The Olfactory System

The human and rodent olfactory systems exploring the sensory world together.

Thomas Brewer

MacMillan Group Meeting

June 13, 2019

Senses and Stimuli

sight

light (electromagnetic wave)
wavelength, intensity

hearing

sound (pressure wave)
frequency, intensity

touch

mechanical pressure
temperature

responsive to physical parameters easily quantified and described

Senses and Stimuli

sight

light (electromagnetic wave)
wavelength, intensity

hearing

sound (pressure wave)
frequency, intensity

touch

mechanical pressure
temperature

chemical senses

taste

sweetness | sourness
saltiness | bitterness | umami

smell

essentially infinite types of stimuli

Senses and Stimuli

diverse stimuli that cannot be classified according to simple physical dimensions

chemical senses

taste

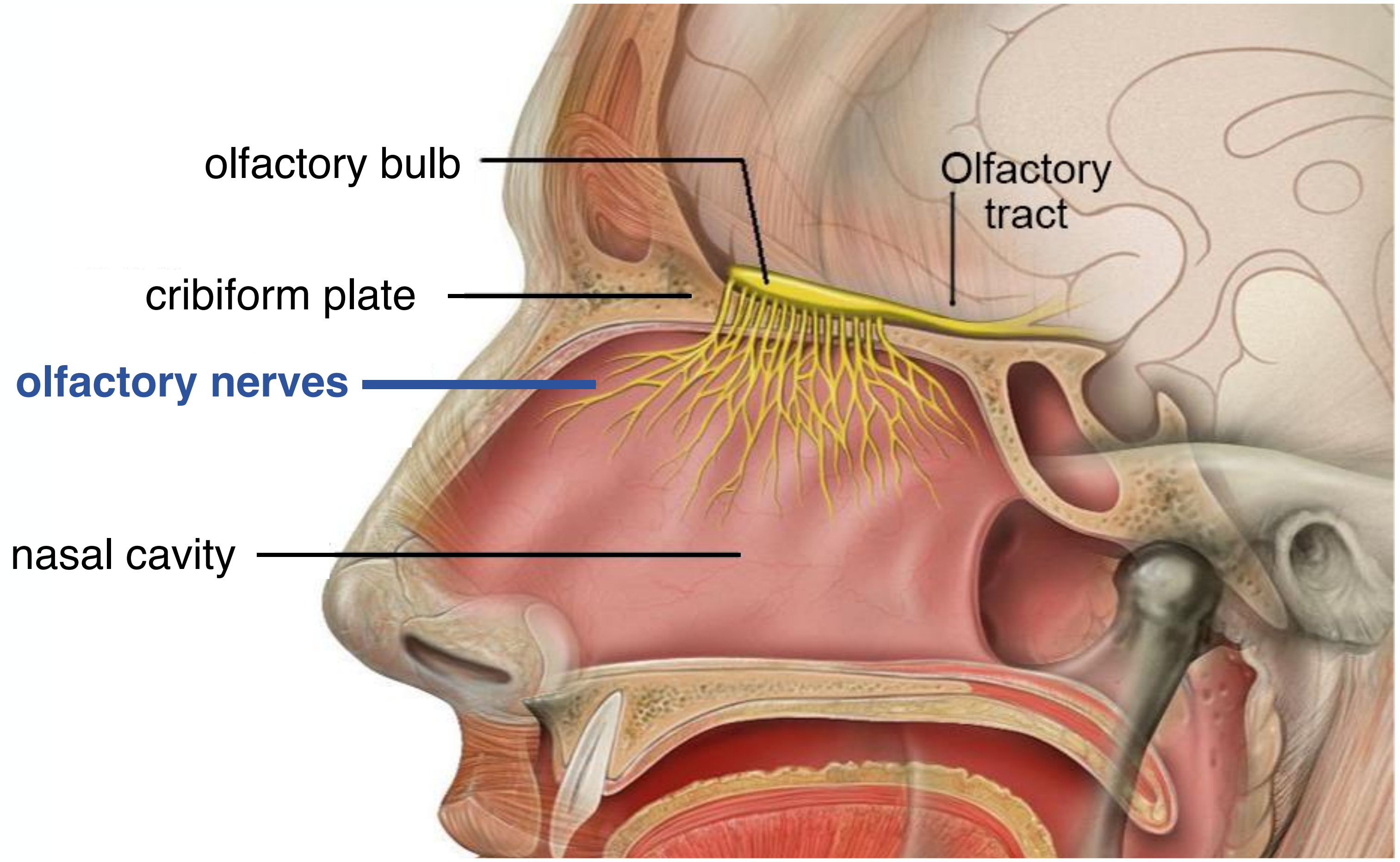
sweetness | sourness
saltiness | bitterness | umami

smell

essentially infinite types of stimuli

How does the olfactory system distinguish over **10^{117}** (low estimate) chemically diverse odorants?

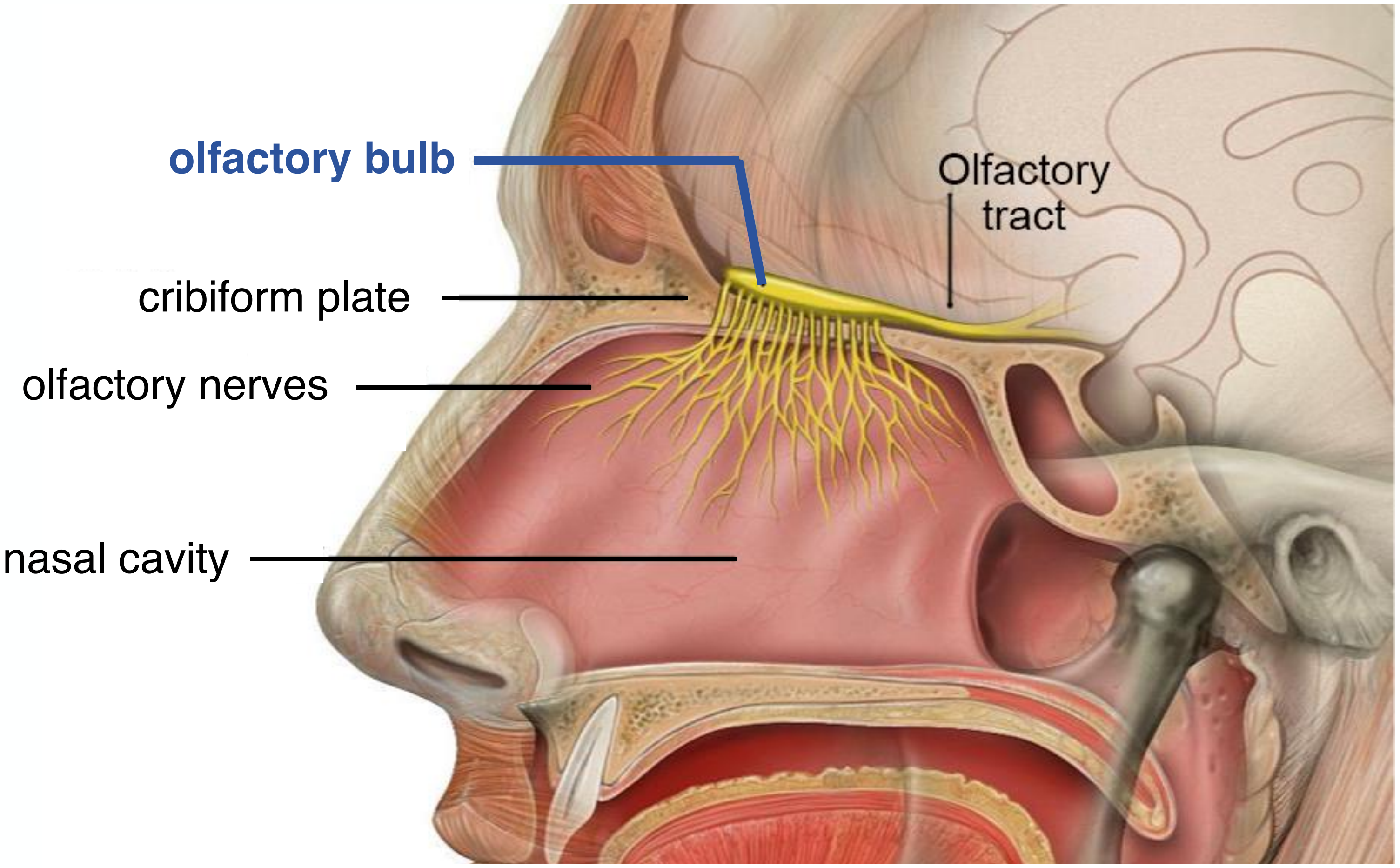
Anatomy of Olfactory System



- ca. 3 x 10^7 bipolar neurons
- contact atmosphere via ~30 olfactory cilia
- olfactory mucosa surrounds cilia

Ohloff, G.; Pickenhagen, W.; Kraft, P. *Scent and Chemistry: The Molecular World of Odors*; Verlag Helvetica Chimica Acta: Zürich, 2012.

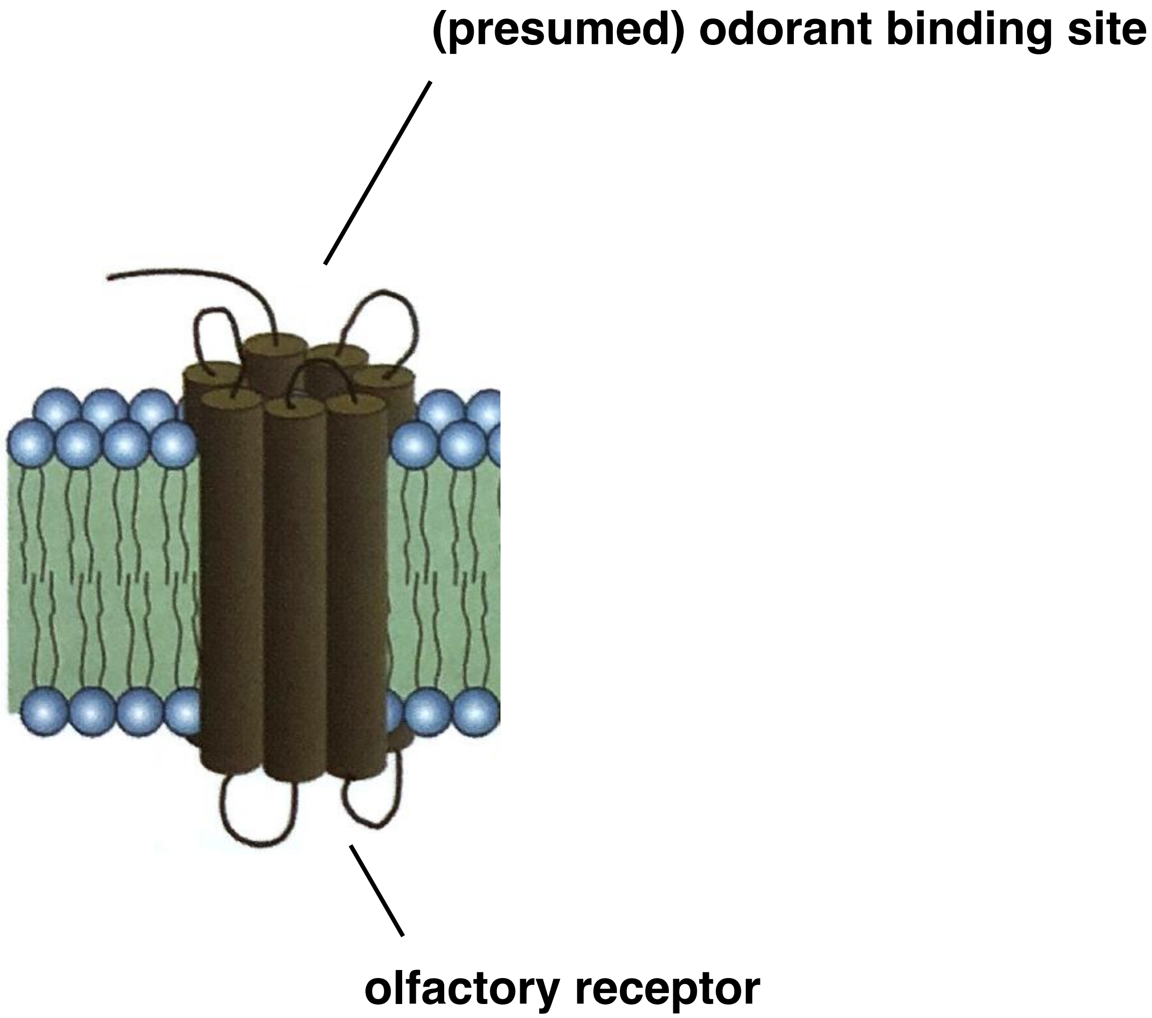
Anatomy of Olfactory System



- axons of ONs converge on globular structures called glomeruli
- activity pattern of glomureli codes odor impression
- information transferred via mitral cells in the olfactory tract
- olfactory cortex decodes signals into olfactory perception

Ohloff, G.; Pickenhagen, W.; Kraft, P. *Scent and Chemistry: The Molecular World of Odors*; Verlag Helvetica Chimica Acta: Zürich, 2012.

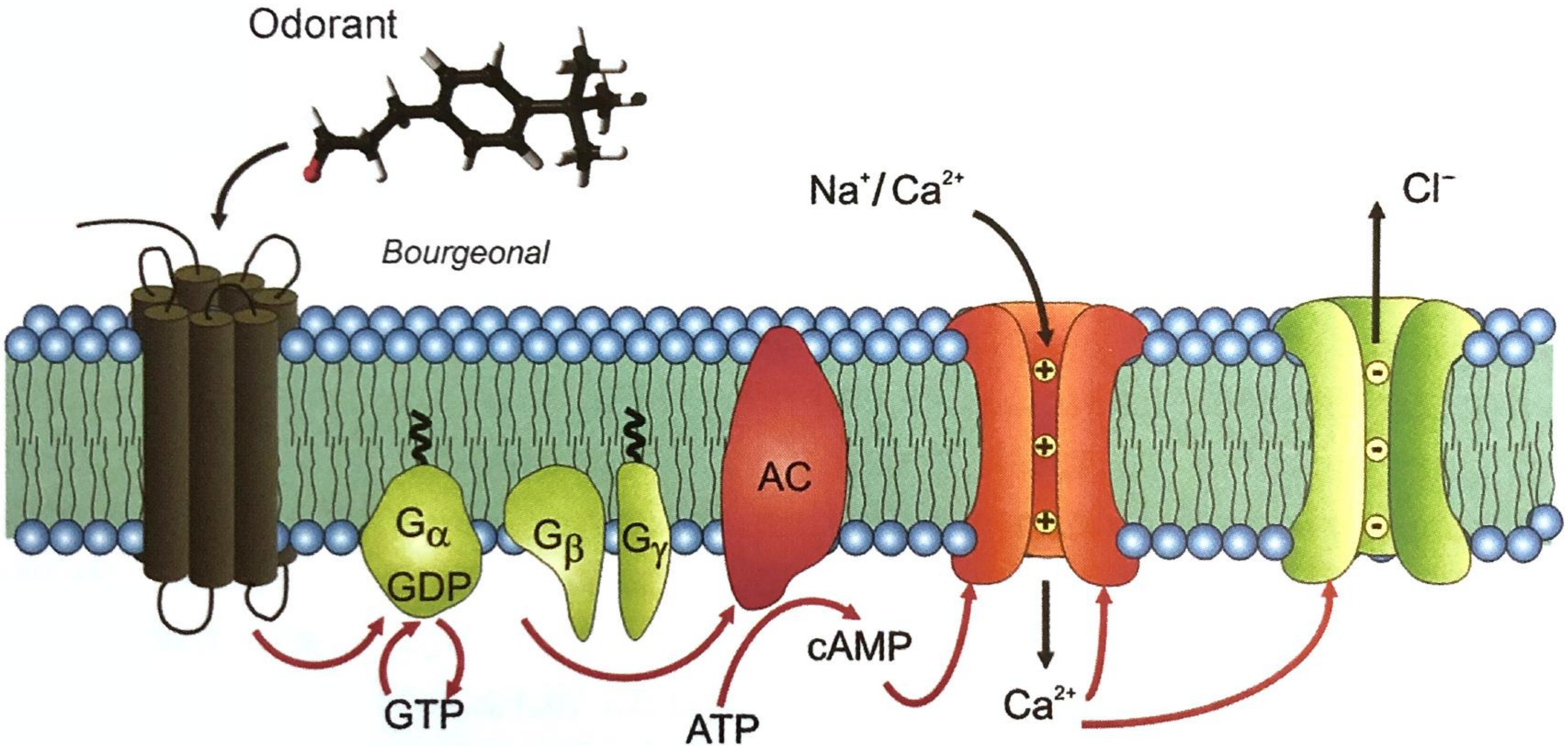
Physiology of Olfaction



- ca. 3% of human genome consists of olfactory receptor (OR) genes
- ~850 genes, ~400 functionally active genes

Ohloff, G.; Pickenhagen, W.; Kraft, P. *Scent and Chemistry: The Molecular World of Odors*; Verlag Helvetica Chimica Acta: Zürich, 2012.

Physiology of Olfaction



Ohloff, G.; Pickenhagen, W.; Kraft, P. *Scent and Chemistry: The Molecular World of Odors*; Verlag Helvetica Chimica Acta: Zürich, 2012.

The Olfactory Code

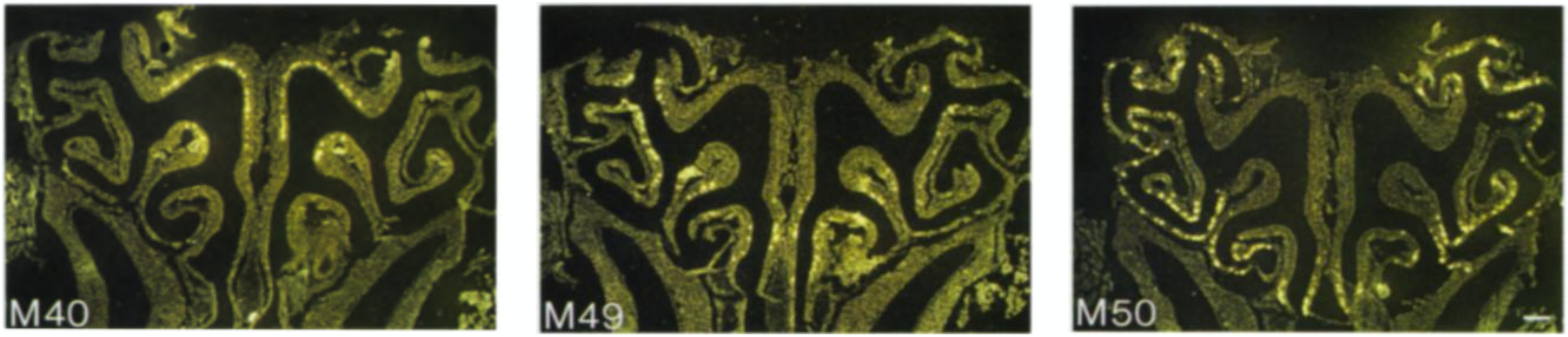
How is olfactory information sorted and regulated?

principles of pre-cortical olfactory coding

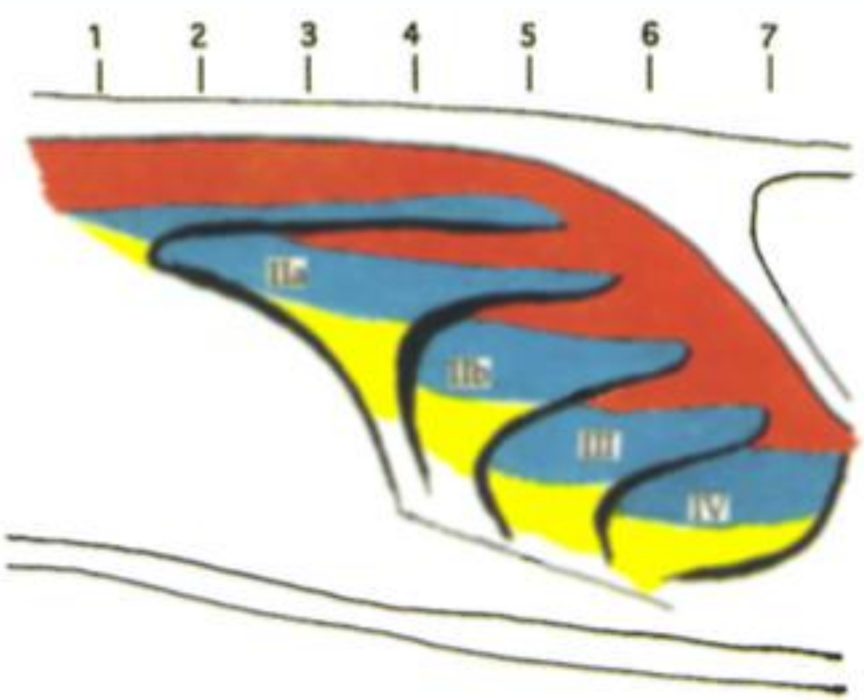
- Level 0: genetic variance of ORs
- Level 1: zonal organization of ORs
- Level 2: evolutionarily distinct receptor classes
- Level 3: perireceptor events

