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Multisensory Olfactory Training for Smell Loss

Par

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Programme de sciences biomédicales, Faculté de médecine

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Cette thèse intitulée

Multisensory Olfactory Training for Smell Loss

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Résumé

La COVID-19 est une maladie infectieuse causée par le virus SARS-CoV-2. L'un de ses symptômes les plus précoces et les plus fréquents est la dysfonction olfactive (OD). Selon le variant du virus, de 2 à 80 % des individus présentent une OD, telle que l'hyposmie ou l'anosmie, et certains développent une OD qualitative, incluant la parosmie ou la phantosmie. Bien que la récupération survienne généralement en quelques semaines, certains cas persistent et entraînent une perte olfactive à long terme.

Des études antérieures ont démontré que l'entraînement olfactif (OT) peut bénéficier aux patients présentant une OD causée par des infections virales des voies respiratoires supérieures. Dans le cadre de l'intégration multisensorielle, différents canaux sensoriels peuvent s'amplifier ou interférer mutuellement selon leur congruence. Étant donné que les entraînements multimodaux montrent des effets supérieurs dans les études sur la plasticité neuronale, l'utilisation de stimuli gustatifs et visuels congruents, en complément à des stimuli olfactifs, pourrait accroître l'efficacité de l'OT. Cette hypothèse peut être évaluée en testant des combinaisons de stimulations doubles et triples dans des tâches de perception multisensorielle.

Le premier objectif de cette thèse est de quantifier la perception de similarité de stimuli olfactifs présentés avec ou sans costimulation gustative et visuelle, et de déterminer l'ampleur de l'effet de la stimulation hétéromodale. Le second objectif consiste à intégrer ces stimuli multisensoriels dans un protocole d'OT modifié afin d'évaluer si la récupération de la perte olfactive peut être optimisée.

Dans l'article 1 (Chapter 2 - Congruency of Multisensory Olfactory Stimuli), nous avons testé 48 jeunes adultes en bonne santé à l'aide de stimuli multisensoriels, comprenant des composantes olfactives, gustatives et visuelles dont les composantes chiniosensorielles étaient administrés par voie rétronasale. Ces stimuli étaient présentés en combinaisons doubles et triples, dans des conditions congruentes et incongruentes. Les stimuli étaient également présentés individuellement afin d'établir des valeurs perceptuelles de base. Les stimuli olfactifs et gustatifs étaient administrés sous forme de gouttes déposées sur la langue (c.-à-d. rétronasale), tandis que les stimuli

visuels étaient présentés sur un écran d'ordinateur. Les participants évaluaient la similarité entre les solutions présentées et l'étiquette olfactive correspondante à l'aide d'échelles visuelles analogiques.

Dans l'article 2 (Chapter 3 - *Can Multisensory Olfactory Training Improve Olfactory Dysfunction Caused by COVID-19?*), nous avons recruté 68 participants présentant une OD persistante depuis au moins deux mois après une infection à la COVID-19. Les participants étaient répartis en trois groupes. Le premier groupe recevait un entraînement olfactif classique (COT), consistant en l'exposition répétée à quatre odorants. Le deuxième groupe suivait un entraînement olfactif multisensoriel (MOT), recevant les mêmes odorants par voie rétronasale sous forme de gouttes sur la langue, accompagnés simultanément de stimuli gustatifs et visuels congruents. Le troisième groupe recevait du propylène glycol inodore dans quatre flacons (groupe témoin/placebo). L'entraînement était réalisé deux fois par jour pendant 12 semaines. La fonction olfactive (UPSIT) et la qualité de vie spécifique à l'olfaction (SQOD-NS) étaient évaluées avant et après l'intervention.

Dans l'article 1, nous avons observé un effet significatif de la congruence sur la perception de similarité ($p < 0,001$). La costimulation gustative avait un effet significativement plus important sur la perception olfactive que la costimulation visuelle ($p = 0,02$). Dans l'Article 2, les deux groupes d'intervention présentaient des améliorations significatives similaires de la fonction olfactive, tandis que le groupe placebo ne montrait aucune amélioration significative. Les scores de qualité de vie se sont améliorés dans les trois groupes après 12 semaines.

En conclusion, la costimulation congruente améliore la perception de similarité, alors que la costimulation incongruente la réduit. La costimulation gustative exerce des effets plus prononcés sur l'olfaction que la costimulation visuelle. Intégrés au MOT, ces stimuli multisensoriels améliorent significativement les capacités olfactives à l'issue de la période d'entraînement.

Mots clés : olfaction, gustation, vision, perception multisensorielle, entraînement olfactif, plasticité.

Abstract

COVID-19 is an infectious disease caused by the SARS-CoV-2 virus. One of its earliest and most common symptoms is olfactory dysfunction (OD). Depending on the COVID-19 variant, 2–80% of individuals experience OD, such as hyposmia or anosmia, and some develop qualitative OD, including parosmia or phantosmia. Although recovery typically occurs within a few weeks, some cases persist and lead to long-term olfactory loss.

Previous studies have shown that olfactory training (OT) can benefit patients with OD caused by viral infections of the upper respiratory tract. In multisensory integration, different sensory channels may enhance or interfere with one another depending on congruency. Because multimodal trainings show improved outcomes in plasticity studies, using congruent gustatory and visual stimuli alongside olfactory stimuli may increase OT effectiveness. This can be evaluated by testing dual and triple stimulation combinations in multisensory perception tasks.

The first objective of this thesis is to quantify likeness perception of olfactory stimuli presented with or without gustatory and visual co-stimulation and to determine the magnitude of the heteromodal stimulation effect. The second aim is to incorporate such multisensory stimuli into a modified OT protocol to assess whether recovery from olfactory loss can be improved.

In Article 1 (Chapter 2 - Congruency of Multisensory Olfactory Stimuli), we tested 48 healthy young participants using retronasal olfactory stimuli consisting of olfactory, gustatory, and visual components. These stimuli were presented in dual and triple combinations under congruent and incongruent conditions. Stimuli were also presented individually to establish baseline perceptual values. Olfactory and gustatory stimuli were delivered as droplets on the tongue (i.e., retronasally), while visual stimuli were shown on a computer screen. Participants rated the likeness of the solutions to the corresponding olfactory label on visual analog scales.

In Article 2 (Chapter 3 - Can Multisensory Olfactory Training Improve Olfactory Dysfunction Caused by COVID-19?), we recruited 68 participants with persistent OD lasting two months or more following COVID-19 infection. Patients were divided into three groups. One group received classical olfactory training (COT), which involved smelling four odorants. The second group

followed multisensory olfactory training (MOT), receiving the same odorants retronasally as droplets on the tongue, accompanied by simultaneous and congruent gustatory and visual stimuli. The third group received odorless propylene glycol in four bottles (control/placebo group). Training was performed twice daily for 12 weeks. Olfactory function (UPSIT) and olfactory-specific quality of life (SQOD-NS) were assessed before and after the intervention.

In Article 1, we observed a significant effect of congruency on likeness perception ($p < 0.001$). Gustatory co-stimulation had a significantly stronger effect on olfactory perception than visual co-stimulation ($p = 0.02$). In Article 2, both intervention groups showed similar significant improvements in olfactory function, while the placebo group showed no significant improvement. Quality-of-life scores improved in all three groups after 12 weeks.

In conclusion, congruent co-stimulation enhances likeness perception, while incongruent co-stimulation reduces it. Gustatory co-stimulation has stronger effects on olfaction than visual co-stimulation. When integrated into MOT, these multisensory stimuli significantly improve olfactory abilities after the training period.

Keywords: Olfaction, gustation, vision, multisensory perception, olfactory training, plasticity.

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List of acronyms and abbreviations

AC: Alternating current

ACE2: Angiotensin-converting enzyme 2

Bcl-2: B-cell lymphoma 2

cAMP: Cyclic adenosine monophosphate

CO2: Carbon dioxide

COT: Classical olfactory training

COVID-19: Coronavirus disease 2019

CNV: Cranial nerve

CREB: cAMP response element-binding protein

CNS: Central nervous system

DAMP: Damage-associated molecular pattern

DP: Daphnée Poupon

EEG: Electroencephalogram

EOG: Electro-olfactogram

ERP: Event-related potentials

FWHM: Full-width-half-max

fMRI: Functional magnetic resonance imaging

GCS: Glasgow coma scale

IPS: Intraparietal sulcus

ISI: Interstimulus interval

MAPK: Mitogen-activated protein kinase

MOT: Multisensory olfactory training

MNI: Montreal Neurological Institute

MPRAGE: Magnetization prepared rapid gradient echo imaging

MRI: Magnetic resonance imaging

mRNA: Messenger ribonucleic acid

NaCl: Sodium chloride

NMP: Negative mucosal potential

OB: Olfactory bulb

OD: Olfactory dysfunction

OE: Olfactory epithelium

OMP: Olfactory marker protein

OR: Odorant receptor

OSN: Olfactory sensory neuron

OT: Olfactory training

PAMP: Pathogen-associated molecular pattern

PRP: Platelet-rich plasma

PTFE: Polytetrafluoroethylene

PVOD: Post-viral olfactory disorders

RmANOVA: Repeated measures of ANOVA

SARS-CoV2: severe acute respiratory syndrome coronavirus 2

STS: Superior temporal sulcus

SZ: Subependymal zone

QOD: Questionnaire of olfactory disorders

S.D.: Standard deviation

SQOD-NS: Shortened Questionnaire of Olfactory Disorders - Negative Statements

Tau: Tubulin associated unit

TBI: Traumatic brain injury

TDI: Threshold-discrimination-identification

TMPRSS2: Transmembrane serine protease 2

UPSIT: University of Pennsylvania Smell Identification Test

URTI: Upper respiratory tract infection

VAS: Visual Analogue Scales

wk: Week

Acknowledgements

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Chapter 1 – Introduction and theoretical background

1.1. Introduction

Multisensory perception occurs when information from more than one sense is combined into a single, multimodal percept (Driver & Spence, 2000). For example, the perception of coffee involves multiple components, including taste, color, texture, and temperature. Multisensory perception is a continuous and powerful process: it reduces ambiguity in perceptual information (van Ee et al., 2009), aids in stimulus detection and identification (Small & Prescott, 2005), provides robust information about the environment (Ernst & Bulthoff, 2004), supports memory (Lehmann & Murray, 2005), enhances overall performance with faster decision-making (Shi & Muller, 2013), and leads to more effective (Shams & Seitz, 2008) and facilitated (Seitz et al., 2006) learning, especially when stimuli are congruent (Kim et al., 2008). Congruency refers to information from different sources that align with one another (Laurienti et al., 2004). For example, when entering a coffee shop, one expects the smell of coffee; when opening a trash can, one expects a foul odor; when eating watermelons, one expects sweetness, and so on.

COVID-19 (Coronavirus disease 2019), a viral infection caused by SARS-CoV2 (severe acute respiratory syndrome coronavirus 2), emerged in Wuhan, China, in December 2019 (Shereen et al., 2020). It rapidly spread worldwide and infected over 750 million people, causing the deaths of more than 7 million individuals as of 29 June 2025 (World Health Organization, 2024). One of the earliest and most reliable predictor symptoms of contracting COVID-19 is the loss of olfactory function (Gerkin et al., 2021). Until the emergence of the Omicron variant, olfactory loss was highly prevalent among COVID-19 patients (Chang et al., 2024), reaching rates as high as 53% (Wu et al., 2022). This percentage decreased to 3.7% once the Omicron variant, an evolved form of SARS-CoV-2 with 2.5 times higher transmissibility (Liu & Rocklov, 2022), became the globally dominant strain (von Bartheld & Wang, 2023). Although COVID-19 patients typically recover olfactory function within a few weeks after losing their sense of smell, up to 10% do not regain olfactory abilities even after six months, resulting in long-term olfactory loss (Kim et al., 2024). Therefore,

there is a need for therapies, and clinicians and researchers have explored various methods to address this issue.

One method for regaining olfactory function after viral infection is olfactory training (OT). In the classical olfactory training (COT) paradigm, patients sniff four different odors (lemon, clove, eucalyptus, and rose) twice daily for 12 weeks (Hummel et al., 2009). This training has been shown to be effective in treating olfactory loss caused by viral infections of the upper respiratory tract, even before the COVID-19 pandemic (Kattar et al., 2021), although the recovery rates remain suboptimal, with only 55% of participants reporting subjective improvement in their olfactory abilities (Khan et al., 2023). In recent years, researchers have attempted to modify OT to increase its success rates. One possible modification is to introduce multisensory elements into the procedure. Although some attempts to modify COT have been made, to date, integrating both gustatory and visual stimuli into the training have never been tried. Our aim is to improve COT by leveraging the strengthening effects of multisensory perception.

1.2. Anatomy, physiology and function of the olfactory system

Olfaction is the least explored human sense, partly because it has historically been regarded as an intuitive sense rather than one that conveys information (Boesveldt & Parma, 2021). Nevertheless, people are constantly surrounded by chemosensory cues, such as the smell of flowers, the perfume of others, and the scent of foods and beverages they consume. Olfactory information is continuously transmitted to the brain, even when it does not reach conscious awareness.

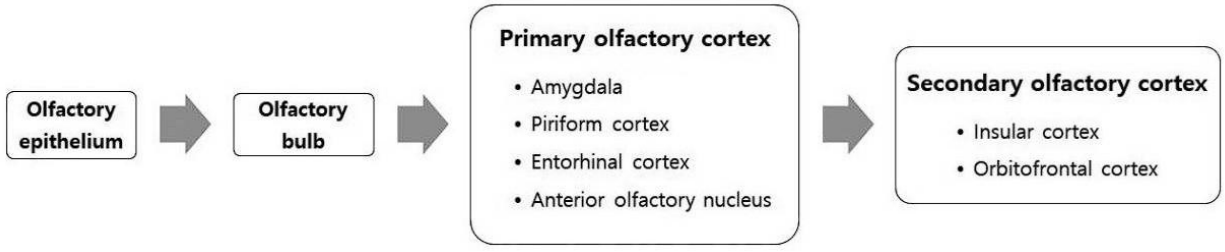


Figure 1.1. Flow of olfactory information from periphery to central nervous system. *The specialized tissue olfactory epithelium captures the olfactory cues and sends them to the olfactory bulb for processing. From here, information is sent to the entorhinal cortex and amygdala for*

integrating and emotionally processing the data, the piriform cortex for perception of smell and anterior olfactory nucleus for early processing of odors. Information is sent to the insular cortex for further sensory processing and emotional regulation and orbitofrontal cortex for higher order cognitive functions such as decision making, mediating responses, value coding, learning or driving social behavior. Figure is taken from (Han et.al., 2023).

Olfaction serves numerous purposes in human life. Odors contribute to ingestive behavior through the detection and identification of food sources, stimulate appetite, facilitate breastfeeding via maternal breast odor, enable recognition and avoidance of environmental hazards such as chemicals or pathogens, identify microbial threats such as vomit or decay, assist in social communication, and influence reproductive behaviors including inbreeding avoidance and mate selection (Stevenson, 2010). Olfaction plays key roles in cognitive and emotional functions, such as emotion processing and emotional regulation (Bratman et al., 2024), as well as personal safety as people with OD cannot detect dangerous odors such as gas leaks or fires (Lee et al., 2024).

The first step in olfactory perception occurs when odorant molecules reach the olfactory mucosa within the olfactory epithelium either orthonasally or retronasally. During orthonasal olfaction, odorants enter the nasal cavity through sniffing or inhaling, whereas in retronasal olfaction, odorant molecules reach the nasal cavity from the mouth through the nasopharynx during chewing or swallowing. Odorant molecules then reach the cilia of olfactory sensory neurons (OSNs), where they bind to chemoreceptors known as odorant receptors (OR). These receptors are expressed in OSNs located within the olfactory mucosa. The binding of odorants triggers chemo-electrical transduction, generating action potentials that are transmitted to the glomeruli in the olfactory bulb (OB) via OSN axons (Breer, 2008, Duchamp-Viret et al., 2016).

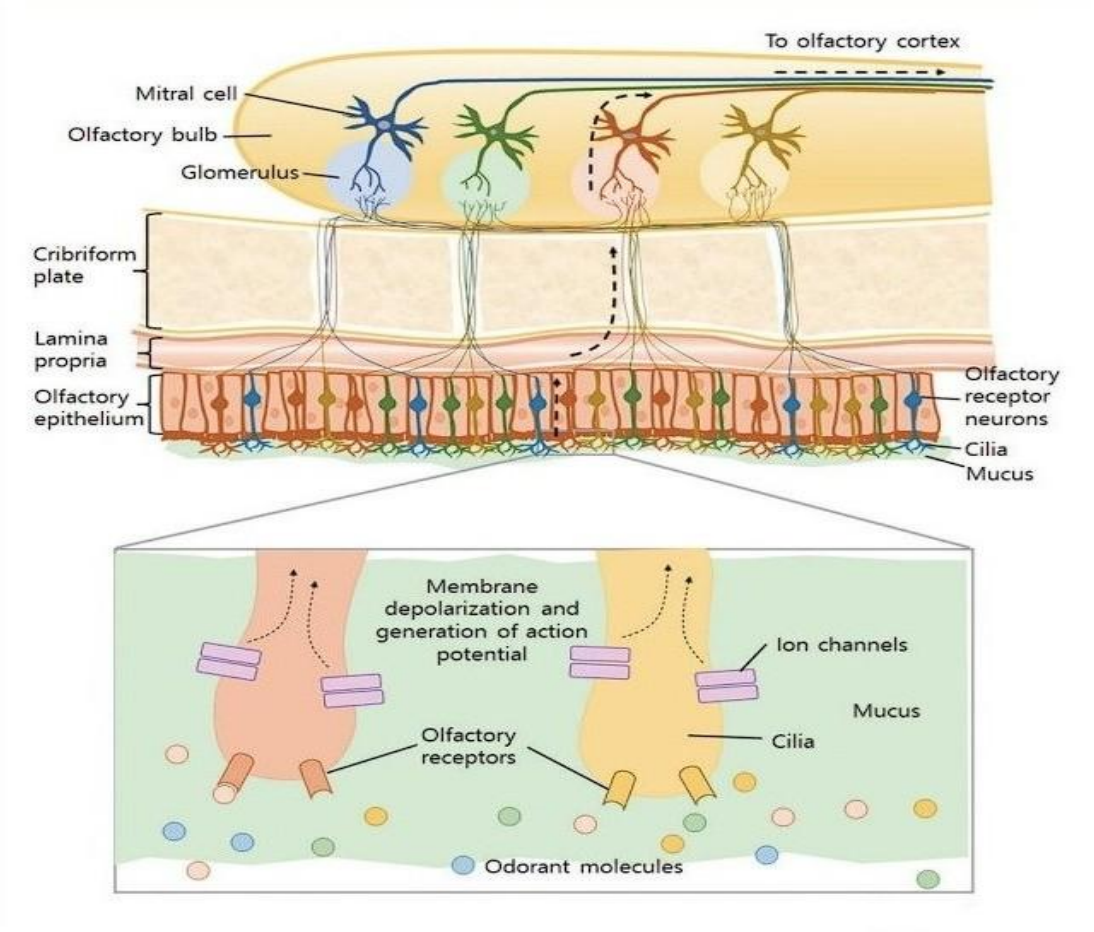


Figure 1.2. Structures of the olfactory system in olfactory epithelium and olfactory bulb. *While mitral cells receive information from the axons of OSNs, they form synapses with glomeruli. Glomeruli are spherical structures that form synapses between the dendrites of mitral cells and terminals of olfactory nerves. The ciribriform plate is the top part of ethmoid bone with tiny holes that allow olfactory nerves to pass through to deliver olfactory information to glomeruli. Lamina propria that is situated under the ciribriform plate is a connective tissue that is constituted of mucosa. Cilias are small, hair-like structures that help in the detection of external stimuli. Figure is taken from (Han et.al., 2023).*

The glomeruli serve as initial synaptic sites where OSN axons converge and synapse with the dendrites of mitral and tufted cells (Chen & Shepherd, 2005). Within the OB, olfactory signals undergo processes such as adjustment and enhancement (Chen & Shepherd, 2005). Mitral cell axons that form the olfactory tract convey output signals to higher brain regions (Breer, 2008). These regions

include the amygdala, primary olfactory cortex which encompasses the piriform cortex, anterior olfactory nucleus, and entorhinal cortex (Han et al., 2023, Duchamp-Viret et al., 2016). From these areas, information is transmitted to limbic structures involved in higher-order cognitive processes, such as emotional and behavioral responses (Shepherd, 2005), olfactory memory (Buchanan et al., 2003), reward processing and learning in the amygdala (Noto et al., 2021), spatial mapping, navigation and memory facilitation in the hippocampus (Silkis, 2023), and attention and learning functions in the thalamus (Courtiol & Wilson, 2015).

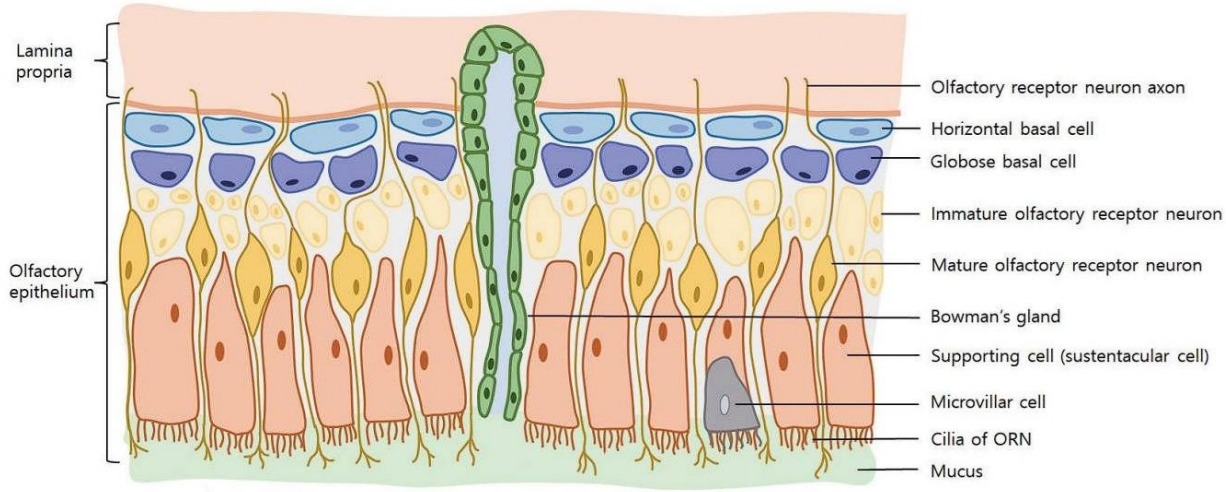


Figure 1.3. A closer look at olfactory mucosa. *Olfactory sensory neurons (also sometimes called olfactory receptor neurons) express olfactory receptors. The axons of the olfactory sensory neurons that express the same receptor converge into a single glomerulus. Two types of basal cells are shown in the image: (1) Globose basal cells are reserves but active progenitors and (2) Horizontal basal cells are also reserves but they are activated by injury to the olfactory system. Bowman's glands secrete mucus that is essential and contains olfactory binding proteins. Supporting cells dispose of dead or dying cells and chemically process the pollutants. Microvillar cells increase the area for more diffusion, help in absorption and secretion. Figure is taken from (Han et.al., 2023).*

Although the olfactory system is the most developed sensory system at birth (Sarnat & Flores-Sarnat, 2019), humans generally perform poorly on odor identification tasks (Stevenson & Case, 2005). This phenomenon is often called "tip of the nose" experience or described as the "feeling

of knowing" (Jonsson & Olsson, 2003). This may occur because odor recognition and verbal labels are weakly linked in semantic memory (Stevenson et al., 2007). Based on this information, one could hypothesize that additional sensory information may facilitate odor identification and potentially enhance performance in OT and related tasks. Because if the information coming from olfactory channel is not enough to identify the stimuli, additional sensory information from gustation and vision can aid in the various olfactory scores such as identification, detection and discrimination.

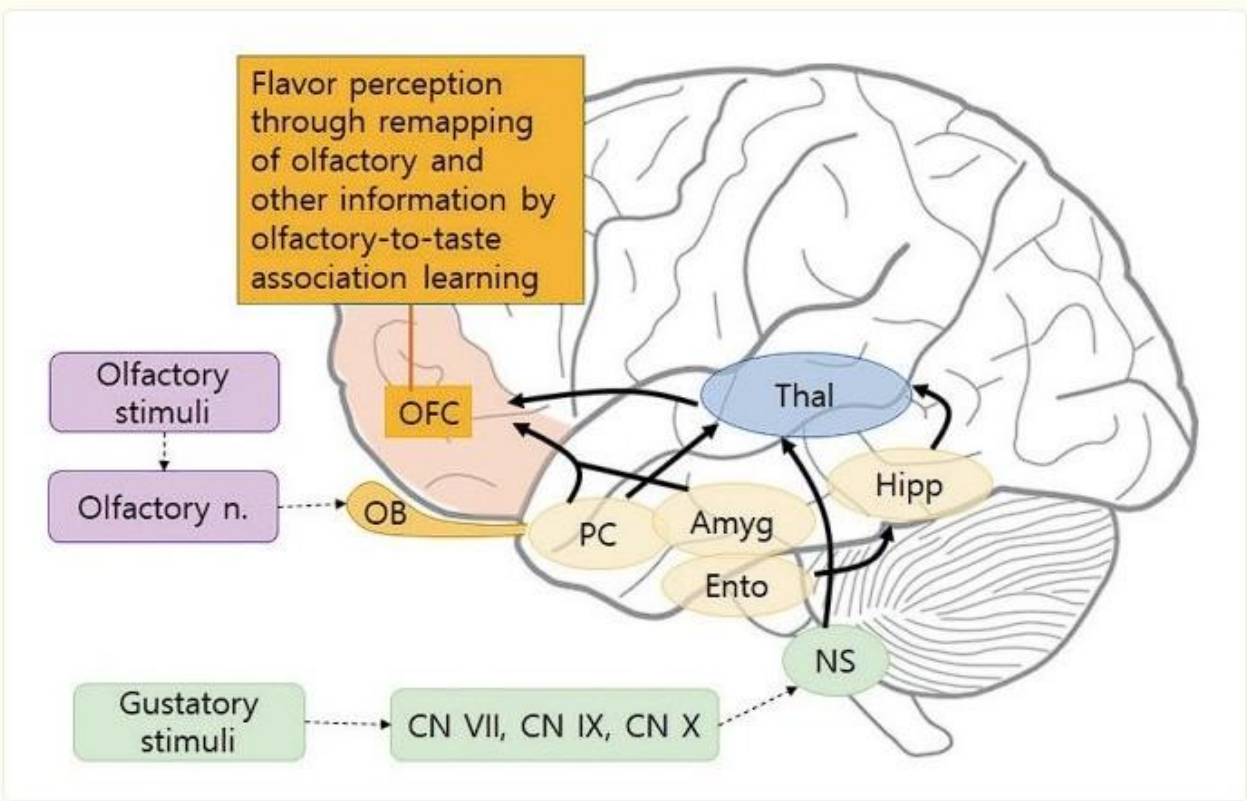


Figure 1.4. Olfactory and gustatory pathways to orbitofrontal cortex to form flavor perception. *Thal: thalamus. Ob: olfactory bulb. PC: piriform cortex. Amyg: amygdala. Hipp: hippocampus. Ento: entorhinal cortex. NS: nucleus of the solitary tract. It is a primary, visceral sensory relay station, receives information from respiratory and gastrointestinal systems. CNVII: Cranial nerve VII; Facial nerve, receives information from the face, responsible for face expression and taste sensation in the anterior tongue. CN IX: Cranial nerve IX; Glossopharyngeal nerve. Receives sensory and taste information from posterior tongue and pharynx. CN X: Cranial nerve X; Vagus nerve. It*

receives sensory information from head, neck, abdominal and digestive tract. Figure is taken from (Han et.al., 2023).

1.3. Olfactory dysfunction

1.3.1. Forms of Olfactory Dysfunction

Olfactory Dysfunction (OD) can be categorized into two main types (1) quantitative OD which refers to the impairments characterized by a loss or reduction in olfactory abilities, and (2) qualitative OD, which involves distorted olfactory perception. Quantitative OD includes two conditions: (1) anosmia, the complete loss of olfactory function, and (2) hyposmia, a significant reduction in the sense of smell. Qualitative OD, also known as dysosmia, can be divided into two subtypes: (1) parosmia, the distorted perception of odors, and (2) phantosmia, the perception of odors in the absence of an external odor source. Parosmia typically results in certain odors being perceived as unpleasant. In very rare cases, distorted olfactory perception may cause normally unpleasant or neutral odors to perceived as pleasant, a condition referred to as euosmia (Landis et al., 2006, Yang & Pinto, 2016).

1.3.2. Prevalence of Olfactory Dysfunction

The prevalence of OD before the COVID-19 pandemic was estimated to be approximately 22% based on a meta-analysis of 25 publications comprising a total of 175.073 participants (Desiato et al., 2021). These data were derived from both survey-based assessments and psychophysical tests, which varied in methodology, task design, and duration.

OD prevalence can also be evaluated according to specific disorder types. Qualitative OD generally shows a lower prevalence compared to quantitative OD, although parosmia appears to emerge more frequently in the context of quantitative impairments (Hummel et al., 2023).

1.3.3. Causes of Olfactory Dysfunction

OD can arise from several conditions, including sinonasal disorders, viral infections of the upper respiratory tract, traumatic brain injuries, idiopathic olfactory loss, neurodegenerative diseases, congenital abnormalities, and age-related factors (Whitcroft et al., 2023).

1.3.3.1. Sinonasal disorders

Sinonasal OD refers to impairments caused by pathologies involving sinuses or nasal pathways. It can be divided into two categories: (1) conductive OD, in which air-flow-related abnormalities obstruct the passage of air to olfactory epithelium or olfactory cleft, and (2) neurosensorial OD, in which damage or dysfunction occurs along the olfactory neural pathway (Schwartz et al., 2019).

Sinonasal-related OD may arise from conditions such as chronic rhinosinusitis, with or without nasal polyps, as well as from surgical interventions that result in damage, scarring or inflammation of the olfactory epithelium. Chronic rhinosinusitis is characterized as an inflammatory condition involving epithelial barrier disturbance caused by microbes, allergens, and inflammatory pathway dysregulation. It typically presents four symptoms: facial pain or pressure, hyposmia or anosmia, nasal obstruction, and nasal drainage. It can persist for at least 12 weeks (Desrosiers et al., 2011, Fokkens et al., 2012).

According to the Canadian Clinical Practice Guidelines, chronic rhinosinusitis may be identified through endoscopic findings such as discolored mucus and edema of the middle meatus or ethmoid region; in chronic rhinosinusitis with polyps, clinicians may observe unilateral or bilateral nasal polyps in the middle meatus (Desrosiers et al., 2011). Nasal polyps are defined as inflammatory lesions occurring within the olfactory airways (Stevens et al., 2016). Potential contributing factors to chronic rhinosinusitis include genetic predispositions, septal deviation, bacterial colonization of the sinonasal tract, environmental exposures such as airborne fungal elements, allergies, and microbial pathogens (Chin et al., 2025).

Sinonasal disorders are the most common cause of OD, with chronic rhinosinusitis affecting approximately 10% of the European population based on self-reported symptoms, and with 84% of individuals with chronic rhinosinusitis exhibiting OD (Whitcroft et al., 2023).

1.3.3.2. Post-viral olfactory disorders (without COVID-19)

Post-viral olfactory disorders (PVOD) are defined as olfactory dysfunctions that occur following viral infections. Viruses known to cause OD include rhinoviruses, influenza and parainfluenza viruses, respiratory syncytial viruses, adenoviruses, enteroviruses, and coronaviruses (Heikkinen & Jarvinen, 2003).

Viruses may impair olfactory function through three primary mechanisms: (1) physical obstruction, caused by local inflammation, swelling of nasal passage lining, or excessive nasal discharge which prevents odor molecules from reaching and binding to olfactory receptors, (2) damage to the olfactory epithelium, including injury to peripheral olfactory nerves and cells, (3) central nervous system involvement, in which viruses penetrate the central nervous system (CNS) and potentially damage olfactory-related regions such as OB, piriform cortex, amygdala, and olfactory tubercle (Rebholz et al., 2020).

Although a systematic review of 38 animal studies reported a reduction in the number of olfactory receptor neurons in the olfactory epithelium and OB, identifying consistent infection sites and specific affected cell types remains challenging. This variability arises from differences in animal models, viral species, and strain variants used across studies (Lee et al., 2021).

Human studies have documented several consequences of PVOD, including increased olfactory cleft volume and width (Altundag et al., 2021), significant gray matter reduction in the orbitofrontal cortex (Yao et al., 2018), hypometabolism in the piriform cortex, amygdala, and parahippocampal regions (Kim et al., 2012), reduced OB volumes (Rombaux, Martinage, et al., 2009), and decreased numbers of olfactory receptor cells and nerve bundles in mucosal biopsies (Yamagishi et al., 1994).

Epidemiological studies also highlight the prevalence of PVOD. In a study of 441 patients at the Taste and Smell Clinic of the Connecticut Chemosensory Clinical Research Center (USA), 19% were diagnosed with PVOD (Goodspeed et al., 1987). Among 375 patients with OD at an outpatient clinic in Japan, 24.3% had post-viral origins (Suzuki et al., 2007).

1.3.3.3. Traumatic brain injury

Traumatic brain injury (TBI) refers to an injury caused by an external force to the head, with severity ranging from a simple blow to the head to penetrating injuries, or in milder cases, concussions (Georges & Das, 2025). Damage resulting from TBI may affect peripheral olfactory structures or orbitofrontal regions, leading to olfactory dysfunction (OD) or reduced olfaction (Tai et al., 2023).

The scope of injury may include: (1) nasal damage, such as direct trauma, mucosal hematoma, edema, scarring involving nasal passages, (2) tearing or trauma to olfactory nerve fibers, which may occur with nasal or skull-based fractures or blunt head forces, (3) brain contusions, including bruising of the OB or orbital frontal pole region (Costanzo & Miwa, 2006), as well as cerebral softening due to bleeding or inflammation in subfrontal and temporal lobes (Yousem et al., 1999). These types of injuries can result in various forms of OD, including anosmia, hyposmia, parosmia, or phantosmia in patients with TBI (Tai et al., 2023).

The severity of TBI is commonly assessed using the Glasgow Coma Scale (GCS), with scores < 9 indicating severe TBI, scores between 9–12 indicating moderate TBI, and scores between 13–15 classified as mild TBI (Marino et al., 2025). The likelihood of OD has been found to correlate strongly with GCS scores, amnesia, and radiological abnormalities (Green et al., 2003). OD has been reported in 13% of patients with mild TBI, 19% of those with moderate TBI, and 25% of those with severe TBI (Howell et al., 2018).

1.3.3.4. Congenital anosmia

Congenital anosmia is defined either as hypoplastic (underdeveloped) or aplastic (absent) OB (Abolmaali et al., 2002). The malformations underlying congenital anosmia may result from genetic or environmental factors and often manifest as multiple defects in organ or bodily morphogenesis, although the precise mechanisms are typically unclear (Corsello & Giuffre, 2012). Reported structural abnormalities include: (1) a bilaterally thicker layer of gray matter in the left piriform cortex and medial orbitofrontal cortex (Frasnelli et al., 2013), (2) a deeper olfactory sulcus levels on the right side compared to the left (Abolmaali et al., 2002), (3) temporal and frontal volume loss (Yousem et al., 1996).

These patients usually present with anosmia or severe hyposmia from birth; however, they are typically diagnosed between the ages of 12 and 16 (Whitcroft et al., 2023). Congenital anosmia appears to be relatively rare, with an estimated prevalence of 1 in 10,000 people (Saw et al., 2022).

1.3.3.5. Neurodegenerative disorders

Neurodegenerative disorders are defined as the progressive loss of neurons, including deterioration of their physicochemical properties and functions in the brain or peripheral organs. Alzheimer's disease, Parkinson's disease, and multiple sclerosis are among the most common neurodegenerative disorders (Gadhave et al., 2024).

Alzheimer's Disease, the most common form of dementia, is characterized by progressive impairment in memory and other cognitive functions such as learning, motor control, and thinking (Gadhave et al., 2024). The underlying pathomechanisms of Alzheimer's Disease include the accumulation of amyloid-β plaques and tau proteins (microtubule-associated protein in neurons), as well as neuronal inflammations and subsequent synaptic-dysfunction (Gouras et al., 2015). Olfactory deterioration is considered an early biomarker for predicting Alzheimer's disease (Wang et al., 2010). As the disease progresses, olfactory function continues to decline, resulting in impaired odor identification scores (Wang et al., 2010). Explanations for the deterioration in olfactory ability include: (1) reduced gray matter volume in the piriform cortex, amygdala, and entorhinal cortex (Jobin et al., 2023), (2) smaller OB volumes and a trend toward reduced primary olfactory cortex (Jobin et al., 2021), (3) reduced blood flow in the primary olfactory cortex and insula (Wang et al., 2010).

Parkinson's disease is a neurodegenerative movement disorder caused by dopaminergic neuronal degeneration in the substantia nigra (Postuma et al., 2015). It is characterized by bradykinesia, tremor, rigidity, or combinations of these symptoms (Postuma et al., 2015). The pathomechanism involves the formation of Lewy bodies, which are abnormal intracellular aggregates composed primarily of α-synuclein protein (Morris et al., 2024). Olfactory degradation is commonly observed in Parkinson's disease, with OD serving as an early biomarker and gradually worsening throughout disease progression (Doty, 2012). Explanations for this decline include: (1) accumulation of Lewy bodies (aggregated α-synuclein) in the OB and anterior olfactory nucleus (Beach et al., 2009), (2) Lewy body pathology in the cells of the olfactory epithelium (Saito et al., 2016), (3) altered functional activity in olfactory regions such as amygdala, orbitofrontal cortex, olfactory gyrus, insula, and parahippocampal gyrus (Su et al., 2015), (4) environmental pathogens such as viruses and toxins (Doty, 2008).

Because olfactory impairment in Alzheimer's disease can serve as a predictor, OD is observed up to 100% of Alzheimer's disease cases (Duff et al., 2002) and in 90% of the Parkinson's disease patients (Doty, 2012).

1.3.3.6. Idiopathic olfactory dysfunction

OD can be classified as idiopathic when all other possible causes have been excluded through testing, clinical examination, or attempted treatments, leaving the disorder without an identifiable underlying reason or pathomechanism (Whitcroft et al., 2023). In some cases, idiopathic OD may serve as a predictor for the later development of neurodegenerative diseases, such as Parkinson's disease (Sommer et al., 2004). The relative frequency of idiopathic OD has been estimated at 8% in the general population (Hummel et al., 2023).

1.3.3.7. Aging

Although aging itself is not a pathology, OD is commonly observed in older people (Gogele et al., 2024). Several factors may contribute to this decline, including comorbidities that increase with age such as those associated with neurodegenerative diseases, as well as atrophy of the olfactory epithelium, reduced mucosal blood flow (Doty & Kamath, 2014), and decreased cortical activity (Morgan & Murphy, 2010). Additionally, some medications frequently used by elder population, including antihistamines, antihypertensives, cardiovascular agents, and anti-infectives can also lead to olfactory complaints (Schiffman, 2018).

The prevalence of age-related OD varies considerably between self-reported assessments and test-based investigations, possibly because older patients may be unaware of the gradual decline in their olfactory abilities. Self- reported estimates of OD are at 1% (Hoffman et al., 1998), whereas psychophysical test-based evaluations indicate a markedly higher prevalence of 24.8% in older populations (Seubert et al., 2017). These discrepancies highlight the limited reliability of self-evaluation for olfactory function.

1.3.4. Consequences of Olfactory Dysfunction

Patients with OD face numerous challenges. Olfaction is an important sensory modality for survival because unpleasant odors can signal potential danger (Koike et al., 2021). Therefore,

olfaction is essential in detecting: (1) house fires, gas leaks, and toxic substances in the environment (Pence et al., 2014), (2) burnt food (Croy et al., 2012) or even (3) individuals who are sick (Tognetti et al., 2023). Losing the sense of smell can make daily life burdensome, as patients must compensate by relying on other senses or devices such as gas detectors (Croy et al., 2014). Consequently, 37% of people with olfactory impairment report experiencing at least one hazardous event related to their dysfunction, such as failing to detect fire, gas leaks, or spoiled food (Santos et al., 2004).

Olfaction also plays a central role in feeding. Humans rely on odors to determine whether food is suitable for ingestion based on prior experiences and the associated outcomes of consuming that food product (Stevenson, 2010). When an ingested food does not match expected olfactory cues, people may reject it, which can help protect against microbial contamination or food poisoning (Stevenson, 2010). Odors can regulate appetite by serving as appetizers before consumption of food (Zoon et al., 2016), promote reliable salivation (Rogers & Hill, 1989), and contribute to feelings of satiety with prolonged exposures (Yeomans, 2006). In infants, olfactory cues guide breastfeeding behavior, as newborns can recognize and orient towards their mother's breast odor promoting suckling and ingestion (Varendi & Porter, 2001).

Olfaction is also crucial for social communication. Humans perceive olfactory cues that convey emotional information such as stress, anxiety and fear (de Groot & Smeets, 2017), happiness (de Groot et al., 2015), and disgust (Zheng et al., 2018). Humans use olfactory cues to determine sex (Schleidt et al., 1981), recognize faces (Damon et al., 2021), and identify family members or kin (Lundstrom et al., 2009). Due to difficulties in perceiving social odor cues, people with OD often worry about their personal hygiene and fear that they may smell unpleasant without being aware of it (Croy et al., 2014). As a result, they may report fewer sexual relationships or feeling less secure in romantic relationships (Croy et al., 2013).

People with OD frequently report that socializing becomes more difficult and that they avoid going to restaurants because they cannot enjoy food or beverages with friends or family (Aschenbrenner et al., 2008). They describe reduced pleasure from eating, which can contribute to eating disorders such as anorexia nervosa or bulimia nervosa (Dazzi et al., 2013), diminished appetite, and unhealthy weight loss (Mattes & Cowart, 1994).

OD can also negatively impact work life (Bramerson et al., 2007), particularly in environments requiring social interaction or teamwork. Careers that rely heavily on olfactory abilities such as chefs, perfumers, sommeliers, firefighters and nurses may be especially affected (Haxel et al., 2012).

Considering these challenges collectively, people with olfactory impairment are at increased risk of developing mental health problems such as anxiety (Chen et al., 2021) or depression (Kohli et al., 2016). Olfaction plays an essential role in daily life, contributing to safety, social interaction, work, eating, and drinking. Therefore, OD should be properly diagnosed and treated to support patients in maintaining quality of life and functional well-being.

1.4. COVID-19

COVID-19 is an infectious disease caused by the SARS-CoV-2 virus (Adil et al., 2021). The onset period of the disease ranges from three to seven days, with an average of five days (Hakki et al., 2022). Common symptoms include fever, cough, fatigue, and headache as well as loss of the sense of smell (Menni et al., 2020). In some cases, infection may be asymptomatic, which can mislead people about their health status and contribute to increased transmission (Ravindra et al., 2022). Diagnosis of COVID-19 is achieved through viral testing methods such as: (1) viral gene detection, (2) human antibody detection, and (3) viral antigen detection (Yuce et al., 2021).

The virus primarily spreads between humans through interactions involving breathing, coughing, or sneezing, both at close and long ranges, particularly indoors (Wang et al., 2021). Various preventive measures are recommended to limit transmission, including social distancing, travel and public gathering restrictions, lockdowns, maintaining hygiene, and wearing masks (Girum et al., 2021).

Angiotensin-converting enzyme 2 (ACE2) serves as the host receptor for SARS-CoV-2 (Hoffmann et al., 2020). ACE2 is expressed primarily in lungs, enterocytes (intestinal absorptive cells), renal tubules in the kidneys, gallbladder, cardiomyocytes (cardiac muscle cells), male reproductive cells, placental cells, ductal cells in the eyes, and blood vessels (Hikmet et al., 2020). Importantly, ACE2 is also expressed by sustentacular cells in the olfactory epithelium, making the sense of

smell vulnerable to infection, although OSNs themselves are not targeted (Bryche et al., 2020). Additionally, the protease transmembrane serine protease 2 (TMPRSS2) facilitates viral entry into host cells (Fuentes-Prior, 2021). TMPRSS2 is an androgen-regulated enzyme encoded by the TMPRSS2 gene (Koyou et al., 2025).

Transmission primarily occurs through contact of respiratory droplets or aerosol with the nasal, conjunctival (thin layer covering the sclera), or oral mucosa (Arienzo et al., 2023). Therefore, the virus mainly affects the respiratory tract (nose, throat, sinuses) and may spread to the lungs, causing respiratory complications such as pneumonia and acute respiratory distress syndrome (Su et al., 2021). Long-term consequences can include chronic lung injury, resulting in symptoms such as dyspnea (shortness of breath), reduced diffusion capacity of carbon monoxide, and radiological anomalies including ground-glass opacity (lighter looking tissue), reticular opacity (decreased soft tissue), bronchial dilation, and air trapping (Cha et al., 2024).

Due to ACE2 expression sites in the gastrointestinal tract, patients may experience gastrointestinal infection leading to irritable bowel syndrome, inflammation, and alterations in the intestinal microbiomes (King, 2024). Significant cardiac complications have also been reported, including myocardial infarction (heart attack), heart failure, Takotsubo cardiomyopathy (broken heart syndrome), myocarditis (inflammation of heart muscle), and arrhythmias (irregular heartbeat) (Krishna et al., 2024). Kidney-related problems may occur as well, such as proteinuria (high levels of protein in urine), hematuria (blood in urine), and renal injury involving tubular damage or collapsing glomerulopathy, with viral particle traces detected in the kidneys (Kaye et al., 2021).

In severe cases, these complications may require hospitalization and can potentially lead to death (Baptista et al., 2023). Hospital admissions increased during the pandemic for conditions involving blood disorders, eye problems, circulatory and digestive system issues, and poisoning in neonatal and pediatric intensive care units as well as emergency and hematology departments (Motiei et al., 2025). Hospitalization risk is heightened by factors such as age (> 50), underlying medical conditions (cardiovascular or lung diseases, diabetes, neurological disorders), asymptomatic infection, and residency in long-term care facilities such as nursing homes, adult daycares, and medical foster homes (Taylor et al., 2024). The median hospital stay was 10 days, but the duration

increased more than 21 days in cases involving pneumonia, chronic liver disease, organ transplants, bacterial sepsis, blood cancers, and blood clots (Lucijanic et al., 2023). Risk factors for fatality due to COVID-19 include chronic obstructive pulmonary disease, kidney injury, diabetes, hypertension, cardiovascular diseases, older age, smoking, and obesity (Dessie & Zewotir, 2021).

COVID-19 symptoms are managed using standard influenza-like treatments or, in severe cases, additional support such as respiratory and circulatory assistance, plasma transfusion, and immunotherapy (Song et al., 2020). Antiviral medications developed specifically for COVID-19 became available in 2021, reducing hospitalization risk and accelerating symptom resolution (Ibrahim et al., 2025).

Researchers began developing vaccines immediately, and effective vaccines became publicly available starting in late 2020 (Prasad & Haslam, 2024). Vaccination has been shown to reduce disease severity, decrease COVID-19 incidence, and lower the risk of hospitalization and death (Brice et al., 2023).

1.4.1. COVID-19 and olfaction

One of the earliest signs of COVID-19 is olfactory loss or impairment (Gerkin et al., 2021, Kim et al., 2020, Chang et al., 2024). Because of this, researchers conducted both animal and human studies to determine the infection sites responsible for olfactory dysfunction. These investigations identified SARS-CoV-2 infections in the OB and olfactory epithelium of golden hamsters (Frere et al., 2022), and in the olfactory cortex and orbitofrontal cortex of rhesus monkeys (Beckman et al., 2022). In humans, post-mortem examinations revealed infection of sustentacular cells in the olfactory mucosa (Khan et al., 2021).

When the virus first enters the body, infection occurs with the assistance of ACE2 and TMPRSS2, leading to active viral replication, a strong inflammatory response, and subsequent cell death. During the immune response phase, cell deaths trigger DAMP (damage-associated molecular pattern) and PAMP (pathogen-associated molecular pattern) recognition, resulting in the release of pro-inflammatory cytokines and chemokines and the recruitment of monocytes, macrophages, and T-cells to eliminate the infected cells. However, if the disease progresses from mild to a severe case, excessive infiltration of immune cells into the lungs can occur, causing dysregulated

cytokine production and allowing viruses to spread to other organs, potentially triggering a pro-coagulant response and organ failure (Bohn et al., 2020).

The infection may spread through the nasal cavity or bloodstream, triggering neuroinflammation in several brain regions such as the hippocampus, insular lobe, or olfactory cortex (Castanares-Zapatero et al., 2022). In persistent taste and smell disorders, neuroepithelial viral invasion and subsequent inflammation have been suggested as mechanisms underlying long-term disturbances (Gasmi et al., 2021). Persistent viral presence and ongoing inflammation within the olfactory epithelium can make recovery from chemosensory dysfunction considerably more difficult (Kandemirli et al., 2021). Persistent SARS-CoV-2 viral presence has also been reported within taste buds (Doyle et al., 2021). Another proposed mechanism for long-term OD is virus-driven changes in neural susceptibility (neurotropism), which may contribute to long-term dysfunction (Castanares-Zapatero et al., 2022).

Chronic inflammation and sustained viral presence can lead to tissue injury, immune dysregulation, and persistent activation of inflammatory pathways (Aid et al., 2025). Additionally, infection of sustentacular cells may produce severe local inflammation that exposes OSNs to cytokines and induces mRNA reorganization, leading to functional impairments (Zazhytska et al., 2022). In normal recovery, infected sustentacular cells are cleared, allowing epithelial regeneration through olfactory basal stem cells, resulting in repair (Gadye et al., 2017).

The persistence and mechanisms of inflammation in OD remain the subject of ongoing research. However, hypotheses such as antibody dysregulation have been proposed (Cabral-Marques et al., 2022, Kim et al., 2024). Sustentacular cell infection and destruction of the olfactory epithelium during the acute phase may progress into long-term OD due to OSN dysfunction, OB injury, and alterations in the olfactory and associated cortex (Chang et al., 2024). Differences in recovery rates among patients, with some experiencing rapid resolution and others developing long-term OD, may be explained by both viral and host factors, including viral variants, genetic background, ethnicity, and age (Chang et al., 2024).

The prevalence of COVID-19-related OD varies considerably across studies and populations, largely due to methodological differences in assessment techniques. In a recent study that

included both subjective complaints and psychophysical measurements of quantitative and qualitative dysfunctions, 89.5% of participants showed quantitative OD, with 50% experiencing parosmia and 43.3% experiencing phantosmia (van Dijk et al., 2025).

1.5. Therapies for olfactory dysfunction post COVID-19

Various therapeutic approaches have been investigated for their potential effectiveness in restoring olfactory function, although OT has emerged as the most effective method for treating olfactory loss resulting from viral infections (Alarfaj et al., 2023). In addition to OT, other suggested therapies include platelet-rich plasma, pharmacological treatments such as corticosteroids, and topical treatments including vitamin A, palmitoylethanolamide, luteolin, and sodium citrate (Hummel et al., 2023).

1.5.1. Platelet-rich plasma

Platelet-rich plasma (PRP) has been introduced as a treatment option for prolonged OD in patients with COVID-19 (Yan et al., 2023). PRP is an autologous blood-derived biological product containing supraphysiologic concentrations of growth factors obtained from the patients' own blood (Yan et al., 2023). Blood is drawn from the patient, and injected into nasal regions such as the septum, or in cases of septal deviation, the closest accessible area to the olfactory cleft (Lechien & Saussez, 2024).

PRP may be beneficial because it contains high levels of leukocytes, fibrin, and bioactive growth factors that contribute to tissue repair. Platelets play a role in hemostasis and release growth factors, while leukocytes (white blood cells) clear pathogens from tissues, and fibrin supports tissue regeneration (Wang et al., 2022). This treatment has been associated with neuronal protection, prevention of neural apoptosis, promotion of vascular and axonal regeneration, regulation of inflammatory processes, and reduction in muscle atrophy (Sanchez et al., 2017). PRP has also been shown to support peripheral nerve regeneration through modulation of inflammatory responses (Wang et al., 2022).

