Ag-Ab reactions
Tests for Ag-Ab reactions

By
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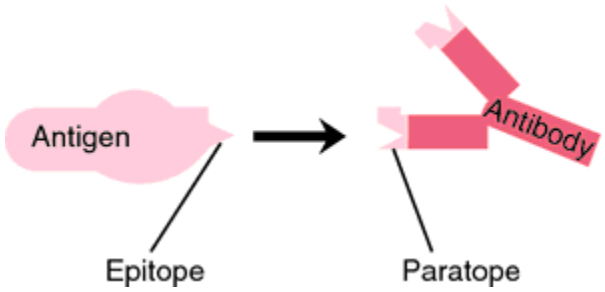
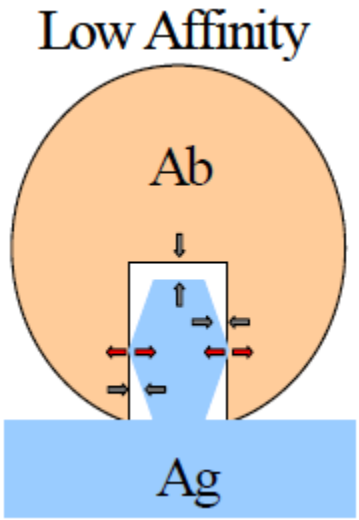
Nature of Ag-Ab reactions

- Lock and key model
- Non covalent interactions
  - Hydrogen bonds
  - Electrostatic bonds
  - Van der Waals forces
  - Hydrophobic bonds
- Multiple bonds involved at binding site
- Reversible bonding



Affinity

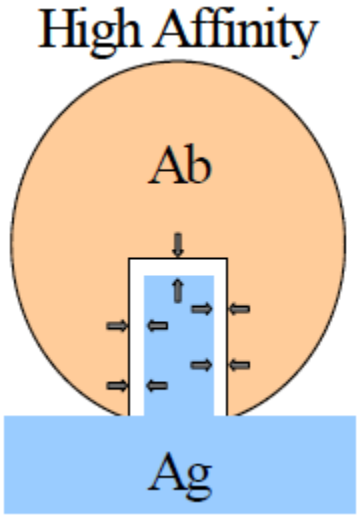
- Strength of reaction between a single epitope (Ag determinant) and paratope (Ab binding site)



$$\mathbf{Ag + Ab \leftrightarrow Ag\text{-}Ab}$$

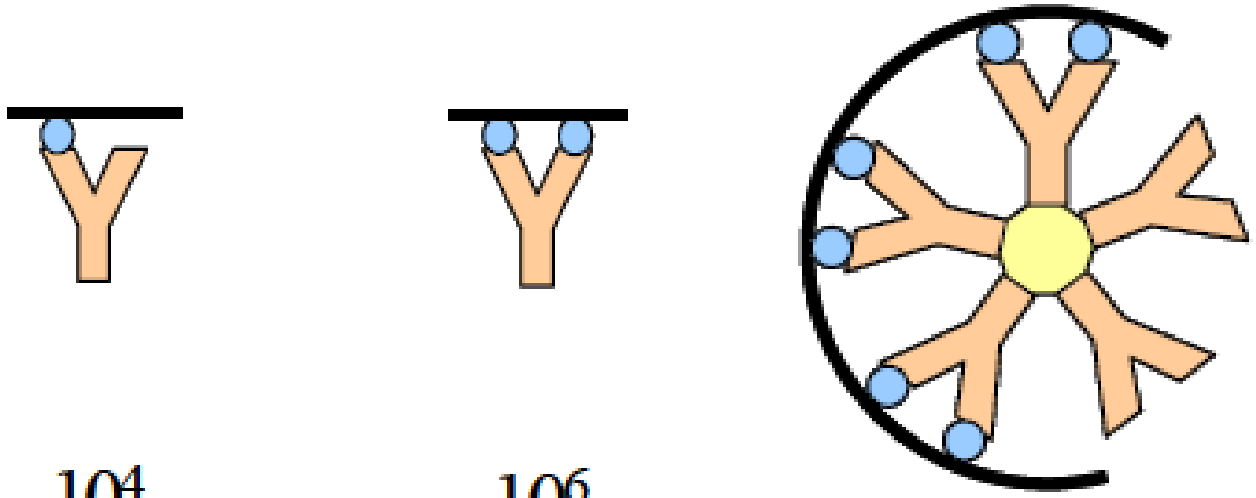
Applying the Law of Mass Action:

