A SPATIAL MODEL FOR p53 NUCLEAR ACCUMULATION

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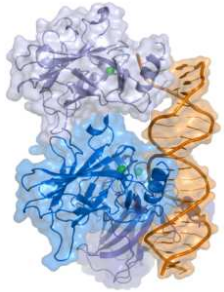
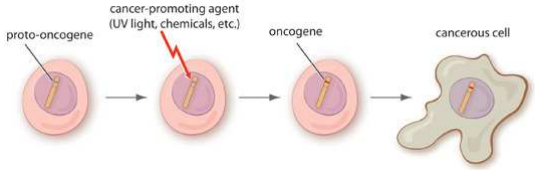
What is p53?

In 1979 a protein of molecular mass of 53 kDa was isolated. It was named **p53**.



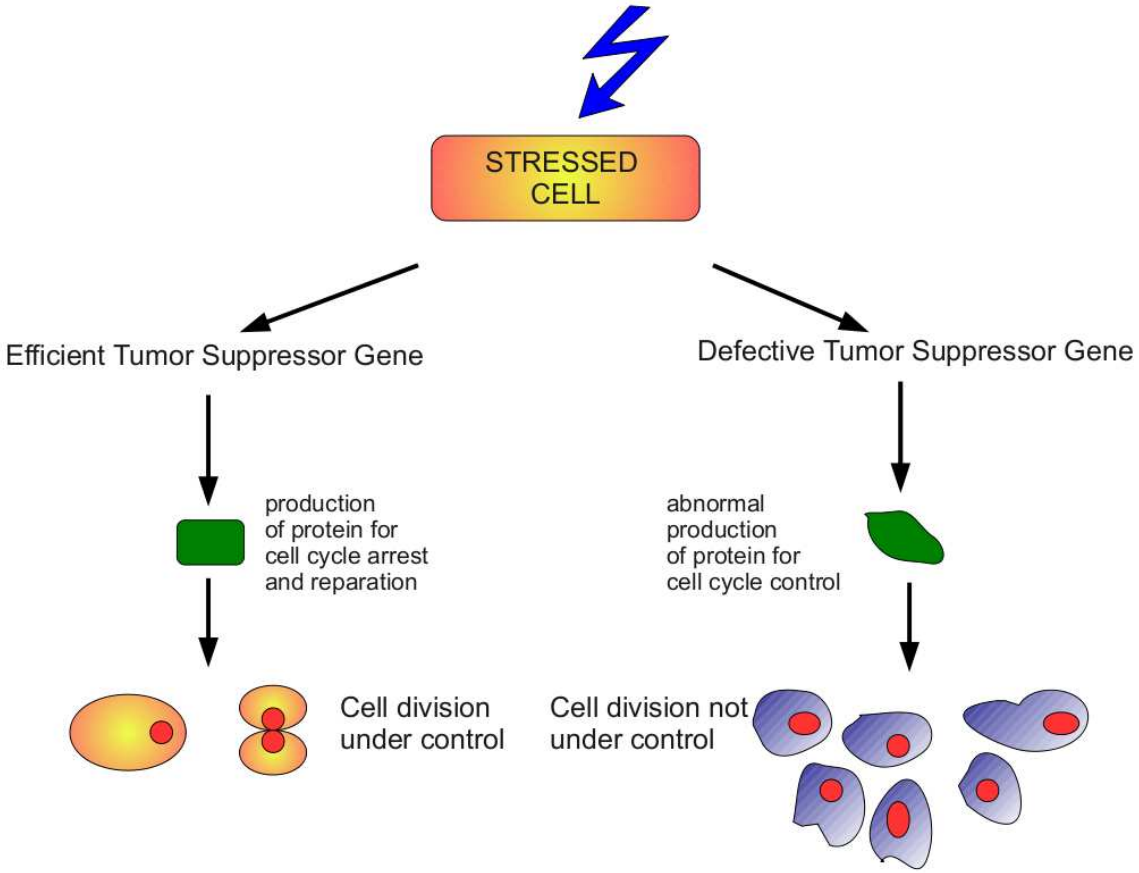
At first biologists believed that p53 was an oncogene, i.e. an **abnormal gene** that predisposes cells to develop into **cancers**.



10 years after they discovered that p53 is a **tumor suppressor** and so it acts to fix the cell or to trigger apoptosis.

What is p53?

The p53 became "popular" only in 1989 when it was realized that in more than **50% of cancers** the p53 gene is **altered**.

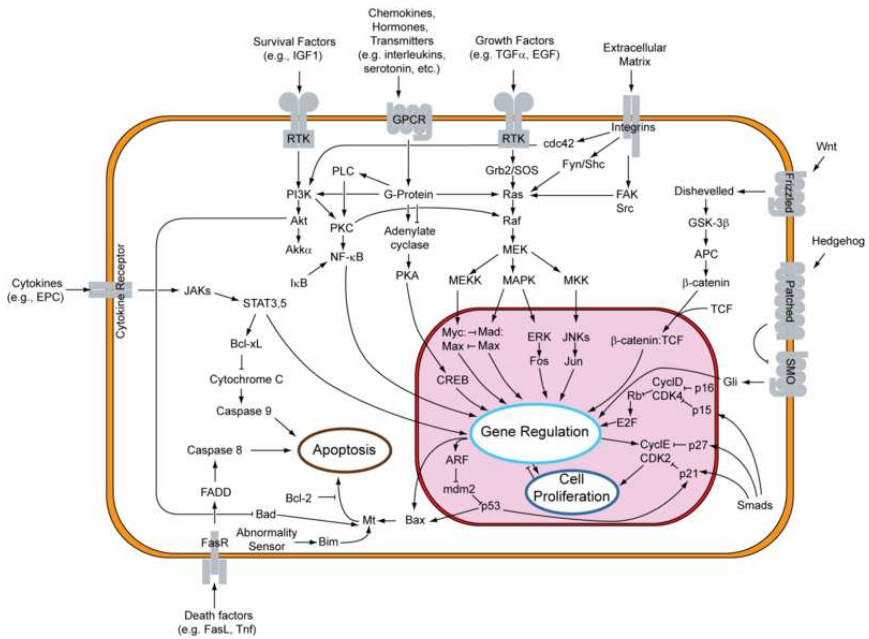


p53 roles: *the Guardian of the Genome*

AFTER A STRESS: p53 acts as a **transcription factor**: it binds to DNA and transcribes the proteins responsible of

- **blocking cell cycle** progress.
- **repair the DNA**.
- **launch apoptosis** (programmed cell death).

