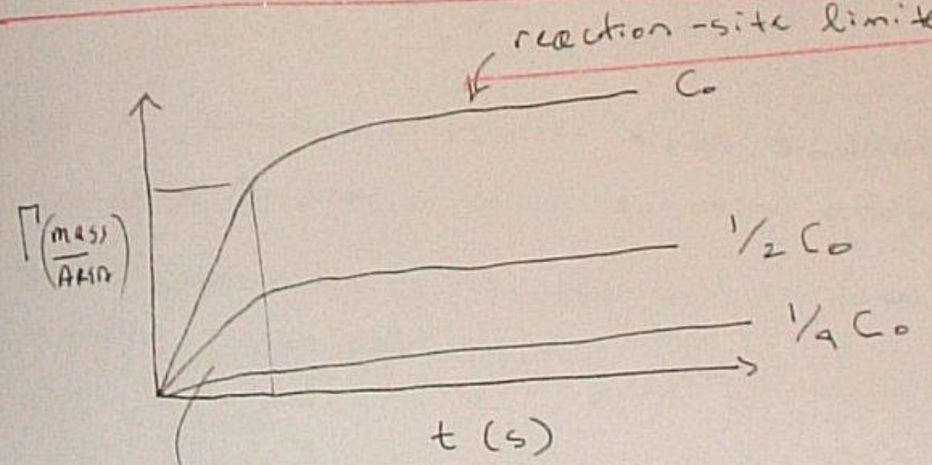
Biological performance is influenced by.

6pts.

1. type of protein adsorbed
2. amount of protein adsorbed, and rate (i.e., kinetics)
3. conformation of the adsorbed protein

b) 5pts.

Place material buffer solution and then add protein. Track protein either via. I^{125}, mass, ellipsometry, fluorescence, etc...



diffusion limited region

c) 10pts

During the successive experiment the protein has "time" to spread since the concentration of protein in solution for a specific part of the experiment is lower than for the direct experiment. Hence, at lower protein concentrations in the successive experiment, a greater area is occupied, and less area is available for the higher protein concentration

MSE/BioE C118 - Biological Performance of Materials

Prof. K. E. Healy
465 Evans Hall

Exam 1: October 16, 2003 **Closed Book Exam**
Please answer all of the questions clearly and box your final answer. Useful equations, data, and physical constants appear at the end of the exam.

NAME: _____

ID NUM _____

MSE/BioE C118

Na [redacted] –

1. You are asked to measure the mechanical properties of a new semi-crystalline ultra high molecular weight poly(ethylene) to be used for orthopaedic total joint replacement implants. You can assume the Maxwell element can satisfactorily describe the viscoelastic behavior of the linear polymer. (25pts.)

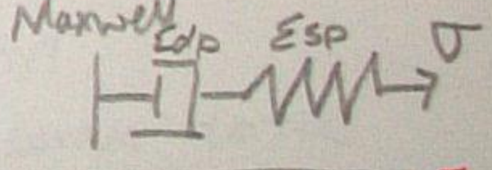
a. First you conduct a typical engineering stress-strain test using a constant rate of tensile strain. Write an expression for the differential equation for the Maxwell element and solve this equation using appropriate boundary conditions.

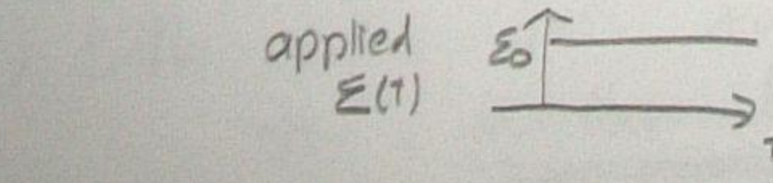
b. Sketch a stress-strain curve and demonstrate the effect of increasing strain rate on stiffness (modulus).

c. Does this model give an accurate qualitative description of linear poly(ethylene) in a stress-strain test?