$$\mathbf{K_{eq} = \frac{[Ag\text{-}Ab]}{[Ag]\ x\ [Ab]}}$$



Avidity

- The overall strength of binding between an Ag with many determinants and multivalent Abs



K_{eq} = 10^4 Affinity

10^6 Avidity

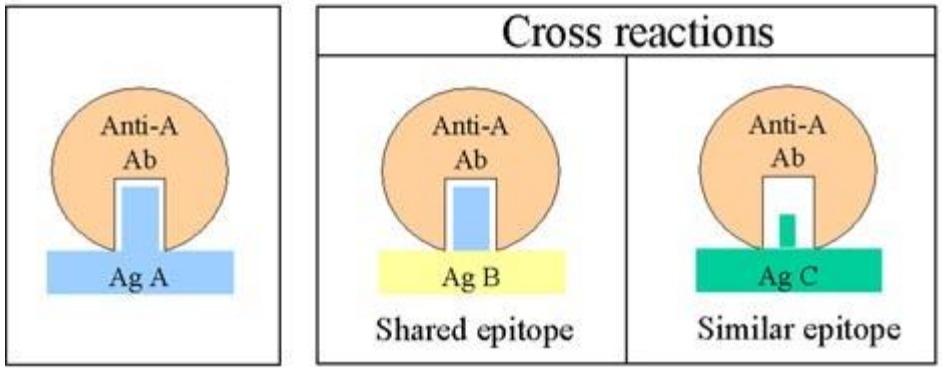
10^{10} Avidity

Specificity

- Ability of an individual antibody combining site with only one antigenic determinant
- The ability of a population of antibody molecules to react with only one antigen
- There is a high degree of specificity in Ag-Ab reaction
- Abs can distinguish differences in
  - Primary structure of an Ag
  - Isomeric forms of an Ag
  - Secondary and tertiary structure of an Ag

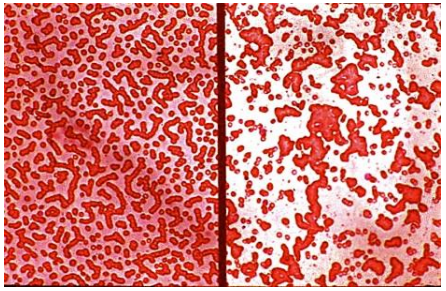
Cross Reactivity

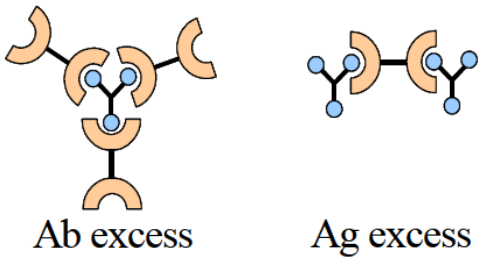
- Ability of an individual antibody combining site with more than one antigenic determinant
- Ability of a population of Ab molecules to react with more than one Ag
- Cross reactions arise because the cross reacting antigen shares an epitope in common with the immunizing antigen or because it has an epitope which is structurally similar to one on the immunizing antigen

