Lectures 11-13:
Electrostatics of Salty Solutions

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Water as a medium of life: H_2O dissociation into H^+ and OH-

- → in pure water: 1 H_2O molecule out of 10^7 is dissociated
- → definition of the $pH = -\log_{10} [H^+]$
- → thus, for pure water pH = 7
- → the presence of ions or other molecules in H_2O can change pH
- → different amino acids can donate or accept H^+, thus their charged state depends on pH and that affects the protein structure
- → example of an influenza virus uptake by a cell:

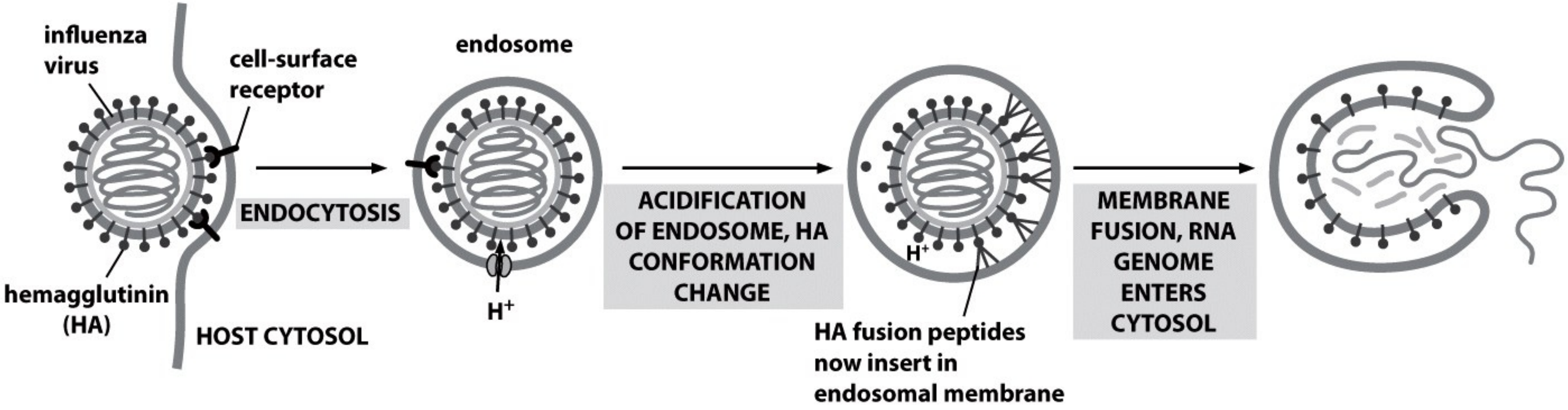


Figure 9.1 Physical Biology of the Cell (© Garland Science 2009)

Variation of pH inside the body:

- pH of the blood is at 7.3
- inside the cell, e.g. lysosome (protein degradation) pH<4
- cells manipulate pH by H^+ transport across the membrane
- in bacterium: 2×10^{10} H_2O molecules

  10^7 ions (mM concentrations): Na^+, K^+, …
- equilibrium constant and pH: charge separation in H_2O dictated by a competition between energetic and entropic effects

$$H_2O \leftrightarrow H^+ + OH-$$

- law of mass action using $\nu_{H_2O} = -1,\ \nu_{H^+} = 1, \nu_{OH^-} = 1$:

$$[H^+]\,[OH^-] / [H_2O] =$$

$$[H^+]_0\,[OH^-]_0 / [H_2O]_0 \exp[-\beta(\mu_{H^+}^{\ 0} + \mu_{OH^-}^{\ 0} - \mu_{H_2O}^{\ 0})]$$

- $[A]_0$ … the concentration of species A in some *standard state*
- $\mu_A^{\ 0}$ … the chemical potential of species A in the standard state
- pure water: presence of H+ is due to dissociation only

$$[H^+] = [OH^-]$$

$$[H_2O] = [H_2O]_0 \text{ (only 1 in } 10^7 \text{ molecules dissociated)}$$

$$[H_2O]_0 = 55\text{ M};\ \ [H^+]_0 = [OH^-]_0 = 1\text{M};$$

$$\mu_{H^+}^{\ 0} + \mu_{OH^-}^{\ 0} - \mu_{H_2O}^{\ 0} = 79.9\text{ kcal/mol}$$

$$[H^+]\,[OH^-] = [H^+]^2 = 1.0 \times 10^{-14}\ \text{M}^2 \rightarrow [H^+] = 10^{-7}\ \text{M}$$

An average distance between the H+ ions as a function of pH

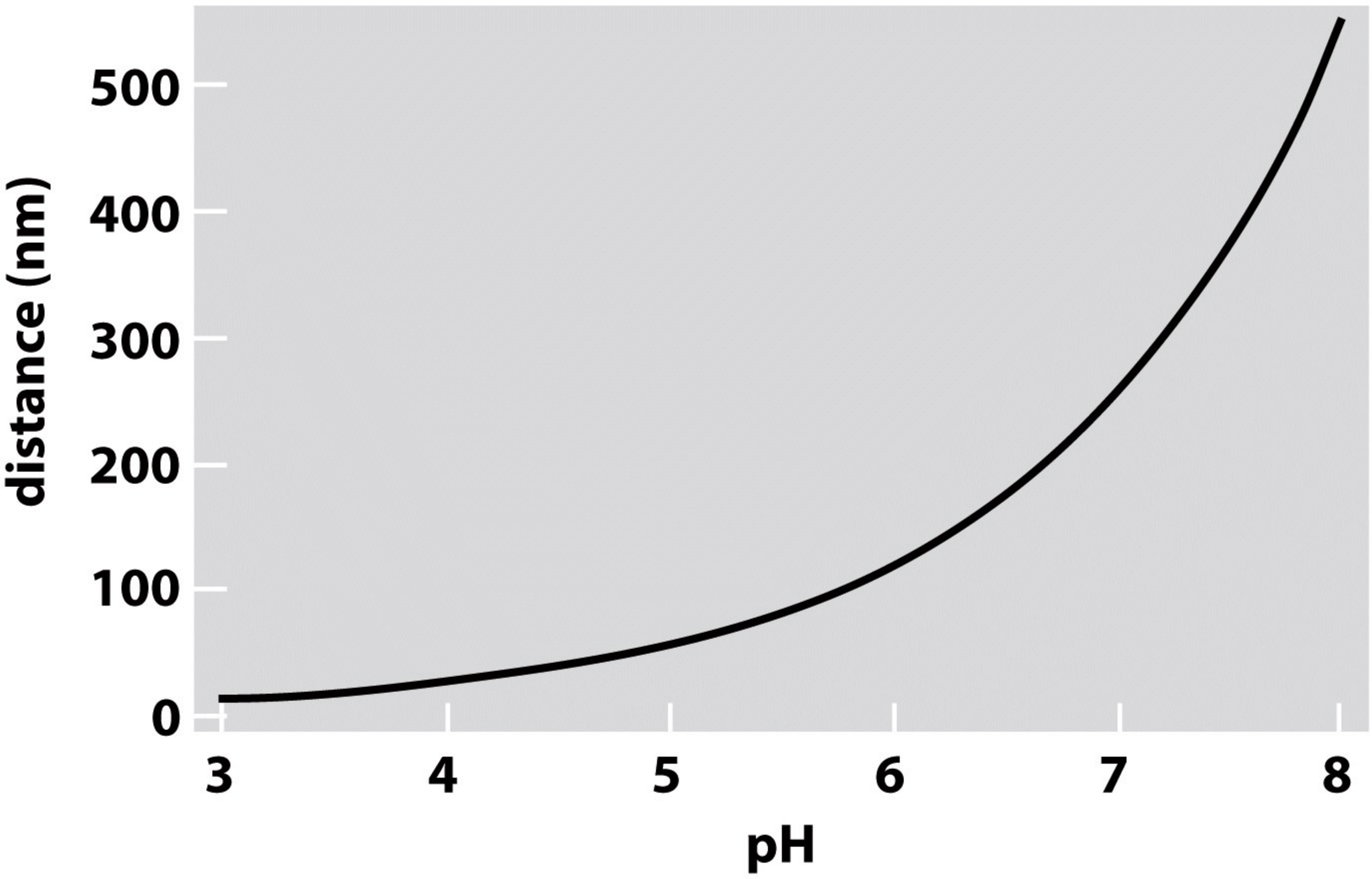


Figure 9.2 Physical Biology of the Cell (© Garland Science 2009)

The charge state of macromolecules M and the pH of the solution

$$\mathrm{HM} \leftrightarrow \mathrm{H}^+ + \mathrm{M}^-$$

Dissociation constant K_d:

$$K_d = [\mathrm{H}^+][\mathrm{M}^-]/[\mathrm{HM}]$$

A measure of the tendency of a macromolecule to undergo the dissociation reaction is known as pK of the molecule:

$$pK = -\log_{10} K_d$$

Henderson-Hasselbalch equation:

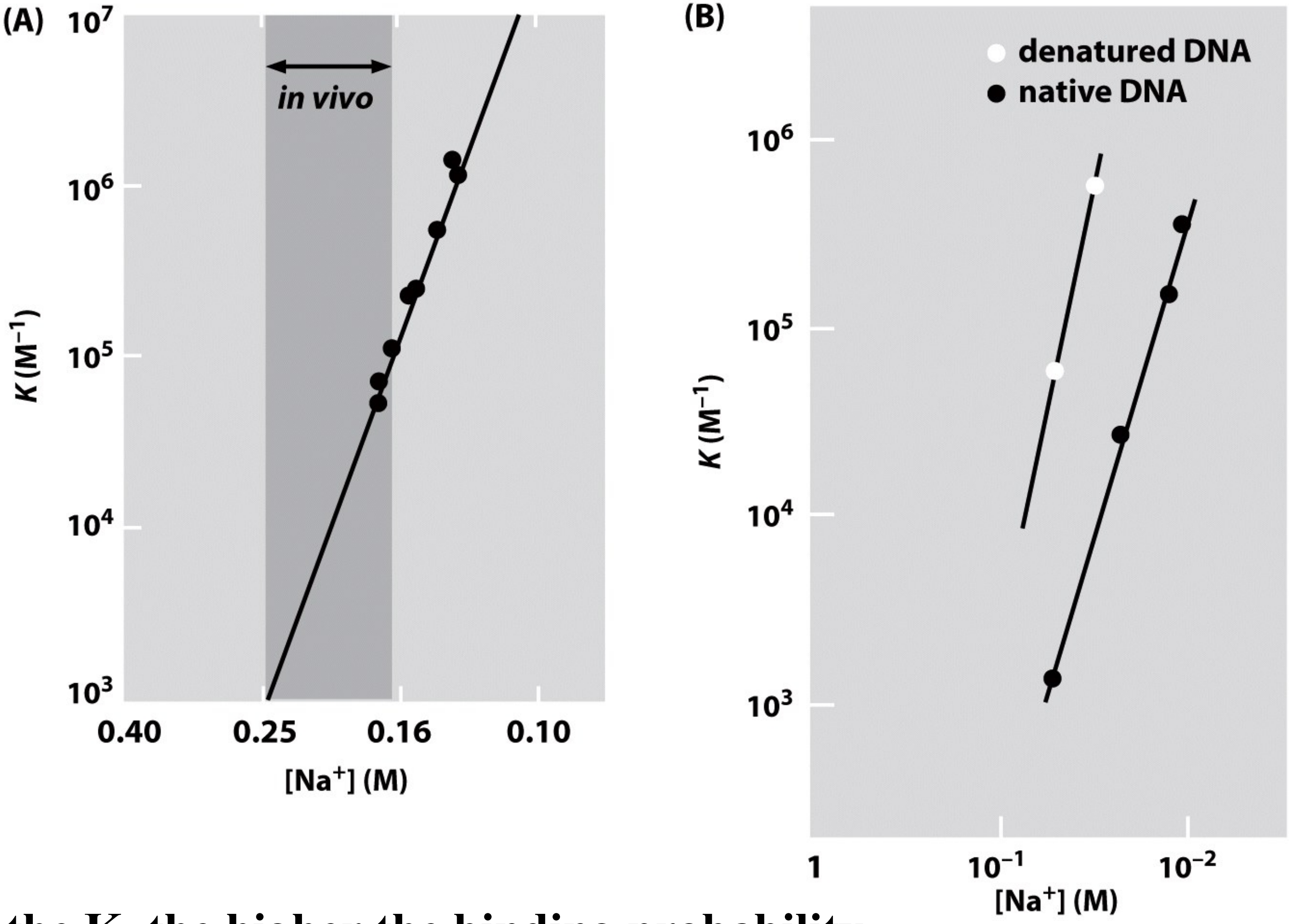
$$pH = pK + \log_{10} \{[\mathrm{M}^-]/[\mathrm{MH}]\}$$

pK is equal to pH, at which half of macromolecules has dissociated.

For example:

- pK =1 for DNA;
- at pH=7, the phosphates on the DNA backbone dissociated (each phosphate group carries 2 negative charges)
- different amino acids have different pK values

Salt concentration dependence of equilibrium constants K: (A) binding of Lac repressor to nonspecific DNA (B) binding of bovine pancreatic ribonuclease to DNA



the larger the K, the higher the binding probability

Figure 9.3 Physical Biology of the Cell (© Garland Science 2009)

Electrostatics for Salty Solutions: Review of EIs between charges

- a force between two charges q_1 and q_2:

$$F = (4\pi\varepsilon_0 D)^{-1}\, q_1\, q_2/r^2$$

- **D** … a dielectric constant (D=1 for vacuum or air, D=80 for water)
- water molecules are polar, they arrange with respect to a local electric field

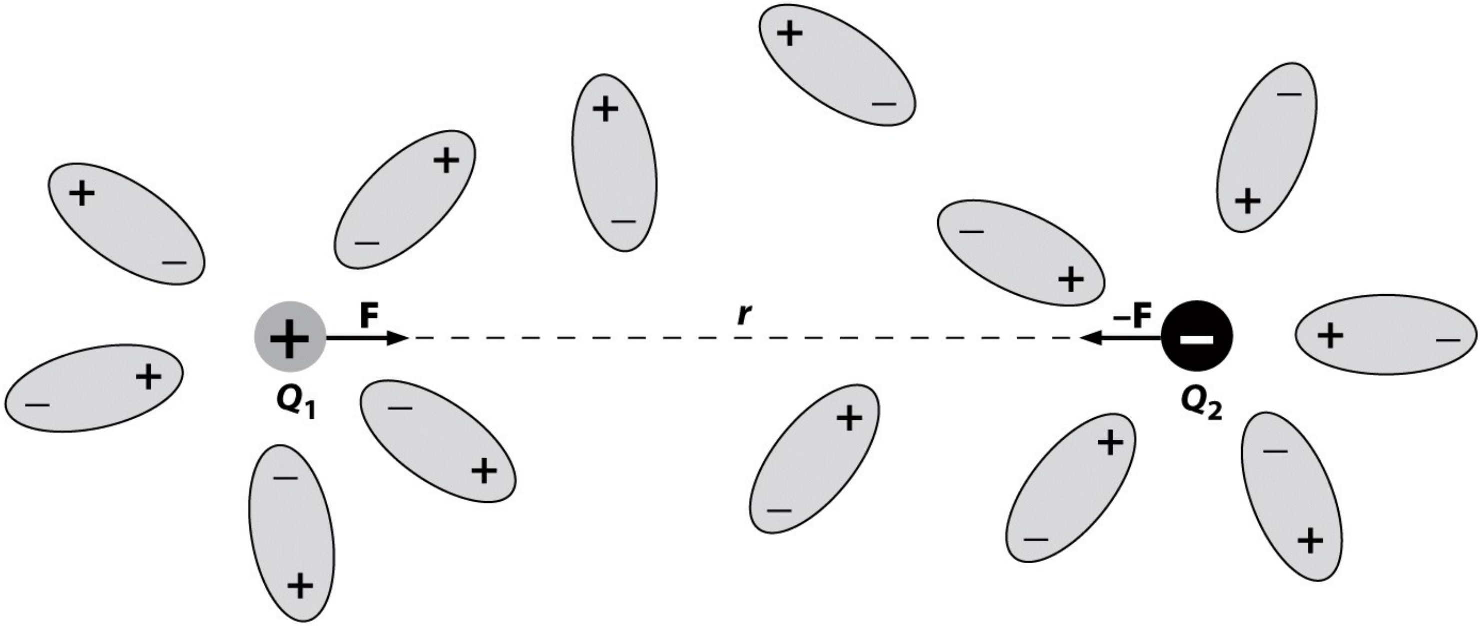


Figure 9.4 Physical Biology of the Cell (© Garland Science 2009)

$$\mathbf{E}(\mathbf{r}) = (4\pi\varepsilon_0 D)^{-1}\ Q/r^2\,\mathbf{e}$$

$$\mathbf{E}_{\mathbf{TOT}}\ (\mathbf{r}) = \sum_{\mathbf{i}} \mathbf{E}_{\mathbf{i}} = \sum_{\mathbf{i}} \mathbf{E}(\mathbf{r} - \mathbf{r}_{\mathbf{i}}) \rightarrow \textbf{superposition}$$

Instead of discrete distribution of charges (which results in individual electric fields), we will introduce continuous charge distribution $\rho(\mathbf{r})$ within a small volume $\Delta\mathbf{V}$

$$\Delta\mathbf{Q} = \rho(\mathbf{r})\,\Delta\mathbf{V}$$

Electric field caused by A) a single charge, B) three charges, C) a local charge density

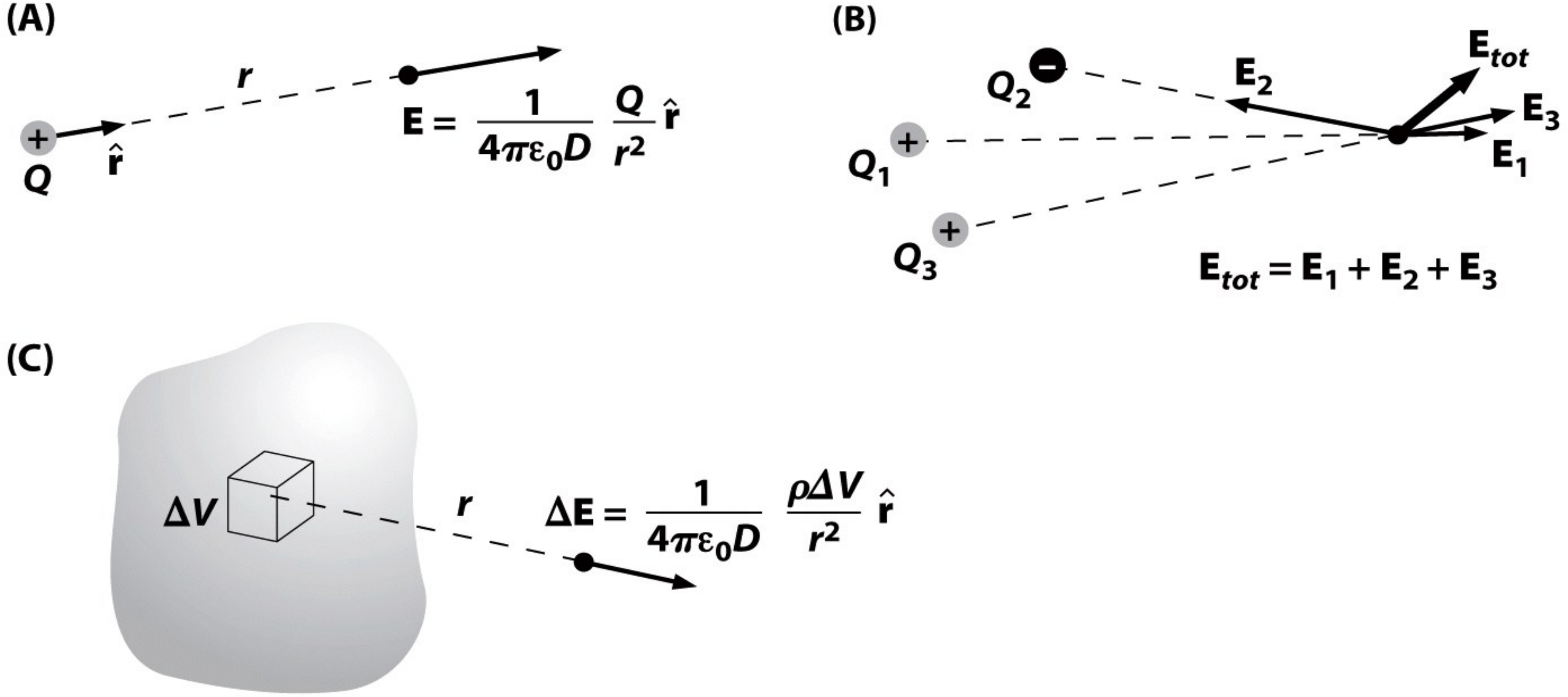


Figure 9.6 Physical Biology of the Cell (© Garland Science 2009)

