Numerical modeling of the receptor driven endocytosis

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The present contribution focuses on the receptor driven endocytosis typical of viral entry into a cell. The process is characterized by a local increase in receptor density necessary to establish contact between the cell and the virus. While the receptors of the virus are fixed on its surface, the receptors of the cell are able to move over its membrane, which leads to a local change in their concentration. In the model developed, the receptor motion is described by the diffusion equation along with two boundary conditions. The boundary conditions represent the balance of fluxes at the front of the contact area, where the velocity is assumed to be proportional to the gradient of the chemical potential, and the energy balance behind and before the front, causing the fronts movement. The model provides a basis to incorporate different phenomena such as for example cooperativity. This property strongly influences the effective binder density necessary to establish contact. The moving boundary problem describing the process is numerically solved by using the finite difference method and applied to study the change of receptor density over the membrane as well as the motion of the adhesion front. Two features are investigated in particular. The process initiation is analyzed in order to obtain information on possibilities for inhibiting the viral entry, and the non-dimensional analysis is performed to minimize the number of necessary process parameters and to check their priority.

1 Introduction

Endocytosis is one of the main mechanisms driving cellular uptake of various particles, including viruses. In general, it holds that the density of receptors on the virus is larger than the one on the cell surface and that the virus receptors are fixed, whereas the receptors of the cell are free to move across the membrane. Due to this characteristic, the virus dictates the required receptor density for bonding. Upon contact, receptors on the cell surface diffuse towards the adhesion area to match the density of virus receptors, the rate of bond forming exceeds the rate of bond breaking, and a strong adhesive contact is established. The case with the equal densities defines the chemical equilibrium of the bonding reaction as well as the lower limit for the adhesion to start.

A typical density profile is shown in Fig. 1b and is characterized by the adhesion zone, in which the density is constant and equal to the receptor density on the virus. At the outer edge of the adhesion zone the profile has a sharp interface, dropping to a value below the initial density of cell receptors. When increasing the distance to the adhesion zone the density progressively grows. Since the size of the cell is significantly larger than the virus, the density recovers its initial value far away from the adhesion front.

An important property for the density profile is the front of the adhesion zone where a jump of the receptor density occurs. This jump is caused by the discrepancy between the equilibrium density and the receptor density ahead of the front, which must be lower than in order to stipulate the receptor diffusion. The position of the front changes in time as the adhesion zone grows and is described by the time dependent function \(a(t)\). Other values typical of the adhesion front are denoted by the subscript + in the subsequent text, for example, \(\xi_+\) denotes the receptor density at the front.
The previous explanation shows that the whole process is regulated by the diffusion of receptors over the cell surface and their gathering in the adhesion zone. Accordingly, the motion of the receptors will be described by the diffusion differential equation

\[
\frac{\partial \xi}{\partial t} = -\frac{\partial j}{\partial x},
\]

which states that the change of receptor density in time has to be equal to the negative spatial change in the flux. Furthermore, following Fick’s first law, the receptor flux \( j \) is proportional to the gradient of density, i.e. \( j = -m \frac{\partial \xi}{\partial x} \). The use of this expression, Eq. (1) leads to the alternative expression of the diffusion equation

\[
\frac{\partial \xi}{\partial t} = m \frac{\partial^2 \xi}{\partial x^2}.
\]

This equation defines the relation between the temporal and the spatial changes of the receptor density weighted by the mobility parameter \( m \). Its evaluation gives insight into the evolution of receptor density for every point in front of the adhesion zone \( a(t) < x < \infty \). Equation (2) is a partial differential equation of second order and requires additional boundary conditions in order to determine the complete particular solution.

The first boundary condition describes the flux of receptors through the adhesive front. Following the Leibniz integration rule of the global form and employing Fick’s first law at the front, this condition is derived from Eq. (1) as

\[
(\xi_{eq} - \xi_+) v_+ = m \left[ \frac{\partial \xi}{\partial x} \right]_{+} = 0.
\]

Here, the first term denotes the amount of receptors required for the advancement of the front, and the second term denotes the amount of receptors provided by the flux [3].

The second condition describes the motion of the front to be caused by the energetic difference ahead and behind the front, each consisting of several contributions, in the form [1]

\[
\left[ -\xi_{eq} C_b + \xi_{eq} \ln \left( \frac{\xi_{eq}}{\xi_0} \right) + \frac{1}{2} B kT^2 \right] + \xi_+ \ln \left( \frac{\xi_+}{\xi_0} \right) + \frac{1}{2} m_r m_{rr} \frac{m}{k T} \frac{1}{\xi_+} \left( \frac{\partial \xi_+}{\partial x} \right)^2 = \frac{1}{2} \xi_{eq} m_r kT v_+^2.
\]

This condition relies on the assumption that the entropy energy of receptors can be expressed in analogy to ideal gas, that the bending energy is a quadratic form of curvature \( \kappa \) and that the binding energy is the linear function of density with the proportionality constant \( C_b \). The following notation applies as well: \( B \) is the bending stiffness, \( m_r \) and \( m_{rr} \) are the receptor mass and the mass of the receptor couple respectively, \( k \) is the Boltzmann constant and \( T \) the absolute temperature. The described moving boundary problem is transferred into a system of nonlinear equations by using the finite difference method and subsequently solved by using the Newton-Raphson scheme.