a)

a) Maxwell model : under a constant rate, tensile strain





$\sigma_{dp} = \eta \frac{d\varepsilon}{dt}$

$\sigma_s = E\varepsilon$

$\sigma = \sigma_s = \sigma_{dp}$

$\varepsilon = \varepsilon_s + \varepsilon_{dp}$, $\dot{\varepsilon} = \dot{\varepsilon}_s + \dot{\varepsilon}_{dp}$, $\boxed{\frac{d\varepsilon}{dt} = \frac{d\sigma}{dt}\frac{1}{E} + \frac{\sigma}{\eta}}$ +5

Initial condition = $\varepsilon_0 = \frac{\sigma_0}{E}$ (instantaneous increase in ε due to spring)

σ $\dot{\sigma}$ $d\varepsilon$ multiply E

MSE/BioE C118 Na[redacted]

2. Poly(L-lactide-co-glycolide) is a semi-crystalline biodegradable copolymer that has found widespread use in medical devices. You are given two compositions of this copolymer to evaluate for an implantable biomedical device. You decide to implant each of these materials into the muscle of a well-established animal model to evaluate their biological performance. When you implant a material in the body you generate a cascade of reactions that leads to the development of a stable tissue-implant interface (i.e., interfacial hierarchy). (25 pts.)

a. What are the relevant bulk and surface characterization studies you would conduct? Be sure to describe what the characterization techniques measure and their limitations.

b. Describe the development of the interfacial hierarchy over time.

c. What type of long-term interactions can occur between the implant and the body?

d. How would degradation of the copolymer affect the interfacial hierarchy?

a) Bulk can be characterized by stress & strain ratio for mechanical properties, or cyclic loading.

Surface → contact angle measures wettability.
Limit + can't tell surface chemistry, hysteresis.
3-20 Å of surface (depe

+6

AFM I can probe surface morphology. see about

MSE/BioE C118

N[redacted]

3. In class, we used DLVO theory to approximate the interaction free energy as a function of distance between a polymer (flat surface) and either proteins or cells in salt solution. ((25 pts.)

+13

a. Explain how you would calculate the total interaction free energy W(D) curve for both a protein and a cell approaching the polymer surface?

+13

b. The electrostatic interaction free energy term depends on the Debye length (κ^{-1}). For a 0.2M $CaCl_2$ (1:2) solution calculate the Debye length (κ^{-1}) at body temperature (37 °C)? How would this κ^{-1} compare to a 1:1 solution like NaCl?

+9

c. Sketch representative curves for the Debye length (κ^{-1}) at various salt concentrations. What parameters of the system affect the Debye length the most.

+10

d. Sketch how altering κ^{-1} affects representative curves for the total interaction free energy W(D) for both a protein and a cell approaching the surface. Make sure to include representative features of the curves and label them clearly.

+5

e. Based on these graphs and DLVO theory, describe initial events occurring at the surface after exposure of the polymer to a 2:1 electrolyte with a concentration of 0.2M solution containing both proteins and cells (e.g., serum). What would allow the cell to overcome the energy barrier at the 2° minimum to approach direct contact with the surface.

