Screening for Immune Deficiency

Charlotte Cunningham-Rundles MD PhD
Mount Sinai School of Medicine

Conflicts of Interest

1. Talecris/Grifols Medical Advisory Board
2. Baxter Heathcare: research funding for project on the Statewide Planning and Research Cooperative System (SPARCS) data base seeking use of IDC codes in primary immune deficiency in NYS.
3. Octapharma: research grant to dissect antibody deficiencies to guide Ig therapy

Who to screen?

Ten Warning *Signs* of Immunodeficiency

(two or more indicates that an evaluation is needed)

- Two or more episodes of pneumonia.
- Unexplained weight loss in adults; failure to thrive in infants.
- Recurrent deep organ or skin abscesses.
- One or more episodes of serious infections such as meningitis, sepsis, cellulitis, osteomyelitis
- Family history in immune deficiency.
- Recurrent ear infections; need for tubes in an adult.
- Oral or cutaneous candidiasis after age one.
- Two or more months on oral antibiotics with little effect.
- Need for IV antibiotics to clear infections.
- Two or more sinus infections in one year.

Unexplained elements in the history

- Lymphadenopathy
- Splenomegaly
- History of auto-immune disease
- Serious infections
- Chronic infections
- Unexplained lung disease
- Scarring herpes zoster
- Myringotomy as an adult
- Prior diagnosis of an immune defect
- Bronchiectasis
- Family history of an immune defect
- Malabsorption
- Weight loss
- Joint pain and swelling

Additional Clinical Signs of Immunodeficiency

- History of autoimmune disease, (especially ITP and AHA)
- Bronchiectasis
- Enlarged spleen and /or lymph nodes
- Scarring herpes zoster
- Diarrhea
- Arthralgia, arthritis
- Weight loss, intermittent fever

Immune Defects in Refractory Sinusitis in Adults

- 79 patients average age 44 with 2.94 (± 2.19 SD) previous operations.
- 50.6% had at least one positive result on skin test to an aeroallergen.
- Low IgG in 14 of 78 patients (18%), low IgA in 13 of 78 (17%), and low IgM in 4 of 78 (5%).
- Common variable immunodeficiency (CVID) was diagnosed in 10% of patients, and selective IgA deficiency was found in 6%.

L. Chee et al, Laryngoscope 2001;111:233-235

Immunologic defects in pediatric patients with refractory sinusitis

- 61 children with chronic sinusitis were evaluated.
- Recurrent otitis media and asthma exacerbation were common.
- 5 had an elevated IgE level; 22 patients had positive prick tests to one or more environmental inhalants.
- 11 had low Ig levels, 6 had low Ig levels and vaccine hyporesponsiveness, and 17 had poor vaccine response only.

Shapiro et al, Pediatrics 1991;87:311-6.

Bronchiectasis in children

- 1994 British study of 4000 children referred for evaluation of chronic suppurative disease (excluding CF) found incidence of bronchiectasis to be 1% (40 children).

- In 63% an etiology was found:

27% immune disorder (11)

17% ciliary dyskinesia (7)

15% congenital malformation (6)

5% foreign body (2)

Bronchiectasis in Adults

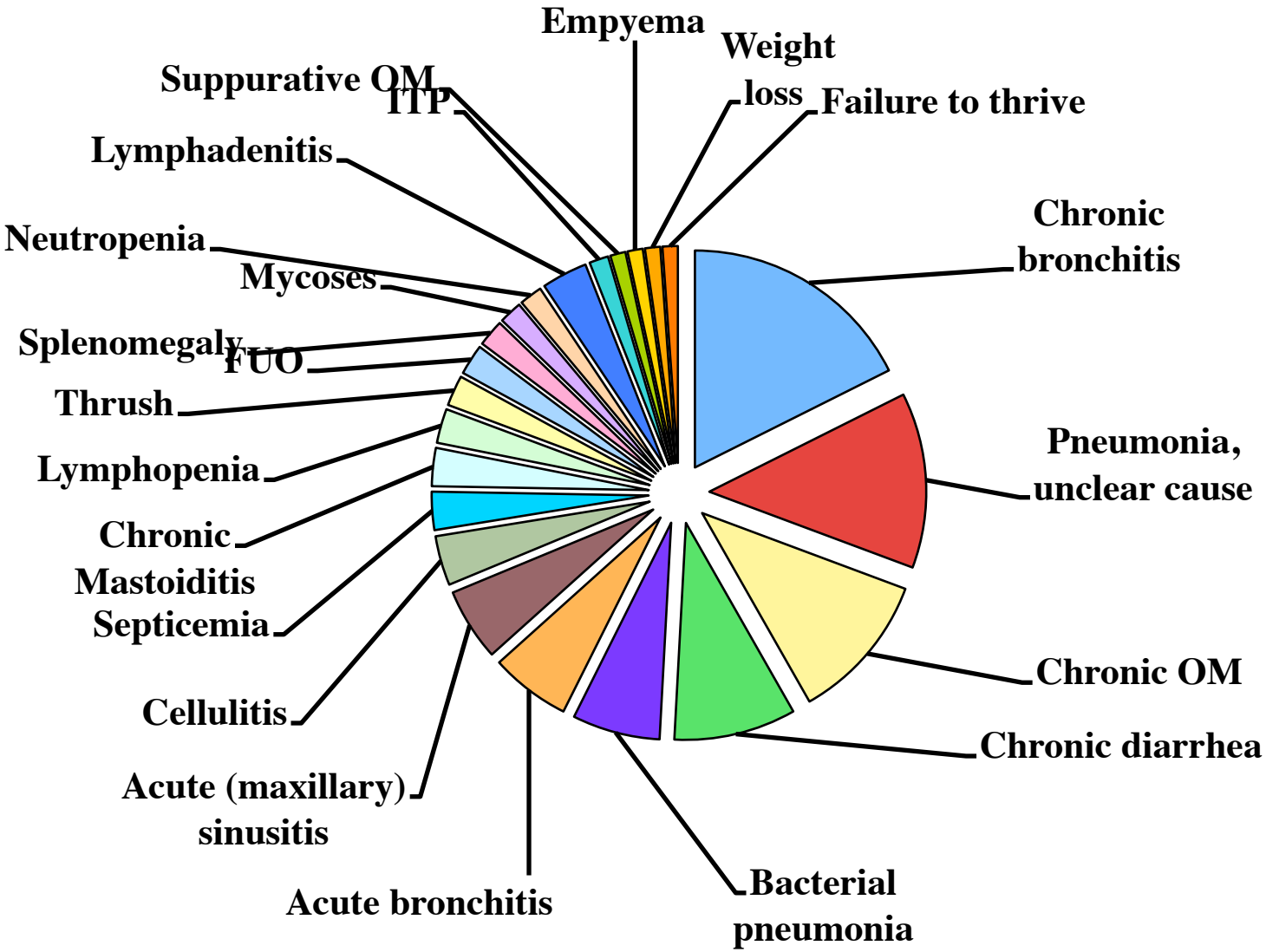
- 150 patients with bronchiectasis, proven by HRCT.
- Mean age = 52.7 years
- Median age at onset symptoms = 14 years
- 11 (7%) ABPA; aspiration 6; CF 4; ciliary defects 3, pan-bronchiolitis 1, congenital defect 1.
- 12 (8%) patients had humoral defects: 1 CVID, 6 isolated IgG subclass deficiency, 3 IgA deficiency, 1 IgM deficiency; 6 had antibody deficiency.

Pasteur et al, Am J.Respir Crit Care Med, 162, 1277-1284, 2000

Pulmonary signs indicate that an immune defect might be present:

- Recurrent pneumonia with low virulence organisms (*H flu, S pneumoniae*, mycoplasma, others)
- Empyema complicating pneumonia
- Unexplained Bronchiectasis
- Unexplained lung abscess,
- Unexplained Obstructive lung disease
- Unexplained Restrictive lung disease
- Lymphocytic interstitial infiltrates
- Bronchospasm with repeated infections.
- Granulomatous disease with recurring infections and low immunoglobulins.

