Pinned polymer loops in external field: modeling meiotic chromosomes

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Chromatin movement and structure are central to many processes in cells such as mitosis, replication, and transcription, where physical properties of the chromatin fiber play an essential role. Motivated by the problem of chromosome alignment and recombination during meiosis we solve for the statistics of a pinned polymer ring in an external force field. We predict how the contact probability between two rings depends on the ratio of the force to the intrinsic noise level and how it changes upon addition of new recombination spots. Due to the underlying loop topology, our theoretical results are readily applicable to the description of bacterial DNA and polymer brushes.

I. INTRODUCTION

The process of meiosis lies at the origin of genetic diversity in Nature. When two haploid cells initiate meiosis their goal is to bring homologous chromosomes in close physical proximity to allow them to recombine. This is an active process during which homologous parts of the chromosomes find each other and proceed to recombination. Recombination points form a tight bond between the chromosomes which are resolved after the first meiotic division of the cell. The second cycle of division finalizes meiosis to produce four cells carrying a unique genetic mixture of both parents [].

Fission yeast *S. bombe* is the model organism of cell biology []. The initial stage of its meiosis is marked by an extended phase of nuclear oscillations where the whole nucleus is dragged from one pole of the elongated zygote to the other (see Fig. 1(a) and Movie S1). It was shown that microtubules and dynein motors actively drive those oscillations []. The exact role of such dramatic nuclear movement, however, remains unclear []. It is hypothesised that nuclear movements are required to align chromosomes before they may proceed to recombination []. Experimental data shows that the distance between two homologous loci gradually decreases during the oscillations till they pair and, contrary, if oscillations are stopped the loci remain far apart during the observation and fail to pair []. It underlines the importance of the emerging physical forces for the alignment of the chromosomes.

As illustrated in Fig.1(b), chromosomes are bound by both ends to the spindle pole body (SPB), which is at the same time the anchoring point of microtubules driving the oscillations. Under the force generated by microtubules and molecular motors, SPB traverses through the nuclear envelope membrane and drags the chromosomes. Due to the generated viscous drag force and loop geometry the chromosomes can align, with a metaphoric similarly to doing washing in a river. In this paper, we investigate how the drag force can affect the spatial configuration of pinned polymers with the loop geometry and how it enhances their contact probability. Remarkably, within the same framework we can take into account additional constraints, such as new recombination spots, and show how they reduce the relative fluctuations of "homologous" polymeric regions. To address this problem we use the ubiquitous freely jointed chain model of the chromosomes [].

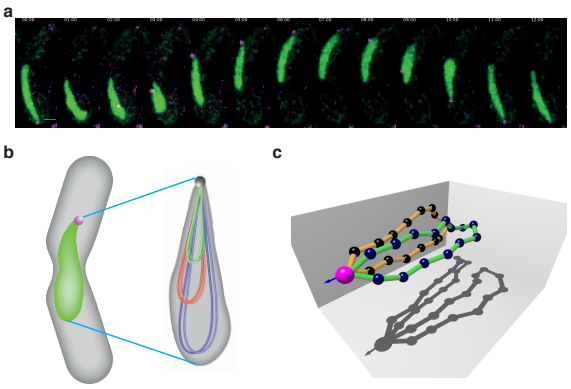

FIG. 1. From meiotic oscillations to the model. (a) Consequent images of moving nucleus during the horsetail oscillations during meiosis in fission yeast. (b) Schematic representation of the homologous chromosomes in loop geometry. For each chromosome its both ends are bound to the spindle pole body and form a bouquet configuration. (c) A sketch of the model showing the polymer loop as a bead rod chain which is dragged through the medium with a constant speed and the force is applied to the single bead.