It has a **huge** network of interactions- hard to model!

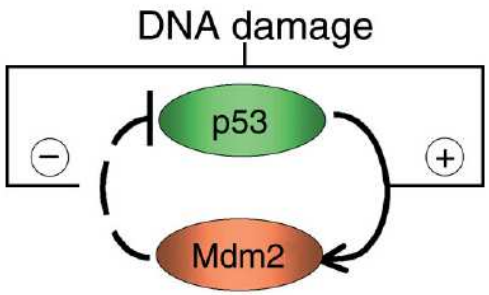


Healthy or Stressed cell

In HEALTHY CELL p53 is **dangerous** (deleterious effects on living organisms)!
Low concentration thanks to **Mdm2**

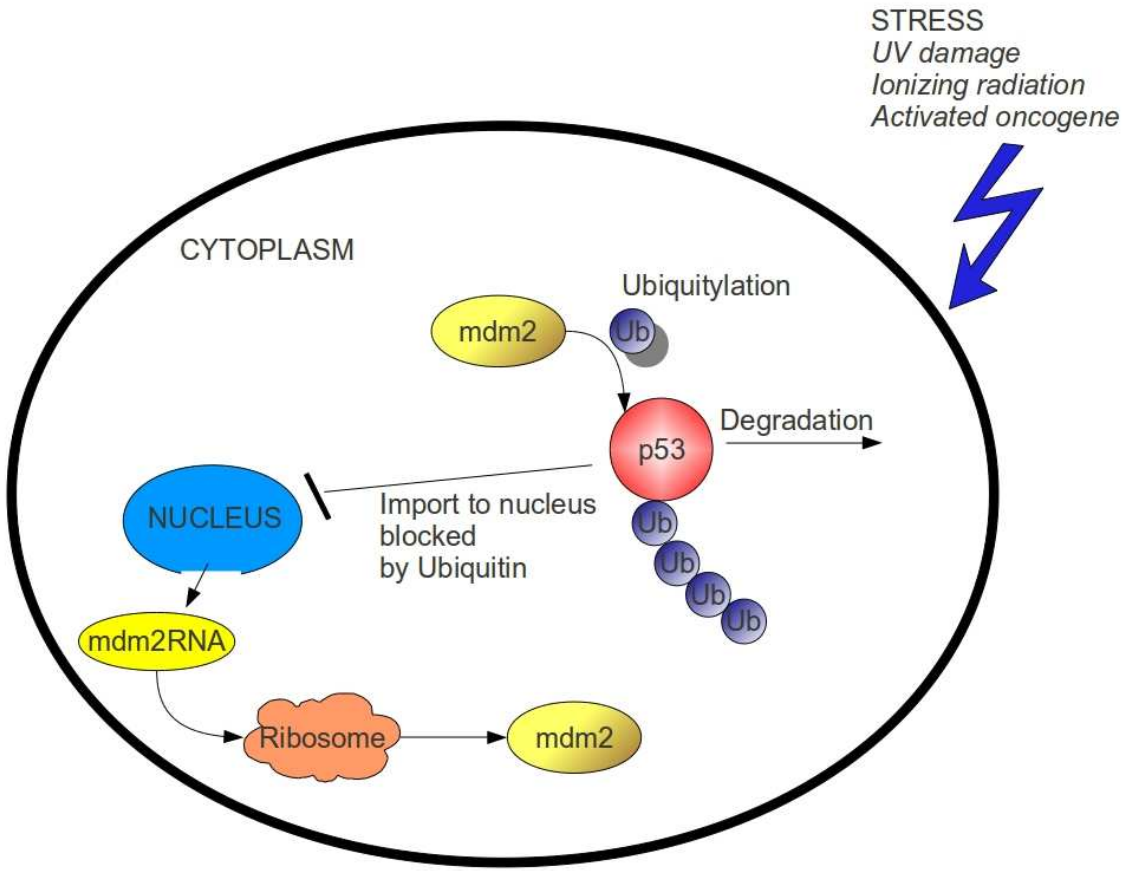
- Mdm2 induces **degradation** of p53 and **blocks** its nuclear import.
- p53 **induces Mdm2 synthesis** .

In STRESSED CELLS p53 concentration rises to **prevent** the transmission of harmful **mutations**.



