicals, interact with receptors and produce sensations, which are signals that go to the central nervous system, said Acree.

*Perception, noted Terry Acree, is both analytical and synthetic.*

*Roger Trumm (Firmenich).*



*Ed Brown (EB Consulting) introduced Roger Trumm.*



*Hedy Kulka (IFF) introduced Terry Acree (Cornell University).*

Flavorist Roger Trumm presented a range of flavor materials to the audience.

Dolores Avezzano (Cargill) introduced the Chemical Sources Association presentation.

Christine Daley (Treatt) discussed the recently relaunched Chemical Sources Association website.

Fred Kiefer (Firmenich), discussed the range of materials on display.

These individual sensations are integrated to produce a "unitary perception," or they can be analyzed so that the person can try to experience the individual sensations. These sensations consist of smell, taste and chemosthesis. Perception, the picture painted by the sensations, is the combination of both qualitative experience (earthiness, sweetness, astringency) and intensity perception (weak, strong, faint). Those can be extracted in individual tests of individual components.

All of these sensations, said Acree, are spurred by relatively small molecules. "Polymers don't seem to be involved in anything, except for some unusual taste properties. In most cases they're under 500 or 1,000 Daltons.

"All of your business is tied up in this picture."

### Detection vs Recognition

"If we take any single stimulant like sugar and add it in increasing concentrations of water, we find that—if we have people rate how intense the sweetness is—as the concentration rises, their scores rise," said Acree. "And you ultimately reach a maximum in sweetness. This is called a dose response curve. This is characteristic of almost all receptor-based systems and living organisms."

He added that there is another method to define this phenomenon. "Instead of asking people to perceive an intensity of a thing, we can simply create some situation in which they [attempt to] distinguish one thing from another. As we increase the concentration of an odorant in such a situation, then the error rate starts to go down. In the beginning the error rate is extremely high. The detectability rate goes up with concentration. It's called a psychometric curve."

The initial curves in these situations register with the perceiver before they know what it is that is being smelled or tasted. "You don't know what you're smelling until after you've reached a certain level of probability of detection," said Acree. "There is a difference between when one can detect something and when one can recognize that sensory input. This difference is defined as a detection threshold and a recognition threshold."

Despite the ability to detect something before it is identified, any flavorist knows that recognition is key in foods and beverages. One wants to recognize what one is consuming. "It's not so much what you detect," said Acree. "What is unrecognizable in foods becomes neither bad nor good." It is neutral and thus unimportant from a sensory point of view. "The important things happen above the threshold of recognition."

### Flavor Perception: Synthetic or Analytic?

The challenge, noted Acree, is to take the composition of a food or flavor system and use chemical tools to 1) analyze what the composition is and 2) relate the elements of that composition to what people perceive in a psychometric test or in terms of behavior.

"Is flavor synthetic or analytic?" Acree asked. "Do all the chemicals [in a mixture] synthesize an idea in your head instantaneously or do you analyze them by detecting each one separately and then coming to some kind of conclusion?" The answer, he said, is that humans do both.

In discussing mixture perception, Acree focused on the example of Concord grapes, which are characterized by methyl anthranilate. Despite this, common Concord grape flavor descriptors include foxy, cotton candy, apple, Kool-Aid and musty. Classic Kool-Aid, noted Acree, is essentially methyl anthranilate, a few additional odorants and sugar, which is then added to water. So, he asked, "What is Concordness?"

In a study by one of Acree's students, panelists were screened for descriptive analysis of fresh Concord grape juice and Kool-Aid. Panelists were asked to determine

## Ingredient Tasting

The Chemical Sources Association's recent meeting at the Valley Regency in Clifton, New Jersey, featured a presentation of flavor materials from Roger Trumm, senior flavorist at Firmenich. All materials were natural or nature-identical, GRAS-certified and GMO-free. Those materials available for tasting by attendees were presented in pectin jellies.

Natural ***hexanal*** (FEMA# 2557) was presented at 20 ppm in acidified sugar syrup. The material was notably clean, Trumm explained, and had no oily aftertaste or fermented grass off note. The hexanal was sweet, fresh, fatty-green, grassy, unripe fruitlike. It was recommended for use at levels of up to 5 ppm in red fruit, apple, peach, orange, kiwi, pear, mango and grape flavors.

