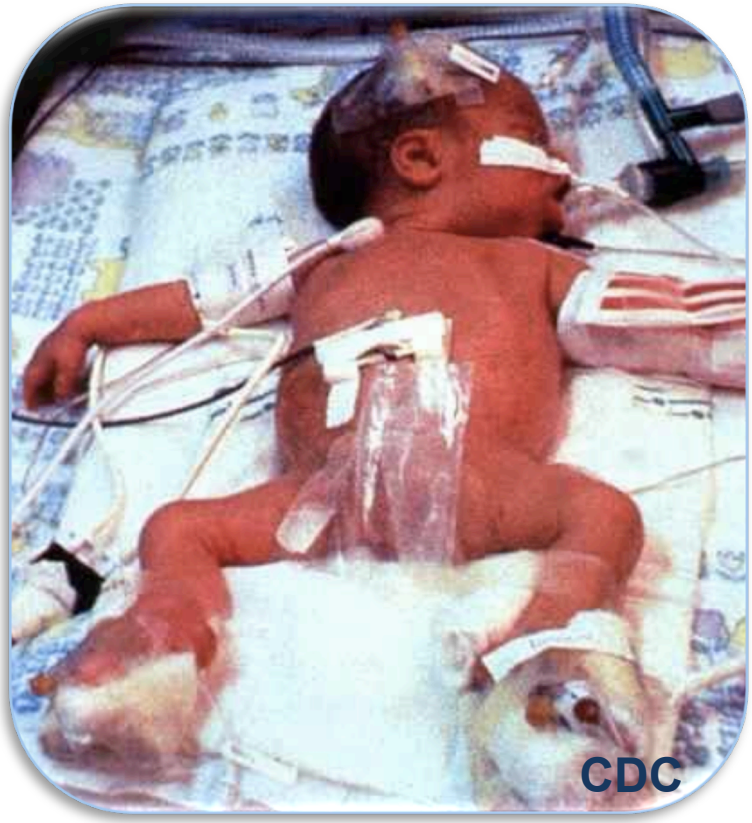


PERINATAL INFECTIONS

The GBS successful practices in prevention

Pierrette Melin

National Reference Centre for GBS
Microbiology, University of Liege
Medical Microbiology, University Hospital of Liege

Definition

Spectrum of infant infections

Mechanism of infection

INTRODUCTION

Perinatal infections

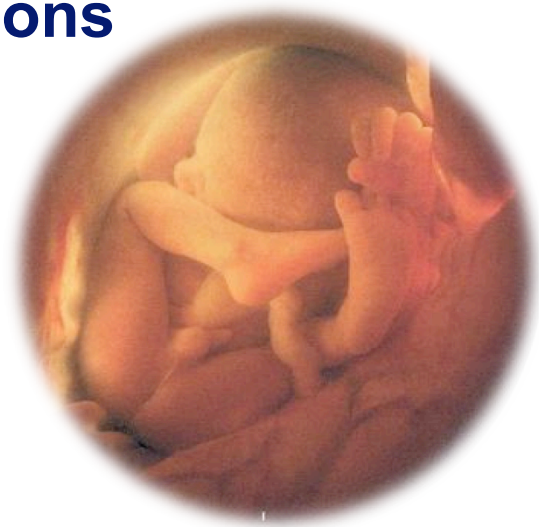
- **Definition**
  - **Bacterial or viral illnesses**
  - **Passed from a mother to her baby**
    - **Usually after rupture of membranes**
    - **In utero**
    - **During delivery process**
  - **Mother, symptomatic or not during pregnancy**

Transmission of Infant infections

Congenital infections

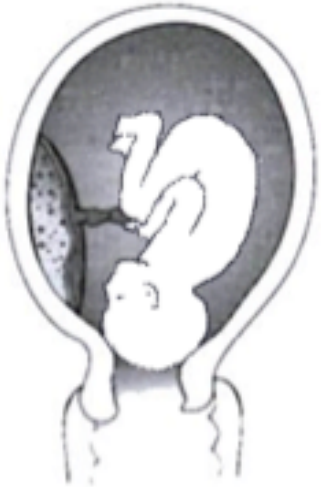
- Growth retardation
- Congenital manifestations
- Fetal loss - stillbirth

Transplacental
Hematogenous



Perinatal infections

- Meningitis
- Septicemia
- Pneumonia
- Preterm labor



By contact, inhalation
(with secretions, blood)
Hematogenous

Neonatal infections

- Meningitis
- Septicemia
- Conjuctivis
- Pneumonia

Breast milk
Person to person
Umbilicus

MAJOR PATHOGENS

Major pathogens

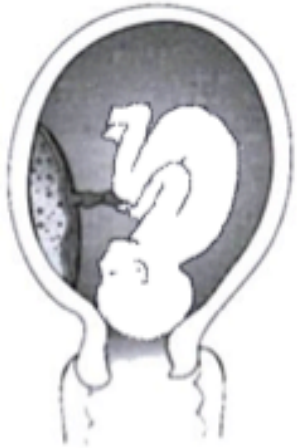
Congenital infections

- Growth retardation
- Congenital manifestations
- Fetal loss – stillbirth

Rubella, CMV, HIV, Toxoplasma gondii, Treponema pallidum, Parvovirus B19, HSV, VZV

Perinatal infections

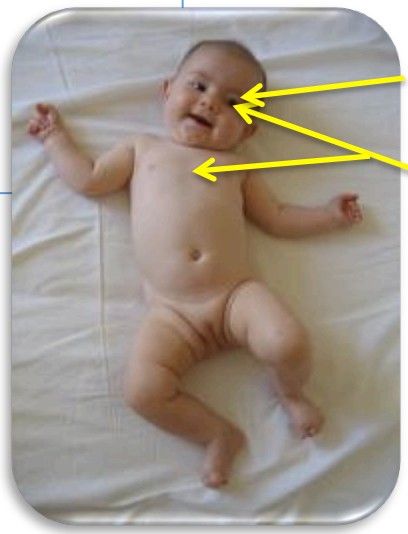
- Meningitis
- Septicemia
- Pneumonia
- Preterm labor



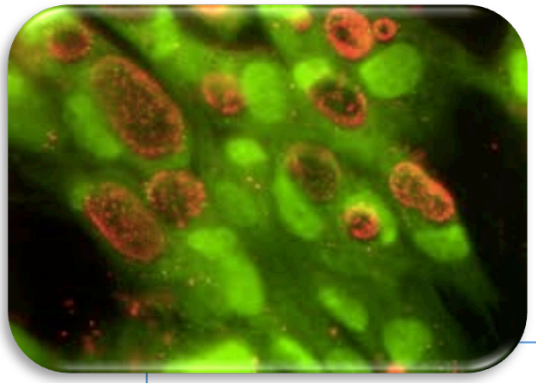
Group B streptococci
N.gonorrhoeae
C.trachomatis
E.coli, Listeria,
HSV,CMV, HIV, HBV

Neonatal infections

- Meningitis
- Septicemia
- Conjuctivis
- Pneumonia

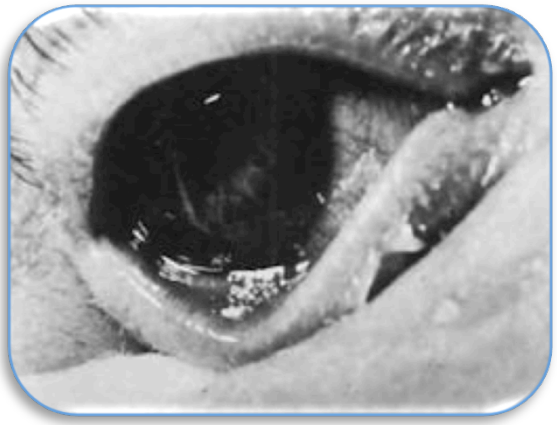


Breast milk
HIV, CMV, HBV
Person to person
GBS, Listeria, E.coli
Umbilicus
S.aureus, tetanos



Chlamydia trachomatis

- **Most common bacterial sexually transmitted disease**
- **No obvious symptoms for majority of women**
- **Infection of mother**
  - → **premature rupture of membranes and early labor**
  - → **ophtalmia neonatorum (20-50%)**
    (within 1st month of life)
  - → **pneumonia**
    (within 1 to 3 months of age)
- **PCR (cervix ok, eyes ?); IF; culture**
- **Screening during pregnancy**
  - **No consensus**



INTERVENTION
Preconceptional / antenatal or perinatal or postnatal

Components of an effective prevention program

- Understanding of biology and epidemiology
- Setting strategic priorities
  - Identify « target » disease and « at risk » populations
  - Conduct cost-effective analysis
    - Burden of disease
      - Incidence, morbidity, mortality, cost of providing care, losss of productivity
    - Cost of preventive intervention
- Investing in material and human resources
- Provide adequate monitoring and evaluation

Is there medical and societal cost-saving ?