II. MODEL

While certainly an oversimplification, the ideal chain model is a powerful tool allowing for the analytical treatment in polymer physics []. In particular, it proves to be effective in the complex geometry with external forces characteristic to meiotic chromosomes in fission yeast. The chromosome is represented by a chain of beads connected by freely jointed rods, where the length of a rod a is given by the Kuhn length of the polymer. By knowing the total number of base pairs in each chromosome

(2.5, 4.5 and 5.6 Mbp []), its compaction ratio ($\sim$ 100 bp/nm []), and Kuhn length ($\sim$ 100 nm []) we can calculate the number of rods N to be in the range 250 to 560. It was shown that during oscillations the SPB is pulled with almost a constant speed $v_0 \simeq 2.5\mu$m/min []. We assume that the force exerted on SPB by the microtubules is balanced by the friction of chromosomes with highly viscous nucleoplasm. In the co-moving frame of the spindle pole body that would result in a constant force field acting on every bead except for the one which we identify with the SPB, see Fig. 1(c). In addition to the regular forces there is a stochastic component resulting from thermal fluctuations but also from other active but intrinsically random processes within the cell, which we will characterize by an effective temperature T. Our goal is to describe the statistics of an ideal chain having a loop geometry and pinned at a single point as a function of a constant uniform force and the noise level. In favour of analytical tractability we will not consider the excluded volume effects, and allow the polymers to freely cross each other.

III. RESULTS

We start with noticing that without the external force the problem is similar to the Brownian bridge setup: a random walk in space which returns to the starting point after N steps, $\mathbf{r}_0 = \mathbf{r}_N = 0$, where the role of time is played by the distance measured along the trajectory or the number of monomers in the chain[1]. The position of each bead $\mathbf{r}_i$ in the chain is normally distributed with a zero mean, but its variance becomes a function of a distance s from the pinned point measured along the polymer. In the proper continuous limit fluctuations of the polymer are described by $\langle \mathbf{r}^2(s) \rangle = s(L-s)/L$, where $L = aN$ is the length of the loop. Fluctuations are maximal in the mid-point of the loop and disappear when approaching the pinned point.

We now add an external force field acting on each bead (except for the pinned one), $\mathbf{F} = -\gamma \mathbf{v}_0$, where $\mathbf{v}_0$ is the velocity of the SPB and we choose it such, that the drag force points to the positive direction of z-axis in the co-moving frame. The potential energy of the chain is then:

$$U = -a \sum_{i=1}^{N} \sum_{j=1}^{i} \mathbf{F} \mathbf{e}_j, \quad \mathbf{e}_j = \frac{1}{a}(\mathbf{r}_j - \mathbf{r}_{j-1}). \tag{1}$$

We are interested in the marginal distribution of the i-th monomer in equilibrium which is determined by the potential energy of each configuration, therefore in the following we will omit the kinetic energy contribution. Because of a very peculiar analogy, we first consider the one-dimensional variant of this general setup and then return to the original 3D setting.

One-dimensional system. Consider the force acting in the positive direction of the axis. In this case the rods connecting the neighbouring beads can point along or against the field with $e_j = \pm 1$. We introduce a shifted and rescaled variable $n_j = (e_j + 1)/2$ to rewrite the potential energy in the following simple form (we also exchanged the summation order in Eq. (1) and utilized the loop condition $\sum_{i=0}^{N} e_i = 1$):

$$U = -2\gamma a v_0 \sum_{j=1}^{N} j n_j. \tag{2}$$

In this expression it is easy to recognize the energy of $N/2$ fermions distributed over N equidistant energy levels with $\delta E = 2\gamma a v_0$ and where $n_i = \{0, 1\}$ is the occupation number. Therefore the problem of a monomer orientation for a pinned polymer loop in the constant external field is equivalent to the problem of distributing $N/2$ fermions over N energy levels[2]. In the limit of large N we take the statistical physics approach, where it is know that the average occupation number is given by the Fermi-Dirac distribution:

$$\langle n_j \rangle = P(n_j = 1) = \frac{1}{1 + \exp\left(\frac{j-\mu}{\widetilde{T}}\right)}, \quad \widetilde{T} = \frac{k_B T}{2\gamma a v_0}. \tag{3}$$