In a comparative study with a control group, after one month of treatment, the PRP group demonstrated more substantial improvements in TDI scores (particularly in threshold scores) than those

in the control group (Steffens et al., 2022). After two months of PRP treatment, patients showed significant improvement in TDI and subjective quality of life scores (QOD) compared to placebo groups (Lechien et al., 2023). At 10 weeks (2.5 months), both PRP and control groups showed significant improvement in TDI scores; however, only the PRP group demonstrated increased QOD scores (Lechien et al., 2024).

In a study following patients with post-COVID-19 OD for one year after receiving either PRP or placebo injections, the PRP group showed significantly greater improvement in objective and subjective scores, indicating potential long-lasting therapeutic effects (Fieux et al., 2025). Systematic reviews and meta-analyses have also confirmed that PRP therapy significantly improves both subjective and objective olfactory outcomes in COVID-19 patients (Albazee et al., 2025).

1.5.2. Pharmacological interventions

Some therapeutic interventions for olfactory dysfunction include pharmacological treatments such as corticosteroids, and topical agents (vitamin A, palmitoylethanolamide, luteolin, sodium citrate), which are applied directly on affected surfaces (Hummel et al., 2023).

The use of corticosteroid nasal sprays has been recommended as earlier studies reported improvements in olfactory threshold, discrimination, and identification scores among hyposmic patients following nasal corticosteroid treatment (Abdelalim et al., 2021). Oral corticosteroids, tested in combination with OT in a prospective cohort study, were also evaluated (Le Bon et al., 2021). While nasal corticosteroids produced significant improvements, oral corticosteroids had minimal effects, and neither demonstrated superiority over OT alone.

Oral supplements such as palmitoylethanolamide and luteolin (both possessing neuroprotective and anti-inflammatory properties) have also been used to address COVID-19-related olfactory impairment (Gellrich et al., 2024). Patients treated with the combination of palmitoylethanolamide and luteolin showed greater improvement in olfactory threshold, discrimination, and identification scores.

Vitamin A has been proposed as a supplement to support repair process in the olfactory epithelium (Taheri et al., 2024), based on evidence demonstrating its role in promoting olfactory sensory

neuron regeneration following PVOD (Hummel, Whitcroft, Rueter, et al., 2017), although its effectiveness appears limited.

Sodium citrate, tested in combination with melatonin and vitamins (including zinc), was evaluated alongside OT in COVID-19 patients; however, it did not provide significant improvement beyond that achieved with OT alone (Garcia et al., 2024). Intranasal sodium citrate on its own has also been tested in patients with post-viral OD and similarly failed to produce significant improvement in Sniffin' Sticks scores (Whitcroft et al., 2021).

1.5.3. Olfactory Training

OT consists of participants sniffing four predetermined odors (lemon, rose, clove, and eucalyptus in the original study) twice daily for 12 weeks (Hummel et al., 2009). Prior to the pandemic, OT had already been shown to improve olfactory abilities in 30% of patients with post-upper respiratory tract infection (URTI)-related olfactory dysfunction (Hummel et al., 2009). As a non-invasive and no-risk intervention, OT remained a primary recommended treatment for OD, (Kattar et al., 2021, Denis et al., 2021) sometimes used in combination with other therapeutic approaches such as intranasal corticosteroids (Asvapoositkul et al., 2023).

1.5.3.1. History of olfactory training

OT is based on the theory that repeated exposure to odors may promote the regeneration and functional recovery of olfactory neurons (Youngentob & Kent, 1995). For example, younger adults demonstrate better performance than children in odor threshold, odor identification, and object-naming tasks in the Boston Naming Test. Researchers have hypothesized that this advantage may be due to greater lifelong exposure to a wider range of odors (Cain et al., 1995). This conceptual foundation of OT can be traced back to 1969, when researchers periodically presented odorous Q-tips to infants and observed selective facilitation and enhanced responsiveness to those odors after repeated exposure (Engen & Bosack, 1969). Modern OT protocols derive their choice of odorants from Hans Henning's "odor prism" theory from 1916, which categorizes odors into six groups: flowery (rose in OT), foul, fruity (lemon in OT), aromatic (clove in OT), burnt, and resinous (eucalyptus in OT) (Hummel et al., 2009, Koyama & Heinbockel, 2022).

1.5.3.2. Mechanisms behind OT

The exact underlying mechanism by which OT improves olfactory function remains unclear. One proposed explanation is that OT promotes neurogenesis within the olfactory epithelium (OE), which contains stem cells capable of generating new OSNs (Schwob & Costanzo, 2010), thereby increasing the number of neurons in the olfactory mucosa (Doty, 2019; Pieniak et al., 2022). OT may also upregulate gene expressions related to synaptic plasticity and neuronal survival in the OB (Tepe et al., 2018). OT might serve as an olfactory marker protein (OMP) promoter, potentially triggering oncogene expression that initiates neuronal turnover by accelerating cell death and replacement (Schwob & Costanzo, 2010).

Another hypothesis involves the trophic dependency of OSNs on the OB. If the OB becomes infected or damaged, rapid OSN cell death may occur; however, repeated stimulation through OT may encourage neurogenesis, modulated by sensory activity and odorant saliency (Schwob & Costanzo, 2010). Repeated odor exposure may stimulate neurogenesis in the OE and promote axonal growth, enabling connection with mitral cells via synapses, ultimately increasing OB volume. Such neurogenesis may occur through heightened activation of olfactory receptors and increased secretion in neurotrophic factors, which promote proliferation and activation of neural precursor cells, ultimately supporting regeneration (Hu, Zhang, et al., 2022). This plastic response may mirror early human developmental mechanism, in which exposure to olfactory stimuli facilitates encoding new information and formation of new synaptic connections (Arisi et al., 2012).

Evidence from animal studies supports these hypotheses. In mice, OT accelerates the expression of olfactory receptors (B. Y. Kim et al., 2020). OT may also stimulate stem cells and glial cells in OE, promoting neurogenesis and suggesting that odorant exposure acts as a regulator of recovery (Kim et al., 2019). Additionally, repeated odor stimulation may prevent OSN apoptosis by activating MAPK (mitogen-activated protein kinase) and CREB (cAMP response element-binding protein) transcriptional pathways, leading to the expression of anti-apoptotic protein such as Bcl-2 (Watt et al., 2004).

Synaptogenesis is another proposed mechanism. Increased sensory activity during OT may modulate and strengthen synaptic connections between OSNs and mitral cells, resulting in increased

OB volume (Eavri & Nedivi, 2013, Zatorre et al., 2012). Progenitor cells in subependymal zone (SZ) can also generate chains of neuroblasts that migrate toward the lateral cerebral ventricle (Kornack & Rakic, 2001). Although the existence of this pathway in humans is debated (Johansson et al., 1999, Sanai et al., 2004), neural stem cells in the SZ of rodents can produce neuroblasts that migrate to the OB and differentiate into olfactory interneurons both in rats and monkeys (Kornack & Rakic, 2001, Lois et al., 1996, Ming & Song, 2011).

Finally, the intrinsic plasticity of the OB may contribute to the effects of OT. Neural stem cells residing in the OB may be stimulated through repeated exposure, leading to increases in OB cell number and volume (Pagano et al., 2000).

1.5.3.3. Forms of olfactory training

The COT protocol consists of four odorants (rose, lemon, eucalyptus, and clove). These odorants are self-administered by patients or participants twice daily for 12 weeks. In the first study introducing this method, 30% of patients demonstrated improvement following COT (Hummel et al., 2009). In a meta-analysis including 16 articles in the qualitative synthesis and 4 in the meta-analysis, researchers found that patients with PVOD had a greater likelihood of recovering olfactory function compared to controls after completing an OT program (Kattar et al., 2021).

Since the introduction of COT, numerous attempts have been made to modify the training paradigm to improve patient outcomes. Some of these modifications have focused on altering the duration of the training program or on changing the types of odor stimuli used in the protocol.

1.5.3.4. Olfactory training with modified training time

One form of modification to OT involves altering the duration of the training period or the number of daily training sessions. Researchers have proposed that extending the duration of OT may strengthen its effects. For example, increasing the training period from 12 weeks to 36 weeks resulted in enhanced outcomes (Altundag et al., 2015). Similarly, prolonged training over 32 weeks (Geissler et al., 2014) and combining OT with 35 weeks of flavor education (Besser et al., 2020) have been associated with improved recovery.

Other modifications have not increased the likelihood of recovery but were found to be equally effective as the standard OT protocol. These include extending the training to 18 weeks (Damm et al., 2014) or increasing the number of daily sessions from two to four (Oleszkiewicz et al., 2022).

1.5.3.5. Olfactory training with modified stimuli

Another form of modification to the OT involves altering the stimuli used in the protocol. This can include changing the concentrations of odorants, introducing new types of stimuli, increasing the number of odorants, or using novel methods for stimulus administration.

Some of these modifications have been shown to enhance the effectiveness of OT. For instance, combining OT with steroids such as budesonide (Nguyen & Patel, 2018) or topical corticosteroids (Fleiner et al., 2012) has been associated with improved outcomes. Increasing the number of odorants to 10 (restricted to plant-based oils) over a 30-day training period also produced beneficial effects (Donelli et al., 2023). Culturally adapted OT using locally familiar odorants has been successful in improving recovery rates (Choi et al., 2021). Additional methods to improve adherence include using an OT ball with four odorants inserted on its sides (Saatci et al., 2020) and holding weekly video calls to encourage consistent participation (Delgado-Lima et al., 2024). Other innovative approaches include using seven different odorants delivered nightly via an odor diffuser for two hours over six months (Woo et al., 2023), as well as distributing randomized single-molecule and complex odorants among test groups (Li et al., 2023).

Several other modifications, while not increasing effectiveness beyond that of standard OT, were still found to be equally effective. Examples include increasing the number of odorants to eight (Genetzaki et al., 2024), using complex odor mixtures instead of simple odorants or changing odorants every two months across training duration of 4 to 12 months (Oleszkiewicz et al., 2018), using seven odorants instead of four (Power Guerra et al., 2024), administering an uncontrolled randomized set of four odorants (Patel et al., 2017), and presenting 64 odorants immersively in a large theater using a huge olfactometer that disperses odorized air to participants (Chao et al., 2024).

1.6. Neuronal Plasticity

Understanding neuronal plasticity within the olfactory system is essential for explaining the mechanisms underlying OT. Neuronal plasticity refers to the brain's ability to modify itself functionally or structurally (von Bernhardi et al., 2017). Historically, neuroplasticity was believed to be unlikely in adults, and the idea of an adult brain reorganizing itself was dismissed until the 1990s (Owji & Shoja, 2020). Today, researchers recognize that the human brain undergoes continuous, lifelong development through processes such as memory formation, skill acquisition, recovery from injury, adaptation, and learning (Marzola et al., 2023). Nevertheless, plasticity manifests differently across the lifespan: (1) during development (prenatal period and childhood), plasticity is highly facilitated as the brain undergoes rapid growth and forms new neuronal connections (Knudsen, 2004), (2) in adulthood, plasticity depends on factors such as age, personal experience, and environmental influences, as the mature brain is more tightly regulated than the developing brain (Marzola et al., 2023).

Neuroplasticity can be categorized into two major types: (1) structural plasticity, involving physical changes in neurons or neural networks, such as altered gray matter volume, changes in neuronal number, neuronal migration, modifications in somato-dendritic and axonal morphology, and changes in synaptic strength or connectivity (Anderson, 2011, Innocenti, 2022). Neuroplastic changes also occur at the cellular level through nerve repair, myelination, and regeneration carried out by Schwann cells, oligodendrocytes, and neural stem cells (Tataranu & Rizea, 2025). (2) Functional plasticity, involving changes in the efficiency, strength, or synchrony of neuronal networks, including formation of new functional connections (Innocenti, 2022, Marzola et al., 2023).

Sensory plasticity is a form of neuroplasticity that includes: (1) associative learning, in which new or altered sensory abilities emerge rapidly in response to specific stimuli, and (2) perceptual learning, in which long-term training produces more gradual (smaller) improvements (McGann, 2015). Plasticity has been studied across multiple sensory systems, including visual (Bang et al., 2023), gustatory (Hill, 2004), and olfactory (Wilson et al., 2004) systems. This thesis focuses specifically on plasticity within the olfactory system.

Plastic modifications can also be categorized by duration: (1) short-term plasticity, which disappears rapidly (within minutes), and (2) long term-plasticity, (often referred to as long-term potentiation), which can persist for years and is considered a fundamental mechanism of learning and memory (Deperrois & Graupner, 2020).

Plastic changes may be positive (restorative), neutral, or negative, with the latter contributing to pathological outcomes (Puderbaugh & Emmady, 2025). During human development, synaptic reorganization (or pruning) is a normal and essential component of a healthy, adaptive brain (Marzola et al., 2023). However, imbalanced pruning has been associated with neurological conditions such as autism and schizophrenia (Monday et al., 2018). Plasticity impairments can also arise from injury (Mavroudis et al., 2024), trauma (Yuan et al., 2025), neurodegenerative disorders such as Alzheimer's disease or mild cognitive impairment (Morais et al., 2025), or, as examined in this thesis, COVID-19-related olfactory dysfunction (Vaira et al., 2020).

1.6.1. Olfactory plasticity

Olfactory plasticity refers to the olfactory system's ability to reorganize itself, modify its structure and function, and recover or restore the sense of smell after impairment (Wilson et al., 2004). Such plasticity may arise from learning and experience, resulting in short or long-term changes, or may occur through non-synaptic changes within the olfactory system (Wilson et al., 2004). A unique feature of olfactory plasticity is that it persists throughout the lifespan, owing to the highly regenerative nature of the olfactory system.

Olfactory sensory neurons undergo lifelong renewal within the olfactory epithelium, and synaptogenesis continues to occur between OSN axons and mitral cells in the glomeruli (Huart et al., 2013). The olfactory epithelium contains neuronal stem cells, progenitor cells, and both mature and immature neurons (Durante et al., 2020), providing a cellular basis for ongoing regeneration.

Olfactory plasticity can also be observed at the electrophysiological level: increases in electro-olfactograms (EOG) amplitudes have been recorded at the olfactory epithelium following olfactory stimulation or training (Hummel et al., 2018). Plasticity-related changes have been detected at the level of neuronal connectivity, with increased connectivity between the orbitofrontal cortex and cingulate cortex (Hosseini et al., 2020). Further enhancements in connectivity have been

found within the broader olfactory network, including the piriform cortex, entorhinal cortex, amygdala, and thalamus following OT (Kollndorfer et al., 2015)

Structural changes can also occur: increases in OB volume have been observed after recovery from olfactory dysfunction (Gudziol et al., 2009), highlighting the capacity of central olfactory structures to undergo measurable plastic reorganization.

1.6.1.1. Olfactory Training-related changes (Plasticity)

Olfactory-related plasticity changes occur in both the peripheral and central nervous systems.

1.6.1.1.1. Behavioral changes

Increased sensitivity to olfactory stimuli can be observed in humans after repeated exposure to odors (Gillmeister et al., 2025) as it is the case in OT. This training can lead to improvements in general olfactory function across various tasks, including odor intensity classification, odor quality classification, and especially odor identification, even in healthy participants (Al Ain et al., 2019). Comparable effects are seen following specific forms of training, such as culinary training, which leads to enhanced discrimination and threshold scores (Niklassen et al., 2025).

Repeated exposure to odorants can also influence affective and cognitive aspects of olfactory perception. Such effects include affective habituation (a progressive decrease in emotional intensity of odors) (Ferdenzi et al., 2014), increased familiarity and hedonic value (Luisier et al., 2019), improved discrimination ability (Jehl et al., 1995), and enhanced performance in similarity-judgement tasks (Jehl et al., 1994). However, the interstimulus interval plays a critical role in these outcomes. Short latencies between odor presentation can lead to habituation effect and reduced amplitudes of chemosensory event-related potentials for both olfactory and trigeminal stimuli (Kassab et al., 2009).

Importantly, the after-effects of exposure, including perceived intensity and salience, remain stable for significantly longer periods for food-related odorants in humans (Fontana et al., 2023). This observation is particularly relevant for our experiment, as we used food odorants. These odorants are less likely to introduce habituation or lose salience throughout the study.

1.6.1.1.2. Changes related to OB

Researchers can investigate olfactory system plasticity by measuring OB volumes, which are closely associated with olfactory function (Huart et al., 2013). OB volumes are significantly smaller in patients with OD (Buschhuter et al., 2008).

Following OT, OB volumes (notably the thickness and length of the bulb) show significant increases compared with pre-training measurements (Negoias et al., 2017). Additionally, EOG signals recorded from the olfactory epithelium are elevated after OT (Hummel et al., 2018).

These findings indicate that structural and functional improvements within the olfactory system occur following OT. Importantly, such effects have been observed not only in patients with olfactory loss but also in healthy participants, demonstrating the capacity of the olfactory system for plasticity.

1.6.1.1.3. Cellular level changes

Plasticity can be observed at the cellular level when mitral-tufted cells and olfactory-receptor fields become more finely tuned to odorants with repeated exposure. After initial exposure, olfactory receptor fields show temporary suppression, but they subsequently recover and shift towards the exposed odorant with even greater response compared to pre-exposure levels (Fletcher & Wilson, 2003). Experience-dependent changes are also visible in the output of mitral cells, reflected in altered excitation and inhibition of synapses within the OB (Wilson et al., 2004). These changes can occur both spatially, through the recruitment of additional glomeruli units, and temporally, through reduced response latencies or increased response amplitudes (Spors & Grinvald, 2002).

1.6.1.1.4. Cortical and subcortical changes

Olfactory plasticity can be observed in various cortical regions following OT. Structural changes such as increased gray matter volume have been reported in the hippocampus or thalamus (Gellrich et al., 2018), as well as in the cerebellum and orbitofrontal cortex (Han et al., 2021). Although the thalamus is not a relay station for olfactory signals, the mediodorsal nucleus of the thalamus receives input from the piriform cortex (Courtiol & Wilson, 2014), while the hippocampus is involved in olfactory memory processes (Li et al., 2025). Increased cortical thickness has

also been observed in the right inferior frontal gyrus, bilateral fusiform gyrus, and right entorhinal cortex following OT (Al Ain et al., 2019).

Neural changes associated with repeated odor exposure can be detected through neuroimaging techniques such as EEG, which reveals event-related potential changes involving primary and secondary olfactory cortices as early as 200 ms after stimulation (Kim et al., 2020). Increased activation has been observed across multiple networks, including: (1) olfactory regions such as anterior entorhinal cortex, inferior prefrontal cortex, primary somatosensory cortex, (2) integrative regions such as insula, orbitofrontal cortex, inferior parietal lobule, middle and superior temporal gyrus, and (3) somatosensory regions such as insula, primary and secondary somatosensory cortex (Kollndorfer et al., 2015). Additionally, OT can lead to increased activations in the medial orbitofrontal cortex and anterior cingulate cortex, as well as enhanced functional connectivity between the piriform cortex and caudate nucleus (Rezaeyan et al., 2025).

1.7. Multisensory Perception

Multisensory perception occurs when information from more than one sensory channel is integrated into a single percept (Driver & Spence, 2000). For instance, the perception of lemon arises from the combination of its smell, taste, flavor, color, texture, and shape. Multisensory perception is a powerful everyday process that reduces the cognitive effort required to process each sensory modality separately, enabling faster construction of percepts and more efficient identification of objects and situations (Lovelace et al., 2003). It facilitates faster responses to environmental stimuli (Laurienti et al., 2004), enhances performance when multisensory cues align closely with the task (Shi & Muller, 2013), and increases perceptual sensitivity (Schifferstein & Verlegh, 1996). In cross-modal integrations, one sensory modality can influence or modulate the perception of another such as odor and taste stimuli modifying each other's perceptual qualities (Prescott, 2012).

For many years, sensory cortices were studied as independent processing units; however, this view has shifted in recent decades. It is now widely accepted that sensory cortices interact and that they can respond to stimuli from different sensory modalities (Driver & Spence, 2000). Cortical areas also interact with subcortical regions through complex neural circuits that contribute

to multisensory integration (Choi et al., 2023). Several polymodal brain regions support this integration, although the specific convergence areas vary depending on the type of sensory information involved; for example, spatial versus speech-related processing (Choi et al., 2023).

Although multisensory integration in olfaction has been studied far less extensively, several investigations provide evidence for cross-modal processing involving smell (Ripp et al., 2018). For example, the anterior cingulate cortex and orbitofrontal cortex respond to pleasantness ratings of odors when they are semantically modulated, such as presenting isovaleric acid with a "cheese" versus "body odor" label (de Araujo et al., 2005). While affective value (pleasantness) and multisensory representations are processed in the anterior cingulate and orbitofrontal cortex, respectively, intensity coding and odor memory processing engage the inferior frontal gyrus (Rolls et al., 2008, Rolls, 2008).

In olfactory-trigeminal integration, multisensory processing involves regions such as the left STS, right IPS, and both left medial and lateral orbitofrontal cortices (Boyle et al., 2007). Posterior STS, orbitofrontal cortex, and the amygdala exhibit strong activation when integrating emotional olfactory and visual stimuli (Novak et al., 2015). Chemosensory anxiety cues increase activation in the insula and middle occipital gyrus, and they also heighten fearful evaluation of faces; for example, when presenting sweat collected during an oral examination compared to sweat collected after exercise alongside fearful facial expressions (Wudarczyk et al., 2016). The anterior insular cortex responds to attention in both taste and olfaction (Veldhuizen & Small, 2011).

Gustatory and olfactory stimuli delivered retronasally converge in the far anterior insular cortex (de Araujo et al., 2003). The orbitofrontal cortex correlates with pleasantness ratings of both taste and smell of food (Rolls, 2008), and neural representation of taste have also been identified in the pregenual cingulate cortex which receives signals from the orbitofrontal cortex (Rolls, 2008). Furthermore, the insula, orbitofrontal cortex, and anterior cingulate cortex respond significantly to congruent taste-odor mixtures but not to incongruent ones (Small et al., 2004). Similarly, the frontal operculum is activated during flavor perception but not during isolated presentation of taste or odor stimuli (Seubert et al., 2015).

1.7.1. Congruency in multisensory perception

Congruency refers to the harmony or comparability between two sensory inputs, based on the assumption that the stimuli naturally belong together (Chen & Spence, 2017). Past experiences and prior knowledge about objects, people, and events determine whether people perceive sensory combinations as congruent or incongruent (Greve et al., 2019). Humans therefore rely on memory to retrieve these experiences to judge what is acceptable (congruent) or unacceptable (incongruent) (Greve et al., 2019). The effects of semantic congruency can be observed in the brain. For example, when people are presented with word-color combinations based on whether the pairs exist in nature, activations occur in the left prefrontal cortex and occipito-temporal cortex (Maril et al., 2011). Congruency effects also emerge in social and marketing contexts. For instance, gender-congruent advertisements (e.g., women presenting female-associated products) elicit greater activation in the posterior cingulate cortex, a region associated with value coding (Casado-Aranda et al., 2018). Collectively, these examples demonstrate that congruency perception is largely a learned ability: sensory congruence is acquired through experience, stored in memory, and retrieved when evaluating sensory combinations.

1.7.2. Multisensory olfactory training

Multisensory training is generally more effective for learning and inducing plasticity than unisensory training, in which only a single sensory modality such as visual-only or auditory-only stimulation is used (Seitz et al., 2006, Lehmann & Murray, 2005). For example, in a study involving people with anosmia, multisensory (audio-visual) stimulation provided greater benefits than unisensory stimulation, including enhanced performance in multisensory integration tasks (Peter et al., 2019).

Building on this principle, multisensory elements have been integrated into COT, leading to increased effectiveness; for instance, presenting visual stimuli (digital images) alongside odorants improved outcomes compared with COT (Khan et al., 2023). Similarly, visually assisting participants using a web application that displayed images and names of odorants enhanced the training effectiveness (Denis et al., 2021). Significant improvements in odor threshold and discrimination were observed when participants watched an instructional video while smelling the

corresponding odorants, suggesting that simultaneous auditory, visual, and olfactory stimulation can amplify the effects of OT (Li et al., 2023).

1.7.3. Gustation

Gustation (the sense of taste) comprises of five basic tastes: sweet, salty, sour, bitter, and umami (Lindemann, 1996). When a substance enters the mouth, it interacts with the gustatory papillae located on the tongue (Azzali et al., 1996). These lingual papillae are small structures composed of epithelial, neural, connective, and vascular tissues, and they contain taste buds (Gravina et al., 2013). Each taste bud consists of 50 to 100 taste receptor cells that extend toward the surface of the tongue, allowing them to detect gustatory molecules in the oral cavity (Kinnamon, 2012).

There are four types of lingual papillae: circumvallate, fungiform, and foliate papillae, which are involved in taste signal transduction, and filiform papillae, which do not participate in taste perception but contributes to touch, pain, and temperature sensation (Gravina et al., 2013). When gustatory stimuli bind to gustatory receptor cells, sensory information is transmitted via cranial nerves VII (facial), IX (glossopharyngeal), and X (vagus) (de Araujo & Simon, 2009). Cranial nerve VII participates in facial expressions and taste perception, cranial nerve IX contributes to taste and throat sensation, cranial nerve X extends to the digestive system and it is responsible from taste sensation from epiglottis and pharynx (de Araujo & Simon, 2009).

These cranial nerves convey gustatory signals to the nucleus of the solitary tract in the medulla (de Araujo & Simon, 2009). From there, the information is relayed to the thalamus and subsequently to the gustatory cortex. Gustatory input further converges with olfactory input in the far anterior insular cortex for perception of flavor and pleasantness (de Araujo et al., 2003).

Gustatory perception serves multiple functions, including helping people avoid toxic substances (Barlow, 2022), select and consume nutritious foods shaping feeding patterns and flavor preferences (Barlow & Klein, 2015). Importantly, gustation interacts with olfaction to construct the multisensory percept of flavor (Small & Prescott, 2005).

1.7.4. Vision

Vision is the sense of sight and a primary tool for navigating the external world. Visual perception begins when light waves reach the cornea. The cornea directs the incoming light toward the back of the eye, focusing it on the fovea centralis, the focal point located at the center of retina (Sanchez Lopez de Nava et al., 2025). The retina contains millions of photoreceptors that are called cones and rods. Cones encode color information, whereas rods are more tuned to low light levels (Sanchez Lopez de Nava et al., 2025).

Signals generated in the retina are transmitted through cranial nerve II (the optic nerve), however, the retinal image is inverted in the "upside-down" form and is corrected by subsequent neural processing in the brain (Baden & Nilsson, 2022). The optic nerve travels to the optic chiasm, located inferior to hypothalamus at the base of the brain (Ireland & Carter, 2025). Beyond this point, the optic tract, an extension of the optic nerve, relays visual information to the lateral geniculate nucleus in the thalamus (Mehra & Moshirfar, 2025). From the LGN, visual input is projected to the primary visual cortex (V1 or striate cortex), located in the occipital lobe (Mehra & Moshirfar, 2025).

From V1, visual information diverges into cortical pathways depending on the cognitive processes involved. The ventral stream supports object recognition, memory-guided actions, and conscious visual perception, whereas the dorsal stream supports spatial processing, attention, and real-time action guidance (Milner, 2017).

Vision plays an important role in short- and long-term visual memory (Schurgin, 2018), mental imagery (Baddeley & Andrade, 2000), mnemonic performance (Keogh & Pearson, 2011), navigation in the external environment (Ekstrom, 2015), maintenance of balance (Villicana et al., 2025), and learning (Bobek & Tversky, 2016).

1.7.5. Olfaction – gustation interaction

Olfaction and gustation have a unique interaction because they jointly construct the percept of flavor (Small & Prescott, 2005). Although "taste" and "flavor" are often used interchangeably in everyday language, i.e., describing odors with taste qualities (Rozin, 1982), gustation alone does

not account for flavor perception. This confusion arises from the olfactory location illusion, in which odor-derived sensations are misattributed as originating in the mouth rather than nose (Prescott, 2012, Rozin, 1982). For this reason, our study used retronasal stimuli (droplets delivered to the back of throat containing both olfactory and gustatory properties) to minimize this perceptual illusion and avoid confusing participants.

Beyond flavor perception, olfaction and gustation interact in additional ways. Congruent odor-taste combinations can alter detection thresholds; for instance, co-stimulation with benzaldehyde (cherry/almond odor) and monosodium glutamate (salty) produces higher thresholds compared to benzaldehyde paired with saccharin (sweet) condition (Dalton et al., 2000). Congruent olfactory-gustatory combinations can also enhance flavor intensity, particularly in patients with chemosensory impairments and such sensory amplification has been proposed as a potential therapeutic tool for treating related sensory deficits (Schiffman & Graham, 2000).

Odors can also influence taste perception directly. For example, congruent strawberry-sucrose co-stimulation facilitates taste detection compared with incongruent ham-sucrose pairing (Djordjevic et al., 2004a). This facilitation can occur even when the odor is only imagined rather than physically presented (Djordjevic et al., 2004a). Congruent odor-taste pairings can also enhance taste identification (White & Prescott, 2007), increase perceived sweetness when strawberry odor accompanies sucrose, decrease perceived sourness when strawberry odor accompanies citric acid (Stevenson et al., 1999), and reduce perceived saltiness when paired with soy sauce (Djordjevic et al., 2004b).

Neurobiological evidence further supports the strong interaction between gustation and olfaction. In rats, functional olfactory receptor cells have been detected in taste papillae, suggesting that gustatory taste cells may serve as an early integration site for odor-taste information (Malik et al., 2019). In macaque monkeys, multimodal neurons in the orbitofrontal cortex respond to congruent odor-taste combinations (Rolls & Baylis, 1994, Prescott, 2004), while gustatory cortex neurons can respond to retronasal odors (Blankenship et al., 2019).

Human neuroimaging studies also reveal substantial overlap in brain regions of taste and smell. Shared activation is observed in the secondary olfactory cortex (orbitofrontal cortex) (Small et al.,

1997), insula (Cerf-Ducastel & Murphy, 2001), and anterior cingulate cortex (Small et al., 2003). In rats, extracellular recordings show that intraoral stimuli can evoke overlapping olfactory, gustatory, and oral-somatosensory responses in both olfactory and gustatory cortices, with convergence occurring in both modality and temporal response patterns (Maier, 2017). This integrative relationship is further demonstrated through lesion studies, where damage to the gustatory cortex impairs odor perception and identification, indicating the interconnectedness of gustatory and piriform cortices (Maier et al., 2015).

1.7.6. Olfaction – vision interaction

Olfactory and visual senses can influence one another across various perceptual conditions and tasks. In congruent visual-olfactory stimulations, visual cues can facilitate olfactory perception (Gottfried & Dolan, 2003), modify perceived odors intensity (Zellner & Kautz, 1990) and pleasantness (Osterbauer et al., 2005), increase accuracy in odor identification (Zellner et al., 1991), and improve odor discrimination performance (Dematte et al., 2009).

Odors can also enhance performance in visual search tasks when the odor is congruent with the visual target; participants locate the visual object more rapidly under these conditions (Seigneuric et al., 2010). Odors may bias in participants' visual judgements, such as increasing the selection of grey or brown colors when certain odors are present (Ward et al., 2023). Moreover, olfactory cues can modulate visual perception even without prior associative learning; for example, lemon odor has been shown to bias participants toward perceiving slower dot movement, whereas vanilla odor leads to the perception of faster movement (Tsushima et al., 2021).

Neuroimaging findings support these interactions. Functional MRI studies demonstrate cross-modal congruency effects in regions such as the right precuneus for memory related visual imagery (Ripp et al., 2018). Increased activity in the orbitofrontal cortex and insular cortex is associated with higher congruency perception between odors and visual stimuli (Osterbauer et al., 2005). Additional regions involved include the hippocampus (Gottfried & Dolan, 2003), right amygdala, and piriform cortex (Schicker et al., 2025). Modulations of response patterns have also been observed in the primary visual cortex (V1) during congruent and incongruent olfactory-visual stimulation (Tsushima et al., 2021). In this experiment, participants reported the speed of

moving dots; they perceived slower motions with a lemon smell and faster motions with a vanilla smell without prior training. This behavior indicates an innate but not acquired behavior. In the fMRI scanning, participants' brain activities in the medial temporal area were significantly higher for vanilla compared to lemon smells.

1.7.7. Olfaction – gustation – vision interaction

The three-way interaction between olfaction, gustation, and vision has been rarely described in literature. In one example, an augmented reality experiment presented participants with three conditions: (1) visual stimuli alone, (2) olfactory stimuli alone, and (3) combined olfactory-visual stimuli. Participants were also given a flavorless coffee creamer and asked to evaluate the overall flavor (rated as correct or incorrect). The multisensory visual-olfactory condition had the strongest influence on participants' beliefs about the coffee flavor, with olfaction contributing more prominently than vision. Accuracy in perceiving the coffee flavor increased in multisensory conditions compared with either unimodal condition, whereas participants were more likely to misidentify the flavor when stimuli were presented unimodally (Halabi & Saleh, 2021).

At the neural level, multimodal neurons responding to gustatory, olfactory, and visual stimuli or to discrimination tasks involving these stimuli have been identified in the orbitofrontal cortex of macaque monkeys through microelectrode single-neuronal recordings (Rolls & Baylis, 1994). Similar integrative processes have been observed in rats trained in flavor discrimination, taste detection, and odor detection tasks before the removal of certain brain regions. Rats were divided into three groups: (1) experimental group, removal of the left OB and right orbitofrontal and insular cortices, (2) unilateral group, the same removals but limited to the right hemisphere, and (3) sham-control group, anesthetized and incised but without tissue removal. While no significant differences were found between groups for odor and taste detection, rats in the experimental group required significantly longer times to complete the flavor task compared to both control and unilateral groups (Schul et al., 1996). These results suggest that cooperative neural processing between olfactory, gustatory, and visual modalities is essential for efficient flavor perception.

Additional evidence supports cross-modal convergences among these senses. The orbitofrontal cortex receives associative and integrative input from visual stimuli (Chaumon et al., 2014), and

the insular cortex has been shown to have reduced cortical thickness in patients with vision loss (Chamard et al., 2022). Together, these findings align with the structures implicated in the study by Schul, et.al., highlighting the potential anatomical convergence of olfactory, gustatory, and visual pathways.

Given these synergistic interactions, dual and triple-modal combinations of olfaction, gustation, and vision may produce stronger sensory integration effects than unisensory stimulation. Therefore, to benefit from these fortifying multisensory interactions, we incorporated all three modalities into our MOT protocol to investigate whether this approach could enhance the recovery outcomes beyond those achieved through COT.

1.8. Methodological Considerations

In Article 1, our experimental design used the absence/presence and congruency/incongruency of three sensory modalities across multiple conditions. In some conditions, gustatory and/or visual stimuli were semantically matched or mismatched with the olfactory stimuli. In the triplet conditions, gustatory and visual stimuli were either both congruent, both incongruent, or one congruent, and the other incongruent relative to the olfactory stimulus. This approach allowed us to collect data from both dual and triple multisensory stimulations within a single experiment, facilitating direct comparisons across conditions. The design also enabled us to evaluate olfactory, gustatory, and visual perceptions separately, as well as in interaction. Importantly, the experiment was optimized to remain concise to avoid participant fatigue associated with prolonged testing sessions.

A noteworthy methodological characteristic of our research is the use of retronasal, rather than orthonasal olfactory stimulation. Participants did not sniff the stimuli; droplets were placed at the back of the throat. This method was intentionally chosen to simulate natural real-life experiences of flavor perception, where olfactory and gustatory information are typically integrated during consumption. Participants therefore evaluated the stimuli as they would during everyday eating or drinking. This method also allowed us to deliver olfactory and gustatory stimuli simultaneously without requiring specialized equipment such as gustometer. This same retronasal approach was

implemented in Article 2, where patients in the MOT group applied droplets onto the tongue rather than sniffing odors.

In Article 2, we not only compared MOT with COT, but we also included a placebo control group. The inclusion of a well-defined control condition strengthens the validity of randomized clinical trials and provides an essential baseline for evaluating treatment effects (Moser, 2020). Although spontaneous recovery from COVID-19-related OD is expected in some cases (Dias et al., 2024), the placebo group in our study did not show significant improvement in olfactory function compared to the multisensory or classical OT groups.

1.9. Aims and Hypotheses

Our overarching aim was to support recovery from OD in the context of COVID-19 by implementing a MOT paradigm. Based on the existing body of literature, we hypothesized that MOT using congruent olfactory, gustatory, and visual stimuli would enhance recovery of olfactory abilities in patients experiencing post-COVID-19 olfactory loss. We hypothesized that introducing gustatory and visual co-stimulations into established OT protocol would better simulate the natural experience of smelling food-related odors and thereby improve training outcomes. For the gustatory stimuli, we selected the four basic tastes. For the olfactory stimuli, we selected four edible odorants: strawberry, cheese, lemon, and coffee to pair with congruent gustatory stimuli: sweet (strawberry), salty (cheese), sour (lemon), and bitter (coffee). Corresponding visual stimuli (images) were chosen to match each odor-taste pair.

Thus, the overall objective of this thesis was to investigate the effects of gustatory and visual congruency during olfactory co-stimulation and explore interactions among these three sensory modalities to develop a MOT protocol. We then aimed to evaluate the effectiveness of this new protocol in patients with long-term olfactory loss following COVID-19 infection.

Objective 1: To elucidate the effects of gustatory and visual co-stimulation on olfactory perception by measuring likeness rating, i.e., how well a label matches an odor, under congruent and incongruent conditions (Chapter 2).

Objective 2: To evaluate the effects of MOT in patients with long-term COVID-19-related OD, and to compare the effectiveness of MOT with COT using both objective and subjective measures (Chapter 3). Objective measure includes UPSIT test with olfactory identification scores, and subjective measure includes SQOD-NS test with quality-of-life scores.

Chapter 2 – Article 1 - Congruency of multisensory olfactory stimuli

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Abstract

We perceive our environment via different sensory channels in a multisensory fashion. During multisensory integration, these channels can enhance or hinder each other depending on congruency. Here, we aimed to investigate how the congruency of gustatory and visual costimulation alter the perception of olfactory stimuli. We hypothesized that congruent costimulation enhances the perception of likeness, i.e. how well stimuli match a label, compared with no and to incongruent costimulation. We also aimed to quantify the effect of gustatory and visual costimulation. We tested 48 healthy young participants. We used retronasal olfactory stimuli (strawberry, cheese, lemon, and coffee) (i) alone or with congruent/incongruent costimulation ((ii) with congruent gustatory (sweet, salty, sour, and bitter), (iii) with congruent visual (images of strawberry, cheese, lemon, and coffee), (iv) with congruent visual and gustatory, (v) with congruent visual and incongruent gustatory, (vi) with congruent gustatory and incongruent visual, (vii) with incongruent

visual and gustatory costimulations). Olfactory and gustatory stimuli were presented by means of droplets on the tongue, i.e. olfactory stimuli were delivered retronasally, while visual stimuli were presented on a computer screen. We asked participants to evaluate the solutions' likeness to the respective olfactory label on visual analog scales. We observed a significant effect of congruency on likeness ($P < 0.001$). Gustatory costimulation had a significantly stronger effect than visual costimulation ($P = 0.02$). Congruent costimulation enhances the evaluation of likeness while incongruent costimulation reduces it, with gustatory costimulation having significantly stronger effects than visual costimulation. This could be useful in multisensory olfactory training paradigms for olfactory loss.

Introduction

We perceive our surroundings as multisensory percepts, based on mental models, preconceived knowledge about the com plex scenes, objects, events from the world (Klasen et al. 2012; Duncan 2025). Multisensory perception is the combination of sensory modalities into one percept (Driver and Spence 2000). Congruent sensory input corresponds to previous experiences and semantic knowledge (Gau and Noppeney 2016). For example, we expect strawberries to be red, straw berry-shaped, sweet, etc., in line with our pre-existing knowledge of this fruit. However, if we are presented with a blue colored or bitter tasting strawberry, the incongruent costimulation interferes resulting in the perception of an object that corresponds less to stored features of the object. Therefore, in congruent costimulation in other modalities leads olfactory stimuli to be perceived as less familiar (Prescott 1999; Labbe et al. 2006), less intense (Zampini et al. 2008), and to be detected only at higher concentrations (Dalton et al. 2000) and more slowly (Wongtrakun et al. 2024) while congruent costimulation in other modalities leads olfactory stimuli to be perceived as more intense, faster, and more accurately (Ernst and Bulthoff 2004; Laurienti et al. 2004; Kim et al. 2008; Roberts et al. 2024). As a consequence, when white wine is colored red, even experienced participants describe its sensory proper ties with labels that are normally attributed to red wine (Morrot et al. 2001). In other words, incongruently colored drinks (e.g. green colored, orange flavored drinks) are identified less accurately (Zampini et al. 2007). In turn, when combined with congruent tastants (e.g. sweet saccharin), odorants (e.g. almond smelling benzaldehyde) are more intense and can be perceived even at levels below perception threshold of

monomodal olfactory stimuli (Dalton et al. 2000). Consequently, sensory training with congruent olfactory-visual stimuli leads to lower detection thresholds as well as better performance in discrimination (Li et al. 2023) and memory tasks (Olofsson et al. 2020).

Olfactory training, i.e. self-administration of a limited number (e.g. 4) of odorants regularly (e.g. twice a day) and repeatedly (e.g. for 12 wk) (Hummel et al. 2009) is the most promising intervention in olfactory dysfunction following viral infection (Hummel et al. 2009; Vance et al. 2024). In its present form, olfactory training is typically carried out with a unimodal olfactory design. Nevertheless, since congruent multisensory stimuli are perceived as more intense, olfactory training with multimodal stimuli may be more efficient than with monomodal stimuli (Filiz et al. 2024).

Therefore, we aimed to investigate how congruency of multisensory stimuli influences perception in individuals with a normal sense of smell. More specifically, we aimed to determine how congruent and incongruent gustatory and visual stimuli affect likeness i.e. how well an olfactory stimulus matches its label. We hypothesized congruent gustatory and/ or visual stimuli to increase likeness, while incongruent gustatory and/or visual stimuli decrease likeness. This also allows for estimating the magnitude of the impact of congruency in the different sensory channels.

Materials and methods

Participants

This research was approved by UQTR's ethics board. Participants provided written consent before the experiment. We recruited 50 healthy participants between 18 and 35. Due to technical issues, we had to exclude the data of two participants; the final sample therefore consisted of 23 women and 25 men (average age: 26 years; standard deviation: 4.1). We assessed demographics of our participants (age, gender, health conditions including allergies, COVID history, history of olfactory dysfunction, and history of nasal surgery).

Stimuli

We used four odorants (strawberry, cheese, lemon, and coffee flavors) and four tastants (sucrose (sweet), sodium chloride (salty), citric acid (sour), and sucrose octa-acetate (bitter)). We opted

not to include umami as a taste quality because of its relative unfamiliarity (See Table 1for an overview of manufacturers; see Table 2 for an overview of the congruency matching of stimuli). Chemosensory (olfactory and gustatory) stimuli were premixed in amber opaque glass vials (30 mL, Fisherbrand Inc, USA). We further presented participants with images of strawberry, cheese, lemon, coffee (Pixabay) on a computer screen by using Psychopy, GNU (GPL v3+, 2023).

Modality	Stimulus	Manufacturer; #	Volume (olfaction)/ Amount (gustation)
Olfaction	Strawberry flavor	Foodarom; MET0003559	3mL
	Cheese flavor	Foodarom; MET0017403	3mL
	Lemon flavor	Foodarom; MET0000055	3ml
	Coffee flavor	Foodarom; MET0017403	1.5mL
Gustation	Sucrose	Acros; 424500010	0.9g
	Sodium chloride	BDH; 127038.119541	0.3g
	Citric acid	Milliard; X000HT86Q5	0.3g
	Sucrose octa-acetate	Sigma Aldrich; W303801	0.0015g

Table 2.1. Characteristics of olfactory and gustatory stimuli. *Manufacturers: Foodarom: Foodarom Glanbia Nutritionals, St. Hubert, QC, Canada; Acros: Acros Organics, Thermo Fischer Scientific, New Jersey, USA; BDH: Inc. LOT, Toronto, Ontario, Canada; Milliard: Milliard Brands, New Jersey, USA; Sigma-Aldrich: Sigma-Aldrich, Oakville, Ontario, Canada. Olfactory/ gustatory stimuli were diluted in 30mL of demineralized water.*

	Gustatory co-stimulation			
Olfactory stimulus	Sweet	Salty	Sour	Bitter
Strawberry	Congruent	Incongruent	Incongruent	Incongruent
Cheese	Incongruent	Congruent	Incongruent	Incongruent
Lemon	Incongruent	Incongruent	Congruent	Incongruent
Coffee	Incongruent	Incongruent	Incongruent	Congruent

Table 2.2. Congruency or incongruency of each co-stimulation.

Conditions

We presented stimuli in seven conditions. Specifically, olfactory stimuli were delivered either without costimulation or with congruent/incongruent, gustatory and/or visual costimulation. The conditions consisted of mixing and matching of four olfactory stimuli (aromas of strawberry, cheese, lemon, and coffee), with five gustatory stimuli (sweet, salty, acid, bitter, and tasteless baseline) and five images as visual stimuli (strawberry, cheese, lemon, coffee, and blank image baseline). Chemosensory stimuli were presented as droplets on the tongue with dropper lids (Filiz et al. 2024). Olfactory stimuli were therefore delivered retronasally. In parallel, participants looked at a computer screen for the presentation of the visual stimuli.

In the following, G indicates the gustatory costimulation; V indicates the visual costimulation; 0 indicates the baseline condition, c indicates a congruent costimulation; i indicates an in congruent costimulation; c and i always refer to the olfactory stimulus. For example, for a congruent stimulation in all three modalities (GcVc) we presented, e.g. strawberry aroma with sweet taste and a strawberry image, while for an incongruent stimulation in all 3 modalities (GiVi), we presented, e.g. cheese flavor with acid taste and a coffee image. For partly incongruent/congruent stimulations we presented, e.g. coffee flavor with bitter taste (congruent) and a cheese image (incongruent), which was coded as GcVi. G0V0 in turn represents the baseline condition without costimulation.

Procedure

We presented stimuli in a randomized order. Upon stimulation, we asked participants to evaluate the likeness of the stimulus to the four labels by the question: “How much like label (label: strawberry/cheese/lemon/coffee) is this mixture?” via four separate visual analog scales (VAS; ranging from 0: “not at all label” to 100: “completely label”) presented on the computer screen. We did not control when participants swallowed or spit out the droplets on their tongue. The visual stimuli stayed on the screen until the participants finished rating the VAS.

Stimuli were separated by a 40s interstimulus interval during which participants rinsed their mouth. Participants received a total amount of 25 different costimulations of stimuli in a total of 97 trials. For a given odorant (e.g. strawberry), we used five gustatory costimulations (one congruent one (Gc), here sweet (sucrose), 3 incongruent ones (Gi), here salty (sodium chloride), sour (citric acid), bitter (sucrose octa-acetate), and as well as a blank (tasteless water; G0)). Analogously, a given odorant was presented with five visual costimulations (one congruent one (Vc), here strawberry image; 3 incongruent ones (Vi), here cheese image, lemon image, coffee image, and 1 blank (gray screen; V0)). We did the analogous with the other odorants. This resulted in 4 (4 odorants) * 5 (5 gustatory costimulations) * 5 (5 visual costimulations) = 100 permutations. However, of these permutations only four are GcVc, the condition of interest, giving too much potential weight to outliers. We therefore repeated each of the four GcVc stimuli three times (12 stimuli), leaving us with 108 stimuli. In turn, since we deemed that the experiment would have taken too long, we took out the 12 stimuli that consisted of congruent gustatory-visual combinations (e.g. sweet/strawberry image) that were incongruent to the olfactory stimulus (e.g. cheese). We did the same with the 24 stimuli for which the costimulation of 1 modality was incongruent while the costimulation of the other modality was a blank (e.g. strawberry flavor with a cheese image and no gustatory costimulation). This resulted in a total of 72 stimuli that were presented to each participant. After each trial, participants rated the likeness to the categories (strawberry, lemon, coffee, and cheese) with four different VAS. However, for further analysis we only regarded the category that corresponded to the odor that had been presented.

The number of trials per condition is presented in Table 3. The experiment took 2 h to complete, and it was divided into 2 sessions with a 15-min break between. The experiment flow is presented in Fig. 1

	GcVc	GcV0	G0Vc	G0V0	GiVc	GcVi	GiVi
Trials	12	4	4	4	12	12	24

Table 2.3. Overview of items in different conditions.

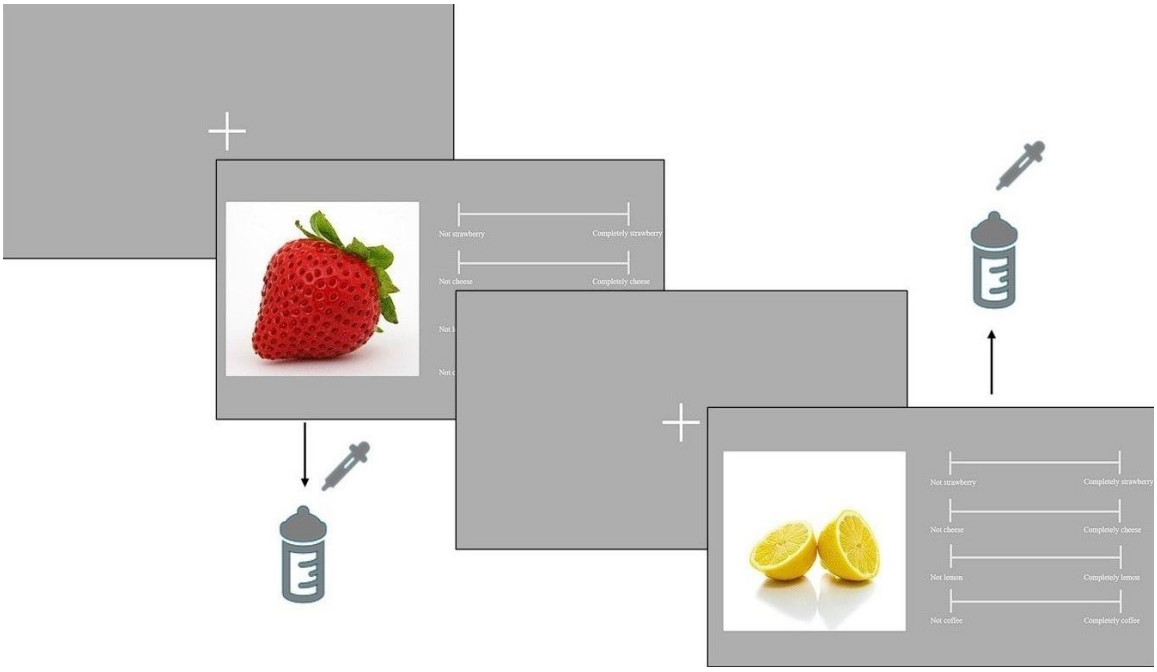


Figure 2.1. Flow chart of the experiment.

Statistical Analysis

We used SPSS 29 (IBM, Armonk, NY) for data analysis.

For the analysis we calculated average scores for the seven conditions across odors. We examined the effects of congruency of the modality of costimulation on likeness by computing 3 repeated measures (rm) ANOVA. In the rmANOVA, we investigated the effect of congruency by including congruent costimulations (4 levels: baseline G0V0 and GcVc, GcV0, and G0Vc). In the second analysis, we investigated the effect of incongruency by including incongruent costimulations (4 levels: baseline G0V0 and GiVc, GcVi, and GiVi). In the third analysis, we compared the effects of congruency and incongruency (3 levels: baseline G0V0 and GcVc, GiVi). In all ANOVA, we used gender as a between subject factor.

For post hoc tests we used Bonferroni corrections for multiple comparisons. We set the alpha value at 0.05.

Results

Average likeness scores are presented in Table 4.

The first rmANOVA with congruent stimuli revealed a significant effect of condition ($F(1, 2.4) = 15.3$, $P < 0.001$) but no significant effect of gender ($F(1, 48) = 1$, $P = 0.3$), and no interaction condition and gender ($F(1, 2.4) = 0.8$, $P = 0.5$).

Next, the rmANOVA with incongruent stimuli revealed a significant effect of condition ($F(1, 2.4) = 31$, $P < 0.001$), but no significant effect of gender ($F(1, 48) = 1.3$, $P = 0.3$) nor an interaction condition and gender ($F(1, 2.4) = 2.6$, $P = 0.07$). Finally, the rmANOVA with the incongruent and congruent costimulations, revealed a significant effect of condition ($F(1, 1.8) = 63.6$, $P < 0.001$) but no significant effect of gender ($F(1, 48) = 1.5$, $P = 0.2$) nor an interaction condition and gender ($F(1, 1.8) = 0.5$, $P = 0.6$).

