Timely investigation into the behaviour of clocks

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A worldwide quest is on, trying to solve the longitudinal problem of travelling and unsynchronised clocks. However, at some time, you become immensely ill and you are confined to your bed for what seems like an eternity. You entertain yourself by watching clock tick, watching its pendulum swing from side to side. You watch another clock tick ... then something about the clocks catches your attention. Perhaps it's cabin fever, but you notice something peculiar with the ticking of the clocks. They're ticking in time. Their pendulums are swinging in time. The clocks are synchronised.

Now, this might seem like something obvious, of course the clocks are synchronised. In today's age, that's why we have clocks. However, to have a full appreciation of this, we need to start our journey back in a time when clocks were only locally synchronised and long distance travel meant that you had to keep time with a new clock. Faster forms of communication was requiring more precision with time and noone could agree on what was the time.

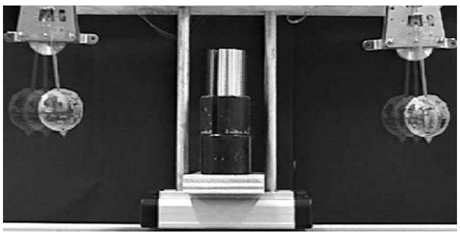

Figure 1: Pendulum clocks by Bennett et al. to reproduce the experiments performed by Huygens [1]. Multiple-exposure images of illustrating the synchronisation behaviour of the pendulums.

It was in 1657 when Christiaan Huygens built the first pendulum clock [2]. This was a great development in timekeeping since previous clock would vary by about 15 minutes per day. Huygen's pendulum clock would only differ by about 15 seconds per day [3]. However, to further improve his clock, Huygens performed a series of experiments in order to determine under what conditions the clocks exhibit synchronisation. These experiments, shown in Figure 1 have recently been redone [1].

Now, to generalise the idea of a pendulum clock, we can consider the clocks as oscillators. The synchronisation of clocks can then be considered as the coupling behaviour of a system of oscillators. An oscillator is any system which exhibits periodic behaviour. The modelling of coupled oscillators is often difficult, but computer simulations show that there will eventually be synchrony [4].

Before diving into the computations, let's introduce some terminology. The synchronisation of oscillator populations can be classed into three categories: synchrony, phase locking and frequency locking. Synchrony is the strongest of the three and is when all oscillators fire in unison. Assumption involved with having with having this type of synchronisation is that all of the oscillators are identical. Phase locking occurs when the phase difference between all oscillators is constant, generally nonzero. Frequency locking occurs when the oscillators share an average frequency, but there may not be a fixed phase relationship.

Simplifying the oscillator model, we could first consider the simplest case of the coupling behaviour between two identical oscillators. We could think of this as the coupling of two metronomes as shown in Figure 2. If we measure the behaviour of one of the oscillators at the same stage in each cycle, we would find that the other oscillator would change over repeated measurements. The response of the oscillator on the other was dependent on the rela-

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tive position to a threshold behaviour. This led to two conjectures: these two oscillators would always eventually synchronise and the oscillators would synchronise even if they were not identical [5].

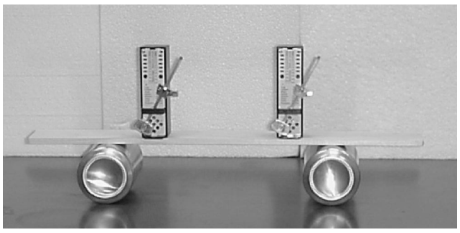


Figure 2: Two metronomes are on a light board balancing on two empty soft drink cans. As the metronomes oscillate, they couple through the board and influence each other's oscillation [2].

With computer simulations, the case of two coupled oscillators could then be extended to an arbitrary number of oscillators. This confirmed that the system of oscillators would always eventually have synchrony [4]. However, there were conditions on this result. It was found that damping and coupling was necessary for synchronisation. These two requirements are usually satisfied in the natural world.

These results for the behaviour of the coupling of arbitrary oscillators confirm observation made from watching the model oscillator of a pendulum clock. This abstraction from clocks to oscillators has allowed an interdisciplinary approach to tackling the same problem. Synchronisation is a phenomenon not only exhibited by pendulum clocks, but also in nature.