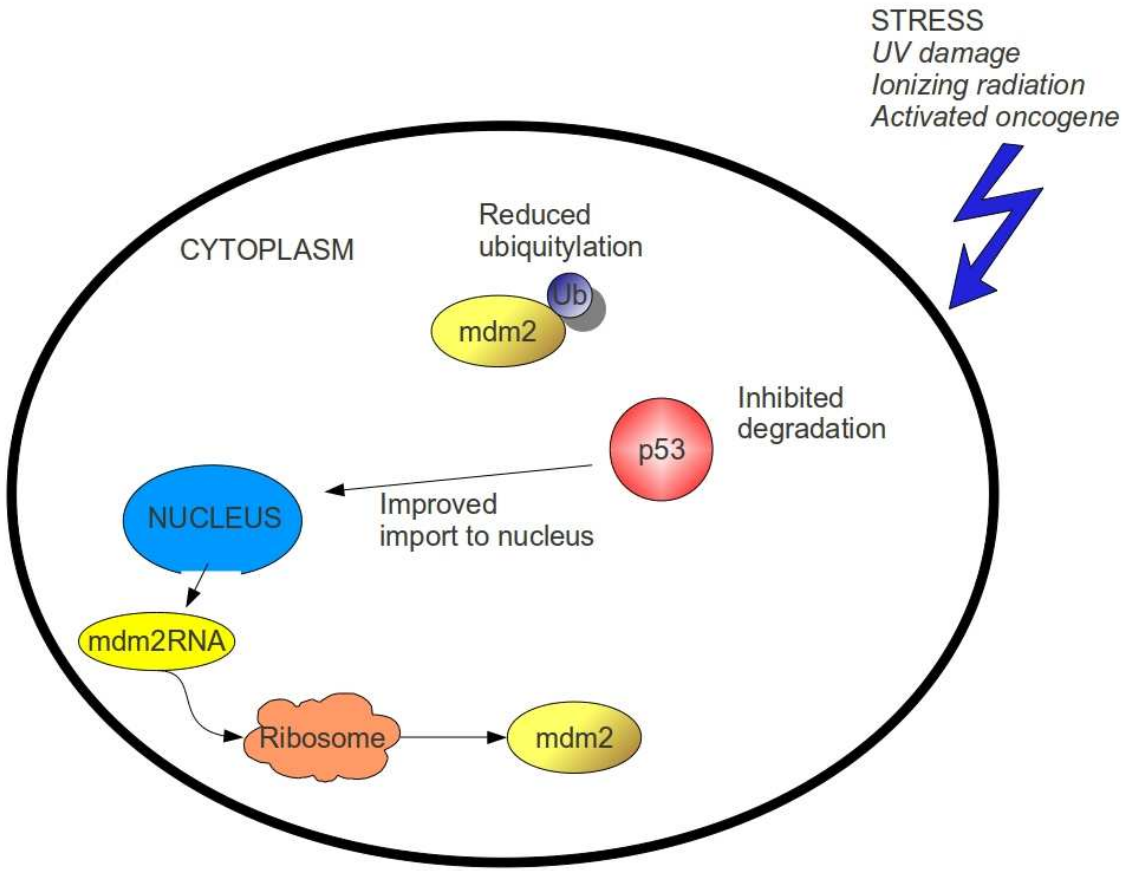
How to switch from a state to the other?

Healthy cells: **blocked import + increased degradation**



How to switch from a state to the other?

Stressed cells: modifications block p53-Mdm2 interactions.
Principal factor: **ATM**



p53 dynamics

the p53-Mdm2 network has an **oscillatory** behavior

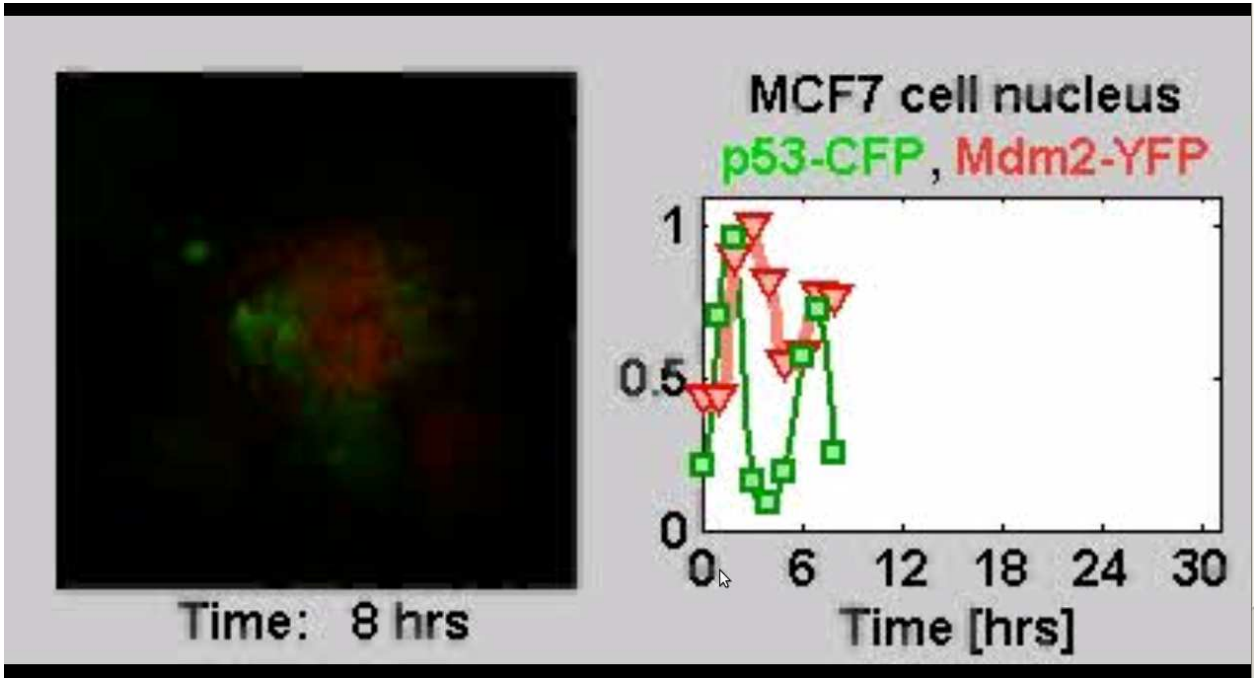


Figure: *in vitro* experiments

Modellers and p53

Why study p53?

- explain oscillations (which mechanism): **HOW**
- understanding its behaviour: **WHY**

Literature models

- **O**rdinary **D**ifferential **E**quations:
  mean cellular concentrations. Depend only on **time**
- Use **delay**: $\frac{du}{dt}(t) = f(t - \tau)$ or negative and positive **feedback**.

