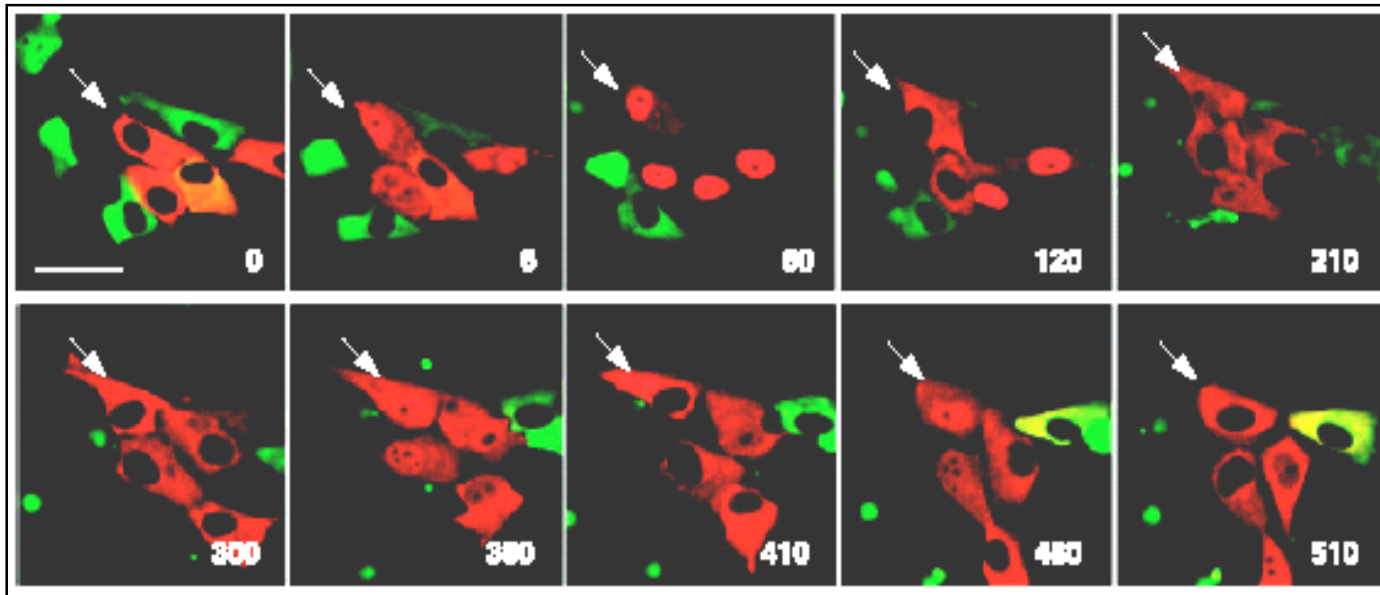


How the number of alleles influences gene expression

Beata Hat
Pawel Paszek
Marek Kimmel
Kazimierz Piechor
Tomasz Lipniacki

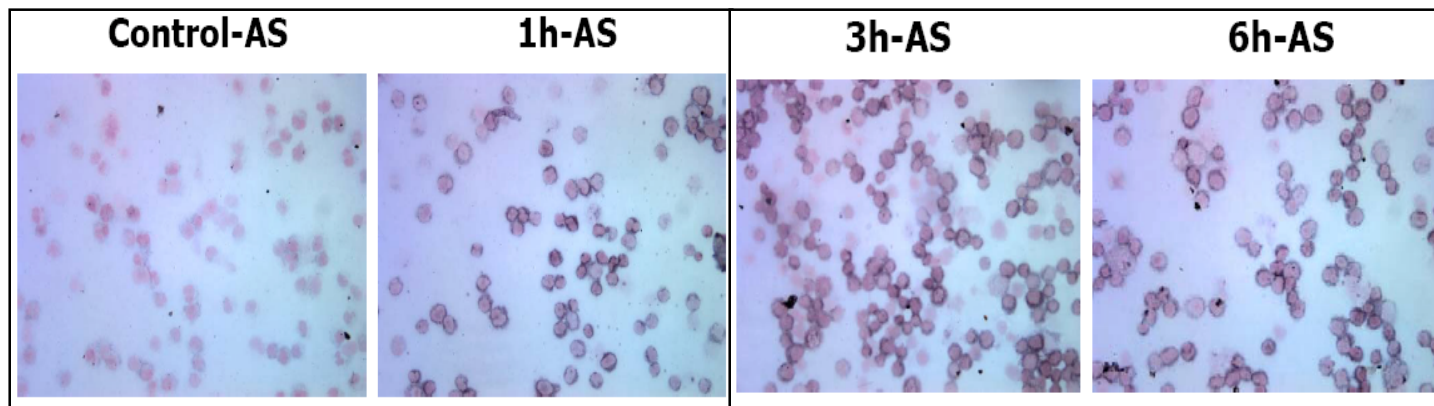
Single cell experiments

Cytoplasm - nucleus oscillations in NF- κ B (red) and I κ B α (green) system after TNF treatment