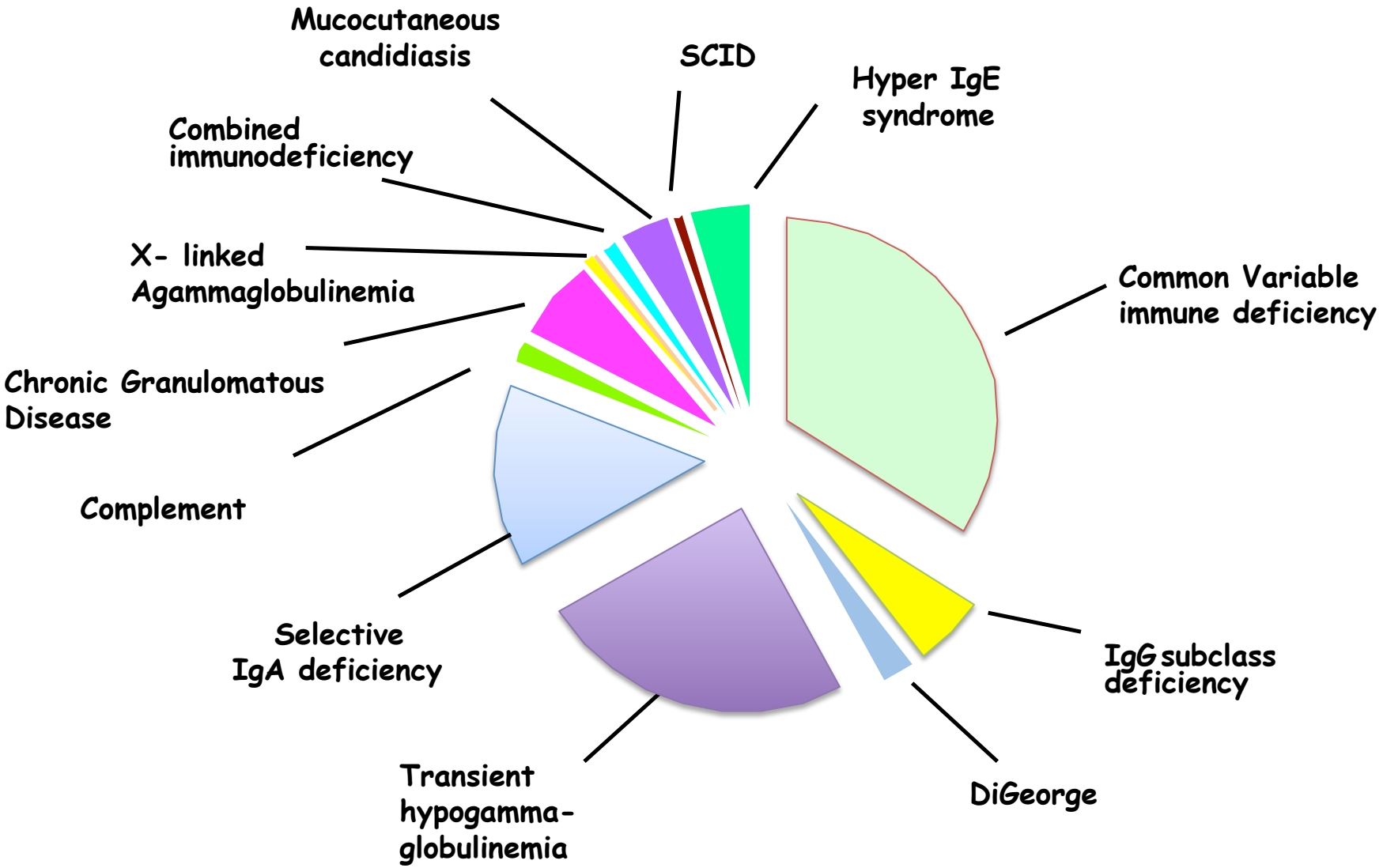
Conditions of 100 immune deficient patients



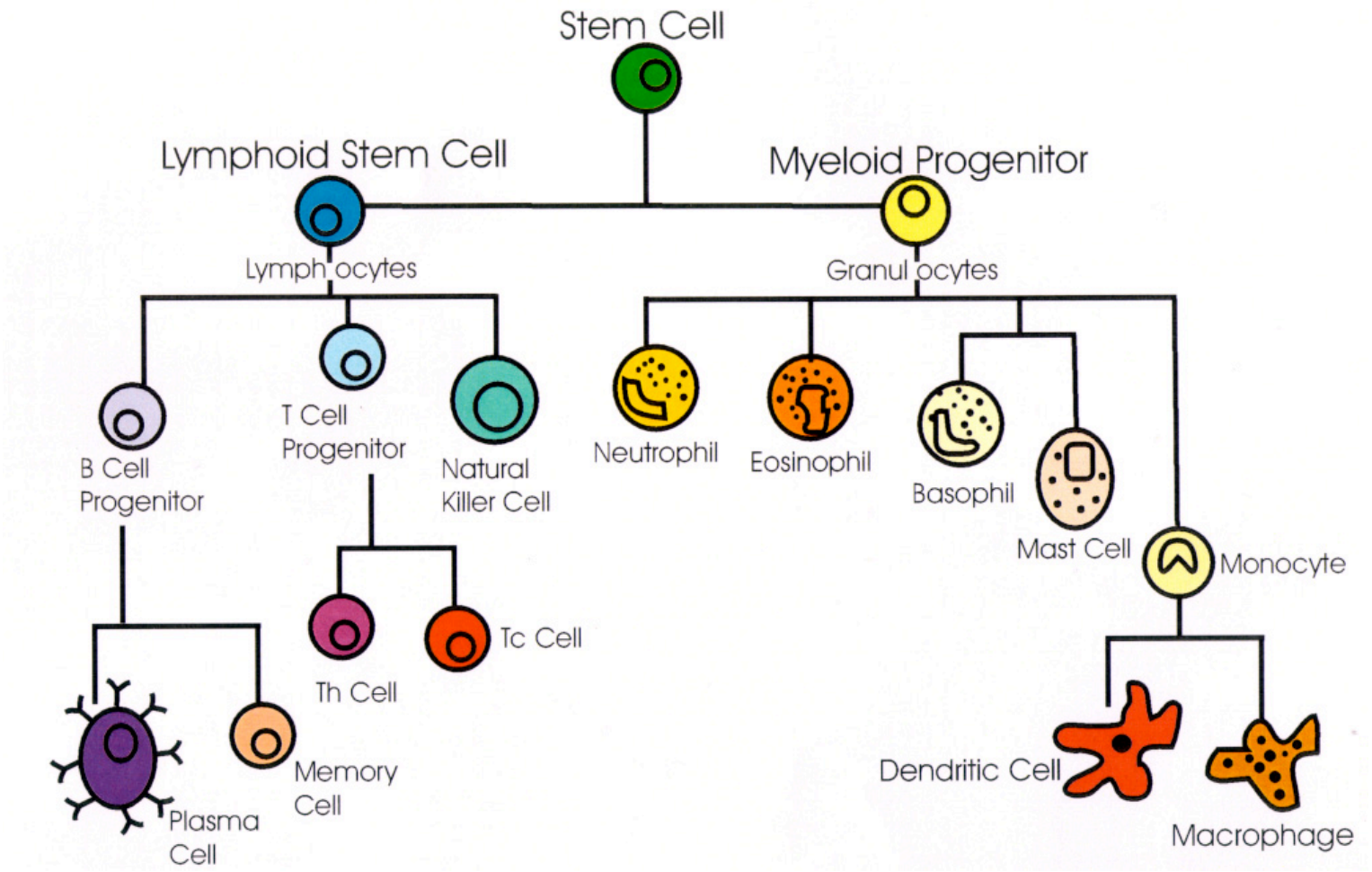
How to approach the workup?

- Initial steps
  - Clues from the family and personal history, physical examination
  - Verify the x-ray and/ or culture/pathology reports
- What is common and what is rare?