### 3 Cooperativity

The energetic description presented provides an efficient and easy to adapt interface for incorporating additional phenomena. This is either achieved by adding a respective term describing the energy contribution or by adapting affected process parameters. An important phenomenon to be incorporated is cooperativity. As soon as receptors of two opposing surfaces create bonds they smooth out the membranes such that the latter become more conform to each other. This makes it easier for additional receptors to create bonds and strengthens the adhesion between the two surfaces [4]. The cooperativity is characterized...
by a reduction of required receptor bonds in order to connect the virus with the cell. The new density is calculated according to
\[
\xi_{\text{eq,req}} = \frac{\kappa_B}{kT} \frac{\ell^2}{w_{\text{we}}} K^2_{\text{pl}} \xi_{\text{eq}}^2 \xi^2,
\]
where \(\kappa_B\) is the effective rigidity, \(c\) is a dimensionless prefactor, \(w_{\text{we}}\) is the interaction range between two opposing receptors and \(K_{\text{pl}}\) is the two dimensional equilibrium constant.

4 Non-dimensionalization

In some physical systems, non-dimensionalization is applied to suggest that it is more convenient to measure certain quantities relative to an appropriate reference quantity. The present model for the viral entry includes the following dimensional quantities which can be expressed in terms of their non-dimensional counterparts
\[
x = l_x x', \quad t = \tau_s t', \quad \xi = \xi_s \xi'.
\]
Here, \(l_x, \tau_s, \xi_s\) are properly chosen scaling parameters for space, time and density, and the prime symbol denotes the dimensional quantities. Furthermore, introducing the abbreviations \(\xi'_+ = \frac{\xi_c}{\xi}, \xi'_- = \frac{\xi_b}{\xi}\) and \(C'_1 = -\xi'_{\text{eq}} C_b + \xi'_{\text{eq}} \ln \xi'_{\text{eq}} + \frac{1}{2} B \xi'_+^2\), the non-dimensional boundary problem can be represented as
\[
\frac{\partial \xi'}{\partial t'} = \bar{m} \frac{\partial^2 \xi}{\partial x'^2}, \quad (\xi'_{\text{eq}} - \xi'_{\text{eq}}) \xi'_{+} - \bar{m} \left[ \frac{\partial \xi'}{\partial x'} \right]_+ = 0, \quad C'_1 = \xi'_{\text{eq}} \ln \xi'_{\text{eq}} - \bar{m} \xi'_+ \left[ \frac{\partial \xi'}{\partial x'} \right]^2 = \bar{m}_{\text{rr}} \xi'_+^2,
\]
where \(\bar{m} = \frac{m_0}{\tau_s^2}\) is the dimensionless mobility, \(\bar{m}_r = \frac{1}{2} \frac{m_0}{\xi^2} m^2_{\text{rr}}\) the non-dimensional cell receptor mass and \(\bar{m}_{\text{rr}} = \frac{1}{2} \frac{m_0}{\xi^2} m^2_{\text{rr}}\) the non-dimensional mass of a receptor couple. Simulations with a focus on the front velocity and on the front density reveal an important influence from the dimensionless mobility \(\bar{m}\) on the velocity, whereas the dimensionless mass \(\bar{m}_r\) mainly influences the receptor density. The variation of the dimensionless equilibrium density \(\xi'_{\text{eq}}\) as well as \(C'_1\) influence both quantities in a similar way, whereas the variation of the dimensionless couple mass \(\bar{m}_{\text{rr}}\) hardly affects the results.