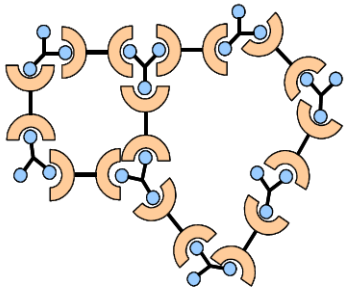


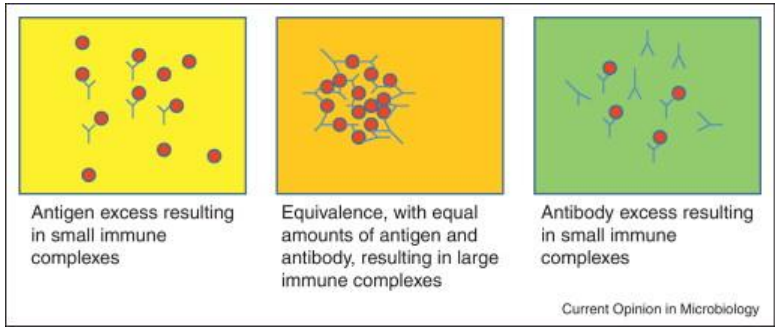
Factors effecting Ag-Ab reactions

- Affinity
- Avidity
- Ag:Ab ratio









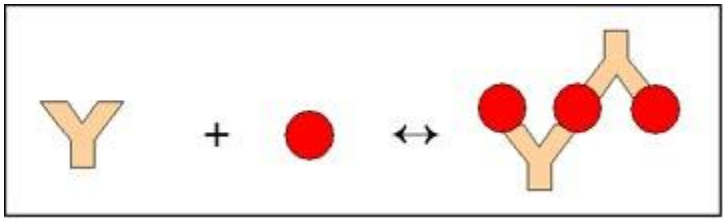
- Physical form of Ag (particulate or soluble)

Tests based on Ag-Ab reactions

- All tests based on Ag-Ab reactions will depend on lattice formation or use ways to detect small immune complexes
- All tests based on Ag-Ab reactions can be used to detect either Ag or Ab
- Mainly two types of tests
  - Agglutination
  - Precipiation

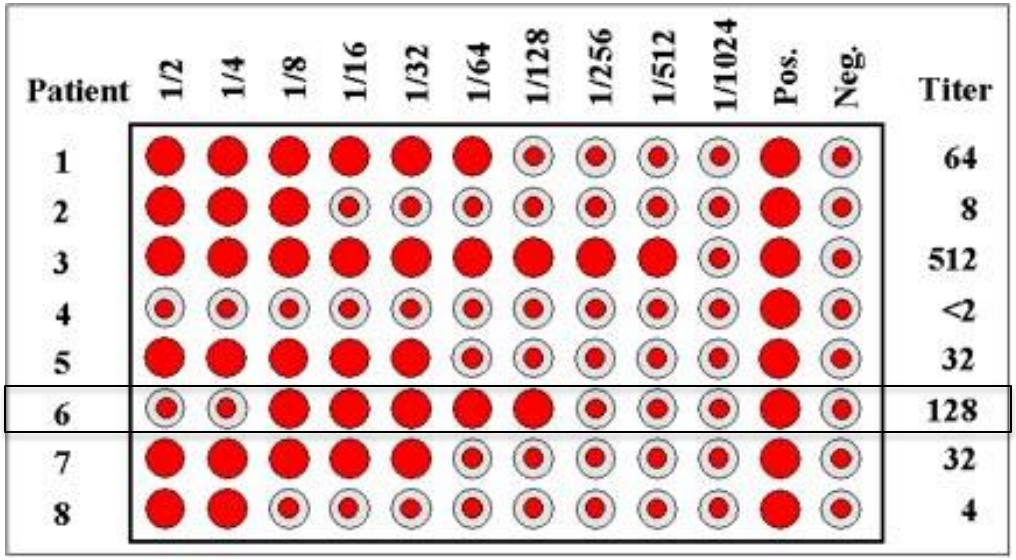
Agglutination tests

- Tests that have as their endpoint the agglutination of a particular Ag
  - Agglutinin (Ab participatin in agglutination)
- Qualitative agglutination test
  - Blood typing



