CMV in pregnancy
- how much testing is enough

Bill Rawlinson
w.rawlinson@unsw.edu.au

"Babies are such a nice way to start people." ~Don Herrold

1. Why is CMV important?
   - Epidemiology
   - Potential outcomes of infection
   - Delayed consequences of infection
2. What is CMV infection in pregnancy?
   - CMV as a herpesvirus
   - The placenta as a barrier to infection
   - Diagnostic testing
3. What issues arise
   - Certainty of diagnosis
   - Certainty of outcome
4. What are the outcomes?
   - Therapies
   - Future pregnancies

What is the problem?

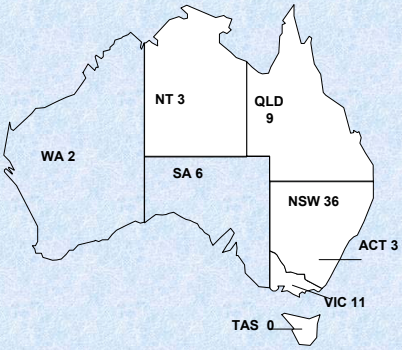
- Congenital CMV (cCMV) is the most common viral cause of congenital malformation in Australia

Primary Modes of CMV Transmission

- **In fetus, perinates, neonates:** Exposure to maternal blood (in utero), cervix, breast milk
- **In children:** Close contact as in day care centers
- **In adults:** Sexual transmission, close contact

Estimated annual incidence of congenital CMV infection & disease in Aust & NSW

Category	Est figure in Australia	Est figure in NSW
Live births	~246,000	86,5000
Primary maternal infection rate (1.5%)	3,690	1,298
CMV infected infants (40%)	1,476	519
Symptomatic infection at birth (15%)	221	78
Those with fatal infection (30%)	66	23
Those with severe sequelae (70%)	155	55
Asymptomatic infection at birth (85%)	1,255	441
Those with later sequelae (15%)	188	66
Total with CMV sequelae	409	144



What is the current status?

- Transmission of cCMV
  - vertically
  - *In utero* infection in 0.2-2.2%
  - 200-800 children born each year in Australia
- Significant numbers of the 250,000 women with concern regarding the risk
- CMV serology is not part of routine antenatal screening

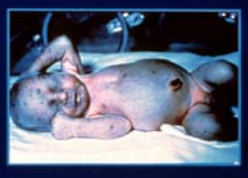
1. Why is CMV important?
   - Epidemiology
   - Potential outcomes of infection
   - Delayed consequences of infection
2. What is CMV infection in pregnancy?
   - CMV as a herpesvirus
   - The placenta as a barrier to infection
   - Diagnostic testing
3. What issues arise
   - Certainty of diagnosis
   - Certainty of outcome
4. What are the outcomes?
   - Therapies
   - Future pregnancies

What is the problem?

- 90% of babies with cCMV will be clinically normal
- Pregnant women cannot be adequately retrospectively diagnosed using current tests

Congenital CMV Infection

- Hepatosplenomegaly
- Jaundice
- Microcephaly
- Prematurity
- Chorioretinitis
- Petechiae
- Mental retardation
- Hearing loss

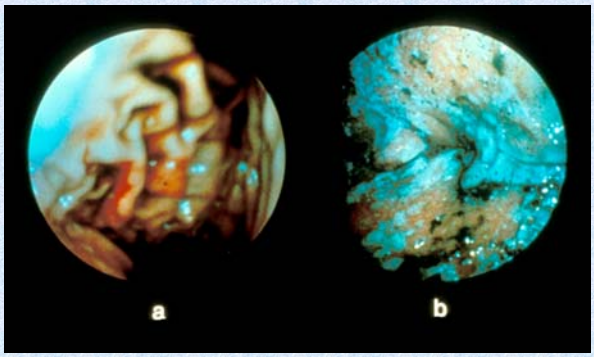


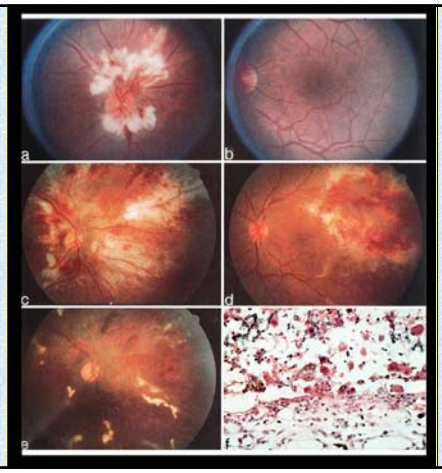
cCMV Aust 1999 - 2002

Clinical symptoms Neurological abnormality	No. affected	% of affected children
Microcephaly	14	23
Intracranial calcification	13	21
Seizures	8	13
Sensorineural hearing loss	11	18
Chorioretinitis	6	10
Developmental delay	14	23
Total affected infants	**27/42**	**44**

