Liquids that form due to dynamics of the molecules that depend on the local density

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RNA molecules in living cells form what look like liquid droplets formed by liquid/liquid phase separation. But unlike the molecules in conventional phase separating mixtures, RNA molecules are transported by molecular motors that consume energy and so are out of equilibrium. Motivated by this we consider what sort of simple rules for the dynamics of model mRNA molecules lead to liquid/liquid phase separation. We find that dynamics that slow as the local density of molecules increases, drive the formation of liquids. We also look at the analogous separation of the two blocks of a block copolymer, in which the monomers of one block have dynamics that depend on the local density of monomers of that block. We find that this block condenses and separates from the monomers of the other block. This is a simple model of the out-of-equilibrium domain formation found in the chromatin in the nucleus of cells.

The contents of living cells are in the liquid state. But these contents are not in one liquid phase, they appear to be in a number of coexisting liquid phases. There are what look like liquid droplets in both the cytoplasm and the nucleus. For example, in the cytoplasm mRNA molecules can undergo what looks like liquid/liquid phase separation, to produce droplets enriched in the mRNA molecules. But these droplets cannot be at thermodynamic equilibrium. They are affected when the molecular motors dynein or kinesin are knocked down. These motors consume energy and actively move mRNA molecules. This suggests that mRNA molecules do not simply diffuse into and out of these liquid droplets, they are actively transported into or out of these droplets.

Inspired by this, we wanted a simple model of density-dependent dynamics that generates condensation. These dynamics should be via hops from one point to another; the hops do not conserve momentum, and there is no well defined velocity. This is our simple model of a motor translating mRNA molecules along microtubules in a cell. It turns out that very simple models show condensation. Rather generically, liquids appear whenever the dynamics of the molecules that depend on the local density of molecules increases. These out-of-equilibrium dynamics are illustrated in a schematic in Fig. 1. The dynamics stabilise liquid droplets as evaporation of molecules from a droplet’s surface is reduced by the slow hopping rate of molecules out of the dense liquid. Attractions slow hopping rates but any non-equilibrium mechanism that also slows hopping out of dense regions will tend to have a similar effect, whether or not that are any attraction energies directly involved. The Edinburgh group and others have extensively studied a system with rather different microscopic dynamics; in their systems the particles have a well-defined velocity. However, despite the differences in the microscopic dynamics, here we are studying models in which the mobility decreases as density increases, just as they have, and so we see qualitatively very similar condensation into liquid-like droplets.

\[ \Delta n \leq 0 \] the move is always accepted as then the energy decrease or stays the same, but if \( \Delta n < 0 \) the move is rejected with probability \( \exp[\epsilon \Delta n / kT] \). Here \( \epsilon \) is an interaction energy.

When the ratio \( \epsilon / kT > 1.7 \) a liquid phase forms. The attractions between the molecules cause them to condense into a liquid phase. Droplets of liquid nucleate and grow until there is a single large droplet coexisting with a vapour.
We now introduce a simple model of dynamics where the rate a molecule hops at depends on the local environment at the start position of its move. At first sight these dynamics may look as though they violate detailed balance, but we will show that they do not. The dynamics are as follows. Select a lattice site at random. If it is occupied by a molecule, attempt to move it to a randomly selected site within a radius \( r_M \). If the chosen site is already occupied the attempt is always rejected. If the chosen site is empty the move is made with probability \( p = \exp[-\alpha_D n_n] \), for \( n_n \) the number of neighbours of the molecule in its starting position. Here \( \alpha_D > 0 \) is a parameter that couples the dynamics to the local density of molecules. In this model the higher the local density of molecules the slower the hopping rate of a molecule. This could be for a number of reasons, such as neighbouring molecules inhibiting motor transport via mechanisms such as direct binding or signalling. Alternatively, even if motors are not involved, then a local density-dependent slow down could be caused by an out-of-equilibrium process such as a phosphorylation/dephosphorylation cycle that modulates binding and so diffusion rates.

