Synchronization of biological clocks

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http://www.youtube.com/watch?v=OBO_PKstYzc
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Synchronization of periodic oscillators by periodic external action of self-sustained oscillators. Finally, we describe a technical system (phase locked loop) that can be interpreted as a particular example of a driven oscillator.

7.1 Phase dynamics

In this section we consider the effect of a weak periodic external force on periodic self-sustained oscillators. The main idea here is that a small force influences only the phase, not the amplitude, so that we can describe the dynamics with a phase equation. In its derivation we follow the method developed by Malkin [1956] and Kuramoto [1984]. Although the method is quite general, the resulting phase equation is very simple and easy to investigate. This will allow us to describe many important properties of synchronization analytically.

7.1.1 A limit cycle and the phase of oscillations

Consider a general $M$-dimensional ($M \geq 2$) dissipative autonomous system of ordinary differential equations

\[
\frac{dx}{dt} = f(x), \quad x = (x_1, \ldots, x_M),
\]

(7.1)

and suppose that this system has a stable periodic (with period $T_0$) solution $x_0(t) = x_0(t + T_0)$. In the phase space (the space of all variables $x$) this solution is an isolated closed attractive trajectory, called the limit cycle (Fig. 7.1). The point in the phase space.

Formally, a driven system can be written as an autonomous one, if one introduces an additional equation for a variable that is equivalent to the time. Physically, such a manipulation does not make the system really autonomous, because the "time variable" cannot be influenced.

Figure 7.1. A stable limit cycle (bold curve), here shown for a two-dimensional dynamical system. Its form can be very different from a circular one; in a high-dimensional phase space it can be even knotted. The neighboring trajectories are attracted to the cycle.
Coupling of oscillators

Adjustment of rhythms: frequency and phase locking

Experiments show that even a weak interaction can synchronize two clocks. That is, two nonidentical clocks which, if taken apart, have different oscillation periods, when coupled adjust their rhythms and start to oscillate with a common period. This phenomenon is often described in terms of coincidence of frequencies as frequency entrainment or frequency locking. Whether they synchronize or not depends on the following two factors.

1. Coupling strength
2. Frequency detuning

The coupling strength of two oscillators is defined as the ratio of the ratio of the difference of their natural frequencies to the sum of the natural frequencies: $\alpha = (f_1 - f_2) / (f_1 + f_2)$. Here $f_1$ and $f_2$ are the natural frequencies of uncoupled oscillators. In contrast to the coupling strength, in experiments with two nonidentical oscillators having their own frequencies $f_1$ and $f_2$, frequency detuning can be easily measured and varied. Indeed, one can tune the frequency of a clock by altering the pendulum length.

Frequency detuning can be measured as $\Delta f = f_1 - f_2$.

The natural frequency $f$ of an oscillating system is defined as $f = 1/T$, where $T$ is the period of oscillation.

From the above definitions it follows that $\alpha = \Delta f / f_1$. Thus, we can find out how weak (or how strong) the interaction is. In an experimental situation it is not always clear how to measure this quantity. In the experiments described above, it depends in a complicated manner on the ability of the supporting structure to move. Indeed, if the beam is absolutely rigid, then the motions of pendula do not influence the support, and therefore there is no way for one clock to act on the other. If the beam is not rigid, but can vibrate longitudinally or bend, then an interaction takes place.

The motion of each pendulum is transmitted through the supporting structure to the other. If the clocks do not interact, the coupling strength is zero. If the beam is slightly flexible, the coupling strength is small but not zero. If the beam is flexible, the coupling strength is large. For intermediate situations it is not always clear how to measure this quantity. In the experiments described above, it depends in a complicated manner on the ability of the supporting structure to move. Indeed, if the beam is absolutely rigid, then the motions of pendula do not influence the support, and therefore there is no way for one clock to act on the other. If the beam is slightly flexible, the coupling strength is small but not zero. If the beam is flexible, the coupling strength is large. For intermediate situations it is not always clear how to measure this quantity. In the experiments described above, it depends in a complicated manner on the ability of the supporting structure to move.

