Виртуално проектиране на лекарства

Drug design

Drug discovery is mostly portrayed as a linear, consecutive process that starts with target and lead discovery followed by lead optimization and pre-clinical in vitro and in vivo studies to determine if such compounds satisfy a number of pre-set criteria for initiating clinical development

(Source: PAREXEL, PAREXEL"S Pharmaceutical R&D Statical SourceBook, 2001, p96)

Проблемът на големите числа във фармацията

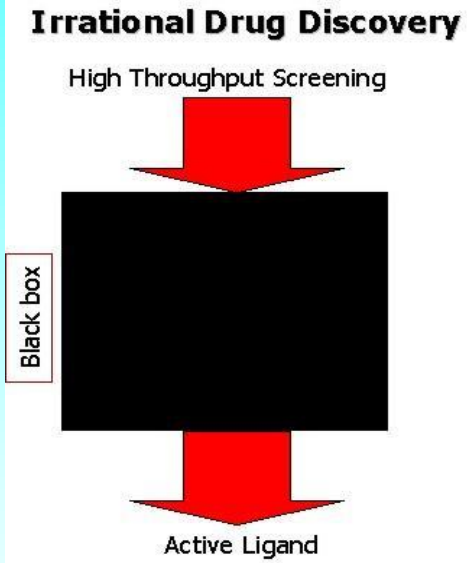
- Има около 10^{80} теоретично възможни биологически активни вещества,
  - 10^{18} от тях биха могли да бъдат вероятни лекарствени препарати
  - 10^{7} са известни химически съединения,
  - 10^{6} са съединения на пазара,
  - още 10^{6} съединения са в базите от информация на фирмите,
  - 10^{5} са химическите вещества в базите от данни на лекарствените фирми ,
  - около $5x10^{4}$ са лекарствата на пазара и
  - 10^{3} са търговски изгодни лекарства.

University of Heidelberg BASF

Drug Research is ....

the Search for a Needle in a Haystack

Irrational vs Rational drug design



High Throughput Screening

$\rightarrow 10^4$ ligands per day

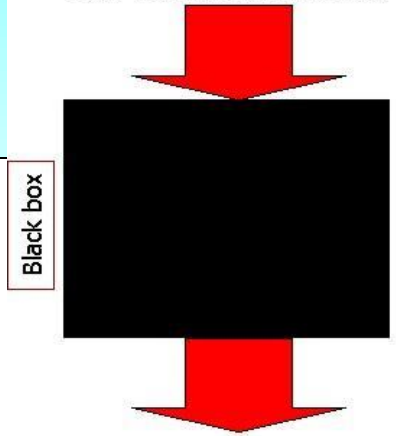
But: Hit Rate 10^{-6} per ligand

Screening

Irrational vs Rational drug design

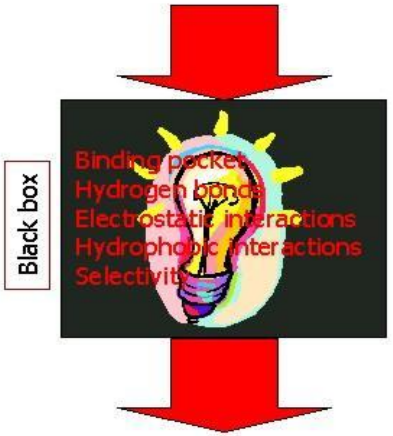
Irrational Drug Discovery

High Throughput Screening



Active Ligand

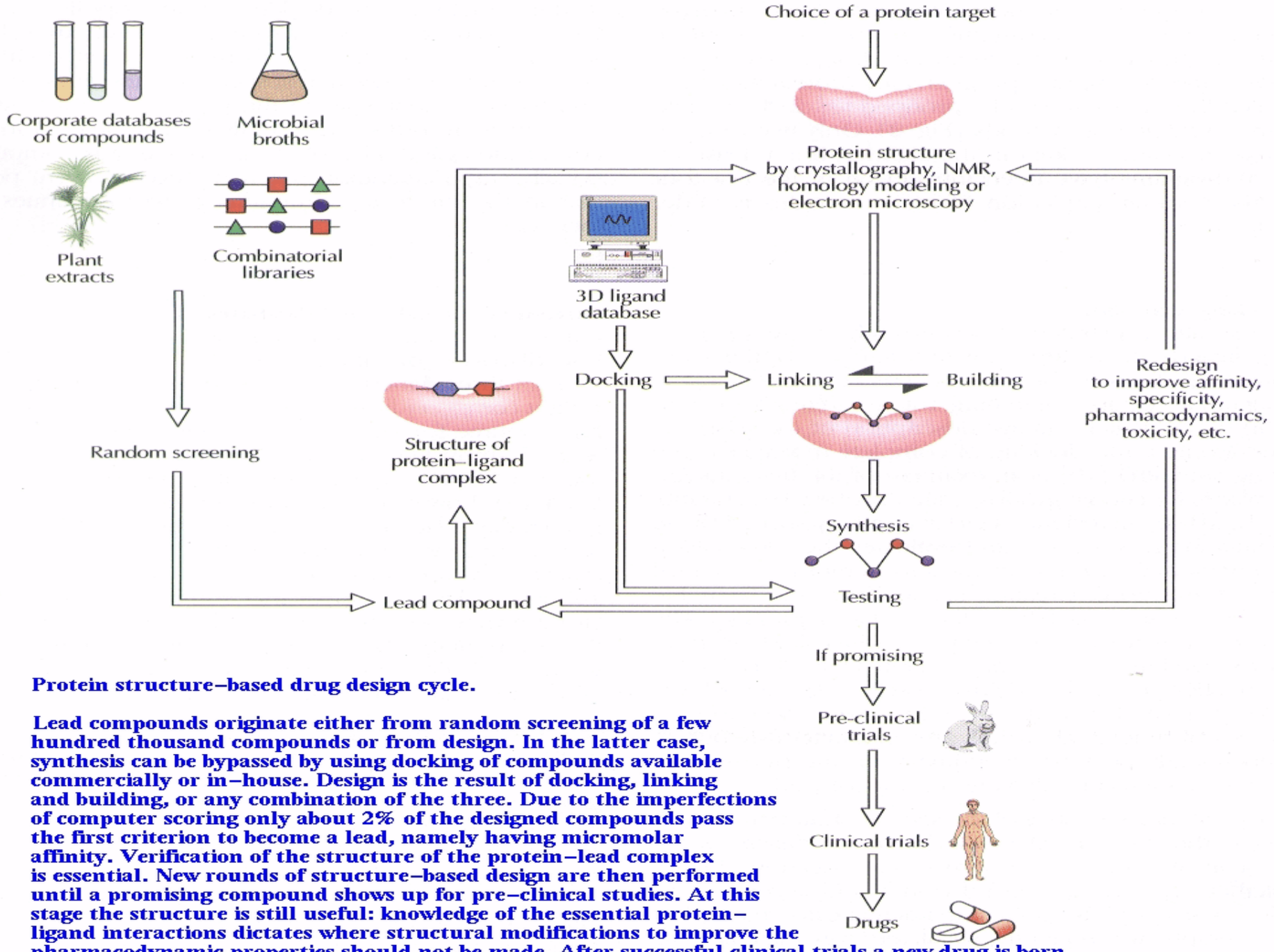
Structure-based Drug Design



Active Ligand

Drug Design

Finding the Right Key for the Lock



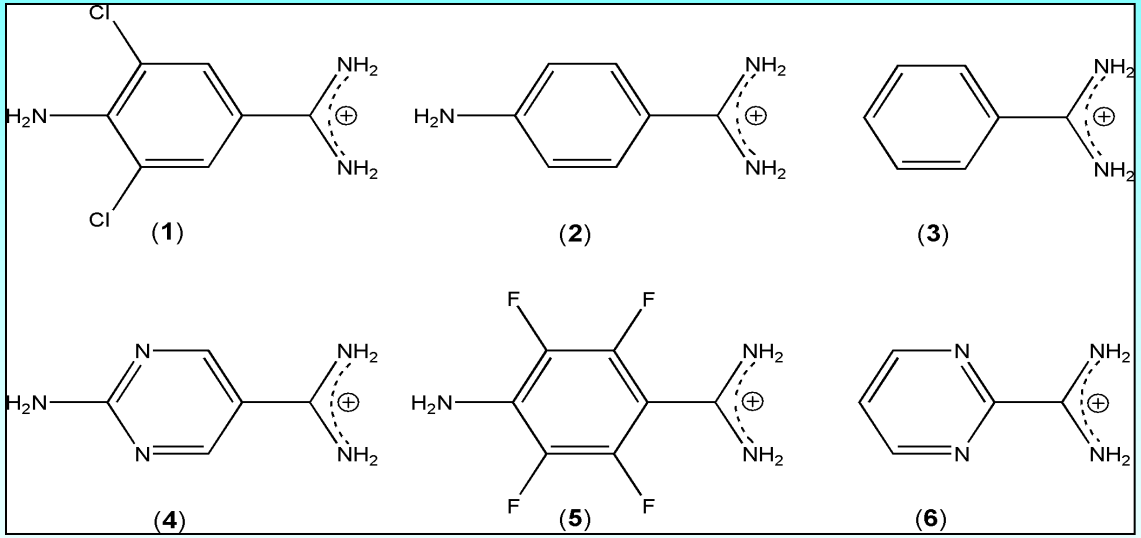
Protein structure–based drug design cycle.

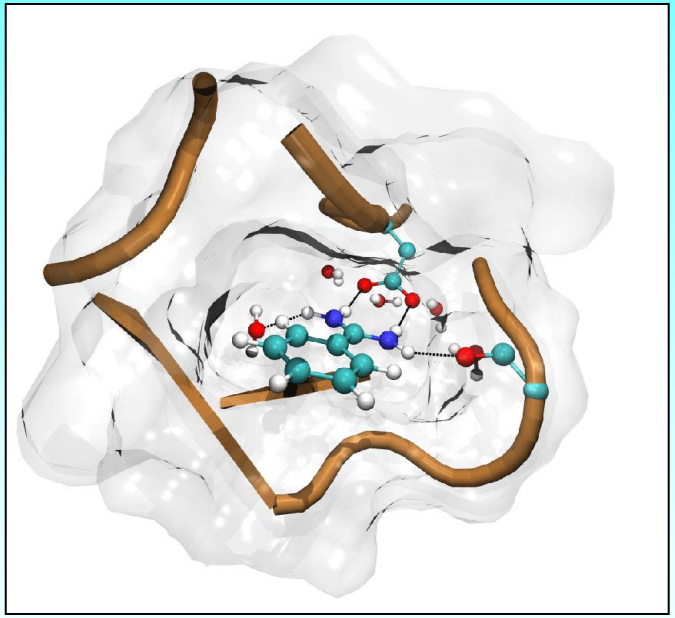
Lead compounds originate either from random screening of a few hundred thousand compounds or from design. In the latter case, synthesis can be bypassed by using docking of compounds available commercially or in–house. Design is the result of docking, linking and building, or any combination of the three. Due to the imperfections of computer scoring only about 2% of the designed compounds pass the first criterion to become a lead, namely having micromolar affinity. Verification of the structure of the protein–lead complex is essential. New rounds of structure–based design are then performed until a promising compound shows up for pre–clinical studies. At this stage the structure is still useful: knowledge of the essential protein–ligand interactions dictates where structural modifications to improve the pharmacodynamic properties should not be made. After successful clinical trials a new drug is born.

Drug design

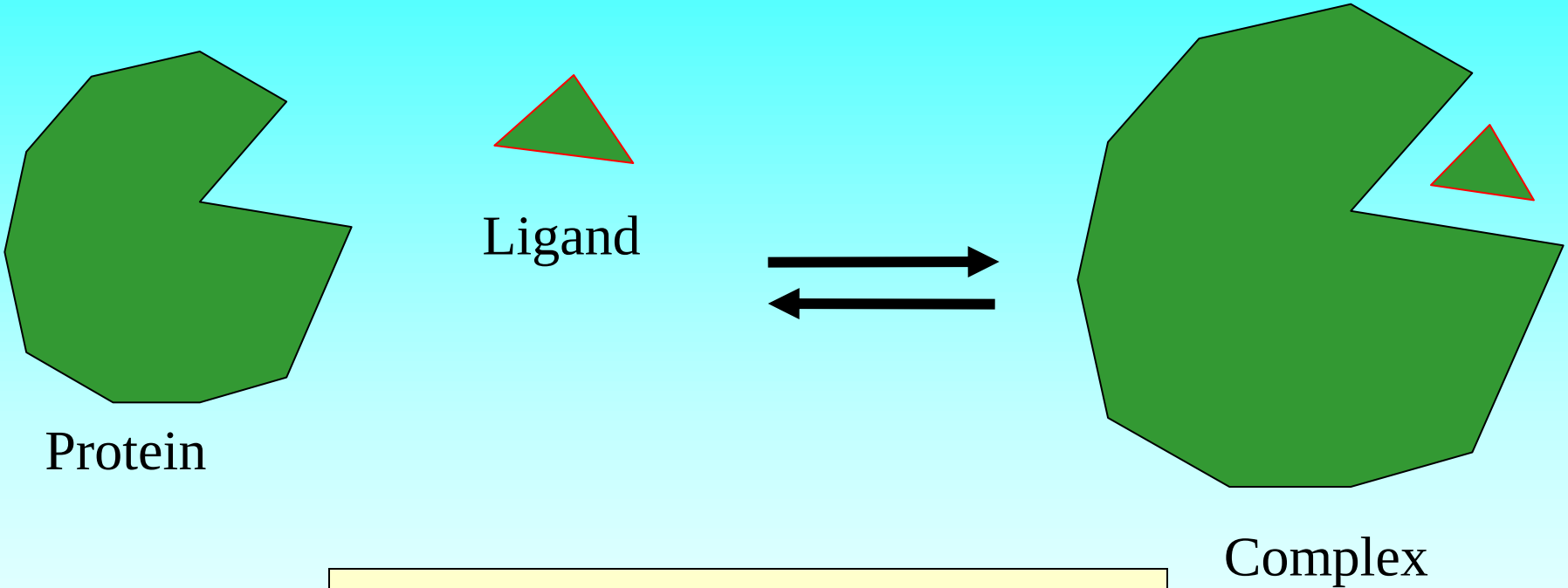
Ligands

Target





Ligand Binding.

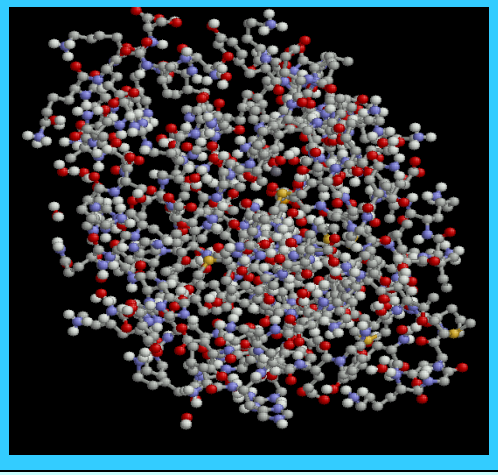


Two Approaches:

1) Binding Free Energy Calculations
2) Empirical Scoring Functions

Computer Simulation - Basic Principles

Model System



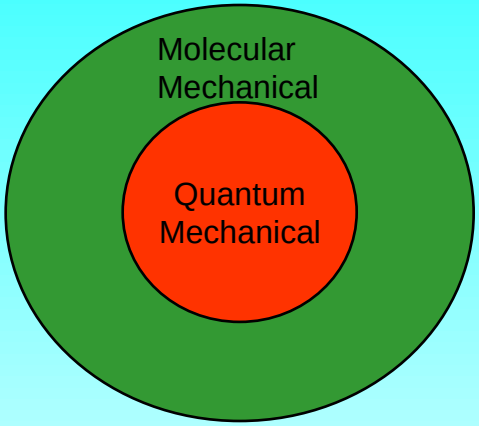
Molecular Mechanics Potential

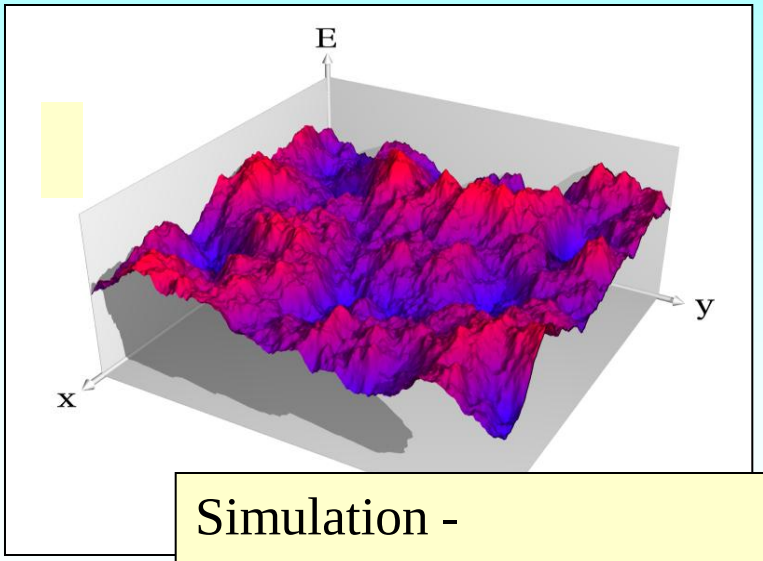
$$V = \sum_{bonds} k_b (b - b_0)^2 + \sum_{angles} k_\theta (\theta - \theta_0)^2 +$$

$$+ \sum_{dihedrals} \sum_{n=1}^{N} K_\phi [1 + \cos(n\phi - \delta)]$$

$$+ \sum_{i,j} 4\varepsilon_{ij} \left[\left(\frac{\sigma_{ij}}{r_{ij}} \right)^{12} - \left(\frac{\sigma_{ij}}{r_{ij}} \right)^{6} \right] + \sum_{i,j} \left(\frac{q_i q_j}{D r_{ij}} \right)$$

