Emergent three-dimensional sperm motility: Coupling calcium dynamics and preferred curvature in a Kirchhoff rod model

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Abstract

Changes in calcium concentration along the sperm flagellum regulates sperm motility and hyperactivation, characterized by an increased flagellar bend amplitude and beat asymmetry, enabling the sperm to reach and penetrate the ovum (egg). The signaling pathways by which calcium increases within the flagellum are well established. However, the exact mechanisms of how calcium regulates flagellar bending are still under investigation. We extend our previous model of planar flagellar bending by developing a fluid-structure interaction model that couples the three-dimensional motion of the flagellum in a viscous, Newtonian fluid with the evolving calcium concentration. The flagellum is modeled as a Kirchhoff rod: an elastic rod with preferred curvature and twist. The calcium dynamics are represented as a one-dimensional reaction-diffusion model on a moving domain, the centerline of the flagellum. The two models are coupled assuming that the preferred curvature and twist of the sperm flagellum depend on the local calcium concentration. To investigate the effect of calcium on sperm motility, we compare model results of flagellar bend amplitude and swimming speed for three cases: planar, helical (spiral with equal amplitude in both directions), and quasi-planar (spiral with small amplitude in one direction). We observe that for the same parameters, the planar swimmer is faster and a turning motion is more clearly observed when calcium coupling is accounted for in the model. In the case of flagellar bending coupled to the calcium concentration, we observe emergent trajectories that can be characterized as a hypotrochoid for both quasi-planar and helical bending.

Keywords: Sperm motility; Kirchhoff rod; Calcium dynamics; Hypotrochoid; Regularized Stokeslets

1 Introduction

Changes in cytosolic calcium concentration in the sperm flagellum have been shown to be essential to achieve fertilization of the ovum (egg) (Guerrero et al. 2010, 2011, Suarez 2008). An increase in calcium concentration is associated with hyperactivated motility (Ho & Suarez 2001a, 2003), which is characterized by an increased flagellar bend amplitude and beat asymmetry. This motility pattern enables the sperm to escape from the oviductal sperm reservoir (Demott & Suarez 1992), to detach from the oviductal epithelium (Demott & Suarez 1992), to switch
from a straight motion to a curved motion and back (Darszon et al., 2008), and to penetrate the egg (Drobni et al., 1988; Quill et al., 2003; Ren et al., 2001).

In particular, in mammalian sperm, the opening of CatSper channels on the principal piece of the flagellum has been associated with an increase in intracellular calcium concentration and hyperactivated motility (Carlson et al., 2003; Ho & Suarez, 2001b; Ho et al., 2002; Ho & Suarez, 2003). Experiments have shown that CatSper-null mutant sperm impairs sperm motility and leads to infertility (Ho et al., 2009). The exact mechanisms through which the increase in calcium concentration influences flagellar bending at the level of dyneins, the active force generators, is not completely known. Calcium is hypothesized to bind to centrin or calmodulin, different calcium binding proteins within the axonemal structure of the flagellum (Lindemann & Kanous, 1995). The increase in calcium does change the bending, but the exact timing and magnitude of force generation is not completely known. However, in a phenomenological model, we can explore different ways to couple evolving calcium dynamics to flagellar bending.

Sperm are navigating in a complex, three-dimensional (3D) fluid environment in order to reach and penetrate the egg. Thus, it is necessary to study sperm motility in 3D (Guerrero et al., 2011). However, many experiments have analyzed the motility of a few sperm where motility is recorded at a fixed depth. In this case, motility patterns observed were restricted in a given two-dimensional (2D) plane. Recently, new technologies have been developed to trace the 3D trajectories of multiple sperm at the same time (Jikeli et al., 2015; Su et al., 2012, 2013). Observed sperm trajectories can vary form planar to quasi-planar, and to helical, possibly forming a chiral ribbon or a flagelloid curve. The particular trajectory observed depends on the species of sperm, on the proximity to the oviductal walls and on the external fluid properties. In particular, Smith et al. (2009) and Woolley (2003) showed that the flagellar beat form can change from 3D to 2D as the fluid viscosity increases.

Different 2D and 3D elastohydrodynamic models have been developed to study flagellar motility and the interaction with the surrounding fluid, where the flagellum is either described at the continuum level (Gadelha et al., 2010; Huang & Olson, 2017; Olson et al., 2013, 2011; Simons et al., 2014, 2015), or at a more detailed level including the mathematical description of the discrete dynein motors, along with the accessory structures such as the microtubules and nexin links in the flagellum (Yang et al., 2008). In these models, the flagellar beat form is an emergent property. Another approach is to prescribe the flagellar beat form (Curtis et al., 2012; Ishimoto & Gaffney, 2016; Ishimoto et al., 2017). In all of these modeling studies, regardless of how the flagellar beat form is modeled, the observed sperm trajectory is an emergent property of the coupled system accounting for the fluid dynamics and the swimmer.

Only a few mathematical models have been developed to describe the time-dependent calcium dynamics inside the flagellum (Li et al., 2014; Olson et al., 2010; Wennemuth et al., 2003). Wennemuth et al. (2003) studied in detail the calcium clearance phenomena along the flagellum via ATP-ase pumps. Later, Olson et al. (2010) developed a model that couples the ATP-ase pumps with the contributions of CatSper channels and a calcium store in the neck. Of the many modeling studies related to sperm motility, only a handful of models have attempted to couple the calcium dynamics to flagellar bending. Previous models include a calcium dependent curvature model where flagellar bending was planar (2D) (Olson et al., 2011; Olson 2013; Simons et al., 2014). Other models have prescribed different waveforms for an activated (low calcium) or hyperactivated (high calcium) sperm to investigate emergent trajectories and interactions with a wall (Curtis et al., 2012; Ishimoto & Gaffney, 2016; Ishimoto et al., 2017).

Here, we develop the first mathematical model that couples the 3D dynamics of the sperm flagellum and the surrounding fluid, modeled respectively as a Kirchhoff rod and a Newtonian viscous fluid, with the CatSper channel mediated calcium dynamics inside the flagellum, via a curvature dependent coupling. The model is then used to investigate the emergent 3D wave-
2 METHODS

2.1 Flagellum and fluid dynamics

Since we are interested in studying the motion of sperm in 3D, we utilize a Kirchhoff rod with preferred curvature and twist to model the elastic flagellum, as in Olson et al. (2013). As depicted in Figure 1, the flagellar centerline is represented by a 3D space curve \( \mathbf{X}(t, s) \) and an associated orthonormal triad \( \{ \mathbf{D}^1(t, s), \mathbf{D}^2(t, s), \mathbf{D}^3(t, s) \} \), \( s \) is the rod spatial coordinate, \( L \) is the length of the unstressed rod, and \( t \) is the temporal coordinate. \( \mathbf{f}(t, s) \) and \( \mathbf{n}(t, s) \) are the external force and torque per unit of length applied to the rod.

