Aging of RNA-chaperone condensates

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1 Introduction

We study the aging of RNA + DEAD-box chaperone condensates. RNA strands are modeled as Gaussian polymers, and DEAD-box proteins are point-like crosslinkers similar to the one in this article [1]. The principle of this model is that the crosslinkers fix the position of the polymers at certain points. The more freedom a polymer has, the easier it is for him to find new cross-linkers. However, the harder it will be to find new cross-linkers. We focus on the binding and unbinding of a single polymer strand bound at its extremities, hereafter called a loop. Depending on the number of cross-linkers in the vicinity of the loop, it can bind to one of them. Each binding or unbinding event leads to a reorganization of the number of crosslinkers in the vicinity. In these notes, we start with a rigorous derivation of the transition between an unbound to a bound state. We then simplify the problem and explain how we will simulate it. Finally, we will derive the equilibrium state from a very simplified version of the model.

2 Rigorous derivation

We consider the transition from the state represented in Fig. 2. We define:

$$\begin{aligned} &\mathcal{C} = \{\mathbf{R}_0, \mathbf{R}_1, \{\mathbf{r}_i\}_{i\in[0,k_0]}, \ell_0\} \\ &\mathcal{C}' = \{\mathbf{R}'_0, \mathbf{R}'_1, \mathbf{R}'_2, \{\mathbf{r0}'_i\}_{i\in[0,k'_0]}, \{\mathbf{r1}'_i\}_{i\in[0,k'_1]}, \ell'_0, \ell'_1\} \end{aligned} \tag{1}$$

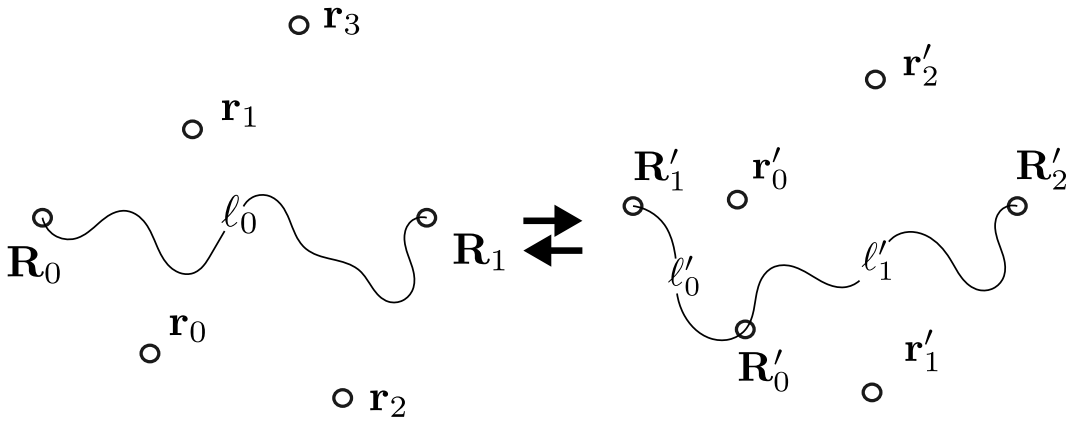
We write the equilibrium probability of being in $\mathcal{C}'$:

$$P(\mathcal{C}') = \frac{\Omega(\mathbf{R}'_0, \mathbf{R}'_1, \ell'_0)\Omega(\mathbf{R}'_1, \mathbf{R}'_2, \ell'_1)e^{\beta E_b}}{\mathcal{Z}} \tag{2}$$

Where $\Omega(\mathbf{R}_i, \mathbf{R}_j, \ell)$ is the number of polymer conformations for a loop bound in $\mathbf{R}_i$ and in $\mathbf{R}_j$ of length ℓ. $\beta = 1/(k_B T)$ and $E_b > 0$ is the binding trap energy. $\mathcal{Z}$ is the partition function of the system defined as:

$$\mathcal{Z} = \Omega(\mathbf{R}_0, \mathbf{R}_1, \ell_0) + \sum_{i=0}^{k} \int \frac{\mathrm{d}\ell}{b} \Omega(\mathbf{R}_0, \mathbf{r}_i, \ell)\Omega(\mathbf{r}_i, \mathbf{R}_1, \ell_0 - \ell)e^{\beta E_b} \tag{3}$$

where b is the characteristic size of a monomer. The sum runs across all crosslinkers, and the integral runs along the polymer.