Highly effective preventive measures

- **Neonatal tetanus**
  - **Maternal tetanus vaccination / booster**
- **Neonatal ophtalmia**
  - **Topical agents** (not efficient against *C.trachomatis*)
    - **Silver nitrate, erythromycin, tetracycline, povidone iodine**
- **Hepatitis B**
  - **Screening and vaccination**
- **HIV**
  - **Anti-retroviral therapy**

Introduction & burden

Guidelines

Screening

vaccine

GROUP B STREPTOCOCCI

Successful practices in prevention

Streptococcus agalactiae or GBS

Gram positive cocci
- **Catalase -**
- **β-hemolytic**
- **CAMP test +**
- **Hippurate +**
- **Esculine-**
- **Orange pigment**

10 capsular serotypes (Ia, Ib, II-IX)

1887, Noccard-Mollereau, bovine mastitis

1933, Group B Antigen

1964, severe neonatal sepsis

➢**1970, N°1 in neonatal infections**

Group B streptococcal diseases in neonates

- Since the 1970s, leading cause of life-threatening infections in newborns
  - Neonatal illness/death
  - Long-term disabilities

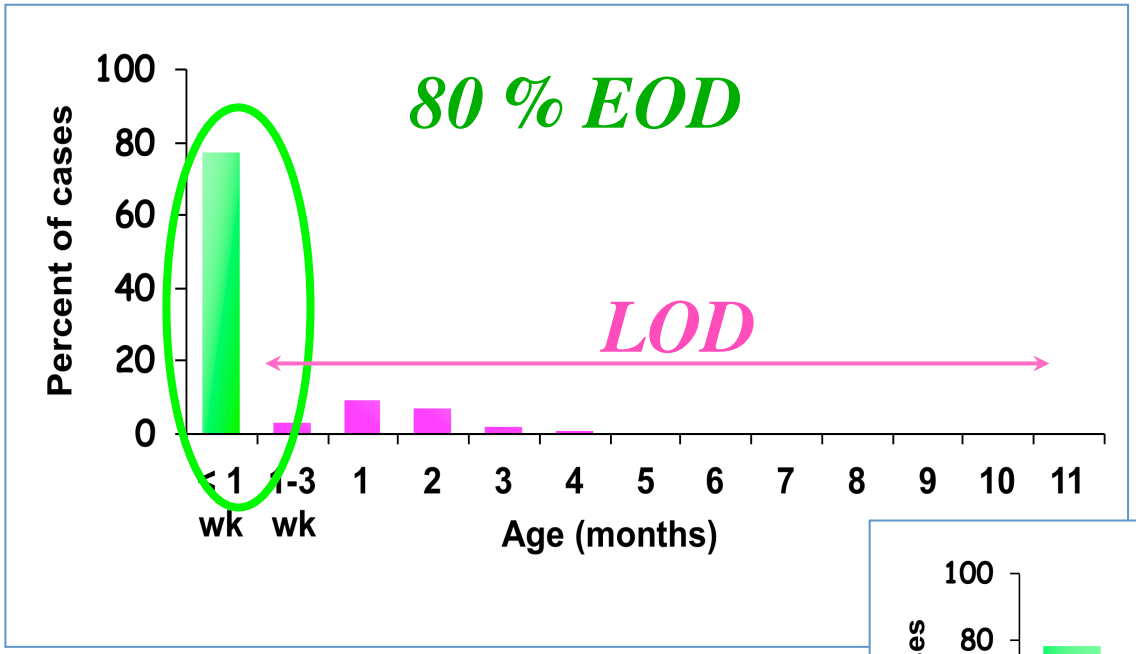
GLOBAL public health major concern !

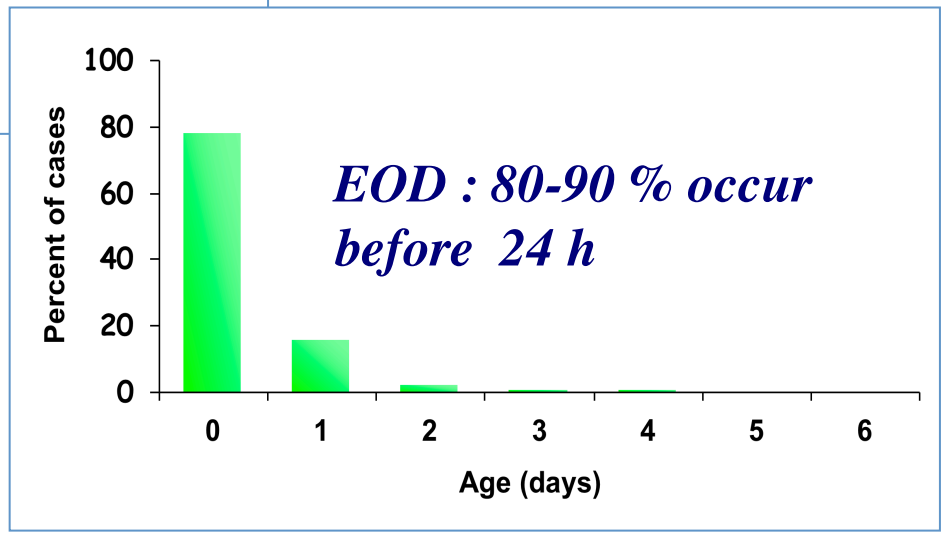
Also in developing countries

- **Maternal morbidity**
  - **Along pregnancy**
  - **Peripartum**
- **Serious diseases among elderly and adults with underlying diseases**
  - **Significant mortality**

GBS Neonatal Infections

A. Schuchat, Clin Microb Rev 1998;11:497-513





Burden of neonatal GBS early onset diseases in European countries

Location	Incidence per 1,000 live-births	*Reference*
Spain	2 (1996) to 0.45 (2008)	*Lopez Sastre et al. Acta Pediatr 2005*
Belgium	2	*Melin, Indian J Med Res 2004*
Eastern Europe	0.2 - 4	*Trijbels-Smeulders, Pediatr Infect Dis J 2004*
Western Europe	0.3 - 2	
The Netherlands	1.9	
Scandinavia	0.76 - 2	
Southern Europe	0.57 - 2	

- Definition?
- Carriage rate ?
- Ethnicity ?
- Sub-reporting?
- Systematic diagnostic approach?
- Virulence?

