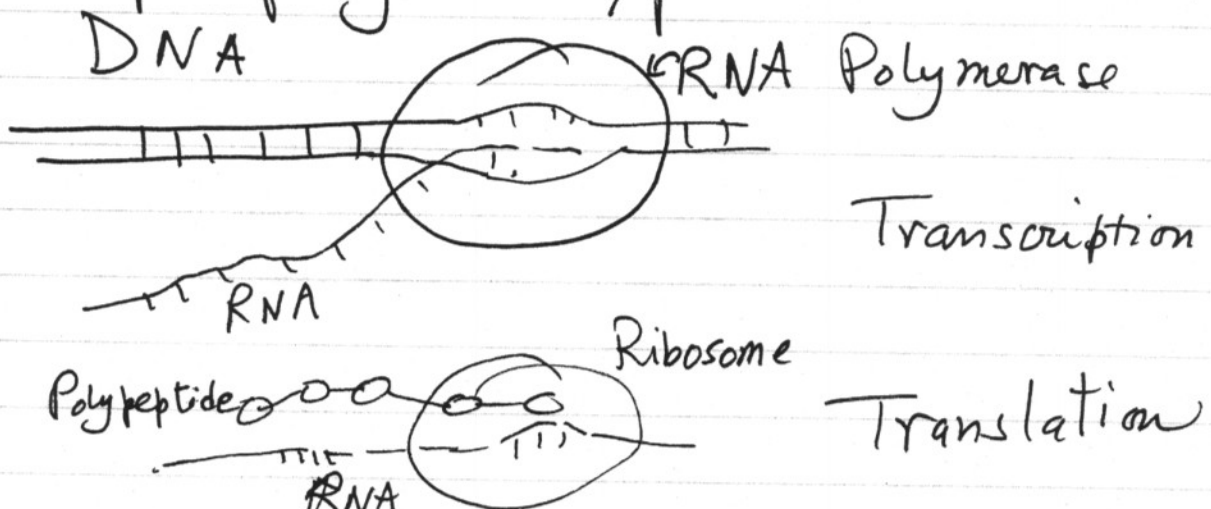10th July, 2007

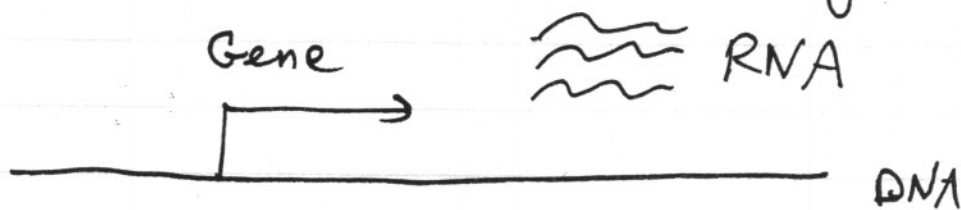# Nonlinear Dynamics for Biochemical Circuits

Anirvan Sengupta

Some elementary concepts for understanding the dynamics of synthetic or natural biological circuits.

We will pretend that we are constructing artificial genetic circuits with specific dynamical properties. Then, we try to analyze the system mathematically to see whether it would function appropriately or not. As we go on, we will introduce various concepts that will be useful in the analysis. You will also learn something about gene expression and transcriptional control in this way.

Steps of gene expression

DNA

RNA Polymerase

Transcription

RNA

Ribosome

Polypeptide

tRNA

Translation

Let us start with the first process

Gene → ≈≈≈ RNA

——————⌐———————————→————————— DNA

We will forget all the complexity and try to model the process of transcription by a rate of RNA production.

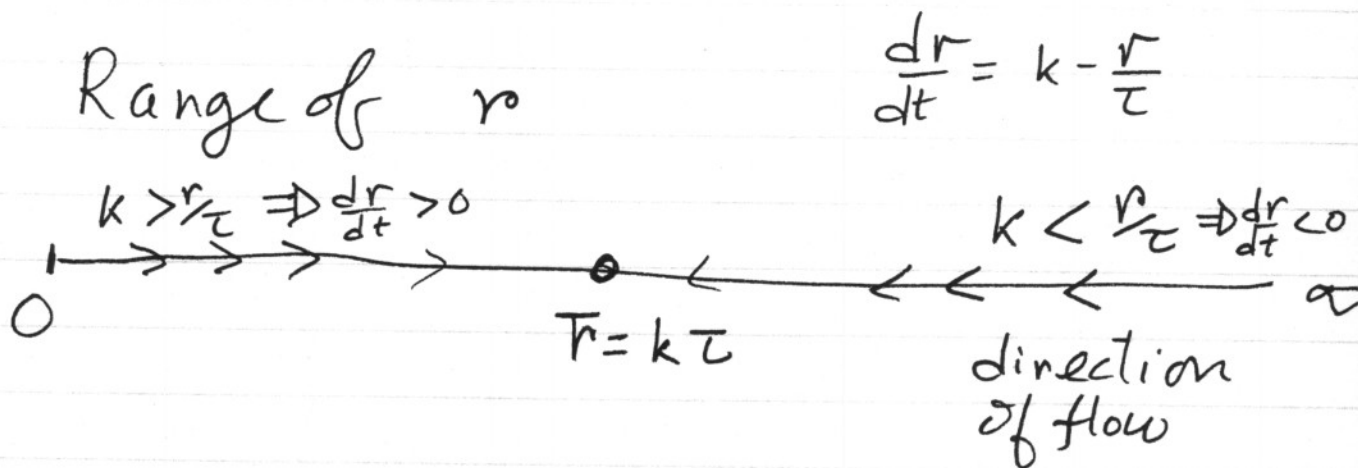$$\frac{d\,r}{dt} = k + \left(\blacksquare\ \text{effects of other processes}\right)$$

RNA amount

↑ Creation rate

Cellular RNA is often degraded quite fast (in minutes in some cases). We could model this by having RNA destruction rate proportional to the amount r.

$$\frac{dr}{dt} = k - \frac{r}{\tau}$$

Now, this is a linear inhomogeneous differential equation that you could solve easily. However, let us see whether we could figure out its long time behavior

without explicitly solving it, but by using simple geometric pictures.