a) DLVO = $W(D) = W_A(D) + W_E(D)$ ✓

$W_A(D)$ ⇒ van der Waals attraction energy can be obtained by

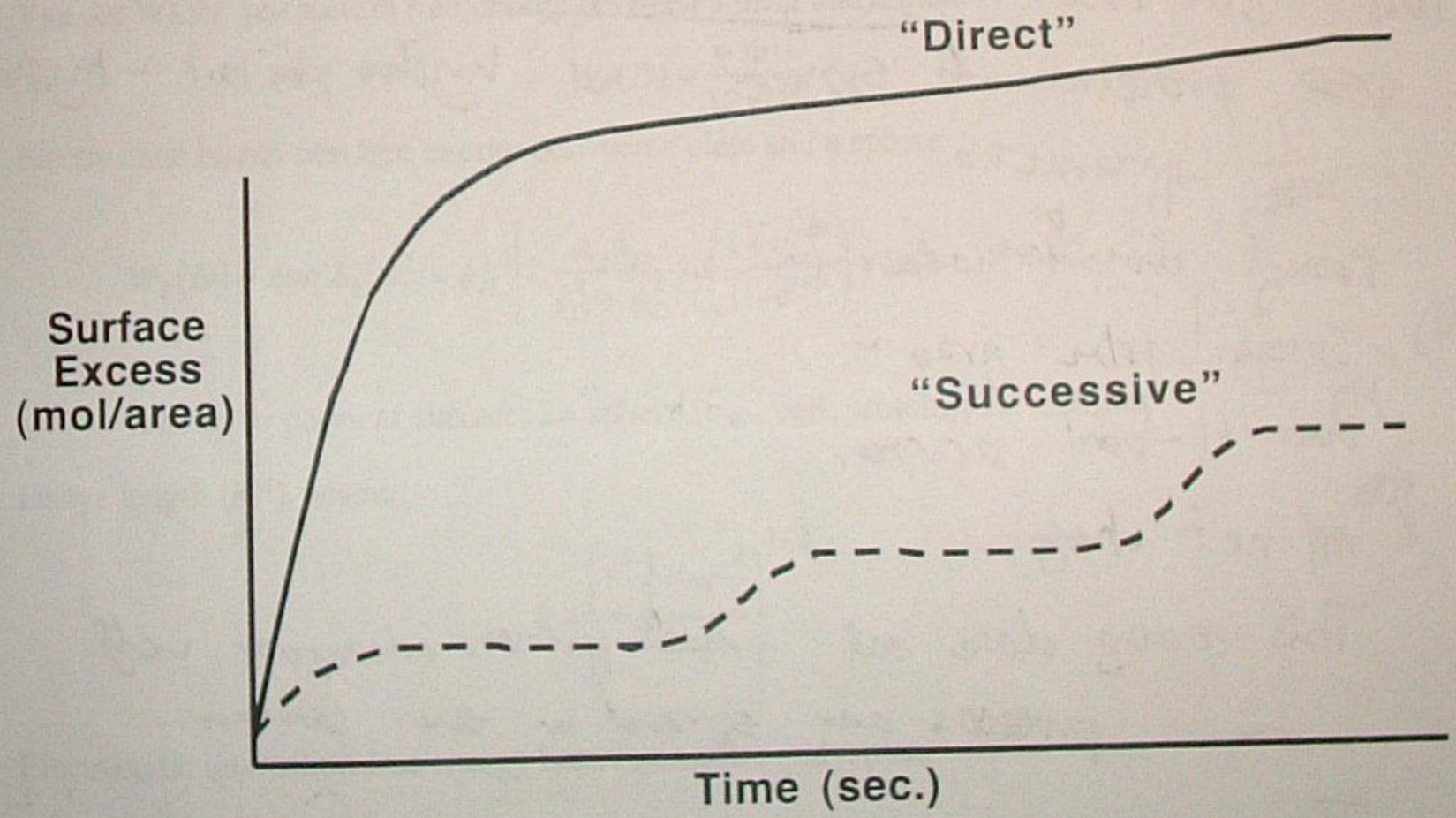
$W_A(D) = \frac{A_{123}R}{6D}$ ✓

4. You are asked to evaluate a coating that purportedly minimizes protein adsorption. (25 pts.)

a) What are the three key aspects of protein adsorption that affect the biological performance of a material?