Data assessing more accurately the true burden are needed

GBS EOD vertical transmission

GBS colonized mothers

60 - 40 %

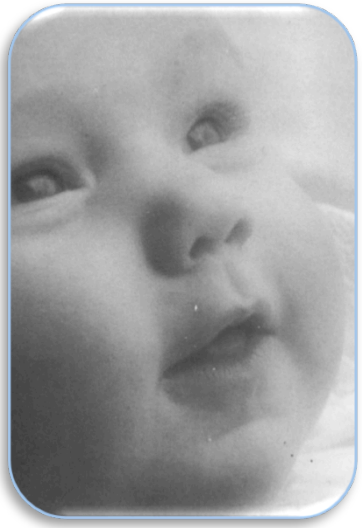
40 - 60 %

Non-colonized newborns

Colonized newborns

Risk factors

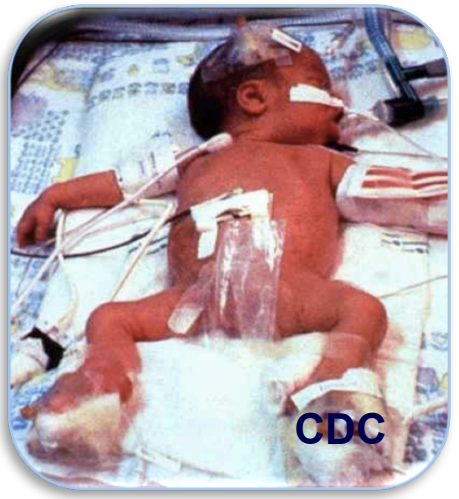
2 - 4 %
GBS EOD



96 - 98 %

Asymptomatic

sepsis
pneumonia
meningitis
long term sequelae



GBS maternal colonization

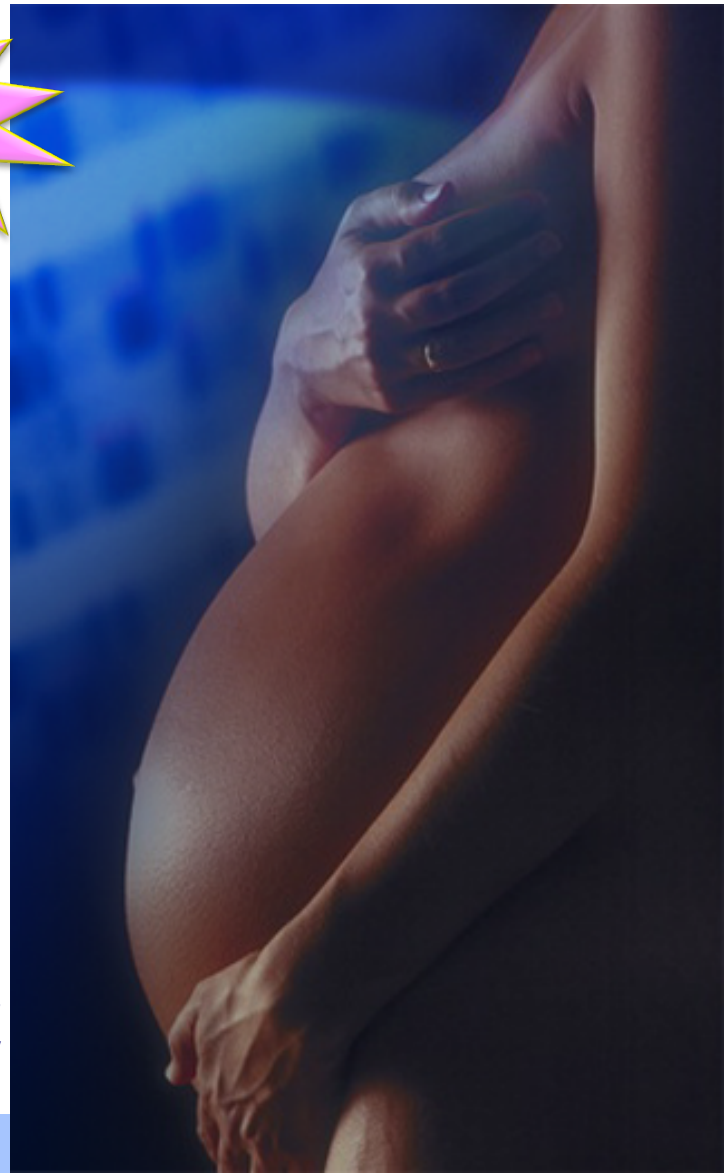
Risk factor for early-onset disease (EOD) :
vaginal GBS colonization at delivery

- **GBS carriers**
  - **10 - 35 % of women**
  - **Clinical signs not predictive**
  - **Dynamic condition**
  - **Intestinal reservoir**
  - **Prenatal cultures late in pregnancy can predict delivery status**

Additional Risk Factors for Early-Onset GBS Disease

- **Obstetric factors:**
  - **Prolonged rupture of membranes,**
  - **Preterm delivery,**
  - **Intrapartum fever**
- **GBS bacteriuria**
- **Previous infant with GBS disease**
- **Immunologic:**
  - **Low specific IgG to GBS capsular polysaccharide**

Risk factors



No difference in occurrence either in GBS Positive or Negative women, except intrapartum fever

Lorquet S., Melin P. & al.

J Gynecol Obstet Biol Reprod 2005

GBS EOD - Belgian data

- **Incidence**
  - **1985 -1990: 3/1000 live births**
  - **1999, estimation : 2/1000 live births**
  - **2010, estimation : < 1/1000 live births**
- **Meningitis : 10 %**
- **Mortality : 5 -10 %**
- **60 % EOD (130 cases) : WITHOUT any maternal/ obstetric risk factor except colonization**
- **Prenatal screening**
  - **Recto-vaginal cultures : 13-35 % GBS Positive**

P. Melin - 2001, 2007 - Reference laboratory for GBS.

Stages in the pathogenesis of GBS neonatal EOD : *Bacterial & individual factors*

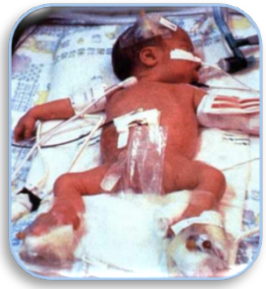
Meningitis

Brain barrier
Pili,
β-hemolysin, …

Colonization : adhesion to epithelial cells
different virulence factors (pili, scpB, …)

Ascendant transmission (amnionitis)

GBS pathogenesis



Sepsis

IL1, IL6, TNF α, PGE2, TxA_2 ,

Bacteria
Peptidoglycan
β-hemolysin, …

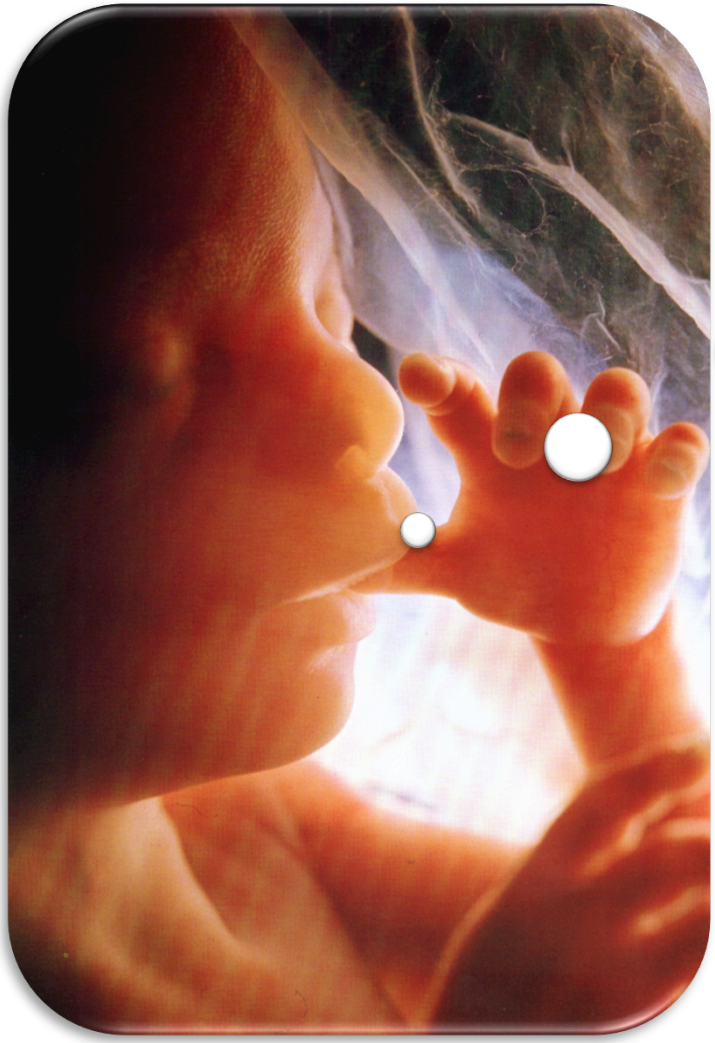
Resistance to phagocytose
- **Capsule**
- **C5a peptidase**
- **…..**

β-hemolysin, invasins (pneumonia)

Phagocytes cells, Antibodies, Complement

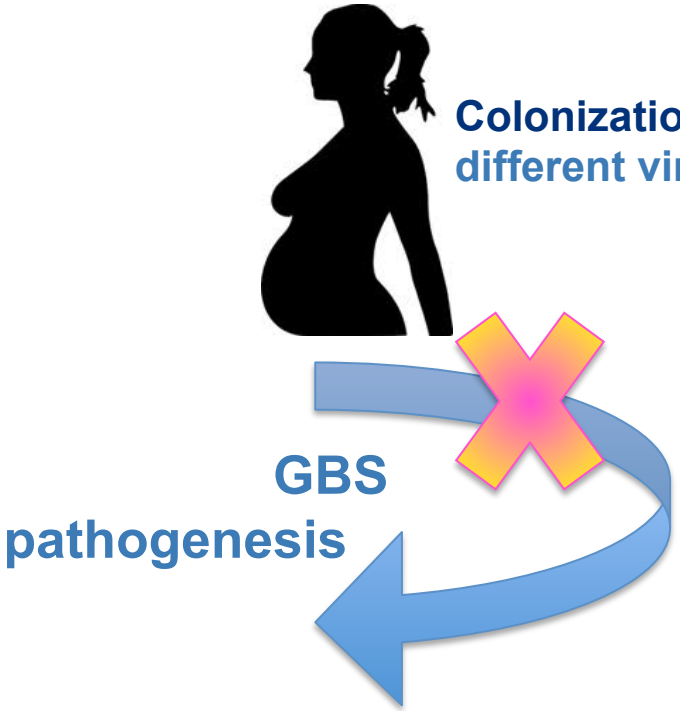
- Universal prenatal screening-based strategy
- Risk-based strategy
- No guideline

GUIDELINES FOR PREVENTION OF GBS PERINATAL DISEASE



Which prevention strategy for GBS perinatal diseases ?

Stages in the pathogenesis of GBS neonatal EOD : *Bacterial & individual factors*



Intrapartum antibioprophylaxis
> 4 (2) hours before delivery

Stages in the pathogenesis of GBS neonatal EOD : *Bacterial & individual factors*

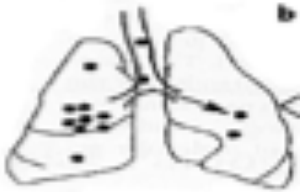
GBS vaccine

« still expected »

Colonization : adhesion to epithelial cells
different virulence factors (pili, scpB, …)

Ascendant transmission (amnionitis)

GBS pathogenesis



β-hemolysin, invasins (**pneumonia**)

Resi… phag…
- Ca…
- C… …ase
- …..