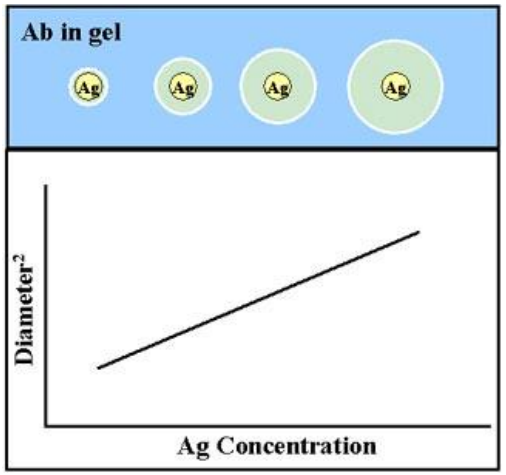
Agglutination tests

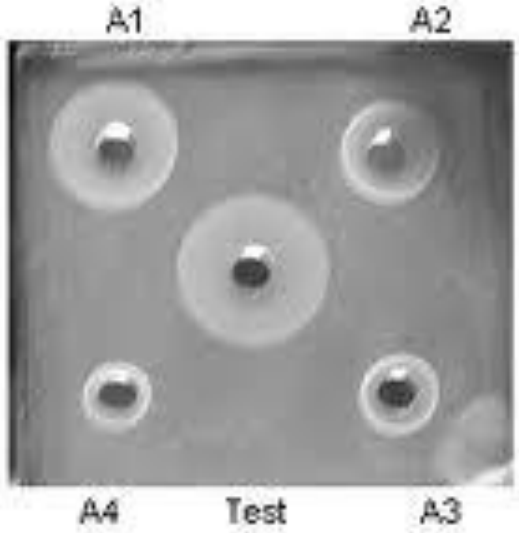
- Quantitative agglutination test
  - Titer
  - Prozone



Precipitation tests

- Radial immunodiffusion
  - Ag in the well
  - Ab in the well

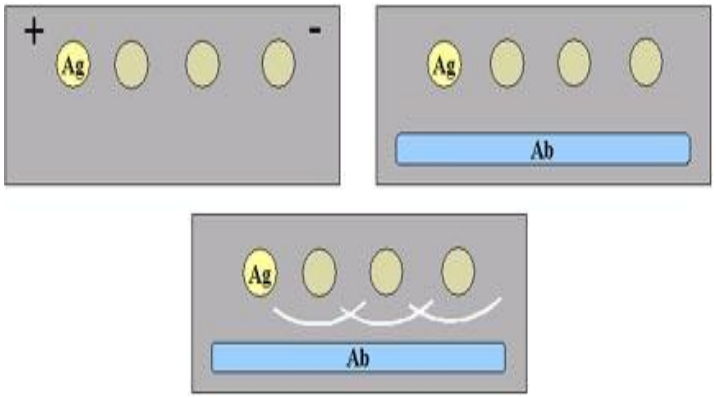


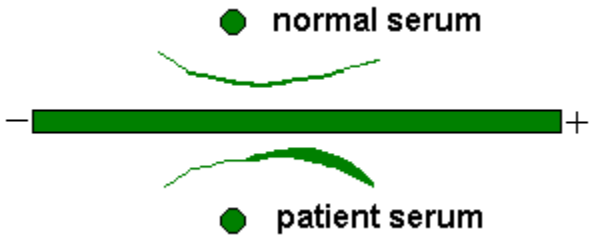


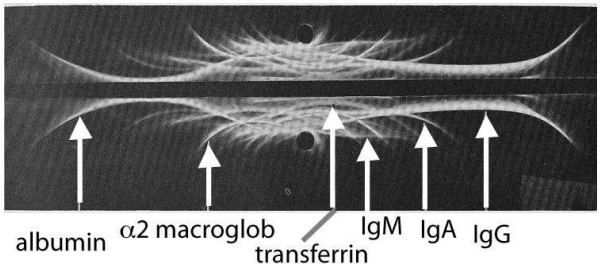
- Diameter of the ring is proportional to the concentration