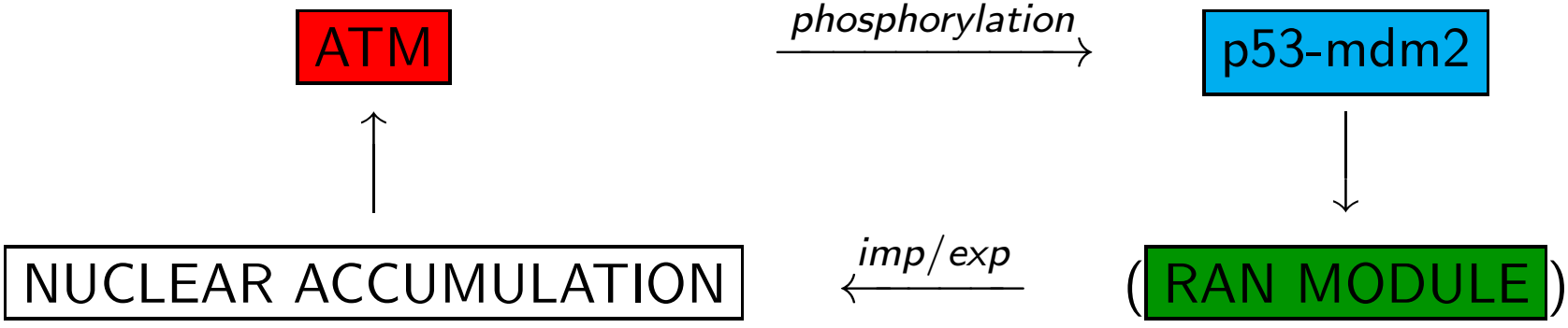
INTRODUCING SPACE

concentrations $u = (\mathbf{x}, t)$depends on **position**

Motivations

- "Operations" in Nucleus and Cytoplasm are not homogenuous (transduction-translation-degradation depends on compartement).
- Temporal dynamics: different space scales (p53's "radius" is 2,4 nm - diameter of a cell can be $30\mu m$)
- Spatial dynamics do not appear without space!

The model


ATM
phosphorylation
p53-mdm2
NUCLEAR ACCUMULATION
imp/exp
RAN MODULE

Variables and Equations

We define the variables "$p53$" and "$p53_p$" (p53 phosphorylated). (Michaelis Menten Kinetics) We use a Quasi-Steady-State-Approximation (QSSA) to represent this enzymatic action on p53.

$$[p53] + [ATM] \underset{k_{-2}}{\overset{k_2}{\rightleftharpoons}} [p53 - ATM] \xrightarrow{k_3} \quad [p53_p] + [ATM]$$

p53 must be:

- inactive (as a transcription factor)
- degraded in the cytoplasm,
- able to **move across the two compartments** (nucleus and cytoplasm)

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- degraded in the cytoplasm,
- able to **move across the two compartments** (nucleus and cytoplasm)

Phosphorylated p53 is supposed to be:

- **active** in the nucleus,
- able to be **imported to the cytoplasm**,
- **unable to be exported** from the nucleus,
- **non degradable**.

We introduce the species (and variable) Mdm2:

- the protein Mdm2 **shuttles** inside and outside of the nucleus.
- it is **degraded** in both compartments.
- it binds and **acts as a ubiquitin** ligase for p53.

We also model Mdm2 RNA, the transcription of which is activated by p53; p53 is active as a tetramer, hence we model its action as:

$$4[p53_p] + [DNA] \underset{k_-}{\overset{k}{\rightleftharpoons}} [4p53_p - DNA] \xrightarrow{k_t} [mdm2_{RNA}] + [DNA]$$

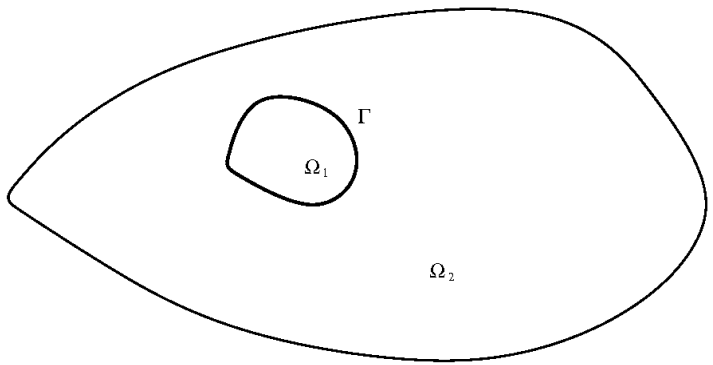
The Model: Nucleus

$$
\begin{cases}
\dfrac{\partial[p53]}{\partial t} = \overbrace{k_{ph}\dfrac{[p53_p]}{K_{ph}+[p53_p]}}^{\text{dephosphorylation}} + \overbrace{d_p\Delta[p53]^{(n)}}^{\text{diffusion}} - \overbrace{k_1[mdm2]\dfrac{[p53]}{K_1+[p53]}}^{\text{ubiquitination}} \\
\qquad\qquad - k_3 A \dfrac{[p53]}{K_{ATM}+[p53]} \\
\dfrac{\partial[mdm2]}{\partial t} = d_m\Delta[mdm2] - \delta_m[mdm2] \\
\dfrac{\partial[mdm2_{RNA}]}{\partial t} = k_{Sm} + \overbrace{k_{Sp}\dfrac{([p53_p])^4}{([p53_p])^4+K_{Sp}}}^{p53-\text{dependent synthesis}} + d_{mRNA}\Delta[mdm2_{RNA}] \\
\qquad\qquad - \delta_{mRNA}[mdm2_{RNA}] \\
\dfrac{\partial[p53_p]}{\partial t} = \overbrace{k_3 A^{(n)}\dfrac{[p53]}{K_{ATM}+[p53]}}^{\text{phosphorylation by ATM}} + d_{p'}\Delta[p53_p]^{(n)} - k_{ph}\dfrac{[p53_p]}{K_{ph}+[p53_p]}
\end{cases}
$$