Pairwise post hoc comparisons are presented in Fig. 2.

On average, baseline olfactory stimuli without costimulation had a likeness of 56.3 (3.6) points. Congruent gustatory costimulation increased likeness by 11.1 (3.2) points, while congruent visual costimulation increased likeness by 4.3 (3.4) points. As a result, combined congruent gustatory and visual costimulation increased likeness by 14.1 (2.8) points, while combined incongruent gustatory and visual costimulation decreased likeness by 8.1 (2.8) points. Mixed congruency, e.g. congruent gustatory and incongruent visual costimulation resulted in an increase of likeness by 9.5 (2.8) points, while incongruent gustatory and congruent visual costimulation decreased likeness by 2.6 (2.8) points.

	Likeness (points)	Standard Deviation
GcVc	70.3	2.8
GcV0	67.3	3.1
G0Vc	60.5	3.4
G0V0	56.2	3.6
GiVc	53.6	2.8
GcVi	65.7	2.8
GiVi	48.1	2.8

Table 2.4. Average likeness scores of different conditions.

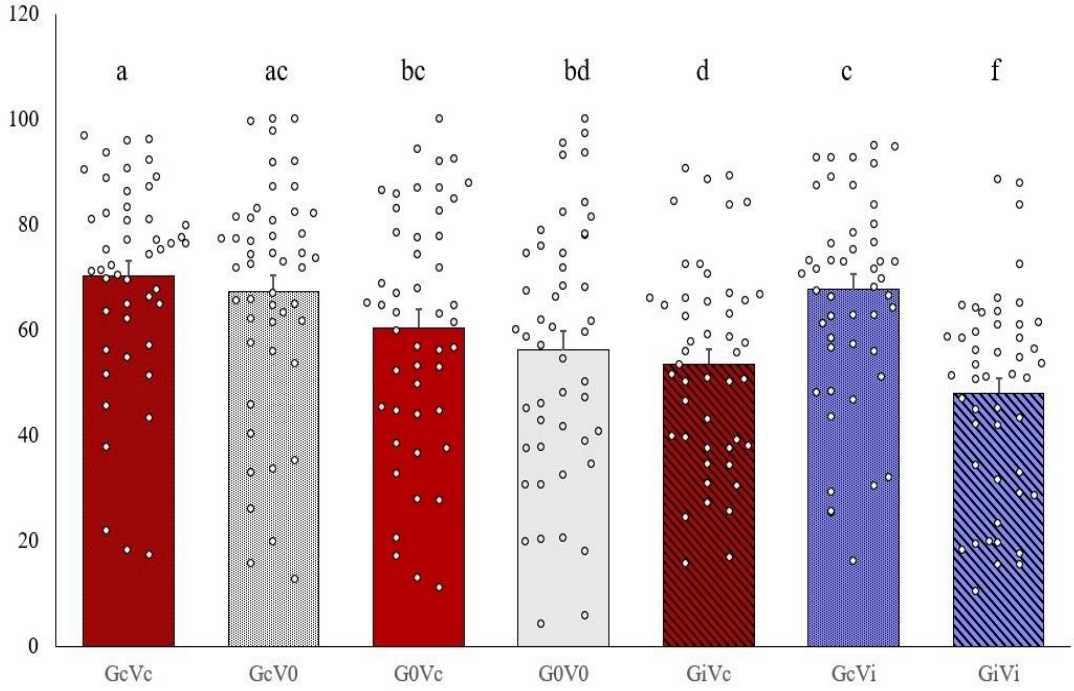


Figure 2.2. Average likeness scores of different conditions. *Error bars represent standard deviation. Significant differences in the pairwise comparisons are indicated by different letters. If two bars contain the same letter, they do not have a significant difference. Bars that do not share the same letter are significantly different.*

Discussion

Here, we report the results of our study on the effects of congruency of gustatory and visual costimulation on retronasal olfactory stimuli. Our main results are (i) congruency increases the perceived likeness of olfactory stimuli; (ii) congruency (incongruency) of gustatory costimulation is more effective in increasing (decreasing) likeness of olfactory stimuli compared with visual costimulation.

We observed that gustatory costimulation affects the likeness of an olfactory stimulus to a label. This is in line with the literature. For example, sweetness is a great enhancer of odor intensity (Fujimaru and Lim 2013). When combined with a sweet taste, retronasally presented cherry flavor were rated as more cherry flavored when compared with a sour taste (Green et al. 2012), and peach flavors more peach flavored (Porto Cardoso and André Bolini 2008); a sweet taste renders a tomato puree to be perceived as more ripe, tropical and fruity (Baldwin et al. 2008). Similar effects have been reported for strawberries (Schwieterman et al. 2014) and citrus (King et al. 2007; Veldhuizen et al. 2017). These effects are not limited to intensity or likeness as participants also respond faster to congruent gustatory-olfactory mixtures compared with monomodal stimuli (Veldhuizen et al. 2010). Participants also report more pleasantness and odor referral to their mouth with the more familiar and congruent stimuli they receive (Fondberg et al. 2018). These results can also be observed when participants were not exposed to the stimuli in training sessions before the experiment, suggesting the effect of congruency in learning associations during experiments (Stevenson et al. 1998). Furthermore, when presented with a congruent gustatory costimulation, olfactory stimuli can be detected at concentrations that are below their monomodal perception thresholds (Dalton et al. 2000; Delwiche and Heffelfinger 2005).

We observed a weaker yet significant effect for visual costimulation. This is again in line with the literature. Red colored tangerine–guava–pineapple mixtures are rated as more fruity in both orthonasal and retronasal odor quality assessments (Koza et al. 2005). Similarly, odorant solutions with matching colors, e.g. red-strawberry or green-mint odorant (Zellner and Whitten 1999), are rated as more intense (Zellner and Kautz 1990). The effect of color congruency of visual congruency extends to higher order olfactory tasks such as odor identification: congruently colored odorants are more easily identified than incongruent ones (Zellner et al. 1991). Similar results are observed when images are presented rather than colors alongside odors. Congruent images lead to increased likeness perception and easier odor detection (Gottfried and Dolan 2003). Congruent visual costimulations with images (e.g. flowers and fruits) led to smaller N400 components in event-related potentials in response to olfactory stimulation (e.g. rose and citrus). This component is thought to reflect the relatedness of stimuli (Grigor et al. 1999; Sarfarazi et al. 1999). In a

similar fashion, visualization of our current results in EEG would be interesting to investigate in the future studies.

Gustatory costimulation had a significantly stronger effect than visual costimulation. This further supports the notion of a strong connection between olfaction and gustation (Czarnecki and Fontanini 2019). The neurobiological underpinning of this may be in the overlap in brain areas responsible for processing of olfaction and gustation such as orbitofrontal cortex (Small et al. 1997), insula and operculum (Small et al. 1999; Cerf-Ducastel and Murphy 2001; Mastinu et al. 2025), anterior cingulate cortex (Small et al. 2003). Similar results can be observed in animal studies, the piriform cortex of rats responds to both olfactory and gustatory stimulation (Maier et al. 2012); in turn, their gustatory cortex also processes particularly retronasal (Blankenship et al. 2019) olfactory stimuli (Maier et al. 2015).

The observations in this study are potentially interesting for olfactory training in a clinical context. Here, olfactory training is a self-administered intervention that consists of sniffing selected odors for 12 wk (Hummel et al. 2009). Previously, we investigated whether a multisensory version of this training with olfactory, gustatory and visual stimuli allows for recovery of olfactory function following COVID-induced olfactory dysfunction. We found multisensory olfactory training to be equally effective as a classical olfactory training (Filiz et al. 2024). This is in line with previous findings with multisensory olfactory trainings in which researchers found significant improvement with olfactory-visual multisensory trainings (Khan et al. 2023; Li et al. 2023) and gustatory-olfactory multisensory trainings (Fjaeldstad 2025). Our current study suggests that the stimuli used in the present study may be better suited for multisensory training because of higher volumes and amounts of olfactory/gustatory stimuli. We had noticed that patients in our previous study struggled to detect odors.

This is an innovative study because most previous research investigated the effect of one additional modality on olfactory perception such as vision on olfaction, e.g. (Morrot et al. 2001; Sakai et al. 2005; Stevenson and Oaten 2008; Dematte et al. 2009) or gustation on olfaction (Green et al. 2012; Lim et al. 2014; Amsellem and Ohla 2016). Here, we investigated both gustation and vision, and this independently and combined, which allows us to compare their individual effects,

but also their synergistic abilities, on flavor perception. Further, while previous research was mostly concerned with the effects of multisensory integration during consumption of food or drinks, e.g. (Fujimaru and Lim 2013; Sukkhown et al. 2019; Wang et al. 2020; Stager et al. 2021) our study was designed with olfactory training protocols in mind (Hummel et al. 2009; Filiz et al. 2024). Our study's stimuli can directly be used in olfactory training protocols. This is particularly interesting given the raised awareness in the public on olfactory dysfunction after viral infections in the aftermath of the pandemic (Asseo et al. 2020; Cheng et al. 2021; Saegner and Austys 2022; Ziakas and Mylonakis 2024). Finally, we feel that our approach that used the same costimulations as congruent or incongruent, depending on the olfactory stimulation, allowing them to serve as controls for themselves rather elegant.

This study has some limitations. First, to limit testing time we did not include certain conditions (e.g. incongruent gustatory stimuli without visual stimulation) rendering the generalizability of findings on incongruent costimulation are relatively limited. Second, we predefined congruent and incongruent costimulations without verifying in participants if this was the case with everyone. This is particularly true since there may be important effects of cultural background on congruency. For example, bitter taste may not be perceived as congruent with coffee aroma if a participant is used to drink their coffee strongly sweetened. This is why we included the same stimuli in the different conditions. By doing so, we considered a stimulus (e.g. sweet) congruent when presented with one odor (e.g. strawberry), but when presented with another condition (e.g. lemon), the same stimulus was considered as incongruent. In other words, the same stimuli served as congruent intervention and as incongruent control. The fact that we observe significant differences between the conditions, actually shows that the stimuli we considered congruent were perceived, on average, as congruent by the participants. This does however not rule out that other stimuli might have had even stronger effects. Future studies should evaluate congruency on a participant-by-participant basis. Third, we did not assess if the stimuli also stimulated the trigeminal system as this was outside the scope of our experiment. The presence of a minor trigeminal costimulation, however, would not change the main message of the paper. Fourth, it is important to point out that we used food-related odors. In the classical olfactory training protocol rose, eucalyptus, clove, and lemon odors are used, which are not typical food odors. One

would not expect non-food odors to be congruent with gustatory costimulations, thereby limiting the applicability of our observations. Previous studies suggest that the choice of odors does not have a predominant importance for the outcome of olfactory training (Altundag et al. 2015; Poletti et al. 2017). Therefore, opting for food odors may be appropriate when including gustatory costimulation. Finally, we excluded umami as a gustatory stimulus since pilot testing suggested that the studied population had difficulties in recognizing it correctly, in line with the literature (Singh et al. 2010; Cecchini et al. 2019).

Conclusion

In conclusion, we show that congruent matching of vision and gustation increases perceived likeness and potentially recognition of multisensory solutions. Gustation especially plays a significant role and matching when presented alongside olfactory stimuli. The results from this study might potentially be useful in multisensory training paradigms such as multisensory olfactory trainings.

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Data Availability

Data cannot be shared for ethical/privacy reasons.

References

Altundag, A., Cayonu, M., Kayabasoglu, G., Salihoglu, M., Tekeli, H., Saglam, O., & Hummel, T. (2015). Modified olfactory training in patients with postinfectious olfactory loss. Laryngoscope, 125(8), 1763-1766. https://doi.org/10.1002/lary.25245

Amsellem, S., & Ohla, K. (2016). Perceived Odor-Taste Congruence Influences Intensity and Pleasantness Differently. Chem Senses, 41(8), 677-684. https://doi.org/10.1093/chemse/bjw078

Asseo, K., Fierro, F., Slavutsky, Y., Frasnelli, J., & Niv, M. Y. (2020). Tracking COVID-19 using taste and smell loss Google searches is not a reliable strategy. Sci Rep, 10(1), 20527. https://doi.org/10.1038/s41598-020-77316-3

Baldwin, E. A., Goodner, K., & Plotto, A. (2008). Interaction of volatiles, sugars, and acids on perception of tomato aroma and flavor descriptors. J Food Sci, 73(6), S294-307. https://doi.org/10.1111/j.1750-3841.2008.00825.x

Blankenship, M. L., Grigorova, M., Katz, D. B., & Maier, J. X. (2019). Retronasal Odor Perception Requires Taste Cortex, but Orthonasal Does Not. Curr Biol, 29(1), 62-69 e63. https://doi.org/10.1016/j.cub.2018.11.011

Cecchini, M. P., Knaapila, A., Hoffmann, E., Boschi, F., Hummel, T., & Iannilli, E. (2019). A cross-cultural survey of umami familiarity in European countries. Food Quality and Preference, 74, 172-178. https://doi.org/https://doi.org/10.1016/j.foodqual.2019.01.017

Cerf-Ducastel, B., & Murphy, C. (2001). fMRI activation in response to odorants orally delivered in aqueous solutions. Chem Senses, 26(6), 625-637. https://doi.org/10.1093/chemse/26.6.625

Cheng, I. K., Heyl, J., Lad, N., Facini, G., & Grout, Z. (2021). Evaluation of Twitter data for an emerging crisis: an application to the first wave of COVID-19 in the UK. Sci Rep, 11(1), 19009. https://doi.org/10.1038/s41598-021-98396-9

Czarnecki, L., & Fontanini, A. (2019). Gustation and Olfaction: The Importance of Place and Time. Current Biology, 29(1), R18-R20. https://doi.org/https://doi.org/10.1016/j.cub.2018.11.038

Dalton, P., Doolittle, N., Nagata, H., & Breslin, P. A. S. (2000). The merging of the senses: integration of subthreshold taste and smell. Nature Neuroscience, 3(5), 431-432. https://doi.org/10.1038/74797

Delwiche, J. F., & Heffelfinger, A. L. (2005). CROSS-MODAL ADDITIVITY OF TASTE AND SMELL. Journal of Sensory Studies, 20(6), 512-525. https://doi.org/https://doi.org/10.1111/j.1745-459X.2005.00047.x

Dematte, M. L., Sanabria, D., & Spence, C. (2009). Olfactory discrimination: when vision matters? Chem Senses, 34(2), 103-109. https://doi.org/10.1093/chemse/bjn055

Driver, J., & Spence, C. (2000). Multisensory perception: Beyond modularity and convergence. Current Biology, 10(20), R731-R735. https://doi.org/https://doi.org/10.1016/S0960-9822(00)00740-5

Duncan, J. (2025). Construction and use of mental models: Organizing principles for the science of brain and mind. Neuropsychologia, 207, 109062. https://doi.org/10.1016/j.neuropsychologia.2024.109062

Ernst, M. O., & Bulthoff, H. H. (2004). Merging the senses into a robust percept. Trends Cogn Sci, 8(4), 162-169. https://doi.org/10.1016/j.tics.2004.02.002

Filiz, G., Berube, S., Demers, C., Cloutier, F., Chen, A., Pek, V., Hudon, E., Bolduc-Begin, J., & Frasnelli, J. (2024). Can Multisensory Olfactory Training Improve Olfactory Dysfunction Caused by COVID-19? Multisens Res, 37(4-5), 299-316. https://doi.org/10.1163/22134808-bja10127

Fjaeldstad, A. W. (2025). Culinary cure? Improved subjective and measured sense of smell after a cooking rehabilitation program for patients with smell loss. International Journal of Gastronomy and Food Science, 39, 101121. https://doi.org/https://doi.org/10.1016/j.ijgfs.2025.101121

Fujimaru, T., & Lim, J. (2013). Effects of Stimulus Intensity on Odor Enhancement by Taste. Chemosensory Perception, 6(1), 1-7. https://doi.org/10.1007/s12078-013-9140-5

Gau, R., & Noppeney, U. (2016). How prior expectations shape multisensory perception. Neuroimage, 124(Pt A), 876-886. https://doi.org/10.1016/j.neuroimage.2015.09.045

Gottfried, J. A., & Dolan, R. J. (2003). The Nose Smells What the Eye Sees: Crossmodal Visual Facilitation of Human Olfactory Perception. Neuron, 39(2), 375-386. https://doi.org/https://doi.org/10.1016/S0896-6273(03)00392-1

Green, B. G., Nachtigal, D., Hammond, S., & Lim, J. (2012). Enhancement of retronasal odors by taste. Chem Senses, 37(1), 77-86. https://doi.org/10.1093/chemse/bjr068

Grigor, J., Van Toller, S., Behan, J., & Richardson, A. (1999). The effect of odour priming on long latency visual evoked potentials of matching and mismatching objects. Chem Senses, 24(2), 137-144. https://doi.org/10.1093/chemse/24.2.137

Hummel, T., Rissom, K., Reden, J., Hahner, A., Weidenbecher, M., & Huttenbrink, K. B. (2009). Effects of olfactory training in patients with olfactory loss. Laryngoscope, 119(3), 496-499. https://doi.org/10.1002/lary.20101

Khan, A. M., Piccirillo, J., Kallogjeri, D., & Piccirillo, J. F. (2023). Efficacy of Combined Visual-Olfactory Training With Patient-Preferred Scents as Treatment for Patients With COVID-19 Resultant Olfactory Loss: A Randomized Clinical Trial. JAMA Otolaryngol Head Neck Surg, 149(2), 141-149. https://doi.org/10.1001/jamaoto.2022.4112

Kim, R. S., Seitz, A. R., & Shams, L. (2008). Benefits of stimulus congruency for multisensory facilitation of visual learning. PLoS One, 3(1), e1532. https://doi.org/10.1371/journal.pone.0001532

King, B. M., Duineveld, C. A. A., Arents, P., Meyners, M., Schroff, S. I., & Soekhai, S. T. (2007). Retronasal odor dependence on tastants in profiling studies of beverages. Food Quality and Preference, 18(2), 286-295. https://doi.org/https://doi.org/10.1016/j.foodqual.2005.11.005

Klasen, M., Chen, Y. H., & Mathiak, K. (2012). Multisensory emotions: perception, combination and underlying neural processes. Rev Neurosci, 23(4), 381-392. https://doi.org/10.1515/revneuro-2012-0040

Koza, B. J., Cilmi, A., Dolese, M., & Zellner, D. A. (2005). Color enhances orthonasal olfactory intensity and reduces retronasal olfactory intensity. Chem Senses, 30(8), 643-649. https://doi.org/10.1093/chemse/bji057

Labbe, D., Damevin, L., Vaccher, C., Morgenegg, C., & Martin, N. (2006). Modulation of perceived taste by olfaction in familiar and unfamiliar beverages. Food Quality and Preference, 17(7), 582-589. https://doi.org/https://doi.org/10.1016/j.foodqual.2006.04.006

Laurienti, P. J., Kraft, R. A., Maldjian, J. A., Burdette, J. H., & Wallace, M. T. (2004). Semantic congruence is a critical factor in multisensory behavioral performance. Exp Brain Res, 158(4), 405-414. https://doi.org/10.1007/s00221-004-1913-2

Li, Z., Anne, A., & Hummel, T. (2023). Olfactory training: effects of multisensory integration, attention towards odors and physical activity. Chemical Senses, 48, bjad037. https://doi.org/10.1093/chemse/bjad037

Lim, J., Fujimaru, T., & Linscott, T. D. (2014). The role of congruency in taste–odor interactions. Food Quality and Preference, 34, 5-13. https://doi.org/https://doi.org/10.1016/j.foodqual.2013.12.002

Maier, J. X., Blankenship, M. L., Li, J. X., & Katz, D. B. (2015). A Multisensory Network for Olfactory Processing. Curr Biol, 25(20), 2642-2650. https://doi.org/10.1016/j.cub.2015.08.060

Maier, J. X., Wachowiak, M., & Katz, D. B. (2012). Chemosensory convergence on primary olfactory cortex. J Neurosci, 32(48), 17037-17047. https://doi.org/10.1523/JNEUROSCI.3540-12.2012

Mastinu, M., Thaploo, D., Warr, J., & Hummel, T. (2025). Cortical Representation of Food-Related Odors in Gustatory Areas Differs According to Their Taste Association: An fMRI Study. Brain Sci, 15(4). https://doi.org/10.3390/brainsci15040418

Morrot, G., Brochet, F., & Dubourdieu, D. (2001). The color of odors. Brain Lang, 79(2), 309-320. https://doi.org/10.1006/brln.2001.2493

Olofsson, J. K., Ekstrom, I., Lindstrom, J., Syrjanen, E., Stigsdotter-Neely, A., Nyberg, L., Jonsson, S., & Larsson, M. (2020). Smell-Based Memory Training: Evidence of Olfactory Learning and Transfer to the Visual Domain. Chem Senses, 45(7), 593-600. https://doi.org/10.1093/chemse/bjaa049

Poletti, S. C., Michel, E., & Hummel, T. (2017). Olfactory Training Using Heavy and Light Weight Molecule Odors. Perception, 46(3-4), 343-351. https://doi.org/10.1177/0301006616672881

Porto Cardoso, J. M., & André Bolini, H. M. (2008). DESCRIPTIVE PROFILE OF PEACH NECTAR SWEETENED WITH SUCROSE AND DIFFERENT SWEETENERS. Journal of Sensory Studies, 23(6), 804-816. https://doi.org/https://doi.org/10.1111/j.1745-459X.2008.00187.x

Prescott, J. (1999). Flavour as a psychological construct: implications for perceiving and measuring the sensory qualities of foods. Food Quality and Preference, 10(4), 349-356. https://doi.org/https://doi.org/10.1016/S0950-3293(98)00048-2

Roberts, K., Jentzsch, I., & Otto, T. U. (2024). Semantic congruency modulates the speed-up of multisensory responses. Sci Rep, 14(1), 567. https://doi.org/10.1038/s41598-023-50674-4

Saegner, T., & Austys, D. (2022). Forecasting and Surveillance of COVID-19 Spread Using Google Trends: Literature Review. Int J Environ Res Public Health, 19(19). https://doi.org/10.3390/ijerph191912394

Sakai, N., Imada, S., Saito, S., Kobayakawa, T., & Deguchi, Y. (2005). The effect of visual images on perception of odors. Chem Senses, 30 Suppl 1, i244-245. https://doi.org/10.1093/chemse/bjh205

Sarfarazi, M., Cave, B., Richardson, A., Behan, J., & Sedgwick, E. M. (1999). Visual event related potentials modulated by contextually relevant and irrelevant olfactory primes. Chem Senses, 24(2), 145-154. https://doi.org/10.1093/chemse/24.2.145

Schwieterman, M. L., Colquhoun, T. A., Jaworski, E. A., Bartoshuk, L. M., Gilbert, J. L., Tieman, D. M., Odabasi, A. Z., Moskowitz, H. R., Folta, K. M., Klee, H. J., Sims, C. A., Whitaker, V. M., & Clark, D. G. (2014). Strawberry flavor: diverse chemical compositions, a seasonal influence, and effects on sensory perception. PLoS One, 9(2), e88446. https://doi.org/10.1371/journal.pone.0088446

Singh, P. B., Schuster, B., & Seo, H. S. (2010). Variation in umami taste perception in the German and Norwegian population. European Journal of Clinical Nutrition, 64(10), 1248-1250. https://doi.org/10.1038/ejcn.2010.133

Small, D. M., Gregory, M. D., Mak, Y. E., Gitelman, D., Mesulam, M. M., & Parrish, T. (2003). Dissociation of neural representation of intensity and affective valuation in human gustation. Neuron, 39(4), 701-711. https://doi.org/10.1016/s0896-6273(03)00467-7

Small, D. M., Jones-Gotman, M., Zatorre, R. J., Petrides, M., & Evans, A. C. (1997). Flavor processing: more than the sum of its parts. Neuroreport, 8(18), 3913-3917. https://doi.org/10.1097/00001756-199712220-00014

Small, D. M., Zald, D. H., Jones-Gotman, M., Zatorre, R. J., Pardo, J. V., Frey, S., & Petrides, M. (1999). Human cortical gustatory areas: a review of functional neuroimaging data. Neuroreport, 10(1), 7-14. https://doi.org/10.1097/00001756-199901180-00002

Stager, L., Roel Lesur, M., & Lenggenhager, B. (2021). What Am I Drinking? Vision Modulates the Perceived Flavor of Drinks, but No Evidence of Flavor Altering Color Perception in a Mixed Reality Paradigm. Front Psychol, 12, 641069. https://doi.org/10.3389/fpsyg.2021.641069

Stevenson, R. J., Boakes, R. A., & Prescott, J. (1998). Changes in Odor Sweetness Resulting from Implicit Learning of a Simultaneous Odor-Sweetness Association: An Example of Learned Synesthesia. Learning and Motivation, 29(2), 113-132. https://doi.org/https://doi.org/10.1006/lmot.1998.0996

Stevenson, R. J., & Oaten, M. (2008). The effect of appropriate and inappropriate stimulus color on odor discrimination. Percept Psychophys, 70(4), 640-646. https://doi.org/10.3758/pp.70.4.640

Sukkhown, P., Pirak, T., Chonpracha, P., Ardoin, R., & Prinyawiwatkul, W. (2019). Seafood Flavor Perception, Liking, Emotion, and Purchase Intent of Coated Peanuts as Affected by Coating Color and Hydrolyzed Squid Peptide Powder. J Food Sci, 84(6), 1570-1576. https://doi.org/10.1111/1750-3841.14638

Vance, D. E., Del Bene, V. A., Kamath, V., Frank, J. S., Billings, R., Cho, D. Y., Byun, J. Y., Jacob, A., Anderson, J. N., Visscher, K., Triebel, K., Martin, K. M., Li, W., Puga, F., & Fazeli, P. L. (2024). Does Olfactory Training Improve Brain Function and Cognition? A Systematic Review. Neuropsychol Rev, 34(1), 155-191. https://doi.org/10.1007/s11065-022-09573-0

Veldhuizen, M. G., Shepard, T. G., Wang, M. F., & Marks, L. E. (2010). Coactivation of gustatory and olfactory signals in flavor perception. Chem Senses, 35(2), 121-133. https://doi.org/10.1093/chemse/bjp089

Veldhuizen, M. G., Siddique, A., Rosenthal, S., & Marks, L. E. (2017). Interactions of Lemon, Sucrose and Citric Acid in Enhancing Citrus, Sweet and Sour Flavors. Chem Senses, 43(1), 17-26. https://doi.org/10.1093/chemse/bjx063

Wang, Q. J., Meyer, R., Waters, S., & Zendle, D. (2020). A Dash of Virtual Milk: Altering Product Color in Virtual Reality Influences Flavor Perception of Cold-Brew Coffee. Front Psychol, 11, 595788. https://doi.org/10.3389/fpsyg.2020.595788

Wongtrakun, J., Zhou, S.-H., Bellgrove, M. A., Chong, T. T. J., & Coxon, J. P. (2024). The Effect of Congruent versus Incongruent Distractor Positioning on Electrophysiological Signals during Perceptual Decision-Making. The Journal of Neuroscience, 44(45), e2079232024. https://doi.org/10.1523/JNEUROSCI.2079-23.2024

Zampini, M., Sanabria, D., Phillips, N., & Spence, C. (2007). The multisensory perception of flavor: Assessing the influence of color cues on flavor discrimination responses. Food Quality and Preference, 18(7), 975-984. https://doi.org/https://doi.org/10.1016/j.foodqual.2007.04.001

Zampini, M., Wantling, E., Phillips, N., & Spence, C. (2008). Multisensory flavor perception: Assessing the influence of fruit acids and color cues on the perception of fruit-flavored beverages. Food Quality and Preference, 19(3), 335-343. https://doi.org/https://doi.org/10.1016/j.foodqual.2007.11.001

Zellner, D. A., Bartoli, A. M., & Eckard, R. (1991). Influence of color on odor identification and liking ratings. Am J Psychol, 104(4), 547-561. https://www.ncbi.nlm.nih.gov/pubmed/1793126

Zellner, D. A., & Kautz, M. A. (1990). Color affects perceived odor intensity. J Exp Psychol Hum Percept Perform, 16(2), 391-397. https://doi.org/10.1037//0096-1523.16.2.391

Zellner, D. A., & Whitten, L. A. (1999). The effect of color intensity and appropriateness on color-induced odor enhancement. Am J Psychol, 112(4), 585-604. https://www.ncbi.nlm.nih.gov/pubmed/10696268

Ziakas, P. D., & Mylonakis, E. (2024). Public interest trends for COVID-19 and pandemic trajectory: A time-series analysis of US state-level data. PLOS Digit Health, 3(3), e0000462. https://doi.org/10.1371/journal.pdig.0000462

Chapter 3 – Article 2 - Can Multisensory Olfactory Training Improve Olfactory Dysfunction caused by COVID 19?

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Abstract

Background: Approximately 30-60% of people suffer from olfactory dysfunction (OD) such as hyposmia or anosmia after being diagnosed with COVID-19; 15-20% of these cases last beyond resolution. Previous studies have shown that olfactory training can be beneficial for patients affected by OD caused by viral infections of the upper respiratory tract.

Objective: The aim of the study is to evaluate whether a multisensory olfactory training involving simultaneously tasting and seeing congruent stimuli is more effective than the classical olfactory training.

Methods: We recruited 68 participants with persistent OD for 2 months or more after COVID-19 infection; they were divided into three groups. One group received olfactory training which involved smelling four odorants (strawberry, cheese, coffee, lemon; classical olfactory training). The other group received the same olfactory stimuli but presented retronasally (i.e., as droplets on their tongue); while simultaneous and congruent gustatory (i.e., sweet, salty, bitter, sour) and visual (corresponding images) stimuli were presented (multisensory olfactory training). The third group received odourless propylene glycol in four bottles (control group). Training was carried out twice daily for 12 weeks. We assessed olfactory function and olfactory specific quality of life before and after the intervention.

Results: The intervention groups showed a similar significant improvement of olfactory function, although there was no difference in the assessment of quality of life.

Conclusion: Both multisensory and classical training can be beneficial for OD following a viral infection, however only the classical olfactory training paradigm leads to an improvement that was significantly stronger than the control group.

Introduction

Acute infection with SARS-CoV-2 is associated with olfactory dysfunction (OD) such as quantitative OD, e.g., hyposmia/anosmia or qualitative OD, e.g., parosmia (Karamali et al., 2022). Although the sense of smell recovers rather quickly in most participants (Jafar et al., 2021), long-term olfactory loss after COVID-19 appears to be widespread. Depending on how olfactory function is assessed and the specific variants of COVID-19, percentages of chronic olfactory dysfunction, i.e., olfactory dysfunction that persists for more than 6 months after the infection seem to differ. For example, in the earlier studies this percentage ranged between 30% (Mazzoli et al., 2021) and 60% (Bussiere et al., 2022). However, with the more recent Omicron variant the number of individuals with olfactory dysfunction was much lower, ranging between 5% (Mella-Torres et al., 2022) and 17% (DeWitt et al., 2023). Persistent olfactory dysfunction may have a heavy burden on the affected individuals. They have a higher risk of being exposed to hazardous situations such as fire, smoke, gas or spoiled food (Croy, Nordin, et al., 2014). Further, olfactory dysfunction affects eating and drinking (Yeomans, 2006), as well as social, sexual or work life (Bramerson et al., 2007). Thus, it comes as a no surprise that olfactory dysfunction is associated with higher rates of depression and anxiety (Kohli et al., 2016), and, in the context of long-term COVID-19, with mood disturbances and cognitive impairment (Llana et al., 2023). Individuals with olfactory disorders might also experience weight gain or loss. (Aschenbrenner et al., 2008). Currently, olfactory training is the intervention of choice for olfactory dysfunction following a viral infection of the upper respiratory tract (Hummel et al., 2009; Vance et al., 2023). During olfactory training, participants typically self-administer four odorants for twice a day for at least twelve weeks (Hummel et al., 2009). The exact underlying mechanism is unclear, but olfactory training may induce plasticity of the olfactory receptor neurons in the olfactory mucosa (Doty, 2019; Pieniak et al., 2022). Hence, olfactory training is also the therapy of choice for olfactory dysfunction following COVID-19 (Altundag et al., 2022; Bérubé et al., 2022; Le Bon et al., 2021; Pires et al., 2022). In daily life, olfactory perception typically occurs in a multisensory context (Auvray & Spence, 2008). For example, when we eat an apple, we taste its sweetness and sourness, we smell its aroma via retronasal olfaction, we feel its texture; in addition, we see its color and hear the crunch while chewing (Croy, Hoffmann, et al., 2014). In fact, flavor perception is the integration of information from

these individual sensory channels into one percept (Auvray & Spence, 2008), during which interactions occur between the single sensory channels (Small et al., 1997). Accordingly, multisensory stimulation and integration leads to changes in activation patterns in olfactory processing centers (Karunanayaka et al., 2015) with consequences on perception and behavior: for example, a congruent taste stimulus increases the intensity of retronasally presented odors (Green et al., 2012; Seo et al., 2013) and decreases detection thresholds (Dalton et al., 2000); together, they are integrated into more pleasant flavors (Fondberg et al., 2018). These effects are not limited to the chemical senses; for example, congruent visual stimuli enhance odor detection (Gottfried & Dolan, 2003) and identification (Zellner et al., 1991); while color and shape-related cues have effects on both visual performance and olfactory discrimination (Dematte et al., 2009; Jadauji et al., 2012). Therefore, multisensory olfactory training involving congruent gustatory and visual stimuli may have a superior effect compared to classical olfactory training with exclusively olfactory stimuli. We therefore hypothesized that multisensory olfactory training could improve olfactory function more than classical olfactory training in patients suffering from Olfactory disorder (OD) post-COVID-19.

Materials and methods

The protocol, its amendments and other documents were approved by the Medical Research Ethics Committee of the CIUSSS MCQ (Centre intégré universitaire de santé et de services sociaux de la Mauricie-et-du-Centre-du-Québec) (MP-2021-486) and UQTR (Université du Québec à Trois-Rivières) (CER-22-288-10.03).

Participants

A total of 68 participants agreed to participate in the study, either via self-referral or by referral from other health professionals. Participants self-identified their genders. Our inclusion criteria were (1) being 18 years or older, (2) being a resident of Quebec, (3) suffering from olfactory dysfunction for 2 months or more after COVID-19. We excluded participants with (1) chronic rhinosinusitis, (2) pre-existing olfactory disorder before COVID-19, (3) nasal sinus surgery, (4) neurologic disorders such as Alzheimer's or Parkinson's disease. For the follow up after the training, 12 participants either dropped out or their data was invalid; therefore, we included 56 participants (41

women, 15 men, mean age = 42.9 (11.3) years) in the overall analysis. Standard deviations will be presented in parentheses from this point in the text.

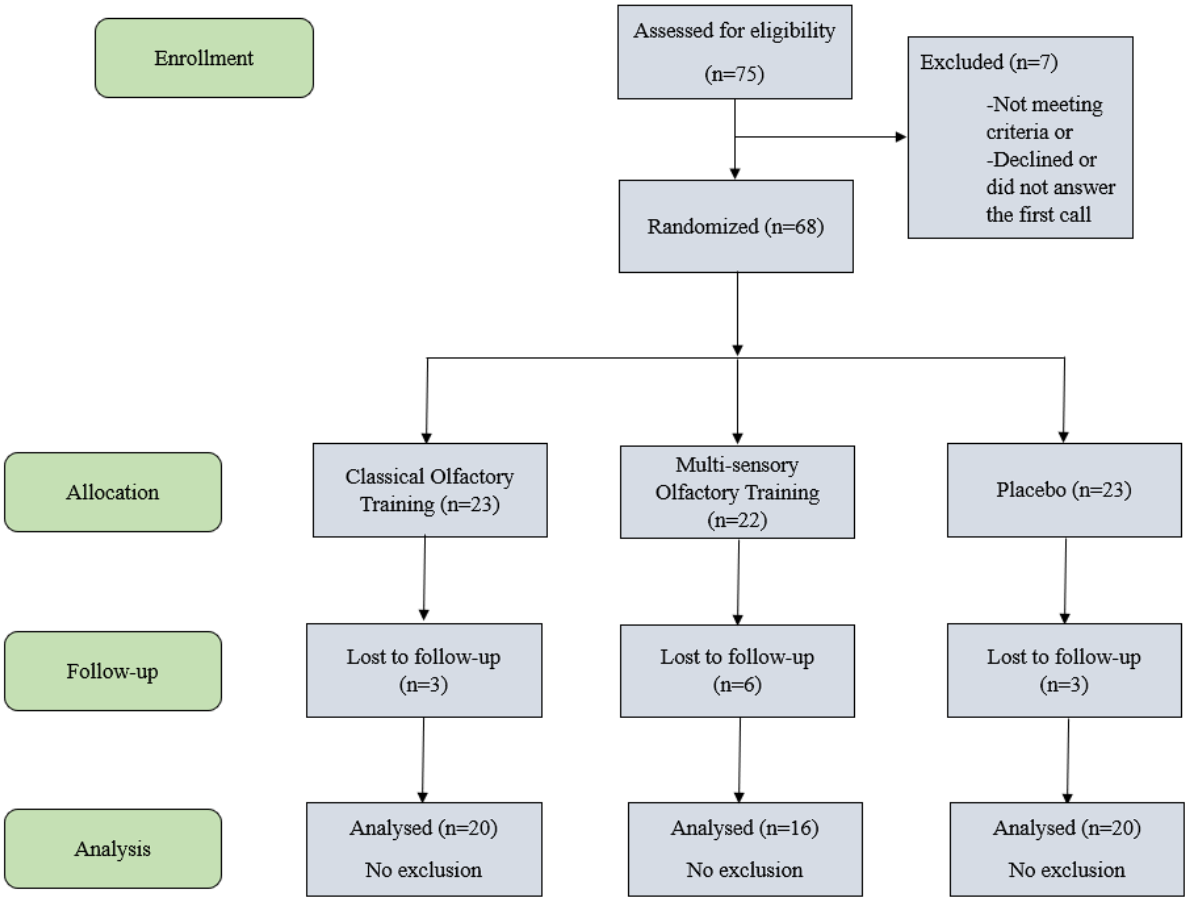


Figure 3.1. Number of participants at each step of the study.

Participants were assigned to two groups ((1) classical olfactory training - COT; (2) multisensory olfactory training – MOT) at a 1:1 ratio in a randomized manner, with no restrictions of groups. Randomization was carried out by a member of the research group who did not take part in the data collection process. Further, we included the data of a control group from an earlier study (Berube et al., 2022). In total, 23, 22 and 23 participants in the COT, MOT, and control group agreed to participate, respectively. Of them, we were able to follow up 20 participants (15 women, 5 men, mean age: 39.5 (9.6) years) in the COT group, 16 participants (11 women, 5 men, mean age: 46.3 (13.8) years) in the MOT group and 20 participants (15 women, 5 men, mean age: 43.5

(10.1) years) in the control group. The interval between the onset of COVID-19 and the start of olfactory training was 269 (78) days, 346 (116) days and 246 (109) days in the COT, MOT, and control groups, respectively. The self-reported average interval between the diagnosis of COVID-19 and olfactory loss was 5.2 (9.7) days.

Olfactory training

Classical olfactory training: In the COT group, participants received an olfactory training kit consisting of amber opaque glass vials (30 ml, Fisherbrand Inc, USA), each of which contained (5 mL, soaked in cotton pads to prevent spilling) one of four different odors (strawberry, cheese, coffee, lemon; all food grade odorants from Foodarom Glanbia Nutritionals, St. Hubert, QC, Canada). Each training session consisted in sniffing each odorant deeply for 10s, with 10s rest intervals between each odorant; we instructed participants to do this for a total of 5 minutes in line with Hummel et al (2009).

Multisensory olfactory training: In the MOT group, participants received an olfactory training kit consisting of the same four amber opaque glass vials (30 ml, Fisherbrand Inc, USA), but with a dropper lid. The bottles contained the same four odorants (20 ml) as in the classical olfactory training group. To this we also added corresponding tastants (sweet to the strawberry odorant; salty to the cheese odorant; bitter to the coffee odorant, sour to the lemon odorant). We opted not to include umami as a taste quality because many participants would have been unfamiliar and therefore would have struggled with it. An overview of the stimuli used can be found in Table 1.

Flavor	Odorant Product#	Odorant Volume	Tastant Product#	Tastant Manufacturer	Tastant amount
Strawberry	Strawberry MET0003559	0.02ml	Sucrose #424500010	Thermo Fisher, St Laurent, QC	0.4mg
Cheese	Cheese MET0017403	0.2ml	Sodium Chloride #127038.119541	BDH Inc. LOT, Toronto, ON	0.08mg
Coffee	Coffee MET0017403	0.2ml	Sucrose octaacetate #W303801	Sigma Aldrich, Oakville, ON	0.008mg
Lemon	Lemon MET0000055	0.2ml	Citric acid #X000HT86Q5	Milliard Brands, Lakewood, NJ	0.3mg

Table 3.1. Odorants and tastes used in the multisensory stimulation. *Odorants are all from Foodarom, St Hubert, QC. Odorants and tastants were dissolved in 20 mL of demineralized water.*

For the MOT group, each training session consisted in placing a drop of a solution from a given bottle on the center of the tongue while simultaneously looking at a card with a corresponding picture for 15s followed by a 30s break. This procedure was repeated 4 times. Control group: The control group received identical bottles as the COT group; however, these bottles were filled with odorless propylene glycol. They followed the same procedure as the COT group in that they were instructed to sniff each (odorless) bottle deeply for 10s, resting 10s for intervals between each bottle, for a total of 5 minutes (sham training). The data from the control group has been reported previously (Berube et al., 2022).

Assessment of olfactory function

Olfactory function was assessed using the University of Pennsylvania Smell Identification Test (UPSIT) (Doty et al., 1984). In short, the UPSIT is a scratch'n'sniff test that consists of identifying 40 microencapsulated odorants on paper booklets that are released upon scratching with 4 response choice per item. We recorded the number of correct responses out of 40 points. We further used the Shortened Questionnaire of Olfactory Disorders – Negative Statements (SQOD-NS) (Mattos et al., 2019), an adapted version of the Questionnaire for Olfactory Disorders (QOD

(Frasnelli & Hummel, 2005)), to assess the impact of olfactory dysfunction on daily life. The questionnaire comprises of 7 statements on the negative outcomes of olfactory dysfunctions that people suffer from. Further, in a separate question we asked participants whether they had perceived any qualitative changes if there was any alteration in their sense of smell to determine if they had parosmia.

Procedure

After randomization, participants were mailed a package containing an olfactory training kit corresponding to their group along with two UPSIT kits. Upon reception of the package, the research team set up a first video call with the participant. During this call we carried out the UPSIT and the SQOD-NS. Olfactory training was performed over a period of 12 weeks (Hummel et al., 2009) with participants self-administering the training twice a day. After six weeks, we called the participants by phone (1) to allow the participant to give us feedback about their olfactory function and (2) to verify and maintain compliance with the training procedure. On week 10, participants were contacted to schedule a meeting by the end of week 12. During the final meeting, UPSIT and SQOD-NS tests were performed again, and participants were debriefed.

Statistical Analysis

We used SPSS 29 (IBM Corp, Armonk, NY) for data analysis. We examined the effects of group (intervention, placebo) and time (before training, after training) on the dependent variables (1) olfactory function (UPSIT score) and (2) impact on daily life (SQOD-NS score) using a repeated measures ANOVA. We corrected post-hoc t-tests with the Bonferroni-Holm procedure to control for multiple comparisons. We carried out chi-square tests to compare the number of participants who exhibited parosmia before and after the training. We set the alpha value at 0.05.

Results

Olfactory function:

Olfactory function (UPSIT scores) increased from 23.9 (5.6) points for the COT group, 23.1 (6.9) points for the MOT group and 23.9 (5.4) for controls before training to 29.2 (4.4) points, 26.2 (7.0)

points and 24.9 (6.0) for COT, MOT, and controls, respectively, after training. The repeated measures ANOVA yielded a significant effect of time ($F(1, 53) = 19.4$, $p < .001$, partial $\eta 2 = .27$) and a significant interaction time * group, ($F(2, 53) = 3.4$, $p = .04$, partial $\eta 2 = .11$), but no effect of group ($F(2, 53) = .9$, $p = .4$).

To disentangle the interaction, we carried out 3 separate paired t-tests, one per group. While both the COT group ($t(19) = -3.834$, $p = .001$, $d = -.857$ and the MOT group ($t(15) = -2.357$, $p = .032$, $d = -.589$) showed significant improvement, there was no significant change in the control group($t(19) = -1.021$, $p = .320$, $d = -.228$).

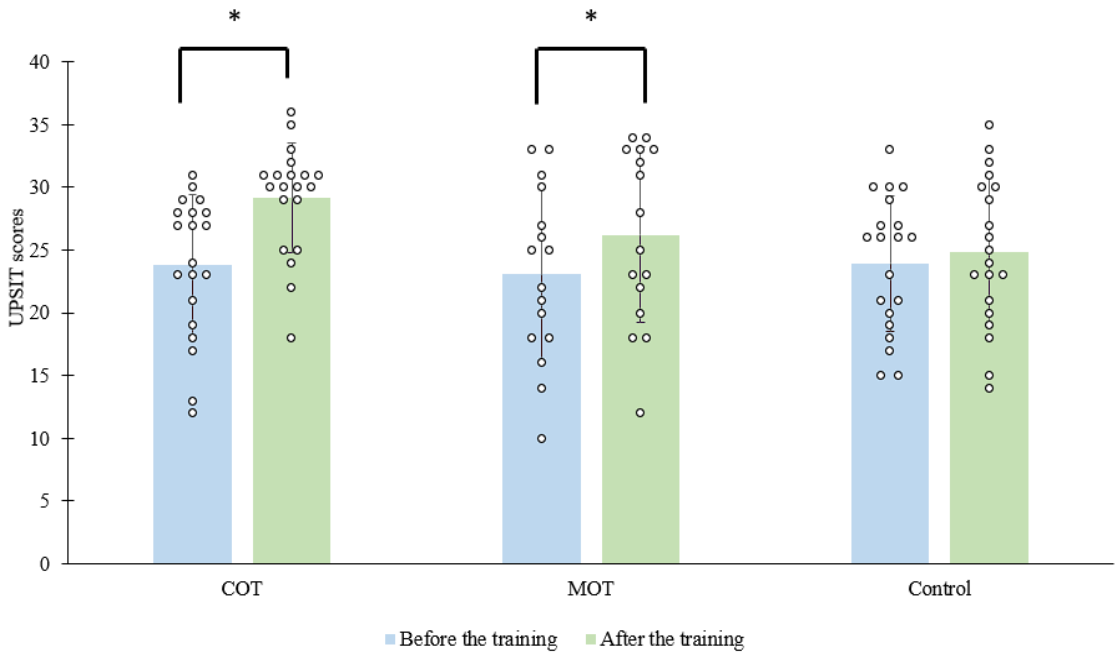


Figure 3.2. UPSIT scores before and after the training. *Scores are for (1) a group following a classical olfactory training protocol, (2) a group following a multisensory olfactory training protocol, and (3) a control group. The asterisks denote a significant difference in smell test scores before and after the training.*

Impact on daily life

(SQOD-NS) scores before training were 10.5 (4.9) points, 10.5 (3.3) points and 12.4 (4.6) points in the COT, MOT, and control groups, respectively. After training, these values were 9.4 (5.7) points, 9.7 (5.04) points and 11.0 (4.3) points for COT, MOT, and controls respectively. The repeated measures ANOVA yielded a significant main effect of time ($F(1, 53) = 6.9$, $p = .011$, partial $\eta 2 = .11$), but no significant effect of group ($F(2,53) = .94$, $p = .4$) nor a significant interaction *time* * *group* ($F(2, 53) = .15$, $p = .9$, partial $\eta 2 = .006$). For SQOD-NS scores, we identified 3 participants from each group (COT, MOT, and controls) as outliers (with a SD>2). After removal of these participants, we repeated the analysis. This yielded a significant effect of *time* ($F(1, 44) = 10.846$, $p = .002$, partial $\eta 2 = .198$) and of group ($F(2, 44) = 9.044$, $p < .001$, partial $\eta 2 = .291$), but no significant interaction *group* * *time*. Post-hoc t-tests revealed that there was a significant difference between the COT and controls ($p = .004$); MOT and controls ($p = .001$) but no significant difference between COT and MOT. When we the carried out post-hoc tests, no group exhibited a significant improvement after correction (COT ($t(16) = 1.725$, $p = .052$, $d = .418$), MOT ($t(12) = 1.891$, $p = .041$, $d = .525$) and controls ($t(16) = 2.182$, $p = .022$, $d = .529$).

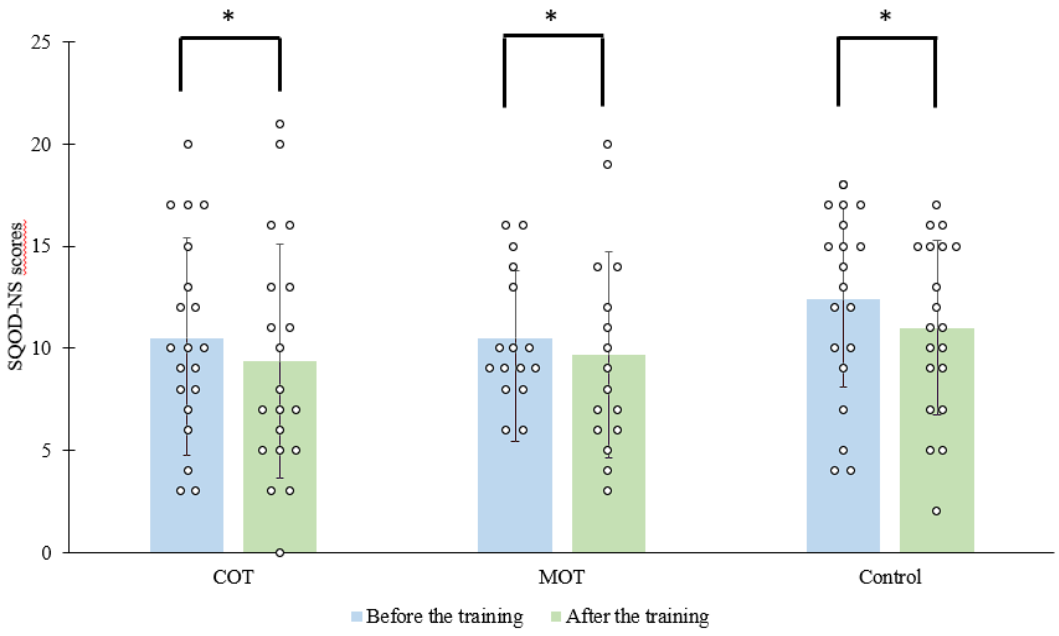


Figure 3.3. SQOD-NS scores before and after the training. *Impact on daily life scores for (1) a group following a classical olfactory training protocol, (2) a group following a multisensory olfactory training protocol, and (3) a control group.*

Parosmia

Before training, 18/20, 14/16 and 16/20 participants reported parosmia in the COT, MOT, and control groups, respectively. These numbers were 17/20, 15/16, and 19/20 after the training, and there was no difference between groups (Chi2 (1, 56) = 0.23, p = 0.9).

Discussion

Here we report the results of our study on different protocols of olfactory training. Our main results are: (1) a 12-week training using either unimodal olfactory or multisensory (i.e., with congruent visual and gustatory stimuli) paradigm improved olfactory function in participants with persistent olfactory dysfunction after COVID-19 but not in the control group that went through a sham training; (2) there was a significant improvement of daily life scores in all groups.

This study shows that olfactory training helps to restore olfactory function in patients with OD following COVID-19, in line with earlier reports. In fact, olfactory training is effective for olfactory dysfunction due to upper respiratory tract infection (URTI) (Hummel et al., 2009; Hummel et al., 2017; Hura et al., 2020; Kattar et al., 2021; Konstantinidis et al., 2013; Ojha & Dixit, 2022; Patel, 2017). The exact mechanisms underlying recovery due to olfactory training are unknown. The regeneration of olfactory receptors in the epithelium has been put forward as a potential mechanism. Accordingly, both repeated exposure in rats (Wang et al., 1993; Youngentob & Kent, 1995) and olfactory training in humans (Hummel et al., 2018) increase electrophysiological signals from the olfactory epithelium. As a consequence, olfactory training increases olfactory bulb volume in both patients with olfactory dysfunction (Gellrich et al., 2018) and healthy individuals (Filiz et al., 2022; Negoias et al., 2017). It further improves different functional and morphometric measures including functional connectivity in the central chemosensory networks (Kollndorfer et al., 2015), as well as grey matter volume (Al Ain et al., 2019; Banks et al., 2016; Delon-Martin et al., 2013; Filiz et al., 2022) and activation levels (Chen et al., 2022) of olfactory processing areas. In an earlier

study on a similar but different cohort (with the same control group), we used an olfactory training protocol (Bérubé et al., 2022). We observed an improvement of quality of life and subjective olfactory function, but no improvement on scores with a validated olfactory test. Again, together with the results of the present study, this suggests that the benefits of olfactory training in OD post-COVID-19 may be relatively limited.