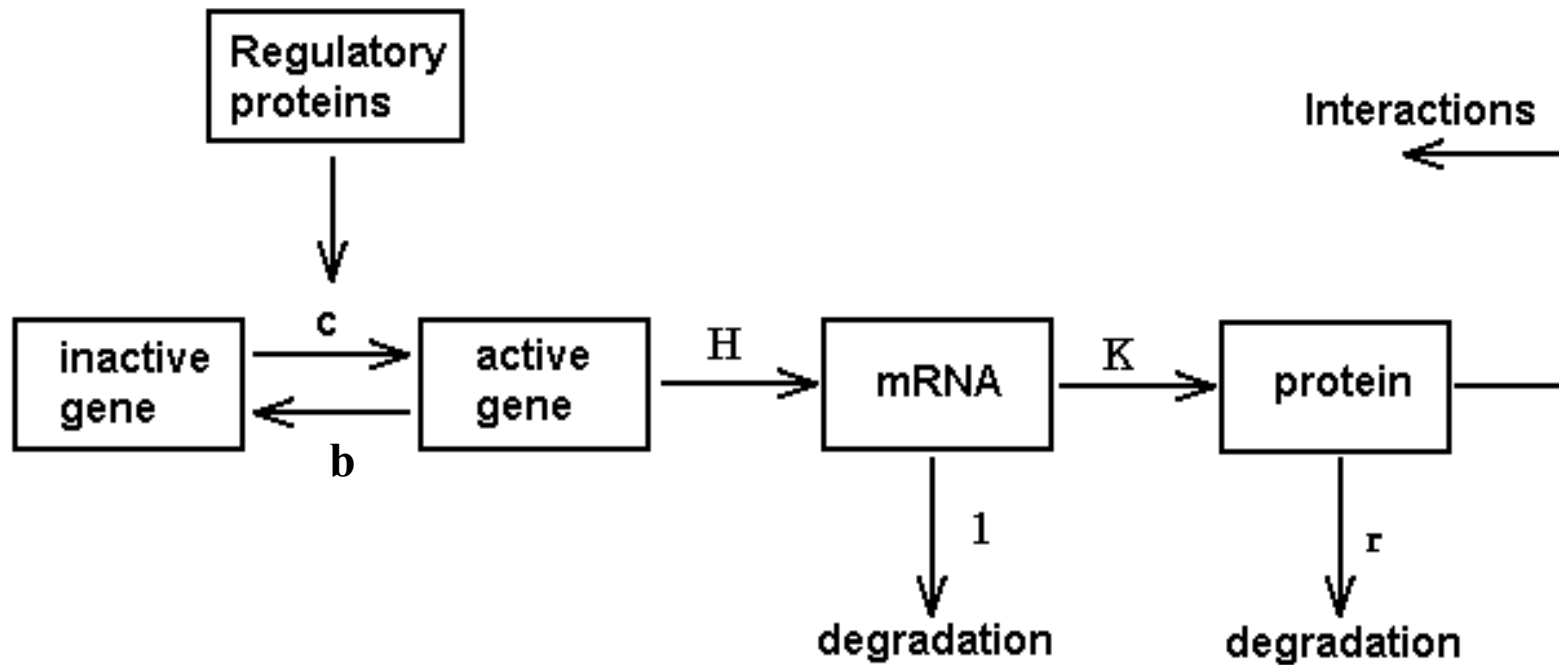
Immortal cancer cell line (SK-N-AS cells), M.R.H. White group

IL8 mRNA level after TNF treatment



Immortal cancer cell line (HeLa cells), A. Brasier group

The main steps in gene expression



The number of molecules involved:



The aim of this work

- How increase in number of gene alleles due to the cancer development or genome duplication in a cell cycle influences its regulation
- How the loss of one allele or its transcriptional inactivity can result in haploinsufficiency disease for autoregulated genes
- To deduce the behavior of „normal” cells from experiments on the transfected cells

A single haploidal gene without feedback regulation



G - gene state

H - transcription rate

K - translation rate

r - protein degradation rate

Continuous approximation



m - # of mRNA molecules

p - # of protein molecules

Probability density functions

$$\frac{\partial}{\partial t} \int_V \rho \, dV + \int_V \rho \mathbf{v} \cdot \nabla \, dV = \int_V \rho \mathbf{v} \cdot \nabla \, dV - \int_V \rho \mathbf{v} \cdot \nabla \, dV$$

$$\frac{\partial}{\partial t} \int_V \rho \, dV + \int_V \rho \mathbf{v} \cdot \nabla \, dV = \int_V \rho \mathbf{v} \cdot \nabla \, dV - \int_V \rho \mathbf{v} \cdot \nabla \, dV$$

The continuity equations for $f(x,y,t)$ and $g(x,y,t)$

$$\frac{\partial f}{\partial t} + \nabla_x f v_x + \nabla_y f v_y + \nabla_z f v_z = \dot{f} - \nabla_x f v_x - \nabla_y f v_y - \nabla_z f v_z$$

$$\frac{\partial g}{\partial t} + \nabla_x g v_x + \nabla_y g v_y + \nabla_z g v_z = \dot{g} - \nabla_x g v_x - \nabla_y g v_y - \nabla_z g v_z$$

$$\frac{\partial f}{\partial t} - \frac{\partial}{\partial x} (f v_x) - \frac{\partial}{\partial y} (f v_y) - \frac{\partial}{\partial z} (f v_z) = \dot{f} - \nabla_x f v_x - \nabla_y f v_y - \nabla_z f v_z$$

$$\frac{\partial g}{\partial t} - \frac{\partial}{\partial x} (g v_x) - \frac{\partial}{\partial y} (g v_y) - \frac{\partial}{\partial z} (g v_z) = \dot{g} - \nabla_x g v_x - \nabla_y g v_y - \nabla_z g v_z$$

Haploidal gene with feedback

- The protein degradation time is much larger than mRNA one (the protein is synthesized directly from the gene and regulates its own expression)