We can then write the total probability of being in a bound state (not only the $\mathcal{C}'$ one):

$$p_{\text{bound}}(\mathcal{C}) = \frac{\sum_{i=0}^{k} \int \frac{\mathrm{d}\ell}{b} \Omega(\mathbf{R}_0, \mathbf{r}_i, \ell) \Omega(\mathbf{r}_i, \mathbf{R}_1, \ell_0 - \ell) e^{\beta E_b}}{\Omega(\mathbf{R}_0, \mathbf{R}_1, \ell_0) + \sum_{i=0}^{k} \int \frac{\mathrm{d}\ell}{b} \Omega(\mathbf{R}_0, \mathbf{r}_i, \ell) \Omega(\mathbf{r}_i, \mathbf{R}_1, \ell_0 - \ell) e^{\beta E_b}} \tag{4}$$

We define the probability that the polymer will find a cross-linker located in $\mathbf{r}$:

$$p_{\text{meet}}(\mathbf{r}, \mathcal{C}) = \frac{\int \frac{\mathrm{d}\ell}{b} \Omega(\mathbf{R}_0, \mathbf{r}, \ell) \Omega(\mathbf{r}, \mathbf{R}_1, \ell_0 - \ell) e^{\beta E_b}}{\Omega(\mathbf{R}_0, \mathbf{R}_1, \ell_0)} \tag{5}$$

This gives for the probability of being bound or unbound:

$$\begin{cases} p_{\text{bound}}(\mathcal{C}) = \dfrac{\sum_{i=0}^{k} p_{\text{meet}}(\mathbf{r}_i, \mathcal{C}) e^{\beta E_b}}{1 + \sum_{i=0}^{k} p_{\text{meet}}(\mathbf{r}_i, \mathcal{C}) e^{\beta E_b}} \\ p_{\text{unbound}}(\mathcal{C}) = \dfrac{1}{1 + \sum_{i=0}^{k} p_{\text{meet}}(\mathbf{r}_i, \mathcal{C}) e^{\beta E_b}} \end{cases} \tag{6}$$

The detailed balance condition gives : $\frac{p_{\text{bound}}}{p_{\text{unbound}}} = \frac{r_{\text{bound}}}{r_{\text{unbound}}}$, which leads to :

$$\begin{cases} r_{\text{bound}}(\mathcal{C}) = \dfrac{\sum_{i=0}^{k} p_{\text{meet}}(\mathbf{r}_i, \mathcal{C})}{\tau_0} \\ r_{\text{unbound}}(\mathcal{C}) = \dfrac{e^{\beta E_b}}{\tau_0} \end{cases} \tag{7}$$

3 Adding rearrangement of cross-linkers

All binding/unbinding events now trigger a rearrangement of the cross-linkers around the loop. The crosslinkers are assumed to be spread throughout the

system with a uniform distribution; thus the probability of having k crosslinkers in the vicinity of a loop is given by a Poisson distribution.

$$P(k) = e^{-\rho V} \frac{(\rho V)^k}{k!} \tag{8}$$

Where ρ is the average number of crosslinkers per unit volume in the system and $V(\mathbf{R}_0, \mathbf{R}_1, \ell_0)$ is the volume accessible by the loop.

We now consider transitions between two ensembles of states, the unbound states defined as :

$$U = \{\mathbf{R}_0, \mathbf{R}_1, \ell_0, \{\{k\}_{\in\mathbb{N}}, \{\mathbf{r}_i\}_{i\in[0,k]}\}\}, \tag{9}$$

where $\mathbf{R}_0, \mathbf{R}_1, \ell_0$ are fixed and $\{\{k\}_{\in\mathbb{N}}, \{\mathbf{r}_i\}_{i\in[0,k]}\}$ correspond to the ensemble of possible values of k and the corresponding position of the k cross-linkers. Similarly, we define the ensemble of bound state:

$$B = \{\mathbf{R}_0, \mathbf{R}_1, \{\ell, \ell_0 - \ell, \mathbf{r}, \{k'_0\}_{\in\mathbb{N}}, \{k'_1\}_{\in\mathbb{N}}, \{\mathbf{r0}_i\}_{i\in[0,k'_0]}\}, \{\mathbf{r1}_i\}_{i\in[0,k'_1]}\}\}, \tag{10}$$

where $\mathbf{R}_0$, $\mathbf{R}_1$ and ℓ_0 are still fixed, but the polymer could be bound to any crosslinker located in $\mathbf{r}$, thus creating a loop of any length ℓ, (the length of the other loop is therefore fixed to $\ell_0 - \ell$). Finally, we consider any cross-linker environment possible for each loop.

