Stochastic and deterministic modelling of the cell-division cycle in primitive eukaryotes

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Abstract

Not much is yet known about the molecular details of the growth control mechanism for the start of DNA replication in eukaryotic cells. Here we simulate three models which describe different hypothesis for the size control in primitive eukaryotes. All the models are characterized by an antagonistic relationship between cyclin-dependent kinase (CDK) and anaphase promoting complex (APC), which degrades the cyclin partner of CDK.

We solve all the models deterministically using an ODE solver. Moreover, we derive a stochastic version of the first model, the primitive APC-CDK controller, and solve it by Monte Carlo methods. To our knowledge a stochastic version of this model has not been presented before.

Our results clearly illustrate the difference between the three different hypothesis on growth controlled cell-division in primitive eukaryotes. The results for the deterministic model are in full agreement with previous simulations in published articles. Also, our stochastic model gives results that are very close to the deterministic version.

Contents

1 Introduction

The cell division cycle is the process of a cell growing to a certain size, replicating its DNA and parting into two nearly identical daughter cells, which then repeat the process. In present day eukaryotes, the division-cycle is controlled by a complex network of chemical reactions, which coordinate cell growth and DNA replication, ensure that each daughter cell receives precisely her share of DNA and prevent overlapping rounds of DNA replication. Here, we explore three models of the cell-division cycle in primitive eukaryotes, put forward in a paper by Novak et al [1]. Although knowledge about the cell division cycle for primitive eukaryotes is somewhat lacking, it is reasonable to assume it to have been considerably simpler than for present day eukaryotes.

A common way to present the cell cycle is to divide it into the following distinct phases [2]:

G1-phase This is the start and finish of the cell division cycle, where the cell is resting and growing. During this phase, the cell has not committed to the replication-division process. The cell has to grow to a certain size before replication can be initiated.

S-phase In this phase, DNA is replicated. Each chromosome consists of a pair of sister chromatids held together by tethering proteins.

G2 A gap phase during which the division cycle is in rest.

M-phase

M-meta The chromosomes are aligned on the metaphase spindle with sister chromatids attached to different poles on the membrane sites. The chromatids will be attached until the preparation for the M-anaphase is completed.

M-ana The tether proteins are removed so the sister chromatids can be segregated to opposite sides. The cell-division is completed when the cell divides into two daughter cells, which both enter into the G1 phase.

In the paper by Novak et al [1], the division cycle is modelled as antagonistic interactions between cyclin-dependent kinase (CDK) and anaphase promoting complex (APC), which labels cyclin for proteolysis. In the first model (equations (1a)- (1e)), a simpler version of the cell cycle is considered, where the S, G2 and M phases are combined into one phase and the mathematical model alternates between two steady states, the G1 phase and the S/M

phase. In the G1 state, APC is high, CDK activity is low and DNA is unreplicated. In the S/M state APC is low, CDK activity is high and DNA is replicated. The transition from G1 to S/M is driven by cell growth and the reverse transition from S/M to G1 is driven by completion of DNA synthesis and division of the cell.

The second model (equation (1b)-(1e) and (3a)-(3c)) introduces a cyclin dependent kinase inhibitor (CKI), which blocks the activity of CDK in the G1 phase. Here, the size control over the start of DNA replication can either be attributed to the inhibition of the APC by CDK or the degradation of CKI by CDK.

In the third model (equations (1b)-(1e) and (4a)-(4d)), the S and M phases are separated by a G2 phase. The size control over the start of DNA replication is due to the transition from G1 to S/G2 or to the transition to S/G2 to M.

In the paper by Novak et al [1], the models are implemented in a deterministic way. As well as implementing the three models deterministically, we have used a stochastic approach for the first model. The remainder of this report is organized as follows. In the next section, we explain the various components of the cell-division cycle and how they interact in different phases of the cycle. After that, we present the deterministic models, together with the results. In section 4 we explain the stochastic modelling process and the result we obtain. Finally, the conclusions are summarized and discussed.

2 Components of the cell-division cycle

In all the models considered in this report, cell growth initiates the transition from the G1 phase to the S-phase, where DNA replication is initiated. It is known that such a size surveillance mechanism is present in modern day eukaryotes (the molecular details of which are yet unknown) and a similar mechanism must have been in place in primitive eukaryotes. When the cell size is modelled, we assume that nuclear volume remains constant while the cell size increases. [1]

Cell cycle events are controlled by a network of molecular signals whose central components are CDK. CDK need cyclin partners to be active and to recognize targets. When active, CDK phosphorylate many target proteins involved in cell cycle events.

