Modelling in Biology

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Contents

Additional references

The interested reader is also referred to the following books, lecture notes and websites for complementary information:

- Strogatz, Steven (1994). *Nonlinear dynamics and chaos: with applications to physics, biology, chemistry, and engineering.* Perseus Books. A really brilliant book on nonlinear dynamics and chaos *available at Imperial library.* **Highly recommended for deepening your understanding of this course.** Youtube channel of recorded lectures of Steven Strogatz, filmed at Cornell University in Spring 2014: https://youtube.com/playlist?list=PLbN57C5Zdl6j_qJA-pARJnKsmROzPnO9V.
- Bois, Justin and Elowitz, Michael. *Biological Circuit Design.* Excellent Python-based (JupyterLab) online course, available at https://biocircuits.github.io.
- Del Vecchio, Domitilla and Murray, Richard. *Biomolecular Feedback Systems*, (in particular chapter 2 for a good recall of cellular biology and corresponding modelling at different levels of abstraction and chapter 5 for the analysis of simple synthetic biology models), available online at http://www.cds.caltech.edu/~murray/BFSwiki/index.php/Main_Page
- Alon, Uri (2007). *An Introduction to Systems Biology: Design Principles of Biological Circuits*, Chapman & Hall/ CRC Mathematical and Computational Biology Series.
- Sontag, Eduardo. *Lecture Notes on Mathematical Systems Biology*, slides available online at http://www.sontaglab.org/FTPDIR/slides_systems_biology_notes_ode_models.pdf and http://www.sontaglab.org/FTPDIR/slides_systems_biology_notes_sde_models.pdf, and notes available online at http://www.sontaglab.org/FTPDIR/systems_biology_notes.pdf. For a quick introduction to molecular cell biology see *Molecular Cell Biology: A Quick Introduction for non-Biologists* available at http://www.sontaglab.org/FTPDIR/introduction_molecular_cell_biology_ver2.pdf.
- Sontag, Eduardo, *Molecular Systems Biology and Control*, available online at http://www.sontaglab.org/FTPDIR/05cdc_ejc_oct05.pdf.
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- http://www.physiome.org/ The Physiome Project is the worldwide effort of several loosely connected research groups to define the physiome via databasing and the development of integrated quantitative and descriptive modeling.

- http://www.ebi.ac.uk/biomodels-main/ BioModels Database is a data resource that allows biologists to store, search and retrieve published mathematical models of biological interests. Models present in BioModels Database are annotated and linked to relevant data resources, such as publications, databases of compounds and controlled vocabularies.
- Milo, Ron, and Philipps, Rob, *Cell Biology by the Numbers*, available online at http://book.bionumbers.org. A great book containing quantitative information and key numbers in cellular biology.
- http://bionumbers.hms.harvard.edu/ A searchable database containing many common biological numbers needed in research along with links to the literature in which these numbers were obtained.
- http://www.scholarpedia.org/article/Main_Page A *peer-reviewed* encyclopedia written by scholars from around the world. Covers concepts from dynamical systems, computational neuroscience, computational intelligence, physics, astrophysics, etc.

Acknowledgement

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Foreword

What is a model?

From the Oxford English dictionary: A model is "a simplified or idealised description, representation or conception of a particular system, situation, or process, often in mathematical terms, that is put forward as a basis for theoretical or empirical understanding, or for calculations, predictions, etc." Models should be as simple as possible, yet as complex as necessary to address a given question of interest.

- "All models are wrong, but some of them are useful", George Box.
- "Everything should be made as simple as possible, but no simpler", Albert Einstein.
- "*Entia non sunt multiplicanda praeter necessitatem*" (entities must not be multiplied beyond necessity), William of Ockham.

In other words, a mathematical model is a representation of the essential aspects of an existing system (or a system to be constructed) which presents knowledge of that system in usable form. Thus, models are *not* replicas of reality, they are simplified representations of it. Simplification allows us to comprehend the *essential features* of a complex process without being burdened and overwhelmed by *unnecessary details*.

Often when engineers analyse a system to be controlled or optimised, they use a mathematical model. In analysis, engineers can build a descriptive model of the system as a hypothesis of how the system could work, or try to estimate how an unforeseeable event could affect the system. Similarly, in control of a system, engineers can try out different control approaches in simulations based on a constructed model.

Modelling, mathematically analysing and simulating complex systems *in silico* is typically much more time and resource effective than actually constructing real-life systems and prototypes for the same purposes.

Typically, the modelling process results from the inspection/investigation of a problem and a series of trade-offs. This analysis identifies the most important processes shaping the problem as well as some less important ones that may be neglected (at least in a first iteration of the model). The effect of each process is then modelled, i.e., described mathematically with some equations (or any tools borrowed from mathematics) and their combination is then mathematically analysed and/or simulated.

Mathematical modelling is typically the result of a trade-off between accuracy and simplicity since large, complex models may be more accurate in theory, but in practice they are hard to simulate and require many parameters that may not be available or identifiable.

The modelling process is considered successful when the obtained model possesses the following characteristics:

- Accurate: the model should attempt to accurately describe current existing observations.
- Predictive: the model should allow to appropriately predict the behaviour of the system (through analysis or simulation) in situations not already observed.
- Reusable: the model can be reused in another, similar case.
- Parsimonious: the model should be as simple as possible. That is, given competing and equally good models, the simplest is preferred.

Models need not match experimental data to be useful. For instance, a model may be built from a research hypothesis and its predictions compared to real data. Mismatches (whether qualitative or quantitative) can then be used to falsify the working hypothesis (see Karl Popper for more on falsification). More generally, a mismatch indicates that something worth looking into is happening. For instance a process that was overlooked might play a larger role than expected.

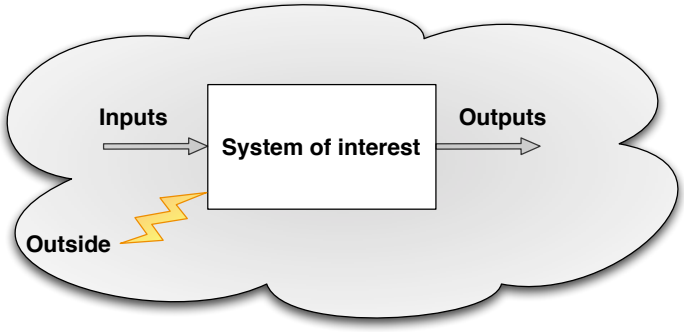
1 Introduction

In this introduction, we will give a broad overview of the essential features of most common mathematical models.

1.1 Essential features of a modelling approach

Isolate your system of interest.

- Identify what is important (and therefore what needs to be included in your model).
- List the quantities that can be observed/measured (they are the outputs).
- List the quantities that can be controlled/acted upon (they are the inputs).
- Define the environment and the constraints it places upon the system.



Remark 1. *You place in the system what is of interest. The parts that can be neglected are placed outside of the system of interest and considered as small perturbations. However, if these perturbations become too large to be neglected, they must be taken into account in the model.*

Modelling of the system of interest

Typically, the model is composed of

- variables
  - independent, e.g., time t
    - 1 indep. var.: ODEs, e.g., time t
    - more than 1 indep. var.: PDEs, e.g., time t and space (x, y, z) (examples include: blood circulation, diffusion, growth)
  - dependent (on the independent variable(s)),
    e.g., concentrations functions of time $\{[E](t), [S](t), [P](t)\}$
- parameters
  - not dependent on independent variables
  - can be varied/changed under experimental conditions (this can lead to a qualitative change in the system behaviour)
- constants
  - fixed, e.g., Avogadro constant, gravitational constant

Based on these concepts, different types of models can be built.

1.2 Types of models

Continuous	**Discrete**
• the independent variables are continuous • ODEs, PDEs	• the independent variables are discrete • Difference equations
Deterministic	**Stochastic**
• var., param. and const. do not contain randomness • they are defined by a unique function	• dynamics contain an element of randomness (described by probabilities, e.g., the variables are random/stochastic processes) • e.g., SDEs
Linear	**Nonlinear**
• $\dot{x} = \frac{dx}{dt} = -kx$ • Linear ODE	• $\dot{x} = \frac{dx}{dt} = -kx + x^3$ • Nonlinear ODE
Autonomous	**Non-autonomous**
• Without control input: $\dot{x} = -kx$	• With control input: $\dot{x} = -kx + u$
Constructive	**Data-driven**
• mechanistic or deductive • also called "equation-based" or "(first) principle-based"	• phenomenological or inductive

Remark 2. *Discrete models are typically used to model discrete events/discontinuous changes, e.g., events/changes which occur at specific time instants (i.e., between two consecutive events nothing changes/happens). They can also be obtained, as we will see, as the result of the discretisation of continuous models.*

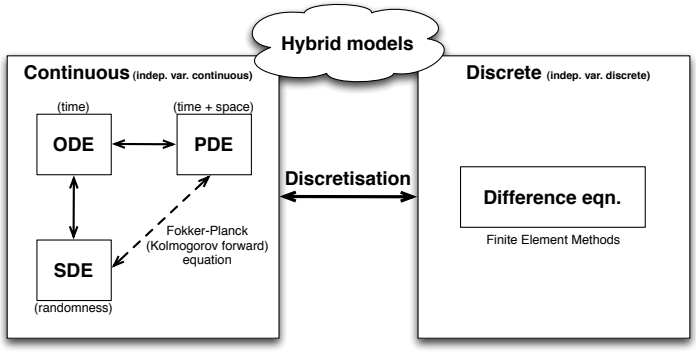
Remark 3. *Stochastic models (e.g., SDEs) are typically used to model diverse phenomena such as fluctuating stock prices, physical systems subject to thermal fluctuations, or intrinsic noise/stochastic effects in cellular biology.*[1]

Remark 4. *Nonlinear, stochastic models are almost unavoidable in biological modelling.*

Remark 5. *We will mostly deal with autonomous, deterministic models obtained through a constructive approach. But we will also briefly introduce stochastic models.*

Remark 6. *Linear deterministic models can be solved analytically. This is typically not the case for nonlinear or stochastic models, which, therefore, are often analysed using bifurcation and phase space analysis tools (which we will cover in this course) and also through computer simulations, e.g., MATLAB.*

1.3 Summary



[1] A good review of stochastic modelling in cellular biology is the paper: D. J. Wilkinson, "Stochastic modelling for quantitative description of heterogeneous biological systems", Nature Reviews Genetics, vol. 10, no. 2, pp. 122-133, Feb. 2009, http://www.nature.com/nrg/journal/v10/n2/full/nrg2509.html

Continuous	ODEs PDEs	Deterministic (L or NL)
	SDEs	Stochastic (L or NL)
Discrete	Difference equations	Deterministic (L or NL)
		Stochastic (L or NL)

Remark 7. *The* Fokker-Planck equation *is a PDE describing the time evolution of the probability density function. It was initially used to give a statistical description of the Brownian motion of a particle in a fluid.*

2 Linear models of order 1

A deterministic, continuous, linear model of order 1

Consider

$$\boxed{\dot{x}(t) = \frac{dx(t)}{dt} = kx(t)}$$

- For $k > 0$, this is known as the *Malthusian population growth* with k denoting the growth rate *per cell*. The Malthusian population growth model is also known as the "simple exponential growth model".

  Suppose that $x(t)$ counts the population of a microorganism in culture, at time t. The Malthusian population growth model is based on the following assumption: *the increase in population size during a (small) time interval is proportional to the population size.*

  Remark 8. *This makes intuitive sense. Let's take an example to illustrate it. Suppose that we have a population of cells and that each cell is experimentally shown to duplicate every* 3 *minutes (which is also called the cell* doubling time*) and assume, for simplicity, that cells never die. Let us call x_0 the number of cells initially present, i.e., present at time $t = 0$ when we start observing them multiply, and $x(t)$ the number of cells at time t. We thus expect to see the following time evolution:*

t (in mins)	$x(t)$
0	x_0
3	$2x_0$
6	$4x_0$
9	$8x_0$
...	...

  This time evolution can be described by a first order linear ordinary differential equation (ODE) of the form: $\boxed{\dot{x}(t) = \left(\frac{\ln(2)}{3}\right) x(t)}$ *whose solution is, as we will see later,* $x(t) = x_0 e^{\frac{\ln(2)}{3}t} = x_0 \left(e^{\ln(2)}\right)^{\frac{t}{3}} = x_0 2^{\frac{t}{3}}$*, with* $\frac{\ln(2)}{3}$ *denoting the growth rate per cell. This solution* $x(t) = 2^{\frac{t}{3}} x_0$ *indeed satisfies the values reported in the table above when t is measured in minutes.*

  Remark 9. *To ease the notation, we will, in the rest of this course, omit the explicit dependence on time when this is obvious from the context and simply write $\dot{x} = kx$ instead of $\dot{x}(t) = kx(t)$.*

  As we will see later, the Malthusian population growth is only valid under the assumption that resources (such as nutrients or space) are available in unlimited quantity, i.e., that there is no competition for resources in the population[2].

- Other examples of **Linear** ODEs:
  - Chemical Engineering: $X \xrightarrow{k} \emptyset$

    This corresponds to a degradation reaction where $k > 0$ is a constant degradation rate. Applying the law of mass action (we will explain it later), the dynamics associated with this reaction are $\dot{[X]} = -k[X]$ where $[X]$ represents the concentration of chemical species X.
  - Electrical Engineering: RLC circuits
  - Mechanical Engineering: Mass-Spring-Damper systems

[2] Strictly speaking, "resources" not only includes nutrients but space (e.g., petri dish) as well.

2.1 Analytical solution of first order linear ODEs

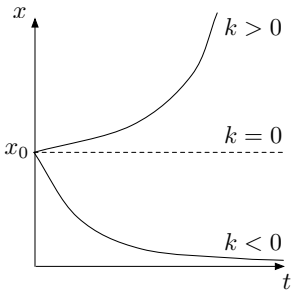
Consider the model:

$$\boxed{\dot{x} = \frac{dx}{dt} = kx, \qquad x(0) = x_0} \tag{1}$$

Its solution is given by

$$\boxed{x(t) = x_0 e^{kt}}$$

where $x_0 = x(0)$ (the initial condition).



Remark 10. *For $k > 0$, the model exhibits exponential growth behaviour. Bacterial populations tend to grow exponentially,* so long as there is no competition for resources (e.g., nutrients or space). *Exponential growth in populations (not only cell populations) in the absence of competition for resources was initially proposed by Malthus in 1798 as an empirical law, obtained by fitting an exponential model to observed data of population growth.*

Remark 11. *We can find the solution of* (1) *by considering the "*Ansatz*" (i.e., an "educated guess" that is verified later by its result) $x(t) = Ae^{kt}$ which indeed verifies eq.* (1)*. (This Ansatz can be obtained by trying to guess what the solution of $\dot{x} = kx$ should be. What we can say is that a solution that satisfies this differential equation should be a function of time which is such that, when you derive it with respect to time, you obtain the function itself multiplied by a constant. An exponential function of time is such a function.)*

Another method to find the solution of eq. (1) *is to rearrange the equation so that both sides can be integrated separately:*

$$\begin{aligned}
\frac{1}{x} dx &= k dt \\
\int \frac{1}{x}\, dx &= \int k\, dt \\
\ln(x(t)) &= kt + \tilde{A}, \quad \textit{where } \tilde{A} \textit{ is an integration constant} \\
x(t) &= e^{kt} \underbrace{e^{\tilde{A}}}_{=A} = Ae^{kt}
\end{aligned}$$

Furthermore, by definition of the initial condition, we have $x(0) = x_0$ which yields

$$x(0) = A \underbrace{e^{k\cdot 0}}_{=1} = A = x_0$$

2.2 Numerical solutions of ODEs: the Euler algorithm

If we want to solve ODEs numerically, i.e., using a computer, we need an algorithm. Here, we will try to find one for solving $\dot{x} = kx$ numerically. This can be done by considering the mathematical definition of the time derivative:

$$\frac{dx}{dt} = kx$$

$$\Leftrightarrow \boxed{\lim_{\Delta t \to 0} \frac{x(t+\Delta t) - x(t)}{\Delta t} = kx(t)}$$

Suppose Δt is fixed to a particular value h (doing this is called *discretising* the continuous ODE model and h is called the *discretisation step*). We then have:

$$\frac{x(t+h) - x(t)}{h} \approx kx(t)$$

$$\boxed{x(t+h) \approx x(t) + hkx(t)} \tag{2}$$

If h is "small", then the recursive algorithm described by eq. (2) and initiated with $x(0) = x_0$ will agree "well" with the analytical solution $x(t) = x_0 e^{kt}$.

Eq. (2) is know as the "Euler algorithm".

Remark 12. *A better alternative to the Euler algorithm for numerical integration is the "Runge-Kutta" family of algorithms (http://en.wikipedia.org/wiki/Runge-Kutta_method). These algorithms typically give more accurate numerical solutions for ODEs and converge faster to a solution for a given desired accuracy. The Runge-Kutta numerical integration algorithms are available in Matlab. In particular, these algorithms are used by the Matlab function* `ode45` *(you can get some information about* `ode45` *by typing* `help ode45` *at the Matlab prompt; if you want to read the code implementing the function* `ode45` *you can type* `type ode45` *at the Matlab prompt).*

2.3 Analytical solution of first order linear difference equations

$$x(t+h) = x(t) + hkx(t)$$

is a discrete-time model which can also be looked at as a linear *difference equation* by taking $h = 1$, and defining for ease of notation $x_t = x(t)$:

$$\boxed{x_{t+1} = \underbrace{(1+k)}_{\alpha} x_t = \alpha x_t} \tag{3}$$

(or equivalently $x_{t+1} - x_t = (\alpha - 1)x_t$.)

Its non-zero solution is given by

$$\boxed{x_t = x_0 \alpha^t}$$

where x_0 is the initial condition.

Remark 13. *The non-zero solution of (3) can be found by iterating (3) from the initial condition and generalising what is being observed in the sequence obtained. This sequence is x_0, $x_1 = \alpha x_0$, $x_2 = \alpha x_1 = \alpha^2 x_0$, $x_3 = \alpha x_2 = \alpha^3 x_0$, $\ldots$, $x_t = \alpha^t x_0$.*

It can also be obtained by considering the Ansatz $x_t = Ar^t$ which indeed verifies (3):

$$\begin{aligned} \cancel{A}r^{t+1} &= \alpha \cancel{A} r^t \\ r^t(r - \alpha) &= 0 \\ &\Rightarrow \boxed{r = \alpha} \text{ or } r = 0 \end{aligned}$$

Furthermore, by definition of the initial condition, we have: $x_0 = A\alpha^0 = A$

Remark 14. *Equation $x_{t+1} = \alpha x_t$ with initial condition x_0 is also know as a "*discrete-time model*". Dicrete-time models are useful to model processes which occur at specific (discrete) time instants. An example for which a discrete-time model can be useful is cell population growth. To see this, define the number of cells at a certain discrete-time point t as the variable $x_t \in \mathbb{N}$. Imagine that each cell divides and duplicates at each discrete-time step $t = 1, 2, \cdots$ and that cells*

never die. In this case the discrete temporal evolution of the number of cells can be described by the following 1st order discrete-time model:

$$x_{t+1} = 2x_t$$

with the initial condition $x_0 \in \mathbb{N}$. The non-zero solution is then given by:

$$x_t = x_0 2^t,$$

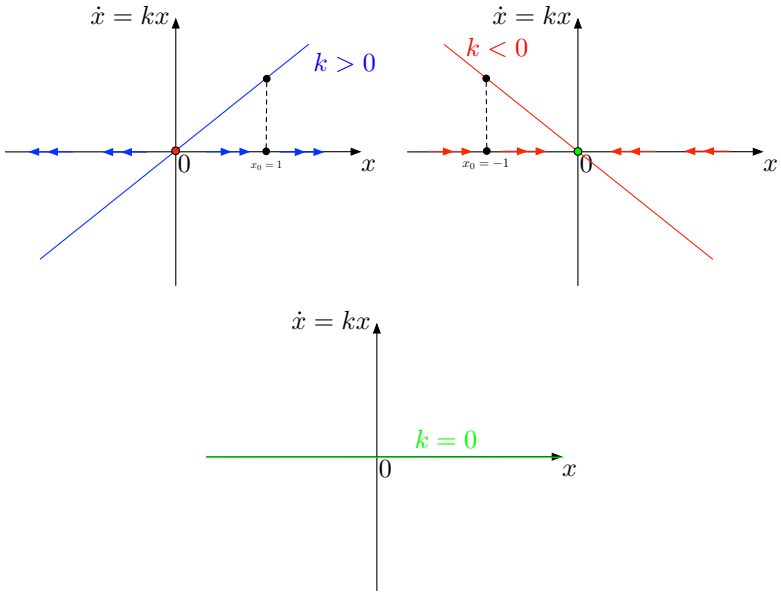
which, for $t = 1, 2, \cdots$, satisfies the values reported in the table appearing in Remark 8.

2.4 Phase line analysis for a linear ODE of order 1

Phase plane

- The phase plane (a.k.a. phase space) is a representation that eliminates time as an explicit variable.
- It is very useful for obtaining a qualitative understanding of the long-term or *asymptotic* behaviour of *nonlinear* ODE models (for which, typically, analytical solutions cannot be found).

Consider $\boxed{\dot{x} = kx}$



1. Consider the case when $k > 0$:
   (a) when $x_0 > 0$: $\dot{x} > 0 \Rightarrow x$ increases until $x \to \infty$;
   (b) when $x_0 < 0$: $\dot{x} < 0 \Rightarrow x$ decreases until $x \to -\infty$;
   (c) when $x_0 = 0$: $\dot{x} = 0 \Rightarrow x$ remains at 0. But if x is slightly perturbed, it will move away from 0 and diverge to $\pm\infty$.

   Thus, the system is unstable for $k > 0$, except when it starts exactly at 0, i.e., when $x_0 = 0$.[3] This agrees well with the analytical solution $x(t) = x_0 e^{kt}$ with $k > 0$.

   This is know as the *Malthusian explosion*.

[3] The fixed point $x^* = 0$ is said to be unstable since any perturbation from the fixed point will cause the solution $x(t)$ to go away from it.

2. When $k < 0$:
   (a) when $x_0 > 0$: $\dot{x} < 0 \Rightarrow x$ decreases until $x \to 0$;
   (b) when $x_0 < 0$: $\dot{x} > 0 \Rightarrow x$ increases until $x \to 0$;
   (c) when $x_0 = 0$: $\dot{x} = 0 \Rightarrow x$ remains at 0.

   If x is slightly perturbed, it will return to 0. Again, this agrees well with the analytical solution $x(t) = x_0 e^{kt}$ with $k < 0$.[4]

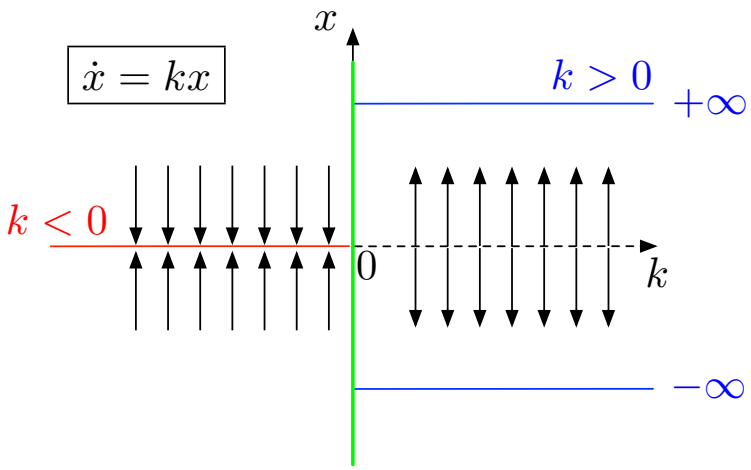
3. When $k = 0$: wherever you start, you stay there since $\dot{x} = 0$, i.e., $x(t) = x_0$, $\forall t$.

Remark 15. *What we have done here is a (global) asymptotic stability analysis of the fixed point $x^* = 0$ on the phase line x. "Asymptotic stability" of a fixed point refers to the $t \to \infty$ asymptotic behaviour of the solution initially perturbed away from the fixed point. We say the fixed point is asymptotically stable if the solution initially perturbed away from this fixed point is* stable *(i.e., it stays in a close neighbourhood of the fixed point) and* attractive *(i.e., in the long-term (i.e., as $t \to \infty$) the solution comes back to the fixed point).*

2.5 Bifurcation diagram for a linear ODE of order 1

Bifurcation diagram

We can summarise the information obtained through the phase plane stability analysis on a *bifurcation diagram*, i.e., a diagram giving the long-term (i.e., asymptotic) behaviour of the system when a parameter is varied. Here the parameter for the ODE model $\dot{x} = kx$ is k.



2.6 Stochastic differential equations (SDEs) of order 1

Consider a stochastic version of the Malthusian growth model:

$$\frac{dx}{dt} = kx + \eta \tag{4}$$

where η is a random variable that represents some *uncertainties* or *stochastic effects* perturbing the system. Eq. (4) is a continuous version of a random walk. Similar types of equations can be used to model the dynamics of diffusion-based phenomena or of stock prices.

Eq. (4) is known as a *Langevin equation*. In the absence of other knowledge about the statistics of the random variable η, η is generally assumed to have a Gaussian or normal probability distribution.