cCMV Aust 1999 - 2002

Clinical symptoms	No. affected	
Abnormality		
Neonatal death	1	2
Maternal symptoms		
Maternal illness	34	55





1. Why is CMV important?
   - Epidemiology
   - Potential outcomes of infection
   - Delayed consequences of infection
2. What is CMV infection in pregnancy?
   - CMV as a herpesvirus
   - The placenta as a barrier to infection
   - Diagnostic testing
3. What issues arise
   - Certainty of diagnosis
   - Certainty of outcome
4. What are the outcomes?
   - Therapies
   - Future pregnancies

Delayed consequences of infection

- Later
  - sensorineural hearing loss (<5yr)
  - neurodevelopmental delay (<5yr)
- Eye disease
- Immunological effects
- Latency

CLINICAL - Late complications

- Rejection
- Bacteraemia
- Fungal infection - aspergillosis
- Vascular effects - atherosclerosis

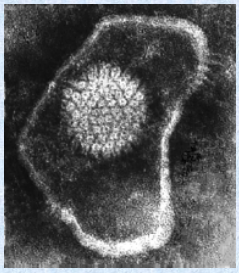
[Nicols 2001]

CMV in Neonates

- Transfusion - seropositive blood
  10/74 CMV infection
  5/74 CMV infection
  - seronegative blood
  0/90 CMV infection
- Higher risk - weight < 1250 g
  - BTF amount > 50 mL
  - mother seronegative
  [Yeager, 1974]
  - prematurity
- Premature seroconversion 1%-10%
  [Galea, 1993]

1. Why is CMV important?
   - Epidemiology
   - Potential outcomes of infection
   - Delayed consequences of infection
2. What is CMV infection in pregnancy?
   - CMV as a herpesvirus
   - The placenta as a barrier to infection
   - Diagnostic testing
3. What issues arise
   - Certainty of diagnosis
   - Certainty of outcome
4. What are the outcomes?
   - Therapies
   - Future pregnancies

Herpesvirus EM



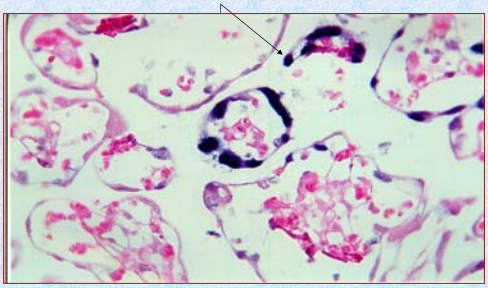
(Linda Stannard, University of Cape Town, S.A.)

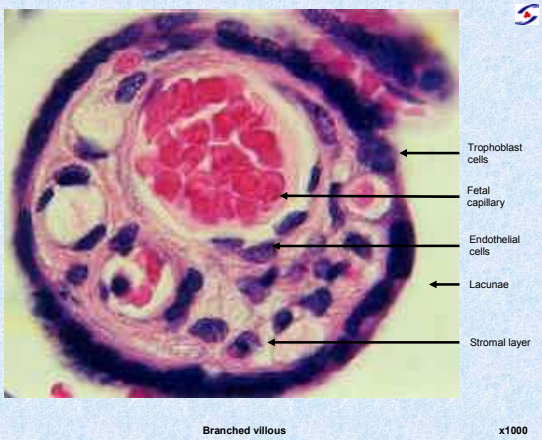
Virus Name	Other Name	Sub-family	Typical illness
HHV1	HSV 1	α	Cold sores
HHV2	HSV 2	α	Genital ulcers
HHV3	VZV	α	Chickenpox, Shingles
HHV4	EBV	β	Glandular fever, Nasopharyngeal carcinoma
HHV5	HCMV	β	Mononucleosis, Pneumonia (immunocompromised)
HHV6	HHV 6	β	Exanthem subitum in children
HHV7	HHV 7	?β	Unknown
HHV8	HHV 8	γ–2	Kaposi's sarcoma