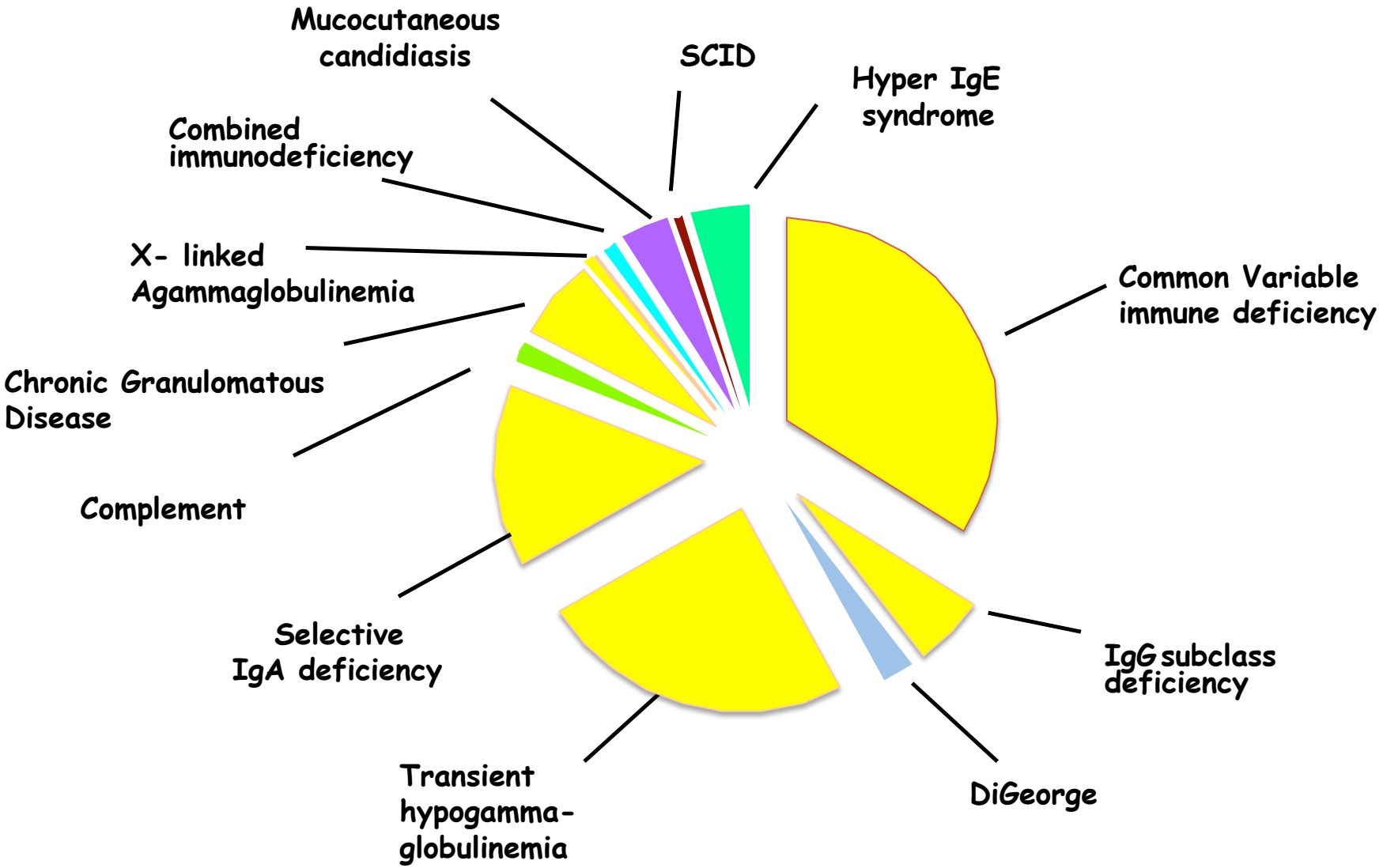
Immunodeficiency Diagnoses:
(450 patients)



Consider what cells of the immune system you will focus upon



Immunodeficiency Diagnoses:
Antibody defects are most common



Primary Immune Deficiencies:
time of diagnosis

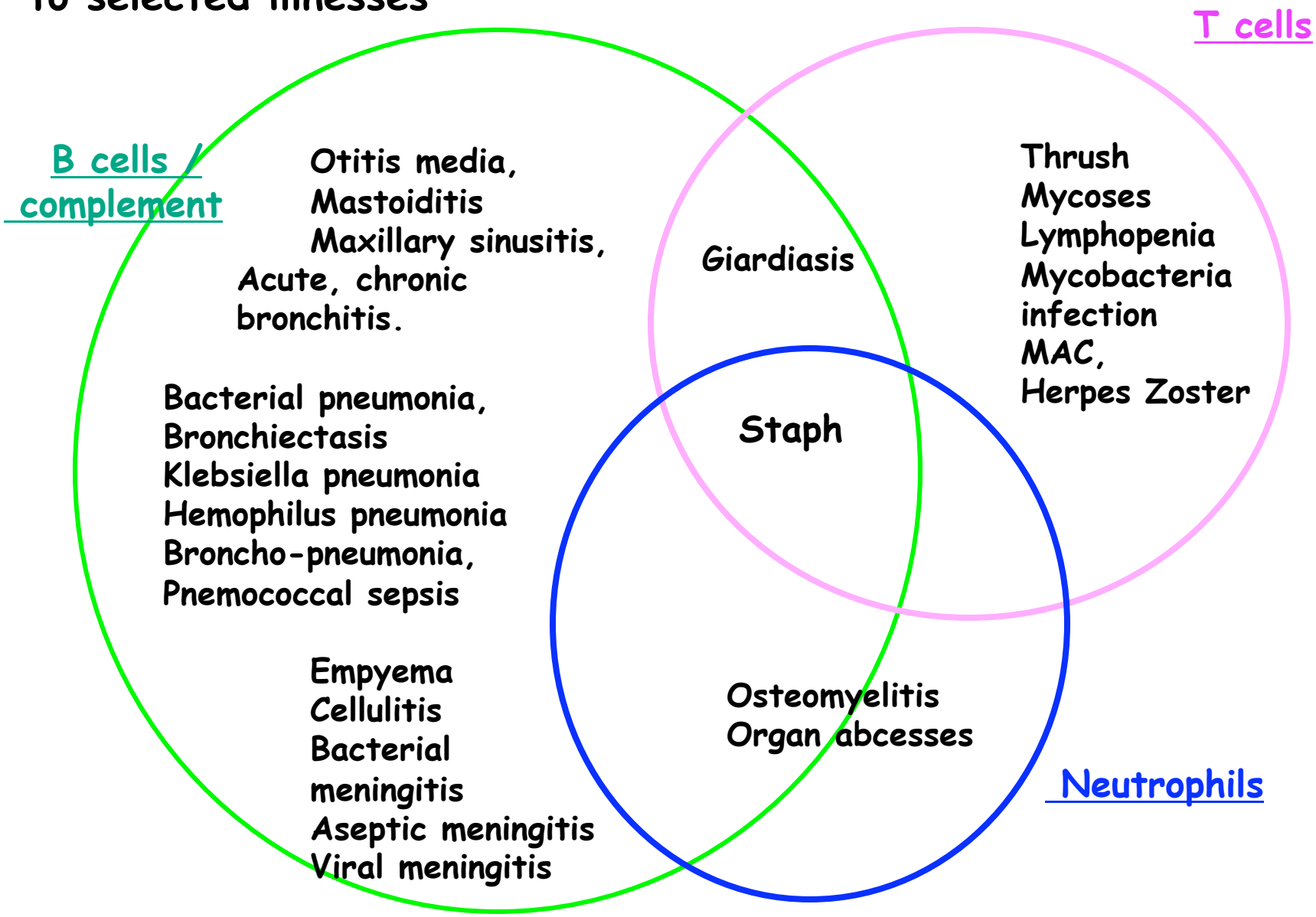
Infants/children

- SCID syndromes
- X Linked
  - Agammaglobulinemia
- Hyper IgM syndromes
- Wiskott Aldrich
- Hyper IgE
- DiGeorge Syndrome
- CGD

Adolescents/adults

- Common variable immunodeficiency
- IgG subclass deficiency
- IgA deficiency
- Complement defects
- Thymoma/agammaglobulinemia
- Mucocutaneous candidiasis
- Neutropenia
- CGD

