



Group B streptococci, a European perspective with results of the

) **DEVANI** project

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Group B streptococci (GBS)

The global picture of neonatal disease

- **Worldwide mortality 0-4 years old** (WHO, Cause of death 2008)
 - 8,3 millions
 - 30-40% within first week of life
- **Neonatal bacterial sepsis**
 - +/- 1 million annually
 - GBS is the leading cause

- **Maternal immunization**
 - **Cornerstone of prevention**
 - Neonatal tetanos and influenza
 - **Potential to protect young infants**

Group B streptococci (GBS)

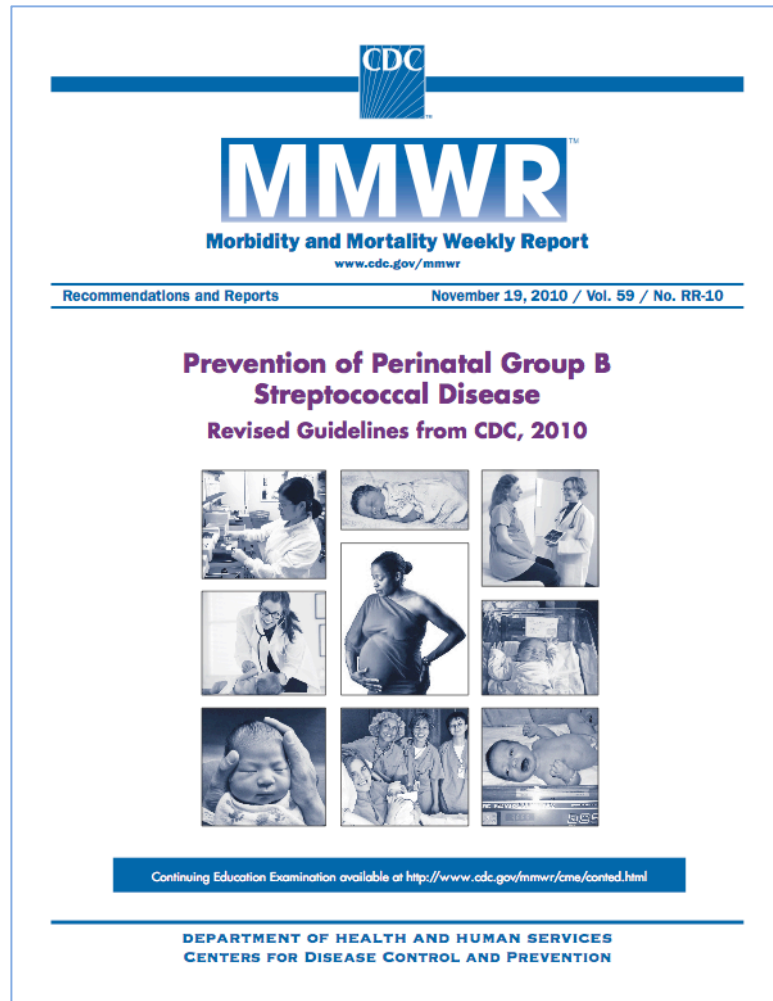
The global picture of neonatal disease

- **In industrialized countries, since 1970's**
 - **Leading cause of pneumonia, sepsis, meningitis**
 - 0.5 to 4 /1000 live births
 - **EOD, mortality 5-10%**
 - **LOD, mortality 3-5%**
 - **Meningitis**
 - 50% permanent sequelae
 - From mild learning or motor disabilities to global cognitive impairment
 - **Maternal colonization: 15-40%**
- **In resource-limited countries**
 - **Many common characteristics with industrialized countries**

**Global public health
major concern !**

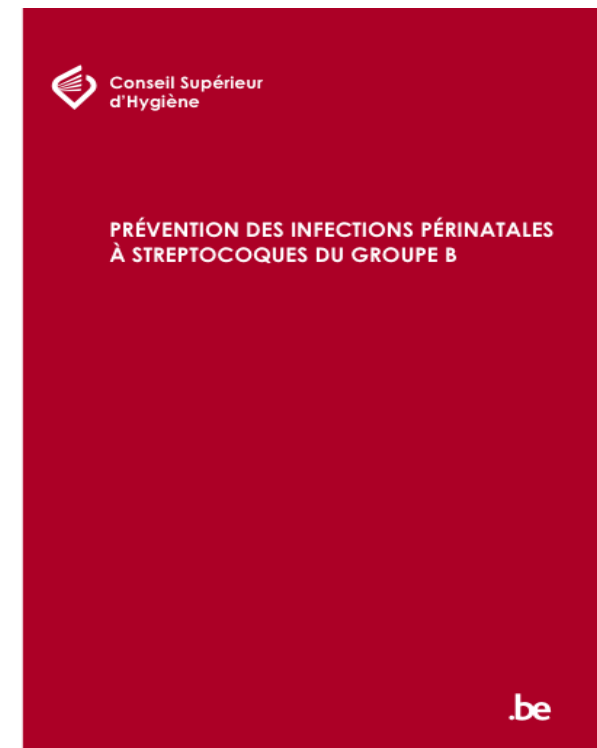
Group B streptococci (GBS)