[4] The fixed point $x^* = 0$ is said to be asymptotically stable since any perturbation from the fixed point will cause the solution $x(t)$ to return back to it.

Eq. (4) can also be rewritten as

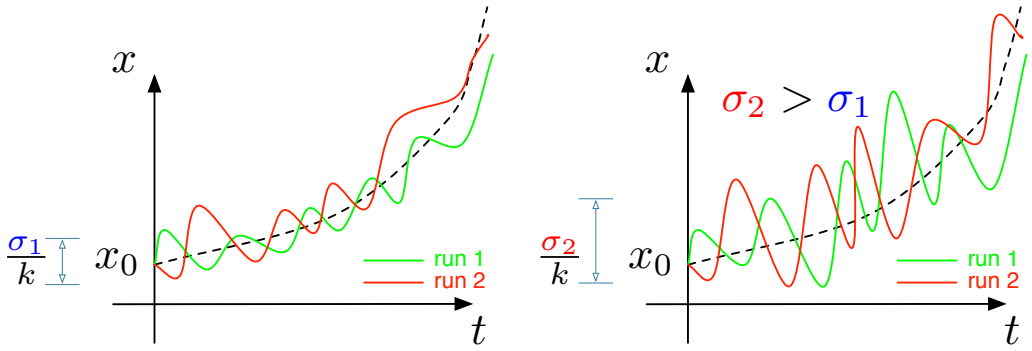
$$dx = [kx]dt + \underbrace{\eta dt}_{\approx \sigma dw} \tag{5}$$

where w represents a standard (one-dimensional) *Wiener process* (also called *Brownian motion*), distributed according to the normal distribution with mean 0 and variance t, i.e. $\mathcal{N}(0,t)$. dw is known as the differential into the future of a Brownian motion, distributed as $\mathcal{N}(0,dt)$. As a consequence, σdw is distributed according to $\mathcal{N}(0,\sigma^2 dt)$.

SDEs such as (5) are typically solved numerically through discretisation using the Euler algorithm:

$$x(t+\Delta t) = \underbrace{[1+k\Delta t]x(t)}_{\text{deterministic part}} + \underbrace{\left(\sigma\sqrt{\Delta t}\right)\texttt{randn}}_{\text{stochastic part}}$$

where the function `randn` (which is available in Matlab) provides a random number sampled from a Gaussian distribution of mean 0 and variance 1. Hereafter, we show simulation "runs" for different values of the variance. These runs are also called *realisations* of the stochastic process.



Remark 16. *If run infinitely many times, the average of the runs will converge to the deterministic solution in both cases. However, when σ is (much) larger (for the same value of k), the uncertainty/variance in each individual run is (much) larger, which in turns makes the prediction of the time evolution for an individual run (much) more uncertain.*

In the financial world the uncertainty/variance of a stock is known as the volatility. *It provides a measure of the risk associated with the (prediction of the price of the) considered stock.*

3 Nonlinear ODE models of order 1

3.1 Non-Malthusian population growth: the logistic equation

Nonlinear ODE models of order 1

First order nonlinear ODE models are written under the generic form:

$$\boxed{\dot{x} = f(x), \quad x \in \mathbb{R}, \quad f(\cdot) : \mathbb{R} \to \mathbb{R}, \text{ "smooth" function}} \tag{6}$$

where $f(\cdot) : \mathbb{R} \to \mathbb{R}$ is a **"smooth" *function***.[5]

Finding the analytical solution of (6), i.e., finding $x(t, x_0)$, is, in general, no longer possible unless a closed form solution can be obtained for $\int \frac{1}{f(x)}\, dx = \int dt$. Since finding the analytical solution of nonlinear ODEs is in general not possible, we will focus in the rest of this chapter on presenting analysis methods that allow to qualitatively understand the behaviour of nonlinear ODE models without the need to find their solution explicitly.

In general, the asymptotic stability analysis of nonlinear models of order 1 is performed using phase line and bifurcation diagrams as we will see next.

Non-Malthusian population growth: the logistic equation

We consider the *non-Malthusian population growth* model in which the reproduction rate takes into account the "competition for resources". Intuitively, the net growth rate or reproduction rate per individual (or per cell) needs to decrease as the population increases due to this competition for resources.

Consider that $x(t)$ represents the number of cells at time instant t. The following ODE model describes population growth under competition for resources:

$$\boxed{\dot{x} = \overbrace{\underbrace{rx\left(1-\frac{x}{k}\right)}_{\text{Resources}}}^{=f(x)} = \left(\underbrace{r\left(1-\frac{x}{k}\right)}_{\substack{\text{non-constant growth rate} \\ \text{per cell}}}\right) x = \underbrace{rx}_{\text{"growth rate"}} \underbrace{-\frac{rx^2}{k}}_{\text{"death rate"}}} \tag{7}$$

where r and k are *positive* parameters.

As we can see, the net growth rate per individual (or per cell) $r\left(1-\frac{x}{k}\right)$ is not constant any more and changes as the number of individuals in the population changes (it decreases as the population increases).

Eq. (7) is also known as the *logistic equation* while k is known as the *carrying capacity* of the environment (we will see why later).

In this particular case and rather exceptionally, a closed form solution to (7) can be found:

$$\boxed{x(t) = \frac{k}{1 + \frac{1}{C}e^{-rt}}, \quad C = \frac{x_0}{k - x_0}}$$

This solution indicates that $x \to k$ as $t \to \infty$.

Remark 17. *The solution of (7) can easily be obtained as follows.*
Consider (7): $\frac{dx}{dt} = rx\left(1-\frac{x}{k}\right)$. *This can be rewritten:*

$$\frac{1}{x\left(1-\frac{x}{k}\right)} dx = r dt$$

Furthermore, we can also write:

$$\frac{1}{x\left(1-\frac{x}{k}\right)} = \frac{a}{x} + \frac{b}{1-\frac{x}{k}}$$

[5] A function is said to be "smooth" if it is $\mathcal{C}^k$, i.e., continuously differentiable at least k times, where $k \geq 2$.

By identification, this yields $a = 1$ *and* $b = \frac{1}{k}$.

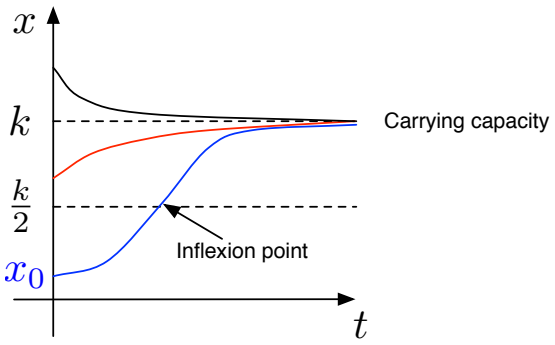
Therefore, eq. (7) *becomes:*

$$\begin{aligned}
\frac{1}{x\left(1-\frac{x}{k}\right)}dx &= rdt \\
\int\left(\frac{1}{x}+\frac{\frac{1}{k}}{1-\frac{x}{k}}\right)dx &= r\int dt \\
\int\left(\frac{1}{x}+\frac{1}{k-x}\right)dx &= r\int dt \\
\ln(x)-\ln(k-x) &= rt+\tilde{C} \\
\ln\left(\frac{x}{k-x}\right) &= rt+\tilde{C} \\
\frac{x}{k-x} &= e^{rt}\underbrace{e^{\tilde{C}}}_{=C} \\
\frac{k-x}{x} &= \frac{e^{-rt}}{C} \\
\frac{k}{x}-1 &= \frac{e^{-rt}}{C} \\
x(t) &= \frac{k}{1+\frac{1}{C}e^{-rt}}
\end{aligned}$$

Furthermore, using the definition of the initial condition, i.e., $x(0) = x_0$*, it is easy to show that* $C = \frac{x_0}{k-x_0}$.

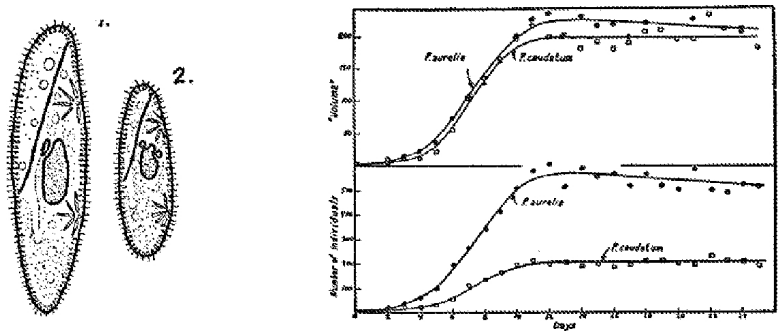
The logistic equation

Time solution: $\boxed{x(t) = \dfrac{k}{1+\frac{1}{C}e^{-rt}}, \quad C = \dfrac{x_0}{k-x_0}}$



Looking at the time solution, we see that any initial condition (except $x_0 = 0$) yields a solution which asymptotically converges to k. We now understand why k is called the *carrying capacity*. It represents the final population size that the resources present in the environment can sustainably carry in the long-term.

Remark 18 (Gause's 1934 Experiments)**.** *G.F. Gause carried out experiments in 1934, involving Paramecium caudatum and Paramecium aurelia, which clearly show logistic growth:*

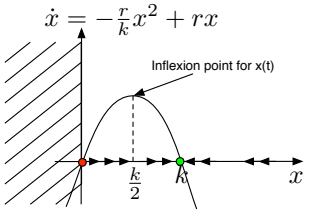


(Time evolution of # individuals and volume of P. caudatum and P. aurelia, cultivated separately, medium changed daily, 25 days.)

3.2 Stability analysis of the logistic equation

$$\boxed{\dot{x} = rx\left(1 - \frac{x}{k}\right)}$$

- Fixed points and flow:
  - Fixed points: $\dot{x}|_{x=x^*} = 0 \Leftrightarrow f(x^*) = 0$
    - Here, $f(x^*) = rx^*\left(1 - \frac{x^*}{k}\right) = 0 \Rightarrow \begin{cases} x^* = 0 \\ x^* = k \end{cases}$
  - Flow:
$$\begin{cases} 0 < x < k \Rightarrow \dot{x} > 0 \Rightarrow x \nearrow \\ x > k \Rightarrow \dot{x} < 0 \Rightarrow x \searrow \end{cases}$$
- Phase plane: $\dot{x}$ vs x



  Since x represents the number of bacteria in a population, x must be positive and thus we are not interested in the region $x < 0$.
- Asymptotic stability of fixed points: $x^* = 0$ is unstable (any slight perturbation from the fixed point $x^* = 0$ will cause the solution to leave the neighbourhood of $x^* = 0$);

  $x^* = k$ is asymptotically stable, i.e.,
  - stable (any slight perturbation from the fixed point $x^* = k$ will cause the solution to remain in the vicinity of $x^* = k$)
  - attractive (any slight perturbation causes the solution to return to $x^* = k$)
- Attractors: $x^* = k$

Remark 19. *Again, we see through the phase plane analysis that the attractor is $x^* = k$. k thus represents the long-term population number that the system can sustainably carry in the long-term – hence the name 'carrying capacity'.*

Remark 20. *Looking at the phase plot of $\dot{x}$ vs x we can confirm the "shape" of the time solution. For $x < \frac{k}{2}$, we see that $x(t)$ increases faster and faster (since $\dot{x}$ increases (i.e., $\ddot{x} > 0$); look at the plot of $\dot{x}$ vs x) until $x = \frac{k}{2}$, after that $x(t)$ increases slower and slower (since $\ddot{x} < 0$) until $x(t) \to k$. In other words, the population initially grows in an accelerating fashion (until reaching half of the carrying capacity) and then the growth slows down until the carrying capacity is reached. This is why we have an inflexion point at $x = \frac{k}{2}$ on the graph of the time solution $x(t)$ vs t.*

3.3 Stability analysis of nonlinear ODE models of order 1

Consider a nonlinear ODE model of order 1:

$$\boxed{\dot{x} = f(x), \quad x \in \mathbb{R}, \quad f(\cdot) : \mathbb{R} \to \mathbb{R}, \text{ "smooth" function}}$$

1. Global stability analysis (only for models of order 1)
   - Find all the fixed points: $\{x^* : f(x^*) = 0\}$ and put them on the phase line x of the plot $\dot{x}$ vs x.
   - Find the flow between the fixed points and indicate them on the phase line x of the plot $\dot{x}$ vs x.
   - Conclude what the stability of the fixed point(s) is.
   - Find the long-term behaviour of the system, i.e., its attractors. Note that $+\infty$ and $-\infty$ can be attractors.
2. Local/linear stability analysis (possible for all orders)
   - Find the fixed points.
   - Linearise the dynamics around *each* fixed point.
   - Study the stability of the corresponding linear systems ($eig(A)$).
   - Link together the local stability information around each fixed point to establish a complete picture of the attractors.

Remark 21. *Performing a global stability analysis using a phase line analysis is very useful for models of order* 1*. The phase plane analysis only gives a local picture for models of order* 2 *(as we will see in the next chapter). Finally, the phase space analysis is quite difficult to perform (and not very useful) for models of order* 3 *and higher.*

Remark 22. *The long-term behaviour (also called the* asymptotic *behaviour) of the system is typically dependent on the initial condition (this is true for models of any order).*

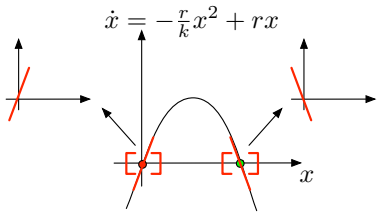
3.4 Linear stability analysis of ODE models of order 1

For **linear** ODE models of any order, global stability analysis and local stability analysis are the same. For nonlinear ODE models of order ≥ 2, local stability analysis is different from global stability analysis.[6]

- Find the fixed points of the system: $f(x^*) = 0$.

[6] For nonlinear ODE models of order 1, local and global stability are the same since in this particular situation the state variable is restricted to move on the real line (this will become clearer as we cover the next chapters).

- Examine the close neighbourhood of the fixed points, i.e., analyse the local stability of the fixed points by considering *small perturbations* around them.



Linear stability analysis of ODE models of order 1

Consider the dynamics of the system when x is "close to" the fixed point x^*, i.e., consider $\dot{x} = f(x)$ when $\boxed{x = x^* + \xi}$ with $\xi = (x - x^*)$ "small", i.e., $\boxed{|\xi| \ll 1}$:

$$\begin{aligned}\frac{dx}{dt} &= \frac{d\xi}{dt} \\ &= f\left(x^* + \xi\right) \\ &= \underbrace{f\left(x^*\right)}_{=0} + \left.\frac{df}{dx}\right|_{x=x^*} \underbrace{\xi}_{\text{"small"}} + \underbrace{\cancel{\mathcal{O}\left(\xi^2\right)}}_{H.O.T.\ (\text{"very small"})} \qquad \text{(Taylor series expansion)}\end{aligned}$$

So, we have:

$$\boxed{\frac{d\xi}{dt} \approx \left.\frac{df}{dx}\right|_{x=x^*} \xi} \text{ (linear system)}$$

$$\Rightarrow \boxed{\xi(t) \approx \xi_0 e^{\left.\frac{df}{dx}\right|_{x=x^*} t}}$$

Local stability analysis (only two possibilities):

- $\left.\frac{df}{dx}\right|_{x=x^*} > 0 \Rightarrow \dot{\xi}\xi > 0 \Rightarrow |\xi| \nearrow \quad \Rightarrow x = x^*$ is unstable
- $\left.\frac{df}{dx}\right|_{x=x^*} < 0 \Rightarrow \dot{\xi}\xi < 0 \Rightarrow |\xi| \searrow \quad \Rightarrow x = x^*$ is locally asymptotically stable, i.e., locally stable and attractive

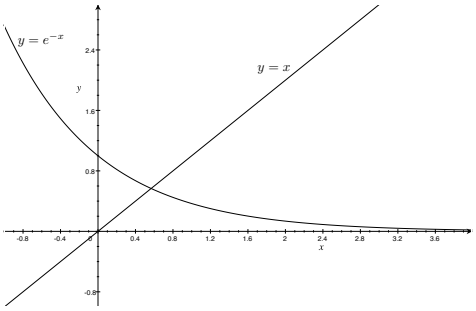
Linear stability analysis of the logistic equation

For $\dot{x} = rx\left(1 - \frac{x}{k}\right)$, we have $\boxed{\frac{df}{dx} = r - \frac{2xr}{k}}$

- $\left.\frac{df}{dx}\right|_{x=0} = r > 0 \Rightarrow \dot{\xi}\xi > 0 \Rightarrow |\xi| \nearrow \quad \Rightarrow x = 0$ is unstable
- $\left.\frac{df}{dx}\right|_{x=k} = -r < 0 \Rightarrow \dot{\xi}\xi < 0 \Rightarrow |\xi| \searrow \quad \Rightarrow x = k$ is locally asymptotically stable, i.e., locally stable and attractive

Remark 23. *Note that if* $\left.\frac{df}{dx}\right|_{x=x^*} = 0 \Rightarrow x^*$ *is* marginally stable *and we* cannot *deduce the stability of the nonlinear system around* $x = x^*$ *using linear stability analysis. This is a consequence of the* Hartman-Grobman Theorem *(also called the linearisation theorem). To illustrate this, consider the system* $\dot{x} = ax^3$ *where* $a \in \mathbb{R}$ *is a parameter and* $x \in \mathbb{R}$ *is the dependent variable. The linearisation of* $\dot{x} = ax^3$ *around its fixed point* $x^* = 0$ *is given by* $\dot{x} = 0$*, which is marginally stable irrespective of the parameter* a*. A graphical phase line analysis, however, shows that* $x^* = 0$ *is asymptotically stable for* $a < 0$ *and unstable for* $a > 0$*. As this example shows, the stability of the fixed point* $x^* = 0$ *of the initial nonlinear ODE* $\dot{x} = ax^3$ *cannot be deduced from the stability of* $\dot{x} = 0$ *(i.e., from the stability of the system linearised around the fixed point).*

Remark 24. *Note that the fixed points can also be obtained graphically. For example, if* $f(x^*) = x^* - e^{-x^*} = 0$*, the solution* $x^* = e^{-x^*}$ *is difficult to find analytically (transcendental equation). However, the solution can be easily approximated as the intersection of two graphs: the graphs* $y = x$ *and the graph* $y = e^{-x}$*.*



Furthermore, MATLAB fuctions such as `fsolve` *or* `solve` *can be used to find the zeros of* $f(x^*) = 0$*. Note that* `fsolve` *only gives you one solution, which is found based on a given initial guess. Thus, to find all the zeros with* `fsolve` *you need to try several initial guesses.*

3.5 Non-dimensionalisation (also called "renormalisation")

The goal of non-dimensionalisation is to reduce the number of parameters appearing in the equations.

Non-dimensionalisation can be very important in biological models where, typically, the number of uncertain parameters can be very large in the original model. Non-dimensionalising the model and therefore reducing the number of parameters is very useful to focus parameter-dependent analyses (such as parameter sensitivity analysis and bifurcation analysis) on a fewer number of parameters.

Consider the following population growth model where $N(t)$ represents the number of cells in the population at time t:

$$\dot{N} = \underbrace{RN\left(1 - \frac{N}{K}\right)}_{\text{comp. for resources}} \underbrace{- \frac{BN^2}{A^2 + N^2}}_{\text{predatory action}} \tag{8}$$

This model contains 4 positive parameters R, K, A, and B.

As we will see in what follows, using non-dimensionalisation, we can reduce the number of parameters from 4 to 2.

In order to reduce the number of parameters in (8), we will proceed as follows:

- Write each variable (dependent and independent – in this example, N and t) as a product of a new variable and a still-to-be-determined positive constant.
- Substitute into the equations, simplify, and collect terms.
- Finally, pick values for the constants so that the equations (in this example, there is only one differential equation, but in other examples there may be several) have as few remaining parameters as possible.

The procedure can be done in many ways (depending on how you collect terms, etc.), so different people may get different solutions. If everything is done properly, the new variables (hereafter $\hat{N}$ and $\hat{t}$) will be "non-dimensional", i.e., they will have no units (hence the name *non-dimensionalisation*).

Let $N(t) = \alpha\hat{N}(t)$ and $t = \beta\hat{t}$. Eq. (8) then writes:

$$\frac{d\left(\alpha\hat{N}(t)\right)}{d\left(\beta\hat{t}\right)} = R\alpha\hat{N}\left(1 - \frac{\alpha\hat{N}}{K}\right) - \frac{B\alpha^2\hat{N}^2}{A^2 + \alpha^2\hat{N}^2}$$

$$\Leftrightarrow \frac{d\left(\hat{N}\left(\hat{t}\right)\right)}{d\hat{t}} = R\beta\hat{N}\left(1 - \frac{\alpha}{K}\hat{N}\right) - \frac{\frac{B}{\alpha}\beta\hat{N}^2}{\frac{A^2}{\alpha^2} + \hat{N}^2} \tag{9}$$

Look at eq. (9). To reduce the total number of parameters, we'd like to make, for example, $\frac{A^2}{\alpha^2} = 1$ and $\frac{B}{\alpha}\beta = 1$ which can be obtained by taking $\alpha = A$ and $\beta = \frac{\alpha}{B} = \frac{A}{B}$. We then have:

$$\frac{d\hat{N}}{d\hat{t}} = \underbrace{\frac{RA}{B}}_{=r}\hat{N}\left(1 - \underbrace{\frac{A}{K}}_{\frac{1}{k}}\hat{N}\right) - \frac{\hat{N}^2}{1 + \hat{N}^2}$$

$$\Leftrightarrow \boxed{\frac{d\hat{N}}{d\hat{t}} = r\hat{N}\left(1 - \frac{1}{k}\hat{N}\right) - \frac{\hat{N}^2}{1 + \hat{N}^2}}$$

As we can see the new parameters $r = \frac{RA}{B}$ and $k = \frac{K}{A}$ combine several parameters of the initial model. Following the described procedure, we notice that, **for each variable (dependent and independent) of the model, we can eliminate one parameter of the model (this property is true in general for any non-dimensionalisation). Therefore, using non-dimensionalisation, the number of parameters can always be reduced by the total number of variables (dependent + independent). Here, we started with 4 parameters and reduced this number to 2 since there were 2 variables in total (the dependent variable N and the independent variable t).**

Once the model has been non-dimensionalised and, consequently, the number of parameters in the model reduced to its minimum, one may wonder how the long-term behaviour of the system changes when these remaining parameters are changed. This question is the basis of *bifurcation analysis* and is introduced in the next section.

3.6 Bifurcations for nonlinear ODE models of order 1

Consider:

$$\boxed{\dot{x} = f(x, r)}$$

where r is a parameter and $f(\cdot) : \mathbb{R} \times \mathbb{R} \to \mathbb{R}$ is a "smooth" *function*.

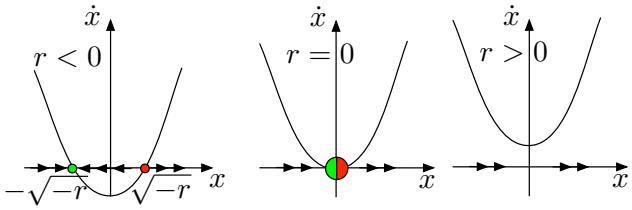
Bifurcation
A bifurcation occurs when a change in the parameter(s) of the model produces a qualitative (or "large") change in the long-term behaviour (of the attractors) of the system, e.g., :

- the number of attractors (e.g., fixed points) changes,
- the type of attractors changes (e.g., from fixed point to limit cycle),
- the stability of attractors (e.g., fixed points or limit cycles) changes.

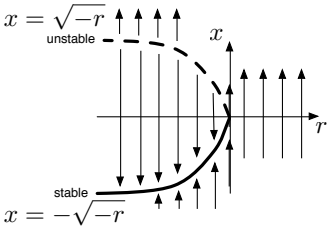
3.6.1 Saddle-node Bifurcation (also called "Blue Sky" Bifurcation)

$$\boxed{\dot{x} = r + x^2, \quad x \in \mathbb{R}}$$

Consider different values for the parameter r:



Saddle-node *bifurcation diagram*:



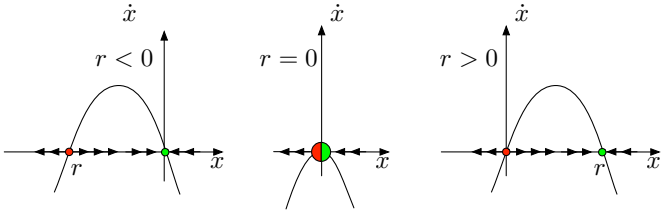
When increasing r from negative to positive values, the two fixed points coalesce at $r = 0$ and disappear for $r > 0$. The saddle-node bifurcation is characterised by a merging and subsequent disappearance (or sudden creation depending how the parameter is varied) of a stable and an unstable fixed point. The name blue sky bifurcation has been coined to refer to the sudden appearance (or disappearance) of two fixed points "out of the blue".

3.6.2 Transcritical Bifurcation

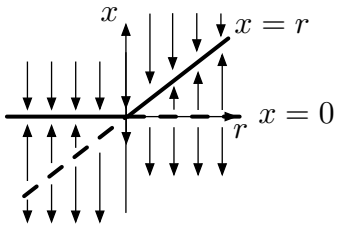
$$\dot{x} = rx - x^2 = x(r - x), \quad x \in \mathbb{R}$$

In this case, there are always two fixed points: one at $x = 0$ and another at $x = r$

Consider different values for the parameter r:



Transcritical bifurcation diagram:



At $r = 0$, there is a reversal in the stability of the fixed points: what used to be stable becomes unstable and vice versa. The transcritical bifurcation is characterised by a merging and subsequent stability reversal of a stable and an unstable fixed point.