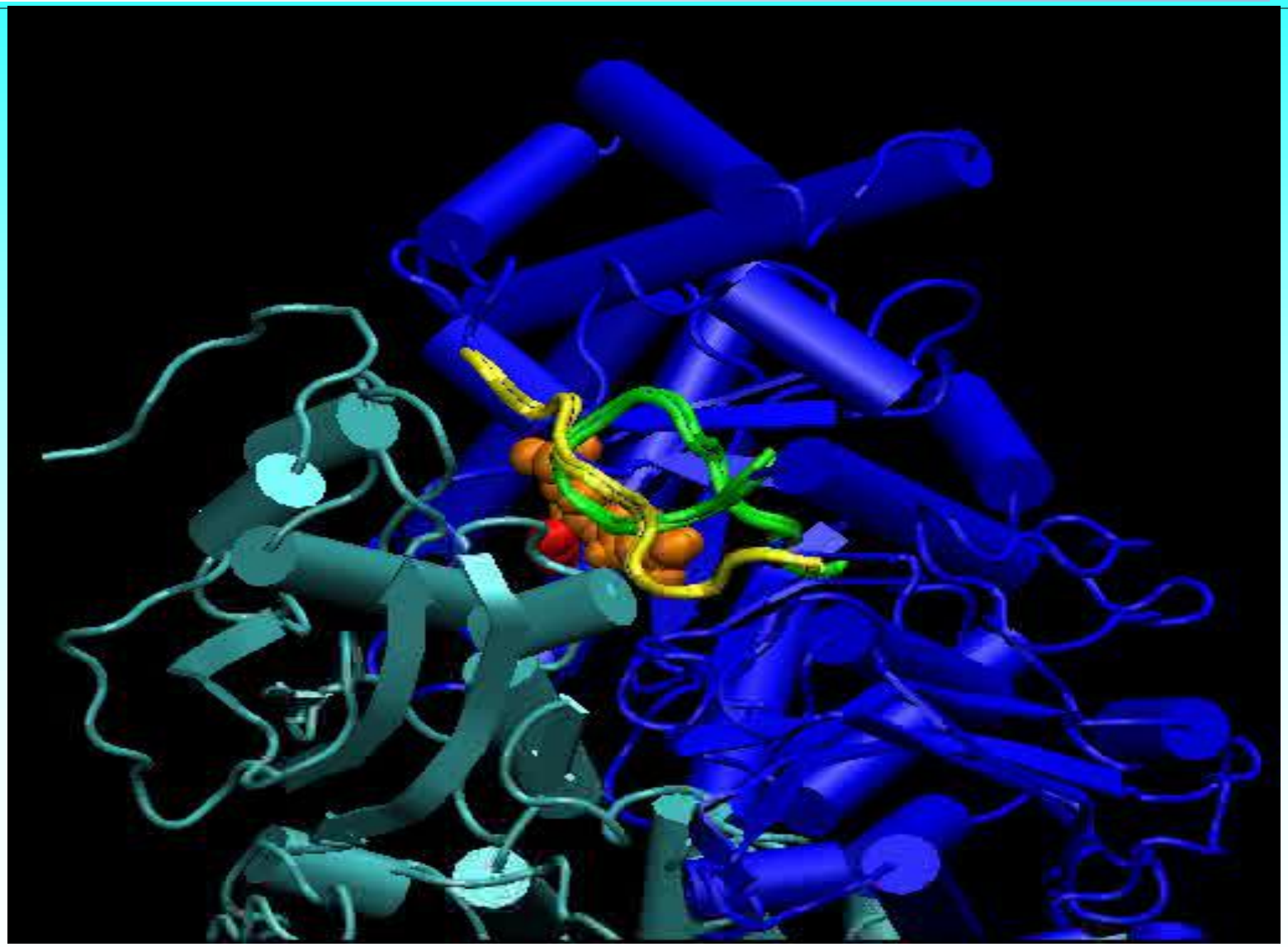
or QM/MM Potential



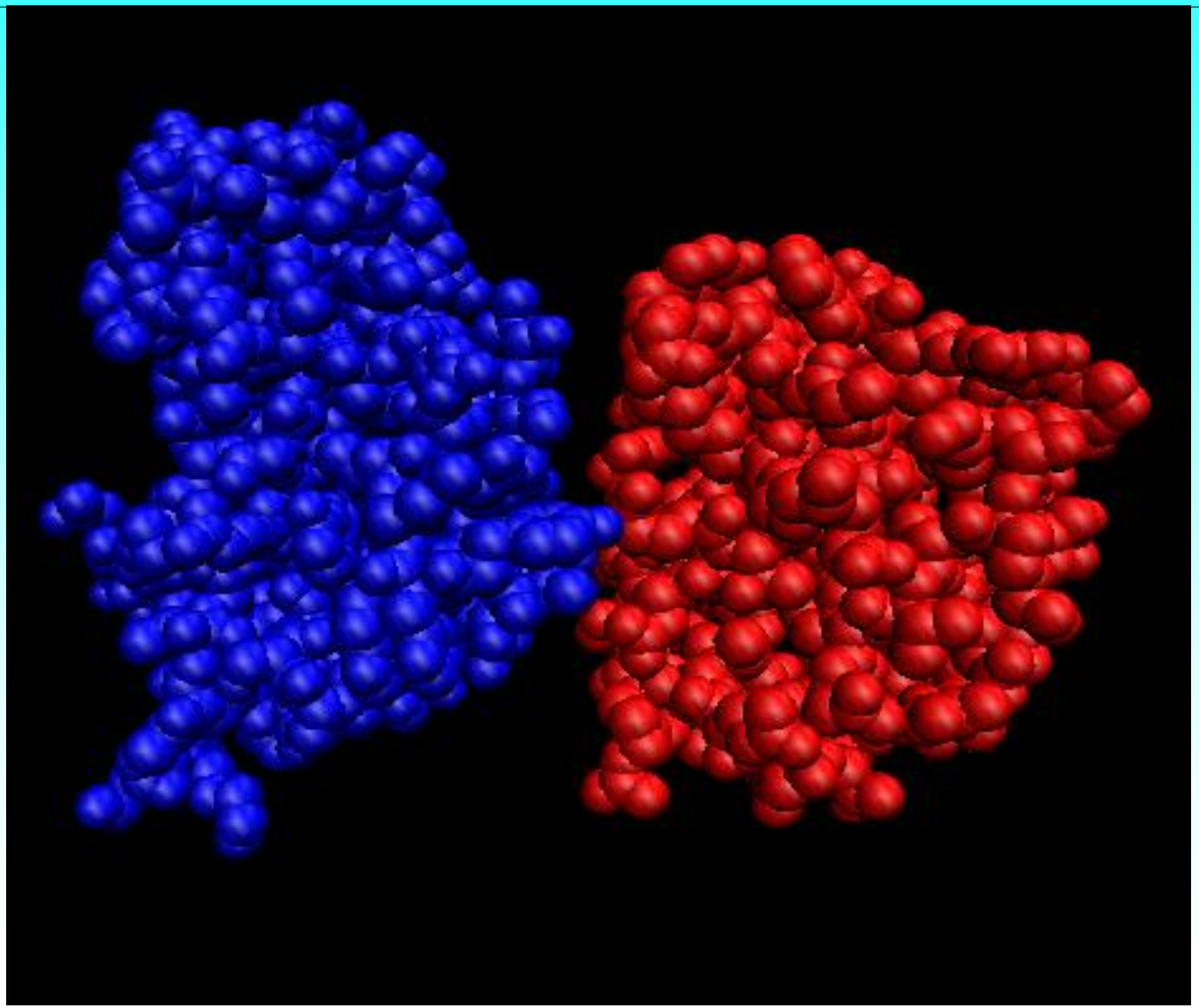


Simulation -
exploring the energy landscape

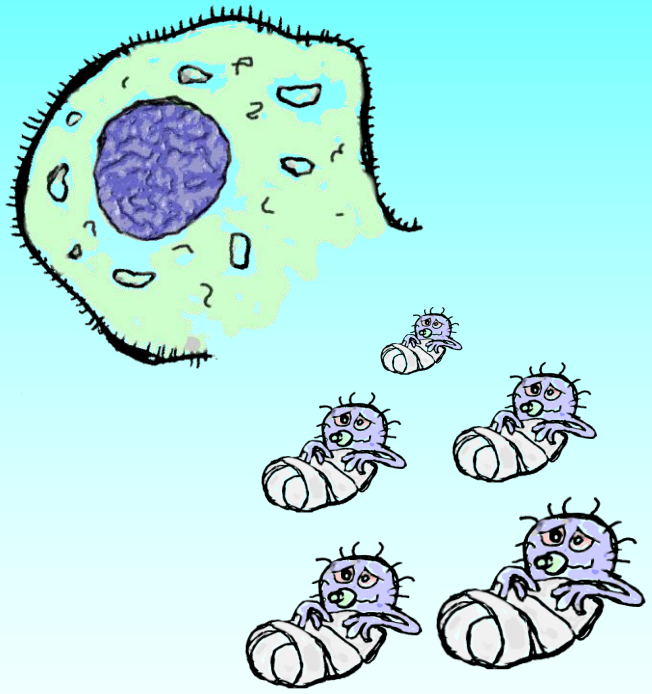
Молекулярна динамика на протеин и лиганд

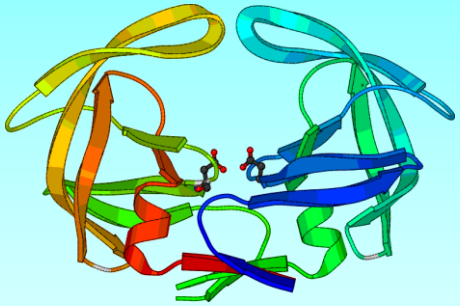


Binding of two proteins



HIV-Protease



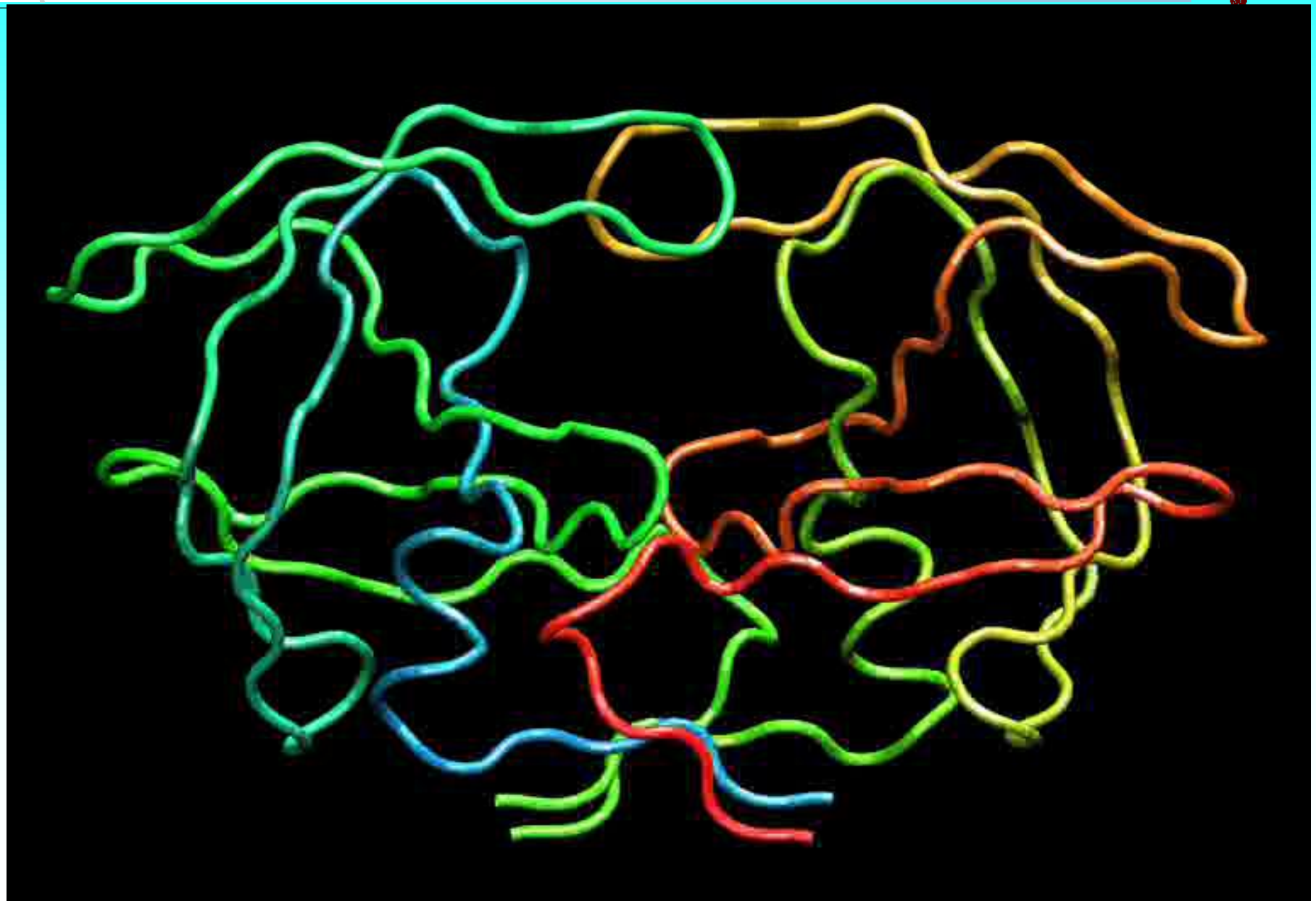


HIV-1 PR

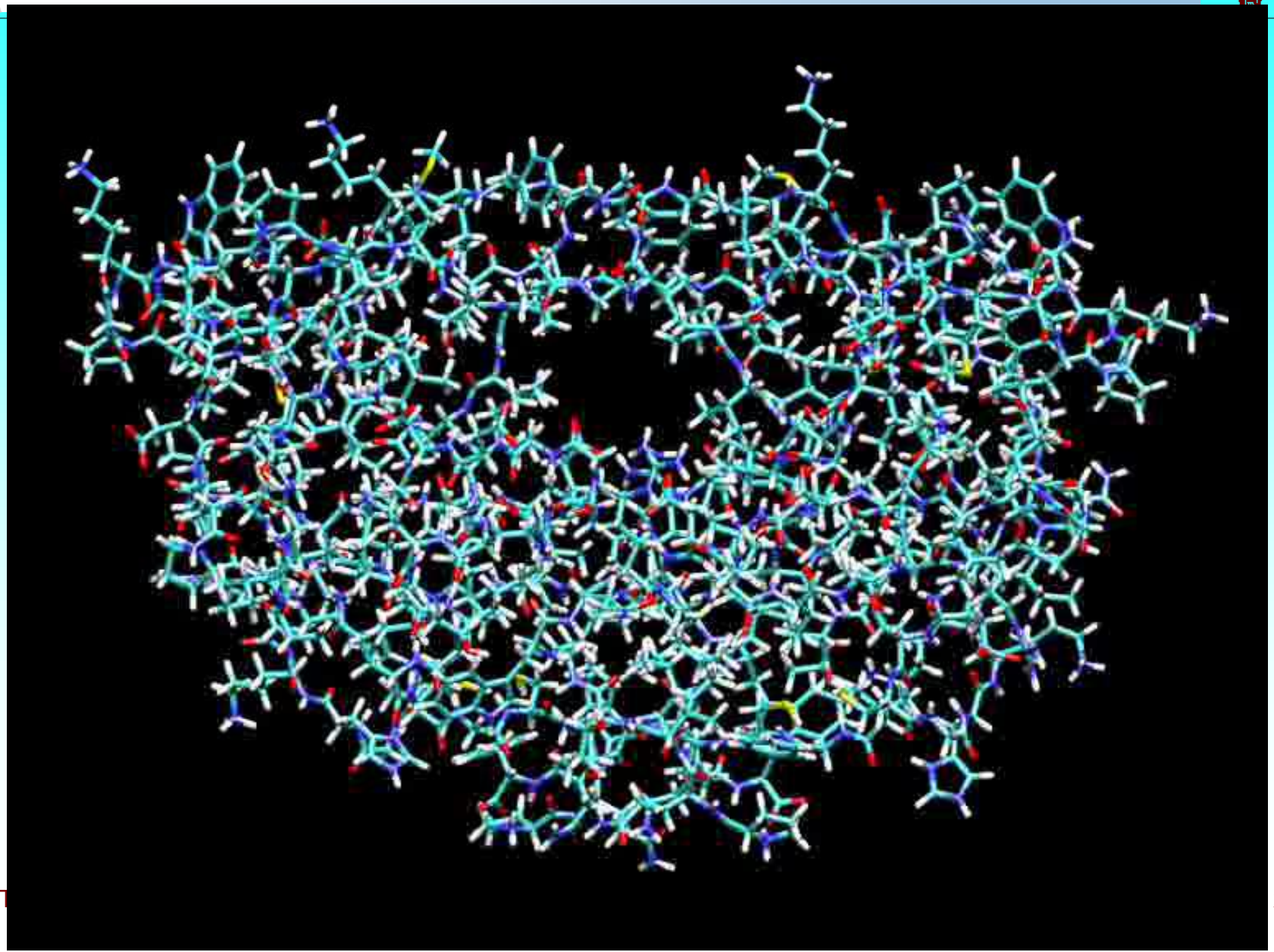
Не зряла и не инфекциозна форма на вирусни частици