Phagocytes cells,
Antibodies, Complement

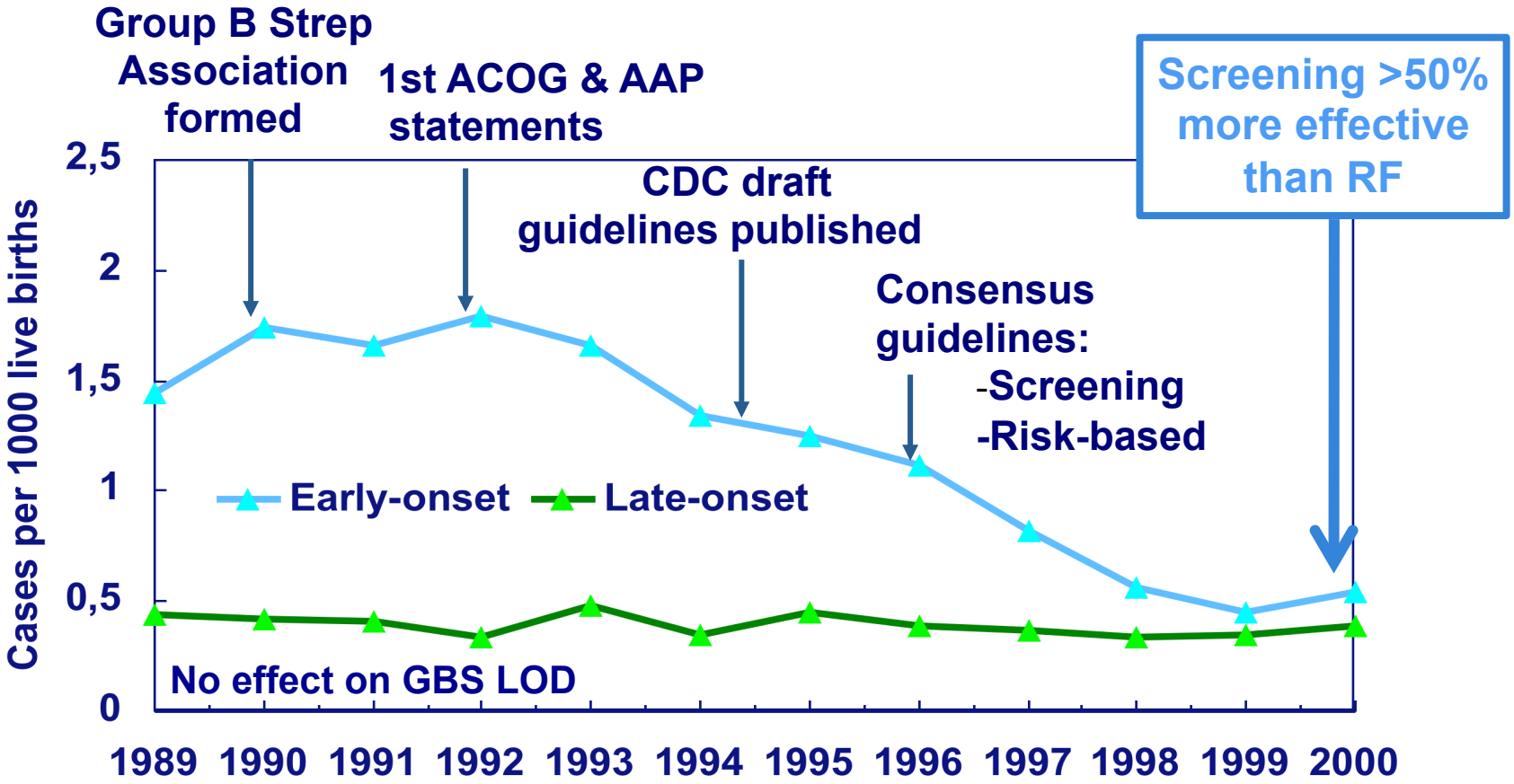
Prevention of perinatal GBS EOD

- **Intrapartum antibiotics**
  - **Highly effective at preventing EOD in women at risk of transmitting GBS to their newborns (≥ 4 h)**

(clinical trials in late 80s)

Risk-based strategy
or
Screening-based strategy

Impact of prevention practices Early- and Late-onset GBS Diseases in the 1990s, U.S.



S. Schrag, New Engl J Med 2000
Schrag S. et al. N Engl J Med 2002; 347:233-9

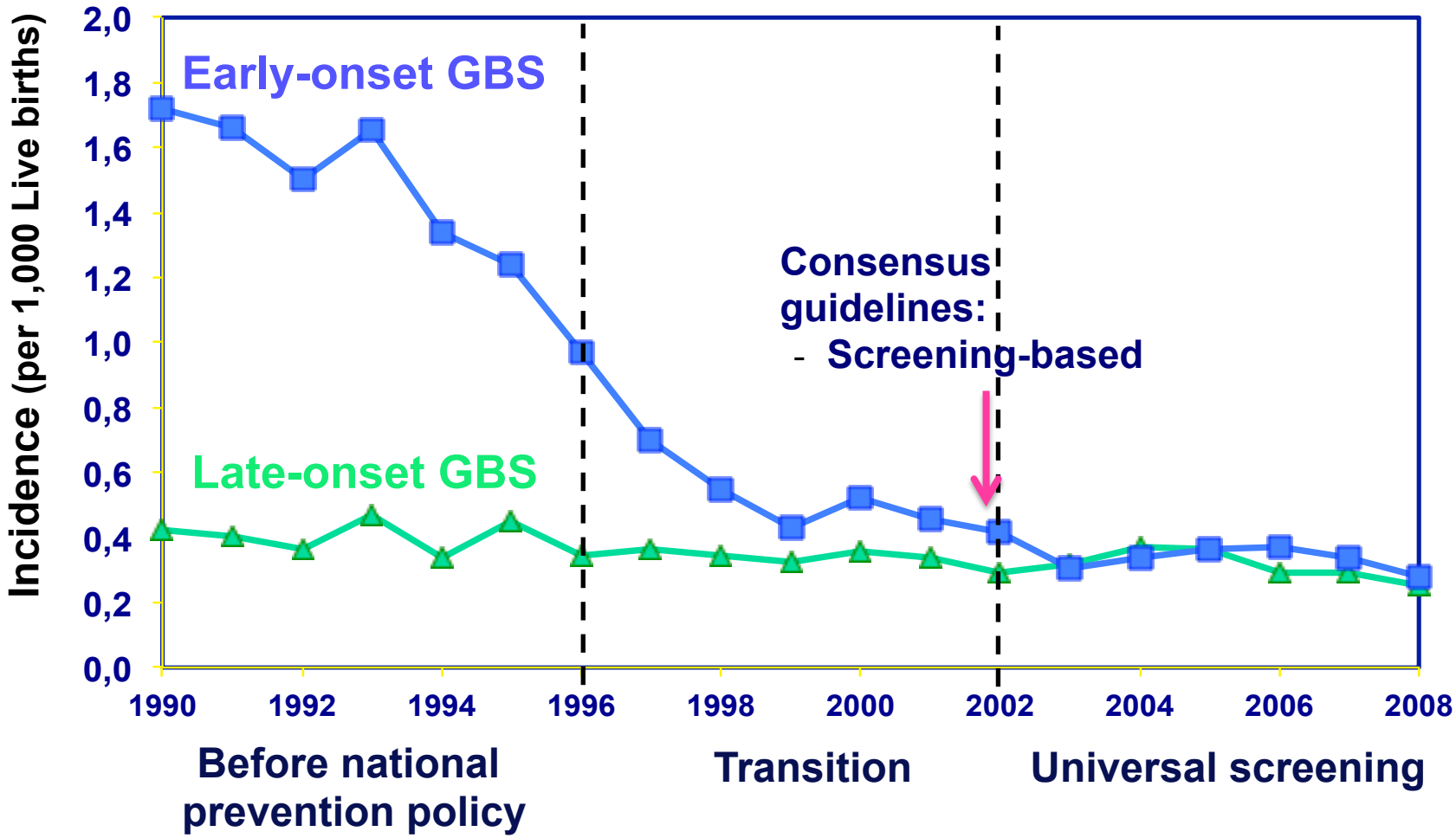
Why is Screening more protective than the risk-based approach ?