CDK plays a central role in initiating and regulating the DNA replication process. In order for the replication to be initiated, the replication origins

must be equipped with a full complement of protein subunits and then CDK is needed to activate the replication. CDK has another important task to fulfill at the start of the cycle as it disables a necessary component of the protein subunits, preventing the replication from starting more than once. [2]

When the chromosomes are aligned on the spindle in M-metaphase, a signal activates the APC which in turn initiates degradation of the glue binding the chromatids together along with the cyclin component of CDK. Then the sister chromatids separate, the cell divides and the daughter cells are ready to begin a new cycle. This final part of the cycle is also coordinated by a surveillance mechanism. This makes sure that the DNA is fully replicated and aligned on the metaphase plate before the separation in the anaphase and before the signal to activate the APC is generated. [2]

Here, we suppose that APC activation is accomplished by an activator, ACT, which opposes the inhibitory effect of CDK on APC. ACT is continuously synthesized and degraded by APC. Newly synthesized ACT is called ACT_P (or less active ACT) and must take part in some chemical reactions before turning into fully active ACT. [1]

Chemical reactions in biology often has some reactants which serve as catalysts or inhibitors. They are usually proteins and such proteins are called enzymes. APC activation is described by Michaelis-Menten (MM) rate laws. MM kinetics describe the rate of enzyme mediated reactions for many enzymes. These kinetics are only valid when the concentration of substrates is higher than the concentration of enzymes. To determine the maximum rate of an enzyme mediated reaction, the substrate concentration is increased until a constant rate of product formation is achieved. This is the maximal velocity of an enzyme. When the substrate concentration is increased, the enzyme is approaching its maximum speed without reaching it. This means that no concentration for a this maximal velocity can be given, instead the value of the enzyme is defined by the concentration at the halved max velocity ($\frac{V_{max}}{2}$). This is called the MM constant. If the substrate is low, reactions very rarely take place. [3]

3 Three Deterministic Models

In the following subsections we present three different deterministic models of the cell-division cycle in primitive eukaryotes (see [1]). The two latter models are both based on the first one, the primitive APC-CDK controller. The main difference between the models is that different mechanisms are assumed to initiate DNA replication.

3.1 The primitive APC-CDK controller

In the first deterministic model, the primitive APC-CDK controller, the mechanism for the control of the cell-division cycle in primitive eukaryotes is modelled as an antagonistic relationship between APC and CDK. CDK inactivates APC, while APC degrades the cyclin subunit of cyclin-CDK dimers. CDK activity is lost by cyclin degradation at a rate dependent on the distribution of APC, which comes in two forms, more active (APC) and less active (APC_P). At the beginning of the G1 phase, APC activity is high and CDK low. As the cell grows the concentration of CDK increases and APC is inactivated. When CDK activity has reached a certain level, DNA replication is initiated. When the DNA is fully replicated and aligned on the metaphase plate the signal to activate APC is given. When the cell division is complete, APC activity is still high while CDK activity is minimal. The model is given by equations (1a) - (1e). [1]

$$\frac{d[CDK]}{dt} = k_1 \cdot \text{size} - \left[k_2'(1-[APC]) + k_2'' \cdot [APC]\right] \cdot [CDK] \tag{1a}$$

$$\frac{d[APC]}{dt} = \frac{(k_3' + k_3'' \cdot [ACT])(1-[APC])}{J_3 + 1 - [APC]} - \frac{(k_4' + k_4'' \cdot [CDK]) \cdot [APC]}{J_4 + [APC]} \tag{1b}$$

$$\frac{d\text{size}}{dt} = \mu \cdot \text{size} \tag{1c}$$

$$\frac{d[ACT]_T}{dt} = k_{as} - \left[k_{ad}'(1-[APC]) + k_{ad}'' \cdot [APC]\right] \cdot [ACT]_T \tag{1d}$$

$$\frac{d[ACT]}{dt} = k_{aa}([ACT]_T - [ACT]) - k_{ai} \cdot [ACT] - [k_{ad}'(1-[APC]) + k_{ad}'' \cdot [APC]] \cdot [ACT]. \tag{1e}$$

In the equations above, [CDK] is the concentration of cyclin-CDK dimers in the cell nucleus and [APC] is the fraction of total APC that is active, where APC_T is the total number of APC molecules. ACT is a hypothetical activator of the APC. $[\text{ACT}]_T$ is the total concentration of ACT.

Despite extensive research, not much is yet known about size control in either primitive or present day eukaryotes. The mechanism proposed in this

model is that cyclin molecules are synthesized in the cell cytoplasm at a rate proportional to the total protein synthetic capacity of the cell ($k1 \cdot size$), bind rapidly to free CDK subunits and are sequestered into the nucleus. Based on the assumption that nuclear volume remains constant as the cell grows, the concentration of active CDK in the nucleus increases. When the activity of CDK reaches a critical value, DNA replication is initiated. [1]

In equation (1c), [size] refers to some appropriate measure of cell size (e.g. ribosome number). Interdivision time or mass doubling time is $= \frac{(ln2)}{\mu}$, where μ is the cell growth rate. Throughout the report, μ =0.0058 min^{-1}, which means that one cell-division cycle will take 120 minutes.

The parameters k_2' and k_2'' are the enzymatic turnover numbers characterizing the less- and more-active forms of APC. As we have already stated, the activation and inactivation of APC is described by Michaelis-Menten law. We have that k_3'' and k_4'' are turnover numbers for activation catalyzed by ACT and for inactivation catalyzed by CDK and where k_3' and k_4' are V_{max} values for the background rates of activation and inactivation. The Michaelis-Menten constants, J_3 and J_4, are assumed to be small relative to APC_T.