Biological synchronisation

The motivation for recent developments in synchronisation has come greatly from the biological sciences. The proof that two oscillators would eventually synchronise was motivated by considering the behaviour of natural pacemaker cells [5][6].

Due to the pattern of a heartbeat, it turns out that the oscillations are pulse-coupled, that is, the observable behaviour of one oscillator is relatively short compared to its whole oscillation. Interaction between two oscillators occurs only when they see each other's short behaviour. This occurs in pacemaker cells as their action would be of the form of an action potential spike. This sort of pulse-coupled oscillation is common in most biological oscillatory behaviour [7][8][9][10][11] yet had not been considered mathematically until the late twentieth century.

A core example of this lies within our own bodies. Gene networks are inherently noisy [12], but synchronisation stabilises the resulting behaviour from a network of intrinsically noisy and unreliable elements [13][14]. Thus synchronisation could provide a means by which biological processes stay regular within a body. For instance, take the human circadian rhythm [7]. This is believed to be an effect of only a few key genes regulating the whole internal clock of a body, describing the sleep-wake cycle and causing discomfort with a misjudgement with jetlag. Even though each cell within a body would have its own rhythm, they synchronise to work together to create the overall rhythm which can be exhibited by the person.

Many processes within the human body have periodic behaviour which require the cooperation of many cells. At the cellular level, we have pacemaker cells in the heart controlling the circulatory system, but also with the release of drugs and hormones. At a larger scale, we blink and breathe in periodic manner. These processes do not require us to forcibly control their pattern, but just occur naturally. At an even larger scale, we tend to walk in a periodic fashion with one leg then the other and so on. This pattern is not just executed in humans, but is resonant throughout a lot of animals. Although these oscillatory behaviours can be alter, for example, we hold our breath, they tend to stabilise to their own rhythm eventually. These cells are synchronised such that they work together.

Within other animals there is also oscillatory behaviour and these behaviours can be synchronised not only within the one animal, but across many other animals as well. Consider the fireflies in Figure 3. These are known for their rhythmically flashing bodies. In the South East Asian region, male fireflies flash on and off with other male fireflies as part of their courting behaviour. This had led to observations of what appears to be whole trees and riverbanks appear to be flashing on and off [8][9][15].

A similar behaviour is observed in crickets with their chirping. Despite each cricket having their own rhythm, they quickly fall in time with each other, creating the ordered chirping sound to which

Figure 3: Male fireflies, *Pteroptyx malaccae*, flash in synchrony with each other, lighting up a mangrove apple tree in Malaysia [15].

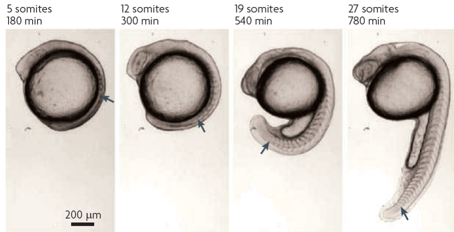
we are familiar [10]. The synchronisation behaviour of algae has led to observations of entire bays flashing periodically [9][11]. Upon investigating the synchronisation from the biological side, it was found that the oscillation and synchronisation was occurring at the subcellular level.

Understanding oscillators in developmental biology

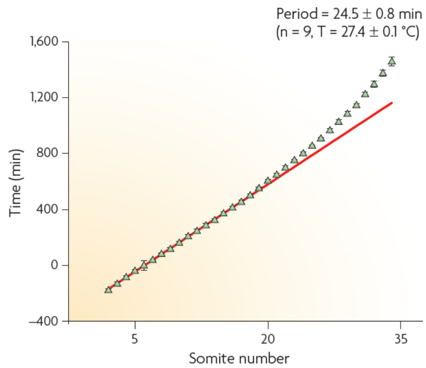
Within the biological world, the main goals of understanding the synchronisation of an oscillator mirror that to the goals of the physical world. Of interest are the synchronisation of oscillations and the role of noise and chaos in biological processes and the origins of complex rhythms [16].