Defects of various immune functions lead to selected illnesses



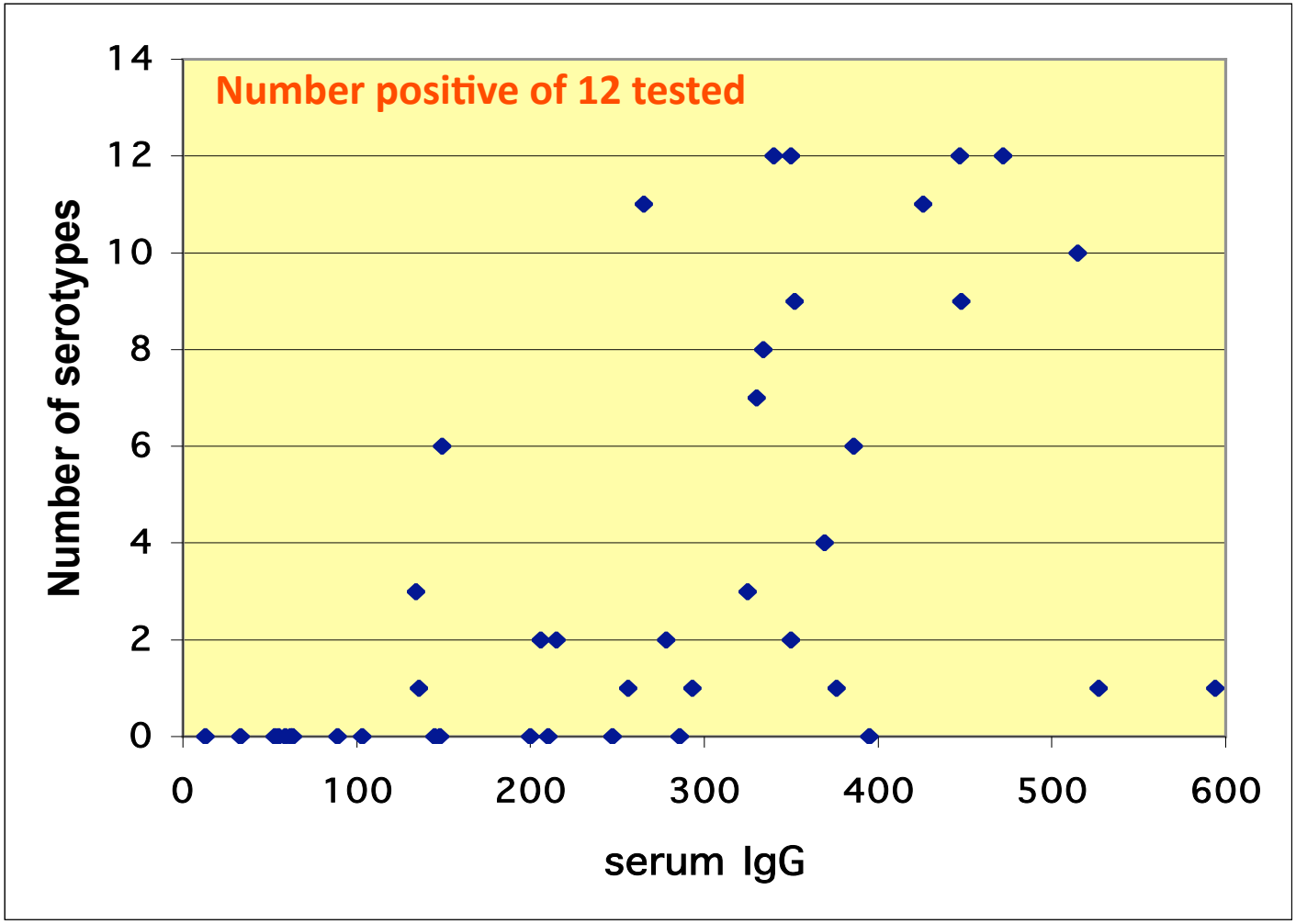
Laboratory Evaluation of Immunodeficiency Initial Workup

- Get a complete history
- Get old records
- Do physical exam. Get height and weight.
- Get blood, culture and X ray results from the past.
- CBC and differential
- Serum Immunoglobulins, IgG, IgA and IgA.
- B cell function: antibody production to several antigens
- T, T cell subsets, B cell and NK cell numbers

IgG	IgA	IgM	anti tet	diptheria	measles	mumps	rubella
33	6.9	15	neg	wk Pos	neg	neg	neg
57	8	18	neg	neg	neg	neg	pos
59	12	53	neg	neg	neg	neg	Pos
89	5	7	neg	neg	neg	neg	neg
103	<15	<5	wkPos	wkPos			
136	<15	5	neg	Pos	neg	Pos	neg
149	20.9	6.7	Pos	Pos	neg	Pos	Pos
179	<15	<5	pos	pos	pos	pos	pos
193	<15	47	pos	pos	neg	neg	neg
200	5.9	24	neg	neg	neg	neg	neg
206	176	47	pos	pos	neg	neg	neg
206	2	18.5	neg	neg	neg	neg	neg
210	6.9	15	Pos	Pos	Pos	Pos	neg
215	15	37	Pos	Pos	neg	neg	Pos
247	17	30	Pos	Pos	Pos	Pos	Pos
265	10	185	Pos	Pos	Pos	Pos	Pos
286	4	34	Pos	neg	neg	neg	neg
293	17	25	Pos	Pos	Pos	Pos	Pos
325	10	30	neg	Pos	Pos	neg	Pos
325	6.9	135	Pos	neg	neg	Pos	Pos
330	10	130	neg	wk Pos	neg	neg	neg
330	44	22	Pos	Pos	Pos	Pos	Pos
334	<15	<5	Pos	Pos	Pos	Pos	Pos
340	13.9	12.8	Pos	Pos	Pos	neg	Pos
350	10	20	neg	Pos	Pos	Pos	Pos
352	27	24	Pos	Pos	neg	Pos	neg
360	47	43	Pos	Pos	Pos	neg	neg
376	7	18	Pos	Pos	Pos	neg	Pos
386	28	47	neg	Pos			
395	6.9	120	neg	neg	neg	neg	Pos
404	19	76	wkPos	neg	neg	neg	Pos
400	69	10	Pos	Pos	neg	Pos	Pos
426	47	32	Pos	Pos	Pos	Pos	neg
454	51	12	Pos	Pos	neg	Pos	Pos
472	18	131	wkPos	wkPos	neg	neg	neg
477	<6	16	Pos	Pos	Pos	Pos	Pos
447	43	21	Pos	Pos	Pos	Pos	Pos
448	7	28	Pos	Pos	Pos	neg	Pos
468	26	24	Pos	Pos	neg	neg	neg
515	43	363	Pos	Pos	Pos	Pos	Pos
527	2	29	Pos	neg	Pos	Pos	Pos
530	42	99	Pos	Pos	Pos	Pos	Pos
574	5.5	48.4	Pos	Pos	Pos	neg	neg
585	22	240	Pos	Pos	Pos	Pos	Pos
594	<7	34	wkPos	wkPos	Pos	Pos	neg

Using IgG alone does not always document antibody deficiency

Post pneumococcal vaccine responses for 40 patients with CVID



Again, serum IgG level is not the end of the story

For Ig replacement: the most important thing

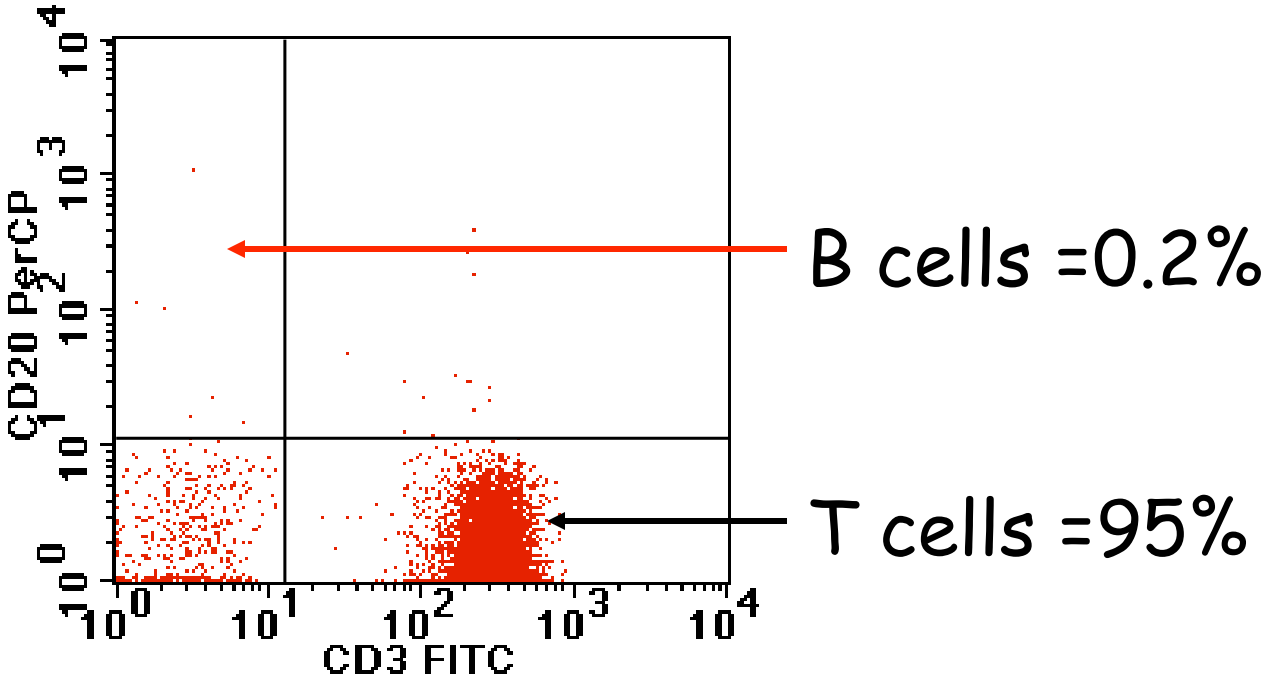
For subjects with lower than normal serum IgG

- Do a complete workup.
- Don't start Ig until this is done.
- Titers to a number of protein vaccines, natural exposures; antibodies to carbohydrate antigens.
- Tetanus, diphtheria, herpes zoster, measles, mumps, rubella, pneumococcal and hemophilus vaccines, isohemagglutinins, etc

Why do the full lymphocyte screen?