The parameter k_{ai} has two contributors, one describing the inactivation of ACT by DNA replication forks and one expressing the inactivated effect of misaligned chromosomes. It is important for the model that k_{ai} is sufficiently large just after the start of DNA replication and then drops back to a low value when the cell cycle reaches its end. [1]

The results of our simulation of the model (equation (1a)-(1e)), using Matlab and ODE45, are given in Figure 1. For parameter values that were used in the simulation, see Table 1 in Appendix A. The Figure displays the growth controlled division cycle in the primitive APC-CDK mechanism over two cell-division cycles. Here we can see how the size of the cell changes with time and observe the interaction of CDK and APC, which is controlled by cell growth. Also, the relationship between ACT and APC can be viewed in the Figure. ACT is the activator of APC and before the ratio of active APC starts to grow at the end of the cell cycle, the concentration of ACT must increase greatly.

The Figure also illustrates that the inter-division time for the cell cycle always is exactly the same as mass doubling time, as stated above. This has to be true for balanced growth and division, since the mother cell divides precisely in half at the end of the division cycle. Hence the time of one cell-division cycle must be equal to the mass doubling time.

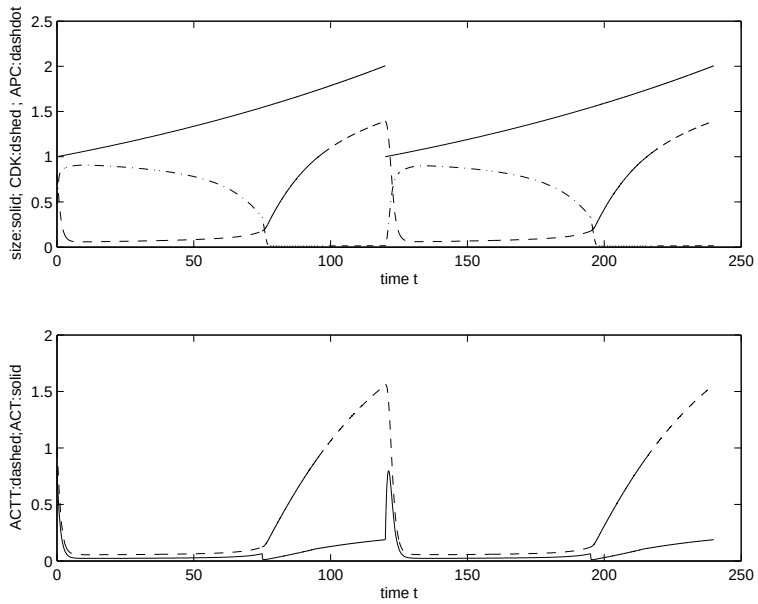

Figure 1: The growth controlled division cycle in the primitive APC-CDK mechanism, simulations of ODE-system (1a)- (1e). For parameter values see Table 1. [1]

Much can be learned about the model by studying its phase-plane portrait. Where the rate of cyclin synthesis is exactly balanced by the rate of cyclin degradation, $\frac{d[CDK]}{dt}$ is zero. The locus of such points in the phase plane is called the CDK nullcline and is described by equation (2a). Equation (2b) represents the APC nullcline.

$$[CDK] = \frac{k_1 \cdot \text{size}}{k_2'(1-[APC]) + k_2'' \cdot [APC]} \tag{2a}$$

$$[CDK] = \frac{(k_3' + k_3'' \cdot [ACT])(1-[APC])}{k_4'' \cdot [APC]} \cdot \frac{J_4 + [APC]}{J_3 + 1 - [APC]} - \frac{k_4'}{k_4''}. \tag{2b}$$

In Figure 2a, the APC and CDK nullclines are plotted for different cell sizes and concentrations of ACT. The two nullclines intersect at three steady states. The first state corresponds to the G1 phase of the cell-division cycle with APC active and CDK inactive (stable node). The second state corresponds to the S/M phase of the cycle with APC inactive and CDK active (stable node). In the third state, both APC and CDK are active (unstable

saddle point). A steady state is called stable if the system returns to the steady state in response to any small perturbation of CDK and APC, otherwise it is unstable. In this scenario the model needs a strong signal to move between the G1 and S/M steady states.