The Peripheral Olfactory Code: Zonal Organization

^{35}S-labeled antisense RNA *in situ* hybridization studies



lateral section



anterior → posterior

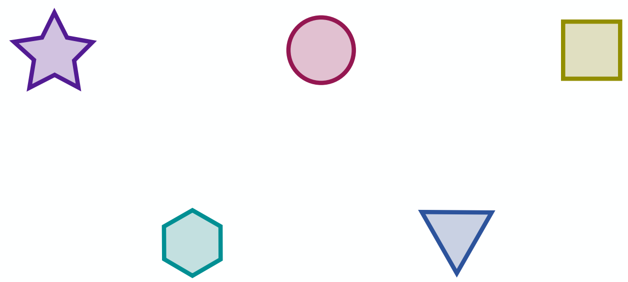
- zonal organization of ORs in olfactory epithelium
- topographically distinct expression zones
- clustering of subfamilies of related ORs
- initial step in information coding

Ressler, K. R.; Sullivan, S. L.; Buck, L. B. *Cell* **1993**, *73*, 5

The Peripheral Olfactory Code: Evolutionarily Distinct Receptors

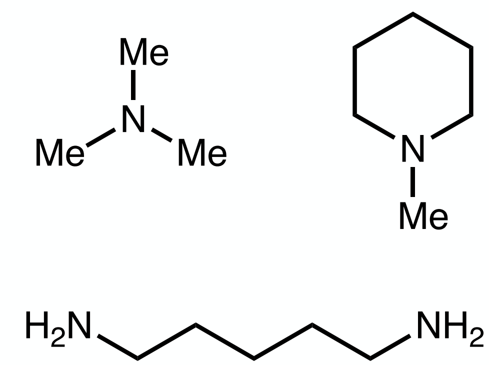
evolutionarily distinct receptor classes

olfactory receptors



everything else

trace amine-associated receptors



volatile amines

- canonical class
- humans express 391 variants
- divergent responses

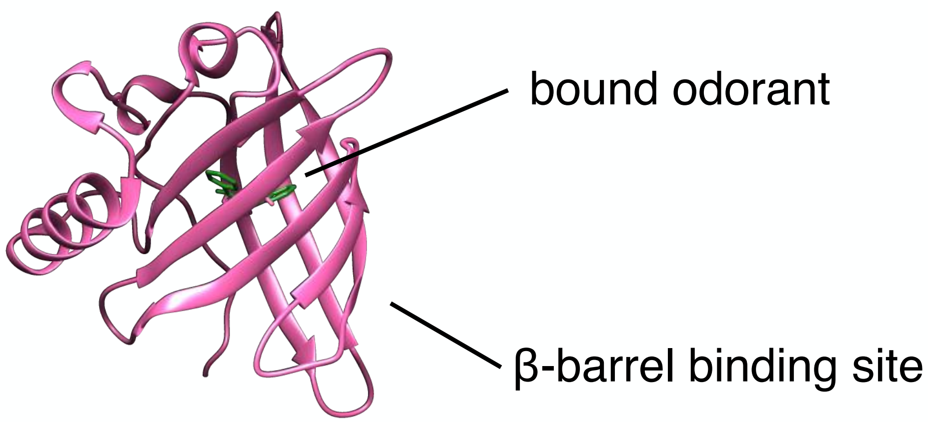
- more recently discovered class
- humans express 6 variants
- largely elicit aversive response

immediately discriminates at least one major group of volatile odorants

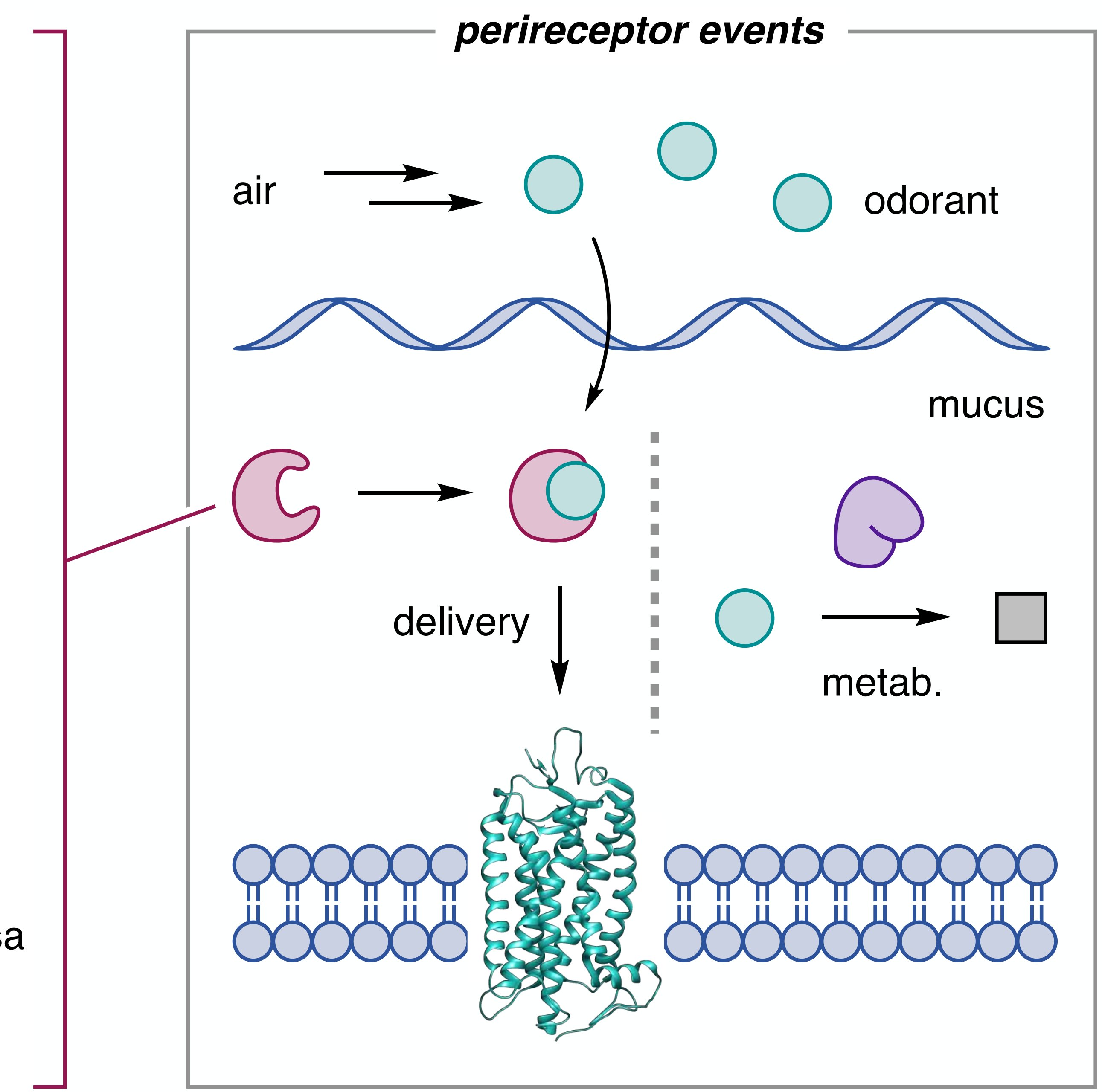
Liberles, S. D.; Buck, L. B. *Nature* **2006**, *442*, 645.
Liberles, S. D. *Curr. Opin. Neurobiol.* **2015**, *34*,

The Peripheral Olfactory Code: Perireceptor Events

odorant-binding proteins



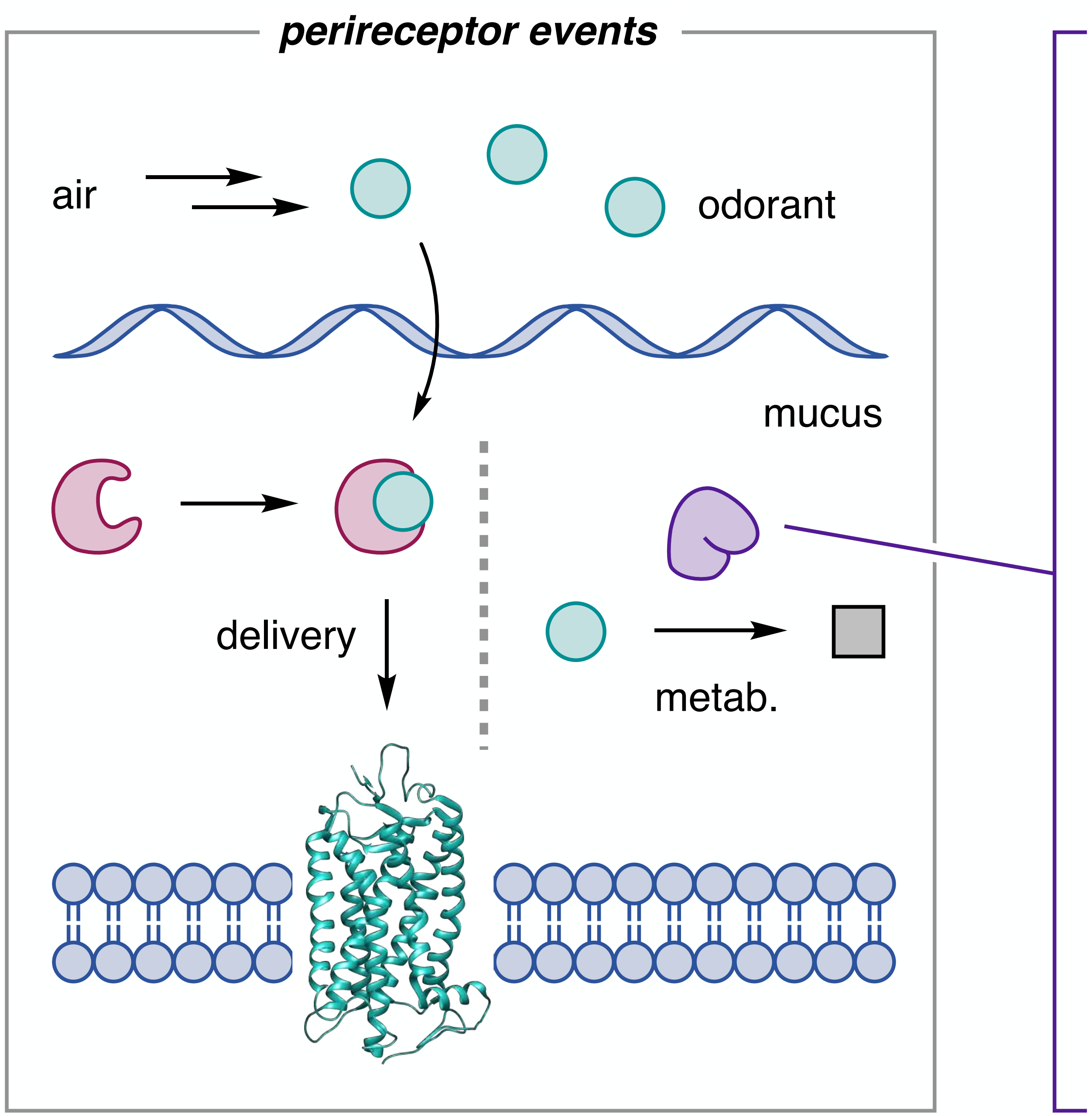
- 10 mM in olfactory mucosa
- ~19 kDa proteins
- bind diverse odorants with K_d ~ μM
- carry hydrophobic odorants through aq. mucosa
- buffer free odorant concentration



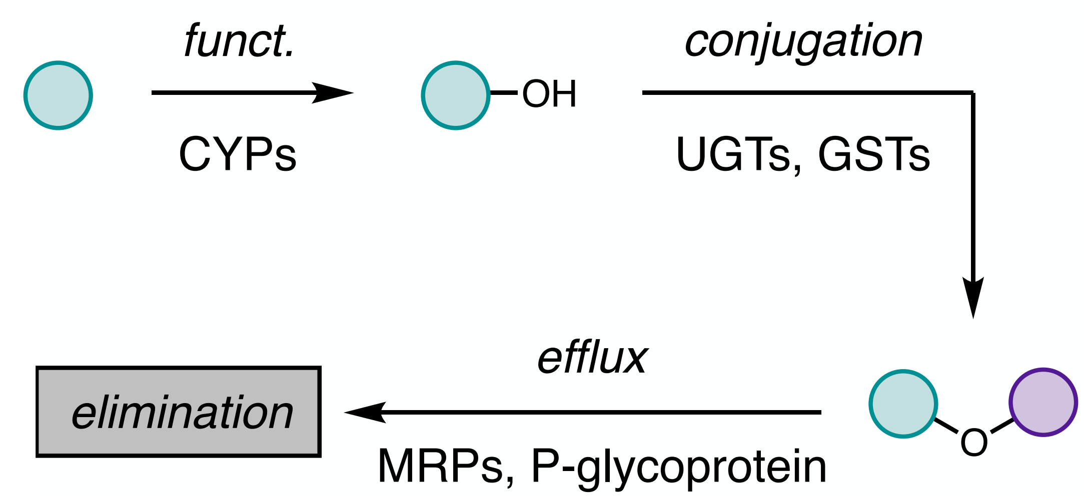
Vogt, R. G.; Riddiford, L. M. *Nature* **1981**, *293*, 161.

Heydel, J.-M., et al. *Anat. Rev.* **2013**, *296*, 13:

The Peripheral Olfactory Code: Perireceptor Events



odorant-metabolizing enzymes



- diverse families of enzymes
- highly expressed in olfactory mucosa
- detoxify harmful airborne molecules
- regulate odorant availability via metabolism

Lazard, D., et al. *Nature* **1991**, *349*, 790.
Heydel, J.-M., et al. *Anat. Rev.* **2013**, *296*, 13

Physiology of Olfaction

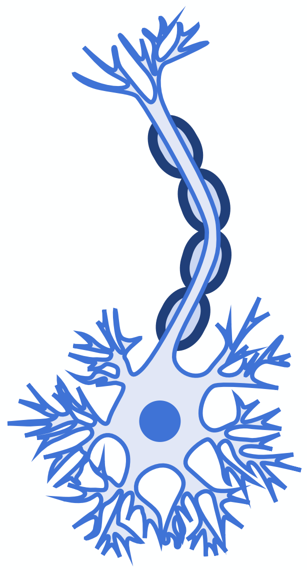
Nobel Prize in Physiology or Medicine for 2004

“for their discoveries of odorant receptors and the organization of the olfactory system”

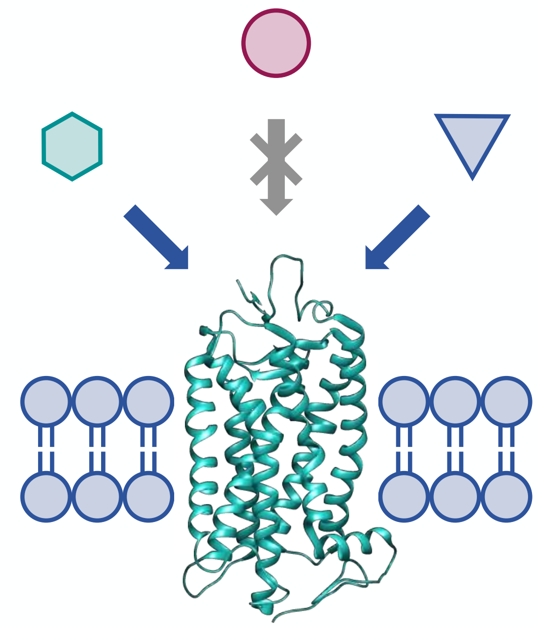
Richard Axel

Linda B. Buck

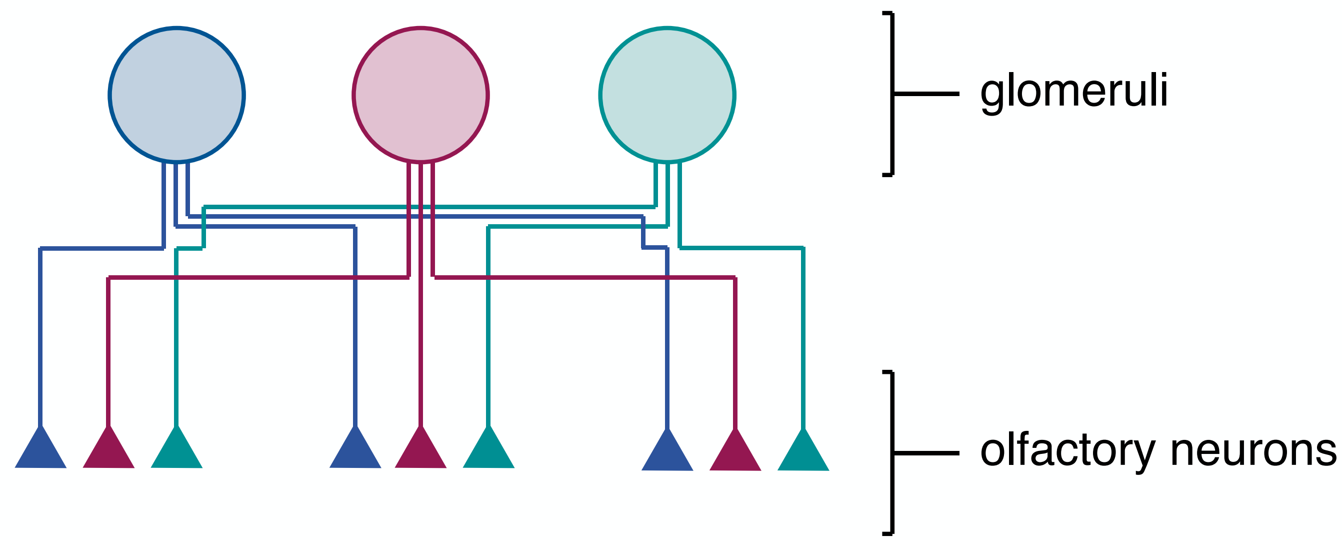
Physiology of Olfaction



each olfactory neuron expresses
one and only one olfactory receptor

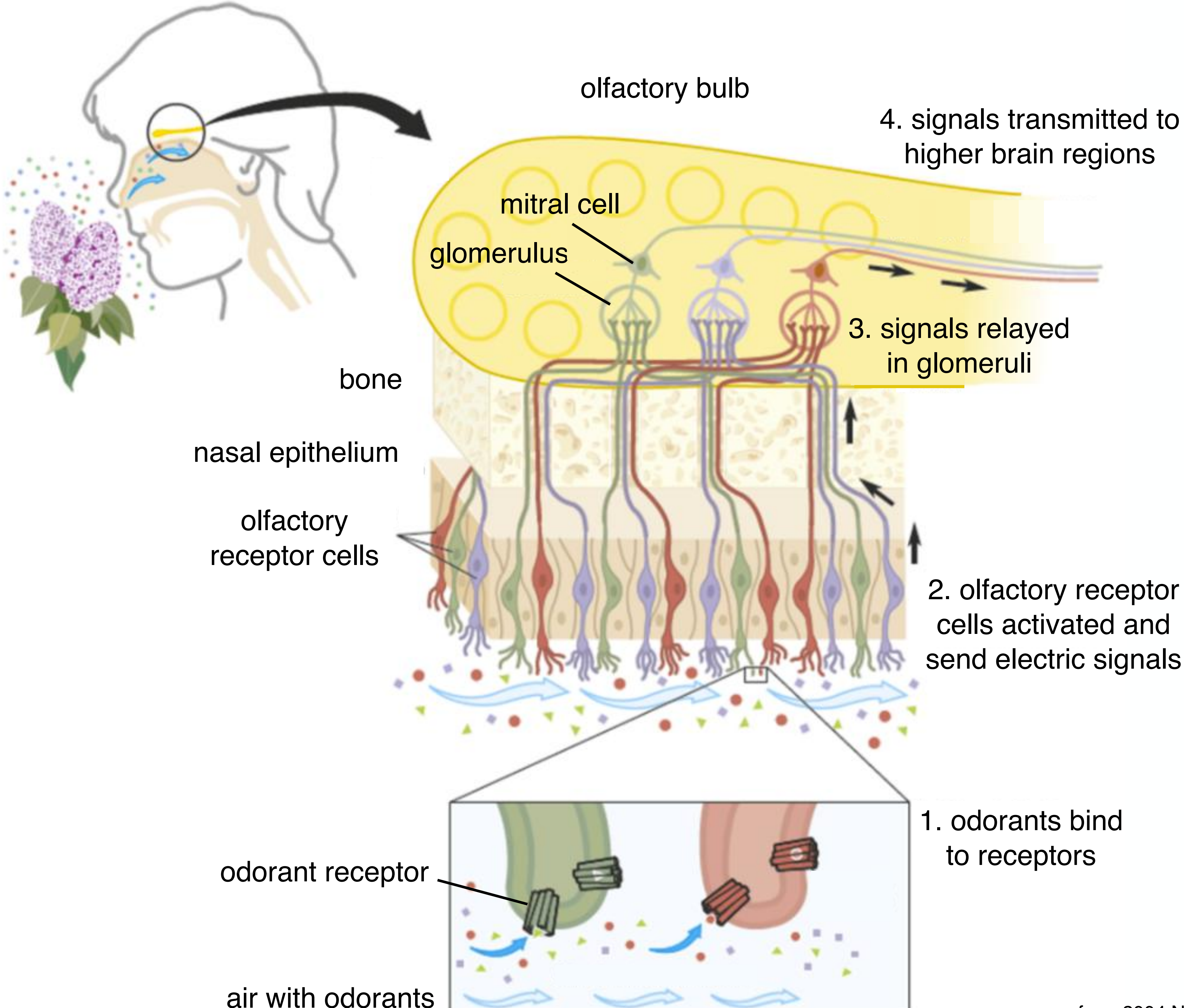


each olfactory receptor can detect
a *limited number of odorants*



olfactory neurons carrying the same type of receptor
converge on the same glomerulus