$y(t)$ - protein level



We have the following continuity equations for $f(y,t)$ and $g(y,t)$

$$\frac{\partial f}{\partial t} = \lambda \frac{\partial f}{\partial y} - \gamma f + \beta \frac{\partial f}{\partial y} - \beta \frac{\partial f}{\partial y} - \beta \frac{\partial f}{\partial y}$$

$$\frac{\partial g}{\partial t} = \lambda \frac{\partial g}{\partial y} - \gamma g + \beta \frac{\partial g}{\partial y} - \beta \frac{\partial g}{\partial y} - \beta \frac{\partial g}{\partial y}$$

$$\frac{\partial f}{\partial t} = \lambda \frac{\partial f}{\partial y} - \gamma f + \beta \frac{\partial f}{\partial y} - \beta \frac{\partial f}{\partial y} - \beta \frac{\partial f}{\partial y}$$

$$\frac{\partial g}{\partial t} = \lambda \frac{\partial g}{\partial y} - \gamma g + \beta \frac{\partial g}{\partial y} - \beta \frac{\partial g}{\partial y} - \beta \frac{\partial g}{\partial y}$$

For $\frac{\partial f}{\partial t} = \frac{\partial g}{\partial t} = 0$ we obtain

$$\lambda \frac{\partial f}{\partial y} - \gamma f + \beta \frac{\partial f}{\partial y} - \beta \frac{\partial f}{\partial y} - \beta \frac{\partial f}{\partial y} = 0$$

$$\lambda \frac{\partial g}{\partial y} - \gamma g + \beta \frac{\partial g}{\partial y} - \beta \frac{\partial g}{\partial y} - \beta \frac{\partial g}{\partial y} = 0$$

Diploidal gene with feedback

$$\frac{d^2 x_1}{dt^2} + \gamma_1 \frac{dx_1}{dt} + \alpha_1 x_1 = \beta_1 \frac{dx_2}{dt} + \gamma_2 x_2$$

$$\frac{d^2 x_2}{dt^2} + \gamma_2 \frac{dx_2}{dt} + \alpha_2 x_2 = \beta_2 \frac{dx_1}{dt} + \gamma_3 x_1$$

$$\frac{d^2 x_3}{dt^2} + \gamma_3 \frac{dx_3}{dt} + \alpha_3 x_3 = \beta_3 \frac{dx_2}{dt} + \gamma_4 x_2$$

$$\frac{d^2 x_1}{dt^2} + \gamma_1 \frac{dx_1}{dt} + \alpha_1 x_1 = \beta_1 \frac{dx_2}{dt} + \gamma_2 x_2$$

$$\frac{d^2 x_2}{dt^2} + \gamma_2 \frac{dx_2}{dt} + \alpha_2 x_2 = \beta_2 \frac{dx_1}{dt} + \gamma_3 x_1$$

$$\frac{d^2 x_3}{dt^2} + \gamma_3 \frac{dx_3}{dt} + \alpha_3 x_3 = \beta_3 \frac{dx_2}{dt} + \gamma_4 x_2$$

In the case without feedback

($\beta_1 = \beta_2 = \beta_3 = 0$ and $\gamma_2 = \gamma_3 = \gamma_4 = 0$)

the functions $x_1(t)$, $x_2(t)$, $x_3(t)$

are given by the convolution formulas

$$x_1(t) = \int_0^t \beta_1 \dot{x}_2(\tau) e^{-\gamma_1(t-\tau)} d\tau + \int_0^t \gamma_2 x_2(\tau) e^{-\gamma_1(t-\tau)} d\tau$$

$$x_2(t) = \int_0^t \beta_2 \dot{x}_1(\tau) e^{-\gamma_2(t-\tau)} d\tau + \int_0^t \gamma_3 x_1(\tau) e^{-\gamma_2(t-\tau)} d\tau$$

$$x_3(t) = \int_0^t \beta_3 \dot{x}_2(\tau) e^{-\gamma_3(t-\tau)} d\tau + \int_0^t \gamma_4 x_2(\tau) e^{-\gamma_3(t-\tau)} d\tau$$

where $\beta_1 = \beta_2 = \beta_3$ and $\gamma_2 = \gamma_3 = \gamma_4$ for $\beta_1 = \beta_2 = \beta_3$

$\beta_1 = \beta_2 = \beta_3$ and $\gamma_2 = \gamma_3 = \gamma_4$ for $\beta_1 = \beta_2 = \beta_3$

Positive Feedback

We consider the external induction of self-activating gene

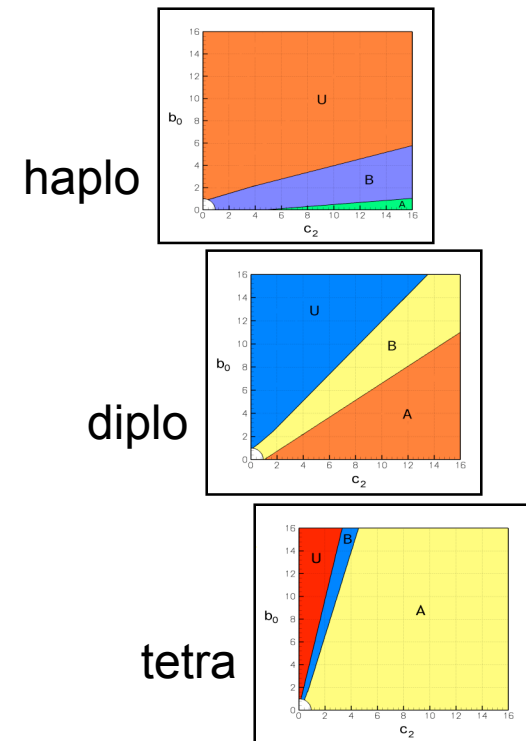


There are three patterns of N - allelic ($N = 1, 2, 4$) gene activation corresponding to different pairs of μ_1 and μ_2 in (μ_1, μ_2) - plane