Nature identical ***1,1-dimethoxyethane*** (FEMA# 3426) was presented in 50 ppm in acidified sugar syrup. This replacer for acetaldehyde in spray dry flavors was fresh and bright. At levels up to 15 ppm it was recommended for use in citrus, tropical and yellow fruit flavors.

Nature identical ***pentyl formate*** (FEMA# 2068) was presented at 30 ppm in acidified sugar syrup. The material, Trumm noted, lacked any dirtiness or acrid quality. The material had the taste of green banana and a fusel oil note. It was notably fresh, bright and sweet. At levels up to 10 ppm it was recommended for use in banana (for fleshy-green effects), berry and rum flavors.

Natural ***2-phenylethanol*** (FEMA# 2858) was presented at 20 ppm in acidified sugar syrup. The material was sweet, with honey and rosy notes, but without greenness and leafiness. It was also bright and clean, with fermented and cocoa back notes and a sundried tomato impression at the end. At levels up to 5 ppm it is recommended for use in honey, red fruit, tomato, pomegranate, cheese, grape, melon and alcoholic drink flavors.

Natural ***2-phenylethyl acetate*** (FEMA# 2857) was presented at 30 ppm in acidified sugar syrup. It had a pronounced sweetness and featured a clean, bright finish and fermented, fatty back notes. At levels up to 5 ppm it was recommended for use in tea flavors at low levels, and guava, whiskey, cider and cherry flavors.

Nature identical ***2-ethoxybutane*** (FEMA# 3131) was presented at 10 ppm in acidified sugar syrup. It featured pronounced woody notes. At levels up to 10 ppm it is appropriate for tropical, blackcurrant and citrus flavors. It can also act as a replacer of acetaldehyde.

Nature identical ***methylphenylcarbinol isobutyrate*** (FEMA# 2687) was presented at 10 ppm in acidified sugar syrup. It was fruity, fleshy, woody, green and tropical. At levels up to 10 ppm it is useful in fig, strawberry, date and tropical berry flavors. It also pairs well with acidity as in guava and grapefruit.

High purity natural ***massoia lactone*** (FEMA# 3744) was presented at 2 ppm in sugar syrup. The material was creamy, coconutlike and spicy, with celery notes in back. Overall, it was full, sweet and clean. At levels up to 5 ppm, it is appropriate for peach, butter, cream, vanilla, pear, apricot, fruit, nut and tobacco flavors. It can also partially replace natural δ-decalactone.

Nature identical ***2-methylhexanoic acid*** (FEMA# 3191) was presented at 100 ppm in acidified sugar syrup. The material featured a pronounced creaminess and butter character and no caprylic fattiness or acrid note. It was also oily and dairylike. At levels of 10 ppm it is recommended for use in kiwi, passion fruit, cheese, raspberry (ripe notes) and berry flavors. It is also appropriate for fruity cooked profiles and for adding a Parmesan note to flavors. 2-Methylhexanoic acid possesses antibacterial properties useful in oral care flavors at a level of about 100 ppm.

Natural ***3-mercapto-2-methylpentanal*** (FEMA# 3996; CAS# 227456-27-1) possessed onion, bacon and scallion notes.

Also available, though not specifically discussed, was natural ***3-mercapto-2-butanone 10 NB*** natural (FEMA# 3298). The material is eggy, meaty, custardlike, fatty, chicken brothlike, and slightly roasted. At levels up to 5 ppm it is useful in savory and meaty profiles with low sulfur or garlic impact. It is also useful in chicken, caramel, smoke, beef, egg, dairy and bread flavors.

***Mercaptomethylbutanol*** (FEMA# 3854) is fruity at low dosages and then becomes sulfury, meaty, beef juice, and gravylike at higher dosage ranges.

which beverage was more Concordlike. There were some who reported that both tasted identical, others who believed Kool-Aid was more Concordlike and finally those that identified Concord as the most Concordlike. Those that fell into the latter group are of course the ideal candidates for the study of Concord grape juice.