Зрели вируси

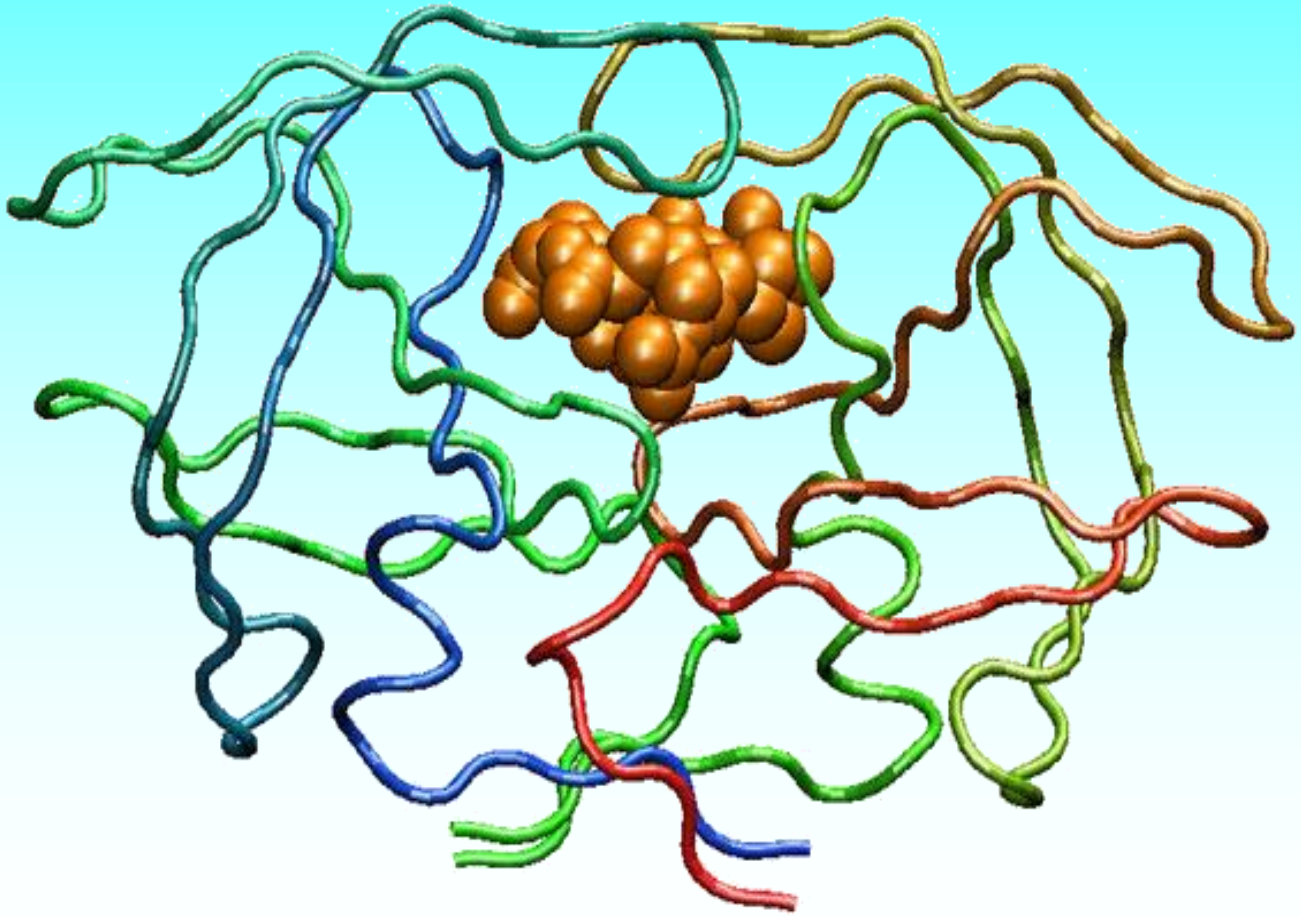
HIV-Protease



HIV-Protease



HIV-1 protease bound to an inhibitor (shown in orange)



Example

Multiple sclerosis

- **Definition:** MS is an autoimmune, chronic, inflammatory, demyelinating desease that affects central neural system.

- more common in women than in men. MS primarily affects adults, with an age of onset typically between 20 and 40 years,

- It affects about 2.5 million people worldwide. (In Bulgaria - 44,5 persons per 100 000).

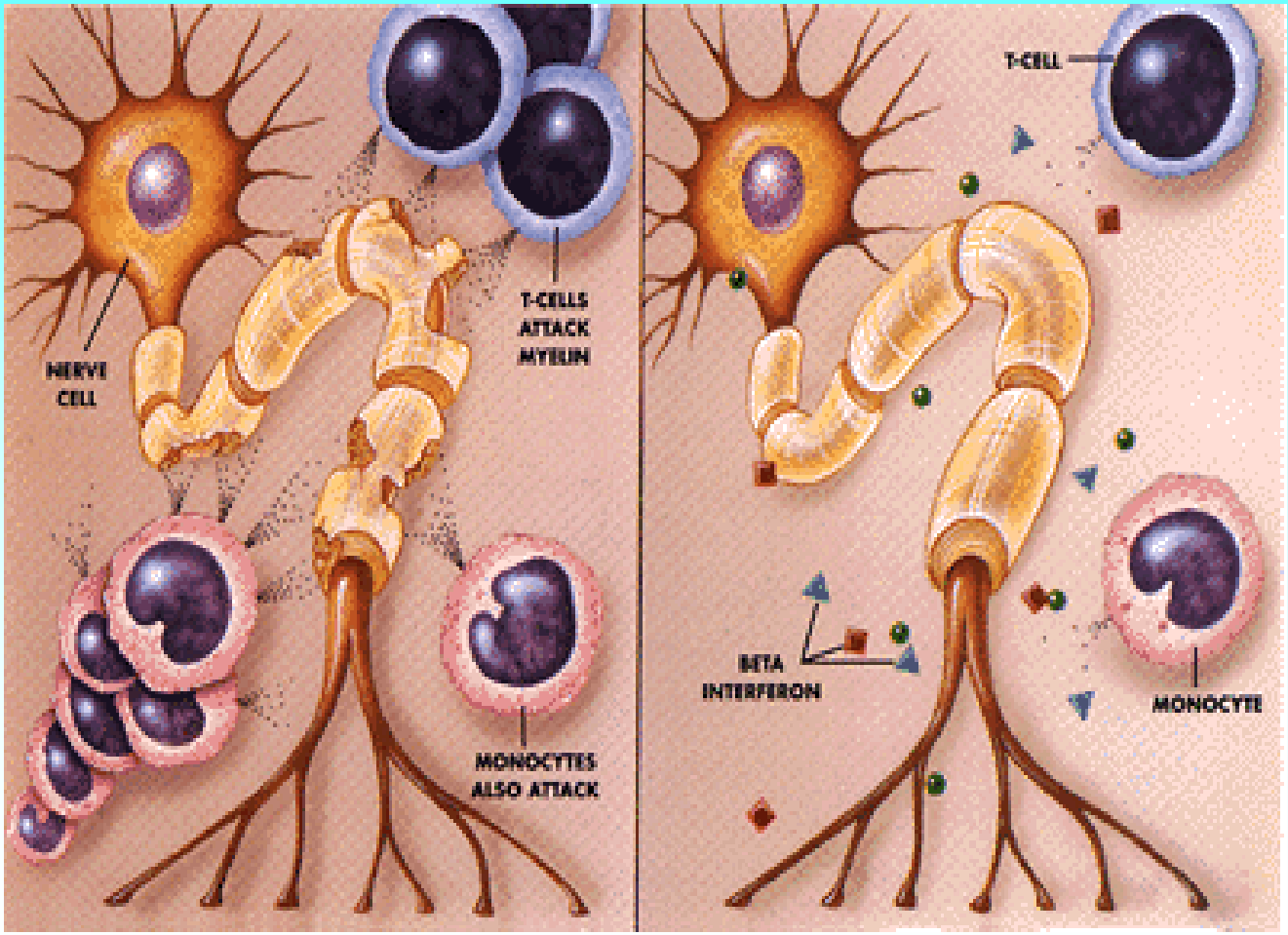
- **MS** starts with formication of limbs followed by muscle weakness and ends with complete disability.

The cause is UNKNOWN.

Multiple sclerosis

Multiple sclerosis is a disease in which the myelin (a fatty substance which covers the axons of nerve cells) degenerates.

T cells recognize myelin as foreign and attack it as if it were an invading virus. That triggers inflammatory processes, stimulating other immune cells and soluble factors like cytokines and antibodies. Leaks form in the blood-brain barrier. These leaks, in turn, cause a number of other damaging effects such as swelling, activation of macrophages, and more activation of cytokines and other destructive proteins such as matrix metalloproteinases. A deficiency of uric acid has been implicated in this process



MS therapy

INF-β is the only which influences the disease positively

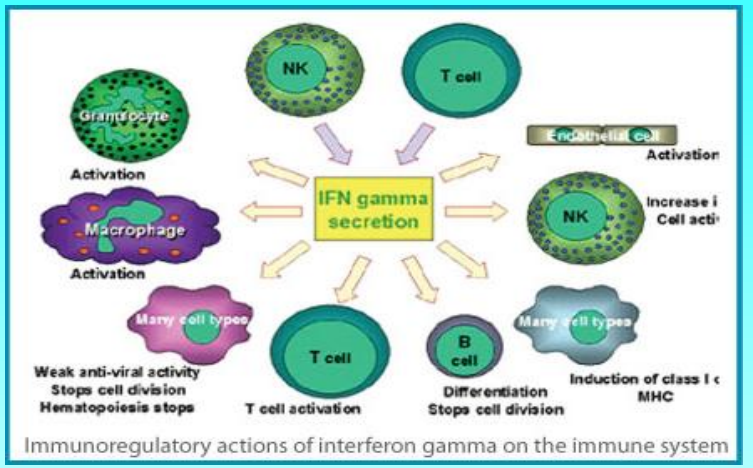
It reduces anti inflammatory cytokines and has an antiviral effect

IFN-β suppresses the **IFN-γ** production and alpha tumor necrotizing factor in the T cells. **IFN-γ** is an inhibitor of myelin synthesis in the oligodendritic cells

Interferon	Означение
Interferon-alfa	**IFN - α**
Interferon-beta	**IFN - β**
Interferon-gama	**IFN - γ**

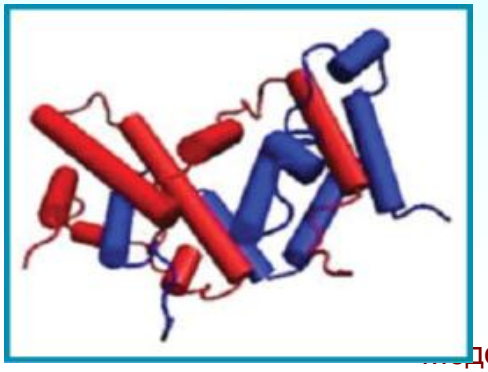
Interferon-Gamma

hIFNγ is a product of activated T lymphocytes and natural killer (NK) cells that was originally described as an antiviral agent



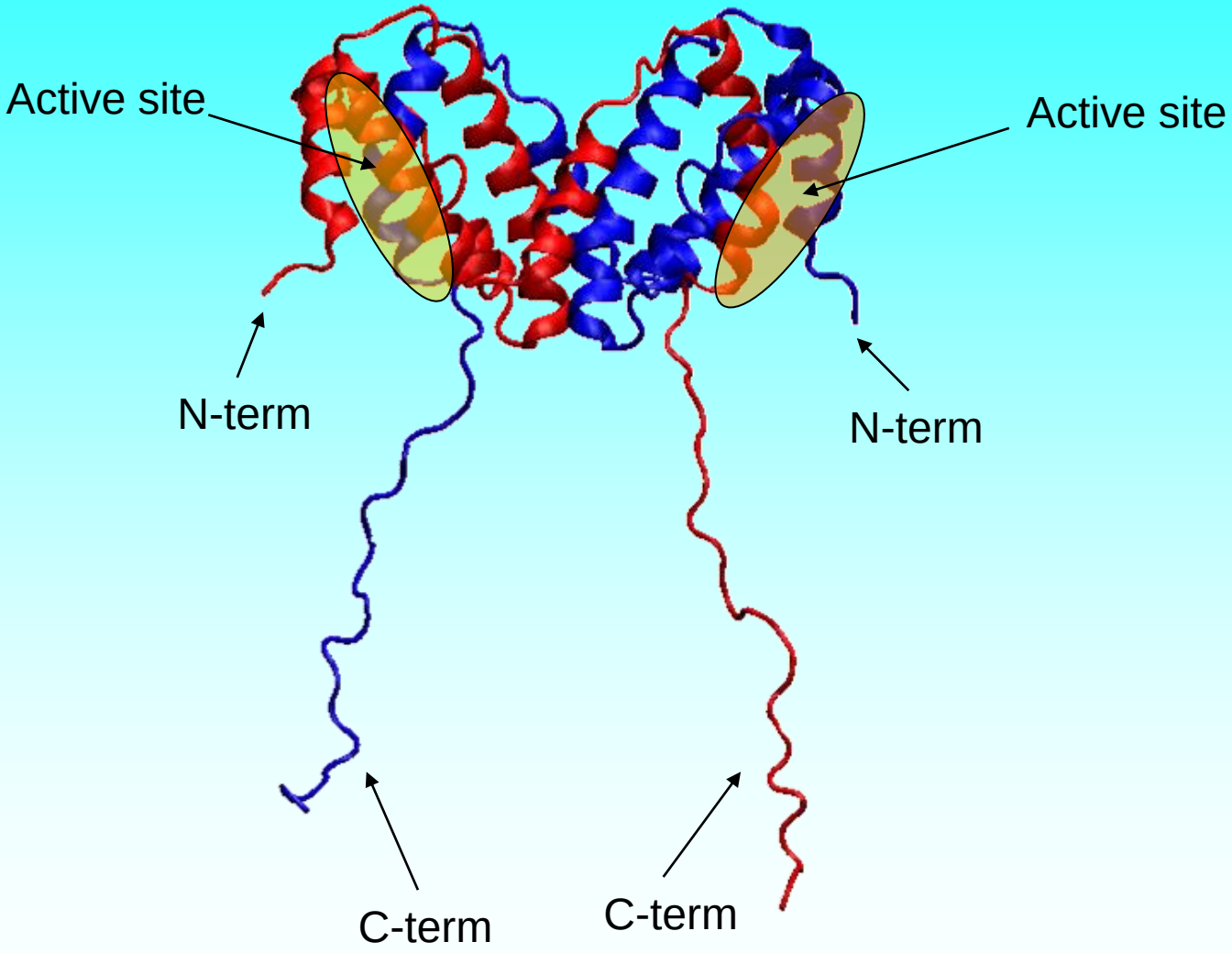
Immunoregulatory actions of interferon gamma on the immune system

The biologically active form of hIFNγ is a homodimer composed of two identical subunits of 143 amino acids each, related by a twofold symmetry axis



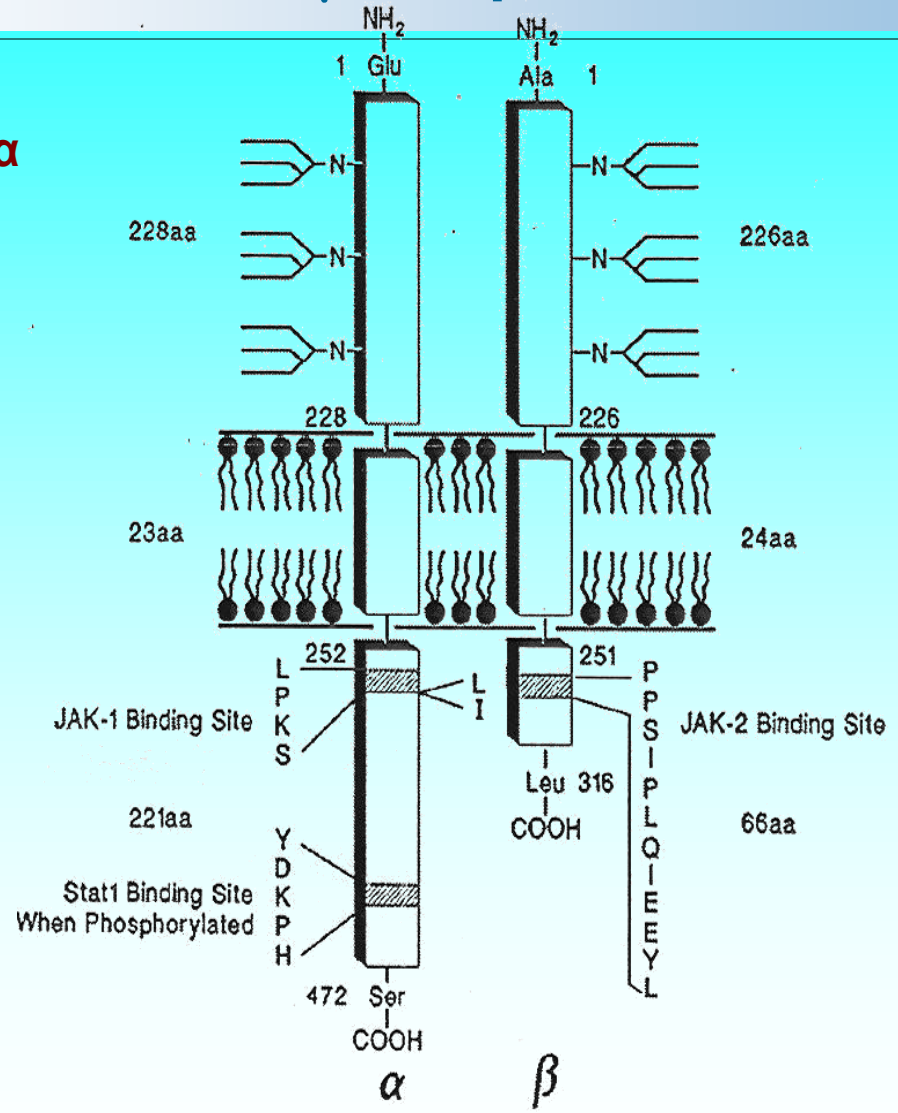
Expression of biological activity appears to be mediated through binding to specific cell-surface receptors

