Impact of aldehyde dehydrogenases and aldo-keto reductases on human olfactory peri-receptor events

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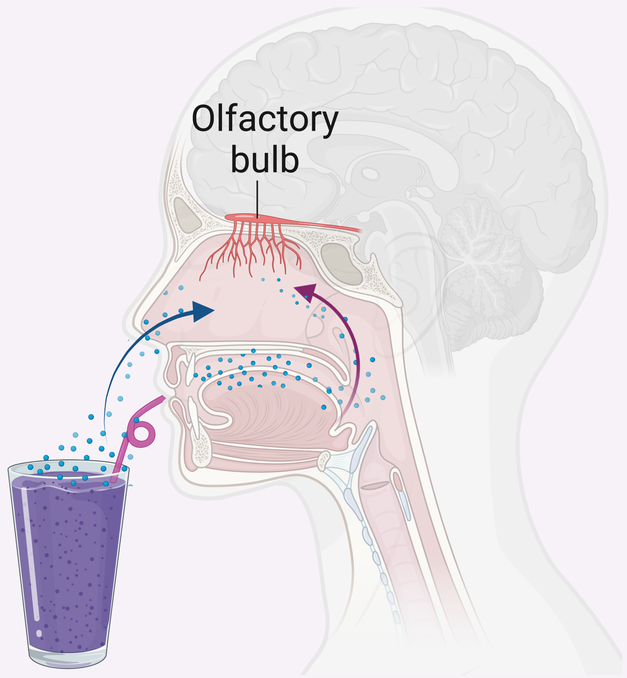
Two aldehyde dehydrogenases (**ALDH1A1** and **ALDH3A1**) and one aldo-keto reductase (**AKR1B10**) were selected for this study, based on a proteomic study showing their strong expression in the human nasal mucus.



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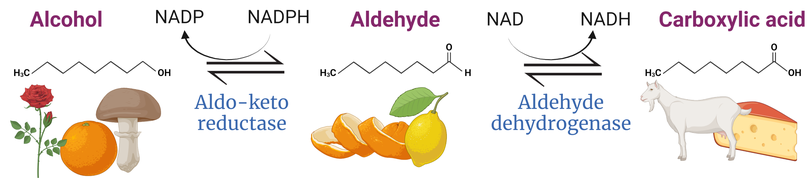
BACKGROUND

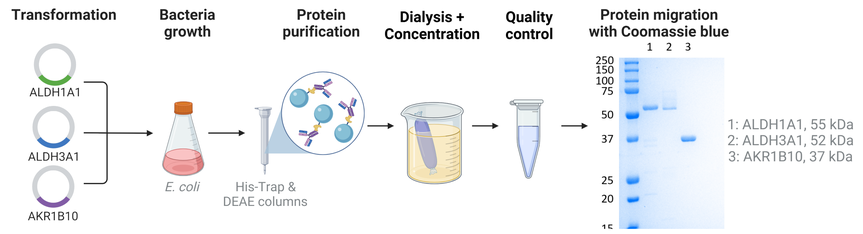
Olfaction strongly contributes to the flavor perception. It is driven by the binding of aromatic compounds to olfactory receptors. However, before and/or after binding, aromatic compounds can be metabolized by enzymes in the olfactory cleft leading to the formation of new compounds and/or signal termination.

Octanal is used as an exemple in this study. Octanal has an odor described as orange peel or citrus, moreover its metabolites: octanol smells a mixture between rose, orange and mushroom whereas octanoic acid is described as goat cheese odor.



METHODS

The localization of the three studied enzymes were investigated in human epitheliums from turbinate and olfactory celft. The recombinant proteins were produced using *E. coli* and purified following different steps summarized in the figure below.
Catalytic parameters of recombinant proteins were measured towards a panel of 20 aldehydes. The complex ALDH3A1/octanal was solved by X-ray crystallography allowing to analyzed the amino acids involved in the catalysis and substrates recognition.
Salivary enzymatic metabolization was studied using GC-MS analysis.



RESULTS

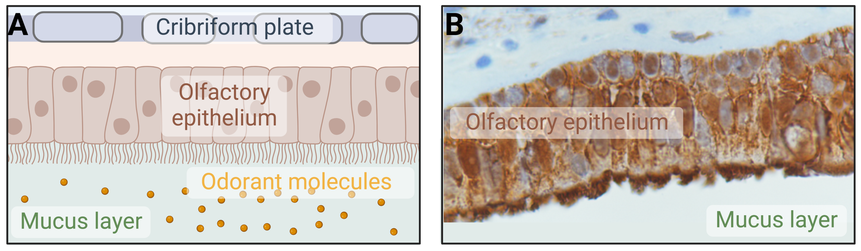


Figure 1. **A:** Schematic representation of human olfactory epithelium. **B**: Immunohistochemistry staining against hALDH1A1 in olfactory epithelium.

Similar stainings (red-brown) were observed for the three studied enzymes in olfactory/respiratory epitheliums using immunohistochemistry. Results indicate a high expression level of these enzymes in epithelial cell types in particular on the apical surface (ciliated cells). This expression allows the interaction between enzymes and odorants.

