ACTIVE vs PASSIVE IMMUNIZATION:
ADVANTAGES AND DISADVANTAGES OF EACH APPROACH

Arturo Casadevall
Albert Einstein College of Medicine
Bronx, New York
DEFINITIONS

• ACTIVE IMMUNIZATION = VACCINES

• PASSIVE IMMUNIZATION = PREFORMED ANTIBODY ADMINISTRATION
ACTIVE IMMUNIZATION - VACCINES

ADVANTAGES
• LONG PROTECTION
• LESS COSTLY (TO ADMINISTER)

DISADVANTAGES
• DEPENDENT ON HOST IMMUNE SYSTEM
  – NOT EVERYONE MAY BE PROTECTED
• PROTECTIVE RESPONSE TAKES TIME
• MAY BE IMPRACTICAL FOR NOSOCOMIAL INFECTION
• LONG DEVELOPMENT TIME
• ? EFFECTS ON MICROFLORA
# PASSIVE IMMUNIZATION

## ADVANTAGES
- Immediate Immunity
- Pharmacologic Control
- Rapid Development Time

## DISADVANTAGES
- Costly
- Temporary Immunity
- Requires IV Administration
- Dose-Response Uncertainty
REAGENTS FOR PASSIVE THERAPY: mAbs

ADVANTAGES
- HIGH SPECIFIC ACTIVITY
- PRECISE PHARMACOLOGIC DEFINITION
- SINGLE ISOTYPE
- LOT TO LOT CONSTANCY

DISADVANTAGES
- COST
- SINGLE SPECIFICITY
  - SELECTION FOR ESCAPE MUTANTS
  - EFFECTIVENESS $\alpha$ 1/ANTIGENIC DIVERSITY
- SINGLE ISOTYPE
  - LIMITED EFFECTOR FUNCTION
  - LIMITED FcR ENGAGEMENT
# Reagents for Passive Therapy: Polyclonal Sera

## Advantages
- Multiple specificities
- Multiple isotypes

## Disadvantages
- Low specific activity
- Lot to lot variation
- Requirement for immunized donors
- Cost
- Concern about inadvertent transmission of disease
NEW INSIGHTS INTO ANTIBODY-MEDIATED PROTECTION FROM STUDIES WITH A FUNGAL PATHOGEN

Or 20 years of work in 18 minutes
C. NEOFORMANS – ONLY ENCAPSULATED EUKARYOTIC PATHOGEN OF HUMANS
CAPSULE IS THE MAJOR VIRULENCE FACTOR

POLYSACCHARIDE CAPSULE

Mannose  Xylose  Glucuronic acid
GLUCURONOXYLOMANNAN
MAJOR VIRULENCE FACTOR
WEILER HOSPITAL, NEW YORK CITY SUMMER 1988

30-SOMETHING CAMBODIAN MAN WITH *C. NEOFORMANS* MENINGITIS DIES AGONIZING DEATH DESPITE RECEIVING LARGE DOSES OF AMPHOTERICIN B DURING A PERIOD OF 3 MONTHS...

COULD ANTIBODY THERAPY BE DEVELOPED?

CIRCA 1990

- WORK ON RELEVANT IMMUNOLOGY
  - CELL MEDIATED IMMUNITY
- IF YOU WANT A SCIENTIFIC CAREER DON'T WORK ON USEFUL THINGS
- GET A DIFFERENT PROJECT – YOU ARE GOING TO DISCOVER WHAT WE KNOW ABOUT PNEUMOCOCCUS
THE GREAT IMMUNOLOGICAL CATASTROPHE

1891-1910

HUMORAL VS PHAGOCYTIC THEORIES

HUMORAL IMMUNITY

1910-1940

ANTIBODY PROTECTS AGAINST ALL MICROBES

CD8+ Cells Enhance Resistance to Pulmonary Serotype 3 Streptococcus pneumoniae Infection in Mice