Physiology of Olfaction



from 2004 Nobel Prize in Physiology or Medicine press release

The Primary Olfactory Code

14 ORs surveyed in intact mouse olfactory neurons

	S1	S3	S6	S18	S19	S25	S41	S46	S50	S51	S79	S83	S85	S86	
Hexanoic Acid					■										rancid, sweaty, sour, goat-like, fatty
Hexanol		■				■									sweet, herbal, woody, Cognac, Scotch whiskey
Heptanoic Acid	■			■	■		■			■	■				rancid, sweaty, sour, fatty
Heptanol		■			■	■									violet, sweet, woody, herbal, fresh, fatty
Octanoic Acid	■			■	■		■	■		■	■	■			rancid, sour, repulsive, sweaty, fatty
Octanol				■	■		■			■					sweet, orange, rose, fatty, fresh, powerful, waxy
Nonanoic Acid	■			■	■		■	■		■		■		■	waxy, cheese, nut-like, fatty
Nonanol				■	■		■			■		■			fresh, rose, oily floral, odor of citronella oil, fatty

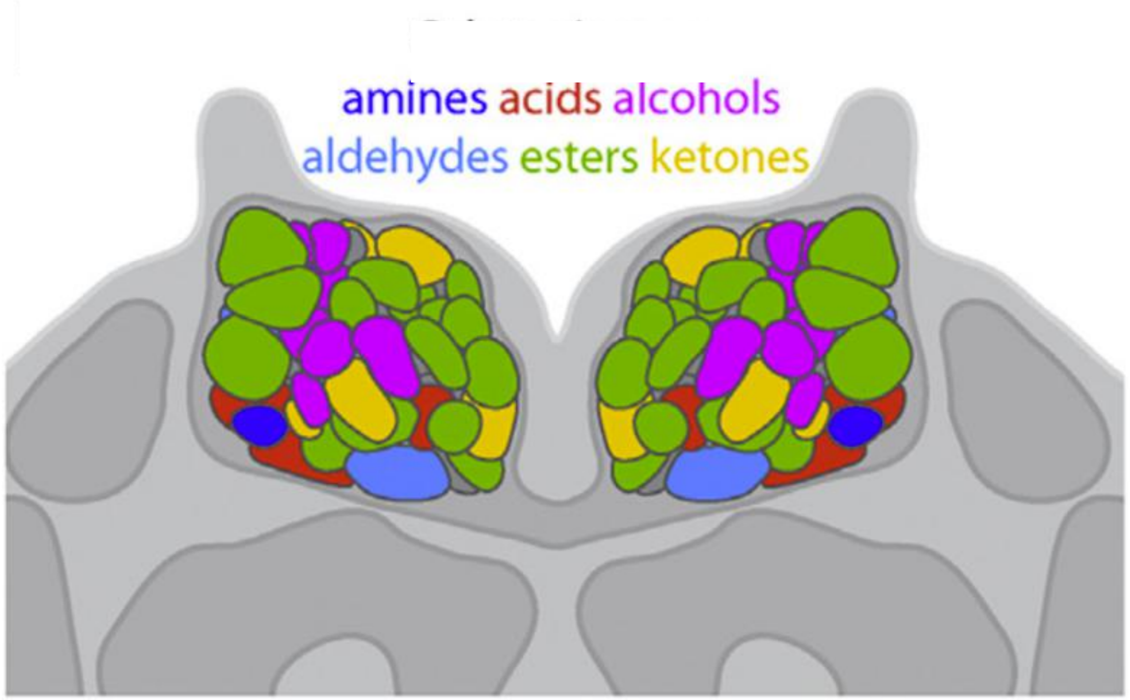
- activation visualized by calcium imaging and RT-PCR
- unique set of ORs are activated to varying degrees by individual odorants

Malnic, B.; Hirono, J.; Sato, T.; Buck, L. B. *Cell* **1999**, *96*, 713.

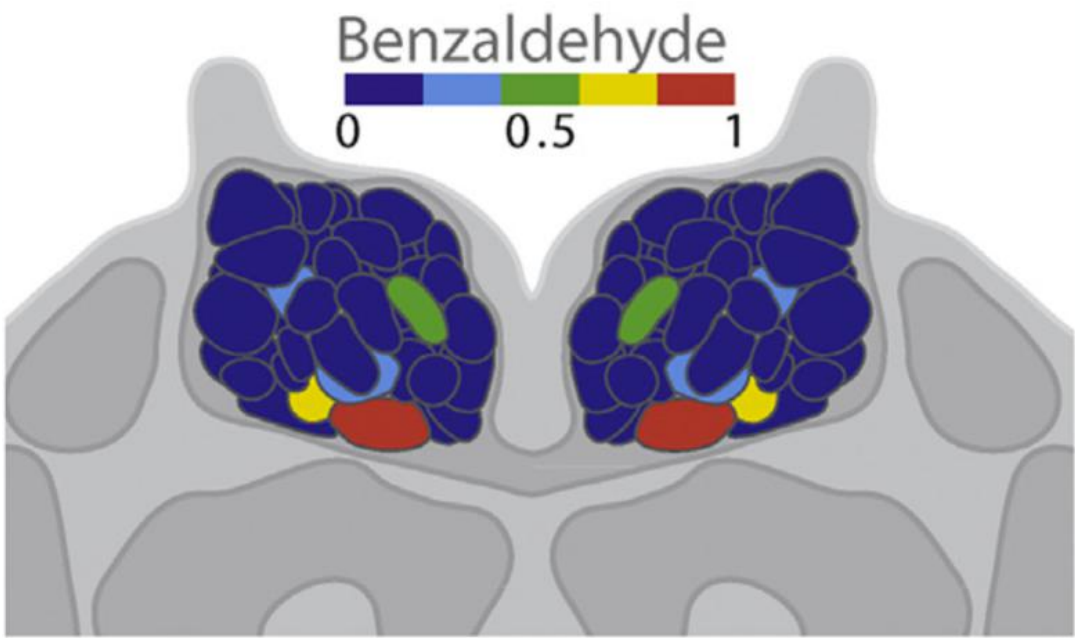
The Primary Olfactory Code

olfactory neuron/glomerular organization leads to an ***odotopic map***

odotopic map in *Drosophila melanogaster*



benzaldehyde odotopic map

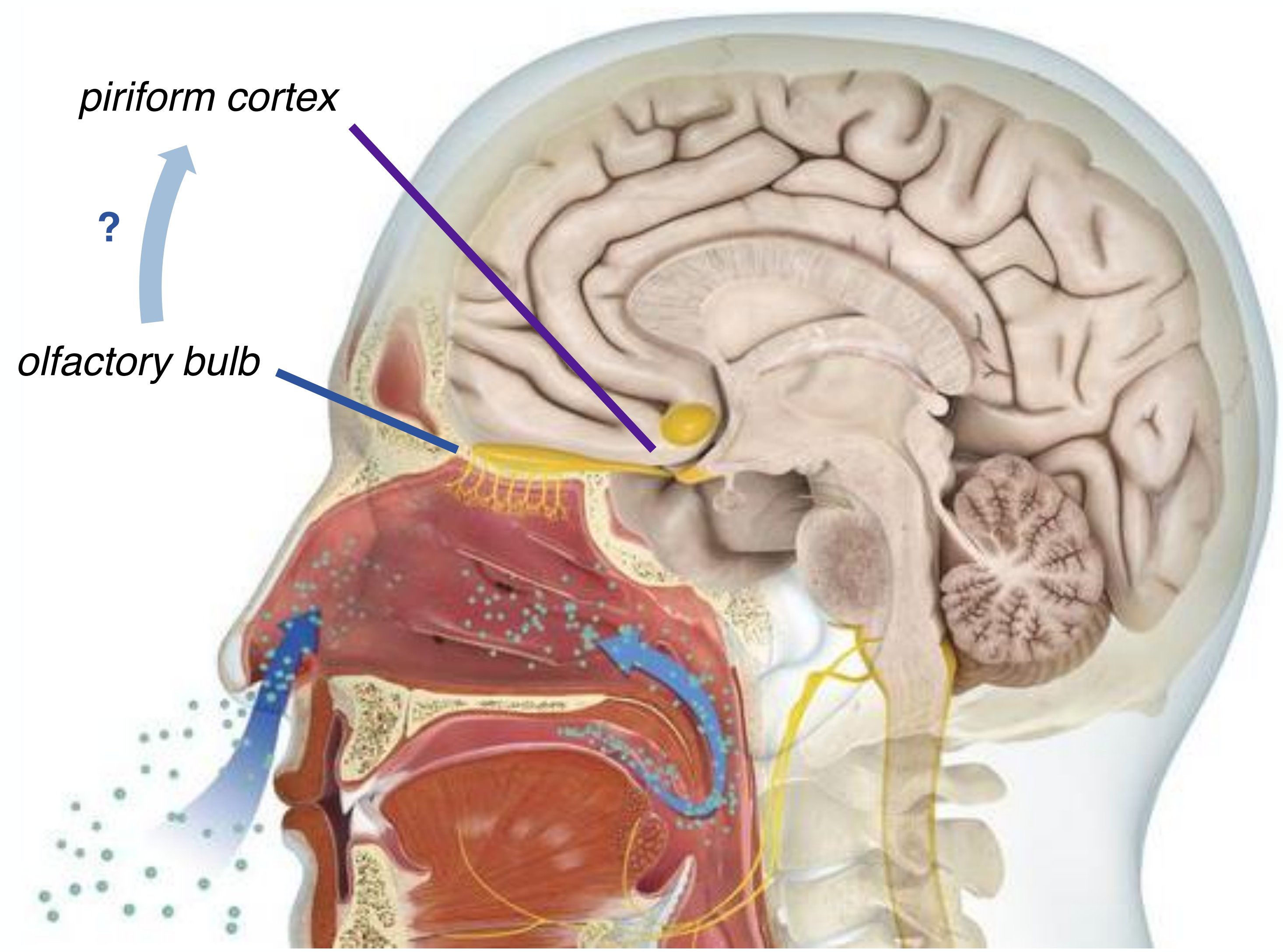


- specific sets of glomeruli activated for specific odorants
- chemically related odorants have related odotopic maps
- true for invertebrate through vertebrate brains

Grabe, V.; Sachse, S. *BioSystems* **2018**, *164*, 94.

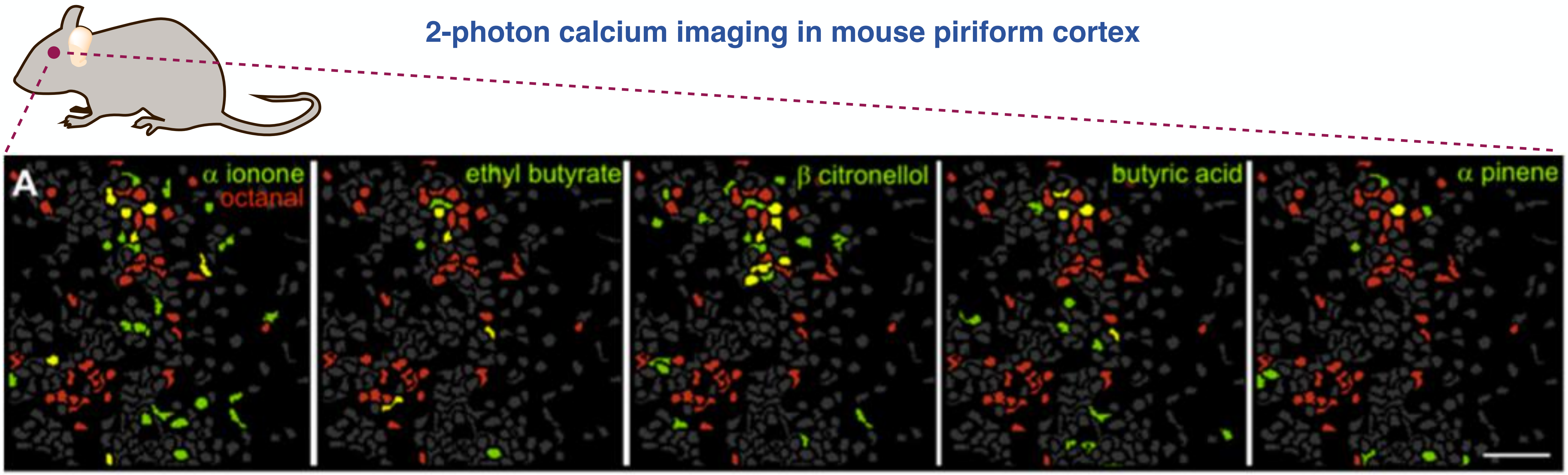
The Primary Olfactory Code

does this spatial and chemotropic organization persist in **higher processing centers**?

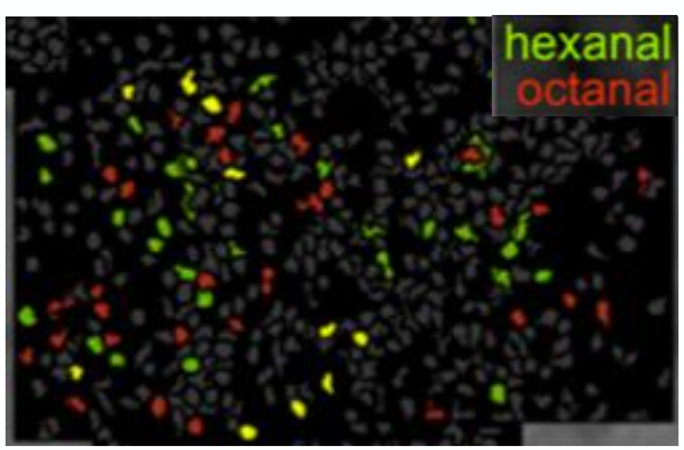


Mapping Odorant Response in Primary Olfactory Cortex

2-photon calcium imaging in mouse piriform cortex



even similar odorants have low overlap



ca. 26% vs. 70% in olfactory bulb

- no spatial clustering
- discontinuous receptive field (neurons respond to dissimilar odorants)
- *piriform cortex discards spatial segregation and chemotropy*
- odorants represented by *unique ensembles of piriform neurons*

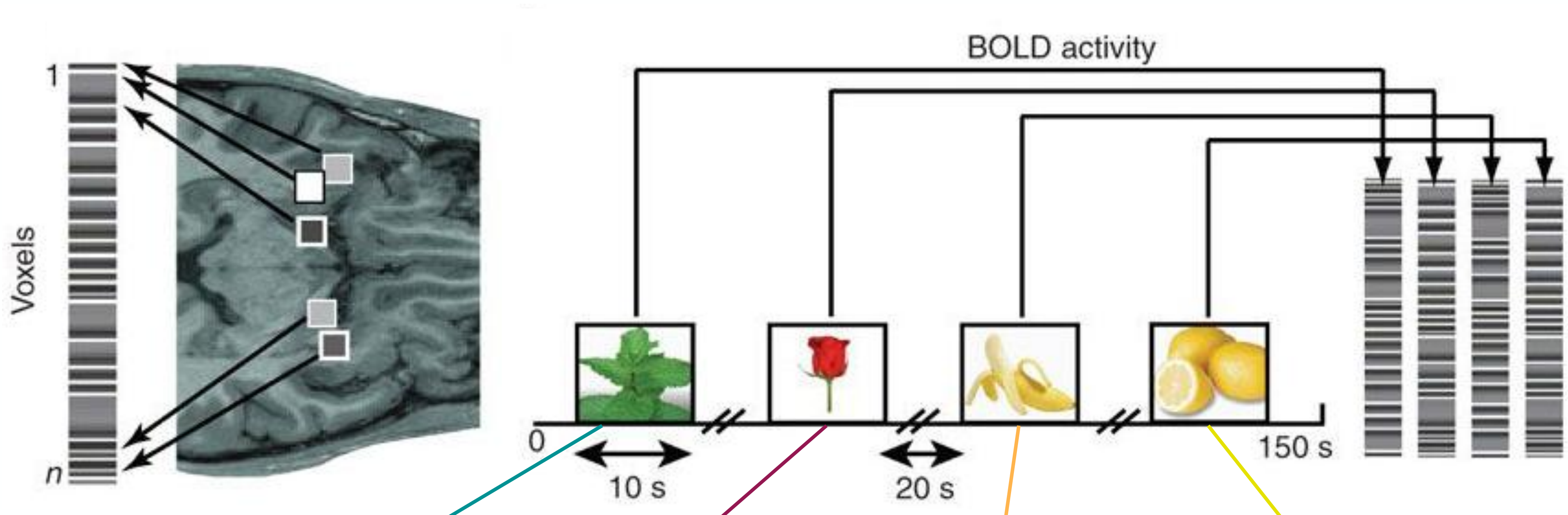
Stettler, D. D.; Axel, R. *Neuron* **2009**, *63*, 854.