Range of r

$$\frac{dr}{dt} = k - \frac{r}{\tau}$$

$k > \frac{r}{\tau} \Rightarrow \frac{dr}{dt} > 0$ 　　　　　　　　　$k < \frac{r}{\tau} \Rightarrow \frac{dr}{dt} < 0$

O 　　　　　　　$r = k\tau$ 　　　　　　direction
of flow

So, in the long run, whatever r we start with, we end up going toward $r = k\tau$. At that r, $\frac{dr}{dt} = 0$. This is called a <u>fixed point</u>. It is a <u>stable</u> fixed point since perturbations taking the system to any other point.

How does a perturbation die?

Let us call $\Delta r = r - k\tau$

$$\frac{d(\Delta r)}{dt} = \frac{dr}{dt} = k - \frac{r}{\tau} = -\frac{1}{\tau}(r - k\tau) = -\frac{1}{\tau}\Delta r$$

$$\Delta r(t) = \Delta r(0) \, e^{-t/\tau}$$

[Now I have ended up solving it despite promising otherwise]

. The crucial point to note is that in the neighborhood of the fixed point we have

$$\frac{d}{dt} \Delta r = \left(-\frac{1}{\tau}\right) \Delta r$$

↑
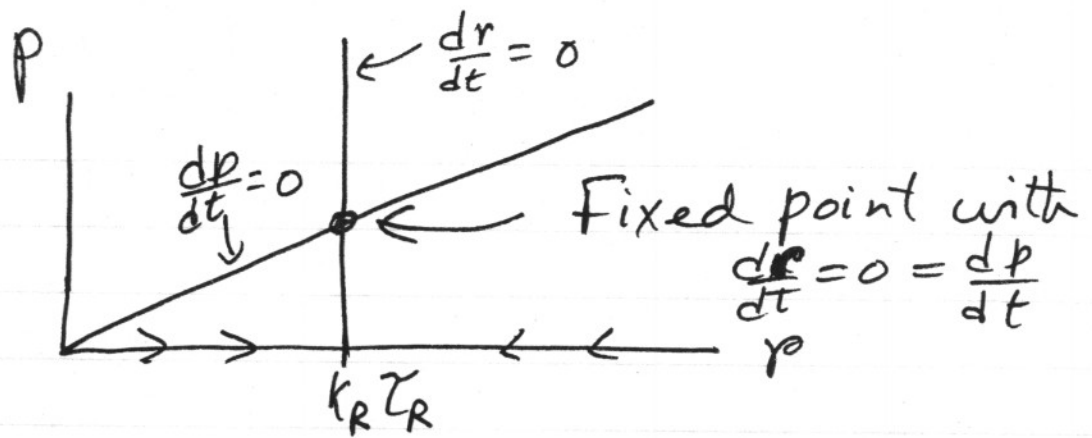negative ~~coefficient~~ coefficient

This goes hand in hand with stability.

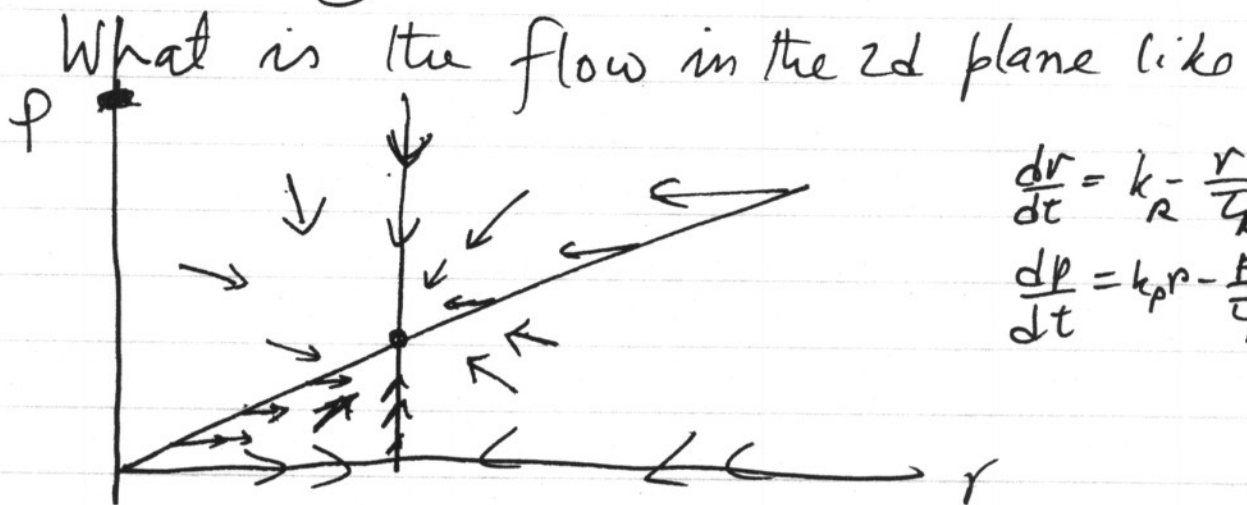Since we are into modeling gene expression, why not go up to the next stage : translation.