Mechanical clocks usually have a mechanism that easily allows one to do this. The process is basically described in Section 1.3.1. We use “entrainment” and “locking” as synonyms (see terminological remarks in Section 1.3.1).
Synchronization of coupled oscillators

Coupled, noisy oscillators can synchronize,

"the odd sympathy of two clocks"
Synchronization of coupled oscillators

http://www.youtube.com/watch?v=W1TMZASCR-I
It is a well-established fact that in the absence of a light–dark cycle the period of the circadian rhythm deviates from 24 hours; it can be either longer or shorter. Under normal conditions, the cycle is entrained by the daily variation of the illuminance. This entrainment was studied in numerous experiments within which the subject was isolated from the normal light–dark cycle and deprived of all other time cues. The results of these experiments can be schematically represented in the way shown in Fig. 3.41. This is a direct indication of the endogenous character of the circadian rhythm, i.e., that there exists a self-sustained clock that governs the rhythm. All available evidence indicates that there is one principal circadian pacemaker in mammals, namely the suprachiasmatic nucleus of the hypothalamus. This nucleus receives entraining information from retinal ganglion cells [Moore 1999].

Quantification of the circadian period in humans has yielded inconsistent results [Czeisler et al. 1999; Sassone-Corsi 1999; Moore 1999]: the average was controversially estimated to be 25 or 23 h, and individual variation from 13 to 65 h was reported in normal subjects. This large variation may be responsible for the behavioral patterns of “early birds” and “owls”. Indeed, from general properties of synchronization we expect that the subjects with an intrinsic period $T_0 < 24$ h should be ahead in phase with respect to the external force, whereas those with a period $T_0 > 24$ h should lag in phase. Similarly, an age-related shortening of the intrinsic period has been observed under various conditions [Aschoff et al. 1982; Czeisler et al. 1986; Moore 1999].

**Figure 3.41.** Schematic diagram of the behavioral sleep–wake rhythm. Here the circadian rhythm is shown entrained for five days by the environmental light–dark cycle and autonomous for the rest of the experiment when the subject is placed under constant light conditions. The intrinsic period of the circadian oscillator is in this particular case greater than 24 hours. Correspondingly, the phase difference between the sleep–wake cycle and daily cycle increases: the internal “day” begins later and later. Such plots are typically observed in experiments with both animals and humans [Aschoff et al. 1982; Czeisler et al. 1986; Moore 1999].
Synchronization of the circadian rhythm with the sleep-wake cycle

Figure 1. Sleep-wake record of Siffre's time-isolation study in Midnight Cave, Texas, 1972. Black bars represent time when subject was asleep. Each sleep episode is plotted twice: beneath the previous episode and also to the right of it. This "double raster-plot" emphasizes the continuity in the data across the artifactual edge at 24:00 h. Internal desynchronization occurs spontaneously after day 37 and again after day 130 (see text).

Strogatz, 1987
Synchronization of the circadian rhythm with the sleep-wake cycle

Model of human sleep-wake cycle by modelers of various physiological oscillations. To mention just a few examples, phase models have been proposed in the context of circadian activity rhythm splitting in rodents \[13, 21, 25\], flashing rhythms of fireflies \[17\], frequency plateaus in the intestine \[16\], and swimming rhythms controlled by central pattern generators in fish \[7, 23\]. The model presented here extends this approach to human sleep and circadian rhythms.

3.2. Model structure

The structure of the PHASE model is summarized in Fig. 4. The phases of the two oscillators are denoted $\theta_1, \theta_2$. Although the phases are real numbers, we often regard them as points on the circle of unit circumference. The governing equations are:

\[
\dot{\theta}_1 = \omega_1 - C_1 \cos 2\pi (\theta_2 - \theta_1) \\
\dot{\theta}_2 = \omega_2 + C_2 \cos 2\pi (\theta_1 - \theta_2)
\]

The overdot signifies time differentiation. All the parameters are taken to be non-negative. The chosen form of the coupling is such that the first oscillator slows down and the second speed up when they are in phase. This property is suggested by the observed modulations of sleep-wake cycle lengths (e.g. Fig. 2d) as the activity and temperature rhythms cross through each other during internal desynchronization.