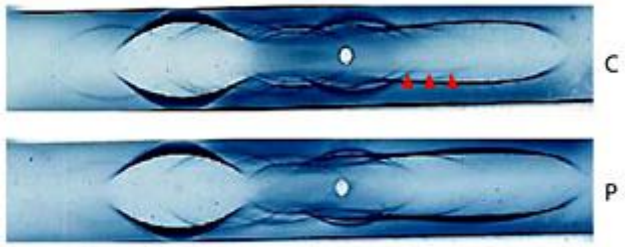
Immuno electrophoresis

- Used to for complex mixtures of Ag (serum)
- Ag are separated by electrophoresis
- Ab is placed in through cut in the agar



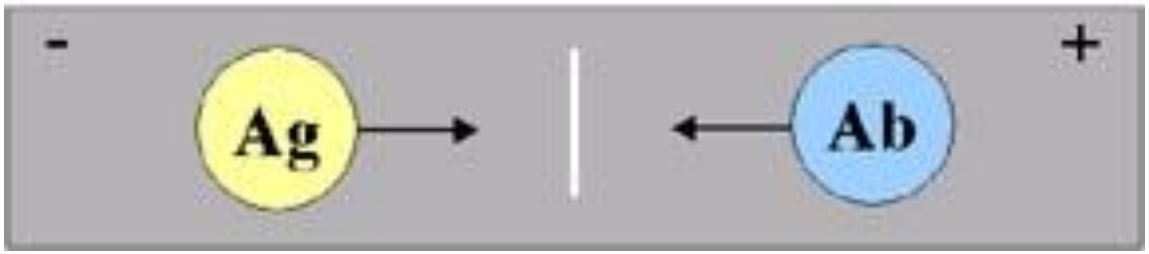






Countercurrent electrophoresis

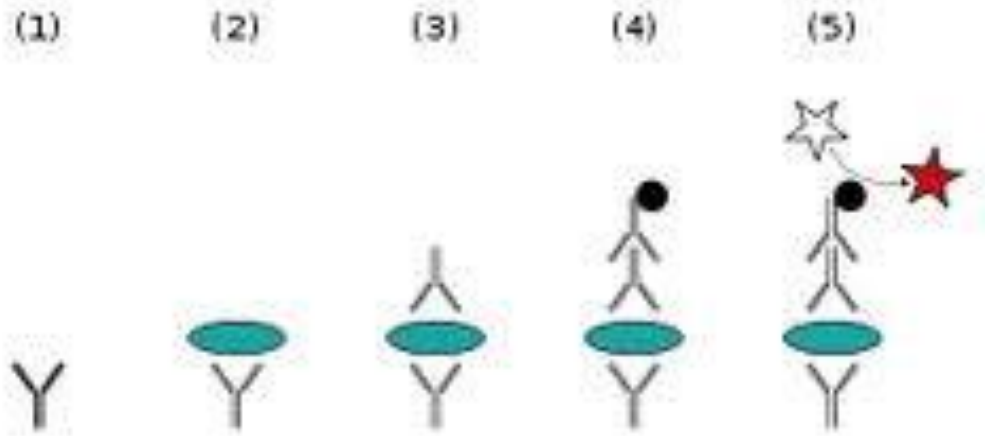
- Ag and Ab migrate toward each other by electrophoresis
- Can be used when Ag and Ab have opposite charges



- Very limited use but gives very fast results
  - (crypto coccal Ag)

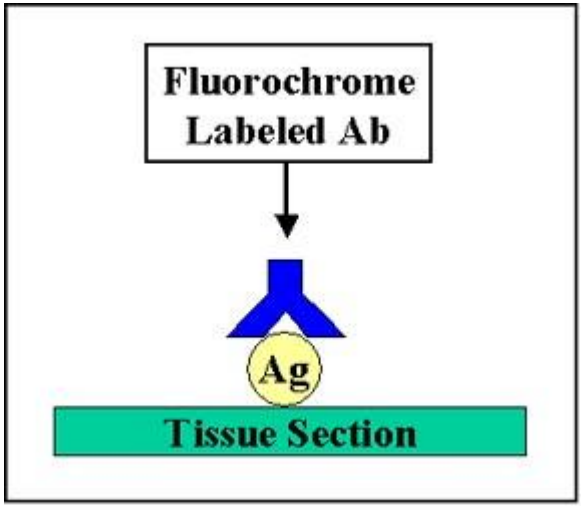
Radioimmunoassays (RIA)