We define the probability that a loop between $\mathbf{R}_0$ and $\mathbf{R}_1$ of total length ℓ_0 is in a state that belong to B

$$p_B(\mathbf{R}_0, \mathbf{R}_1, \ell_0) = \frac{1}{\mathcal{Z}} \int \mathrm{d}^3\mathbf{r} \sum_{k=0}^{\infty} P(k) \underbrace{kP(\mathbf{r})}_{\text{proba of having a } \times \text{ in } \mathbf{r}\text{, with k trial}} \int_0^{\ell_0} \frac{\mathrm{d}\ell}{b} \Omega(\mathbf{R}_0, \mathbf{r}, \ell)\Omega(\mathbf{r}, \mathbf{R}_1, \ell_0 - \ell) e^{\beta E_b} \tag{11}$$

with

$$\mathcal{Z} = \Omega(\mathbf{R}_0, \mathbf{R}_1, \ell_0) + \int \mathrm{d}^3\mathbf{r} \sum_{k=0}^{\infty} P(k) \underbrace{kP(\mathbf{r})}_{\text{proba of having a } \times \text{ in } \mathbf{r}\text{, with k trial}} \int_0^{\ell_0} \frac{\mathrm{d}\ell}{b} \Omega(\mathbf{R}_0, \mathbf{r}, \ell)\Omega(\mathbf{r}, \mathbf{R}_1, \ell_0 - \ell) e^{\beta E_b} \tag{12}$$

Similarly as before, we define the probability that the polymer meets a crosslinker in $\mathbf{r}$:

$$p_{\text{meet}}(\mathbf{R}_0, \mathbf{R}_1, \ell_0, \mathbf{r}) = \frac{\int_0^{\ell_0} \frac{\mathrm{d}\ell}{b} \Omega(\mathbf{R}_0, \mathbf{r}, \ell)\Omega(\mathbf{r}, \mathbf{R}_1, \ell_0 - \ell)}{\Omega(\mathbf{R}_0, \mathbf{R}_1, \ell_0)} \tag{13}$$

Therefore we can re-write the probability of being bound, using the abreviation : $(\mathbf{R}_0, \mathbf{R}_1, \ell_0) = \mathcal{C}$. And using the property : $\sum_{k=0}^{\infty} kP(k) = \rho V$, and $P(\mathbf{r}) = 1/V$

$$p_B(\mathcal{C}) = \frac{\rho \int \mathrm{d}^3\mathbf{r} p_{\text{meet}}(\mathcal{C}, \mathbf{r}) e^{\beta E_b}}{1 + \rho \int \mathrm{d}^3\mathbf{r} p_{\text{meet}}(\mathcal{C}, \mathbf{r}) e^{\beta E_b}} \tag{14}$$

3.1 Gaz polymer

To compute p_{meet} we assimilate the polymer to a gaz of particles of size b into a volume $V(\mathbf{R}_0, \mathbf{R}_1, \ell_0)$. There are ℓ_0/b monomers in the polymer, and the volume is given by:

$$V(\mathbf{R}_0, \mathbf{R}_1, \ell_0) = \frac{\pi}{6}\ell_0((\mathbf{R}_1 - \mathbf{R}_0)^2 + \ell_0^2) \tag{15}$$

if a cross-linker is at a position $\mathbf{r}$ in V, the probability that each monomer is at $\mathbf{r}$ is also $1/V$, in total:

$$p_{\text{meet}}(\mathcal{C}, \mathbf{r}) = \ell_0/b(a^3/V(\mathcal{C})) = \frac{6}{\pi b}\frac{a^3}{((\mathbf{R}_1 - \mathbf{R}_0)^2 + \ell_0^2)} \tag{16}$$

