Fig. S1. Detailed scheme for the model of the cyclin/Cdk network driving the mammalian cell cycle. The model includes the four main cyclin/Cdk complexes, which control cell-cycle progression. The model also incorporates the pRB/E2F pathway, which controls progression or arrest in the cell cycle. At the beginning of the cell cycle, the GF promotes the synthesis of cyclin D (see details in Fig. S2A). Cyclin D can form a complex with the kinase subunit Cdk4–6. The active forms of cyclin D/Cdk4–6 and cyclin E/Cdk2 ensure progression in G1 and elicit the G1/S transition (Fig. S2B), by phosphorylating and inhibiting pRB. The inhibition of pRB ensures the activation of the transcription factor E2F that allows cell-cycle progression by promoting the synthesis of G1 cyclins. During S and G2 (Fig. S2C), cyclin A/Cdk2 inhibits, by phosphorylation, the Cdh1 protein that promotes the degradation of cyclin B. The negative feedback loops exerted, via Cdc20 activation, by cyclin B/Cdk1 on itself and cyclin A/Cdk2 (Fig. S2D), and the negative feedback loop exerted by cyclin A/Cdk2 on E2F (Fig. S2A) allow the reset of the cell cycle and the start of a new round of oscillations. Inhibitory phosphorylation by the kinase Wee1 and activating dephosphorylation by the Cdc25 phosphatases regulate the activity of Cdk1 and Cdk2. The activity of the cyclin/Cdk complexes can also be regulated by reversible association with the protein inhibitor p21 or p27 (see section 1 of SI Appendix for more details).
regulated by phosphorylation–dephosphorylation. A Cdc25 phosphatase (Pb) activates cyclin B/Cdk1 through dephosphorylation, whereas the kinase Wee1 inactivates it through phosphorylation. A positive feedback loop is again present between cyclin E/Cdk2 and its Cdc25 phosphatase (Pe), given that cyclin E/Cdk2 activates Cdc25 by phosphorylation, whereas Cdc25 activates cyclin E/Cdk2. The cdk E/Cdk2 complex can be also regulated by a reversible formation of a complex with the cdk inhibitor p27/p21. The accumulation of cyclin E/Cdk2 leads to phosphorylation of its inhibitor p21/p27. This phosphorylation brings about the specific degradation of p21/p27, under control by the Skp2 protein, which belongs to the proteasome. Skp2 also governs the degradation of cyclin E, which ensures that the activity of cyclin E/Cdk2 decreases at this phase of the cycle. (C) Detailed scheme for the cyclin A/Cdk2 module controlling the S-G2 transition. The synthesis of cyclin A is induced by E2F and inhibited directly and indirectly by pRB and pRBp. Like cyclin E, cyclin A can form a reversible complex with Cdk2. The activity of this complex can be regulated by phosphorylation–dephosphorylation; the Cdc25 phosphatase Cdc25 activates the complex cyclin A/Cdk2 by dephosphorylation, whereas the kinase Wee1 inactivates it by phosphorylation. A positive feedback loop exists between cyclin E/Cdk2 and its Cdc25 phosphatase (Pe), given that cyclin E/Cdk2 activates Cdc25 by phosphorylation, whereas Cdc25 activates cyclin E/Cdk2. The cdk E/Cdk2 complex can be also regulated by a reversible formation of a complex with the cdk inhibitor p21/p27. Cyclin A/Cdk2 controls the S-G2 transition by eliciting the phosphorylation and subsequent degradation of the transcription factor E2F (see A). Cyclin A/Cdk2 and cyclin B/Cdk1 both phosphorylate and thereby inhibit the protein Cdk1. This inhibition leads to the accumulation of cyclin B and to the G2/M transition (see D). (D) Detailed scheme for the cyclin B/Cdk1 module controlling the G2/M transition. In the preceding module (see C), cyclin A/Cdk2 inhibits by phosphorylation the protein Cdk1. This protein cannot anymore promote the degradation of cyclin B, so that the level of cyclin B increases. Cyclin B reversibly forms a complex with Cdk1. The fact that cyclin B/Cdk1 also phosphorylates and inhibits Cdk1 results in mutual inhibition between Cdk1 and Cdk1, which amounts to yet another positive feedback loop. Like for cyclin E/Cdk2 and cyclin A/Cdk2, the activity of the complex cyclin B/Cdk1 can be