Flow Cytometric Evaluation:

No B Cells:



Low or absent B cells (no CD20+ cells) = XLA

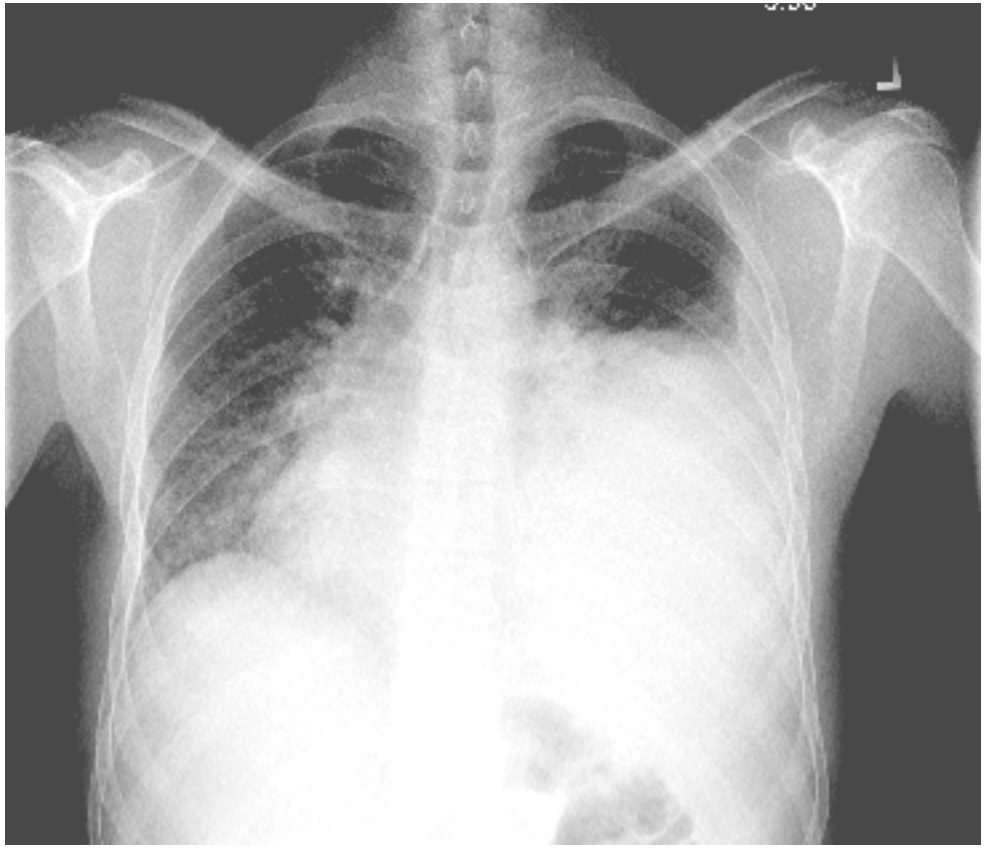
Or what?

51 year old architect

1. Long history of sinusitis but no hospitalizations or serious infections
2. Felt poorly and fainted
3. Went to his internist who noted that the chest was dull on the left
4. Referred for chest Xray
5. A mass suspicious for a thymoma
6. Removed at MSSM
7. 8 months later, he went to an allergist about the sinusitis.

8. Allergist tested serum immune globulins:
9. IgG= 30 IgA =0 IgM =5
10. Diagnosis?

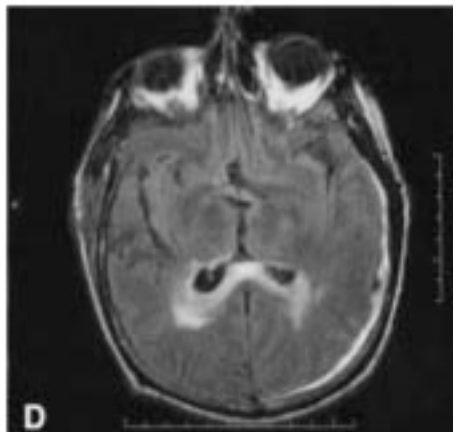
1 kg thymoma



51 cases of Good Syndrome

All had recurrent sinopulmonary infections

Haemophilus influenzae (11)



CMV disease (8)

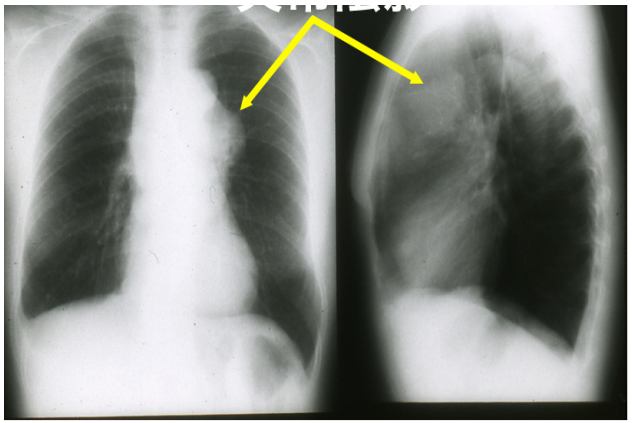
Bacteremia (7)

Oral, esophageal, mucocutaneous candidiasis (11)

Chronic diarrhea (5) with documented pathogens

Urinary tract infections (4)

P. carinii pneumonia (4)



Tuberculosis (2)

Kaposi sarcoma (1)

Disseminated varicella (1)

Candidemia (1)

Clostridium perfringens wound infection (1)

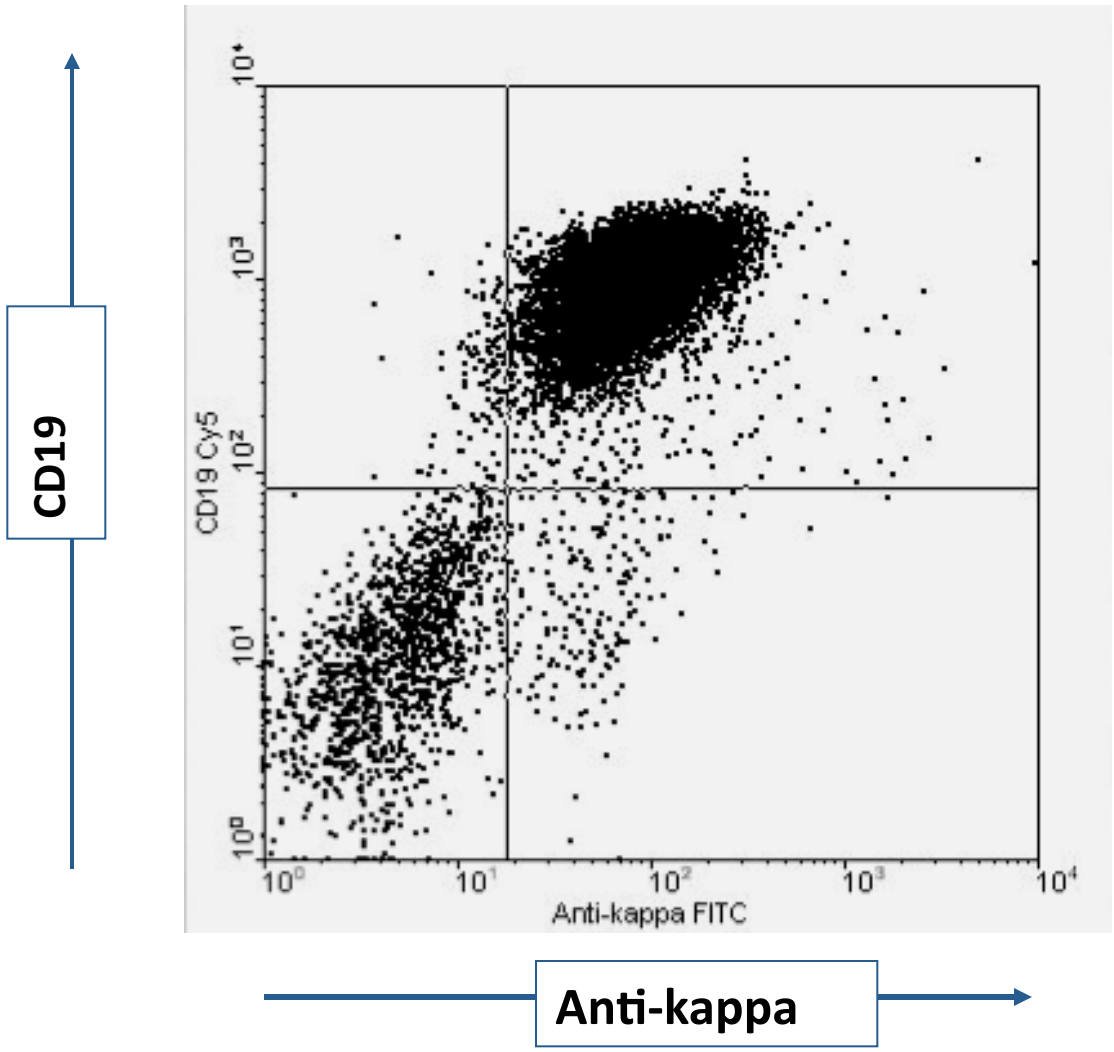
Mycoplasma arthritis (1)

Tarr et al, Medicine, 2001

Referral for Mild Hypogammaglobulinemia

- 43 year old woman with URIs and recurrent sinus infections, referred for this and IgG1 subclass deficiency. (IgG1= 250)
- IgG = 601, IgA= 55, IgM= 43
- CBC 51% lymphocytes; 5,000
- T=48%
- B%=40% (normal 5-15); 1731 (75-375)

Too many B cells: B cell clonal issue, CLL: test for light chains kappa/lambda



Unusual lab data

- 51 year old man referred for unusual IgG subclass tests, done for recurrent sinusitis.
- IgG1= 301 mg/dl; IgG2=1,241, IgG3= 13; IgG4=1.