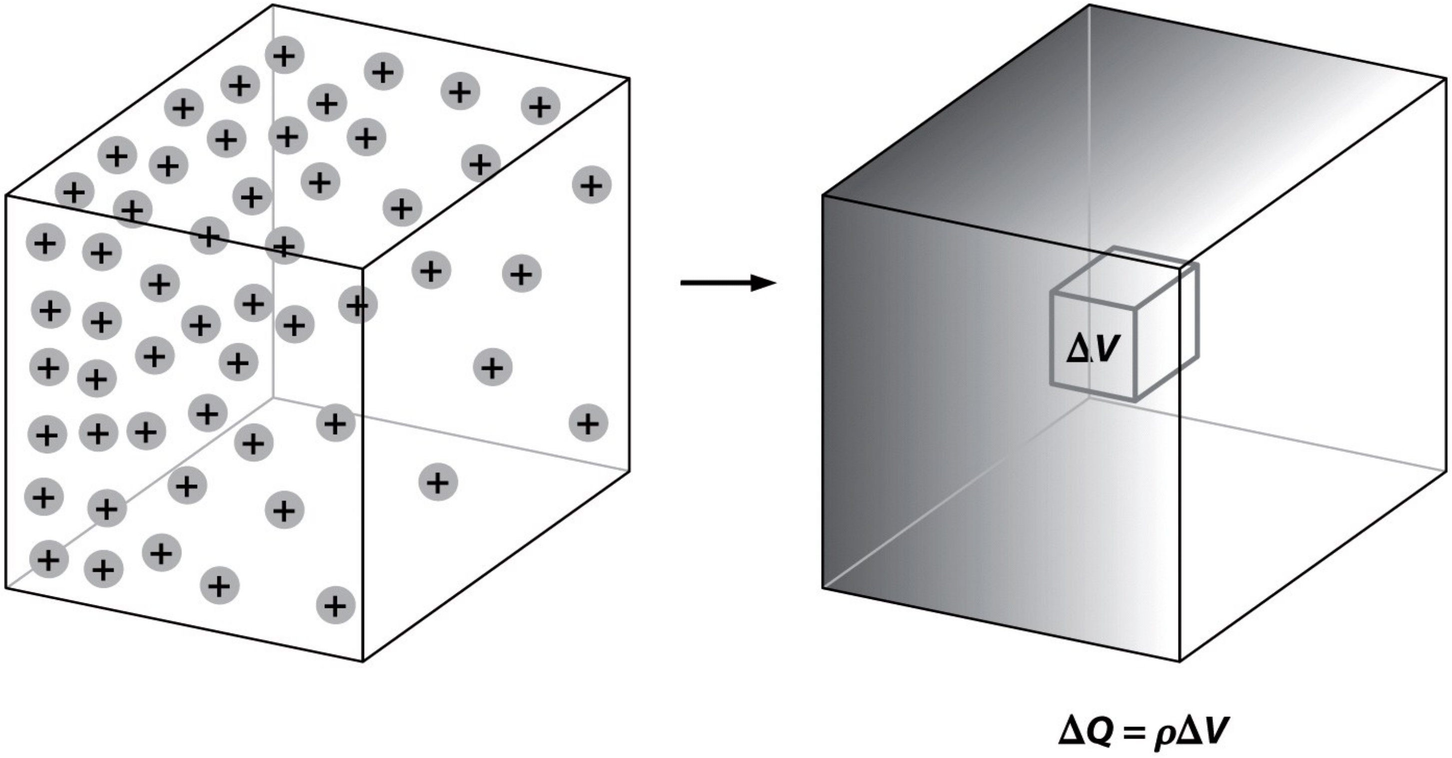


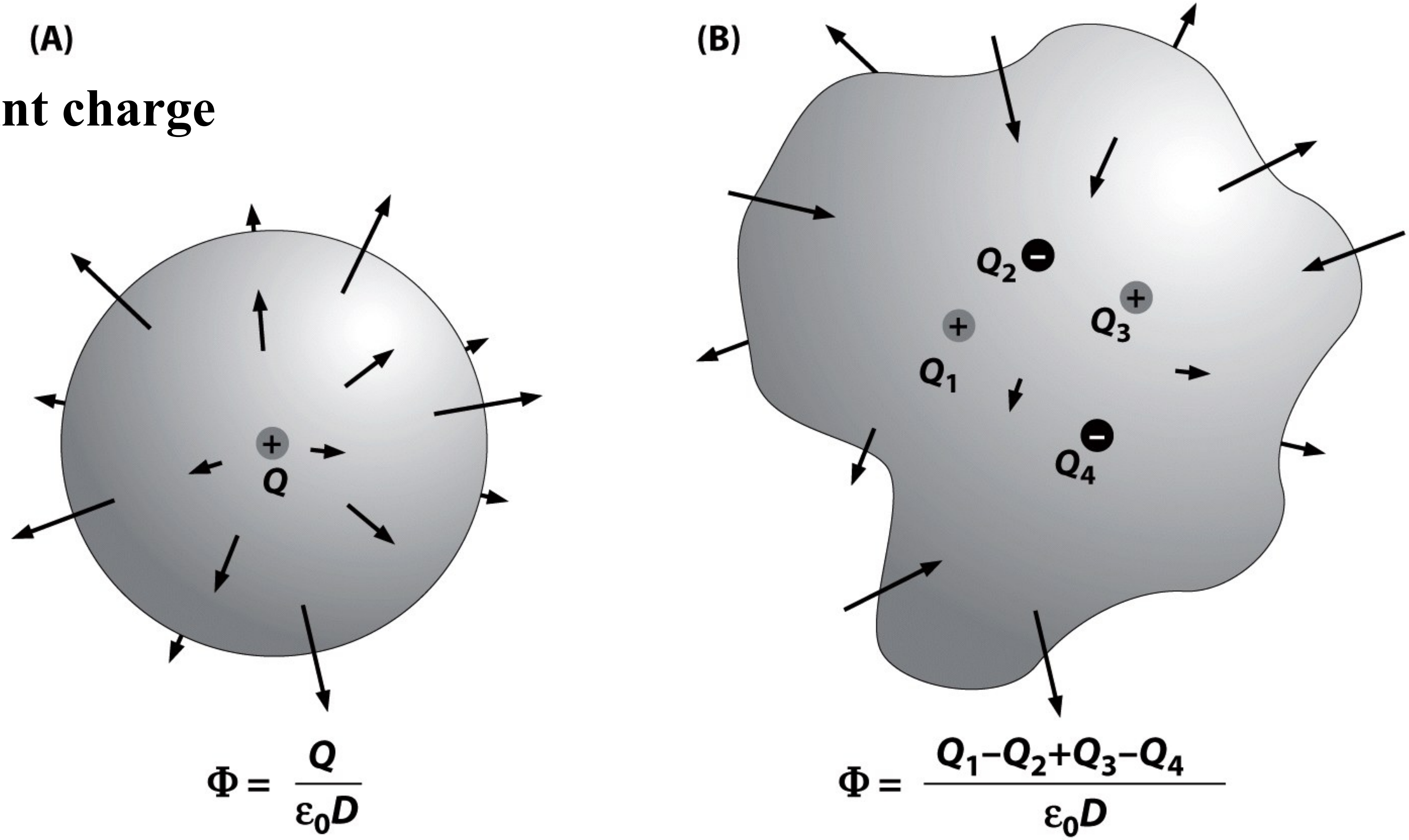
Figure 9.5 Physical Biology of the Cell (© Garland Science 2009)

The relationship between the charge density and the electric field using the concept of the flux Φ of the electric field (flux =the # of electric field lines per unit area, perpendicular to the field direction):

$$\Phi = \int \mathbf{E}(\mathbf{r})\ \mathbf{n}\ dA = \int (4\pi\varepsilon_0 D)^{-1}\ Q/r^2\ \mathbf{e}_r\ \mathbf{e}_r\ dA = Q/(\varepsilon_0 D)$$

Electric Flux and Gauss's Law

point charge



$$\Phi = \frac{Q}{\varepsilon_0 D}$$

$$\Phi = \frac{Q_1 - Q_2 + Q_3 - Q_4}{\varepsilon_0 D}$$

Figure 9.7 Physical Biology of the Cell (© Garland Science 2009)

Let the charge density depend only on x: Calculate the flux through a closed surface:

$$\Phi = E_x(x + \Delta x)\, \Delta y\, \Delta z - E_x(x)\, \Delta y\, \Delta z = dE_x/dx\, \Delta x\, \Delta y\, \Delta z$$

so the Gauss's law is expressed as:

$$dE_x(x)\,/dx = \rho(x)/D\varepsilon_0$$

or in general $E=E(x,y,z)$
for each component x,y,z:

$$\partial E_x/\partial x + \partial E_y/\partial y + \partial E_z/\partial z = \rho(x)/D\varepsilon_0$$

the most general form of Gauss's law.