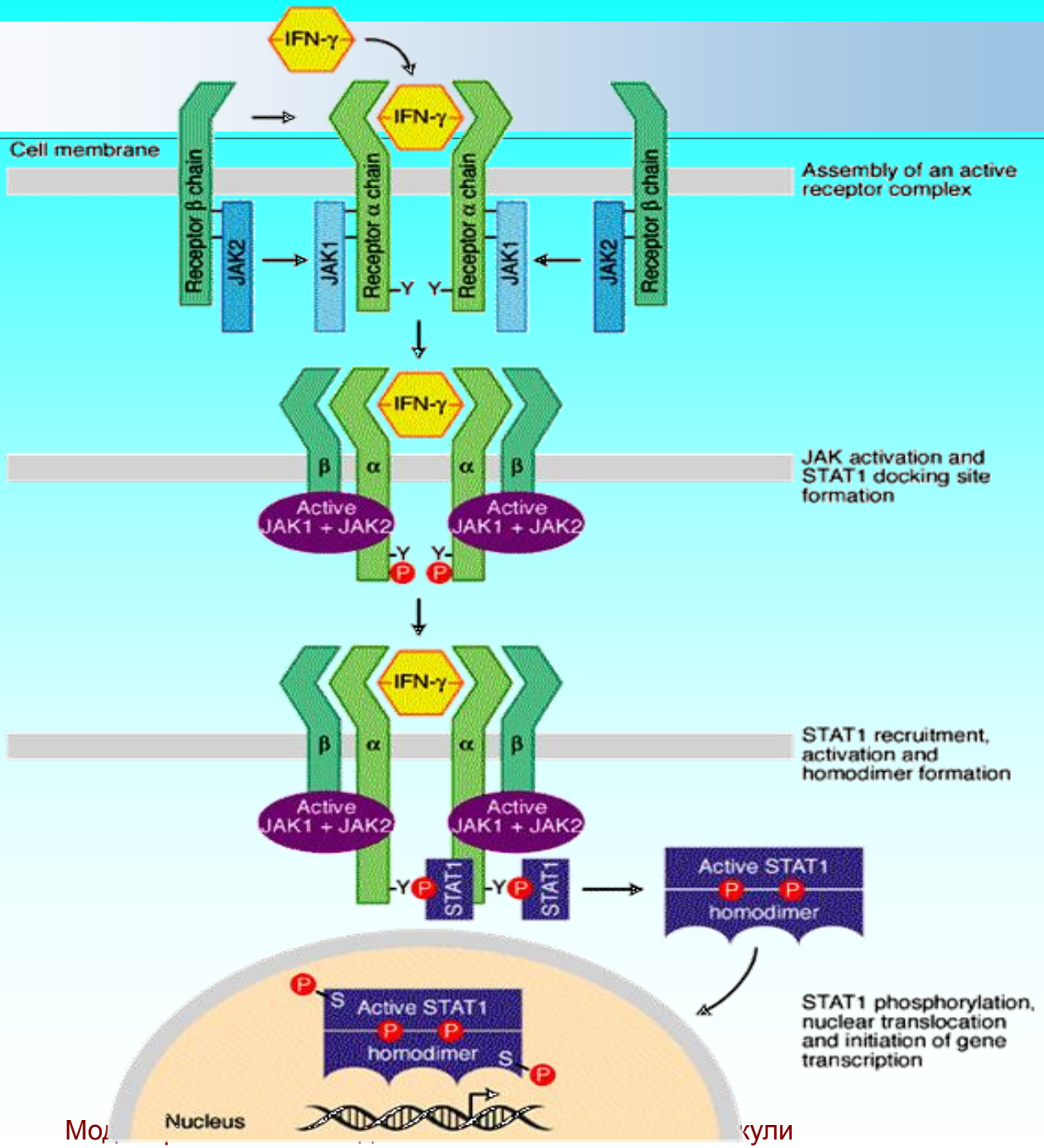
Interferon Gamma



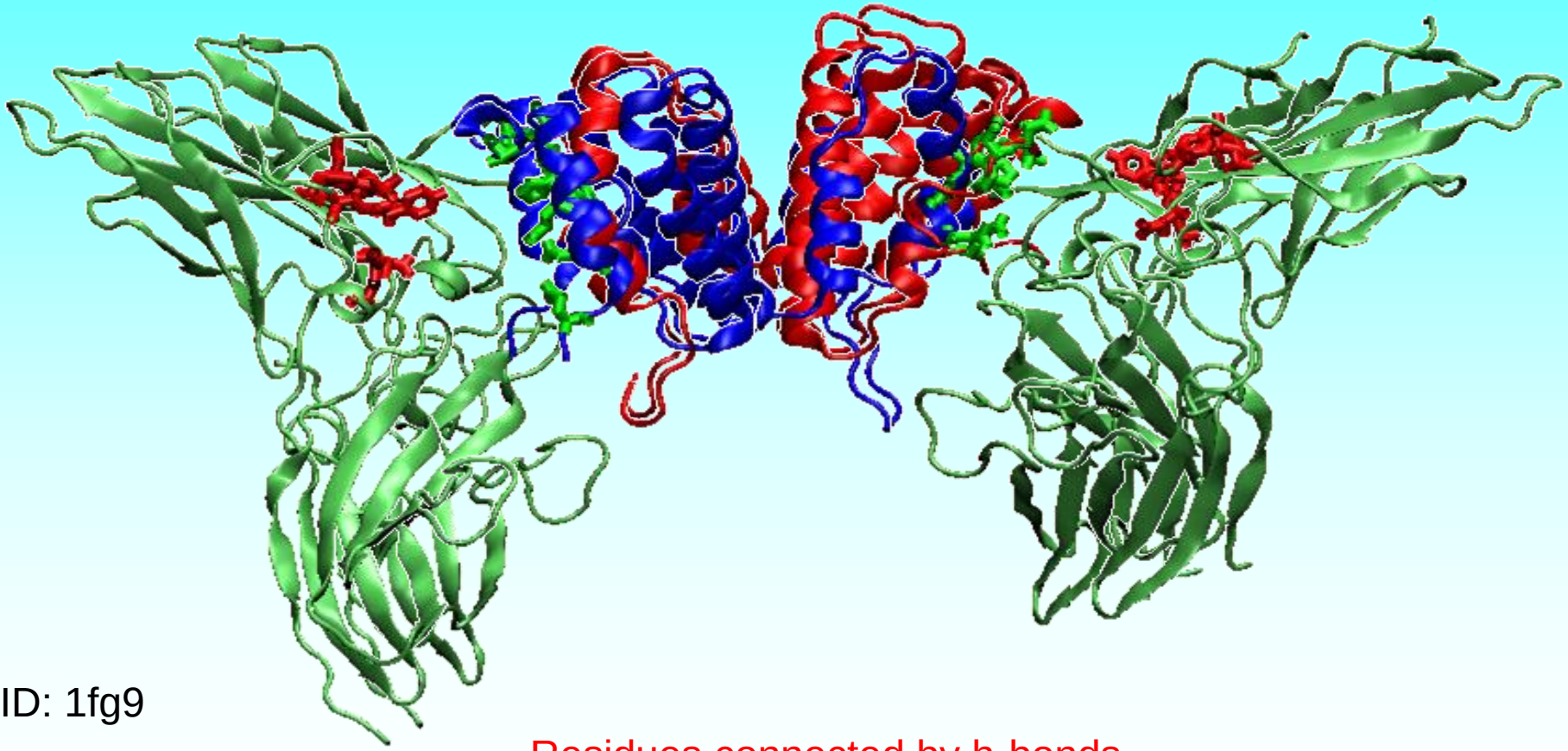
INF-γ receptors

IFN-γ is binding with two receptors, called **IFN-γRα** and **IFN-γRβ**.




IFN-γ
IFN-γ
Cell membrane
Receptor β chain
JAK2
JAK1
Receptor α chain
Y Y
Receptor α chain
JAK1
JAK2
Receptor β chain
Assembly of an active receptor complex
IFN-γ
β α α β
Active JAK1 + JAK2
Active JAK1 + JAK2
Y Y
P P
JAK activation and STAT1 docking site formation
IFN-γ
β α α β
Active JAK1 + JAK2
Active JAK1 + JAK2
Y P
STAT1
Y P
STAT1
STAT1 recruitment, activation and homodimer formation
Active STAT1
P P
homodimer
P
S
Active STAT1
P P
homodimer
S P
Nucleus
STAT1 phosphorylation, nuclear translocation and initiation of gene transcription