The Model: Cytoplasm

$$
\begin{cases}
\dfrac{\partial [p53]}{\partial t} = \mathbb{1}_R k_S + k_{ph}\dfrac{[p53_p]}{K_{ph}+[p53_p]} + d_p \Delta[p53] - k_1[mdm2]\dfrac{[p53]}{K_1+[p53]} \\
\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad - k_3 A \dfrac{[p53]}{K_{ATM}+[p53]} \\
\dfrac{\partial [mdm2]}{\partial t} = d_m \Delta[mdm2] + \overbrace{\mathbb{1}_R[mdm2_{RNA}]}^{\textbf{translation}} - \delta_m[mdm2] \\
\dfrac{\partial [mdm2_{RNA}]}{\partial t} = d_{mRNA}\Delta[mdm2_{RNA}] - \mathbb{1}_R[mdm2]_{RNA} \\
\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad\qquad - \delta_{mRNA}[mdm2_{RNA}] \\
\dfrac{\partial [p53_p]}{\partial t} = k_3 A \dfrac{[p53]}{K_{ATM}+[p53]} + d_{p'}\Delta[p53_p] + k_3 A \dfrac{[p53]}{K_{ATM}+[p53]}
\end{cases}
$$

The Model: boundary conditions



we impose the continuity of the fluxes, not of concentrations: there is a **membrane**!

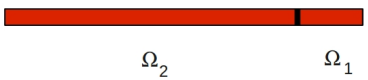
The Spatial Environment(s!)

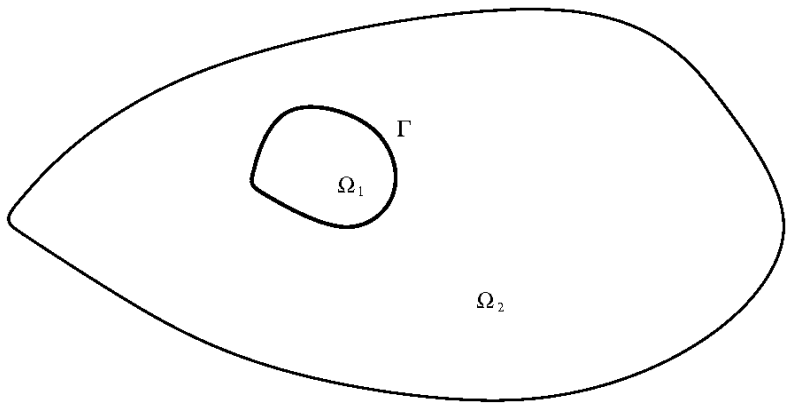
The spatial environment is the CELL! We represent it by:

- either a compartmental model (ODE system version)

$$\boxed{\text{NUCLEUS}} \longleftrightarrow \boxed{\text{CYTOPLASM}}$$

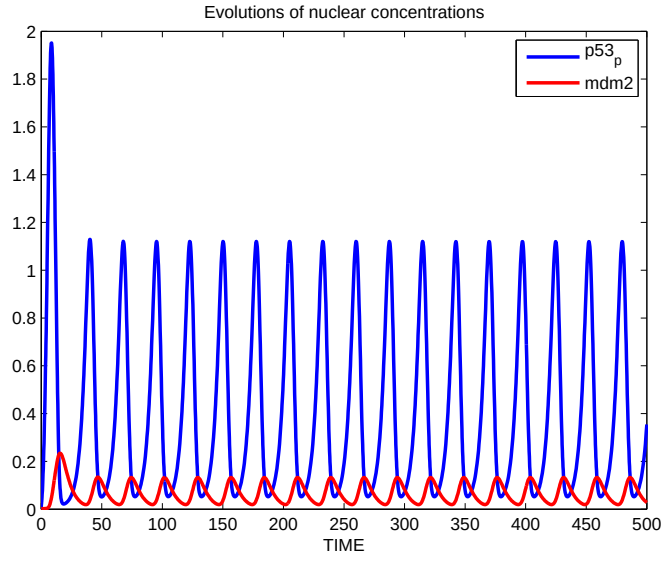
- or a spatial model (PDE system version): 1D and 2D domains