Fig. S2. Detailed description of the four modules of the cdk network. (A) Detailed description of the cyclin D/Cdk4–6 module controlling progression in G1. The GF activates the synthesis of the cdk inhibitor AP1, which promotes synthesis of cyclin D. Cyclin D forms a reversible complex with Cdk4–6. The activity of this complex can be regulated by phosphorylation–dephosphorylation. The active form of cyclin D/Cdk4–6 can also form a complex with the cdk inhibitor p27/p21, but this binding does not result in the inhibition of cyclin D/Cdk4–6, contrary to what is observed for Cdk1 and Cdk2 (see section 1.4 of SI Appendix). Cyclin D/Cdk4–6 promotes the phosphorylation and subsequent inactivation of pRB. This phosphorylation allows the activation of the transcription factor E2F. The non-phosphorylated and monophosphorylated forms of pRB can form a complex with E2F, thereby inhibiting its transcriptional activity. A second phosphorylation of pRB by cyclin E/Cdk2 totally inhibits pRB, because the twice-phosphorylated form of pRB cannot bind to E2F. This second phosphorylation thus fully activates the transcription factor E2F. (B) Detailed description of the cyclin E/Cdk2 module controlling the G1/S transition. The synthesis of cyclin E is induced by E2F, and inhibited by pRB and pRBp, both directly and indirectly through formation of inactive complexes between pRB and pRBp with E2F. Cyclin E reversibly forms a complex with Cdk2. The activity of this complex is regulated by phosphorylation–dephosphorylation; the phosphatase Cdc25 activates the complex cyclin E/Cdk2 by dephosphorylation, whereas the kinase Wee1 inactivates it by phosphorylation. A positive feedback loop exists between cyclin E/Cdk2 and its Cdc25 phosphatase (Pe), given that cyclin E/Cdk2 activates Cdc25 by phosphorylation, whereas Cdc25 activates cyclin E/Cdk2. The cdk E/Cdk2 complex can be also regulated by a reversible formation of a complex with the cdk inhibitor p21/p27. The accumulation of cyclin E/Cdk2 leads to phosphorylation of its inhibitor p21/p27. This phosphorylation brings about the specific degradation of p21/p27, under control by the Skp2 protein, which belongs to the proteasome. Skp2 also governs the degradation of cyclin E, which ensures that the activity of cyclin E/Cdk2 decreases at this phase of the cycle. (C) Detailed scheme for the cyclin A/Cdk2 module controlling the S-G2 transition. The synthesis of cyclin A is induced by E2F and inhibited directly and indirectly by pRB and pRBp. Like cyclin E, cyclin A can form a reversible complex with Cdk2. The activity of this complex can be regulated by phosphorylation–dephosphorylation; the Cdc25 phosphatase activates the complex cyclin A/Cdk2, whereas the kinase Wee1 inactivates it by phosphorylation. We consider a positive feedback loop between cyclin A/Cdk2 and its Cdc25 phosphatase (Pa). Indeed, Cdc25 activates cyclin A/Cdk2, and cyclin A/Cdk2 activates its phosphatase Cdc25 by phosphorylation. The complex cyclin A/Cdk2 can also be regulated by reversible formation of a complex with the cdk inhibitor p21/p27. Cyclin A/Cdk2 controls the S-G2 transition by eliciting the phosphorylation and subsequent degradation of the transcription factor E2F (see A). Cyclin A/Cdk2 and cyclin B/Cdk1 both phosphorylate and thereby inhibit the protein Cdk1. This inhibition leads to the accumulation of cyclin B and to the G2/M transition (see D). (D) Detailed scheme for the cyclin B/Cdk1 module controlling the G2/M transition. In the preceding module (see C), cyclin A/Cdk2 inhibits by phosphorylation the protein Cdk1. This protein cannot anymore promote the degradation of cyclin B, so that the level of cyclin B increases. Cyclin B reversibly forms a complex with Cdk1. The fact that cyclin B/Cdk1 also phosphorylates and inhibits Cdk1 results in mutual inhibition between Cdk1 and Cdk1, which amounts to yet another positive feedback loop. Like for cyclin E/Cdk2 and cyclin A/Cdk2, the activity of the complex cyclin B/Cdk1 can be
Fig. S3. A balance between E2F and pRB controls the oscillatory dynamics of the Cdk network. (A) Antagonistic effects of the tumor suppressor pRB and the transcription factor E2F on oscillations in the Cdk network. The diagram illustrates the dynamic behavior of the network as a function of the rate of synthesis of E2F ($v_{\text{se2f}}$) and the rate of synthesis of pRB ($v_{\text{spRB}}$). The Cdk network evolves either to a stable steady state or sustained oscillations. The red dot corresponds to the oscillations shown in Fig. 2B. The successive arrows from points 1 to 5 correspond to the increments considered in B. Note that oscillations can occur when the rate of pRB synthesis goes to zero (even in the absence of GF). (B) A balance between E2F and pRB controls progression in the cell cycle, as shown by changes in the rates of synthesis of pRB and E2F, $v_{\text{spRB}}$ and $v_{\text{se2f}}$ (in $\mu$M h$^{-1}$). Thus, increasing $v_{\text{spRB}}$ from point 1 ($v_{\text{spRB}} = 0.05$, $v_{\text{se2f}} = 0.01$) to 2 ($v_{\text{spRB}} = 0.5$, $v_{\text{se2f}} = 