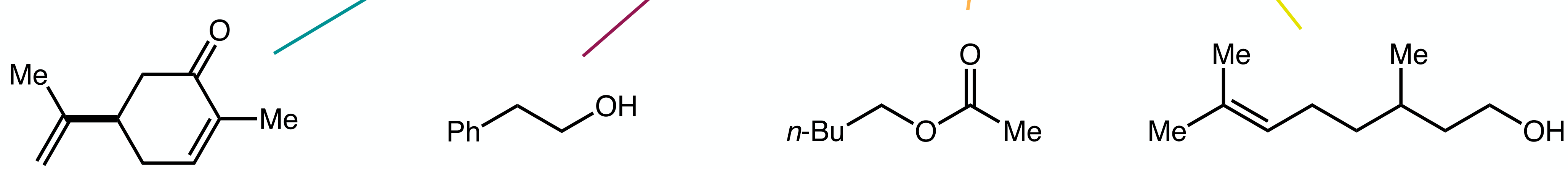
Mapping Odorant Response in Primary Olfactory Cortex

does patterned response in piriform cortex code for ***smell*** rather than ***structure*?**

Mapping Odorant Response in Primary Olfactory Cortex

functional magentic resonance imaging (fMRI) study in human cortex





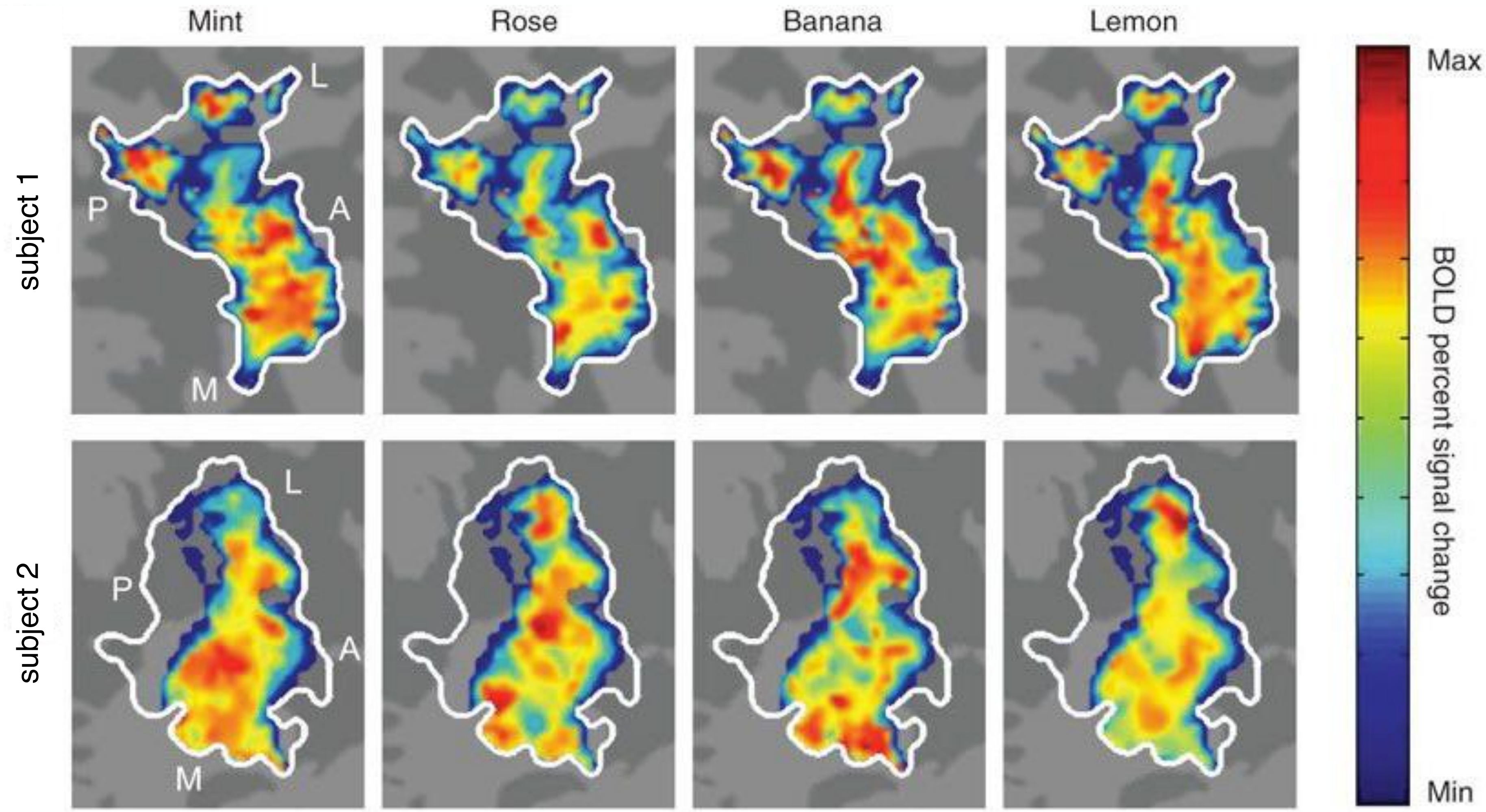
(–)-*R*-carvone phenethyl alcohol amyl acetate citronellol

- spatial patterns of voxel activity were measured using fMRI
- subjects exposed to individual odorants from 4 categories (mint, rose, banana, lemon)
- later, subjects exposed to 3 distinct odorants from 3 categories (mint, woody, citrus)

Howard, J. D.; Plailly, J.; Grueschow, M.; Haynes, J.-D.; Gottfried, J. A. *Nat. Neurosci.* **2009**, *12*, 932.

Mapping Odorant Response in Primary Olfactory Cortex

odorant-specific spatial maps in posterior piriform cortex (PPC)



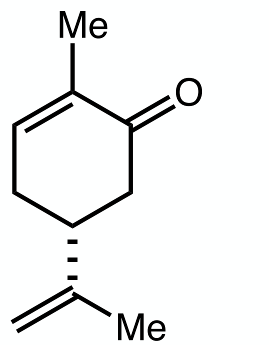
- pattern spatially distributed and unique for each odorant
- no local clustering
- no topographical consistency between subjects

Howard, J. D.; Plailly, J.; Grueschow, M.; Haynes, J.-D.; Gottfried, J. A. *Nat. Neurosci.* **2009**, *12*, 932.

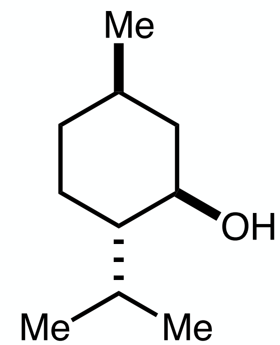
Mapping Odorant Response in Primary Olfactory Cortex

panel of odorants used to study olfactory 'object' recognition

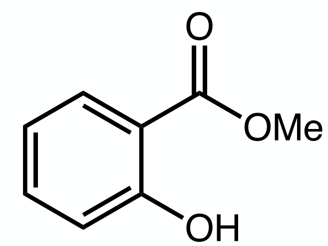
minty



(–)-*R*-carvone

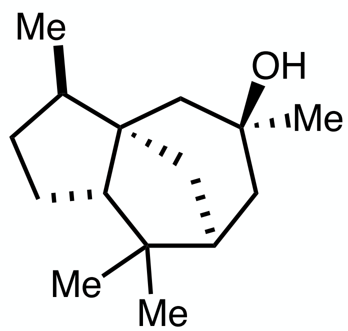


(–)-menthol

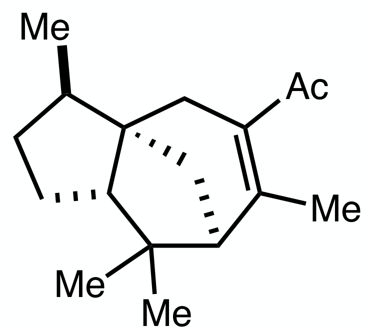


methyl salicylate

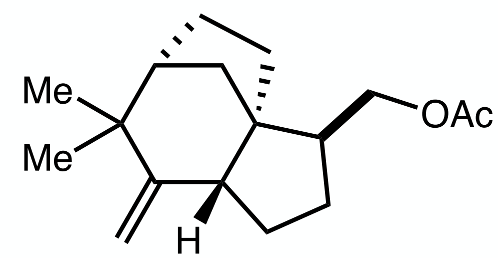
woody



(+)-cedrol

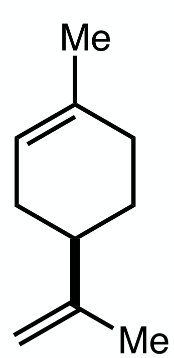


methyl cedryl ketone

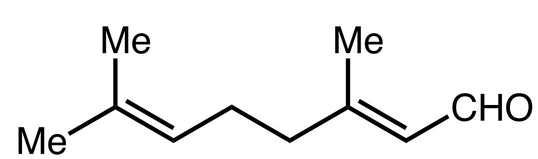


vetiver acetate

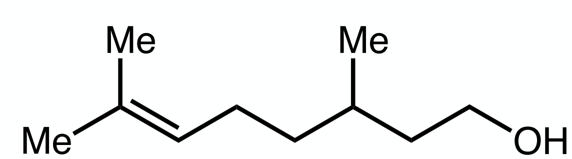
citrus



(+)-*R*-limonene



citral

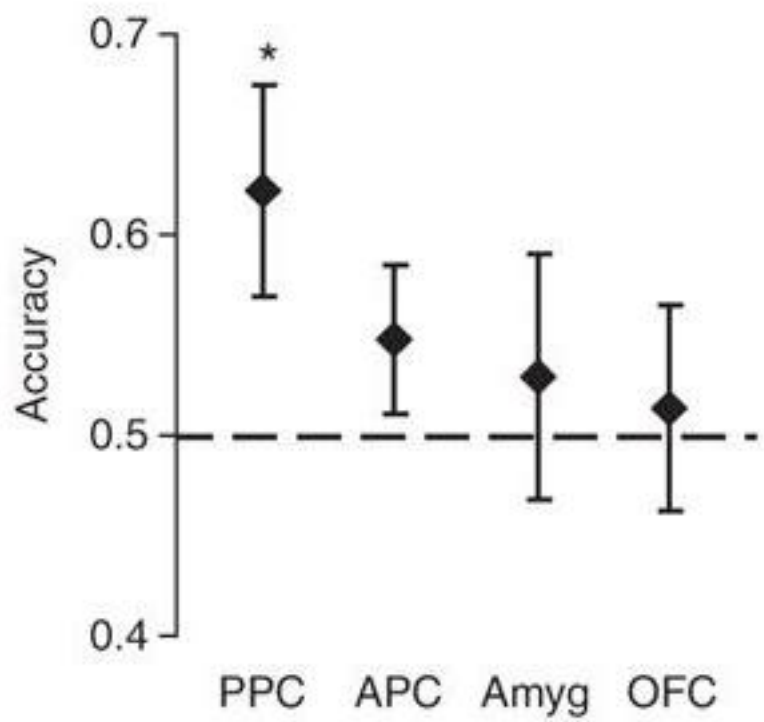


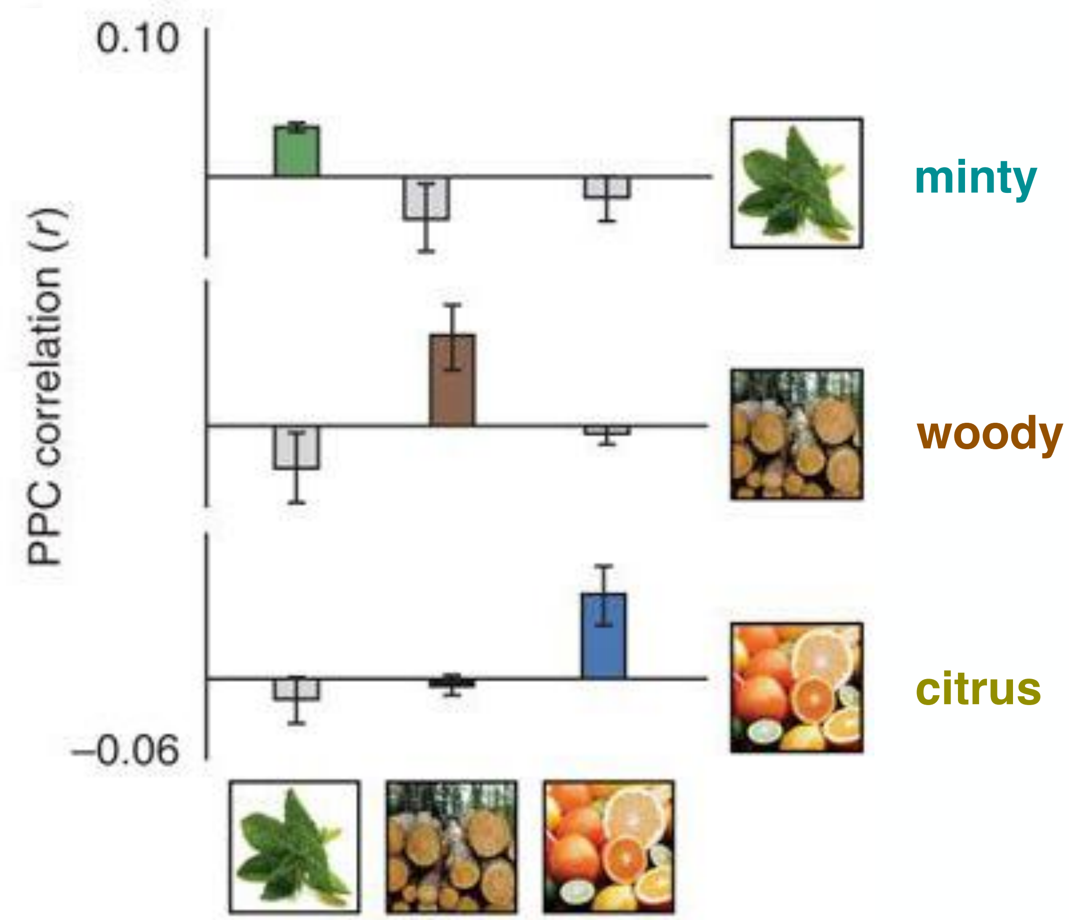
citronellol

Howard, J. D.; Plailly, J.; Grueschow, M.; Haynes, J.-D.; Gottfried, J. A. *Nat. Neurosci.* **2009**, *12*, 932.

Mapping Odorant Response in Primary Olfactory Cortex

fMRI patterns in posterior piriform cortex (PPC) match across related smells





- strong pattern correlation between similar-smelling odorants
- PPC patterns are predictive of *smell* rather than *structure*
- demonstrates that olfaction performs feature extraction and object synthesis

Howard, J. D.; Plailly, J.; Grueschow, M.; Haynes, J.-D.; Gottfried, J. A. *Nat. Neurosci.* **2009**, *12*, 932.

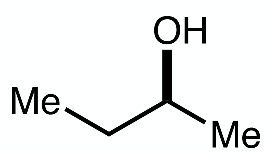
Mapping Odorant Response in Primary Olfactory Cortex

how plastic are posterior piriform cortex olfactory object representations?

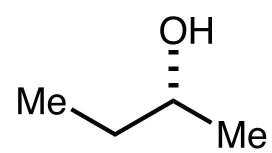
Plasticity of Odorant Response in Primary Olfactory Cortex

can traditional (Pavlovian) conditioning train the PPR to distinguish stereoisomers?

“oily-vinous”

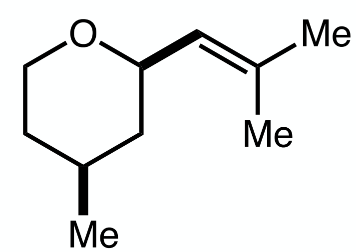


(*S*)-2-butanol

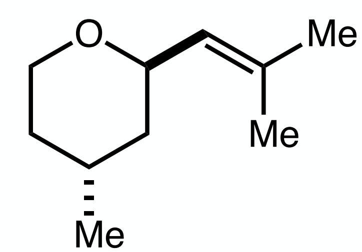


(*R*)-2-butanol

“green”



(–)-rose oxide



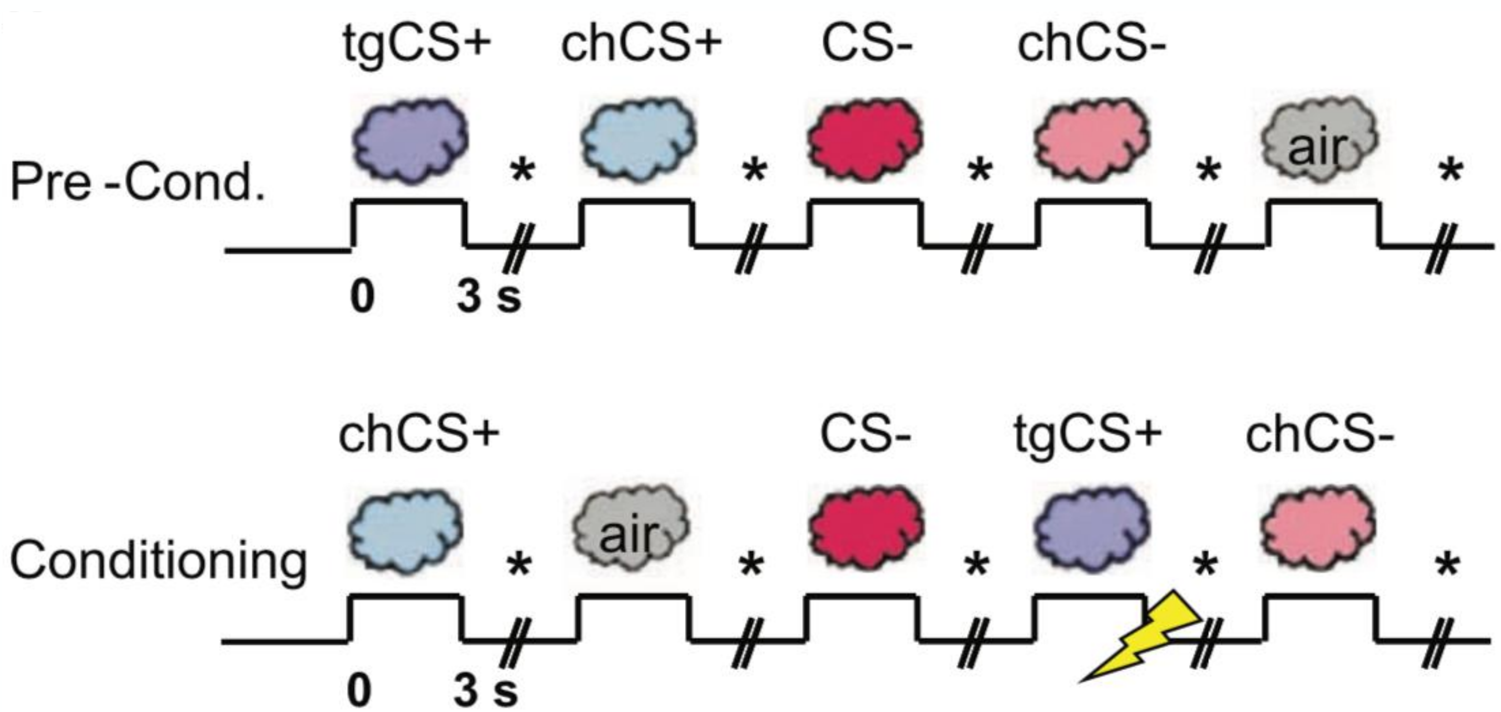
(+)-rose oxide

initially indistinguishable stereiosmer pairs examined

Li, W.; Howard, J. D.; Parrish, T. B.; Gottfried, J. A. *Science.* **2008**, *319*, 1842.

Plasticity of Odorant Response in Primary Olfactory Cortex

experimental paradigm / learning task

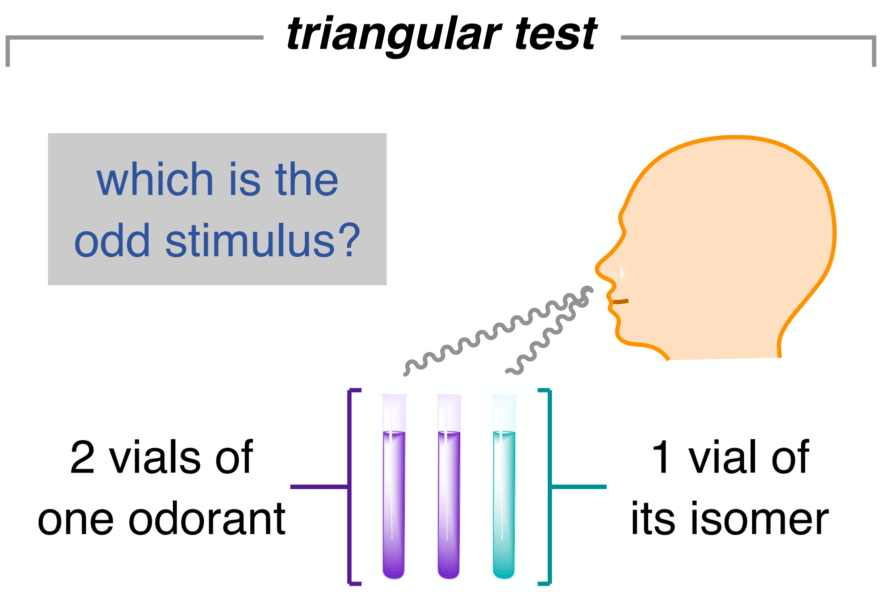


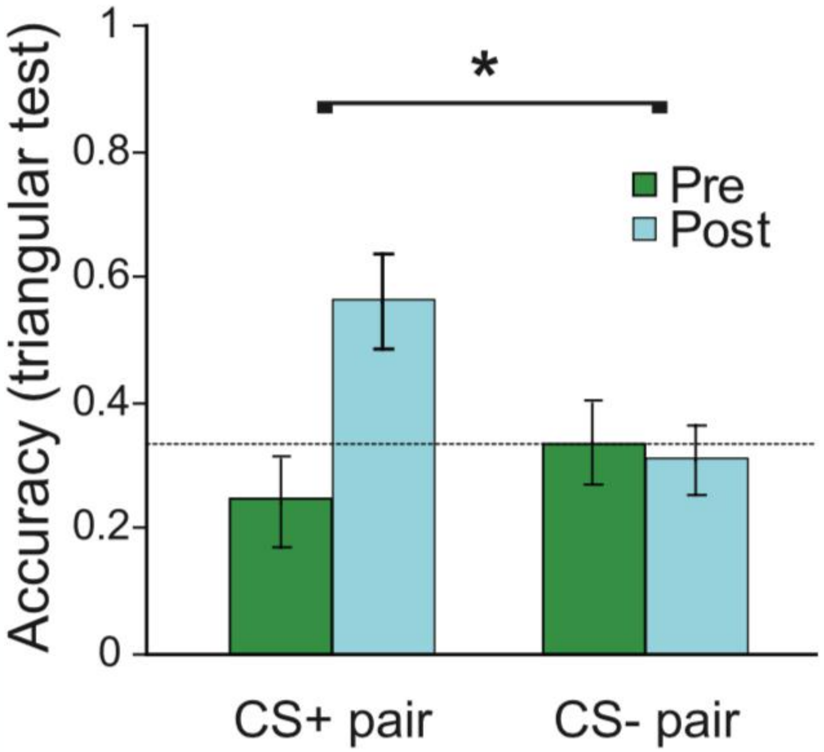
tgCS+	(*S*)-2-butanol
chCS+	(*R*)-2-butanol
CS–	(–)-rose oxide
chCS–	(+)-rose oxide

- tgCS+ co-administered with electric shock for aversive conditioning
- CS– and chCS– serve as controls

Li, W.; Howard, J. D.; Parrish, T. B.; Gottfried, J. A. *Science.* **2008**, *319*, 1842.