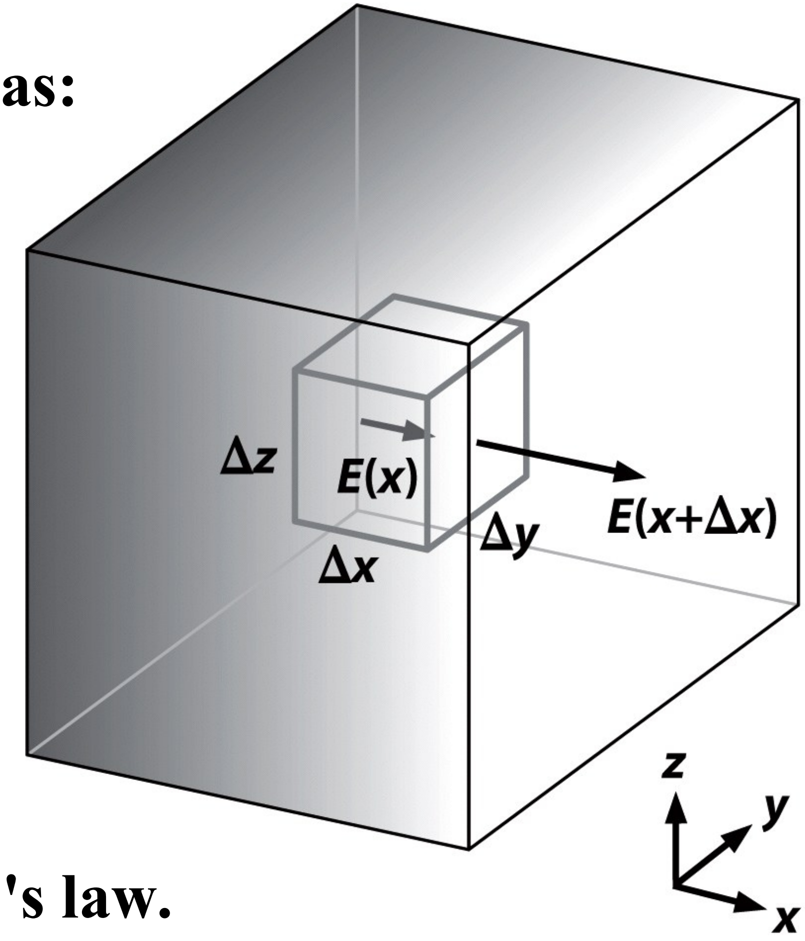


Figure 9.8 Physical Biology of the Cell (© Garland Science 2009)

From the electric field E(r) description to the electrostatic potential V(r)

V(r) … electrostatic potential = work per unit charge done to bring a test charge from infinity to r very slowly

Example in 1D:

E(x) as a result of a charge distribution $\rho(x)$:

$$V(x) = \int_x^\infty -(E_x(x')\, e_x) \cdot -(e_x\, dx')$$
$$= \int_x^\infty E_x(x')\, dx'$$

$$E_x(x) = dV(x)/dx$$

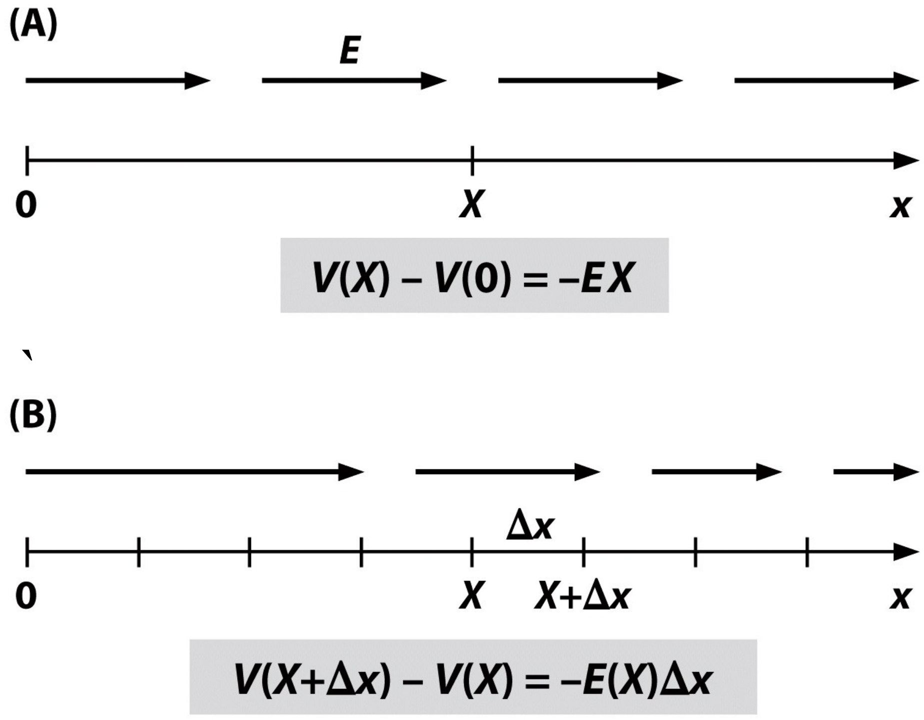


Figure 9.9 Physical Biology of the Cell (© Garland Science 2009)

For each component of E(r):

$$(E_x(x,y,z), E_y(x,y,z), E_z(x,y,z)) =$$
$$(-\partial V(x,y,x)/\partial x, -\partial V(x,y,x)/\partial y, -\partial V(x,y,x)/\partial z)$$

Express the relationship between V(r) and E(r) into Gauss's law:

$$\partial^2 V(x,y,x)/\partial x^2 + \partial^2 V(x,y,x)/\partial y^2 + \partial^2 V(x,y,x)/\partial z^2$$
$$= -\rho(x,y,x)/(D\varepsilon_0)$$

We derived the Poisson equation that relates electrostatic potential to the charge density.

$$\nabla^2 V(\mathbf{r}) = -\rho(\mathbf{r})/(D\varepsilon_0)$$

∇^2 **… Laplacian of the scalar function** $V(\mathbf{r})$

Energy cost associated with assembly a charged sphere

- for a general charge distribution:

$$\mathbf{U}_{\mathbf{EL}} = \tfrac{1}{2} \sum \mathbf{q}_i \mathbf{V}_i = \tfrac{1}{2} \int V(\mathbf{r})\, \rho(\mathbf{r})\, d^3\mathbf{r}$$

(bring charge 1 to the vicinity of charge 2 and the other way around → ½)

- for a sphere (to the right):

$$d\mathbf{U}_{\mathbf{EL}} = V(\mathbf{r})\, dq$$

$$V(r) = \int_r^{\infty} E(r')dr' = q/(4\pi\varepsilon_0 D) \int_r^{\infty} r'^{-2}\, dr'$$

$$= q/(4\pi\varepsilon_0 Dr)$$

$$d\mathbf{U}_{\mathbf{EL}} = V(r)\, dq$$

$$= 1/(4\pi\varepsilon_0 Dr)\, \rho\, 4/3\pi r^3\, \rho 4\pi r^2 dr \rightarrow$$

$$U_{EL} = \int_{\mathbf{0}}^{R} 16\pi^2\, \rho^2/(12\pi\varepsilon_0 D)\, r^4 dr$$

$$= 3Q^2/(20\pi\varepsilon_0 DR)$$

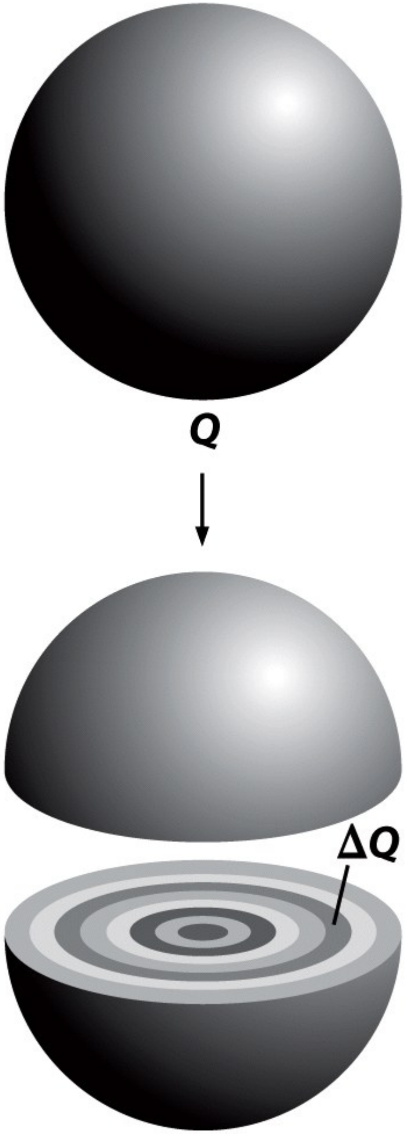


Figure 9.10 Physical Biology of the Cell (© Garland Science 2009)

What is an energy cost of separation of two charges (a) in vacuum and (b) in water?