A

Odorant (scent)	Structure	k_{cat}/K_M ($min^{-1}.\mu M^{-1}$)		
		ALDH1A1	ALDH3A1	AKR1B10
Pentanal (fermented fruity)		30 ± 14	12 ± 2	2.0 ± 0.5
Hexanal (green)		15 ± 10	102 ± 35	25 ± 6
Heptanal (green)		12 ± 4	129 ± 51	13 ± 2
Octanal (lemon)		16 ± 4	209 ± 87	15 ± 4
Nonanal (rose)		12 ± 7	164 ± 101	11 ± 3
Decanal (orange)		4 ± 2	33 ± 17	2 ± 1
Undecanal (citrus)		16 ± 14	24 ± 13	2.6 ± 0.5

B

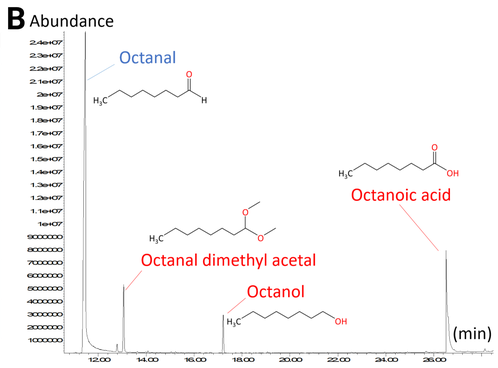


Figure 2. **A:** Kinetic parameters of ALDH1A1, ALDH3A1, and AKR1B10 towards some aldehydic odorants. **B:** Chromatogram of an octanal solution metabolized by saliva. Substrate is indicated in blue and metabolites are indicated in red.

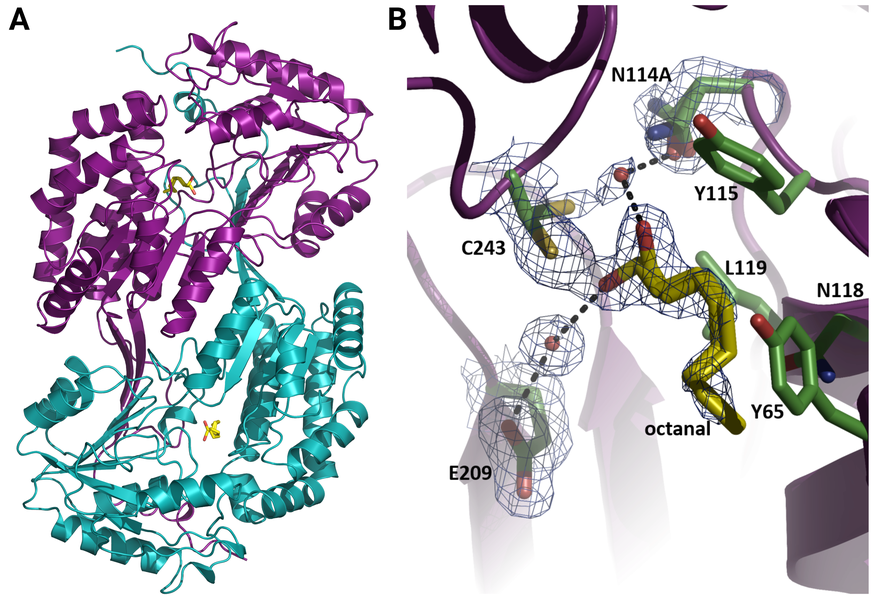


Figure 3. **A:** Crystal structure of ALDH3A1 bound to octanal (yellow) solved in this study. **B**: ALDH3A1 active site bound to octanal near the catalytic cysteine 243. Octanal, Asn 114, and Cys 243 are all in two conformations. Side chains residues are shown as green sticks.

The X-ray structure of the complex ALDH3A1/octanal was solved at 1.8 Å. ALDH3A1 is a homodimeric protein (monomer A in cyan, monomer B in magenta). The active site of both monomer is occupied by an octanal molecule (in yellow). Interpretation of the electron density maps in the active site region near the catalytic Cys 243 led to the conclusion that octanal is present as two alternative conformations. Considering the carbon atoms' positions, these two conformations are very close, with hydrophobic interactions stabilizing the aliphatic moiety of octanal by the surrounding residues' side chains (Tyr 65, Tyr 115, Asn 118, Leu 119).

CONCLUSION

The results of this study demonstrate: first the **abundance of the three tested enzymes in olfactory/respiratory epithelium and nasal mucus** allowing them to interact easily with aldehyde odorants. Secondly, this study highlights the ability of these enzymes to **metabolize aldehyde compounds** leading to new molecules with potentially new sensory properties or contributing to terminate the signal. Finally, the 3-dimensional structure of ALDH3A1 give further information on the **amino acids involved in the catalytic reaction** to better understand the catalysis and substrate specificities.

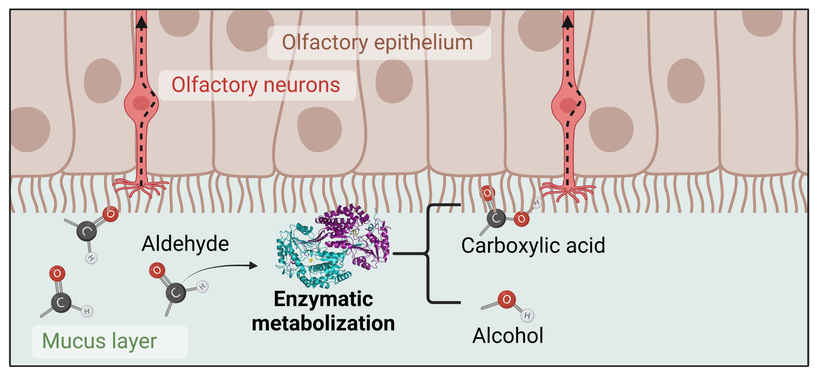


Figure 4. Schematic representation of enzymatic metabolization of aldehydes into alcoholic or carboxylic acid in olfactory vicinity.

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