Temperature oscillator

Sleep-wake oscillator

Low temperature

Wake-up

$\theta_1 = 0$

$\theta_2 = F \sim \frac{1}{3}$

Sleep onset

Coupling

C1

C2

Strogatz, 1987
Inner ear hair cells
Inner ear hair cells
Inner ear hair cells coupled to cyber clones

A

B

Real-time simulation

X

Δ

\[ X \]

\[ F_{\text{EXT}} \]

\[ F_{\text{K}} \]

\[ F_{\text{EXT}} \]

\[ F_{\text{EXT}} \]

\[ F_{\text{EXT}} \]

\[ F_{\text{EXT}} \]

\[ -F_{\text{1}} \]

\[ -F_{\text{2}} \]

\[ -F_{\text{3}} \]

Enhancement of Mechanical Amplification.

Coupling a hair bundle to two cyber clones by dynamic force clamp.

Gentamicin, an aminoglycoside antibiotic that blocks transduction channels of inner ear hair cells.

The cross-correlation coefficients of three coupled cyber clones (Fig. 2).

Spectral density of both hair-bundle and time-dependent force measurements of hair-bundle sensitivity with and without coupling at a low force of 10 pN.

Frequency selectivity. In our experiments, the gain saturated at a similar magnitude was also observed in the experiments for a low force of 10 pN.

Gain was linearly related to the quality factor of an oscillatory cyber clone increased almost linearly with the number of cyber clones (Fig. 4).

Discussion

We used simulations to anticipate the behavior of a larger module of coupled hair bundles.

A similiar magnitude was also observed in the experiments for a low force of 10 pN.

Gain was linearly related to the quality factor of an oscillatory cyber clone increased almost linearly with the number of cyber clones (Fig. 4). All these observations were in quantitative agreement with pure computer simulations.

Relative movements of adjacent hair bundles yield elastic force with a low force.

Under such conditions, the compressive nonlinearity extended a low force.

The cross-correlation coefficients (Fig. 2).

The gain of individual hair bundles is limited by intrinsic fluctuations of stiffness (Fig. 2).

Dissipation of the fiber of the fiber.

The spectral density of spontaneous oscillations (Fig. 4).

The gain of 400 or 52 dB resulted in a gain reduction of the hair bundle from the hair-bundle sensitivity to weak stimuli and thus enlarges the range of stimulus sensitivity to weak stimuli.
Synthetic oscillators coupled by quorum sensing

Danino et al. 2010
Self-organized uterus contractions

\[ \dot{V}_e = F_e(V_e, g) + n_p C_r(V_p - V_e), \]
\[ \dot{V}_p = F_p(V_p) - C_r(V_p - V_e), \]
\[ \frac{\partial V_e}{\partial t} = F_e(V_e, g) + n_p C_r(V_p - V_e) + D \nabla^2 V_e, \]

Singh et al., 2012
Synthetic oscillators coupled by quorum sensing

Moving edge (Supplementary Fig. 3 and Supplementary Movie 3). As cell-density space–time plot shows that a higher density of cells is first the cell density is lower (between 118–200 min). The corresponding during this first burst (273 min), the bright band shows that cells the colony, with a bright band near the centre (Fig. 3c, 228 min).

Microfluidic chamber (Fig. 3c, d and Supplementary Movie 5). In and right appears as a green–yellow concave line. The larger slope is blue, indicating no fluorescence. Then at 100 min, there is an and negative slopes meet (300–400 m, corresponding to the burst in 8–35 m). Corresponding space–time diagram showing the 35 m, Snapshots of the GFP fluorescence superimposed over brightfield images

35 m | 8–35 m, Corresponding space–time diagram showing the 35 m | 8–35 m, Corresponding space–time diagram showing the

In the mid-seventeenth century, Chirstiaan Huygens serendipitously observed that two pendulum clocks oscillated in synchrony when (which, in the case of the pendula, he deduced as vibrations in the

Huygens is credited as the first to systematically characterize the syn-

Inverted microscope (TE2000-U, Nikon Instruments Inc.), and chip tempera-

The dynamic response to external signals. Along these lines, our results

Along these lines, our results

The modelling of this coupling, and the

Non-trivial phenomenology of the spatiotemporal quorum clock through extracellular AHL. The model consists of a one-

AiiA and LuxI. Both of these proteins then oscillations (Supplementary Information). Although conceptually the

Cell density has an important role in experimental data. Furthermore, cell density has an important role in