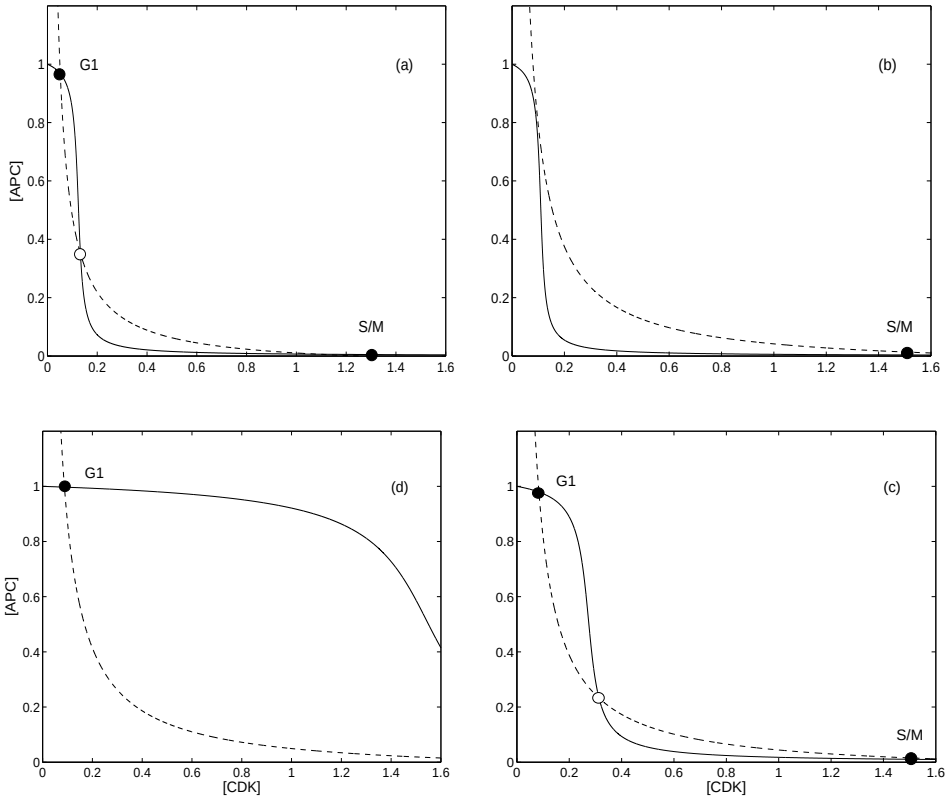

Figure 2: APC and CDK nullclines plotted for different cell sizes and values of ACT. • corresponds to a stable node and ∘ to an unstable node. a) size=1 and ACT=0.05 b) size=1.6 and ACT=0.05 c) size=1.65 and ACT=1.75 d) size=1.75 and ACT=1.

At the beginning of the cell-division cycle, the system stays in the stable point representing the G1 phase, see Figure 2a. As size increases the CDK nullcline moves up, making the G1 node and the saddle point coincide and disappear, see Figure 2b. When the cell has grown such that size $>$ size$_{critical}$ only one stable steady state, S/M, remains to which the system must tend. When the cell grows large enough, the G1 state disappears and S/M is the

only stable state available.

When the concentration of ACT increases and activates APC, the APC nullcline shifts right and the three steady states are created again, see Figure 2c. A further increase in ACT and cell growth shifts the APC nullcline even further right, the S/M steady state is lost and the cell is pushed back to the G1 state, see Figure 2d. This marks the finish of the cell-division cycle.

3.2 Model with CDK inhibitor

As we have already stated, the mechanism for size control proposed in the first model is completely hypothetical. An alternative approach is to use evidence from fission yeast, which indicates that cell size at the start of DNA replication is related to the activity of a cyclin-dependent kinase inhibitor, CKI. CKI binds to cyclin-CDK dimers in the G1 phase and blocks their activities. Here, the cell is held in G1 with low CDK activity by synthesizing CKI. When CKI is degraded, pre-formed CDK is unmasked to initiate DNA synthesis. In this model, just like the first one, the division-cycle is growth controlled. In this case, however, size control over the start of DNA replication can either be attributed to the inhibition of the APC by CDK or the degradation of CKI by CDK. This model is given by equation (1b)-(1e), together with equation (3a)-(3c).

$$\begin{aligned}\frac{d[CDK]}{dt} &= k_1 \cdot \text{size} - \left[k_2'(1-[APC]) + k_2'' \cdot [APC]\right] \cdot [CDK] + \\ &+ (k_6' + k_6'' \cdot [CDK]) \cdot [TRI] - l_1 \cdot [CDK] \cdot [CKI] + \\ &+ l_2 \cdot [TRI]\end{aligned} \tag{3a}$$

$$\begin{aligned}\frac{d[CKI]}{dt} &= k_5 + [v_2(1-[APC]) + v_2'' \cdot [APC]] \cdot [TRI] - \\ &- (k_6' + k_6'' \cdot [CDK]) \cdot [CKI] - l_1 \cdot [CDK] \cdot [CKI] + \\ &+ l_2 \cdot [TRI]\end{aligned} \tag{3b}$$

$$\begin{aligned}\frac{d[TRI]}{dt} &= -[v_2'(1-[APC]) + v_2'' \cdot [APC]] \cdot [TRI] - \\ &- (k_6' + k_6'' \cdot [CDK]) \cdot [TRI] + \\ &+ l_1 \cdot [CDK] \cdot [CKI] - l_2 \cdot [TRI]\end{aligned} \tag{3c}$$

Here, [CDK], [CKI] and [TRI] denote the concentrations of cyclin-CDK dimers, CKI monomers and cyclin-CDK-CKI trimmers, respectively. ν_2'' is

the rate constant for degradation of cyclin from trimers, which should be less than k_2''. For parameter values used in the simulation of the model, see Table 1.

Figure 3 illustrates the primitive APC-CDK-CKI mechanism. One can see that this cycle also is growth controlled by the mass doubling time. The choice of parameter values is very important here, it will determine which critical size is operative and which is cryptic. For example, if the cell first reaches the critical size necessary to destabilize the CKI-CDK interaction, size control will be operative and APC-CDK size control will be cryptic.