New referral

- 28 year old man from NC moved to NYC for business; on IVIG for CVID for 3 years.
- Had diagnosis of sinusitis prior to Ig
- He brought original lab data: IgG=67, IgA= 1,261, IgM=180 mg/dl.

New referral

- 28 year old man from NC moved to NYC for business; on IVIG for CVID for 3 years.
- Had diagnosis of sinusitis prior to Ig
- He brought original lab data: IgG=67, IgA= 1,261, IgM=180 mg/dl.

- Repeat: IgG= 1400, IgA= 59, IgM=192

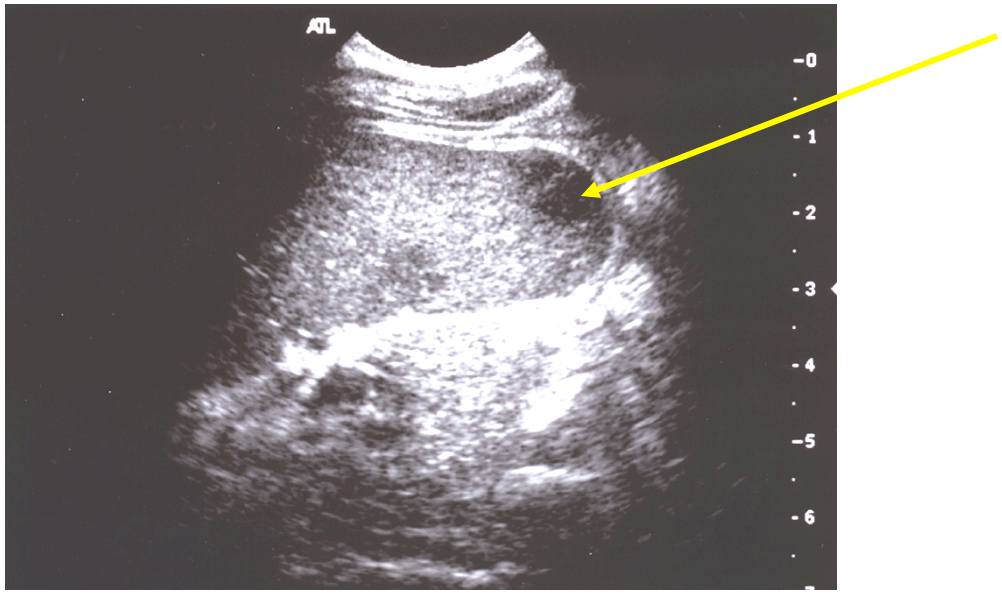
What about other immune defects?

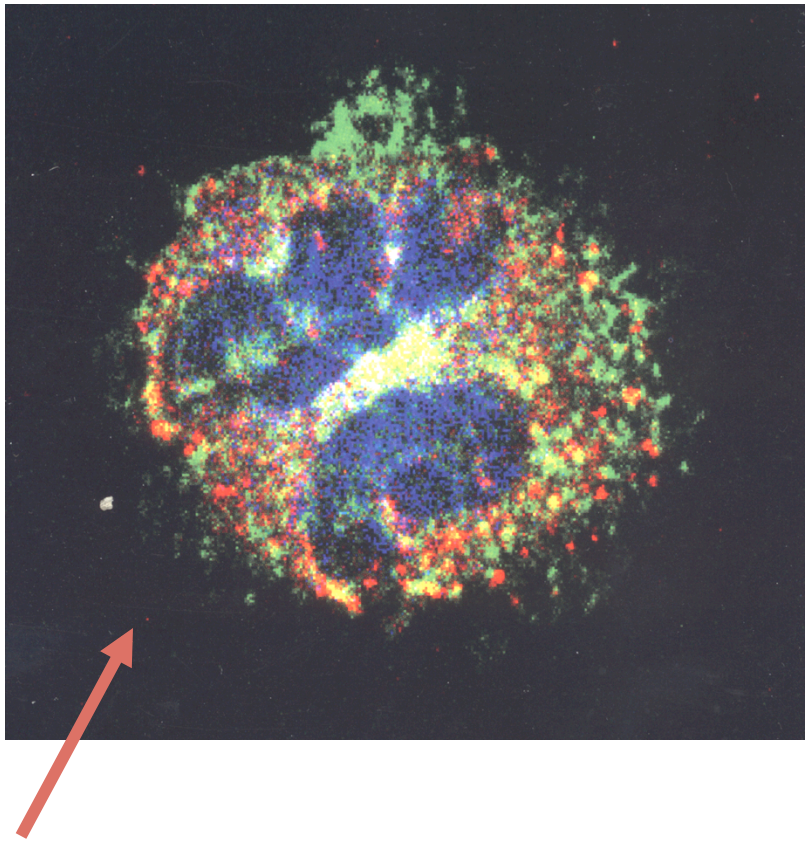
Laboratory Evaluation of Immunodeficiency Intermediate Work-up

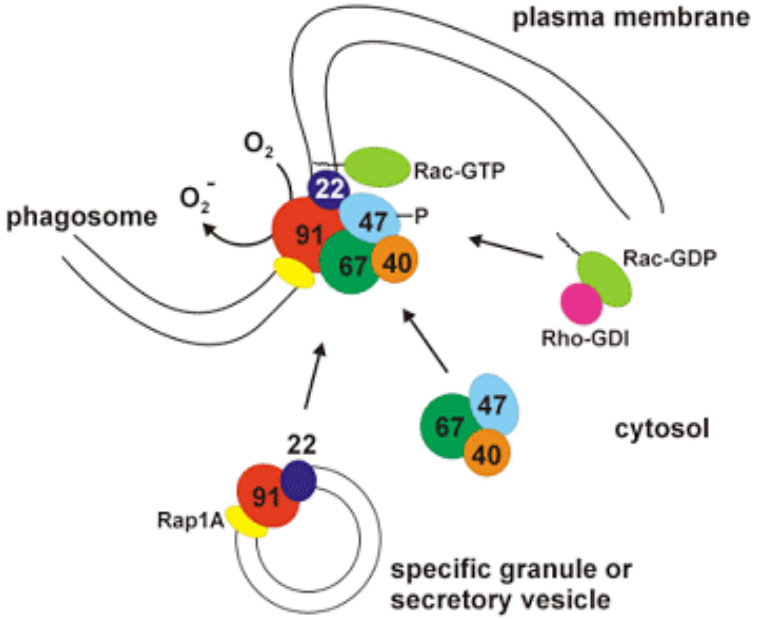
- T cell proliferation using non specific stimulators: mitogens phytohemagglutinin, Concanavalin A, pokeweed mitogen
- T cell proliferation using antigens such as tetanus, candida.
- More B cell functions: antibody production, before and after vaccine challenge. (tetanus, diphtheria, Hemophilus, pneumococcus, etc. Isohemagglutinins
- Neutrophil function.
- Complement pathway functions (CH50 and AH50)
- Fluorescent *in situ* hybridization for DiGeorge syndrome
- Flow cytometer studies for specific markers, hyper IgM syndrome, CD18 etc

SO: male with increased abdominal girth and fever

- 18 month old male with rectal abscesses at 2 months
- Admitted to hospital, increased abdo size + fever
- Sonogram - fluid in the abdomen and lesions in spleen