Our density-dependent dynamics map onto the standard Ising lattice gas, as can be seen as follows. To do this we need to consider the transition probability between state \( i \) and state \( j \), \( p_{ij} \); these states \( i \) and \( j \) correspond to two positions of a molecule on the lattice. For transition between a state \( i \) with a molecule surrounded by \( n_n(i) \) neighbours and a state \( j \) where the molecule has \( n_n(j) \) neighbours, the ratio of the transition probabilities \( i \) to \( j \), and \( j \) to \( i \) are

\[
\frac{p_{ij}}{p_{ji}} = \frac{\exp[-\alpha_D n_n(i)]}{\exp[-\alpha_D n_n(j)]} = \exp[-\alpha_D(n_n(i) - n_n(j))] \quad (1)
\]

These satisfy detailed balance, and map to the Ising lattice gas with \( \epsilon/kT = \alpha_D \). Thus, for example, our model has the usual Ising critical point at \( \alpha_D = 1.76 \), and a reduced surface tension \( \gamma' = \gamma/kT \) given by Onsager’s expression \( 25 \) but with \( \alpha_D \) replacing \( \epsilon/kT \):

\[
\gamma' = \alpha_D/2 - \ln[1 + \exp(-\alpha_D/2)]/1 - \exp(-\alpha_D/2).\]

The result of a simulation run at \( \alpha_D = 2.5 \) is shown in Fig. 2. We see that, as it must, it shows the usual Ising lattice gas vapour/liquid coexistence. We fix \( r_M = 8 \), as varying it over a wide range of values (2 to 24) is found to have little effect on the dynamics, and cannot change the phase behaviour as varying \( r_M \) does not alter the mapping to the Ising lattice gas.

In terms of studying vapour/liquid coexistence in energy-consuming systems, the studies closest to this work are those of the Edinburgh group \( 19\) who model active Brownian particles with motilities that depend on the local density of these particles, but no attractions. Redner et al. \( 15 \) and Mognietti et al. \( 20 \) also study systems with local slow down in the dynamics that show condensation. In the Edinburgh group’s work, the particles condense into a liquid phase coexisting with a dilute phase. Our models behave in a qualitatively identical way, the fact that for the model here high densities decrease a hopping rate whereas it decreases a velocity in the model studied in Edinburgh appears to make little difference.

However, our motivation is different. The Edinburgh group focus on showing that a specific out-of-equilibrium model shows equilibrium-like vapour-liquid phase separation, whereas we are interested in constructing the simplest possible models model the liquid droplets inside cells. There are also similarities between our model, the models studied by the Edinburgh group and others, and cooling granular media. In cooling granular media locally high rates of inelastic collisions in dense regions slow the particles in these dense regions, which in turn causes clustering that is partly analogous to condensation into a liquid-like phase. See Paul and Dill \( 20 \) and references therein for recent work in this area. However, there are differences, these clusters are not at steady state and there may well not be a well-defined surface tension there \( 20 \).

mRNA-rich droplets in the cytoplasm of cells are not the only example of self-organised domains in cells. There is a huge amount of self-organised structure in the chromatin in the nuclei of cells. Chromatin is the DNA of our genes, together with associated proteins and RNA \( 24,14,44,11 \). Examples of this structure are domains called nucleoli where ribosomes are made \( 21,11 \). Cajal bodies where RNA splicing occurs \( 21,10 \), and what is effectively microphase separation between chromatin that is being transcribed and chromatin that is not active \( 2,10,12 \).

So chromatin, which includes huge polymers of DNA, exhibits behaviour reminiscent of the microphase separation seen in block copolymers with immiscible blocks \( 14 \). The nucleus is microphase separated in the sense that lengths of chromatin that have a common feature, e.g., are making ribosomal RNA in the case of nucleoli, have separated out from the rest of the DNA. However, the dynamics of the chromatin depends on energy consuming processes \( 23,24 \), unlike in block copolymers where the monomers move via thermal diffusion. In conventional block copolymers the microphase separation is driven by intermolecular attractions between like monomers being stronger than between unlike monomers. The mechanism may be different for chromatin.
Nuclear self-organisation is a very active field of research \cite{24,13,31} and we cannot answer all the many unsolved questions here. However we can see if density-dependent dynamics of the monomers of a block copolymer, can result in microphase separation. This would be a very simple model of separation between active and inactive chromatin, or between a nucleolus and the surrounding chromatin that is not involved in the making of ribosomes.