{A} - mode in which gene remains Active
(i.e. $\mu_1 \mu_2 \geq \frac{1}{\mu_1}$) for all μ_1

{B} - mode in which gene activates for some μ_1
and distribution $\mu_1 \mu_2$ is transiently Bimodal

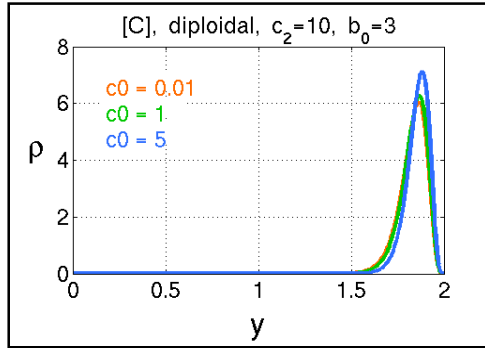
{U} - mode in which gene activates for some μ_1
and its distribution $\mu_1 \mu_2$ remains Unimodal



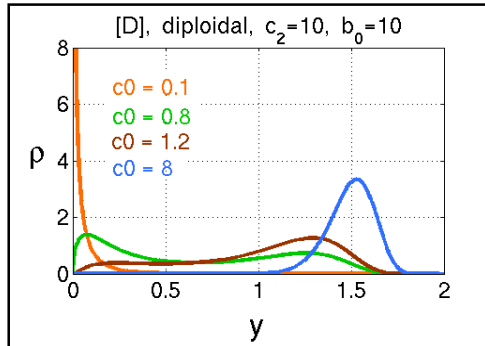
Protein distributions

3 modes of diploidal gene activation

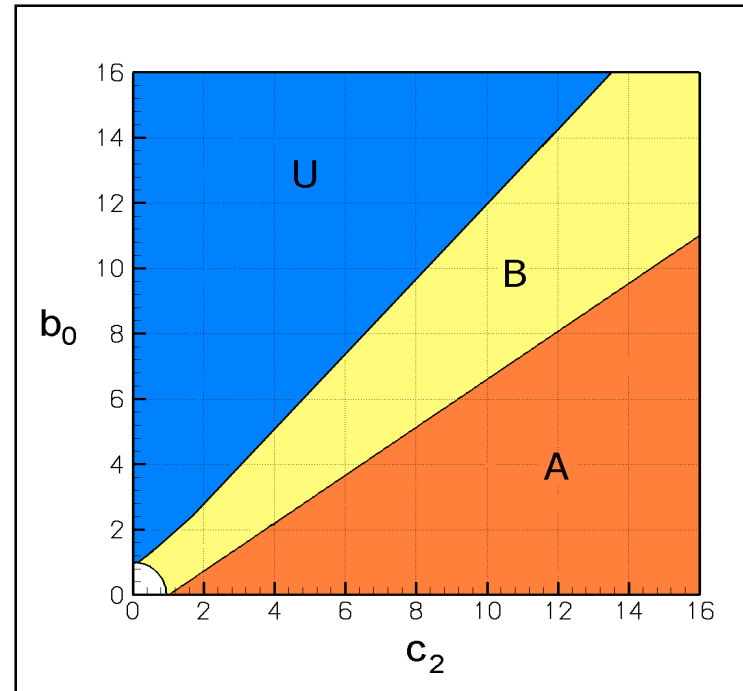
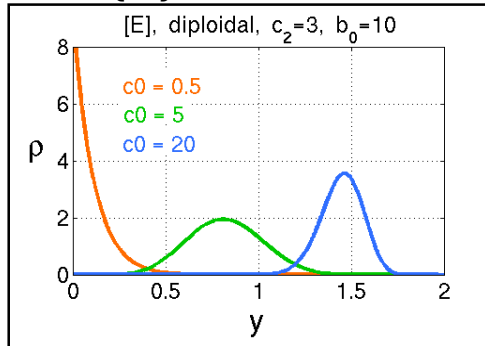
{A} - mode



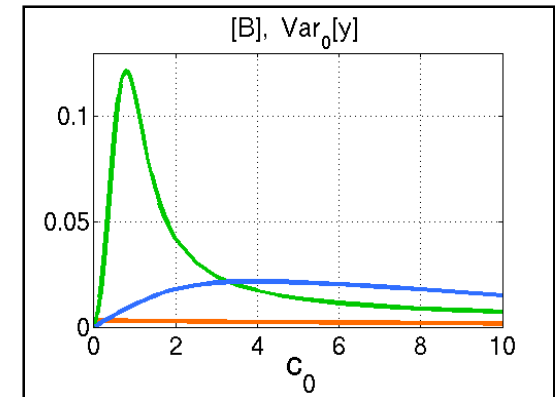
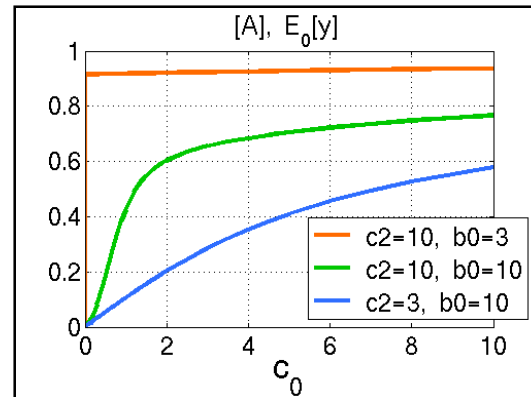
{B} - mode