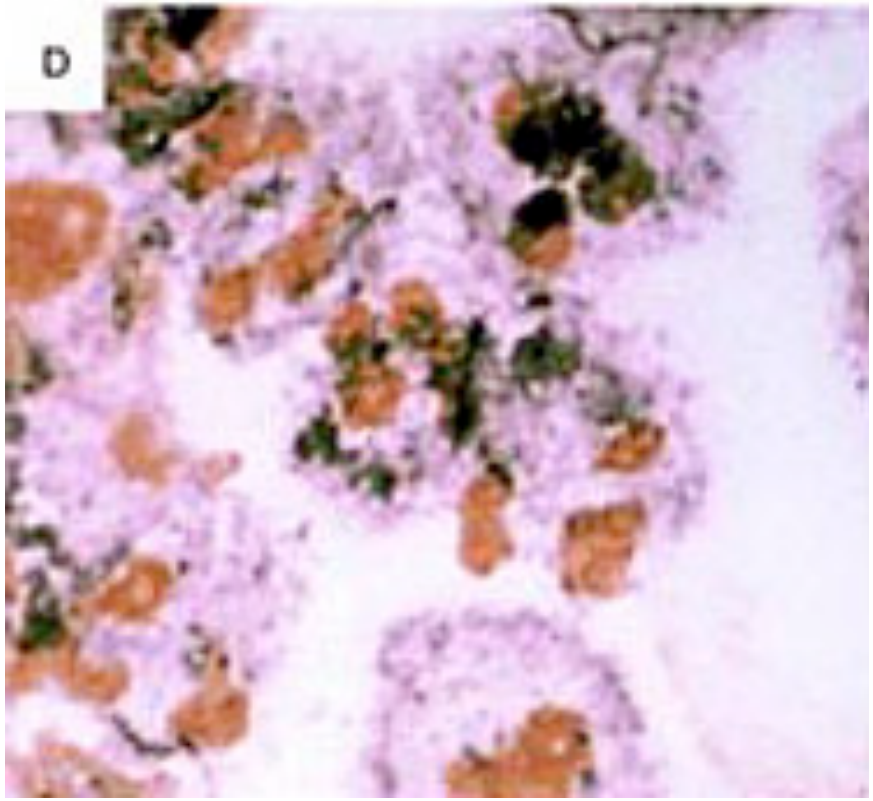


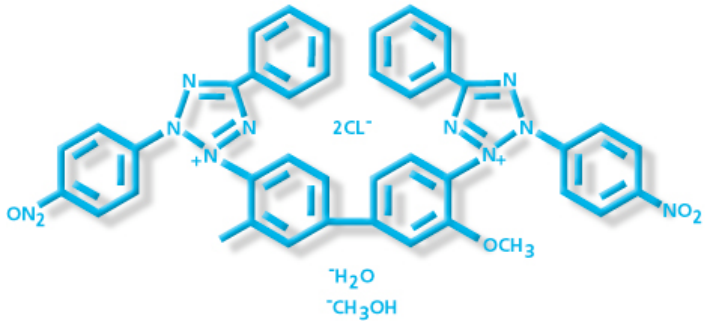


Confocal image of human neutrophil undergoing respiratory burst.

The bilobed nucleus (blue) is surrounded by Rac GTPase (red), and p22 ^{phox} (green.) The NADPH oxidase complex is assembled and activated Intra-cellularly, shown as regions of yellow where Rac and p22 ^{phox} colocalize.

Reduction of nitro blue tetrazolium dye by neutrophils

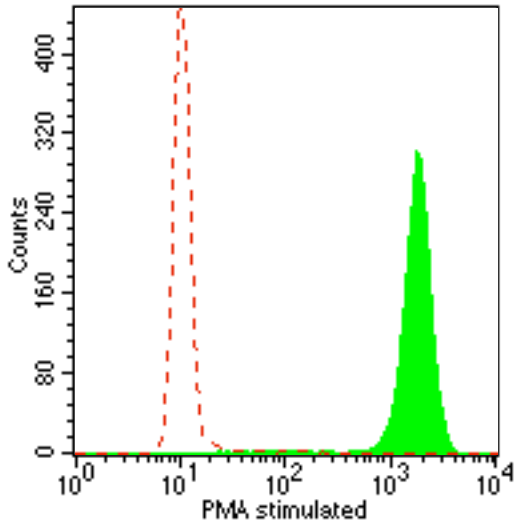


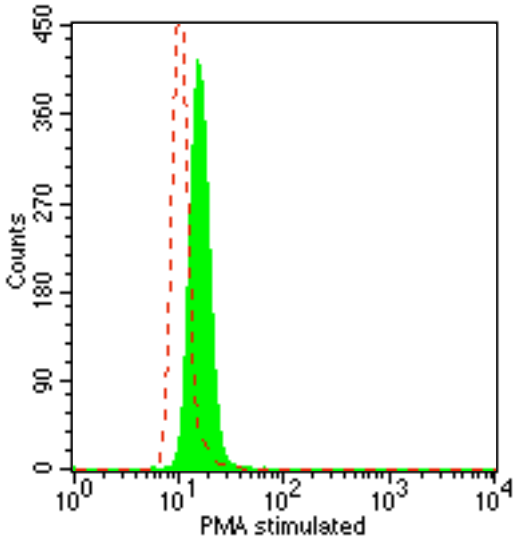


Yellow → blue

DHR Assay to Diagnose CGD

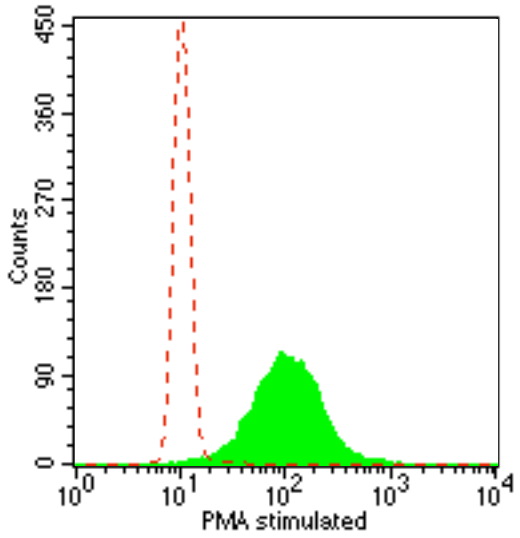
Normal

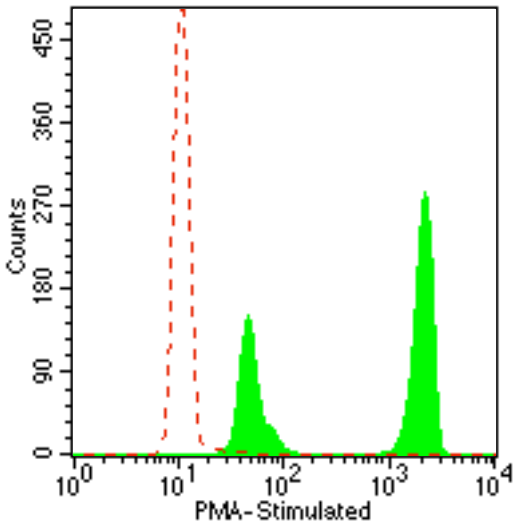




Phox^{91} deficient CGD (X-linked)

Phox^{47} deficient CGD (AR)

