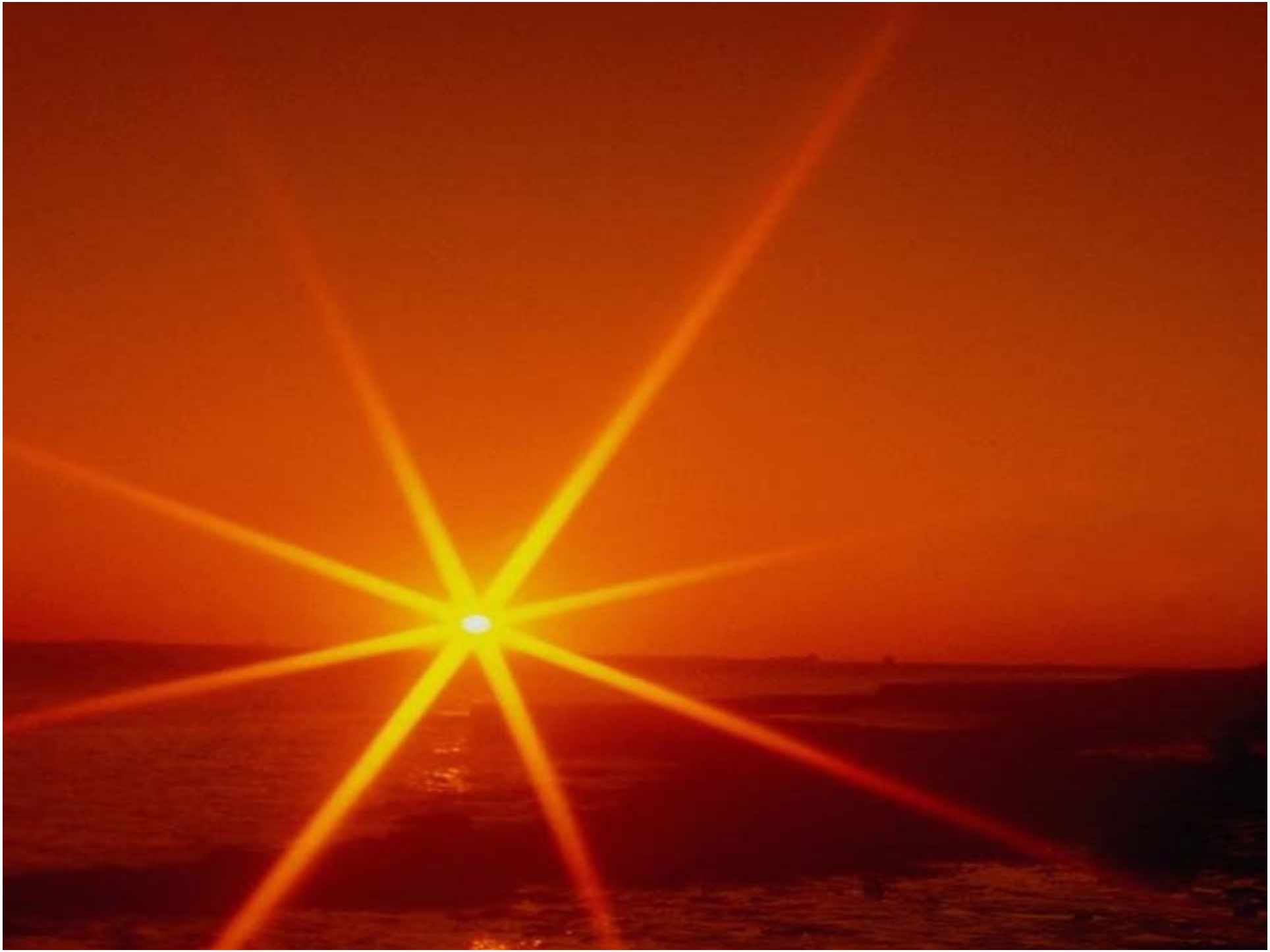


بسم الله الرحمن الرحيم

Laboratory Evaluation of Deficiencies in

Humoral Factors

of the

