

# INTRODUCTION

An important function of the olfactory system is to discriminate and identify behaviorally relevant odors from a complex background that often consists of more than hundreds odorous stimuli. Accordingly, for better understanding olfactory system, it is essential to investigate how the system makes representations of complex odor stimuli in olfactory nervous system, and processes them.

### *1 Excitatory transduction cascade*

The initial events in olfactory perception occur in olfactory receptor neurons (ORNs), which are embedded in the olfactory epithelium (Figure 1). The ORN is a bipolar nerve cell. From its apical pole, it extends a single dendrite to the epithelial surface. Numerous cilia protrude from this dendrite to the mucus, providing an extensive perceptive surface for the detection of odorants. From the opposite pole, the neuron sends a single axon to the olfactory bulb of the brain. Odorant that dissolved in the nasal mucus binds to specific odorant receptors (ORs) on the plasma membrane of the cilia of ORNs and thereby induces a cascade of signal transduction events that culminate in the generation of action potentials in the sensory axon and the transmission of signals to the olfactory bulb of the brain (reviewed by Mori and Yoshihara, 1995).

Recently, a large multigene family was identified in rats, which appeared to code for hundreds of different ORs expressed in ORNs (Buck and Axel, 1991). The ORs belong to a large superfamily of G-protein-coupled receptors; and, homogeneous families of OR genes have now been identified in a variety of vertebrate species (for review, see Buck, 1996). Raming et al. (1993) have shown that expression of at least one member of this gene family endows those cells with the ability to selectively respond to a particular subset of

odorants.

The binding of odorants to ORs induces a transduction cascade that leads to depolarization of the ORNs and action potential generation in the olfactory axons. The mechanism of excitatory olfactory receptor transduction has been intensely studied. Figure 3 shows a schematic diagram of models of olfactory transduction cascade. It is now well established that odorants bind to ORs and cause the activation of adenylate cyclase via a receptor-coupled G-protein (Sklar et al., 1986; Lowe et al., 1989; Bakalyar and Reed, 1990; Breer et al., 1990). As a result, intracellular cAMP levels increase, causing the opening of a cyclic nucleotide-gated, non-selective cation channel (CNG-channel) on the plasma membrane of the cilia (Nakamura and Gold, 1987; Kurahashi, 1990; Lowe and Gold, 1991; Firestein et al., 1991; Frings and Lindemann, 1991; Kleene and Gesteland, 1991a; Kramer and Siegelbaum, 1992). The cation influx through the open CNG-channels produces a membrane depolarization, giving rise to action potentials (Firestein and Werblin, 1989; Kurahashi, 1989). In addition, calcium influx through the CNG-channels activates chloride channels that leads to an inward Cl<sup>-</sup> current that further depolarizes the cell (Kleene, 1993; Kurahashi and Yau, 1993; Lowe and Gold, 1993; Reuter et al. 1998).

## *2 Inhibitory responses and mixture suppression*

On the other hand, studies on wide variety of vertebrate species (frog; Duchamp et al., 1974; salamander; Getchell and Shepherd, 1978a, 1978b; catfish; Kang and Caprio, 1995; rat; Duchamp-Viret et al., 1999) have shown that odorants can elicit inhibitory responses as well as excitatory responses in ORNs. These odorant-evoked inhibitory and excitatory responses can be observed even in a single cell (Duchamp et al., 1974; Morales et al., 1994;

Kang and Caprio, 1995), suggesting that ORNs possess both excitatory and inhibitory mechanisms. While the mechanism of the excitatory response is well established, little is known about inhibitory mechanism (reviewed by Getchell, 1986; Ache and Zhainazarov, 1995).

In behavioral (Monkey; Laska and Hudson, 1993; Rat; Laing et al., 1989b; Lobster; Daniel and Derby, 1991) and psychological (Cain, 1975; Laing et al., 1984; Berglund and Olsson, 1993a, 1993b; Laing et al., 1989a) studies, it has been shown that responses to odorants were smaller when the odorants were applied simultaneously than when the odorants were applied individually. The mutual suppression by odorants was shown to take place in individual olfactory receptor neurons (Bell et al., 1987). Thus one of factors proposed to account for this odor suppression in behavioral and psychological level is the mutual suppression by odorants in olfactory receptor neurons (Jinks and Laing, 1999a, 1999b).

A series of olfactory studies on lobster seems to convincingly demonstrate this story (Michel et al., 1991; Michel and Ache, 1992; Fadool and Ache, 1992; reviewed by Ache and Zhainazarov, 1995); lobster olfactory neurons have dual transduction system, inhibitory and excitatory system (Figure 4). Besides, it was shown that the mutual suppression is due to coexistence of the inhibitory and excitatory pathways, which have different odor-specificity. However, knowledge on the mutual suppression in vertebrate olfactory receptor neurons has been limited. Because transduction system of vertebrate olfactory receptor neurons is different from that of invertebrate cells, the lobster model has not been accepted in vertebrate animals (Ache and Zhainazarov, 1995).

In order to explore other mechanisms of mutual suppression, I examined the possibility that odorant suppression of the depolarizing conductance,

which is induced through the excitatory transduction cascade, causes hyperpolarization in newt olfactory receptor neurons. Odorant suppression of the current was first reported by Kurahashi et al. (1994). They showed that responses to double pulses of odorants, indicating that odorants have inhibitory as well as excitatory effects in newt olfactory receptor neurons. In this study, I also observed that simultaneous application of odorants suppressed depolarizing responses to odorant, and further examined the underlying mechanisms and properties of inhibition of depolarization caused by other odorants. The results in this study provide an evidence for a hypothesis that the mutual suppression takes place in ORNs in also vertebrates. The response pattern of olfactory receptor neurons in the olfactory epithelium was formed by inhibitory as well as excitatory effects of odorants, which played a role in mutual suppression in vertebrate (Yamada and Nakatani, in press).