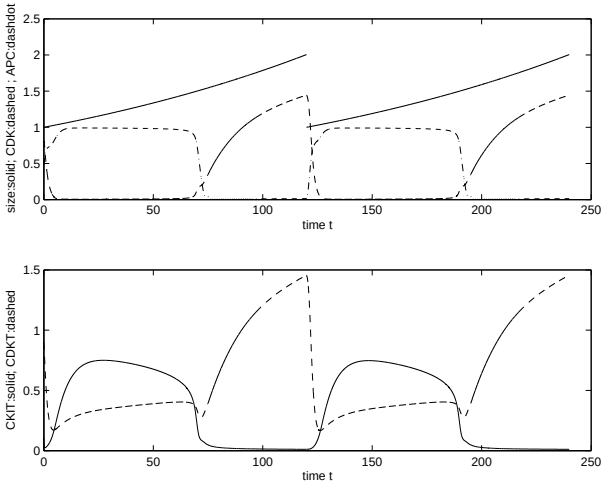

Figure 3: The growth controlled division cycles in the primitive APC-CDK-CKI mechanism, simulation of the system in equation (1b)-(1e) and (3a)-(3c). The size control over the start of DNA replication can be attributed either by the inhibition of APC by CDK or the degradation-inducing inactivation of CKI by CDK. For parameter values see Table 1. [1]

3.3 G2-phase added to the model

In the first two models, no distinction is made between the initiation of the S and M phases. To separate S and M phases we introduce a G2 phase, it is necessary to delay the onset of mitosis until some time after DNA replication is complete. In [1], a model is proposed where these phases are separated.

The full model is given by equation (1b) - (1e), together with equations (4a) - (4d) with the following two changes. In equation (1b), [CDK] is replaced by MPF.[1] Also, CDK is now an activator of ACT as well as an

[1]Here, MPF is the weighted activity of the unphosphorylated and phosphorylated

inhibitor of the APC.

$$\frac{d[CDK]_T}{dt} = k_1 \cdot \text{size} - \left[k_2'(1-[APC]) + k_2'' \cdot [APC]\right] \cdot [CDK]_T \quad (4a)$$

$$\frac{d[CDK]_A}{dt} = k_1 \cdot \text{size} - \left[k_2'(1-[APC]) + k_2'' \cdot [APC]\right] \cdot [CDK]_A - \quad (4b)$$
$$-k_{wee} \cdot [CDK]_A + k_{c25}([CDK]_T - [CDK]_A)$$

$$\frac{d[Wee1]}{dt} = \frac{k_{wr}(1-[Wee1])}{J_{wr}+1-[Wee1]} - \frac{k_w \cdot MPF \cdot [Wee1]}{J_w + [Wee1]} \quad (4c)$$

$$\frac{d[Cdc25]}{dt} = \frac{k_{25} \cdot MPF \cdot (1-[Cdc25])}{J_{25}+1-[Cdc25]} - \frac{k_{25r} \cdot [Cdc25]}{J_{25r}+[Cdc25]}, \quad (4d)$$

where

$$\begin{aligned} [CDK]_T &= [CDK]_A + [CDK]_P \\ k_{wee} &= v'_{wee}(1-[Wee1]) + v''_{wee} \cdot [Wee1] \\ k_{c25} &= v'_{c25}(1-[Cdc25]) + v''_{c25} \cdot [Cdc25]. \end{aligned}$$

[Wee1] and [Cdc25] represent the active fractions of the Wee1 and Cdc25 enzymes. Activation and inactivation of Wee1 and Cdc25 is accounted for by equations (4c) and (4d). Here, J_w and J_{wr} represent the Michaelis constants relative to the total concentrations of [Wee1] and [Cdc25], which remain constant throughout the cycle.

In Figure 4, the growth-controlled division cycle in the primitive APC-CDK_T-CDK_A mechanism is illustrated. The size control can operate either at the transition from G1 to S/G2 or at the transition from S/G2 to M. The parameters are set so that G1 size control is cryptic, for parameter values see Table 1.

4 The Stochastic Model

In section 3.1 we described a deterministic model for the primitive APC-CDK controller. Instead of writing the model as ordinary differential equations it

forms of cyclin-CDK, with $MPF = [CDK]_A + \alpha \cdot ([CDK]_T - [CDK]_A)$ and $[CDK]_T = [CDK]_A + [CDK]_P$. In our simulations we let $\alpha = 0.06$. CDK_T is the total concentration of CDK.

