MODELLING THE DYNAMICS OF ODOUR TRANSPORT IN THE OLFACTORY EPITHELIUM

A.J. TAYLOR[1], F. Wulfert[1], O.E. Jensen[2], A. Borysik[1], and D.J. Scott[1]

[1] *School of Biosciences, University of Nottingham, Sutton Bonington Campus, LE12 5RD, UK;*
[2] *School of Mathematical Sciences, University of Nottingham, University Park, Nottingham NG7 2RD, UK*

Abstract

The role of Odour Binding Protein (OBP) in the mammalian olfaction process is not entirely clear. The binding behaviour of odorants with OBP has been studied, mainly over long time periods and under equilibrium conditions. However, little is known about the dynamics of odour transport from the nasal gas phase to the odour receptors (and vice versa). Direct measurement is technically difficult due to the micro scale of the liquid mucus phase and the small amounts of volatiles involved. Using data from the literature, the physico-chemical mechanisms, length scales and timing of odour mass transfer can be inferred and a theoretical mass transfer model of the process built. Hypotheses can then be tested with the model to determine mass transfer behaviour in the system and, from the results, the contribution of the different factors that drive odour uptake from the gas phase to the receptor, can be assessed.

Introduction

OBPs are found in the olfactory mucus layer of many species [1]. The proteins bind a wide range of hydrophobic odour compounds and the mechanism of binding has been studied using techniques like X-ray crystallography data [2] and molecular dynamics [3]. The exact role of OBP in transferring odour from the gas phase to the receptors is the subject of some debate and both passive and active modes have been proposed [3]. The passive mode assumes OBP simply acts as an agent to help hydrophobic aroma molecules solubilise in the mucus phase and access the Olfactory Receptors (ORs). In the active mode, it is hypothesised that OBP binds with an odour ligand and then the complex interacts with the OR. Recently, we proposed a mechanism where OBP serves to maintain the odour signal to the ORs which has the potential benefit of amplifying the signal as well as prolonging its duration [4]. The proposed scheme is shown in (Figure 1).

Experiments to test this hypothesis are difficult to formulate. Most experiments to study OBP-odour binding are based on liquid *in-vitro* systems and are often carried out over long time periods using equilibrium or displacement techniques to obtain information on fundamental biophysical processes like dissociation constants (Kd). Dynamic measurements in our laboratory have provided data on the dynamic competition between odorants, the extent of binding and the time to load and unload OBP, although the length scales were much greater (several hundred μm) than those found *in-vivo* [4]. Direct measurement of odour transport in a system that recreates the length- and time-scales found in the olfactory epithelium is difficult. Mass transfer in the olfactory epithelium occurs across very thin films (several μm thick) and over

very short (about 5 second) time scales. These factors create sensitivity and speed issues for most analytical techniques and there is no readily available approach which might provide useful data for odour molecules. Instead, a simple mathematical model was developed to determine the effect of the different rate constants on the mass transfer of odorants (Figure 1). Existing mass transfer models for odorants in the nose [5, 6] do not include an OBP factor and, thus, a model, relating only to the scheme shown in Figure 1, was developed.

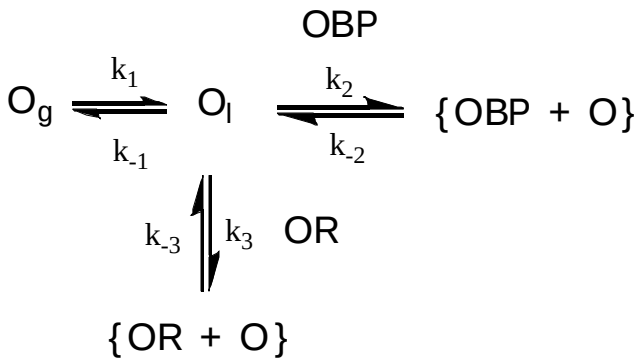

Figure 1. *Scheme showing the transport of odours from the gas phase (O_g) into the liquid phase (O_l) with binding to OR or OBP. The on and off rate constants are denoted by k and –k respectively.*

Experimental

Standard mass transfer equations were used to build a model based on well-established interfacial mass transfer principles [7]. The model was hosted in Matlab and accessed through a user interface to allow values to be input. The key principles were: gas-liquid mass transfer across the interface is driven by partition; mass transfer in the liquid layer is entirely through diffusion, binding to OBP follows the known patterns from *in-vitro* systems, binding to OR is assumed to follow general ligand binding (and release) behaviour. The system was set up so that the rate constants could be input as dimensionless numbers to study the relative influence of each factor.

Results

Effect of air-water partition (K_{gl}) on mass transfer. To test the model, the input values (the k values in Figure 1) were all set to zero except for k_1 and $-k_1$ which were set to give ratios typical of K_{gl} values for odours (10^{-2} to 10^{-5}) and the odour input was set to sinusoidal to mimic the breathing cycle in humans. Figure 2 shows the effect of K_{gl} where values around 10^{-1} produce fluctuations of odour concentration in the liquid layer which are offset from the input sinusoidal gas patterns due to the time taken for mass transfer. As K_{gl} changes towards 10^{-4}, the mass transfer is driven more towards the liquid phase and the odour is rapidly and almost completely removed from the gas phase. The gas phase trace is the lower one in each box and changes from a clear “up and down” sinusoidal trace (K_{gl} 10^{-1}) to a flat line trace (K_{gl} 10^{-4}). Thus partition coefficient is one of the potential factors determining the rate and extent of odour mass transfer as typical values lie between 10^{-2} and 10^{-5}.