1. Why is CMV important?
   - Epidemiology
   - Potential outcomes of infection
   - Delayed consequences of infection
2. What is CMV infection in pregnancy?
   - CMV as a herpesvirus
   - The placenta as a barrier to infection
   - Diagnostic testing
3. What issues arise
   - Certainty of diagnosis
   - Certainty of outcome
4. What are the outcomes?
   - Therapies
   - Future pregnancies

CMV Vertical Transmission

- Location





CMV in placenta

- 12% (11/94)
- Cell types +++ trophoblasts (m)
  + stromal fibroblasts (m)
  +/- endothelial cells (f)

CMV in placenta

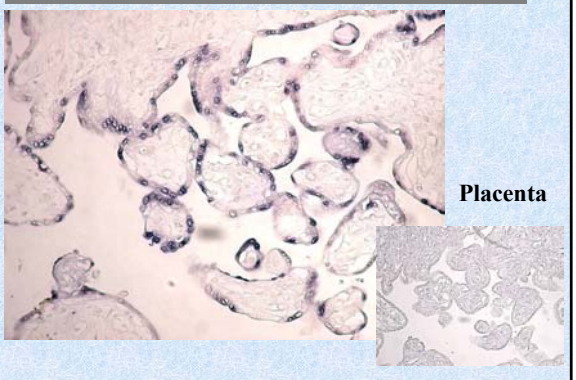
- Placental infection
  - reactivation
  - higher viral load
  - timing (T1 - T2 - T3)
  - transient maternal leucocytes
  - DC

CMV in placenta

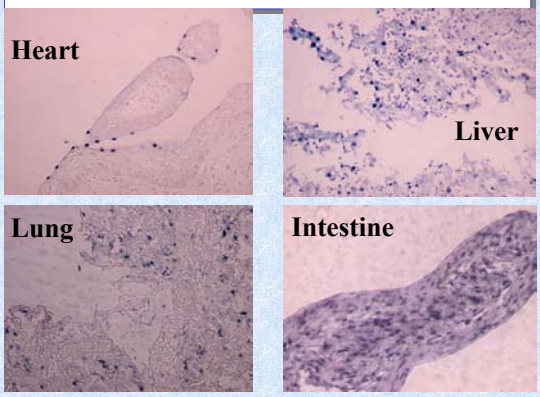
- Transmission to placenta
  - spermatozoum / ovum (undifferentiated, receptors)
  - cervix
  - maternal leucocytes
- Trophoblasts
  - epithelial
  - ↓ HLA II
  - Vimentin neg
  - Annexin II pos

[Mathys, 1991; Witz, 1999; Trincado, 2002]

Placenta - IS-PCR



Stillbirth - IS-PCR

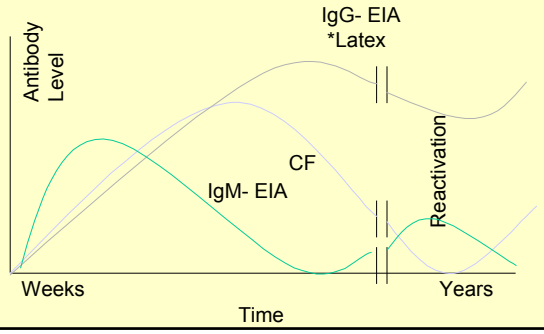


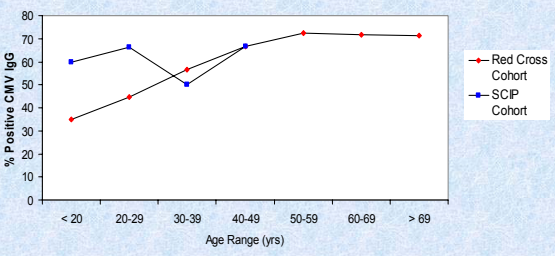
1. Why is CMV important?
   - Epidemiology
   - Potential outcomes of infection
   - Delayed consequences of infection
2. What is CMV infection in pregnancy?
   - CMV as a herpesvirus
   - The placenta as a barrier to infection
   - Diagnostic testing
3. What issues arise
   - Certainty of diagnosis
   - Certainty of outcome
4. What are the outcomes?
   - Therapies
   - Future pregnancies