Sarah E. Weber, Haijun Tian, and Liise-anne Pirofski

1940-1960s

HUMORAL IMMUNITY

VIRUSES

TOXINS

ENCAPSULATED BACTERIA

HUMORAL IMMUNITY

1970-1990s

CELLULAR IMMUNITY

MYCOBACTERIA

FUNGI

INTRACELLULAR PATHOGENS

FOUR ‘HUMORS’

‘HUMORAL IMMUNITY’

GALEN

The New England Journal of Medicine

The Efficacy of a Salmonella Typhi Vi Conjugate Vaccine in Two-to-Five-Year-Old Children

FENG YING C. LIN, M.D., M.P.H., VO ANH HO, M.D., HA BA KHEM, M.D., DANG Duc TRACH, M.D., PH. D., PHAN VAN BAY, M.D., TRAN CONG THANH, M.D., ZUZANA KOSSACZKA, PH. D., DOLORES A. BREYLA, M.P.H., JOSEPH SHLOACH, PH. D., JOHN B. ROBBINS, M.D., RACHEL SCHNEERSON, M.D., AND SHOUXUN C. SU, PH. D.
METHODS FOR ESTABLISHING USEFULNESS OF ANTIBODY-MEDIATED PROTECTION

1. PASSIVE TRANSFER EXPERIMENTS (1891)

IMMUNE SERA + SUSCEPTIBLE HOST INFECTIOUS CHALLENGE PROTECTION?

2. CORRELATE ANTIBody TITER WITH RESISTANCE (1900s)

SUSCEPTIBILITY $\propto 1/titer$

3. ASSOCIATE SUSCEPTIBILITY WITH ANTIBODY DEFICIT (1940s)

PROBLEM: THESE METHODS SUGGEST SPECIFIC ANTIBODY IS NOT IMPORTANT IN DEFENSE AGAINST MANY PATHOGENS e.g. Mycobacterium tuberculosis Cryptococcus neoformans Listeria monocytogenes Many others….

LOGICAL ERROR: JUST BECAUSE THE ASSAY SUGGESTED THAT ANTIBODY WAS NOT PROTECTIVE DID NOT MEAN THAT ANTIBODY COULD NOT BE PROTECTIVE

ERROR INVOLVED MAKING A POSITIVE INERENCE FROM NEGATIVE DATA
PROBLEM:
EXPERIMENTS WITH POLYCLONAL SERA ARE INCONCLUSIVE

A DIFFERENT APPROACH

PROTECTION STUDIES

NON-PROTECTIVE mAb
YYYYYY

PROTECTIVE mAb
YYYYYY

NON-PROTECTIVE mAbs INHIBITED EFFICACY OF PROTECTIVE mAbs
ANTIBODY-MEDIATED PROTECTION AGAINST *M. TUBERCULOSIS*

A Novel Human IgA Monoclonal Antibody Protects against Tuberculosis

Sucharitha Balu,* Rajko Reljic,† Melanie J. Lewis,‡ Richard J. Pleass,*Richard McIntosh,¶ Cees van Kooten,‖ Marjolein van Egmond,♯ Stephen Challacombe,* Jenny M. Woof,‡ and Juraj Ivanyi*

CONFIRMATION (5 independent labs):

- PETHE ET AL. NATURE 2001:412;190
- HAMASUR ET AL. VACCINE 2003:20:4081
- WILLIAMS ET AL. IMMUNOLOGY 2004:111;328
- CHAMBERS ET AL. FEMS IMMUNOL.MED.MICROBIOL. 2004:41;93
- HAMESURE ET AL. CLIN.EXP.IMMUNOL 2005: 138:30-8

PROC.NATL.ACAD.SCI 1998:95;15688
THE CMI vs. HUMORAL PARADIGM UNTENABLE

HUMORAL IMMUNITY
VIRUSES
TOXINS
ENCAPSULATED BACTERIA

CELLULAR IMMUNITY
MYCOBACTERIA
FUNGI
SOME BACTERIA
VIRUSES
TOXINS (SUPERANTIGENS)