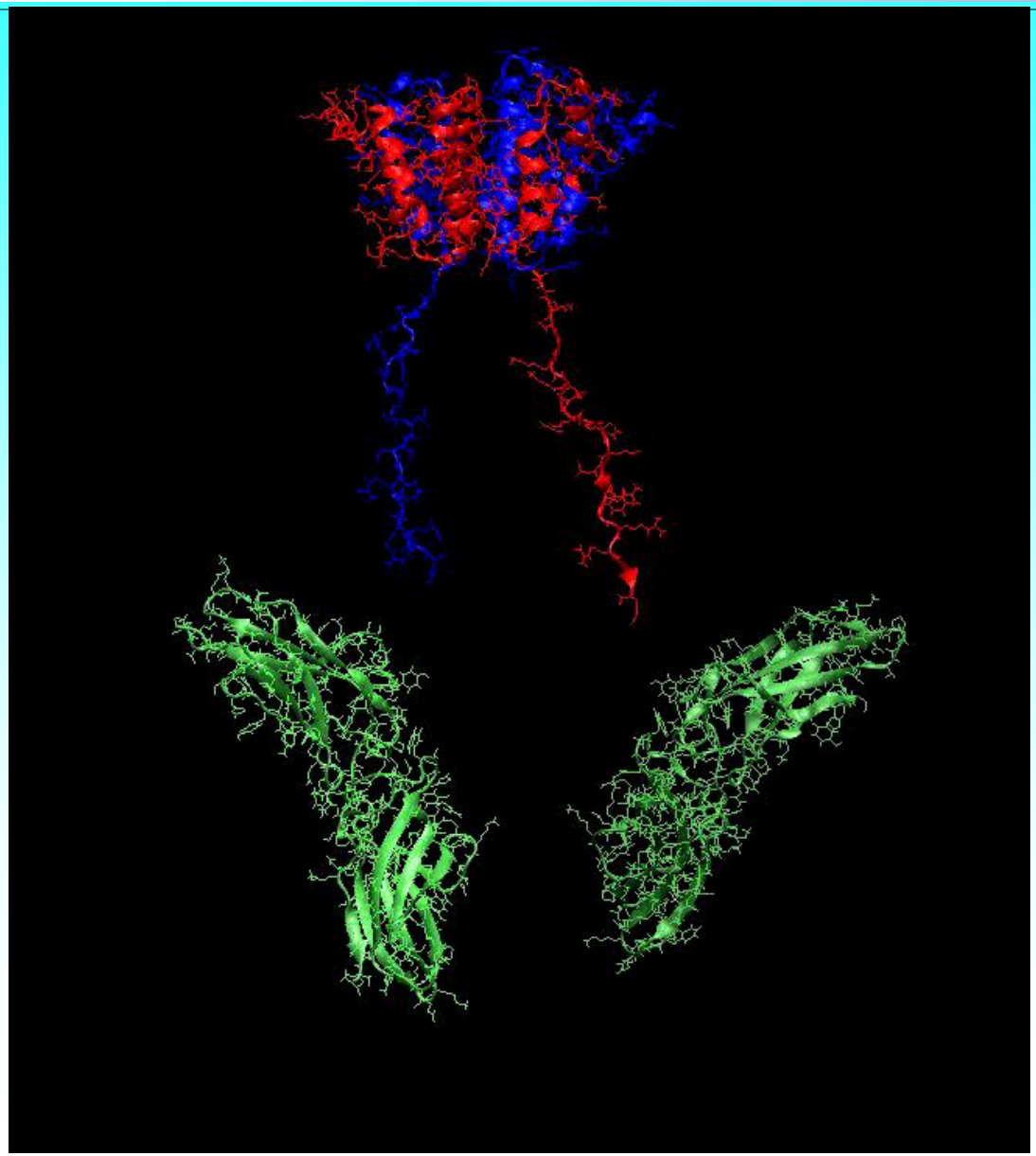
Interferon-gamma and its alpha receptor



PDB ID: 1fg9

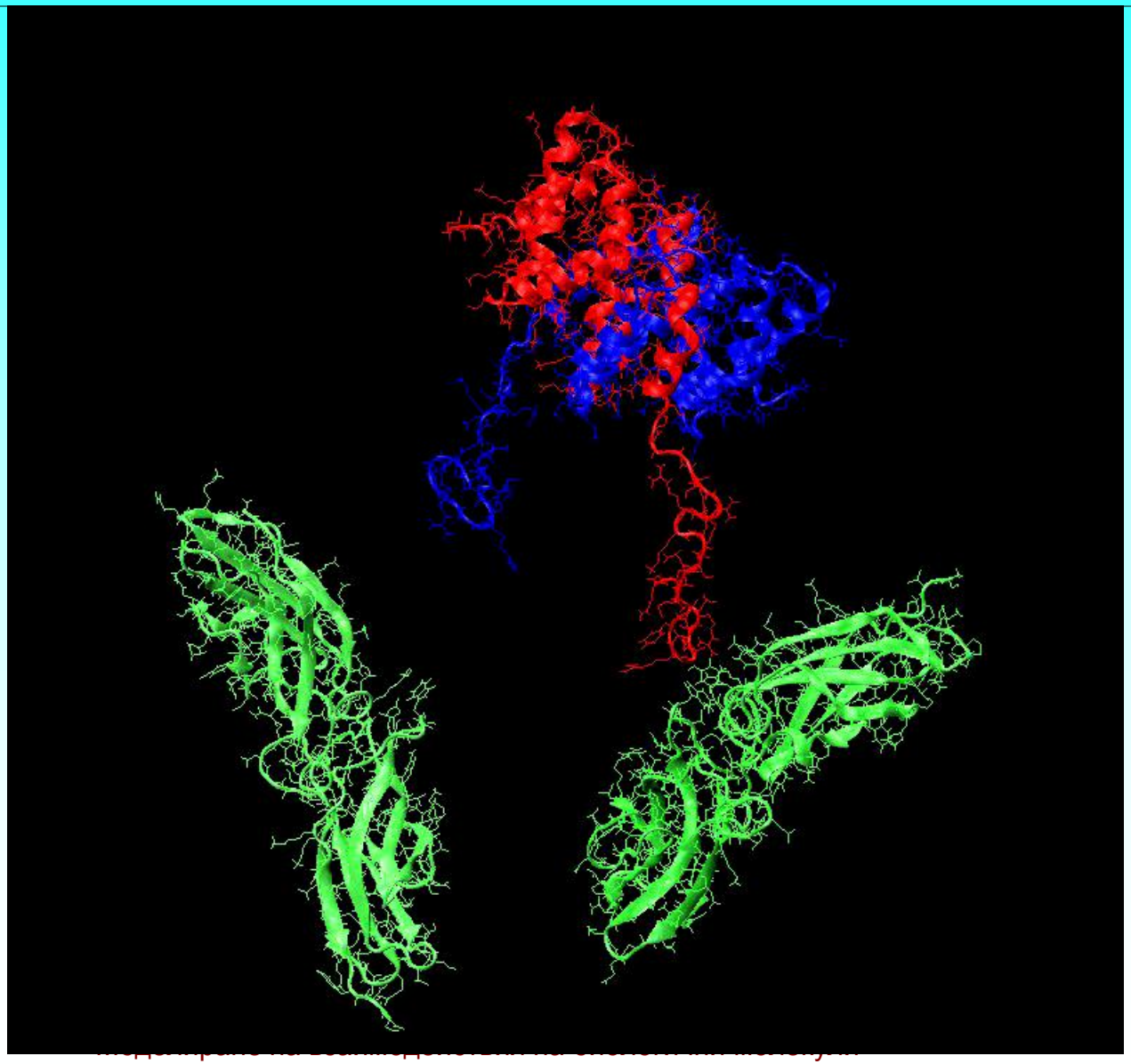
Residues connected by h-bonds

INF-γ simulations

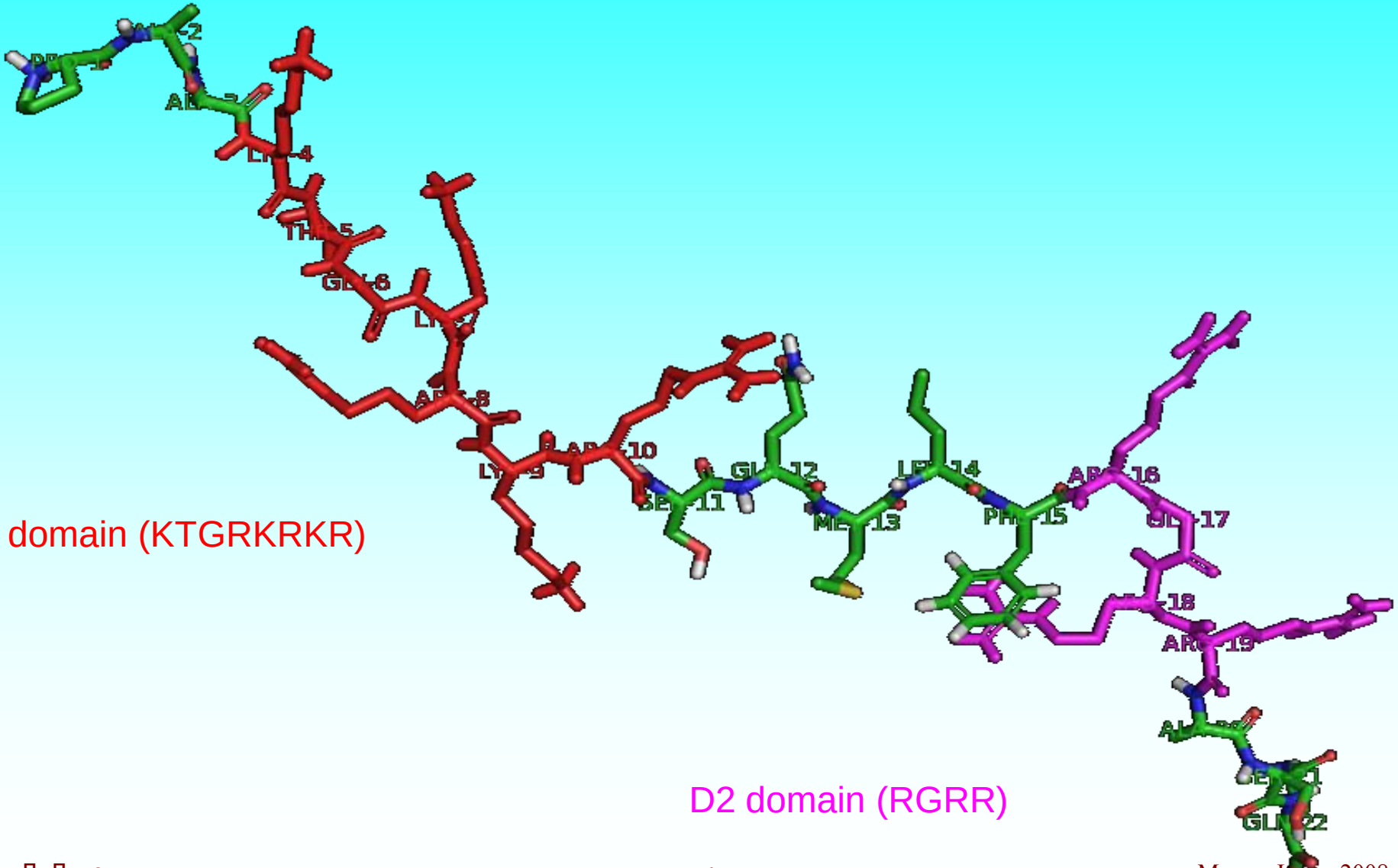


GROMACS

INF-γ simulations

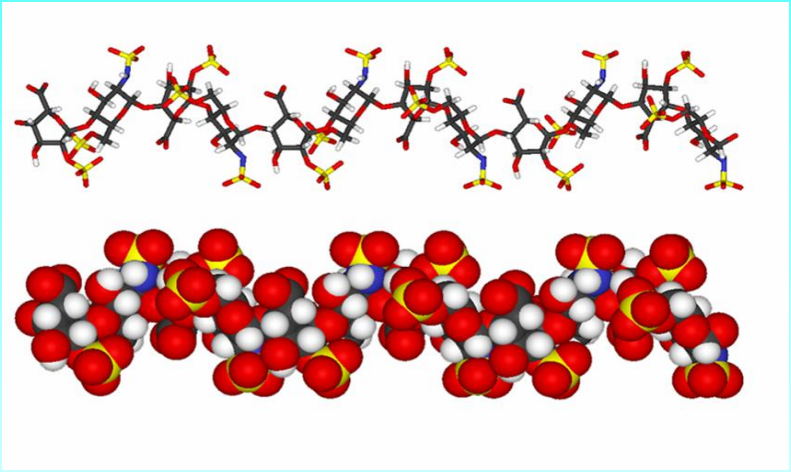


INF-γ C-term



INF-γ C-term

Heparin derived oligosaccharide

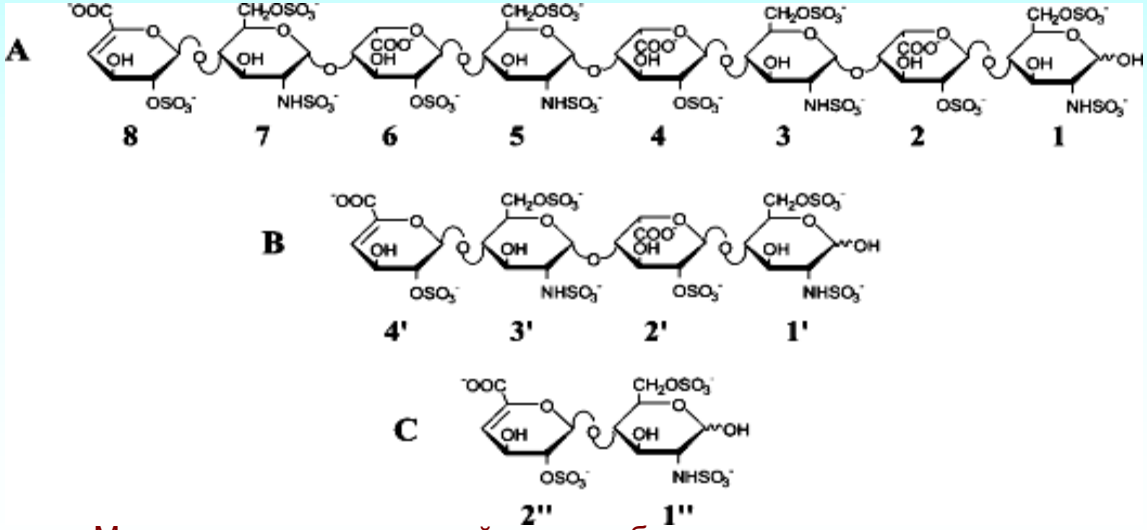


Biochem J. 2004 November 15; 384(Pt 1): 93–99.

NMR characterization of the interaction between the C-terminal domain of interferon-γ and heparin-derived oligosaccharides

Cécile Vanhaverbeke,*1 Jean-Pierre Simorre,* Rabia Sadir,† Pierre Gans,*2 and Hugues Lortat-Jacob†

PDB ID: 1hpn



d8

d4

d2

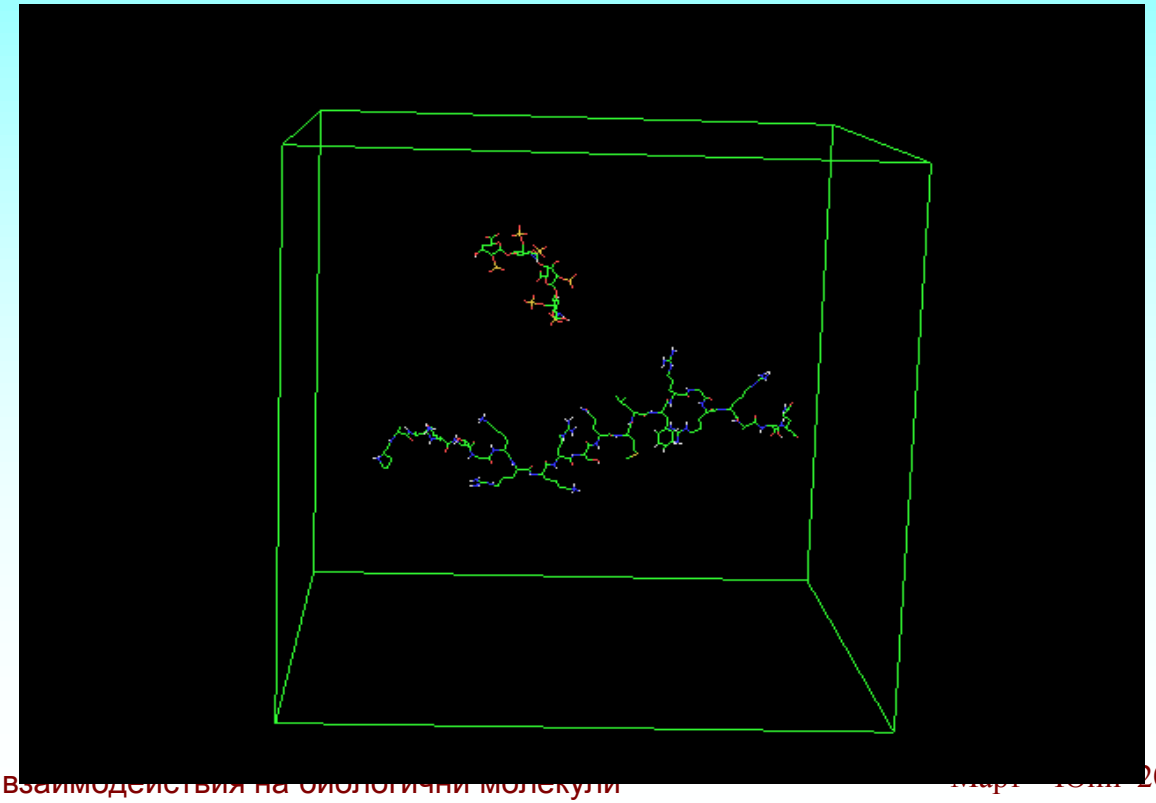
INF-γ C-term

Force field: GROMACS (gmx)

Software used: GROMACS

Topology builders:
pdb2gmx (GRAMCS)
&
PRODRG2 (http://davapc1.bioch.dundee.ac.uk/programs/prodrg/)

Simulation box: cubic with periodic boundary conditions



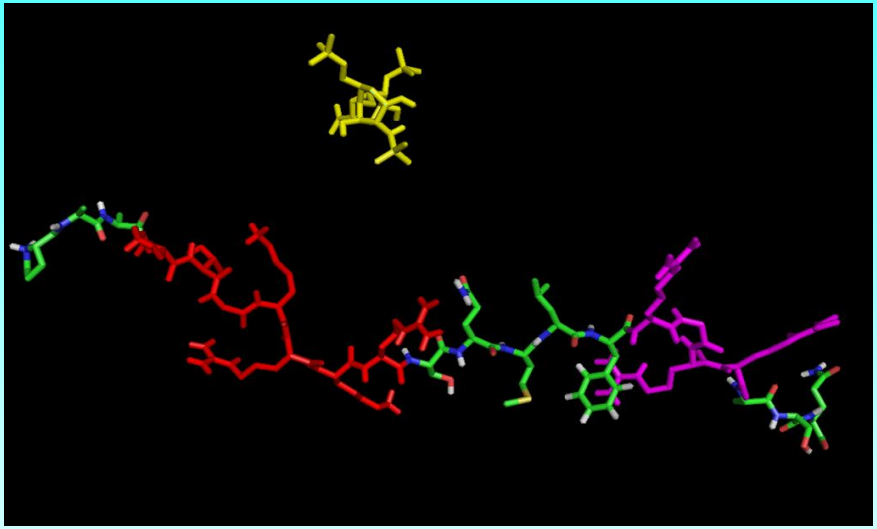
INF-γ C-term

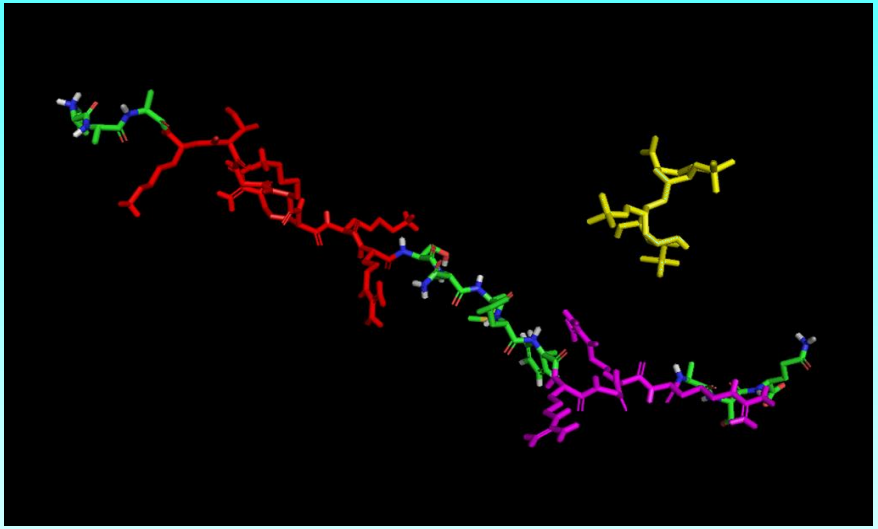
Configuration 1 (close to D1)

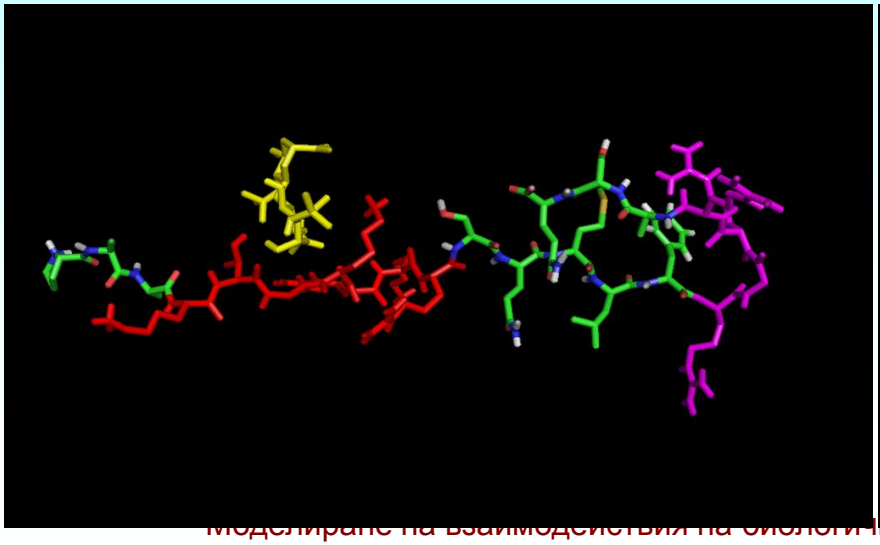
Configuration 2 (close to D2)

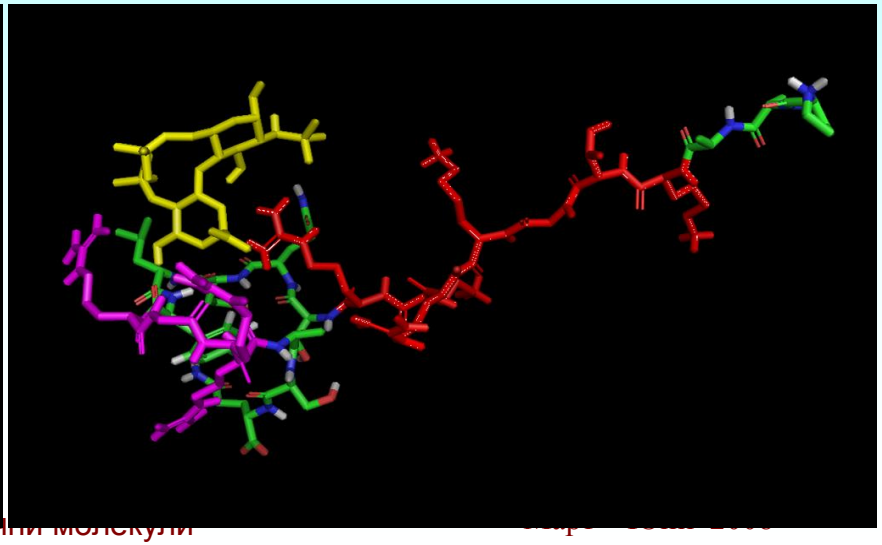
T = 0 ps

T = 1000 ps







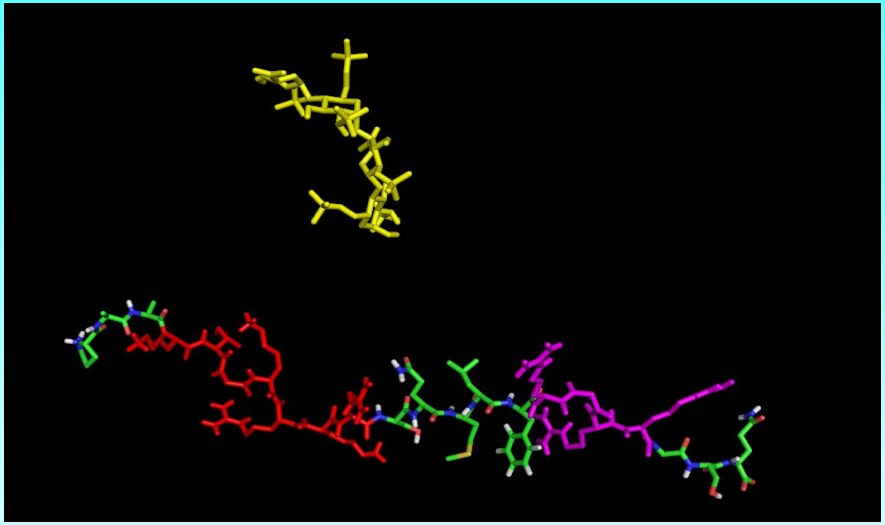


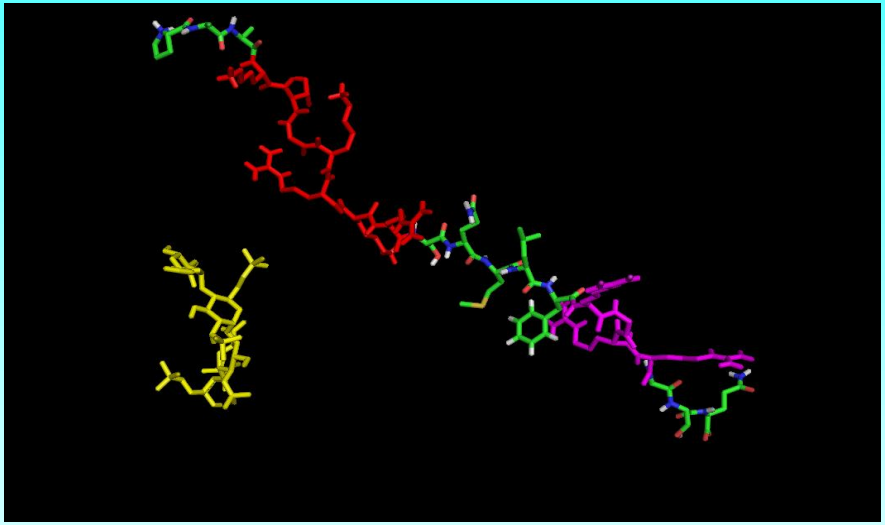
Configuration 1 (close to D1)