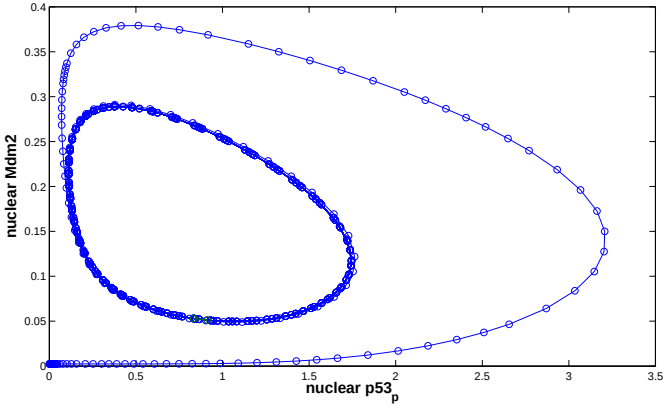


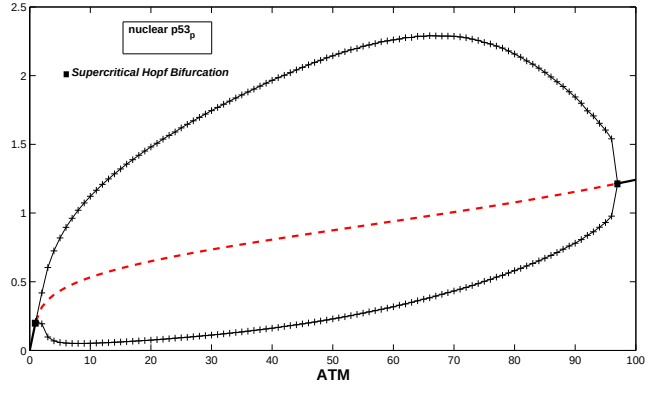


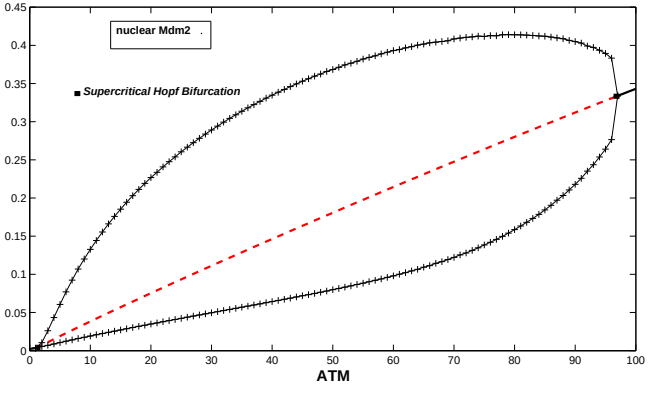
Where Ω_1 =Nucleus, Ω_2 =Cytoplasm and Γ the common boundary, $\Gamma = \Omega_1 \cap \Omega_2$.

First Simulations as ODE system









Simulations in a 1D PDE system

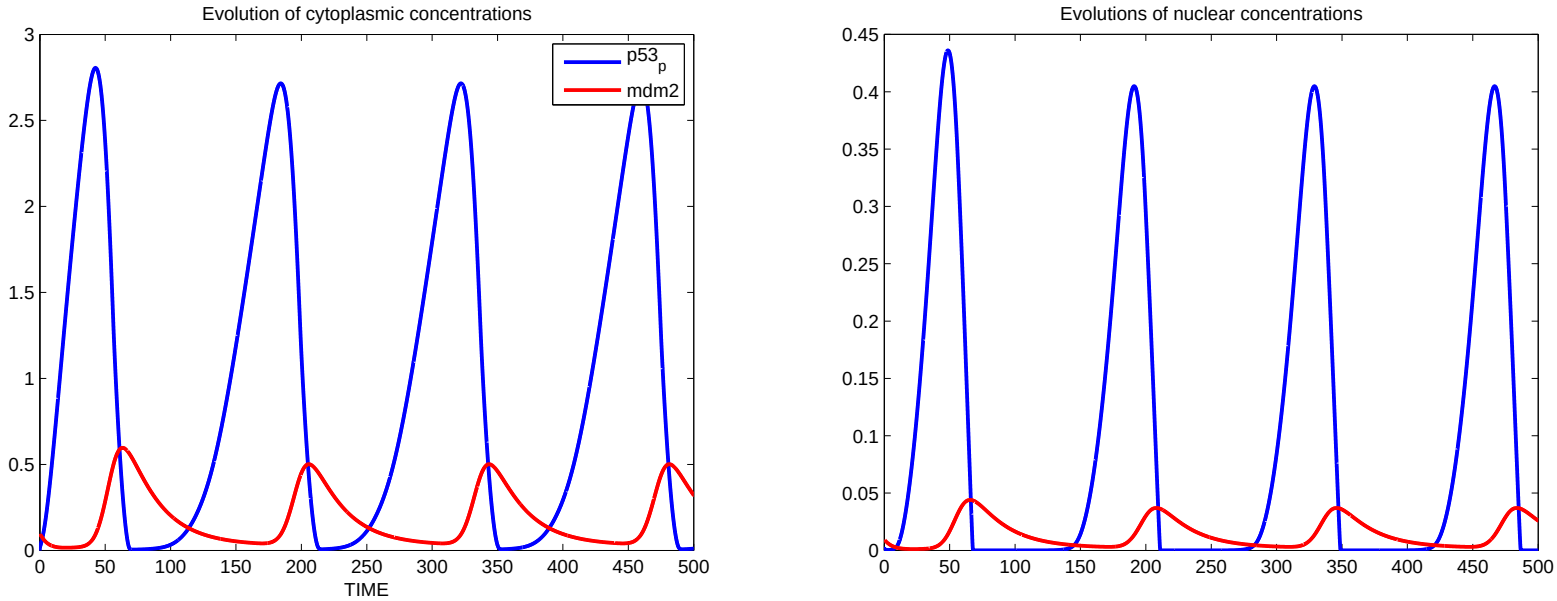
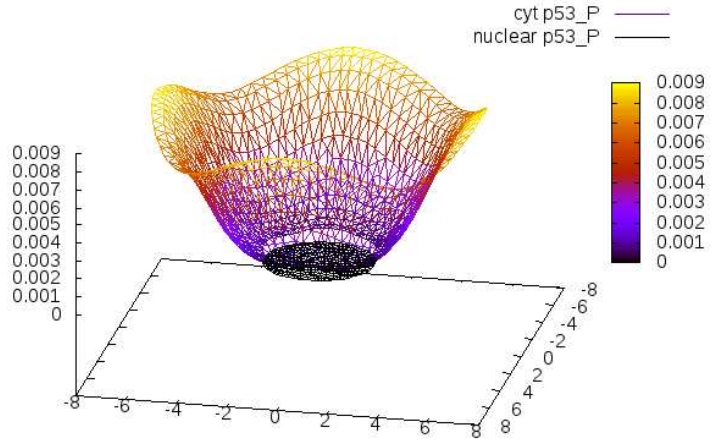