The following elastic energy penalty is considered

\[
E(t) = \frac{1}{2} \int_0^L \sum_{i=1}^3 \left( a_i \left( \frac{\partial \mathbf{D}^i}{\partial s} \cdot \mathbf{D}^k - \Omega_i \right)^2 + b_i \left( \frac{\partial \mathbf{X}}{\partial s} \cdot \mathbf{D}^i - \delta_{i3} \right)^2 \right) ds, \tag{2.1}
\]

where \((i, j, k)\) is any cyclic permutation of \((1,2,3)\) and \(\delta_{ij}\) is the Kronecker delta. The material parameters for the flagellum (Kirchhoff rod) include the bending moduli \(a_1\) and \(a_2\), the twisting modulus \(a_3\), shear moduli \(b_1\) and \(b_2\), and extensional modulus \(b_3\). We consider an axially symmetric and isotropic rod, hence \(a_1 = a_2\). The preferred strain twist vector is defined as \(\Omega = \left( \Omega_1, \Omega_2, \Omega_3 \right)\) where \(\Omega_1\) and \(\Omega_2\) are the geodesic and normal curvature, respectively,

\[
\Omega = \sqrt{\Omega_1^2 + \Omega_2^2} \tag{2.2}
\]

is the preferred curvature, and \(\Omega_3\) is the preferred twist. In this framework, the rod will tend to minimize its energy and differences between the rod configuration and its preferred shape forms and trajectories when coupling calcium and curvature, comparing to the 2D case. The planar bending case is fully characterized for the Kirchhoff rod model and compared to previous results when using an Euler elastica representation. Further, we investigate helical bending in the case of equal bending amplitudes (spiral bending) and the case of unequal bending amplitudes (quasi-planar since one amplitude is significantly smaller). The quasi-planar and helical bending cases exhibit emergent trajectories that can be described as a hypotrochoid, similar to the flagelloid curve observed in experiments by Woolley (2003).
will generate force and torque along the centerline. As described in Section 2.2, the preferred strain twist vector will be time and spatially dependent and coupled to the evolving calcium concentration inside the sperm flagellum. In order to study fully 3D movement and couple mechanics to chemical concentrations, we simplify the sperm representation by neglecting the head or cell body, as in previous modeling studies (Olson et al., 2013; Simons et al., 2014).

Starting from the energy penalty in (2.1), and using a similar variational argument to the one detailed in Peskin (2002) and Lim et al. (2008), the following balance of linear and angular momentum equations can be derived

\[ 0 = f + \frac{\partial F}{\partial s}, \quad (2.3) \]
\[ 0 = n + \frac{\partial N}{\partial s} + \left( \frac{\partial X}{\partial s} \times F \right), \quad (2.4) \]

where \( f \) and \( n \), illustrated in Figure 1, are the external force and torque per unit of length applied to the rod, respectively, and \( F \) and \( N \) are the average internal force and momentum transmitted across a cross-section of the rod, respectively. The constitutive equations for internal force and torque can be expressed as

\[ F = \sum_{i=1}^{3} b_i \left( \frac{\partial X}{\partial s} \cdot D^i - \delta_{3i} \right) D^i, \quad (2.5) \]
\[ N = \sum_{i=1}^{3} a_i \left( \frac{\partial D^i}{\partial s} \cdot D^k - \Omega_i \right) D^i. \quad (2.6) \]

We remark that, by minimizing the energy formulation considered in (2.1), we are weakly imposing the following constraints

1. the actual strain twist vector \((\Omega_1^*, \Omega_2^*, \Omega_3^*) = \left( \frac{\partial D^2}{\partial s} \cdot D^3, \frac{\partial D^3}{\partial s} \cdot D^1, \frac{\partial D^1}{\partial s} \cdot D^2 \right)\) is equal to the preferred strain twist vector \((\Omega_1, \Omega_2, \Omega_3)\), hence the actual curvature

\[ \Omega^* = \sqrt{(\Omega_1^*)^2 + (\Omega_2^*)^2} = \sqrt{\left( \frac{\partial D^2}{\partial s} \cdot D^3 \right)^2 + \left( \frac{\partial D^3}{\partial s} \cdot D^1 \right)^2} \quad (2.7) \]

is equal to the preferred curvature \(\Omega\) in (2.2);

2. \(D^3\) is aligned with the tangent vector, both \(D^1\) and \(D^2\) are orthogonal to the tangent vector, and the rod is inextensible, i.e. \( \left| \frac{\partial X}{\partial s} \right| = 1 \).

These constraints are imposed weakly, hence they tend to be maintained approximately instead of exactly. Moreover, \( a_i \) and \( b_i \), that physically represent the material properties of the flagellum, act as Lagrange multipliers for these constraints. Note that the first constraint is reflected in the internal torque constitutive equation (2.6), while the second constraint is reflected in the internal force constitutive equation (2.5). The standard Kirchhoff rod model, i.e. the strongly constrained model, can be obtained from the weakly constrained model by setting \( b_1 = b_2 = b_3 \) (see Lim et al., (2008)), as is the case in this work.

Sperm motility occurs in a regime where viscous forces dominate and acceleration is negligible. Hence, the fluid surrounding the flagellum is modeled as a viscous and incompressible
2 METHODS

Newtonian fluid using Stokes equations

\[ 0 = -\nabla p + \mu \nabla \cdot \mathbf{v} + \mathbf{f}^r, \quad (2.8) \]
\[ 0 = \nabla \cdot \mathbf{v}, \quad (2.9) \]

where \( p \) and \( \mathbf{v} \) are the fluid pressure and velocity, respectively, \( \mu \) is the fluid viscosity and \( \mathbf{f}^r \) is the force per unit of volume that the sperm exerts on the fluid.

To solve this fluid-structure interaction problem we use the method of regularized Stokeslets, described in detail in Cortez (2001) and Olson et al. (2013). The main idea is to derive the fundamental solution of the Stokes problem in the case of a regularized point force or a regularized point torque. Then, the global solution is obtained by adding the various contributions along the flagellum, taking advantage of the linearity of the Stokes equations. The point force or torque applied at the point \( \mathbf{X}_0 \) is regularized using the following radially symmetric blob function

\[ \phi_\varepsilon(\mathbf{x}, \mathbf{X}_0) = \frac{15\varepsilon^4}{8\pi (|\mathbf{x} - \mathbf{X}_0|^2 + \varepsilon^2)^{7/2}}, \quad (2.10) \]

where \( \mathbf{x} \) is any point in the fluid. The blob function approaches the Dirac delta distribution as \( \varepsilon \to 0 \) and satisfies \( \int_{\mathbb{R}^3} \phi_\varepsilon(\mathbf{x}, \mathbf{X}_0) d\mathbf{x} = 1 \). Note that the regularization parameter \( \varepsilon \) determines the support of the blob function \( \phi_\varepsilon \) and its value can be chosen so that the support of \( \phi_\varepsilon \) corresponds to the physical radius of the cross-section of the rod.