The growing colony of cells. This phenomenology is also in excellent

The concentration and wave propagation, we developed a computational model

The modelling of this coupling, and the

Theoretical analysis: Engineering spatiotemporal clocks with synthetic oscillators

AiiA and LuxI. Both of these proteins then

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The segmentation clock
Vertebrates have segmented bodies
Segments form one by one, from head to tail

forming segments are known as somites

zebrafish somitogenesis

Schröter et al., 2008
Cyclic gene expression patterns reflect the activity of the clock

Masamizu et al., 2006
Cyclic gene expression patterns reflect the activity of the clock.
Delta/Notch coupling introduces COMMUNICATION DELAYS

Somitogenesis period ~ 25 min

Delta trafficking ~ tens of min

[Diagram showing the processes of Delta and Notch signaling with corresponding mRNA and DNA regulation]
gene regulatory network
negative feedback. Genetic oscillations produced by single-cell oscillators driven by delayed oscillators. The arrow indicates the direction of embryonic elongation. Segments are red/white. Small circles with arrows represent single cell patterns (blue/white) controlled by long-length scale gradients across studies have changed the stability of Hes7 protein or the delays modifications, and the decay of the products (Fig. 4). Elegant biochemical events, such as splicing and post-translational transport of molecules in and out of the nucleus, additional oscillator is set by delays that arise from a combination of the Tyson, 2008). When oscillating, the period of the single-cell balancing of the timescales of the reactions involved (Novak and Glossary, Box 1) in the feedback, sufficient non-linearity and a 2003) require a negative-feedback loop, a time delay (see Box 1) at the single-cell level is supported by observations and Importantly, Hes7-binding N-box regulatory elements are found in segmentation is severely defective (Bessho et al., 2003). Corroborated in vivo for Hes7 by similar manipulations in wild-life and activity in these cells indicate that unstable Hes1 protein 2002). Biochemical data from manipulating Hes1 production, half-periodically represses its own expression. This regulatory logic was 2005). The disruption of cyclic gene expression, indicating that they are currently no models that explain this change. Thus, although many questions remain unanswered (see Box 6), it seems likely that individual PSM cells possess a noisy single-cell segmentation clock period (Schröter and Oates, 2010), directly persistent oscillation of several Wnt and FGF pathway genes in Henry et al., 2002; Oates and Ho, 2002;Sieger et al., 2006). The theories of an oscillating circuit (Oswald and Oates, 2011). Why do transient signaling perturbations cause changes to segment level patterns? Do individual oscillators slow across the PSM, as predicted by tissue-arrest mechanism? What are the quantitative characteristics of the FGF, Wnt and retinoic acid gradients that span the PSM? How do the signaling systems spanning the PSM act on the pace-making circuits of individual cells to arrest/sustain their oscillation? How do the signaling systems spanning the PSM act on the pace-making circuits of individual cells to arrest/sustain their oscillation? How do PSM cells in embryos other than zebrafish stay discontinuous signaling? Do neighboring cells send smooth coupling signals of equal of change in the phase of cells as they synchronize? In a wild-type zebrafish embryo, as predicted? Are oscillating cells noisier and slower in Delta-Notch mutants than Are there multiple parallel Hes/Her feedback circuits and, if so, what predictably engineered in vivo? Can a mutant segmentation clock with an altered period be regulated?
\[
\frac{dp(t)}{dt} = am(t - T_p) - bp(t),
\]

\[
\frac{dm(t)}{dt} = f(p(t - T_m)) - cm(t)
\]
between neighbors is lost: there is not only a defect in synchrony. In these embryos, it is not surprising that synchrony continues noisily, presumably corresponding to the latter case, with oscillation continuing noisily.

B. Aerne, personal communication) thus, according to the dissociation rate of the repressor, behavior approximates that of the deterministic system. Far from disrupting oscillation, noise helps it to occur. Thus, oscillation continues but is episodic, with periods fluctuating in a noisy manner. In fact, when these are noisy, they still show the standard periodicity.