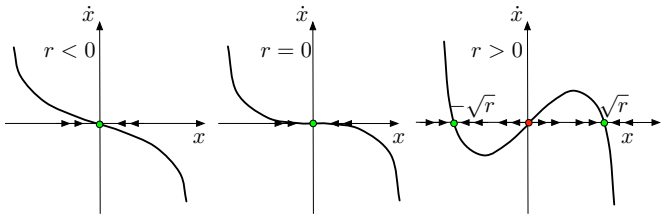
Remark 25. *This is the type of bifurcation we would observe for the logistic equation* (7) *if we considered k as a parameter that could also take negative values.*

3.6.3 Pitchfork Bifurcation

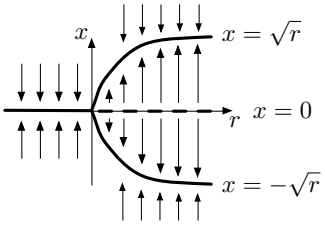
$$\boxed{\dot{x} = rx - x^3 = x\left(r - x^2\right), \quad x \in \mathbb{R}}$$

In this case, there is always one fixed point at $x = 0$. Furthermore, if $r > 0$, there are two other fixed points at $x = \sqrt{r}$ and $x = -\sqrt{r}$.

Consider different values for the parameter r:



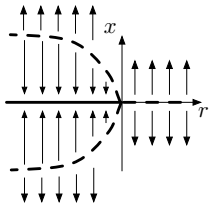
Pitchfork bifurcation diagram (supercritical):



If, when changing the value of the parameter, two new *stable* fixed points appear while the third fixed point is now unstable instead of stable, the corresponding pitchfork bifurcation is said to be *supercritical*.

On the contrary, if, when changing the value of the parameter, two new *unstable* fixed points appear while the third fixed point is now stable instead of unstable, the corresponding pitchfork bifurcation is said to be *subcritical*.

Pitchfork bifurcation diagram (subcritical): $\boxed{\dot{x} = rx + x^3 = x\left(r + x^2\right), \quad x \in \mathbb{R}}$



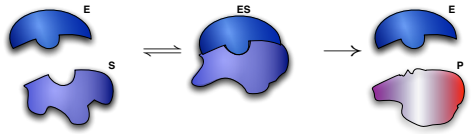
Remark 26. *The pitchfork bifurcation is common to systems that present symmetry. For example, the dynamics that we have considered here, i.e.,* $\dot{x} = x\left(r \pm x^2\right)$ *do not change under the change of variable* $\tilde{x} = -x$.

3.7 Summary of behaviours for NL ODE models of order 1

- Motions (solutions) are on the real line, i.e., $x \in \mathbb{R}$
- Attractors are either the fixed points or $\pm\infty$ (no oscillatory or other types of behaviour)
- Three types of bifurcation can occur:
  - Saddle node
  - Transcritical
  - Pitchfork (subcritical or supercritical)

3.8 Enzymatic reactions and the law of mass action

Now that we know how to analyse nonlinear ODEs of order 1, let us apply this knowledge to the analysis of enzymatic reactions. In the next section, we will use the law of mass action to propose a simple 4th order nonlinear ODE model for enzymatic reactions. We will then reduce this model to a first order nonlinear ODE model that we will analyse using a phase line approach.



In molecular biology, certain types of proteins, called *enzymes* act as catalysts for biochemical reactions. In particular, they facilitate such reactions, converting substrates into products, while remaining basically unchanged. In a simple enzymatic reaction, the enzyme E attaches to the substrate S, thereby forming a complex ES. The complex formation produces bond changes and distortions in the substrate shape which facilitate the creation of the product P. In other words, the enzymes act as "pliers" that place an appropriate stress to help break a bond; they also may bring substrates together, or they may help place a chemical group on a substrate.

Like all catalysts, enzymes work by lowering the activation energy for a reaction, thus dramatically increasing the rate of the reaction. Most enzyme reaction rates are millions of times faster than those of comparable uncatalyzed reactions.

Remark 27. *Almost all processes in a biological cell need enzymes to occur at significant rates. Since enzymes are selective for their substrates and speed up only a few reactions from among many possibilities, the set of enzymes made in a cell determines which metabolic pathways occur in that cell.*

Remark 28. *The modelling of (bio-)chemical reactions is typically done using the* law of mass action *which gives ODEs for the concentrations of the species involved in the reactions. This law states that,* when two or more reactants are involved in a reaction, their reaction rates (at constant temperature) are proportional to the product of their concentrations*. For example, the following chemical reaction:*

$$X + Y \xrightarrow{\alpha} Z$$

yields, using the law of mass-action, the following dynamics:

$$\frac{d[X]}{dt} = -\alpha[X][Y] \tag{10}$$
$$\frac{d[Y]}{dt} = -\alpha[X][Y] \tag{11}$$
$$\frac{d[Z]}{dt} = +\alpha[X][Y] \tag{12}$$
$$\tag{13}$$

where $[X]$ (resp. $[Y]$, $[Z]$) represents the concentration of chemical species X (resp. Y, Z).

The law of mass action relies on two assumptions:

- *the medium must be well mixed: this allows to obtain ODEs instead of PDEs since, under this assumption, the concentrations may depend on time but not on space.*
- *the number of each species must be large: this allows to obtain ODEs instead of SDEs. If the number of species is small then stochastic models (e.g., SDEs) taking into account individual random collisions between species must be used.*

Enzymatic reaction:

$$E + S \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} ES \xrightarrow{k_2} E + P$$

Law of mass action: For a simple enzymatic reaction we have 4 species ⇒ 4 ODEs

$$\frac{d[ES]}{dt} = k_1[E][S] - k_{-1}[ES] - k_2[ES] \tag{14}$$

$$\frac{d[E]}{dt} = -k_1[E][S] + k_{-1}[ES] + k_2[ES] \tag{15}$$

$$\frac{d[S]}{dt} = -k_1[E][S] + k_{-1}[ES] \tag{16}$$

$$\frac{d[P]}{dt} = k_2[ES] \tag{17}$$

Remark 29. *Note that the biochemical degradation of the 4 species involved in the reactions has been neglected. This can be justified by the general stability of the species coupled with the typical short time-scale of enzymatic reactions (less than an hour for the reactions to be complete whereas degradation of the species involved in the enzymatic reaction typically takes tens of hours or several days).*

3.8.1 Elimination of variables – model reduction: the Michaelis-Menten and the Hill equations

Elimination of variables – model reduction through time scale separation

The goal here is to reduce the number of dependent variables in the system by using the fact that certain quantities are conserved or by making certain assumptions (e.g., the *quasi-stationary* assumption as we will see shortly).

We will illustrate the procedure of elimination of variables on the particular example of the enzymatic reaction, and show how the *Michaelis-Menten* model can be obtained as a result of this.

1. Conservation laws

   - (14) + (15) ⇒ $\frac{d[ES]}{dt} + \frac{d[E]}{dt} = 0$

     $$\Rightarrow [ES] + [E] = [E]_0 \tag{18}$$

   - (15) - (16) - (17) ⇒ $\frac{d[E]}{dt} - \frac{d[S]}{dt} - \frac{d[P]}{dt} = 0$

     $$\Rightarrow [E] = [S] + [P] + \kappa \tag{19}$$

     where $[E]_0$ is a constant representing the initial concentration of enzyme before the reaction starts[7], and κ is an integration constant.

   Thus, just looking for conserved quantities, we have reduced the initial 4^{th} order model to a 2^{nd} order model.

2. Quasi-stationary approximation (time scale separation)

   Leonor Michaelis and Maud Leonora Menten formulated in 1913 an approach that allows one to reduce the problem even further, by doing an approximation called the "quasi-stationary"

[7] This conservation law is not surprising. Indeed, the enzyme is either free or present in the ES complex, but it is neither produced nor consumed according to the model we are considering here for enzymatic reactions (remember that we have neglected enzyme degradation and we also did not consider enzyme production, which could for example occur through expression of the gene that codes for the enzyme).

approximation. This approximation is typically used in multiple time scale systems where some parts of the dynamics are very fast while others are rather slow.

In an enzymatic reaction, the time scales of the different reactions are typically very different. This means that certain reactions will reach their equilibrium much faster than others. A common assumption is then to neglect the short transient needed to reach equilibrium for the fast reactions, and thus to consider that the corresponding species reach their steady state almost instantaneously. This is known as the *quasi-stationary approximation*.

In particular, in an enzymatic reaction, the enzyme-substrate complex reaches its equilibrium state very fast. A typical assumption is thus to assume that the enzyme-substrate complex is almost instantly at steady state and therefore stays more or less constant at the time scale of the product producing reaction.

- $\frac{d[ES]}{dt} \approx 0$

(14) $\Rightarrow [ES] \approx \underbrace{\frac{k_1}{k_{-1}+k_2}}_{=\tilde{K}_M}[E][S] = \tilde{K}_M[E][S]$

(18) $\Rightarrow [ES] \approx \tilde{K}_M\left([E]_0 - [ES]\right)[S] \Rightarrow [ES] \approx \frac{\tilde{K}_M[E]_0[S]}{1+\tilde{K}_M[S]}$

(17), (18), (19) $\Rightarrow \frac{d[S]}{dt} = -\frac{d[P]}{dt} - \underbrace{\frac{d[ES]}{dt}}_{\approx 0} \approx -k_2[ES]$

Using the conservation laws and the quasi-stationary approximation, we can thus reduce the model (14)-(17) and end up with a first order nonlinear ODE model:

$$\Rightarrow \boxed{\frac{d[S]}{dt} \approx -\frac{d[P]}{dt} \approx -V_{\max}\frac{[S]}{K_M+[S]}} \quad \text{(the } \textit{Michaelis-Menten} \text{ equation)}$$

with

$$V_{\max} = k_2[E]_0, \quad K_M = \frac{k_{-1}+k_2}{k_1}$$

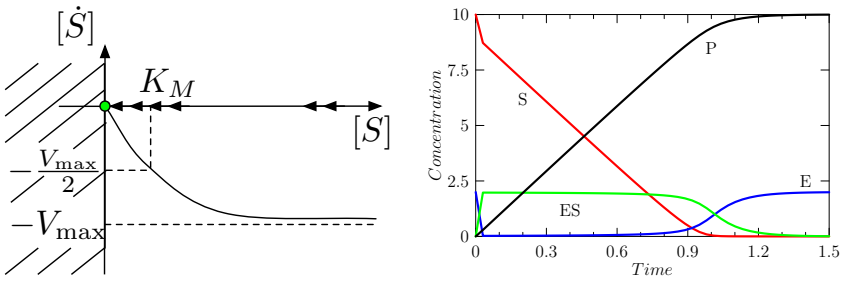
Remark 30. *Note that V_{max} is proportional to $[E]_0$, a fact that is commonly exploited in practice (all it takes to increase the speed of the reaction is to increase the initial amount of enzymes accordingly).*

Remark 31. *For small substrate concentrations, i.e., small [S], we see that* $\frac{d[S]}{dt} \approx -\frac{V_{max}}{K_M}[S] = -\frac{k_2[E]_0}{K_M}[S]$.

3.8.2 The Michaelis-Menten equation

Let's now analyse the behaviour of the Michaelis-Menten ODE by performing a phase line analysis of it.

$$\boxed{\frac{d[S]}{dt} \approx -\frac{d[P]}{dt} \approx -V_{\max}\frac{[S]}{K_M+[S]}} \quad \text{(the } \textit{Michaelis-Menten} \text{ equation)}$$



On the phase line, we see that when the concentration of the substrate $[S]$ is large, the rate of depletion of $[S]$ (i.e., $[\dot{S}]$) is almost constant. On the contrary, we see that the rate of depletion of substrate $[\dot{S}]$ becomes smaller when $[S]$ is smaller (since $\left|[\dot{S}]\right|$ becomes smaller).

Next to the phase line, we can see the plot of the time evolution of $[E]$, $[S]$, $[ES]$, and $[P]$. This plot has been obtained by numerically integrating (14)-(17) in Matlab. In the central portion of the plots one can observe the relatively constant concentrations of $[ES]$ and $[E]$ (which corresponds to $\frac{d[ES]}{dt} = -\frac{d[E]}{dt} \approx 0$). At the same time, the rate of change of $[S]$ and $[P]$ are constant over this period.

Justifying the quasi-stationary assumption through non-dimensionalisation and time-scale separation

Reminder: the goal of non-dimensionalisation is to reduce the number of parameters appearing in the equations.

Consider (14) and (16). Using (18) we get:

$$\begin{cases} \frac{d[ES]}{dt} = k_1[E]_0[S] - (k_1[S] + k_{-1} + k_2)[ES] \\ \frac{d[S]}{dt} = -k_1[E]_0[S] + (k_1[S] + k_{-1})[ES] \end{cases}$$

Dividing both equations by $[E]_0[S]_0$, and posing $x = \frac{[ES]}{[E]_0}$, $y = \frac{[S]}{[S]_0}$, and $\epsilon = \frac{[E]_0}{[S]_0}$ leads to:

$$\begin{cases} \frac{1}{[S]_0}\frac{dx}{dt} = k_1 y - \left(k_1 y + \frac{k_{-1}+k_2}{[S]_0}\right) x \\ \frac{1}{[E]_0}\frac{dy}{dt} = -k_1 y + \left(k_1 y + \frac{k_{-1}}{[S]_0}\right) x \end{cases}$$

Finally, posing $\tau = k_1[E]_0 t$, we obtain

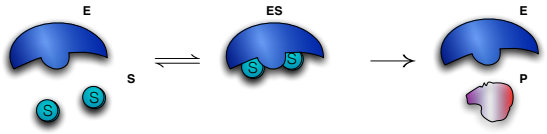
$$\begin{cases} \epsilon\frac{dx}{d\tau} = y - \left(y + \frac{k_{-1}+k_2}{k_1[S]_0}\right) x \\ \frac{dy}{d\tau} = -y + \left(y + \frac{k_{-1}}{k_1[S]_0}\right) x \end{cases}$$

It is common for enzymatic reactions to have an initial concentration of substrates that is much larger than the initial concentration of enzymes, i.e., $\epsilon = \frac{[E]_0}{[S]_0} \ll 1$.[8] Therefore, we see that the dynamics of x is much faster than the dynamics of y, since $\frac{dx}{d\tau} = \frac{1}{\epsilon}(\dots)$ and $\frac{dy}{d\tau} = (\dots)$. The assumption that x, i.e., $[ES]$, reaches its steady state very quickly (almost instantaneously with respect to the dynamics of y, i.e., $[S]$), is therefore justified and the Michaelis-Menten assumption holds.

[8] It can be shown that another choice of non-dimensional variables can lead to the less conservative condition $\epsilon = \frac{[E]_0}{[S]_0+K_M} \ll 1$. With this condition, we can see that one doesn't need $[E]_0 \ll [S]_0$ for the quasi-stationary approximation to hold. It is enough that K_M is very large, i.e., that the rate of formation of complex k_1 is very small compared to $k_{-1} + k_2$ (sum of dissociation rates). An interesting paper about this is: L. A. Segel and M. Slemrod, "The Quasi-Steady-State Assumption: A Case Study in Perturbation", SIAM Review, vol. 31, no. 3, pp. 446–477, Sep. 1989.

Enzymatic cooperative reactions – The Hill equation

Sometimes several substrates need to bind the enzyme for the enzymatic reaction to take place. When this is the case, the enzymatic reaction is said to be *cooperative*.



A model for the enzymatic reaction with cooperativity is:

$$E + nS \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} ES \xrightarrow{k_2} E + P$$

where ES represents the enzyme-n-substrates complex and n is called the *cooperativity coefficient*.

Law of mass action: 4 species $\Rightarrow$ 4 ODEs

$$\frac{d[ES]}{dt} = k_1[E][S]^n - k_{-1}[ES] - k_2[ES] \tag{20}$$

$$\frac{d[E]}{dt} = -k_1[E][S]^n + k_{-1}[ES] + k_2[ES] \tag{21}$$

$$\frac{d[S]}{dt} = n\left(-k_1[E][S]^n + k_{-1}[ES]\right) \tag{22}$$

$$\frac{d[P]}{dt} = k_2[ES] \tag{23}$$

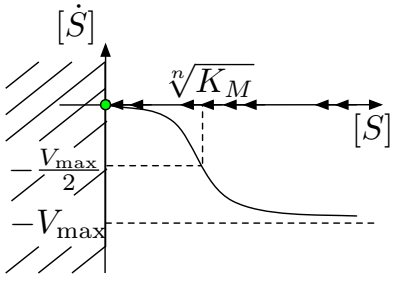
3.8.3 The Hill equation

Using a similar model reduction approach as for the non-cooperative enzymatic reactions we saw before (Michaelis-Menten), it is easy to see that the following 1^{st} order nonlinear ODE model is obtained:

$$\boxed{\frac{d[S]}{dt} \approx -\frac{d[P]}{dt} \approx -V_{\max}\frac{[S]^n}{K_M + [S]^n}} \quad \text{(the } \textit{Hill} \text{ equation)}$$

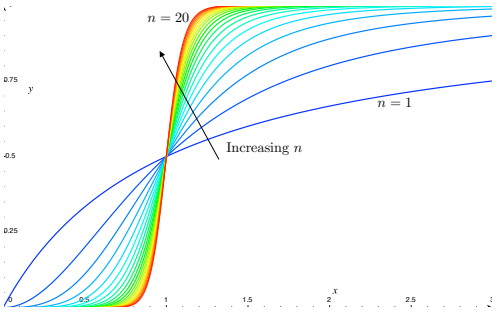
with

$$V_{\max} = nk_2[E]_0, \quad K_M = \frac{k_{-1} + k_2}{k_1}$$



The Hill equation: effect of the cooperativity coefficient n

The Hill function is defined as $h(x) = V_{\max}\frac{x^n}{K_M + x^n}$. The effect of the Hill coefficient n is illustrated hereafter for $V_{\max} = 1$ and $K_M = 1$:



For very large values of n, the Hill function approximates a step function[9], i.e., a function $h(x)$ defined as

$$h(x) = \begin{cases} 0, & \text{if } x \leq \sqrt[n]{K_M} \\ V_{\max}, & \text{if } x > \sqrt[n]{K_M} \end{cases}$$

This is very useful for a cell which can then use this type of "step-regulated" reaction as a switch since for low concentrations (i.e., $x \leq \sqrt[n]{K_M}$) nothing happens, while for high concentrations (i.e., $x > \sqrt[n]{K_M}$) the enzymatic reaction happens at its maximal rate $V_{\max}$.

Remark 32. *In the above simplified model for enzymatic reactions with cooperativity, we have neglected the fact that the binding of the n molecules of substrate to the enzyme does not take place at once but in a succession of steps:*

$$E + S \underset{k_{-1,1}}{\overset{k_{1,1}}{\rightleftharpoons}} ES$$

$$ES + S \underset{k_{-1,2}}{\overset{k_{1,2}}{\rightleftharpoons}} ES_2$$

$$\vdots$$

$$ES_{n-1} + S \underset{k_{-1,n}}{\overset{k_{1,n}}{\rightleftharpoons}} ES_n \xrightarrow{k_2} E + P$$

Remark 33. *If the first, second, ..., n^{th} steps (see Remark 32) are much faster than the last (product producing) step, the pure Hill function is justified. In general however, one gets:*

$$\frac{d[S]}{dt} = -V_{max} \frac{P([S])}{K_M + Q([S])} \tag{24}$$

where both $P([S])$ and $Q([S])$ are polynomials of the form:

$$P([S]) = [S]^n + \alpha_{n-1}[S]^{n-1} + \ldots + \alpha_1[S]$$
$$Q([S]) = [S]^n + \beta_{n-1}[S]^{n-1} + \ldots + \beta_1[S]$$

It has however remained common to use a classic Hill function to model cooperative enzymatic reactions, i.e., $[\dot{S}] = -V_{max} \frac{[S]^n}{K_M + [S]^n}$. However, to fit data to this latter model you might need to use a Hill function with a non-integer Hill coefficient (e.g., $n = 2.8$) instead of an integer Hill coefficient. This is due to the fact that a model of the form $[\dot{S}] = -V_{max} \frac{[S]^n}{K_M + [S]^n}$ is a mere approximation of the actual dynamics.

Remark 34. *Another aspect which we have neglected here is the fact that the n binding sites on the enzyme are usually not equivalent, which makes the biochemical model even more complicated. The corresponding dynamic can however still be approximated with an equation of the form of eq. (24).*

[9] This is only an approximation as the Hill function is "smooth" for any value of the Hill coefficient "n" whereas the step function is discontinuous at a point.

4 Linear ODE models of order 2 and higher

4.1 Some examples

4.1.1 A chemical example of a linear ODE model of order 2

Consider the chemical reaction:

$$X \underset{k}{\overset{k}{\rightleftharpoons}} Y$$

Using the law of mass action, the corresponding ODEs write:

$$[\dot{X}] = -k[X] + k[Y] \tag{25}$$

$$[\dot{Y}] = k[X] - k[Y], \quad k > 0 \tag{26}$$

To solve (25)-(26) analytically, we define the vector $\boldsymbol{x} = \begin{pmatrix} x_1 \\ x_2 \end{pmatrix} = \begin{pmatrix} [X] \\ [Y] \end{pmatrix}$ and rewrite the equation under the form $\dot{\boldsymbol{x}} = A\boldsymbol{x}$. We then use a change of variables in order to *diagonalise* the matrix A.

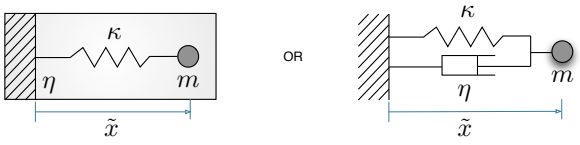
Remark 35. *The order of the model is defined as the number of* ***dependent*** *variables appearing in the ODE, i.e., the dimension of the vector* $\boldsymbol{x}$*. In our present case, the model is of order* 2 *since there are* 2 *dependent variables:* $[X]$ *and* $[Y]$*.*

Before we proceed with the diagonalisation of A, we introduce a few more examples of second order linear ODEs useful in (bio-)mechanics. The procedure to find the analytical solution of the linear ODE system (25)-(26) through the diagonalisation of A is explained in Section 4.2.

4.1.2 A mechanical example of a linear ODE model of order 2

In this section, we consider a classical mechanical example: the mass-spring-damper system. Mass-spring-damper systems are very useful in biomechanics to model the dynamics of musculo-skeletal systems such as, for example, the dynamics of your arms in reaching tasks or of your legs in walking, running or hopping motions.

Consider the mass-spring-damper system:



where m is the mass (in kilograms), κ is the spring constant (in newtons per meter), η is the damping coefficient (in newton-seconds per meter or kilograms per second), and x is the displacement (in meters) with respect to the resting position of the spring $\tilde{x}_0$, i.e., $x = \tilde{x} - \tilde{x}_0$ where $\tilde{x}$ is the displacement (in meters) of the mass relative to a fixed point of reference.

The equation of motion of the mass-spring-damper system is given by[10]:

$$m\ddot{x} = -\kappa x - \eta \dot{x} \tag{27}$$

which is equivalent to:

$$\boxed{\frac{m}{\eta}\frac{d^2x}{dt^2} + \frac{dx}{dt} + \frac{\kappa}{\eta}x = 0} \tag{28}$$

[10] For a quick reminder about the physics and equations of motion of the mass-spring-damper system, visit http://en.wikipedia.org/wiki/Damping#Example:_mass-spring-damper.

This is a second order linear ODE[11]. However, under some assumption, this second order linear ODE model can be reduced to a first order linear ODE model.

Reduction to a first order model: In the overdamped limit, i.e., when $\eta \gg m$, eq. (27) reduces to

$$\boxed{\dot{x} = -kx, \quad k = \frac{\kappa}{\eta}\sqrt{\frac{\eta}{m}} > 0}$$

This is easy to see by considering the following change of variables:

$$\tau^2 = \frac{m}{\eta}t^2 \Leftrightarrow \tau = \sqrt{\frac{m}{\eta}}t \tag{29}$$

Eq. (29) implies:

$$\frac{d\tau}{dt} = \sqrt{\frac{m}{\eta}}$$

$$\frac{dx}{dt} = \frac{dx}{d\tau}\frac{d\tau}{dt} = \sqrt{\frac{m}{\eta}}\frac{dx}{d\tau} \tag{30}$$

$$\frac{d^2x}{dt^2} = \frac{m}{\eta}\frac{d^2x}{d\tau^2} \tag{31}$$

Applying (29), (30), and (31) to (28), we obtain (when $\eta \gg m$):

$$\underbrace{\frac{m^2}{\eta^2}}_{\to 0}\frac{d^2x}{d\tau^2} + \sqrt{\frac{m}{\eta}}\frac{dx}{d\tau} + \frac{\kappa}{\eta}x = 0$$

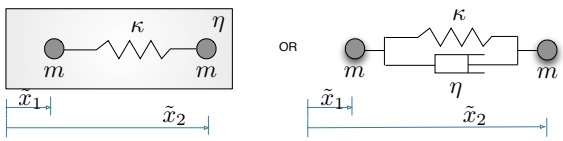
$$\frac{dx}{d\tau} + \underbrace{\frac{\kappa}{\eta}\sqrt{\frac{\eta}{m}}}_{=k}x = 0$$

$$\boxed{\frac{dx}{d\tau} = -kx, \quad k > 0}$$

The solution and analysis of this first order linear ODE model has been described previously (see the analysis of the Malthusian population growth model). Thus we see that the mass-spring-damper system has a simple exponentially decaying behaviour in the overdamped limit.