{U} - mode

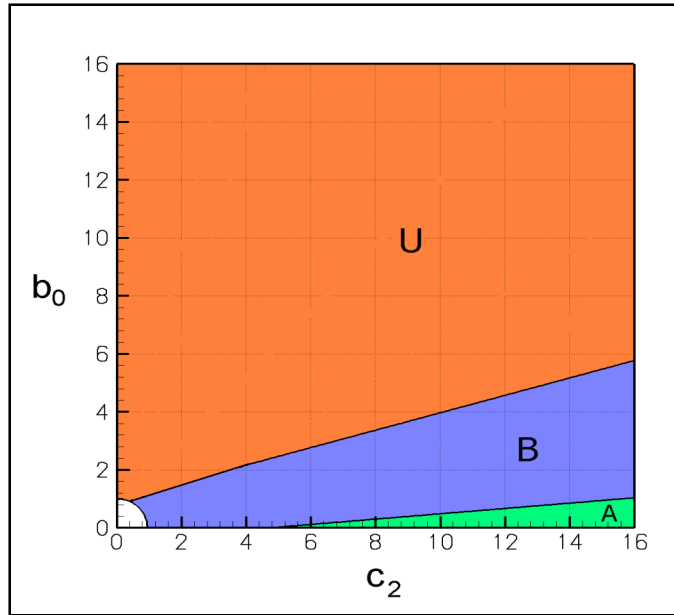


Mean and variance (per gene copy)