5 Process initiation

The virus radius gives important information on the process, especially with regard to its initiation. Since Eq. (4) provides a relation for the kinetic energy, it directly follows that the difference between the energy behind and ahead of the front has to be non-negative. In the beginning of the process, the density distribution is still uniform and the front has not yet been established. In this case, the energy ahead of the front does not contribute to the total amount, while the energy behind the front can be interpreted as an initial barrier that must be overcome in order to start the process. A study of the limiting case, where the front velocity approaches to zero, yields the expression for the maximal radius
\[
R_{\text{max}} = \sqrt{\frac{B}{2}} \left/ \sqrt{\xi_{\text{eq}} B_{\text{c}} - \xi_{\text{eq}} \ln \left( \frac{\xi_{\text{eq}}}{\xi_0} \right), \quad C_b - \ln \left( \frac{\xi_{\text{eq}}}{\xi_0} \right) > 0.}
\]
The maximum radius strongly depends on the receptor density ratio, which is limited from both sides. Its upper limit is given by \(\frac{\xi_{\text{eq}}}{\xi_0} \leq 1\) since \(\xi_0 \leq \xi_{\text{eq}}\) and its lower limit by \(\frac{\xi_{\text{eq}}}{\xi_0} \geq e^{-C_B}\) as imposed by Eq. (8)b. Numerical simulations show a rapid increase in the maximum radius as the density ratio approaches its lower limit. The latter is also known as the critical density value for that reason.

6 Numerical results

The model presented offers a basis for a variety of different setups to be investigated. It currently considers spherical as well as helical virus geometries. The first simulation presented here investigates the performance of the uptake process. Special attention is paid to the density profile on the cell and to the current state of the viral uptake. A helical virus of size \(D = 0.05\) \(\mu\text{m}\) is assumed to come into contact with a much larger cell, such that the cell curvature is negligible. Due to the axial symmetry of the virus, the problem is treated in a 1D representation which assumes the unit width of the active domain. The initial density of cell receptors is set to \(\xi_0 = 1000 \text{ \(\mu\text{m}^{-2}\)}\), whereas the initial density of virus receptors is set to \(\xi_{\text{eq}} = 4800 \text{ \(\mu\text{m}^{-2}\)}\). Time increment \(\Delta t = 1e^{-4}\) s and space increment \(\Delta x = 1e^{-3}\) \(\mu\text{m}\) are used for the numerical simulations. Figure 2 shows the density profile at five stages throughout the simulation until the virus has almost fully entered the cell. The diagrams show a rapid reduction in the receptor density at the front at the beginning of the process. This rapid decline in density at the front slows down in the course of the further process. The \(x\)-distance determines the contact area between virus and cell. Since the virus geometry is known, monitoring the \(x\)-distance of the adhesion front on the cell surface enables the tracking of the current progress of the uptake process.
The model proposed has shown that there are certain limit cases which have to be overcome in order to initiate the process. Figure 3 shows the change of the process duration for combinations of two parameters. First, the interaction between the initial cell density $\xi_0$ and the mobility $m$ is analyzed (Fig. 3a). For combinations with values below a certain threshold the process does not start. However, once this threshold is surpassed, an increase in either of these parameters result in a strong decrease of the required time. Especially the area with small values for the mobility is interesting. Multiple viruses are known to bind to cell receptors with small mobilities, such as the HIV-virus connecting to receptors with mobility $m = 0.05 \mu m^2/s$ or the Semliki Forest virus connecting to a receptor with $m = 0.01 \mu m^2/s$. The combinations for mobility $m$ and binding range $l_{\text{we}}$, crucial for cooperativity, is shown in second plot (Fig. 3b).

Figure 3: (a) Process duration depending on mobility $m$ and the initial receptor density $\xi_0$. (b) Process duration depending on mobility $m$ and the binding range $l_{\text{we}}$. Plots also show the admissibility ranges for chosen parameter sets.

7 Conclusion

The present study focuses on the simulation of the viral entry into a cell via receptor driven endocytosis. The model proposed includes the diffusion differential equation in combination with two boundary conditions describing the flux balance and the energy balance at the adhesion front. The underlying formulation allows for an efficient extension of the approach to simulate additional phenomena such as cooperativity. Multiple setups are considered providing insight into the performance of the viral uptake as well as its limiting behavior. Furthermore, the non-dimensional formulation allows for physical interpretations of several process parameters. An extension to a three dimensional setup for the simulation of viruses with no axial symmetry or with non-homogeneous receptor densities is envisaged as a topic of future work.

Acknowledgements We gratefully acknowledge the financial support by the German Research Foundation (DFG) for the scientific project KL 2678/7-1. We furthermore acknowledge support by the DFG and the Open Access Publication Fund of TU Berlin. Open access funding enabled and organized by Projekt DEAL.

References

[1] T. Wiegold, S. Klinge, R. P. Gilbert and G. A. Holzapfel, Comput. Math. Appl., 84, 224-243 (2021), DOI: 10.1016/j.camwa.2020.12.012
[2] H. Gao, W. Shi and L. B. Freund, Proc. Natl. Acad. Sci. USA 102(27), 9469–9474, (2005)
[3] L. B. Freund and Y. Lin, J Mech Phys Solid 52, 2455-2472 (2004)
[4] H. Krobath, B. Różycki, R. Lipowsky and T. R. Weikl, Soft Matter, 5, 3354-3361 (2009)