The global picture of neonatal disease



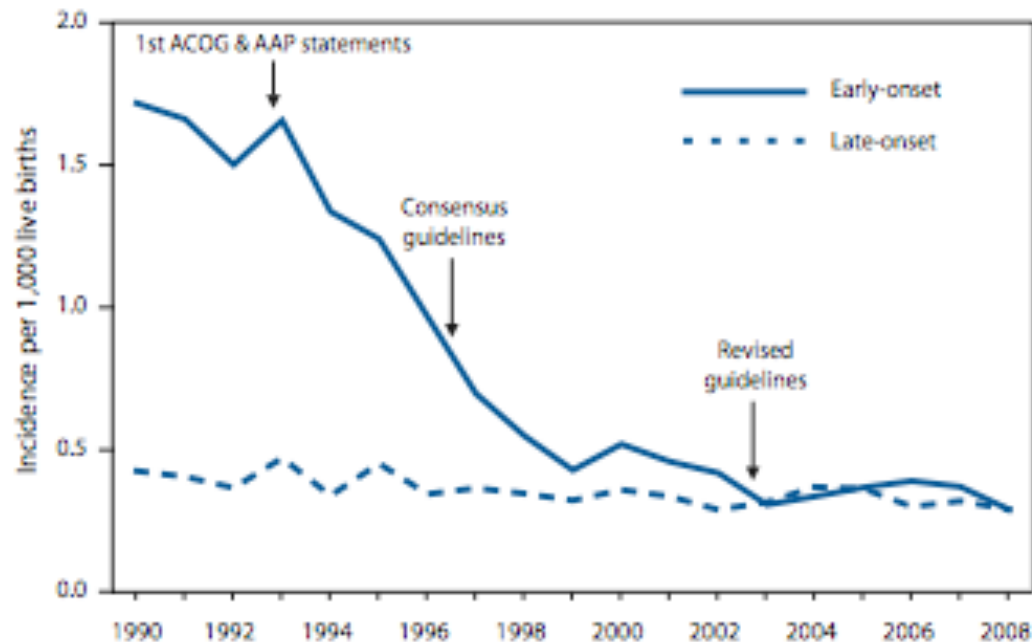
CDC, USA, MMWR, Vol 59 (RR-10) Nov.2010
Endorsed by ACOG, AAP, ACNM, AAFP and ASM

CSS, Belgium July 2003 (Revision ongoing)



Group B streptococci (GBS)

The global picture of neonatal disease



Incidence of GBS EOD and LOD, 1990 to 2008, ABC surveillance areas, USA

- **Prevention through IAP**
 - **In industrialized countries**
 - Substantial declines of EOD
 - Remaining burden
 - No effect on LOD
 - Several concerns
 - **In resource limited countries**
 - Not an option
 - Intrapartum vaginal and newborn chlorhexidine washes proven ineffective

European strategies for prevention of GBS EOD

- **Prevention through IAP**
 - **Screening-based strategy**
 - Spain, 1998, revised 2003
 - France, 2001
 - Belgium, 2003, revision ongoing 2011
 - Germany, 1996, revised 2008
 - Switzerland, 2007
 - **Risk-based strategy**
 - UK, the Netherlands
- **No guidelines**
 - Bulgaria, ...

GBS Vaccines

- **GBS neonatal disease**
 - **Mainly CPS type III followed by Ia, V, Ib, II**
 - **Substantial perinatal morbidity and mortality**
 - **Especially in the first 48 hrs of life**
 - **Concern about IAP**
 - **Higher levels of maternal specific CPS Ab // reduction of risk of neonatal disease**

- **GBS Vaccines**
 - **Uniquely suited for maternal immunization**
 - **To prevent GBS disease in young infants**

Since the 1980's: GBS Vaccines, Challenges

Capsular polysaccharide (CPS) vaccines

- **10 serotypes Ia, Ib – IX**
 - **Variability of CPS distribution**
 - Type of infections: EOD, LOD, in adults
 - Geographically and along time
- **Conjugated vaccines**
- **Multivalent vaccines Ia, Ib, II, III, V**
- **Clinical studies (*Phase I and II*)**
 - Immunogenicity ; Safety ; Efficacy (scheduled / ongoing)
 - Ia, Ib, III conjugated to CRM197 (Novartis) clinical trials in Belgium

→ **Well tolerated and immunogenic**
Functional Abs (opsonization, phagocytosis, killing, protecting)

Since the 1980's: GBS Vaccines, Challenges

GBS Protein-based vaccines

- **Antigen = common surface protein**
 - Cross protection against different CPS
 - Better immunogenicity
 - Humoral response T-cell dependant → Long lasting immunity

- **Among several candidates**
 - +/- ubiquitous among all GBS
 - BPS (Group B protective surface protein), C5a peptidase
 - **Sip** (Surface immunogenic protein)
Brodeur B et al, Infect Imm 2000
 - **Pili proteins** (*PI-1, PI-2a, PI-2b*)
Maione D et al, Science 2006