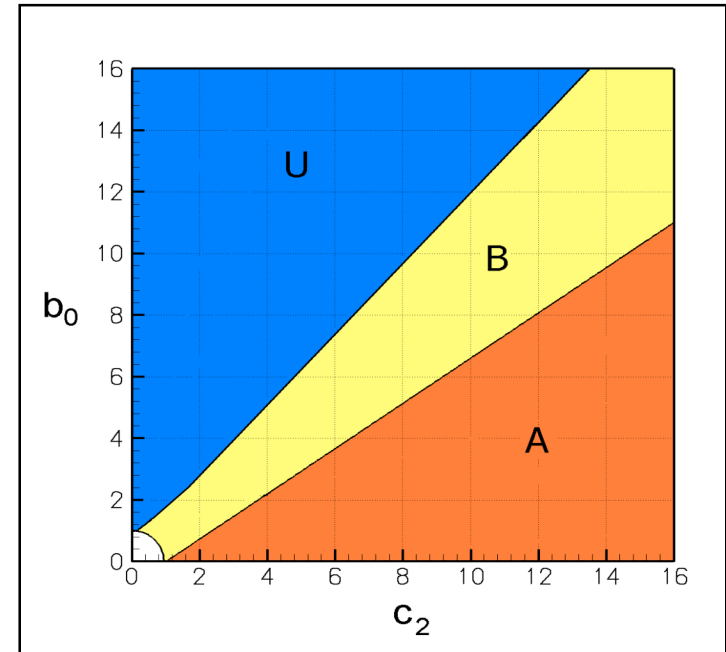
11/15

haploidal gene

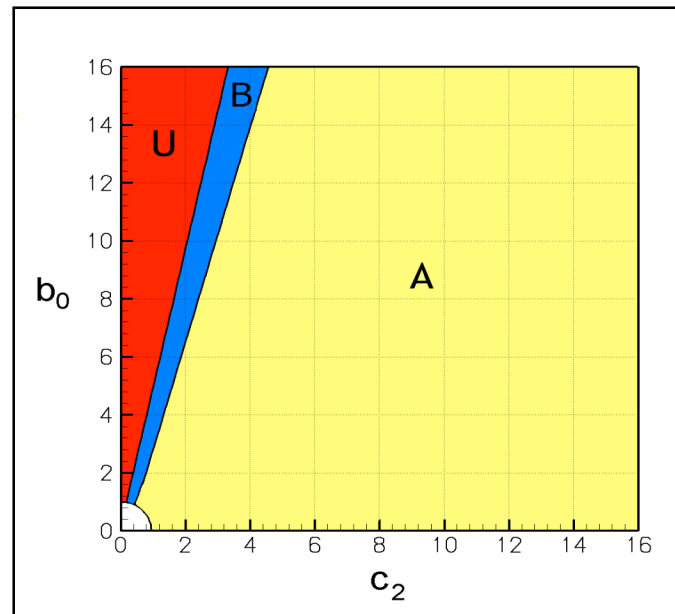


3 regions in (b_0, c_2) - plane corresponding to 3 modes of activation

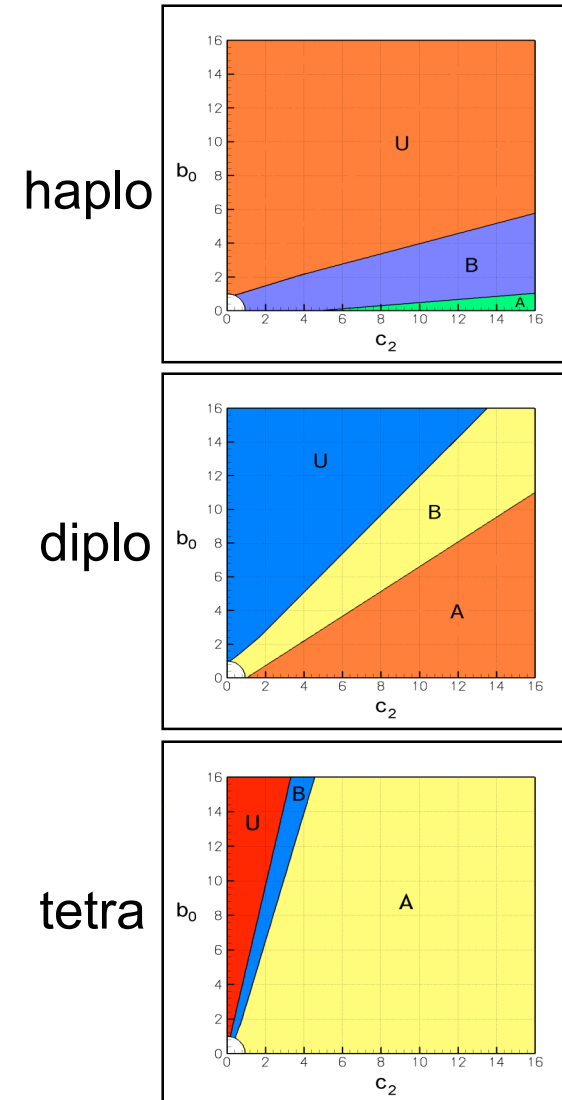
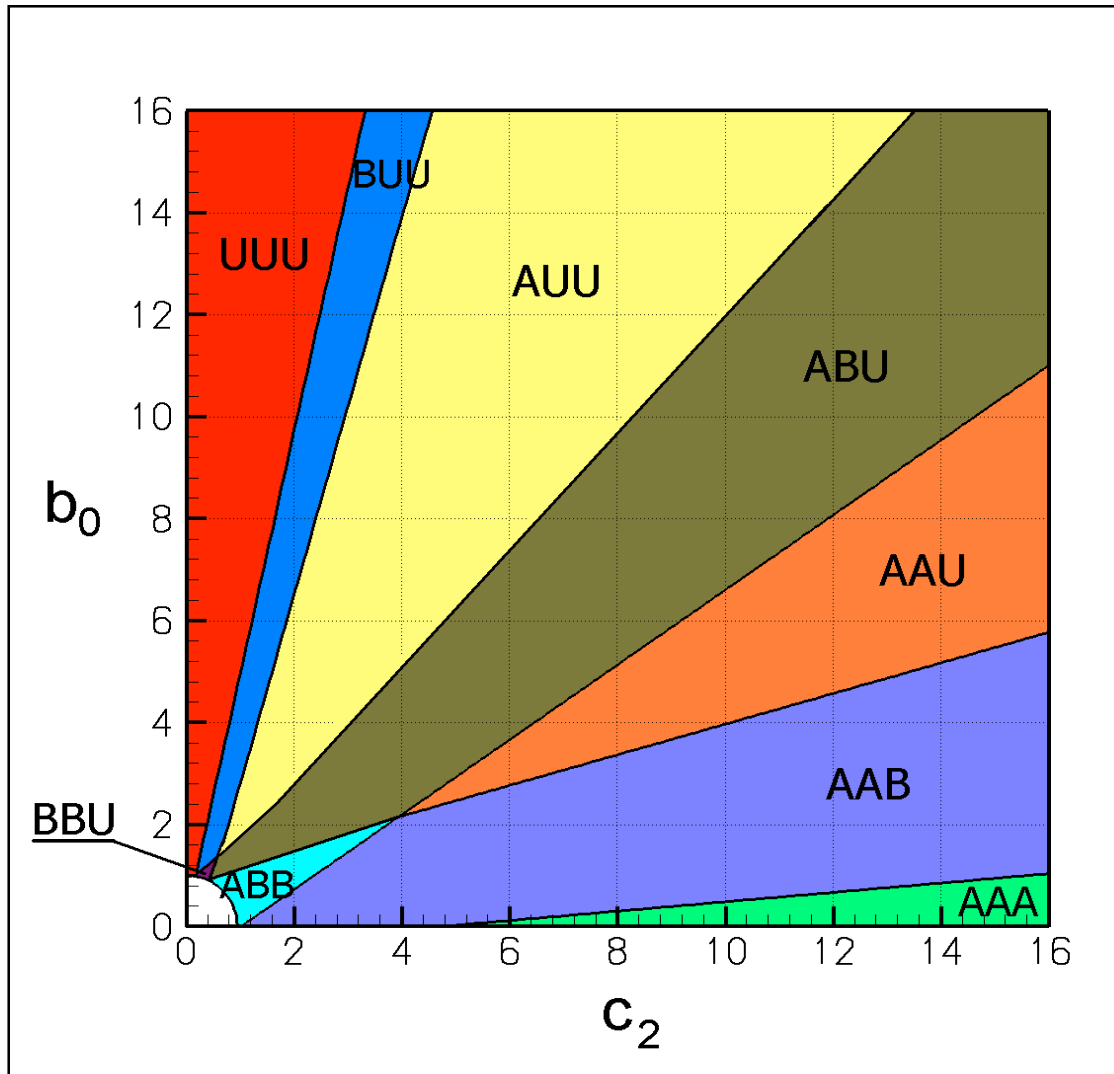
diploidal