NEW APPROACH
USE MAbs

PROTECTIVE MAbs MADE TO:
CRYPTOCOCCUS NEOFORMANS (FUNGUS)
CANDIDA ALBICANS (FUNGUS)
LISTERIA MONOCYTOGENES (INTRACELLULAR BACTERIA)
HISTOPLASMA CAPSULATUM (INTRACELLULAR FUNGUS)
MYCOBACTERIUM TUBERCULOSIS (INTRACELLULAR BACTERIA)
EHRLICHIA CHAFFENSIS (INTRACELLULAR BACTERIA)
LEISHMANIA MEXICANA (INTRACELLULAR PARASITE)

TWO EMERGING EXPLANATIONS
‘GOOD & BAD’
C. neoformans
C. albicans
M. tuberculosis

INADEQUATE AMOUNTS
L. monocytogenes
H. capsulatum
WHY IS IT SO HARD TO PROTECT AGAINST SOME MICROBES WITH ANTIBODY?

THE THINGS THAT MATTER....

- SPECIFICITY
- ISOTYPE
- AMOUNT
- DEPENDENCE ON HOST

CELL-MEDIATED IMMUNITY

GENETICS

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<th>IgG</th>
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<tr>
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<tr>
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<td>INHIBITS PS RELEASE</td>
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<tr>
<td>ISOTYPE</td>
<td>NA</td>
<td>IgG1, IgG2a, IgG2b</td>
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<tr>
<td>NEED C3</td>
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TWO STORIES:

IgM – EFFICACY REQUIRES ‘RIGHT’ SPECIFICITY AND COMPLEMENT

IgG - EFFICACY REQUIRES ‘RIGHT’ ISOTYPE AND HOST
WHAT IS THE DIFFERENCE BETWEEN PROTECTIVE AND NON-PROTECTIVE ANTIBODIES?

PROTECTIVE

- mAb 12A1
  - IgM

NON-PROTECTIVE

- mAb 13F1
  - IgM

Clinical Trial

- 12A1 IgM
- 15E8 IgM
- 12F4 IgG1
- 14E1 IgG1
- 13F1 IgM
- 18B7 IgG1
- 25G12 IgA

- VH7183 Jh2 (7 aa D)
- Vk5.1 Jk1

POSITIONS 33 AND 57 IN PROTECTIVE AND NON-PROTECTIVE CLASS II MAbs TO C. NEOFORMANS POLYSACCHARIDE

ANTIBODY BINDING SITE FROM X-RAY CRYSTAL STRUCTURE

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<tr>
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<th>33</th>
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<td>3B10</td>
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<td>F</td>
<td>R</td>
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<tr>
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<td>F</td>
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<td>K</td>
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<td>L</td>
<td>K</td>
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<tr>
<td>13F1</td>
<td>Y</td>
<td>S</td>
<td>NO</td>
<td>PUNCTATE</td>
</tr>
<tr>
<td>21D2</td>
<td>Y</td>
<td>S</td>
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TYROSINE

PHENYLALANINE

COMPLEMENT-INDEPENDENT IgM-MEDIATED PHAGOCYTOSIS OF C. NEOFORMANS

mAb 12A1

mAb 13F1

CD18 BINDING SITE

CR3

MACROPHAGE

Taborda et al. Immunity 2002
PROTECTIVE AND NON-PROTECTIVE IgM mAbs MEDIATE C3 LOCALIZATION IN DIFFERENT PARTS OF CAPSULE

LIGHT

IgM

C3

MERGE

FUNGAL CELLS IN VIVO COATED WITH IgM IN ‘PUNCTATE PATTERN

COMPLEMENT IS DEPOSITED IN VIVO AWAY FROM SURFACE

Zaragoza et al Infect Immun 2004
C. neoformans BIOFILM FORMATION

Adhesion Microcolony Maturation
PROTECTIVE ANTIBODIES BLOCK BIOFILM FORMATION

No Ab

MAb 18B7

MAb 3671 (control)