Another mechanical example of a linear ODEs model of order 2

Consider the following mass-spring-damper system:



In the overdamped limit, i.e., when $\eta \gg m$, the equations of motion are:

$$\dot{x}_1 = -k(x_1 - x_2) \tag{32}$$

$$\dot{x}_2 = -k(x_2 - x_1), \quad k > 0 \tag{33}$$

[11] This can be seen by rewriting the system as:

$$\dot{x} = y$$

$$\dot{y} = -\frac{\kappa}{m}x - \frac{\eta}{m}y$$

which indicates that the order of the system is 2.

Remark 36. *Without the overdamped limit assumption, the corresponding linear ODE model is of order* 4.

Again, to solve (32)-(33) analytically, we define the vector $\boldsymbol{x} = \begin{pmatrix} x_1 \\ x_2 \end{pmatrix}$ and rewrite the equation under the form $\dot{\boldsymbol{x}} = A\boldsymbol{x}$. We then use a change of variables in order to *diagonalise* the matrix A.

4.2 Diagonalisation, eigenvalues and eigenvectors

The system of equations (25)-(26) (or (32)-(33))
can be rewritten as:

$$\begin{pmatrix} \dot{x}_1 \\ \dot{x}_2 \end{pmatrix} = k \underbrace{\begin{pmatrix} -1 & 1 \\ 1 & -1 \end{pmatrix}}_{=A} \begin{pmatrix} x_1 \\ x_2 \end{pmatrix} \Leftrightarrow \dot{\boldsymbol{x}} = kA\boldsymbol{x} \tag{34}$$

To solve (34), we diagonalise A, i.e., we find its *eigenvalues* and *eigenvectors*.[12]

The diagonalisation allows to decouple the equations[13], and therefore to reduce the problem to finding the solution of 1^{st} order linear ODEs (which we now know how to do).

1. **Eigenvalues:** Solutions of $\boxed{\det(A - \lambda I) = 0}$. Here, we have: $\lambda_1 = 0$ and $\lambda_2 = -2$.
2. **Eigenvectors (normalised):** Solutions of $\boxed{A\boldsymbol{v} = \lambda \boldsymbol{v}}$, for each eigenvalue λ. Here, we have: $\boldsymbol{v}_1 = \frac{1}{\sqrt{2}} \begin{pmatrix} 1 \\ 1 \end{pmatrix}$ corresponding to $\lambda_1 = 0$ and $\boldsymbol{v}_2 = \frac{1}{\sqrt{2}} \begin{pmatrix} 1 \\ -1 \end{pmatrix}$ corresponding to $\lambda_2 = -2$.

The eigenvalues are found easily. They are the solutions of $\det(A - \lambda I) = \det \begin{pmatrix} -1-\lambda & 1 \\ 1 & -1-\lambda \end{pmatrix} = 0$. This gives the algebraic equation $(1+\lambda)(1+\lambda) - 1 = \lambda^2 + 2\lambda = 0$, whose solutions are $\lambda_1 = 0$ and $\lambda_2 = -2$.

Remark 37. *For linear ODE models of order* 2*, the eigenvalues can also be easily found by noting the following properties:* $\det(A) = \lambda_1\lambda_2$ *and trace*$(A) = \lambda_1 + \lambda_2$*. Here we have* $\lambda_1\lambda_2 = 0$ *and* $\lambda_1 + \lambda_2 = -2$ *which directly leads to* $\lambda_1 = 0$ *and* $\lambda_2 = -2$*.*

The eigenvectors are found separately for each eigenvalue:

1. For $\lambda_1 = 0$, we search the solution of $A\boldsymbol{v}_1 = \begin{pmatrix} -1 & 1 \\ 1 & -1 \end{pmatrix} \begin{pmatrix} v_a \\ v_b \end{pmatrix} = 0 \begin{pmatrix} v_a \\ v_b \end{pmatrix}$, i.e., the solution of
$$\begin{cases} -v_a + v_b = 0 \\ v_a - v_b = 0 \end{cases}$$
which leads to $v_a = v_b$. Now, under the normalisation constraint $v_a^2 + v_b^2 = 1$, we obtain the normalised eigenvector $\boldsymbol{v}_1 = \frac{1}{\sqrt{2}} \begin{pmatrix} 1 \\ 1 \end{pmatrix}$.
2. Similarly, for $\lambda_2 = -2$, we need to solve $A\boldsymbol{v}_2 = \begin{pmatrix} -1 & 1 \\ 1 & -1 \end{pmatrix} \begin{pmatrix} v_c \\ v_d \end{pmatrix} = -2 \begin{pmatrix} v_c \\ v_d \end{pmatrix}$, which, after normalisation, gives $\boldsymbol{v}_2 = \frac{1}{\sqrt{2}} \begin{pmatrix} 1 \\ -1 \end{pmatrix}$.

[12] The key requirement to be able to diagonalise a square matrix A of dimension $n \times n$ is to be able to find n linearly independent eigenvectors for A (see theorem on diagonalisation of real matrices in any textbook on linear algebra). A sufficient condition for this is that the eigenvalues of A are distinct, which indeed is the case here since $\lambda_1 \neq \lambda_2$. If it is not possible to find n linearly independent eigenvectors, one can still find an analytical expression for the solution, however the procedure followed to do so is somewhat more involved (it typically requires to perform a *Jordan block decomposition* of the matrix A) and the analytical solution $\boldsymbol{x}(t)$ has a different form.

[13] This means that, for all i, the equation for $\dot{x}_i$ only depends on x_i and not on any of the other state variables.

From the eigenvectors of A, we construct a new matrix V having the eigenvectors of A as columns:

$$V = \begin{pmatrix} | & | \\ \boldsymbol{v}_1 & \boldsymbol{v}_2 \\ | & | \end{pmatrix} = \frac{1}{\sqrt{2}} \begin{pmatrix} 1 & 1 \\ 1 & -1 \end{pmatrix}$$

We then have (theorem on diagonalisation of matrices):

$$V^{-1}AV = \Lambda = \begin{pmatrix} \lambda_1 & 0 \\ 0 & \lambda_2 \end{pmatrix} = \begin{pmatrix} 0 & 0 \\ 0 & -2 \end{pmatrix} \tag{35}$$

Now, recall the initial model was $\dot{\boldsymbol{x}} = kA\boldsymbol{x}$. Multiplying this latter equation by V^{-1} on the left gives:

$$V^{-1}\frac{d}{dt}\boldsymbol{x} = \frac{d}{dt}\underbrace{(V^{-1}\boldsymbol{x})}_{=\boldsymbol{X}} = k\underbrace{V^{-1}AV}_{=\Lambda}\underbrace{(V^{-1}\boldsymbol{x})}_{=\boldsymbol{X}} = k\Lambda\boldsymbol{X}$$

$$\Rightarrow \boxed{\frac{d\boldsymbol{X}}{dt} = k\Lambda\boldsymbol{X},} \quad \boldsymbol{X} = V^{-1}\boldsymbol{x} \tag{36}$$

Eq. (36) is now a system of decoupled linear ODEs (each of order 1) for which we can easily compute the solutions in terms of X_1 and X_2.

$$\begin{pmatrix} \dot{X}_1 \\ \dot{X}_2 \end{pmatrix} = k\begin{pmatrix} 0 & 0 \\ 0 & -2 \end{pmatrix}\begin{pmatrix} X_1 \\ X_2 \end{pmatrix} = k\begin{pmatrix} \lambda_1 & 0 \\ 0 & \lambda_2 \end{pmatrix}\begin{pmatrix} X_1 \\ X_2 \end{pmatrix} \tag{37}$$

$$\Leftrightarrow \begin{cases} \dot{X}_1 = k\lambda_1 X_1 \\ \dot{X}_2 = k\lambda_2 X_2 \end{cases}$$

$$\Rightarrow \begin{cases} X_1(t) = X_1(0)e^{k\lambda_1 t} \\ X_2(t) = X_2(0)e^{k\lambda_2 t} \end{cases}$$

The last step is to transform back into the original coordinates using $\boldsymbol{X} = V^{-1}\boldsymbol{x}$ which implies $\boldsymbol{x} = V\boldsymbol{X}$. Using $\boldsymbol{x} = V\boldsymbol{X}$, i.e., $\boldsymbol{x} = \begin{pmatrix} | & | \\ \boldsymbol{v}_1 & \boldsymbol{v}_2 \\ | & | \end{pmatrix}\begin{pmatrix} X_1 \\ X_2 \end{pmatrix} = \boldsymbol{v}_1X_1 + \boldsymbol{v}_2X_2$, we obtain

$$\boldsymbol{x}(t) = \boldsymbol{v}_1X_1(t) + \boldsymbol{v}_2X_2(t) \Leftrightarrow \boxed{\boldsymbol{x}(t) = \boldsymbol{v}_1X_1(0)e^{k\lambda_1 t} + \boldsymbol{v}_2X_2(0)e^{k\lambda_2 t}}$$

with $\lambda_1 = 0$ and $\lambda_2 = -2$ being the eigenvalues of A and $\boldsymbol{v}_1 = \frac{1}{\sqrt{2}}\begin{pmatrix} 1 \\ 1 \end{pmatrix}$ and $\boldsymbol{v}_2 = \frac{1}{\sqrt{2}}\begin{pmatrix} 1 \\ -1 \end{pmatrix}$ being the corresponding eigenvectors.

For a linear, 2^{nd} order ODE of the form $\dot{\boldsymbol{x}} = A\boldsymbol{x}$, $\boldsymbol{x} \in \mathbb{R}^2$, where A is diagonalisable, the solution is a linear combination of exponentials of the form $e^{\lambda_i t}$ where λ_i are the eigenvalues of A, i.e., the general solution is of the form $\boxed{\boldsymbol{x}(t) = \boldsymbol{v}_1X_1(0)e^{\lambda_1 t} + \boldsymbol{v}_2X_2(0)e^{\lambda_2 t}} = \boldsymbol{c}_1e^{\lambda_1 t} + \boldsymbol{c}_2e^{\lambda_2 t}$.

Remark 38. *Note that in the mass-spring-damper case (32)-(33), X_1 and X_2 have a physical meaning*[14]*: Indeed, from $\boldsymbol{X} = V^{-1}\boldsymbol{x}$, with, in this particular case, $V = V^{-1}$ (check by verifying $VV = I$), we have:*

$$X_1(t) = \frac{1}{\sqrt{2}}(x_1 + x_2)$$

which is proportional to the total displacement of the centre of mass with respect to the resting position of the spring, and

$$X_2(t) = \frac{1}{\sqrt{2}}(x_1 - x_2)$$

[14]This is typically not true, i.e., $\boldsymbol{X}$ typically does not have a physical meaning.

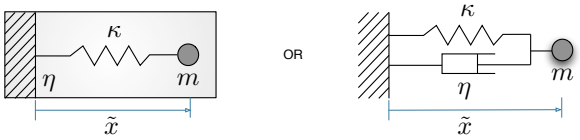
which is proportional to the stretch or compression of the spring with respect to the resting position of the spring.

Remark 39. *From (32)-(33) and Remark 38 we have:*

1. $\lambda_1 = 0 \Rightarrow \dot{X}_1 = 0 = \frac{1}{\sqrt{2}}(\dot{x}_1 + \dot{x}_2) \Rightarrow$ *the centre of mass cannot move (because no net force is applied).*
2. $\lambda_2 = -2 \Rightarrow \dot{X}_2 = -2kX_2 = \frac{1}{\sqrt{2}}(\dot{x}_1 - \dot{x}_2)$ *with* $k > 0 \Rightarrow \dot{x}_1 - \dot{x}_2 < 0$ *if* $X_2(t) > 0$*, i.e., the spring will be compressed if the distance between the two masses is larger than the resting distance, or* $\dot{x}_1 - \dot{x}_2 > 0$ *if* $X_2(t) < 0$*, i.e., the spring will be stretched if the distance between the masses is smaller than the resting distance.*

4.2.1 The mass-spring-damper system

Let us consider the mass-spring-damper system:



for which the equation of motion is

$$m\ddot{x} + \eta\dot{x} + \kappa x = 0 \tag{38}$$

To solve (38), we put the model in the form $\dot{\boldsymbol{x}} = A\boldsymbol{x}$ and diagonalise A:

$$\begin{cases} \dot{x} = y \\ \ddot{x} = \dot{y} = -\frac{\kappa}{m}x - \frac{\eta}{m}y \end{cases} \Rightarrow \begin{pmatrix} \dot{x} \\ \dot{y} \end{pmatrix} = \underbrace{\begin{pmatrix} 0 & 1 \\ -\frac{\kappa}{m} & -\frac{\eta}{m} \end{pmatrix}}_{=A} \begin{pmatrix} x \\ y \end{pmatrix}$$

The eigenvalues of A are $\lambda_\pm = \frac{-\frac{\eta}{m} \pm \sqrt{\frac{\eta^2}{m^2} - 4\frac{\kappa}{m}}}{2}$. The general solution is thus $\boxed{\boldsymbol{x}(t) = \boldsymbol{c}_+ e^{\lambda_+ t} + \boldsymbol{c}_- e^{\lambda_- t}}$ where $\boldsymbol{c}_\pm$ are proportional to the eigenvectors associated with $\lambda_\pm$.

Remark 40. *Note that* **if** $\lambda_\pm$ *are complex conjugate numbers, i.e.,* $\boxed{\lambda_\pm = \alpha \pm i\beta}$ **then** *the general solution writes* $\boxed{\boldsymbol{x}(t) = \boldsymbol{c}_+ e^{\alpha t} e^{i\beta t} + \boldsymbol{c}_- e^{\alpha t} e^{-i\beta t} = e^{\alpha t}\left(\boldsymbol{c}_+ e^{i\beta t} + \boldsymbol{c}_- e^{-i\beta t}\right)}$*. The real exponential* $e^{\alpha t}$ *can grow (if* $\alpha > 0$*) or decay (if* $\alpha < 0$*). On the contrary, the complex exponentials* $e^{\pm i\beta t}$ *correspond to pure oscillations at frequency* $\frac{\beta}{2\pi}$ *(or period* $\frac{2\pi}{\beta}$*) since* $e^{\pm i\beta t} = \cos(\beta t) \pm i\sin(\beta t)$ *(this relation is called "Euler's formula").*

Remark 41. *Note that we could here have also solved (38) using the Ansatz* $x(t) = Ce^{\alpha t}$*. Indeed, using the Ansatz* $x(t) = Ce^{\alpha t}$ *gives the equation* $m\alpha^2 + \eta\alpha + \kappa = 0$ *which is the same equation as the one obtained for the calculation of the eigenvalues of A, i.e.,* $\det(A - \lambda I) = 0$*.*

4.3 General solution for linear ODEs models of any order

What we have just seen is true in general for linear ODEs models of any order, i.e., if $\dot{\boldsymbol{x}} = A\boldsymbol{x}$ with $\boldsymbol{x} \in \mathbb{R}^n$ and A is diagonalisable[15], then the solution is of the form $\boxed{\boldsymbol{x}(t) = \sum_{i=1}^{n} \boldsymbol{v}_i X_i(0) e^{\lambda_i t} = \sum_{i=1}^{n} \boldsymbol{c}_i e^{\lambda_i t}}$ where $\lambda_i \in \mathbb{C}$ are the *eigenvalues of* A and $\boldsymbol{v}_i \in \mathbb{C}^n$ are the *corresponding eigenvectors of* A.

[15] A real matrix A is always diagonalisable if all its eigenvalues are distinct. If this is not the case, the analytical solution can still be always written but it takes a different form. See Strogatz book for more details.

5 Nonlinear ODE models of order 2

5.1 Stability analysis of nonlinear ODE models of order 2

Consider a nonlinear ODE model of order 2:

$$\boxed{\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x}), \quad \boldsymbol{x} \in \mathbb{R}^2 \Leftrightarrow \begin{pmatrix} \dot{x}_1 \\ \dot{x}_2 \end{pmatrix} = \begin{pmatrix} f_1(x_1, x_2) \\ f_2(x_1, x_2) \end{pmatrix}, \quad f(\cdot) : \mathbb{R}^2 \to \mathbb{R}^2, \text{ "smooth" function}}$$

1. Global stability analysis (difficult for models of order ≥ 2)

   For models of order 1, the global stability analysis is quite easy since the motion of the system is on the real line and thus we can predict the long-term behaviour of the system from the fixed points, the flows, and the initial condition. For models of order 2 (or higher), this is typically much more difficult to do since the motion of the system is on a plane (or in $\mathbb{R}^n$) and thus it may not be obvious where the trajectories will go from a given initial condition.

2. Local stability analysis (possible for all orders)

   - Find the fixed points: $\{\boldsymbol{x}^* : \boldsymbol{f}(\boldsymbol{x}^*) = \boldsymbol{0}\}$.
   - Linearise the dynamics around *each* fixed point.
   - Study the stability of the corresponding linear systems.
   - Draw the local flows around each fixed point:
   - Try to link together the local stability information around each fixed point to establish a global picture of the attractors in the state space. Two important properties of autonomous, time-invariant, ODE models are important here:
     - *Nullclines*: the curves in the phase plane corresponding to *individual* first derivatives being zero ($\dot{x}_1 = 0$ or $\dot{x}_2 = 0$), i.e., the curves $f_1(x_1, x_2) = 0$ and $f_2(x_1, x_2) = 0$. Nullclines are very useful for models of order 2 to graphically find the location of fixed points and to sketch the vector field (also called flow) on a phase plane. Indeed, the fixed points are located at the intersection of the nullcllines. Furthermore, by definition of a nullcline, the vector field on a nullcline always has one of its component equal to zero. For systems of order 2, this means that on a nullcline, the vector field or flow can only be either horizontal or vertical (depending on which component of the vector field is zero on the considered nullcline). Have a look at Appendix A for more information. Note that nullclines are less useful for graphically finding fixed points or sketching the vector field for models of order 3 or higher.
     - *Trajectories in the phase plane (phase space for models of order 3 or higher) cannot cross, except at the fixed points.* The non-crossing property is a consequence of the theorem of existence and uniqueness of solutions of time-invariant ODEs, i.e., ODEs where time does not appear explicitly in the dynamics: $\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x})$ (time-invariant ODE) and not $\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x}, t)$ (time-varying ODE). It basically corresponds to the idea that, for these types of systems, you cannot have different pasts that lead to the same future without this future being a fixed point, or, said differently, that you cannot have different futures (or trajectories) starting from the same state (or point in the state space). This non-crossing property makes it sometimes easier to build a global picture of the attractors and their corresponding basins of attraction for models of order 2. This property is however less useful for models of order 3 or higher.

   Have a look at the phase planes for the pendulum depicted on page 42 to get an idea of a global picture of a phase plane that can be obtained by piecing together the local phase plane information gathered around each fixed point.

5.2 Linearisation of nonlinear ODE models of order 2

- Find the fixed points, i.e., the points $\boldsymbol{x}^*$ s.t. $\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x}^*) = \boldsymbol{0} \Leftrightarrow \begin{cases} f_1(x_1^*, x_2^*) = 0 \\ f_2(x_1^*, x_2^*) = 0 \end{cases}$

  Remark 42. *The MATLAB functions* `fsolve` *or* `solve` *can be used to find the zeros of a system of algebraic equations* $\begin{cases} f_1(x_1^*, \ldots, x_n^*) = 0 \\ \vdots \\ f_n(x_1^*, \ldots, x_n^*) = 0 \end{cases}$. *Remember that* `fsolve` *only gives you one solution which is found based on a given initial guess. Thus, to find all the zeros with* `fsolve` *you need to try several initial guesses.*

- Linearise the dynamics around *each* fixed point (using Taylor):
  Consider $\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x})$ with $\boldsymbol{x} = \boldsymbol{x}^* + \boldsymbol{\xi}$ where $\boldsymbol{\xi} \in \mathbb{R}^2$ s.t. $\|\boldsymbol{\xi}\| \ll 1$:

$$\dot{\boldsymbol{\xi}} = \dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x}^* + \boldsymbol{\xi}) = \underbrace{\boldsymbol{f}(\boldsymbol{x}^*)}_{=\boldsymbol{0}} + \underbrace{\begin{pmatrix} \frac{\partial f_1(x_1,x_2)}{\partial x_1}\Big|_{\boldsymbol{x}=\boldsymbol{x}^*} & \frac{\partial f_1(x_1,x_2)}{\partial x_2}\Big|_{\boldsymbol{x}=\boldsymbol{x}^*} \\ \frac{\partial f_2(x_1,x_2)}{\partial x_1}\Big|_{\boldsymbol{x}=\boldsymbol{x}^*} & \frac{\partial f_2(x_1,x_2)}{\partial x_2}\Big|_{\boldsymbol{x}=\boldsymbol{x}^*} \end{pmatrix}}_{\triangleq J(\boldsymbol{x}^*)} \boldsymbol{\xi} + \underbrace{\cancel{\mathcal{O}\left(\|\boldsymbol{\xi}\|^2\right)}}_{H.O.T.}$$

  We thus obtain:

$$\dot{\boldsymbol{\xi}} = J(\boldsymbol{x}^*)\,\boldsymbol{\xi} \tag{39}$$

  where $J(\boldsymbol{x}^*)$ is the *Jacobian* matrix evaluated at the fixed point $\boldsymbol{x}^*$, i.e., a constant matrix whose (i,j) element is $J_{ij}(\boldsymbol{x}^*) = \frac{\partial f_i(\boldsymbol{x})}{\partial x_j}\Big|_{\boldsymbol{x}=\boldsymbol{x}^*}$.

- Study the stability of (39) at each fixed point $\boldsymbol{x}^*$
  $\Rightarrow$ Diagonalise $J(\boldsymbol{x}^*)$ (i.e., $\tilde{\boldsymbol{\xi}} = V^{-1}\boldsymbol{\xi}$)

Remark 43. *An important result (the Hartman-Grobman theorem) justifies the study of linearisations. It states that solutions of the nonlinear system* $\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x})$ *in the vicinity of the steady state* $\boldsymbol{x}^*$ *look "qualitatively" just like solutions of the linearised equation* $\dot{\boldsymbol{\xi}} = J(\boldsymbol{x}^*)\,\boldsymbol{\xi}$ *do in the vicinity of the point* $\boldsymbol{\xi} = \boldsymbol{0}$.[16]

5.3 Diagonalisation of the Jacobian for ODE models of order 2

Consider

$$J(\boldsymbol{x}^*) = \begin{pmatrix} a & b \\ c & d \end{pmatrix}$$

The eigenvalues of $J(\boldsymbol{x}^*)$ are given by solving

$$\det\left(J(\boldsymbol{x}^*) - \lambda I\right) = 0$$

which gives the algebraic equation

$$\lambda^2 - \underbrace{(a+d)}_{=\tau}\lambda + \underbrace{(ad-bc)}_{=\Delta} = 0$$

where

- τ is the *trace* of $J(\boldsymbol{x}^*)$, i.e., the sum of the diagonal elements of $J(\boldsymbol{x}^*)$ $(= \lambda_+ + \lambda_-)$
- Δ is the *determinant* of $J(\boldsymbol{x}^*)$ $(= \lambda_+\lambda_-)$

[16] The theorem assumes that *none* of the eigenvalues of $J(\boldsymbol{x}^*)$ has a zero real part (i.e., as we will see later, that the linearised fixed point $\boldsymbol{\xi} = \boldsymbol{0}$ is *not* a center).

Eigenvalues:

$$\lambda^2 - \tau\lambda + \Delta = 0$$
$$\Rightarrow \boxed{\lambda_\pm = \frac{\tau \pm \sqrt{\tau^2 - 4\Delta}}{2}}$$

Therefore, diagonalising $J(\boldsymbol{x}^*)$, we can see that the general solution is of the form:

$$\boxed{\boldsymbol{\xi}(t) = \boldsymbol{c}_+ e^{\lambda_+ t} + \boldsymbol{c}_- e^{\lambda_- t}}, \quad \boldsymbol{\xi}(t) \in \mathbb{R}^2, \boldsymbol{c}_+, \boldsymbol{c}_- \in \mathbb{C}^2, \lambda_+, \lambda_- \in \mathbb{C} \tag{40}$$

where $\lambda_\pm$ are the eigenvalues of $J(\boldsymbol{x}^*)$ and $\boldsymbol{c}_\pm$ are proportional to the corresponding eigenvectors.