Plasticity of Odorant Response in Primary Olfactory Cortex

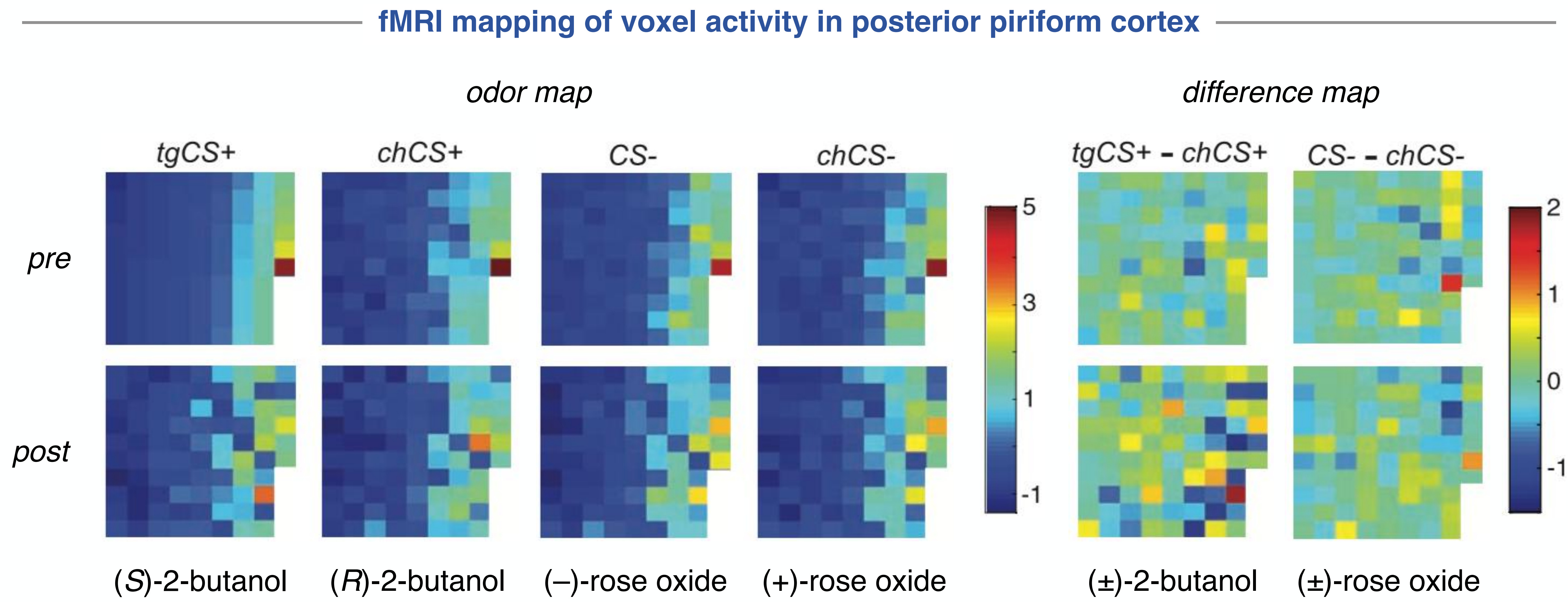




- subjects conditioned for *7 minutes*
- subjects gained the ability to discriminate tgCS+ [(*S*)-2-butanol] from chCS+ [(*R*)-2-butanol]
- control CS– pair (±)-rose oxide remained indistinguishable

Li, W.; Howard, J. D.; Parrish, T. B.; Gottfried, J. A. *Science.* **2008**, *319*, 1842.

Plasticity of Odorant Response in Primary Olfactory Cortex



- activity patterns selectively reorganized for CS+ pair
- activity patterns remained highly coupled for CS– pair
- conditioning alters the *sensory processing of the odorant itself* (not just behavioral output)

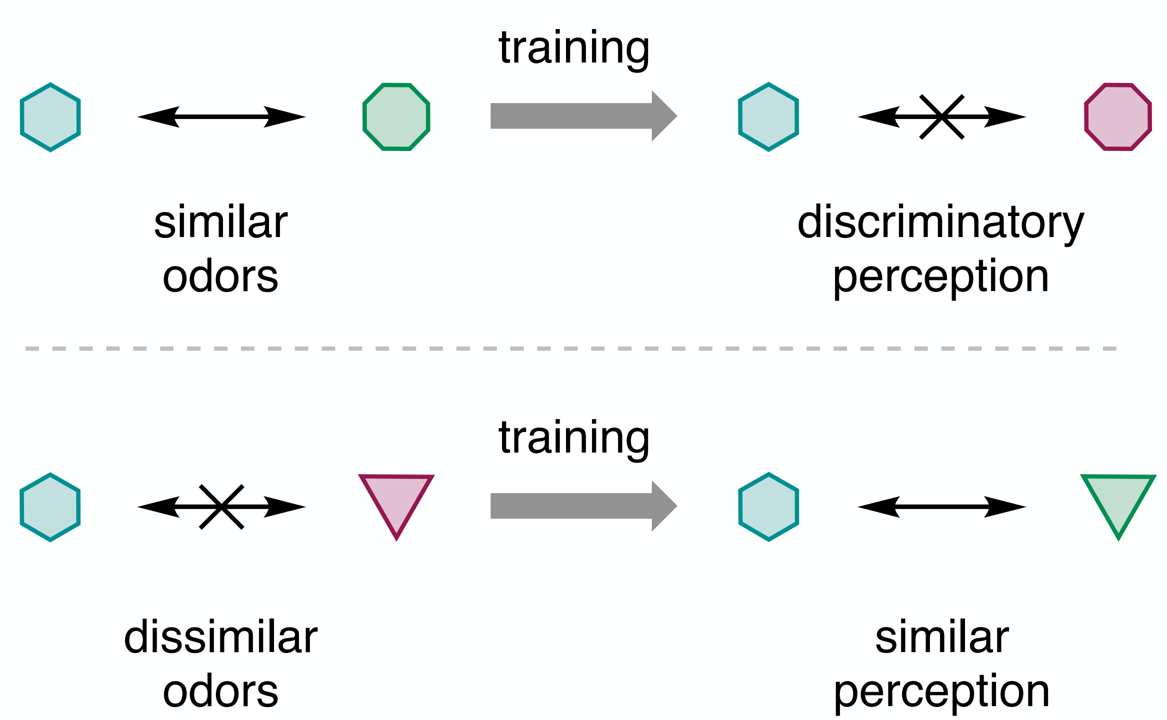
Li, W.; Howard, J. D.; Parrish, T. B.; Gottfried, J. A. *Science.* **2008**, *319*, 1842.

Plasticity of Odorant Response in Primary Olfactory Cortex

further questions

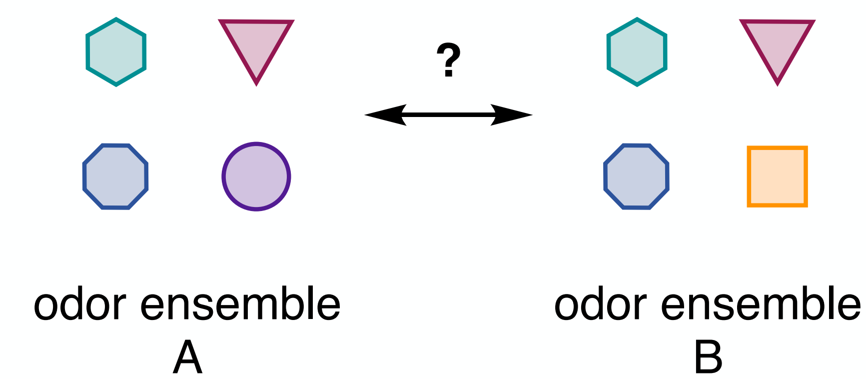
- 1. Can plasticity be bidirectional (discriminatory and nondiscriminatory)?
- 2. Can the olfactory cortex perform pattern completion?

bidirectional plasticity



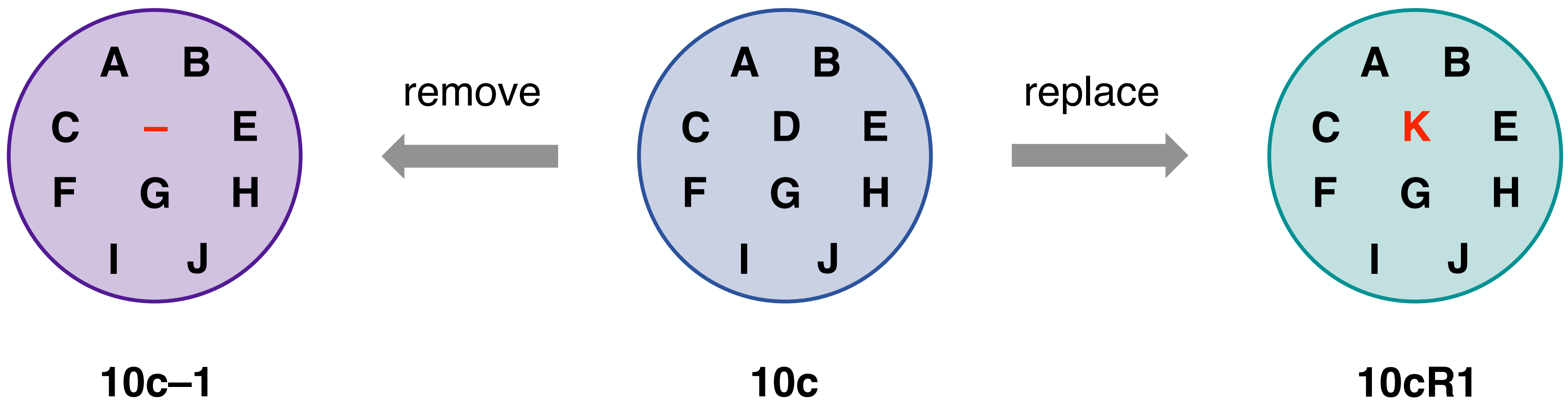
pattern recognition

In tihs scnetnee, mnay of the ltetres in wdros hvae been jbmueld. Hevewor, it is sltil rtaleivley esay to raed bausece the barin rogicsenes penttras.

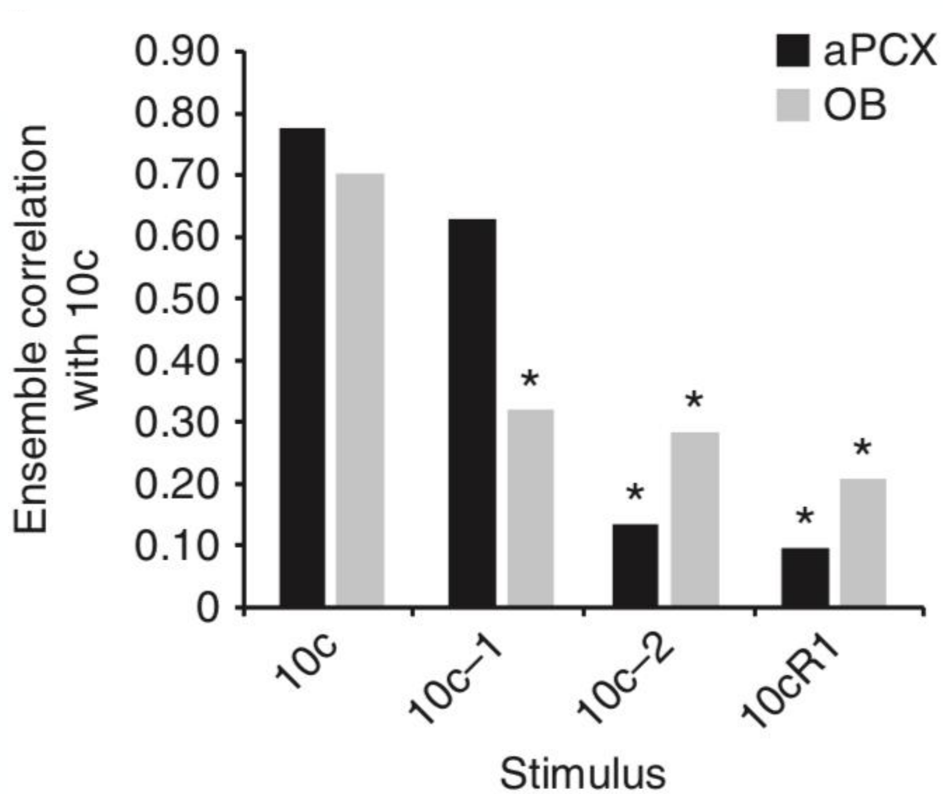


Plasticity of Odorant Response in Primary Olfactory Cortex

odorant mixtures studied as stimuli for plasticity



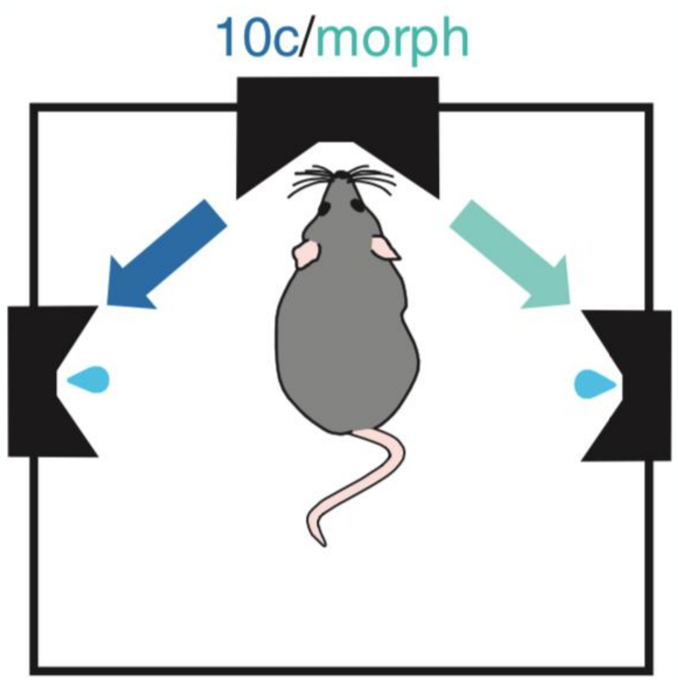
initial mapping of olfactory bulb (OB) and anterior piriform cortex (aPCX) correlations



- OB easily differentiates all mixtures
- aPCX cannot differentiate 10c and 10c–1

Chapuis, J.; Wilson, D. A. *Nat. Neurosci.* **2012**, *15*, 155.

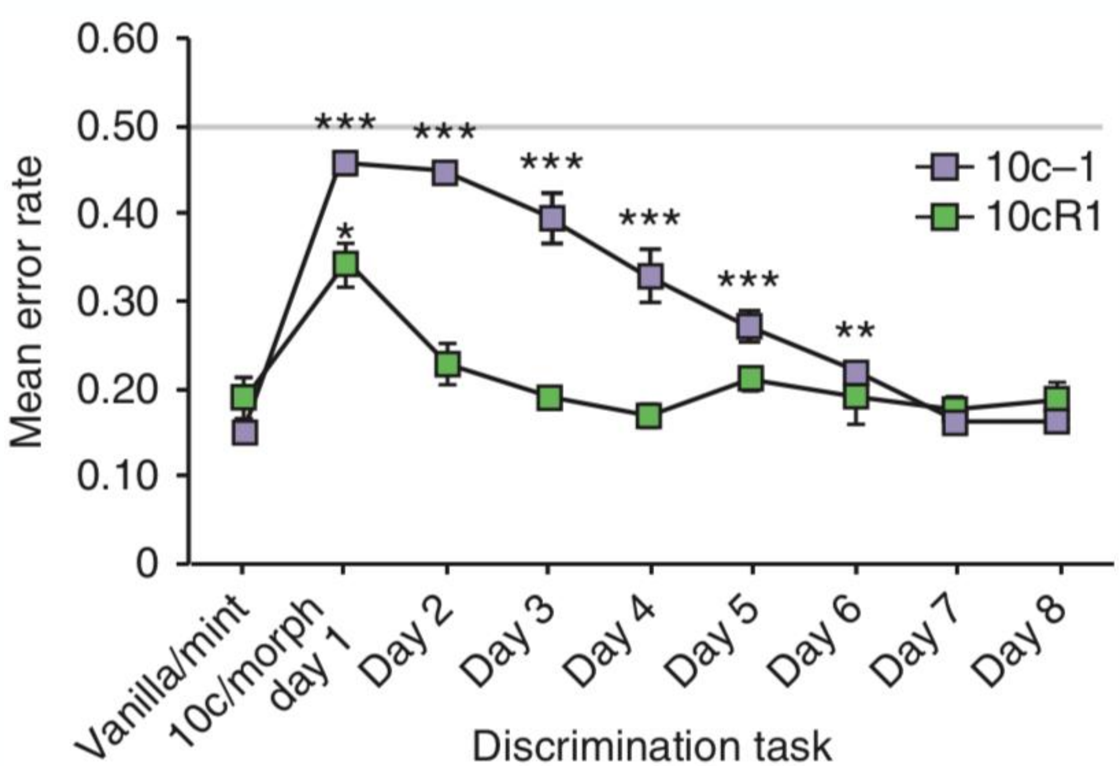
Plasticity of Odorant Response in Primary Olfactory Cortex



training paradigm

- two-alternative forced-choice task
- stimulus paired with left or right reward (water)
- error rate evaluted
- 50% = chance, negative deviation implies discrimination

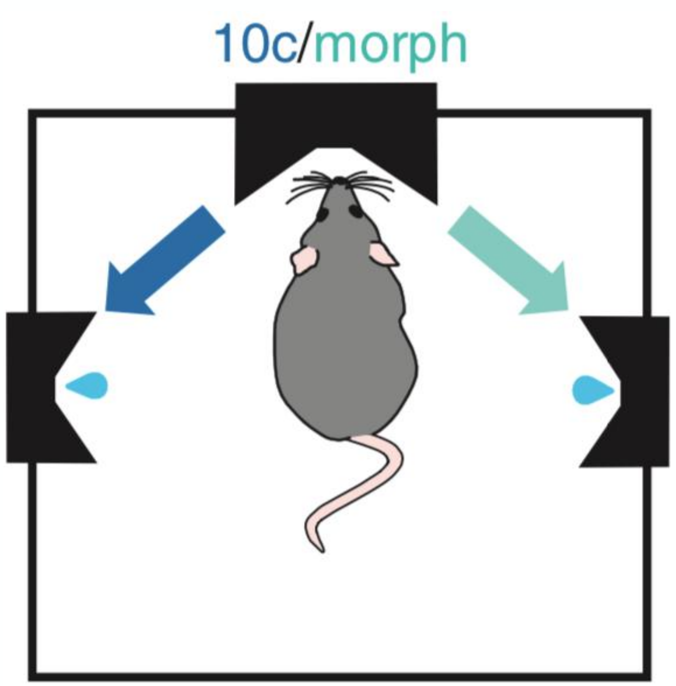
learned enhancement in sensory acuity (discrimination)



- rats quickly learn to discriminate 10cR1 (“easy”)
- rats slowly learn to discriminate 10c–1 (“difficult”)
- learned acuity maintained over >2 weeks

Chapuis, J.; Wilson, D. A. *Nat. Neurosci.* **2012**, *15*, 155.