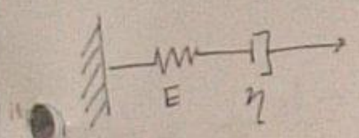
b) You conduct a protein adsorption experiment to address the aspects defined in a) to determine the magnitude of protein adsorption. How would you perform this experiment and what would you measure? What would a typical protein adsorption curve look like? Make sure to identify appropriate regions of the curve.

c) You then decide to conduct a protein adsorption experiment where you increase the concentration of protein successively after the surface excess (mol/area) has reached equilibrium. You obtain the curves below comparing "direct" versus "successive" protein adsorption isotherms. Why does the successive experiment have much less protein adsorbed to the surface? Is the coating hydrophobic or hydrophilic?



d) What are the "new" themes for surfaces that resist protein adsorption? Does this coating meet all of these characteristics? Do you think this is a good coating to resist protein adsorption?

——11 Element



constant strain rate

$\dot{\varepsilon} = \frac{d\varepsilon}{dt} = \text{const}$

$\sigma = \sigma_{ep} = \sigma_{dp}$

$\varepsilon = \varepsilon_{ep} + \varepsilon_{dp}$

$\sigma = \sigma_{ep} = E\varepsilon_{ep}$ $\quad \frac{d\varepsilon_{ep}}{dt} = \frac{1}{E}\frac{d\sigma}{dt}$

$\sigma_{dp} = \sigma = \eta \frac{d\varepsilon_{dp}}{dt}$

$\frac{d\varepsilon_{dp}}{dt} = \frac{\sigma}{\eta}$

$\frac{d\varepsilon}{dt} = \frac{d\varepsilon_{ep}}{dt} + \frac{d\varepsilon_{dp}}{dt}$

$\frac{d\varepsilon}{dt} = \frac{1}{E}\frac{d\sigma}{dt} + \frac{\sigma}{\eta}$

First order linear differential equation

$\frac{dy}{dx} + a_1(x)\, y = h(x)$ $\quad p = e^{\int a_1(x)\,dx}$ (integrating factor)

solution: $y = \frac{1}{p}\int p\, h(x)\,dx + \frac{c}{p}$

$\frac{1}{E}\frac{d\sigma}{dt} + \frac{\sigma}{\eta} = \frac{d\varepsilon}{dt}$

$\frac{d\sigma}{dt} + \frac{E\sigma}{\eta} = E\frac{d\varepsilon}{dt}$ $\quad \therefore a_1(t) = E/\eta$ $\quad h(t) = E\frac{d\varepsilon}{dt}$

$p = e^{\int E/\eta\, dt} = e^{Et/\eta}$

solution: $\sigma = e^{-Et/\eta}\int e^{Et/\eta} E\frac{d\varepsilon}{dt}\cdot dt + ce^{-Et/\eta}$ $\quad u = E/\eta$, $du = \frac{E}{\eta}dt$

$= e^{-Et/\eta}\cdot E\cdot\frac{d\varepsilon}{dt}\left(\frac{\eta}{E}\right)\int \frac{E}{\eta} e^{Et/\eta}\,dt + ce^{-Et/\eta}$

$= e^{-Et/\eta}\cdot\eta\cdot\frac{d\varepsilon}{dt}e^{Et/\eta} + ce^{-Et/\eta}$

$\sigma = \frac{d\varepsilon}{dt}\eta + ce^{-Et/\eta}$

boundary conditions: $t=0$, $\sigma=0$

$c = -\frac{d\varepsilon}{dt}\cdot\eta$

$\sigma = \eta\cdot\frac{d\varepsilon}{dt}\left(1 - e^{-Et/\eta}\right)$

$\sigma = \eta\,\dot{\varepsilon}\left[1 - e^{-\left(Et/\eta\dot{\varepsilon}\right)}\right]$ for the stress-strain curve