0.01$) in A results in cell-cycle arrest. Then, increasing $v_{\text{se2f}}$ from point 2 ($v_{\text{spRB}} = 0.5$, $v_{\text{se2f}} = 0.01$) to 3 ($v_{\text{spRB}} = 0.5$, $v_{\text{se2f}} = 0.1$) restores the oscillations. Further increasing $v_{\text{spRB}}$ from 0.5 to 5 (and maintaining $v_{\text{se2f}}$ at 0.1, i.e., moving from point 3 to 4 in A) suppresses the oscillations. A final increase of $v_{\text{se2f}}$ from 0.1 to 5 (i.e., moving from point 4 to 5) again results in the resumption of stable oscillations. This balance between the antagonistic effects of pRB and E2F is robust and can be observed over several orders of magnitude for these parameters, as shown in A. The diagram in A has been established by numerical integration of Eqs. 1–39 in SI Appendix for the parameter values listed in Table S2.
Fig. S4. Oscillatory dynamics of the Cdk network and the transition to cell proliferation. (A and B) Dynamic behavior as a function of the parameters $k_{ce}$ and $k_{cd1}$ governing the rates of synthesis of cyclin E and cyclin D. Two distinct types of dynamical behavior are observed: the Cdk network evolves either to a stable steady state, generally associated with quiescence, or sustained oscillations, associated with cell proliferation. (C) The dynamic behavior of the Cdk network is shown as a function of the rate of synthesis of the phosphatase Cdc25 acting on cyclin A/Cdk2 ($v_{spai}$) and the rate of synthesis of Cdh1 ($v_{scdh1a}$), which promotes the degradation of cyclin B and prevents the degradation of cyclin E by inducing the degradation of Skp2 (see Figs. S1 and S2 B and D). Here again the Cdk network reaches a stable (nonoscillatory) steady state or undergoes sustained oscillations, associated with cell proliferation. The vertical arrow on the right indicates a region of the oscillatory domain in parameter space where oscillations in Cdk2 occur in the absence of oscillations in Cdk1, a situation that corresponds to endoreplication. The two arrows originating from a point (black dot) corresponding to a stable, nonoscillatory state illustrate how an increase in the activity of phosphatase Cdc25 or a decrease in the level of the protein Cdh1 can bring about the transition to cell proliferation, as observed in a number of tumors. The curves in A–C are generated as in Fig. 2B, for the same set of parameter values. In B, $V_{v_{spai}} = V_{v_{spRB}} = 0.2$ (instead of 0.8) $\mu$M h$^{-1}$.
Fig. S5. The ATR/Chk1 DNA replication checkpoint. (A) Scheme of the ATR/Chk1 DNA replication checkpoint. At the beginning of the DNA replication phase, cyclin E/Cdk2 activates, by phosphorylation, the anchor factor Cdc45. This anchor factor allows DNA polymerase α to bind to DNA. At the initiation of DNA replication, DNA polymerase α synthesizes an RNA primer. Activation of the kinase ATR follows its binding to this RNA primer. The active form of ATR promotes the phosphorylation and subsequent activation of the kinase Chk1. Once activated, the kinase Chk1 inhibits, by phosphorylation, the phosphatases Cdc25 responsible for the activation of cyclin E/Cdk2, cyclin A/Cdk2, and cyclin B/Cdk1. The inhibition of cyclin E/Cdk2 and cyclin A/Cdk2 during DNA replication creates a checkpoint, which limits the activation of Cdc45 and thereby curtails excessive initiation of DNA synthesis at the multiple points of origin of DNA replication. At the end of DNA replication, cyclin E/Cdk2 is further inhibited because of the degradation of cyclin E, brought about by the rise in Skp2, which follows from the inactivation of Cdh1 by cyclin A/Cdk2 (Fig. S1). The anchor factor Cdc45 will not be active anymore because of the lack of cyclin E/Cdk2, so that the activity of DNA polymerase α will decrease, and so will the concentration of RNA primer and the activity of the kinases ATR and Chk1. Moreover, the ATR/Chk1 checkpoint promotes the inhibition of the phosphatase Cdc25 that activates cyclin B/Cdk1. The resulting inhibition of cyclin B/Cdk1 prevents cells to enter into mitosis as long as DNA replication is not completed. (B) Detailed scheme of the ATR/Chk1 DNA replication checkpoint incorporated in the model for the mammalian cell cycle. This ATR/Chk1 DNA replication checkpoint is present during normal cell-cycle progression and can be overexpressed after DNA damage (see section 3 of SI Appendix). The overexpression of this checkpoint can be responsible for slowing down or possibly arresting the cell cycle as long as DNA damage is not repaired. The braking effect of the checkpoint is illustrated in Fig. 2C.