Innate & Adaptive Immune Systems

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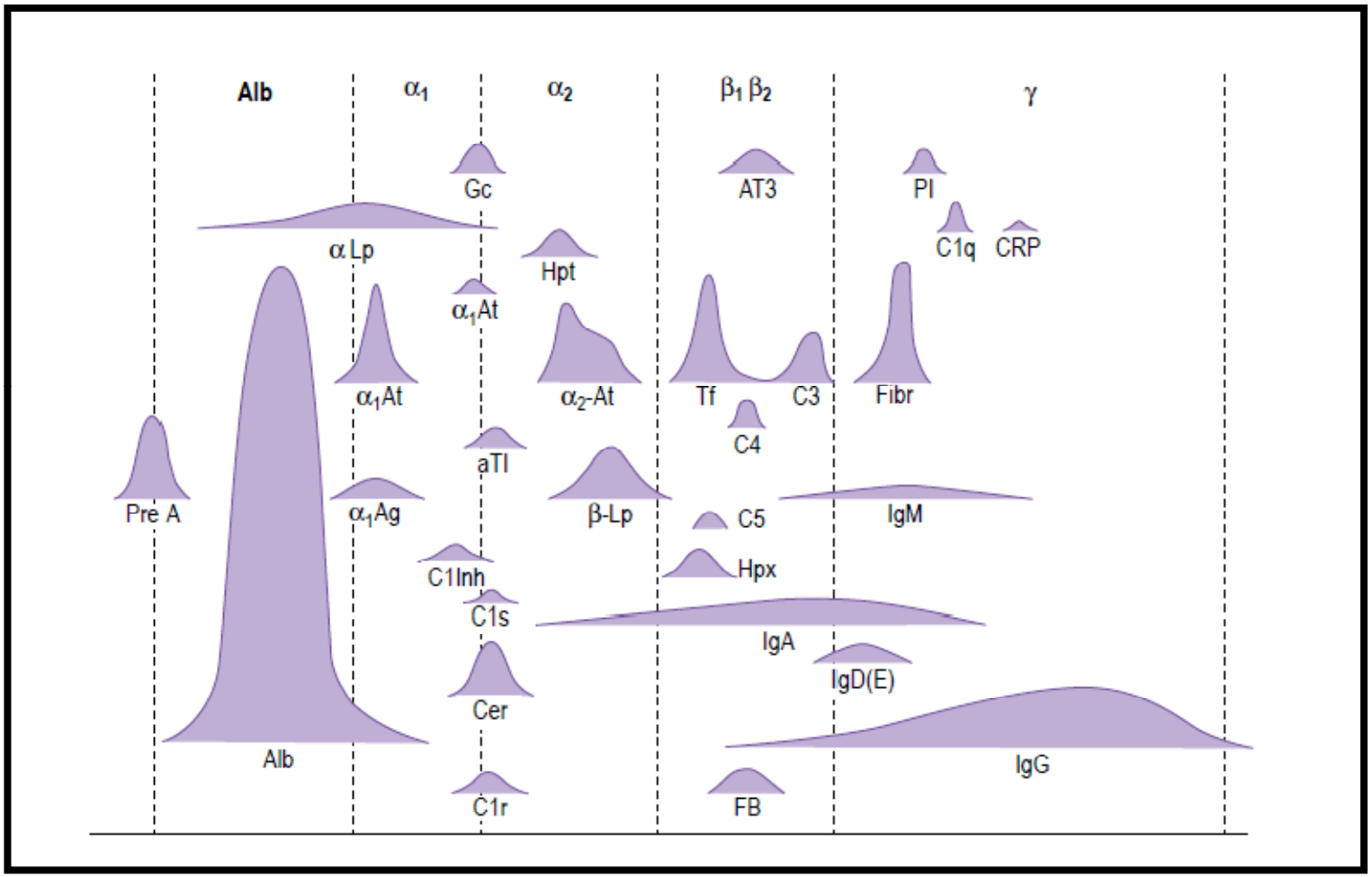
by **Humoral Factors** I mean:

- Acute Phase Reactants (e.g. CRP)
- Cytokines (e.g. Interferones)
- Complement Components
  - **(Activatory & Inhibitory) ; (Functional & Structural)**
- Gamma Globulins, Antibodies, Immunoglobulins
  - **SPE,**
  - **Ig Level, Class & Subclasses (first IgA, then G & M)**
  - **Titer, (with CH50!) etc.**
  - **Natural Ab(only IgM),Anti-Protein vs. Anti-Glucid(IgM&IgG),**
  - **Basal level & post-challenge state,**
  - **Affinity, Memory, Protective, etc.**
  - **in serum or secretions**
- Ab Producing Cells (by FCM or Other Techniques)

FCM Technique
(cellular or Humoral ?!)

- Approximately ٨-10% of circulating lymphocytes are B cells,
- which are absent in:
  - X-linked agammaglobulinemia (XLA,(
- and present in:
  - CVID,
  - IgA deficiency,
  - Hyper-IgM syndromes.
- Patients found to be agammaglobulinemic should have their blood B cells enumerated by flow cytometry using B-cell-specific CD antigens (usually CD١٩ or CD.(٢٠


Alb
α1
α2
β1 β2
γ
Gc
AT3
PI
C1q
CRP
α Lp
Hpt
α1At
α1At
α2-At
Tf
C3
Fibr
C4
aTI
Pre A
α1Ag
β-Lp
C5
IgM
C1Inh
Hpx
C1s
IgA
IgD(E)
Cer
Alb
IgG
C1r
FB

Immunoglobulin Quality

- The evaluation of **immunoglobulin quality** is complex and can be difficult to assess.
- **Quality considerations involve:**
  - antibody repertoire & antigen-specific immune responses;
  - Class-Specific Immunoglobulin (Ig vs Ab);
  - specific avidities for antigens;
  - development of immunologic memory;
  - etc.

- This is of critical relevance because subjects incapable of generating **protective antibody** responses are more susceptible to infection and, under many circumstances, can benefit from immunoglobulin replacement therapy.

Indications & Applications

- **Which Patient? Which Test?**
  - Hx(Fam. & Med.);PE(signs & symmptoms); Clin Sense!
  - Reproducible, cost Effective, Informative, etc.
- **When Tested?**
  - >6mo? >2yr? Before IVIG! etc.
- **Where to be Tested?**
  - The same Lab along with the previous sample
- **Which Lab. Method to be Selected?**
  - Agglu? SRID? Nephelo? Turbidi? ELISA? Flow?
- **How to be Assessed?**
  - Screening,Eclusive/Supportive,Inclsv/Confirmatory?
  - Baseline or post challenge?
- **Regarding the Many Interpretation Notes!**
  - False Pos (Hi C3 for Pre-analytical Errors)
  - False Neg (Lo C3 @ snapshot due to H/I def.)
  - ASO neg ; HbsAg+ while HBsAb- ; Tolerized??
  - HD Widal Test ; Protein Loss (GI / UT) ;
  - Hook Effect(ELISA IgE)&Zone Phenomenon(SRID IgM)

Test Selection

- A variety of laboratory-based tools (as simple as **Agglutination** Methods to more complex methods such as **FCM** & Molecular Methods) are available for the evaluation of suspected PIDDs with deficits in humoral immunity. These include :
  - direct genetic diagnosis of single-gene disorders,
  - flow cytometric analysis of lymphocyte subpopulations,
  - quantitative and qualitative evaluation of serum immunoglobulins.
- Age, sex, environmental exposures, medications, and geography can influence some of these measures, but these tests are, in the vast majority of cases, **objective** and **useful** for providing definitive diagnoses.

Rationale 4 Isohaemagglutinin Assays

- Natural antibodies are found in every serum. These IgM antibodies are produced by non-conventional B 1 (CD5+ Lo IgD) cells.
- They tend to have specificity for bacterial antigens as well as autoantigens. In general, they are both of low affinity and polyreactivity.
- These antibodies represent a second important and immediate defense against infectious organisms.