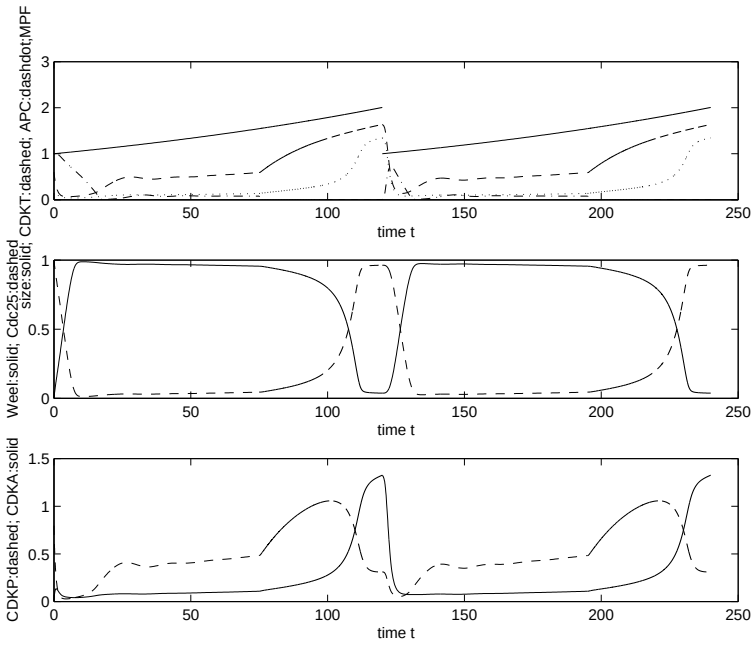

Figure 4: The S- and M-phases are separated by introduction of the G2 phase. The size control can operate either at the transition from G1 to S/G2 or at the transition from S/G2 to M. The parameters are set so that G1 size control is cryptic. [1]

can be converted to a system of chemical reactions and be simulated in a stochastic manner. Below, we introduce a stochastic version of the primitive APC-CDK controller that we derive from equations (1a) to (1e) and implement. To our knowledge, a stochastic version of the model has not been presented before.

The first step of the stochastic modelling procedure was to convert the ordinary differential equations (1a) - (1e) into chemical reactions. Note, that we have assumed that the cell grows deterministically, as described by equation (1c). From the four ODEs we derived 14 chemical reactions, equations (5) - (18), with 5 reactants; CDK, APC, APC_P, ACT_P and ACT_P. The reaction constants are displayed above the reaction arrows.

$$\xrightarrow{k_1 \cdot size \cdot vol} CDK \tag{5}$$

$$CDK + APC_P \xrightarrow{\frac{k_2'}{APC_T}} APC_P \tag{6}$$

$$CDK + APC \xrightarrow{\frac{k_2''}{APC_T}} APC \tag{7}$$

$$APC_P \xrightarrow{\frac{k_3'}{J_3+1-\frac{APC}{APC_T}}} APC \tag{8}$$

$$APC_P + ACT \xrightarrow{\frac{k_3''}{J_3+1-\frac{APC}{APC_T} \; vol}} APC + ACT \tag{9}$$

$$APC \xrightarrow{\frac{k_4'}{J_4+\frac{APC}{APC_T}}} APC_P \tag{10}$$

$$APC + CDK \xrightarrow{\frac{\mathrm{k_4''}}{\mathrm{J_4+\frac{APC}{APC_T}} \; \mathrm{vol}}} APC_P + CDK \tag{11}$$

$$\xrightarrow{k_{as} \cdot vol} ACT_P \tag{12}$$

$$ACT_P + APC_P \xrightarrow{\frac{k_{ad}'}{APC_T}} APC_P \tag{13}$$

$$ACT_P + APC \xrightarrow{\frac{k''_{ad}}{APC_T}} APC \tag{14}$$

$$ACT_P \xrightarrow{k_{aa}} ACT \tag{15}$$

$$ACT \xrightarrow{k_{ai}} ACT_P \tag{16}$$

$$ACT + APC_P \xrightarrow{\frac{k'_{ad}}{APC_T}} APC_P \tag{17}$$

$$ACT + APC \xrightarrow{\frac{k''_{ad}}{APC_T}} APC. \tag{18}$$

For the numerical simulation of the stochastic model we follow a procedure described in a paper by D.T. Gillespie [4]. In order to apply the methodology presented in that paper, the chemical species cannot be represented in terms of concentrations, but must be written as the number of molecules of the species in question. This conversion was made for the reactions above. In the deterministic model [APC] stands for the percentage of total APC (APC_T) which is active. Thus, APC is replaced by $\frac{APC}{APC_T}$ when the chemical reactions are derived. As can be seen, APC_T becomes a part of the reaction constants. The same procedure is followed with $[APC]_P$.[2] APC_T is assumed to be a constant number of molecules. The parameter *vol* in the reaction constants stands for the volume of the cell nucleus and is assumed to remain constant throughout the cell division cycle. Other parameters which occur as a part of the reaction constants are as described in section 3.1 and their values can be found in Table 1.