GBS Protein-based Vaccines

Reverse vaccinology approach
Knowledge of complete GBS genome

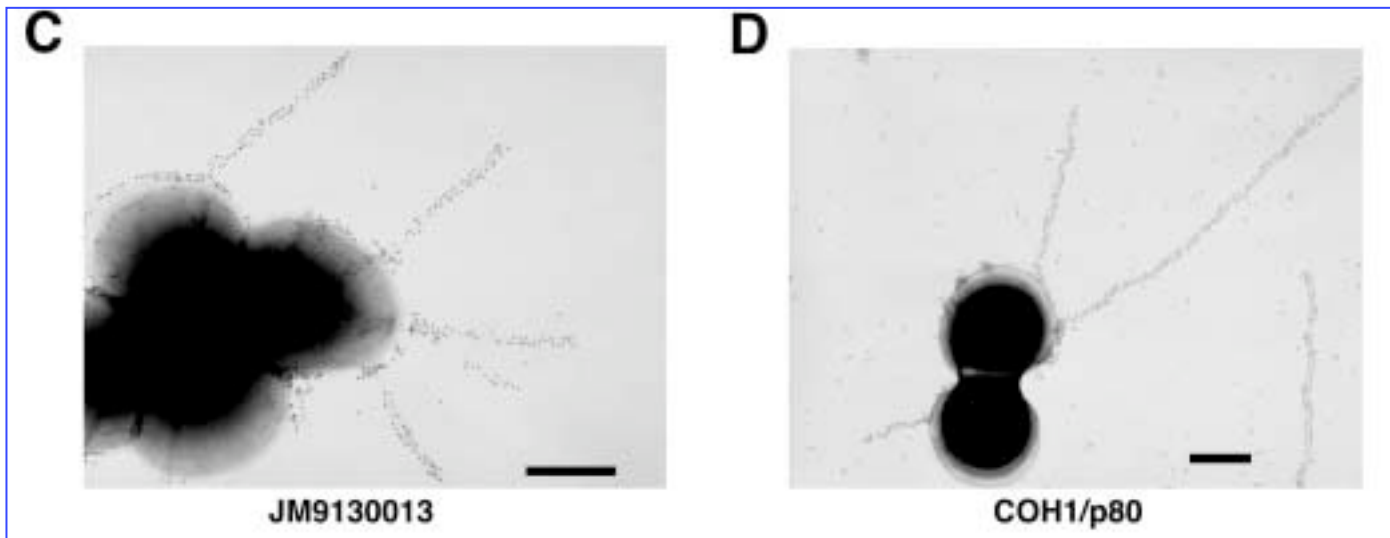
- **Comparaison of genomes from 8 different GBS serotypes**

D.Maione et al, Science 2006

- **312 surface proteins were cloned**
- **4 Provide a high protective humoral response in mouse**
 - **Sip**
 - **Three other proteins = « pilus like structures »**

GBS « pilus like structure »

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





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Vaccine Against Neonatal Infections

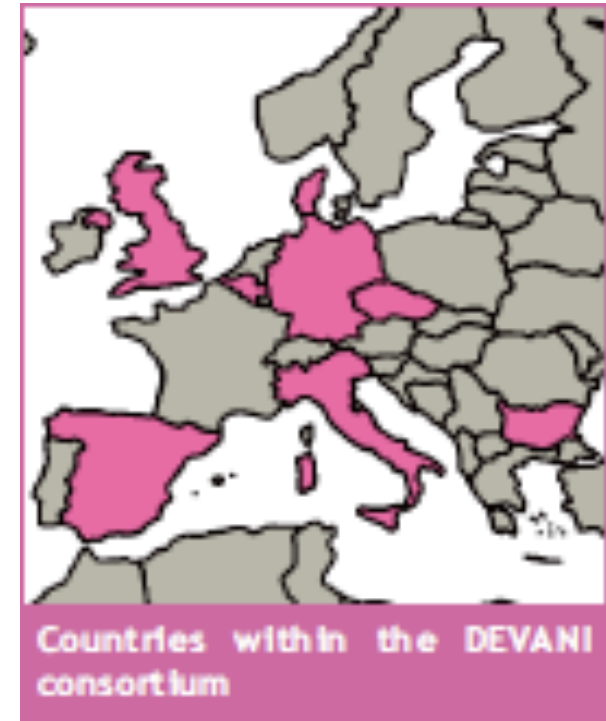
Design of a vaccine to immunize neonates against GBS infections through a durable maternal immune response



PROJECT (01.2008 - 06.2011)

- **Development of a vaccine against pili proteins & major CPS serotypes**
- **Development of a mouse model of GBS meningitis**
- **European epidemiology**
 - **Genito-rectal colonizing strains**
 - **Invasive neonatal strains and diseases**
- **Identification of protective levels of specific antibodies**

Consortium of 8 European countries



Material and methods (Targets)

- **200 GBS neonatal diseases (EOD & LOD)**
 - Strain isolated from blood, CSF or another normal sterile site and perinatal mother's serum
 - 25 per country
- **400 GBS negative mothers of healthy babies**
 - Serum
 - 50 per country
- **800 GBS positive mothers of healthy babies**
 - Strain and perinatal mother's serum
 - 100 per country

For each patient included in the study (2009-2010)

Case Report Form (eplatform web.database)

Signed consent form

Epidemiology

Material and methods

- **Determination of capsular type**
 - Serotyping by latex microagglutination (SSI, Dk)
 - Genotyping by multiplex PCR (*Poyart C, 2007 and Kong F, 2008 or Imperi M, 2010*)
 - Set up of an international EQA (*Afshar et al, JCM 2011*)
- **Assessment of presence of pili genes**
 - PCR PI-1, PI-2a and PI-2b (*Baldassari L et al, submitted*)
- **MLST** (*Jones N. et al., JCM 2003*)
- **FACS analysis**
 - *Pili expression*
- **GBS serology**
 - *Abs Ia, Ib, III and V*
 - *Abs PI-1, PI-2a and PI-2b*