Congenital Diagnosis

- Clinical suspicion
  - IgM seropositive
  - IgG seropositive
  - IgG avidity
  - Maternal urine culture
  - Maternal blood NAT
  - Affected child
- Amniocentesis
  - NAT
  - Q-NAT
  - Culture
- Affected child
  - Usually after 4 wks age
  - Dried blood spot

CMV Antibody

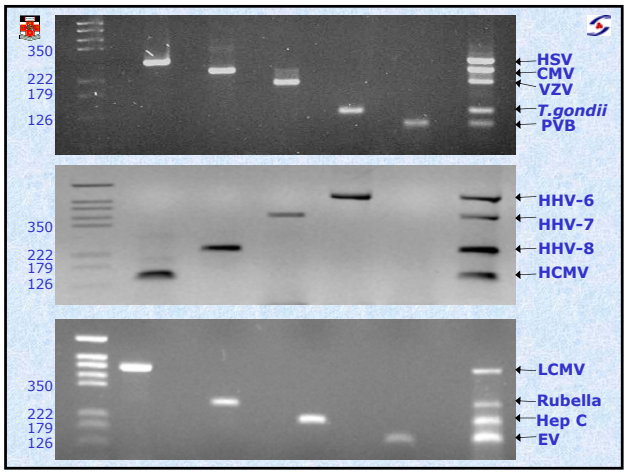


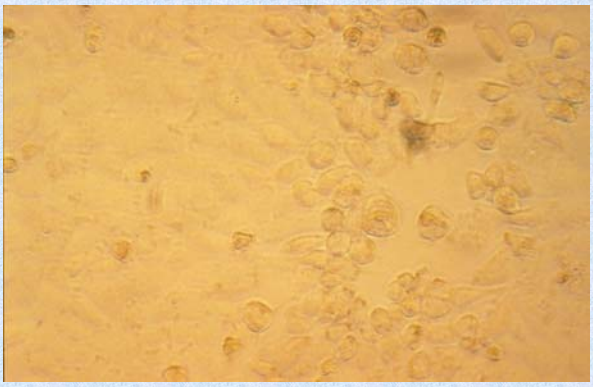


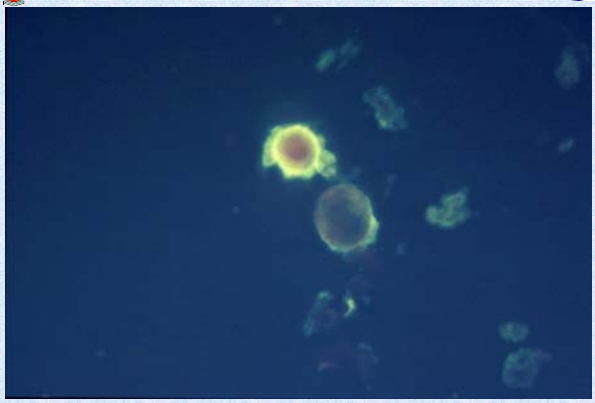
Multiplex PCR

- Simultaneous detection of pathogens without the need for guidance by clinical manifestations
- High sensitivity and specificity
- Rapid
- Smaller sample size required

DNA1	DNA2	RNA	PRO1
CMV, HSV1	HHV6, HHV7	Rubella, EV	GpB Streptococcus
VZV, toxoplasma	Adenovirus, AAV	HCV, LCMV	Ureaplasma, mycoplasma
Parvovirus B19	Mouse mammary tumour virus	Influenza	*C. trachomatis*