While the effectiveness of olfactory training in olfactory dysfunction due to URTI is now well established (Hwang et al., 2023; Vance et al., 2023), researchers investigated several parameters to further improve its impact. These modifications included duration of the training, odor variety, odor intensity, pharmaceutical support, and other. For example, the use of steroids to support olfactory training yielded mixed results, as one study showed additional benefit (Fleiner et al., 2012), while another did not (Schepens et al., 2022). Other interventions such as the use of odorants of higher molecular weight (Poletti et al., 2017) and the use of odorants in higher concentrations (Damm et al., 2014) appeared to be more promising. Increasing the number of odorants from four to eight (Pires et al., 2022) did not change the effectiveness of olfactory training, but changing odor sets during the training did (Altundag et al., 2015).

However, the parameter with the highest impact on a positive outcome of olfactory training appears to be the duration of the olfactory training protocol. For example, patients with olfactory dysfunction following COVID-19 who followed olfactory training for more than 28 days showed greater long-term improvements compared to those who took the training for less than 28 days (Denis et al., 2021). Along the same lines, 24 weeks of olfactory training in patients with olfactory loss due to URTI yielded significant improvements in olfactory identification and discrimination abilities after 3 and 6 months, but not after 1 month (Qiao et al., 2019). Similarly, olfactory training in patients with olfactory loss due to URTI yielded significantly better outcomes after 32 weeks (Geissler et al., 2014) and 56 weeks (Konstantinidis et al., 2016) when compared to 12 weeks. Even longer follow up periods (6, 12, and 18 months) lead to further improvement in patients with COVID-19 related olfactory dysfunction (Lechien et al., 2023). Protocols of such long duration may be more easily be carried out when stimuli are presented as drops onto the tongue rather than sniffing from bottles, and this is for two reasons. First, for individuals with olfactory dysfunction, it may be difficult to judge when an odor has evaporated from a bottle, while it is rather

obvious when there is no more liquid in the bottle. Second, administrating odorants as drops onto to the tongue may help to generate olfactory stimuli of higher concentration, which is crucial for the positive outcome of olfactory training (Damm et al., 2014). In fact, when intranasally measuring odorant concentrations released from intraoral sources, peak amplitudes were only reached after several seconds after intake, supporting the notion that intraoral administration may lead to prolonged stimulation of the olfactory system (Frasnelli et al., 2005).

With long intervention periods, compliance with the training protocol becomes an issue (Vance et al., 2023). It is not surprising that extremely long training protocols to up to a year and a half are particularly useful in patients with strong adherence to the protocol (Lechien et al., 2023). Rendering olfactory training easier, e.g., using an olfactory ball, i.e., a ball containing 4 different holes to hold 4 different olfactory stimuli significantly increased adherence to the protocol and improved outcome (Saatci et al., 2020). Although we did not assess patient compliance, offering multisensory stimuli for olfactory training may increase adherence; future studies should evaluate this potential.

In the present study, we integrated both visual and gustatory stimuli into the already existing olfactory training. Visual and olfactory stimuli have been combined for olfactory training before. For example, combining odors with digital images showed the largest clinically meaningful improvements in olfactory dysfunction due to COVID-19 (Denis et al., 2021; Khan et al., 2022). Considering the multisensory interactions between olfaction and vision, it is no surprise that MOT has a potential to increase the efficiency in both olfaction and cognitive processes (Vance et al., 2023). This argument could further be supported by the fact that olfactory and the ventral visual processing streams converge in olfactory regions e.g., orbitofrontal cortex (Kuang & Zhang, 2014; Rolls, 2019; Rolls et al., 1996) or piriform cortex (Qureshy et al., 2000) and (visual) occipital cortex (Qureshy et al., 2000). Similarly to the olfactory-visual interactions, the olfactory and gustatory senses influence each other mutually (Czarnecki & Fontanini, 2019) by having shared stimuli (e.g., food) and converging central pathways in the orbitofrontal cortex (Czarnecki & Fontanini, 2019; Rolls, 2016) and insula (Mazzola et al., 2017).

In this study, we did not find any superiority of a multisensory training paradigm over a classical olfactory protocol. While potentially this may be due to the small sample size, it also suggests that there is no major advantage of a multisensory over an olfactory paradigm. However, several studies show the benefits of multisensory training over using unimodal sensory training in various settings such as audio-visual integration (Seitz et al., 2006; von Kriegstein & Giraud, 2006), postural stability (Hu & Woollacott, 1994), dyslexia (Kast et al., 2007) and auditory impairments restored with cochlear implants (Isaiah et al., 2014). While such a superiority of multisensory training is not evident in our study, one could imagine a scenario in which multisensory stimuli (e.g., candies) could be associated with higher compliance than pure olfactory training. This should be investigated in future studies.

Even though the olfactory percept is multi-dimensional, we were not able to capture a particular effect of multisensory training beyond the effect from classical olfactory training. This may be due to some of our study's limitations. First, we included a relatively small sample yielding limited statistical power. Additionally, in all groups we recruited more women than men. While this may reflect gender-related differences in the impact of COVID-19 on olfactory abilities (Bussiere et al., 2021) in line with other URTI studies (Liu et al., 2016) or studies with healthy participants (Sorokowski et al., 2019), it could potentially skew our results as women typically score higher in olfactory identification and memory tasks (Doty & Cameron, 2009).

Further, participants self-administered the training, hence it is not possible to know with certainty if participants actually followed the suggested routine as recall bias or social desirability might have affected the results (Vance et al., 2023). It is therefore important to put adherence rules or tasks in place to track such data, e.g., keeping journals or reports (Vance et al., 2023). Next, as this study was carried out during the pandemic, public health restrictions forced us to test participants remotely. This came with the major disadvantage that we were only able to evaluate olfactory identification scores as endpoints and could not measure olfactory threshold or discrimination. In this context, it is important to point out that multisensory olfactory training appears to mainly lead to improvement in olfactory threshold and discrimination scores (Li et al., 2023).

Furthermore, the interval between COVID-19 onset and initiation of OT was not equal between groups; on average, participants in the MOT group started the training 80 days later than the COT group. This may have influenced the outcome. In fact, a previous study on individuals with smell loss due to an URTI found that the probability of spontaneous recovery decreased with time (Reden et al., 2006). However, the observation period in that study ranged from 1 to more than 200 months and was therefore much more heterogenous than in ours. Nevertheless, future studies will have to carefully match participants to evaluate the effect of this potential confounder. Finally, we instructed participants to sniff the odorants for 10s in the COT condition, the only instructions regarding timing in the MOT condition was to look at the image for 15 s. It is therefore possible that the duration of the exposure to the odorant in the MOT condition was shorter or longer (Frasnelli et al., 2005) than in the COT condition. It is easier to control the timing parameters in a lab setting (Al Ain et al., 2019), and future studies should control for this parameter.

In conclusion, we show that both a multisensory olfactory training and a classical olfactory training can lead to improved olfactory function in participants with chronic olfactory dysfunction following COVID-19.

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References

Al Ain, S., Poupon, D., Hetu, S., Mercier, N., Steffener, J., & Frasnelli, J. (2019). Smell training improves olfactory function and alters brain structure. Neuroimage, 189, 45-54. https://doi.org/10.1016/j.neuroimage.2019.01.008

Altundag, A., Cayonu, M., Kayabasoglu, G., Salihoglu, M., Tekeli, H., Saglam, O., & Hummel, T. (2015). Modified olfactory training in patients with postinfectious olfactory loss. Laryngoscope, 125(8), 1763-1766. https://doi.org/10.1002/lary.25245

Altundag, A., Yilmaz, E., & Kesimli, M. C. (2022). Modified Olfactory Training Is an Effective Treatment Method for COVID-19 Induced Parosmia. Laryngoscope, 132(7), 1433-1438. https://doi.org/10.1002/lary.30101

Aschenbrenner, K., Scholze, N., Joraschky, P., & Hummel, T. (2008). Gustatory and olfactory sensitivity in patients with anorexia and bulimia in the course of treatment. J Psychiatr Res, 43(2), 129-137. https://doi.org/10.1016/j.jpsychires.2008.03.003

Auvray, M., & Spence, C. (2008). The multisensory perception of flavor. Conscious Cogn, 17(3), 1016-1031. https://doi.org/10.1016/j.concog.2007.06.005

Banks, S. J., Sreenivasan, K. R., Weintraub, D. M., Baldock, D., Noback, M., Pierce, M. E., Frasnelli, J., James, J., Beall, E., Zhuang, X., Cordes, D., & Leger, G. C. (2016). Structural and Functional MRI Differences in Master Sommeliers: A Pilot Study on Expertise in the Brain [Original Research]. Frontiers in Human Neuroscience, 10. https://www.frontiersin.org/articles/10.3389/fnhum.2016.00414

Bérubé, S., Demers, C., Bussière, N., Cloutier, F., Pek, V., Chen, A., Bolduc-Bégin, J., & Frasnelli, J. (2022). Olfactory Training Impacts Olfactory Dysfunction Induced by COVID-19: A Pilot Study. ORL. https://doi.org/10.1159/000528188

Bramerson, A., Nordin, S., & Bende, M. (2007). Clinical experience with patients with olfactory complaints, and their quality of life. Acta Otolaryngol, 127(2), 167-174. https://doi.org/10.1080/00016480600801357

Bussiere, N., Mei, J., Levesque-Boissonneault, C., Blais, M., Carazo, S., Gros-Louis, F., De Serres, G., Dupre, N., & Frasnelli, J. (2021). Chemosensory Dysfunctions Induced by COVID-19 Can Persist up to 7 Months: A Study of Over 700 Healthcare Workers. Chem Senses, 46. https://doi.org/10.1093/chemse/bjab038

Bussiere, N., Mei, J., Levesque-Boissonneault, C., Blais, M., Carazo, S., Gros-Louis, F., Laforce, R., De Serres, G., Dupre, N., & Frasnelli, J. (2022). Persisting chemosensory impairments in 366 healthcare workers following COVID-19: an 11-month follow-up. Chem Senses, 47. https://doi.org/10.1093/chemse/bjac010

Chen, B., Espin, M., Haussmann, R., Matthes, C., Donix, M., Hummel, T., & Haehner, A. (2022). The Effect of Olfactory Training on Olfaction, Cognition, and Brain Function in Patients with Mild Cognitive Impairment. J Alzheimers Dis, 85(2), 745-754. https://doi.org/10.3233/JAD-215257

Croy, I., Hoffmann, H., Philpott, C., Rombaux, P., Welge-Luessen, A., Vodicka, J., Konstantinidis, I., Morera, E., & Hummel, T. (2014). Retronasal testing of olfactory function: an investigation and comparison in seven countries. Eur Arch Otorhinolaryngol, 271(5), 1087-1095. https://doi.org/10.1007/s00405-013-2684-9

Croy, I., Nordin, S., & Hummel, T. (2014). Olfactory disorders and quality of life--an updated review. Chem Senses, 39(3), 185-194. https://doi.org/10.1093/chemse/bjt072

Damm, M., Pikart, L. K., Reimann, H., Burkert, S., Goktas, O., Haxel, B., Frey, S., Charalampakis, I., Beule, A., Renner, B., Hummel, T., & Huttenbrink, K. B. (2014). Olfactory training is helpful in postinfectious olfactory loss: a randomized, controlled, multicenter study. Laryngoscope, 124(4), 826-831. https://doi.org/10.1002/lary.24340

Delon-Martin, C., Plailly, J., Fonlupt, P., Veyrac, A., & Royet, J.-P. (2013). Perfumers' expertise induces structural reorganization in olfactory brain regions. Neuroimage, 68, 55-62. https://doi.org/10.1016/j.neuroimage.2012.11.044

Denis, F., Septans, A. L., Periers, L., Maillard, J. M., Legoff, F., Gurden, H., & Moriniere, S. (2021). Olfactory Training and Visual Stimulation Assisted by a Web Application for Patients With Persistent Olfactory Dysfunction After SARS-CoV-2 Infection: Observational Study. J Med Internet Res, 23(5), e29583. https://doi.org/10.2196/29583

Doty, R. L. (2019). Treatments for smell and taste disorders: A critical review. Handb Clin Neurol, 164, 455-479. https://doi.org/10.1016/B978-0-444-63855-7.00025-3

Doty, R. L., & Cameron, E. L. (2009). Sex differences and reproductive hormone influences on human odor perception. Physiol Behav, 97(2), 213-228. https://doi.org/10.1016/j.physbeh.2009.02.032

Doty, R. L., Shaman, P., & Dann, M. (1984). Development of the University of Pennsylvania Smell Identification Test: a standardized microencapsulated test of olfactory function. Physiol Behav, 32(3), 489-502. https://doi.org/10.1016/0031-9384(84)90269-5

Filiz, G., Poupon, D., Banks, S., Fernandez, P., & Frasnelli, J. (2022). Olfactory bulb volume and cortical thickness evolve during sommelier training. Hum Brain Mapp, 43(8), 2621-2633. https://doi.org/10.1002/hbm.25809

Fleiner, F., Lau, L., & Goktas, O. (2012). Active olfactory training for the treatment of smelling disorders. Ear Nose Throat J, 91(5), 198-203, 215. https://doi.org/10.1177/014556131209100508

Fondberg, R., Lundstrom, J. N., Blochl, M., Olsson, M. J., & Seubert, J. (2018). Multisensory flavor perception: The relationship between congruency, pleasantness, and odor referral to the mouth. Appetite, 125, 244-252. https://doi.org/10.1016/j.appet.2018.02.012

Frasnelli, J., & Hummel, T. (2005). Olfactory dysfunction and daily life. Eur Arch Otorhinolaryngol, 262(3), 231-235. https://doi.org/10.1007/s00405-004-0796-y

Geissler, K., Reimann, H., Gudziol, H., Bitter, T., & Guntinas-Lichius, O. (2014). Olfactory training for patients with olfactory loss after upper respiratory tract infections. Eur Arch Otorhinolaryngol, 271(6), 1557-1562. https://doi.org/10.1007/s00405-013-2747-y

Gellrich, J., Han, P., Manesse, C., Betz, A., Junghanns, A., Raue, C., Schriever, V. A., & Hummel, T. (2018). Brain volume changes in hyposmic patients before and after olfactory training. Laryngoscope, 128(7), 1531-1536. https://doi.org/10.1002/lary.27045

Gottfried, J. A., & Dolan, R. J. (2003). The nose smells what the eye sees: crossmodal visual facilitation of human olfactory perception. Neuron, 39(2), 375-386. https://doi.org/10.1016/s0896-6273(03)00392-1

Hu, M. H., & Woollacott, M. H. (1994). Multisensory training of standing balance in older adults: I. Postural stability and one-leg stance balance. J Gerontol, 49(2), M52-61. https://doi.org/10.1093/geronj/49.2.m52

Hummel, T., Rissom, K., Reden, J., Hahner, A., Weidenbecher, M., & Huttenbrink, K. B. (2009). Effects of olfactory training in patients with olfactory loss. Laryngoscope, 119(3), 496-499. https://doi.org/10.1002/lary.20101

Hummel, T., Stupka, G., Haehner, A., & Poletti, S. C. (2018). Olfactory training changes electrophysiological responses at the level of the olfactory epithelium. Rhinology, 56(4), 330-335. https://doi.org/10.4193/Rhin17.163

Hummel, T., Whitcroft, K. L., Andrews, P., Altundag, A., Cinghi, C., Costanzo, R. M., Damm, M., Frasnelli, J., Gudziol, H., Gupta, N., Haehne, A., Holbrook, E., Hong, S. C., Hornung, D., Huttenbrink, K. B., Kamel, R., Kobayashi, M., Konstantinidis, I., Landis, B. N., . . . Welge-Luessen, A. (2017). Position paper on olfactory dysfunction. Rhinol Suppl, 54(26), 1-30. https://doi.org/10.4193/Rhino16.248

Hura, N., Xie, D. X., Choby, G. W., Schlosser, R. J., Orlov, C. P., Seal, S. M., & Rowan, N. R. (2020). Treatment of post-viral olfactory dysfunction: an evidence-based review with recommendations. Int Forum Allergy Rhinol, 10(9), 1065-1086. https://doi.org/10.1002/alr.22624

Hwang, S. H., Kim, S. W., Basurrah, M. A., & Kim, D. H. (2023). The Efficacy of Olfactory Training as a Treatment for Olfactory Disorders Caused by Coronavirus Disease-2019: A Systematic Review and Meta-Analysis. Am J Rhinol Allergy, 19458924221150977. https://doi.org/10.1177/19458924221150977

Isaiah, A., Vongpaisal, T., King, A. J., & Hartley, D. E. (2014). Multisensory training improves auditory spatial processing following bilateral cochlear implantation. J Neurosci, 34(33), 11119-11130. https://doi.org/10.1523/JNEUROSCI.4767-13.2014

Jafar, A., Lasso, A., Shorr, R., Hutton, B., & Kilty, S. (2021). Olfactory recovery following infection with COVID-19: A systematic review. PLoS One, 16(11), e0259321. https://doi.org/10.1371/journal.pone.0259321

Jiramongkolchai, P., Jones, M. S., Peterson, A., Lee, J. J., Liebendorfer, A., Klatt-Cromwell, C. N., Schneider, J. S., Drescher, A. J., Ogden, M. A., Brunworth, J. D., Kallogjeri, D., Kukuljan, S., Peelle, J. E., & Piccirillo, J. F. (2021). Association of Olfactory Training With Neural Connectivity in Adults With Postviral Olfactory Dysfunction. JAMA Otolaryngol Head Neck Surg, 147(6), 502-509. https://doi.org/10.1001/jamaoto.2021.0086

Karamali, K., Elliott, M., & Hopkins, C. (2022). COVID-19 related olfactory dysfunction. Curr Opin Otolaryngol Head Neck Surg, 30(1), 19-25. https://doi.org/10.1097/MOO.0000000000000783

Karunanayaka, P. R., Wilson, D. A., Vasavada, M., Wang, J., Martinez, B., Tobia, M. J., Kong, L., Eslinger, P., & Yang, Q. X. (2015). Rapidly acquired multisensory association in the olfactory cortex. Brain Behav, 5(11), e00390. https://doi.org/10.1002/brb3.390

Kast, M., Meyer, M., Vogeli, C., Gross, M., & Jancke, L. (2007). Computer-based multisensory learning in children with developmental dyslexia. Restor Neurol Neurosci, 25(3-4), 355-369. https://www.ncbi.nlm.nih.gov/pubmed/17943011

Kattar, N., Do, T. M., Unis, G. D., Migneron, M. R., Thomas, A. J., & McCoul, E. D. (2021). Olfactory Training for Postviral Olfactory Dysfunction: Systematic Review and Meta-analysis. Otolaryngol Head Neck Surg, 164(2), 244-254. https://doi.org/10.1177/0194599820943550

Khan, A. M., Piccirillo, J., Kallogjeri, D., & Piccirillo, J. F. (2022). Efficacy of Combined Visual-Olfactory Training With Patient-Preferred Scents as Treatment for Patients With COVID-19 Resultant Olfactory Loss: A Randomized Clinical Trial. JAMA Otolaryngol Head Neck Surg. https://doi.org/10.1001/jamaoto.2022.4112

Kohli, P., Soler, Z. M., Nguyen, S. A., Muus, J. S., & Schlosser, R. J. (2016). The Association Between Olfaction and Depression: A Systematic Review. Chem Senses, 41(6), 479-486. https://doi.org/10.1093/chemse/bjw061

Kollndorfer, K., Fischmeister, F. P., Kowalczyk, K., Hoche, E., Mueller, C. A., Trattnig, S., & Schopf, V. (2015). Olfactory training induces changes in regional functional connectivity in patients with long-term smell loss. Neuroimage Clin, 9, 401-410. https://doi.org/10.1016/j.nicl.2015.09.004

Konstantinidis, I., Tsakiropoulou, E., Bekiaridou, P., Kazantzidou, C., & Constantinidis, J. (2013). Use of olfactory training in post-traumatic and postinfectious olfactory dysfunction. Laryngoscope, 123(12), E85-90. https://doi.org/10.1002/lary.24390

Konstantinidis, I., Tsakiropoulou, E., & Constantinidis, J. (2016). Long term effects of olfactory training in patients with post-infectious olfactory loss. Rhinology, 54(2), 170-175. https://doi.org/10.4193/Rhino15.264

Le Bon, S. D., Konopnicki, D., Pisarski, N., Prunier, L., Lechien, J. R., & Horoi, M. (2021). Efficacy and safety of oral corticosteroids and olfactory training in the management of COVID-19-related loss of smell. Eur Arch Otorhinolaryngol, 278(8), 3113-3117. https://doi.org/10.1007/s00405-020-06520-8

Lechien, J. R., Vaira, L. A., & Saussez, S. (2023). Effectiveness of olfactory training in COVID-19 patients with olfactory dysfunction: a prospective study. Eur Arch Otorhinolaryngol, 280(3), 1255-1263. https://doi.org/10.1007/s00405-022-07665-4

Liu, J., Pinto, J. M., Yang, L., Li, L., Sun, J., Miao, X., Li, K., Chen, G., & Wei, Y. (2016). Gender difference in Chinese adults with post-viral olfactory disorder:a hospital-based study. Acta Otolaryngol, 136(9), 976-981. https://doi.org/10.3109/00016489.2016.1172729

Llana, T., Mendez, M., Garces-Arilla, S., Hidalgo, V., Mendez-Lopez, M., & Juan, M. C. (2023). Association between olfactory dysfunction and mood disturbances with objective and subjective cognitive deficits in long-COVID. Front Psychol, 14, 1076743. https://doi.org/10.3389/fpsyg.2023.1076743

Mattos, J. L., Edwards, C., Schlosser, R. J., Hyer, M., Mace, J. C., Smith, T. L., & Soler, Z. M. (2019). A brief version of the questionnaire of olfactory disorders in patients with chronic rhinosinusitis. Int Forum Allergy Rhinol, 9(10), 1144-1150. https://doi.org/10.1002/alr.22392

Mazzoli, M., Molinari, M. A., Tondelli, M., Giovannini, G., Ricceri, R., Ciolli, L., Picchetto, L., & Meletti, S. (2021). Olfactory function and viral recovery in COVID-19. Brain Behav, 11(3), e02006. https://doi.org/10.1002/brb3.2006

Ojha, P., & Dixit, A. (2022). Olfactory training for olfactory dysfunction in COVID-19: A promising mitigation amidst looming neurocognitive sequelae of the pandemic. Clin Exp Pharmacol Physiol, 49(4), 462-473. https://doi.org/10.1111/1440-1681.13626

Patel, Z. M. (2017). The evidence for olfactory training in treating patients with olfactory loss. Curr Opin Otolaryngol Head Neck Surg, 25(1), 43-46. https://doi.org/10.1097/MOO.0000000000000328

Pieniak, M., Oleszkiewicz, A., Avaro, V., Calegari, F., & Hummel, T. (2022). Olfactory Training-Thirteen Years of Research Reviewed. Neurosci Biobehav Rev, 104853. https://doi.org/10.1016/j.neubiorev.2022.104853

Pires, I. A. T., Steffens, S. T., Mocelin, A. G., Shibukawa, D. E., Leahy, L., Saito, F. L., Amadeu, N. T., Lopes, N. M. D., Garcia, E. C. D., Albanese, M. L., De Mari, L. F., Ferreira, I. M., Veiga, C. A., Jebahi, Y., Coifman, H., Fornazieri, M. A., & Hamerschmidt, R. (2022). Intensive Olfactory Training in Post-COVID-19 Patients: A Multicenter Randomized Clinical Trial. Am J Rhinol Allergy, 36(6), 780-787. https://doi.org/10.1177/19458924221113124

Poletti, S. C., Michel, E., & Hummel, T. (2017). Olfactory Training Using Heavy and Light Weight Molecule Odors. Perception, 46(3-4), 343-351. https://doi.org/10.1177/0301006616672881

Qiao, X. F., Wang, G. P., Li, X., Bai, Y. H., & Zheng, W. (2019). Analysis of the clinical effect of olfactory training on olfactory dysfunction after upper respiratory tract infection. Acta Otolaryngol, 139(7), 643-646. https://doi.org/10.1080/00016489.2019.1614224

Saatci, O., Altundag, A., Duz, O. A., & Hummel, T. (2020). Olfactory training ball improves adherence and olfactory outcomes in post-infectious olfactory dysfunction. Eur Arch Otorhinolaryngol, 277(7), 2125-2132. https://doi.org/10.1007/s00405-020-05939-3

Schepens, E. J. A., Blijleven, E. E., Boek, W. M., Boesveldt, S., Stokroos, R. J., Stegeman, I., & Kamalski, D. M. A. (2022). Prednisolone does not improve olfactory function after COVID-19: a randomized, double-blind, placebo-controlled trial. BMC Med, 20(1), 445. https://doi.org/10.1186/s12916-022-02625-5

Seitz, A. R., Kim, R., & Shams, L. (2006). Sound Facilitates Visual Learning. Current Biology, 16(14), 1422-1427. https://doi.org/https://doi.org/10.1016/j.cub.2006.05.048

Seo, H. S., Iannilli, E., Hummel, C., Okazaki, Y., Buschhuter, D., Gerber, J., Krammer, G. E., van Lengerich, B., & Hummel, T. (2013). A salty-congruent odor enhances saltiness: functional magnetic resonance imaging study. Hum Brain Mapp, 34(1), 62-76. https://doi.org/10.1002/hbm.21414

Small, D. M., Jones-Gotman, M., Zatorre, R. J., Petrides, M., & Evans, A. C. (1997). Flavor processing: more than the sum of its parts. Neuroreport, 8(18), 3913-3917. https://doi.org/10.1097/00001756-199712220-00014

Sorokowski, P., Karwowski, M., Misiak, M., Marczak, M. K., Dziekan, M., Hummel, T., & Sorokowska, A. (2019). Sex Differences in Human Olfaction: A Meta-Analysis. Front Psychol, 10, 242. https://doi.org/10.3389/fpsyg.2019.00242

Vance, D. E., Del Bene, V. A., Kamath, V., Frank, J. S., Billings, R., Cho, D. Y., Byun, J. Y., Jacob, A., Anderson, J. N., Visscher, K., Triebel, K., Martin, K. M., Li, W., Puga, F., & Fazeli, P. L. (2023). Does Olfactory Training Improve Brain Function and Cognition? A Systematic Review. Neuropsychol Rev, 1-37. https://doi.org/10.1007/s11065-022-09573-0

Chapter 4 – General discussion

In this thesis, we present the results of two studies: “Congruency of Multisensory Olfactory Stimuli” (Article 1) and “Can Multisensory Olfactory Training Improve Olfactory Dysfunction caused by COVID-19?” (Article 2).

Our main findings are as follows: (1) congruency significantly influenced likeness perception, (2) olfactory stimuli presented with congruent gustatory and/or visual co-stimulation resulted in better label matching, whereas incongruent co-stimulation produced the opposite effect, (3) gustatory co-stimulation exerted a stronger influence on olfactory perception than visual co-stimulation, (4) although COVID-19-related OD showed a natural tendency to improve over time, both MOT and COT enhanced olfactory abilities more effectively than placebo.

4.1. Impact on Research and Literature

4.1.1. Olfactory training

A 12-week OT program (whether classical or multisensory) improves olfactory abilities of COVID-19 patients with long-term olfactory loss compared with a placebo group that receives no real training. Additionally, all groups demonstrated significant improvements in daily-life (quality-of-life) scores.

The findings from our study align with the current literature on OT, including research conducted with post-viral olfactory disorder (PVOD) patients (Hummel et al., 2009). Patients with post-viral OD show progressive improvements in TDI scores after 16 and 32 weeks of training (Geissler et al., 2014). Alongside improved TDI scores, increased sensitivity to odors has also been observed in COVID-19 patients following OT paradigms (Yaylaci et al., 2023).

OT can also improve olfactory abilities in healthy participants. Structural neural changes have been reported following only a six-week training period, including increased cortical thickness in the right inferior frontal gyrus, bilateral fusiform gyrus, and right entorhinal cortex (Al Ain et al., 2019). The importance of multisensory interaction has also been demonstrated in healthy

participants: a 12-week MOT with congruent visual-olfactory stimuli resulted in greater TDI improvements compared with olfactory stimulation alone (Li et al., 2023).

Age-related effects have also been documented. In healthy children, COT enhances odor identification and threshold scores, with children who undergo COT outperforming those who do not (Mori et al., 2015, Mahmut et al., 2021, Pieniak et al., 2022). In older adults, COT similarly increases Sniffin' Sticks scores (Birte-Antina et al., 2018).

OT can enhance olfactory abilities in domain specialists as well. Although these training do not follow classical OT paradigms, they involve long-term odor exposure, which may explain their improved olfactory performance. For example, sommelier students display superior odor identification (Poupon et al., 2019) and recognition (Parr et al., 2002) skills. Wine experts perceive odorants as more intense compared with controls (Marino-Sanchez et al., 2010), and experienced beer drinkers outperform novices in discrimination tasks (Peron & Allen, 1988). However, some argue that these advantages may be domain-specific, as coffee and wine experts show only modest advantage over non-specialists in identifying or describing stimuli within their respective areas (Croijmans & Majid, 2016).

Our newly developed MOT improved both olfactory abilities and quality-of-life scores. Although modifications to OT in PVOD patients have been introduced in various forms such as changing the duration of training (Altundag et al., 2015), altering the number of daily sessions (Oleszkiewicz et al., 2022), adjusting stimulus concentrations (Damm et al., 2014), using local odorants (Choi et al., 2021), modifying the number of odorants (Genetzaki et al., 2024), adding adjunct therapies such as corticosteroids (Fleiner et al., 2012), vitamin A (Hummel, Whitcroft, Rueter, et al., 2017), or budesonide (Nguyen & Patel, 2018), and incorporating methods to increase adherence (Saatci et al., 2020) or visual or audiovisual supports (Khan et al., 2023, Li et al., 2023), no prior training paradigm has focused specifically on congruent triplets of olfaction-gustation-vision.

Our MOT did not demonstrate greater benefits than COT. Several factors may explain this outcome: (1) small sample size, especially after participant dropout, reduced the statistical power of our study, (2) remote delivery of training instructions (phone or Zoom calls), increased the likelihood of user errors or inconsistent applications, (3) adherence was not monitored, which is a

critical factor influencing outcomes in OT paradigms, (4) participants in the MOT group began training, on average, nearly three months later than those in the COT group, providing them with more time for spontaneous recovery.

In the future, researchers should consider testing healthy participants with MOT paradigm to acquire baseline evaluation of the stimuli. Further implementation of sensitivity measurements with thresholds of stimuli (both with healthy participants and OD patients) could give more insights into how effective stimuli would be in the real paradigm. After such measurements, stimuli intensity could be adjusted for better detection. Additionally, increasing the training time from 12 weeks to further training times is an option to evaluate whether MOT would be more effective in longer terms. Finally, adjusting the age range of participants to include younger individuals, as in the case of Article 1 could give us more insights about the effectiveness of MOT.

4.1.2. Multisensory integration between olfaction, gustation and vision

We report that congruent multisensory stimuli significantly increase the likeness perception of olfactory stimuli, whereas incongruent stimuli reduce likeness perception. Furthermore, congruent or incongruent gustatory stimuli exert a stronger influence on likeness ratings than visual stimuli. This means that even in the presence of incongruent visual stimuli, congruent gustatory stimuli can partially counteract the incongruence effect of vision and still lead to significantly higher likeness scores. These findings reinforce the well-established strong connections between olfaction and gustation (Oleszkiewicz et al., 2019) demonstrating that congruent gustatory co-stimulation provides a measurable perceptual advantage when paired with odorants. Our results also confirm robust, albeit comparatively weaker, interactions between visual and olfactory perception.

The stronger olfaction-gustation link, relative to the olfaction-vision relationship, may arise from the fact that both senses construct flavor perception together, a vital component of food consumption and survival (Auvray & Spence, 2008). Olfactory and gustatory processing share overlapping neural substrates, including the orbitofrontal cortex (Small et al., 2007), insula (Mazzola et al., 2017), and frontal operculum (Seubert et al., 2015). Multivariate pattern analyses further show overlapping representations of retronasal odors and congruent tastes (Khorisantono et al.,

2025). Additionally, patients with congenital olfactory impairments exhibit significantly reduced activation in the medial orbitofrontal cortex and anterior insula in response to sweet, salty, and bitter during taste identification tasks (Gagnon et al., 2014), suggesting interdependence between these sensory systems. Perhaps the most direct behavioral evidence of this interdependence is the observation that olfactory loss compromises taste perception (Migneault-Bouchard et al., 2020).

Animal studies provide support for these interactions. In mice, the gustatory cortex is required for processing retronasal odorants, but not orthonasal odorants (Blankenship et al., 2019). In rats, single-unit recordings from both the primary olfactory cortex and primary gustatory cortex reveal responses to intraoral stimuli, indicating that both cortices can process retronasal information (Maier, 2017). Furthermore, experimentally blocking the gustatory epithelium in awake rats reduces neuronal activity in the piriform cortex, a central hub of olfactory processing (Maier et al., 2012). These findings collectively suggest early sensory convergence and bidirectional modulation between the gustatory and olfactory systems.

Because much of the detailed knowledge comes from animal models, additional human studies could greatly enhance our understanding of olfactory function, dysfunction, and plasticity, especially the investigations of OSN in the olfactory epithelium and OB. Future work could focus on both healthy individuals and patients with olfactory dysfunction (across etiologies) to assess potential recovery mechanisms. Promising approaches include non-invasive physiological measurements such as electro-olfactogram recordings before and after classical or multisensory OT to compare modulatory effects. Additionally, single-cell recordings from OE (Durante et al., 2020) could help identify which olfactory receptors contribute to multisensory integration. Such studies could help refine future iterations of MOT. Our experiments were limited to behavioral approaches; hence electrophysiological evidence could elucidate the processes that happen in the brain and highlight how MOT effects the brain during and after the training.

In the future, researchers should consider employing non-invasive brain imaging methods such as fMRI or EEG. By using fMRI, experimenters could show the areas in the brain that are activated by MOT compared to COT. This would also help in understanding whether MOT recruit more brain

regions as in original multisensory perception mechanisms. By using EEG, experimenters can investigate similar mechanisms whether MOT would lead to an increase in signals compared to COT. Invasive methods such as single cell-recordings could be an option in patients that are already going through surgery.

4.1.3. Congruency

In our studies, the strong multisensory interactions among olfaction, gustation, and vision are tested within the framework of congruency. We demonstrated that congruence plays a significant role in multisensory perception, particularly in likeness perception. In dual stimulations, congruent gustatory or visual stimuli accompanying olfactory stimuli produced higher likeness ratings compared to incongruent pairings. In triplet stimulations, presenting both congruent gustatory and visual stimuli alongside olfactory stimuli produced even higher likeness scores than dual congruent conditions. Conversely, incongruent triplet stimulations resulted in even lower likeness ratings compared with dual incongruent conditions.

These findings are consistent with previous literature showing that multisensory perception can either enhance or inhibit performance across a range of experimental tasks, including detection (Campanella et al., 2012), associative learning (Eordegh et al., 2019), classification (Wu et al., 2020), and categorization (Li et al., 2020). A plausible explanation is that congruent stimuli match individuals' memory representations, making retrieval and decision-making more efficient. In contrast, incongruent stimuli generate a conflict between sensory input and prior knowledge, requiring additional cognitive processing and potentially delaying responses.

To further understand the cognitive mechanisms of congruency, a future experiment could incorporate EEG to measure decision-making speed and accuracy in response to congruent versus incongruent olfactory-gustatory-visual pairings. Such an approach would clarify the extent to which incongruence slows neural processing and alters the accuracy of perceptual decisions. On the behavioral side, using an olfactometer in combination with a gustometer could provide precise reaction-time measurements and insights into participants' attentional focus when evaluating congruency.

One way to examine innate versus learned congruency is to expose people to unfamiliar stimuli. For example, using an exotic fruit that participants have never encountered, either in smell or taste, may reduce the influence of expectations. While people may still make similarity-based judgements, it becomes more difficult to determine congruency because no strong memory-based assumptions exist about whether the fruit should be sweet, salty, sour, or bitter.

4.1.4. Olfactory plasticity

In mammals, repeated exposure to odorants produces strong olfactory plasticity effects (Sorg & Hochstatter, 1999). It is well established that OSNs undergo continuous renewal within the olfactory epithelium, where both mature and immature stem cells are present (Huart et al., 2013, Durante et al., 2020). This OSN renewal could be examined before and after OT to confirm the effectiveness of OT in humans (or alternatively in animal models). Such examinations, however, would prove to be difficult and unethical unless they are completed during already scheduled surgeries. A multisensory approach may further allow comparisons between patients undergoing COT and those undergoing MOT, providing further insight into whether multimodal stimulation influences OSN expression. Whether this influence would be positively effective or not is still debatable; however one could expect more sensory stimulations from olfactory, gustatory and visual channels to aid in plasticity.

Additionally, the coding of odorants by OSN could be examined by dissecting the OE in animal models genetically closer to humans, such as primates. This approach would enable researchers to investigate how MOT influences cellular-level changes within the OE and OSNs, including neuronal number, renewal, activation patterns, and gene expression.

Olfactory plasticity is also observable at the electrophysiological level. Increased electro-olfactograms amplitudes have been recorded at the OE following OT (Hummel et al., 2018). Multisensory paradigms could be used to test whether MOT induces stronger or more rapid changes than COT by comparing pre- and post-training EOG signals between groups. OB volume measurements may also reveal whether MOT promotes greater structural growth than COT. If there is a greater growth following MOT this could be due to simply stronger and more widespread stimulation of the brain regions due to stimulation from olfaction, gustation and vision.

Plasticity-related changes occur at the cellular level, particularly in mitral cell output and synaptic excitation or inhibition within the OB both spatially and temporally (Spors & Grinvald, 2002). Mitral-tufted cells and olfactory receptor fields become more finely tuned after repeated exposure to odorants (Wilson et al., 2004, Fletcher & Wilson, 2003). Understanding how multisensory training influences these mechanisms that occur at the cellular level would require studies involving human participants. Current knowledge about cellular-level OT effects comes primarily from animal research. While these findings inform our understanding of human olfaction, direct comparisons are limited due to structural differences between humans and various models. Additionally, since different animal models are used across studies, one must keep in mind that cross species comparisons have to be interpreted cautiously.

Investigating OSNs in humans before and after OT could clarify how training affects OSN expression, activation, and renewal. Post-mortem studies are limited due to cell death after extraction. Therefore, primate (chimpanzees, macaque, or bonobo monkeys) remains an accessible alternative. Rolls & Baylis (1994) found multimodal neurons in the monkey orbitofrontal cortex that respond to olfactory, gustatory, and visual stimuli. Additional primate studies show that neurons in the hypothalamus respond visually to food (Rolls et al., 1976), due to projections from orbitofrontal neurons that encode both sight and taste in rhesus monkeys (Thorpe et al., 1983). Gustatory neuronal responses can also be recorded in the medial orbitofrontal cortex of cynomolgus monkeys (Pritchard et al., 2005), and neurons in this region encode taste, texture, and temperature information during oral stimulation (Kadohisa et al., 2005). These findings support the orbitofrontal cortex as a major site of multisensory integration and even a candidate for secondary taste cortex (Rolls et al., 1990).

Rodent studies provide additional mechanistic insights. Repeated exposure to formaldehyde increases odor sensitivity in rats (Sorg & Hochstatter, 1999). Reliable OB transmission in mice requires 30-second inter-odor intervals to avoid habituation (Subramanian et al., 2024). Longer exposure to odorants increases long-term odor preference memory (24h vs 48h), decreases odor thresholds, and enhances neural responsiveness in the anterior piriform cortex (Fontaine et al., 2013). Neurons responsive to both gustatory and olfactory stimuli can be found in the posterior piriform cortex of awake rats (Idris et al., 2023). Single-unit recordings also show that intraoral

delivery of odor-taste mixtures produces unique neural representations in the thalamus, distinct from unimodal odor or taste stimuli (Fredericksen & Samuelsen, 2022). Such multimodal neural representations may strengthen perceptual outcomes by enabling faster, more robust information processing. The strengthening effect may arise from the bidirectional flow of information between the OB and cortex (Gire et al., 2013), which could be amplified through combined olfactory, gustatory, and visual stimulation, given that these sensory modalities interact across multiple integrative brain regions.

To explore multisensory effects, gustatory and olfactory combined stimulations could also be examined; however, direct stimulations of the cortex in humans are invasive and feasible only in rare circumstances (e.g., during neurosurgical procedures). For example, the orbitofrontal cortex responds to olfactory, gustatory, and somatosensory (emotional) stimuli during direct cortical stimulation in neurosurgical epilepsy patients Fox et al. (2018).

After OT or repeated odor exposure, increased activation and connectivity in olfactory, integrative, and somatosensory networks have been documented (Kollndorfer et al., 2015, Rezaeyan et al., 2025). If olfactory stimuli alone enhance connectivity, it is reasonable to hypothesize that adding gustatory and visual stimuli would elicit stronger or more widespread activation patterns. MRI represents a non-invasive tool to test this hypothesis. Future studies could apply MOT alongside portable olfactometers and gustometers to examine training-related structural and functional brain changes. Regions of interests should include not only olfactory areas, but also gustatory and visual cortices. Functional MRI could reveal connectivity differences before and after MOT and identify overlapping activation patterns unique to multisensory processing. Stronger brain activity with multimodal presentations could be observed because of multiple brain regions recruitment. Behavioral tasks such as threshold, discrimination, or identification could be integrated to evaluate how multisensory inputs influence both performance and decision-making speed.

Increases in gray matter volume and cortical thickness have been reported following OT in multisensory brain regions (Gellrich et al., 2018, Han et al., 2021, Al Ain et al., 2019). MOT may stimulate these regions even more robustly by recruiting overlapping olfactory, gustatory, and visual

processing areas simultaneously. This can strengthen the effects of OT. Researchers can further investigate the structural consequences of MOT with MRI.

In summary, multisensory stimulation can effectively promote plasticity in humans. Although we did not employ neurophysiological or neuroimaging methods in this study, our introduction of multisensory components into OT demonstrated behavioral evidence of plasticity. Patients who completed MOT showed significant improvements in olfactory abilities, as reflected in their post-training UPSIT scores.

4.2. Limits, advantages and future considerations

The methodology used in this thesis is new, and the studies presented here address areas of literature that have been comparatively underexplored. However, despite these strengths, the experiments also contain some limitations.

4.2.1. Stimuli

We selected our stimuli based on previously published research. For the odorants, strawberry (Prescott et al., 2004), cheese (Andriot et al., 2024), lemon (Tsushima et al., 2021), and coffee (Stafford et al., 2020) have all been commonly used in earlier studies. The same applies to gustatory components: sucrose (Marks et al., 2012), sodium chloride (Liu et al., 2024), citric acid (Stevenson et al., 1999), and sucrose octaacetate (Isogai & Wise, 2016), all of which have been safely administered in human experiments. Our stimuli had the advantage of being food-grade, thereby simulating naturalistic exposure to odorants. Such edible stimuli tend to be more salient (Morquecho-Campos et al., 2019).

However, a limitation is that although participants were recruited from the local population and evaluated the stimuli in various combinations, individuals from different cultural backgrounds, with varying personal preferences or prior experiences, may interpret congruent or incongruent pairings differently (Gau & Noppeney, 2016). A stimulus combination considered congruent in our design might be perceived as incongruent by others due to cultural differences (Chen et al., 2024), or personal predispositions (Gao et al., 2025). Future studies should therefore account for cultural and individual variability when selecting stimuli. One approach would be to conduct

preliminary evaluations in different demographic groups to identify stimuli that are reliably perceived as congruent or incongruent within the target population.

Another limitation that concerns stimulus, intensity differences between the MOT (Article 2) and the experimental study with healthy participants (Article 1). Although both sets of stimuli were based on prior studies, they might not have been well-optimized (especially in intensity) for OT purposes. The gustatory-olfactory stimuli used in the clinical study (Article 2) were of lower intensity than those used in the healthy participant study (Article 1). This mismatch occurred because initial pilot testing for the clinical study relied on participants' ratings of how "natural" the stimuli smelled or tasted. While conceptually reasonable, this method may not be reliable in populations with OD, who already struggle to perceive "natural" smells and tastes. Consequently, intensities were increased in the healthy participant study but not in the clinical training study. This might have potentially caused the weaker improvements in people's olfactory abilities with the MOT paradigm.

Using higher-intensity stimuli during MOT may have yielded stronger effects. The discrepancy in stimulus strength may explain why MOT did not outperform COT. Future studies should consider using more concentrated and clinically optimized stimuli to enhance training effectiveness.

A revised MOT protocol could incorporate the tested triplets of olfactory-gustatory-visual stimuli from the multisensory perception experiment (Article 1). Additionally, testing the stimuli in individuals with olfactory dysfunction prior to training may help identify optimal concentrations and ensure that the stimuli are both perceptible and meaningful for clinical populations. Future research could also include non-edible stimuli to test if edible stimuli make the training more salient or effective for patients.

4.2.2. Adherence

Previous research has shown that adherence is a critical factor in the success of OT, as it can determine whether individuals' olfactory abilities improve over time (Boscolo-Rizzo et al., 2024). In our study, we did not systematically track adherence to the OT procedure, and this represents an important limitation that future researchers should address. We attempted to mitigate this

issue by contacting participants at the halfway point of the training (six weeks) to confirm whether they wished to continue and to address any questions or concerns they had about the procedure.

Another methodological challenge was that the OT was conducted during the COVID-19 pandemic. As a result, strict regulations prevented face-to-face interactions with participants. This restriction meant that all instructions, guidance, and communication were delivered remotely via Zoom or phone calls. It is possible that the lack of in-person contact introduced confounding factors such as reduced adherence, improper application of the training protocol, or misinterpretation of instructions. Conducting the experiment in a controlled laboratory environment would likely minimize these issues, as researchers could have provided direct demonstrations, monitored performance, and verified correct implementation of the procedures.

4.2.3. Experimental task

In Article 1, our experimental task involved administering droplets to the back of participants' throats while simultaneously presenting images on a computer screen. Participants evaluated likeness scores by responding to the question, "How much strawberry/cheese/lemon/coffee do you think this is?" We deliberately avoided using the words "smell" or "taste" to prevent confusion between olfaction and gustation, as these senses are frequently mixed up by participants (Rozin, 1982).

Participants provided their ratings using Visual Analogue Scales (VAS) displayed on the screen. A VAS consists of a horizontal continuous line, typically ranging from 0 to 10, but in our experiment the scale ranged from 0 to 100. VAS offers the advantage of avoiding the restrictive, ordinal nature of Likert scales (Sung & Wu, 2018). Importantly, participants did not see the numerical values on the scale; numbers were only available in the recorded output, ensuring responses were driven by perceptual judgement rather than numerical anchoring.

A limitation of the experimental paradigm in Article 1 is that we measured only likeness perception. Future research could benefit from incorporating a broader range of tasks to achieve a more comprehensive understanding of multisensory interactions. These could include identification tasks, sensitivity measures, detection threshold assessment, response time, and accuracy measures. Such additions could contribute significantly to the multisensory perception literature

by providing deeper insights about the process and effects of multisensory perception. Furthermore, combining behavioral data with neuroimaging techniques, such as EEG or fMRI, would allow researchers to visualize neural correlates underlying multisensory processing. These methods could help validate behavioral findings and contribute to more robust arguments.

4.2.4. Scope of training

MOT improves olfactory abilities; however, because we recruited only COVID-19 patients in this study, the findings cannot be generalized to OD caused by other conditions. Nevertheless, given that COT has been shown to be effective across a wide range of etiologies, including sinonasal disorders, post-traumatic OD, idiopathic impairments, neurodegenerative diseases, and age-related olfactory decline (Vance et al., 2024), it is reasonable to assume that MOT might also improve olfactory function in these populations. This represents a promising direction for future research.

While it may be tempting to extend the potential effectiveness of MOT to OD caused by other viral infections, researchers should proceed with caution. COVID-19 may differ from other viruses in important ways, including infection rate, degree of inflammatory damage, and the scale and prognosis of olfactory impairment (Landis et al., 2023, Chang et al., 2024). These differences could influence how patients respond to multisensory interventions. Therefore, future studies should investigate whether MOT yields comparable benefits in OD induced by non-COVID viral infections or other etiologies.

4.2.5. Measurement

All measurements in the OT study (Article 2) were conducted via Zoom or phone calls. This limited our testing capabilities, as patients could not visit our laboratory. Consequently, we relied solely on the UPSIT test, which is a self-administered and relatively simple tool for assessing olfactory function. Although the UPSIT is a robust and widely validated measure of olfactory performance, it assesses only odor identification (Doty et al., 1984). It would have been desirable to include additional measures, such as Sniffin' Sticks test, which evaluates odor threshold, discrimination, and identification (Hummel et al., 1997). However, using Sniffin' Sticks remotely was impractical

because the test includes numerous odor pens and is not cost-effective or feasible to ship to participants' homes.

Despite this limitation, the relevance of our findings remains supported by the fact that both UPSIT and Sniffin' Sticks demonstrate strong to very strong test-retest reliability (Bech et al., 2025). As pandemic-related restrictions are no longer in place, future studies could more easily incorporate additional olfactory assessments, including laboratory-based tests requiring in-person administration.

Gustometers offer a controlled method for administering taste solutions through computer-regulated pumps and plastic tubing (Andersen et al., 2019). They allow precise control over timing and concentration of gustatory stimuli (Andersen et al., 2019) and support measurement of gustatory event-related potentials (Hummel et al., 2010). The use of gustometer would also enable measurement of response times, an important indicator in multisensory perception processing (Hecht et al., 2008) and would make it possible to examine how gustatory and visual cues influence olfactory performance in terms of task speed. In addition, gustometer provides advantages in stimulus randomization and delivery (Gudziol & Guntinas-Lichius, 2019).

In Article 1, we did not use a gustometer and therefore measured only participants' likeness perception, limiting the range of data we could collect. Future studies should consider employing a gustometer to investigate additional measures such as reaction times, response accuracy, and task-related speed during multisensory olfaction-gustation-vision processing.

4.2.6. Sex ratio in the experiments

The ratio of women to men is an important consideration in experimental research, particularly in tasks where sex-based differences may influence performance (Legato et al., 2016), and for ensuring that the sample adequately represents the general populations (Galea et al., 2020).

In Article 1, we were able to recruit nearly equal numbers of men and women (23 women, 25 men). This balanced distribution strengthens the generalizability of our findings regarding multisensory perception and provides a more accurate representation of both sexes.

In contrast, the MOT study (Article 2) included a higher proportion of women than men, with an approximately 3:1 ratio (41 women, 15 men). This imbalance should be considered when interpreting the results, as women often perform better than men in olfactory tasks such as odor threshold, discrimination, and identification (Sorokowski et al., 2019). Therefore, the overrepresentation of women in this study may have contributed to elevated olfactory performance outcomes. Future research should consider recruiting balanced number of both men and women in olfactory training studies.

4.2.7. Age of participants

In Article 2, patients affected by long-term OD tended to be older. Because olfactory problems and sensory degradation increase with age (Kondo et al., 2020), this may have contributed to the recruitment of participants who already exhibited lower baseline of olfactory scores. Although we attempted to balance the sample by including both younger and older participants, the mean averages across all training groups remained within the middle-aged range (average ages were: 39.5 (9.6) for COT, 46.3 (13.8) for MOT, and 43.5 (10.1) for placebo groups).