The dynamic coupling between the surrounding fluid flow and the elastic flagellum is expressed by the force \( \mathbf{f}^r \) in (2.8), which by Newton’s third law, depends on the rod external force \( \mathbf{f} \) and torque \( \mathbf{n} \). Using the method of regularized Stokeslets, we can write the force \( \mathbf{f}^r \) exerted by the rod on the point in the fluid \( \mathbf{x} \) as

\[ \mathbf{f}^r(t, \mathbf{x}) = \int_{\Gamma(t)} \left( -\mathbf{f}(t, s) + \frac{1}{2} \nabla \times (-\mathbf{n}(t, s)) \right) \phi_\varepsilon(\mathbf{x}, \mathbf{X}(t, s)) ds, \quad (2.11) \]

where the curve \( \Gamma(t) = \mathbf{X}(t, s) \) (for more details see Olson et al. (2013)). Note that in (2.8)-(2.9) we consider the incompressible steady Stokes equation, however the forces \( \mathbf{f}^r \) that the rod exerts on the fluid in (2.11) is time-dependent. The kinematic coupling between the surrounding fluid flow and the motion of the elastic flagellum is imposed by the following no-slip boundary conditions on the fluid linear velocity \( \mathbf{v} \) and angular velocity \( \mathbf{w} \)

\[ \frac{\partial \mathbf{X}}{\partial t} = \mathbf{v}(\mathbf{X}), \quad \frac{\partial \mathbf{D}^i}{\partial t} = \mathbf{w} \times \mathbf{D}^i, \quad i = 1, 2, 3. \quad (2.12) \]

Accordingly, at each instant in time, given the force exerted by the rod on the fluid, we can solve for the resulting fluid flow and update the rod location assuming it moves with the local fluid velocity.

2.2 Calcium and curvature dynamics

In mammalian sperm, the asymmetry and magnitude of flagellar bending is known to vary as a function of the calcium concentration inside the flagellum (Ho & Suarez, 2001a; Ho et al., 2002; Lindemann & Goltz, 1988; Marquez & Suarez, 2007; Tash & Means, 1982). We will use a previously developed one-dimensional reaction-diffusion model to account for the relevant calcium dynamics (Olson et al., 2010). The increase in intracellular calcium concentration is initiated by the opening of CatSper channels (Carlson et al., 2003), which allows calcium to
flow from the external fluid bath to the inside of the sperm flagellum. The sperm flagellum is composed of five pieces: the head, neck, mid-piece, principal piece, and end piece (Cummins & Woodall, 1985; Pesch & Bergmann, 2006). Channels such as CatSper (Chung et al., 2014) and pumps such as the calcium ATP-ase pump (Okunade et al., 2004) are localized along the length of the principal piece, whereas the Redundant Nuclear Envelope (RNE), a calcium store (Ho & Suarez, 2001b, 2003), is found in the neck.

The calcium concentration $Ca(t, s)$ at time $t$ and location $s$ along the centerline of the Kirchhoff rod $\mathbf{X}(t, s)$ is governed by

$$
\frac{\partial}{\partial t} \left( Ca(t, s) \left| \frac{\partial \mathbf{X}(t, s)}{\partial s} \right| \right) = D_{Ca} \frac{\partial}{\partial s} \left( \frac{\partial Ca(t, s)}{\partial s} / \left| \frac{\partial \mathbf{X}(t, s)}{\partial s} \right| \right) + J(t, s, Ca(t, s)) \left| \frac{\partial \mathbf{X}(t, s)}{\partial s} \right|,
$$

where $D_{Ca}$ is the calcium diffusion coefficient. The calcium flux $J$ varies along the sperm length and incorporates the fluxes in the principal piece (CatSper channels and pumps) and in the neck (RNE contribution), depends non-linearly on $Ca$, and is coupled to the evolving concentration of inositol 1,4,5-trisphosphate or IP$_3$ (for more details see Olson et al. (2010, 2011)). Note that we assume a large and constant calcium concentration in the fluid surrounding the sperm. The motion and deformation of the rod is taken into account in (2.13) via the term $|\partial \mathbf{X}(t, s)/\partial s|$, representing the Jacobian of the transformation from the straight rod to the current rod configuration (for more details see Lai et al. (2008); Stone (1990)). Figure 2(a) shows the calcium dynamics in time for the case of a fixed straight rod (i.e. $|\partial \mathbf{X}(t, s)/\partial s| = 1$) at three different points along the the sperm length $L$: head (H) - at 0.5% of $L$, in the mid-piece (MP) - at 16% of $L$, and in the principal piece (PP) - at 24% of $L$. (b) Plot of $f$, given in (2.16), as a function of calcium concentration $Ca$ depending on the sign of the preferred normal curvature $\Omega_2$.

The movement of the flagellum will be coupled to the evolving calcium dynamics through the preferred amplitude, in the same fashion as Olson et al. (2011) and Simons et al. (2014), but here we allow for a fully 3D preferred beat form with the Kirchhoff rod model, whereas the previous work used an Euler elastica model with strictly planar or quasi-planar beating. As in Huang & Olson (2017), we assume the following preferred reference configuration of the flagellum in time

$$
\mathbf{X}(t, s) = [x, y, z] = [x(t, s), A \sin(k x(t, s) - \sigma t), B \cos(k x(t, s) - \sigma t)]^T,
$$

Figure 2: Calcium dynamics. (a) Calcium ($Ca$) concentration dynamics in time for the case of a fixed, straight rod at three different points along the sperm length $L$: head (H) - at 0.5% of $L$, in the mid-piece (MP) - at 16% of $L$, and in the principal piece (PP) - at 24% of $L$. (b) Plot of $f$, given in (2.16), as a function of calcium concentration $Ca$ depending on the sign of the preferred normal curvature $\Omega_2$. 

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$$
with the following preferred wave parameters: $A$ and $B$ are the wave amplitudes, $k$ is equal to $2\pi / \lambda$ the wavelength, and $\sigma / 2\pi$ is the beat frequency. Note that depending on the values of $A$ and $B$, the wave can be planar, quasi-planar, or helical; this is representative of the range of beat forms observed in experiments (Dresdner & Katz, 1981; Drobnis et al, 1988; Guerrero et al, 2011; Ooi et al, 2014; Suarez & Dai, 1992; Woolley & Vernon, 2001). We remark that this assumed reference configuration captures different beat forms but does not try to exactly model flagellar bending at the level of dynein activation or microtubule sliding (Vernon & Woolley, 2004; Woolley, 2010).

Since increases in calcium are associated with larger amplitude bending (Ho & Suarez, 2001a; Lindemann & Goltz, 1988; Marquez & Suarez, 2007; Tash & Means, 1982), we assume that the rod amplitudes $A$ and $B$ depend on the calcium concentration along the rod as follows

$$A(t, s) = A_0 f(Ca(t, s)), \quad \text{and/or} \quad B(t, s) = B_0 f(Ca(t, s)),$$

where $A_0$ and $B_0$ are the constant baseline amplitudes and

$$f(Ca(t, s)) = \frac{2}{1 + \exp \left( \frac{-c_1(Ca(t, s) - \hat{Ca})}{c_2 - \hat{Ca}} \right)},$$

with $\hat{Ca}$ representing the baseline calcium concentration in the flagellum. The function $f(Ca)$ is equal to 1 at the baseline, i.e. when $Ca = \hat{Ca}$, and asymptotically approaches 2 as $Ca$ increases, see Figure 2(b). Hence, the values of $A$ and $B$ can at most double their baseline values $A_0$ and $B_0$, respectively, depending on the calcium concentration, in agreement with experiments (Marquez & Suarez, 2007).