$$\frac{dr}{dt} = k_R - \frac{r}{\tau_R}$$

$$\frac{dp}{dt} = k_p \, r - \frac{p}{\tau_p}$$

(p is the amount of protein. Its production depends on r)

P $\frac{dr}{dt} = 0$

$\frac{dp}{dt} = 0$

Fixed point with $\frac{dr}{dt} = 0 = \frac{dp}{dt}$

$k_R \tau_R$ p

Curves on which $\frac{dx_i}{dt} = 0$ are known as nullclines. Since we have linear equations they are straight lines.

What is the flow in the 2d plane like

$\frac{dr}{dt} = k_R - \frac{r}{\tau_R}$

$\frac{dp}{dt} = k_p r - \frac{p}{\tau_p}$

On the $p = k_R \tau_R$ line, the flow of p is easy to figure out. On the p nullcline the flow is horizontal. In the four secants the flow direction could be found by interpolation.

Seems things are converging to the fixed point. How could we be sure?

The fixed point is given by

$$\bar{r} = k_R \tau_R \qquad \bar{p} = k_p \tau_p \bar{r} = k_R k_p \tau_R \tau_p$$

For Δr, $\Delta p \; (= p - \bar{p})$ the equations are

$$\frac{d}{dt} \Delta r = -\frac{t}{\tau_R} \Delta r$$

$$\frac{d}{dt} \Delta p = k_p \Delta r \quad -\frac{1}{\tau_p} \Delta p$$

$$\frac{d}{dt} \begin{pmatrix} \Delta r \\ \Delta p \end{pmatrix} = \begin{pmatrix} -\frac{t}{\tau_R} & 0 \\ k_p & -\frac{1}{\tau_p} \end{pmatrix} \begin{pmatrix} \Delta r \\ \Delta p \end{pmatrix}$$

$$= M \begin{pmatrix} \Delta r \\ \Delta p \end{pmatrix}$$

Perhaps we could find linear combinations so that there is no mixing. This is equivalent to diagonalizing the matrix M.

Call $\begin{pmatrix} \Delta r \\ \Delta p \end{pmatrix} = X$

$$\frac{dx}{dt} = Mx \qquad \text{If} \quad M = S^{-1}\Lambda S$$

Λ being diagonal
$$\begin{pmatrix} \lambda_1 & 0 \\ 0 & \lambda_2 \end{pmatrix}$$

$$\frac{dx}{dt} = S^{-1}\Lambda S x$$

$$\frac{d}{dt}(Sx) = \Lambda (Sx)$$

Call components of Sx u, v

$$Sx = \begin{pmatrix} u \\ v \end{pmatrix} \qquad u, v \text{ are linear combinations of } \Delta r, \Delta p$$

$$\frac{d}{dt}\begin{pmatrix} u \\ v \end{pmatrix} = \begin{pmatrix} \lambda_1 & 0 \\ 0 & \lambda_2 \end{pmatrix}\begin{pmatrix} u \\ v \end{pmatrix}$$

$$\Rightarrow \quad \frac{du}{dt} = \lambda_1 u \qquad \frac{dv}{dt} = \lambda_2 v$$

$$\Rightarrow u \sim e^{\lambda_1 t} \qquad \Rightarrow v \sim e^{\lambda_2 t}$$

Under what condition would $u, v \to 0$
(same as $\Delta r, \Delta p \to 0$) at $t \to \infty$?

Real part of λ_1, λ_2 are negative!

$\Rightarrow$ $\lambda_1 + \lambda_2$ is negative   and $\lambda_1 \lambda_2$ is positive!

Turns out it is also the sufficient condition.

In our particular case

$$\lambda_1 + \lambda_2 = \text{Trace} = -\left(\frac{1}{\tau_R} + \frac{1}{\tau_p}\right) \rightarrow \text{negative}$$

$$\lambda_1 \lambda_2 = \det = \left(-\frac{1}{\tau_R}\right)\left(-\frac{1}{\tau_p}\right) \rightarrow \text{positive}$$

Characteristic equation.

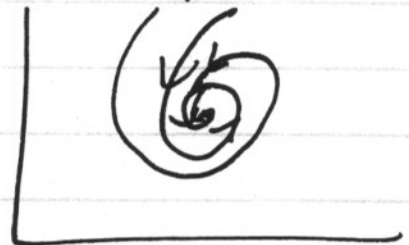$$f(\lambda) = \lambda^2 - (\text{Trace})\lambda + \text{Det}$$

or

⇓

Two negative roots

⇓

Complex Conjugate roots with negative real part.

In our problem $\lambda_1, \lambda_2 = -\frac{1}{\tau_1}, -\frac{1}{\tau_p}$

Question: What if one or both eigenvalues had a two real root?

You might be wondering when would anything non-linear showup in this lecture. We need to discuss the role of gene expression regulating proteins before we get there.

Previously we ~~XXX~~ dealt with constant RNA production rates. However, transcription rates are regulated, often by proteins binding to the "promoter". Such proteins are also called transcription factors.