Remember that to diagonalise $J(\boldsymbol{x}^*)$ appearing in $\dot{\boldsymbol{\xi}} = J(\boldsymbol{x}^*)\boldsymbol{\xi}$ you need to proceed as follows:

1. Compute the eigenvalues and corresponding eigenvectors of $J(\boldsymbol{x}^*)$
2. Construct a matrix V having the eigenvectors of $J(\boldsymbol{x}^*)$ as columns
3. Perform the change of variables $\tilde{\boldsymbol{\xi}} = V^{-1}\boldsymbol{\xi}$

This then leads to:

$$\dot{\tilde{\boldsymbol{\xi}}} = V^{-1}\dot{\boldsymbol{\xi}} = \left(V^{-1}J(\boldsymbol{x}^*)V\right)\tilde{\boldsymbol{\xi}} = \Lambda\tilde{\boldsymbol{\xi}}$$

where Λ is a diagonal matrix with the eigenvalues of $J(\boldsymbol{x}^*)$ on the diagonal (in the same order as the corresponding eigenvectors in the columns of V). The diagonalisation allows to decouple the equations, and therefore to reduce the problem to finding the solution of 1^{st} order linear ODEs (which we know how to do).

5.4 Local stability analysis for ODE models of order 2

The general solution for a linear ODE of order 2 is:

$$\boxed{\boldsymbol{\xi}(t) = \boldsymbol{c}_+ e^{\lambda_+ t} + \boldsymbol{c}_- e^{\lambda_- t}, \quad \lambda_\pm = \frac{\tau \pm \sqrt{\tau^2 - 4\Delta}}{2}, \quad \boldsymbol{c}_\pm \propto \text{eigenvec. assoc. with } \lambda_\pm} \tag{41}$$

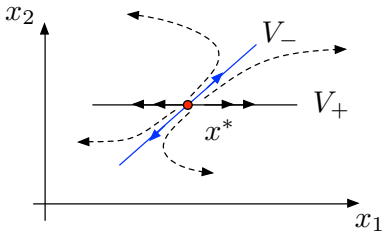
The local behaviours are dictated by the signs of $\tau(=\lambda_+ + \lambda_-)$, $\Delta(=\lambda_+\lambda_-)$, and $\tau^2 - 4\Delta\left(=(\lambda_+ - \lambda_-)^2\right)$.

1. $\Delta > 0$: $\sqrt{\tau^2 - 4\Delta} < |\tau|$
   - $\tau > 0$:
     (a) $\tau^2 - 4\Delta > 0$: $\lambda_+ > \lambda_- > 0$ (A)
     (b) $\tau^2 - 4\Delta < 0$: $\lambda_\pm$ complex conjugate with pos. real part (B)
   - $\tau < 0$:
     (a) $\tau^2 - 4\Delta > 0$: $\lambda_- < \lambda_+ < 0$ (C)
     (b) $\tau^2 - 4\Delta < 0$: $\lambda_\pm$ complex conjugate with neg. real part (D)
   - $\tau = 0$: $\lambda_\pm$ purely imaginary (E)
2. $\Delta < 0$: $\lambda_- < 0 < \lambda_+$ (F)

In the diagonalised coordinates, i.e., for $\tilde{\boldsymbol{\xi}} = V^{-1}\boldsymbol{\xi}$:

$$\boxed{\tilde{\xi}_1(t) = \tilde{\xi}_1(0)e^{\lambda_+ t}, \quad \tilde{\xi}_2 = \tilde{\xi}_2(0)e^{\lambda_- t}, \quad \lambda_\pm = \frac{\tau \pm \sqrt{\tau^2 - 4\Delta}}{2}}$$

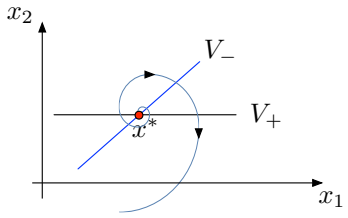
(A) $\lambda_+ > \lambda_- > 0$: Exponential growth in both directions: **Repelling or unstable node**



where V_+ and V_- represent the eigenvectors of $J(\boldsymbol{x}^*)$.

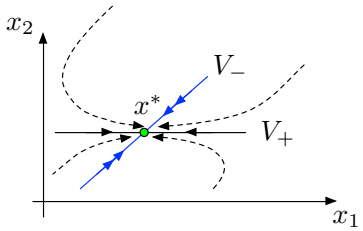
Remark 44. *To understand this phase plane picture remember that* $\boldsymbol{\xi} = V\tilde{\boldsymbol{\xi}}$ *where* $V = \begin{pmatrix} | & | \\ V_+ & V_- \\ | & | \end{pmatrix}$ *with* V_+ *and* V_- *being the columns of* V *and representing the eigenvectors associated with* λ_+ *and* λ_- *respectively. The relation* $\boldsymbol{\xi} = V\tilde{\boldsymbol{\xi}}$ *expresses* $\boldsymbol{\xi}$ *in the basis formed by the eigenvectors* V_+ *and* V_- *since* $\boldsymbol{\xi} = \tilde{\xi}_1 V_+ + \tilde{\xi}_2 V_- (= \boldsymbol{c}_+ e^{\lambda_+ t} + \boldsymbol{c}_- e^{\lambda_- t}$ *with* $\boldsymbol{c}_+ = \tilde{\xi}_1(0)V_+$ *and* $\boldsymbol{c}_- = \tilde{\xi}_2(0)V_-)$. *Note that the divergence is faster in the eigenvector direction associated with the largest eigenvalue.*

(B) $\lambda_\pm$ complex conjugate with pos. real part: $\tilde{\xi}_{1,2}(t) = \tilde{\xi}_{1,2}(0)e^{\frac{\tau}{2}t}e^{\pm i\omega t}$ with $\tau > 0$, $\omega = \frac{\sqrt{|\tau^2 - 4\Delta|}}{2}$: **Unstable spiral**

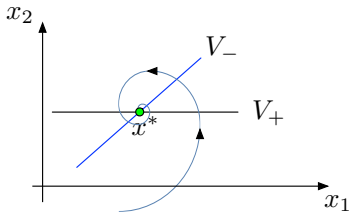


Remark 45. *Non-real complex conjugate eigenvalues* $\lambda = a \pm ib$ *are associated to oscillations. They correspond to terms in solutions that involve complex exponentials* $e^{\lambda t}$. *Since one has the general formula* $e^{\lambda t} = e^{at \pm ibt} = e^{at}(\cos(bt) \pm i\sin(bt))$, *solutions, when re-written in real-only form, contain terms of the form* $e^{at}\cos(bt)$ *and* $e^{at}\sin(bt)$, *and therefore diverge to* ∞ *(with growing oscillations of "period"* $\frac{2\pi}{b}$*) provided that* $a > 0$, *that is to say, that the real part of* λ *is positive. Another way to see this is to notice that asking that* $e^{\lambda t} \to \infty$ *is the same as requiring that the magnitude* $\left|e^{\lambda t}\right| \to \infty$. *Furthermore, since* $\left|e^{\lambda t}\right| = e^{at}\sqrt{(\cos(bt))^2 + (\sin(bt))^2} = e^{at}$, *we see that* $a > 0$ *is the condition needed in order to ensure that* $\left|e^{\lambda t}\right| \to \infty$.

(C) $\lambda_- < \lambda_+ < 0$: Exponential decay in both directions: **Attracting or stable node**

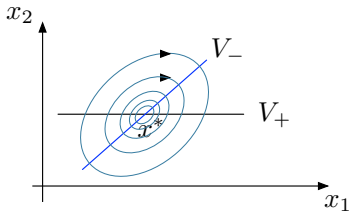


(D) $\lambda_\pm$ complex conjugate with neg. real part: $\tilde{\xi}_{1,2}(t) = \tilde{\xi}_{1,2}(0)e^{\frac{\tau}{2}t}e^{\pm i\omega t}$ with $\tau < 0$, $\omega = \frac{\sqrt{|\tau^2 - 4\Delta|}}{2}$: **Stable spiral**



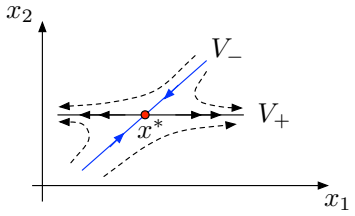
Remark 46. *As in Remark 45, since the solutions, when re-written in real-only form, contain terms of the form* $e^{at}\cos(bt)$ *and* $e^{at}\sin(bt)$*, they converge to zero (with decaying oscillations of "period"* $\frac{2\pi}{b}$*) provided that* $a < 0$*, that is to say, that the real part of* λ *is negative. Another way to see this is to notice that asking that* $e^{\lambda t} \to 0$ *is the same as requiring that the magnitude* $\left|e^{\lambda t}\right| \to 0$*. Since* $\left|e^{\lambda t}\right| = e^{at}\sqrt{(\cos(bt))^2 + (\sin(bt))^2} = e^{at}$*, we see that* $a < 0$ *is the condition needed in order to ensure that* $|e^{\lambda t}| \to 0$*.*

(E) $\lambda_\pm$ purely imaginary: $\tilde{\xi}_{1,2}(t) = \tilde{\xi}_{1,2}(0)e^{\pm i\omega t}$ with $\omega = \sqrt{\Delta}$: periodic oscillations: **Center**



Remark 47. *In this case, the system is said to be* marginally stable*. This type of stability is very fragile to perturbations since it only occurs when* $\tau = 0$*, and the slightest perturbation will typically either render* x^* *stable and attractive, or unstable.*

(F) $\lambda_- < 0 < \lambda_+$: exp. decay in one dir. and exp. growth in the other: **Saddle point**

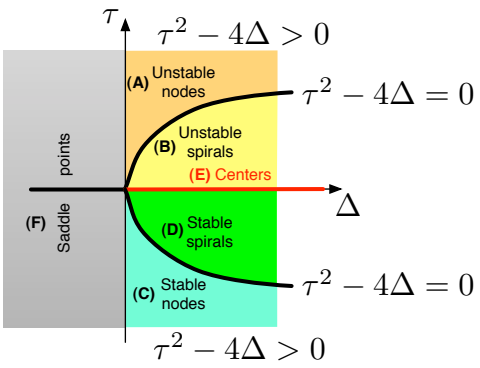


Under the assumption that $|\lambda_+| > |\lambda_-|$, the divergence along V_+ is faster than the convergence along V_-.

Remark 48. *Summarising what we have seen, we now understand how the local stability analysis of a nonlinear ODE of the form* $\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x})$ *is performed. This is done by linearising this nonlinear ODE around each of its fixed points* $\boldsymbol{x}^*$*, thereby obtaining linearised ODEs of the form* $\dot{\boldsymbol{\xi}} = J(\boldsymbol{x}^*)\boldsymbol{\xi}$ *where* $\boldsymbol{\xi} = \boldsymbol{x} - \boldsymbol{x}^*$ *with* $\|\boldsymbol{\xi}\|$ *"small". As we have seen, the local behaviour around a fixed point* $\boldsymbol{x}^*$ *is dependent on the eigenvalues and eigenvectors of* $J(\boldsymbol{x}^*)$*. In particular, diagonalising*[17] $J(\boldsymbol{x}^*)$ *through a change of coordinates* $\tilde{\boldsymbol{\xi}} = V^{-1}\boldsymbol{\xi}$*, we obtain a decoupled system of first order ODEs of the form* $\dot{\tilde{\xi}}_{1,2} = \lambda_\pm \tilde{\xi}_{1,2}$ *whose solution is* $\tilde{\xi}_{1,2}(t) = \tilde{\xi}_{1,2}(0)e^{\lambda_\pm t}$*. Now, in general, the eigenvalues* $\lambda_\pm$ *will be complex numbers:* $\lambda_\pm = a \pm ib$ *(with* $i^2 = -1$ *and* $a, b \in \mathbb{R}$*) and thus the general solution can be written as:* $\tilde{\xi}_{1,2}(t) = \tilde{\xi}_{1,2}(0)e^{at}e^{\pm ibt}$*. The complex exponential* $e^{\pm ibt} = \cos(bt) \pm i\sin(bt)$ *accounts for "oscillations" of the solution around* $\boldsymbol{x}^*$ *whereas the real exponential* e^{at} *accounts for exponential increases* $(a > 0)$ *or decreases* $(a < 0)$ *in the amplitude of the solution.*

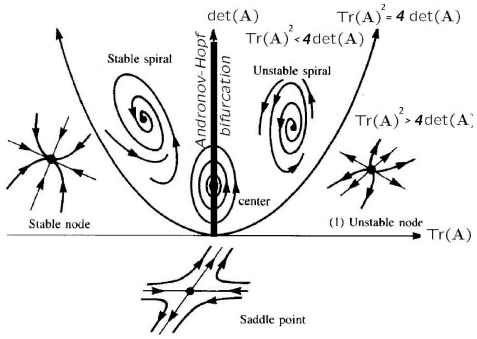
[17] Note that there are conditions for this to be possible.

5.4.1 Summary of the possible local behaviours for models of order 2



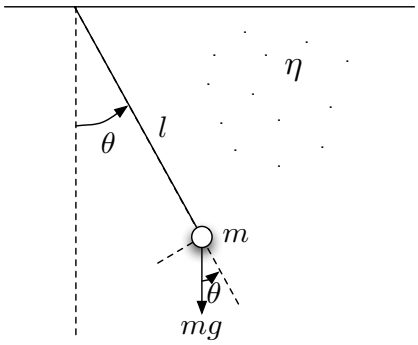
As a general property for models of any order, the considered fixed point is stable if *all* the eigenvalues of the Jacobian evaluated at this fixed point have negative real parts. On the contrary, if one of the eigenvalues has a positive real part then the corresponding fixed point will be unstable.

The phase plane behaviours, with the corresponding local flows around the considered fixed point, are given in the following picture:



Once you have a local picture around all the fixed points, the global behaviour of a system of order 2 can be obtained by remembering that for models of the form $\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x})$ trajectories do not cross. This non-crossing property makes it sometimes easier to build a global picture of the attractors and their corresponding basins of attraction for models of order 2.

For example, the global phase portrait obtained by piecing together the local flows around the fixed points of a simple ODE model of a pendulum is given in the following figure:

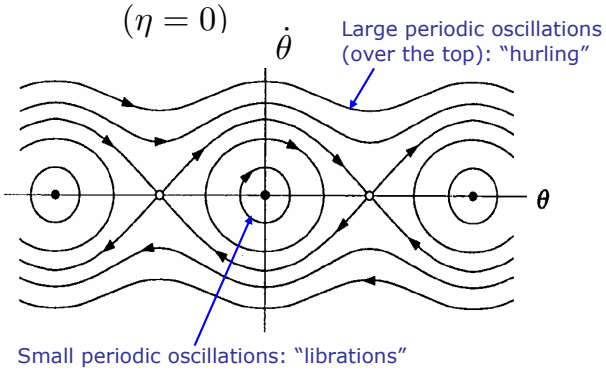


$$ml\ddot{\theta} = -\eta' l\dot{\theta} - mg\sin(\theta)$$

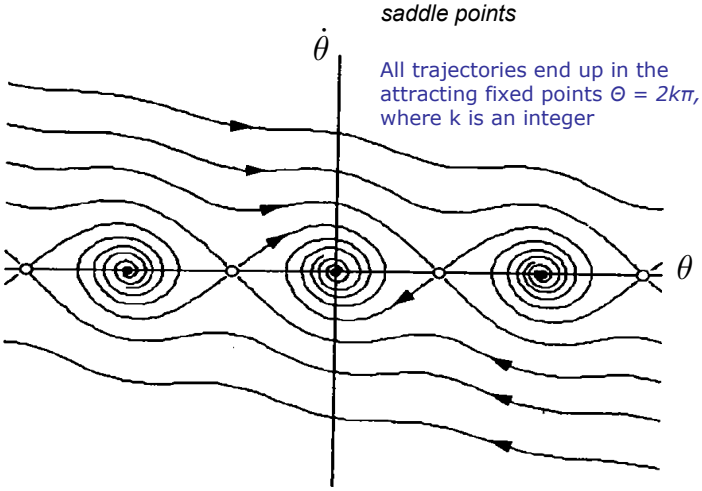
$$\Leftrightarrow \boxed{\ddot{\theta} + \eta\dot{\theta} + k\sin(\theta) = 0}$$

with $\eta = \frac{\eta'}{l}$ and $k = \frac{g}{l}$

1. Undamped pendulum: *Centers and saddle points*



2. Underdamped pendulum: *Attracting spirals and saddle points*



As an exercise you can try to perform a global stability analysis (by piecing together info obtained by local stability analysis around the various fixed points) of the pendulum ODE model:

$$\ddot{\theta} + \eta\dot{\theta} + k\sin(\theta) = 0$$

and see if you obtain the same global phase portrait as in the above picture. (See also the book of Strogatz for more information.)

5.5 Periodic behaviour and limit cycles

5.5.1 Linear oscillations and their limitations

Periodic behaviors (i.e., periodic oscillations) are very important in biology, appearing in diverse areas such as neural signaling, circadian rythms, heart beats, etc. In the case of linear ODEs models of order 2, periodic oscillations can only be obtained if the system has a fixed point which is a center. The typical set of linear ODEs in that situation is of the form:

$$\begin{cases} \dot{x} = y \\ \dot{y} = -x \end{cases} \tag{42}$$

whose trajectories are circles. More generally, linear ODE models of order 2 with eigenvalues that are purely imaginary[18] lead to ellipsoidal trajectories in the phase plane.

An example of system leading to this type of equations is the mass-spring system with no damping.

The mass-spring-damper model with $\eta = 0$

Consider the model described in (38). If there is no damping, i.e., if $\eta = 0$ (e.g., the fluid in which the mass-spring system is immersed is non-viscous) then (38) becomes:

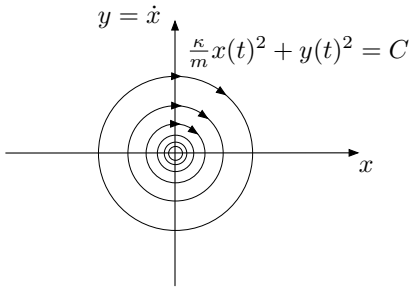
$$m\ddot{x} + \kappa x = 0 \Rightarrow \begin{cases} \dot{x} = y \\ \dot{y} = -\frac{\kappa}{m}x \end{cases} \tag{43}$$

For (43), an analytical solution can be obtained as follows:

$$\begin{aligned} \frac{dy}{dx} &= -\frac{\kappa}{m}\frac{x}{y} \\ \int y\,dy &= -\frac{\kappa}{m}\int x\,dx \\ \frac{y^2}{2} + C_1 &= -\frac{\kappa}{m}\frac{x^2}{2} + C_2 \\ \Rightarrow \frac{\kappa}{m}x(t)^2 + y(t)^2 &= C = 2(C_1 + C_2), \quad \forall t \end{aligned} \tag{44}$$

The equation $\frac{\kappa}{m}x(t)^2 + y(t)^2 = C$ corresponds to the definition of an ellipse in the $x - y$ plane (the phase plane).

Consider $m = 1$ and $\kappa = 1$. The phase plane is then:



$\frac{\kappa}{m}x(t)^2 + y(t)^2 = C \Leftrightarrow \underbrace{\frac{\kappa}{2}x(t)^2}_{\text{Potential Energy}} + \underbrace{\frac{m}{2}\dot{x}(t)^2}_{\text{Kinetic Energy}} = \tilde{C}$ which makes sense since with no damping ($\eta = 0$), the total energy of the system is conserved. We say that we have a *conservative* system.

Remark 49. *Conservative systems cannot have attractors. Also, ODE models of order* 1*, even nonlinear, cannot have periodic oscillations. Do you see why? (Think about the non crossing property of solutions.)*

Remark 50. *In general, closed trajectories in the phase plane correspond to "purely periodic" behaviours (periodic time trajectories).*

Remark 51. *C is defined by the initial condition since $\frac{\kappa}{m}x(0)^2 + y(0)^2 = C$.*

Remark 52. *How do we know in which direction we rotate on the phase plane? Well, this can be easily seen from the ODEs evaluated on the x and y axes:*

[18] Such linear systems are said to be *marginally stable.*

- *On the x-axis (i.e., when $y = 0$), we have $y = 0 \Rightarrow \dot{x} = 0$, $\dot{y} = -\frac{\kappa}{m}x$, which means that there is no change in x, and the change in y is upward for $x < 0$ and downward for $x > 0$.*
- *A similar argument can be used on the y-axis.*

Linear (or "harmonic") oscillations have 2 serious limitations:

1. They are "fragile" or non-robust to small perturbations in the model
2. The oscillation characteristics (amplitude and phase) depend on the initial condition

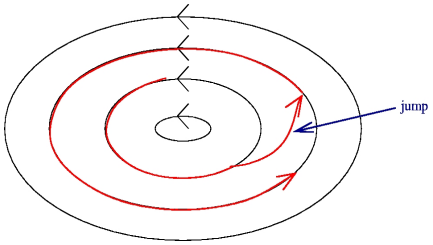
A first serious limitation of such linear oscillators is that they are not robust to small perturbations in the model. Suppose that there is a small perturbation in the equations given in (42):

$$\begin{cases} \dot{x} = y \\ \dot{y} = -x + \epsilon y \end{cases}$$

where $\epsilon \neq 0$ is small, i.e., $|\epsilon| \ll 1$. The trajectories are not periodic anymore since the corresponding fixed point is now a stable ($\epsilon < 0$) or unstable ($\epsilon > 0$) spiral. You can check this by computing the eigenvalues and looking at the sign of their real part (the eigenvalues are $\lambda_{\pm} = \frac{\epsilon \pm \sqrt{\epsilon^2 - 4}}{2}$ with ϵ "small").

When dealing with electrical or mechanical systems, it is often possible to construct things with precise components and low error tolerance. In biology, in contrast, things are too "messy" and oscillators, if they are to be reliable, must be more "robust" than simple harmonic oscillators.

Another disadvantage of simple linear oscillations is that if, for some reason, the state "jumps" to another position then the system will simply start oscillating along a different orbit and never come back to the original trajectory[19]:

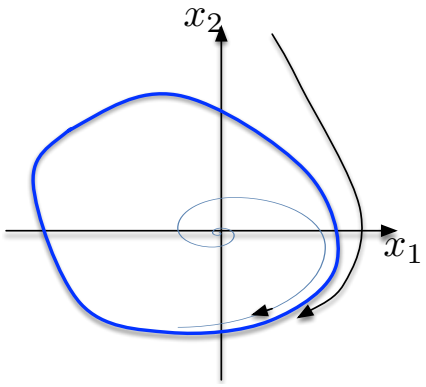


To put it in different terms, the particular oscillation depends on the initial conditions. Biological systems, in contrast, tend to reset themselves (e.g., your internal circadian clock adjusting after a jetlag).

5.5.2 Limit cycles

A stable *limit cycle* is a periodic trajectory which attracts other solutions to it (at least those starting "close to" the limit cycle).

[19] To illustrate this consider again the mass-spring-damper system with zero damping (see (43)). In that case, we saw that the trajectories are constrained by: $\frac{\kappa}{m}x(t)^2 + y(t)^2 = C$ with $C = \frac{\kappa}{m}x(0)^2 + y(0)^2$. This clearly shows that if we change the initial condition (i.e. if we force the trajectory to "jump" to a different point in the phase plane), then a different closed trajectory will ensue.



Thus, a member of a family of "parallel" periodic solutions (as for linear centers) is not called a limit cycle, because other close-by trajectories remain at a fixed distance away, and do not converge to it. Stable, attractive limit cyles are "robust" in ways that linear periodic solutions are not:

- If a (small) perturbation moves the state to a different initial state away from the limit cycle, the system will return to the limit cycle by itself.
- If the dynamics changes a little (e.g., small perturbation), a limit cycle will still exist, close to the original one.

Remark 53. *Limit cycles can only occur in nonlinear ODEs. They cannot happen in linear ODEs.*

Remark 54. *In particular, if, no matter where we start, we end up on the limit cycle, then the limit cycle is a* global attractor *of our system. On the contrary, if the limit cycle is only attracting solutions starting in a particular region of the phase plane around it, then it is a* local attractor *and the corresponding region is called the* basin of attraction *of the attractor (i.e., of the limit cycle).*

5.5.3 Limit cycles: an example

In order to understand the definition, and to have an example that we can use for various purposes later, we will consider the following system:

$$\dot{x} = -\omega y + x\left(\mu - x^2 - y^2\right) \tag{45}$$
$$\dot{y} = \omega x + y\left(\mu - x^2 - y^2\right) \tag{46}$$

where μ *is a parameter* while $\omega \neq 0$ is a constant.