where we defined a dimensionless temperature which characterizes the ratio of random forces to the friction force. The chemical potential has to be selected such that the total number of particles is equal to $N/2$: $\mu = (N+1)/2$. We can now directly translate this result to the average position of $i-$th particle:

$$\langle r_i \rangle = a \sum_{j=1}^{i} \langle e_j \rangle = 2a \sum_{j=1}^{i} \langle n_j \rangle - i, \tag{4}$$

which in the limit of large temperature $\widetilde{T} \gg 1$ is approximated by a simple expression (see Supplementary Material):

$$\langle r_i \rangle \simeq 2a\widetilde{T} \ln \left[\frac{\exp\left(\frac{N}{2\widetilde{T}}\right)}{\exp\left(\frac{i}{2\widetilde{T}}\right) + \exp\left(\frac{N-i}{2\widetilde{T}}\right)} \right]. \tag{5}$$

This expression excellently reproduces the average position of each polymer segment and therefore the average polymer configuration. We compare the analytical result of Eq.(4) with Metropolis MC simulations in Fig.

[1] The difference is that Brownian bridge is defined for a time continuous Brownian motion, but the equivalence to the random walk problem can be demonstrated in the proper limit.

[2] The number of positive and negative steps is equal due to the loop condition, therefore it is exactly half of the monomers aligned in positive direction which we identify with fermion particles.

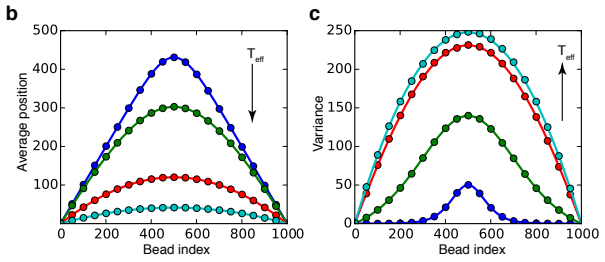

FIG. 2. Configuration of a polymer loop, one dimension. (a) Average positions of the beads as a function of their index along the polymer and different temperature. For high temperatures it is close to zero as in the force free case, and for low temperature the polymer is completely stretched. (b) Fluctuations are maximal for higher temperature and gradually decrease with increasing force. Symbols are numerical simulations and the lines correspond to the theoretical predictions.

2(a). In a very strong force field (low rescaled temperature), the polymer is completely stretched whereas for high temperature average position of each bead is close to zero, recovering the force-free result.

However, to describe how two identical polymer loops behave relative to each other (trivially, the average distance between them is zero) we need to quantify their fluctuations. A sample path of a random walk which takes steps according to the Fermi-Dirac probability distribution Eq. (3) will return to the origin only on average. Therefore we need to enforce this constrain in the same way it is done for a standard Brownian bridge problem.

Brownian bridge in external field. Consider a random walk which takes independent steps with probabilities given by Fermi-Dirac distribution Eq.(3). For an arbitrary piece of trajectory from a bead k to a bead l ($l > k$) the variance accumulates as a sum of variances of each single step:

$$\sigma^2_{k\to l} := \mathrm{var}[r_l - r_k] = 4a^2 \sum_{j=k+1}^{l} \mathrm{var}[n_j]$$
$$= 4a^2 \sum_{j=k+1}^{l} P(n_j = 1)[1 - P(n_j = 1)]. \quad (6)$$

The corresponding displacement $(r_l - r_k)$ is normally distributed around the mean calculated by using Eq. (4). These formulas define the propagator $\rho\,(r_l = r|r_k = 0)$ with variance and mean which depend on k and s according to Eqs. (4) and (6) . The solution of the loop problem is a conditional distribution $\rho_{0\to s\to N} = \rho\,(r_s = r|r_1 = 0, r_N = 0)$ which can be calculated by the conditional probability formula

$$\rho_{0\to s\to N} = \frac{\rho(r_s = r|r_1 = 0)\rho(r_{N-s} = r|r_1 = 0)}{\rho(r_N = 0|r_0 = 0)}. \quad (7)$$