$t=0, \varepsilon=0$

$\frac{d\varepsilon}{dt}$ const

...LVE SOLUTION 1A)

$$\frac{d\varepsilon}{dt} = \frac{1}{E}\frac{d\sigma}{dt} + \frac{\sigma}{\eta} \qquad \frac{d\varepsilon}{dt} = \dot{\varepsilon} \cdot \text{CONSTANT (INDEPENDENT OF TIME OR } \sigma)$$

$$\dot{\varepsilon} = \frac{1}{E}\frac{d\sigma}{dt} + \frac{\sigma}{\eta} \longrightarrow \dot{\varepsilon} - \frac{\sigma}{\eta} = \frac{1}{E}\frac{d\sigma}{dt}$$

$$\int_0^t E\,dt = \int_0^\sigma \frac{d\sigma}{\dot{\varepsilon} - \frac{\sigma}{\eta}}$$

$$Et = -\eta \ln\left(\dot{\varepsilon} - \frac{\sigma}{\eta}\right)\Big|_0^\sigma$$

$$Et = -\eta \ln\left(\dot{\varepsilon} - \frac{\sigma}{\eta}\right) + \eta \ln(\dot{\varepsilon})$$

$$-\frac{Et}{\eta} = \ln\left(\dot{\varepsilon} - \frac{\sigma}{\eta}\right) - \ln(\dot{\varepsilon})$$

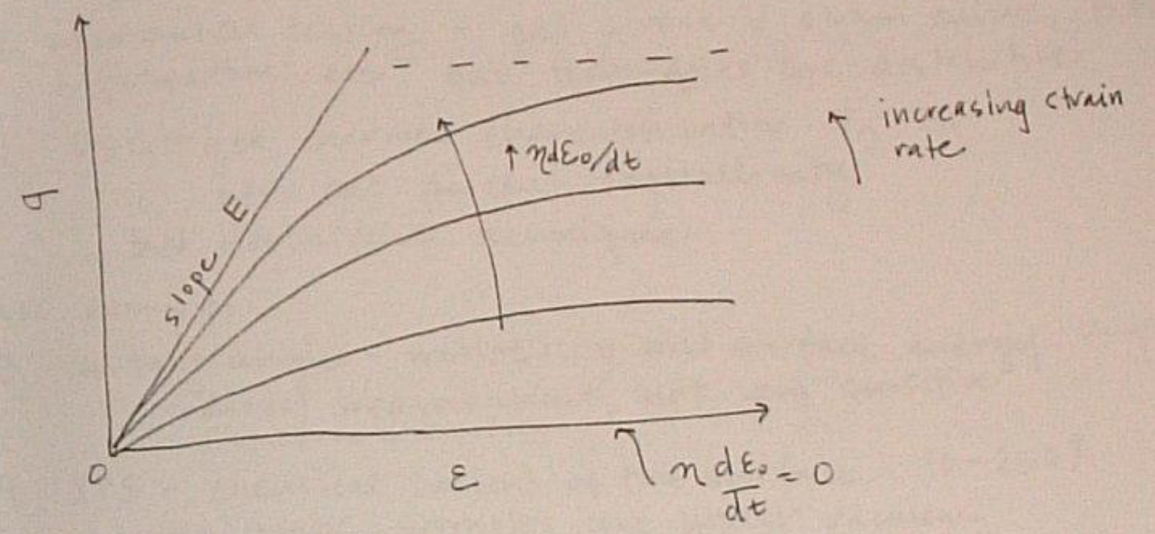
$$e^{-\frac{Et}{\eta}} = \frac{\dot{\varepsilon} - \frac{\sigma}{\eta}}{\dot{\varepsilon}} = 1 - \frac{\sigma}{\dot{\varepsilon}\eta}$$

$$\frac{\sigma}{\dot{\varepsilon}\eta} = 1 - e^{-\frac{Et}{\eta}}$$

$$\sigma = \dot{\varepsilon}\eta\left[1 - e^{-\frac{Et}{\eta}}\right]$$

$$\chi = \frac{\eta}{E}$$

$$\sigma = \eta \frac{d\varepsilon_0}{dt}\left[1 - e^{\left(-E\varepsilon/\eta \frac{d\varepsilon_0}{dt}\right)}\right]$$