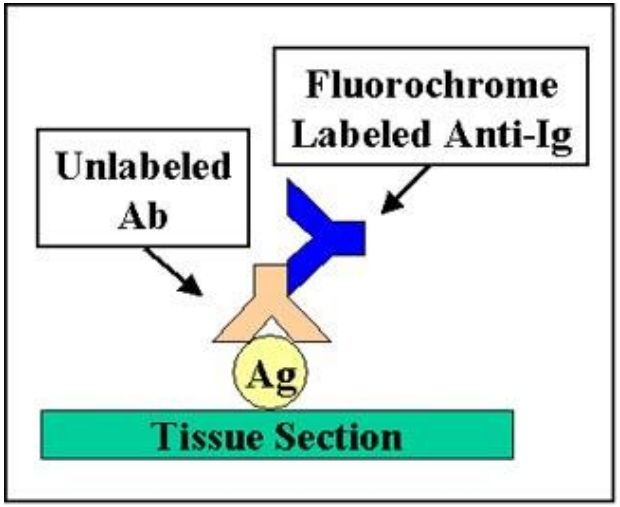
- DO NOT REQUIRE LATTICE FORMATION
- Enzyme-Linked Immunosorbent Assays (ELISA)



Tests for cell associated Ags

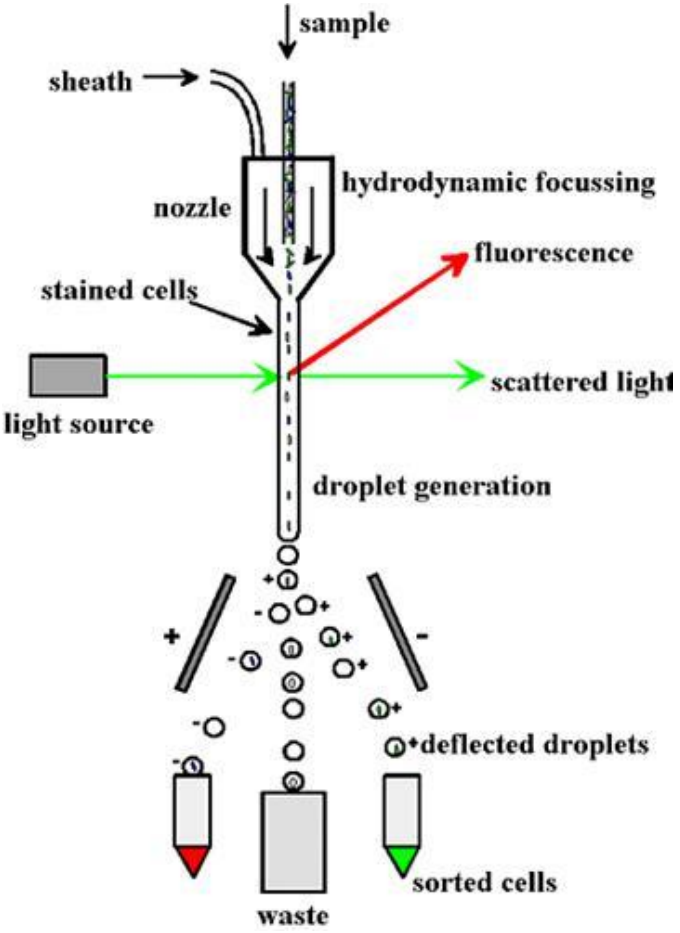
- Fluoresence
  - Direct
  - Indirect

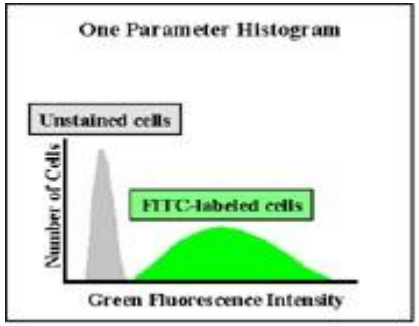


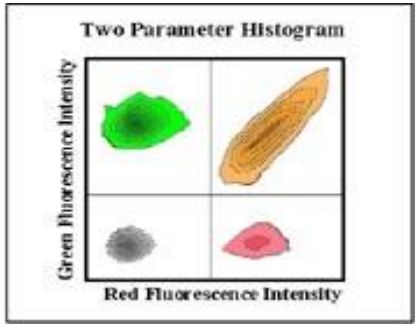