Nothing bound

rate k_0

Multimer binding

$\xleftarrow{\hspace{1em}} m \xrightarrow{\hspace{1em}}$

rate k_m

The transcription rate

$$k(S) = k_0 \times \frac{1}{1 + \left(\frac{S}{K_D}\right)^m} + k_m \times \frac{\left(\frac{S}{K_D}\right)^m}{1 + \left(\frac{S}{K_D}\right)^m}$$
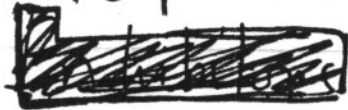
$k_0 < k_m$, S activator
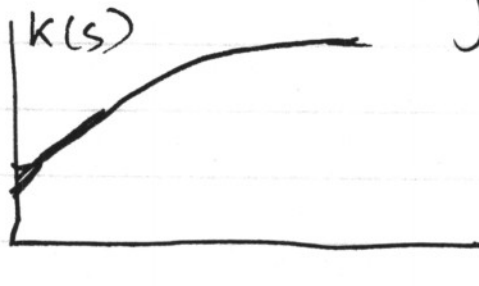
$k_0 > k_m$, S repressor

Activators

$k(s)$

k_m
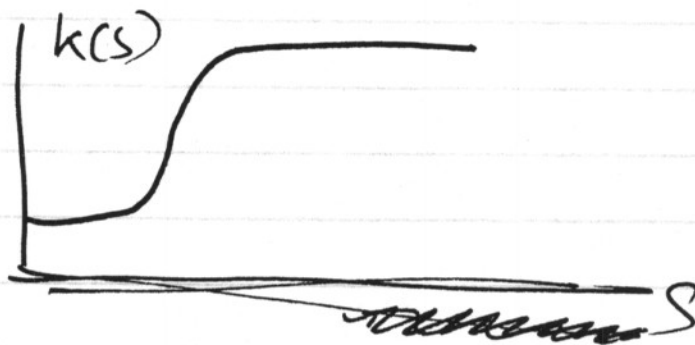
k_0

s

Repressors

k_0

k_m

s

m is the Hill coefficient.

Low m

$k(s)$

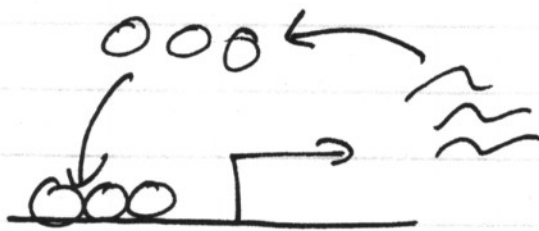s

High m

$k(s)$

s

$$\frac{dr}{dt} = k(s) - \frac{r}{\tau_R}$$

$$\frac{dp}{dt} = k_p r - \frac{p}{\tau_p}$$

If s changes with time, p changes in response.

The cell uses This to respond to changing conditions. The gene expression pattern alters in response to external cues.

However, we might be interested in making something more funky. What if the protein that the gene makes in itself a transcription factor? What if we make it hook up to its own promoter

This is a common theme in biology: feedback regulation. We consider two cases

Activator $\longrightarrow$ Positive Feedback
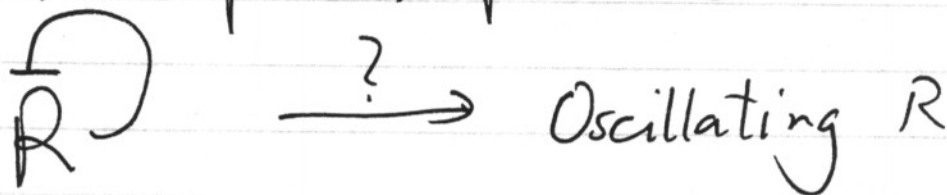
Repressor $\longrightarrow$ Negative Feedback.

Now, let us answer two questions.

1) With a self-activator, could we make a toggle switch?

2) ▬▬▬ With a self-repressor, could we get an oscillator?

Naively, both seems plausible. For the activator, having more of it helps produce more but having none of it produces very little.

$$\circlearrowleft A \xrightarrow{\ ?\ } \begin{array}{c} \text{High A state} \\ \text{Low A state.} \end{array}$$

Similarly, for the repressor, one might argue as follows. Having a high amount of it leads to shutdown of gene expression. Perhaps this could make the amount of protein drop significantly. Once that happens, the repression would be gone and more protein produced
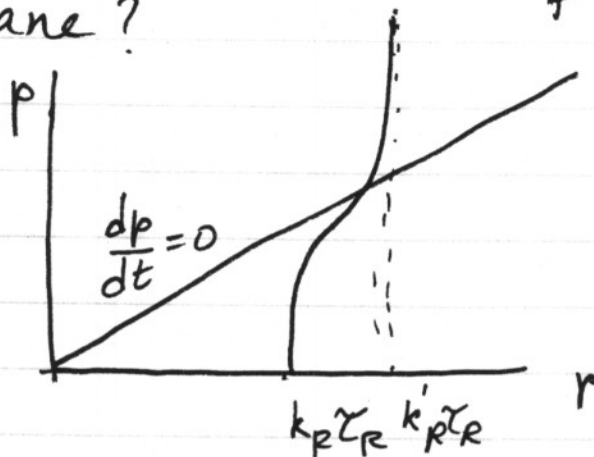
$$\overline{R} \xrightarrow{\ ?\ } \text{Oscillating } R$$

I) Positive feedback

~~████████~~ → Toggle Switch ?