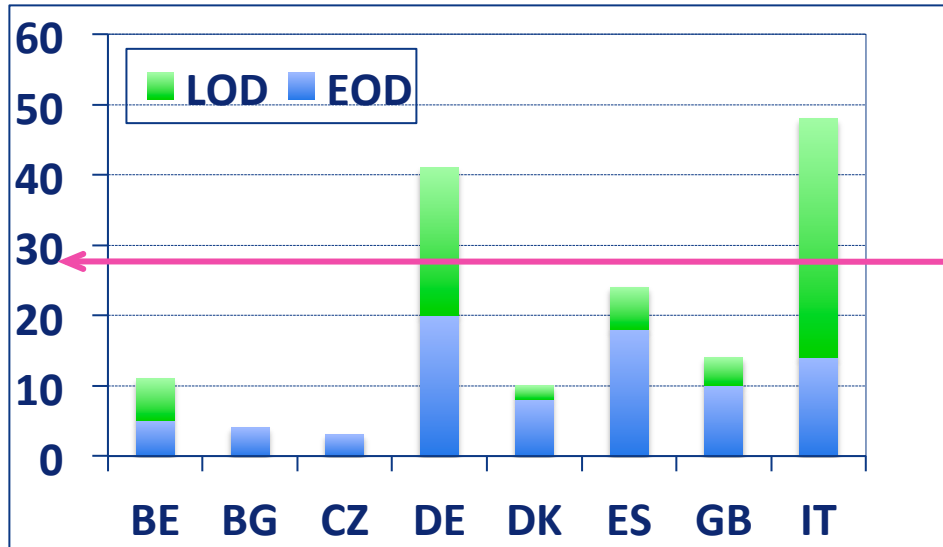
Descriptive and statistical analysis

- **Description and comparison of populations**
 - **Demographic - anamnestic – clinical – biological data – CPS - Pili - MLST**
 - Europe and countries
 - Pregnant women of healthy babies vs mothers of EOD/ LOD
 - Neonatal cases: EOD and LOD

- **CPS – Pili – MLST relations**

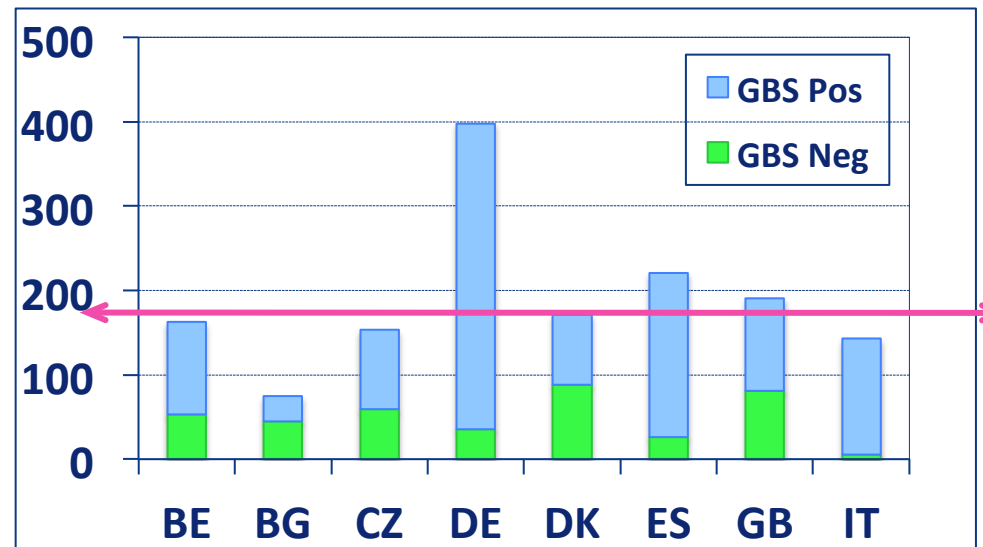
- *Serological relations*
 - *Protective thresholds*

Subject accountability



159 GBS neonatal infections
EOD / LOD = 1.12

1525 healthy infant's mothers
1122 GBS Pos
7 controls / NI case



PROVISIONAL ANALYSIS

“Pregnant women”

	Healthy babies' mothers (1525: 1122 pos)	GBS EOD's mothers (78)	P value
GBS prenatal screening			
% (Pos)	89.5%, 1365 (954)	47.4% (48.6%)	
Vagino-rectal swab	80%	33.3%	
IAP if GBS pos	60%	27%	
GBS intrapartum screening			
% (Pos)	16.7% (58%)	16.5% (92.3%)	
Maternal age at delivery			
Mean (years)	30.8 (15-48)	35.9 (26-40)	
Notified Risk Factor for neonatal GBS EOD			
ROM > 18h	5%	17.9 %	<0.001
T° >= 38°C	1%	11.5%	<0.001
GBS bacteriuria	3.9%	11.4%	0.02
Previous GBS sibling	0.3%	1.3%	
No RF	88.7%	51.3%	<0.001

“Pregnant women”

Type of delivery

	Healthy babies' mothers	GBS EOD's mothers
Vaginal	51.9%	68.1%
Planned C-section	12.9%	3.8%
Non-elective C-section	11.0%	27.8% (P<0.01)
Unknown	24.3%	1.3%