(c) Yes, the model does qualitatively account for the observed viscoelastic properties of linear polymers during engineering stress-strain tests.

must list at least three techniques (both bulk + surface intended)

bulk examples:

1. mechanical testing - get stress v strain curves, fatigue information etc but techniques are destructive
2. DSC - get thermal characterization T_g, T_m
   also get percent crystallinity
   but destructive technique

surface examples:

3. contact angle - wettability and surface energy, very cheap
   indirect measurement, not very sensitive of surface chemistry
4. XPS - chemical content of the surface 10-250 Å
   expensive, samples are under vacuum
5. AFM - topographic images of the surface, quantitative
   very low resolution, difficult to operate

(b) interfacial hierarchy

4pts. involves the progression of events starting from the initial molecular (ions, small molecules) adsorption, through protein or macromolecular adsorption, to cell adhesion and ultimately tissue organization and adaptation over the long-term

initial events: ion adsorption, protein and lipid adsorption, protein exchange, conformational change

cellular events: cell adhesion, attachment, spreading
phenotypic expression

tissue level events: tissue formation in response to implant reorganization over time with chemical, electrical, and mechanical stimuli

(c) long-term interactions:

1 pt. host responds to material

material responds to host

mat	host
corrosion	inflammation
wear	tissue remodeling
degradation	immunological
fatigue	
fracture	

(d) dynamic interaction?

3pts. affect each other and new materials are exposed to each other

W(D) interation free energy is the sum of the electrostatic repulsion:

$$W_E = \pi \varepsilon \varepsilon_0 R (\phi_{13}^2 + \phi_{23}^2)\left[2\frac{\phi_{13}\phi_{23}}{\phi_{13}^2+\phi_{23}^2}\ln\left(\frac{1+e^{-\kappa D}}{1-e^{-\kappa D}}\right) + \ln(1-e^{-2\kappa D})\right]$$

and the Van der Waals attraction: interaction free energies.

$$W_A = -\frac{AR}{6D} \qquad W(D) = W_E(D) + W_A(D)$$

(b) $\kappa = \left(\frac{e^2 \sum_i \rho_{i\infty} z_i^2}{kT\varepsilon\varepsilon_0}\right)^{1/2}$

$e = 1.602 \times 10^{-9}\ C$
$k = 1.381 \times 10^{-23}\ J/K$
$\varepsilon = 8.854 \times 10^{-12}\ C^2/J\cdot m$
$\varepsilon_{H_2O}(T = 37°C) = 74.8$

$$\rho_{CaCl_2} = C \cdot N_A = \left(\frac{0.2\,mol}{L}\right)\left(\frac{1000\,L}{m^3}\right)\left(\frac{6.02\times10^{23}\ molec}{mol}\right) = 1.204\times10^{26}\ \frac{molec\,CaCl_2}{m^3}$$

$\rho_{Ca^{+2}} = \rho_{CaCl_2}$

$\rho_{Cl^{-1}} = 2\rho_{CaCl_2}$

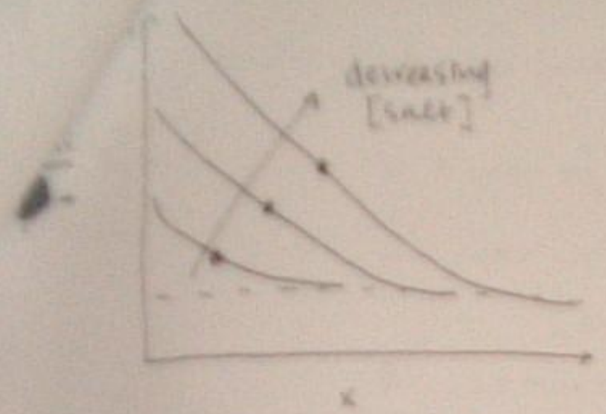
$$\kappa = \frac{(1.602\times10^{-19})^2\left[(1.204\times10^{26})(+2)^2 + (2)(1.204\times10^{26})(-1)^2\right]}{(1.381\times10^{-23}\ J/K)(310)(74.8)(8.854\times10^{-12})}$$