$$\frac{dr}{dt} = \frac{k_R + k_R' \, p^m}{1 + p^m} - \frac{r}{\tau_R}$$
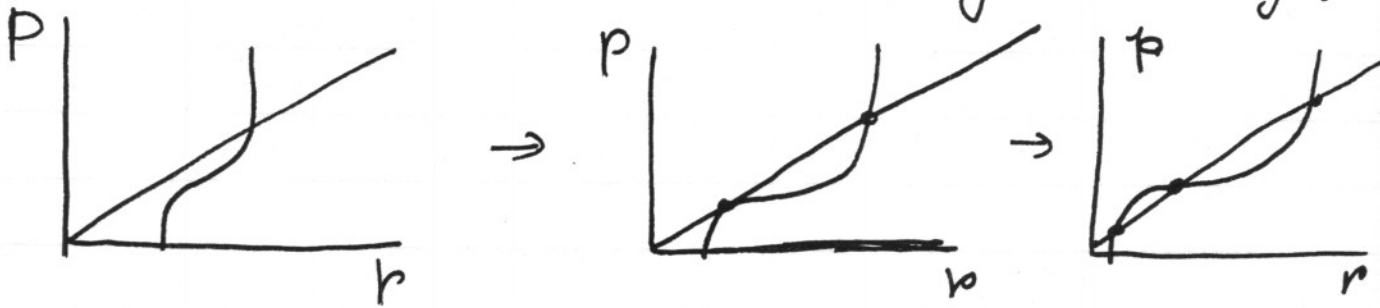
$$\frac{dp}{dt} = k_p r - \frac{p}{\tau_p}$$

$k_R' \gg k_R$. The variable p has been scaled so that the denominator is $1 + p^m$.

What about the nullclines in the r, p plane?

$\frac{dp}{dt} = 0$

$k_p \tau_R \quad k_p' \tau_R$

system
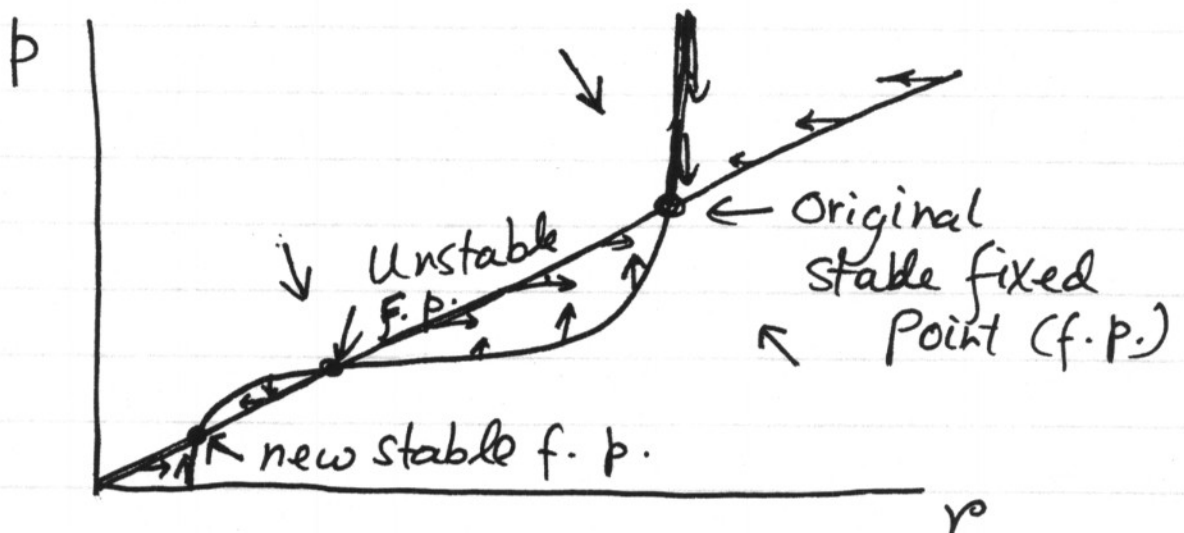
Could this have more than one stable fixed points? First we need more than one fixed points

What if we keep reducing k_R? Perhaps we get

This would be an example of a _saddle-node bifurcation_. Let us look closely at it.

Out of a clear blue sky, we suddenly get a ~~fixed point~~ appearing that splits into two fixed points. It will turn out that one of them is stable and another unstable.

← Original stable fixed point (f.p.)

Unstable F.P.

new stable f.p.

The bifurcation happens at the point where the nullclines touch.

[In general, bifurcations lead to qualitative change

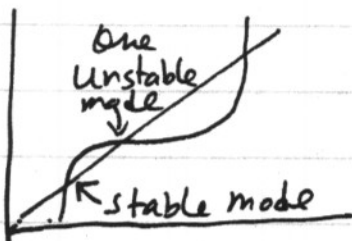Say they touch at r^*, p^*. Let us see what the dynamic close to the point is.

$$\Delta r = r - r^* \quad , \quad \Delta p = p - p^*$$

$$\frac{d}{dt} \Delta r = -\frac{1}{\tau_R} \Delta r + \frac{m(k_R' - k_R) p^{*\,m-1}}{(1 + p^{*\,m})^2} \Delta p$$

$$\frac{d}{dt} \Delta p = k_p \Delta r - \frac{1}{\tau_p} \Delta p$$

So $\quad \dfrac{d}{dt} \begin{pmatrix} \Delta r \\ \Delta p \end{pmatrix} = M \begin{pmatrix} \Delta r \\ \Delta p \end{pmatrix}$

If you look carefully, the condition for touching is also the condition of the determinant of M vanishing, or the condition of M having a zero eigenvalue.