Fig. S6. Oscillations in the Cdk network in the presence of only Cdk1 or in the absence of pRB. (A) As described in section 4 of SI Appendix, the kinetic equations can be modified in the case where Cdk1 is the only Cdk present and can substitute for Cdk4–6 and Cdk2 in forming complexes with cyclins D, E, and A. The curves showing self-sustained oscillations in the complexes formed by Cdk1 with cyclins D, E, A, and B were obtained for the parameter values of Table S2, with $v_{spRB} = 0.125 \mu M h^{-1}$, $\alpha = 40$, $k_+ = 0.4 \mu M^{-1} h^{-1}$, $k_{cat} = 0.54 h^{-1}$, $k_{cat} = 0.065 \mu M^{-1} h^{-1}$, and Cdk$_{tot} = 2 \mu M$. (A) Self-sustained oscillations can occur in the Cdk network in the absence of pRB, in the presence or even in the absence of GF. The curves show the time evolution of cyclin A/Cdk2, cyclin E/Cdk2, and cyclin B/Cdk1 for the parameter values of Table S2, with $v_{spRB} = 0$ (no synthesis of pRB) and $v_{se2f} = 0.003$ (instead of 0.15) $\mu M h^{-1}$ (this reduction in the rate of synthesis of E2F is needed to limit the level of cyclin A/Cdk2 in the course of oscillations in the absence of the inhibitory effect of pRB).
Fig. S7. Entrainment of the mammalian cell cycle by the circadian clock. (A) Scheme for the control of the mammalian cell cycle by the circadian clock. The complex CLOCK–BMAL1 is a regulator of the circadian clock machinery. This complex activates the transcription of the kinase Wee1, which inhibits cell-cycle progression by phosphorylating the kinases Cdk2 and Cdk1. The kinase Cdk1 can also inhibit, by phosphorylation, kinase Wee1. When the cell cycle is coupled to the circadian clock, the complex CLOCK–BMAL1, varying in a circadian manner, will inhibit, through the kinase Wee1, progression in the cell cycle. Thereby, cell cycles with an autonomous period different from 24 h can be entrained to oscillate with a circadian period. (B) In Eq. 38 in SI Appendix, in addition to the constant, basal value of the rate of synthesis of the kinase Wee1, $v_{sWee1}$, we include a term ($k_sMW$) that reflects the circadian activation exerted by the circadian clock complex CLOCK–BMAL1 on Wee1 gene transcription. The variable $MW$ in Eq. 45 in SI Appendix represents the amount of Wee1 mRNA expressed under control of the circadian clock complex CLOCK–BMAL1, at a maximum rate $v_{sW}$. In the absence of coupling to the circadian clock, $v_{sW}$ is set equal to zero. In the presence of coupling, $v_{sW}$ increases from 0 to 0.2 μM·h$^{-1}$ for $t \geq 150$ h (see section 5 of SI Appendix). The period of Cdk1 oscillations then shifts from $\sim 19$ to 24 h, reflecting entrainment of the cell cycle by the circadian clock. The curves showing the time evolution of the total amount of Wee1 mRNA (see section 5 of SI Appendix) and cyclin B/Cdk1 before and after (vertical arrow at time = 150 h) coupling to the circadian clock have been obtained by numerical integration of Eqs. 1–39 and 45 in SI Appendix for the parameter values of Table S2. In B the circadian variation of CLOCK–BMAL1 is generated by means of a model for the mammalian circadian clock, in the conditions of figure 2C in ref. 40.

**Other Supporting Information Files**

SI Appendix (PDF)
Table S1 (PDF)
Table S2 (PDF)