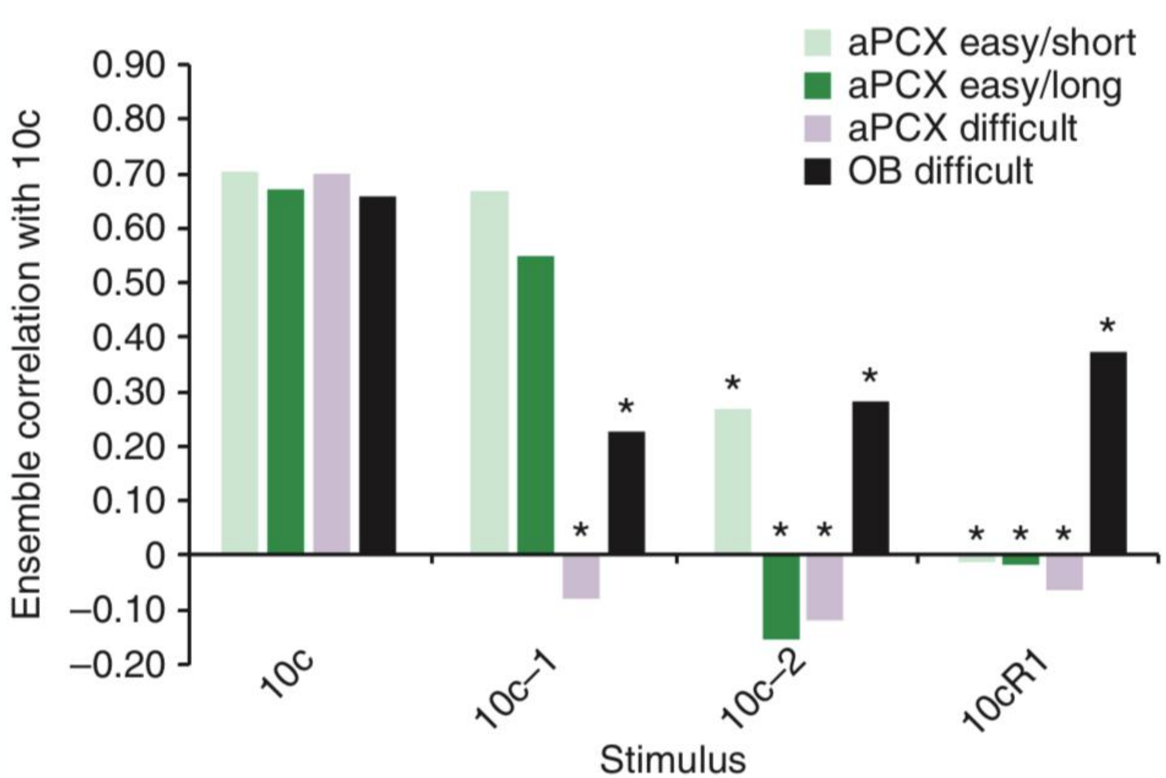
Plasticity of Odorant Response in Primary Olfactory Cortex



training paradigm

- two-alternative forced-choice task
- stimulus paired with left or right reward (water)
- error rate evaluted
- 50% = chance, negative deviation implies discrimination

learned enhancement in sensory acuity (discrimination)

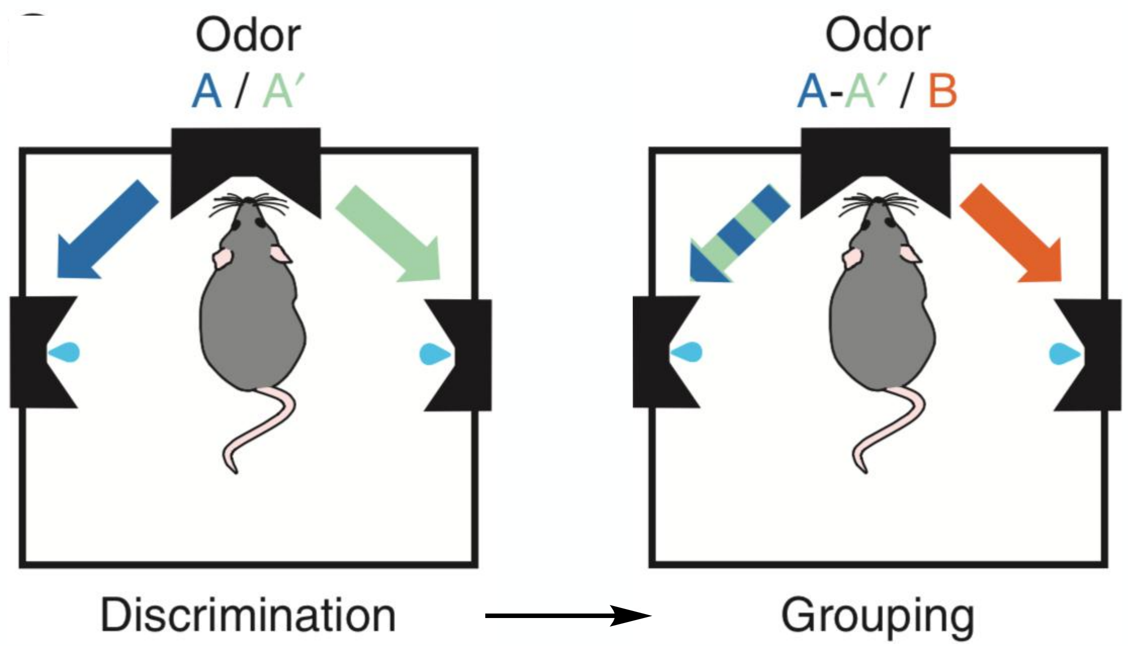


- aPCX ensembles enhance pattern separation ability
- only “difficult” conditioning exhibits significant change
- OB ensembles unchanged vs naive rats

Chapuis, J.; Wilson, D. A. *Nat. Neurosci.* **2012**, *15*, 155.

Plasticity of Odorant Response in Primary Olfactory Cortex

training paradigm



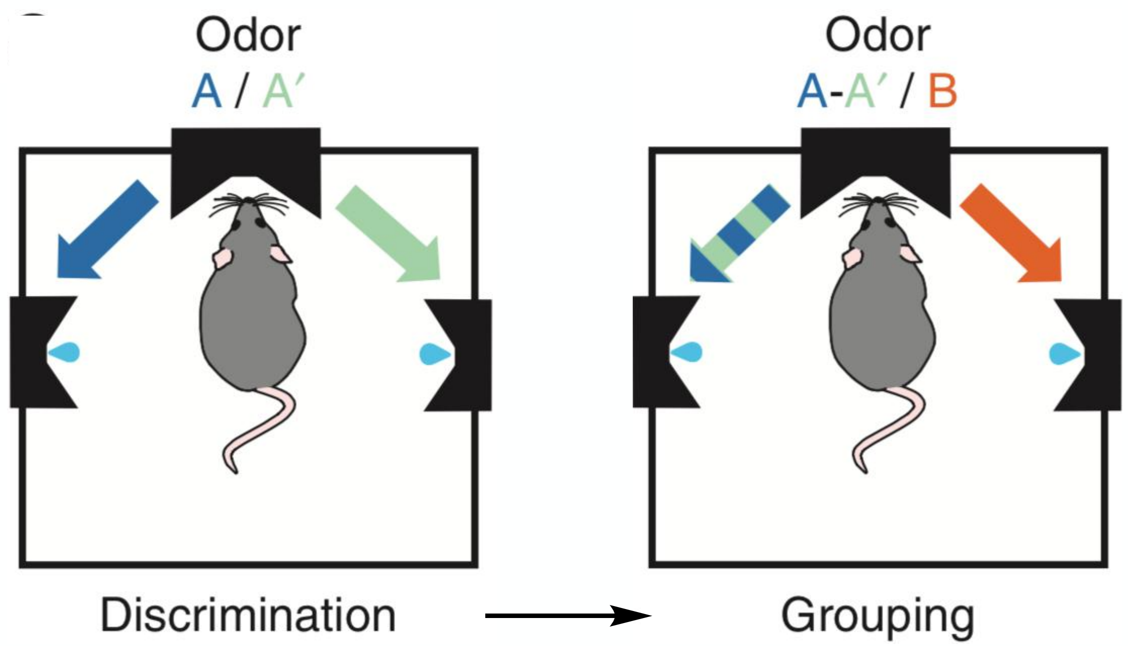
- rats trained first to differentiate two stimuli (A v A′)
- rats then trained to group two stimuli (A & A′)
- ‘close’: A = 10c, A′ = 10cR1
- ‘distant’: A = 10c, A′ = limonene

can rats learn to impair olfactory cortical acuity by enhancing pattern completion?

Chapuis, J.; Wilson, D. A. *Nat. Neurosci.* **2012**, *15*, 155.

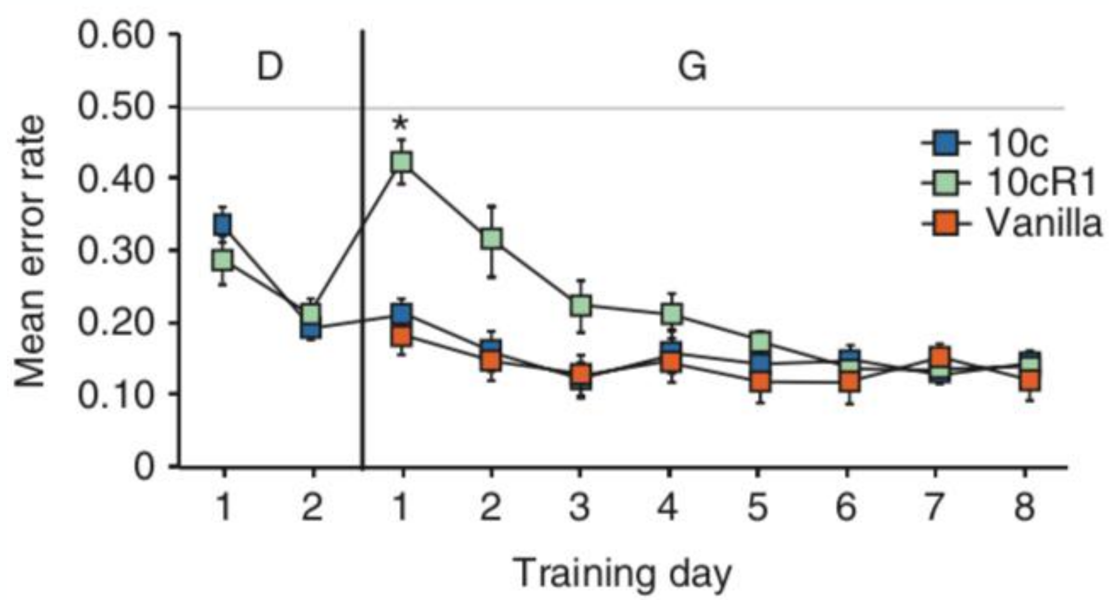
Plasticity of Odorant Response in Primary Olfactory Cortex

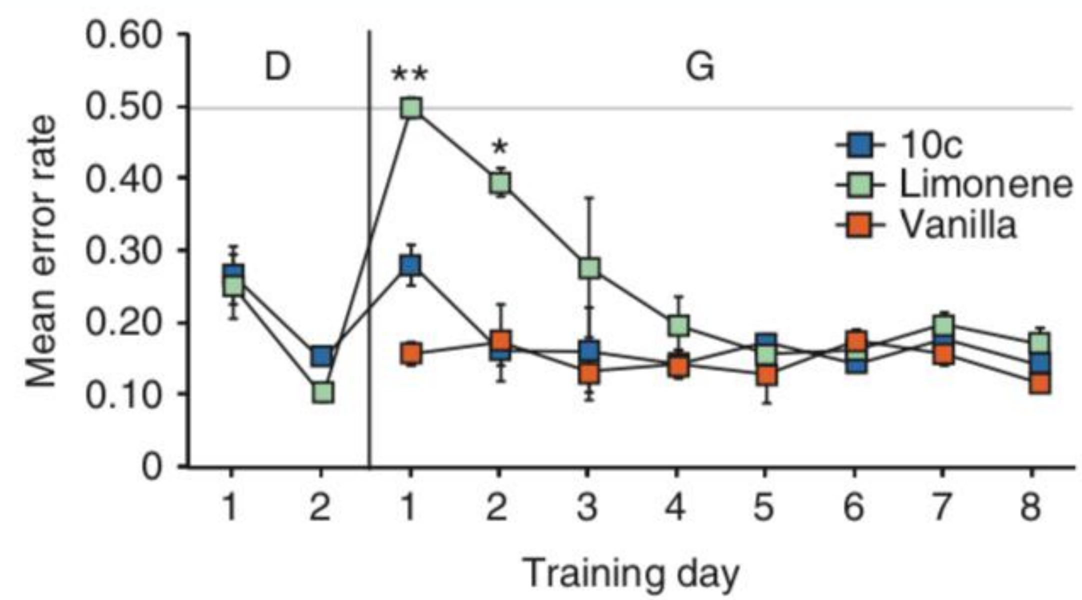
training paradigm



- rats trained first to differentiate two stimuli (A v A′)
- rats then trained to group two stimuli (A & A′)
- ‘close’: A = 10c, A′ = 10cR1
- ‘distant’: A = 10c, A′ = limonene

behavioral performance



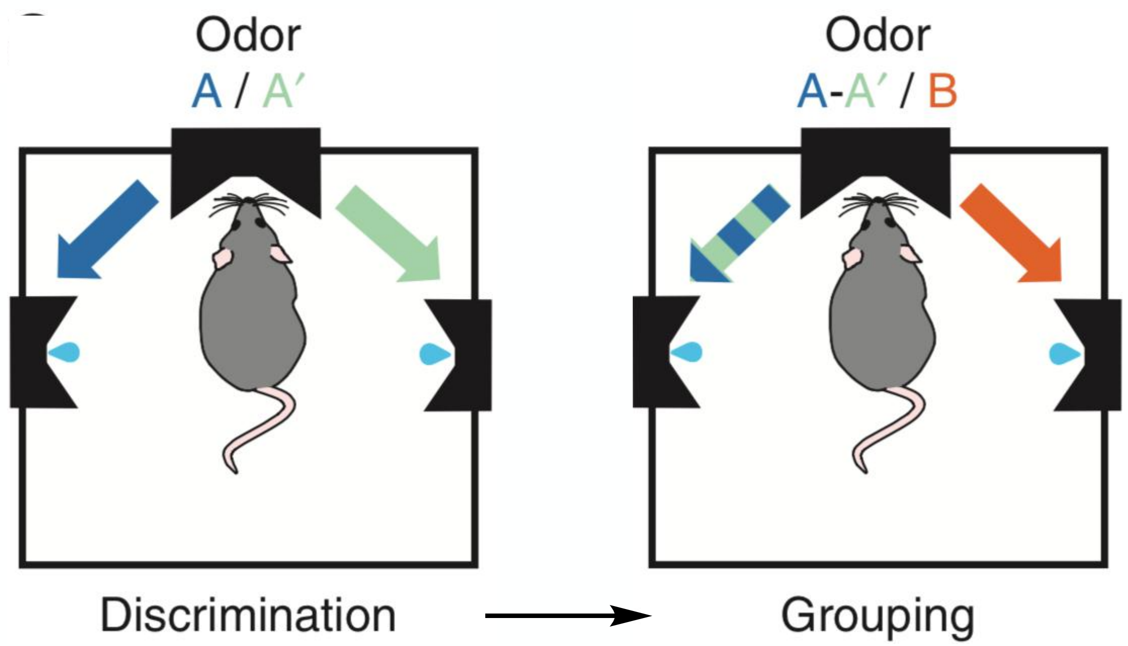


- rats successfully differentiate and then group both sets of stimuli

Chapuis, J.; Wilson, D. A. *Nat. Neurosci.* **2012**, *15*, 155.

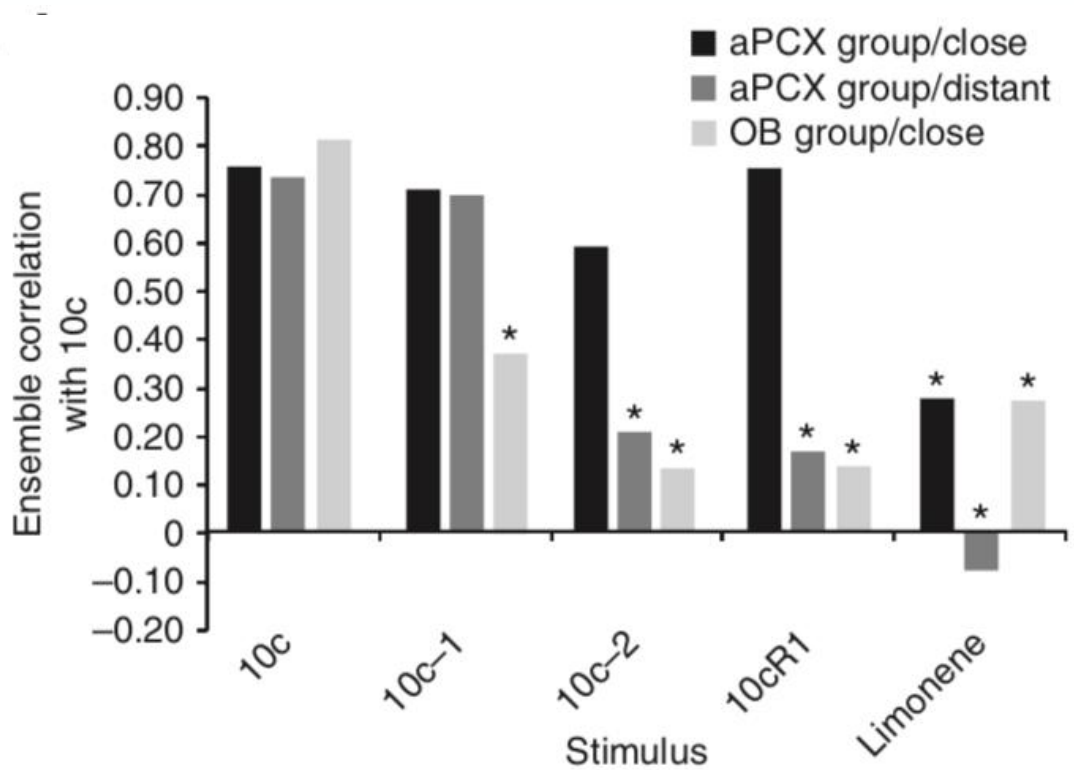
Plasticity of Odorant Response in Primary Olfactory Cortex

training paradigm



- rats trained first to differentiate two stimuli (A v A′)
- rats then trained to group two stimuli (A & A′)
- ‘close’: A = 10c, A′ = 10cR1
- ‘distant’: A = 10c, A′ = limonene

aPCX cell ensembles strongly affected in a task-specific manner



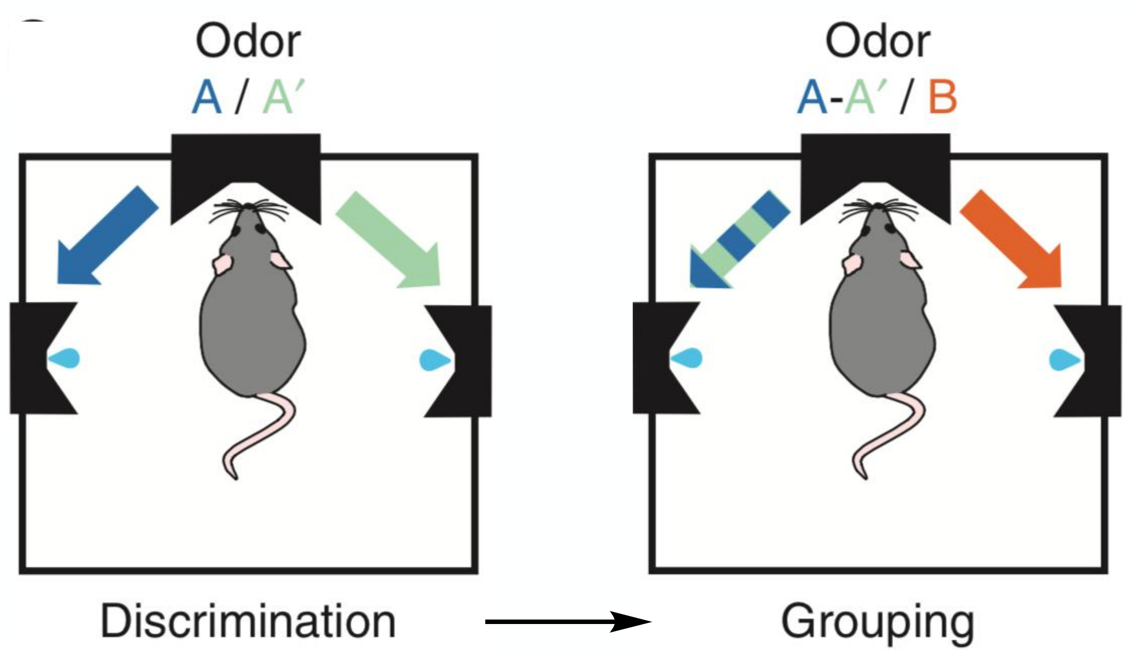
‘close’ conditioning

- enhanced pattern completion for *all transformed versions of 10c*
- aPCX merged representations of formerly discriminable mixtures
- pattern completion limited to similar mixtures (i.e., not limonene)

Chapuis, J.; Wilson, D. A. *Nat. Neurosci.* **2012**, *15*, 155.