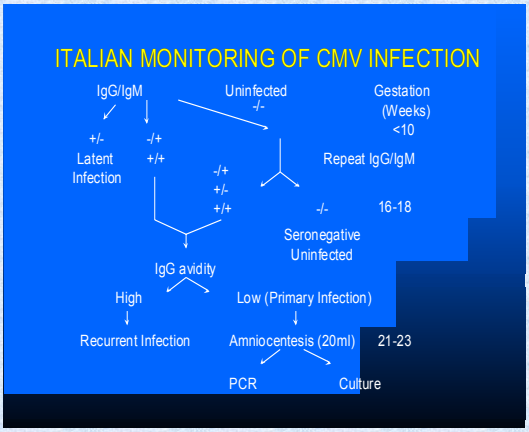


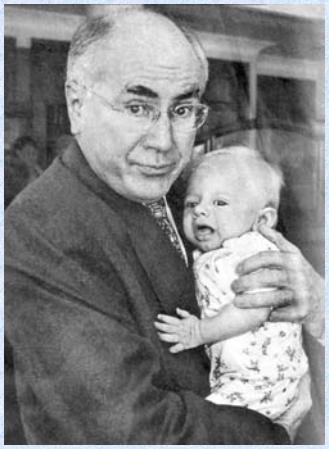




Improved Diagnostics

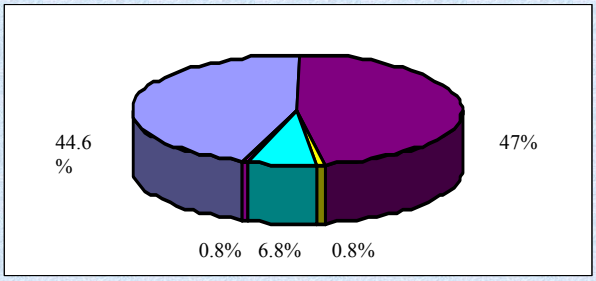
- NAD – commercial
- Quantitation
- Genotyping
- Better reference standards
- Better interpretation
- Peripheral measures to replace amniocentesis





1. Why is CMV important?
   - Epidemiology
   - Potential outcomes of infection
   - Delayed consequences of infection
2. What is CMV infection in pregnancy?
   - CMV as a herpesvirus
   - The placenta as a barrier to infection
   - Diagnostic testing
3. What issues arise
   - Certainty of diagnosis
   - Certainty of outcome
4. What are the outcomes?
   - Therapies
   - Future pregnancies

SCIP serostatus
-/- +/- +/+/+ +/+/-



Serology 600 pregnant women 2002-2004

CMV IgG	CMV IgM	CMV IgG Avidity	≤20 wks gestation	>20 wks gestation	Total
-	-	ND	169	90	259
+	-	ND	202	106	308
+	+	Low	5	2	7
+	+	High	20	6	26

Low avidity <35%

CMV Antibody

	IgG	Avidity	IgM	CF
Congenital	+	+/-	+	+
Primary infection	+/-	+	+	+
Secondary infection	+	-	+/-	+

1. Why is CMV important?
   - Epidemiology
   - Potential outcomes of infection
   - Delayed consequences of infection
2. What is CMV infection in pregnancy?
   - CMV as a herpesvirus
   - The placenta as a barrier to infection
   - Diagnostic testing
3. What issues arise
   - Certainty of diagnosis
   - Certainty of outcome
4. What are the outcomes?
   - Therapies
   - Future pregnancies

Cytomegalovirus

0.5 - 2.2% primary infection in pregnancy

↓

10 - 30% placental transmission to fetus

↓

30% symptomatic - *CID, retardation, blindness etc.*

18% long-term sequelae - *hearing loss, learning difficulties*

Quantitation of CMV

- **Benefits**
  - Surrogate measure of resistance
  - Better correlation with disease
  - Measure of viral load in blood vs other tissue
  - Association with prognosis in some diseases
  - Simplified sample
- Problems
  - Cost
  - Lack of correlation with some disease
  - Lower sensitivity that qualitative
  - Availability
  - Sample size for testing

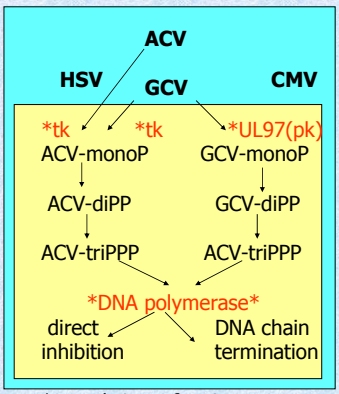
1. Why is CMV important?
   - Epidemiology
   - Potential outcomes of infection
   - Delayed consequences of infection
2. What is CMV infection in pregnancy?
   - CMV as a herpesvirus
   - The placenta as a barrier to infection
   - Diagnostic testing
3. What issues arise
   - Certainty of diagnosis
   - Certainty of outcome
4. What are the outcomes?
   - Therapies
   - Future pregnancies