Where a is the typical size of a crosslinker Using this expression in eq.14 gives :

$$\begin{cases} p_B = \dfrac{\rho a^3 \ell_0/b e^{\beta E_b}}{1 + \rho a^3 \ell_0/b e^{\beta E_b}} \\ p_U = \dfrac{1}{1 + \rho a^3 \ell_0/b e^{\beta E_b}} \end{cases} \tag{17}$$

The detailed balance condition gives : $P_U/P_B = r_{b\to u}/r_{u\to b}$, we choose:

$$\begin{cases} r_{b\to u} = \dfrac{e^{-\beta E_b}}{\tau_0} \\ r_{u\to b} = \dfrac{\rho a^3 \ell_0/b}{\tau_0} \end{cases} \tag{18}$$

One concern remains : the transition between a bound state and an unbound state with no crosslinkers around the loop. There would be no possibility to come back from this unbinding event directly. This can play the role of an absorbing boundary condition that could be very useful.

3.2 No average over k

We now consider a process where k changes each time a binding / unbinding event occurs. Notice that this process do not satisfy detailed balance. Indeed, the unbound situation with 0 surrounding k can be reached from a bounded situation with non-zero k_0', k_1', however the reverse binding event is impossible. The unbound case with 0 bonds is an absorbing state, thus the equilibrium distribution is this unbound state with 0 surrounding cross-linkers. Our process obeys the same transition rates, on top of which we add the probability of having k cross-linkers:

$$\begin{cases} r_{u\to b}(k, k_0', k_1') = \dfrac{k p_{\text{meet}}(\mathcal{C})}{\tau_0} P(k)P(k_0')P(k_1') \\ r_{b\to u}(k, k_0', k_1') = \dfrac{e^{\beta E_b}}{\tau_0} P(k)P(k_0')P(k_1') \end{cases} \tag{19}$$

We notice that we recover Eq. 18 when summing the rates over k,k_0',k_1'.

3.2.1 Polymer gas

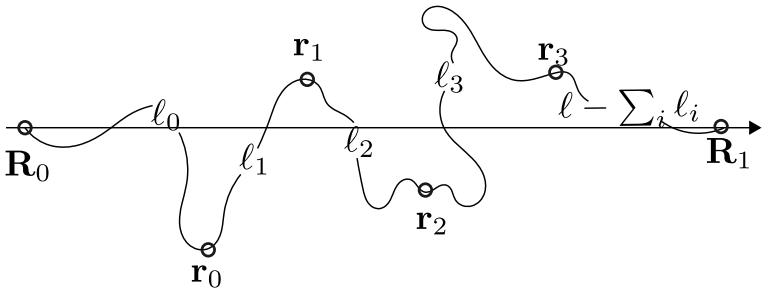
In the polymer gas model, we have according to Eq. 16 $p_{\text{meet}} = a^3/V\ell/b$ which leads to :

$$\begin{cases} p_{\mathcal{U}}(k) = \dfrac{(1 - k\ell/ba^3/V)P(k)}{(1 - \rho a^3\ell/b) + a^3\rho\ell/be^{\beta E_b}} \\ p_{\mathcal{B}}(k'_0, k'_1) = \dfrac{e^{\beta E_b}\ell/b\rho a^3 P(k'_0)P(k'_1)}{(1 - \rho a^3\ell/b) + a^3\rho\ell/be^{\beta E_b}} \end{cases} \tag{20}$$

4 interlude reminding of the plan

So far we focused on a single polymer loop. The point of this first step is to get an idea of when the system will be stucked due to the lack of freedom imposed by multiple binding. It seems that p_B so far is independant of V, thus we want to focus on the event when a single loop reach an absorbing point in which, the loop is unbound and with zeros crosslinkers surrounding it. The only way the system can evolve further is to unbind. The other output possible for a loop is a consecutive binding. We can compare the probability that a loop continue to bind, as compare to the previous absorbing state. The scaling with $|\mathbf{R}_1 - \mathbf{R}_0|/\ell_0$ or ℓ_0 alone should leads to a minimum size of the loop. We then consider a Poland-Sheraga type of model, in which a polymer loop can bind at precise, and regular binding points. The spacing between these different binding points should fit the previous minimum size of a loop. To bind further, we finally consider that the concentration of cross-linkers further increases as the order parameter : $|\mathbf{R}_1 - \mathbf{R}_0|/\ell_0$ goes to 1. That's the self consistent equation which should lead to 'infinite aging.