The basic idea for the computational procedure described in [4] is to use Monte Carlo techniques to simulate the stochastic process described by the reaction probability function;

$$\begin{aligned} P(\tau,\mu) = {} & \text{probability at time } t \text{ that the next reaction} \\ & \text{will occur in the time interval } (t+\tau, t+\tau+d\tau) \\ & \text{and will be a reaction of type } \mu, \end{aligned}$$

with

$$P(\tau,\mu) = h_\mu c_\mu \cdot \exp(-\sum_{\nu=1}^{14} h_\nu c_\nu \tau). \tag{19}$$

[2] $[APC]_P = 1 - [APC]$

Here, c_μ is the reaction constant for reaction μ and h_μ stands for the number of combinations of molecules that can take part in reaction μ. Our system consists of chemical reactions of the type

$$* \longrightarrow \text{reaction products} \tag{20a}$$
$$S_j \longrightarrow \text{reaction products} \tag{20b}$$
$$S_j + S_k \longrightarrow \text{reaction products} \tag{20c}$$

where (20a) is a spontaneous reaction. In general, h_μ is a function of the number of molecules of the reactant species taking part in reaction μ. For our system, we have;

$$h_\mu = 1 \qquad \text{for type (20a) reactions} \tag{21a}$$
$$h_\mu = X_j \qquad \text{for type (20b) reactions} \tag{21b}$$
$$h_\mu = X_j X_k \qquad \text{for type (20c) reactions.} \tag{21c}$$

The Monte Carlo step consists of generating a pair (τ,μ) according to the joint probability function (19). We employed a technique called the first-reaction method. In this method a so-called tentative reaction time is calculated for all the 14 chemical reactions;

$$\tau_\nu = \frac{1}{h_\nu c_\nu} \cdot \ln \frac{1}{r_\nu}, \tag{22}$$

where r_ν is a uniformly distributed random number on the unit interval. From these tentative reactions we choose the reaction which will happen first. Thus, in each step the time t is advanced by τ_μ and reaction μ is performed. This procedure is repeated until either the stopping time is reached or no reactants remain.

Figure 5 displays the results of one simulation of the stochastic model described by the chemical reactions (5)-(18). Other simulations yields similar results. The Figure illustrates the growth controlled division cycle in the primitive APC-CDK mechanism. The behavior of the model is clearly similar to the deterministic model and the antagonistic relationship between APC and CDK is apparent. The fluctuations in the figure are caused by the stochastic simulation process.

Figure 6 illustrates how often the different reactions occur over a simulation of slightly more than two cell-division cycles, or 280 minutes. It is clear from the figure that the number of times that the reactions happen differs greatly. For example, reaction (10) never takes place since its reaction constant is zero, see Table 1, and hence the probability of that reaction ever

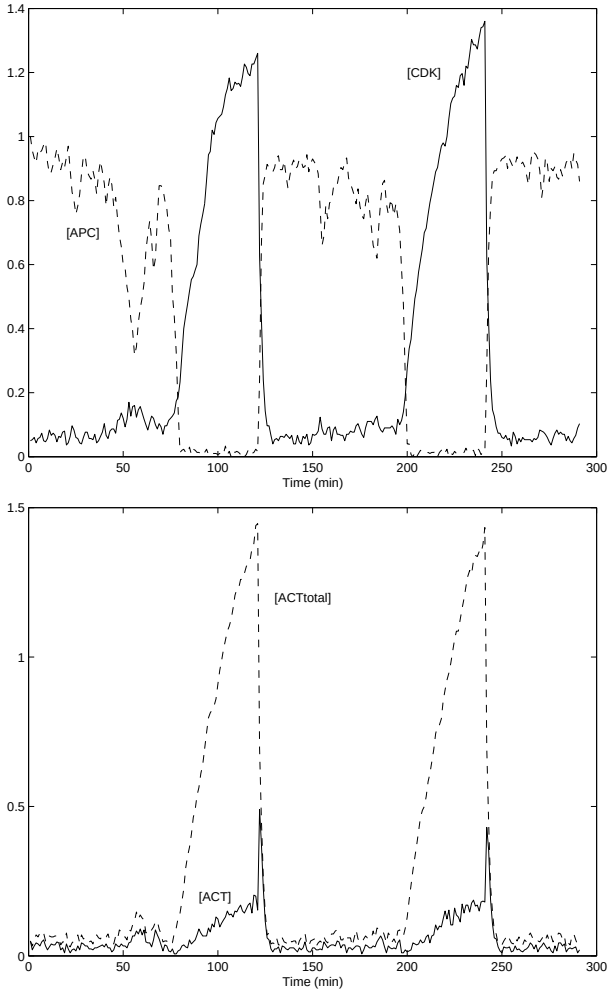

Figure 5: The results of one simulation of the stochastic model (5)-(18) over slightly more than two division cycles, or 280 minutes. The concentration of CDK and ratio of APC relative to APC_T are illustrated in the top plot and the concentration of ACT and ACT_T in the bottom plot.

occurring is also zero. Reactions (13) and (17) do not happen very often, which is due to the fact that their reaction constant is very small which affects the probability of them to occur. However, as we have mentioned before, the probability for a reaction also depends on the number of possible combinations of molecules for the chemical species taking part in the reaction.