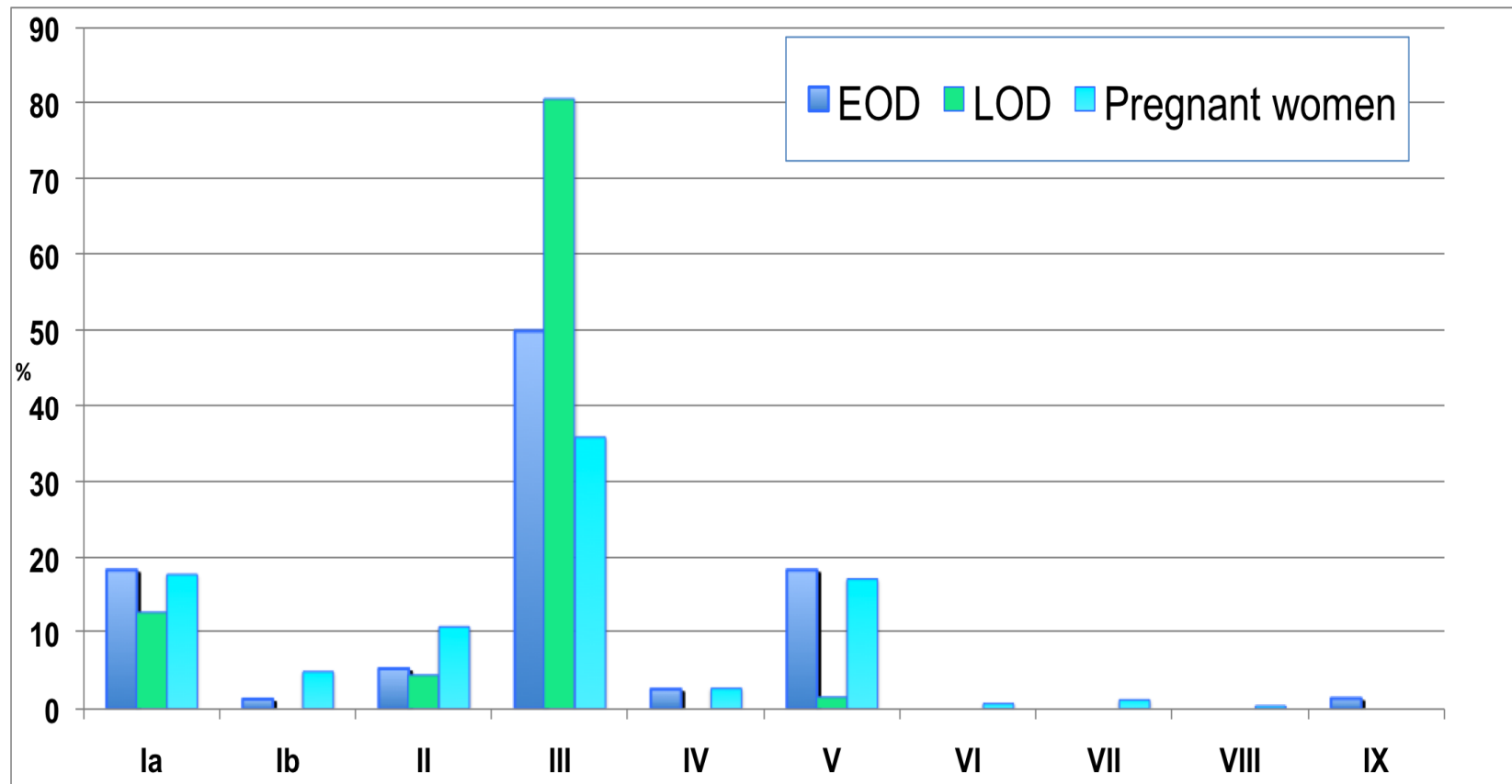
Mothers of newborns with GBS disease

	GBS EOD's mothers (78)	GBS LOD's mothers (72)
GBS prenatal screening		
% (Pos)	47.4% (48.6%)	61.1% (45.5%)
Vagino-rectal swab	33.3%	56.8%
IAP if GBS pos	27%	26%
GBS intrapartum screening		
% (Pos)	16.5% (92.3%)	14.1% (60%)
Maternal age at delivery		
Mean (years)	35.9 (26-40)	31.2 (20-44)
Notified Risk Factor for neonatal GBS EOD		
ROM > 18h	17.9%	8.6%
T° >= 38°C	11.5%	1.4%
GBS bacteriuria	11.4%	8.3%
Previous GBS sibling	1.3%	0%
No RF	51.3%	52.1%