In terms of the Kirchhoff model presented in Section 2.1, we need to define a preferred strain twist vector that will define flagellar bending and is also coupled to the calcium concentration. Using the reference or preferred beat form $\hat{X}$ in (2.14) and with the following reference orthonormal triad: $\hat{D}^3$ as the tangent, $\hat{D}^1$ as the normal, and $\hat{D}^2$ as the binormal with respect to $\hat{X}$, we can calculate the preferred strain twist vector $(\Omega_1, \Omega_2, \Omega_3)$ using $\Omega_i = \partial_s \hat{D}^j \cdot \hat{D}^k$, where $(i, j, k)$ is a cyclic permutation of $(1, 2, 3)$. For this rod configuration, the corresponding strain twist vector is given as (Huang & Olson, 2017)

$$\Omega_1 = \frac{Bk^2(1 + A^2k^2) \cos(k x(t, s) - \sigma t)}{\sqrt{K_c(K_c + K_s)^{3/2}}},$$

$$\Omega_2 = \frac{-Ak^2 \sin(k x(t, s) - \sigma t)}{\sqrt{K_c(K_c + K_s)}},$$

$$\Omega_3 = \frac{ABk^3 \sin^2(k x(t, s) - \sigma t)}{(K_c)(K_c + K_s)},$$

where

$$K_c = 1 + A^2k^2 \cos^2(k x(t, s) - \sigma t) \quad \text{and} \quad K_s = B^2k^2 \sin^2(k x(t, s) - \sigma t).$$

The rod is initialized from (2.14) at $t = 0$ and it is given an evolving strain and twist vector by (2.17); the amplitude in (2.15) also varies and is coupled to the evolving calcium concentration. Thus, we have a propagating curvature wave, similar to observations of human sperm flagellar bending (Smith et al, 2009). Additionally, as calcium concentrations increase, experiments have shown that flagellar bending shows an asymmetry: the bend grows in the principal
bend direction and ends early in the reverse bend \cite{Tash1982}. We include this asymmetry in our model by having the parameter $c_2$ in (2.16) vary the speed at which the function $f$ approaches its maximum, as shown in Figure 2(b), coupling $c_2$ to the sign of the curvature $\Omega_2$ as follows

$$c_2 = \begin{cases} 
  c_{2,p} & \text{if } \Omega_2(t, s) > 0, \\
  c_{2,n} & \text{if } \Omega_2(t, s) \leq 0,
\end{cases}$$

(2.19)

similar to \cite{Olson2011} and \cite{Simons2014}. In order to introduce an asymmetry in flagellar bending, both in the case of a 3D wave (i.e. $A \neq 0$ and $B \neq 0$), and in the case of a planar wave (i.e. $B = 0$), we vary the parameter $c_2$ in (2.19) based on the sign of $\Omega_2$, which is the only component of the reference preferred strain vector in (2.17) that does not depend on $B$.

### 2.3 Numerical algorithm for coupling

At time $t = 0$, the rod is initialized using the preferred reference configuration in (2.14) for a given set of wave parameters and is discretized into $k = 1, \ldots, M$ points with constant spacing $\Delta s$. Similarly, the orthonormal triad is initialized as $\hat{D}_3 = [\partial_s x, \partial_s y, \partial_s z]^T$, $\hat{D}_1 = \left(1/\sqrt{\left(\partial_s x\right)^2 + \left(\partial_s y\right)^2}\right)[\partial_s x, \partial_s y, 0]^T$, $\hat{D}_2 = \hat{D}_3 \times \hat{D}_1$ and discretized at the same $M$ points.

To solve for the new configuration of the rod at time $\Delta t$:

1. compute the calcium concentration along the rod by solving (2.13) as in \cite{Olson2010, Olson2011}: using the symmetric Crank-Nicolson scheme detailed in \cite{Lai2008}, that ensures that the total mass of calcium is numerically conserved in the case of flux $J = 0$;

2. given the calcium concentration, determine the preferred amplitude in (2.15) for $A$ and $B$ at each discretized point along the rod;

3. determine the preferred strain and twist $\Omega_i$ in (2.17) for each of the $M$ points, assuming $A$ and $B$ constant at each point;

4. calculate the point forces $f_k$ and torques $n_k$ along the rod using (2.3)-(2.6) as in \cite{Olson2013};

5. solve for the fluid flow in (2.8)-(2.9) using regularized fundamental solutions: the local linear velocity $\mathbf{v}$ and angular velocity $\mathbf{w}$ are given as

$$\mathbf{v}(x) = \frac{1}{\mu} \sum_{k=1}^{M} \mathcal{S}[-f_k \Delta s] + \frac{1}{\mu} \sum_{k=1}^{M} \mathcal{R}[-n_k \Delta s],$$

$$\mathbf{w}(x) = \frac{1}{2} \nabla \times \mathbf{v} = \frac{1}{\mu} \sum_{k=1}^{M} \mathcal{R}(-f_k \Delta s) + \frac{1}{\mu} \sum_{k=1}^{M} \mathcal{D}(-n_k \Delta s),$$

(2.20)

(2.21)

where $\mathcal{R}$, $\mathcal{S}$, and $\mathcal{D}$ are the regularized rotlet, Stokeslet, and dipole for the blob function given in (2.10), as detailed in \cite{Olson2013};

6. update the rod location $X$ and orthonormal triads $D^i$ using the local linear and angular velocity via the no-slip conditions in (2.12), we solve for these using the forward Euler method detailed in \cite{Lim2008} and \cite{Olson2013}. 

3 Results and Discussion

We investigate the effect of the calcium and curvature coupling described in Section 2.2 on sperm motility. The following flagellar beat forms are considered:

- **2D dynamics**: planar wave with $A_0 = 3 \, \mu m$ and $B_0 = 0 \, \mu m$;
- **3D dynamics**: 
  1. helical wave with $A_0 = B_0 = 3 \, \mu m$;
  2. quasi-planar wave with $A_0 = 3 \, \mu m$ and $B_0 = 1 \, \mu m$.

Moreover, we consider three possible cases of calcium and curvature coupling:

- **a) no-coupling (No Ca)**: $A$ and $B$ are fixed to the baseline values, i.e. $A = A_0$ and $B = B_0$;
- **b) symmetric coupling (Ca sym)**: $A$ and $B$ vary symmetrically with respect to curvature, i.e. in Eq. (2.16) $c_2 = c_{2,p} = c_{2,n} = 1$;
- **c) asymmetric coupling (Ca asym A & B)**: $A$ and $B$ vary asymmetrically with respect to curvature, i.e. in Eq. (2.16) $c_{2,p} \neq c_{2,n}$ as in Table 3;
- **d) asymmetric coupling only A (Ca asym A)**: $A$ varies asymmetrically with respect to curvature, while $B$ is kept constant to its baseline value, i.e. $B = B_0$.

Note that in the planar wave case, since $B_0 = 0$, couplings c) and d) are equivalent, and we will refer to them as the asymmetric coupling case (Ca asym).

The material and geometric parameters of the sperm flagellum, the calcium model parameters, as well as the numerical algorithm parameters are summarized in Table 3, with references given where applicable. In this section, we omit results for calcium concentration dynamics along the flagellum since they are analogous to the case of a fixed rod shown in Figure 2(a), with the minor addition of infinitesimal oscillations due to the deformation of the rod, see Olson et al. (2011) for more details.