As in the case of the standard Brownian bridge problem, $\rho_{0\to s\to N}$ is again a Gaussian distribution with a mean $\langle r_s\rangle$ and variance

$$\sigma^2_{0\to s\to N} = \frac{\sigma^2_{0\to s}\sigma^2_{0\to N-s}}{\sigma^2_{0\to N}}. \quad (8)$$

However, in case of the external force and Fermi-Dirac distribution of steps, the variances in the above equation depend on the starting and end points in a non-trivial way. To illustrate this, we consider the limit $\widetilde{T} \gg 1$, when the sums can be approximated by integrals (see Supplemental Material) and obtain

$$\sigma^2_{k\to l} \simeq 4a^2\widetilde{T}\left[\frac{1}{1+\exp\left(\frac{k-N/2}{\widetilde{T}}\right)} - \frac{1}{1+\exp\left(\frac{l-N/2}{\widetilde{T}}\right)}\right] \quad (9)$$

By substituting Eq.(9) into Eq.(8) we arrive to an analytical expression for the variance of the polymer position as a function of a distance from the pinned point. In Fig. 2(b) we see that analytical results excellently match the simulations even for $\widetilde{T} \gtrsim 1$. As in the force-free case the fluctuations are strongest in the middle of the loop. The main anticipated result is the overall decrease of the fluctuations with increasing force, which confirms the positive effect of the external force on polymer alignment. We now generalize the results for the three-dimensional case.

Back to three dimensions. We choose the $z-$axis to point along the force field and rewrite the potential energy in terms of the z-projection of the rod connecting two beads $e_{j,z}$:

$$U = -\gamma a v_0 \sum_{i=1}^{N}\sum_{j=1}^{i} e_{j,z}. \quad (10)$$

Following the standard procedure of statistical mechanics we write down the grand-canonical partition function:

$$\mathcal{P} = \prod_{j=1}^{N}\mathcal{P}_j = \prod_{j=1}^{N}\frac{1}{2}\int_{-1}^{1} de_{j,z}\exp\left(-\frac{(j-\mu)e_{j,z}}{\widetilde{T}}\right) \quad (11)$$

The partition function $\mathcal{P}_j$ can be easily calculated

$$\mathcal{P}_j = \frac{\widetilde{T}\sinh\frac{(j-\mu)}{\widetilde{T}}}{j-\mu} \quad (12)$$

and allows us to compute the average and variance of $e_{j,z}$ as

$$\langle e_{j,z}\rangle = \widetilde{T}\partial_\mu \ln\mathcal{P}_j = \coth\frac{\mu-j}{\widetilde{T}} - \frac{\widetilde{T}}{\mu-j}$$
$$\mathrm{var}[e_{j,z}] = \widetilde{T}^2\partial^2_\mu \ln\mathcal{P}_j = \frac{\widetilde{T}^2}{(j-\mu)^2} - \mathrm{csch}^2\frac{j-\mu}{\widetilde{T}} \quad (13)$$

By using the loop condition $\sum_{i=1}^{N}\langle e_{j,z}\rangle = 0$ we can determine the chemical potential again to be $\mu = (N+1)/2$.

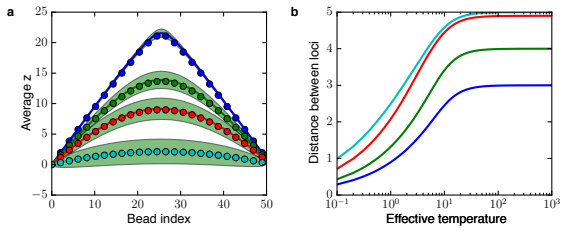

FIG. 3. From a single to two loops (a) Average positions (dots and lines) and fluctuations (shaded regions) of the beads in 3D for different temperatures. Stronger force leads to a more stretched polymer and reduced fluctuations, whereas low force leads to a zero average position and strongest fluctuations. Only the coordinate along the field is shown. (b) Fluctuations of the distance between two polymer loops for four different beads position as a function of temperature. Fluctuations are systematically decreasing with decreasing the temperature, but the absolute value depends on the exact position of the bead. Symbols are numerical simulations and the lines correspond to the theoretical predictions.