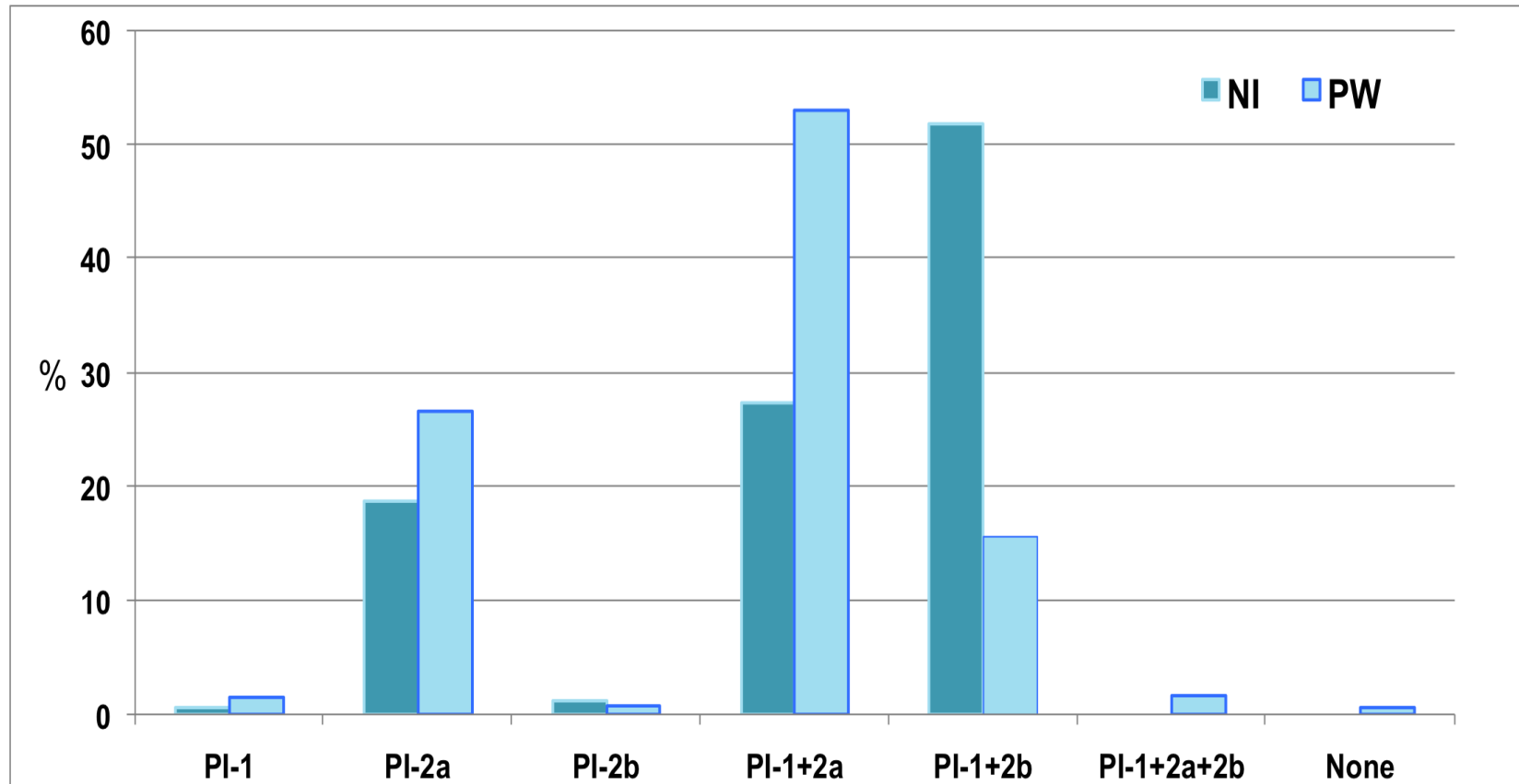
Neonatal Invasive GBS Diseases

	GBS EOD (5.1% death)	GBS LOD (1.5% death)	P
Number (%)	78 (52)	72 (48)	
Age at onset	< 1 day (88%)	38 d (6-109)	
Birth weight	2.9 kg (1-4.9!)	2.7 kg (0.7-4.1)	
Gestational age < 37 weeks	37.7 wks (26-42) 21.5%	36.2 wks (24-43) 35.7%	0.05
Sex M/F	1.16	0.89	0.42
Predominant manifestation at onset	Respiratory distress (38% of cases)	Fever (63% of cases)	<0.001
Type of infection			
-Bacteremia without focus	26.8%	11%	
-Sepsis/Shock	70.7%	75.3%	
-Meningitis	8.5%	30.1%	<0.001
-Pneumonia	13.4%	2.7%	0.017
-Others	2.4%	9.6%	
-Birth in Twins	5.1%	15.7%	0.03

Distribution of CPS serotypes among GBS from neonatal infections and among healthy babies' mothers