In contrast, all participants in Article 1 were between 18 and 35 years old and reported having normal olfactory abilities. This discrepancy between the two studies should be considered when interpreting and comparing our results. The older age of participants in Article 2 may have contributed to the lower scores as olfactory abilities reduce with age, whereas the younger age of participants in Article 1 could have contributed to higher scores in likeness perception, as younger adults typically perform better in olfactory tasks. However, it should be noted that COT, MOT and controls had average UPSIT scores of 23.9, 23.1, and 23.9 points respectively before the training. These numbers were 29.2, 26.2, and 24.9 for COT, MOT, and controls respectively after the training. Within the normative data, people with anosmia are expected to have below 18 scores in UPSIT test and above 32.7 for normosmia (Doty et al., 1984). In other words, on average participants exhibited hyposmia both before and after the training.

4.3. Conclusion

In summary, this thesis contributes significantly to the literature in two major areas: (1) multisensory perception, by delivering olfactory-gustatory-visual co-stimulations in both dual or triplet formats within the same experimental framework, thereby offering new insights into the relatively unexplored domain of multisensory combinations involving olfaction, (2) olfactory plasticity, by incorporating these multisensory components into COT and demonstrating significant improvements in olfactory abilities among patients with long-term, COVID-19-related OD.

The findings presented here reveal a strong influence of gustatory and visual cues on olfactory perception within the context of congruency and likeness judgements. We show that congruent gustatory or visual stimuli presented alongside olfactory stimuli increase likeness perception, allowing participants to make more accurate perceptual evaluations. Conversely, incongruent gustatory or visual stimuli reduce likeness ratings. Furthermore, when both congruent gustatory and visual cues accompany olfactory stimuli, participants provide even higher likeness ratings than in dual-modality congruent conditions. The opposite pattern also holds: simultaneously presenting both incongruent gustatory and visual cues leads to even lower likeness ratings than dual incongruent presentations.

Together, the results from both studies may prove valuable for future multisensory research involving gustatory, visual, and olfactory stimuli. The tested stimulus combinations from the multisensory perception experiment (Article 1) could be further optimized and incorporated into future MOT protocols across diverse cultural contexts and patient populations.

Moving forward, future research should aim to: (1) improve stimulus selection and intensity through more extensive pilot testing, especially in clinical populations, (2) implement stricter adherence-tracking tools to ensure participants follow OT protocol accurately, (3) employ neuroimaging techniques, such as EEG or fMRI, to visualize neural responses to MOT and to understand underlying mechanisms, and (4) use gustometers, to deliver precisely timed and controlled gustatory stimuli, enabling the measurement of additional dependent variables such as response times and decision-making speed. Incorporating such methodological advancements could

deepen our understanding of the interactions between olfaction, gustation, and vision, and contribute to the development of more effective MOT programs.

References

Abdelalim, A. A., Mohamady, A. A., Elsayed, R. A., Elawady, M. A., & Ghallab, A. F. (2021). Corticosteroid nasal spray for recovery of smell sensation in COVID-19 patients: A randomized controlled trial. Am J Otolaryngol, 42(2), 102884. https://doi.org/10.1016/j.amjoto.2020.102884

Abolmaali, N. D., Hietschold, V., Vogl, T. J., Huttenbrink, K. B., & Hummel, T. (2002). MR evaluation in patients with isolated anosmia since birth or early childhood. AJNR Am J Neuroradiol, 23(1), 157-164. https://www.ncbi.nlm.nih.gov/pubmed/11827889

Adil, M. T., Rahman, R., Whitelaw, D., Jain, V., Al-Taan, O., Rashid, F., Munasinghe, A., & Jambulingam, P. (2021). SARS-CoV-2 and the pandemic of COVID-19. Postgrad Med J, 97(1144), 110-116. https://doi.org/10.1136/postgradmedj-2020-138386

Aid, M., Boero-Teyssier, V., McMahan, K., Dong, R., Doyle, M., Belabbaci, N., Borducchi, E., Collier, A.-r. Y., Mullington, J., & Barouch, D. H. (2025). Long COVID involves activation of proinflammatory and immune exhaustion pathways. Nature Immunology. https://doi.org/10.1038/s41590-025-02353-x

Al Ain, S., Poupon, D., Hetu, S., Mercier, N., Steffener, J., & Frasnelli, J. (2019). Smell training improves olfactory function and alters brain structure. Neuroimage, 189, 45-54. https://doi.org/10.1016/j.neuroimage.2019.01.008

Alarfaj, A. A., Aldrweesh, A. K., Aldoughan, A. F., Alarfaj, S. M., Alabdulqader, F. K., & Alyahya, K. A. (2023). Olfactory Dysfunction following COVID-19 and the Potential Benefits of Olfactory Training. J Clin Med, 12(14). https://doi.org/10.3390/jcm12144761

Albazee, E., Alajmi, S. A., Alkandari, A. M., Aladwani, A. N., Alenezi, Y. Y., Alsaeed, M. A., Uqlah, B., & Abu-Zaid, A. (2025). Platelet-Rich Plasma for COVID-19-Related Olfactory Dysfunction: A Systematic Review and Meta-analysis of Randomized Controlled Trials. Cureus, 17(10), e94386. https://doi.org/10.7759/cureus.94386

Altundag, A., Cayonu, M., Kayabasoglu, G., Salihoglu, M., Tekeli, H., Saglam, O., & Hummel, T. (2015). Modified olfactory training in patients with postinfectious olfactory loss. Laryngoscope, 125(8), 1763-1766. https://doi.org/10.1002/lary.25245

Altundag, A., Temirbekov, D., Haci, C., Yildirim, D., & Cayonu, M. (2021). Olfactory Cleft Width and Volume: Possible Risk Factors for Postinfectious Olfactory Dysfunction. Laryngoscope, 131(1), 5-9. https://doi.org/10.1002/lary.28524

Andersen, C. A., Alfine, L., Ohla, K., & Hochenberger, R. (2019). A new gustometer: Template for the construction of a portable and modular stimulator for taste and lingual touch. Behav Res Methods, 51(6), 2733-2747. https://doi.org/10.3758/s13428-018-1145-1

Anderson, B. J. (2011). Plasticity of gray matter volume: the cellular and synaptic plasticity that underlies volumetric change. Dev Psychobiol, 53(5), 456-465. https://doi.org/10.1002/dev.20563

Andriot, I., Septier, C., Peltier, C., Noirot, E., Barbet, P., Palme, R., Arnould, C., Buchin, S., & Salles, C. (2024). Influence of Cheese Composition on Aroma Content, Release, and Perception. Molecules, 29(14). https://doi.org/10.3390/molecules29143412

Arienzo, A., Gallo, V., Tomassetti, F., Pitaro, N., Pitaro, M., & Antonini, G. (2023). A narrative review of alternative transmission routes of COVID 19: what we know so far. Pathog Glob Health, 117(8), 681-695. https://doi.org/10.1080/20477724.2023.2228048

Arisi, G. M., Foresti, M. L., Mukherjee, S., & Shapiro, L. A. (2012). The role of olfactory stimulus in adult mammalian neurogenesis. Behavioural Brain Research, 227(2), 356-362. https://doi.org/https://doi.org/10.1016/j.bbr.2011.03.050

Aschenbrenner, K., Hummel, C., Teszmer, K., Krone, F., Ishimaru, T., Seo, H. S., & Hummel, T. (2008). The influence of olfactory loss on dietary behaviors. Laryngoscope, 118(1), 135-144. https://doi.org/10.1097/MLG.0b013e318155a4b9

Asvapoositkul, V., Samuthpongtorn, J., Aeumjaturapat, S., Snidvongs, K., Chusakul, S., Seresirikachorn, K., & Kanjanaumporn, J. (2023). Therapeutic options of post-COVID-19 related olfactory dysfunction: a systematic review and meta-analysis. Rhinology, 61(1), 2-11. https://doi.org/10.4193/Rhin22.221

Auvray, M., & Spence, C. (2008). The multisensory perception of flavor. Conscious Cogn, 17(3), 1016-1031. https://doi.org/10.1016/j.concog.2007.06.005

Azzali, G., Gennari, P. U., Maffei, G., & Ferri, T. (1996). Vallate, foliate and fungiform human papillae gustatory cells. An immunocytochemical and ultrastructural study. Minerva Stomatol, 45(9), 363-379. https://www.ncbi.nlm.nih.gov/pubmed/8950860

Baddeley, A. D., & Andrade, J. (2000). Working memory and the vividness of imagery. J Exp Psychol Gen, 129(1), 126-145. https://doi.org/10.1037//0096-3445.129.1.126

Baden, T., & Nilsson, D. E. (2022). Is our retina really upside down? Curr Biol, 32(7), R300-R303. https://doi.org/10.1016/j.cub.2022.02.065

Bang, J. W., Hamilton-Fletcher, G., & Chan, K. C. (2023). Visual Plasticity in Adulthood: Perspectives from Hebbian and Homeostatic Plasticity. Neuroscientist, 29(1), 117-138. https://doi.org/10.1177/10738584211037619

Baptista, A., Vieira, A. M., Capela, E., Juliao, P., & Macedo, A. (2023). COVID-19 fatality rates in hospitalized patients: A new systematic review and meta-analysis. J Infect Public Health, 16(10), 1606-1612. https://doi.org/10.1016/j.jiph.2023.07.006

Barlow, L. A. (2022). The sense of taste: Development, regeneration, and dysfunction. WIREs Mech Dis, 14(3), e1547. https://doi.org/10.1002/wsbm.1547

Barlow, L. A., & Klein, O. D. (2015). Developing and regenerating a sense of taste. Curr Top Dev Biol, 111, 401-419. https://doi.org/10.1016/bs.ctdb.2014.11.012

Beach, T. G., Adler, C. H., Lue, L., Sue, L. I., Bachalakuri, J., Henry-Watson, J., Sasse, J., Boyer, S., Shirohi, S., Brooks, R., Eschbacher, J., White, C. L., 3rd, Akiyama, H., Caviness, J., Shill, H. A., Connor, D. J., Sabbagh, M. N., Walker, D. G., & Arizona Parkinson's Disease, C. (2009). Unified staging system for Lewy body disorders: correlation with nigrostriatal degeneration, cognitive impairment and motor dysfunction. Acta Neuropathol, 117(6), 613-634. https://doi.org/10.1007/s00401-009-0538-8

Bech, M., Jakobsen, K. K., Andersen, I. S. B., Pedersen, C. K., Gronlund, M. W., & von Buchwald, C. (2025). Which is superior, the University of Pennsylvania Smell Identification Test or the threshold, Discrimination and Identification Test for testing human olfaction? A systematic review. Acta Otolaryngol, 1-4. https://doi.org/10.1080/00016489.2025.2458170

Beckman, D., Bonillas, A., Diniz, G. B., Ott, S., Roh, J. W., Elizaldi, S. R., Schmidt, B. A., Sammak, R. L., Van Rompay, K. K. A., Iyer, S. S., & Morrison, J. H. (2022). SARS-CoV-2 infects neurons and induces neuroinflammation in a non-human primate model of COVID-19. Cell Rep, 41(5), 111573. https://doi.org/10.1016/j.celrep.2022.111573

Besser, G., Oswald, M. M., Liu, D. T., Renner, B., & Mueller, C. A. (2020). Flavor education and training in olfactory dysfunction: a pilot study. Eur Arch Otorhinolaryngol, 277(7), 1987-1994. https://doi.org/10.1007/s00405-020-05950-8

Birte-Antina, W., Ilona, C., Antje, H., & Thomas, H. (2018). Olfactory training with older people. Int J Geriatr Psychiatry, 33(1), 212-220. https://doi.org/10.1002/gps.4725

Blankenship, M. L., Grigorova, M., Katz, D. B., & Maier, J. X. (2019). Retronasal Odor Perception Requires Taste Cortex, but Orthonasal Does Not. Curr Biol, 29(1), 62-69 e63. https://doi.org/10.1016/j.cub.2018.11.011

Bobek, E., & Tversky, B. (2016). Creating visual explanations improves learning. Cogn Res Princ Implic, 1(1), 27. https://doi.org/10.1186/s41235-016-0031-6

Boesveldt, S., & Parma, V. (2021). The importance of the olfactory system in human well-being, through nutrition and social behavior. Cell Tissue Res, 383(1), 559-567. https://doi.org/10.1007/s00441-020-03367-7

Bohn, M. K., Hall, A., Sepiashvili, L., Jung, B., Steele, S., & Adeli, K. (2020). Pathophysiology of COVID-19: Mechanisms Underlying Disease Severity and Progression. Physiology (Bethesda), 35(5), 288-301. https://doi.org/10.1152/physiol.00019.2020

Boscolo-Rizzo, P., Hummel, T., Menini, A., Maniaci, A., Uderzo, F., Bigolin, L., & Tirelli, G. (2024). Adherence to olfactory training improves orthonasal and retronasal olfaction in post-COVID-19 olfactory loss. Rhinology, 62(6), 681-688. https://doi.org/10.4193/Rhin24.233

Boyle, J. A., Frasnelli, J., Gerber, J., Heinke, M., & Hummel, T. (2007). Cross-modal integration of intranasal stimuli: a functional magnetic resonance imaging study. Neuroscience, 149(1), 223-231. https://doi.org/10.1016/j.neuroscience.2007.06.045

Bramerson, A., Nordin, S., & Bende, M. (2007). Clinical experience with patients with olfactory complaints, and their quality of life. Acta Otolaryngol, 127(2), 167-174. https://doi.org/10.1080/00016480600801357

Bratman, G. N., Bembibre, C., Daily, G. C., Doty, R. L., Hummel, T., Jacobs, L. F., Kahn, P. H., Jr., Lashus, C., Majid, A., Miller, J. D., Oleszkiewicz, A., Olvera-Alvarez, H., Parma, V., Riederer, A. M., Sieber, N. L., Williams, J., Xiao, J., Yu, C. P., & Spengler, J. D. (2024). Nature and human well-being: The olfactory pathway. Sci Adv, 10(20), eadn3028. https://doi.org/10.1126/sciadv.adn3028

Breer, H. (2008). The sense of smell: reception of flavors. Ann N Y Acad Sci, 1126, 1-6. https://doi.org/10.1196/annals.1433.010

Brice, Y., Morgan, L., Kirmani, M., Kirmani, M., & Udeh, M. C. (2023). COVID-19 Vaccine Evolution and Beyond. Neurosci Insights, 18, 26331055231180543. https://doi.org/10.1177/26331055231180543

Bryche, B., St Albin, A., Murri, S., Lacote, S., Pulido, C., Ar Gouilh, M., Lesellier, S., Servat, A., Wasniewski, M., Picard-Meyer, E., Monchatre-Leroy, E., Volmer, R., Rampin, O., Le Goffic, R., Marianneau, P., & Meunier, N. (2020). Massive transient damage of the olfactory epithelium associated with infection of sustentacular cells by SARS-CoV-2 in golden Syrian hamsters. Brain Behav Immun, 89, 579-586. https://doi.org/10.1016/j.bbi.2020.06.032

Buchanan, T. W., Tranel, D., & Adolphs, R. (2003). A specific role for the human amygdala in olfactory memory. Learn Mem, 10(5), 319-325. https://doi.org/10.1101/lm.62303

Buschhuter, D., Smitka, M., Puschmann, S., Gerber, J. C., Witt, M., Abolmaali, N. D., & Hummel, T. (2008). Correlation between olfactory bulb volume and olfactory function. Neuroimage, 42(2), 498-502. https://doi.org/10.1016/j.neuroimage.2008.05.004

Cabral-Marques, O., Halpert, G., Schimke, L. F., Ostrinski, Y., Vojdani, A., Baiocchi, G. C., Freire, P. P., Filgueiras, I. S., Zyskind, I., Lattin, M. T., Tran, F., Schreiber, S., Marques, A. H. C., Placa, D. R.,

Fonseca, D. L. M., Humrich, J. Y., Muller, A., Giil, L. M., Grasshoff, H., . . . Shoenfeld, Y. (2022). Autoantibodies targeting GPCRs and RAS-related molecules associate with COVID-19 severity. Nat Commun, 13(1), 1220. https://doi.org/10.1038/s41467-022-28905-5

Cain, W. S., Stevens, J. C., Nickou, C. M., Giles, A., Johnston, I., & Garcia-Medina, M. R. (1995). Life-Span Development of Odor Identification, Learning, and Olfactory Sensitivity. Perception, 24(12), 1457-1472. https://doi.org/10.1068/p241457

Campanella, S., Delle-Vigne, D., Kornreich, C., & Verbanck, P. (2012). Greater sensitivity of the P300 component to bimodal stimulation in an event-related potentials oddball task. Clin Neurophysiol, 123(5), 937-946. https://doi.org/10.1016/j.clinph.2011.10.041

Casado-Aranda, L. A., Van der Laan, L. N., & Sanchez-Fernandez, J. (2018). Neural correlates of gender congruence in audiovisual commercials for gender-targeted products: An fMRI study. Hum Brain Mapp, 39(11), 4360-4372. https://doi.org/10.1002/hbm.24276

Castanares-Zapatero, D., Chalon, P., Kohn, L., Dauvrin, M., Detollenaere, J., Maertens de Noordhout, C., Primus-de Jong, C., Cleemput, I., & Van den Heede, K. (2022). Pathophysiology and mechanism of long COVID: a comprehensive review. Ann Med, 54(1), 1473-1487. https://doi.org/10.1080/07853890.2022.2076901

Cerf-Ducastel, B., & Murphy, C. (2001). fMRI activation in response to odorants orally delivered in aqueous solutions. Chem Senses, 26(6), 625-637. https://doi.org/10.1093/chemse/26.6.625

Cha, M. J., Solomon, J. J., Lee, J. E., Choi, H., Chae, K. J., Lee, K. S., & Lynch, D. A. (2024). Chronic Lung Injury after COVID-19 Pneumonia: Clinical, Radiologic, and Histopathologic Perspectives. Radiology, 310(1), e231643. https://doi.org/10.1148/radiol.231643

Chamard, C., Maller, J. J., Menjot, N., Debourdeau, E., Nael, V., Ritchie, K., Carriere, I., & Daien, V. (2022). Association Between Vision and Brain Cortical Thickness in a Community-Dwelling Elderly Cohort. Eye Brain, 14, 71-82. https://doi.org/10.2147/EB.S358384

Chang, K., Zaikos, T., Kilner-Pontone, N., & Ho, C. Y. (2024). Mechanisms of COVID-19-associated olfactory dysfunction. Neuropathol Appl Neurobiol, 50(2), e12960. https://doi.org/10.1111/nan.12960

Chao, Y. T., Aden, F., Goktas, O., Schmidt, F., Goktas, G., Jurkov, M., Georgsdorf, W., & Hummel, T. (2024). Can immersive olfactory training serve as an alternative treatment for patients with smell dysfunction? Laryngoscope Investig Otolaryngol, 9(3), e1270. https://doi.org/10.1002/lio2.1270

Chaumon, M., Kveraga, K., Barrett, L. F., & Bar, M. (2014). Visual predictions in the orbitofrontal cortex rely on associative content. Cereb Cortex, 24(11), 2899-2907. https://doi.org/10.1093/cercor/bht146

Chen, P., Chung-Fat-Yim, A., Guo, T., & Marian, V. (2024). Cultural background and input familiarity influence multisensory emotion perception. Cultur Divers Ethnic Minor Psychol, 30(3), 487-496. https://doi.org/10.1037/cdp0000577

Chen, W. R., & Shepherd, G. M. (2005). The olfactory glomerulus: a cortical module with specific functions. J Neurocytol, 34(3-5), 353-360. https://doi.org/10.1007/s11068-005-8362-0

Chen, X., Guo, W., Yu, L., Luo, D., Xie, L., & Xu, J. (2021). Association Between Anxious Symptom Severity and Olfactory Impairment in Young Adults with Generalized Anxiety Disorder: A Case-Control Study. Neuropsychiatr Dis Treat, 17, 2877-2883. https://doi.org/10.2147/NDT.S314857

Chen, Y. C., & Spence, C. (2017). Assessing the Role of the 'Unity Assumption' on Multisensory Integration: A Review. Front Psychol, 8, 445. https://doi.org/10.3389/fpsyg.2017.00445

Chin, C. J., Scott, J. R., & Lee, J. M. (2025). Diagnosis and management of chronic rhinosinusitis. CMAJ, 197(6), E148-E154. https://doi.org/10.1503/cmaj.241101

Choi, B. Y., Jeong, H., Noh, H., Park, J. Y., Cho, J. H., & Kim, J. K. (2021). Effects of Olfactory Training in Patients With Postinfectious Olfactory Dysfunction. Clin Exp Otorhinolaryngol, 14(1), 88-92. https://doi.org/10.21053/ceo.2020.00143

Choi, I., Demir, I., Oh, S., & Lee, S. H. (2023). Multisensory integration in the mammalian brain: diversity and flexibility in health and disease. Philos Trans R Soc Lond B Biol Sci, 378(1886), 20220338. https://doi.org/10.1098/rstb.2022.0338

Corsello, G., & Giuffre, M. (2012). Congenital malformations. J Matern Fetal Neonatal Med, 25 Suppl 1, 25-29. https://doi.org/10.3109/14767058.2012.664943

Costanzo, R. M., & Miwa, T. (2006). Posttraumatic olfactory loss. Adv Otorhinolaryngol, 63, 99-107. https://doi.org/10.1159/000093753

Courtiol, E., & Wilson, D. A. (2014). Thalamic olfaction: characterizing odor processing in the mediodorsal thalamus of the rat. J Neurophysiol, 111(6), 1274-1285. https://doi.org/10.1152/jn.00741.2013

Courtiol, E., & Wilson, D. A. (2015). The olfactory thalamus: unanswered questions about the role of the mediodorsal thalamic nucleus in olfaction. Front Neural Circuits, 9, 49. https://doi.org/10.3389/fncir.2015.00049

Croijmans, I., & Majid, A. (2016). Not All Flavor Expertise Is Equal: The Language of Wine and Coffee Experts. PLoS One, 11(6), e0155845. https://doi.org/10.1371/journal.pone.0155845

Croy, I., Bojanowski, V., & Hummel, T. (2013). Men without a sense of smell exhibit a strongly reduced number of sexual relationships, women exhibit reduced partnership security - a reanalysis of previously published data. Biol Psychol, 92(2), 292-294. https://doi.org/10.1016/j.biopsycho.2012.11.008

Croy, I., Negoias, S., Novakova, L., Landis, B. N., & Hummel, T. (2012). Learning about the functions of the olfactory system from people without a sense of smell. PLoS One, 7(3), e33365. https://doi.org/10.1371/journal.pone.0033365

Croy, I., Nordin, S., & Hummel, T. (2014). Olfactory disorders and quality of life--an updated review. Chem Senses, 39(3), 185-194. https://doi.org/10.1093/chemse/bjt072

Dalton, P., Doolittle, N., Nagata, H., & Breslin, P. A. (2000). The merging of the senses: integration of subthreshold taste and smell. Nat Neurosci, 3(5), 431-432. https://doi.org/10.1038/74797

Damm, M., Pikart, L. K., Reimann, H., Burkert, S., Goktas, O., Haxel, B., Frey, S., Charalampakis, I., Beule, A., Renner, B., Hummel, T., & Huttenbrink, K. B. (2014). Olfactory training is helpful in

postinfectious olfactory loss: a randomized, controlled, multicenter study. Laryngoscope, 124(4), 826-831. https://doi.org/10.1002/lary.24340

Damon, F., Mezrai, N., Magnier, L., Leleu, A., Durand, K., & Schaal, B. (2021). Olfaction in the Multisensory Processing of Faces: A Narrative Review of the Influence of Human Body Odors. Front Psychol, 12, 750944. https://doi.org/10.3389/fpsyg.2021.750944

Dazzi, F., Nitto, S. D., Zambetti, G., Loriedo, C., & Ciofalo, A. (2013). Alterations of the olfactory-gustatory functions in patients with eating disorders. Eur Eat Disord Rev, 21(5), 382-385. https://doi.org/10.1002/erv.2238

de Araujo, I. E., Rolls, E. T., Kringelbach, M. L., McGlone, F., & Phillips, N. (2003). Taste-olfactory convergence, and the representation of the pleasantness of flavour, in the human brain. Eur J Neurosci, 18(7), 2059-2068. https://doi.org/10.1046/j.1460-9568.2003.02915.x

de Araujo, I. E., Rolls, E. T., Velazco, M. I., Margot, C., & Cayeux, I. (2005). Cognitive modulation of olfactory processing. Neuron, 46(4), 671-679. https://doi.org/10.1016/j.neuron.2005.04.021

de Araujo, I. E., & Simon, S. A. (2009). The gustatory cortex and multisensory integration. Int J Obes (Lond), 33 Suppl 2(Suppl 2), S34-43. https://doi.org/10.1038/ijo.2009.70

de Groot, J. H., Smeets, M. A., Rowson, M. J., Bulsing, P. J., Blonk, C. G., Wilkinson, J. E., & Semin, G. R. (2015). A sniff of happiness. Psychol Sci, 26(6), 684-700. https://doi.org/10.1177/0956797614566318

de Groot, J. H. B., & Smeets, M. A. M. (2017). Human Fear Chemosignaling: Evidence from a Meta-Analysis. Chem Senses, 42(8), 663-673. https://doi.org/10.1093/chemse/bjx049

Delgado-Lima, A. H., Bouhaben, J., & Delgado-Losada, M. L. (2024). Maximizing Participation in Olfactory Training in a Sample with Post-COVID-19 Olfactory Loss. Brain Sci, 14(7). https://doi.org/10.3390/brainsci14070730

Dematte, M. L., Sanabria, D., & Spence, C. (2009). Olfactory discrimination: when vision matters? Chem Senses, 34(2), 103-109. https://doi.org/10.1093/chemse/bjn055

Denis, F., Septans, A. L., Periers, L., Maillard, J. M., Legoff, F., Gurden, H., & Moriniere, S. (2021). Olfactory Training and Visual Stimulation Assisted by a Web Application for Patients With Persistent Olfactory Dysfunction After SARS-CoV-2 Infection: Observational Study. J Med Internet Res, 23(5), e29583. https://doi.org/10.2196/29583

Deperrois, N., & Graupner, M. (2020). Short-term depression and long-term plasticity together tune sensitive range of synaptic plasticity. PLoS Comput Biol, 16(9), e1008265. https://doi.org/10.1371/journal.pcbi.1008265

Desiato, V. M., Levy, D. A., Byun, Y. J., Nguyen, S. A., Soler, Z. M., & Schlosser, R. J. (2021). The Prevalence of Olfactory Dysfunction in the General Population: A Systematic Review and Meta-analysis. Am J Rhinol Allergy, 35(2), 195-205. https://doi.org/10.1177/1945892420946254

Desrosiers, M., Evans, G. A., Keith, P. K., Wright, E. D., Kaplan, A., Bouchard, J., Ciavarella, A., Doyle, P. W., Javer, A. R., Leith, E. S., Mukherji, A., Robert Schellenberg, R., Small, P., & Witterick, I. J. (2011). Canadian clinical practice guidelines for acute and chronic rhinosinusitis. J Otolaryngol Head Neck Surg, 40 Suppl 2, S99-193. https://www.ncbi.nlm.nih.gov/pubmed/21658337

Dessie, Z. G., & Zewotir, T. (2021). Mortality-related risk factors of COVID-19: a systematic review and meta-analysis of 42 studies and 423,117 patients. BMC Infect Dis, 21(1), 855. https://doi.org/10.1186/s12879-021-06536-3

Dias, M., Shaida, Z., Haloob, N., & Hopkins, C. (2024). Recovery rates and long-term olfactory dysfunction following COVID-19 infection. World J Otorhinolaryngol Head Neck Surg, 10(2), 121-128. https://doi.org/10.1002/wjo2.163

Djordjevic, J., Zatorre, R. J., & Jones-Gotman, M. (2004a). Effects of perceived and imagined odors on taste detection. Chem Senses, 29(3), 199-208. https://doi.org/10.1093/chemse/bjh022

Djordjevic, J., Zatorre, R. J., & Jones-Gotman, M. (2004b). Odor-induced changes in taste perception. Exp Brain Res, 159(3), 405-408. https://doi.org/10.1007/s00221-004-2103-y

Donelli, D., Antonelli, M., & Valussi, M. (2023). Olfactory training with essential oils for patients with post-COVID-19 smell dysfunction: A case series. Eur J Integr Med, 60, 102253. https://doi.org/10.1016/j.eujim.2023.102253

Doty, R. L. (2008). The olfactory vector hypothesis of neurodegenerative disease: is it viable? Ann Neurol, 63(1), 7-15. https://doi.org/10.1002/ana.21327

Doty, R. L. (2012). Olfactory dysfunction in Parkinson disease. Nat Rev Neurol, 8(6), 329-339. https://doi.org/10.1038/nrneurol.2012.80

Doty, R. L. (2019). Treatments for smell and taste disorders: A critical review. Handb Clin Neurol, 164, 455-479. https://doi.org/10.1016/B978-0-444-63855-7.00025-3

Doty, R. L., & Kamath, V. (2014). The influences of age on olfaction: a review. Front Psychol, 5, 20. https://doi.org/10.3389/fpsyg.2014.00020

Doty, R. L., Shaman, P., & Dann, M. (1984). Development of the University of Pennsylvania Smell Identification Test: a standardized microencapsulated test of olfactory function. Physiol Behav, 32(3), 489-502. https://doi.org/10.1016/0031-9384(84)90269-5

Doyle, M. E., Appleton, A., Liu, Q. R., Yao, Q., Mazucanti, C. H., & Egan, J. M. (2021). Human Type II Taste Cells Express Angiotensin-Converting Enzyme 2 and Are Infected by Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2). Am J Pathol, 191(9), 1511-1519. https://doi.org/10.1016/j.ajpath.2021.05.010

Driver, J., & Spence, C. (2000). Multisensory perception: beyond modularity and convergence. Curr Biol, 10(20), R731-735. https://doi.org/10.1016/s0960-9822(00)00740-5

Duchamp-Viret, P., Lacroix, M.-C., Kuszewski, N., & Baly, C. (2016). 3 - Olfactory perception and integration. In P. Etiévant, E. Guichard, C. Salles, & A. Voilley (Eds.), Flavor (pp. 57-100). Woodhead Publishing. https://doi.org/https://doi.org/10.1016/B978-0-08-100295-7.00003-7

Duff, K., McCaffrey, R. J., & Solomon, G. S. (2002). The Pocket Smell Test: successfully discriminating probable Alzheimer's dementia from vascular dementia and major depression. J Neuropsychiatry Clin Neurosci, 14(2), 197-201. https://doi.org/10.1176/jnp.14.2.197

Durante, M. A., Kurtenbach, S., Sargi, Z. B., Harbour, J. W., Choi, R., Kurtenbach, S., Goss, G. M., Matsunami, H., & Goldstein, B. J. (2020). Single-cell analysis of olfactory neurogenesis and

differentiation in adult humans. Nat Neurosci, 23(3), 323-326. https://doi.org/10.1038/s41593-020-0587-9

Eavri, R., & Nedivi, E. (2013). Synaptogenesis in the Adult CNS – Neocortical Plasticity. Cellular Migration and Formation of Neuronal Connections, 713-721. doi:10.1016/B978-0-12-397266-8.00110-1

Ekstrom, A. D. (2015). Why vision is important to how we navigate. Hippocampus, 25(6), 731-735. https://doi.org/10.1002/hipo.22449

Elkholi, S. M. A., Abdelwahab, M. K., & Abdelhafeez, M. (2021). Impact of the smell loss on the quality of life and adopted coping strategies in COVID-19 patients. Eur Arch Otorhinolaryngol, 278(9), 3307-3314. https://doi.org/10.1007/s00405-020-06575-7

Engen, T., & Bosack, T. N. (1969). Facilitation in olfactory detection. J Comp Physiol Psychol, 68(3), 320-326. https://doi.org/10.1037/h0027437

Eordegh, G., Oze, A., Bodosi, B., Puszta, A., Pertich, A., Rosu, A., Godo, G., & Nagy, A. (2019). Multisensory guided associative learning in healthy humans. PLoS One, 14(3), e0213094. https://doi.org/10.1371/journal.pone.0213094

Ernst, M. O., & Bulthoff, H. H. (2004). Merging the senses into a robust percept. Trends Cogn Sci, 8(4), 162-169. https://doi.org/10.1016/j.tics.2004.02.002

Ferdenzi, C., Poncelet, J., Rouby, C., & Bensafi, M. (2014). Repeated exposure to odors induces affective habituation of perception and sniffing. Front Behav Neurosci, 8, 119. https://doi.org/10.3389/fnbeh.2014.00119

Fieux, M., Castro, B. R., Jang, S. S., Yan, C. H., & Patel, Z. M. (2025). Long-term Outcomes of PRP Injections for Post-viral Olfactory Loss: A Prospective Cohort Study. Int Forum Allergy Rhinol, 15(4), 420-427. https://doi.org/10.1002/alr.23505

Fleiner, F., Lau, L., & Goktas, O. (2012). Active olfactory training for the treatment of smelling disorders. Ear Nose Throat J, 91(5), 198-203, 215. https://doi.org/10.1177/014556131209100508

Fletcher, M. L., & Wilson, D. A. (2003). Olfactory bulb mitral-tufted cell plasticity: odorant-specific tuning reflects previous odorant exposure. J Neurosci, 23(17), 6946-6955. https://doi.org/10.1523/JNEUROSCI.23-17-06946.2003

Fokkens, W. J., Lund, V. J., Mullol, J., Bachert, C., Alobid, I., Baroody, F., Cohen, N., Cervin, A., Douglas, R., Gevaert, P., Georgalas, C., Goossens, H., Harvey, R., Hellings, P., Hopkins, C., Jones, N., Joos, G., Kalogjera, L., Kern, B., . . . Wormald, P. J. (2012). European Position Paper on Rhinosinusitis and Nasal Polyps 2012. Rhinol Suppl, 23, 3 p preceding table of contents, 1-298. https://www.ncbi.nlm.nih.gov/pubmed/22764607

Fontaine, C. J., Harley, C. W., & Yuan, Q. (2013). Lateralized odor preference training in rat pups reveals an enhanced network response in anterior piriform cortex to olfactory input that parallels extended memory. J Neurosci, 33(38), 15126-15131. https://doi.org/10.1523/JNEUROSCI.2503-13.2013

Fontana, L., Albayay, J., Fernandez-Prieto, I., & Zampini, M. (2023). Olfactory habituation to food and nonfood odours. Q J Exp Psychol (Hove), 76(6), 1209-1219. https://doi.org/10.1177/17470218221115046

Fox, K. C. R., Yih, J., Raccah, O., Pendekanti, S. L., Limbach, L. E., Maydan, D. D., & Parvizi, J. (2018). Changes in subjective experience elicited by direct stimulation of the human orbitofrontal cortex. Neurology, 91(16), e1519-e1527. https://doi.org/10.1212/WNL.0000000000006358

Frasnelli, J., Fark, T., Lehmann, J., Gerber, J., & Hummel, T. (2013). Brain structure is changed in congenital anosmia. Neuroimage, 83, 1074-1080. https://doi.org/10.1016/j.neuroimage.2013.07.070

Fredericksen, K. E., & Samuelsen, C. L. (2022). Neural Representation of Intraoral Olfactory and Gustatory Signals by the Mediodorsal Thalamus in Alert Rats. J Neurosci, 42(43), 8136-8153. https://doi.org/10.1523/JNEUROSCI.0674-22.2022

Frere, J. J., Serafini, R. A., Pryce, K. D., Zazhytska, M., Oishi, K., Golynker, I., Panis, M., Zimering, J., Horiuchi, S., Hoagland, D. A., Moller, R., Ruiz, A., Kodra, A., Overdevest, J. B., Canoll, P. D., Borczuk, A. C., Chandar, V., Bram, Y., Schwartz, R., . . . tenOever, B. R. (2022). SARS-CoV-2 infection in

hamsters and humans results in lasting and unique systemic perturbations after recovery. Sci Transl Med, 14(664), eabq3059. https://doi.org/10.1126/scitranslmed.abq3059

Fuentes-Prior, P. (2021). Priming of SARS-CoV-2 S protein by several membrane-bound serine proteinases could explain enhanced viral infectivity and systemic COVID-19 infection. J Biol Chem, 296, 100135. https://doi.org/10.1074/jbc.REV120.015980

Gadhave, D. G., Sugandhi, V. V., Jha, S. K., Nangare, S. N., Gupta, G., Singh, S. K., Dua, K., Cho, H., Hansbro, P. M., & Paudel, K. R. (2024). Neurodegenerative disorders: Mechanisms of degeneration and therapeutic approaches with their clinical relevance. Ageing Res Rev, 99, 102357. https://doi.org/10.1016/j.arr.2024.102357

Gadye, L., Das, D., Sanchez, M. A., Street, K., Baudhuin, A., Wagner, A., Cole, M. B., Choi, Y. G., Yosef, N., Purdom, E., Dudoit, S., Risso, D., Ngai, J., & Fletcher, R. B. (2017). Injury Activates Transient Olfactory Stem Cell States with Diverse Lineage Capacities. Cell Stem Cell, 21(6), 775-790 e779. https://doi.org/10.1016/j.stem.2017.10.014

Gagnon, L., Vestergaard, M., Madsen, K., Karstensen, H. G., Siebner, H., Tommerup, N., Kupers, R., & Ptito, M. (2014). Neural correlates of taste perception in congenital olfactory impairment. Neuropsychologia, 62, 297-305. https://doi.org/10.1016/j.neuropsychologia.2014.07.018

Galea, L. A. M., Choleris, E., Albert, A. Y. K., McCarthy, M. M., & Sohrabji, F. (2020). The promises and pitfalls of sex difference research. Front Neuroendocrinol, 56, 100817. https://doi.org/10.1016/j.yfrne.2019.100817

Gao, Y., Xue, K., Odegaard, B., & Rahnev, D. (2025). Automatic multisensory integration follows subjective confidence rather than objective performance. Commun Psychol, 3(1), 38. https://doi.org/10.1038/s44271-025-00221-w

Garcia, E. C. D., Rosa, L. R., Santos, A., Ishimatsu, G. K. A., Lopes, N. M. D., & Fornazieri, M. A. (2024). Investigating the efficacy of melatonin, topical sodium citrate, and multivitamin with zinc as a potential treatment for postinfectious loss of smell. Braz J Otorhinolaryngol, 90(6), 101496. https://doi.org/10.1016/j.bjorl.2024.101496

Gasmi, A., Tippairote, T., Mujawdiya, P. K., Gasmi Benahmed, A., Menzel, A., Dadar, M., & Bjorklund, G. (2021). Neurological Involvements of SARS-CoV2 Infection. Mol Neurobiol, 58(3), 944-949. https://doi.org/10.1007/s12035-020-02070-6

Gau, R., & Noppeney, U. (2016). How prior expectations shape multisensory perception. Neuroimage, 124(Pt A), 876-886. https://doi.org/10.1016/j.neuroimage.2015.09.045

Geissler, K., Reimann, H., Gudziol, H., Bitter, T., & Guntinas-Lichius, O. (2014). Olfactory training for patients with olfactory loss after upper respiratory tract infections. Eur Arch Otorhinolaryngol, 271(6), 1557-1562. https://doi.org/10.1007/s00405-013-2747-y

Gellrich, J., Czaplewski, L., Hahner, A., & Hummel, T. (2024). Palmitoylethanolamide and Luteolin for Postinfectious Olfactory Disorders: How Clinically Meaningful Is Its Effect? ORL J Otorhinolaryngol Relat Spec, 86(3-4), 118-123. https://doi.org/10.1159/000539651

Gellrich, J., Han, P., Manesse, C., Betz, A., Junghanns, A., Raue, C., Schriever, V. A., & Hummel, T. (2018). Brain volume changes in hyposmic patients before and after olfactory training. Laryngoscope, 128(7), 1531-1536. https://doi.org/10.1002/lary.27045

Genetzaki, S., Nikolaidis, V., Markou, K., & Konstantinidis, I. (2024). Olfactory training with four and eight odors: comparison with clinical testing and olfactory bulb volumetrics. Eur Arch Otorhinolaryngol, 281(1), 497-502. https://doi.org/10.1007/s00405-023-08283-4

Georges, A., & Das, J. M. (2025). Traumatic Brain Injury (Archive). In StatPearls. https://www.ncbi.nlm.nih.gov/pubmed/29083790

Gerkin, R. C., Ohla, K., Veldhuizen, M. G., Joseph, P. V., Kelly, C. E., Bakke, A. J., Steele, K. E., Farruggia, M. C., Pellegrino, R., Pepino, M. Y., Bouysset, C., Soler, G. M., Pereda-Loth, V., Dibattista, M., Cooper, K. W., Croijmans, I., Di Pizio, A., Ozdener, M. H., Fjaeldstad, A. W., . . . Author, G. G. (2021). Recent Smell Loss Is the Best Predictor of COVID-19 Among Individuals With Recent Respiratory Symptoms. Chem Senses, 46. https://doi.org/10.1093/chemse/bjaa081

Gillmeister, A., Pieniak, M., & Hummel, T. (2025). Investigating plasticity of the olfactory system through the lens of specific anosmia. Cortex, 192, 53-63. https://doi.org/10.1016/j.cortex.2025.08.012

Gire, D. H., Restrepo, D., Sejnowski, T. J., Greer, C., De Carlos, J. A., & Lopez-Mascaraque, L. (2013). Temporal processing in the olfactory system: can we see a smell? Neuron, 78(3), 416-432. https://doi.org/10.1016/j.neuron.2013.04.033

Girum, T., Lentiro, K., Geremew, M., Migora, B., Shewamare, S., & Shimbre, M. S. (2021). Optimal strategies for COVID-19 prevention from global evidence achieved through social distancing, stay at home, travel restriction and lockdown: a systematic review. Arch Public Health, 79(1), 150. https://doi.org/10.1186/s13690-021-00663-8

Gogele, M., Emmert, D., Fuchsberger, C., & Frasnelli, J. (2024). Factors influencing olfactory function in an adult general population sample: the CHRIS study. Chem Senses, 49. https://doi.org/10.1093/chemse/bjae011

Goodspeed, R. B., Gent, J. F., & Catalanotto, F. A. (1987). Chemosensory dysfunction. Clinical evaluation results from a taste and smell clinic. Postgrad Med, 81(1), 251-257, 260. https://doi.org/10.1080/00325481.1987.11699680

Gottfried, J. A., & Dolan, R. J. (2003). The nose smells what the eye sees: crossmodal visual facilitation of human olfactory perception. Neuron, 39(2), 375-386. https://doi.org/10.1016/s0896-6273(03)00392-1

Gouras, G. K., Olsson, T. T., & Hansson, O. (2015). beta-Amyloid peptides and amyloid plaques in Alzheimer's disease. Neurotherapeutics, 12(1), 3-11. https://doi.org/10.1007/s13311-014-0313-y

Gravina, S. A., Yep, G. L., & Khan, M. (2013). Human biology of taste. Ann Saudi Med, 33(3), 217-222. https://doi.org/10.5144/0256-4947.2013.217

Green, P., Rohling, M. L., Iverson, G. L., & Gervais, R. O. (2003). Relationships between olfactory discrimination and head injury severity. Brain Inj, 17(6), 479-496. https://doi.org/10.1080/0269905031000070242

Greve, A., Cooper, E., Tibon, R., & Henson, R. N. (2019). Knowledge is power: Prior knowledge aids memory for both congruent and incongruent events, but in different ways. J Exp Psychol Gen, 148(2), 325-341. https://doi.org/10.1037/xge0000498

Gudziol, H., & Guntinas-Lichius, O. (2019). Electrophysiologic assessment of olfactory and gustatory function. Handb Clin Neurol, 164, 247-262. https://doi.org/10.1016/B978-0-444-63855-7.00016-2

Gudziol, V., Buschhuter, D., Abolmaali, N., Gerber, J., Rombaux, P., & Hummel, T. (2009). Increasing olfactory bulb volume due to treatment of chronic rhinosinusitis--a longitudinal study. Brain, 132(Pt 11), 3096-3101. https://doi.org/10.1093/brain/awp243

Hakki, S., Zhou, J., Jonnerby, J., Singanayagam, A., Barnett, J. L., Madon, K. J., Koycheva, A., Kelly, C., Houston, H., Nevin, S., Fenn, J., Kundu, R., Crone, M. A., Pillay, T. D., Ahmad, S., Derqui-Fernandez, N., Conibear, E., Freemont, P. S., Taylor, G. P., . . . investigators, A. s. (2022). Onset and window of SARS-CoV-2 infectiousness and temporal correlation with symptom onset: a prospective, longitudinal, community cohort study. Lancet Respir Med, 10(11), 1061-1073. https://doi.org/10.1016/S2213-2600(22)00226-0

Halabi, O., & Saleh, M. (2021). Augmented reality flavor: cross-modal mapping across gustation, olfaction, and vision. Multimed Tools Appl, 80(30), 36423-36441. https://doi.org/10.1007/s11042-021-11321-0

Han, P., Musch, M., Abolmaali, N., & Hummel, T. (2021). Improved Odor Identification Ability and Increased Regional Gray Matter Volume After Olfactory Training in Patients With Idiopathic Olfactory Loss. Iperception, 12(2), 20416695211005811. https://doi.org/10.1177/20416695211005811

Han, S. A., Kim, J. K., Cho, D. Y., Patel, Z. M., & Rhee, C. S. (2023). The Olfactory System: Basic Anatomy and Physiology for General Otorhinolaryngologists. Clin Exp Otorhinolaryngol, 16(4), 308-316. https://doi.org/10.21053/ceo.2023.00185

Haxel, B. R., Nisius, A., Fruth, K., Mann, W. J., & Muttray, A. (2012). [Deficits in medical counseling in olfactory dysfunction]. HNO, 60(5), 432-438. https://doi.org/10.1007/s00106-011-2448-z (Defizite der arztlichen Beratung bei Riechstorungen.)