Tests for cell associated Ags

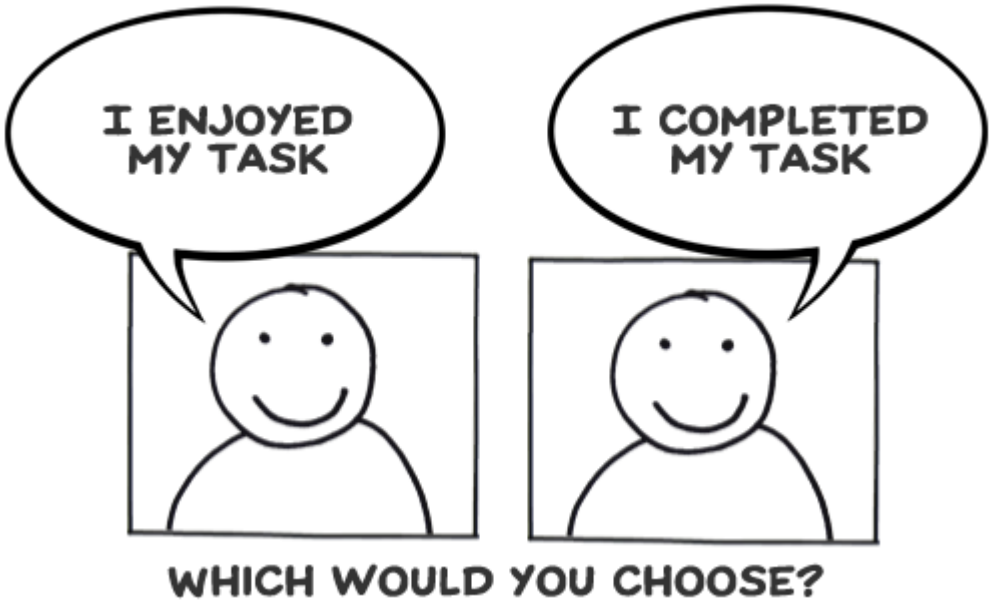
- Flowcytometry
- Fluorescent activated cell sorter (FACS)





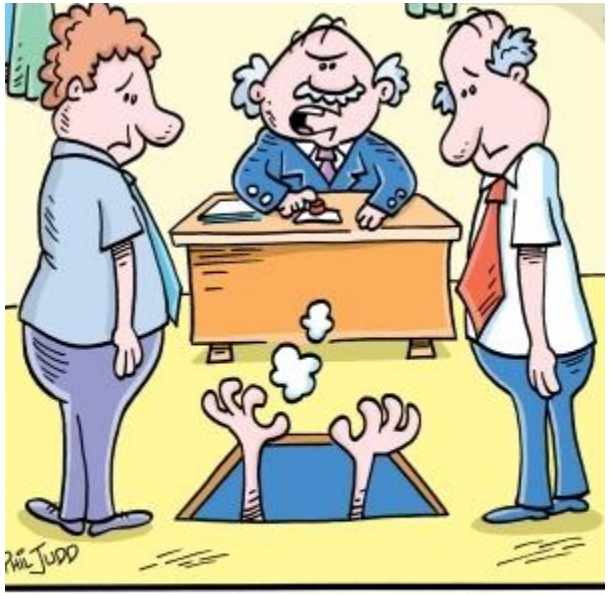


Completed / enjoyed



Suggestions & complaints

Comments encouraged!



"Does anyone else have any complaints?"