3.1 2D flagellar dynamics

Figure 3 shows sperm trajectories over a 15 second interval for simulations corresponding to the different cases of calcium-curvature coupling. Note that in the planar wave case $\Omega_1 = \Omega_3 = 0$ since $B = 0$, hence the preferred curvature of the rod is $\Omega = |\Omega_2|$. For this curvature case, in both the symmetric and asymmetric coupling cases, we observe that the sperm has a higher linear velocity ($\mu m \, s^{-1}$) compared to the no-coupling case: No Ca - 28.9, Ca symm - 40.4, and Ca asym - 34.9. The linear velocities extracted from the model results are in agreement with the experimental measurements for human sperm reported in Smith et al. (2009). However, the symmetric coupling is not enough to reproduce the calcium-dependent turn in the sperm trajectory observed in vivo (Marquez & Suarez, 2007). Only when the beat asymmetry is included in the coupling, the numerical results are able to reproduce the turning phenomenon. This is similar to previous computational results for a 2D model of a sperm using an Euler elastica representation of the flagellum (Olson et al. 2011). For this reason, we will only consider the asymmetric coupling cases in the rest of the results section.

To fully understand the dynamics related to the turning phenomenon in the asymmetric coupling case, we compute the external force applied by the sperm to the fluid along the sperm centerline. For the centerline, we use the line passing through the head and the center of mass...
### 3 Results and Discussion

| Parameter                              | Value          | Units         | Reference                     |
|----------------------------------------|----------------|---------------|-------------------------------|
| Amplitude range for $A_0$ and $B_0$    | [0, 3]         | µm            |                               |
| Beat frequency, $\sigma$               | $20(2\pi)$     | Hz            |                               |
| Wavelength, $2\pi/k$                   | 30             | µm            |                               |
| Length, $L$                            | 60             | µm            |                               |
| Head range                             | [0, 0.5]%$L$   | µm            |                               |
| Neck range                             | [0.5, 2.5]%$L$ | µm            |                               |
| Mid-piece range                        | [2.5, 20.5]%$L$| µm            |                               |
| Principal piece range                  | [20.5, 93]%$L$ | µm            |                               |
| End piece range                        | [93, 100]%$L$  | µm            |                               |
| Fluid viscosity, $\mu$                 | $1 \times 10^{-6}$ | $g \mu m^{-1} s^{-1}$ | (water at room temperature) |
| Bending moduli, $a_1 = a_2$            | 1              | g $\mu m^3 s^{-2}$ | [Pelle et al., 2009]          |
| Twist moduli, $a_3$                    | 1              | g $\mu m^3 s^{-2}$ | [Okuno & Hiramoto, 1979]      |
| Shear moduli, $b_1 = b_2$              | 0.6            | g $\mu m s^{-2}$ | [Allbritton et al., 1992]     |
| Extension modulus, $b_3$               | 0.6            | g $\mu m s^{-2}$ | [Sneyd et al., 1995]          |
| Calcium diffusion coefficient, $D_{Ca}$| 20             | $\mu m^2 s^{-1}$|                               |
| Calcium baseline concentration, $\hat{C}_{a}$ | 0.1           | $\mu M$      | [Wennemuth et al., 2003]      |
| Calcium parameter, $c_1$               | ln(9)          | 1             |                               |
| Calcium parameter, $c_{2,p}$           | 0.7            | $\mu M$      |                               |
| Calcium parameter, $c_{2,n}$           | 1              | $\mu M$      |                               |
| Rod spatial discretization, $\Delta s$ | 0.2            | $\mu m$      |                               |
| Temporal discretization, $\Delta t$   | $1 \times 10^{-6}$ | s           |                               |
| Regularization parameter, $\varepsilon$| $5\Delta s$   | $\mu m$      |                               |

Table 1: The material and geometric parameters of the sperm flagellum, the calcium model parameters, as well as the the numerical algorithm parameters.

Figure 3: Planar wave. Sperm trajectories in time $t$ from 0 to 15s for the case of no calcium-curvature coupling (No Ca), symmetric coupling (Ca sym) and asymmetric coupling (Ca asym). A filled in circle is used to denote the head and swimming direction.

Figure 4 shows the time evolution of the average centerline force in the head (top panel) and in the end piece (bottom panel) of the sperm for the case of no-coupling and asymmetric coupling. In the no-coupling case, the average force shows, both in the head and end
RESULTS AND DISCUSSION

Figure 4: Planar wave. Average centerline component of the external force applied by the flagellum to the fluid in time in the head (top panel) and in the end piece (bottom panel) for no-coupling (No Ca, black) and asymmetric coupling (Ca asym, red) cases. For \( t = 0 \) to 0.2s, the curves lie on top of each other.

In Figure 4, the force in the head starts to switch sign from positive to negative, while the average force in the end piece keeps oscillating between positive and negative values. Moreover, at \( t = 5s \), both forces show an increase in amplitude and a decrease in frequency of oscillation. The turning point is followed by a more pronounced asymmetry in the oscillations, a decrease in amplitude and an increase in frequency for \( t = 10s \) and \( t = 15s \), for both flagellum regions considered. Note that the average force in the head is one order of magnitude bigger than the average force at the end piece. We remark that in the planar case, the sperm trajectories lie in the 2D \( xy \)-plane, hence the average external torque component along the centerline is zero. In comparison to a 2D model of a sperm using an Euler elastica representation, the forces we present in the direction of the centerline are on the same order of magnitude (Simons et al., 2014).

In Figures 5 and 6, we investigate the effect of varying the flagellum material properties on the motion of the sperm. We can consider the moduli \( a_i \) as both numerical and material parameters. As numerical parameters, the moduli are Lagrange multipliers that enforce how strictly the curvature and twist are enforced. In addition, these moduli give an effective stiffness to the elastic rod that represents the sperm flagellum. From experiments, we know that the stiffness varies by several orders of magnitude for different species of sperm, where mammalian sperm are generally stiffer than invertebrate sperm (Lindemann et al., 1973; Schmitz & Lindemann, 2004; Pelle et al., 2009). Thus, we run simulations where the bending and twist moduli \( a_i \), in Table 3, have been reduced by a factor of 5 and by a factor of 10. In this parameter regime, the larger bending and twist moduli correspond to a stiffer elastic rod. Figure 5 shows the head trace over 15 seconds in the case of no-coupling (left) and in the case of asymmetric coupling.
Figure 5: Planar wave. Sperm head trajectories in time $t$ varying the bending and twist moduli $a_i$: moduli as in Table 3 (black), reduced by a factor of 5 (red) and reduced by a factor of 10 (blue). No-coupling case (No Ca, left) and asymmetric coupling case (Ca asym, right). The zoomed in portion highlights that the trajectories oscillate in time.

(right) for the three bending and twist moduli values considered. Despite the different material properties considered, the head shows, in general, a linear trajectory in the no-coupling case and a clockwise turning trajectory in the asymmetric coupling case. Moreover, in the asymmetric coupling case, the trajectory radius of curvature is directly proportional to the bending and twist moduli, and the linear speed is also proportional to $a_i$ in both coupling cases considered. While the head trace shows a general linear or turning trajectory, in reality, as shown in the zoomed area of Figure 5, the head trace is an oscillating curve in time. The amplitude and frequency of these head oscillations vary with the material properties and the coupling condition considered, while the shape of the oscillations vary with the material properties but seems to be independent from the coupling condition.