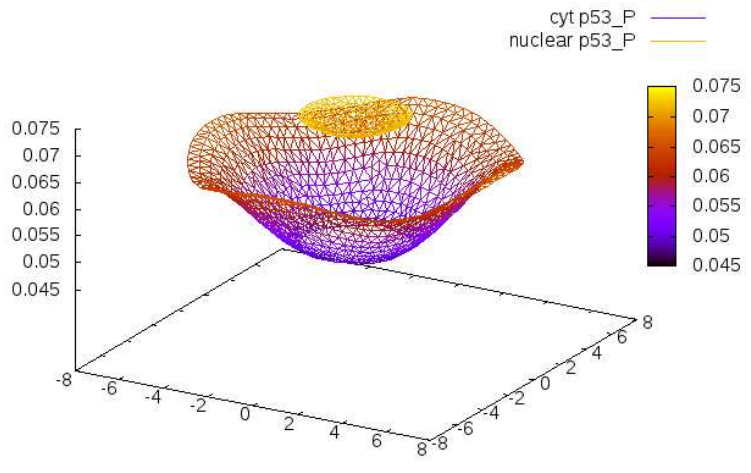
Figure: Simulations of the PDE system; **Left**: temporal evolution of nuclear concentrations. **Right**: temporal evolution of cytoplasmic concentrations

Simulations in a 2D PDE system: circular domain

Concentration of $\mathbf{p53_p}$ after a few simulations: nucleus is still empty.

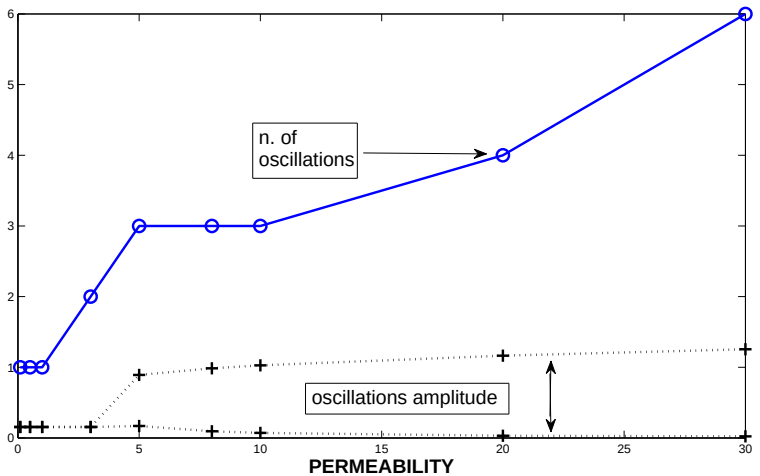


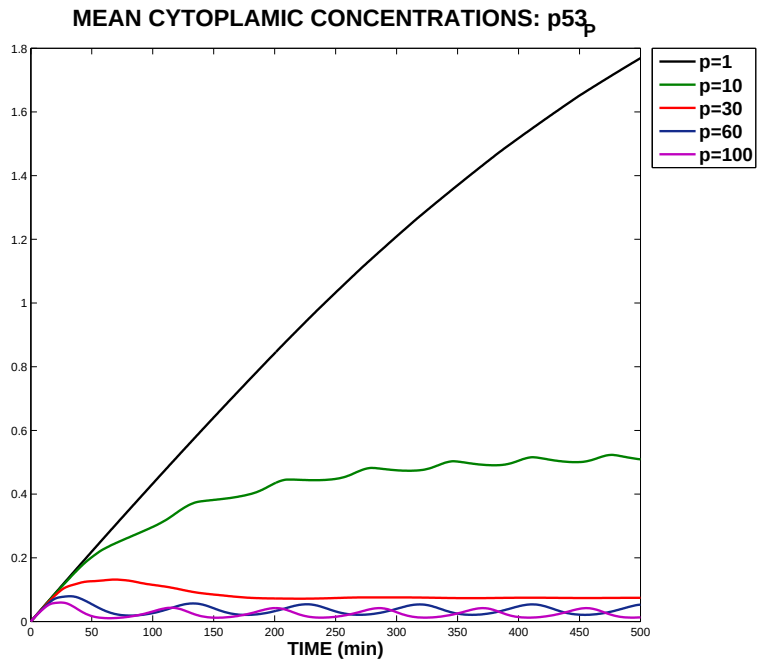
After a longer time $\mathbf{p53_p}$ has accumulated in the nucleus.



Spatial variability

Oscillations depends on diffusion constants. Period depends on permeability, volume...