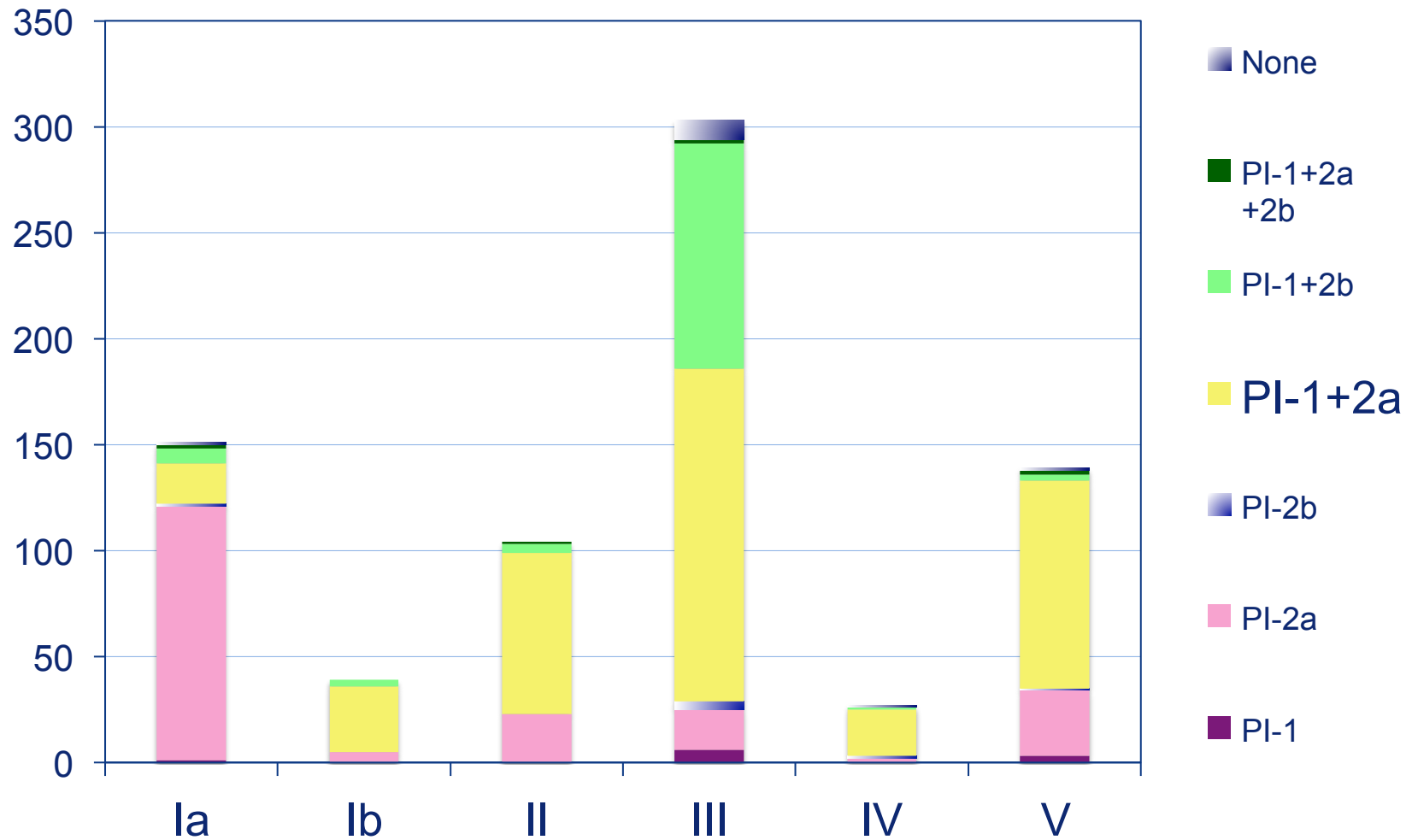
Distribution of Pili genes among GBS from neonatal infections and among healthy babies' mothers



NI: 100% with pili gene(s), most common pattern is PI-1+b2

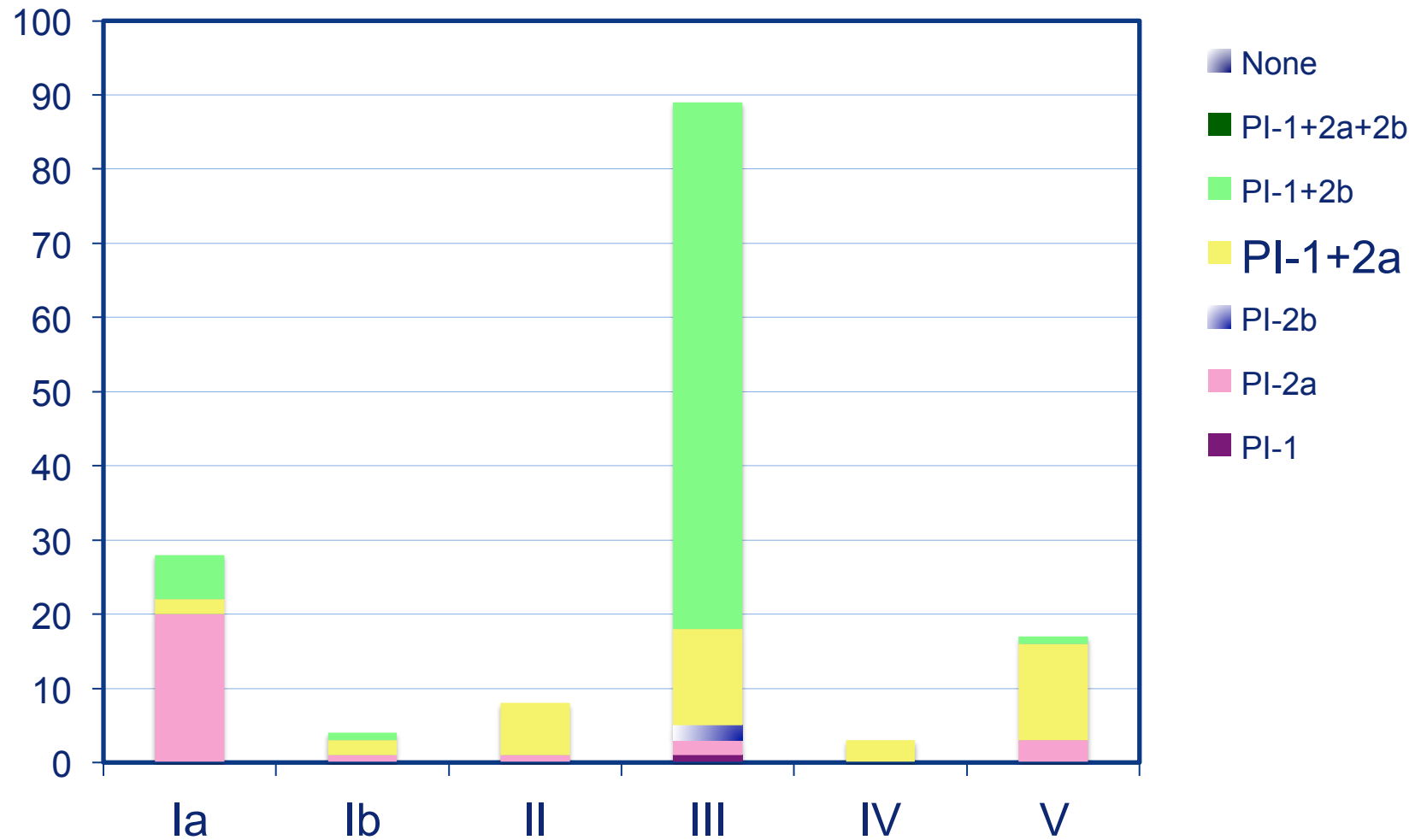
PW: 0.6% without pili genes, most common is PI-1+2a