- in water the charges are largely free (non-interacting)

Bjerrum length l_B = length at which the electrostatic interaction energy between two oppositely charged charges is equal to the thermal energy k_BT: $e^2/(4\pi\varepsilon_0 D l_B) = k_BT$ or

$$l_B = e^2/(4\pi\varepsilon_0 D\, k_BT) \sim 0.7 \text{ nm (in water with D=80)}$$

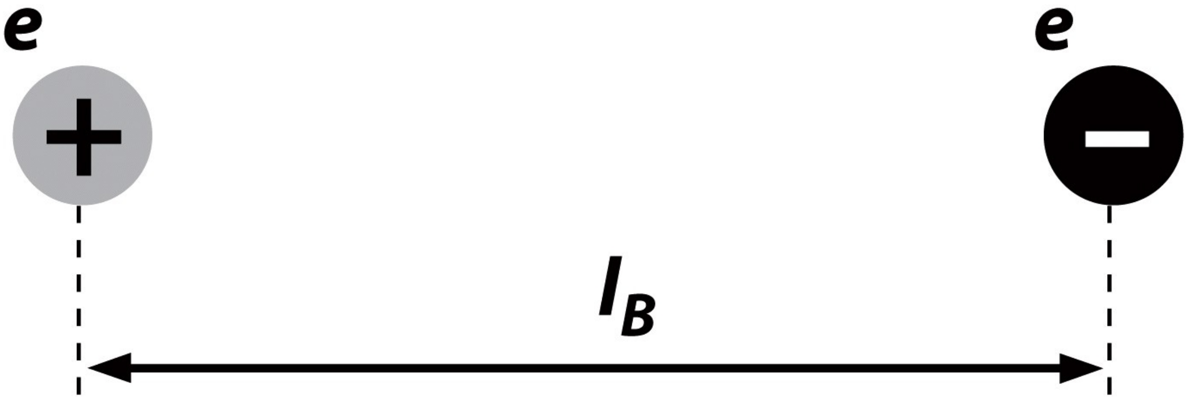


Figure 9.11 Physical Biology of the Cell (© Garland Science 2009)

A simple globular protein = a ball of radius R made of amino acids: small beads with radii r

- all hydrophobic residues: inside the ball

- all polar (hydrophilic) residues: on the surface

- each of polar residues can release one unit charge into the solution

What is the self energy of the sphere if all charge is concentrated on a shell of radius R?

- if the # of polar residues as a function of R is Q_T, then:

$$U = Q_T^2/(4\pi\varepsilon_0 D 2R)$$

$$Q_T = Ne = (4\pi R^2/\pi r^2)\ e$$

$$U = e^2/(4\pi\varepsilon_0 D)\ 8\ R^3/r^4$$

$$= k_B T\ 8R^3 l_B/(Dr^4)$$

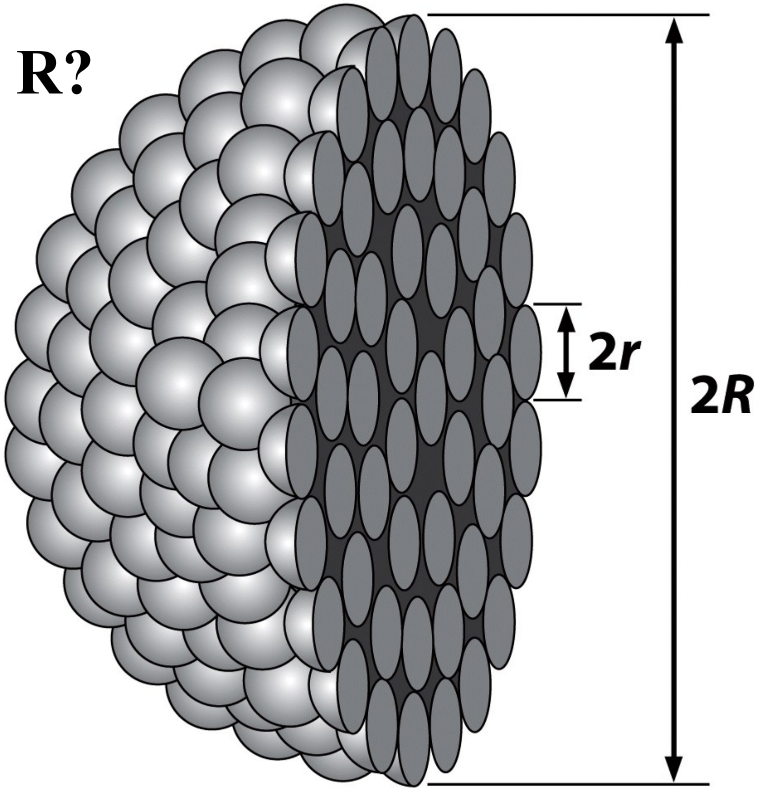


Figure 9.12 Physical Biology of the Cell (© Garland Science 2009)

Energy cost to charge a protein:

- $l_B \sim 0.7$ nm
- $r \sim 0.5$ nm
- $U = k_B T\, R^3\, 8 \times 0.7/(0.5)^4$ for R [nm]

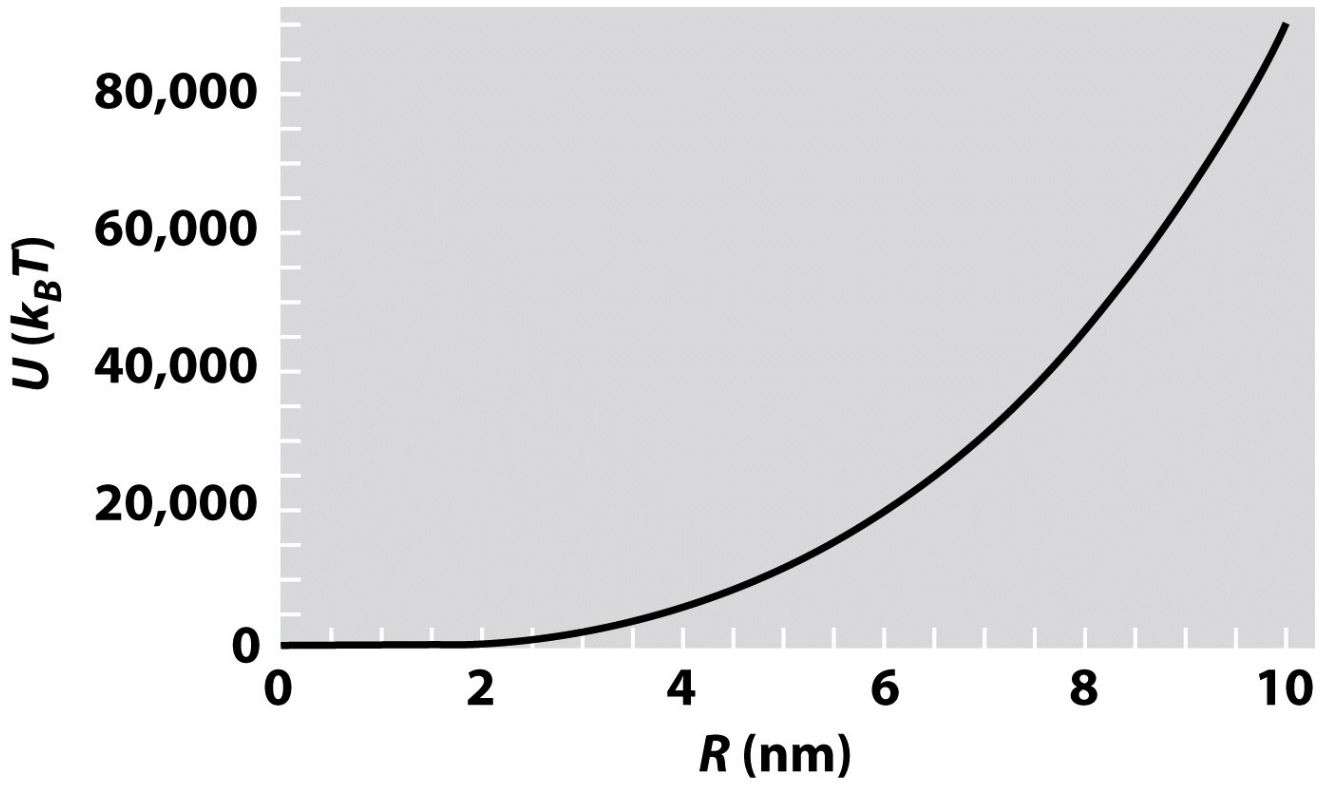


Figure 9.13 Physical Biology of the Cell (© Garland Science 2009)

Electrostatics in Salty Solutions:
Negatively Charged DNA in Ionic Solution with Counterions

- salty solutions are physiologically relevant

- DNA backbone is negatively charged

- positive ions in solution form a screening cloud because it lowers their electrostatic energy

- the thickness of the “cloud”: interplay between lower EI energy and entropy loss

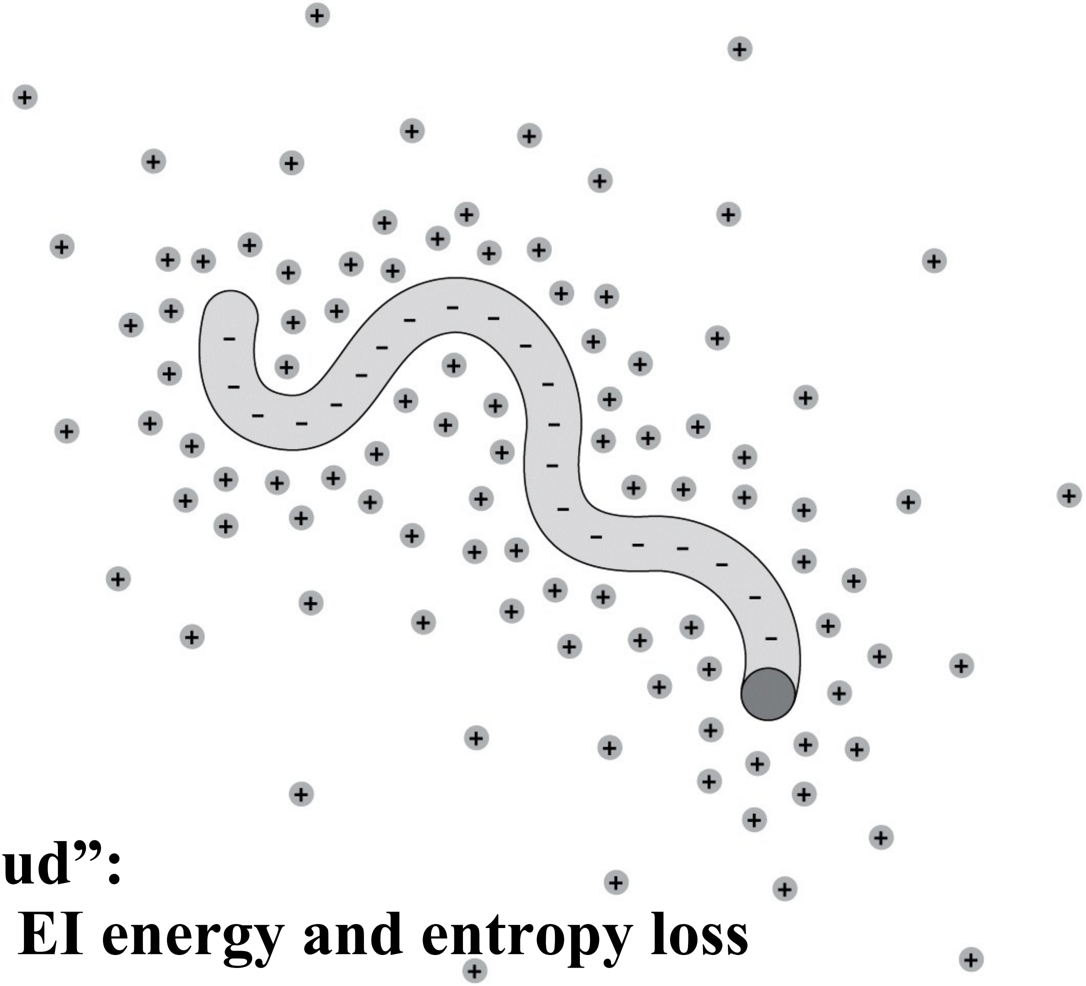


Figure 9.14 Physical Biology of the Cell (© Garland Science 2009)