tetraploidal

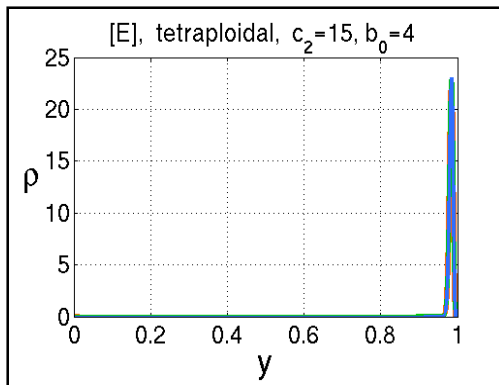
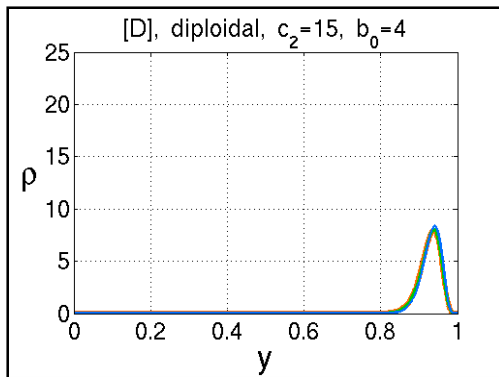
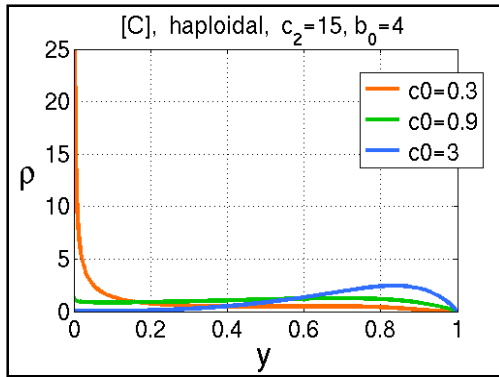


9 possible patterns of 1-,2- and 4-copy-gene-system activation

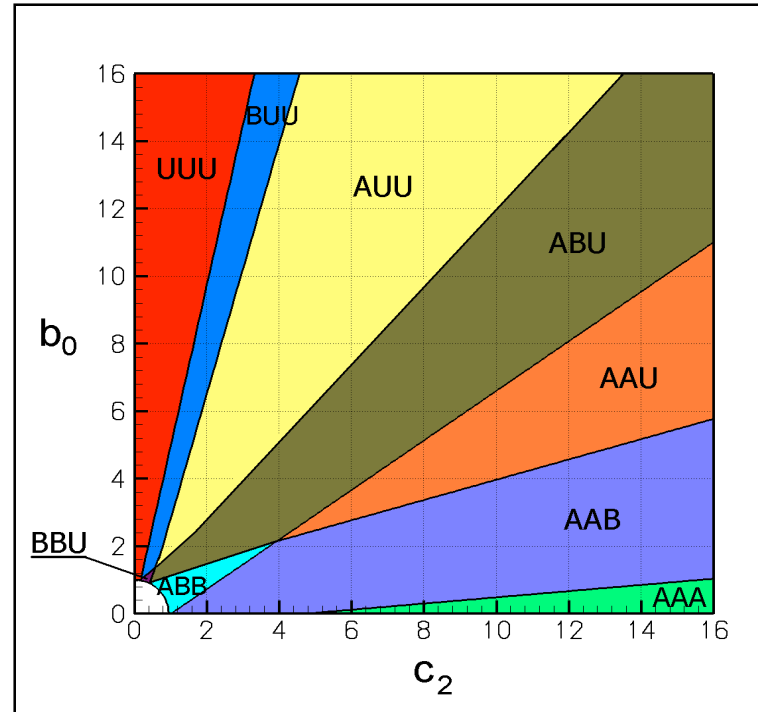


{ABU} sector denotes that tetraploidal gene is in the {A}-mode, diploidal - in the {B}-mode, haploidal - in the {U}-mode

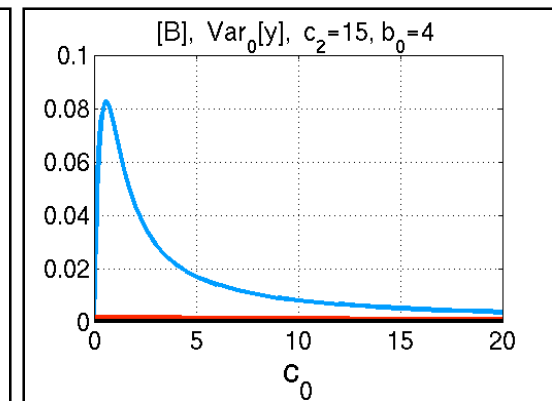
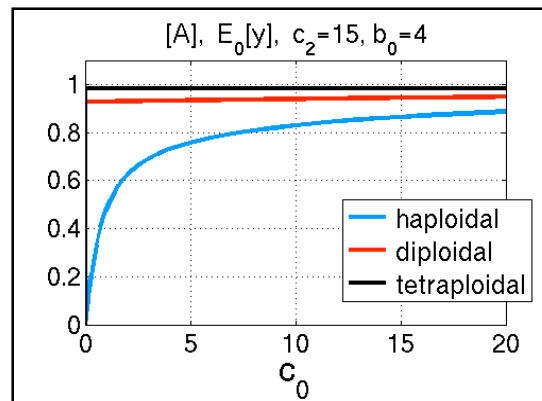
Protein distributions