Relation pili / CPS among GBS from PW



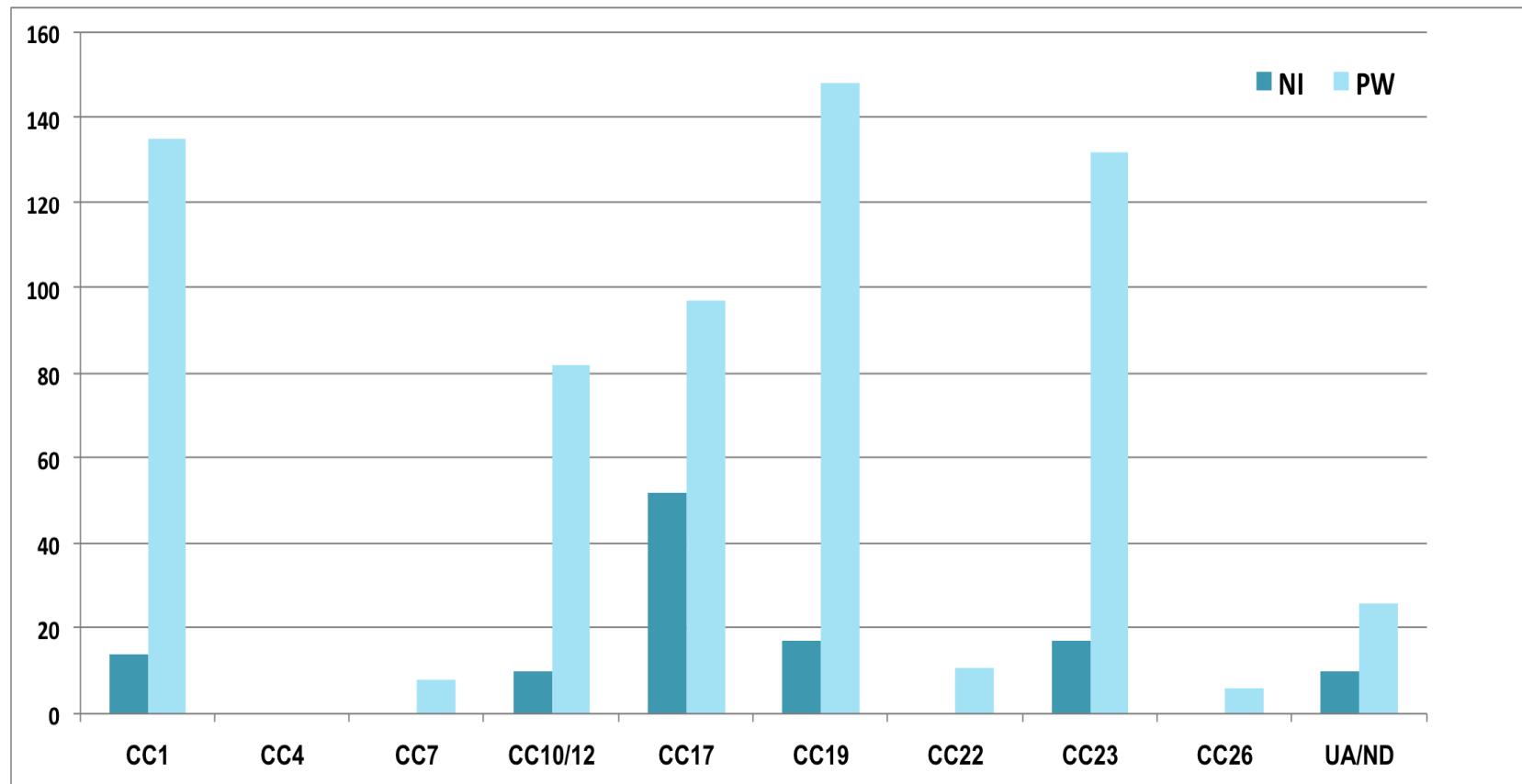
Association between certain serotypes and pili gene pattern

Relation pili / CPS among GBS from newborns



MLST – Clonal analysis of GBS

646 GBS from PW and 121 from NI



In PW: 66 Sequence types (ST) for 9 clonal complexes (CC)

Five CC include 92% of isolates tested

In NI: 6 CC; the most frequent is CC17, the hypervirulent clone

DEVANI Project

Preliminary conclusions

- **Set up of a mouse meningitis model**
- **In European countries**
 - **Difference of prevention strategies**
 - **Difference of resource for routine diagnostic of severe neonatal infection**
- **In Belgium, difficult to include cases even if they occurred**
- **Standardization of typing methods**
- **Among neonatal infections:**
 - **Higher prevalence of GBS CPS III, pili pattern PI-1+2b and CC17**
- **Assessment of presence of pili genes**
 - **100% in NI et 99% in PW**
- **MLST et CPS more heterogenous among GBS from PW**
- **No significant difference in CC distribution /country**
- **Serological analysis ongoing**



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