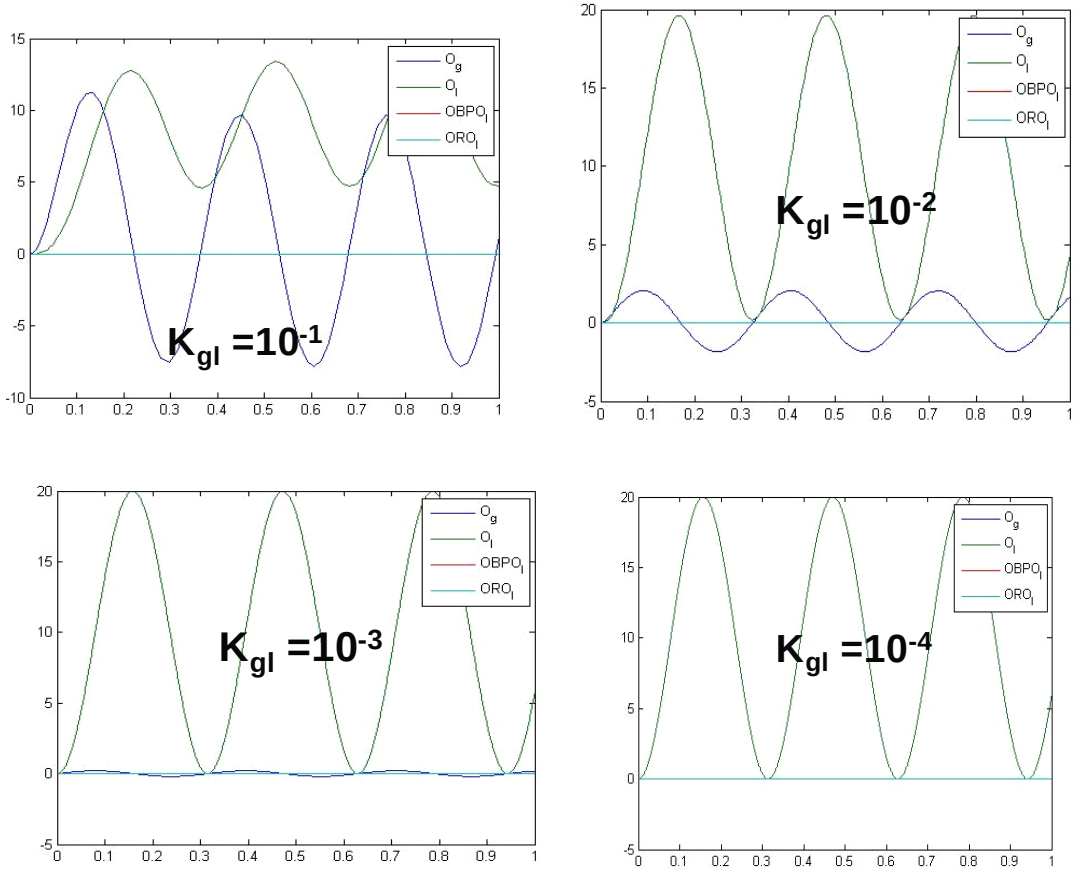

Figure 2. *Odour concentrations in the gas and liquid phases as a function of K_{gl} values. X axis; time, Y axis; concentration, both in arbitrary units.*

Factors favouring prolongation of the odour signal in the liquid phase. The model was then used to determine which of the various rate constants were key in prolonging the concentration of an odour (K_{gl} 10^{-4}) to the ORs. A range of values were input into the model to produce the sort of trace shown in Figure 3, where the change in concentration of OBP complexes and the gas and liquid distribution of odour can be plotted as a function of time. In this case, the values of the rate constants were modified to optimise the concentration of the {OR+O} form as the hypothesis in Figure 1 suggests this is the form which will produce the greatest odour signal. This concept is based on the results of experiments which describe that, prolonging the stimulus to a receptor can amplify the signal output, as well as increasing the signal duration [8, 9]. For example, Firestein [8], demonstrated in salamander, that increasing the duration of an odour stimulus from 0.5 s to 1.2 s not only increased the duration of the output signal from the olfactory system but also increased the signal intensity by a factor of 5. From the mass transfer model, the key factor in prolonging the signal to the ORs was found to be the off rate of odours from OBP (k_{-2}) and the task now is to measure the on and off rates of odours onto OBP experimentally to determine whether the values are related to those found in the theoretical modelling studies. If the model outputs are useful and related to the olfactory events, then a revised version of the model is planned where the values can be input as real concentrations of odours and OBP along with their associated diffusion coefficients, and can be adjusted for temperature and the presence of mucus, to give a more sophisticated model. Data can then be compared with observed data from the literature to determine whether the model is robust.