Hecht, D., Reiner, M., & Karni, A. (2008). Multisensory enhancement: gains in choice and in simple response times. Exp Brain Res, 189(2), 133-143. https://doi.org/10.1007/s00221-008-1410-0

Heikkinen, T., & Jarvinen, A. (2003). The common cold. Lancet, 361(9351), 51-59. https://doi.org/10.1016/S0140-6736(03)12162-9

Hikmet, F., Mear, L., Edvinsson, A., Micke, P., Uhlen, M., & Lindskog, C. (2020). The protein expression profile of ACE2 in human tissues. Mol Syst Biol, 16(7), e9610. https://doi.org/10.15252/msb.20209610

Hill, D. L. (2004). Neural plasticity in the gustatory system. Nutr Rev, 62(11 Pt 2), S208-217; discussion S224-241. https://doi.org/10.1111/j.1753-4887.2004.tb00101.x

Hoffman, H. J., Ishii, E. K., & MacTurk, R. H. (1998). Age-related changes in the prevalence of smell/taste problems among the United States adult population. Results of the 1994 disability supplement to the National Health Interview Survey (NHIS). Ann N Y Acad Sci, 855, 716-722. https://doi.org/10.1111/j.1749-6632.1998.tb10650.x

Hoffmann, M., Kleine-Weber, H., Schroeder, S., Kruger, N., Herrler, T., Erichsen, S., Schiergens, T. S., Herrler, G., Wu, N. H., Nitsche, A., Muller, M. A., Drosten, C., & Pohlmann, S. (2020). SARS-CoV-2 Cell Entry Depends on ACE2 and TMPRSS2 and Is Blocked by a Clinically Proven Protease Inhibitor. Cell, 181(2), 271-280 e278. https://doi.org/10.1016/j.cell.2020.02.052

Hosseini, K., Zare-Sadeghi, A., Sadigh-Eteghad, S., Mirsalehi, M., & Khezerloo, D. (2020). Effects of olfactory training on resting-state effective connectivity in patients with posttraumatic olfactory dysfunction. Acta Neurobiol Exp (Wars), 80(4), 381-388. https://www.ncbi.nlm.nih.gov/pubmed/33350991

Howell, J., Costanzo, R. M., & Reiter, E. R. (2018). Head trauma and olfactory function. World J Otorhinolaryngol Head Neck Surg, 4(1), 39-45. https://doi.org/10.1016/j.wjorl.2018.02.001

Hu, B., Zhang, J., Gong, M., Deng, Y., Cao, Y., Xiang, Y., & Ye, D. (2022). Research Progress of Olfactory Nerve Regeneration Mechanism and Olfactory Training. Ther Clin Risk Manag, 18, 185-195. https://doi.org/10.2147/TCRM.S354695

Huart, C., Rombaux, P., & Hummel, T. (2013). Plasticity of the human olfactory system: the olfactory bulb. Molecules, 18(9), 11586-11600. https://doi.org/10.3390/molecules180911586

Hummel, T., Genow, A., & Landis, B. N. (2010). Clinical assessment of human gustatory function using event related potentials. J Neurol Neurosurg Psychiatry, 81(4), 459-464. https://doi.org/10.1136/jnnp.2009.183699

Hummel, T., Liu, D. T., Muller, C. A., Stuck, B. A., Welge-Lussen, A., & Hahner, A. (2023). Olfactory Dysfunction: Etiology, Diagnosis, and Treatment. Dtsch Arztebl Int, 120(9), 146-154. https://doi.org/10.3238/arztebl.m2022.0411

Hummel, T., Rissom, K., Reden, J., Hahner, A., Weidenbecher, M., & Huttenbrink, K. B. (2009). Effects of olfactory training in patients with olfactory loss. Laryngoscope, 119(3), 496-499. https://doi.org/10.1002/lary.20101

Hummel, T., Sekinger, B., Wolf, S. R., Pauli, E., & Kobal, G. (1997). 'Sniffin' sticks': olfactory performance assessed by the combined testing of odor identification, odor discrimination and olfactory threshold. Chem Senses, 22(1), 39-52. https://doi.org/10.1093/chemse/22.1.39

Hummel, T., Stupka, G., Haehner, A., & Poletti, S. C. (2018). Olfactory training changes electrophysiological responses at the level of the olfactory epithelium. Rhinology, 56(4), 330-335. https://doi.org/10.4193/Rhin17.163

Hummel, T., Whitcroft, K. L., Andrews, P., Altundag, A., Cinghi, C., Costanzo, R. M., Damm, M., Frasnelli, J., Gudziol, H., Gupta, N., Haehne, A., Holbrook, E., Hong, S. C., Hornung, D., Huttenbrink, K. B., Kamel, R., Kobayashi, M., Konstantinidis, I., Landis, B. N., . . . Welge-Luessen, A. (2017). Position paper on olfactory dysfunction. Rhinol Suppl, 54(26), 1-30. https://doi.org/10.4193/Rhino16.248

Hummel, T., Whitcroft, K. L., Rueter, G., & Haehner, A. (2017). Intranasal vitamin A is beneficial in post-infectious olfactory loss. Eur Arch Otorhinolaryngol, 274(7), 2819-2825. https://doi.org/10.1007/s00405-017-4576-x

Ibrahim, S., Siemieniuk, R. A. C., Oliveros, M. J., Islam, N., Diaz Martinez, J. P., Izcovich, A., Qasim, A., Zhao, Y., Zaror, C., Yao, L., Wang, Y., Vandvik, P. O., Roldan, Y., Rochwerg, B., Rada, G., Prasad, M., Pardo-Hernandez, H., Mustafa, R. A., Fashami, F. M., . . . Brignardello-Petersen, R. (2025). Drug

treatments for mild or moderate covid-19: systematic review and network meta-analysis. BMJ, 389, e081165. https://doi.org/10.1136/bmj-2024-081165

Idris, A., Christensen, B. A., Walker, E. M., & Maier, J. X. (2023). Multisensory integration of orally-sourced gustatory and olfactory inputs to the posterior piriform cortex in awake rats. J Physiol, 601(1), 151-169. https://doi.org/10.1113/JP283873

Innocenti, G. M. (2022). Defining neuroplasticity. Handb Clin Neurol, 184, 3-18. https://doi.org/10.1016/B978-0-12-819410-2.00001-1

Ireland, A. C., & Carter, I. B. (2025). Neuroanatomy, Optic Chiasm. In StatPearls. https://www.ncbi.nlm.nih.gov/pubmed/31194427

Isogai, T., & Wise, P. M. (2016). The Effects of Odor Quality and Temporal Asynchrony on Modulation of Taste Intensity by Retronasal Odor. Chem Senses, 41(7), 557-566. https://doi.org/10.1093/chemse/bjw059

Jehl, C., Royet, J. P., & Holley, A. (1994). Very short term recognition memory for odors. Percept Psychophys, 56(6), 658-668. https://doi.org/10.3758/bf03208359

Jehl, C., Royet, J. P., & Holley, A. (1995). Odor discrimination and recognition memory as a function of familiarization. Percept Psychophys, 57(7), 1002-1011. https://doi.org/10.3758/bf03205459

Jobin, B., Boller, B., & Frasnelli, J. (2021). Volumetry of Olfactory Structures in Mild Cognitive Impairment and Alzheimer's Disease: A Systematic Review and a Meta-Analysis. Brain Sci, 11(8). https://doi.org/10.3390/brainsci11081010

Jobin, B., Boller, B., Frasnelli, J., & group, C.-Q. (2023). Smaller grey matter volume in the central olfactory system in mild cognitive impairment. Exp Gerontol, 183, 112325. https://doi.org/10.1016/j.exger.2023.112325

Johansson, C. B., Svensson, M., Wallstedt, L., Janson, A. M., & Frisén, J. (1999). Neural Stem Cells in the Adult Human Brain. Experimental Cell Research, 253(2), 733-736. https://doi.org/https://doi.org/10.1006/excr.1999.4678

Jonsson, F. U., & Olsson, M. J. (2003). Olfactory metacognition. Chem Senses, 28(7), 651-658. https://doi.org/10.1093/chemse/bjg058

Kadohisa, M., Rolls, E. T., & Verhagen, J. V. (2005). Neuronal representations of stimuli in the mouth: the primate insular taste cortex, orbitofrontal cortex and amygdala. Chem Senses, 30(5), 401-419. https://doi.org/10.1093/chemse/bji036

Kandemirli, S. G., Altundag, A., Yildirim, D., Tekcan Sanli, D. E., & Saatci, O. (2021). Olfactory Bulb MRI and Paranasal Sinus CT Findings in Persistent COVID-19 Anosmia. Acad Radiol, 28(1), 28-35. https://doi.org/10.1016/j.acra.2020.10.006

Kassab, A., Schaub, F., Vent, J., Huttenbrink, K. B., & Damm, M. (2009). Effects of short inter-stimulus intervals on olfactory and trigeminal event-related potentials. Acta Otolaryngol, 129(11), 1250-1256. https://doi.org/10.3109/00016480802644605

Kattar, N., Do, T. M., Unis, G. D., Migneron, M. R., Thomas, A. J., & McCoul, E. D. (2021). Olfactory Training for Postviral Olfactory Dysfunction: Systematic Review and Meta-analysis. Otolaryngol Head Neck Surg, 164(2), 244-254. https://doi.org/10.1177/0194599820943550

Kaye, A. D., Okeagu, C. N., Tortorich, G., Pham, A. D., Ly, E. I., Brondeel, K. C., Eng, M. R., Luedi, M. M., Urman, R. D., & Cornett, E. M. (2021). COVID-19 impact on the renal system: Pathophysiology and clinical outcomes. Best Pract Res Clin Anaesthesiol, 35(3), 449-459. https://doi.org/10.1016/j.bpa.2021.02.004

Keogh, R., & Pearson, J. (2011). Mental imagery and visual working memory. PLoS One, 6(12), e29221. https://doi.org/10.1371/journal.pone.0029221

Khan, A. M., Piccirillo, J., Kallogjeri, D., & Piccirillo, J. F. (2023). Efficacy of Combined Visual-Olfactory Training With Patient-Preferred Scents as Treatment for Patients With COVID-19 Resultant Olfactory Loss: A Randomized Clinical Trial. JAMA Otolaryngol Head Neck Surg, 149(2), 141-149. https://doi.org/10.1001/jamaoto.2022.4112

Khan, M., Yoo, S. J., Clijsters, M., Backaert, W., Vanstapel, A., Speleman, K., Lietaer, C., Choi, S., Hether, T. D., Marcelis, L., Nam, A., Pan, L., Reeves, J. W., Van Bulck, P., Zhou, H., Bourgeois, M., Debaveye, Y., De Munter, P., Gunst, J., . . . Van Gerven, L. (2021). Visualizing in deceased COVID-

19 patients how SARS-CoV-2 attacks the respiratory and olfactory mucosae but spares the olfactory bulb. Cell, 184(24), 5932-5949 e5915. https://doi.org/10.1016/j.cell.2021.10.027

Khorisantono, P. A., Veldhuizen, M. G., & Seubert, J. (2025). Tastes and retronasal odours evoke a shared flavour-specific neural code in the human insula. Nature Communications, 16(1), 8252. https://doi.org/10.1038/s41467-025-63803-6

Kim, B. Y., Park, J., Kim, E., & Kim, B. (2020). Olfactory Ensheathing Cells Mediate Neuroplastic Mechanisms After Olfactory Training in Mouse Model. Am J Rhinol Allergy, 34(2), 217-229. https://doi.org/10.1177/1945892419885036

Kim, B. Y., Park, J. Y., Kim, E. J., Kim, B. G., Kim, S. W., & Kim, S. W. (2019). The neuroplastic effect of olfactory training to the recovery of olfactory system in mouse model. Int Forum Allergy Rhinol, 9(7), 715-723. https://doi.org/10.1002/alr.22320

Kim, G. U., Kim, M. J., Ra, S. H., Lee, J., Bae, S., Jung, J., & Kim, S. H. (2020). Clinical characteristics of asymptomatic and symptomatic patients with mild COVID-19. Clin Microbiol Infect, 26(7), 948 e941-948 e943. https://doi.org/10.1016/j.cmi.2020.04.040

Kim, K., Bae, J., Jin, Y., & Moon, C. (2020). Odor habituation can modulate very early olfactory event-related potential. Sci Rep, 10(1), 18117. https://doi.org/10.1038/s41598-020-75263-7

Kim, R. S., Seitz, A. R., & Shams, L. (2008). Benefits of stimulus congruency for multisensory facilitation of visual learning. PLoS One, 3(1), e1532. https://doi.org/10.1371/journal.pone.0001532

Kim, S., Finlay, J. B., Ko, T., & Goldstein, B. J. (2024). Long-term olfactory loss post-COVID-19: Pathobiology and potential therapeutic strategies. World J Otorhinolaryngol Head Neck Surg, 10(2), 148-155. https://doi.org/10.1002/wjo2.165

Kim, Y. K., Hong, S. L., Yoon, E. J., Kim, S. E., & Kim, J. W. (2012). Central presentation of postviral olfactory loss evaluated by positron emission tomography scan: a pilot study. Am J Rhinol Allergy, 26(3), 204-208. https://doi.org/10.2500/ajra.2012.26.3759

King, L. R. (2024). Gastrointestinal manifestations of long COVID. Life Sci, 357, 123100. https://doi.org/10.1016/j.lfs.2024.123100

Kinnamon, S. C. (2012). Taste receptor signalling - from tongues to lungs. Acta Physiol (Oxf), 204(2), 158-168. https://doi.org/10.1111/j.1748-1716.2011.02308.x

Knudsen, E. I. (2004). Sensitive periods in the development of the brain and behavior. J Cogn Neurosci, 16(8), 1412-1425. https://doi.org/10.1162/0898929042304796

Kohli, P., Soler, Z. M., Nguyen, S. A., Muus, J. S., & Schlosser, R. J. (2016). The Association Between Olfaction and Depression: A Systematic Review. Chem Senses, 41(6), 479-486. https://doi.org/10.1093/chemse/bjw061

Koike, K., Yoo, S. J., Bleymehl, K., Omura, M., Zapiec, B., Pyrski, M., Blum, T., Khan, M., Bai, Z., Leinders-Zufall, T., Mombaerts, P., & Zufall, F. (2021). Danger perception and stress response through an olfactory sensor for the bacterial metabolite hydrogen sulfide. Neuron, 109(15), 2469-2484 e2467. https://doi.org/10.1016/j.neuron.2021.05.032

Kollndorfer, K., Fischmeister, F. P., Kowalczyk, K., Hoche, E., Mueller, C. A., Trattnig, S., & Schopf, V. (2015). Olfactory training induces changes in regional functional connectivity in patients with long-term smell loss. Neuroimage Clin, 9, 401-410. https://doi.org/10.1016/j.nicl.2015.09.004

Kondo, K., Kikuta, S., Ueha, R., Suzukawa, K., & Yamasoba, T. (2020). Age-Related Olfactory Dysfunction: Epidemiology, Pathophysiology, and Clinical Management. Front Aging Neurosci, 12, 208. https://doi.org/10.3389/fnagi.2020.00208

Kornack, D. R., & Rakic, P. (2001). The generation, migration, and differentiation of olfactory neurons in the adult primate brain. Proc Natl Acad Sci U S A, 98(8), 4752-4757. https://doi.org/10.1073/pnas.081074998

Koyama, S., & Heinbockel, T. (2022). Chemical Constituents of Essential Oils Used in Olfactory Training: Focus on COVID-19 Induced Olfactory Dysfunction. Front Pharmacol, 13, 835886. https://doi.org/10.3389/fphar.2022.835886

Koyou, H. L., Salleh, M. N., Jelemie, C. S., Badrin, M. J. Q., Prastiyanto, M. E., & Ramachandran, V. (2025). TMPRSS2: A Key Host Factor in SARS-CoV-2 Infection and Potential Therapeutic Target. Medeni Med J, 26(4), 101-109. https://doi.org/10.4274/MMJ.galenos.2025.40460

Krishna, B. A., Metaxaki, M., Sithole, N., Landin, P., Martin, P., & Salinas-Botran, A. (2024). Cardiovascular disease and covid-19: A systematic review. Int J Cardiol Heart Vasc, 54, 101482. https://doi.org/10.1016/j.ijcha.2024.101482

Landis, B. N., Frasnelli, J., & Hummel, T. (2006). Euosmia: a rare form of parosmia. Acta Otolaryngol, 126(1), 101-103. https://doi.org/10.1080/00016480510043954

Landis, B. N., Hsieh, J. W., Konstantinidis, I., Holbrook, E. H., Huart, C., Mori, E., & Hummel, T. (2023). What Is Different in COVID-19-Associated Olfactory Loss from Classical Postinfectious Olfactory Impairment? ORL J Otorhinolaryngol Relat Spec, 85(5), 245-247. https://doi.org/10.1159/000533228

Laurienti, P. J., Kraft, R. A., Maldjian, J. A., Burdette, J. H., & Wallace, M. T. (2004). Semantic congruence is a critical factor in multisensory behavioral performance. Exp Brain Res, 158(4), 405-414. https://doi.org/10.1007/s00221-004-1913-2

Le Bon, S. D., Konopnicki, D., Pisarski, N., Prunier, L., Lechien, J. R., & Horoi, M. (2021). Efficacy and safety of oral corticosteroids and olfactory training in the management of COVID-19-related loss of smell. Eur Arch Otorhinolaryngol, 278(8), 3113-3117. https://doi.org/10.1007/s00405-020-06520-8

Lechien, J. R., Le Bon, S. D., & Saussez, S. (2023). Platelet-rich plasma injection in the olfactory clefts of COVID-19 patients with long-term olfactory dysfunction. Eur Arch Otorhinolaryngol, 280(5), 2351-2358. https://doi.org/10.1007/s00405-022-07788-8

Lechien, J. R., & Saussez, S. (2024). Injection of Platelet Rich Plasma in the Olfactory Cleft for COVID-19 Patients With Persistent Olfactory Dysfunction: Description of the Technique. Ear Nose Throat J, 103(1_suppl), 115S-119S. https://doi.org/10.1177/01455613221124773

Lechien, J. R., Saussez, S., Vaira, L. A., De Riu, G., Boscolo-Rizzo, P., Tirelli, G., Michel, J., & Radulesco, T. (2024). Effectiveness of Platelet-Rich Plasma for COVID-19-Related Olfactory Dysfunction: A Controlled Study. Otolaryngol Head Neck Surg, 170(1), 84-91. https://doi.org/10.1002/ohn.460

Lee, J. C., Nallani, R., Cass, L., Bhalla, V., Chiu, A. G., & Villwock, J. A. (2021). A Systematic Review of the Neuropathologic Findings of Post-Viral Olfactory Dysfunction: Implications and Novel Insight for the COVID-19 Pandemic. Am J Rhinol Allergy, 35(3), 323-333. https://doi.org/10.1177/1945892420957853

Lee, L., Luke, L., Boak, D., & Philpott, C. (2024). Impact of olfactory disorders on personal safety and well-being: a cross-sectional observational study. Eur Arch Otorhinolaryngol, 281(7), 3639-3647. https://doi.org/10.1007/s00405-024-08529-9

Legato, M. J., Johnson, P. A., & Manson, J. E. (2016). Consideration of Sex Differences in Medicine to Improve Health Care and Patient Outcomes. JAMA, 316(18), 1865-1866. https://doi.org/10.1001/jama.2016.13995

Lehmann, S., & Murray, M. M. (2005). The role of multisensory memories in unisensory object discrimination. Cognitive Brain Research, 24(2), 326-334. https://doi.org/https://doi.org/10.1016/j.cogbrainres.2005.02.005

Li, Y., Seger, C., Chen, Q., & Mo, L. (2020). Left Inferior Frontal Gyrus Integrates Multisensory Information in Category Learning. Cereb Cortex, 30(8), 4410-4423. https://doi.org/10.1093/cercor/bhaa029

Li, Z., Anne, A., & Hummel, T. (2023). Olfactory training: effects of multisensory integration, attention towards odors and physical activity. Chem Senses, 48. https://doi.org/10.1093/chemse/bjad037

Li, Z., Li, S. B., Tan, S., Liu, L. L., Yan, C., & Zou, L. Q. (2025). Neural correlates of olfactory working memory in the human brain. Neuroimage, 306, 121005. https://doi.org/10.1016/j.neuroimage.2025.121005

Lindemann, B. (1996). Taste reception. Physiol Rev, 76(3), 719-766. https://doi.org/10.1152/physrev.1996.76.3.719

Liu, G., Zong, G., Doty, R. L., & Sun, Q. (2016). Prevalence and risk factors of taste and smell impairment in a nationwide representative sample of the US population: a cross-sectional study. BMJ Open, 6(11), e013246. https://doi.org/10.1136/bmjopen-2016-013246

Liu, S., Gu, Y., Zheng, R., Sun, B., Zhang, L., & Zhang, Y. (2024). Progress in Multisensory Synergistic Salt Reduction. Foods, 13(11). https://doi.org/10.3390/foods13111659

Liu, Y., & Rocklov, J. (2022). The effective reproductive number of the Omicron variant of SARS-CoV-2 is several times relative to Delta. J Travel Med, 29(3). https://doi.org/10.1093/jtm/taac037

Lois, C., Garcia-Verdugo, J. M., & Alvarez-Buylla, A. (1996). Chain migration of neuronal precursors. Science, 271(5251), 978-981. doi:10.1126/science.271.5251.978

Lovelace, C. T., Stein, B. E., & Wallace, M. T. (2003). An irrelevant light enhances auditory detection in humans: a psychophysical analysis of multisensory integration in stimulus detection. Brain Res Cogn Brain Res, 17(2), 447-453. https://doi.org/10.1016/s0926-6410(03)00160-5

Lucijanic, M., Marelic, D., Stojic, J., Markovic, I., Sedlic, F., Kralj, I., Rucevic, D., Busic, N., Javor, P., Lucijanic, T., Mitrovic, J., & Luksic, I. (2023). Predictors of prolonged hospitalization of COVID-19 patients. Eur Geriatr Med, 14(3), 511-516. https://doi.org/10.1007/s41999-023-00787-w

Luisier, A. C., Petitpierre, G., Clerc Berod, A., Garcia-Burgos, D., & Bensafi, M. (2019). Effects of familiarization on odor hedonic responses and food choices in children with autism spectrum disorders. Autism, 23(6), 1460-1471. https://doi.org/10.1177/1362361318815252

Lundstrom, J. N., Boyle, J. A., Zatorre, R. J., & Jones-Gotman, M. (2009). The neuronal substrates of human olfactory based kin recognition. Hum Brain Mapp, 30(8), 2571-2580. https://doi.org/10.1002/hbm.20686

Mahmut, M. K., Pieniak, M., Resler, K., Schriever, V. A., Haehner, A., & Oleszkiewicz, A. (2021). Olfactory training in 8-year-olds increases odour identification ability: a preliminary study. Eur J Pediatr, 180(7), 2049-2053. https://doi.org/10.1007/s00431-021-03970-y

Maier, J. X. (2017). Single-neuron responses to intraoral delivery of odor solutions in primary olfactory and gustatory cortex. J Neurophysiol, 117(3), 1293-1304. https://doi.org/10.1152/jn.00802.2016

Maier, J. X., Blankenship, M. L., Li, J. X., & Katz, D. B. (2015). A Multisensory Network for Olfactory Processing. Curr Biol, 25(20), 2642-2650. https://doi.org/10.1016/j.cub.2015.08.060

Maier, J. X., Wachowiak, M., & Katz, D. B. (2012). Chemosensory convergence on primary olfactory cortex. J Neurosci, 32(48), 17037-17047. https://doi.org/10.1523/JNEUROSCI.3540-12.2012

Malik, B., Elkaddi, N., Turkistani, J., Spielman, A. I., & Ozdener, M. H. (2019). Mammalian Taste Cells Express Functional Olfactory Receptors. Chem Senses, 44(5), 289-301. https://doi.org/10.1093/chemse/bjz019

Maril, A., Avital, R., Reggev, N., Zuckerman, M., Sadeh, T., Ben Sira, L., & Livneh, N. (2011). Event congruency and episodic encoding: a developmental fMRI study. Neuropsychologia, 49(11), 3036-3045. https://doi.org/10.1016/j.neuropsychologia.2011.07.004

Marino-Sanchez, F. S., Alobid, I., Cantellas, S., Alberca, C., Guilemany, J. M., Canals, J. M., De Haro, J., & Mullol, J. (2010). Smell training increases cognitive smell skills of wine tasters compared to the general healthy population. The WINECAT Study. Rhinology, 48(3), 273-276. https://doi.org/10.4193/Rhino09.206

Marino, A. N., Mui, M., Dobryakova, E., & Sandry, J. (2025). Classification Systems for Moderate-Severe Traumatic Brain Injury: Review and Recommendations. Current Physical Medicine and Rehabilitation Reports, 13(1), 32. https://doi.org/10.1007/s40141-025-00504-7

Marks, L. E., Shepard, T. G., Burger, K., & Chakwin, E. M. (2012). Flavor-intensity perception: effects of stimulus context. Physiol Behav, 105(2), 443-450. https://doi.org/10.1016/j.physbeh.2011.08.039

Marzola, P., Melzer, T., Pavesi, E., Gil-Mohapel, J., & Brocardo, P. S. (2023). Exploring the Role of Neuroplasticity in Development, Aging, and Neurodegeneration. Brain Sci, 13(12). https://doi.org/10.3390/brainsci13121610

Mattes, R. D., & Cowart, B. J. (1994). Dietary assessment of patients with chemosensory disorders. J Am Diet Assoc, 94(1), 50-56. https://doi.org/10.1016/0002-8223(94)92041-9

Mavroudis, I., Ciobica, A., Bejenariu, A. C., Dobrin, R. P., Apostu, M., Dobrin, I., & Balmus, I. M. (2024). Cognitive Impairment following Mild Traumatic Brain Injury (mTBI): A Review. Medicina (Kaunas), 60(3). https://doi.org/10.3390/medicina60030380

Mazzola, L., Royet, J. P., Catenoix, H., Montavont, A., Isnard, J., & Mauguiere, F. (2017). Gustatory and olfactory responses to stimulation of the human insula. Ann Neurol, 82(3), 360-370. https://doi.org/10.1002/ana.25010

McGann, J. P. (2015). Associative learning and sensory neuroplasticity: how does it happen and what is it good for? Learn Mem, 22(11), 567-576. https://doi.org/10.1101/lm.039636.115

Mehra, D., & Moshirfar, M. (2025). Neuroanatomy, Optic Tract. In StatPearls. https://www.ncbi.nlm.nih.gov/pubmed/31751030

Menni, C., Valdes, A. M., Freidin, M. B., Sudre, C. H., Nguyen, L. H., Drew, D. A., Ganesh, S., Varsavsky, T., Cardoso, M. J., El-Sayed Moustafa, J. S., Visconti, A., Hysi, P., Bowyer, R. C. E., Mangino, M., Falchi, M., Wolf, J., Ourselin, S., Chan, A. T., Steves, C. J., & Spector, T. D. (2020). Real-time tracking of self-reported symptoms to predict potential COVID-19. Nat Med, 26(7), 1037-1040. https://doi.org/10.1038/s41591-020-0916-2

Migneault-Bouchard, C., Hsieh, J. W., Hugentobler, M., Frasnelli, J., & Landis, B. N. (2020). Chemosensory decrease in different forms of olfactory dysfunction. J Neurol, 267(1), 138-143. https://doi.org/10.1007/s00415-019-09564-x

Milner, A. D. (2017). How do the two visual streams interact with each other? Exp Brain Res, 235(5), 1297-1308. https://doi.org/10.1007/s00221-017-4917-4

Ming, G. L., & Song, H. (2011). Adult neurogenesis in the mammalian brain: significant answers and significant questions. Neuron, 70(4), 687-702. https://doi.org/10.1016/j.neuron.2011.05.001

Monday, H. R., Younts, T. J., & Castillo, P. E. (2018). Long-Term Plasticity of Neurotransmitter Release: Emerging Mechanisms and Contributions to Brain Function and Disease. Annu Rev Neurosci, 41, 299-322. https://doi.org/10.1146/annurev-neuro-080317-062155

Morais, R. F., Pires, R., Jesus, T., Lemos, R., Duro, D., Lima, M., Baldeiras, I., Oliveira, T. G., & Santana, I. (2025). Cognitive impairment in neurodegenerative diseases: A trans-diagnostic approach using a lesion-symptom mapping analysis. Neuroscience, 573, 214-227. https://doi.org/10.1016/j.neuroscience.2025.03.034

Morgan, C. D., & Murphy, C. (2010). Differential effects of active attention and age on event-related potentials to visual and olfactory stimuli. Int J Psychophysiol, 78(2), 190-199. https://doi.org/10.1016/j.ijpsycho.2010.07.008

Morquecho-Campos, P., Bikker, F. J., Nazmi, K., de Graaf, K., Laine, M. L., & Boesveldt, S. (2019). Impact of food odors signaling specific taste qualities and macronutrient content on saliva secretion and composition. Appetite, 143, 104399. https://doi.org/10.1016/j.appet.2019.104399

Morris, H. R., Spillantini, M. G., Sue, C. M., & Williams-Gray, C. H. (2024). The pathogenesis of Parkinson's disease. Lancet, 403(10423), 293-304. https://doi.org/10.1016/S0140-6736(23)01478-2

Moser, P. (2020). Out of Control? Managing Baseline Variability in Experimental Studies with Control Groups. Handb Exp Pharmacol, 257, 101-117. https://doi.org/10.1007/164_2019_280

Motiei, M., Hassanzadeh Rad, A., Badeli, H., & Bayat, R. (2025). Hospitalization dynamics during COVID-19: Insights into disease trends and patient outcomes. PLoS One, 20(4), e0321269. https://doi.org/10.1371/journal.pone.0321269

Negoias, S., Pietsch, K., & Hummel, T. (2017). Changes in olfactory bulb volume following lateralized olfactory training. Brain Imaging Behav, 11(4), 998-1005. https://doi.org/10.1007/s11682-016-9567-9

Nguyen, T. P., & Patel, Z. M. (2018). Budesonide irrigation with olfactory training improves outcomes compared with olfactory training alone in patients with olfactory loss. Int Forum Allergy Rhinol, 8(9), 977-981. https://doi.org/10.1002/alr.22140

Niklassen, A. S., Fernandes, H. M., Linnet, E., Therkildsen, N. B., Hummel, T., Ovesen, T., & Fjaeldstad, A. W. (2025). The plasticity of olfactory learning in culinary students and matched controls. Brain Imaging Behav. https://doi.org/10.1007/s11682-025-01055-0

Noto, T., Zhou, G., Yang, Q., Lane, G., & Zelano, C. (2021). Human Primary Olfactory Amygdala Subregions Form Distinct Functional Networks, Suggesting Distinct Olfactory Functions. Front Syst Neurosci, 15, 752320. https://doi.org/10.3389/fnsys.2021.752320

Novak, L. R., Gitelman, D. R., Schuyler, B., & Li, W. (2015). Olfactory-visual integration facilitates perception of subthreshold negative emotion. Neuropsychologia, 77, 288-297. https://doi.org/10.1016/j.neuropsychologia.2015.09.005

Oleszkiewicz, A., Bottesi, L., Pieniak, M., Fujita, S., Krasteva, N., Nelles, G., & Hummel, T. (2022). Olfactory training with Aromastics: olfactory and cognitive effects. Eur Arch Otorhinolaryngol, 279(1), 225-232. https://doi.org/10.1007/s00405-021-06810-9

Oleszkiewicz, A., Hanf, S., Whitcroft, K. L., Haehner, A., & Hummel, T. (2018). Examination of olfactory training effectiveness in relation to its complexity and the cause of olfactory loss. Laryngoscope, 128(7), 1518-1522. https://doi.org/10.1002/lary.26985

Oleszkiewicz, A., Park, D., Resler, K., Draf, J., Schulze, A., Zang, Y., Hahner, A., & Hummel, T. (2019). Quality of Life in Patients With Olfactory Loss Is Better Predicted by Flavor Identification Than by Orthonasal Olfactory Function. Chem Senses, 44(6), 371-377. https://doi.org/10.1093/chemse/bjz027

Osterbauer, R. A., Matthews, P. M., Jenkinson, M., Beckmann, C. F., Hansen, P. C., & Calvert, G. A. (2005). Color of scents: chromatic stimuli modulate odor responses in the human brain. J Neurophysiol, 93(6), 3434-3441. https://doi.org/10.1152/jn.00555.2004

Otte, M. S., Haehner, A., Bork, M. L., Klussmann, J. P., Luers, J. C., & Hummel, T. (2023). Impact of COVID-19-Mediated Olfactory Loss on Quality of Life. ORL J Otorhinolaryngol Relat Spec, 85(1), 1-6. https://doi.org/10.1159/000523893

Owji, S., & Shoja, M. M. (2020). The History of Discovery of Adult Neurogenesis. Clin Anat, 33(1), 41-55. https://doi.org/10.1002/ca.23447

Pagano, S. F., Impagnatiello, F., Girelli, M., Cova, L., Grioni, E., Onofri, M., Cavallaro, M., Etteri, S., Vitello, F., Giombini, S., Solero, C. L., & Parati, E. A. (2000). Isolation and Characterization of Neural Stem Cells from the Adult Human Olfactory Bulb. Stem Cells, 18(4), 295-300. https://doi.org/10.1634/stemcells.18-4-295

Parr, W. V., Heatherbell, D., & White, K. G. (2002). Demystifying wine expertise: olfactory threshold, perceptual skill and semantic memory in expert and novice wine judges. Chem Senses, 27(8), 747-755. https://doi.org/10.1093/chemse/27.8.747

Patel, Z. M., Wise, S. K., & DelGaudio, J. M. (2017). Randomized Controlled Trial Demonstrating Cost-Effective Method of Olfactory Training in Clinical Practice: Essential Oils at Uncontrolled Concentration. Laryngoscope Investig Otolaryngol, 2(2), 53-56. https://doi.org/10.1002/lio2.62

Pence, T. S., Reiter, E. R., DiNardo, L. J., & Costanzo, R. M. (2014). Risk factors for hazardous events in olfactory-impaired patients. JAMA Otolaryngol Head Neck Surg, 140(10), 951-955. https://doi.org/10.1001/jamaoto.2014.1675

Peron, R. M., & Allen, G. L. (1988). Attempts to train novices for beer flavor discrimination: a matter of taste. J Gen Psychol, 115(4), 403-418. https://doi.org/10.1080/00221309.1988.9710577

Peter, M. G., Porada, D. K., Regenbogen, C., Olsson, M. J., & Lundstrom, J. N. (2019). Sensory loss enhances multisensory integration performance. Cortex, 120, 116-130. https://doi.org/10.1016/j.cortex.2019.06.003

Pieniak, M., Oleszkiewicz, A., Avaro, V., Calegari, F., & Hummel, T. (2022). Olfactory training - Thirteen years of research reviewed. Neurosci Biobehav Rev, 141, 104853. https://doi.org/10.1016/j.neubiorev.2022.104853

Postuma, R. B., Berg, D., Stern, M., Poewe, W., Olanow, C. W., Oertel, W., Obeso, J., Marek, K., Litvan, I., Lang, A. E., Halliday, G., Goetz, C. G., Gasser, T., Dubois, B., Chan, P., Bloem, B. R., Adler, C. H., & Deuschl, G. (2015). MDS clinical diagnostic criteria for Parkinson's disease. Mov Disord, 30(12), 1591-1601. https://doi.org/10.1002/mds.26424

Poupon, D., Fernandez, P., & Frasnelli, J. (2019). Sommelier Students Display Superior Abilities to Identify but Not to Detect or Discriminate Odors Early in their Training. Chemosensory Perception, 12(2), 106-114. https://doi.org/10.1007/s12078-019-09256-w

Power Guerra, N., Kruschwitz, E., Krautwurst, D., & Hummel, T. (2024). Four odorants for olfactory training are enough: a pilot study. Eur Arch Otorhinolaryngol, 281(12), 6445-6458. https://doi.org/10.1007/s00405-024-08930-4

Prasad, V., & Haslam, A. (2024). COVID-19 vaccines: history of the pandemic's great scientific success and flawed policy implementation. Monash Bioeth Rev, 42(1), 28-54. https://doi.org/10.1007/s40592-024-00189-z

Prescott, J. (2004). Psychological Processes in Flavour Perception. In Flavor Perception (pp. 256-278). https://doi.org/https://doi.org/10.1002/9780470995716.ch9

Prescott, J. (2012). Multimodal Chemosensory Interactions and Perception of Flavor. In M. M. Murray & M. T. Wallace (Eds.), The Neural Bases of Multisensory Processes. https://www.ncbi.nlm.nih.gov/pubmed/22593874

Prescott, J., Johnstone, V., & Francis, J. (2004). Odor-taste interactions: effects of attentional strategies during exposure. Chem Senses, 29(4), 331-340. https://doi.org/10.1093/chemse/bjh036

Pritchard, T. C., Edwards, E. M., Smith, C. A., Hilgert, K. G., Gavlick, A. M., Maryniak, T. D., Schwartz, G. J., & Scott, T. R. (2005). Gustatory neural responses in the medial orbitofrontal cortex of the old world monkey. J Neurosci, 25(26), 6047-6056. https://doi.org/10.1523/JNEUROSCI.0430-05.2005

Puderbaugh, M., & Emmady, P. D. (2025). Neuroplasticity. In StatPearls. https://www.ncbi.nlm.nih.gov/pubmed/32491743

Ravindra, K., Malik, V. S., Padhi, B. K., Goel, S., & Gupta, M. (2022). Asymptomatic infection and transmission of COVID-19 among clusters: systematic review and meta-analysis. Public Health, 203, 100-109. https://doi.org/10.1016/j.puhe.2021.12.003

Rebholz, H., Braun, R. J., Ladage, D., Knoll, W., Kleber, C., & Hassel, A. W. (2020). Loss of Olfactory Function-Early Indicator for Covid-19, Other Viral Infections and Neurodegenerative Disorders. Front Neurol, 11, 569333. https://doi.org/10.3389/fneur.2020.569333

Rezaeyan, A., Asadi, S., Kamrava, S. K., & Zare-Sadeghi, A. (2025). Olfactory training affects the correlation between brain structure and functional connectivity. Neuroradiol J, 38(4), 450-463. https://doi.org/10.1177/19714009241303129

Ripp, I., Zur Nieden, A. N., Blankenagel, S., Franzmeier, N., Lundstrom, J. N., & Freiherr, J. (2018). Multisensory integration processing during olfactory-visual stimulation-An fMRI graph theoretical network analysis. Hum Brain Mapp, 39(9), 3713-3727. https://doi.org/10.1002/hbm.24206

Rogers, P. J., & Hill, A. J. (1989). Breakdown of dietary restraint following mere exposure to food stimuli: interrelationships between restraint, hunger, salivation, and food intake. Addict Behav, 14(4), 387-397. https://doi.org/10.1016/0306-4603(89)90026-9

Rolls, E. T. (2008). Functions of the orbitofrontal and pregenual cingulate cortex in taste, olfaction, appetite and emotion. Acta Physiol Hung, 95(2), 131-164. https://doi.org/10.1556/APhysiol.95.2008.2.1

Rolls, E. T., & Baylis, L. L. (1994). Gustatory, olfactory, and visual convergence within the primate orbitofrontal cortex. J Neurosci, 14(9), 5437-5452. https://doi.org/10.1523/JNEUROSCI.14-09-05437.1994

Rolls, E. T., Burton, M. J., & Mora, F. (1976). Hypothalamic neuronal responses associated with the sight of food. Brain Res, 111(1), 53-66. https://doi.org/10.1016/0006-8993(76)91048-9

Rolls, E. T., Grabenhorst, F., Margot, C., da Silva, M. A., & Velazco, M. I. (2008). Selective attention to affective value alters how the brain processes olfactory stimuli. J Cogn Neurosci, 20(10), 1815-1826. https://doi.org/10.1162/jocn.2008.20128

Rolls, E. T., Yaxley, S., & Sienkiewicz, Z. J. (1990). Gustatory responses of single neurons in the caudolateral orbitofrontal cortex of the macaque monkey. J Neurophysiol, 64(4), 1055-1066. https://doi.org/10.1152/jn.1990.64.4.1055

Rombaux, P., Martinage, S., Huart, C., & Collet, S. (2009). Post-infectious olfactory loss: a cohort study and update. B-ENT, 5 Suppl 13, 89-95. https://www.ncbi.nlm.nih.gov/pubmed/20084809

Rozin, P. (1982). "Taste-smell confusions" and the duality of the olfactory sense. Percept Psychophys, 31(4), 397-401. https://doi.org/10.3758/bf03202667

Saatci, O., Altundag, A., Duz, O. A., & Hummel, T. (2020). Olfactory training ball improves adherence and olfactory outcomes in post-infectious olfactory dysfunction. Eur Arch Otorhinolaryngol, 277(7), 2125-2132. https://doi.org/10.1007/s00405-020-05939-3

Saito, Y., Shioya, A., Sano, T., Sumikura, H., Murata, M., & Murayama, S. (2016). Lewy body pathology involves the olfactory cells in Parkinson's disease and related disorders. Mov Disord, 31(1), 135-138. https://doi.org/10.1002/mds.26463

Sanai, N., Tramontin, A. D., Quinones-Hinojosa, A., Barbaro, N. M., Gupta, N., Kunwar, S., Lawton, M. T., McDermott, M. W., Parsa, A. T., Manuel-Garcia Verdugo, J., Berger, M. S., & Alvarez-Buylla, A. (2004). Unique astrocyte ribbon in adult human brain contains neural stem cells but lacks chain migration. Nature, 427(6976), 740-744. https://doi.org/10.1038/nature02301

Sanchez Lopez de Nava, A., Somani, A. N., & Salini, B. (2025). Physiology, Vision. In StatPearls. https://www.ncbi.nlm.nih.gov/pubmed/30860728

Sanchez, M., Garate, A., Delgado, D., & Padilla, S. (2017). Platelet-rich plasma, an adjuvant biological therapy to assist peripheral nerve repair. Neural regeneration research, 12(1), 47-52. https://doi.org/10.4103/1673-5374.198973

Saniasiaya, J., & Prepageran, N. (2021). Impact of olfactory dysfunction on quality of life in coronavirus disease 2019 patients: a systematic review. J Laryngol Otol, 135(11), 947-952. https://doi.org/10.1017/S0022215121002279

Santos, D. V., Reiter, E. R., DiNardo, L. J., & Costanzo, R. M. (2004). Hazardous events associated with impaired olfactory function. Arch Otolaryngol Head Neck Surg, 130(3), 317-319. https://doi.org/10.1001/archotol.130.3.317

Sarnat, H. B., & Flores-Sarnat, L. (2019). Chapter 3 - Development of the human olfactory system. In R. L. Doty (Ed.), Handbook of Clinical Neurology (Vol. 164, pp. 29-45). Elsevier. https://doi.org/https://doi.org/10.1016/B978-0-444-63855-7.00003-4

Saw, C., Friesen, N. D., & Bartley, A. (2022). An Extremely Rare Cause of Isolated Congenital Anosmia. Case Rep Otolaryngol, 2022, 9692716. https://doi.org/10.1155/2022/9692716

Schicker, D., Khorisantono, P. A., Rramani Dervishi, Q., Lim, S. X. L., Saruco, E., Pleger, B., Schultz, J., Ohla, K., & Freiherr, J. (2025). Smell the Label: Odors Influence Label Perception and Their Neural Processing. J Neurosci, 45(14). https://doi.org/10.1523/JNEUROSCI.1159-24.2024

Schifferstein, H. N., & Verlegh, P. W. (1996). The role of congruency and pleasantness in odor-induced taste enhancement. Acta Psychol (Amst), 94(1), 87-105. https://doi.org/10.1016/0001-6918(95)00040-2

Schiffman, S. S. (2018). Influence of medications on taste and smell. World J Otorhinolaryngol Head Neck Surg, 4(1), 84-91. https://doi.org/10.1016/j.wjorl.2018.02.005

Schiffman, S. S., & Graham, B. G. (2000). Taste and smell perception affect appetite and immunity in the elderly. Eur J Clin Nutr, 54 Suppl 3, S54-63. https://doi.org/10.1038/sj.ejcn.1601026

Schleidt, M., Hold, B., & Attili, G. (1981). A cross-cultural study on the attitude towards personal odors. J Chem Ecol, 7(1), 19-31. https://doi.org/10.1007/BF00988632

Schul, R., Slotnick, B. M., & Dudai, Y. (1996). Flavor and the frontal cortex. Behav Neurosci, 110(4), 760-765. https://www.ncbi.nlm.nih.gov/pubmed/8864267

Schurgin, M. W. (2018). Visual memory, the long and the short of it: A review of visual working memory and long-term memory. Atten Percept Psychophys, 80(5), 1035-1056. https://doi.org/10.3758/s13414-018-1522-y

Schwartz, J. S., Tajudeen, B. A., & Kennedy, D. W. (2019). Diseases of the nasal cavity. Handb Clin Neurol, 164, 285-302. https://doi.org/10.1016/B978-0-444-63855-7.00018-6

Schwob, J. E., & Costanzo, R. M. (2010). Regeneration of the Olfactory Epithelium. The Senses: A Comprehensive Reference, 4, 591-612. doi:10.1016/B978-012370880-9.00115-8

Seigneuric, A., Durand, K., Jiang, T., Baudouin, J. Y., & Schaal, B. (2010). The nose tells it to the eyes: crossmodal associations between olfaction and vision. Perception, 39(11), 1541-1554. https://doi.org/10.1068/p6740

Seitz, A. R., Kim, R., & Shams, L. (2006). Sound facilitates visual learning. Curr Biol, 16(14), 1422-1427. https://doi.org/10.1016/j.cub.2006.05.048

Seubert, J., Laukka, E. J., Rizzuto, D., Hummel, T., Fratiglioni, L., Backman, L., & Larsson, M. (2017). Prevalence and Correlates of Olfactory Dysfunction in Old Age: A Population-Based Study. J Gerontol A Biol Sci Med Sci, 72(8), 1072-1079. https://doi.org/10.1093/gerona/glx054

Seubert, J., Ohla, K., Yokomukai, Y., Kellermann, T., & Lundstrom, J. N. (2015). Superadditive opercular activation to food flavor is mediated by enhanced temporal and limbic coupling. Hum Brain Mapp, 36(5), 1662-1676. https://doi.org/10.1002/hbm.22728

Shams, L., & Seitz, A. R. (2008). Benefits of multisensory learning. Trends Cogn Sci, 12(11), 411-417. https://doi.org/10.1016/j.tics.2008.07.006

Shepherd, G. M. (2005). Outline of a theory of olfactory processing and its relevance to humans. Chem Senses, 30 Suppl 1, i3-5. https://doi.org/10.1093/chemse/bjh085

Shereen, M. A., Khan, S., Kazmi, A., Bashir, N., & Siddique, R. (2020). COVID-19 infection: Origin, transmission, and characteristics of human coronaviruses. J Adv Res, 24, 91-98. https://doi.org/10.1016/j.jare.2020.03.005

Shi, Z., & Muller, H. J. (2013). Multisensory perception and action: development, decision-making, and neural mechanisms. Front Integr Neurosci, 7, 81. https://doi.org/10.3389/fnint.2013.00081

Silkis, I. G. (2023). The Role of the Hippocampus in the Perception and Recall of Odors. A Hypothetical Neural Mechanism. Neuroscience and Behavioral Physiology, 53(7), 1255-1268. https://doi.org/10.1007/s11055-023-01521-6

Small, D. M., Bender, G., Veldhuizen, M. G., Rudenga, K., Nachtigal, D., & Felsted, J. (2007). The role of the human orbitofrontal cortex in taste and flavor processing. Ann N Y Acad Sci, 1121, 136-151. https://doi.org/10.1196/annals.1401.002

Small, D. M., Gregory, M. D., Mak, Y. E., Gitelman, D., Mesulam, M. M., & Parrish, T. (2003). Dissociation of neural representation of intensity and affective valuation in human gustation. Neuron, 39(4), 701-711. https://doi.org/10.1016/s0896-6273(03)00467-7

Small, D. M., Jones-Gotman, M., Zatorre, R. J., Petrides, M., & Evans, A. C. (1997). Flavor processing: more than the sum of its parts. Neuroreport, 8(18), 3913-3917. https://doi.org/10.1097/00001756-199712220-00014

Small, D. M., & Prescott, J. (2005). Odor/taste integration and the perception of flavor. Exp Brain Res, 166(3-4), 345-357. https://doi.org/10.1007/s00221-005-2376-9

Small, D. M., Voss, J., Mak, Y. E., Simmons, K. B., Parrish, T., & Gitelman, D. (2004). Experience-dependent neural integration of taste and smell in the human brain. J Neurophysiol, 92(3), 1892-1903. https://doi.org/10.1152/jn.00050.2004

Sommer, U., Hummel, T., Cormann, K., Mueller, A., Frasnelli, J., Kropp, J., & Reichmann, H. (2004). Detection of presymptomatic Parkinson's disease: combining smell tests, transcranial sonography, and SPECT. Mov Disord, 19(10), 1196-1202. https://doi.org/10.1002/mds.20141

Song, Y., Zhang, M., Yin, L., Wang, K., Zhou, Y., Zhou, M., & Lu, Y. (2020). COVID-19 treatment: close to a cure? A rapid review of pharmacotherapies for the novel coronavirus (SARS-CoV-2). Int J Antimicrob Agents, 56(2), 106080. https://doi.org/10.1016/j.ijantimicag.2020.106080

Sorg, B. A., & Hochstatter, T. (1999). Behavioral sensitization after repeated formaldehyde exposure in rats. Toxicol Ind Health, 15(3-4), 346-355. https://doi.org/10.1177/074823379901500309

Sorokowski, P., Karwowski, M., Misiak, M., Marczak, M. K., Dziekan, M., Hummel, T., & Sorokowska, A. (2019). Sex Differences in Human Olfaction: A Meta-Analysis. Front Psychol, 10, 242. https://doi.org/10.3389/fpsyg.2019.00242

Spors, H., & Grinvald, A. (2002). Spatio-temporal dynamics of odor representations in the mammalian olfactory bulb. Neuron, 34(2), 301-315. https://doi.org/10.1016/s0896-6273(02)00644-x

Stafford, L. D., Damant, K., Ashurst, S., & Parker, M. O. (2020). Higher olfactory sensitivity to coffee odour in habitual caffeine users. Exp Clin Psychopharmacol, 28(2), 245-250. https://doi.org/10.1037/pha0000293

Steffens, Y., Le Bon, S. D., Lechien, J., Prunier, L., Rodriguez, A., Saussez, S., & Horoi, M. (2022). Effectiveness and safety of PRP on persistent olfactory dysfunction related to COVID-19. Eur Arch Otorhinolaryngol, 279(12), 5951-5953. https://doi.org/10.1007/s00405-022-07560-y

Stevens, W. W., Schleimer, R. P., & Kern, R. C. (2016). Chronic Rhinosinusitis with Nasal Polyps. J Allergy Clin Immunol Pract, 4(4), 565-572. https://doi.org/10.1016/j.jaip.2016.04.012

Stevenson, R. J. (2010). An initial evaluation of the functions of human olfaction. Chem Senses, 35(1), 3-20. https://doi.org/10.1093/chemse/bjp083

Stevenson, R. J., & Case, T. I. (2005). Olfactory imagery: a review. Psychon Bull Rev, 12(2), 244-264. https://doi.org/10.3758/bf03196369

Stevenson, R. J., Case, T. I., & Mahmut, M. (2007). Difficulty in evoking odor images: the role of odor naming. Mem Cognit, 35(3), 578-589. https://doi.org/10.3758/bf03193296

Stevenson, R. J., Prescott, J., & Boakes, R. A. (1999). Confusing tastes and smells: how odours can influence the perception of sweet and sour tastes. Chem Senses, 24(6), 627-635. https://doi.org/10.1093/chemse/24.6.627

Su, M., Wang, S., Fang, W., Zhu, Y., Li, R., Sheng, K., Zou, D., Han, Y., Wang, X., & Cheng, O. (2015). Alterations in the limbic/paralimbic cortices of Parkinson's disease patients with hyposmia under resting-state functional MRI by regional homogeneity and functional connectivity analysis. Parkinsonism Relat Disord, 21(7), 698-703. https://doi.org/10.1016/j.parkreldis.2015.04.006

Su, W. L., Lu, K. C., Chan, C. Y., & Chao, Y. C. (2021). COVID-19 and the lungs: A review. J Infect Public Health, 14(11), 1708-1714. https://doi.org/10.1016/j.jiph.2021.09.024

Subramanian, N., Leong, L. M., Boukani, P. S. M., & Storace, D. A. (2024). Recent odor experience selectively modulates olfactory sensitivity across the glomerular output in the mouse olfactory bulb. bioRxiv. https://doi.org/10.1101/2024.07.21.604478

Sung, Y. T., & Wu, J. S. (2018). The Visual Analogue Scale for Rating, Ranking and Paired-Comparison (VAS-RRP): A new technique for psychological measurement. Behav Res Methods, 50(4), 1694-1715. https://doi.org/10.3758/s13428-018-1041-8

Suzuki, M., Saito, K., Min, W. P., Vladau, C., Toida, K., Itoh, H., & Murakami, S. (2007). Identification of viruses in patients with postviral olfactory dysfunction. Laryngoscope, 117(2), 272-277. https://doi.org/10.1097/01.mlg.0000249922.37381.1e

Taheri, A., Naderi, M., Jonaidi Jafari, N., Emadi Koochak, H., Saberi Esfeedvajani, M., & Abolghasemi, R. (2024). Therapeutic effects of olfactory training and systemic vitamin A in patients with COVID-19-related olfactory dysfunction: a double-blinded randomized controlled clinical trial. Braz J Otorhinolaryngol, 90(5), 101451. https://doi.org/10.1016/j.bjorl.2024.101451

Tai, K., Leland, E. M., Seal, S. M., Schneider, A. L. C., Rowan, N. R., & Kamath, V. (2023). Olfactory Dysfunction Following Moderate to Severe Traumatic Brain Injury: A Systematic Review and Meta-Analysis. Neuropsychol Rev, 33(4), 717-732. https://doi.org/10.1007/s11065-022-09563-2

Tataranu, L. G., & Rizea, R. E. (2025). Neuroplasticity and Nervous System Recovery: Cellular Mechanisms, Therapeutic Advances, and Future Prospects. Brain Sci, 15(4). https://doi.org/10.3390/brainsci15040400

Taylor, C. A., Whitaker, M., Patton, M. E., Melgar, M., Kirley, P. D., Kawasaki, B., Yousey-Hindes, K., Openo, K. P., Ryan, P. A., Kim, S., Como-Sabetti, K., Solhtalab, D., Barney, G., Tesini, B. L., Moran, N. E., Sutton, M., Talbot, H. K., Olsen, K., & Havers, F. P. (2024). Trends in COVID-19-Attributable Hospitalizations Among Adults With Laboratory-Confirmed SARS-CoV-2-COVID-NET, June 2020 to September 2023. Influenza Other Respir Viruses, 18(11), e70021. https://doi.org/10.1111/irv.70021

Tepe, B., Hill, M. C., Pekarek, B. T., Hunt, P. J., Martin, T. J., Martin, J. F., & Arenkiel, B. R. (2018). Single-Cell RNA-Seq of Mouse Olfactory Bulb Reveals Cellular Heterogeneity and Activity-Dependent Molecular Census of Adult-Born Neurons. Cell Rep, 25(10), 2689-2703 e2683. https://doi.org/10.1016/j.celrep.2018.11.034

Thorpe, S. J., Rolls, E. T., & Maddison, S. (1983). The orbitofrontal cortex: neuronal activity in the behaving monkey. Exp Brain Res, 49(1), 93-115. https://doi.org/10.1007/BF00235545

Tognetti, A., Williams, M. N., Lybert, N., Lekander, M., Axelsson, J., & Olsson, M. J. (2023). Humans can detect axillary odor cues of an acute respiratory infection in others. Evol Med Public Health, 11(1), 219-228. https://doi.org/10.1093/emph/eoad016

Tsushima, Y., Nishino, Y., & Ando, H. (2021). Olfactory Stimulation Modulates Visual Perception Without Training [Original Research]. Frontiers in Neuroscience, Volume 15 - 2021. https://www.frontiersin.org/journals/neuroscience/articles/10.3389/fnins.2021.642584

Vaira, L. A., Hopkins, C., Salzano, G., Petrocelli, M., Melis, A., Cucurullo, M., Ferrari, M., Gagliardini, L., Pipolo, C., Deiana, G., Fiore, V., De Vito, A., Turra, N., Canu, S., Maglio, A., Serra, A., Bussu, F., Madeddu, G., Babudieri, S., . . . De Riu, G. (2020). Olfactory and gustatory function impairment in COVID-19 patients: Italian objective multicenter-study. Head Neck, 42(7), 1560-1569. https://doi.org/10.1002/hed.26269

van Dijk, B., Postma, E. M., Boek, W. M., Kamalski, D. M. A., & Boesveldt, S. (2025). Prevalence and recovery rate of persistent olfactory dysfunction in COVID-19: the COVORTS study. Rhinology, 63(3), 334-342. https://doi.org/10.4193/Rhin24.331

van Ee, R., van Boxtel, J. J., Parker, A. L., & Alais, D. (2009). Multisensory congruency as a mechanism for attentional control over perceptual selection. J Neurosci, 29(37), 11641-11649. https://doi.org/10.1523/JNEUROSCI.0873-09.2009

Vance, D. E., Del Bene, V. A., Kamath, V., Frank, J. S., Billings, R., Cho, D. Y., Byun, J. Y., Jacob, A., Anderson, J. N., Visscher, K., Triebel, K., Martin, K. M., Li, W., Puga, F., & Fazeli, P. L. (2024). Does Olfactory Training Improve Brain Function and Cognition? A Systematic Review. Neuropsychol Rev, 34(1), 155-191. https://doi.org/10.1007/s11065-022-09573-0

Varendi, H., & Porter, R. H. (2001). Breast odour as the only maternal stimulus elicits crawling towards the odour source. Acta Paediatr, 90(4), 372-375. https://www.ncbi.nlm.nih.gov/pubmed/11332925

Veldhuizen, M. G., & Small, D. M. (2011). Modality-specific neural effects of selective attention to taste and odor. Chem Senses, 36(8), 747-760. https://doi.org/10.1093/chemse/bjr043

Villicana, J., Joseph, A., Goldstein, O., Luu, K., Matta, E., Ngo, D., Nakawaki, K., Siebein, K., Sustek, R., Tang, K., Shiraishi, M., Mayorga, A., Moon, S. H., Rajagopal, S., Chauhan, R. K., & Soangra, R. (2025). The Role of Peripheral Vision in Enhancing Balance and Postural Stability: Insights from Central Vision Obstruction. Biomed Sci Instrum, 61(1), 1-8. https://www.ncbi.nlm.nih.gov/pubmed/40599631

von Bartheld, C. S., & Wang, L. (2023). Prevalence of Olfactory Dysfunction with the Omicron Variant of SARS-CoV-2: A Systematic Review and Meta-Analysis. Cells, 12(3). https://doi.org/10.3390/cells12030430

von Bernhardi, R., Bernhardi, L. E., & Eugenin, J. (2017). What Is Neural Plasticity? Adv Exp Med Biol, 1015, 1-15. https://doi.org/10.1007/978-3-319-62817-2_1