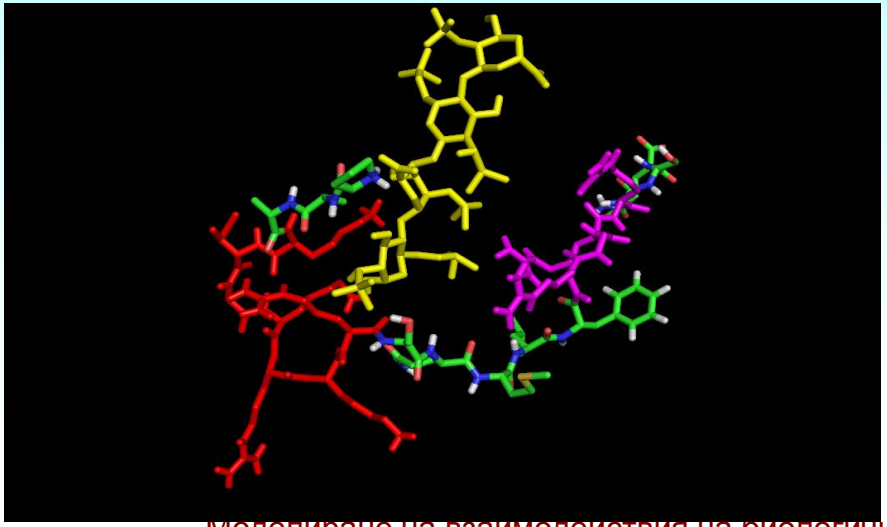
Configuration 2 (close to D1)