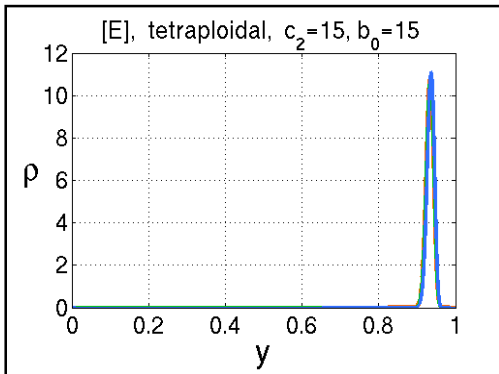
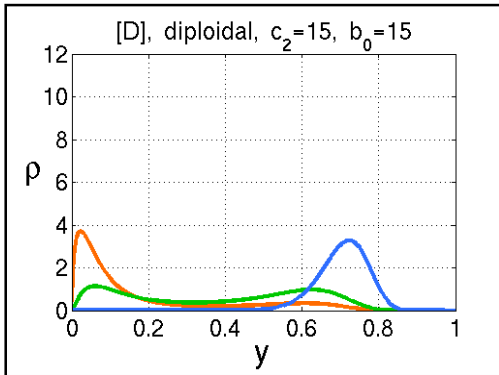
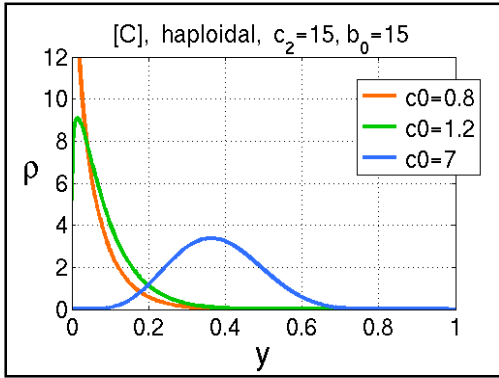
Mode {AAB}, ■ - ■■, ■■ - ■■



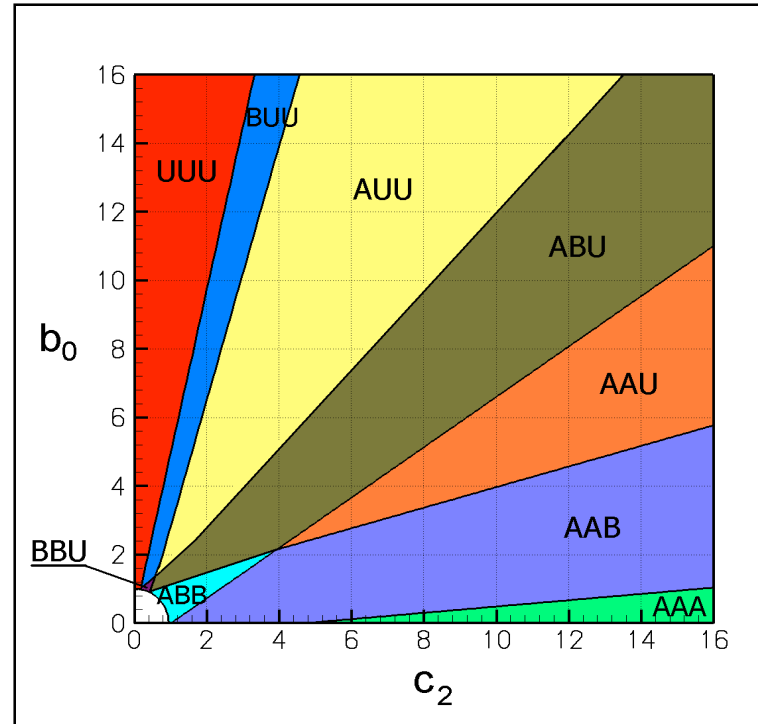
Mean and variance of the protein (per gene copy)



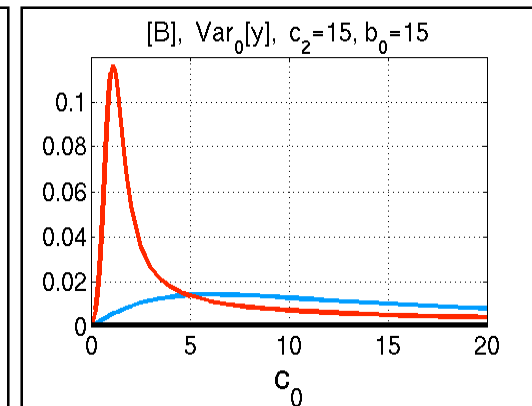
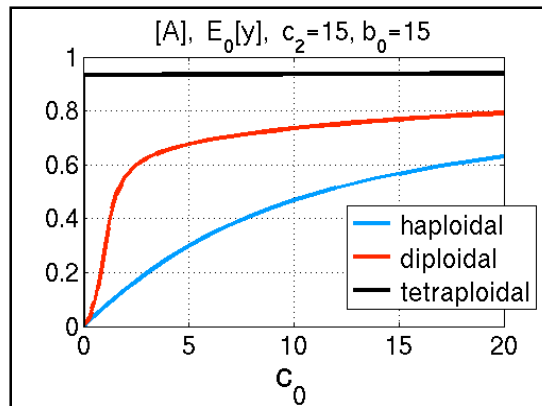
Protein distributions



Mode {ABU}, ■ - ■■, ■ - ■■



Mean and variance of the protein (per gene copy)



Take Home Conclusions

Considering the simultaneous activation of a haploid, diploid and tetraploid gene there exist nine modes of gene activation

- allele loss may stop the persistent gene activity and lead to disease if the constant level of gene product is required
- gene duplication may result in a persistent activity and lead to disease when haploid or diploid gene is „designed” to act as a switch