ESTIMATE: The size of the screening cloud λ_D

- **concentration of + ions:** $c_{\text{positive}} = c_\infty + \frac{1}{2}\Delta c$

where $\Delta c = c_{\text{positive}} - c_{\text{negative}}$

- **the net charge of the screening cloud:**

$$Q = e\,\Delta c\,A\,\lambda_D$$

(A … surface area of the biomolecule)

- **the total charge of a biomolecule and the surrounding cloud is zero**

$$\Delta c = \frac{Q}{e}\frac{1}{A\lambda_D} \qquad \frac{Q}{e} = \text{no. charges}$$

- **electric field:** $E = \frac{Q}{D\varepsilon_0 A}$

- **electric potential:** $V = \frac{Q\lambda_D}{2D\varepsilon_0 A}$

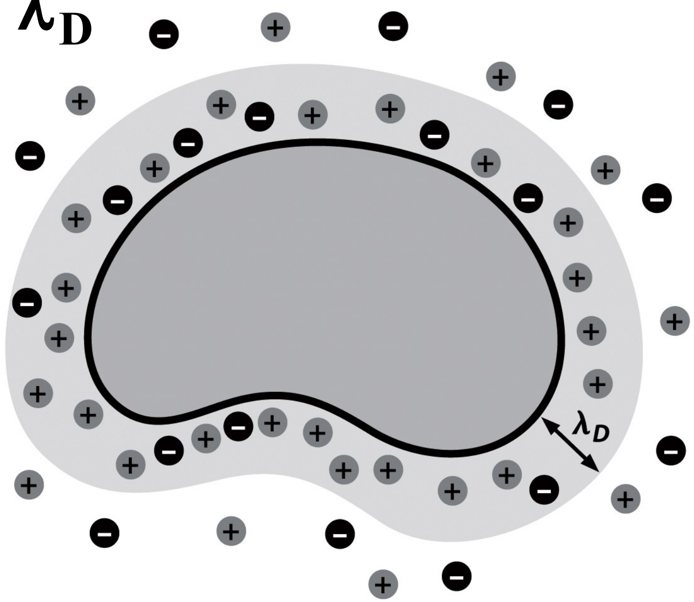


Figure 9.15a Physical Biology of the Cell (© Garland Science 2009)

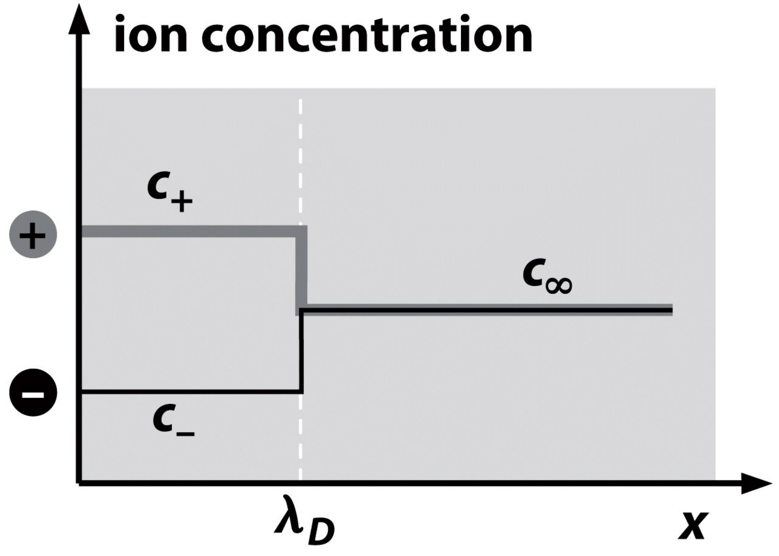


Figure 9.15b Physical Biology of the Cell (© Garland Science 2009)

In equilibrium the chemical potential away from the cloud is:

$$\mu = \mu_0 + k_B T \ln(c_\infty/c_0)$$

The chemical potential of an ion inside the cloud:

$$\mu = \mu_0 + k_B T \ln[(c_\infty + ½\, \Delta c)/c_0] + e\, V$$

$$= \mu_0 + k_B T \ln(c_\infty/c_0)$$

The two chemical potentials are the same in equilibrium:

$$k_B T \ln(c_\infty/c_0) = k_B T \ln[(c_\infty + ½\, \Delta c)/c_0] + eQ\lambda_D/(2D\varepsilon_0 A)$$

So assuming that $\Delta c \ll c_\infty$ we can write

$$\ln(c_\infty + ½\, \Delta c) - \ln(c_\infty) = \ln(1 + ½\, \Delta c/c_\infty) \sim ½\, \Delta c/c_\infty$$

and obtain *the Debye screening length* λ_D expressed by I_B:

$$\lambda_D = (4\pi I_B c_\infty)^{-1/2}$$

Remember that $I_B = e^2/(4\pi\varepsilon_0 D k_B T)$ … Bjerrum length

The Poisson-Boltzmann Equation

- Poisson Eq.: relates the EI potential to the charge density
- Boltzmann distribution: probability of different microstates
- far from the biomolecule:

$$c_{\text{positive}} = c_{\text{negative}} = c_\infty$$

- consider a negatively charged membrane with σ as a charge per unit area: only variation in the x-direction

$$c_{\text{positive}}(x) = c_\infty e^{-\beta z e V(x)}$$

$$c_{\text{negative}}(x) = c_\infty e^{+\beta z e V(x)}$$

- ±zeV(x) … the EI energy of ± ions

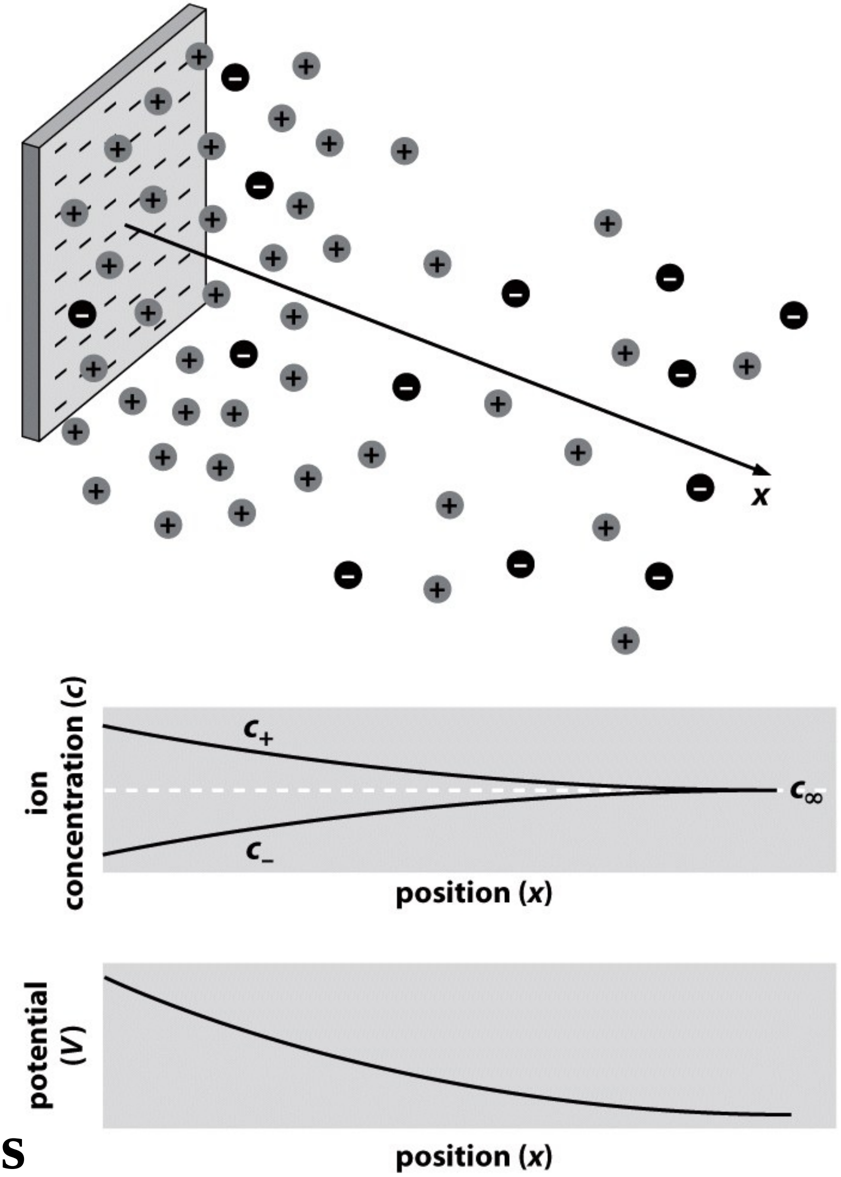


Figure 9.16 Physical Biology of the Cell (© Garland Science 2009)