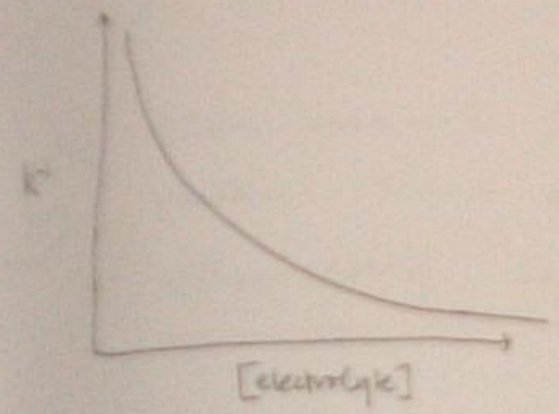
$$\left(\frac{[C^2][1/m^3]}{[J/K][K][C^2/J\cdot m]}\right)^{1/2} = m^{-1}$$

$\kappa = 2.56\times10^{9}\ m^{-1}$

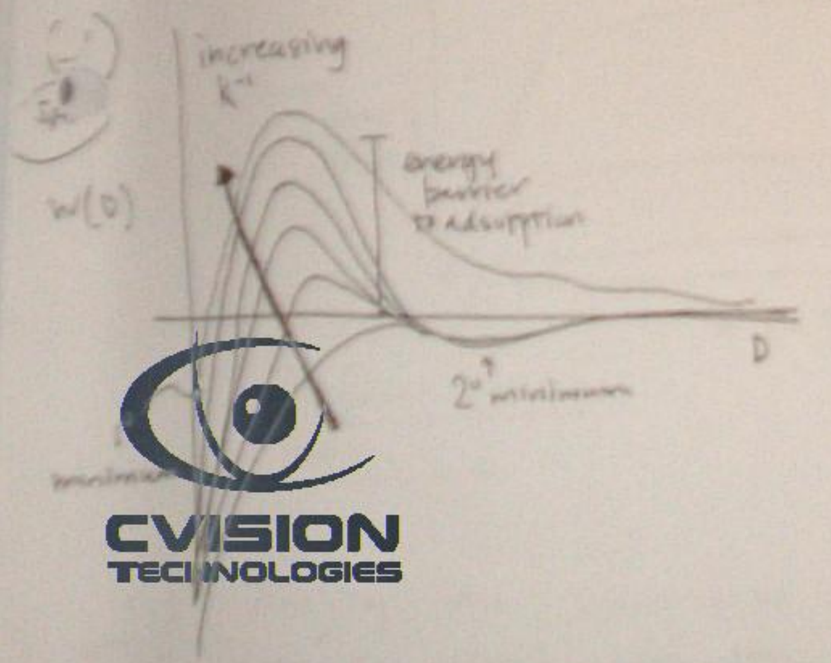
$\kappa^{-1} = 0.391\ nm$ @ 37 °C

debye length for a 1:1 solution is larger than for a 2:1 solution





- κ^{-1} is most sensitive to electrolyte concentration and type



(e) In a solution of both cells and proteins, initially the cells will be held in the 2° minimum while the proteins will adsorb to the surface. The protein adsorption will lower the potential of the surface, and decrease the energy barrier for the cell. The cell is then able to reach the 1° minimum and irreversibly adsorb to the surface.

c) or different amount of ~~total~~ protein adsorbed during the direct and successive experiments.

d) Neo "themes." The surface contains

(δ pts.)

1. Polar functional groups
2. Incorporate hydrogen bond accepting groups
3. N= net charge

Although Whitesides reports that surfaces should not contain hydrogen bond donating groups, there are some examples (i.e., the cell surface) that invalidate this rule.