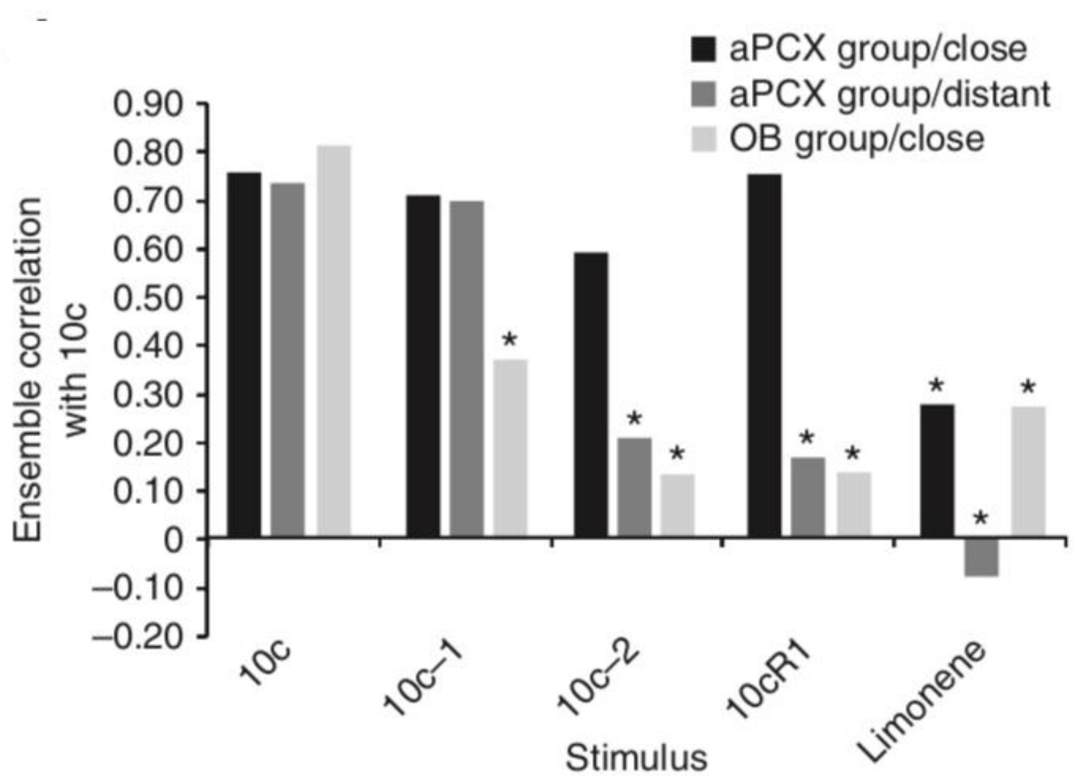
Plasticity of Odorant Response in Primary Olfactory Cortex

training paradigm



- rats trained first to differentiate two stimuli (A v A′)
- rats then trained to group two stimuli (A & A′)
- ‘close’: A = 10c, A′ = 10cR1
- ‘distant’: A = 10c, A′ = limonene

aPCX cell ensembles strongly affected in a task-specific manner



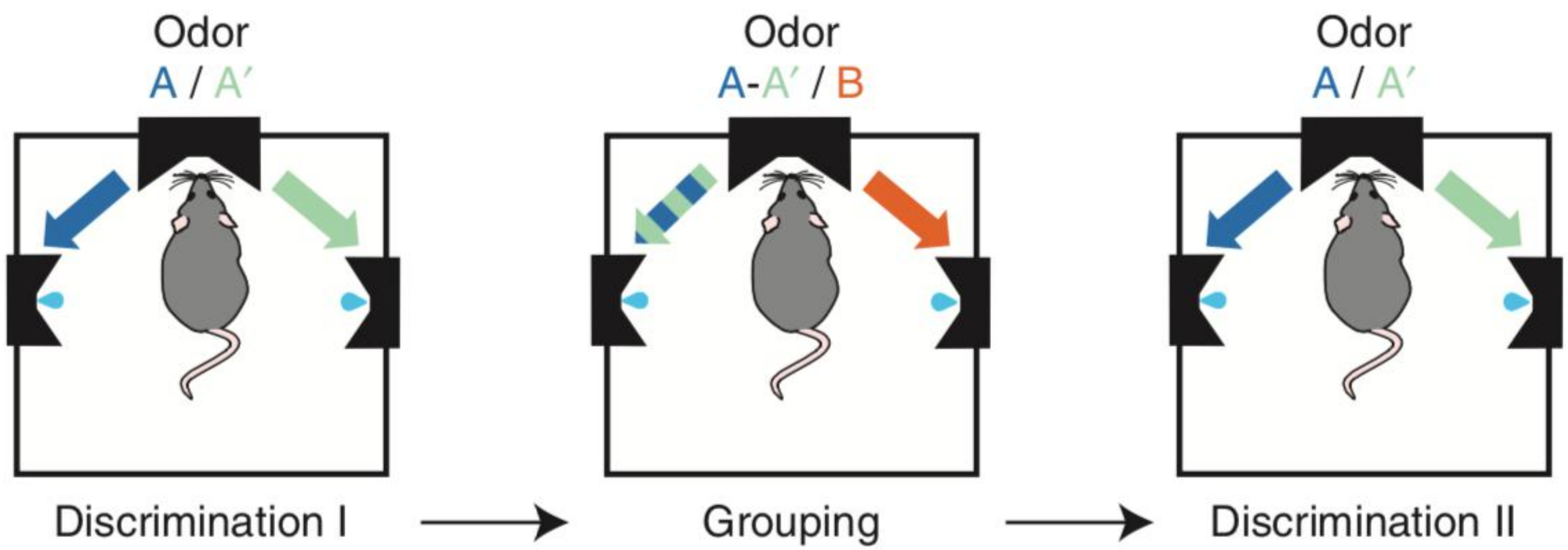
‘distant’ conditioning

- does not entrain pattern completion
- either A or A′ signal reward rather than merged representation
- 10c variants retain aPCX ensembles of naive rats

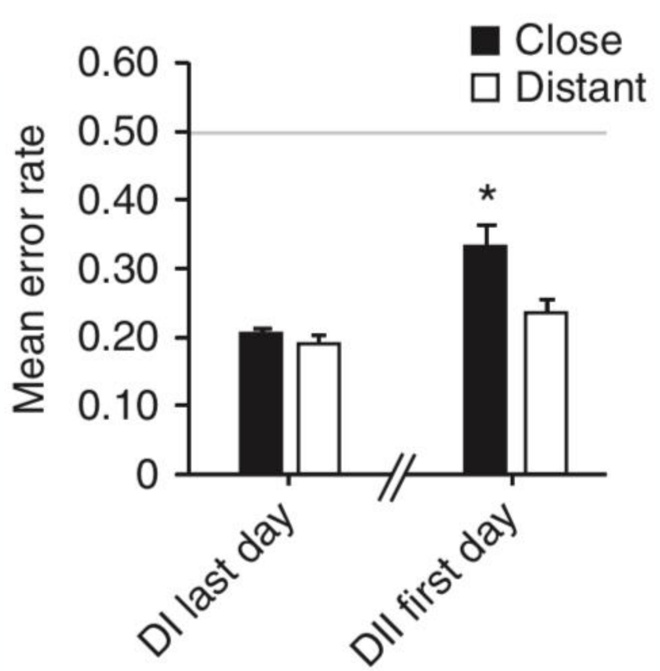
Chapuis, J.; Wilson, D. A. *Nat. Neurosci.* **2012**, *15*, 155.

Plasticity of Odorant Response in Primary Olfactory Cortex

training paradigm



rats challenged to differentiate two stimuli (A & A′) after grouping



- entrained pattern completion leads rats to treat 10c and 10cR1 as the *same odor object*
- control ('distant') training does not impact performance

Chapuis, J.; Wilson, D. A. *Nat. Neurosci.* **2012**, *15*, 155.

Plasticity of Odorant Response in Primary Olfactory Cortex

how does long-term learning affect olfactory cortex?

perfumer student[a]

cortical changes?

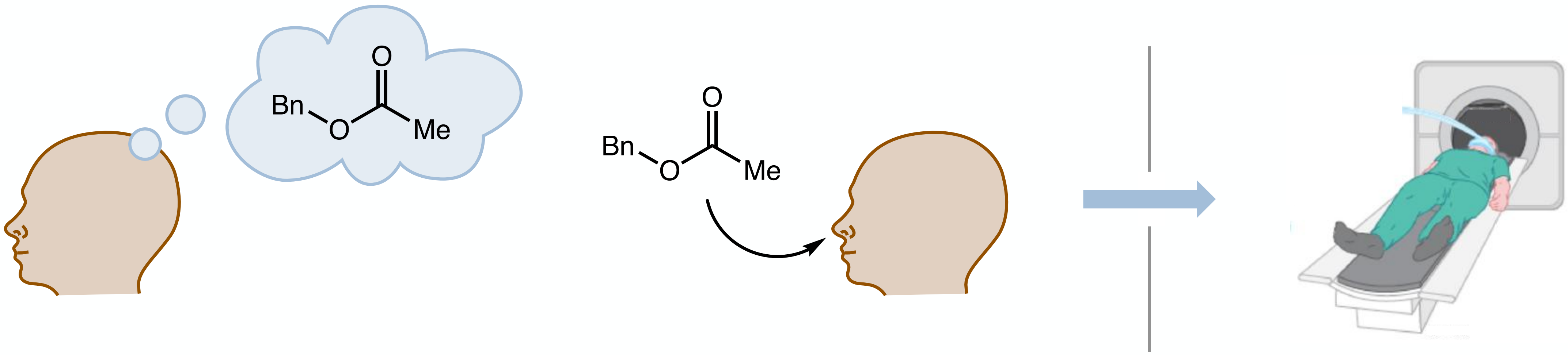
master perfumer[b]

[a]at Givaudan's Perfumery School | [b]Rosendo Mateu

Plailly, J.; Delon-Martin, C.; Royet, J.-P.. *Hum. Brain Mapp.* **2012**, *33*, 224.

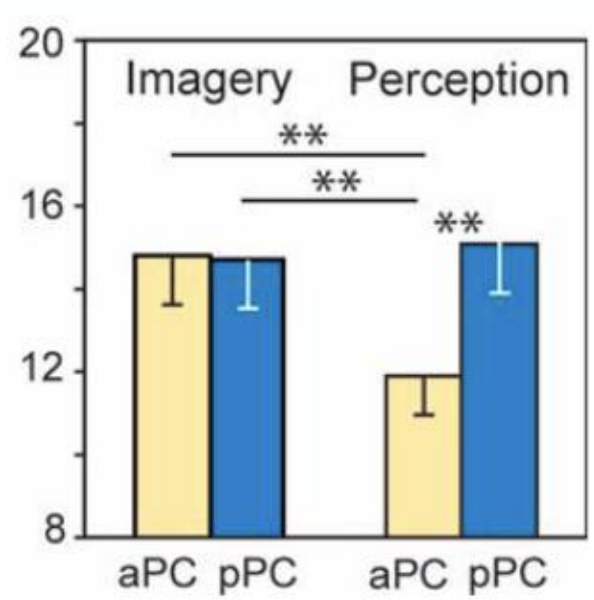
Plasticity of Odorant Response in Primary Olfactory Cortex

experimental design



olfactory imagery *olfactory perception* *both analyzed by fMRI*

olfactory imagination vs perception

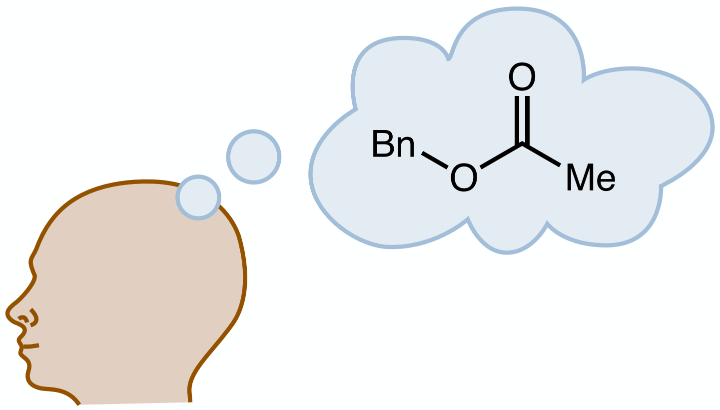


- mental imagery can induce activation of the primary olfactory cortex
- activity patterns remarkably similar in posterior piriform cortex

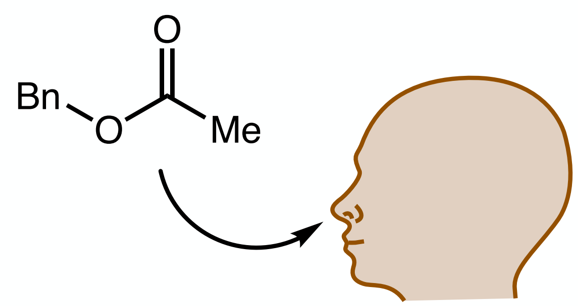
Plailly, J.; Delon-Martin, C.; Royet, J.-P.. *Hum. Brain Mapp.* **2012**, *33*, 224.

Plasticity of Odorant Response in Primary Olfactory Cortex

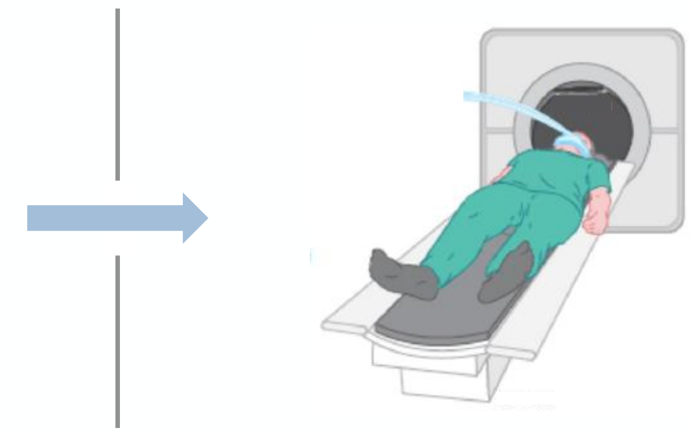
experimental design



olfactory imagery

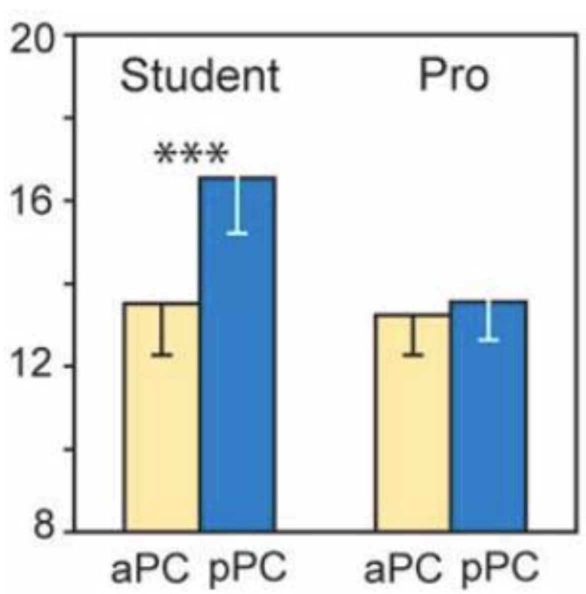


olfactory perception



both analyzed by fMRI

student vs professional perfumer in olfactory imagination

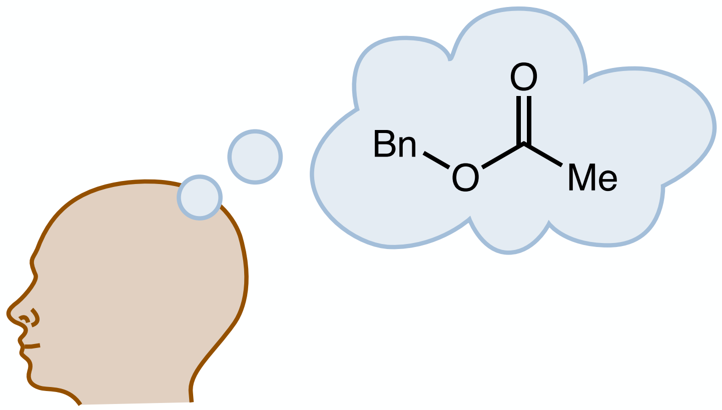


- similar overall activation in aPC
- markedly decreased activation in pPC for professional

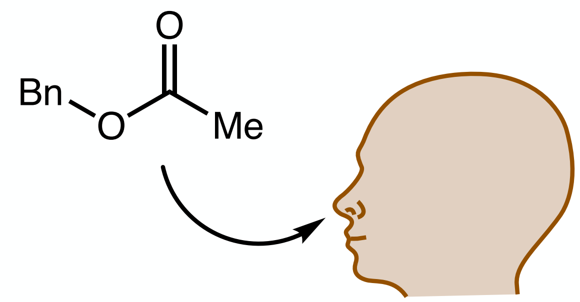
Plailly, J.; Delon-Martin, C.; Royet, J.-P.. *Hum. Brain Mapp.* **2012**, *33*, 224.

Plasticity of Odorant Response in Primary Olfactory Cortex

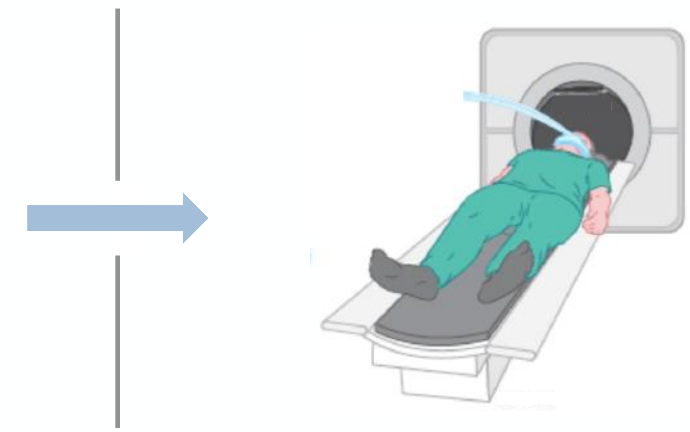
experimental design



olfactory imagery



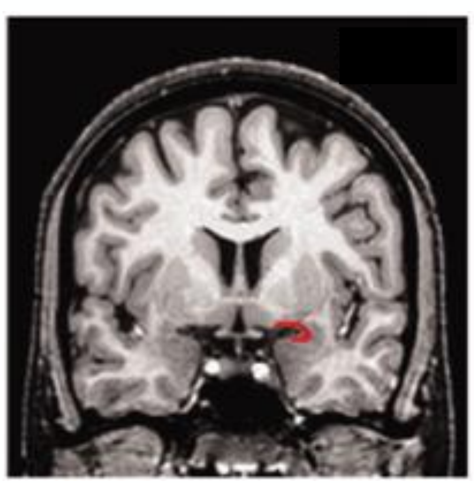
olfactory perception

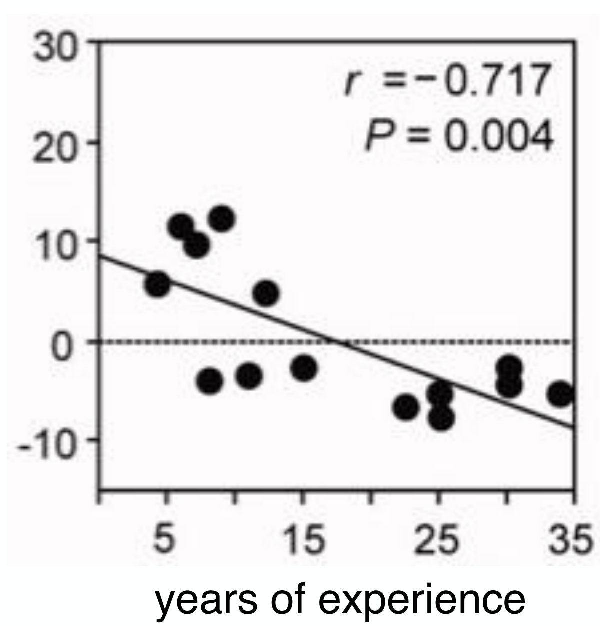


both analyzed by fMRI

changes with experience for professional perfumers in olfactory imagination

piriform cortex activation

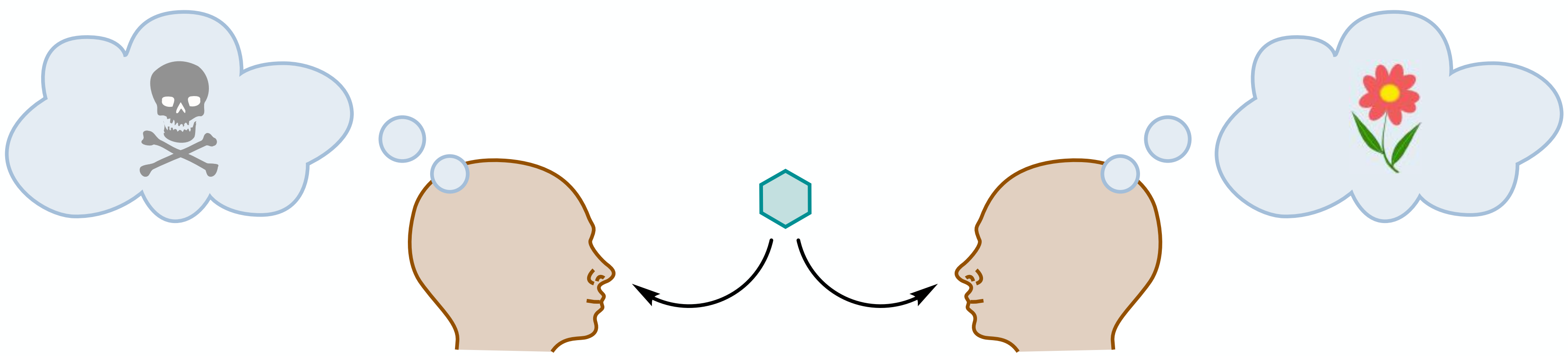




- negative correlation between experience and PC activation
- also observed in hippocampus
- experts develop more efficient neural strategies

Plailly, J.; Delon-Martin, C.; Royet, J.-P.. *Hum. Brain Mapp.* **2012**, *33*, 224.