One last comment. If we had Hill coefficient $m = 1$ could we get ~~this~~ this scenario? The answer is no!

We have the following equation for the fixed point.

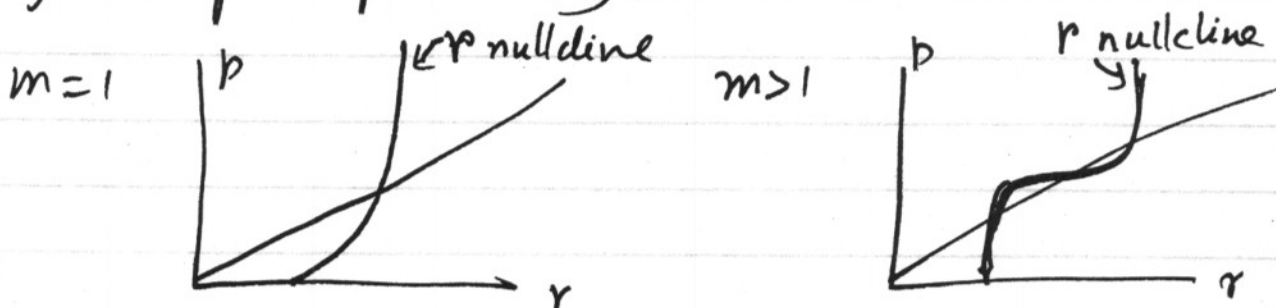$$\frac{k_R + k_R' \, p}{1 + p} - \frac{r}{\tau_R} = 0$$

and

$$k_p \, r - \frac{p}{\tau_p} = 0$$

Eliminating r

$$\frac{k_R + k_R' \, p}{1 + p} = \frac{p}{k_p \, \tau_p \, \tau_R}$$

This leads to a quadratic equation. which will not have the three solutions.

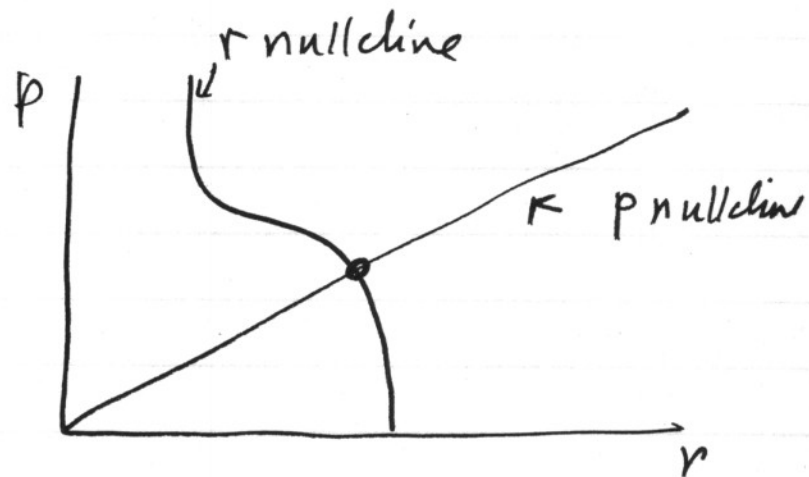So, we need $m > 1$ (in words, some degree of cooperativity)

2) Negative feedback $\longrightarrow$ Oscillation?

$$\frac{dr}{dt} = \frac{k_R + k_R' p^m}{1 + p^m} - \frac{r}{\tau_R}$$

$$\frac{dp}{dt} = k_p r - \frac{p}{\tau_p}$$

$k_R' < k_R$

p nullcline, r nullcline

Would this fixed point ever become unstable?

$$\frac{d}{dt}\begin{pmatrix} \Delta r \\ \Delta p \end{pmatrix} = M \begin{pmatrix} \Delta r \\ \Delta p \end{pmatrix}$$

negative

$$M = \begin{pmatrix} -\frac{1}{\tau_R} & -\frac{m(k_R - k_R')\bar{p}^{m-1}}{(1 + \bar{p}^m)^2} \\ k_p & -\frac{1}{\tau_p} \end{pmatrix}$$
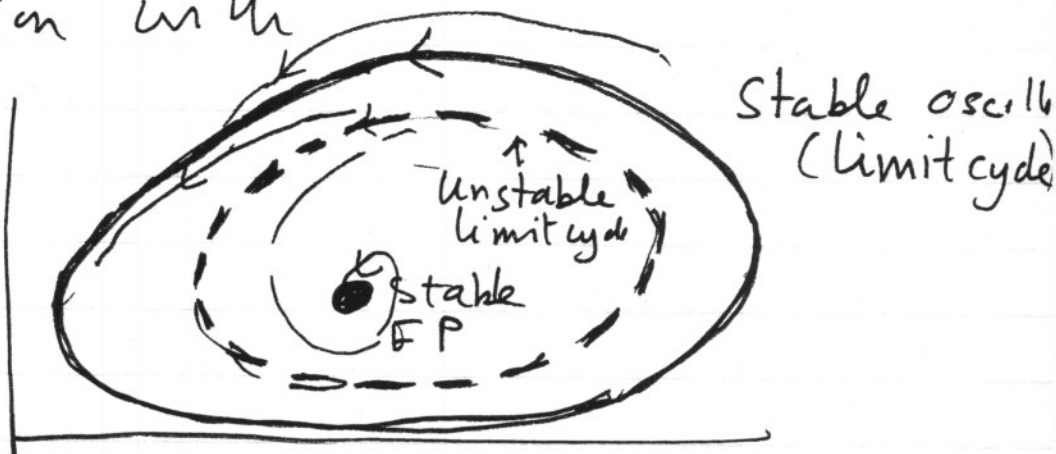
Note $\text{tr}(M) < 0$, $\det(M) > 0$

$\Rightarrow$ Always stable!