In Figure 6, the flagellar configurations are illustrated for different values of bending and twist moduli $a_i$, over the time interval 14s to 15s. The configurations are translated and rotated so that the head is at the origin and the centerline lies on the horizontal axis, where spatial units are shown in microns. Each row corresponds to a calcium-curvature coupling case, and each column corresponds to different flagellum material properties. The results show that the emergent flagellar wave amplitude is directly proportional to the bending and twisting moduli variation. For all the values of bending and twist moduli considered, there is an increase in the wave amplitude when the asymmetric coupling is considered, and this increase is proportional to the corresponding no-coupling configuration. Moreover, the asymmetric coupling generates an increased tilt of the end piece with respect to the centerline, and this phenomenon is more prominent as the bending and twisting moduli decrease. We also note that trajectories and observed waveforms for varying moduli in the Kirchhoff rod model are similar to those for an Euler elastica model (Olson et al., 2011).

3.2 3D dynamics

3.2.1 Helical wave.

In the helical wave case, Figure 7 shows the overlap of flagellar configurations (thin gray lines) and head trajectory (thick colored line) over the time interval of 1s, form 9s (blue) to 10s (red), for three different coupling cases: No Ca, Ca asym A & B and Ca asym A. On the right is
Figure 6: Planar wave. Evolution of flagellar centerline (translated and rotated to the horizontal axis) for $t = 14s$ to $15s$, spatial units shown are in microns. No-coupling (No Ca, top row), asymmetric coupling (Ca asym, bottom row), bending and twist moduli as in Table 3 (left, black), moduli reduced by a factor of 5 (center, red) and moduli reduced by a factor of 10 (right, blue). The black circle at the origin denotes the head or first point on the flagellum. Depicted the corresponding flagelloid curve (f-curve) traced by the head on the $yz$-plane. The f-curve is defined as the path followed by a fixed point on the flagellum (Woolley, 1998). In the No Ca and Ca asym A & B coupling cases, the flagellar configurations remains helical in time, with increased amplitudes in the Ca asym A & B case, and the f-curves traced by the head are almost circular. In the Ca asym A coupling case, the flagellar configurations show an irregular beat pattern and the head f-curve resembles a hypotrochoid with eight singular points, with increased amplitudes in comparison to the no-coupling case. We note that the linear velocity of the Ca asym A & B case is 12% lower than the no coupling case, due to the higher amplitude and almost perfect helical shape, see Table 3.2.4. While, in the Ca asym A & B case, the irregularity of the flagellum shape together with the increase in wave amplitudes produces an increase of 53% in the linear velocity compared to the no-coupling case, see Table 3.2.4.

Variations in the actual curvature $\Omega^*$ (2.7) along the flagellum spacial coordinate $s$ in the time interval from $9s$ to $9.2s$ are reported in Figure 8 for the same three coupling cases investigated in Figure 7. In the No Ca and Ca asym A & B coupling cases the flagellum curvature is almost constant, and this is consistent with the helical flagellum configuration reported in Figure 7 since helices by definition have constant curvature and twist. However, in the Ca asym A case the curvature varies periodically along the flagellum, and this is consistent with the irregular flagellum beating reported in Figure 7. In all three coupling cases, the normalized absolute difference between the actual curvature $\Omega^*$ and the preferred curvature $\Omega$ (2.2) is less than $1 \times 10^{-2}$.

3.2.2 Quasi-planar wave.

In the case of a quasi-planar wave propagating along the flagellum, Figure 9 shows flagellar configurations (thin gray lines) and the head trajectory (thick colored line) over the time interval
Figure 7: *Helical wave.* 3D overlap of flagellar configurations (thin gray lines) and head trajectory (thick colored line) over the time interval of 1s, form 9s (blue) to 10s (red), on the left. Corresponding flagelloid curve traced by the head on the $yz$-plane, on the right. No-coupling case (No Ca, top), asymmetric coupling case (Ca asym $A \& B$, center) and asymmetric coupling only $A$ case (Ca asym $A$, bottom).

of 1s, form 9s (blue) to 10s (red). The three different coupling cases considered are: *No Ca*, *Ca asym $A \& B$* and *Ca asym $A$*. On the right is depicted the corresponding f-curve traced by the head on the $yz$-plane. In all of the coupling cases, the flagellum configuration shows an irregular beat form, due to the baseline geometric non-linearity introduced by the quasi-planer wave, i.e $A_0 \neq B_0$. At the same time, the three cases differ from each other in terms of the emergent shapes of the head f-curve, achieved wave amplitude, and linear velocity. In the *No Ca* and *Ca asym $A \& B$* cases, the head f-curves resemble a hypotrochoid with four singular points, while in the *Ca asym $A$* case, the head f-curves resemble a hypotrochoid with three singular points. Moreover, the two asymmetric coupling cases show visible increased amplitudes in comparison to the no-coupling case. The linear velocity of the quasi-planar wave cases are reported in Table 3.2.4. The *Ca asym $A \& B$* and *Ca asym $A$* couplings produce an increase in the linear velocity of 37% and 76% compared to the no-coupling case, respectively.

In the quasi-planar case, for all three coupling cases considered in Figure 9 the curvature
3 RESULTS AND DISCUSSION

No Ca
Actual curvature $\Omega^*$ [1/µm]

Ca asym A & B
Actual curvature $\Omega^*$ [1/µm]

Ca asym A
Actual curvature $\Omega^*$ [1/µm]

Figure 8: *Helical wave.* Actual curvature $\Omega^*$ variations along the flagellum spacial coordinate $s$ in the time interval from 9s to 9.2s. No-coupling case (No Ca, left), asymmetric coupling case (Ca asym A & B, center) and asymmetric coupling only A case (Ca asym A, right).

is not constant along the flagellum length and periodically oscillates in time. In particular, Figure 10 shows the comparison between preferred curvature $\Omega$ (2.2) and preferred twist $\Omega_3$ (solid lines) with the actual curvature $\Omega^*$ (2.7) and actual twist $\Omega_3^*$ (dashed lines), for the No Ca (black) and the Ca asym A (red) cases, at time $t = 10s$. The maximum curvature and twist in the Ca asym A is almost twice that of the no-coupling case, and both cases present a phase lag between the oscillations of the preferred and computed curvature and twist.

3.2.3 Hypotrochoid approximation of head f-curves.

As briefly mentioned in the previous sections, the head f-curves presented in Figures 7 and 9 resemble hypotrochoid curves. Hypotrochoid curves can be defined as the trajectories of a point $P$ subjected to a movement composed of two circular motions in opposite directions. The hypotrochoid equation in the complex $xy$-plane can be expressed as

$$z(\gamma) = x(\gamma) + i y(\gamma) = \tilde{R} \exp(i\gamma) + d \exp\left(-i\frac{\omega_2}{\omega_1}\gamma\right),$$