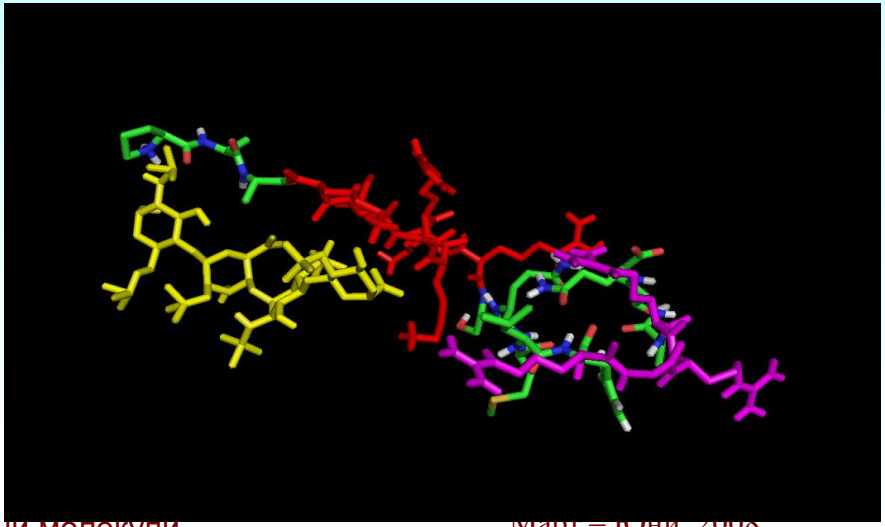
T = 0 ps

T = 2000 ps

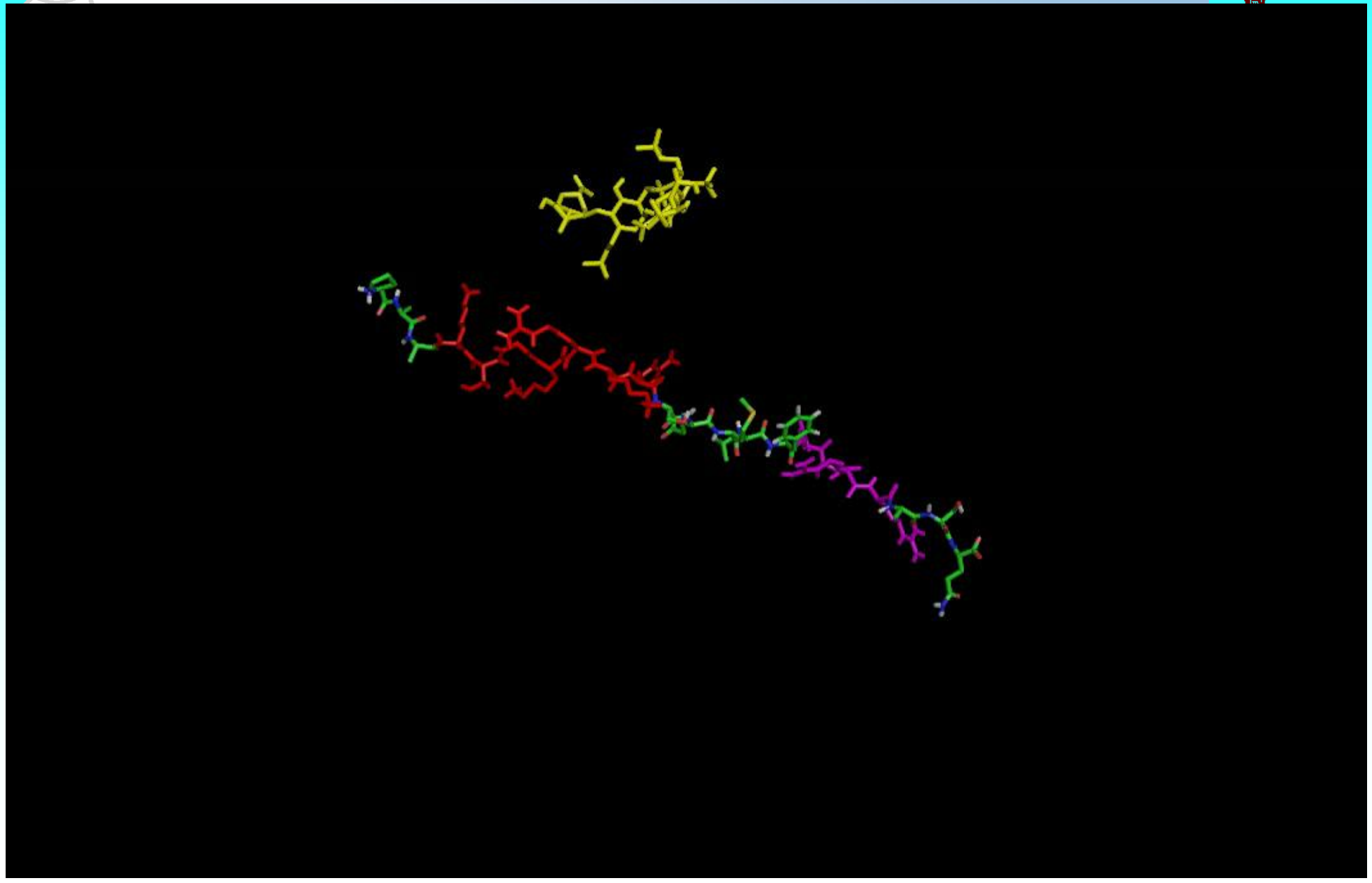








INF-γ C-term



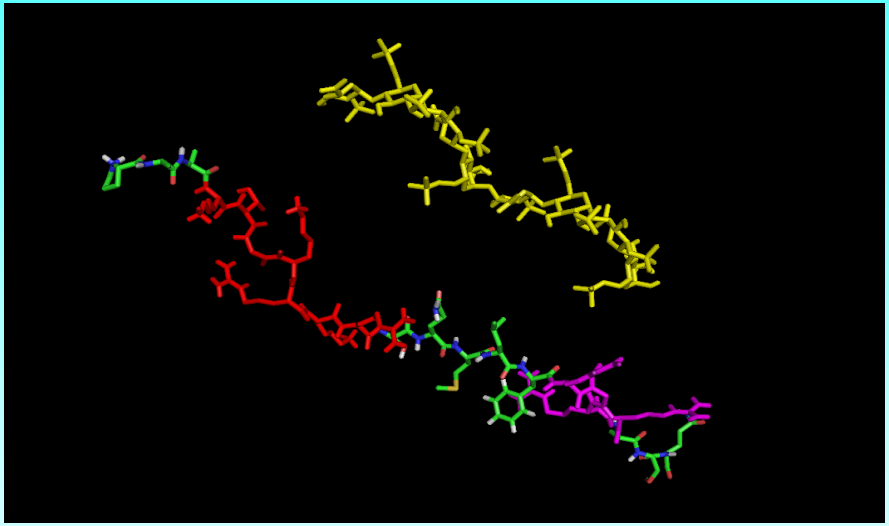
INF-γ C-term

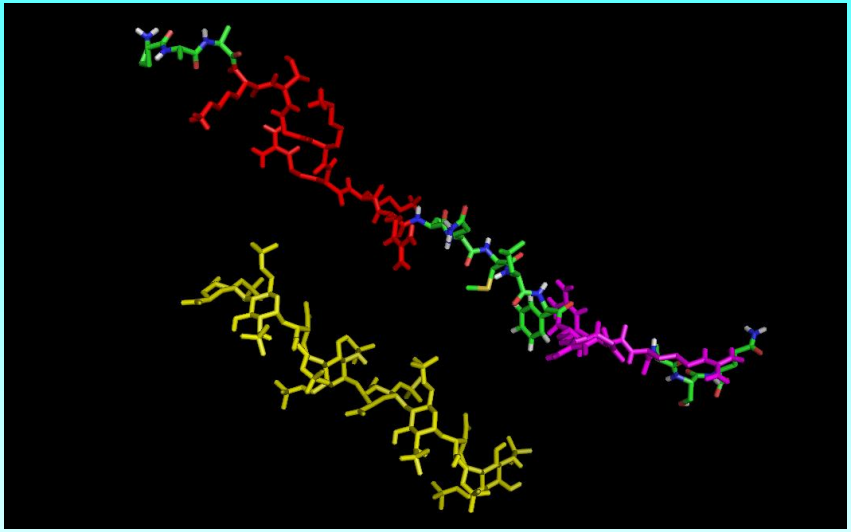
Configuration 1

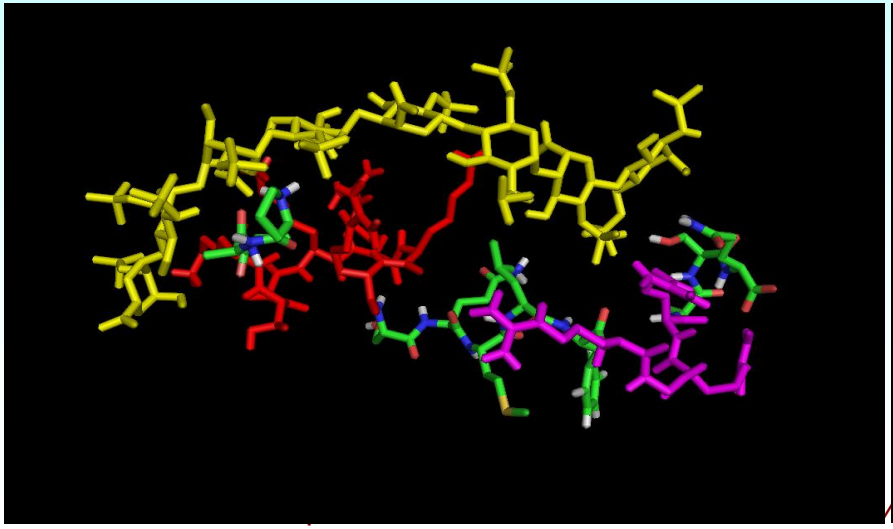
Configuration 2

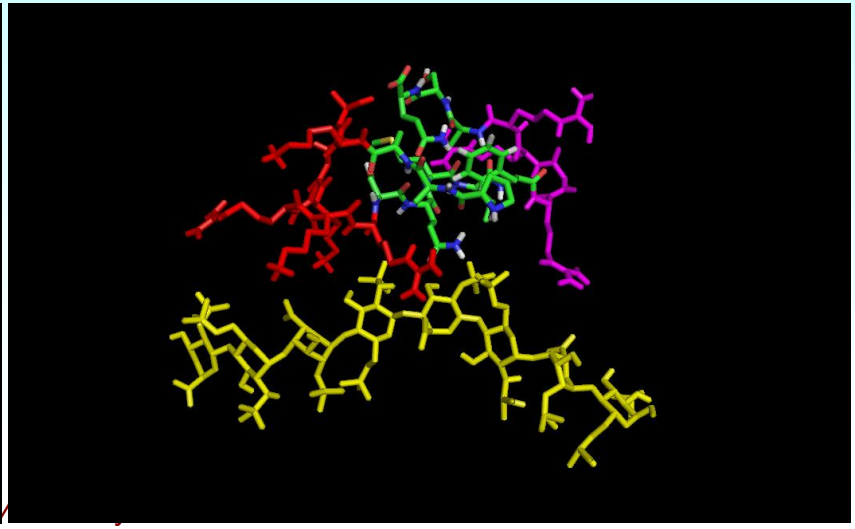
T = 0 ps

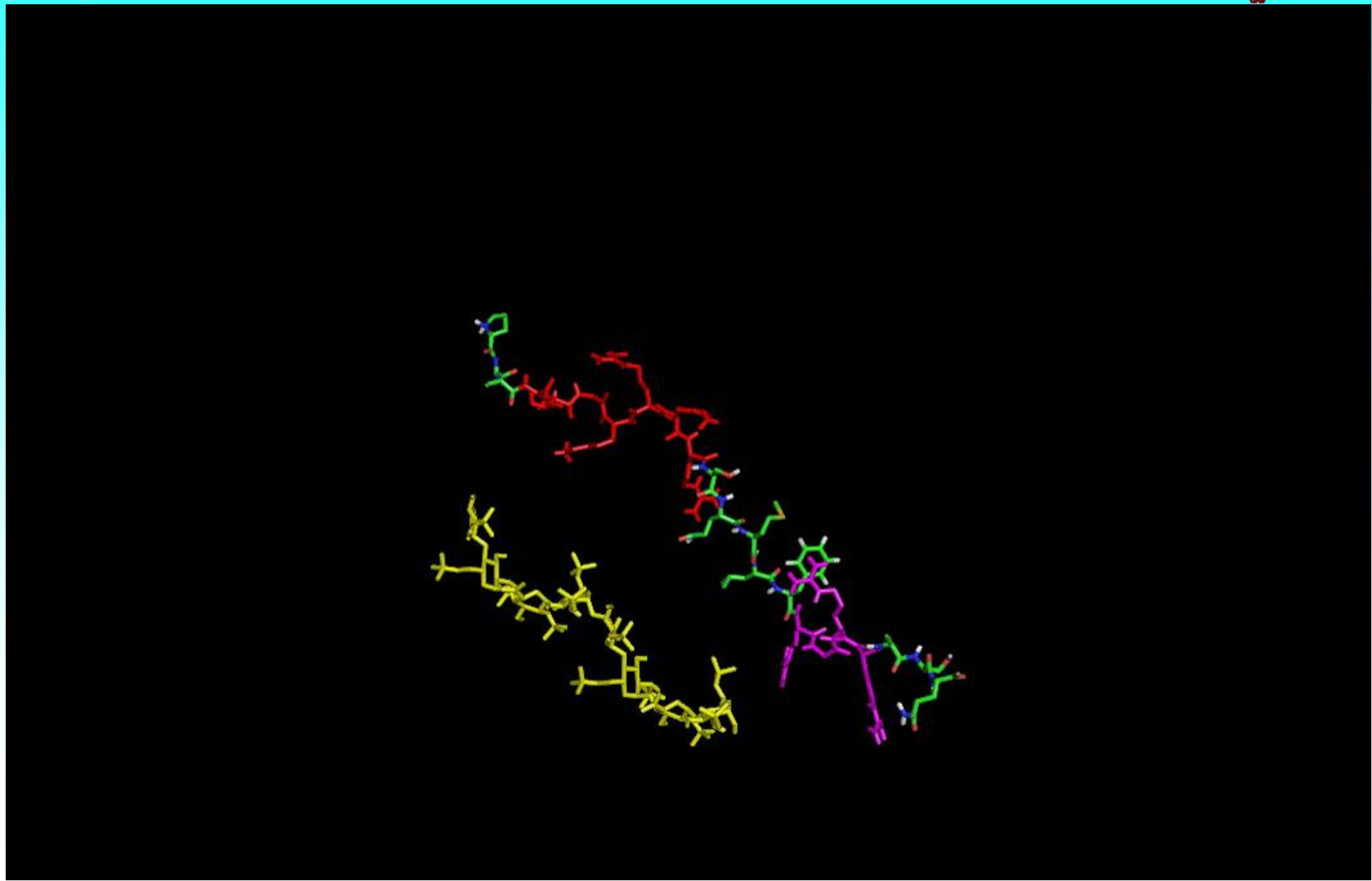
T = 2000 ps











GRID @ University of Sofia

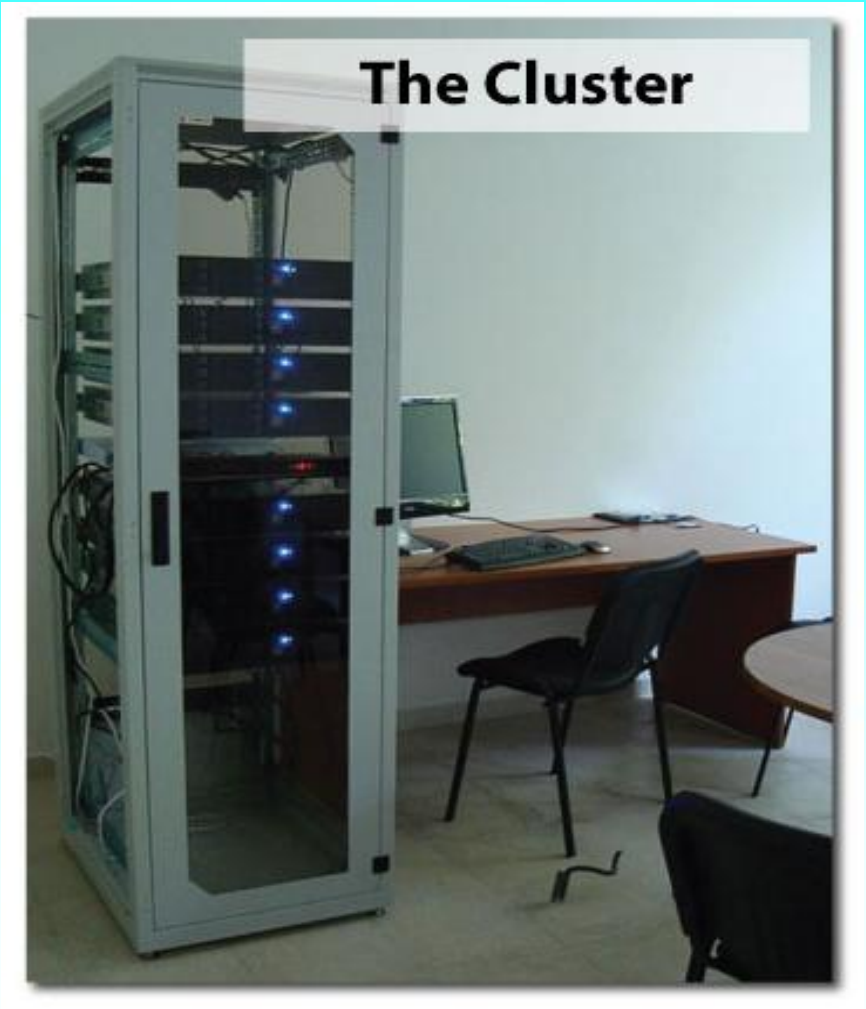
# servers	Services	Archietcture	CPU	RAM	Storage	OS
1	UI	i686	2 x 400MHz Pentium II	384MB	7 GB	Scientific Linux CERN Release 3.0.8 (SLC)
1	CE	i686	2 x 2.2 GHz AMD Athlon™ 64 X2 Dual Core	4 GB	250 GB	Scientific Linux CERN Release 3.0.8 (SLC)
1	SE	i686	2 x 2.8GHz Xeon	1GB	160 GB (RAID1)	Scientific Linux CERN Release 3.0.8 (SLC)
1	MON	i686	2 x 2.8GHz Xeon	1GB	160 GB (RAID1)	Scientific Linux CERN Release 3.0.8 (SLC)
1	WN	i686	2 x 1600MHz AMD Opteron	512MB	60 GB	Scientific Linux CERN Release 4.5 (SLC)
1	WN	i686	2 x 2.1GHz AMD Athlon	1GB	160 GB	Scientific Linux CERN Release 4.5 (SLC)
6	WN	i686	4 x 1.8GHz Dual Core AMD Opteron™	4 GB	140GB	Scientific Linux CERN Release 4.5 (SLC)

Computer Cluster – Physon Faculty of Physics

4 compute nodes:
- dual Intel Xeon E5335 (4 cores @ 2 GHz)
- 12 GB (16 GB @ node001) ECC DDR2-667 RAM
- 250 GB SATA2 HDD
- 2 x 1 Gbps Fast Ethernet
- 1 x 20 Gbps 4x DDR InfiniBand
- Scientific Linux 4.4 64-bit
- Sun N1 Grid Engine executives

NFS/NIS server:
- Intel Core2 Duo E6600 (2 cores @ 2,4 GHz)
- 2 GB ECC DDR2-667 RAM
- 4 x 500 GB SATA2 HDD (total of 1,75 TB in ZFS RAIDZ1 array)
- 2 x 1 Gbps Fast Ethernet
- Sun Solaris
- Sun N1 Grid Engine master

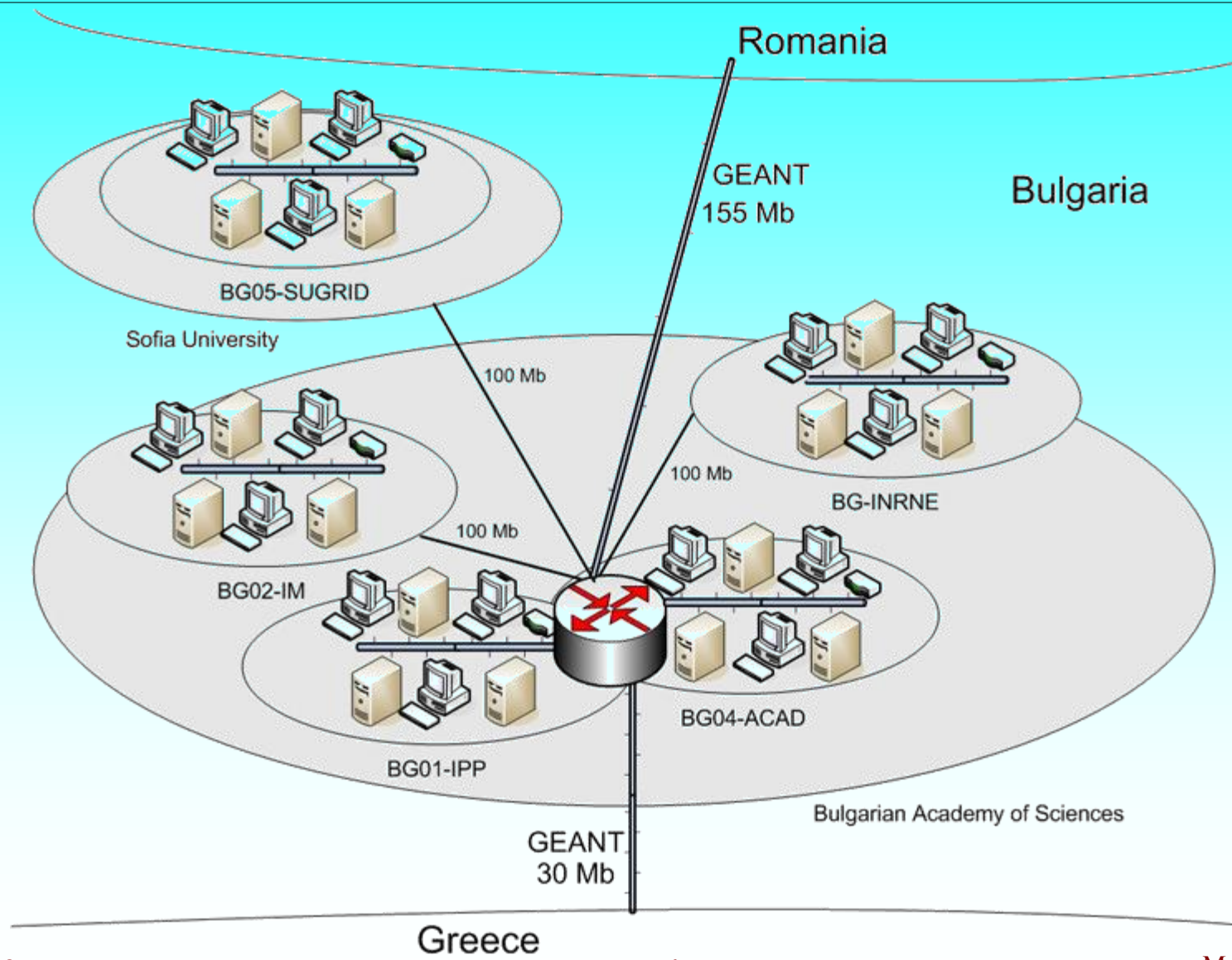
Technical University



- ☑ 8 x AMD Opteron 64 DualCore Processors
- ☑ 8 x 2GB RAM
- ☑ 8 x 2*160GB Hitachi SATA HDD
- ☑ 8 x Asus M2N-LR Mainboards
- ☑ ViewSonic 22" WideScreen LCD
- ☑ KVM Switch - DLink DKVM-8E
- ☑ Network Switch - 24port 1GBit/s 3COM 3c17300A

Institute for parallel processing

BG Grid


Romania
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GEANT
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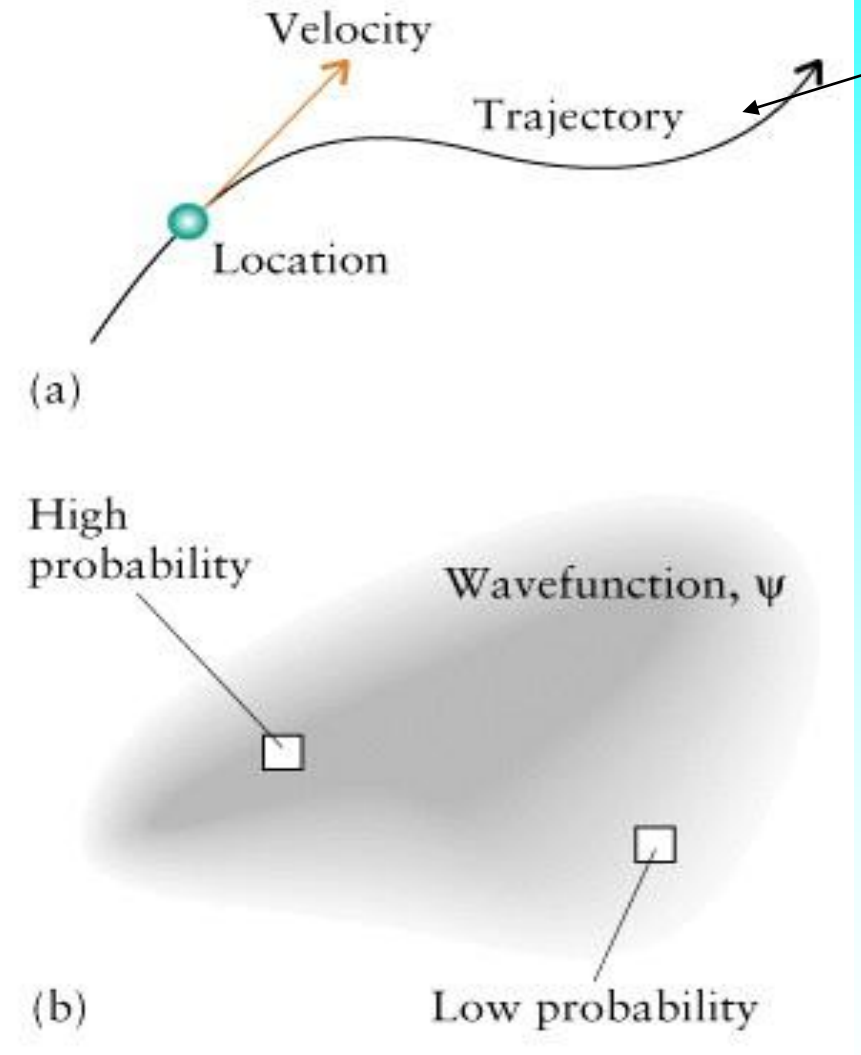
Greece

Молекулна динамика
Основни принципи

Механика



$\vec{x}(t)$ Закон за движение

Уравнение на Нютон

$$\vec{v}(t) = \frac{d}{dt}\vec{x}(t) \qquad \vec{F} = m\frac{d^2}{dx^2}\vec{x}(t)$$

Уравнение на Шрьодингер

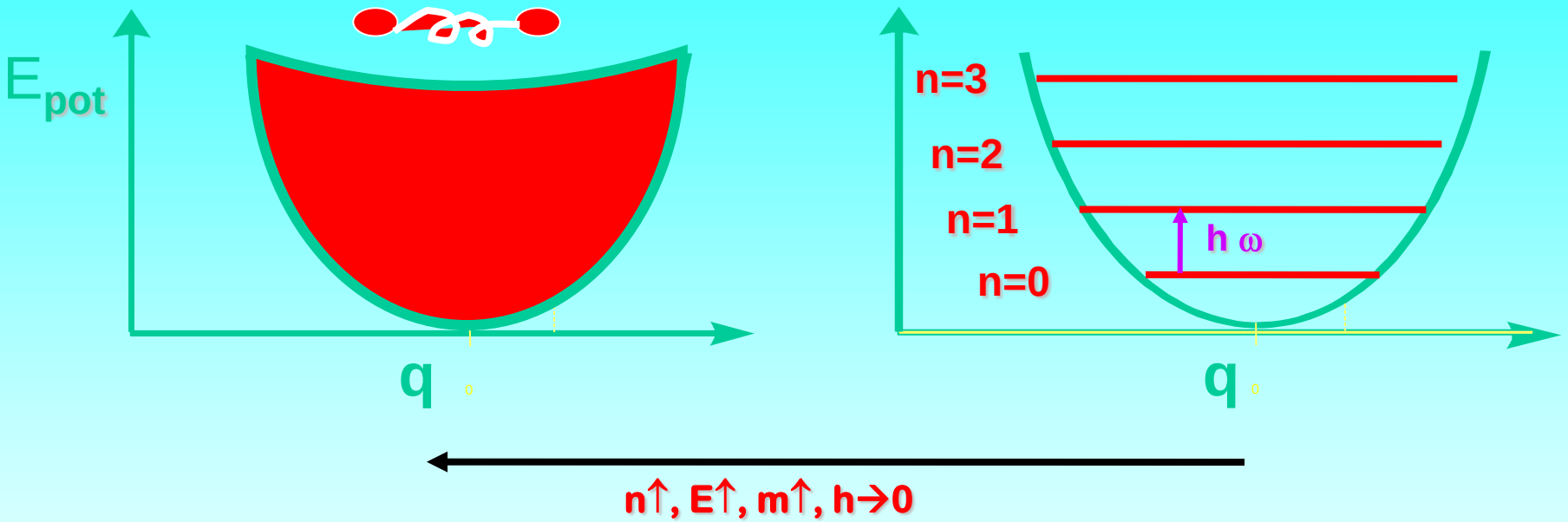
$$i\hbar\frac{\partial\Psi}{\partial t} = \hat{H}\Psi = -\frac{\hbar^2}{2m}\nabla^2\Psi + V(\boldsymbol{r},t)\Psi$$

$\Delta x.\Delta p \geq \hbar / 2$ Принцип на Хайзенберг

$P(x,t) = \Psi^*(x,t)\Psi(x,t)$ Вероятност системата да се намира в (r,t)

$\langle A\rangle = \int \Psi * \hat{A}\Psi \, d\boldsymbol{r}$ Средна стойност на A

A – физична величина



$$\frac{dx}{dt} = \frac{p}{m}$$

Нютон

Теорема на Еренфест

$$\frac{d\langle x\rangle}{dt} = \frac{\langle p\rangle}{m}$$

$$\frac{dp}{dt} = -\frac{dV(x)}{dx}$$

$$\frac{d\langle p\rangle}{dt} = -\langle\frac{dV}{dx}\rangle$$

Молекулна механика

- Молекулна механично описание на структурите – похват основан на класическата механика
- Прости уравнения описващи потенциала на взаимодействие между атомите в молекулите и между молекулните взаимодействия - молелулно-механично силово поле
- Метод за минимизиране на енергията на молекулни системи чрез вариране на геометрията
- Метод за пресмятане на времевата еволюция на атомите и молекулните системи - молекулна динамика (МД)