The total charge density $\rho(x)$: $\rho(x)=zec_{\text{positive}}(x)-zec_{\text{negative}}(x)$
is related to the electric potential through the Poisson equation:

$$\frac{d^2V(x)}{dx^2}=\frac{-\rho(x)}{D\varepsilon_0}$$

Resulting in the non-linear differential equation for salty solution which is *the Poisson-Boltzmann equation*:

$$\frac{d^2V(x)}{dx^2}=\frac{zec_\infty}{D\varepsilon_0}\cdot\left[e^{+\beta zeV(x)}-e^{-\beta zeV(x)}\right]$$

When the charge density on a biomolecule does not produce a too large electric potential $V(x)$, we can use a linearized form:

$$\frac{d^2V(x)}{dx^2}=\frac{2z^2e^2c_\infty}{D\varepsilon_{.}k_BT}\,V(x)$$

Known as *the Debye-Huckel equation*. Solution is well-known:

$$V(x)=Ae^{-x/\lambda_D}+Be^{+x/\lambda_D}\qquad\lambda_D=\sqrt{\frac{D\varepsilon_0k_BT}{2z^2e^2c_\infty}}$$

Debye screening length

Solution:

$$V(x)=\frac{\sigma\lambda_D}{D\varepsilon_0}e^{-x/\lambda_D}$$

$$\rho(x)=\frac{-\sigma}{\lambda_D}e^{-x/\lambda_D}$$

For a charged protein in a salt solution with charge density c_∞ = 200 mM (typical for potassium ions inside a cell),

λ_D=0.7 nm

Viruses as Charged Spheres in a Salty Solution: Debye-Hückel Model

What are viruses?

- ***virus*: an example of a macromolecular assembly of sizes ~10s to ~100s of nanometers**

- **highly ordered and symmetrical objects made of proteins and nucleic acids**

- ***capsid*: protein shell (repetitive packing of the same protein unit resulting in icosahedron: 12 vertices, 20 faces each with 3 edges)**

- ***viral genome*: once in a host cell, command the construction of its own inventory of parts that further assemble into virus copies**

Examples of Viral Capsid Structures

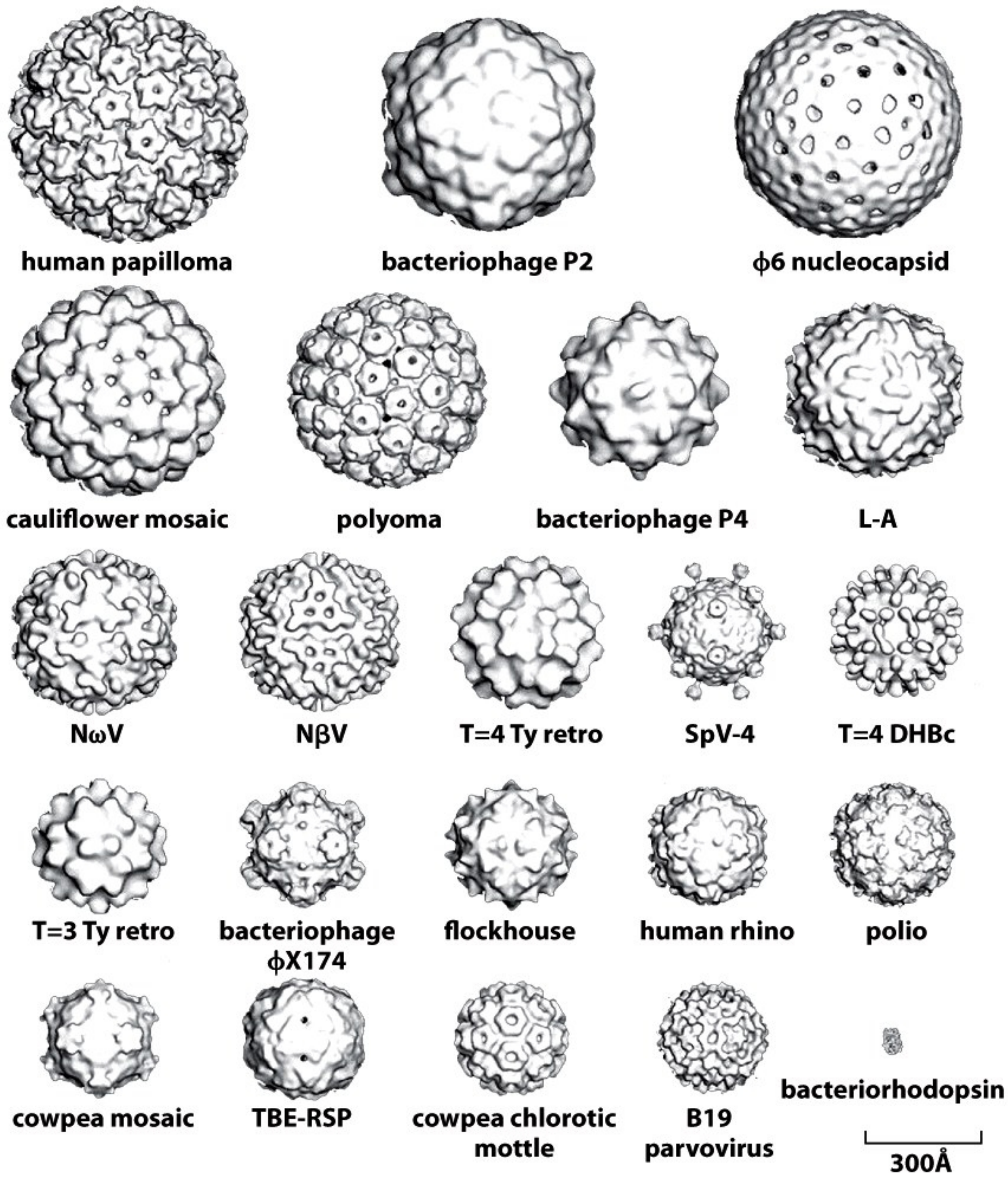


Figure 2.19 Physical Biology of the Cell (© Garland Science 2009)

HIV virus (electron micrograph)

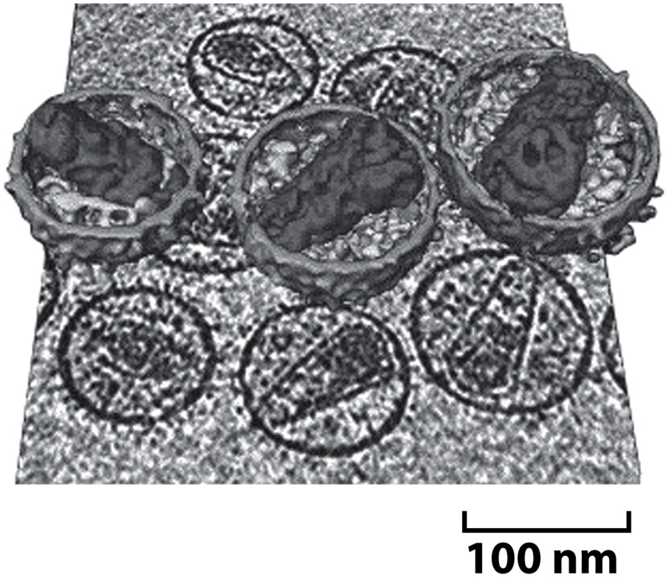


Figure 2.20 Physical Biology of the Cell (© Garland Science 2009)

The energy cost for assembling a spherical shell of charge Q and radius R is: $\frac{1}{2} Q V(R)$

$V(R)$ **... the potential on the surface of the sphere**

For spherically symmetric charge distribution, the flux:

$$\Phi(r) = E_r(r) 4 \pi r^2$$

The charge within a sphere of radius r is: $q(r) = \int_0^r \rho(r') 4 \pi r'^2 dr'$

Using Gauss's law relating the flux and the charge: $\Phi(r) = \frac{q(r)}{D \varepsilon_0}$

We then get the Poisson equation for a spherically symmetric case:

$$\frac{1}{r} \frac{d^2 [r V(r)]}{dr^2} = \frac{-\rho(r)}{D \varepsilon_0}$$

Using linearized version of the Boltzmann equation:

$$\rho(r) \approx \frac{-2 z^2 e^2 c_\infty}{k_B T} V(r)$$

We then derive the Debye-Hückel equation:

$$\frac{d^2[rV(r)]}{dr^2}=\frac{rV(r)}{\lambda_D^2}$$

λ_D … **the Debye screening length**

Known solution for the function $rV(r)$ **is:** $V(r)=A\frac{e^{-r/\lambda_D}}{r}$

The constant A **is determined by taking into account that**

$$\left(\frac{dV(r)}{dr}\right)_{r=R}=E(R)=\frac{Q}{4\pi\varepsilon_0 DR^2}\qquad A=\frac{Qe^{R/\lambda_D}}{4\pi D(1+\frac{R}{\lambda_D})}$$

$$V(R)=\frac{1}{4\pi\varepsilon_0 D}\left(\frac{Q}{R}\right)\frac{\lambda_D}{R+\lambda_D}$$

The electrostatic energy of the spherical shell (virus capsid):

$$U(R)=\frac{1}{2}QV(R)=\frac{1}{2}k_B T\left(\frac{Q}{e}\right)^2\frac{l_B\lambda_D}{R(R+\lambda_D)}$$