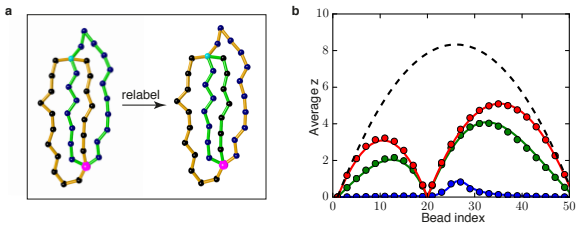

FIG. 4. Centromere as an additional constrain. (a) The sketch shows re-definition of the loops allowing to treat the soft constrain of an additional contact point between the polymer loops. (b) Fluctuations of the distance between homologous loci of the loops at different temperatures. The dashed line shows the maximal fluctuations of the force free case.

For the symmetry reasons the remaining two average projections of the rod are zero: $\langle e_{j,x}\rangle = \langle e_{j,y}\rangle = 0$. The fixed length of the rod at the same time allows to connect the fluctuations in orthogonal directions to the fluctuations along the force: $\mathrm{var}[e_{j,x}] = \mathrm{var}[e_{j,y}] = (1 - \langle e_{j,z}^2\rangle)/2$. We now use the information on averages and variances to construct three independent Gaussian random walk processes happening along each axis. For each axis we proceed with the same steps as for the one dimensional case to impose the loop constrain. In a similar way the fluctuations of polymer position can be calculated from the fluctuations of individual rods' orientations (see Supplemental Material for details). This time we compare the analytical results with a 3D molecular dynamics simulation of bead-rod model and observe an excellent agreement (see Fig.3 a). As in the one-dimensional case, fluctuations are decreasing with increasing the force. The magnitudes of fluctuations are different in the directions along and orthogonal to the force (See Supplemental Material Fig. S1). It is clearly seen for low temperatures, where it is easier to excite transversal fluctuations which cost less energy than perturbing the chain along the force field.

With this information at hand we can quantify the relative fluctuations of two homologous loops. Because of the Gaussian statistics of each loop the relative distance between the loops has the double variance of a single one and fluctuations in all directions needs to be summed up. We can now follow how the fluctuations in the distance change with temperature and the relative position along the loop, as shown in Fig. 3b. By setting a threshold distance one can read out what force is required to bring the polymers sufficiently sloes to each other. In general one needs higher forces to align the monomers which are closer to the middle of the loop. Therefore it is plausible to assume that cells developed additional mechanisms to facilitate pairing.

IV. INTERACTING POLYMER LOOPS

One of such mechanisms can be the higher binding affinity of centromeric chromosomes region (close to the centre of chromosomes) during meiosis []. In addition, during recombination the homologous chromosomes locally form tight junctions called chiasma which hold chromosomes together and are resolved only at later stages of meiosis. To mimic such interactions in the model, we can introduce a *soft* constrain: two polymer loops are pinned to a fixed point, but they are also bound to each other in one additional spot positioned at the same distance along each polymer. This point, which we will refer to as a centromere, however, is free to move in space.