In studying what happens in mixtures, Acree noted that panelists submitting to qualitative descriptive analysis (QDA) of Concord grape juice using standard procedures would tend to use the term "Concord grape juice," reflecting a mindset that favors perception rather than analysis. In fact, most of what makes Concord grape juice so intense is its taste: sweetness, acidity, astringency and thickness. Only three odor descriptors stand out, according to Acree: fruity, berry and prune. And so, flavor can be described two ways: via sensation (foxy, grapelike) or individually (elemental, or analytic, processing). There is a temporal component to this, said Acree. One analyzes a flavor over time. The immediate reaction is configural, or synthetic: Concord grape. "These are odor images that your brain constructs." Over time, however, individual elements can be identified and analyzed as discrete qualities, descriptors or notes, typically related to specific chemicals.

## Individual Components vs Mixtures

Analyzing a mixture via a gas chromatography olfactometry (GC-O) allows the flavor chemist to pick up on components. However, Acree said, they are being experienced outside of the mixture and are consequently of limited value in understanding mixtures. Acree's analogy likens using GC-O to understand mixtures to trying to understand a play by looking at a list of its cast members. "If you want to know what the play is you have to sit and watch the play."

As one dilutes a pure odorant, its odor does not change, continued Acree. This flies in the face of conventional flavorist wisdom, which states the reverse. How can both be true?

Acree explained that so much smelling done by flavorists involves mixtures. As ratios among components change, the overall character changes. The exception, said Acree, were chemicals presented in very high concentrations. "You activate all kinds of receptors and start to get different kinds of odors," he said of those cases. "The odor you usually get if you get it high enough is the smell of a synthetic organic chemistry lab." Other exceptions include molecules that have epitopes that react with more than one receptor type, including 2-methyl isoborneol; at high concentrations it smells of camphor, while at low concentrations it smells earthy-musty. The material, explained Acree, interacts with two different receptors that have different thresholds, which accounts for this phenomenon.

## Understanding How Perception of Mixtures Works

In studies of wines, four cultivars can have 60-plus odorants, many of which are present in all of the cultivars, but at varying levels. About four to eight descriptors are typically attached to these four cultivars. Acree noted the clear disconnect between the number of odorants a chemist can find in a mixture and the number of descriptors they will elicit from consumers. This, he said, had been reproduced in many different experimental settings.

In some research, panelists were trained to identify individual odorants. When the odorants were presented alone, 80% identified it correctly. When two materials were combined, 35% of panelists identified both correctly. When three were combined, just 14% of panelists identified all three chemicals. When four materials were combined, there was no statistical significance to ability of panelists to identify individual components.

In the flavor perception of sauvignon blanc, across many panels and types of wine, the dominant descriptor was passion fruit (GC-O identified (S)-2-hexyl acetate), followed by urine (4-mercapto-4-methyl pentan-2-one) and a bell pepper note (isobutyl-2-methoxypyrazine). The "wrinkle" in the research, said Acree, was the notable presence of isoamyl acetate in the wines, which has a strong bananalike character. This was a dominant odor character in the wines being studied, yet "banana" was never noted by panelists. Why? Panelists were never given banana as a descriptor option. As Acree concluded, "They don't describe what they don't seek."

In understanding what makes a Riesling wine taste as it does, researchers have discovered TDN, a norisoprenoid with a diesel-kerosene odor. The component plays an important role among other notes such as citrus and pineapple-grapefruit. By adding TDN to no-oak chardonnay, which has a very neutral taste, the following was found:

- At 1 ppb TDN there was no detection
- At 2 ppb TDN there was a detectable difference between those with and those without the material
- At 10 ppb TDN the fruitiness in the wine fell away
- At 20 ppb TDN the wine smelled like a typical Riesling
- At 100 ppb TDN, the wine took on an "atypical aging" character reminiscent of overly aged or light-exposed Riesling (Rieslings should be drunk within 10 years of production because TDN levels build up to off-putting levels over time)

The detection threshold of TDN is 10–20 ppb (in water and ethanol), which is the level found in most Rieslings. Yet in the no-oak chardonnay experiment, it was detectable below this. The conclusion? Adding a chemical to a mixture of other chemicals lowers its threshold.

Acree closed his talk with a discussion of mixture suppression and self- and cross-adaption among chemicals, in addition to emerging GC techniques that will promise new and better understanding of mixtures.

#### References


1. DM Small and J Prescott, Odor/taste integration and the perception of flavor. *Experimental Brain Research*, 166, 345–357 (2005)