Молекулна динамика

Уравниения на Нютон за ядрата

$$F_n = m_n \ddot{x}_n = \nabla_{R_n} V\left(x_1, x_2, \cdots, x_N\right)$$

Числено интегриране

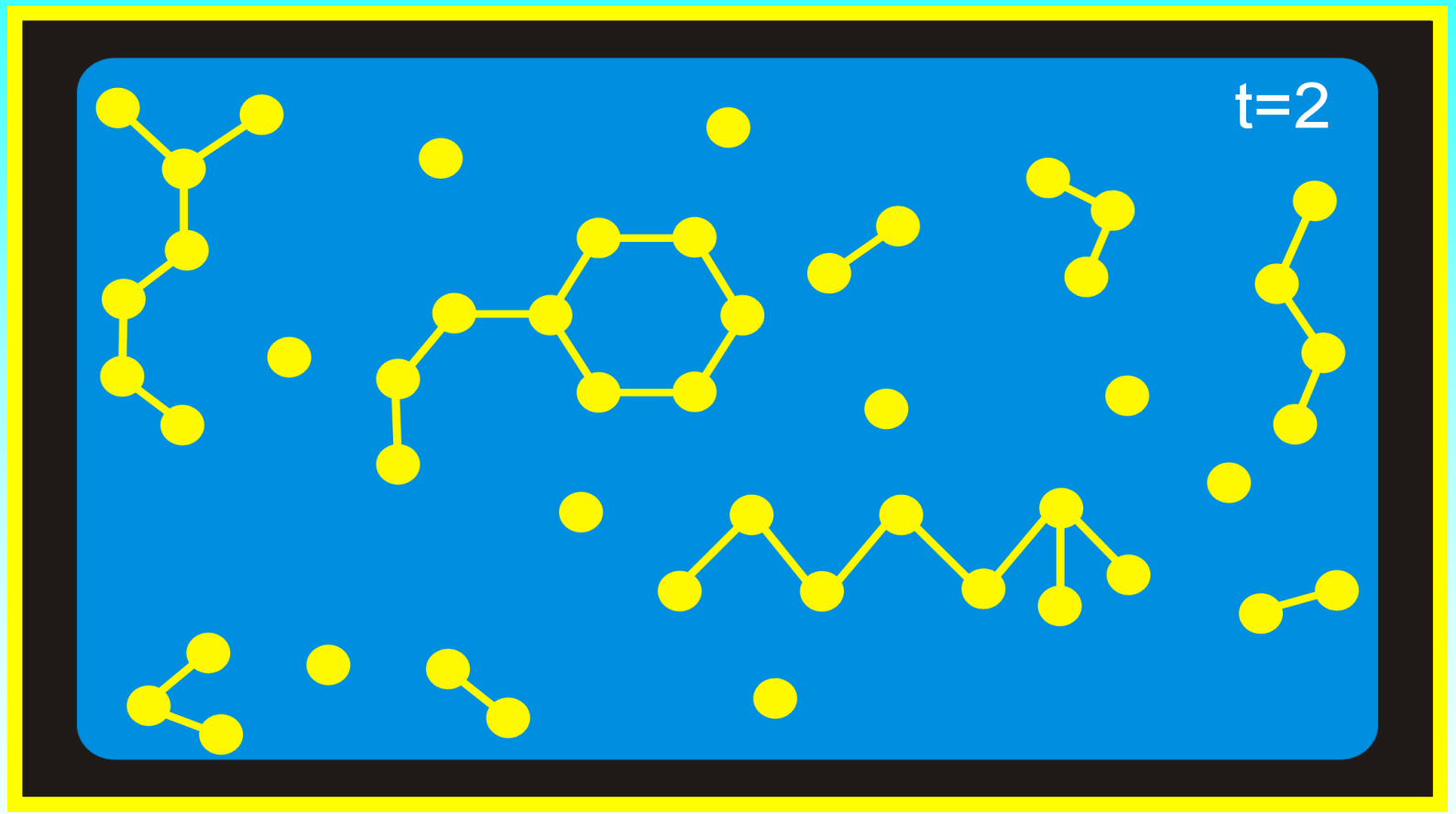
$$x_n(t) = x_n(t_0) + \dot{x}_n(t_0)(t - t_0) + \frac{1}{2}\ddot{x}_n^2(t - t_0)^2$$

$V\left(x_1, x_2, \cdots, x_N\right)$ – ***Потенциална енергия***

- Параметризиране на потенциалната енергия – молекулно-механично силово поле

$$V\left(x_1, x_2, \cdots, x_N\right) = \sum_k v_k\left(x; p_k\right)$$

Молекулна динамика



- Числено интегриране на уравненията на Нютон

$$\vec{x}_i(2\Delta t) = \vec{x}_i(0) + \vec{v}_i(0)\Delta t + \frac{\vec{F}_i(0)}{2m_i}\Delta t^2 + \frac{\vec{F}_i(\Delta t)}{2m_i}\Delta t^2$$

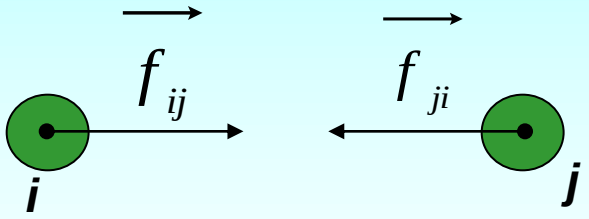
Модели

- ***Характеризират се от природата на взаимодействията между частиците***
- ***Използват се консервативни двучастични сили – изпълнен е закона за запазване на енергията и силата, действаща на една частица е суперпозиция от силите, с които и действат всяка една от другите частици в системата***

$$\vec{f}_{ij} = -\frac{\partial}{\partial \vec{x}_i} U(\vec{x}_i, \vec{x}_j)$$

$$\vec{f}_{ij} = -\vec{f}_{ji}$$

$$\vec{F}_i = \sum_{\substack{j=1 \\ i \neq j}}^{N} \vec{f}_{ij}$$



$$X_{eq} \equiv (\vec{x}_1, \vec{x}_2, \ldots, \vec{x}_N),$$

такава че

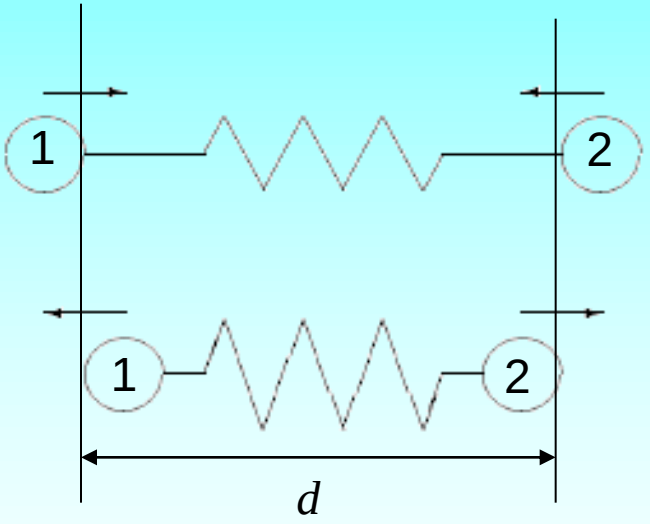
$$\vec{F}_i = 0 \text{ за } \forall\ i$$

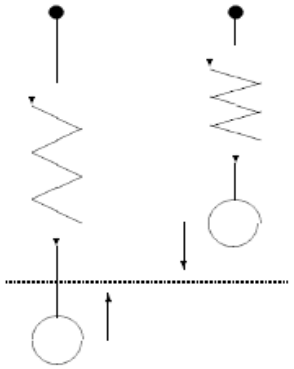
се нарича

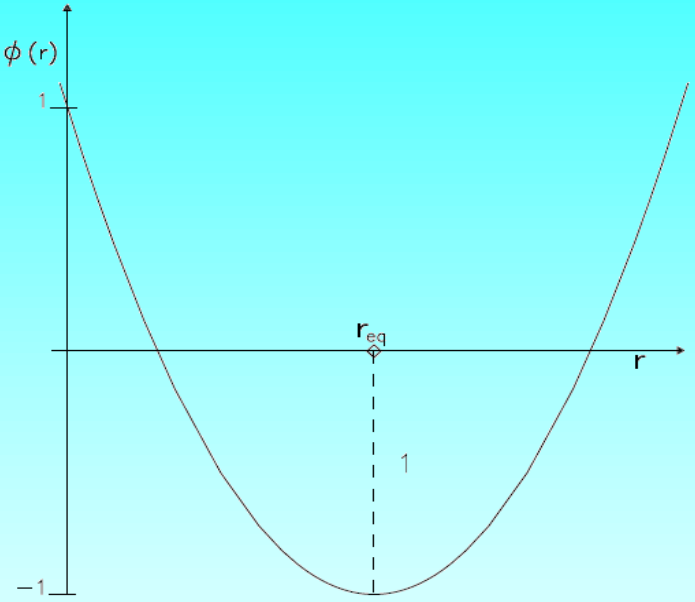
равновесна точка

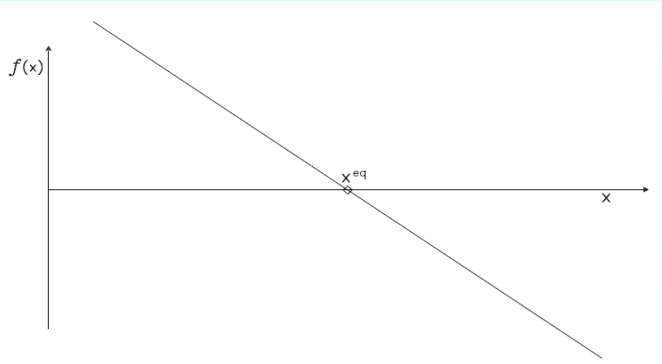
Закон на Хук

$$\phi(x) = \frac{k}{2}(x - x^{eq})^2 + \phi^{min}$$
$$f(x) = -k(x - x^{eq}),$$



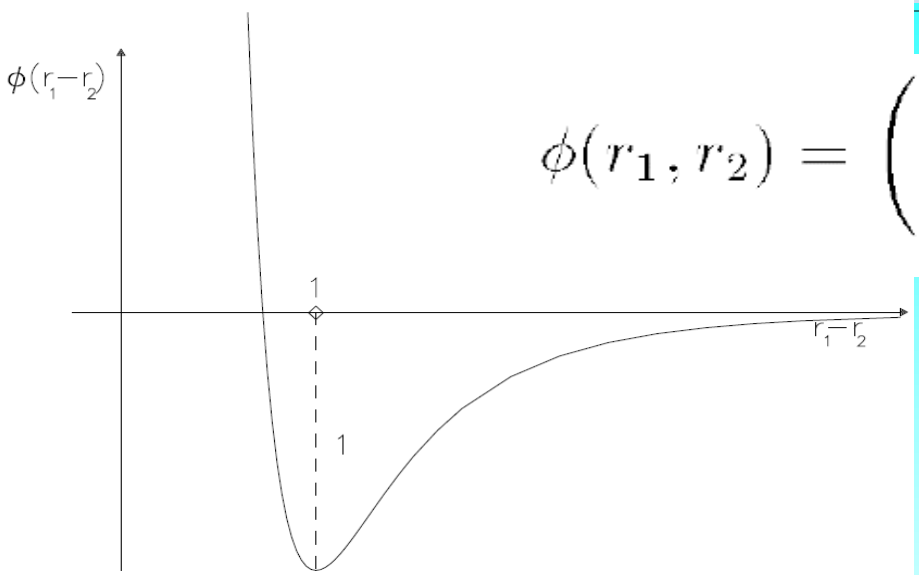




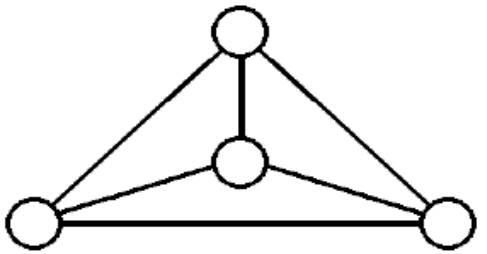


$$f_1(x_1, x_2) = -k(x_1 - x_2 + d),$$
$$f_2(x_1, x_2) = k(x_1 - x_2 + d).$$

Ленард-Джонс



$$\phi(r_1, r_2) = \left(\frac{1}{\|r_1 - r_2\|^{12}} - \frac{2}{\|r_1 - r_2\|^{6}} \right)$$



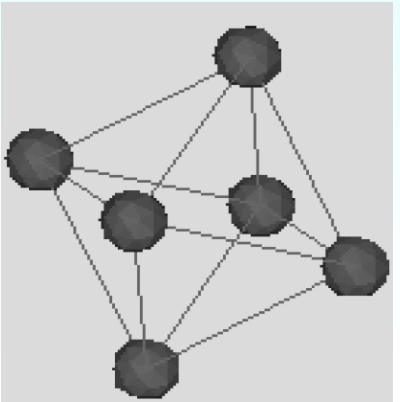
Четиричастична конфигурация с минимална енергия

$$f_{1,x} = 12 \left(\frac{1}{\|r_1 - r_2\|^{14}} - \frac{1}{\|r_1 - r_2\|^{8}} \right) (x_1 - x_2)$$

$$f_{1,y} = 12 \left(\frac{1}{\|r_1 - r_2\|^{14}} - \frac{1}{\|r_1 - r_2\|^{8}} \right) (y_1 - y_2)$$

$$f_{1,z} = 12 \left(\frac{1}{\|r_1 - r_2\|^{14}} - \frac{1}{\|r_1 - r_2\|^{8}} \right) (z_1 - z_2)$$

Шестчастична конфигурация с минимална енергия

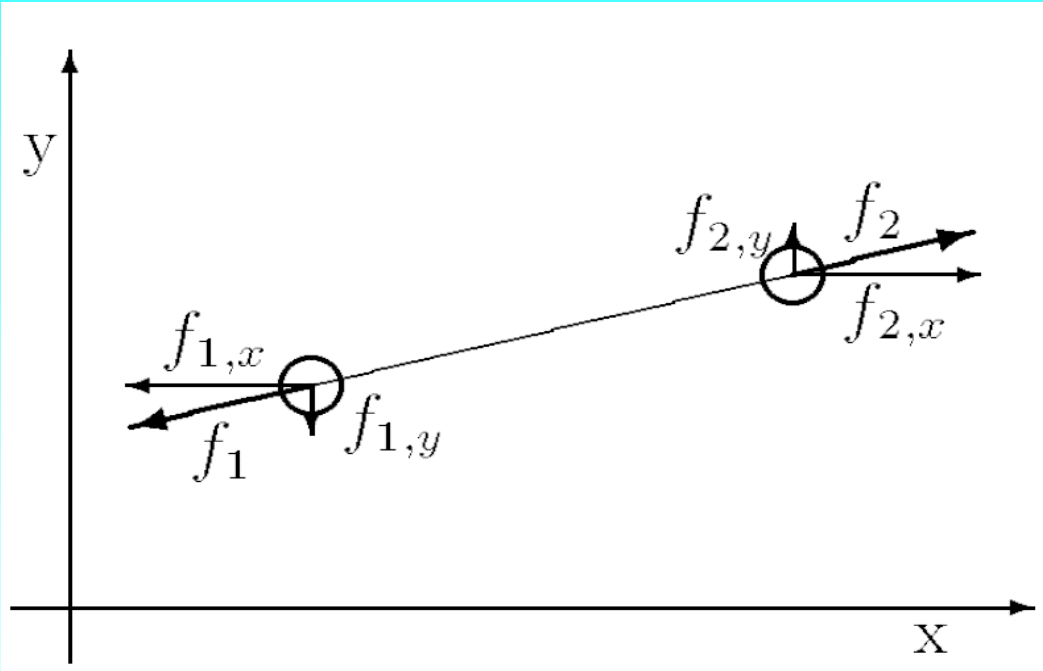


Уравнение на движение - 2D случай

$$m\ddot{r} = f$$

$$r = \begin{pmatrix} r_1 \\ r_2 \end{pmatrix} = \begin{pmatrix} x_1 \\ y_1 \\ x_2 \\ y_2 \end{pmatrix}$$

$$m \begin{pmatrix} \ddot{x}_1 \\ \ddot{y}_1 \\ \ddot{x}_2 \\ \ddot{y}_2 \end{pmatrix} = \begin{pmatrix} f_{1,x} \\ f_{1,y} \\ f_{2,x} \\ f_{2,y} \end{pmatrix}$$



$$m \begin{pmatrix} \ddot{r}_1 \\ \ddot{r}_2 \\ \vdots \\ \ddot{r}_n \end{pmatrix} = \begin{pmatrix} f_1 \\ f_2 \\ \vdots \\ f_n \end{pmatrix}$$

За n-частици

$$m\ddot{x} = f(x)$$

$$\begin{aligned} m\dot{u} &= f(x) \\ \dot{x} &= u \end{aligned}$$

Начални условия

$$x(0) = x^{(0)}$$

$$\dot{x}(0) = \dot{x}^{(0)}$$

Радиус вектора и скоростта след стъпка h

$$\begin{aligned} x(t+h) &\approx x(t) + hu(t) \\ u(t+h) &\approx u(t) + \frac{h}{m} f(x(t)) \end{aligned}$$

Алгоритъм на Верле

$$\ddot{x} \approx \frac{x(t+h) - 2x(t) + x(t-h)}{h^2}$$

$$x(t+h) \approx 2x(t) - x(t-h) + h^2 f(x(t))$$