Wang, C. C., Prather, K. A., Sznitman, J., Jimenez, J. L., Lakdawala, S. S., Tufekci, Z., & Marr, L. C. (2021). Airborne transmission of respiratory viruses. Science, 373(6558). https://doi.org/10.1126/science.abd9149

Wang, J., Eslinger, P. J., Doty, R. L., Zimmerman, E. K., Grunfeld, R., Sun, X., Meadowcroft, M. D., Connor, J. R., Price, J. L., Smith, M. B., & Yang, Q. X. (2010). Olfactory deficit detected by fMRI in early Alzheimer's disease. Brain Res, 1357, 184-194. https://doi.org/10.1016/j.brainres.2010.08.018

Wang, S., Liu, X., & Wang, Y. (2022). Evaluation of Platelet-Rich Plasma Therapy for Peripheral Nerve Regeneration: A Critical Review of Literature. Front Bioeng Biotechnol, 10, 808248. https://doi.org/10.3389/fbioe.2022.808248

Ward, R. J., Ashraf, M., Wuerger, S., & Marshall, A. (2023). Odors modulate color appearance. Front Psychol, 14, 1175703. https://doi.org/10.3389/fpsyg.2023.1175703

Watt, W. C., Sakano, H., Lee, Z. Y., Reusch, J. E., Trinh, K., & Storm, D. R. (2004). Odorant stimulation enhances survival of olfactory sensory neurons via MAPK and CREB. Neuron, 41(6), 955-967. https://doi.org/10.1016/s0896-6273(04)00075-3

Whitcroft, K. L., Altundag, A., Balungwe, P., Boscolo-Rizzo, P., Douglas, R., Enecilla, M. L. B., Fjaeldstad, A. W., Fornazieri, M. A., Frasnelli, J., Gane, S., Gudziol, H., Gupta, N., Haehner, A.,

Hernandez, A. K., Holbrook, E. H., Hopkins, C., Hsieh, J. W., Huart, C., Husain, S., . . . Hummel, T. (2023). Position paper on olfactory dysfunction: 2023. Rhinology, 61(33), 1-108. https://doi.org/10.4193/Rhin22.483

Whitcroft, K. L., Gunder, N., Cuevas, M., Andrews, P., Menzel, S., Haehner, A., & Hummel, T. (2021). Intranasal sodium citrate in quantitative and qualitative olfactory dysfunction: results from a prospective, controlled trial of prolonged use in 60 patients. Eur Arch Otorhinolaryngol, 278(8), 2891-2897. https://doi.org/10.1007/s00405-020-06567-7

White, T. L., & Prescott, J. (2007). Chemosensory cross-modal stroop effects: congruent odors facilitate taste identification. Chem Senses, 32(4), 337-341. https://doi.org/10.1093/chemse/bjm001

Wilson, D. A., Best, A. R., & Sullivan, R. M. (2004). Plasticity in the olfactory system: lessons for the neurobiology of memory. Neuroscientist, 10(6), 513-524. https://doi.org/10.1177/1073858404267048

Winter, A. L., Henecke, S., Lundstrom, J. N., & Thunell, E. (2023). Impairment of quality of life due to COVID-19-induced long-term olfactory dysfunction. Front Psychol, 14, 1165911. https://doi.org/10.3389/fpsyg.2023.1165911

Woo, C. C., Miranda, B., Sathishkumar, M., Dehkordi-Vakil, F., Yassa, M. A., & Leon, M. (2023). Overnight olfactory enrichment using an odorant diffuser improves memory and modifies the uncinate fasciculus in older adults. Front Neurosci, 17, 1200448. https://doi.org/10.3389/fnins.2023.1200448

World Health Organization. (2024). WHO COVID-19 dashboard. Retrieved June 29, 2025 from https://data.who.int/dashboards/covid19/cases?n=c

Wu, D., Wang, V. Y., Chen, Y. H., Ku, C. H., & Wang, P. C. (2022). The prevalence of olfactory and gustatory dysfunction in covid-19 - A systematic review. Auris Nasus Larynx, 49(2), 165-175. https://doi.org/10.1016/j.anl.2021.07.007

Wu, J., Fu, Q., & Rose, M. (2020). Stimulus modality influences the acquisition and use of the rule-based strategy and the similarity-based strategy in category learning. Neurobiol Learn Mem, 168, 107152. https://doi.org/10.1016/j.nlm.2019.107152

Wudarczyk, O. A., Kohn, N., Bergs, R., Goerlich, K. S., Gur, R. E., Turetsky, B., Schneider, F., & Habel, U. (2016). Chemosensory anxiety cues enhance the perception of fearful faces - An fMRI study. Neuroimage, 143, 214-222. https://doi.org/10.1016/j.neuroimage.2016.09.002

Yamagishi, M., Fujiwara, M., & Nakamura, H. (1994). Olfactory mucosal findings and clinical course in patients with olfactory disorders following upper respiratory viral infection. Rhinology, 32(3), 113-118. https://www.ncbi.nlm.nih.gov/pubmed/7530857

Yan, C. H., Jang, S. S., Lin, H. C., Ma, Y., Khanwalkar, A. R., Thai, A., & Patel, Z. M. (2023). Use of platelet-rich plasma for COVID-19-related olfactory loss: a randomized controlled trial. Int Forum Allergy Rhinol, 13(6), 989-997. https://doi.org/10.1002/alr.23116

Yang, J., & Pinto, J. M. (2016). The Epidemiology of Olfactory Disorders. Curr Otorhinolaryngol Rep, 4(2), 130-141. https://doi.org/10.1007/s40136-016-0120-6

Yao, L., Yi, X., Pinto, J. M., Yuan, X., Guo, Y., Liu, Y., & Wei, Y. (2018). Olfactory cortex and Olfactory bulb volume alterations in patients with post-infectious Olfactory loss. Brain Imaging Behav, 12(5), 1355-1362. https://doi.org/10.1007/s11682-017-9807-7

Yaylaci, A., Azak, E., Onal, A., Akturk, D. R., & Karadenizli, A. (2023). Effects of classical olfactory training in patients with COVID-19-related persistent loss of smell. Eur Arch Otorhinolaryngol, 280(2), 757-763. https://doi.org/10.1007/s00405-022-07570-w

Yeomans, M. R. (2006). Olfactory influences on appetite and satiety in humans. Physiol Behav, 87(4), 800-804. https://doi.org/10.1016/j.physbeh.2006.01.029

Youngentob, S. L., & Kent, P. F. (1995). Enhancement of odorant-induced mucosal activity patterns in rats trained on an odorant identification task. Brain Res, 670(1), 82-88. https://doi.org/10.1016/0006-8993(94)01275-m

Yousem, D. M., Geckle, R. J., Bilker, W., McKeown, D. A., & Doty, R. L. (1996). MR evaluation of patients with congenital hyposmia or anosmia. AJR Am J Roentgenol, 166(2), 439-443. https://doi.org/10.2214/ajr.166.2.8553963

Yousem, D. M., Geckle, R. J., Bilker, W. B., Kroger, H., & Doty, R. L. (1999). Posttraumatic smell loss: relationship of psychophysical tests and volumes of the olfactory bulbs and tracts and the temporal lobes. Acad Radiol, 6(5), 264-272. https://doi.org/10.1016/s1076-6332(99)80449-8

Yuan, X., Xu, Q., Du, F., Gao, X., Guo, J., Zhang, J., Wu, Y., Zhou, Z., Yu, Y., & Zhang, Y. (2025). Development and validation of a model to predict cognitive impairment in traumatic brain injury patients: a prospective observational study. EClinicalMedicine, 80, 103023. https://doi.org/10.1016/j.eclinm.2024.103023

Yuce, M., Filiztekin, E., & Ozkaya, K. G. (2021). COVID-19 diagnosis -A review of current methods. Biosens Bioelectron, 172, 112752. https://doi.org/10.1016/j.bios.2020.112752

Zatorre, R. J., Fields, R. D., & Johansen-Berg, H. (2012). Plasticity in gray and white: neuroimaging changes in brain structure during learning. Nature Neuroscience, 15(4), 528-536. https://doi.org/10.1038/nn.3045

Zazhytska, M., Kodra, A., Hoagland, D. A., Frere, J., Fullard, J. F., Shayya, H., McArthur, N. G., Moeller, R., Uhl, S., Omer, A. D., Gottesman, M. E., Firestein, S., Gong, Q., Canoll, P. D., Goldman, J. E., Roussos, P., tenOever, B. R., Jonathan, B. O., & Lomvardas, S. (2022). Non-cell-autonomous disruption of nuclear architecture as a potential cause of COVID-19-induced anosmia. Cell, 185(6), 1052-1064 e1012. https://doi.org/10.1016/j.cell.2022.01.024

Zellner, D. A., Bartoli, A. M., & Eckard, R. (1991). Influence of color on odor identification and liking ratings. Am J Psychol, 104(4), 547-561. https://www.ncbi.nlm.nih.gov/pubmed/1793126

Zellner, D. A., & Kautz, M. A. (1990). Color affects perceived odor intensity. J Exp Psychol Hum Percept Perform, 16(2), 391-397. https://doi.org/10.1037//0096-1523.16.2.391

Zheng, Y., You, Y., Farias, A. R., Simon, J., Semin, G. R., Smeets, M. A., & Li, W. (2018). Human chemosignals of disgust facilitate food judgment. Sci Rep, 8(1), 17006. https://doi.org/10.1038/s41598-018-35132-w

Zoon, H. F., de Graaf, C., & Boesveldt, S. (2016). Food Odours Direct Specific Appetite. Foods, 5(1). https://doi.org/10.3390/foods5010012

Annexes

Below are two other scientific papers published that are not part of this thesis. The first manuscript is about the plasticity and olfactory abilities of Sommelier students before and after their training. The second manuscript is a book chapter that includes various methods used in trigeminal testing.

Article - Olfactory bulb volume and cortical thickness evolve during sommelier training

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Ethical statement: This study was approved by the Ethics Committee of the Institut Universitaire de Gériatrie de Montréal (IUGM) research center, the Ethics Committee of the Institut de Tourisme et d'Hôtellerie du Québec in Montreal, and the Ethics Committee of the University of Quebec in Trois-Rivières, Canada. All participants gave informed written consent to participate.

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Abstract

Brain plasticity is essential for experts to acquire the abilities they need. Sommeliers are olfaction experts who display differences in olfactory regions in the brain that correlate with greater olfactory abilities. While most studies on this topic are cross-sectional, we used a longitudinal design and invited 17 sommelier students at the start and end of their training then to compare them to 17 control students to study the effects of training-related brain plasticity. After a year and a half, 5 Sommelier students and 4 control students dropped out, leading to 12 Sommelier students vs 13 controls. More specifically, we used magnetic resonance imaging (MRI) to measure cortical thickness. We also measured olfactory bulb volume, as this structure plays a crucial role in olfactory processing and its volume positively correlates to olfactory performance which has never been examined in sommeliers. We used Sniffin' Sticks test to evaluate olfactory performance. During training, olfactory bulb volume increased in sommelier students while there was no significant change in the control group. We also observed that thickness of right entorhinal cortex increased, and cortical thickness decreased in other regions of interests. Our olfactory tests did not

reveal any significant changes in sommelier students. In conclusion, this is the first longitudinal study to report an increase in olfactory bulb volume in olfaction experts and our aim was to show the effects of ecological training-related brain plasticity. The mixed results about cortical thickness might be explained by "overproduction-pruning" model of brain plasticity, according to which the effects of training-related plasticity are non-linear and simultaneously involve different processes.

Keywords: plasticity, brain, olfaction, sommelier, olfactory bulb, cortical thickness, magnetic resonance imaging

Introduction

Brain plasticity allows experts to acquire the skills they need: through training, they acquire and refine abilities that come with changes in brain structure and function. Effects of training-related brain plasticity can be observed in top athletes, musicians, or in professionals whose jobs require specific skills. For example, structural differences can be observed in hippocampus of London taxi drivers because the hippocampus is involved in spatial memory. With years of experience, taxi drivers memorize the London map better thus need less encoding of new spatial information so they start using their mental map of the city (Maguire *et al.*, 2000). Likewise, brain changes facilitate visuo-spatial processing and coordination in professional badminton players (Di *et al.*, 2012), grant musicians refined hand motor skills (Amunts *et al.*, 1997), allow radiologists to process and interpret radiographs more effectively (Harley *et al.*, 2009). These training-related changes in the brain can be structural or functional; they can occur in both cortex and white matter. We can detect this type of changes with neuroimaging techniques such as magnetic resonance imaging (MRI). It is important to know whether these changes can also be detected as a result of ecological training which will give us insight about olfactory plasticity. This has the potential to improve the olfactory training protocols for individuals with the loss of olfactory function.

The olfactory bulb (OB) is an ovoid structure located under the frontal lobe of brain. It constitutes the first relay of olfactory processing as it receives input directly from the olfactory epithelium: olfactory receptor neurons project to the ipsilateral OB and synapse with second-order neurons

known as mitral and tufted cells that will convey the information deeper into brain (Gottfried, 2010; Huart *et al.*, 2013; K. Mori *et al.*, 1999).

By using MRI, the volume of this structure can be measured. In healthy people with a normal sense of smell, significant positive correlations between OB volumes and olfactory performance were reported: a bigger OB is associated with better olfactory abilities (Buschhuter *et al.*, 2008; Seubert *et al.*, 2013). Similar correlations were observed in patients with olfactory dysfunction: impairment of olfactory dysfunction is positively correlated with smaller OB volume (Liu *et al.*, 2017; Rombaux *et al.*, 2006). Cause of olfactory dysfunction can be congenital in some rare cases, but most of the time the impairment is acquired. Patients with a unilateral complete nasal obstruction are subject to a decrease of ipsilateral OB volume. For example, absence of olfactory input on one side during a few months resulted in a smaller ipsilateral OB, while contralateral OB was not affected (Askar *et al.*, 2015). OB volume changes are not only caused by olfactory dysfunction: age, for example, is another factor that can impact OB volume. Similar to olfactory performance, OB volume decreases with age (Hang *et al.*, 2015).

Olfactory training typically consists of smelling odours every day for a few months, and it is a potential way for patients to recover from olfactory dysfunction/loss. It can improve olfactory performance, and these changes correlate with OB volume increases (Haehner *et al.*, 2008; Rombaux *et al.*, 2009). Olfactory training can have the same effect in people with a normal sense of smell. In one experiment, participants were tested at the beginning and end of a four-month olfactory training but only one of their nostrils was trained. OB on both the trained and contralateral side nostrils became bigger. This confirms the correlation between olfactory function and OB volume. It also indicates that the underlying mechanism is complex and involves top-down processes which allows OB that is not on the stimulated side to grow along with a stimulated OB (Negoias *et al.*, 2017). Besides receiving input from olfactory epithelium, OB neuronal activity is modulated by centrifugal input from cerebral structures such as primary olfactory cortex, amygdala, hippocampus, locus coeruleus and raphe nuclei (Lazarini & Lledo, 2011). Changes in OB volume could be due to neurogenesis that happens in adult olfactory system or synaptogenesis between olfactory receptor neurons and mitral cells in OB (Curtis *et al.*, 2007; Eavri & Nedivi, 2013; Lotsch *et al.*, 2014) (for a review, see (Huart *et al.*, 2013, 2019; Zatorre *et al.*, 2012)).

Measuring cortical thickness is an efficient way to evaluate changes in grey matter. Numerous studies reported that training and expertise in different domains can impact cortical thickness. Auditory cortex as well as frontal regions involved in high cognitive function were reported to be thicker in musicians (Bermudez *et al.*, 2009). In another study, a nine-month training of social skills resulted in cortical thickness changes in well-known socio-affective and socio-cognitive brain networks (Valk *et al.*, 2017). Similarly, the ability to perceive and identify odors is also related to brain anatomy as: olfactory performance and cortical thickness in both olfactory and non-olfactory regions are correlated (Frasnelli *et al.*, 2010). Another study targeting sommeliers reported that they display a thicker entorhinal cortex which plays a key role in olfactory processing (Banks *et al.*, 2016). Being an expert is not needed to observe the effects of training-related brain plasticity: training novices for six weeks in different olfactory tasks and testing them before and after the training showed that cortical thickness increased in regions such as right entorhinal cortex, right inferior frontal gyrus and bilateral fusiform gyrus (Al Ain *et al.*, 2019).

Objectives and hypotheses

Many studies about brain plasticity in sommeliers and perfumers are cross-sectional, which is an efficient way to compare them with a control group, but it does not allow to see the evolution of brain over time. In olfactory field, only in a few studies, researchers used a longitudinal design to examine the effects of training-related brain plasticity with MR imaging. The most popular study doing this consisted of testing adults with a normal sense of smell before and after a six-week-long olfactory training (Al Ain *et al.*, 2019). Though the OB has been the subject of many studies, there has been no report of OB volume in olfactory specialists.

We therefore aimed at examining the effects of sommelier training on the brain and olfactory function in an explorative study. We tested sommelier students at the start and end of their 18 months training and compared them with a group of students whose training did not involve any sense of smell. We used Magnetic Resonance Imaging (MRI) to measure OB volume and cortical thickness. We hypothesized that (1) Sommelier training leads to OB volume increases; (2) Sommelier training leads to changes of cortical thickness.

Materials and methods

Participants

Initially 17 Sommelier students and 17 control students were recruited in the study. Sommelier students were recruited from the Institut de Tourisme et d'Hôtellerie du Québec. Participants did not have any known smell or taste loss/dysfunction. At time point-1 Sommelier group composed of 7 women aged 26.1 ± 4.7 years, and 10 men (aged Mean = 25.7, S.D. = 5.0 years, range: 21-34). The control group consisted of 17 students from the University of Montreal or the University of Quebec in Montreal. These participants were chosen to match sommelier students in age and gender. Therefore they were composed of 7 women aged 26.6 ± 4.3 years, and 10 men (aged Mean = 25.6, S.D. = 5.7 years, range: 20-35). One sommelier student was excluded from the study because of pregnancy, defined by the Ethics Committee as an exclusion criterion for the MRI scan. At follow up, 5 Sommelier students and 4 control participants dropped out of the study resulting in 12 Sommelier students and 13 control participants. Therefore at follow up, sommelier group consisted of 5 women and 7 men (aged Mean = 26,6, S.D. = 4,3 years, range: 21-34) and control group consisted of 4 women and 8 men (aged, Mean = 27,2, S.D. = 4,8 years, range: 20-35).

	Sommeliers	Controls	Statistic	p-value
Baseline				
Sex (f/m)	7/10	7/10	χ^2	1
Age	26.0	26.2	t test	0.9
Follow up				
Sex (f/m)	5/7	4/8	χ^2	0.67
Age	26.6	27.3	t test	0.7

Table 5.1. Average ages across Sommelier and control groups at baseline and follow-up. *Mean ages were compared with independent samples t-test. P-values of the tests were added.*

Sommelier training

The participants in the sommelier group underwent the International Service and Sommelier Training at The Institut de Tourisme et d'Hôtellerie du Québec in Montreal (https://www.ithq.qc.ca/en/school/future-students/programs/program/international-service-and-sommelier-training/), which is the prerequisite for becoming a professional sommelier. The training consists of 1200 hours of classes; olfactory training takes place in most of these classes as only 45 hours do not involve any sensory analysis. There is also a minimum of 905 hours work experience obtained during different compulsory internships that include 4 months in an English-speaking establishment outside of Quebec, 3 months at a Michelin-starred or Relais & Châteaux restaurant in France, and a month at a vineyard in France.

Students in the control group came from different fields of study, e.g. administration, psychology, life sciences, economics, humanities, which did not involve any practical olfactory training. We did not monitor whether students were involved in any olfactory-training like activities during the period of training.

Brain Imaging

MRI images were acquired at the Unité de Neuroimagerie Fonctionnelle (UNF) at IUGM. UNF provides access to a Prisma Fit 3 Tesla MRI scanner from Siemens.

Olfactory bulb volume

To measure OB volume, we used a standard protocol resulting in 2-mm-thick T2-weighted images in Turbo Spin Echo (TSE) mode. Images were obtained in the coronal plane and there was no gap between the 2-mm-thick slices, with voxel size: 0.16 x 0.16 x 2 mm^3. This method was described as the most suitable method for OB volumetry (Huart *et al.*, 2013; Seubert *et al.*, 2013). OB volumes were measured for right and left OB by DP without blinding the process. To maintain objectivity, measurements were done twice and made sure that the difference between the two volumes were less than 10%.

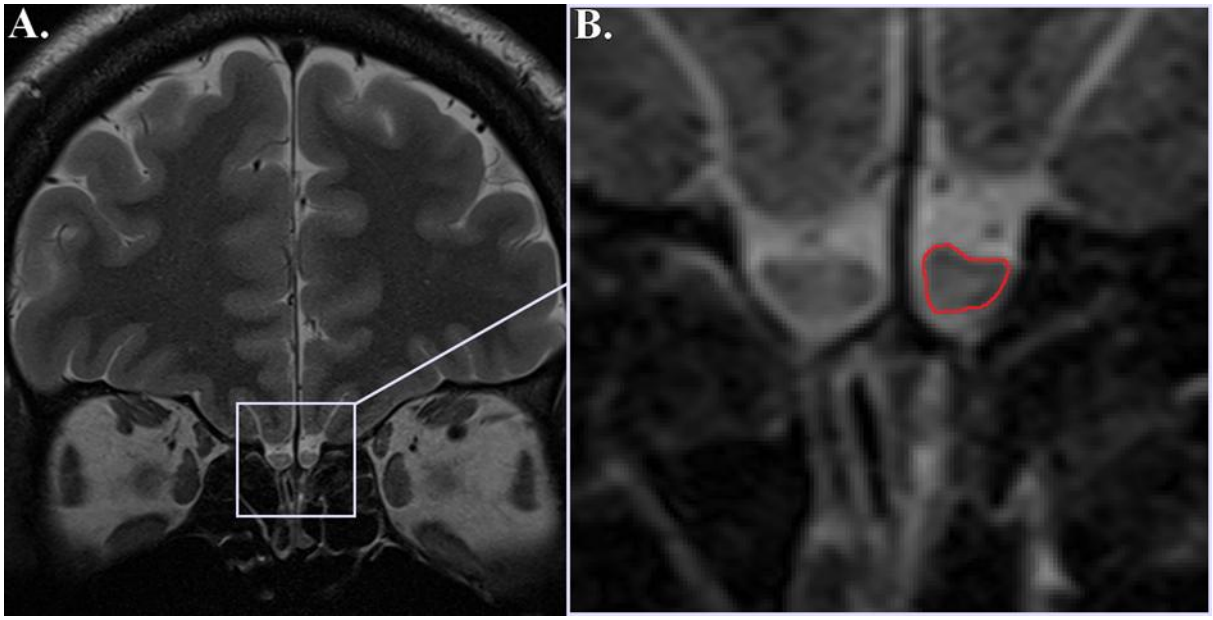


Figure 5.1. Coronal slice of the brain. *It is as seen on MIPAV software, A. zoomed out, B. zoomed in on the olfactory bulbs. The red line delineates the left olfactory bulb.*

T1-weighted MRI

To obtain whole brain volume and measure cortical thickness, we acquired a T1-weighted structural volume using an MPRAGE sequence. This sequence provides 176 contiguous sagittal slices with an isotropic spatial resolution of 1 mm^3 (repetition time 2300 ms, echo time 2.26 ms, flip angle 8°, in-plane field of view 256 mm). An automated reconstitution of a tridimensional brain image was performed using Freesurfer 6.0 for Linux (http://surfer.nmr.mgh.harvard.edu), which provided us with the whole brain volume.

Olfactory performance

Olfactory performance was assessed using an extended version of the Sniffin' Sticks test (Hummel *et al.*, 1997; Hummel *et al.*, 2007). Sniffin' Sticks are felt-tip pens which are filled with odorants instead of ink. The experimenter presents the odorants to the participant by removing the cap and placing the pen's tip approximately 2 cm in front of both nostrils.

We assessed olfactory performance by using the Sniffin'Sticks test measure (1) odor threshold, (2) odor discrimination, and (3) odor identification. We further used the same test to carry out an

odor memory test, as described earlier (Al Ain et.al., 2019). In short, participants were presented with a total of 16 odors; 8 of this were novel, while 8 others had been presented during the identification test. Participants had to indicated whether they had already smelled these odors; we counted the number of correct responses. Using the Sniffin' Sticks sets provided for each of these tests and following the established procedures (for details, see Frasnelli et al., 2010; Hummel et al., 1997; Hummel et al., 2007). In most studies, the Sniffin' Sticks test is performed birhinally, i.e., both nostrils are tested simultaneously, but it has been shown that olfactory performance assessment is more complete when it is performed unirhinally, i.e., when nostrils are tested separately, especially for the threshold task (Poupon *et al.*, 2017). Therefore, we tested nostrils separately for the odor threshold task, which is the most perceptual task, i.e., this task does not involve as many cognitive factors as the other tasks (Hedner *et al.*, 2010). To do this, participant closed a given nostril with a finger during each odor presentation, and one nostril was tested after the other. The order of nostrils was randomized. We then obtained a threshold estimate for each nostril.

Olfactory detection threshold

We assessed odor thresholds for phenylethyl alcohol (PEA) using a single staircase, three-alternative forced choice procedure: we presented participants with triplets of pens, one of them containing the odorant in a given concentration, the two other containing solvent. Participants had to identify the pen containing the odorant. In this task, we tested two nostrils separately: the participant closed a given nostril with a finger during each odor presentation, and one nostril was tested after the other. The order of the nostrils was randomized (Hummel *et al.*, 1997). Obtained scores ranged between 1 and 16 for each nostril.

Odor discrimination

To assess odor discrimination, 32 triplets of pens (two pens containing the same odorant, and a third pen containing a different one) were presented. Participants had to identify the target pen, i.e. the pen containing the different odorant. The 32-triplets discrimination test is an extended version of the commercially available 16-triplets test (Frasnelli *et al.*, 2010; Haehner *et al.*, 2009). Odors are listed in Hummel et al., 1997. Obtained scores ranged from 0 to 32.

Odor identification

Odor identification was assessed for 16 common odors. Two sets of Sniffin' Sticks are available for this task. The first one that we used at the beginning of training is composed of following odors: orange, leather, cinnamon, peppermint, banana, lemon, licorice, turpentine, garlic, coffee, apple, cloves, pineapple, rose, anise, fish. The second one that we used at the end of training is composed of 16 other common odors: pear, coke, lilac, grapefruit, grass, raspberry, honey, ginger, coconut, lavender, melon, peach, mushrooms, smoked meat, chocolate, onion (Haehner *et al.*, 2009). Notes corresponding to all of these odors can be perceived in wine.

Each odorant was presented a first time and participants had to identify it without any cue (free identification). The second time, for each individual odor, a list of four descriptors were presented; participants had to identify the odorant by picking one of them (cued identification). Lists of descriptors for each odorant have been established by the creators of Sniffin' Sticks test. Therefore, they were same for all participants. In total, we obtained two scores: free and cued identification, each ranging from 0 to 16 and corresponding to number of odors that were correctly identified. In the free identification task, participants scored only if the identification was fully correct, e.g. naming lemon or leather for orange would both count as zero.

Olfactory memory

We assessed olfactory memory by using two sets of 16 pens designed for the identification task. Only 8 pens from each set were used for this task; half of the participants were tested with pens labelled with even numbers, the other half with odd numbers. The order of pens was randomized. Participants had to tell whether they had smelled the odorant during the identification task. This task took about 40 minutes.

The score for this task consisted in the sensitivity index d' that we calculated using the signal detection theory (MacMillan & Creelman, 2005): we determined the numbers of hits (i.e. the participant said an odor was present in the identification task and that odor was present) and false alarms (i.e. the participant said an odor was present in the identification task but it was not). From that, we calculated sensitivity index d': d'= z (hit rate)-z (false alarm rate)

The sensitivity index d' indicates the ability to detect whether odors were present in the identification task: d'=1 roughly corresponds to 69% of correct answers (hits and correct rejections), d'=2 roughly corresponds to 95% of correct answers.

We obtained a total of six scores per participant in the olfactory tasks: two scores in the threshold task (right and left nostrils), one score in the discrimination task, two scores in the identification tasks (free and cued), one score in the olfactory memory task.

Analysis

The analysis of cortical thickness was performed with FreeSurfer 6.0 for Linux (http://surfer.nmr.mgh.harvard.edu).

OB volume and olfactory data were analyzed using SPSS 23.0 for Windows. Alpha was set at 0.05 and we used Bonferroni-Holm corrections for multiple comparisons.

OB volume

We used the MIPAV (Medical Image Processing, Analysis, and Visualization) to measure OB volume by manually contouring the OB surface on each coronal slice, from anterior to posterior, with pixel size 0.16 x 0.16 mm^2. The first slice (most anterior one) we consider is the one on which OB becomes visible. A sudden decrease in the diameter of OB marks the posterior end of the structure and it allows to identify the last slice to be used in the measurement. Once OB surfaces are delineated on each slice, all surfaces are added up, and multiplied by the slice thickness (2 mm) to obtain the OB volume in mm^3. This approach is commonly used in studies examining OB volumes, and it was proven to be a reliable and an accurate method (Huart *et al.*, 2013; Seubert *et al.*, 2013; Yousem *et al.*, 1998).

For the analysis, OB volume was our dependent variable. We performed repeated measures of ANOVA with two within-subject factors: *time* (2 levels: beginning of training "T1", and end of training after 18 months "T2"), and *side* (2 levels: left and right). *Group* (2 levels: sommelier students and control participants) was defined as between-subject factor. We also used *whole brain volume* as covariate.

We performed post-hoc repeated measures of ANOVAs in sommeliers and controls separately, with *time* and *side* as within-subject factors so that we could investigate if there was a group-specific evolution of OB volume between T1 and T2.

Cortical thickness

Measuring cortical thickness consists of reconstituting a tridimensional image of the brain; measuring the distance between these two surfaces, modelling white surface (at the limit between white and grey matter) and pial surface (between grey matter and cerebrospinal fluid).

The automated reconstitution of a tridimensional image of brain performed by FreeSurfer involves skull stripping, volumetric labeling, intensity normalization, white matter segmentation, surface extraction and gyral labeling. For each hemisphere, each surface is made of about 140,000 vertices that are defined by X, Y and Z coordinates. Vertices of white and pial surfaces have the same identity: each vertex of white surface has a corresponding vertex in the pial surface, which allows us to calculate the distance between two surfaces (cortical thickness).

Because we have longitudinal data, we used FreeSurfer's longitudinal stream which consists of three pre-processing steps. The first step is a cross-sectional processing corresponding to the reconstitution of a tridimensional image of brain as described in the previous paragraph and this is performed independently for each time point. The output is used in the second step to create a within-subject template corresponding to the average anatomy of the participant across time. The third step uses the within-subject template to create, final results for each time point that are more accurate and reliable than the independent cross-sectional runs. Once this preprocessing was done for each participant, we computed the longitudinal data from cortical thickness measures at T1 (first time point, at the beginning of training) and T2 (second time point, at the end of training). These longitudinal data included:

- the average thickness across time: (thickness T1 + thickness T2) / 2
- the rate of change in mm/year (rate): (thickness T2 – thickness T1) / (T2 – T1)
- the symmetrized percent change (SPC): 100 * rate / avg

Additional postprocessing steps included smoothing, using a 5 FWHM kernel and resampling onto FreeSurfer average subject FSaverage.

Finally, a group analysis was performed using a general linear model (GLM) with *SPC* as our dependent variable and *group* as our between-subject factor. A correction for multiple comparisons can be done by Monte Carlo cluster-wise simulation. Results were thresholded at $p < 0.05$ when corrected for multiple comparisons, or at $p < 0.0001$ for predicted regions. FreeSurfer stores significance as -log10(p-value); a significance of 4 and more corresponds to $p < 0.0001$ uncorrected.

See https://surfer.nmr.mgh.harvard.edu/fswiki/FsTutorial/LongitudinalTutorial for more details.

Olfactory performance

We performed two analyses. Since we measured left and right OB volumes and tested nostrils separately in the threshold task, first we analyzed olfactory threshold on each side. We included the other olfactory tasks in a second analysis.

Olfactory threshold

Olfactory threshold was our dependent variable. We performed repeated measures ANOVA with two within-subject factors: *time* (2 levels: beginning of training "T1", and end of training 18 months later "T2"), and *side* (2 levels: left and right). *Group* (2 levels: sommelier students and control participants) was defined as between-subject factor.

Then, we performed post-hoc repeated measures ANOVAs in sommeliers and controls separately. We used time and *side* as within-subject factors to investigate if there was a group-specific evolution of olfactory threshold between T1 and T2.

Overall olfactory performance

This analysis included more olfactory scores. Because there were strong correlations between left and right thresholds; free and cued identification scores, we only kept the better threshold for this analysis which reflects the score obtained when both nostrils are tested simultaneously (Frasnelli *et al.*, 2002). The score obtained in the cued identification task is most commonly used in studies with the Sniffin' Sticks. We performed a repeated measures ANOVA with two within-

subject factors: *time* (2 levels: beginning of training "T1", and end of training 18 months later "T2"), and *test* (4 levels: better threshold, discrimination, cued identification, olfactory memory). *Group* (2 levels: sommelier students and control participants) was defined as between-subject factor.

Correlations between brain and olfaction

We calculated evolutions ΔT2-T1 for each dependent variable and performed Pearson correlations to examine if there was any correlation between left and right olfactory thresholds and left and right OB volumes, between other olfactory tasks and total OB volume (left + right OB volumes), and between olfactory scores and cortical thickness.

Results

Olfactory bulb volume

Figure 1 depicts a coronal slice of a brain that is zoomed on the OB as seen on the MIPAV software. OB volumes are represented on Figure 2.

We found a significant interaction *time*group* ($F_{1,22}$ = 16.246, p = 0.001) and a significant effect of *whole brain volume* ($F_{1,22}$ = 16.842, p <0.001) on OB volumes. There was no significant main effect of *time* ($F_{1,22}$ = 2.339, p = 0.140), *group* ($F_{1,22}$ = 1.015, p = 0.325), *side* ($F_{1,22}$ = 0.007, p = 0.934) or any interaction.

To disentangle the interaction, we compared OB volumes at T1 and T2 in each group separately. In the sommeliers group, we observed a significant main effect of *time* with OB volume being bigger at T2 than T1 ($F_{1,11}$ = 12.028, p = 0.005). In contrast, there was no significant effect of *time* in controls ($F_{1,12}$ = 0.474, p = 0.504); the evolution of OB volume over time was not significant in control group (Figure 2).

We then investigated the effect of *whole brain volume* which is correlated with OB volume (left OB at T1: r = 0.456, p = 0.016; right OB at T1: r = 0.371, p = 0.033; left OB at T2: r = 0.578, p = 0.006; right OB at T2: r = 0.622, p = 0.004). There was no effect of group on the whole brain volume ($F_{1,33}$ = 0.939, p = 0.340).

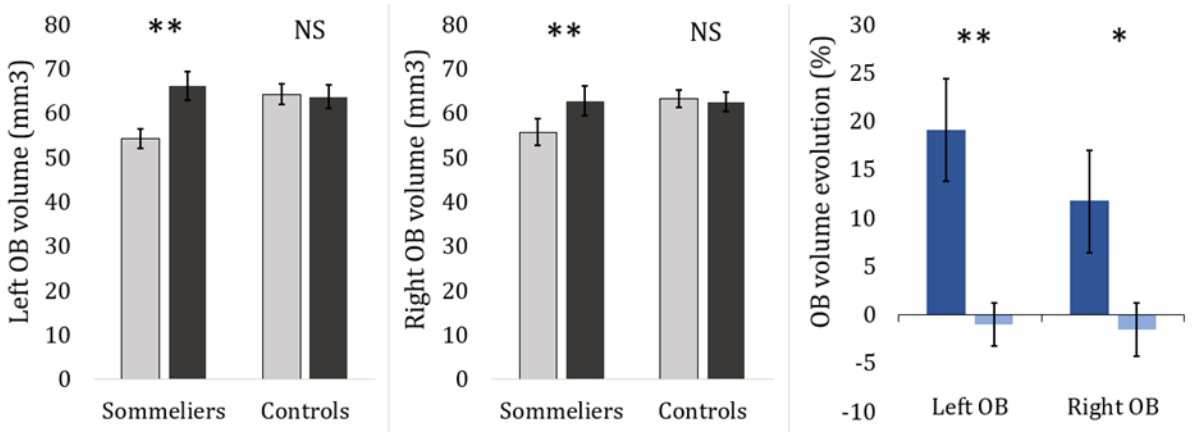


Figure 5.2. OB volumes T1 and T2. *A. and B. Left and right olfactory bulb volume (in mm3) at the start of training (T1) and at the end of training (T2) in sommelier students (dark) and controls (light). C. Evolution of OB volumes during training (in %) in sommelier students (dark) and controls (light). Standard error of the means can be observed in the graph.*

Cortical thickness

When we applied a correction for multiple comparisons, we found no significant cluster. When we lowered the threshold to $p < 0.0001$ uncorrected, we observed that sommelier training had an effect on cortical thickness in several small clusters: in sommelier students, there was an increase of cortical thickness in right entorhinal cortex, and a decrease of cortical thickness in left inferior temporal gyrus, triangular portion of right inferior frontal gyrus (pars triangularis), left superior parietal and superior frontal gyri (for more details, see Table 1 and Figure 3).

Region	Coordinates			Size	Sig.
	x	y	z		
L inferior temporal gyrus	-52.7	-61.9	-3.9	28	-5.71
R entorhinal cortex	28.8	-7.9	-32.8	16	5.38
R pars triangularis	52.8	28	3.3	3	-4.23
L superior parietal gyrus	-29.7	-52.6	51.7	8	-4.23
L superior frontal gyrus	-21.2	23.9	47.5	2	-4.12

Table 5.2. Effects of sommelier training on cortical thickness*. Effect of group on the following structures was significant at a $p < 0.0001$ uncorrected level. Coordinates (x, y, z) are in the MNI*

space. The size corresponds to the number of vertices where differences were observed. Sig = -log10(p-value); a significance of 4 and more corresponds to p < 0.0001, a positive significance indicates an increase of cortical thickness in sommeliers compared to controls while a negative significance indicates a decrease.

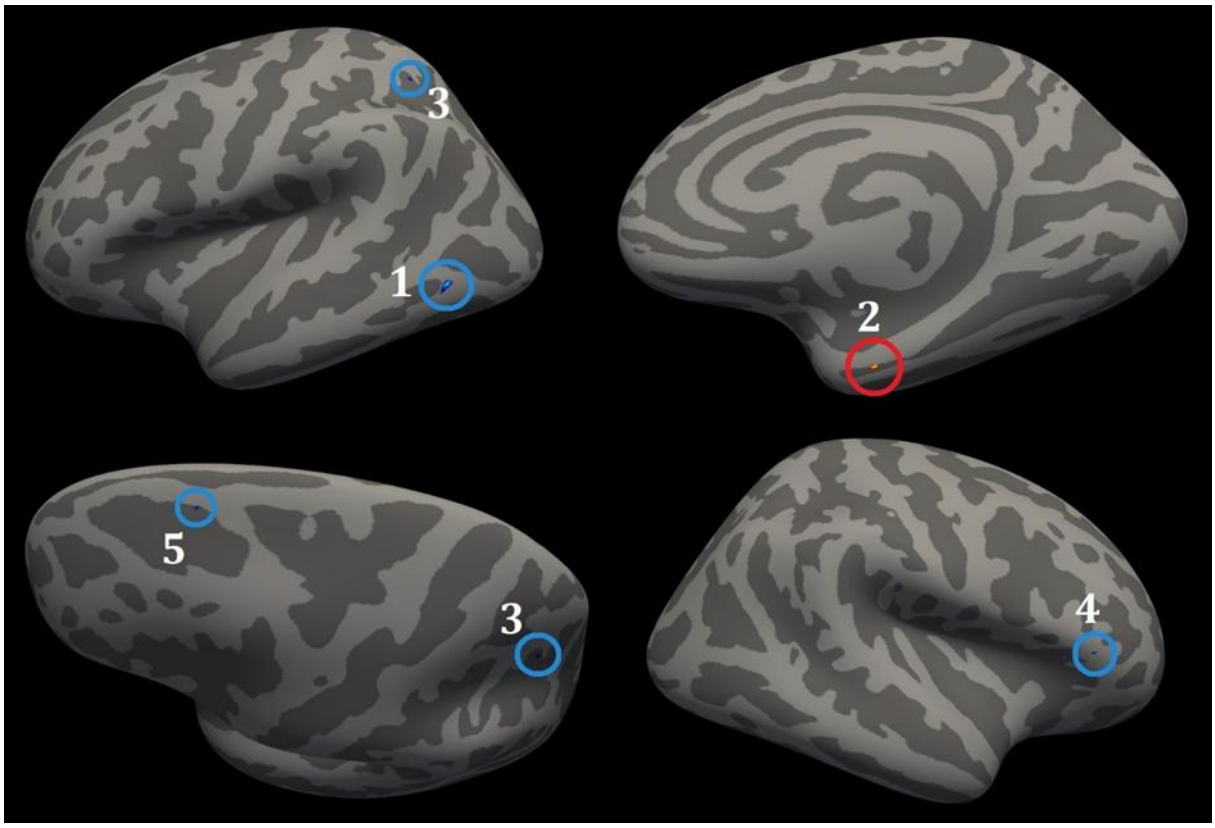


***Figure 5.3**. **Effect of sommelier training on cortical thickness.** Comparison of symmetrized percent change (SPC) over training between sommelier and control students: blue clusters indicate that cortical thickness in sommelier students decreased during training while the red and yellow cluster indicates an increase of cortical thickness. A. Lateral view of the left hemisphere. B. Inferomedial view of the right hemisphere. C. Superolateral view of the left hemisphere. D. Lateral view of the right hemisphere. p < 0.0001 uncorrected: 1. Inferior temporal gyrus, 2. Entorhinal cortex, 3. Superior parietal gyrus, 4. Pars triangularis, 5. Superior frontal gyrus.*

Olfactory performance

Olfactory scores are depicted in Figure 4.

In our first analysis, we found a significant interaction of *time*group* ($F_{1,23}$ = 8.951, p = 0.007) and a significant effect of *time* with thresholds at T2 better than T1 ($F_{1,23}$ = 7.510, p = 0.012). There was no effect of *group* ($F_{1,23}$ = 1.435, p = 0.243), *side* ($F_{1,23}$ = 0.241, p = 0.628) or any interaction.

To disentangle the interaction, we compared olfactory thresholds at T1 and T2 in each group separately. In sommeliers, there was no main effect of *time* ($F_{1,11}$ = 0.033, p = 0.860) or any other variable and interaction. In control group, there was a significant main effect of *time* with olfactory thresholds being better at T2 than T1 ($F_{1,12}$ = 29.775, p < 0.001; Figure 4).

In our second analysis, for other olfactory tasks, we found no significant effect of *group* ($F_{1,23}$ = 0.176, p = 0.678) or *time* ($F_{1,23}$ = 0.906, p = 0.351), interaction of *time*group* ($F_{1,23}$ = 3.591, p = 0.071). This means that the olfactory scores did not significantly evolve between T1 and T2. There was no overall significant difference between groups.

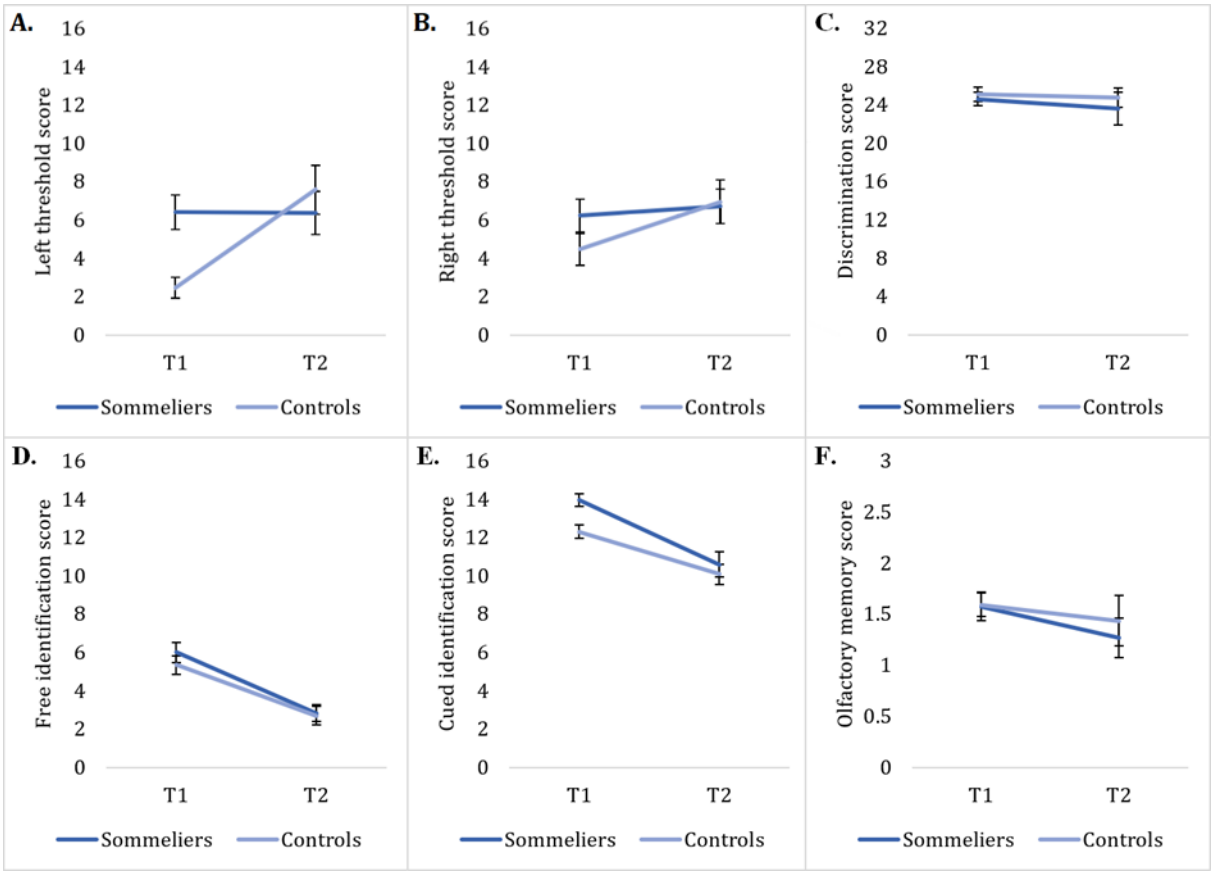


Figure 5.4. Olfactory performance at T1 and T2. *Dark color represents sommelier students and light color represents controls. Scores obtained in the detection threshold task with the left nostril (A.) and the right nostril (B.), in the discrimination task (C.), the free identification task (D.), the cued identification task (E.) and the olfactory memory task (F.).*

Correlations between OB volume and olfactory performance

There was no correlation between the OB volume and olfactory threshold at T1 and T2 (left: T1: r = -0.241, p = 0.176; T2: r = -0.111, p = 0.598; right: T1: r = -0.012, p = 0.947; T2: r = 0.084, p = 0.690).

We also found no correlation between evolutions of OB volume and olfactory threshold (left: sommeliers: r = 0.259, p = 0.416; controls: r = -0.234, p = 0.442; right: sommeliers: r = -0.315, p = 0.319; controls: r = -0.443, p = 0.129).

There was no significant correlation between OB volumes or cortical thickness and other olfactory tests.

Discussion

We found that sommelier training led to an increase of OB volume and to changes in cortical thickness in five different regions associated with olfactory processing. Also, our olfactory tests did not reveal any improvement of the olfactory function; thus, changes in the brain were not correlated with enhanced scores on the Sniffin' Sticks test.

Olfactory bulb volume

We report data about OB volume in experts measured twice during a longitudinal study that is conducted with sommelier students. This study allowed us to examine the effect of training on OB volume.

The increase of OB volume as sommelier students become olfaction experts is a great example of ecological training-related brain plasticity. This result is in line with the literature about OB volume in patients and healthy people with a normal sense of smell. Since greater OB volume is associated with a better sense of smell (Buschhuter *et al.*, 2008; Liu *et al.*, 2017; Rombaux *et al.*,

2006; Seubert *et al.*, 2013), and olfactory training results with the increase of OB volume (Haehner *et al.*, 2008; Negoias *et al.*, 2017; Rombaux *et al.*, 2009), it is plausible that becoming an expert in olfaction leads to an increase of OB volume.

We showed that training led to an increase of OB volume. Four mechanisms have been proposed to explain OB plasticity, i.e. how the OB can grow with training.

Synaptogenesis is a first possible mechanism: activity modulates the connections between neurons, i.e. the synapses between olfactory receptor neurons and mitral cells lead to an increase or decrease in the number of synapses, thus modulating the size of the structure (Eavri & Nedivi, 2013; Zatorre *et al.*, 2012).

Other mechanisms consist of an increase in the number of neurons. Neurogenesis is a remarkable ability of the olfactory system. Continuous neurogenesis occurring at the level of the olfactory epithelium is a second proposed mechanism. The olfactory epithelium contains stem cells that can differentiate into new olfactory receptor neurons (Schwob & Costanzo, 2010). This regeneration is essential to maintain a functional sense of smell since olfactory receptor neurons are directly exposed to the environment and thus can be damaged. By repeatedly activating the olfactory system, one can hypothesize that olfactory training stimulates neurogenesis in the olfactory epithelium. Olfactory receptor neurons then grow axons which synapse with mitral cells in the OB. This could explain the increase in OB volume.

A third possible mechanism relies on neurogenesis in the supraventricular zone of the lateral ventricle: neural stem cells produce neuroblasts which migrate toward the OB and differentiate into olfactory interneurons in the OB. These additional cells could explain the OB growth. However, this mechanism which was first demonstrated in adult rodents and monkeys (Kornack & Rakic, 2001; Lois *et al.*, 1996; Ming & Song, 2011) is still debated in adult humans. If neural stem cells have been observed along the lateral ventricle in humans (Johansson *et al.*, 1999; Sanai *et al.*, 2004), their ability to produce neuroblasts which migrate to the OB is still a matter of debate (Curtis *et al.*, 2007; Sanai *et al.*, 2007; Sanai *et al.*, 2011).

Finally, intrinsic bulbar plasticity constitutes a fourth possible mechanism underlying OB plasticity: neural stem cells are present in the adult human OB and it was hypothesized that they could

be responsible for an increase in the number of cells, and thus OB growth (Pagano *et al.*, 2000). However, while the study of functional genomics suggests neurogenesis in the OB (Lotsch *et al.*, 2014), no OB neurogenesis was detected (Bergmann *et al.*, 2015).

Cortical thickness

We observed an increase of cortical thickness in sommeliers' right entorhinal cortex, and a decrease in the inferior temporal gyrus, the superior parietal and the superior frontal gyrus (all left) as well as the triangular portion of right inferior frontal gyrus. It is important to point out that the group difference in cortical thickness were only significant with a liberal threshold of $p<0.0001$, uncorrected, while a Bonferroni correction yielded no significant difference. Our approach is in line with earlier research on the association between structural measures in olfactory specialists/ training (Banks et al., 2016, Al Ain et al., 2019, Delon-Martin et al., 2013), as using a conservative threshold such as Bonferroni correction comes with the risk of false negative results. We therefore primarily discuss predicted olfactory regions, i.e., regions that have shown an effect in earlier studies. This is only true for the right entorhinal cortex which increased in sommelier students. A similar increase was reported following a 6 week long olfactory training (Al Ain *et al.*, 2019). In fact, the entorhinal cortex – one of the primary olfactory regions (Patel & Pinto, 2014), – has a larger volume in sommeliers, and its cortical thickness is positively correlated with years of sommelier experience (Banks *et al.*, 2016).

Other regions with significant differences were not predicted, and conclusions are therefore speculative. We observed a decrease in cortical thickness in left inferior temporal gyrus (ITG), triangular portion of right inferior frontal gyrus (tIFG), left superior parietal gyrus (SPG) and left superior frontal gyrus (SFG) in sommelier students. These regions are not typically found to be involved in olfactory processing. We did not expect cortical thickness to decrease because greater olfactory abilities are usually associated with larger brain structures and thicker cortices, as several studies reported before (Al Ain *et al.*, 2019; Banks *et al.*, 2016; Buschhuter *et al.*, 2008; Frasnelli *et al.*, 2010; Hummel *et al.*, 2003; Seubert *et al.*, 2013). Most of these studies are cross-sectional and compare two groups of individuals instead of examining the evolution of cortical thickness like we did, but the longitudinal study (Al Ain *et al.*, 2019) also seemed to validate the idea that cortical

thickness increases with training: all the changes they observed after their six-week-long olfactory training were local increases of cortical thickness, including the two brain regions where we observed a decrease: left ITG and right tIFG.