Schrag S. et al. N Engl J Med 2002; 347:233-9

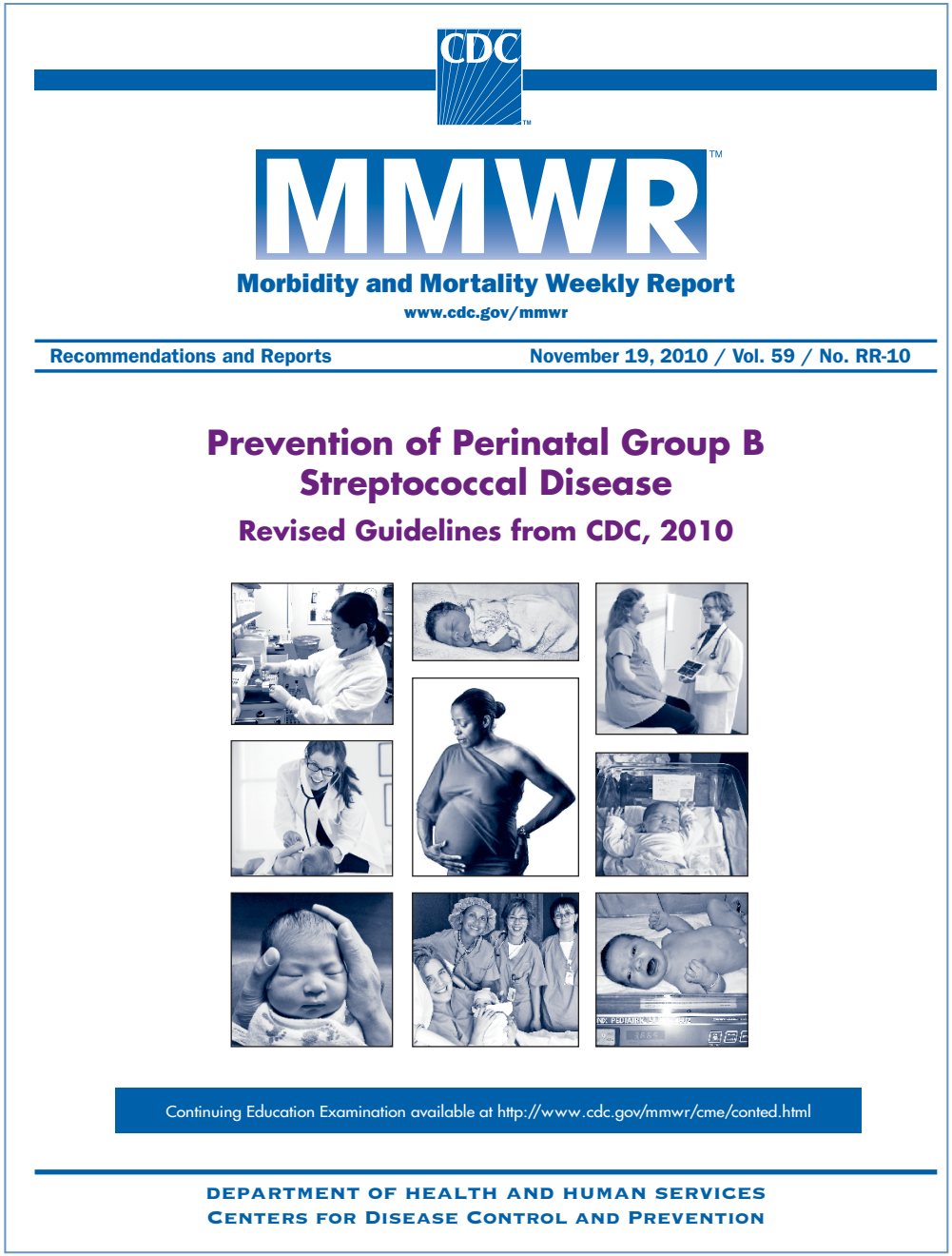
Broader coverage of « at-risk » population

- **Captures colonized women without obstetric RF**
- **High level of compliance with recommendations**
- **Enhanced compliance with risk-based approach cannot prevent as many cases as universal screening**

Impact of prevention practices
Early- and Late-onset GBS Diseases, U.S.



Incidence of early- and late-onset invasive group B streptococcal disease in selective Active Bacterial Core surveillance areas, 1989-2008 (CDC 2010)