One could in principle have a

Situation with

Stable oscill.
(limit cycle)
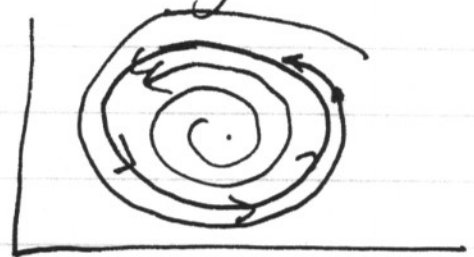
Unstable
limit cycle

stable
F P

However, this is not the case
in the current model.

The naive plausibility argument
does not work !

A few words about non linear
oscillators. Unlike linear oscillators
which can have any amplitude, non linear
oscillators give rise to limit cycles.
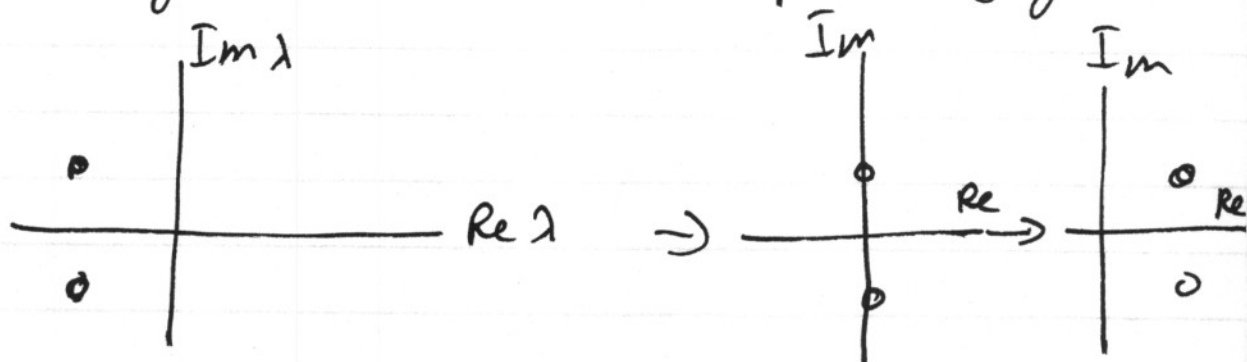

Linear Oscillator

Non linear
oscillator

Limit cycles are "isolated" close trajectories.
Isolated means other close by trajectories either
spiral away or ~~to~~ towards it.

One of the routes by which
stable limit cycles come into existence
is by having a stable fixed point
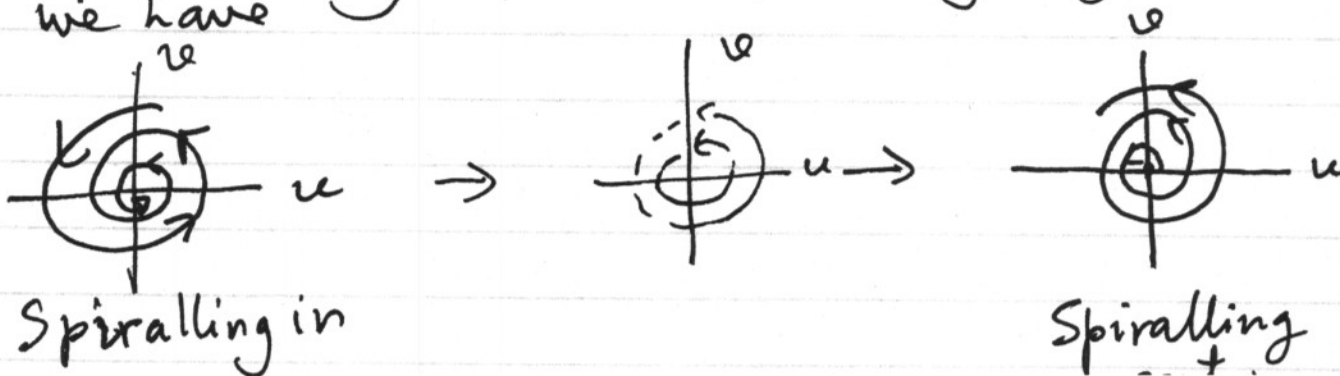going unstable. This is related to
Hopf bifurcation.