The idea of the synchronisation of oscillating cells has become important within developmental biology. For example, it has been found that during embryonic development of vertebrates, there is periodically oscillatory expression of particular proteins. A change in the pattern of the oscillation of gene expression resulted in different lengths and different numbers of vertebrae when the animal was born as shown in Figure 4 [17][18].

The noisy and chaotic behaviour of gene expression is being investigated with both the mathematical and experimental biology approaches [12][14]. These investigations have led to not only a further understanding of real world oscillators, but have also led to developments of technology and



(a) Zebrafish embryos developing vertabrae strctures over time.



(b) Plot of time against the number vertebrae showing that there is a linear realation between time, so period, of oscillation and vertebrae development. Note that there is only slight deviation towards the end of growth.

Figure 4: In zebrafish, *Danio rerio*, vertebrae (somites) develop at a rate proportional to the period of oscillation of gene expression[17] .

improved techniques of looking into the behaviour of single cells [19].

Synthetic Biology to understand and manipulate genetic oscillators

Since these biological clocks are a result of their genetic outputs, there are currently efforts to replicate on the genetic level to create these biological clocks. Synthetic biology is an emerging discipline which involves the creation of synthetic genetic circuits [20]. Such a process would allow a suspected arrangement of genes to be tested to see if it could generate the oscillatory behaviour expected from a clock. This develops not only the understanding of biological clocks, but also replicates the often messy system in a more tangible and simplified manner by finding the minimal gene circuitry required for generating a biological clock.

There are five characteristics that a synthetic genetic circuit much possess for it to be a feasible biological clock. (1) The circuit must be able to function in a population of many growing cells. (2) The behaviour of the clock must be resistant to minor disturbances in the environment. (3) The clock must be self-contained and must be simple to measure the oscillatory output. (4) The clock must not interfere with other process occurring in its environment. (5) The clock must be suitable for regulating any gene [21].

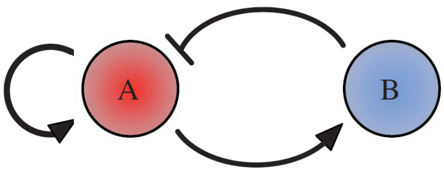


Figure 5: Simplest genetic circuitry to produce and oscillation [22]. The production of Gene A produces more Gene A and Gene B. More Gene B inhibits the production of Gene A, thus creating a cycle of fluctuating gene expression.

Through mathematical analysis of different arrangements of genetic networks, it was found that the way in which particular genes are connected is crucial in demonstrating the final oscillatory output. The simplest way to achieve oscillation is through a delayed negative feed back loop [20][22]. This requires one gene to produce the other, but the other gene inhibits the first gene as can be seen in Figure 5. However this circuits fails after a long time as the oscillations dissipate into their steady states.

This simple circuit can be modified to create more tunable and robust oscillator networks [23][24]. Such networks can produce oscillations with periods on the order of days, hours or minutes. This behaviour replicates that of a clock and we can appreciate that these genetic circuits can be simplified to a level where they can be created from scratch.

These investigations in genetic clocks are not limited to the big research laboratories. Undergraduates in a synthetic biology competition, iGEM [25], are investigating the feasibility of recreating a biological clock from scratch [26].

These circuitries to sustain a single oscillating cell can then be applied a system of cells with global intercellular coupling such that the behaviour can be observed on a larger scale [13]. This would be intended to recreate the synchronisation seen in pendulum clocks, thus recreating the exact behaviour of a clock in a biological system.

Summary

So from the journey beginning 400 years ago, we had a problem to solve which was that there was no way for anyone to agree on the time. The solution to this was the pendulum clock: a clock that would take advantage of the regular behaviour seen in a pendulum and synchronise itself to other pendulum clock. Continuing the idea of a pendulum just being an oscillator, rigorous mathematical models have been created to explain this behaviour. These mathematical models have then been applied to observations in the natural world. Further investigations in these natural oscillators are finding that synchronisation and regularity are key to many biological functions. This allows a further union of disciplines to stand the test of time.

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