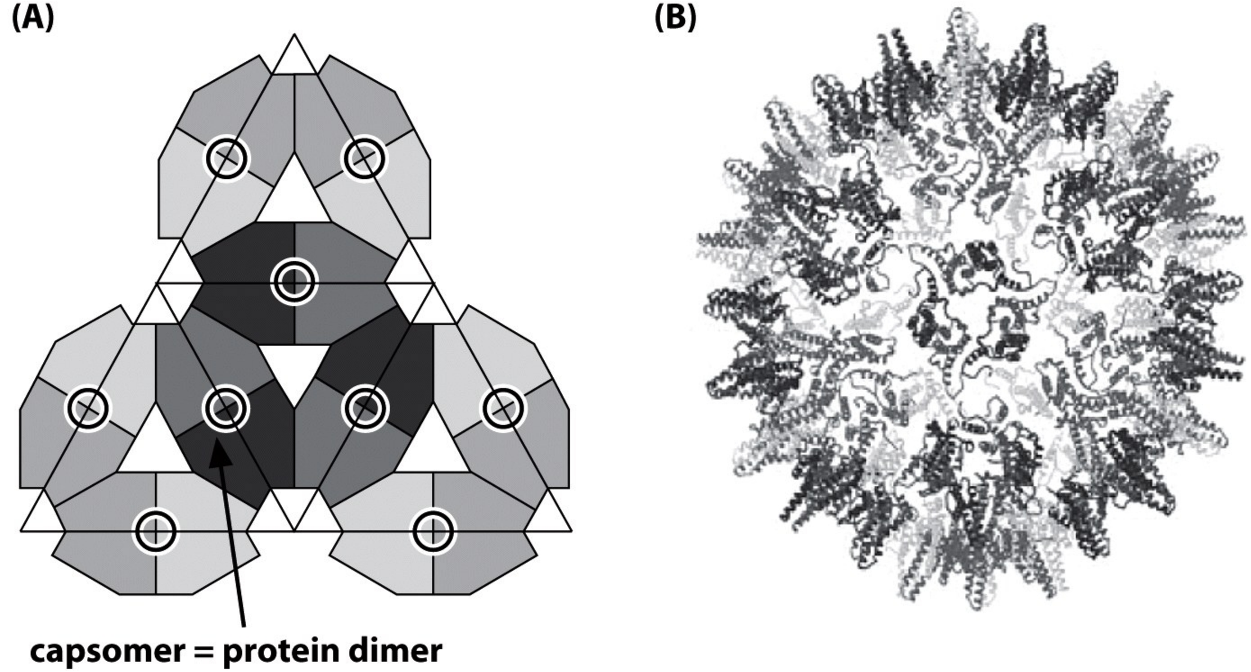


Figure 9.17 Physical Biology of the Cell (© Garland Science 2009)

Capsomers arranged into a triangular face of the icosahedral capsid are held together by hydrophobic forces that need to overcome the electrostatic interactions ($Q = N z e$ & $R + \lambda_D \sim R$):

$$\Delta G_{capsid} = \Delta G_{contact} + \frac{1}{2} k_B T \frac{N^2 z^2 l_B \lambda_D}{R^2}$$

Salt concentration dependence is via the Debye screening length λ_D.

Equilibrium constant can be measured and compared to:

$$\ln K_{capsid} = \frac{-\Delta G_{capsid}}{k_B T} = \frac{-\Delta G_{contact}}{k_B T} - \frac{1}{2} N^2 z^2 \frac{l_B \lambda_D}{R^2}$$

- ➔ **measured equilibrium constant = concentration of capsids in solution as a function versus concentration of capsomers squared**

- ➔ **temperature dependence: (hydrophobic effect)**

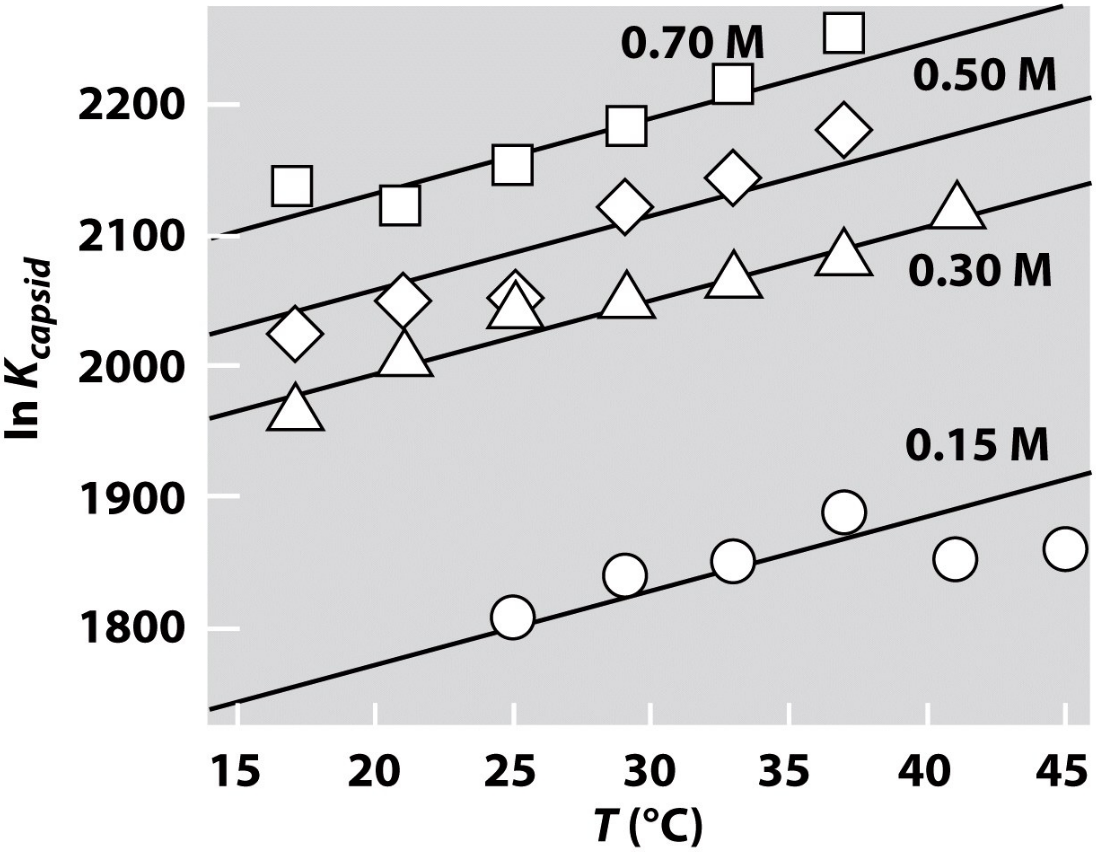
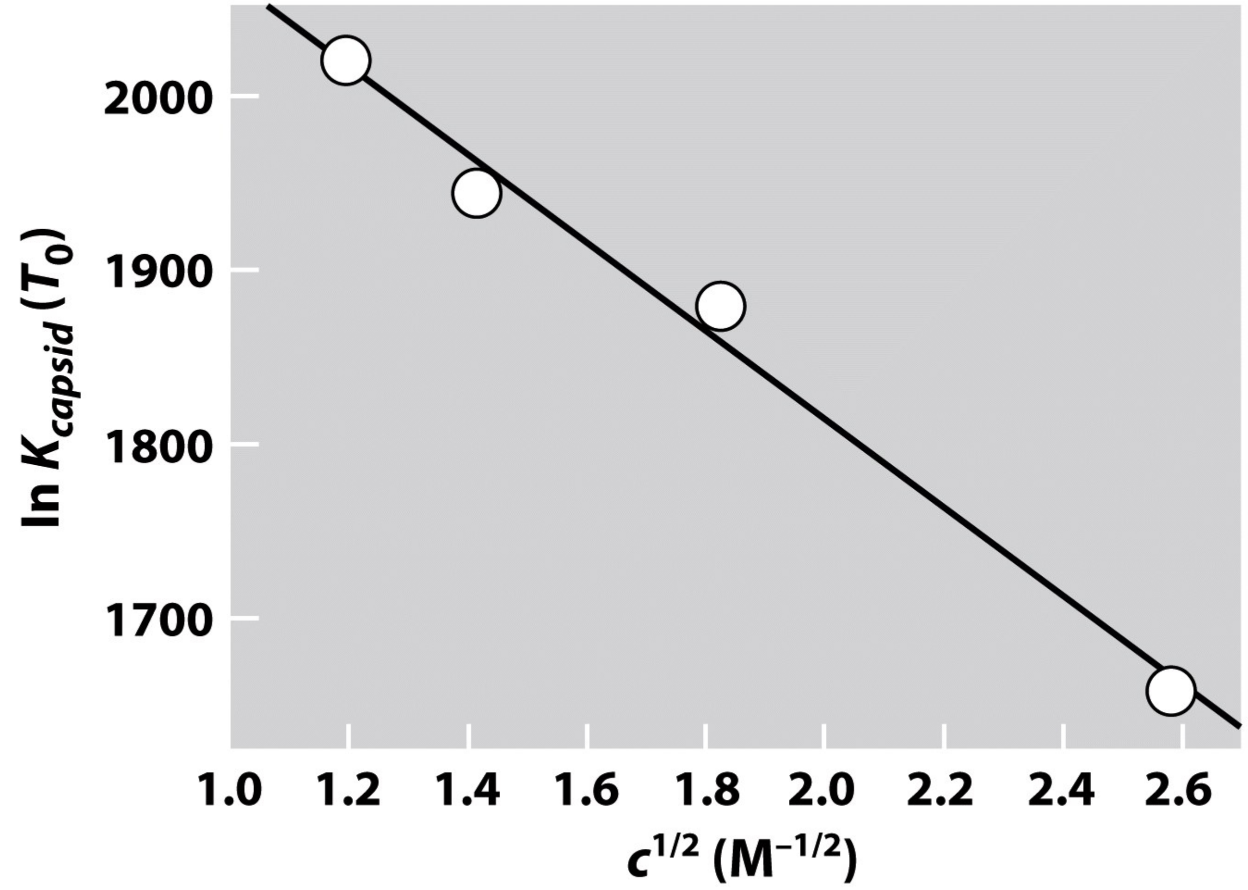


Figure 9.18 Physical Biology of the Cell (© Garland Science 2009)

Equilibrium constant depends on the salt concentration:

$$\ln K_{\mathrm{capsid}} \propto \frac{1}{\sqrt{c}}$$



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