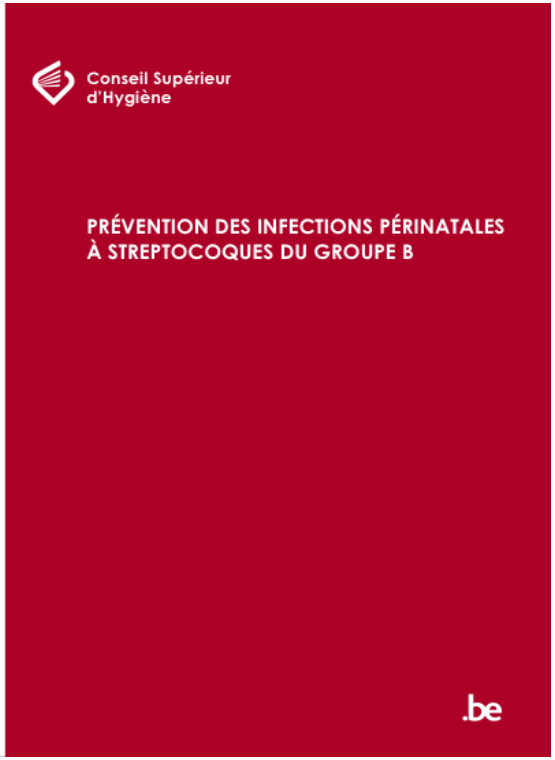


CDC, USA, MMWR, Vol 59 (RR-10) August 2010

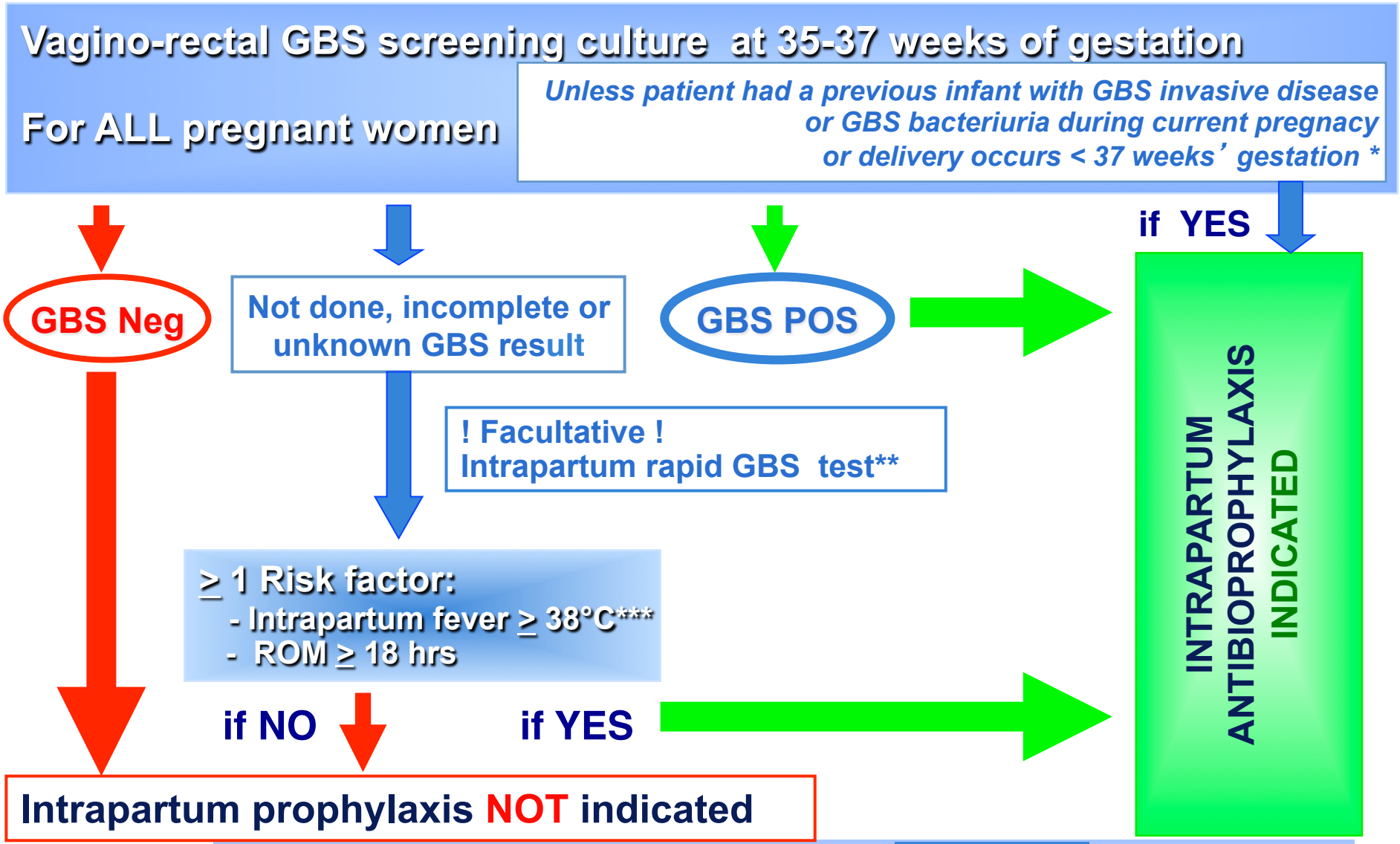
Endorsed by

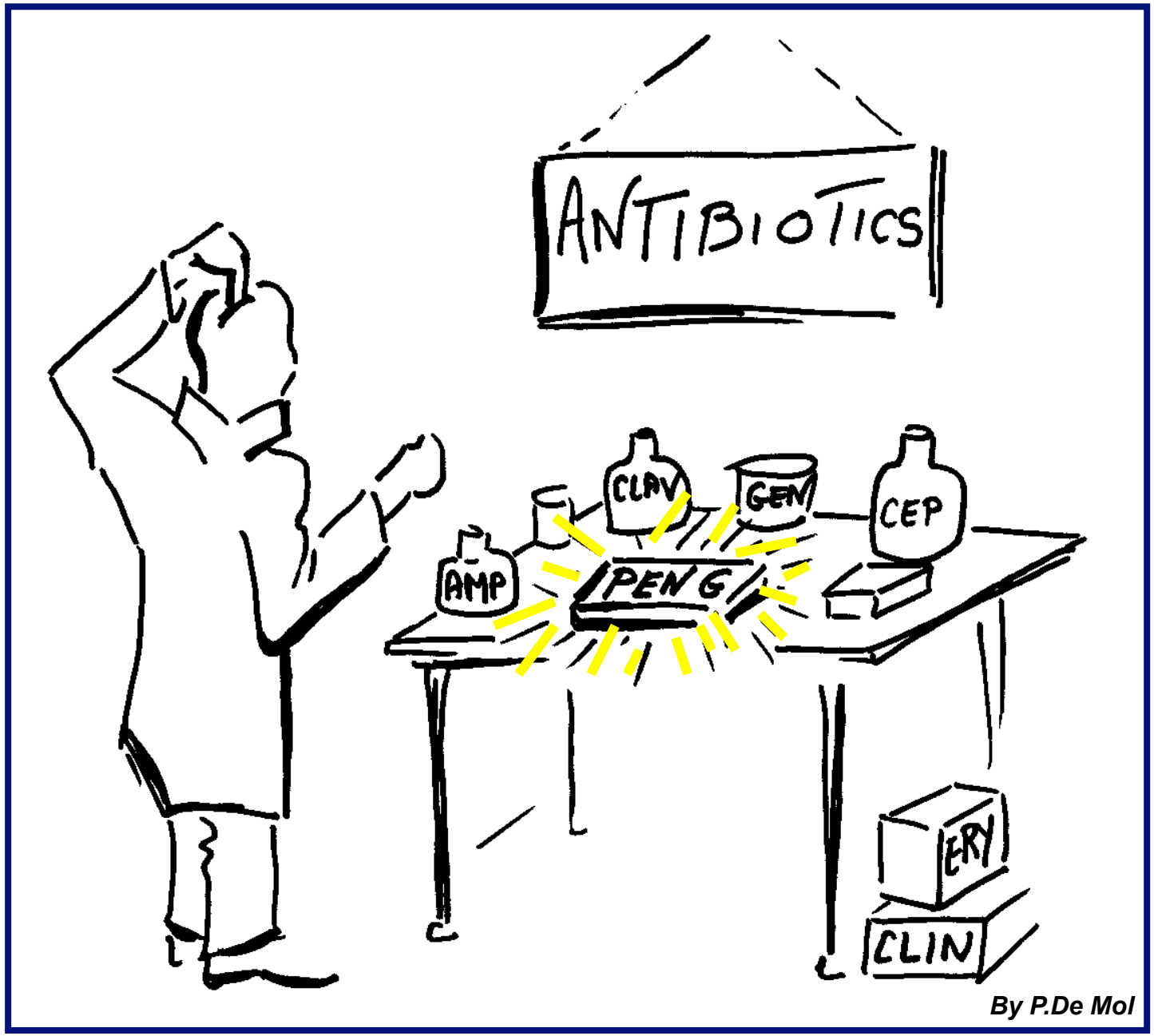
- ***AAP***
- ***ACOG***

SHC, Belgium July 2003
Revision ongoing



Universal screening-based strategy for prevention of GBS perinatal disease




ANTIBIOTICS
CLAV
GEN
CEP
AMP
PEN G
ERY
CLIN
By P.De Mol

Remaining burden of GBS EOD
Missed opportunities

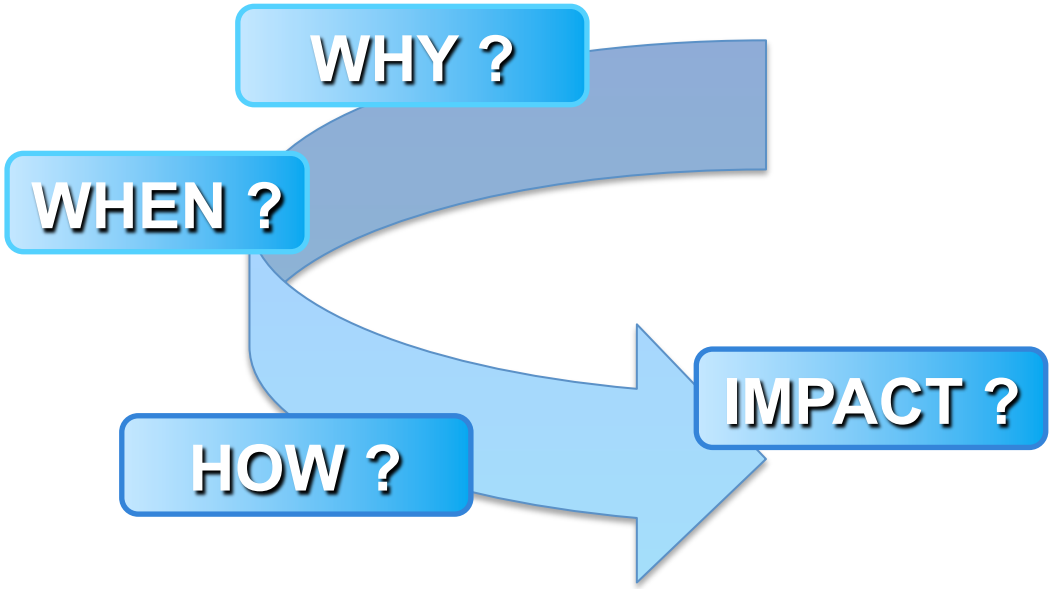
In spite of universal screening prevention strategy

In spite the great progress

Cases still occur

- **Among remaining cases of EOD**
  - **Some may be preventable cases**
    - **Missed opportunities for (appropriate) IAP**
    - **False negative screening**

Van Dyke MK, Phares CR, Lynfield R et al. N Engl J Med 2009
CDC revised guidelines 2010
Poyart C, Reglier-Poupet H, Tazi et al. Emerg Infect Dis 2008
DEVANI project, unpublished data 2011



SCREENING FOR GBS COLONIZATION

Antenatal GBS culture-based screening

Goal of GBS screening

To predict GBS vaginal (rectal) colonization at the time of delivery

- **Critical factors influencing accuracy**
  - **Swabbed anatomic sites**
  - **Timing of sampling**
  - **Screening methods**
    - **Culture**
      - ***Procedure***
      - ***Media***
    - **Non-culture**

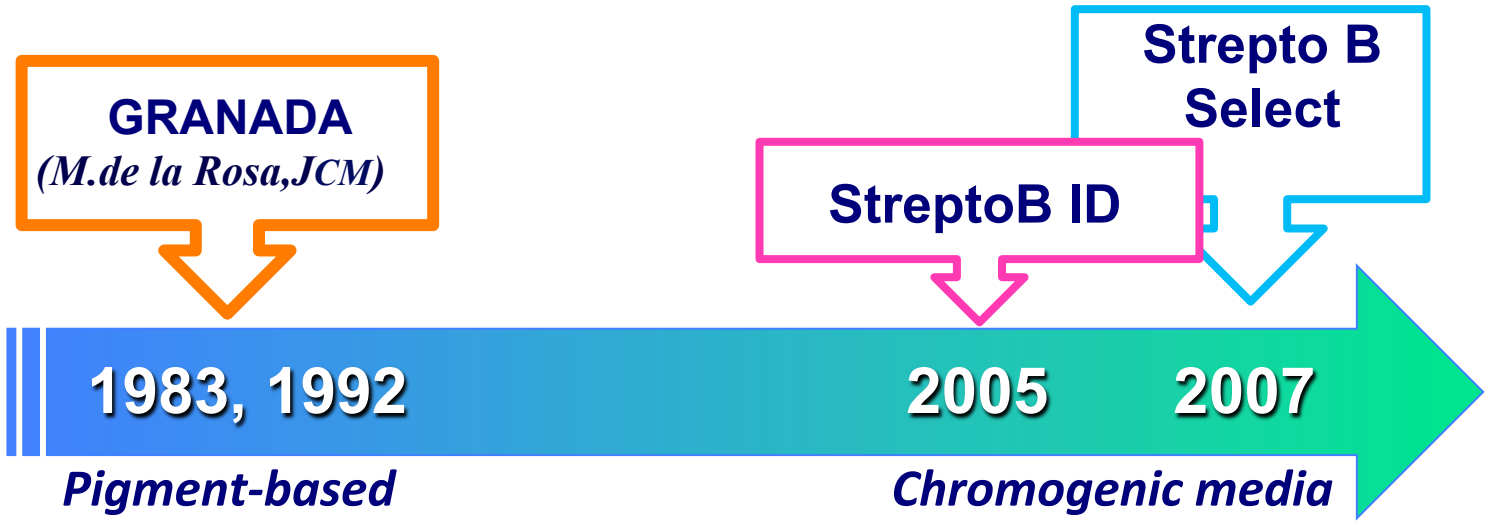
From direct plating on blood agar
Evolution of culture methods