Simulations in a 2D system : cellular domain

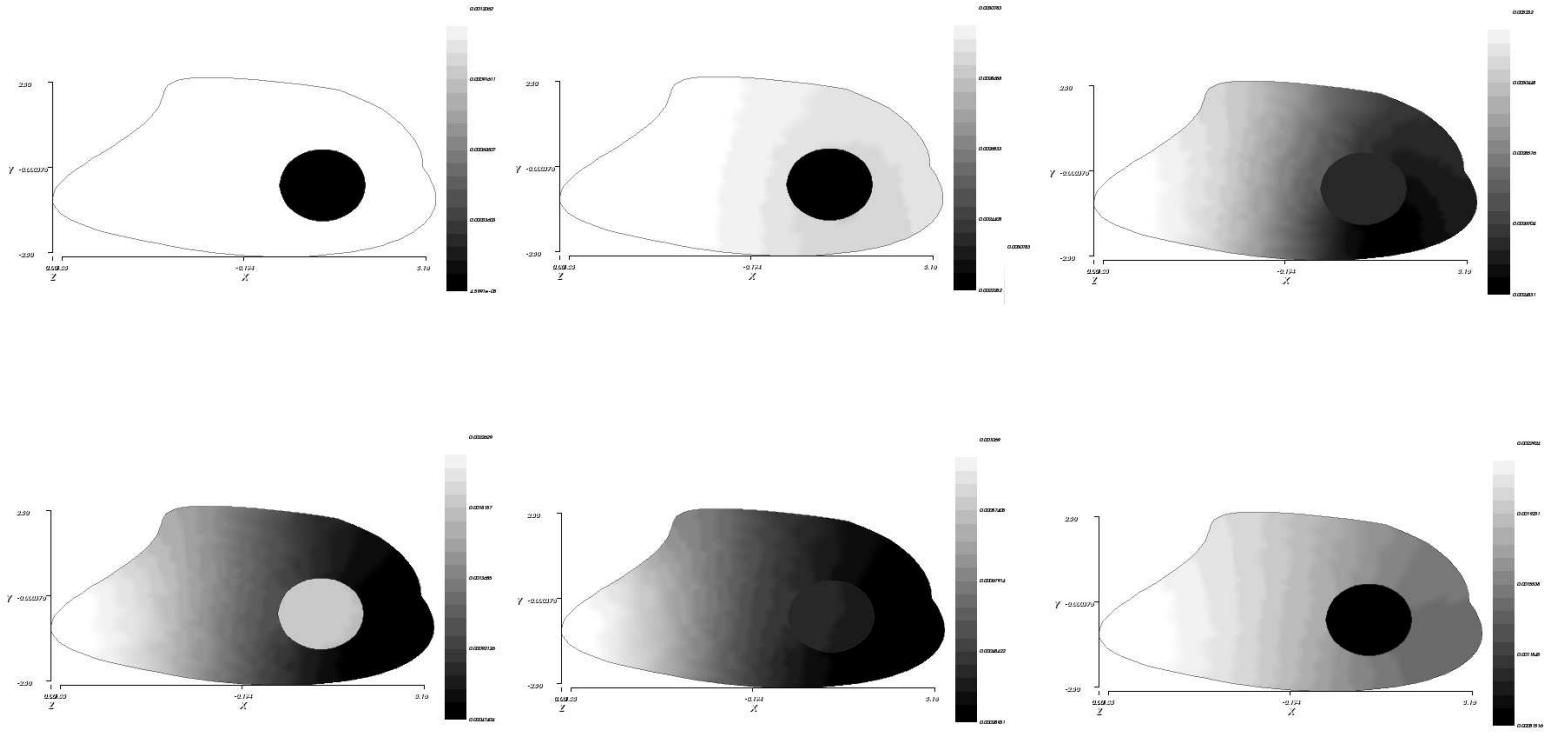


Figure: *in silico* experiments

Results and conclusions

- Physiologically based model of p53 intracellular dynamics
- Amenable model to identify parameters starting from oscillations (for instance measuring delays between nuclear and cytoplasmic peaks)
- Robust oscillations (over different geometry): dynamics depends on **spatial** coefficients
- This model may allow us to investigate the effects of p53 on cell cycle

Questions and Perspectives

Use the model to understand how p53 acts on

- celle cycle arrest
- apoptosis
- DNA repair

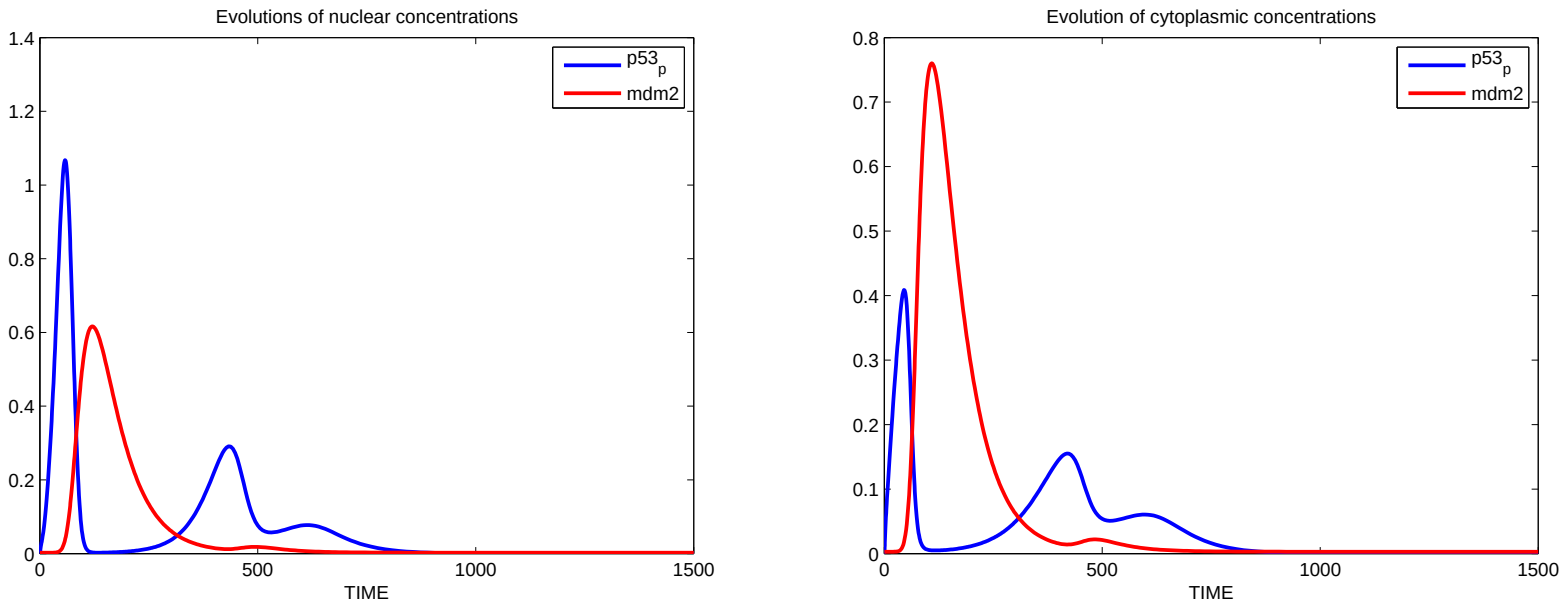


Figure: A basic example of DNA repair: a few oscillations occur. Here the bifurcation parameter ATM is a variable of the system

Also we need to compare the model with real biological data!

A SPATIAL MODEL FOR p53 NUCLEAR ACCUMULATION
how to get to the nucleus

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