We introduce a simple out-of-equilibrium block copolymer model. The polymer consists of a linear chain of \( M \) monomers on a 2D square lattice. The monomers are held together by bonds. If the \( x \) and \( y \) coordinates of monomer \( i \) are \( x_i \) and \( y_i \) then the bonds along the backbone are enforced by insisting that \( |x_{i+1} - x_i| \leq 1 \) and \( |y_{i+1} - y_i| \leq 1 \) for \( i = 1, M - 1 \). The monomers are of two types: Type E which just interact via excluded volume interactions, and type D which in addition have dynamics that depend on the local density.

A simulation cycle consists of \( M \) attempted moves, in each of which one of the \( M \) monomers is selected and an attempt is made to move it to one of the eight neighbouring lattice sites. These eight are the sites above, above right, right, below right, etc., of the central site. If a type E monomer has been selected, that move is always successful unless it would move the monomer to an already occupied site, in which case it is always rejected. If a type D monomer is selected then the move is also rejected if it would move the monomer to an already occupied site. But if the site is vacant, the move is only made with a probability \( p = \exp(-\alpha_{DP} n_{MN}) \). Here \( n_{MN} \) is the total number of monomers (excluding the monomers it is bonded to) in the eight lattice sites that surround the site the molecule would be moved from. The parameter \( \alpha_{DP} \) controls the density dependence of the dynamics. The polymer is simulated on a large lattice; periodic boundary conditions are not used. Note that this model for density dependent dynamics also satisfies detailed balance, for the same reason as for our monomeric model. The model has hard constraints, both restricting the molecules to no more than one per site and restricting the bond length between successive monomers.

For two states \( i \) and \( j \) that are allowed as they satisfy these requirements the transition probabilities satisfy \( p_{ij}/p_{ji} = \exp[-\alpha_{DP} (n_{MN}(i) - n_{MN}(j))] \), and so satisfy detailed balance.

For a block copolymer of the two types of monomer, the two halves of the polymer separate, with the type D monomers forming a condensed globule, while the type E monomers form an extended chain. A snapshot of this is shown in Fig. 3.

Note that our model for microphase separation is different to Ganai et al.\cite{12}’s model for separation of chromatin that is being transcribed from chromatin that is not being transcribed. In their work domain formation is driven by a large increase in effective temperature in chromatin that is being transcribed, without a direct mechanism for density-dependence of the dynamics. Within our model, transcriptionally active regions of chromatin separate if the active dynamics of these regions are such that their motion slows when in contact with other active regions. Similarly, applied to a nucleolus, our model predicts that the nucleolus separates from the surrounding chromatin due to ribosome-producing chromatin slowing its mesoscale motion in the presence of other ribosome-producing chromatin — perhaps because it is sharing factors or due to direct attractive interactions.

We have studied very simple models and found that molecules whose hopping rate decreases strongly when the local density is high, condense into liquid droplets. The simplicity of our model, and the fact that condensation is also seen in very different models in which the velocity decreases when the local density is high\cite{14,15,19,21}, suggests that this condensation is quite generic. The model is an equilibrium one in the sense that it obeys detailed balance, but is for out-of-equilibrium systems that consume energy. Thus to the extent that it models the microscopic dynamics of mRNA molecules in cells correctly, their behaviour will be qualitatively that of an equilibrium liquid. Although the model maps to the Ising lattice gas, an equilibrium model, the energy driving the dynamics does not have to be \( kT \). For mRNA molecules in cells, a better candidate, at least for long distance motion,\cite{15} is the much larger forces that molecular motors can provide. This has consequences. For example the effective surface tension that limits the fluctuations of the liquid surface is then not \( \sim kT/\mu^2 \), it is presumably \( \sim w_M/\mu^2 \), with \( w_M \) the work done by one or a few motors on one of the molecules of the liquid. Here \( \mu \) is a molecular diameter.

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