Untreated Outcomes

- Asymptomatic 90%
- Symptomatic
- Issues
  - Infertility
  - Developmental delay up to 5 yrs

Antiviral Agents

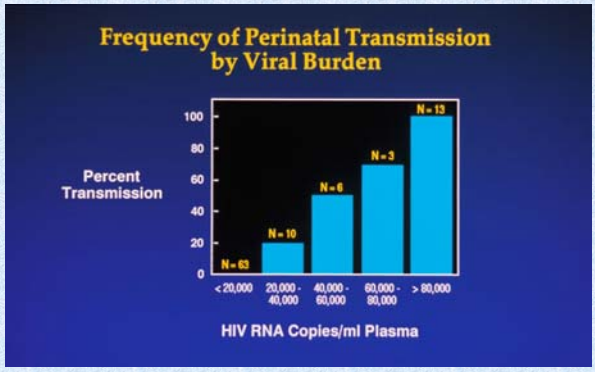
Antiviral agent	Analogue of	Mode of action	Virus
Aciclovir (ACV)	nucleoside	Inhibits DNA pol DNA chain termination	HSV VZV
Ganciclovir (GCV)	nucleoside	"	CMV HSV VZV
Foscarnet (PFA)	PPi	Inhibits DNA polymerase	CMV HSV VZV



* usual sites of resistance

Principles

- Virus intimately related to the cell
- Development
  - Combinatorial chemistry
  - Fortuitous
  - Specific agents for specific virus
- Acceptable agents
  - Target essential process
  - low toxicity



1. Why is CMV important?
   - Epidemiology
   - Potential outcomes of infection
   - Delayed consequences of infection
2. What is CMV infection in pregnancy?
   - CMV as a herpesvirus
   - The placenta as a barrier to infection
   - Diagnostic testing
3. What issues arise
   - Certainty of diagnosis
   - Certainty of outcome
4. What are the outcomes?
   - Therapies
   - Future pregnancies

Issues

- Should all pregnant women be screened for CMV?
- How best to monitor the affected child
- What is the best form of CMV treatment for congenital CMV
  - Prophylaxis?
  - Combination therapy?
- When will CMV vaccines be available?

Some solutions tried

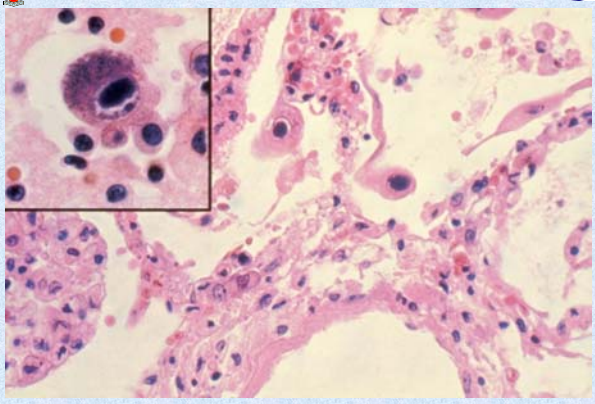
- D/R matching
- Blood transfusion screening
- Hyperimmune Ig
- ACV prophylaxis
- VCV prophylaxis
- GCV prophylaxis
- GCV pre-emptive therapy
- VGCV prophylaxis and therapy
- Combined therapy

Pathogen	Symptoms
Varicella-Zoster virus	Limb hypoplasia
Herpes simplex virus	Neurodevelopmental disability
Parvovirus B19	Hydrops fetalis
Toxoplasma gondii	Similar to CMV
Hepatitis C	Chronic hepatitis
Enterovirus	Fetal death, ?diabetes
Lymphocytic choriomeningitis virus	Similar to CMV
Human herpesvirus type 6	
Human herpesvirus type 7	
Human herpesvirus type 8	

Prevention of post-transfusion CMV - leucoreduction or screening

- Australian high risk pts
  - BMT
  - stem cell tx
  - newly diagnosed leukaemics
  - pts likely to proceed to BMT
  - CMV seronegative allograft recipients
  - neonates
  - HIV seropositive (unproven)

[Vox Sanguinis 2002]



Every day in Australia at least one child is born with congenital CMV infection, every month, two children are born with perinatal HSV disease, it is likely there are >1000 more children born every year with undiagnosed congenital or perinatal viral infection