To solve this problem we redefine the loops as shown on Fig.4. The first loop starts along the chromosome 1, goes to the centromere, and then returns to the origin along the chromosome $1'$ by a path of the same length. If the centromere is located at a bead position c, the length of the new loop is $2ac$. For this new loop we choose the statistics of individual rods as defined by the respective chromosomes 1 and $1'$, but impose a new loop condition. Now what we want to know is the statistics of the distance between two homologous chromosome parts. If the point of interest is a bead $h < c$ then its homologous partner has an index $2c - h$ on the new loop. The average distance between these two beads is zero, but its fluctuations we can find following the same lines as before, for example for the z component of the distance $d_z = (\mathbf{r}_h - \mathbf{r}_{2c-h})_z$:

$$\mathrm{var}[d_z] = 2\frac{\bar{\sigma}^2_{0\to h,z}\bar{\sigma}^2_{h\to c,z}}{\bar{\sigma}^2_{0\to c,z}} \tag{14}$$

As one can notice there is an additional factor of 2 and the corresponding variances carry a bar sign, pointing to the fact that they have to be calculated according to the statistics of the redefined loops (slightly more involved but analogous to previous results, see Supplementary Material).

In Fig. 4(b) we can clearly see that by adding a new soft constrain at the centre of the chromosomes dramatically decreases the fluctuations of the distances between

the two chromosomes in the centromeric region and as a result over the whole length of the chromosome (fluctuations without the constrain are showed by the dashed line). The real position of the centromere in fission yeast is slightly off the centre and our theory allows to quantify the fluctuations for an arbitrary position of the binding spot (see Supplementary Material Fig. S2).

V. DISCUSSION

To understand the effect of external force on the alignment of chromosomes we investigated the statistical properties of the ideal chain anchored by both ends to the single point and subjected to the uniform force field. We found the equilibrium configuration of the chain and also explicitly described its fluctuations as a function of the applied force. By combining the basic approach of equilibrium statistical physics and random walks we generalized the Brownian bridge problem to account for the external force. In the one-dimensional case the orientation of the chain monomers is described by a Fermi-Dirac statistics, with analogous results in higher dimensions. Remarkably, by adding additional contact points between the loops, the problem remains analytically solvable.

Looking back to the original biological problem. We considered an idealized model which, however, captures many essential aspects of the original problem of chromosome alignment during meiosis: the loop geometry, a point force balanced by the viscous drag of the whole chain, intrinsic noise, tunable total and persistence lengths of a polymer, multiple loops, and additional soft constrains of the centromere or recombination spots. Certainly, there are challenging aspects that require further work. Probably most important one is the inclusion of a periodic force, in which our approach maybe only valid for certain quasi-stationary stage of each oscillation cycle. Relaxation dynamics or how fast a stationary state can be reached is another interesting problem. We should also mention the importance of confinement by cell walls and the effect of the nuclear envelope which screens the chromosomes from the cytoplasm flow. Nucleus is also a very dense environment as it has to accommodate three pairs of chromosomes, where each chromosome is present as a pair of sister chromatids. Even for numerical simulations the problem in its full physical complexity is a challenging task.

Our results provide a tractable way of elucidating the major effects which originate from a competition of stochasticity and external driving in the proper geometrical setting. They can predict that the central parts of the chromosomes are in general further apart, and how the centromeres can serve an additional role to minimizie the separation of chromosomes in this region.

Recombination as a sequence of brownian bridges It is not enough just to bring homologous loci together, a proper combination of proteins has to assemble to allow for double strand DNA breaks, homology search and chiasma formation, which is a dynamic process. Therefore it is essential to allow for sufficient time of pairing to proceed with recombination. Similarly to the centromere, new appearing recombination spots in turn also bring chromosomes closer together leading to further recombinations. It is known that during meiosis in fission yeast there are around 90 (check!) such locations over three chromosomes []. Our generalisation of the Brownian bridge problem can tackle the statistics of such multiple soft constrains. It can be further generalised to capture the phenomenological dynamics of the sequence of the recombination events that depends on the distribution of the chiasmata locations.

Other applications Polymer rings in general is an active area of research in biopolymers and polymer physics. They were actively studied in different contexts, including their topological properties, knotting, melts, crumpling, and their response to external shear flows. Other examples of ring polymers are bacterial DNA and polymer brushes. Our results provide a novel theoretical framework to quantify the behavior of ring polymers accounting for effects of pinning, polymer-polymer interactions, and external forcing.

ACKNOWLEDGMENTS