Use of selective enrichment broth

- To maximize the isolation of GBS
- To avoid overgrowth of other organisms

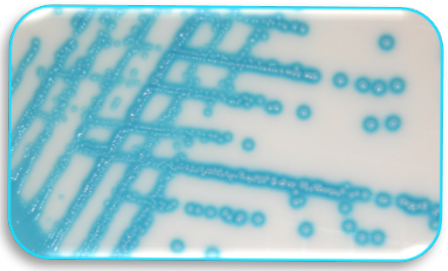
Use of differential agar media

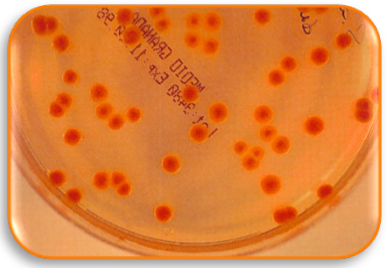
Recommended by some European guidelines (+ CDC 2010)

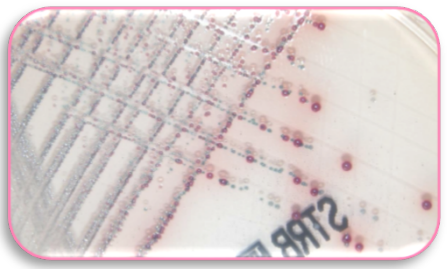


Which agar or which combination?

+/- Blood agar







Workload - costs - extra-testing - non β-hemolytic GBS detection to be considered

Crucial conditions to optimize SCREENING

- **WHEN** — **35-37 weeks**
- **WHO** — **ALL the pregnant women**
- **Specimen** — **Vaginal + rectal swab(s)**
- **Collection** — **WITHOUT speculum**
- **Transport** — **Transport/collection device/condition**
  (non nutritive medium: Amies/Stuart
  (type of swab)(Length and T°)
- **Request form** — **To specify prenatal « GBS » screening**
- **Laboratory procedure**

(CDC 2010 - Belgian SCH 2003)

Crucial conditions to optimize SCREENING

- WHEN — 35-37 weeks
- WHO — ALL the pregnant women
- Specimen — Vaginal + rectal swab(s)
- Collection — WITHOUT speculum
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(CDC 2010 - Belgian SCH 2003)

Crucial conditions to optimize SCREENING

Transport-collection system & transport-storage condition

Preliminary results (2012, NRC GBS)

- **Use of a selective enrichment Lim broth** ***(BD, Copan, bioMérieux)***
  - **At RT° up to 35°C**
  - **Between 4-8°C**

- **Use of a selective enrichment Granada medium** ***(bioMérieux)***
  - **At RT° up to 35°C**
  - **Between 4-8°C**

Crucial conditions to optimize SCREENING

Transport-collection system & transport-storage condition

***Preliminary results** (2012, NRC GBS)*

- **Use of a selective enrichment Lim broth** *(BD, Copan, bioMérieux)*
  - **At RT° up to 35°C**
    - **Rapid important amplification of GBS initial inoculum**
    - **Sustained viability > 4 days**
  - **Between 4-8°C**
    - **≥ 24 hours, continuous decrease of life GBS**

- **Use of a selective enrichment Granada medium** *(bioMérieux)*
  - **At RT° up to 35°C**
    - **Rapid important amplification of GBS initial inoculum**
    - **Sustained viability at RT°**
    - **Abrupt lost of viability at 35°C ≥ 48-72h**
  - **Between 4-8°C**
    - **≥ 24 hours, continuous decrease of life GBS**

Prenatal culture-based screening: Limiting factors

- **Positive and negative predictive values**
  - **False-negative results**
    - **Failure of GBS culture (oral ATB, feminine hygiene, delay before culture) or new acquisition**
    - **Up to 1/3 of GBS positive women at time of delivery**
    - **Continuing occurrence of EO GBS cases**
  - **False-positive**
    - **Positive prenatal screening /negative at time of delivery**
    - **Unnecessary IAP**

Need for more accurate predictor of intrapartum GBS vaginal colonization

Prenatal culture-based screening: Limiting factors

- **Positive and negative predictive values**
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Need for more accurate predictor of intrapartum GBS vaginal colonization

Prenatal culture-based screening combined with *illumigene*® Group B Streptococcus assay

A loop mediated isothermal amplification (LAMP) assay by Meridian Bioscience, Inc

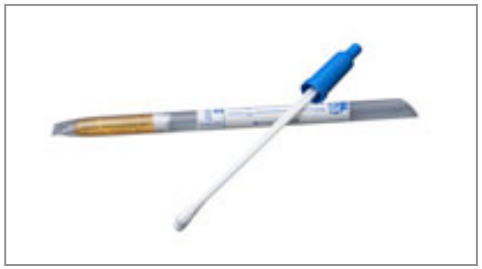
- **Broth enrichment followed by *illumigene*® GBS**
  - Speed and accuracy
  - DNA detection

Evaluation of the *illumigene*® GBS

Cf. Poster M5, Dodemont M., Vanhouteghem K. et al.

242 vagino-rectal swabs

CHU Lg
& AZ St-Lucas Gent



And

Granada agar

18 – 48h *at 35°C/ana*

LIM broth

Overnight subculture at 35°C and

Granada agar

Strep B select

illumigene GBS

18 – 48h ana

at 35°C air

Primary culture GBS Pos (1-4+)/Neg

Enrichment GBS Pos (1-4+)/Neg

illumigene GBS Pos/Neg/Invalid

Evaluation of the *illumigene*® GBS

		GBS culture		
		Positive	Negative	
illumigene GBS	Positive	45	2	47
	Negative	5	188	193
		50	190	240

GBS Positive cultures: 20.7%

illumigene GBS *vs* GBS reference culture (all discrepancies were retested)

Sensitivity	90.0 %
Specificity	98.9 %
PPV	95.7 %
NPV	97.4 %
Efficiency	97.1 %

Evaluation of the *illumigene*® GBS

		GBS culture		
		Positive	Negative	
illumigene GBS	Positive	45	2 : PCR pos	47
	Negative	2 positive 3 very rare GBS	188	193
		50	190	240

GBS Positive cultures: 20.7%

illumigene GBS *vs* GBS reference culture /GBS DNA

Sensitivity	90.0 %	→ 95.7%
Specificity	98.9 %	→ 100%
PPV	95.7 %	→ 100 %
NPV	97.4 %	→ 99 %
Efficiency	97.1 %	

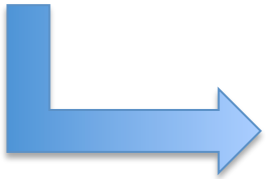
Evaluation of the *illumigene*® GBS

- Speed and accuracy
- Easy to perform, short hands-on-time
- Good comparison to reference culture method

  - 100% specificity and positive predictive value
  - High sensitivity and negative predictive value
  - Identification of >= 0.8% additional GBS positive specimen
  - Overall cost and logistic to be considered

Prenatal culture-based screening: Limiting factors

- **Unknown GBS status at presentation for delivery**
  - **Screening performed but result not available**
  - **Women with no prenatal care**



Risk based strategy

- **60% at GBS risk not identified**
- **> 10% of unnecessary IAP**

Need for rapid accurate predictor of intrapartum GBS vaginal colonization

Alternative to GBS prenatal screening: intrapartum screening
Theranostic approach

Turnaround time

collect specimen at admission

Specimen Analysis "POCT" ?