Vaccine Challenge Tests

- A major diagnostic intervention in the consideration of many patients with primary immunodeficiency diseases (PIDs) is the application and interpretation of vaccination.
- Antibody response to antigenic challenge with vaccines provide valuable information on immune function.
- Numerous vaccines are commonly used in children ,as well as others that are available for specialized applications.
- Both can potentially be used to facilitate consideration of PIDD.

- Protein vaccines – Tetanus and diphtheria are representative protein antigens to which most patients have been exposed through primary and booster vaccination.
- The response to the conjugate vaccine for Hemophilus influenzae type B (Hib), while it includes antibodies against Hemophilus influenza surface polysaccharide, is determined by the response to the protein component of the conjugate and thus represents a protein response.

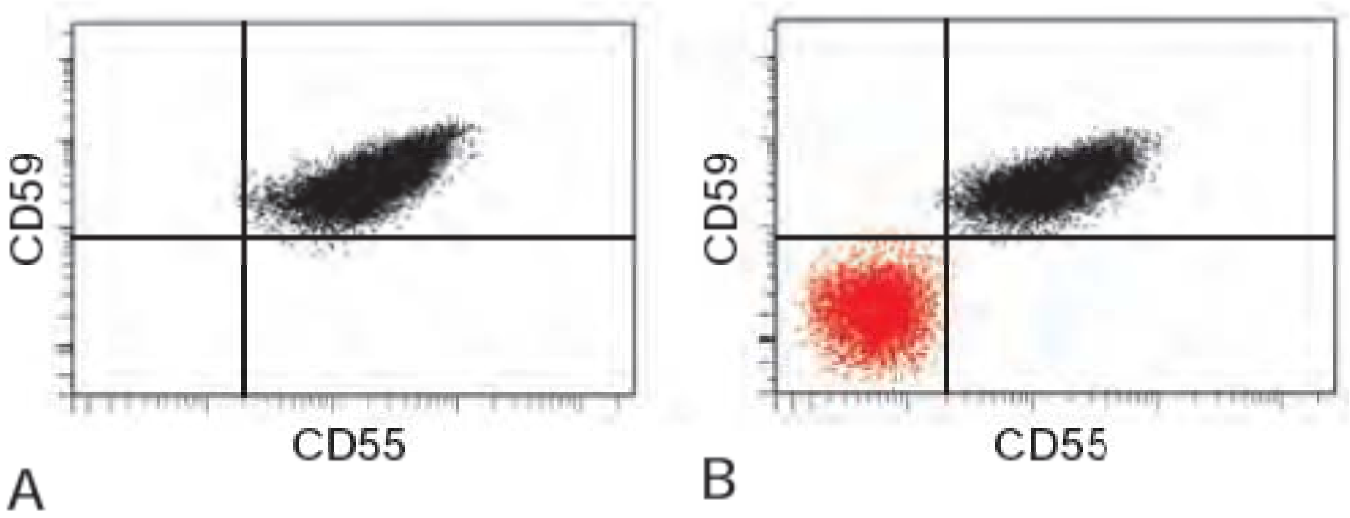
- Polysaccharide vaccines – These include capsular polysaccharides from pathogenic strains of pneumococcus to which patients may have been exposed through infection or vaccination.
- Occasionally, antibodies to polysaccharide antigens in the meningococcal vaccine can also be measured, although the "normal" response to meningococcal vaccines is not as well-characterized .

LABORATORY ISSUES

For the VACCINE Challenge Tests:

- both pre- and postvaccination titers
- by the same laboratory
- by the same assay method
  - nephelometry
  - turbidimetry
  - SRID
  - ELISA, FELISA & other quasi-ELISA methods)

Paroxysmal nocturnal hemoglobinuria (PNH)



A- Normal individual shows that the red cells express both phosphatidyl Inositol Glycan (PIG)-linked membrane proteins, CD۵۵ and CD۵۹, on their surfaces.

B- Patient with PNH shows a population of red cells that is deficient in both CD۵۵ and CD.۵۹

As is typical of PNH, a second population of CD۵۵+/CD۵۹+ red cells that is derived from residual normal hematopoietic stem cells is also present.

از توجه شما متشکرم


جمعه خوبي داشته باشيد!