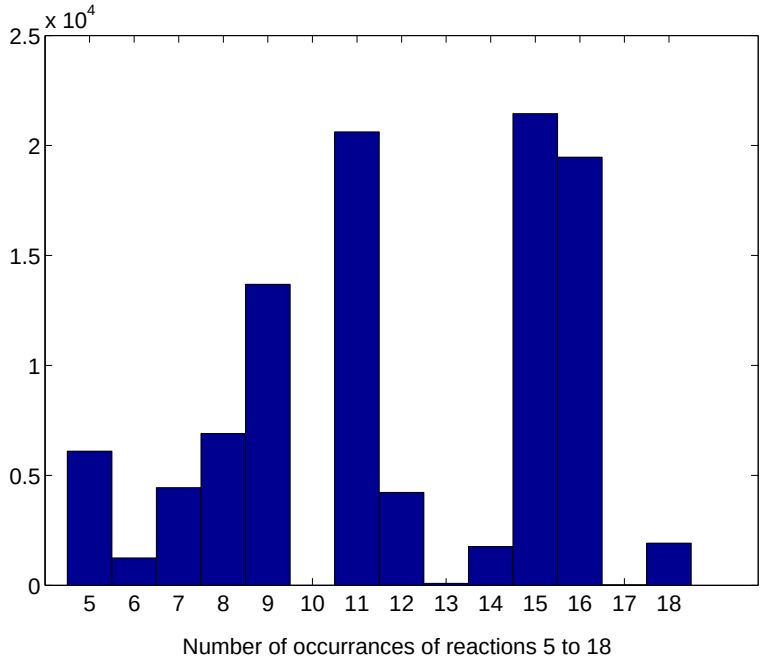

Figure 6: The figure illustrates how many times the different reactions (equation (5)-(18)) take place in 280 minutes, which represents slightly more than two cell-division cycles.

5 Discussion and conclusions

We have presented and solved three different models for the cell-division cycle in primitive eukaryotic cells, proposed in [1]. All the models are characterized by an antagonistic relationship between cyclin-CDK and APC, which labels the cyclin for proteolysis. The difference between the models is that each of them represents a different hypothesis for the growth control mechanism over start of DNA replication. In the first and most simple model, the primitive APC-CDK controller, DNA replication starts when the concentration of CDK in the cell nucleus has reached a critical level. In the second

model, size control can either be attributed to the inhibition of APC by CDK or to the degradation-inducing phosphorylation of CKI by CDK. The third model introduces a G2 phase separating the S and M phases. In this model size control can operate either at the transition from G1 to S/G2 or at the transition from S/G2 to M. Not much is known about size control in either present day or primitive eukaryotes. However, the hypothesis represented by these models can be used as a basis for further research into that area.

We used two different methods to solve the models, a deterministic and a stochastic approach. We applied the deterministic procedure to all the three models where we used ODE45, a Matlab ordinary differential equation solver, to solve the equations. We wrote our own Matlab program to solve the stochastic model and followed a Monte Carlo computational procedure described in [4]. We only used the stochastic solution process for the first model, the primitive APC-CDK controller.

The main advantage of the stochastic version of the model is that it does not require an ODE solver and is therefore much easier to implement and work with. Also, it is advantageous to have two different ways in which the model can be simulated.

The results that we obtained from the models were according to expectations and matched those presented in [1]. The results of the stochastic model are, in essence, the same as for the deterministic model, although they are characterized by a certain variability which is normal for all stochastic models.

Our most difficult and challenging problem was to construct the stochastic model. Especially to convert the ODE's into chemical reactions and finding the reaction constants. To our knowledge, a stochastic version of this model has not been constructed before. Since the results of our stochastic model are in agreement with the deterministic model, we are confident that we have built the stochastic model correctly.

In sum, we are pleased with the work that we have done on the simulation of the three models and the construction of the stochastic version of the primitive APC-CDK controller. There are many possible ways to extend this research, two of which we are particularly interested in. The first one is to examine the behavior of the models for parameter values that are different from those that we have tried. The second is to create a stochastic version of the second and third model as well. However, in order to provide

a contribution to the biological theory represented by the models, a much more extensive knowledge of cellular biology is needed.

A Parameters

Table 1: The parameter values in the different models.

Parameter	Figure 1 & 5	Figure 3	Figure 4
k_1	0.05	0.05	0.05
k_2'	0.05	0.05	0.05
k_2"	1	0.05	0.05
k_3'	0.1	0.001	0.02
k_3"	3	0.05	2
k_4'	0	0.05	0.05
k_4"	2	0.05	0.05
k_{as}	0.05	0.05	0.05
k_{ad}'	0.005	0.05	0.05
k_{ad}"	1	0.05	0.05
k_{aa}	1	0.05	0.05
μ	0.0058	0.0058	0.0058
J_3	0.05	0.05	0.01
J_4	0.05	0.05	0.01
v_2'	-	0.05	-
v_2"	-	0.15	-
k_5	-	0.15	-
k_6'	-	0.15	-
k_6"	-	9	-
l_1	-	200	-
l_2	-	1	-
k_{aa}'	-	-	0.001
k_{aa}"	-	-	1
$k_w = k_{25}$	-	-	0.5
$k_{wr} = k_{25}$	-	-	0.2
k_{wee}'	-	-	0.01
k_{wee}"	-	-	0.8
k_{r25}'	-	-	0.02
k_{r25}"	-	-	0.5
J_w	-	-	0.1
J_{wr}	-	-	0.1
J_{25}	-	-	0.1
J_{25r}	-	-	0.1
α	-	-	0.06

References

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