5 The total system



We now consider a full system of loops, as represented in Fig. 5. We denote a state of the system : $\mathcal{C} = \mathbf{R}_0, \mathbf{R}_1, \ell, \{\mathbf{r}_i, \ell_i\}_{i\in[0,N]}$. The transition rates are

essentially the same as before:

$$\begin{cases} r_{u\to b}(\ell_i) = \dfrac{\rho a^3 \ell_i / b}{\tau_0} \\ r_{b\to u} = \dfrac{E^{\beta E_b}}{\tau_0} \end{cases} \tag{21}$$

6 Appendix

6.1 No average over k wrong (I think)

We consider two spaces of states : $\mathcal{U} = \mathcal{R}_0, \mathcal{R}_1, \ell, \{k\}$, and $\mathcal{B} = \mathcal{R}_0, \mathcal{R}_1, \ell, \{k'_0\}, \{k'_1\}$. The probability of being in a state in $\mathcal{U}$ can be written as :

$$p_{\mathcal{U}}(k) = \frac{\Omega(\mathcal{U}, k) P(k)}{\mathcal{Z}}, \tag{22}$$

where $\Omega(\mathcal{U}, k)$ is the number of configuration of the loop $(\mathbf{R}_0, \mathbf{R}_1, \ell)$ that is <u>not</u> bounded to any of the k cross-linkers. Using Eq. 5 we write :

$$\Omega(\mathcal{U}, k) = \Omega(\mathbf{R}_0, \mathbf{R}_1, \ell)(1 - k < p_{\text{meet}}(\mathbf{r}, \mathcal{C} >_{\mathbf{r}}), \tag{23}$$

where $< p_{\text{meet}}(\mathbf{r}, \mathcal{C} >_{\mathbf{r}}$ is the probability that the polymer meets a cross-linker averaged over all possible positions of the cross-linker. We write it simply p_{meet} in the following. Similarly to Eq. 22, we define:

$$p_{\mathcal{B}}(k'_0, k'_1) = \frac{e^{\beta E_b} < k < \Omega(\mathcal{C}, \mathbf{r}) >_{\mathbf{r}} >_k P(k'_0) P(k'_1)}{\mathcal{Z}}, \tag{24}$$

where $< k < \Omega(\mathcal{C}, \mathbf{r}) >_{\mathbf{r}} >_k$ corresponds to the number of configuration of the loop averaged over all position and all values of k. Finally we define the partition function of the system :

$$\mathcal{Z} = \Omega(\mathbf{R}_0, \mathbf{R}_1, \ell) \left(\sum_{k=0}^{\infty} (1 - k p_{\text{meet}}) P(k) + \sum_{k'_0=0}^{\infty} \sum_{k'_1=0}^{\infty} e^{\beta E_b} p_{\text{meet}} P(k'_0) P(k'_1) \right) \tag{25}$$

Finally we get:

$$\begin{cases} p_{\mathcal{U}}(k) = \dfrac{(1 - k p_{\text{meet}}) P(k)}{(1 - \rho V p_{\text{meet}}) + \rho V p_{\text{meet}} e^{\beta E_b}} \\ p_{\mathcal{B}}(k'_0, k'_1) = \dfrac{e^{\beta E_b} \rho V p_{\text{meet}} P(k'_0) P(k'_1)}{(1 - \rho V p_{\text{meet}}) + \rho V p_{\text{meet}} e^{\beta E_b}} \end{cases} \tag{26}$$

and detailed balance gives:

$$\begin{cases} r_{\mathcal{U}\to\mathcal{B}}(k, k'_0, k'_1) = \dfrac{1}{\tau_0} \dfrac{\rho V p_{\text{meet}}}{1 - k p_{\text{meet}}} P(k'_0) P(k'_1) \\ r_{\mathcal{B}\to\mathcal{U}}(k, k'_0, k'_1) = \dfrac{e^{-\beta E_b}}{\tau_0} P(k) \end{cases} \tag{27}$$

References

[1] Miriam Linsenmeier, Maria Hondele, Fulvio Grigolato, Eleonora Secchi, Karsten Weis, and Paolo Arosio. Dynamic arrest and aging of biomolecular condensates are regulated by low-complexity domains, RNA and biochemical activity, February 2021.