Suppose we have a system with
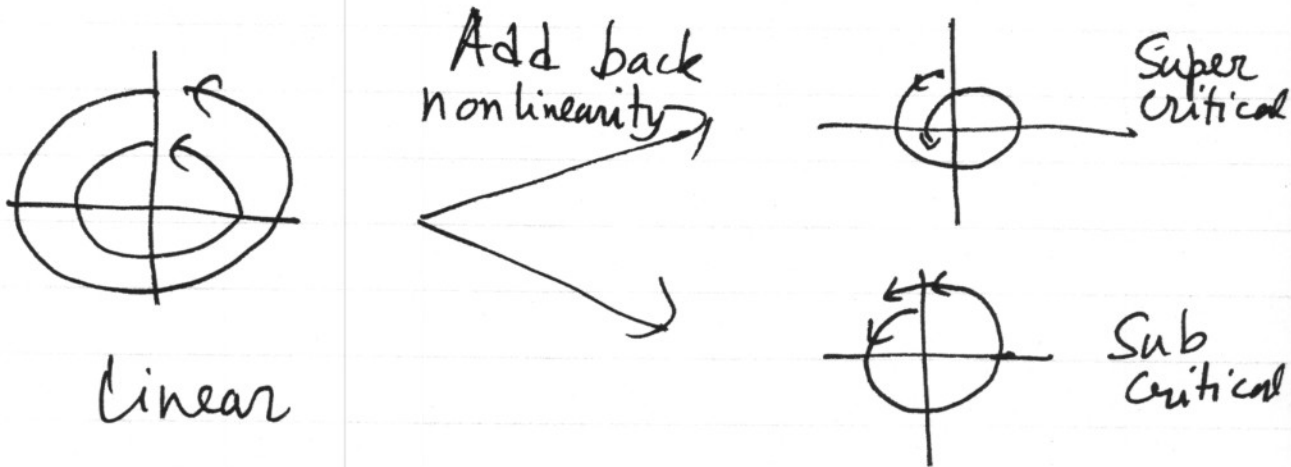a pair eigenvalues, which are complex conjugate.

Stable   ⟶ Marginal ⟶ Unstable

As we change some parameter. Assume
that the ▮ rest of the eigne values stay stable.
Hopf bifurcation happens when the eigenvalue is purely imaginary.
Around the fixed point, in the subspace
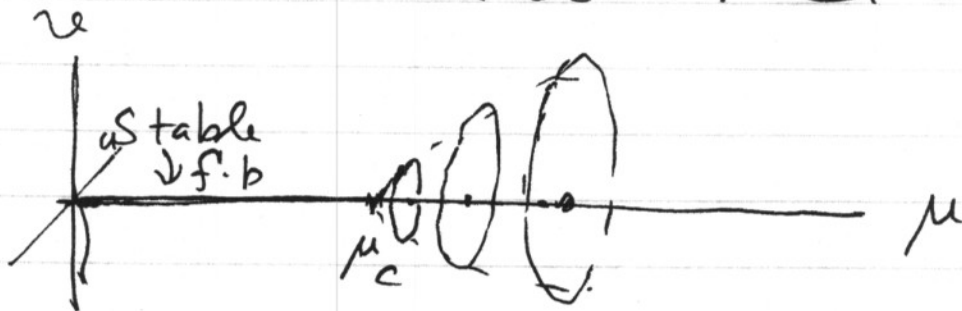spanned by the corresponding eigenvectors,
we have

Spiralling in   Spiralling

What does it spiral out to?
Answering that requires going be linear
analysis. At the bifurcation point
one might ask, what of is the effect
on non-linearities

Add back
non linearity

Super
critical

Sub
Critical

Linear

At super critical ▓▓▓ Hopf bifurcation
non linearity is shrinking the amplitude.

For these, when the fixed point goes
unstable, linear terms want amplitude
to grow and the non-linear term wants
is to shrink. The result is ▓▓▓ the formation

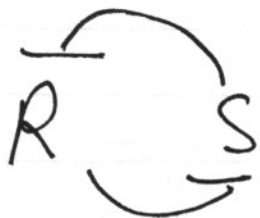unstable
v f.b

μ_c

μ

of a small limit ▓▓ cycle that grows

Could we make a circuit with repressors that would oscillate? We saw

does not work !

At most makes a bistable switch
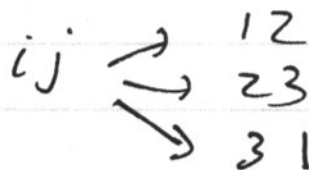[was made by the Collins group at BU]

However

does work for certain parameters.

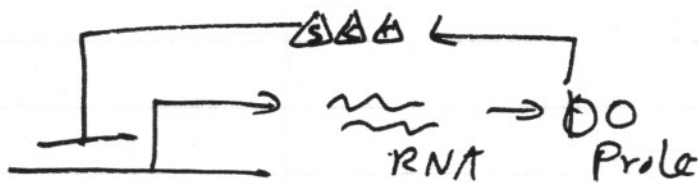This was made and analyzed by Elowitz and Leibler, the now famous repressilator.

 Home work: Compute Hopf bifurcation condition in a symmetric repressilator.

$$\frac{d r_i}{dt} = \frac{k_R}{1 + p_j^m} - \frac{r_i}{\tau_R}$$

$$\frac{d p_i}{dt} = k_0 r_i - \frac{p_i}{?}$$

$$ij \rightsquigarrow \begin{matrix} 1 2 \\ 2 3 \\ 3 1 \end{matrix}$$

A theorist's favorite that also oscillates is the Goodwin model

$$\frac{dr}{dt} = \frac{k_R}{1+s^m} - \frac{r}{\tau_R}$$

$$\frac{dp}{dt} = k_p r - \frac{p}{\tau_p}$$

$$\frac{ds}{dt} = k_s s - \frac{s}{\tau_s}$$

This oscillates for unphysically high m ($m > 8$). This limit cycle forms through a supercritical Hopf bifurcation, as the negative feedback (say k_R) cranks up.