- Fixed points:

$$\begin{cases} -\omega y^* + x^*\left(\mu - x^{*2} - y^{*2}\right) = 0 \\ \omega x^* + y^*\left(\mu - x^{*2} - y^{*2}\right) = 0 \end{cases}$$

$$\Rightarrow -\cancel{\omega}\frac{y^*}{x^*} = \cancel{\omega}\frac{x^*}{y^*} \Leftrightarrow y^{*2} + x^{*2} = 0 \Rightarrow \boxed{(x^*, y^*) = (0,0)}$$

- Linearisation around $(0,0)$:

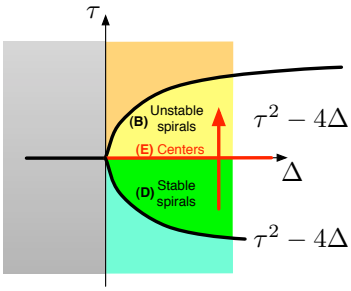
$$J(x,y) = \begin{pmatrix} \left(\mu - x^2 - y^2\right) + x(-2x) & -\omega - 2xy \\ \omega - 2xy & \left(\mu - x^2 - y^2\right) + y(-2y) \end{pmatrix}$$

Local stability analysis of $(0,0)$:

$$J(0,0) = \begin{pmatrix} \mu & -\omega \\ \omega & \mu \end{pmatrix}$$

$$\Rightarrow \boxed{\lambda_{\pm} = \mu \pm i\omega}$$

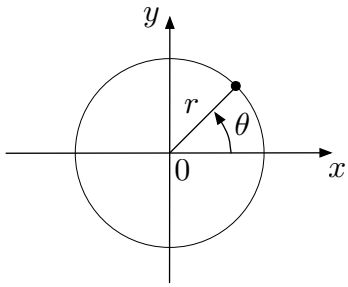
$$\begin{cases} \tau = 2\mu \\ \Delta = \mu^2 + \omega^2 > 0 \\ \tau^2 - 4\Delta = \cancel{4\mu^2} - \cancel{4\mu^2} - 4\omega^2 < 0 \end{cases}$$



$$\begin{cases} \mu > 0, & \text{Unstable spiral} \\ \mu = 0, & \text{Center} \\ \mu < 0, & \text{Stable spiral} \end{cases}$$

Global stability analysis for the limit cycle example (rare)

To perform the global analysis explicitly (which typically is very hard to do; the example we have chosen here is an exception in that respect), we rewrite (45)-(46) in polar coordinates:



$$\begin{pmatrix} x \\ y \end{pmatrix} \leftrightarrow \begin{pmatrix} r \\ \theta \end{pmatrix}$$

$$\begin{cases} r^2 = x^2 + y^2 \\ \tan(\theta) = \frac{y}{x} \end{cases}$$

i.e., transform $\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x})$ with $\boldsymbol{x} = \begin{pmatrix} x \\ y \end{pmatrix}$ into $\dot{\boldsymbol{p}} = \boldsymbol{F}(\boldsymbol{p})$ with $\boldsymbol{p} = \begin{pmatrix} r \\ \theta \end{pmatrix}$.

Differentiating $r^2 = x^2 + y^2$ with respect to time, we obtain:

$$\begin{aligned} \cancel{2}r\dot{r} &= \cancel{2}x\dot{x} + \cancel{2}y\dot{y} \\ r\dot{r} &= x\left[\cancel{-\omega y} + x\left(\mu - r^2\right)\right] + y\left[\cancel{\omega x} + y\left(\mu - r^2\right)\right] \\ \cancel{r}\dot{r} &= \cancel{r}^1\left(\mu - r^2\right) \\ \dot{r} &= r\left(\mu - r^2\right) \end{aligned} \tag{47}$$

Differentiating $\theta = \arctan\left(\frac{y}{x}\right)$ with respect to time, we obtain:

$$\begin{aligned}
\dot{\theta} &= \frac{d}{dt}\left(\arctan\left(\frac{y}{x}\right)\right) \\
&= \frac{d}{d\left(\frac{y}{x}\right)}\left(\arctan\left(\frac{y}{x}\right)\right)\frac{d\left(\frac{y}{x}\right)}{dt} \\
&= \frac{1}{1+\left(\frac{y}{x}\right)^2}\frac{x\dot{y}-\dot{x}y}{x^2} \\
&= \frac{x\left[\omega x + \cancel{y\left(\mu - r^2\right)}\right] - y\left[-\omega y + \cancel{x\left(\mu - r^2\right)}\right]}{r^2} \\
&= \omega\frac{\cancel{r^2}}{\cancel{r^2}} \\
\dot{\theta} &= \omega
\end{aligned} \tag{48}$$

This gives:

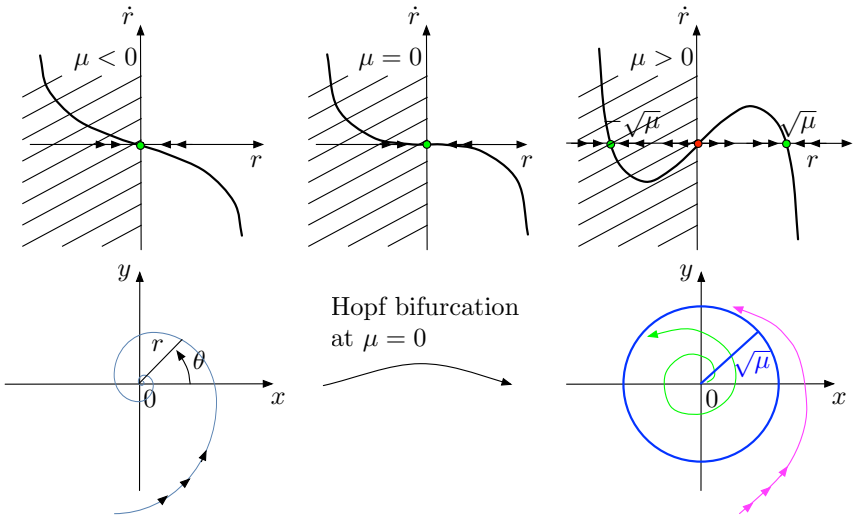
$$\dot{r} = r\left(\mu - r^2\right) \tag{49}$$

$$\dot{\theta} = \omega \tag{50}$$

Global stability analysis for the limit cycle example (cont')

The system of ODEs (49)-(50) is decoupled since $\dot{r}$ depends only on r, and $\dot{\theta} = \omega$. We can thus analyse these two ODEs separately using our knowledge for ODEs of order 1.

From $\dot{\theta} = \omega$, we have $\theta(t) = \omega t + \theta_0$, i.e., the phase grows linearly with time. From $\dot{r} = r\left(\mu - r^2\right) = r\mu - r^3$, we see that the number of fixed points depends on μ.

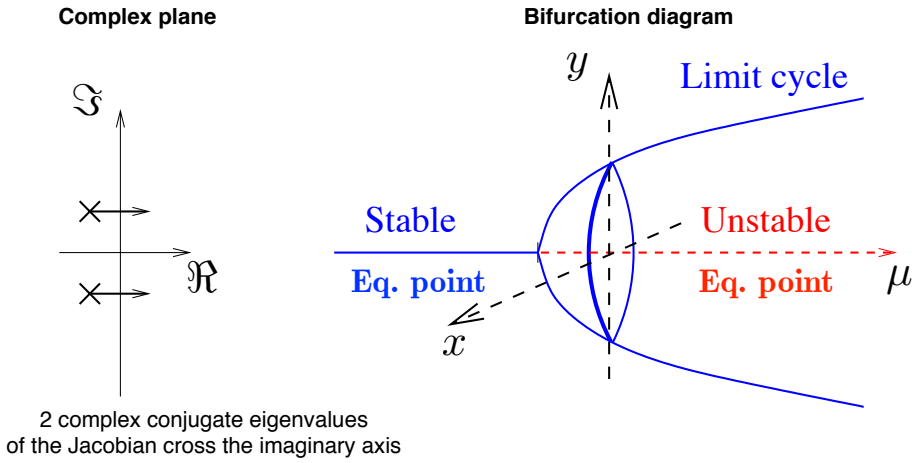


When $\mu \leq 0$, the origin is the only fixed point, and every solution converges to $r = 0$. This means that the full planar system is so that all trajectories spiral into the origin.

When $\mu > 0$, the origin becomes unstable, as we can see from the phase plane of $\dot{r}$ vs r. In fact, $\dot{r}$ is negative for $r > \sqrt{\mu}$ and positive for $0 < r < \sqrt{\mu}$, so that the fixed point $r = \sqrt{\mu}$ is an almost global attractor (except for an initial condition at $r = 0$). This means that the full planar system is so that all trajectories (except those starting at $r = 0$) spiral into the circle of radius $\sqrt{\mu}$, which is, therefore, a limit cycle. Note that the oscillation has magnitude $\sqrt{\mu}$ and frequency $\frac{\omega}{2\pi}$.

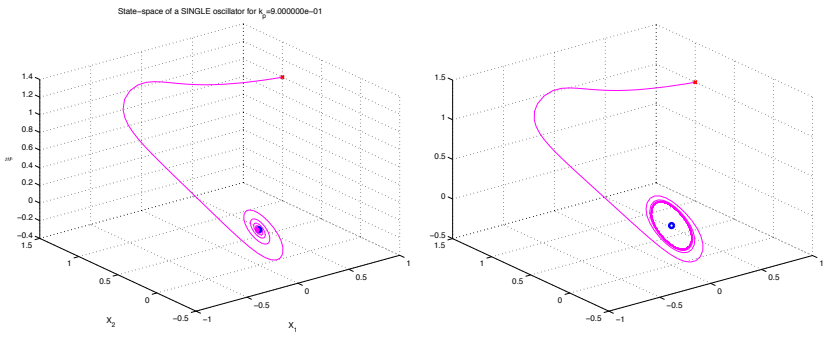
When we increase the values of μ from negative to positive we can see that, at $\mu = 0$, we transition from a situation where there is a change in the type of attractors. For $\mu < 0$, the

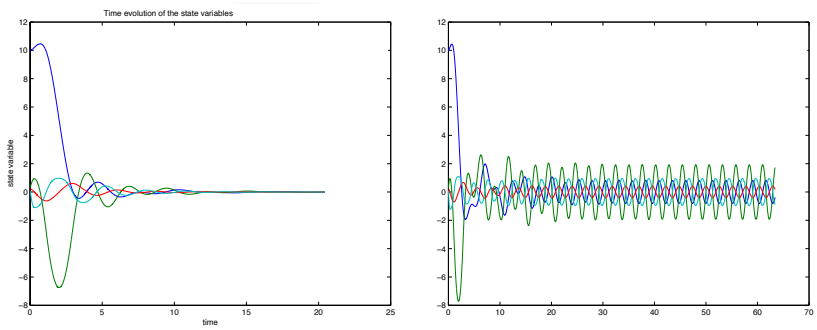
only attractor is the fixed point at the origin while for $\mu > 0$, the only attractor is the limit cycle of amplitude $\sqrt{\mu}$. When, changing the value of a parameter, such a change occurs, a *Hopf* bifurcation happens at the critical bifurcation value (i.e., the value where the qualitative change of behaviour occurs; here this is $\mu = 0$). The signature of a Hopf bifurcation is the emergence, when a parameter is varied, of a limit cycle *with increasing amplitude* (at $\mu = 0$ the amplitude of the limit cycle is 0, while it is $\sqrt{\mu}$ for $\mu \gtrsim 0$). This typically happens when at the critical bifurcation value (here, $\mu = 0$), two complex conjugate eigenvalues of the Jacobian J cross the imaginary axis (i.e., at the critical bifurcation value, two eigenvalues of J are purely imaginary). Note that this property of the eigenvalues of J is not enough to ensure the emergence of a limit cycle as this is a property that would also be true for linear systems and limit cycles can only occur for nonlinear systems. We omit here the additional technical conditions that need to be satisfied to ensure the emergence of a limit cycle. More details on these conditions and on the Hopf bifurcation can be found in the book by Strogatz.



Remark 55. *In biology, periodic oscillations are typically the result of limit cycles.*

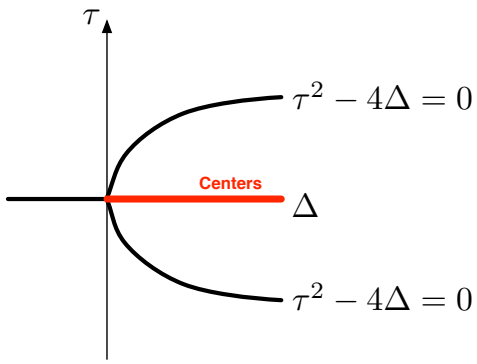
Example of a limit cycle in a model of order 3





5.6 Local (linear) and global stability are not equivalent for nonlinear ODE models of order 2 and higher

Remark 56. *In nonlinear ODE models of order* 2 *or larger, linear stability analysis is* not *equivalent to global stability analysis. This is especially true for center points:*



In particular, the stability of a fixed point in a nonlinear system can be deduced from the local stability analysis of the system linearised around this fixed only if *this point is not a center (the precise formulation of this statement is known as the* Hartman-Grobman theorem*). If this point* is *a center then the stability property of the corresponding fixed point of the nonlinear system can only be deduced by explicitly taking into account the nonlinarities (this is typically done by performing a* center manifold stability analysis *– don't worry, we will not cover this in this course).*

To better understand the previous remark, consider the following example:

$$\dot{x} = -y + ax\left(x^2 + y^2\right) \tag{51}$$
$$\dot{y} = x + ay\left(x^2 + y^2\right) \tag{52}$$

where a is a parameter.

- *Fixed points:*
$$(x^*, y^*) = (0, 0)$$
- *Linearisation around* $(0, 0)$*:*
$$J(x, y) = \begin{pmatrix} 3ax^2 + ay^2 & -1 + 2axy \\ 1 + 2axy & ax^2 + 3ay^2 \end{pmatrix}$$

- *Local stability analysis of* $(0,0)$*:*

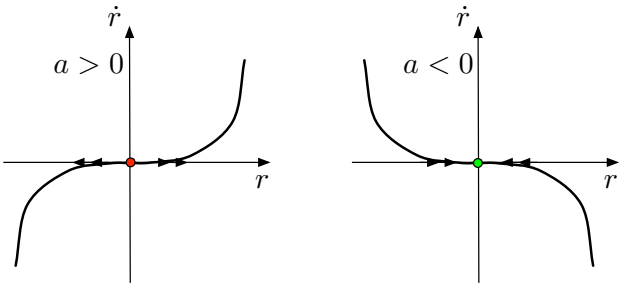
$$J(0,0) = \begin{pmatrix} 0 & -1 \\ 1 & 0 \end{pmatrix} \Rightarrow \boxed{\lambda_{\pm} = \pm i}, \text{ or } \begin{cases} \tau = 0 \\ \Delta = 1 \end{cases}$$

The local stability analysis of $(0,0)$ *indicates that the fixed point* $(0,0)$ *is a center for any value of the parameter* a*. As we will see next the stability of* $(0,0)$ *in the original system* (51)-(52) *is actually very different (i.e., in the original system the fixed point* $(0,0)$ *is not a center).*

Rewriting the nonlinear ODEs (51) *and* (52) *in polar coordinates, we obtain:*

$$\dot{r} = ar^3$$
$$\dot{\theta} = 1$$

which, as was the case for (49) *and* (50)*, are decoupled. In particular, we see that* $\theta(t) = t + \theta_0$ *and thus the phase variable* $\theta(t)$ *grows linearly with time.*



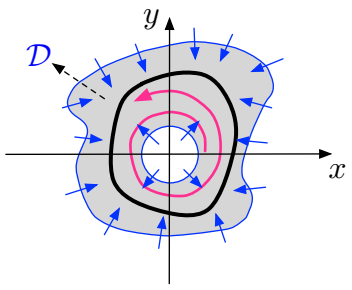
Now looking at the dynamics of r*, i.e.,* $\dot{r} = ar^3$ *and drawing the corresponding phase plane, we can see that the stability of the fixed point* $(0,0)$ *depends on "a" and that, except for* $a = 0$*,* $(0,0)$ *is actually never a center of the original nonlinear system.*

Typically, it is quite difficult to actually prove that a limit cycle exists. But for models of order 2, there is a very powerful criterion called the Poincaré-Bendixson Theorem.

5.7 The Poincaré-Bendixson Theorem

The basic idea is the following:

For an ODE model of order 2, if you are able to find a region in the phase plane which does not contain any fixed point and is attractive, then this region must contain a limit cycle.



A loose statement of the Poincaré-Bendixson Theorem

Suppose $\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x})$, $\boldsymbol{x} \in \mathbb{R}^2$, is a continuously differentiable vector field[20]

and there exists a bounded subset $\mathcal{D}$ of the phase plane such that

[20] A vector valued function is said to be continuously differentiable if the partial derivatives of each of its component functions with respect to each of the functions' arguments all exist and are continuous functions of their arguments.

- no trajectory can exit $\mathcal{D}$, in which case $\mathcal{D}$ is said to be "forward-invariant" with respect to $\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x})$ or a "trapping region" of $\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x})$,
- and there are no fixed points inside $\mathcal{D}$.

Then, there exists at least one limit cycle in $\mathcal{D}$ and any trajectory that enters $\mathcal{D}$ converges to a limit cycle.

The main idea behind the proof of this theorem comes from the fact that trajectories cannot cross and that our movements are restricted to a plane since $\boldsymbol{x} \in \mathbb{R}^2$. Since a trajectory starting in $\mathcal{D}$ cannot escape $\mathcal{D}$ or cross itself (remember that trajectories cannot cross except at fixed points, which $\mathcal{D}$ does not contain by definition), this trajectory must approach a closed orbit, i.e., a limit cycle.

Remark 57. *The Poincaré-Bendixson theorem is only valid for models of order* 2. *It does* not *hold for models of order* 1 *(which cannot have limit cycles) or for models of order* 3 *or higher (where the non-crossing property is insufficient to restrict the behaviour to a limit cycle).*

The Poincaré-Bendixson theorem is a very powerful tool to establish the existence of attractive limit cycles in models of order 2. The main difficulty is to find a set $\mathcal{D}$ that satisfies the assumptions of the theorem.

The Poincaré-Bendixson Theorem: How can we find $\mathcal{D}$?

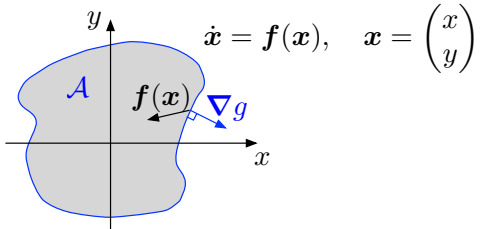
Let $\mathcal{A}$ be defined as

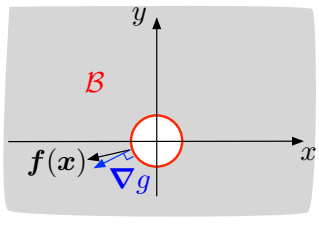
$$\mathcal{A} = \left\{ \boldsymbol{x} = \begin{pmatrix} x \\ y \end{pmatrix} \in \mathbb{R}^2 : g(\boldsymbol{x}) \leq 0 \right\}$$

for some continuously differentiable function $g : \mathbb{R}^2 \to \mathbb{R}$. Then the set $\mathcal{A}$ is forward invariant if and only if

$$\frac{dg(\boldsymbol{x})}{dt} = \frac{\partial g(\boldsymbol{x})}{\partial \boldsymbol{x}} \frac{d\boldsymbol{x}}{dt} = \frac{\partial g(\boldsymbol{x})}{\partial \boldsymbol{x}} \boldsymbol{f}(\boldsymbol{x}) \leq 0 \qquad \forall \boldsymbol{x} : g(\boldsymbol{x}) = 0.$$

The above has a rather simple interpretation. If $\boldsymbol{x}$ starts inside $\mathcal{A}$ and, as time t evolves, $\boldsymbol{x}(t)$ ever "reaches" the boundary of $\mathcal{A}$, which is defined as $\bar{\mathcal{A}} = \{\boldsymbol{x} \in \mathbb{R}^2 : g(\boldsymbol{x}) = 0\}$, then $g(\boldsymbol{x}(t))$ cannot increase any further since $\frac{dg(\boldsymbol{x}(t))}{dt} \leq 0, \forall \boldsymbol{x} \in \bar{\mathcal{A}}$. Thus $\boldsymbol{x}$ must remain in $\mathcal{A}$.





$\mathcal{A} = \{(x, y) \in \mathbb{R}^2 : g(x, y) \leq 0\}$ $\qquad$ $\mathcal{B} = \{(x, y) \in \mathbb{R}^2 : b(x, y) \geq 0\}$

$\bar{\mathcal{A}} = \{(x, y) \in \mathbb{R}^2 : g(x, y) = 0\}$ $\qquad$ $\bar{\mathcal{B}} = \{(x, y) \in \mathbb{R}^2 : b(x, y) = 0\}$

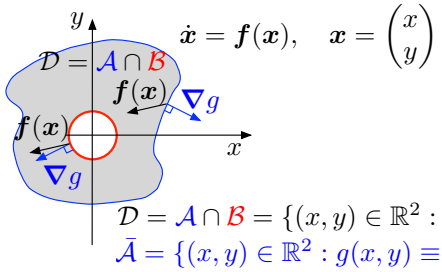
Alternatively, one can view the above geometrically by noting that the vector normal to the boundary of $\mathcal{A}$, at a point on the boundary $\boldsymbol{x}$, (pointing outwards $\mathcal{A}$, see Remark 59) is given by $\boldsymbol{\nabla} g(\boldsymbol{x}) = \left(\frac{\partial g(\boldsymbol{x})}{\partial \boldsymbol{x}} \right)^T$. Thus, $\frac{\partial g(\boldsymbol{x})}{\partial \boldsymbol{x}} \boldsymbol{f}(\boldsymbol{x}) = \boldsymbol{\nabla} g(\boldsymbol{x})^T \boldsymbol{f}(\boldsymbol{x}) \leq 0, \forall \boldsymbol{x} \in \bar{\mathcal{A}}$ implies that the angle between $\boldsymbol{f}(\boldsymbol{x})$, the direction in which $\boldsymbol{x}$ is moving, and $\boldsymbol{\nabla} g(\boldsymbol{x})$, the vector normal to the boundary of $\mathcal{A}$, is greater than 90°. Thus $\boldsymbol{f}(\boldsymbol{x})$ is always "pointing to somewhere inside of $\mathcal{A}$" and hence $\boldsymbol{x}$ can never leave $\mathcal{A}$.

Remark 58. *Equivalently, the set* $\mathcal{B} = \{\boldsymbol{x} \in \mathbb{R}^2 : b(\boldsymbol{x}) \geq 0\}$, *where* $b : \mathbb{R}^2 \to \mathbb{R}$ *is a continuously differentiable function, is forward invariant if and only if*

$$\frac{db(\boldsymbol{x})}{dt} = \frac{\partial b(\boldsymbol{x})}{\partial \boldsymbol{x}} \frac{d\boldsymbol{x}}{dt} = \frac{\partial b(\boldsymbol{x})}{\partial \boldsymbol{x}} \boldsymbol{f}(\boldsymbol{x}) \geq 0 \qquad \forall \boldsymbol{x} : b(\boldsymbol{x}) = 0.$$

Remark 59. *The gradient of a scalar function, $g(x,y)$, is given by* $\boldsymbol{\nabla} g(x,y) = \begin{pmatrix} \frac{\partial}{\partial x} g(x,y) \\ \frac{\partial}{\partial y} g(x,y) \end{pmatrix}$. *The gradient of a scalar function evaluated at a given point (x,y) is defined as the vector whose direction is that of the greatest rate of increase of that scalar function at that point, and whose magnitude is that rate of increase.*

By noting that *the intersection of two forward invariant sets (or regions) is also forward-invariant*, one can use several different forward invariant sets to construct more refined forward invariant sets. This is easy to prove: if $\mathcal{A}$ and $\mathcal{B}$ are forward invariant sets with respect to $\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x})$ and $\boldsymbol{x}$ is initialised in the intersection $\mathcal{D} = \mathcal{A} \cap \mathcal{B}$, then it may never leave $\mathcal{A}$ (since $\mathcal{A}$ is forward invariant by assumption) and it may never leave $\mathcal{B}$ (since $\mathcal{B}$ is forward invariant by assumption). Therefore, $\boldsymbol{x}$ may never leave the intersection of $\mathcal{A}$ and $\mathcal{B}$ which implies that the intersection set, $\mathcal{D}$, is also forward invariant.



$$\mathcal{D} = \mathcal{A} \cap \mathcal{B} = \{(x,y) \in \mathbb{R}^2 : g(x,y) \leq 0, b(x,y) \geq 0\}$$
$$\bar{\mathcal{A}} = \{(x,y) \in \mathbb{R}^2 : g(x,y) \equiv 0\}$$
$$\bar{\mathcal{B}} = \{(x,y) \in \mathbb{R}^2 : b(x,y) \equiv 0\}$$

The Poincaré-Bendixson Theorem: an example

Consider the second order model in example in (45)-(46) for $\mu > 0$:

$$\dot{x} = -\omega y + x\left(\mu - x^2 - y^2\right)$$
$$\dot{y} = \omega x + y\left(\mu - x^2 - y^2\right)$$

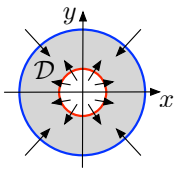
Suppose that we suspect there is a limit cycle encircling the origin. To prove its existence we first have to find a trapping region that contains it. Consider as a candidate trapping region the ball of radius R centred at the origin, that is $\mathcal{A}_R = \{(x,y) \in \mathbb{R}^2 : g(x) = x^2 + y^2 - R^2 \leq 0\}$. Then,

$$\boldsymbol{\nabla} g(x,y)^T \begin{pmatrix} \dot{x} \\ \dot{y} \end{pmatrix} = \begin{pmatrix} 2x & 2y \end{pmatrix} \begin{pmatrix} -\omega y + x\left(\mu - x^2 - y^2\right) \\ \omega x + y\left(\mu - x^2 - y^2\right) \end{pmatrix} = 2(x^2 + y^2)(\mu - x^2 - y^2)$$

By definition, if $(x,y) \in \bar{\mathcal{A}}_R$ (remember, $\bar{\mathcal{A}}_R$ denotes the boundary of $\mathcal{A}_R$), then $x^2 + y^2 = R^2$. So, for all $\boldsymbol{x} \in \bar{\mathcal{A}}_R$, we have $\boldsymbol{\nabla} g(x,y)^T \boldsymbol{f}(\boldsymbol{x}) = 2R^2(\mu - R^2) \leq 0$ for all $R \geq \sqrt{\mu}$. Thus, any ball centred at the origin which has a radius bigger than or equal to $\sqrt{\mu}$ is forward invariant.