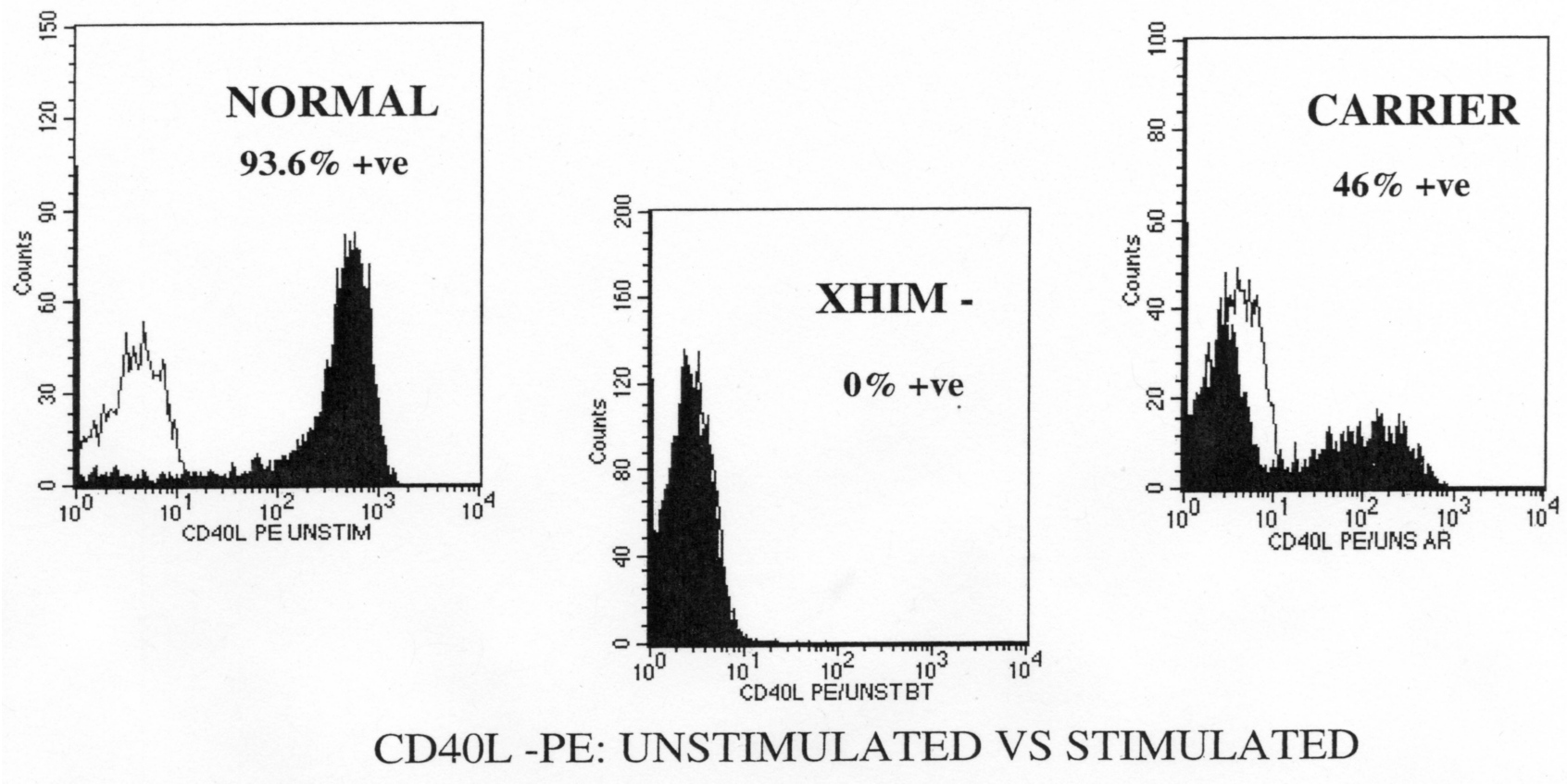




X-linked CGD carrier

Slide from T Fleisher NIH

CD40L (CD154) Expression in HIGM1



Evaluation based on PMA/ionomycin activated CD4 T cells

One more case

Recurrent pneumonia

- 53 year old woman with X ray documented pneumonias in 2005, 2007, 2009 and 2010. RUL, RML
- Not hospitalized. Resolves with levaquin.
- Previous history of sinusitis; ENT said to be nl.
- CT showed minimal RML bronchiectasis
- PFTs: 100% function
- Infectious disease work up negative.
- Past medical history: migratory arthritis hypertension, parathyroidectomy for hyperparathyroidism and partial thyroidectomy in 2005.

Case: recurrent pneumonia

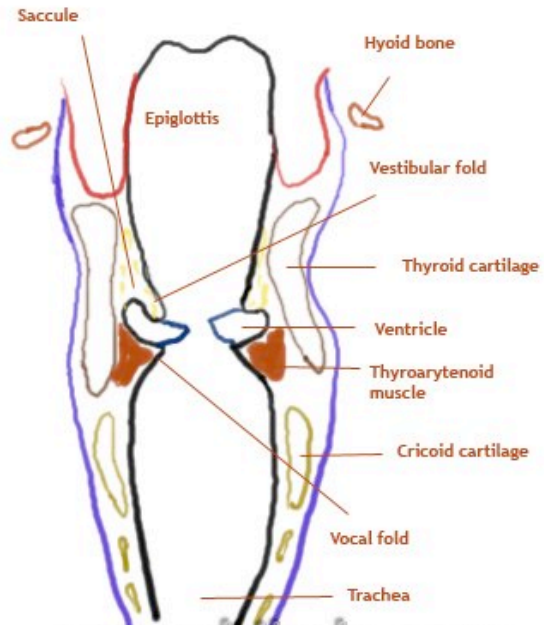
- CBC and general lab tests nl.
- IgG 1000; IgA 165; IgM 284
- IgG subclasses 1-4 nl
- Antibody titers to pneumococci + 10/14 serotypes.
- Antibodies to tetanus, diptheria, MMR and varicella all +.

ENT specialist

Flexible fiberoptic endoscopic evaluation with sensory testing:

- Severe post glottic edema
- Aryepiglottic folds, arytenoids: severe edema
- Ventricle: obliteration R ar
- Sensory: severe deficits

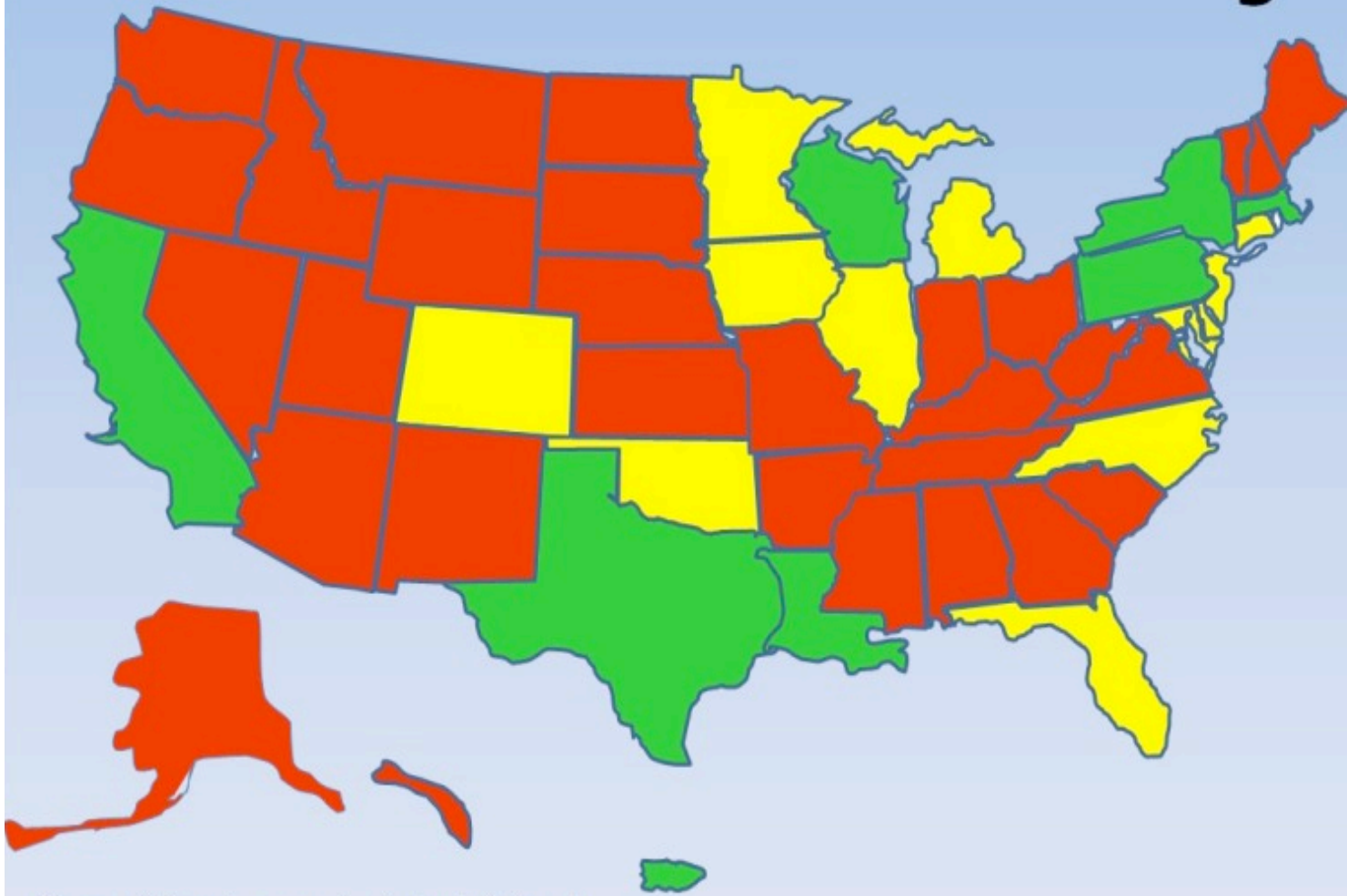
due to chronic reflux



New methods for screening

- New born Screening for SCID and severe T cell defects ongoing
- Targeting likely genes
- Whole genome studies

SCID Newborn Screening



Green - States where screening is already taking place

Yellow - States that have voted to begin screening through legislation or their Newborn Screening Advisory Committees

Red - States that we are still working on

IMMUNE DEFICIENCY FOUNDATION

Conclusions