Percentage of C. neoformans biofilms metabolic activity

B3501

H99

OD 492 nm

Percentage of C. neoformans biofilms metabolic activity

No Ab

50 µg/ml 18B7

100 µg/ml 18B7

50 µg/ml 3671

100 µg/ml 3671

p<0.05

PROTECTIVE

NON-PROTECTIVE

IRRELEVANT
Exopolymeric matrix

No Ab

Biofilm

Ab cross-links capsule

Interference with biofilm formation

HOW DOES ANTIBODY INTERFERE WITH FUNGAL PHYSIOLOGY?
IgG AND C3 OPSONIZATION HAVE DIFFERENT OUTCOMES IN EXIT DISPERSION
ANTIBODY BINDING CAN ALTER MICROBIAL GENE EXPRESSION

McClelland, Nicola, Prados, Casadevall

J Clin Invest 2010

JANOFF & FRANK, JCI 2010 (IN PRESS) COMMENTARY
Antibodies to *Streptococcus pneumoniae* Capsular Polysaccharide Enhance Pneumococcal Quorum Sensing

Masahide Yano, Shruti Gohil, J. Robert Coleman, Catherine Manix, and Liise-anne Pirofski

Department of Microbiology and Immunology, Albert Einstein College of Medicine, Bronx, New York, USA; and Division of Infectious Diseases, Department of Medicine, Albert Einstein College of Medicine, Montefiore Medical Center, Bronx, New York, USA.

Mid-log phase H37Rv defined MM culture

10 μg/ml Mab

45 min

Expression ratio: Capsular Mab/isotype IgG
MORE IS NOT BETTER: PROZONE-LIKE EFFECTS

PNEUMOCOCCUS 1935

A SINGLE mAb CAN BE PROTECTIVE, NON-PROTECTIVE OR DISEASE ENHANCING DEPENDING ON ITS CONCENTRATION AND INOCULUM

C. NEOFORMANS 2001
LARGE AMOUNTS OF ANTIBODY BOUND TO CAPSULE PROTECT AGAINST NITRIC OXIDE FUNGICIDAL ACTIVITY

EXCESS ANTIBODY

MECHANISM 1: REDUCED KILLING BY MICROBICIDAL OXIDANTS IN VITRO

MECHANISM 2: DIFFERENCES IN CYTOKINE EXPRESSION IN HIGH AND LOW Ab CONDITIONS

Ab PROTECTION
HIGH IL-4, IL-6, IL-12, TNF-a

NO PROTECTION
LOW IL-6 AND IFN-g;
HIGH IL-10 AND IL-12

MECHANISM 3: DIFFERENCES IN C3 DEPOSITION IN HIGH AND LOW Ab CONDITIONS

TABORDA & CASADEVALL, J. IMMUNOL. 2001
TABORDA ET AL. J. IMMUNOL. 2003
HOW DO ANTIBOIES WORK?
ANTIBODY-MEDIATED PROTECTION ASSOCIATED WITH CHANGES IN INFLAMMATORY RESPONSE

- REDUCED IFN-ГAMMA
- INCREASED IL-10
- REDUCED IL-4
- INCREASED B7-2
- INCREASED % GRANULOCYTES

NET RESULT: DAMAGE
CRYPTOCOCCOSIS IN B CELL DEFICIENT AND FcRIII-/- MICE

B CELL DEFICIENT MICE

ACTION | EFFECT | CONSEQUENCE | OUTCOME
---|---|---|---
PASSIVE Ab | ADD Ab | LESS IFN-g | PROTECTION
B CELL KO | NO Ab | MORE IFN-g | NO PROTECTION
FcRIII-/- | NO FcRIII | MORE IFN-g | NO PROTECTION