Can we now conclude there exists a limit cycle around the origin? No! Not yet. As we know, the system considered in (45)-(46) has a fixed point at $(0,0)$ and so the second condition of the Poincaré-Bendixson Theorem is not satisfied. We can however attempt to define a set $\mathcal{D}$ that "cuts out" a region around the origin. We do so by showing that the *outside* of some ball centred at the origin and with small radius $r \leq R$ is forward invariant. Consider such a set, $\mathcal{B}_r = \{(x,y) \in \mathbb{R}^2 : b(x) = x^2 + y^2 - r^2 \geq 0\}$. Similarly as before, we have that if $(x,y) \in \bar{\mathcal{B}}_r$, then $x^2 + y^2 = r$ and $\boldsymbol{\nabla} b(x,y)^T \begin{pmatrix} \dot{x} & \dot{y} \end{pmatrix}^T = 2r^2(\mu - r^2) \geq 0$ for any $r \leq \sqrt{\mu}$. Thus, the outside of any ball centred at the origin with radius smaller or equal to $\sqrt{\mu}$ is forward invariant (see Remark 58).

Choosing any $R \geq \sqrt{\mu}$ and $r \leq \sqrt{\mu}$ we have shown that the doughnut $\mathcal{D} = \mathcal{A}_R \cap \mathcal{B}_r = \{(x,y) \in \mathbb{R}^2 : r^2 \leq x^2 + y^2 \leq R^2, r \leq \sqrt{\mu} \leq R\}$ is forward invariant. In addition, it contains no fixed points, so applying the Poincaré-Bendixson Theorem we can conclude that there exists a limit cycle in $\mathcal{D}$ and that any trajectory that enters $\mathcal{D}$ converges to a limit cycle. Indeed, in this case, we can go a step further and by "squeezing" the doughnut, that is setting $R = r = \sqrt{\mu}$, we can deduce that the limit cycle is the circle defined by $x^2 + y^2 = \mu$.



The Poincaré-Bendixson Theorem: some final remarks

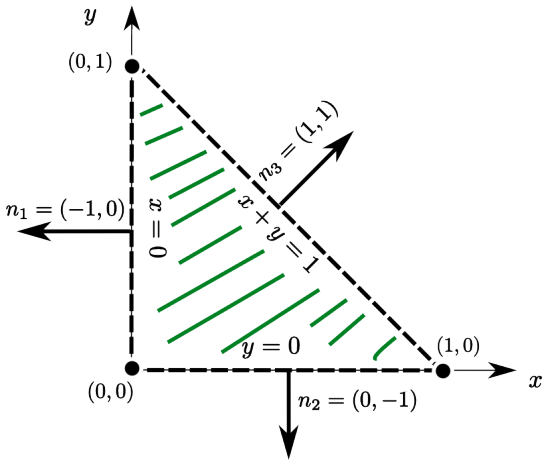
The first time we considered the system defined by (45)-(46), we established that if $\mu > 0$, then $(0,0)$ locally behaves like an unstable focus. Unstable focuses and unstable nodes share the property that all trajectories not initialized on them, diverge away from them. Thus, it is not surprising that we were able to "cut out" the fixed point from the trapping region (that is, find a forward invariant $\mathcal{B}_r$). Indeed, it can be shown that *one can always remove unstable focuses and unstable nodes from trapping regions.* Thus, in the example we didn't even have to find $\mathcal{B}_r$ to conclude that a limit cycle exists. All that we had to do was linearise around the equilibrium, verify that it behaves locally as an unstable focus (or node) and, once this is done, we can safely ignore the fixed point when constructing the trapping region.

For some systems, the simplest trapping regions that can be found are polytopes (e.g., triangles, rectangles, etc). Unfortunately, these sets cannot be described by a continuously differentiable function as was assumed above (that is, they cannot be written as polytope $\mathcal{P} = \{\boldsymbol{x} \in \mathbb{R}^2 : g(\boldsymbol{x}) \leq 0\}$ where $g(\boldsymbol{x})$ is continuously differentiable). However, a rather simple approach still allows to construct trapping regions in that case: Suppose that the boundary of polytope $\mathcal{P}$ is given by m line segments $l_1, l_2, \ldots, l_m$ and let n_i denote the vector normal to l_i (pointing towards the outside of $\mathcal{P}$). Then, similarly as before, one can show that the polytope is a trapping region by showing that $\boldsymbol{f}(\boldsymbol{x})$ evaluated at the boundary of $\mathcal{P}$ always points "inside" $\mathcal{P}$, that is by showing that $\boldsymbol{x} \in l_i \Rightarrow \boldsymbol{n_i}^T \boldsymbol{f}(\boldsymbol{x}) \leq 0$ for all $i = 1, 2, \ldots, m$.

For example, consider the system described by

$$\dot{x} = x(-x^2 + y^3 - y - 1) + 1$$
$$\dot{y} = y(x - 1 - x^2 - y^2)$$

and the right-angle triangle, $\mathcal{T}$, whose sides are given by $l_1 = \{(0, \alpha) : 0 \leq \alpha \leq 1\}$, $l_2 = \{(\alpha, 0) : 0 \leq \alpha \leq 1\}$ and $l_3 = \{(\alpha, 1-\alpha) : 0 \leq \alpha \leq 1\}$.



The normal vectors are given by $\boldsymbol{n_1} = (-1, 0)^T$, $\boldsymbol{n_2} = (0, -1)^T$ and $\boldsymbol{n_3} = (1, 1)^T$. Then all we need to do is check that $\boldsymbol{f}(x, y) = (\dot{x}, \dot{y})^T$ points inside $\mathcal{T}$, along each of the sides of $\mathcal{T}$:

$$(x,y) \in l_1 \Rightarrow \boldsymbol{n_1}^T \boldsymbol{f}(x,y) = -\dot{x}|_{(x,y)\in l_1} = -x(-x^2+y^3-y-1)-1|_{(x,y)\in l_1} = -(0)(-0^2+\alpha^3-\alpha-1)-1 = -1 \leq 0,$$

$$(x,y) \in l_2 \Rightarrow \boldsymbol{n_2}^T \boldsymbol{f}(x,y) = -\dot{y}|_{(x,y)\in l_2} = -y(x-1-x^2-y^2)|_{(x,y)\in l_2} = -(0)(\alpha-1-\alpha^2-0^2) = 0,$$

$(x,y) \in l_3 \Rightarrow \boldsymbol{n_3}^T \boldsymbol{f}(x,y) = (\dot{x}+\dot{y})\,|_{(x,y)\in l_3} = x(-x^2+y^3-y-1)+1+y(x-1-x^2-y^2)|_{(x,y)\in l_3}$

$$= 1-x^3-y^3-x-y-yx^2+xy^3|_{(x,y)\in l_3}$$

$= 1-(x+y)(x^2+1)+y^3(x-1)|_{(x,y)\in l_3} = 1-(1)(\alpha^2+1)+(1-\alpha)^3(\alpha-1) = -(1-\alpha)^4-\alpha^2 \leq 0.$

(where the last two equalities were obtained considering $x=\alpha$ and $y=1-\alpha$ since $x+y=1$ for all $(x,y)\in l_3$)

Thus, $\mathcal{T}$ is forward invariant.

5.8 Summary of behaviours for nonlinear ODE models of order 1 & 2

- Models of order 1
  - Attractors: fixed points, or ∞
  - Local (linear) and global stability analysis are equivalent
  - Bifurcations: Saddle node, Transcritical, or Pitchfork
- Models of order 2
  - Attractors: fixed points, limit cycles, or ∞
  - Local stability analysis (linearisation) around fixed points $\neq$ global stability analysis
  - Bifurcation: all those of order 1 + Hopf + others

6 Nonlinear ODE models of order 3 and higher

$$\boxed{\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x}), \quad \boldsymbol{x} \in \mathbb{R}^d, \quad d \geq 3, \quad f(\cdot) : \mathbb{R}^d \to \mathbb{R}^d, \text{ "smooth" function}} \tag{53}$$

Behaviours for ODE models of order 3 and higher
In one sentence: everything that happens in lower order models + 2 other phenomena:

- quasi-periodicity
- deterministic chaos

Some good introductions to quasi-periodicity and deterministic chaos can be found in the **book of Strogatz** and on the following webpages:

- **Quasi-periodic oscillations:**
  - http://www.scholarpedia.org/article/Quasiperiodic_Oscillations
  - http://en.wikipedia.org/wiki/Quasiperiodic_motion
- **Deterministic chaos:**
  - http://en.wikipedia.org/wiki/Chaos_theory
  - http://www.scholarpedia.org/article/Chaos_in_neurons
  - http://en.wikipedia.org/wiki/Lorenz_system
  - http://planetmath.org/LorenzEquation
  - http://mathworld.wolfram.com/LorenzAttractor.html

If you want to see some really cool videos on experimental nonlinear dynamics and chaos, I also recommend having a look at the following videos from Strogatz:

- **Lorenz Waterwheel:**
  http://uk.youtube.com/watch?v=7iNCfNBEJHo
- **Double pendulum:**
  http://uk.youtube.com/watch?v=anwl6OZ1UuQ
- **Airplane wing vibrations (and instabilities!):**
  http://uk.youtube.com/watch?v=_Ys8qGxr--M
- **Chemical oscillations (Belousov-Zhabotinsky):**
  http://uk.youtube.com/watch?v=8R33KWPmqlo
- **Synchronised chaotic circuits and communications:**
  http://uk.youtube.com/watch?v=J-ca_bqWp4I
- **Musical chaos:**
  - http://uk.youtube.com/watch?v=dL4VKuKNgXI
  - http://uk.youtube.com/watch?v=Wz3cmlVwI30

6.1 Summary for nonlinear ODE systems of order 1, 2, 3, and higher

	1D	**2D**	**3D and higher**
Attractors	Only F.P. or $\pm\infty$	Same as 1D + limit cycles	Same as 2D + quasi-periodic attractor + chaotic attractor
Behaviours	Decay or explosion	Same as 1D + robust periodic oscillations	Same as 2D + quasi-periodicity + chaos
Bifurcations	Saddle-Node Transcritical Pitchfork	Same as 1D + Hopf + others	Same as 2D + many more

Remark 60. *Discrete-time systems (also called maps) have a much wilder behaviour than continuous-time systems. For example, one dimensional discrete-time systems (also called one dimensional maps) such as $x_{k+1} = rx_k(1 - x_k)$ can exhibit limit cycles or even chaotic behaviours even though $x_k \in \mathbb{R}, \forall k$.*

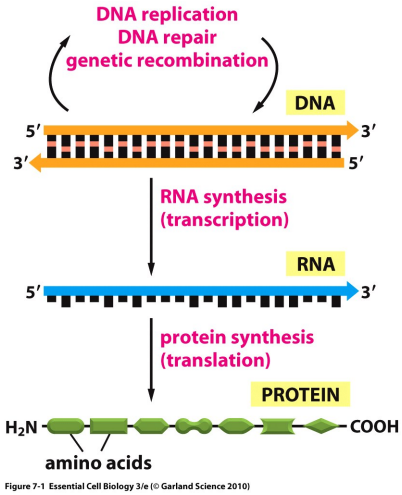
7 Modelling gene regulation networks

7.1 The central dogma in molecular biology

Even if the entire genome of some model organisms can be (and indeed has been) decoded, the links between genetic information and biological function are still far from being understood. In what follows we briefly introduce and provide some modelling examples of gene regulation mechanisms.

We first start with a brief reminder of the *central dogma* of molecular biology.

The central dogma



Proteins are the workhorses of the cell. Most of the life-critical tasks of the cell are accomplished by proteins, each protein having its own function and specificity. The information required to synthesise proteins is found in a sequential genetic information carrying polymer called DNA (deoxyribonucleic acid). A gene is a section of DNA, typically delimited by a promoter at its beginning and a terminator at its end. The protein synthesis process is done in two phases: transcription and translation.

- **Transcription**: Transcription is the process by which the information contained in a section of DNA (e.g., a gene) is transferred to a newly assembled piece of messenger RNA (mRNA). Transcription is triggered by the binding of RNA Polymerase to specific regions on the DNA called promoters, and is enabled/facilitated or impeded by a range of promoter-specific proteins called transcription factors.
- **Translation**: During translation, proteins are produced by ribosomes that bind to a specific site of the mRNA (the ribosome binding site – RBS) and then move along the mRNA chain converting triplets of bases or "codons" on the mRNA into the appropriate peptide chain of amino acids defining the desired protein. The mRNA is read by the ribosome as triplet of bases, usually beginning with an AUG, or an initiator methionine codon downstream of the ribosome binding site. Complexes of initiation factors and elongation factors bring aminoacylated transfer RNAs (tRNAs) into the ribosome-mRNA complex, matching the codon in the mRNA to the anti-codon in the tRNA, thereby adding the correct amino acid in the growing peptide sequence encoding the emerging protein. As the amino acids are linked together into the growing peptide chain, they begin folding into the correct conformation. This folding continues until the nascent polypeptide chain is released from the ribosome as a mature protein.

Remark 61. *In eukaryotic cells, the site of transcription (the cell nucleus) is usually separated from the site of translation (the cytoplasm), so the mRNA must be transported out of the nucleus into the cytoplasm, where it can be bound by ribosomes.*

In prokaryotic cells, there is no nucleus and translation and transcription are coupled: the mRNA chains are being translated immediately as they are being synthesised.

Remark 62. *Please note that not all RNA are translated. Certain RNA can perform other important functions. For instance, ribosomes are made of non-translated RNAs (rRNAs).*

7.2 Constitutive gene expression

As we have seen, the central dogma can be summarized as:

$$\text{Gene} \xrightarrow[\text{Transcr.}]{} \text{mRNA} \xrightarrow[\text{Transl.}]{} \text{Protein}$$

$$\text{mRNA} \xrightarrow[\text{Degrad.}]{} \emptyset$$

$$\text{Protein} \xrightarrow[\text{Degrad.}]{} \emptyset$$

When gene expression is unregulated, it is said to be *constitutive*, and the gene is always on. Using the law of mass action[21], a model for constitutive expression is given as:

$$\dot{m} = k_1 - d_1 m \tag{54}$$

$$\dot{p} = k_2 m - d_2 p \tag{55}$$

where $m = [mRNA]$ and $p = [Protein]$, where $[X]$ represents the concentration of X.

- k_1 is the constitutive transcription rate. It is considered to be constant, and it represents the number of mRNA molecules produced per gene, per unit of time. (Here we consider that there is only one copy of the gene in the cell (e.g., chromosomal gene); if there were several copies (e.g., plasmid located gene) we would multiply k_1 by the copy number N to obtain to total transcription rate of the considered gene).
- d_1 is the mRNA degradation rate. The typical half-life for mRNA, in *E. coli*, has been measured to be between 2 min. and 8 min. (average 5 min.)
- k_2 is the translation rate. It is considered to be constant, and it represents the number of protein molecules produced per mRNA molecule, per unit of time.
- d_2 is the protein degradation rate. In practice, the degradation rate of a protein is made of two terms. The first term corresponds to the propensity of the protein to break down per unit of time. The second term called the dilution term corresponds to the variation of the cell volume (through cell expansion and division) per unit of time (remember that p represents protein concentration and thus in that situation the volume of the cell influences the protein concentration). Typically the division time of *E.coli* ranges from 20-40 mins.

The model (54)-(55) is a linear ODE model of order 2. It can be written under the form:

$$\begin{pmatrix} \dot{m} \\ \dot{p} \end{pmatrix} = \begin{pmatrix} -d_1 & 0 \\ k_2 & -d_2 \end{pmatrix} \begin{pmatrix} m \\ p \end{pmatrix} + \begin{pmatrix} k_1 \\ 0 \end{pmatrix} \tag{56}$$

This model is of the form $\dot{\boldsymbol{x}} = A\boldsymbol{x} + \boldsymbol{b}$.

- Fixed point: $(m^*, p^*) = \left(\frac{k_1}{d_1}, \frac{k_1 k_2}{d_1 d_2}\right)$

[21] This is based on empirical studies since strictly speaking it does not really make sense to use the law of mass action for gene expression.

- Stability of the fixed point: the stability of a model of the form $\dot{\boldsymbol{x}} = A\boldsymbol{x} + \boldsymbol{b}$ (such as the model (56)) is also given by the eigenvalues of the A. This can easily be seen by rewriting the system under the form $\dot{\boldsymbol{x}} = A\left(\underbrace{\boldsymbol{x} + A^{-1}\boldsymbol{b}}_{=\tilde{\boldsymbol{x}}}\right)$ which, posing $\tilde{\boldsymbol{x}} = \boldsymbol{x} + A^{-1}\boldsymbol{b}$, gives $\dot{\tilde{\boldsymbol{x}}} = A\tilde{\boldsymbol{x}}$. So we have: $\lambda_1 = -d_1$ and $\lambda_2 = -d_2$: Stable node (or, if you want to use the τ-Δ approach: $\tau = \lambda_1 + \lambda_2 = -d_1 - d_2 < 0$, $\Delta = \lambda_1\lambda_2 = d_1 d_2 > 0$, $\tau^2 - 4\Delta = (d_1 - d_2)^2 \geq 0$: Stable node).

Remark 63. *Before moving to other types of gene expressions, it is interesting to explore the application of the quasi-stationary assumption on the expression of mRNA. Typically the concentration of mRNA reaches steady state very quickly, compared to the protein concentration. This suggests that we could neglect the fast transient required by the mRNA concentration to reach its steady state and thus use the following quasi-stationary approximation: $\dot{m} \approx 0$. Under this assumption, we see that the corresponding model is now of order 1 and given by $\dot{p} = c - d_2 p$ with $c = \frac{k_1 k_2}{d_1}$. This model has a stable fixed point at $p^* = \frac{c}{d_2}$ which is the same as previously.*

Few genes are known to have a purely constitutive expression. Most genes have their expression controlled by some outside signals (DNA-binding proteins, temperature, metabolites, RNA molecules, etc.). In the next section we will particularly focus on the study of DNA-binding proteins, called transcription factors. These proteins, when binding to the promoter region of the gene, can either have an activation effect on the gene (positive gene regulation action), or a repression effect (negative gene regulation action). In prokaryotes, control of transcriptional initiation is considered to be the major point of regulation in gene expression.

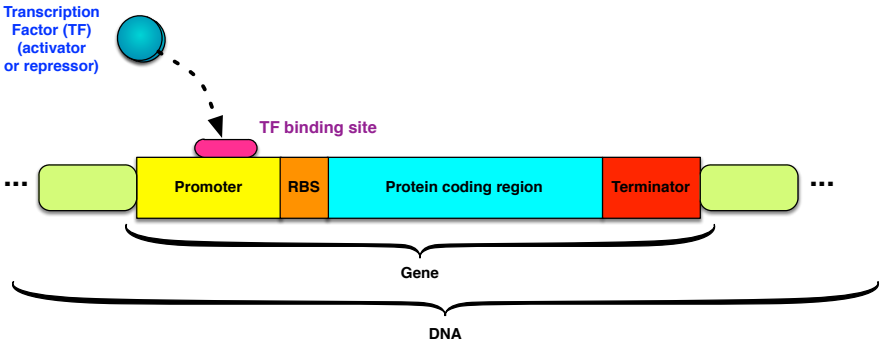
Remark 64. *At least two arguments seem to indicate the need for regulation of gene expression:*

- *Each cell of a multicellular organism carries the same genetic information. Although the genetic information is the same in all differentiated cells of a multicellular organism, the set of genes expressed is different in each differentiated cell (Indeed, a neuron, a skin cell or a liver cell do not exhibit the same phenotypes. This is a consequence of these cells expressing different genes based on the same genetic code).*
- *Cells are subject to a large range of environmental perturbations/changes (e.g., pH, nutrients, space, etc.) and have to react to them in order to survive. Such constraints can only be met through appropriate spatio-temporal regulation of gene expression.*

Gene expression must therefore be controlled by some mechanisms which determine at each instant of time which gene (or set of genes) should or should not be expressed.

7.3 Gene transcription regulation

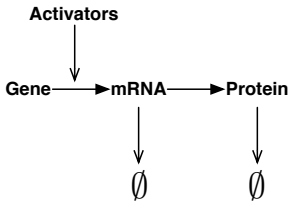
At the transcription level, gene expression can be controlled by certain proteins called *transcription factors*:



Transcription factors are transcription regulation proteins which can bind certain sections of the promoter, called *transcription factor binding sites*, thereby either preventing (inhibiting) or facilitating (activating) the transcription of the gene, i.e., the binding of mRNA polymerase to the promoter. When gene transcription is controlled by transcription factors, we talk about *inducible gene expression*.

7.3.1 Gene transcription regulation by activators

Consider the case of a gene whose transcription is **activated** by the cooperative binding of activators to the transcription factor binding site of the gene.



The following model is commonly used to describe activator controlled gene transcription:

$$\dot{m} = k_1 \frac{A^n}{K^n + A^n} - d_1 m \tag{57}$$

$$\dot{p} = k_2 m - d_2 p \tag{58}$$

where $m = [mRNA]$, $p = [Protein]$, $A = [Activator]$, k_1 = maximal transcription rate, K = activation coefficient, n = Hill coefficient (= number of activators that need to cooperatively bind the promoter to trigger the activation of gene expression).

Exercise 1. *As an exercise, plot the activating Hill function* $\frac{A^n}{K^n+A^n}$ *as a function of* A *for* $K = 1$ *and both* $n = 1$ *and* $n = 2$. *Explain why this shape is appropriate to model transcriptional activation of gene expression as a function of the amount of activator* A.

Remark 65. *The activating (i.e., monotonically increasing) Hill function appearing in the dynamics of* $\dot{m}$ *can be derived from considering the DNA/activators complex to very quickly reach its steady state value (similarly to the enzyme-substrate complex quasi-stationary assumption in the Michaelis-Menten enzymatic reaction). Indeed, the set of reactions for the mRNA dynamics can be seen as:*

$$nA + D \underset{\beta}{\overset{\alpha}{\rightleftharpoons}} C \ (+mRNAPol) \xrightarrow{\tilde{k}_1} m + D + nA \ (+mRNAPol)$$

$$m \xrightarrow{d_1} \emptyset$$

where $A = Activator$, $D = DNA$, $C = DNA/activators\ complex$, *and* $m = mRNA$. *Using the law of mass action to write the ODEs and the following two assumptions:*

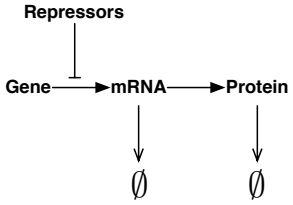
- *the quasi-stationary approximation* $\dot{C} \approx 0$, *and*
- *the conservation law* $D + C = D_{total}$ *where* D_{total} *is a positive constant (expressing that the total amount of DNA is conserved),*

it is easy to show that the dynamics for the concentration of mRNA is of the form given in (57). *The derivation is given in Appendix B.*

As with enzymatic reactions, the real set of reactions describing the activation process is much more complicated than shown above. It has however remained common to use a Hill function to model activation. As a consequence of this unaccounted complexity of the regulated activation process, instead of using an integer exponent as the model suggests, non-integer Hill coefficients might need to be used (e.g., $n = 2.8$*) to fit data appropriately.*

7.3.2 Gene transcription regulation by repressors

Consider the case of a gene whose transcription is **repressed** by the cooperative binding of repressors to the transcription factor binding site of the gene.



The following ODE model describes repressor-controlled gene transcription:

$$\dot{m} = k_1 \frac{K^n}{K^n + R^n} - d_1 m \tag{59}$$

$$\dot{p} = k_2 m - d_2 p \tag{60}$$

where $m = [mRNA]$, $p = [Protein]$, $R = [Repressor]$, k_1 = maximal transcription rate, K = repression coefficient, n = Hill coefficient (= number of repressors that need to cooperatively bind the promoter to trigger the inhibition of gene expression).

Remark 66. *Repressor proteins often bind to DNA as dimers (i.e., $n \approx 2$) or pairs of dimers (effectively tetramers, i.e., $n \approx 4$).*

Exercise 2. *As an exercise, plot the repressing Hill function $\frac{K^n}{K^n+R^n}$ as a function of R for $K = 1$ and both $n = 1$ and $n = 2$. Explain why this shape is appropriate to model transcriptional repression of gene expression as a function of the amount of repressor R.*

Remark 67. *Similarly to what we saw for the activator case, the repressing (i.e., monotonically decreasing) Hill function can be derived from considering the DNA/repressors complex to very quickly reach its steady state value. Indeed, the set of reactions for the mRNA dynamics can be seen as:*

$$nR + D \underset{\beta}{\overset{\alpha}{\rightleftharpoons}} C$$

$$D\ (+mRNAPol) \xrightarrow{\tilde{k}_1} m + D\ (+mRNAPol)$$

$$m \xrightarrow{d_1} \emptyset$$

where $R = Repressor$, $D = DNA$, $C = DNA/repressors\ complex$, and $m = mRNA$. Using the law of mass action, the quasi-stationary approximation $\dot{C} \approx 0$ and the conservation law $D + C = D_{total}$ where D_{total} is a positive constant, it is easy to show that the dynamics for the concentration of mRNA is of the form given in (59). The derivation is given in Appendix C.