The Period of Oscillation Is Determined by the Sum of the Transcriptional and Post-translational Regulational Delays

In the absence of noise, the model reduces to a simple delay differential equation model for the following general purpose:

\[
\frac{dp(t)}{dt} = am(t - T_p) - bp(t)
\]

\[
\frac{dm(t)}{dt} = f(p(t - T_m)) - cm(t)
\]

where the time dependencies of the variables are now given to a good approximation (within 0.1% of the actual value). The period is then given by the formula:

\[
T = \frac{1}{k_{off}} - \frac{T_{m}}{k_{on}} - T_{p}
\]

where the time dependencies of the variables are now given to a good approximation. The period is then given by the formula.

\[
T = \frac{1}{k_{off}} - \frac{T_{m}}{k_{on}} - T_{p}
\]
Cell movement

Mara et al., 2007

Uriu et al., 2007
Synchrony dynamics

\[ Z(t) = Z(t_0) \cdot e^{-\lambda(t-t_0)/2} \]
delayed coupling theory
The delayed coupling theory is a tissue level description of the segmentation clock.

2D - hexagonal lattice

the blue cell ■, has six neighbors □
We describe autonomous oscillators as phase oscillators.

\[ \frac{\partial \theta_i(t)}{\partial t} = \omega_i(t) \]

autonomous oscillators
Delayed coupling theory: local synchronization

\[ \frac{\partial \theta_i(t)}{\partial t} = \omega_i(t) \]

autonomous oscillators

delayed coupling

noise

The delayed coupling theory describes cyclic gene expression patterns at the tissue level.

1. phase oscillators (± noise)
2. coupling of oscillators
3. delay in the coupling
4. moving boundaries
5. frequency profile
The period is regulated by autonomous oscillators, coupling strength and delays.

steady state solution

\[ \Omega = \omega_A - \varepsilon \sin(\Omega \tau) \]

collective frequency (somitogenesis rate)
autonomous frequency

stability condition: \( \cos(\Omega \tau) > 0 \)
Predictions: effects of changes in coupling

1. Altered period
   - Altered segment length
   - Altered oscillatory expression patterns

2. Existence of instabilities
experimental test of predictions
Segmentation period is regulated by intercellular coupling

Segment length changes accordingly

First period mutants of the segmentation clock!!!

Collective period is tunable through coupling:

\[ \Omega = \omega_A - \varepsilon \sin(\Omega \tau) \]

Coupling strength tunes the collective period.
Parameter estimation

\[ \Omega = \omega_A - \varepsilon \sin(\Omega \tau) \]

- collective frequency
- coupling strength
- autonomous frequency
- coupling delay

autonomous period \( T_A = 28 \text{ min} \)

coupling delay \( \tau = 21 \text{ min} \)

coupling strength \( \varepsilon = 0.07 / \text{ min} \)
Altered gene expression patterns reflect a longer period in \textit{mindbomb} mutants.

\begin{align*}
\text{wt and het: } & \rho = 0.69, \ s = 0.150 \\
\text{mib: } & \rho = 0.72, \ s = 0.174
\end{align*}
Altered gene expression patterns reflect a longer period in \textit{mindbomb} mutants

Altered gene expression patterns reflect a longer period in mindbomb mutants

\[ x \approx \sigma \log \left[ \frac{\sinh(\lambda/2\sigma)}{\pi \nu^{-1} (1 + \eta)^{-1} + (\lambda/2\sigma) e^{-L/\sigma}} \right]. \]
The delayed coupling theory predicts regions of instability.
Patterns become increasingly disrupted as the delay is reduced.

\[ \tau = 20.75 \text{ min} \]

\[ \tau = 17.87 \text{ min} \]

\[ \tau = 17.38 \text{ min} \]

\[ \tau = 16.93 \text{ min} \]

\[ \tau = 14.91 \text{ min} \]

*wildtype*
The autocorrelation function characterizes disorder in gene expression patterns.

\[ C(\delta) = \langle I(x)I(x + \delta) \rangle \]

autocorrelation function
The instability leaves a characteristic signature in the autocorrelation function.

\[ \tau = 20.75 \text{ min} \]
\[ \tau = 17.87 \text{ min} \]
\[ \tau = 17.38 \text{ min} \]
\[ \tau = 16.93 \text{ min} \]
\[ \tau = 14.91 \text{ min} \]
Delays in the segmentation clock are sensitive to Mib levels.
Over-expression of Mindbomb moves the segmentation clock to an instability

Conclusions

Coupling between oscillators changes the collective period of the segmentation clock

first segmentation period mutants

first example of delayed coupling in natural systems
Thank you!

David J. Jörg