SEVERAL NEW MECHANISMS FOR ANTIBODY-MEDIATED PROTECTION

• MEDIATING PHAGOCYTOSIS THROUGH A NON-Fc AND C3 INDEPENDENT MECHANISM
• INHIBITING BIOFILM FORMATION
• MODULATING INFLAMMATION TO IMPROVE GRANULOMA FORMATION
• DIRECT PHYSIOLOGICAL EFFECTS ON MICRORGANISMS
PROTECTIVE ANTIBODIES

DIRECT EFFECTORS

INDEPENDENT OF CMI

TOXIN NEUTRALIZATION
PHAGOCYTOSIS & KILLING
COMPLEMENT ACTIVATION
VIRAL NEUTRALIZATION
ADCC

DEPENDENT ON CMI

CYTOKINE CHANGES
CELL ACTIVATION
ANTIGEN PRESENTATION
CLEARANCE OF ANTIGENS

INDIRECT EFFECTORS

REDUCE MICROBIAL INNOCULUM

REDUCE HOST DAMAGE

S. PNEUMONIAE
C. DIPTHERIA
H. INFLUENZAE
WEST NILE VIRUS
B. ANTHRACIS
VARIOLA VIRUS

‘EASY’

C. NEOFORMANS
M. TUBERCULOSIS
H. CAPSULATUM

‘HARD’
BASIC RELATIONSHIP FOR ‘DAMAGE-RESPONSE FRAMEWORK’

- HOST DAMAGE
- HOST RESPONSE
- DISEASE THRESHOLD
- WEAK
- STRONG

Dr. Liise-anne Pirofski
PROFESSOR
CHIEF, ID DIVISION
ALBERT EINSTEIN/MONTEFIORE

ANTIBODIES ARE NOT INTRINSICALLY GOOD OR BAD: FUNCTION DEPENDS ON THE CONTEXT OF THE HOST RESPONSE

A. SITUATION FOR AN ANTI-INFLAMMATORY Ab

B. SITUATION FOR A PRO-INFLAMMATORY Ab
SOME TAKE HOME MESSAGES

FORGET ABOUT THE OLD CELLULAR VS HUMORAL DIVIDE: THINK ONLY IN TERMS OF PROTECTIVE vs. NON-PROTECTIVE

HOWEVER…’PROTECTIVE’ AND ‘NON-PROTECTIVE’ ARE RELATIVE TERMS THAT DEPEND ON HOST AND OTHER FACTORS

ANTIBODY EFFICACY IS A COMPLEX FUNCTION

\[ \text{PROTECTIVE EFFICACY} = f(\text{SPECIFICITY})(\text{AMOUNT})(\text{ISOTYPE})(\text{HOST GENETICS}) \]

CONSTANT REGION CAN INFLUENCE ANTIBODY FINE SPECIFICITY

ISOTYPE Ab FUNCTION IS PROBABLY PATHOGEN SPECIFIC

FOR \textit{C. NEOFORMANS} EFFICACY: \( \text{IgG2a} > \text{IgG1} > \text{IgG2b} \gg \text{IgG3} \)

FOR \textit{M. TUBERCULOSIS} EFFICACY: \( \text{IgG3} \gg \text{IgG2a} \)
A NEGATIVE EXPERIMENT/TRIAL IS JUST A NEGATIVE EXPERIMENT – AVOID LOGICAL ERRORS BY NOT MAKING POSITIVE INFERENCES FROM NEGATIVE DATA

YOU CAN PROBABLY MAKE A PROTECTIVE ANTIBODY AGAINST ANY PATHOGEN

EVERY ANTIBODY AND EVERY PATHOGEN ARE DIFFERENT – ALL THOUGHT SOME THEMES ARE GENERALIZABLE EACH ANTIBODY-PATHOGEN SYSTM IS UNIQUE

CONSIDERING THE COMPLEXITY I HAVE TOLD YOU ABOUT – IT IS AMAZING, BUT VERY ENCOURAGING THAT WE HAVE ALREADY SUCCEEDED IN MAKING SOME USEFUL VACCINES