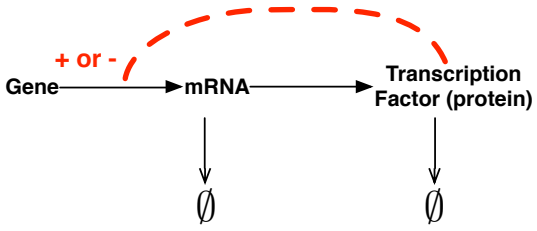
Again, the real set of reactions describing the repression process is much more complicated than shown above and this might result in the need to use non-integer values for the Hill coefficient n to fit data appropriately.

When genes interact with each other in a nonlinear fashion, new behaviours emerge. These behaviours do not require many genes to appear as we will see in the next sections.

7.4 Regulation of gene transcription: auto-activation and auto-inhibition

Auto-activation and auto-inhibition

Auto-activation or auto-inhibition occurs when the promoter of the considered gene is regulated by the transcription factor (protein) that it encodes.



$$\dot{m} = k_1 f(p) - d_1 m \tag{61}$$
$$\dot{p} = k_2 m - d_2 p \tag{62}$$

where $f(p) = f^+(p) = \frac{p^n}{K^n+p^n}$ (monnotonically increasing Hill function) for an auto-activating action of the transcription factor p, and $f(p) = f^-(p) = 1 - f^+(p) = \frac{K^n}{K^n+p^n}$ (monotonically decreasing Hill function) for an auto-inhibiting action of the transcription factor p.

Remark 68. *The activating Hill function $f^+(p) = \frac{p^n}{K^n+p^n}$ has two parameters:*

- *K which is the* activation coefficient *(units of concentration). In this case, K is equal to the concentration of transcription factor p needed to activate by 50% the overall gene expression, i.e., $f^+(K) = \frac{1}{2}$.*
- *n which is the* Hill coefficient*. It represents the cooperativity of the considered reaction. In this case, n corresponds to the number of p that need to cooperatively bind the promoter to trigger the activation of gene expression.*

Similar parameter definitions apply for the repressing Hill function $f^-(p) = 1 - f^+(p)$.

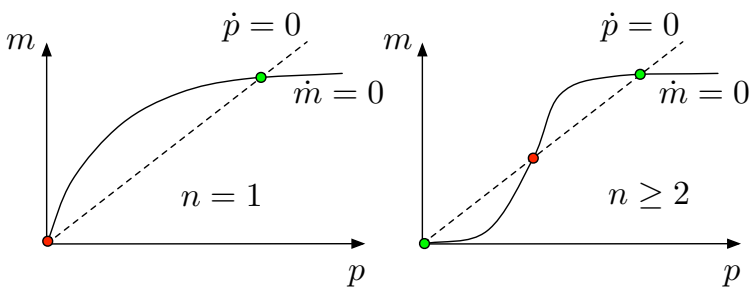
The model (61)-(62) is a nonlinear ODE model of order 2.

7.4.1 Auto-activation

In the auto-activation case, the model (61)-(62) writes:

$$\dot{m} = k_1 \frac{p^n}{K^n + p^n} - d_1 m \tag{63}$$
$$\dot{p} = k_2 m - d_2 p \tag{64}$$

We perform the analysis of the model (61)-(62) directly by looking at its phase plane.



The fixed points are located at the intersection of the two nullclines $\dot{m} = 0$ and $\dot{p} = 0$ (see Appendix A for the definition of a nullcline). The stability of the fixed points can be checked by (a) considering the direction of the vector field *on* each of the nullclines and *within* the regions delimited by the nullclines and (b) by considering the sign of the real part of the eigenvalues of the Jacobian evaluated at each fixed point (i.e., the linearisation of the system around each fixed point) (or if you prefer the τ-Δ approach, by considering the signs of the trace $\tau(= \lambda_1 + \lambda_2)$ and the determinant $\Delta(= \lambda_1\lambda_2)$ of the Jacobian).

Exercise 3. *Consider the ODEs in (63) and (64). Draw the flow (1) on the nullclines and (2) within each of the regions delimited by the nullclines. Note that on each nullcline the vector field can only be either vertical or horizontal. Within each of the regions delimited by the nullclines, the vector field can be deduced as a vectorial combination of the vector fields at the boundaries of these regions, i.e., on the nullclines (see also Appendix A).*

Can you infer the stability of the various fixed points from the vector field information that you have represented on the phase plane?

Exercise 4. *Perform a complete linear stability analysis around each fixed point for the model given in (63)-(64) (i.e., compute the eigenvalues of the Jacobian of the linearised system associated with each fixed point). Check that their local stability property agrees with the flow picture that you have drawn in the previous exercise.*

Remark 69. *To understand the link between the linearisation around a fixed point and the slopes of the nullclines intersecting at that fixed point, remember that linearising the nullclines around a fixed point corresponds to considering the slopes of the nullclines at the considered fixed point. For example, if we take the red fixed points in the phase planes, we see that the slope of the Hill nullcline ($\dot{m} = 0$) at these points (let's call it α) is larger than the slope of the linear nullcline ($\dot{p} = 0$) at these points (let's call it β). We have* $\alpha = \frac{\textit{linearisation of } k_1 \frac{p^n}{K^n+p^n} \textit{ at the FP}}{d_1} > \beta = \frac{d_2}{k_2}$. *We thus have* $\tau = \lambda_1 + \lambda_2 = -d_1 - d_2 < 0$ *while* $\Delta = \lambda_1\lambda_2 = d_1 d_2 - k_2 * \left(\textit{linearisation of } k_1 \frac{p^n}{p^n+K^n} \textit{ at the FP}\right) < 0$. *Therefore, the red fixed points are saddle points. A similar argument applied to the green fixed points shows that these are stable nodes.*

As we can see, if the cooperativity coefficient of the Hill activating function, i.e., n, is equal to 1, the model can have at most one stable fixed point[22]. On the contrary, if $n \geq 2$, then two stable fixed points can coexist (one corresponding to the gene being expressed and the other to the gene not being expressed) and the system is said to be *bistable*. Which fixed point is asymptotically reached depends on the initial condition of the system.

7.4.2 Auto-repression

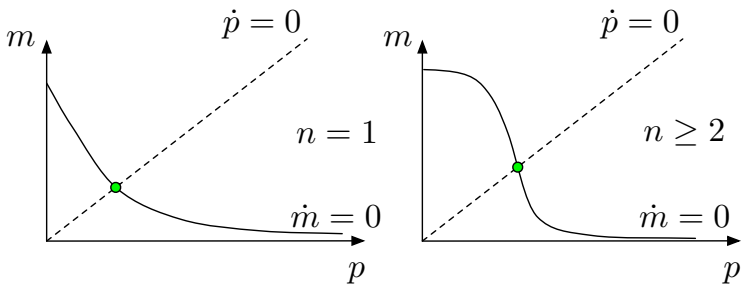
In the auto-repression case, the model (61)-(62) writes:

$$\dot{m} = k_1 \frac{K^n}{K^n + p^n} - d_1 m \tag{65}$$

$$\dot{p} = k_2 m - d_2 p \tag{66}$$

Once again, we perform the analysis of the model (61)-(62) directly by looking at its phase plane.

[22] The number of fixed points depends on the parameters of the model. These parameters determine the shape of the nullclines. For example, the shape of the nullcline $\dot{m} = 0$ is dictated by the parameters n, K, and k_1, while the slope of the nullcline $\dot{p} = 0$ is dependent on the ratio of parameters $\frac{d_2}{k_2}$.



As we can see, in this case there always exists a single stable fixed point, for any value of the cooperativity coefficient n.

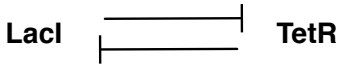
Exercise 5. *Consider the ODEs in* (65) *and* (66)*. Draw the flow (1)* on *the nullclines and (2) within each of the regions delimited by the nullclines. Can you infer the stability of the various fixed points from this information?*

Exercise 6. *Perform a complete linear stability analysis around each fixed point for the model given in* (65)-(66) *and check that their local stability property agrees with the flow picture that you have drawn in the previous exercise.*

7.5 Synthetic Biology gene regulation models

7.5.1 The toggle switch

The toggle switch is a synthetic biology construct composed of two genes which mutually repress each other. In the initial construct proposed by a Boston research group (Gardner, Cantor and Collins, Nature, 2000), one of the genes is *LacI* while the other one is *TetR*.



Similarly to what we have seen in the previous section, a simple ODE model for this gene regulatory network can be given as:

$$\begin{aligned}
\dot{m}_L &= k_{L,1}\frac{K_T^{n_T}}{K_T^{n_T}+p_T^{n_T}} - d_{L,1}m_L \\
\dot{p}_L &= k_{L,2}m_L - d_{L,2}p_L \\
\dot{m}_T &= k_{T,1}\frac{K_L^{n_L}}{K_L^{n_L}+p_L^{n_L}} - d_{T,1}m_T \\
\dot{p}_T &= k_{T,2}m_T - d_{T,2}p_T
\end{aligned}$$

where m_L (resp. m_T) is the concentration of *LacI* (resp. *TetR*) mRNA, and p_L (resp. p_T) is the concentration of *LacI* (resp. *TetR*) protein.

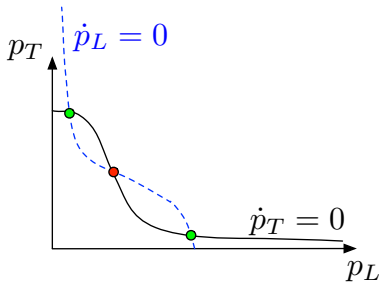
Using a quasi-stationary assumption for the mRNA dynamics, i.e., $\dot{m}_L \approx 0$ and $\dot{m}_T \approx 0$, we obtain a model of order 2 whose equations are:

$$\dot{p}_L = k_{L,2}\frac{k_{L,1}}{d_{L,1}}\frac{K_T^{n_T}}{K_T^{n_T}+p_T^{n_T}} - d_{L,2}p_L \tag{67}$$

$$\dot{p}_T = k_{T,2}\frac{k_{T,1}}{d_{T,1}}\frac{K_L^{n_L}}{K_L^{n_L}+p_L^{n_L}} - d_{T,2}p_T \tag{68}$$

The number and stability of the fixed points can be analysed by performing a nullcline analysis since the system is two-dimensional. *For certain values of the parameters, the nullclines intersect in three points, which determine the steady states of this system.*

For example, for $n_L = n_T = 2$, $k_{L,1} = k_{T,1} = 10$, and all other parameters equal to 1 the phase plane looks like this:



Specifically, by setting $\dot{p}_L = 0$ and $\dot{p}_T = 0$, we obtain the nullclines shown on the above depicted phase-plane. The nullclines partition the plane into 6 regions. By determining the sign of $\dot{p}_L$ and $\dot{p}_T$ in each of these 6 regions, one can determine the direction towards which the vector field is pointing in each of these regions[23].

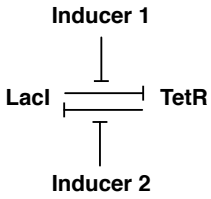
Exercise 7. *Perform a phase plane analysis of the* 2^{nd} *order toggle switch model given in* (67)-(68). *To this end, choose parameter values such that the toggle switch has 3 fixed points, and plot the vector field on the nullclines and within each of the* 6 *regions delimited by the nullclines. Can you infer the stability of the* 3 *fixed points based on this information?*

Exercise 8. *Assuming that the parameter values have been chosen so that the toggle switch has 3 fixed points, perform a local stability analysis around each fixed point. To this end, linearise the system around each fixed point and analyse the local stability of these fixed points. Check that your stability results are coherent with the ones you obtained through the phase plane analysis in the previous exercise.*

Let's consider the case for which the parameter values are chosen such that the toggle switch system has 3 fixed points. From the direction of the vector field on the nullclines, one can deduce that the middle steady state (i.e., the one for which $p_L^* = p_T^*$ if all the corresponding parameters of p_L and p_T are the same) is unstable. Performing a local stability analysis, one can check that the other two fixed points are stable. This is thus a bistable system, i.e., the system converges to one steady steady or the other depending on whether the initial condition is in the basin of attraction of one of the stable fixed point or the other (note that the stable eigenvector of the saddle point (in red on the phase plane) is a separatrix of the phase plane which delimits the basins of attraction of the other two stable fixed points). In one of the fixed points, only LacI is expressed at a high level while in the other, only TetR is expressed at a high level.

Once the system has converged to one of the two fixed points, it cannot switch to the other unless an external stimulation is applied that moves the initial condition into the basin of attraction of the other steady state. Typically this perturbation is applied by increasing the concentration of an inducer, i.e., a molecule that inhibits the action of a repressor (either Inducer 1 or Inducer 2 in the diagram given hereafter). Using the appropriate inducer, the system can be switched from one fixed point to the other.

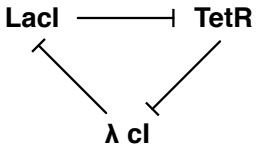
[23] The phase plane analysis can be confirmed easily by visualising the vector field on the plane. For this the matlab function `quiver` can be used. A Matlab tool that allows to visualise the vector field of a system of order 2 is PPLANE which can be found online at: https://uk.mathworks.com/matlabcentral/fileexchange/61636-pplane. You can visualise the phase plane of the toggle switch with the following default parameter values: $n_T = n_L = 2$, $k_{L,1} = k_{T,1} = 10$, and all other parameter values equal to 1. With these parameter values, optimal visualisation of the corresponding toggle switch vector field and nullclines in the phase plane can be obtained by considering p_L and p_T both in the range $[0, 10]$.



Note that a bistable system, when subject to noise, can give rise to noise-induced oscillations since the noise can act as a perturbation on the system which can push the system from the basin of attraction of one of the fixed points into the basin of attraction of the other. This noise-induced oscillations property of the toggle switch has been observed experimentally in the wet-lab.

7.5.2 The repressilator

The repressilator is a synthetic biology construct composed of three genes which mutually repress each other in sequence according to a ring structure. In the initial construct proposed by the research group of Elowitz (Elowitz and Leibler, Nature, 2000), the three chosen genes were *LacI*, *TetR*, and λ *cI*. The repressilator of Elowitz and Leibler exhibits sinusoidal, limit cycle oscillations with periods of hours.



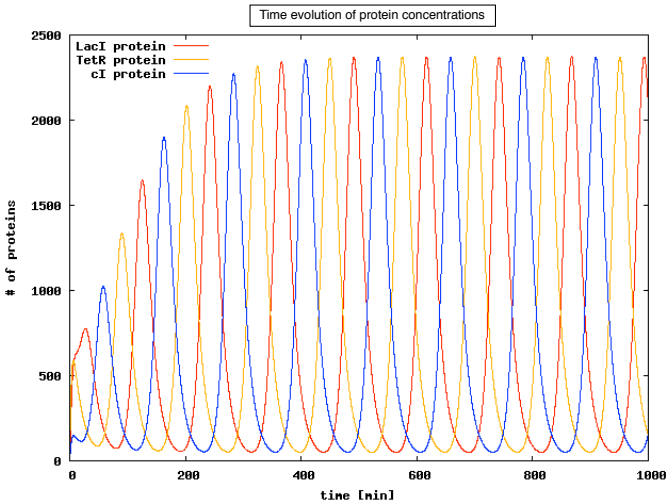
The corresponding model is of order 6 and can be written as (after non-dimensionalisation):

$$\dot{m}_i = -m_i + \frac{\alpha}{1+p_j^n} + \alpha_0 \tag{69}$$

$$\dot{p}_i = -\beta\,(p_i - m_i) \tag{70}$$

where $(i,j) = \{(LacI, cI), (TetR, LacI), (cI, TetR)\}$.

For certain values of the parameters, this system exhibits limit cycle oscillations as can be seen on the following simulation (from the BioModels website: http://www.ebi.ac.uk/biomodels-main/BIOMD0000000012).



Remark 70. *Note that when the order of the model becomes larger than* 2*, its mathematical analysis may become quite difficult. In such situations, numerical integration of the ODE model (computer simulations) for various initial conditions together with bifurcation analysis (for example using the Matlab toolbox* Matcont*) may become our first tools of choice for the analysis of high dimensional models in order to gain some initial understanding of the underlying dynamics.*

Remark 71. *The repressilator belongs to the class of* cyclic feedback systems *for which results have been proven mathematically, even when the number of genes in the ring is arbitrarily large*[24]*. In particular, the Mallet-Paret and Smith Theorem and the Hastings Theorem can be applied to cyclic feedback systems such as the repressilator to infer that if the system has a unique fixed point which is unstable, then it admits a periodic solution.*

7.6 Concluding remarks

So far, we have considered gene regulatory networks modelled using ODEs. This type of models implicitly assumes that the underlying quantities, i.e., concentrations or molecule numbers, vary in a continuous (i.e., real-valued) and deterministic fashion. However, at the molecular level these assumptions are mere approximations which might not accurately reflect the underlying dynamics, especially when the number of molecules involved is "small", such as is the case for transcription factors, which, in certain circumstances, can be expressed at low levels, i.e., a few tens of molecules, or for chromosomal DNA, for which a single copy exists in the cell. Intrinsic stochasticity at the molecular level may not be neglected any more when small numbers of molecules are involved (since in this case the "averaging" out of stochastic effects due to the application of the law of large numbers does not hold any more). Therefore, stochastic models have to be considered. For gene regulation stochastic models, the dependent variable $\boldsymbol{x}$ is a random variable which represents the number of molecules of the considered species at time instant t. The model then expresses the dynamics of the joint probability $p(\boldsymbol{x}, t)$ of having x_1 number of molecules of the first species at time t, x_2 number of molecules of the second species at time t, etc. (this type of model is called the *Master Equation* of the system[25]). The analysis of such stochastic models is then realised by mathematically deriving the most important moments (e.g., mean and variance of $\boldsymbol{x}$) if possible and also through *stochastic simulation algorithms* (SSA) such as the *Gillespie* stochastic integration algorithm. These stochastic simulations are typically more realistic but they also require more computer power to run and are more difficult to analyse. We will not cover stochastic gene regulation models in this course.

[24] A good paper to read for this is by Hal Smith, "*Oscillations and multiple steady states in a cyclic gene model*", J. Math. Biol. (1985), 25: 169-190.

[25] In physics a master equation is a set of ODEs describing the time evolution of the probability of a system to occupy each one of a discrete set of states.

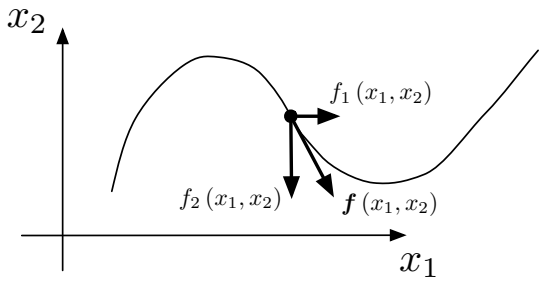
A Phase plane for ODE models of order 2

Phase plane for ODE models of order 2

An ODE model of order 2 can be written under the general form

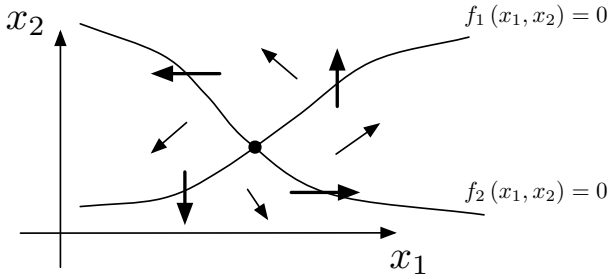
$$\dot{x}_1 = f_1(x_1, x_2)$$
$$\dot{x}_2 = f_2(x_1, x_2)$$

A trajectory in its phase plane looks like this:



At each point (x_1, x_2) in the phase plane, the vector $\boldsymbol{f}(x_1, x_2) = \begin{pmatrix} f_1(x_1, x_2) \\ f_2(x_1, x_2) \end{pmatrix}$ determines the direction in which the trajectory evolves and is thus tangent to this trajectory at the considered point (x_1, x_2).[26]

The curves $f_1(x_1, x_2) = 0$ and $f_2(x_1, x_2) = 0$ are called *nullclines*. Along these curves, one of the component of the vector field is zero. For ODE models of order 2, this means that on a nullcline, the vector field (or flow) can only be either horizontal or vertical (depending on which component of the vector field is zero on the considered nullcline). Fixed points are located at the intersection of the nullclines.



The nullclines delineate regions in the phase plane within which the vector field points towards one of the four directions which can be deduced from the vectorial composition of the vector field directions at the boundaries of the considered region:

- $\dot{x}_1 > 0$ and $\dot{x}_2 > 0$ (North East direction)
- $\dot{x}_1 > 0$ and $\dot{x}_2 < 0$ (South East direction)
- $\dot{x}_1 < 0$ and $\dot{x}_2 > 0$ (North West direction)
- $\dot{x}_1 < 0$ and $\dot{x}_2 < 0$ (South West direction)

[26] An amazing vector field visualisation tool to show how the weather changes is available at https://earth.nullschool.net: the animated weather map shows for each point on earth how weather currents change in real time. This is a direct illustration of a phase plane of a dynamical system $\dot{\boldsymbol{x}} = f(\boldsymbol{x})$ used to predict the weather on earth surface.

The local stability analysis around a fixed point gives you a local picture of the vector field around this fixed point (see section 5.4.1). Once you have a local picture around all the fixed points, the global behaviour of a system of order 2 can be obtained by remembering that for models of the form $\dot{\boldsymbol{x}} = \boldsymbol{f}(\boldsymbol{x})$ trajectories do not cross.

B Gene transcription regulation by activators

In this section, we show how the dynamics (57) can be obtained from the following biochemical reactions:

$$nA + D \underset{\beta}{\overset{\alpha}{\rightleftharpoons}} C \ (+\text{mRNAPol}) \xrightarrow{\tilde{k}_1} m + D + nA \ (+\text{mRNAPol})$$
$$m \xrightarrow{d_1} \emptyset$$

where $A = Activator$, $D = DNA$, $C =$ DNA-activators complex, and $m = mRNA$. Using the law of mass action to write the ODEs, we obtain:

$$\dot{m} = \tilde{k}_1 C - d_1 m$$
$$\dot{C} = \alpha A^n D - (\beta + \tilde{k}_1) C$$

Now, using the following two assumptions:

- the quasi-stationary approximation $\dot{C} \approx 0$, and
- the conservation law $D + C = D_{total}$ where D_{total} is a positive constant (expressing that the total amount of DNA is conserved),

we obtain:

- $\dot{C} \approx 0 \Rightarrow (\beta + \tilde{k}_1) C = \alpha A^n D$
- $D + C = D_{total} \Rightarrow (\beta + \tilde{k}_1) C = \alpha A^n \left(D_{total} - C\right)$

This last equation yields:

$$C = \frac{\alpha D_{total} A^n}{\beta + \tilde{k}_1 + \alpha A^n}$$

which, when inserted in the equation of $\dot{m}$, gives:

$$\dot{m} = \tilde{k}_1 D_{total} \frac{A^n}{\frac{\beta + \tilde{k}_1}{\alpha} + A^n} - d_1 m = k_1 \frac{A^n}{K^n + A^n} - d_1 m$$

with $k_1 = \tilde{k}_1 D_{total}$ and $K^n = \frac{\beta + \tilde{k}_1}{\alpha}$.

C Gene transcription regulation by repressors

In this section, we show how the dynamics (59) can be obtained from the following biochemical reactions:

$$nR + D \underset{\beta}{\overset{\alpha}{\rightleftharpoons}} C$$
$$D \ (+\text{mRNAPol}) \xrightarrow{\tilde{k}_1} m + D \ (+\text{mRNAPol})$$
$$m \xrightarrow{d_1} \emptyset$$

where $R = Repressor$, $D = DNA$, $C =$ DNA-repressors complex, and $m = mRNA$. Using the law of mass action to write the ODEs, we obtain:

$$\dot{m} = \tilde{k}_1 D - d_1 m$$
$$\dot{C} = \alpha R^n D - \beta C$$

Now, using the following two assumptions:

- the quasi-stationary approximation $\dot{C} \approx 0$, and
- the conservation law $D + C = D_{total}$ where D_{total} is a positive constant (expressing that the total amount of DNA is conserved),

we obtain:

- $\dot{C} \approx 0 \Rightarrow \alpha R^n D = \beta C$
- $D + C = D_{total} \Rightarrow \alpha R^n D = \beta \left(D_{total} - D \right)$

This last equation yields:

$$D = \frac{\beta D_{total}}{\beta + \alpha R^n}$$

which, when inserted in the equation of $\dot{m}$, gives:

$$\dot{m} = \tilde{k}_1 D_{total} \frac{\frac{\beta}{\alpha}}{\frac{\beta}{\alpha} + R^n} - d_1 m = k_1 \frac{K^n}{K^n + R^n} - d_1 m$$

with $k_1 = \tilde{k}_1 D_{total}$ and $K^n = \frac{\beta}{\alpha}$.