(3.1)

where $i$ is the imaginary unit, $\gamma$ is the curve parametrization parameter, $\tilde{R}$ is the radius of the counterclockwise rotation of frequency 1 rad/s, and $d$ is the radius of the clockwise rotation of frequency $\omega_2/\omega_1$. For more details on hypotrochoid curves and their parametric representations, please refer to the Appendix. Note that in the sperm motility framework, $\omega_1$ represents the sperm roll frequency while $\omega_2$ represents the counter-rotation flagellar frequency (Woolley, 1998). The shape of the hypotrochoid depends on the ratio between the frequencies $\omega_1$ and $\omega_2$, in particular, the number of singular points $n$ can be determined as

$$n = \frac{\omega_2}{\omega_1} + 1.$$

To better quantify the various head f-curve shapes reported in Figures 7 and 9 we follow the method reported in Woolley (1998) to approximate the head f-curve via a hypotrochoid curve, and the results of the approximation are reported in Figure 11 and in Table 3.2.3. More details on the approximation procedure can be found in the Appendix. Figure 11 shows the comparison between the simulation results for one full rotation of the head f-curve (colored curve) and the approximated hypotrochoid curve (black curve) in the case of a helical wave (left), and of a quasi-planar wave (right), considering the asymmetric coupling only A case. In Table 3.2.3 we report the fitted parameter values for the hypotrochoid in the case of helical and quasi-planar
waves and, for the three curvature-calcium coupling cases considered in Figures 7 and 9. The model results, the counter-rotation of the flagellum is significantly present only if a difference between the preferred amplitudes $A$ and $B$ is introduced. In our model, $A \neq B$ can be obtained in two
3 RESULTS AND DISCUSSION

Figure 10: *Quasi-planar wave.* Comparison at \( t = 10s \) between preferred (solid lines) and actual (dashed lines) configurations of the rod: curvature in the top panel and twist in the bottom panel for the case of no-coupling (No Ca, black) and in the case of asymmetric coupling only \( A \) (Ca asym \( A \), red).

Figure 11: *Helical vs Quasi-planar waves.* Comparison between simulation results for a full rotation of the head f-curve around the center of mass (colored line) and the corresponding hypotrochoid curve (black line) for the case of a helical wave (left) and a quasi-planar wave (right), considering the asymmetric coupling only \( A \) case.

ways: a baseline geometrical difference in the quasi-planar wave case where \( A_0 \neq B_0 \), and a calcium coupling difference in the \( Ca \ Asym \ A \) case, where only \( A \) varies with calcium and \( B \) is kept constant to its baseline value.

3.2.4 Comparison of helical & quasi-planar waves.

To investigate the fluid-structure interaction between the flagellum and the surrounding fluid, in Figure 12 we report the fluid velocity fields (black arrows) and pressure \((p)\) distributions at time \( t = 10s \). We choose either a horizontal or vertical plane containing the flagellum centerline in the case of helical and quasi-planar waves, accounting for the \( Ca \ Asym \ A \) coupling. The horizontal centerline plane is defined as the plane passing through the sperm centerline with normal orthogonal to the \( y \)-axis, while the vertical centerline plane is defined as the plane
Table 2: The fitted hypotrochoid parameter values varying the flagellar waveform and varying the calcium-curvature coupling condition. No-coupling (No Ca), asymmetric coupling (Ca asym A\&B) and asymmetric coupling only A (Ca asym A). †Value imposed a priori in order to obtain a circle.

| Wave        | Coupling case | $\bar{R}$ [µm] | $d$ [µm] | $\omega_1$ [rad/s] | $\omega_2$ [rad/s] | $n$  |
|-------------|---------------|----------------|----------|--------------------|--------------------|------|
| Helical     | No Ca         | 1.091          | 0.003    | 29.7               | 29.7               | 2.0† |
|             | Ca asym A\&B | 1.451          | 0.019    | 20.3               | 215.6              | 11.6 |
|             | Ca asym A    | 1.272          | 0.180    | 28.5               | 210.5              | 8.4  |
| Quasi-planar| No Ca         | 0.796          | 0.297    | 62.5               | 169.0              | 3.7  |
|             | Ca asym A\&B | 1.049          | 0.375    | 51.6               | 172.2              | 4.3  |
|             | Ca asym A    | 0.945          | 0.510    | 68.5               | 145.9              | 3.1  |

Figure 12: Helical vs Quasi-planar waves. Fluid velocity fields (black arrows) and pressure ($p$) distributions at time $t = 10s$ in the flagellum horizontal (left column) or vertical (right column) centerline planes in the case of a helical wave (top row) and a quasi-planar wave (bottom row), for the asymmetric coupling only A case. A filled in sphere is used to denote the head and swimming direction passing through the sperm centerline with normal orthogonal to the $z$-axis. The pressure distribution range is approximately $-40$ to $40$ gµms$^{-2}$ in the helical wave case, and from $-80$ to $80$ gµms$^{-2}$ in the quasi-planar wave case. In both wave cases, the velocity fields show vortices along the flagellum length, where the fluids rotates from regions of negative pressure to regions of positive pressure.

In Table 3.2.4 we report the values of head linear velocity, maximum actual curvature $\Omega^*$, and maximum distance from the centerline over the 1s interval form 9s to 10s in the case of no-coupling, asymmetric coupling and asymmetric coupling only A, for the helical and quasi-
Conclusions

This paper presents the first mathematical model that couples the 3D dynamics of the sperm flagellum and surrounding fluid, with the calcium dynamics inside the flagellum. The coupling is achieved by assuming that the flagellum preferred curvature and twist depend on the local calcium concentration. We compare the model results for 2D and 3D flagellar reference beat forms: planar, helical and quasi-planar. Linear velocities, forces and trajectories extracted from the model results are in agreement with experimental data and previously developed mathematical models [Olson et al., 2011; Simons et al., 2014; Smith et al., 2009; Woolley, 1998, 2003; Woolley & Vernon, 2001].

In particular, the planar swimmer results present (i) a more clear turning motion when asymmetric calcium coupling is considered, (ii) significantly higher linear velocity than the helical swimmer, in agreement with Chwang & Wu (1971), supporting the hypothesis that helical motion might be an important strategy for chemotaxis sampling [Guerrero et al., 2011; Su et al., 2012, 2013]. In the case of 3D flagellar beat forms, if an asymmetry between the flagellar amplitudes $A$ and $B$ is introduced, the flagelloid curves (f-curves) extracted from the

| Coupling case | Linear Velocity [μm/s] | Max. Curvature [1/μm] | Max. Distance [μm] |
|---------------|------------------------|------------------------|--------------------|
|               | Helical | Quasi-planar | Helical | Quasi-planar | Helical | Quasi-planar |
| No Ca         | 8.6    | 22.1       | 0.09    | 0.13       | 2.86    | 2.87       |
| Ca asym $A & B$ | 7.6    | 30.4       | 0.11    | 0.20       | 3.94    | 3.91       |
| Ca asym $A$   | 13.2   | 38.9       | 0.16    | 0.22       | 3.82    | 3.95       |

Table 3: Comparison of head linear velocity, maximum curvature, and maximum distance form the centerline for the various flagellar wave and calcium-curvature coupling conditions. No-coupling (No Ca), asymmetric coupling (Ca asym $A & B$) and asymmetric coupling only $A$ (Ca asym $A$).
model resemble hypotrochoid curves. In this paper we investigate two sources of asymmetry: a geometrical asymmetry in the quasi-planar wave case, and a calcium coupling asymmetry in the Ca Asym A case. The shape of the hypotrochoid f-curve, i.e. the number $n$ and the presence or absence of cusps and self intersections, varies with the choice of preferred flagellar amplitudes $A_0$ and $B_0$, and with the calcium coupling considered, and it is maintained along the length of the flagellum. We remark that hypotrochoid f-curves similar to the one extracted from the model results have been obtained in the experimental measurements reported by Woolley (1998, 2003).