However, in other fields than olfaction, there were reports of learning-dependent decreases of cortical thickness. For example, following a 9 months training of social skills (Valk *et al.*, 2017) or after a week-long training aiming at improving processing speed (Takeuchi *et al.*, 2011). This suggests that cortical thinning could have a role in learning. Another idea is that the progression of learning-dependent changes is nonlinear: this was the theory supported by a team who observed that over a 7 weeks long training during which right-handed participants practiced writing and drawing with left hand, cortical thickness increased in the first four weeks but then decreased despite continued practice and increasing task proficiency (Wenger *et al.*, 2017). This led to the 'overproduction - pruning' model of plasticity according to which, number of synapses increases greatly at the beginning – resulting in increased cortical thickness – and then, behaviorally-relevant connections are stabilized while connections that prove to be functionally irrelevant are eliminated by pruning (Lindenberger *et al.*, 2017) – associated with reduced cortical thickness.

This model is supported by the evidence of two-photon microscopy in mice during motor training: rapid formation of new dendritic spines was followed by a slower process of spine elimination while newly formed and retained dendritic spines were stabilized and probably function as the physiological substrate for skill acquisition and improvement (Xu *et al.*, 2009). It is in line with previous findings supporting the idea that changes in the brain appear quickly: in a study where participants were tested several times during a 5 week long juggling training, increases of cortical thickness were visible after only a week, leading the authors to suggest that learning a new task has more impact on brain structure than continued training of an already-learned task (Driemeyer *et al.*, 2008). Finally, this model would explain why we found that cortical thickness decreased in brain regions where Al Ain et al. observed an increase: in Al Ain et al.'s study, olfactory training lasted only 6 weeks and participants were tested before it started; in our study, training lasted 18 months and we tested sommelier students when their training had already started, mostly during the second month. Because the timing was different, it is possible that Al Ain et al. observed the increase of cortical thickness that happens at the beginning of training during a first phase of

overproduction, while we first tested our participants when they were (possibly) already near the end of this first phase and thus, we observed a decrease in second phase during which more synapses would be eliminated by pruning than the ones that were newly formed. Those dynamic changes over time support the idea that training-related brain plasticity has complex nonlinear effects that involves several processes.

Apart from the entorhinal cortex which is known as an olfactory processing area, the brain regions where we observed changes in cortical thickness are not typically associated with olfaction.

Olfactory performance

While we expected olfactory performance to improve in sommelier students, scores obtained by sommelier students in olfactory tasks were not significantly better at T2 than at T1. Because OB volume increased during sommelier training, but their olfactory scores did not evolve, we found no correlation between evolutions of OB volume and olfactory performance. In the control group, there was no significant differences between T1 and T2 for most tests, except in the threshold task in which we observed a surprising improvement of the performance. The improvement of olfactory abilities during an olfactory training cannot always be revealed with the Sniffin' Sticks: studies showed that smelling four odors every day for a few months led to an improvement of olfactory sensibility but this effect was specific to the odors used during training and could not be detected with the Sniffin' Sticks threshold task (Dalton *et al.*, 2002; E. Mori *et al.*, 2015). This kind of training also led to an improvement of the ability to identify the four odors but the effect was not generalized and thus undetectable with the Sniffin' Sticks identification task (E. Mori *et al.*, 2015).

It is interesting to mention that these are the results observed in participants with a normal sense of smell, but olfactory training is also used in patients with olfactory dysfunction and it has been reported multiple times that smelling four odors every day during a few months leads to an improvement that is not specific to the odors that were used: scores in the Sniffin' Sticks test improved with training, especially in the discrimination and identification tasks (Altundag *et al.*, 2015; Damm *et al.*, 2014; Fleiner *et al.*, 2012; Geissler *et al.*, 2014; Haehner *et al.*, 2013; Hummel *et al.*, 2009; Konstantinidis *et al.*, 2013). However, in participants with a normal sense of smell, a

generalized effect of training was also observed. In a study where olfactory training consisted of tasks that were more complex than just passively smelling odors on a daily basis: the effect of training was measurable with the Sniffin' Sticks test, mostly with the identification task (Al Ain *et al.*, 2019). In our study, since sommelier training involves various complex exercises, we could have expected similar results with greater scores in the identification task, especially because it has also been shown that wine experts perform better than novices in high-order olfactory tasks such as identification, olfactory memory or discrimination of odorants within mixtures; but not in more basic tasks such as olfactory detection thresholds (Parr *et al.*, 2002; Poupon *et al.*, 2018; Poupon *et al.*, 2019). One key argument may explain the potentially conflicting results for the olfactory bulb (Sommelier students but not controls exhibit an increase in volume) and the behavioral test (controls but not sommelier students exhibit improved scores); the Sniffin'Sticks test was designed to detect olfactory dysfunction and to distinguish between individuals with olfactory dysfunction from those with no such problem. One may therefore argue that there is a ceiling effect for olfactory specialists, and this test might not be suited for accurately discriminating between people with a normal sense of smell and olfactory experts. We have shown that other tasks may be better suited to distinguish between individuals with normal olfactory function and olfactory specialists, e.g., the identification of components within a mixture of odours (Poupon et al., 2018). Further, anecdotally, we noticed that sommelier students have a more analytical approach: during the identification task, they took more time to answer. In the free identification task, while participants from the control group usually gave one answer, sommelier students used different descriptors as they could smell hints of different odors in each pen. Even when they were faced with a list of four descriptors in the cued identification task, they were more hesitant than the control group as they perceived notes corresponding to several of the four descriptors. This was most prominent for the chocolate odor, for which the list of descriptors also included vanilla and biscuit. In other case, mostly fruity odors, it was common to hear them say that none of the four descriptors fitted as the odor was too intense and not natural enough, especially compared to the refined nuances they are used to smelling in wine. In this context, it is important that the odorants used in the Sniffin' Sticks are artificial odors rather than the actual odor source. An alternative explanation could be put forward to explain the lack of association between olfactory

bulb volume changes and changes in olfactory scores, namely that olfactory bulb volume is not specifically associated with olfactory function. This explanation is however unlikely: first, the association between olfactory bulb volume and olfactory function is well established (Buschhuter et al., 2008; Liu et al., 2017; Rombaux et al., 2006; Seubert et al., 2013, Haehner et al., 2008; Negoias et al., 2017; Rombaux et al., 2009). Secondly, the sommelier students do get better at sensory and thus olfactory evaluations over the course of the 18 months of training, as novices would not be able to pass the final exams. There are other behavioral tests which would have been a better choice such as the identification of components in a mixture. We have shown that sommeliers outperform controls in this task (Poupon et al., 2018). Unfortunately, this test was not included in the present study.

Further studies should consider these points.

Discussion and limitations

We did not observe any significant changes in the olfactory performance of Sommeliers. This could be due to the fact that Sniffin Sticks is a clinical test which may not be suitable for professionals' performance evaluation. People report that this test smells unnatural. Additionally, the changes in the olfactory performance might be in different shape or tasks. On the contrary, we found out that control group's olfactory threshold tests performed better, although we do not know the reason behind this result, it could be that they became more sensitive to the odours and learnt throughout the training. Perhaps, Sommeliers had sensitive noses all along or they had already been accustomed to these odours all along.

It is also worth to note that we have relatively small sample for an imaging study. While the sample at baseline consisted of 34 participants, nine participants dropped out over the observation period of 18 months yielding a final sample size of 25. Our results have therefore to be interpreted cautiously.

Further studies should examine OB volume in sommeliers with more or less experience to test, for example, whether the OB keeps getting bigger over years, or if at some point a limit is reached and the OB stops growing. Future longitudinal studies should also be designed while keeping in

mind the idea that the effects of training-related plasticity are not linear to further test the over-production-pruning model: having more than two time points would be ideal to fully observe the effects of training-related brain plasticity.

Conclusion

In conclusion, this study aims to explore the effects of training-related brain plasticity in brain. Unlike other studies in which olfactory training consists of smelling a few odors every day during several weeks, the olfactory training we evaluated here is not as experimental since it is a sommelier training leading students to become professionals. OB volume increased during their training which constitutes the first data we have about OBs in olfaction experts. We also observed local increases or decreases of cortical thickness that support the overproduction-pruning model of plasticity according to which changes in the brain are nonlinear. It is worth to note that the positive changes in entorhinal cortex and the negative changes in other regions might be a question of timing. It could be that not every region evolves at the same rate.

References

R., Volle, E., Seassau, M., Duffau, H., Kinkingnehun, S., Samson, Y., Zhang, S., & Dubois, B Al Ain, S., Poupon, D., Hetu, S., Mercier, N., Steffener, J., & Frasnelli, J. (2019). Smell training improves olfactory function and alters brain structure. Neuroimage, 189, 45-54. doi:10.1016/j.neuroimage.2019.01.008

Altundag, A., Cayonu, M., Kayabasoglu, G., Salihoglu, M., Tekeli, H., Saglam, O., & Hummel, T. (2015). Modified olfactory training in patients with postinfectious olfactory loss. Laryngoscope, 125(8), 1763-1766. doi:10.1002/lary.25245

Amunts, K., Schlaug, G., Jancke, L., Steinmetz, H., Schleicher, A., Dabringhaus, A., & Zilles, K. (1997). Motor cortex and hand motor skills: structural compliance in the human brain. Hum Brain Mapp, 5(3), 206-215. doi:10.1002/(sici)1097-0193(1997)5:3<206::aid-hbm5>3.0.co;2-7

Askar, S. M., Elnashar, I. S., El-Anwar, M. W., Amer, H. S., El Shawadfy, M. A., Hosny, S. M., Bazeed, M. F., Ahmed, A. F., & Ghanem, M. A. (2015). Ipsilateral reduced olfactory bulb volume in patients

with unilateral nasal obstruction. Otolaryngol Head Neck Surg, 152(5), 959-963. doi:10.1177/0194599815573196

Banks, S. J., Sreenivasan, K. R., Weintraub, D. M., Baldock, D., Noback, M., Pierce, M. E., Frasnelli, J., James, J., Beall, E., Zhuang, X., Cordes, D., & Leger, G. C. (2016). Structural and Functional MRI Differences in Master Sommeliers: A Pilot Study on Expertise in the Brain. Front Hum Neurosci, 10, 414. doi:10.3389/fnhum.2016.00414

Bergmann, O., Spalding, K. L., & Frisen, J. (2015). Adult Neurogenesis in Humans. Cold Spring Harb Perspect Biol, 7(7), a018994. doi:10.1101/cshperspect.a018994

Bermudez, P., Lerch, J. P., Evans, A. C., & Zatorre, R. J. (2009). Neuroanatomical correlates of musicianship as revealed by cortical thickness and voxel-based morphometry. Cereb Cortex, 19(7), 1583-1596. doi:10.1093/cercor/bhn196

Buschhuter, D., Smitka, M., Puschmann, S., Gerber, J. C., Witt, M., Abolmaali, N. D., & Hummel, T. (2008). Correlation between olfactory bulb volume and olfactory function. Neuroimage, 42(2), 498-502. doi:10.1016/j.neuroimage.2008.05.004

Curtis, M. A., Kam, M., Nannmark, U., Anderson, M. F., Axell, M. Z., Wikkelso, C., Holtas, S., van Roon-Mom, W. M., Bjork-Eriksson, T., Nordborg, C., Frisen, J., Dragunow, M., Faull, R. L., & Eriksson, P. S. (2007). Human neuroblasts migrate to the olfactory bulb via a lateral ventricular extension. Science, 315(5816), 1243-1249. doi:10.1126/science.1136281

Dade, L. A., Zatorre, R. J., & Jones-Gotman, M. (2002). Olfactory learning: convergent findings from lesion and brain imaging studies in humans. Brain, 125(Pt 1), 86-101.

Dalton, P., Doolittle, N., & Breslin, P. A. (2002). Gender-specific induction of enhanced sensitivity to odors. Nat Neurosci, 5(3), 199-200. doi:10.1038/nn803

Damm, M., Pikart, L. K., Reimann, H., Burkert, S., Goktas, O., Haxel, B., Frey, S., Charalampakis, I., Beule, A., Renner, B., Hummel, T., & Huttenbrink, K. B. (2014). Olfactory training is helpful in postinfectious olfactory loss: a randomized, controlled, multicenter study. Laryngoscope, 124(4), 826-831. doi:10.1002/lary.24340

Delon-Martin, C., Plailly, J., Fonlupt, P., Veyrac, A., & Royet, J. P. (2013). Perfumers' expertise induces structural reorganization in olfactory brain regions. NeuroImage, 68, 55–62. https://doi.org/10.1016/j.neuroimage.2012.11.044

Di, X., Zhu, S., Jin, H., Wang, P., Ye, Z., Zhou, K., Zhuo, Y., & Rao, H. (2012). Altered resting brain function and structure in professional badminton players. Brain Connect, 2(4), 225-233. doi:10.1089/brain.2011.0050

Driemeyer, J., Boyke, J., Gaser, C., Buchel, C., & May, A. (2008). Changes in gray matter induced by learning--revisited. PLoS One, 3(7), e2669. doi:10.1371/journal.pone.0002669

du Boisgueheneuc, F., Levy,. (2006). Functions of the left superior frontal gyrus in humans: a lesion study. Brain, 129(Pt 12), 3315-3328. doi:10.1093/brain/awl244

Eavri, R., & Nedivi, E. (2013). Synaptogenesis in the Adult CNS – Neocortical Plasticity. Cellular Migration and Formation of Neuronal Connections, 713-721. doi:10.1016/B978-0-12-397266-8.00110-1

Fleiner, F., Lau, L., & Goktas, O. (2012). Active olfactory training for the treatment of smelling disorders. Ear Nose Throat J, 91(5), 198-203, 215.

Frasnelli, J., Livermore, A., Soiffer, A., & Hummel, T. (2002). Comparison of lateralized and binasal olfactory thresholds. Rhinology, 40(3), 129-134.

Frasnelli, J., Lundstrom, J. N., Boyle, J. A., Djordjevic, J., Zatorre, R. J., & Jones-Gotman, M. (2010). Neuroanatomical correlates of olfactory performance. Exp Brain Res, 201(1), 1-11. doi:10.1007/s00221-009-1999-7

Geissler, K., Reimann, H., Gudziol, H., Bitter, T., & Guntinas-Lichius, O. (2014). Olfactory training for patients with olfactory loss after upper respiratory tract infections. Eur Arch Otorhinolaryngol, 271(6), 1557-1562. doi:10.1007/s00405-013-2747-y

Gottfried, J. A. (2010). Central mechanisms of odour object perception. Nat Rev Neurosci, 11(9), 628-641. doi:10.1038/nrn2883

Haehner, A., Rodewald, A., Gerber, J. C., & Hummel, T. (2008). Correlation of olfactory function with changes in the volume of the human olfactory bulb. Arch Otolaryngol Head Neck Surg, 134(6), 621-624. doi:10.1001/archotol.134.6.621

Haehner, A., Mayer, A. M., Landis, B. N., Pournaras, I., Lill, K., Gudziol, V., & Hummel, T. (2009). High test-retest reliability of the extended version of the "Sniffin' Sticks" test. Chem Senses, 34(8), 705-711. doi:10.1093/chemse/bjp057

Haehner, A., Tosch, C., Wolz, M., Klingelhoefer, L., Fauser, M., Storch, A., Reichmann, H., & Hummel, T. (2013). Olfactory training in patients with Parkinson's disease. PLoS One, 8(4), e61680. doi:10.1371/journal.pone.0061680

Hang, W., Liu, G., Han, T., Zhang, J., & Zhang, Q. (2015). [A correlation study on olfactory bulb volumes with ages and olfactory function in healthy adults]. Zhonghua Er Bi Yan Hou Tou Jing Wai Ke Za Zhi, 50(9), 744-748.

Harley, E. M., Pope, W. B., Villablanca, J. P., Mumford, J., Suh, R., Mazziotta, J. C., Enzmann, D., & Engel, S. A. (2009). Engagement of fusiform cortex and disengagement of lateral occipital cortex in the acquisition of radiological expertise. Cereb Cortex, 19(11), 2746-2754. doi:10.1093/cercor/bhp051

Hedner, M., Larsson, M., Arnold, N., Zucco, G. M., & Hummel, T. (2010). Cognitive factors in odor detection, odor discrimination, and odor identification tasks. J Clin Exp Neuropsychol, 32(10), 1062-1067. doi:10.1080/13803391003683070

Huart, C., Rombaux, P., & Hummel, T. (2013). Plasticity of the human olfactory system: the olfactory bulb. Molecules, 18(9), 11586-11600. doi:10.3390/molecules180911586

Huart, C., Rombaux, P., & Hummel, T. (2019). Neural plasticity in developing and adult olfactory pathways - focus on the human olfactory bulb. J Bioenerg Biomembr, 51(1), 77-87. doi:10.1007/s10863-018-9780-x

Hummel, T., Sekinger, B., Wolf, S. R., Pauli, E., & Kobal, G. (1997). 'Sniffin' sticks': olfactory performance assessed by the combined testing of odor identification, odor discrimination and olfactory threshold. Chem Senses, 22(1), 39-52.

Hummel, T., Damm, M., Vent, J., Schmidt, M., Theissen, P., Larsson, M., & Klussmann, J. P. (2003). Depth of olfactory sulcus and olfactory function. Brain Res, 975(1-2), 85-89.

Hummel, T., Kobal, G., Gudziol, H., & Mackay-Sim, A. (2007). Normative data for the "Sniffin' Sticks" including tests of odor identification, odor discrimination, and olfactory thresholds: an upgrade based on a group of more than 3,000 subjects. Eur Arch Otorhinolaryngol, 264(3), 237-243. doi:10.1007/s00405-006-0173-0

Hummel, T., Rissom, K., Reden, J., Hahner, A., Weidenbecher, M., & Huttenbrink, K. B. (2009). Effects of olfactory training in patients with olfactory loss. Laryngoscope, 119(3), 496-499. doi:10.1002/lary.20101

Hummel, T., Fliessbach, K., Abele, M., Okulla, T., Reden, J., Reichmann, H., Wullner, U., & Haehner, A. (2010). Olfactory FMRI in patients with Parkinson's disease. Front Integr Neurosci, 4, 125. doi:10.3389/fnint.2010.00125

Jadauji, J. B., Djordjevic, J., Lundstrom, J. N., & Pack, C. C. (2012). Modulation of olfactory perception by visual cortex stimulation. J Neurosci, 32(9), 3095-3100. doi:10.1523/jneurosci.6022-11.2012

Johansson, C. B., Svensson, M., Wallstedt, L., Janson, A. M., & Frisen, J. (1999). Neural stem cells in the adult human brain. Exp Cell Res, 253(2), 733-736. doi:10.1006/excr.1999.4678

Klingberg, T. (2006). Development of a superior frontal-intraparietal network for visuo-spatial working memory. Neuropsychologia, 44(11), 2171-2177. doi:10.1016/j.neuropsychologia.2005.11.019

Konstantinidis, I., Tsakiropoulou, E., Bekiaridou, P., Kazantzidou, C., & Constantinidis, J. (2013). Use of olfactory training in post-traumatic and postinfectious olfactory dysfunction. Laryngoscope, 123(12), E85-90. doi:10.1002/lary.24390

Kornack, D. R., & Rakic, P. (2001). The generation, migration, and differentiation of olfactory neurons in the adult primate brain. Proc Natl Acad Sci U S A, 98(8), 4752-4757. doi:10.1073/pnas.081074998

Kupers, R., Beaulieu-Lefebvre, M., Schneider, F. C., Kassuba, T., Paulson, O. B., Siebner, H. R., & Ptito, M. (2011). Neural correlates of olfactory processing in congenital blindness. Neuropsychologia, 49(7), 2037-2044. doi:10.1016/j.neuropsychologia.2011.03.033

Kupers, R., & Ptito, M. (2014). Compensatory plasticity and cross-modal reorganization following early visual deprivation. Neurosci Biobehav Rev, 41, 36-52. doi:10.1016/j.neubiorev.2013.08.001

Lazarini, F., & Lledo, P. M. (2011). Is adult neurogenesis essential for olfaction? Trends Neurosci, 34(1), 20-30. doi:10.1016/j.tins.2010.09.006

Lindenberger, U., Wenger, E., & Lovden, M. (2017). Towards a stronger science of human plasticity. Nat Rev Neurosci, 18(5), 261-262. doi:10.1038/nrn.2017.44

Liu, H., Hang, W., Liu, G., & Han, T. (2017). [Olfactory bulb volume in patients with posttraumatic olfactory dysfunction]. Zhonghua Er Bi Yan Hou Tou Jing Wai Ke Za Zhi, 52(4), 273-277. doi:10.3760/cma.j.issn.1673-0860.2017.04.007

Lois, C., Garcia-Verdugo, J. M., & Alvarez-Buylla, A. (1996). Chain migration of neuronal precursors. Science, 271(5251), 978-981. doi:10.1126/science.271.5251.978

Lotsch, J., Schaeffeler, E., Mittelbronn, M., Winter, S., Gudziol, V., Schwarzacher, S. W., Hummel, T., Doehring, A., Schwab, M., & Ultsch, A. (2014). Functional genomics suggest neurogenesis in the adult human olfactory bulb. Brain Struct Funct, 219(6), 1991-2000. doi:10.1007/s00429-013-0618-3

MacMillan, N., & Creelman, C. (2005). Detection theory: a user's guide. Taylor & Francis.

Maguire, E. A., Gadian, D. G., Johnsrude, I. S., Good, C. D., Ashburner, J., Frackowiak, R. S., & Frith, C. D. (2000). Navigation-related structural change in the hippocampi of taxi drivers. Proc Natl Acad Sci U S A, 97(8), 4398-4403. doi:10.1073/pnas.070039597

Ming, G. L., & Song, H. (2011). Adult neurogenesis in the mammalian brain: significant answers and significant questions. Neuron, 70(4), 687-702. doi:10.1016/j.neuron.2011.05.001

Mishkin, M., Ungerleider, L. G., & Macko, K. A. (1983). Object vision and spatial vision: Two cortical pathways. . Trends in Neurosciences.(6), 414–417.

Mori, E., Petters, W., Schriever, V. A., Valder, C., & Hummel, T. (2015). Exposure to odours improves olfactory function in healthy children. Rhinology, 53(3), 221-226. doi:10.4193/Rhin14.192

Mori, K., Nagao, H., & Yoshihara, Y. (1999). The olfactory bulb: coding and processing of odor molecule information. Science, 286(5440), 711-715.

Negoias, S., Pietsch, K., & Hummel, T. (2017). Changes in olfactory bulb volume following lateralized olfactory training. Brain Imaging and Behavior, 11(4), 998-1005. doi:10.1007/s11682-016-9567-9

Pagano, S. F., Impagnatiello, F., Girelli, M., Cova, L., Grioni, E., Onofri, M., Cavallaro, M., Etteri, S., Vitello, F., Giombini, S., Solero, C. L., & Parati, E. A. (2000). Isolation and characterization of neural stem cells from the adult human olfactory bulb. Stem Cells, 18(4), 295-300. doi:10.1634/stemcells.18-4-295

Pardini, M., Huey, E. D., Cavanagh, A. L., & Grafman, J. (2009). Olfactory function in corticobasal syndrome and frontotemporal dementia. Arch Neurol, 66(1), 92-96. doi:10.1001/archneurol.2008.521

Parr, W. V., Heatherbell, D., & White, K. G. (2002). Demystifying wine expertise: olfactory threshold, perceptual skill and semantic memory in expert and novice wine judges. Chem Senses, 27(8), 747-755.

Patel, R. M., & Pinto, J. M. (2014). Olfaction: anatomy, physiology, and disease. Clin Anat, 27(1), 54-60. doi:10.1002/ca.22338

Peng, P., Gu, H., Xiao, W., Si, L. F., Wang, J. F., Wang, S. K., Zhai, R. Y., & Wei, Y. X. (2013). A voxel-based morphometry study of anosmic patients. Br J Radiol, 86(1032), 20130207. doi:10.1259/bjr.20130207

Plailly, J., Bensafi, M., Pachot-Clouard, M., Delon-Martin, C., Kareken, D. A., Rouby, C., Segebarth, C., & Royet, J. P. (2005). Involvement of right piriform cortex in olfactory familiarity judgments. Neuroimage, 24(4), 1032-1041. doi:10.1016/j.neuroimage.2004.10.028

Poupon, D., Hummel, T., Haehner, A., Welge-Luessen, A., & Frasnelli, J. (2017). Nostril Differences in the Olfactory Performance in Health and Disease. Chem Senses, 42(8), 625-634. doi:10.1093/chemse/bjx041

Poupon, D., Fernandez, P., Archambault Boisvert, S., Migneault-Bouchard, C., & Frasnelli, J. (2018). Can the Identification of Odorants Within a Mixture Be Trained? Chem Senses, 43(9), 721-726. doi:10.1093/chemse/bjy060

Poupon, D., Fernandez, P., & Frasnelli, J. (2019). Sommelier Students Display Superior Abilities to Identify but Not to Detect or Discriminate Odors Early in their Training. Chem Perception. doi:10.1007/s12078-019-09256-w

Rombaux, P., Mouraux, A., Bertrand, B., Nicolas, G., Duprez, T., & Hummel, T. (2006). Retronasal and orthonasal olfactory function in relation to olfactory bulb volume in patients with posttraumatic loss of smell. Laryngoscope, 116(6), 901-905. doi:10.1097/01.mlg.0000217533.60311.e7

Rombaux, P., Duprez, T., & Hummel, T. (2009). Olfactory bulb volume in the clinical assessment of olfactory dysfunction. Rhinology, 47(1), 3-9.

Sanai, N., Tramontin, A. D., Quinones-Hinojosa, A., Barbaro, N. M., Gupta, N., Kunwar, S., Lawton, M. T., McDermott, M. W., Parsa, A. T., Manuel-Garcia Verdugo, J., Berger, M. S., & Alvarez-Buylla, A. (2004). Unique astrocyte ribbon in adult human brain contains neural stem cells but lacks chain migration. Nature, 427(6976), 740-744. doi:10.1038/nature02301

Sanai, N., Berger, M. S., Garcia-Verdugo, J. M., & Alvarez-Buylla, A. (2007). Comment on "Human neuroblasts migrate to the olfactory bulb via a lateral ventricular extension". Science, 318(5849), 393; author reply 393. doi:10.1126/science.1145011

Sanai, N., Nguyen, T., Ihrie, R. A., Mirzadeh, Z., Tsai, H. H., Wong, M., Gupta, N., Berger, M. S., Huang, E., Garcia-Verdugo, J. M., Rowitch, D. H., & Alvarez-Buylla, A. (2011). Corridors of migrating neurons in the human brain and their decline during infancy. Nature, 478(7369), 382-386. doi:10.1038/nature10487

Schwob, J. E., & Costanzo, R. M. (2010). Regeneration of the Olfactory Epithelium. The Senses: A Comprehensive Reference, 4, 591-612. doi:10.1016/B978-012370880-9.00115-8

Seubert, J., Freiherr, J., Frasnelli, J., Hummel, T., & Lundstrom, J. N. (2013). Orbitofrontal cortex and olfactory bulb volume predict distinct aspects of olfactory performance in healthy subjects. Cereb Cortex, 23(10), 2448-2456. doi:10.1093/cercor/bhs230

Stickel, S., Weismann, P., Kellermann, T., Regenbogen, C., Habel, U., Freiherr, J., & Chechko, N. (2019). Audio-visual and olfactory-visual integration in healthy participants and subjects with autism spectrum disorder. Hum Brain Mapp, 40(15), 4470-4486. doi:10.1002/hbm.24715

Takeuchi, H., Taki, Y., Hashizume, H., Sassa, Y., Nagase, T., Nouchi, R., & Kawashima, R. (2011). Effects of training of processing speed on neural systems. J Neurosci, 31(34), 12139-12148. doi:10.1523/jneurosci.2948-11.2011

Ungerleider, L. G., & Haxby, J. V. (1994). 'What' and 'where' in the human brain. Curr Opin Neurobiol, 4(2), 157-165.

Valk, S. L., Bernhardt, B. C., Trautwein, F. M., Bockler, A., Kanske, P., Guizard, N., Collins, D. L., & Singer, T. (2017). Structural plasticity of the social brain: Differential change after socio-affective and cognitive mental training. Sci Adv, 3(10), e1700489. doi:10.1126/sciadv.1700489

Welge-Lussen, A., Wattendorf, E., Schwerdtfeger, U., Fuhr, P., Bilecen, D., Hummel, T., & Westermann, B. (2009). Olfactory-induced brain activity in Parkinson's disease relates to the expression of event-related potentials: a functional magnetic resonance imaging study. Neuroscience, 162(2), 537-543. doi:10.1016/j.neuroscience.2009.04.050

Wenger, E., Kuhn, S., Verrel, J., Martensson, J., Bodammer, N. C., Lindenberger, U., & Lovden, M. (2017). Repeated Structural Imaging Reveals Nonlinear Progression of Experience-Dependent Volume Changes in Human Motor Cortex. Cereb Cortex, 27(5), 2911-2925. doi:10.1093/cercor/bhw141

Westermann, B., Wattendorf, E., Schwerdtfeger, U., Husner, A., Fuhr, P., Gratzl, O., Hummel, T., Bilecen, D., & Welge-Lussen, A. (2008). Functional imaging of the cerebral olfactory system in patients with Parkinson's disease. J Neurol Neurosurg Psychiatry, 79(1), 19-24. doi:10.1136/jnnp.2006.113860

Xu, T., Yu, X., Perlik, A. J., Tobin, W. F., Zweig, J. A., Tennant, K., Jones, T., & Zuo, Y. (2009). Rapid formation and selective stabilization of synapses for enduring motor memories. Nature, 462(7275), 915-919. doi:10.1038/nature08389

Yousem, D. M., Geckle, R. J., Bilker, W. B., & Doty, R. L. (1998). Olfactory bulb and tract and temporal lobe volumes. Normative data across decades. Ann N Y Acad Sci, 855, 546-555. doi:10.1111/j.1749-6632.1998.tb10624.x

Zatorre, R. J., Jones-Gotman, M., & Rouby, C. (2000). Neural mechanisms involved in odor pleasantness and intensity judgments. Neuroreport, 11(12), 2711-2716.

Book Chapter – Different methods to assess trigeminal system

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Abstract

The chemical senses work both together and independently when eating and drinking. When fibers of the trigeminal nerve are stimulated, they evoke sensations such as irritation, pain, cooling, or burning which can be perceived as pleasant or unpleasant; the sensory system allowing for these perceptions is the trigeminal system. Typically, chemical substances do not activate the trigeminal system alone, as they usually also evoke an olfactory perception simultaneously, and this is already at lower concentrations. It is therefore challenging to assess sensitivity of the trigeminal system. Here, we present a protocol that includes four different techniques enabling to assess trigeminal sensation at different levels of processing: (A) perception thresholds; (B) detection through a lateralization task; (C) peripheral neural activity via negative mucosal potential recordings; and (D) central neural activity via trigeminal event-related potentials.

Keywords: Trigeminal system, Lateralization test, Threshold, Negative mucosal potential, Event related potentials, Flavor

Introduction

The trigeminal system is one of the three chemosensory systems, next to smell and taste. This system has a very important function during food intake and in the affective states evoked by the food that stimulate it. It is linked to the trigeminal nerve, the fifth cranial nerve. This is the thickest cranial nerve with three eponymous branches that are called ophthalmic (CNV1), maxillary (CNV2), and mandibular (CNV3) nerve.

The sensory part of the trigeminal nerve is responsible for the somatosensory sensations we receive from the skin and mucosa of our face, tongue, neck, and throat (Sanders, 2010). It therefore also allows for the perception of sensations such as tingling, burning, cooling caused by the interaction of chemical substances with receptors located on nerve fibers (Frasnelli et al., 2010). Examples for this are the preference of cooling sensations evoked by menthol in sweets or ice cream, as well as the burning sensation from capsaicin in spicy dishes (Liu & Simon, 1994). Some humans prefer to eat spicy or pungent food with an important interindividual and intercultural difference.

In fact, flavor is the combination of trigeminal, olfactory, and gustatory (and other) inputs (Laing & Jinks, 1996). Although the three chemosensory systems are nominally independent from each other as they rely on distinct receptors and neural pathways, during daily life activities such as the consumption of food or drinks, two or more of the three chemosensory systems are usually stimulated together. Because of the overlapping stimulus space as well as common central nervous processing areas, the three chemical senses interact mutually and enhance or suppress each other (Tremblay & Frasnelli, 2018).

To assess sensitivity of the trigeminal system, one has to use stimuli that are either purely trigeminal, i.e., they stimulate the trigeminal system exclusively, such as carbon dioxide and capsaicin, or mixed olfactory–trigeminal, i.e., they stimulate both the trigeminal system and the olfactory system, such as menthol, eucalyptol, or cinnamaldehyde. In the latter cases, it is challenging to separate the olfactory sensation from the trigeminal one. Different methods have been put forward to overcome this challenge. First, we can (1) use substances which solely stimulate the trigeminal systems to eliminate the interference from other senses such as olfaction. Second, we can (2) use methods that solely rely on activation of the trigeminal system. As an alternative to these two approaches, if the aim is to assess the capacity of a given substance to activate the trigeminal system, one can test (3) participants with a working olfactory system who are instructed to focus on the trigeminal aspect of a chemosensory stimulus (e.g., “please rate how pungent this odor is.”) (Frasnelli et al., 2009, Laska, 2001), (see Note 1), or (4) individuals with no working sense of smell, i.e., individuals with anosmia (Doty et al., 1978, Laska et al., 1997), (see Note 2).

In this chapter, we propose a protocol that integrates four different methods to assess trigeminal sensation at different levels of processing: (A) psychophysical detection threshold measurements; (B) psychophysical lateralization task; (C) peripheral neural activity by recordings of negative mucosal potentials; and (D) central neural activity by measured evoked potentials related to trigeminal events. The collected measures can be correlated with subjective measures related to the emotional or hedonic perception of these trigeminal stimulations in order to better understand the interplay between trigeminal system and emotions in the food domain.

Materials

Threshold for trigeminal stimuli

Oral testing (capsaicin)

Prepare five solutions of capsaicin in 10% ethanol and 90% distilled water: Solution 1: 3.16 10 5%, Solution 2: 1 10 4%, Solution 3: 3.16 10 4%, Solution 4 ¼ 1 10 3%, and Solution 5 ¼ 3.16 10 3% w/w. Prepare one blank solution containing 10% ethanol and 90% distillated water.

Nasal testing (CO2)

Carbon dioxide is usually provided in pure form in gas tanks. It must be diluted with air, typically with the help of an olfactometer (see Note 3).

Lateralization task

Two amber bottles (60 mL); add a cotton ball. One bottle is filled with 15 mL of 50% eucalyptol in propylene glycol. The other bottle is filled with 15 mL odorless propylene glycol (Frasnelli & Hummel, 2003) (see Note 4). A custom-made lid that snugly fits onto the nostril is put on the bottles. It has two openings one of which is inside the nostril, the other outside (Frasnelli, Oehrn, et al., 2009).

Negative mucosal potential

Carbon dioxide (Kobal & Hummel, 1988) 60% v/v (see Notes 4 and 5). A silver wire electrode of 0.3 mm diameter. A saturated NaCl solution. A 5 V battery. A PTFE tube (0.8 mm outer diameter, inner diameter 0.4 mm). 1% Agar in Ringer solution. An AC amplifier (bandwidth 0.1–100 Hz). Olfactometer.

Trigeminal event-related potentials

Carbon dioxide (Kobal & Hummel, 1988) 60% v/v (see Note5). EEG electrodes. An AC amplifier (bandwidth 0.1–100 Hz) (see Notes 6 and 7).

Methods

Thresholds for trigeminal stimuli

Oral testing (capsaicin)

1. The participant is seated in a quiet room.

2. The experimenter starts at the lowest concentration and increases the intensity toward higher concentrations.

3. The experimenter applies the capsaicin solution (applied volume is 0.02mL) topically onto na-solabial folds in a single-blind simultaneous split-face application in a randomized order so that the side capsaicin/blank solution is applied to are not repetitively same (Jourdain et al., 2005). The blank solution is used as a control contralaterally.

4. The participant is asked to indicate the presence of a painful sensation.

5. When the subject indicates a painful sensation lasting more than 30 s, the test is stopped. At this point, their detection threshold for capsaicin is determined.

Nasal testing (CO_2)

1. The participant is seated in a well-ventilated room.

2. White noise is applied on the background with or without headphones to prevent subjects from hearing the switching sound of the stimulus presentation device.

3. CO_2 is presented by means of an olfactometer. Concentrations are 30–70% v/v (in increments of 5% v/v); typical stimulus duration is 200 ms.

4. Respecting an interstimulus interval of 40s will minimize habituation; therefore, stimuli can be applied ascending/descending staircases (Hummel & Kobal, 1999).

5. Starting with the weakest concentration, experimenters present three stimuli in a row (40 s interstimulus interval) to participants (in random order, two blanks containing pure air and one CO_2 containing stimulus).

6. Participants indicate which stimulus contains CO2.

7. The correct identification of two correct stimuli in a row triggers subsequent presentation of a weaker stimulus; the incorrect identification of a stimulus triggers the presentation of a stronger stimulus.

8. Testing continues until seven staircase reversals are obtained; the average of the last four reversals will serve as an estimate for the CO2 threshold.

Lateralization task

1. The participant is seated in a well-ventilated room.

2. The participant is simultaneously presented with one bottle to one nostril, while the other receives the other bottle (see Note 8).

3. The experimenter fits the bottles' lids to each nostril snugly so that the air space of one bottle reaches only this nostril.

4. The participant is instructed to take one sniff.

5. The participant indicates which nostril has been stimulated (see Note 9).

6. This is repeated 40 times in a pseudorandomized counterbalanced order (20 trials with stimulation of one nostril; 20 trials with stimulation of the other nostril).

7. The number of correct localizations is counted and used as an estimate of trigeminal sensitivity (see Notes 10–12) (Fig. 1).

Negative mucosal potential

1. Preparation of the electrode: The silver electrode is chloridized using the 5 V battery for 10 min in a water bath with saturated NaCl solution.

2. Insulation of the electrode: 10% Agar is sucked into the PTFE tubing; the electrode is then inserted into the tubing by ascertaining an agar bridge.

3. Placement of the electrode: under endoscopic control, the electrode tip is placed onto the respiratory mucosa of the nose. A frame similar to lensless glasses is used to keep the electrode in place (see Note 13).

4. The reference electrode is placed at the bridge of the nose, contralateral to the recording site.

5. The participant is seated in a well-ventilated room.

6. Stimulus duration of 500 ms.

7. Interstimulus interval is 40 s.

8. The electrode can be replaced if there is no signal.

9. A minimum of three repetitions is needed.

10. Recordings are averaged offline.

11. Averages will show a one-phased signal; the latency and the amplitude are measured.

Trigeminal event-related potentials

1. The participant is seated in a faraday cage or equivalent to prevent interference from outside, in a well-ventilated room.

2. Electrodes are placed on regions of interests over the scalp using the international 10/20 system.

3. Stimuli are presented by means of an olfactometer that does not interfere with EEG signals.

4. White noise is applied on background with or without headphones to prevent subjects from hearing the switching sound of the stimulus presentation device.

5. Stimulus duration is 200 ms (see Note 14).

6. Interstimulus intervals (ISI) of 30–40 s.

7. At least 10 recordings per condition have to be recorded in order to average out background noise (see Note 15).

8. Responses are averaged offline; the latency and amplitudes of ERP peaks are measured, either from baseline or from peak to peak. ERP responses follow a distinct pattern with positive (P) and negative (N) peaks that appear in typical order in general. Typical peaks are P1, N1, P2, and P3.

Notes

1. This simple approach has the disadvantage that since most trigeminal stimuli do also activate the olfactory system, there will be a non-negligible confusion between the trigeminal and olfactory percept, e.g., to separate the freshness of peppermint from its minty smell.

2. While in theory, this allows for the assessment of the trigeminal impact of a given substance with no interference from the sense of smell, this approach has another drawback: acquired anosmia is typically associated with reduced trigeminal sensitivity (Frasnelli et al., 2007).

3. The term olfactometer, in this context, describes a tool that allows for some form of automated stimulation with volatile substances. Since the development of the first olfactometer by Dutch physiologist Hendrik Zwaardemaker in 1887, several devices have been introduced that allow for stimulation with chemical stimuli using high precision in terms of stimulus concentration, duration, and with rapid onset. Furthermore, most devices can be controlled by a computer. They are available in different degrees of complexity and therefore price ranges, but only few are commercially available. A sophisticated olfactometer was introduced in the 1980s by Gerd Kobal (Kobal, 1985). This device produces a constant flow of humidified and heated (both controlled) air that can be odorized. Its switching device allows for controlling stimulus duration in the range of milliseconds with rapid rise times and offset. A tool with these characteristics is needed if one aims to investigate electrophysiological responses to trigeminal stimulation, as it is free of mechanical co-stimulation. Modern versions of these instruments allow for birhinal stimulation with up to 8 independent stimulus channels. This device can be acquired from the German manufacturer Burghart. (https://www.burghart-mt.de/en/). Another olfactometer was developed by Tyler Lorig roughly 15 years later (Lorig et al., 1999). This device had a shorter onset time but does not control humidity or heat of the delivered air f low. Such a tool is mainly used in the context of fMRI, where lower airflows are needed and therefore challenges inherent to the delivery of air with room temperature and humidity are less pronounced. A similar olfactometer was introduced in 2010

by Johan Lundstrom and colleagues (Lundstrom et al., 2010). This device again does not provide humidification or heating and may therefore not be useful if one aims at recording electrophysiological responses. However, crucially, the publication comes with a detailed construction manual with the off-the-shelf components, so that this device can be built from scratch with a limited budget in the range of 20.000 $. Additional olfactometers, mainly to be used for behavioral studies or in fMRI settings, are manufactured by Sensonics Inc (https://sensonics.com/) and Osmic Enterprises (http:// www.osmicenterprises.com/).

4. Alternative stimuli can be used: nicotine (Renner & Schreiber, 2012), eucalyptol menthol (Frasnelli & Hummel, 2003), cinnamaldehyde, or others, which can be used in neat concentrations or dilutions, equally CO2 (Frasnelli, Oehrn, et al., 2009). In fact, most odorants activate the trigeminal system in higher concentrations, i.e., they are mixed olfactory–trigeminal stimuli (Doty et al., 1978). This group of stimuli includes typically used stimuli such as eucalyptol (Muschenich et al., 2019), menthol, and cinnamaldehyde (Klein et al., 2011). Even phenyl ethanol, a substance commonly used in olfactory research with a distinct rose odor activates the trigeminal system in higher concentrations (Frasnelli et al., 2011). For mixed olfactory–trigeminal stimuli, the concentration needed to activate the trigeminal system is usually several orders of magnitude above the concentration needed to activate the olfactory system (Tremblay & Frasnelli, 2018). An exception to this rule is ammonia, which activates trigeminal system even at low concentrations (Cometto-Muñiz & Cain, 1984).

5. The negative mucosal potential (NMP) is an electrophysiological signal from the surface of the respiratory mucosa that responses to chemosensory (specifically nociceptive) stimulation (Kobal, 1985). As such, it is free of olfactory components. The NMP therefore assesses the functionality of the periphery of the trigeminal system. It is a monophasic signal and the result of the receptor potentials to painful chemosensory stimuli (Hummel et al., 1996). It is also linked to the activation of C fibers and Aδ fibers of the trigeminal nerve. This technique needs experienced researchers.

6. Event-related potentials are the result of the concerted electro physiological response of the brain to sensory stimulation (Helfrich & Knight, 2019, Kobal & Hummel, 1994). This technique relies on stimulus delivery by an olfactometer with controlled humidity and heat. When CO2 is

used, pure trigeminal ERP can be recorded, alternatively, mixed olfactory–trigeminal stimuli will yield mixed ERP.

7. The long ISI yields ERP experiments long and, together with the need of sophisticated olfactometers, only feasible in highly specialized research lab settings. ERP allow to assess trigeminal processing with unmatched temporal resolution, but rather weak spatial resolution. Stimulus duration can vary between (100–500 ms).

8. In passive stimulation, squeezable bottles are used and participants do not actively sniff, they continue breathing normally. Instead, the bottles are compressed which delivers the air puff to the nostrils (Frasnelli et al., 2009).

9. To avoid left-right confusion, they can be instructed to raise the hand on the side of the perceived eucalyptol.

10. In an alternative method, staircase approach might be used to decide the final threshold (Wysocki et al., 2003).

11. Trigeminal system acts as a mass detector rather than a concentration detector (Cometto-Muñiz & Cain, 1984, Cometto-Muniz & Cain, 1998); therefore, the same stimulus intensity can be achieved by varying stimulus concentration and stimulus duration. Therefore, some recent approaches apply stimuli of varying duration (Frasnelli et al., 2003).

12. Using a mixture of components might affect the thresholds since when two or more ingredients are combined, they might enhance or hinder each other's effects (Hummel et al., 2013).

13. Electrode placement can be marked.

14. Stimulus duration can vary between (100–500 ms).

15. Additionally, since one challenge inherent to trigeminal stimulation is that activation of the trigeminal system may lead to blinking, sneezing, or coughing that all of which induce artifacts on the readings, eye movements can be recorded to later contaminated recordings from further analysis.

References

Sanders RD (2010) The trigeminal (V) and facial (VII) cranial nerves: head and face sensation and movement. Psychiatry (Edgmont) 7(1):13–16. https://www.ncbi.nlm.nih.gov/ pubmed/20386632

Frasnelli J, Schuster B, Hummel T (2010) Olfactory dysfunction affects thresholds to tri geminal chemosensory sensations. Neurosci Lett 468(3):259–263. https://doi.org/10.1016/j.neulet.2009.11.008

Liu L, Simon SA (1994) A rapid capsaicin activated current in rat trigeminal ganglion neurons. Proc Natl Acad Sci U S A 91(2): 738–741. https://doi.org/10.1073/pnas.91. 2.738

Laing D, Jinks A (1996) Flavour perception mechanisms. Trends Food Sci Technol 7(12): 387–389. https://doi.org/10.1016/s0924 2244(96)10049-2

Tremblay C, Frasnelli J (2018) Olfactory and trigeminal systems interact in the periphery. Chem Senses 43(8):611–616. https://doi. org/10.1093/chemse/bjy049

Frasnelli J, Charbonneau G, Collignon O, Lepore F (2009) Odor localization and sniff ing. Chem-Senses34(2):139–144. https://doi. org/10.1093/chemse/bjn068

Laska M (2001) Perception of trigeminal chemosensory qualities in the elderly. Chem Senses 26(6):681–689. https://doi.org/10. 1093/chemse/26.6.681

Doty RL, Brugger WE, Jurs PC, Orndorff MA, Snyder PJ, Lowry LD (1978) Intranasal trigeminal stimulation from odorous volatiles: psychometric responses from anosmic and normal humans. Physiol Behav 20(2):175–185. https://doi.org/10.1016/0031-9384(78) 90070-7

Laska M, Distel H, Hudson R (1997) Trigeminal perception of odorant quality in congenitally anosmic subjects. Chem Senses 22(4): 447–456. https://doi.org/10.1093/chemse/ 22.4.447

Frasnelli J, Hummel T (2003) Age-related decline of intranasal trigeminal sensitivity: is it a peripheral event? Brain Res 987(2):201–206. https://doi.org/10.1016/s0006-8993(03) 03336-5

Frasnelli J, Oehrn C, Jones-Gotman M (2009) Effects of oral irritation on olfaction. Food Chem 113(4):1003–1007. https://doi.org/ 10.1016/j.foodchem.2008.08.052

Kobal G, Hummel C (1988) Cerebral chemosensory evoked potentials elicited by chemical stimulation of the human olfactory and respiratory nasal mucosa. Electroencephalogr Clin Neurophysiol 71(4):241–250. https://doi. org/10.1016/0168-5597(88)90023-8

Jourdain R, Bastien P, de Lacharriere O, Rubinstenn G (2005) Detection thresholds of capsaicin: a new test to assess facial skin neuro sensitivity. J Cosmet Sci 56(3):153–166. https://www.ncbi.nlm.nih.gov/pubmed/1 6116520

Hummel T, Kobal G (1999) Chemosensory event-related potentials to trigeminal stimuli change in relation to the interval between repetitive stimulation of the nasal mucosa. Eur Arch Otorhinolaryngol 256:16–21. https:// doi.org/10.1007/s004050050115

Frasnelli J, Schuster B, Hummel T (2007) Interactions between olfaction and the trigeminal system: what can be learned from olfactory loss. Cerebral Cortex (New York, N.Y.: 1991) 17(10):2268–2275. https://doi.org/10. 1093/cercor/bhl135

Kobal G (1985) Pain-related electrical potentials of the human nasal mucosa elicited by chemical stimulation. Pain 22(2):151–163. https://doi.org/10.1016/0304-3959(85) 90175-7

Lorig TS, Elmes DG, Zald DH, Pardo JV (1999) A computer-controlled olfactometer for fMRI and electrophysiological studies of olfaction. Behav Res Methods Instrum Com put 31(2):370–375. https://doi.org/10. 3758/bf03207734

Lundstrom JN, Gordon AR, Alden EC, Boesveldt S, Albrecht J (2010) Methods for building an inexpensive computer-controlled olfactometer for temporally-precise experiments. Int J Psychophysiol 78(2):179–189. https://doi.org/10.1016/j.ijpsycho.2010. 07.007

Renner B, Schreiber K (2012) Olfactory and trigeminal interaction of menthol and nicotine in humans. Exp Brain Res 219(1):13–26. https://doi.org/10.1007/s00221-012 3063-2

Müschenich FS, Sijben R, Gallenmüller F, Singer M, Rodriguez-Raecke R, Di Francesco ME, Wiesmann M, Freiherr J (2019) Eucalyptol masks the olfactory but not the trigeminal sensation of ammonia. Chem Senses 44(9): 733–741. https://doi.org/10.1093/chemse/ bjz065

Klein AH, Carstens MI, Zanotto KL, Sawyer CM, Ivanov M, Cheung S, Carstens E (2010) Self- and cross-desensitization of oral irritation by menthol and cinnamaldehyde (CA) via peripheral interactions at trigeminal sensory neurons. Chem Senses 36(2):199–208. https://doi.org/10.1093/chemse/bjq115

Frasnelli J, Albrecht J, Bryant B, Lundstrom JN (2011) Perception of specific trigeminal chemosensory agonists. Neuroscience 189:377 383. https://doi.org/10.1016/j.neurosci ence.2011.04.065

Cometto-Muniz JE, Cain WS (1984) Temporal integration of pungency. Chem Senses 8(4): 315–327. https://doi.org/10.1093/chemse/ 8.4.315

Hummel T, Schiessl C, Wendler J, Kobal G (1996) Peripheral electrophysiological responses decrease in response to repetitive painful stimulation of the human nasal mucosa. Neurosci Lett 212:37–40. https://doi.org/ 10.1016/0304-3940(96)12767-1

Helfrich RF, Knight RT(2019) Cognitive neurophysiology: event-related potentials. Handb Clin Neurol 160:543–558. https://doi.org/ 10.1016/B978-0-444-64032-1.00036-9

Kobal G, Hummel T (1994) Olfactory (chemosensory) event-related potentials. Toxicol Ind Health 10(4–5):587–596. https://www. ncbi.nlm.nih.gov/pubmed/7778117

Wysocki CJ, Cowart BJ, Radil T (2003) Nasal trigeminal chemosensitivity across the adult life span. Percept Psychophys 65(1):115–122. https://doi.org/10.3758/bf03194788

Cometto-Muniz JE, Cain WS (1998) Trigeminal and olfactory sensitivity: comparison of modalities and methods of measurement. Int Arch Occup Environ Health 71(2):105–110. https://doi.org/10.1007/s004200050256

Frasnelli J, Lotsch J, Hummel T (2003) Event related potentials to intranasal trigeminal stimuli change in relation to stimulus concentration and stimulus duration. J Clin Neurophysiol 20(1):80–86. https://doi.org/ 10.1097/00004691-200302000-00011

Hummel T, Olgun S, Gerber J, Huchel U, Frasnelli J (2013) Brain responses to odor mixtures with sub-threshold components. Front Psychol 4:786. https://doi.org/10.3389/ fpsyg.2013.00786