Subjectivity of Odor



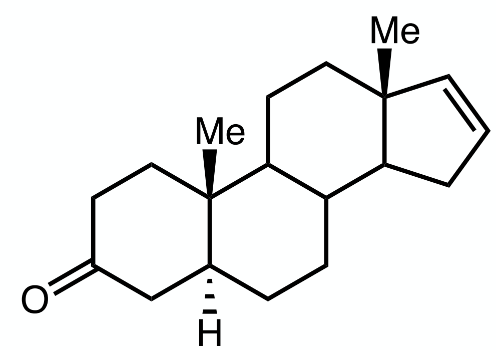
what affects changes in olfactory perception between individuals?

- 1) experience and learning (neural plasticity)
- 2) expectancy (pyschological)
- 3) environmental factors (e.g., humidity)
- 4) nasal metabolism (perireceptor events)
- **5) genetics?**

Subjectivity of Odor

case study on androstenone

sweet, floral



sweaty, urinous

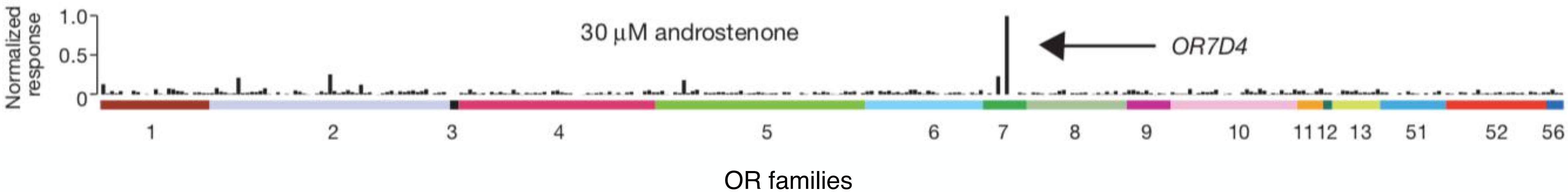
odorless

- odorous steriod derived from testosterone
- perceived with wide variation between individuals
- does such dramatic perceptional variation have genetic underpinnings?

Keller, A.; Zhuang, H.; Chi, Q.; Vosshall, L. B.; Matsunami, H. *Nature.* **2007**, *449*, 468.

Subjectivity of Odor

selective activation of OR7D4 by androstenone

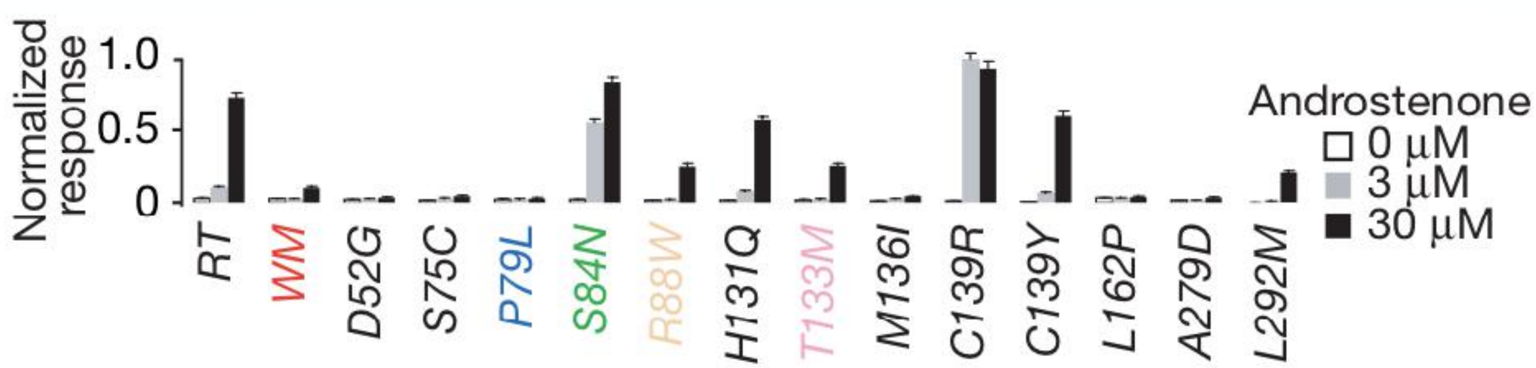


in vitro luciferase assay identified OR7D4 as a selective OR for androstenone

common SNPs for OR7D4

Residue change	Allele frequency
refseq	0.786
P79L	0.040
S84N	0.013
R88W	0.157
T133M	0.157

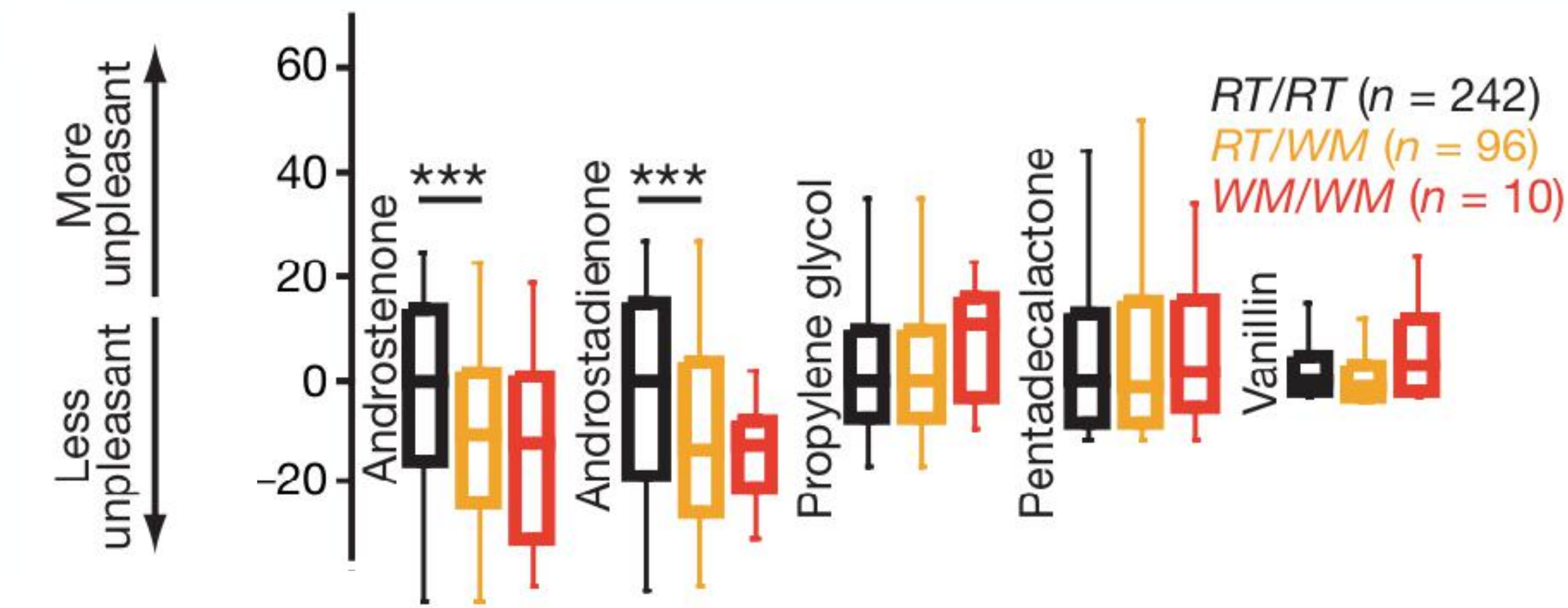
- OR7D4 has several common single-nucleotide polymorphisms (SNPs)
- wildtype = “RT”; common double SNP (R88W, T133M) = “WM”
- androstenone response by WM severely diminished



Keller, A.; Zhuang, H.; Chi, Q.; Vosshall, L. B.; Matsunami, H. *Nature.* **2007**, *449*, 468.

Subjectivity of Odor

perceptual differences in groups carrying OR7D4 SNPs

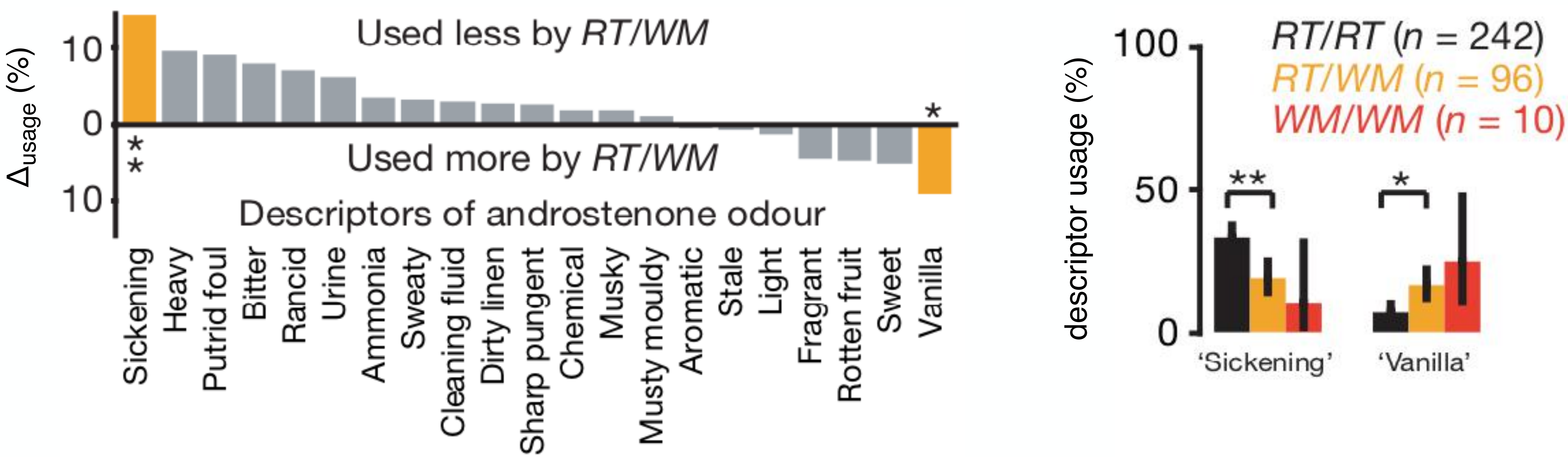


- RT/WM and WM/WM groups rated androstenone as less unpleasant than RT/RT

Keller, A.; Zhuang, H.; Chi, Q.; Vosshall, L. B.; Matsunami, H. *Nature.* **2007**, *449*, 468.

Subjectivity of Odor

perceptual differences in groups carrying OR7D4 SNPs



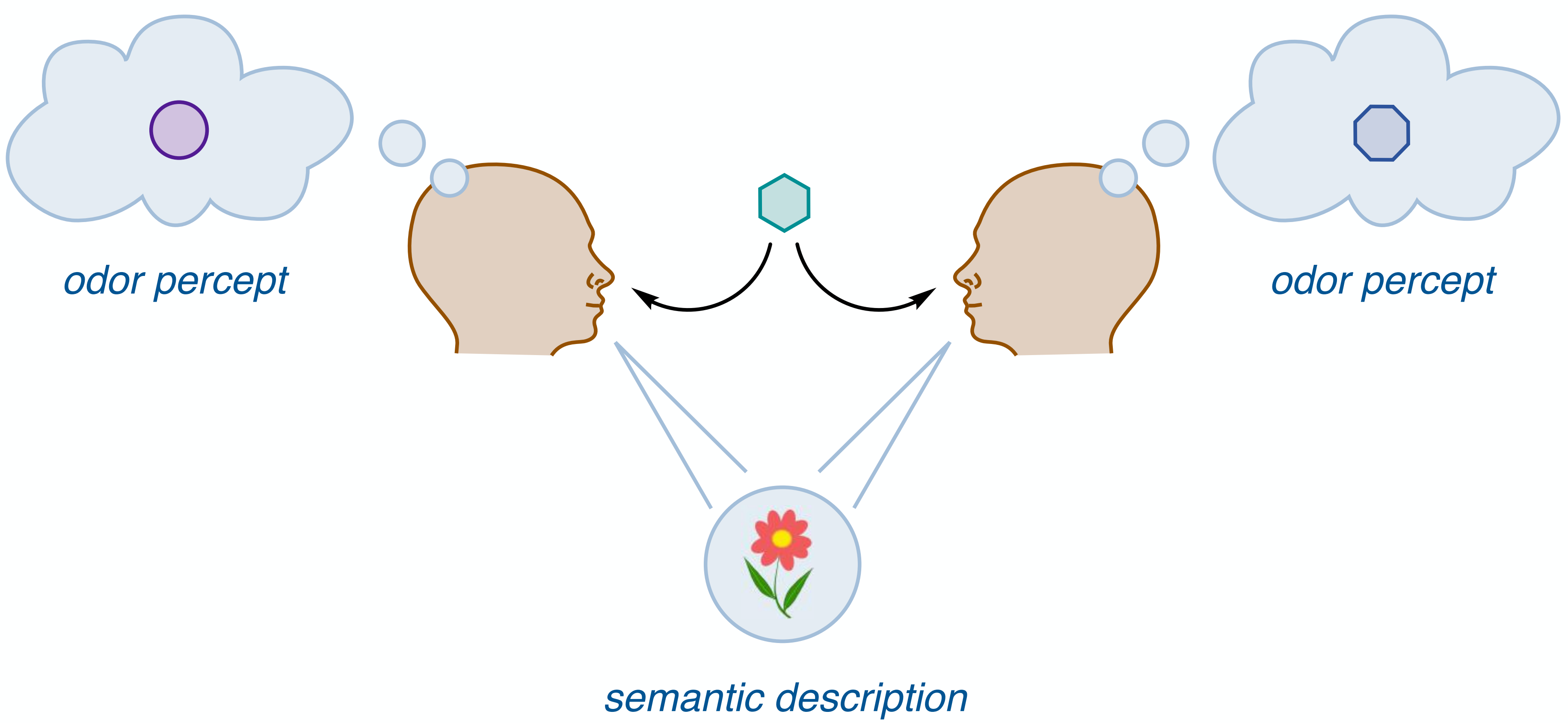
- RT/WM and WM/WM groups described androstenone as more 'vanilla' and less 'sickening'
- genotypic variation in OR7D4 accounts for a significant proportion of differential perception of andostenone

Keller, A.; Zhuang, H.; Chi, Q.; Vosshall, L. B.; Matsunami, H. *Nature.* **2007**, *449*, 468.

Subjectivity of Odor

How prevalent are SNPs in ORs?

- ***86%*** *of ORs have SNPs that* ***affect functionality***
- *functionality of OR alleles of any two indivudals differs by* ***42%***



Sell, C. S. *Chemistry and the Sense of Smell*; John Wiley & Sons: Hoboken, 2014.

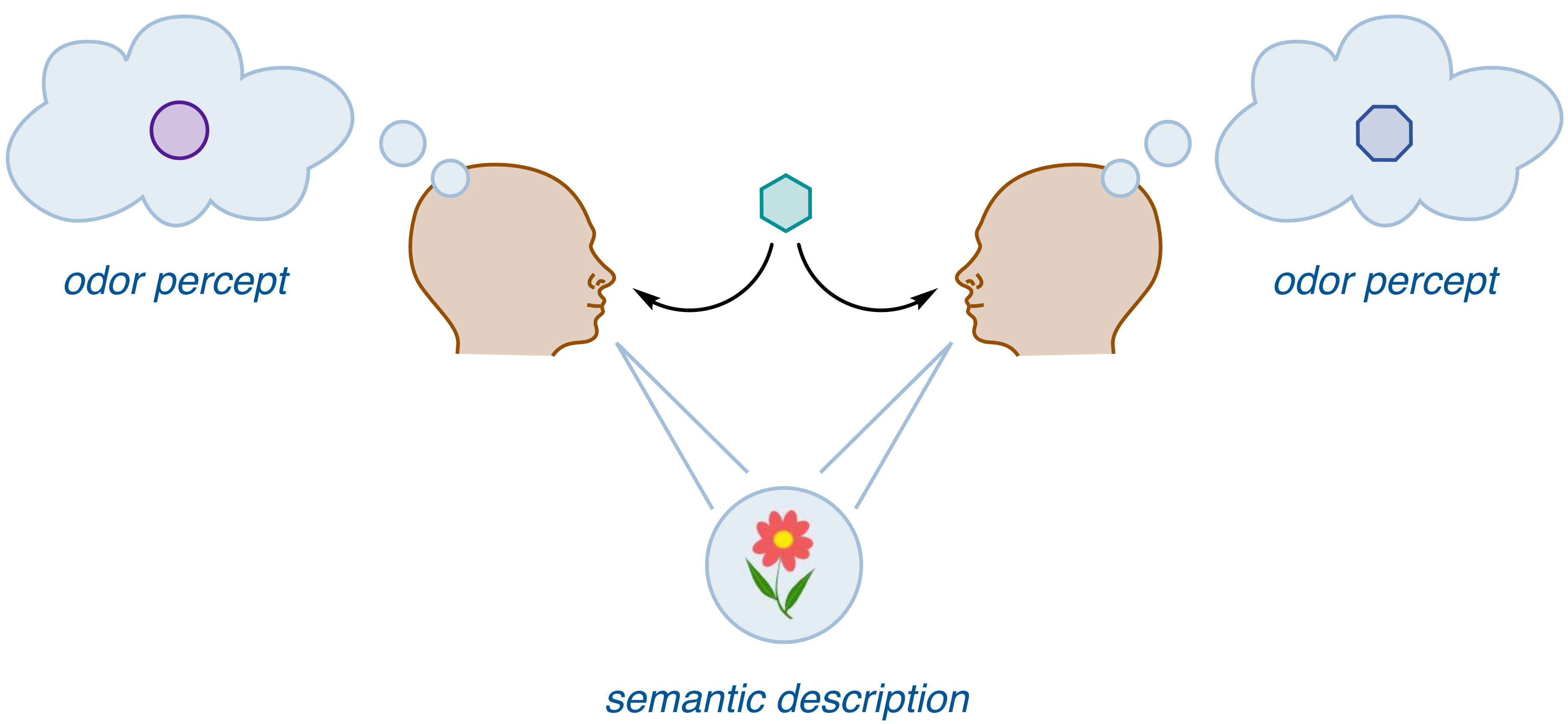
Subjectivity of Odor

"Smell is not present in the molecules that stimulate the smell receptors."

–Gordon Shepherd

"Odour…is a property of the person perceiving it and not of the molecules being perceived."

–Charles Sell



Sell, C. S. *Chemistry and the Sense of Smell*; John Wiley & Sons: Hoboken, 2014.