Throughout the development of the model, several assumptions were made. We assumed a simplified sperm representation neglecting the head. Experiments have shown that headless mammalian sperm can still swim in fluid flows (Miki & Clapham, 2013). Moreover, since the head is small compared to the flagellum lengthscale, Ishimoto & Gaffney (2016) performed a dimensional analysis and showed that neglecting the head induces accurate angular velocity and linear velocity of the correct scale. Therefore, accounting for the head in the model should not significantly affect the overall sperm motility, however further studies are required to investigate the head effect on sperm trajectories and f-curves.

We also assumed that hyperactivated motility of mammalian sperm is associated with an evolving calcium concentration that is mediated by CatSper channels, ATP-ase pumps, and a calcium store (redundant nuclear envelope) where calcium release is coupled to the evolving concentration of inositol 1,4,5-trisphosphate or $IP_3$. However, in other species, different calcium signaling mechanisms might play an important role. For example, in sea urchin sperm, an increase in internal calcium is associated with an increase in internal cGMP (cyclic guanosine monophosphate) and the calcium channels could be CatSper or other voltage sensitive calcium channels (Darszon et al., 2008; Seifert et al., 2015; Wood et al., 2003). Hence, in order to
Figure 14: Sketch of a theoretical hypotrochoid curve (black) obtained by tracing a point $P$ attached to a small circle (blue) of radius $r$ rolling around the inside of a bigger circle (red) of radius $R$, where $d$ is the distance between the point $P$ and the center of the small circle. $\gamma$ is the angle formed by the horizontal axis and the center of the small rolling circle. The blue dashed line represents the trace of the center of the small circle, i.e. a circle of radius $\tilde{R} = R - r$. $R = 2$, $r = 0.5$ and $d = 0.7$.

study the effect of calcium dynamics on sperm motility in different species, the calcium model used here should be adapted to account for the species specific calcium signaling pathways and associated channels. We remark that, in addition to the CatSper signaling pathways related to hyperactivation in mammalian sperm, there are other mechanisms that could guide the sperm to the egg and change the flagellar waveform. These include chemotaxis, i.e. the effect of a chemical concentration gradient such as an egg protein in marine invertebrate sperm or progesterone in human sperm ([Lishko et al., 2011] [Wood et al., 2003]), rheotaxis, i.e. the effect of a background flow ([Miki & Clapham, 2013]), and thermotaxis, i.e. the effect of the fluid temperature gradient ([Boryshpolets et al., 2015]). It would be an interesting research direction to expand the model in the current study to include the effect of a background flow and an evolving gradient of chemoattractants or calcium in the fluid.

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Appendix - Hypotrochoid approximation

Hypotrochoid curves are usually defined as the curve created by tracing a point $P$ rigidly attached to a small circle of radius $r$ rolling around the inside of a bigger circle of radius $R$, where $d$ is the distance between the point $P$ and the center of the small circle, as sketched in Figure 14. The parametric equations for a hypotrochoid in the $xy$-plane can be expressed as

$$x(\gamma) = (R - r) \cos \gamma + d \cos \left(\frac{R - r}{r} \gamma\right), \quad y(\gamma) = (R - r) \sin \gamma - d \sin \left(\frac{R - r}{r} \gamma\right),$$

$(4.1)$
where $\gamma$ is the angle formed by the horizontal axis and the center of the small rolling circle. Equation (4.1) can be derived directly from (3.1), given

$$\tilde{R} = R - r$$
and
$$n = \frac{R}{r} = \frac{\omega_2}{\omega_1} + 1. \quad (4.2)$$

Hence, hypotrochoid curves can also be described as the trajectories of a point $P$ subjected to a movement composed of two circular motions in opposite directions: the center of the small circle is rotating around the origin with a counterclockwise circular motion of radius $\tilde{R} = R - r$ and frequency 1 rad/s (represented by the blue dashed line in Figure 14), and the point $P$, rigidly attached to the small circle, is rotating around the center of the small circle with a clockwise circular motion of radius $d$ and frequency $\omega_2/\omega_1$. As mentioned in Section 3.2.3, this last characterization of hypotrochoid curves can be directly interpreted in the sperm motility framework. Note that the curve corresponding to (3.1) is equivalent to the curve generated by a counterclockwise circular motion of radius $\tilde{R}$ and frequency $\omega_1$ together with a clockwise circular motion of radius $d$ and frequency $\omega_2$; $P$ is just going across them with a different speed as the parameter $\gamma$ varies.

Fixing $n$, the value of $d$ determines if singular points are present, i.e. cusps or self intersections (crunodes). We remark that in Section 3.2.3 we refer to $n$ as the number of singular points of the curve, however this is a slight abuse of notation since, fixing $n$, for some values of $d$ the curve does not present any singular point and can be described as a rounded approximation of a $n$-sided regular polygon. Note that $d$ admits positive and negatives values: at $\gamma = 0$, if $d > 0$ the point $P$ is chosen to the right of the small circle center, otherwise if $d < 0$ the point $P$ is chosen to the left. Moreover, the point $P$ can be chosen to be either inside ($|d| < r$), outside ($|d| > r$) or on the circumference of the small circle ($|d| = r$). The case of $d = r > 0$ corresponds to hypocycloid curves, while the limit case of $d = 0$ corresponds to a circle of radius $\tilde{R}$.

Fitting sperm trajectory data to hypotrochoid curves involves different mathematical challenges. Starting from finding the best minimization algorithm and initial value to approximate the three parameters, $(\tilde{R}, d, n)$ or $(R, r, d)$, that uniquely identify the curve. To our knowledge, there is only one result available in the literature on least-squares hypotrochoid curve fitting in Sinnreich (2016), where a method for a particular case of hypotrochoid curve is presented, i.e. rounded approximation of regular polygons with no cusps and no self-intersections. A further challenge comes form the fact that for each data point the corresponding $\gamma$ is also an unknown, since by definition $\gamma$ is in general not equal to the point polar angle $\theta$. We can relate $\theta$ and $\gamma$ using the inverse tangent of the ratio between the $y$ and $x$ coordinates of the data point. However, this correspondence is not one-to-one when self intersections are present in the hypotrochoid curve. For this reason, we chose to use the method reported in [Woolley (1998)] to approximate the simulation f-curves with a hypotrochoid curve. This method exploits the definition of hypotrochoid curve as two circular motions, and uses geometrical and physical principles to estimate the curve parameters without utilizing a minimization algorithm, and can be used for approximating any kind of hypotrochoid curves, including ones that exhibit self-intersections.

Given the head f-curves data points, we follow [Woolley (1998)] to find the approximating hypotrochoid curve as detailed below

1. starting at time $t = 9s$, the roll frequency $\omega_1$ is estimated as the frequency of a full rotation of the head around its center of mass;
2. $n$ is estimated as the ratio between $2\pi$ and the mean angular separation between two singular points over a full rotation;
3. the flagellar frequency is $\omega_2 = \omega_1(n - 1)$;

4. let $d_{min}$ and $d_{max}$ be respectively the average minimum and maximum distance from the center of mass over one rotation, then the remaining hypotrochoid parameters can be estimated as follows

$$d = \frac{d_{max} - d_{min}}{2}, \quad R = \frac{\omega_1 + \omega_2}{\omega_2} (d_{min} + d), \quad r = \frac{\omega_1}{\omega_2} (d_{min} + d).$$

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3D Sperm Motility: Calcium-Curvature Coupling

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