Results

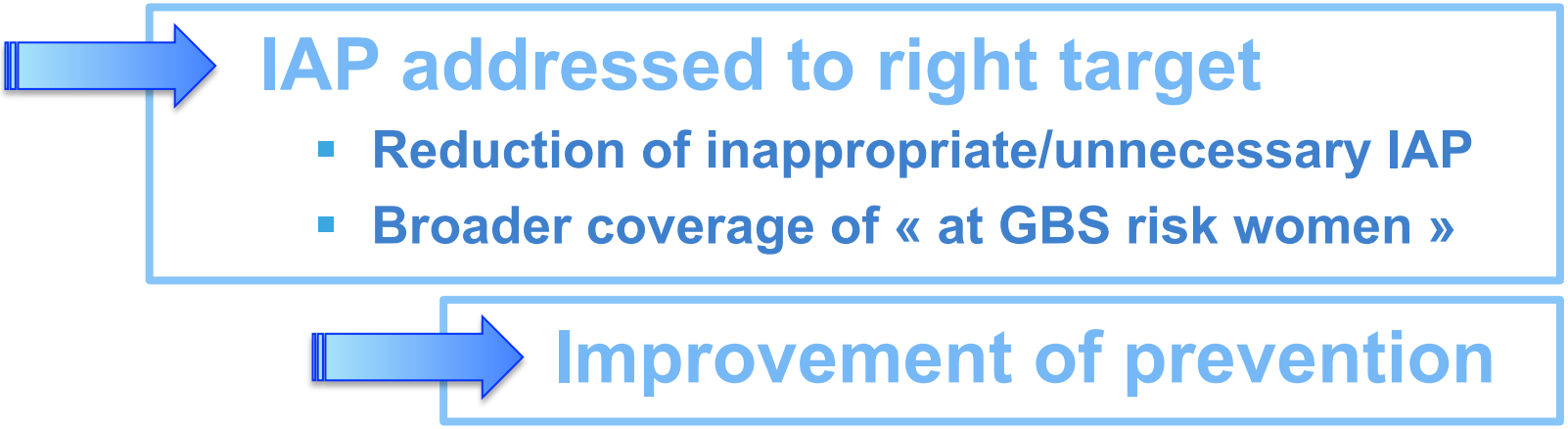
Optimal management of patient

30-45 minutes, 24 hrs/7 d, robust

Benitz et al. 1999, Pediatrics, Vol 183 (6)

Intrapartum screening theranostic approach: expected advantages

- Inclusion of women without prenatal screening/care
- Identification of women with change of GBS status after 35-37 wks gestation
- Increased accuracy of vaginal GBS colonization status at time of labor & delivery



Real Time PCR for intrapartum screening

- **Advance in PCR techniques & development of platforms**
  - **BD GeneOhm™ Strep B Assay (+/- 1 hr) (in laboratory)**
  - **Xpert GBS, Cepheid (35-45 min)** (can be performed as a POCT)

Real-time PCR, very promising

- **Rapid, robust & accurate technology**
- **Still an expensive technology (specific equipment)**
  - Cost effective ?
    - *Need for more cost-effective clinical study*
- **Logistic**
  - 24 hours 7 days
  - In the lab?
  - In the obstetrical department as a POCT ?
- **In combination with prenatal screening strategy ?**
  - CDC 2010 : for women with premature delivery or no prenatal care
- **No antimicrobial result**
  - In the future detection of R genes, but mixed microbiota !

Prevention of GBS EOD and LOD

VACCINE

Vaccine - Background

- **Correlate between maternal low level off CPS type Ab at time of delivery and risk for development of GBS EOD**

Baker C et Kasper D, 1976, NEJM

Vaccine for pregnant women:

Likely the most effective, sustainable and cost effective approach

GBS Vaccines, since the 1980s Challenges

Capsular polysaccharide vaccines

- 10 serotypes
  - Different distributions
    - EOD, LOD, invasives infections in adults
    - Geographically and along time
- Conjugated vaccines
- Multivalent vaccines Ia, Ib, III, V
- Clinical studies
  - Immunogenicity : ok
  - Safety : ok
  - Efficacy: scheduled/ongoing

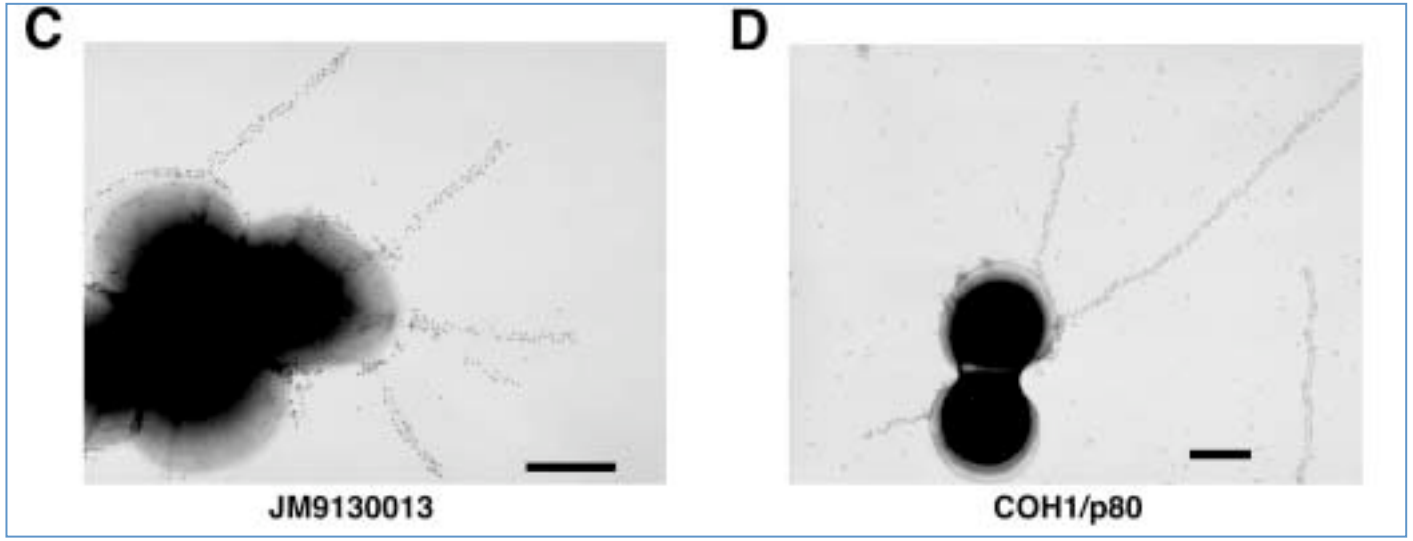
GBS Vaccines

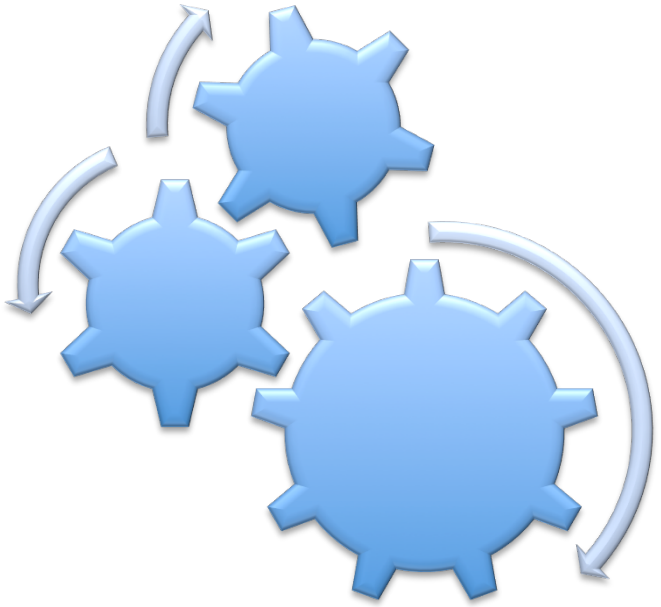
GBS Protein-based Vaccine

- **Ag = Surface proteins**
  - **Cross protection against different serotypes**
  - **Better immunogenicity**
    - **Humoral response T-cell dependent**
      = long lasting immunity

GBS Vaccines
GBS « pilus like structure »

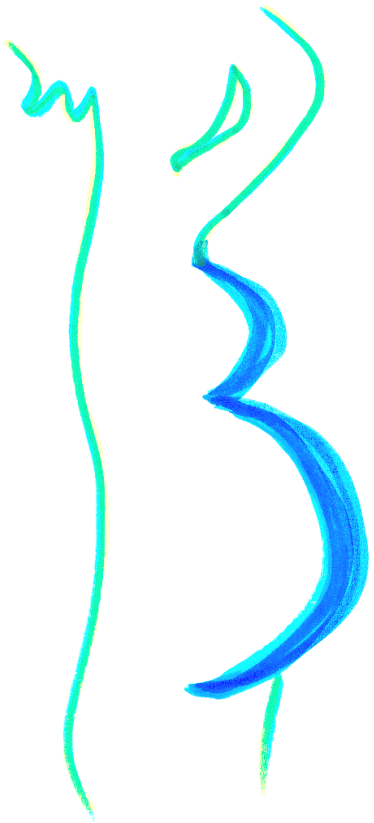
- Highly immunogenic proteins
- Elicit protective and functional antibodies
- Virulence factor
  - Adhesion
  - Transcytose through cells





CONCLUSION
Take home messages

GBS Summary



- EOD & LOD, a public health concern
  - IAP, an effective prevention
- "Screening" Prevention strategies
  - Improvement of culture-based GBS prenatal screening
  - Culture-LAMP combined GBS prenatal screening
  - Room for a rapid intrapartum screening (POCT)
- Development of a vaccine
  - Against pili proteins and major capsular polysaccharidic serotypes