Clinical Sensitivity of Saliva and Dried Blood Spot PCR and Infant Outcomes from a Congenital CMV Newborn Screening Study: 2016-2022

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Congenital CMV Public Health and Policy Conference

Salt Lake City, Utah

Tuesday, 10 October 2023

11:15 -11:40 AM

Disclosures

- Financial Support
  - Centers for Disease Control and Prevention (CDC)
  - National Vaccine Program Office
  - Minnesota Department of Health Newborn Screening Program
  - University of South Carolina's Disability Research and Dissemination Center (DRDC) through its Cooperative Agreement (6U19DD001218) with CDC
  - IDSA "G.E.R.M." Award (RK)
- UMN receives research support from Moderna vaccine
- I will not discuss off-label use of medications

Should We Include cCMV in the Recommended Uniform Screening Panel (RUSP)

YES

- Diagnostic evaluation
- Anticipatory monitoring
- Hearing loss may be delayed, progressive in nature
- Antiviral therapy
- Neurodevelopmental evaluation

NO

- Most infants asymptomatic
- Overuse of antivirals
- Undue parental anxiety
- Cost issues (though is cost-effective)
- Ethical concerns
- Does not fit RUSP paradigm

Cannon, PMID 24760655
Griffiths, PMID 31237046
Ronchi, PMID 28277819, 31575999
Fowler, PMID 28049114
Ganti, PMID 27723885
Gievers, PMID 32591436

Universal Congenital CMV Screening

- Importance of timing of specimen acquisition
- **WHAT** to Use for Screening?
  - Dried blood spots (DBS)
  - Urine
  - Saliva
- CHIMES study
  - DBS PCR **insufficient sensitivity** (20,448 subjects; range 28-34%)
  - Saliva PCR demonstrated **high sensitivity** (34,989 subjects; 97.4-100%)
- Saliva-based PCR has been focal point of policy discussions in newborns
  - False positives
  - Cost

- Universal Screening
- Targeted Screening
- Enhanced Targeted Screening

Haller, PMID 32361556
Yamada, PMID 32273174
Pellegrinelli, PMID 32164599

Boppana, PMID 20388893
Gantt, PMID 27723885
Gievers, PMID 32591436

Reconsideration of DBS PCR-based Detection

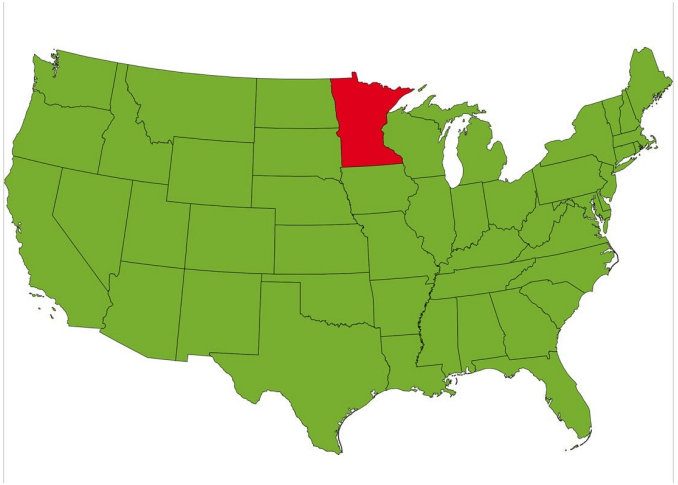
- Concerns raised over perceived risk of false positive assays (*J Pediatr* 1962;61:610–616)
- Vigorous opposition from physicians and medical societies routine screening to be "socialized medicine" and an infringement on the private practice of medicine
- Consultants to the California State Department of Health concluded that the Guthrie assay "required further evaluation" and that knowledge needed to be "more complete" before screening could be justified (doi: 10.1001/jama.1964.03070250079042)
- In 1964, the House of Delegates of the American Medical Association voted to oppose any form of "legislation requiring compulsory testing" for phenylketonuria (doi: 10.1001/jama.1964.03070250079042)

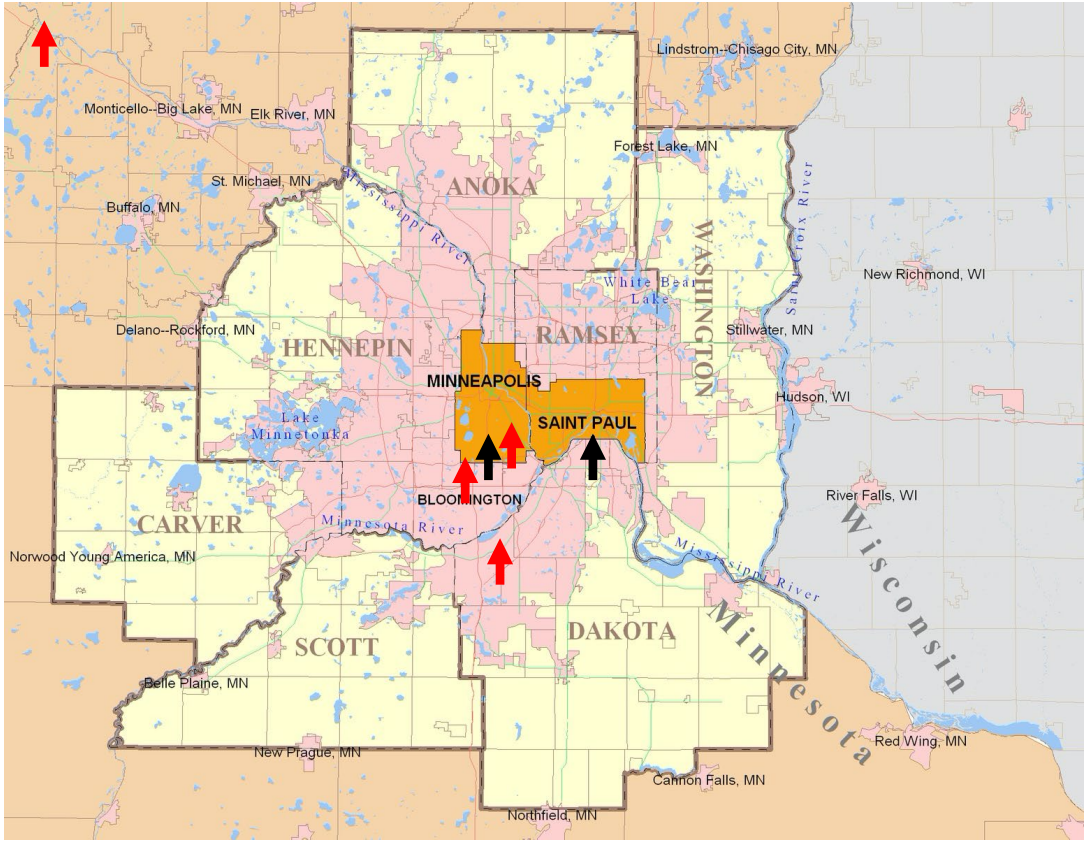
Improved Extraction and DNA Recovery

- Improved DNA recovery methodologies have been described in recent years
- Improved extraction buffers for DBS DNA recovery
- Increased sensitivity of PCR techniques/platforms

Hypothesis: *Do improved methodologies translate to better sensitivity of DBS PCR, supporting re-consideration of the DBS as a tool for universal screening for cCMV infection?*

Compare: *Analytic sensitivity of saliva and DBS for CMV detection for newborn screening in a prospective, unselected population-based study in Minnesota.*





JAMA Pediatrics | Original Investigation

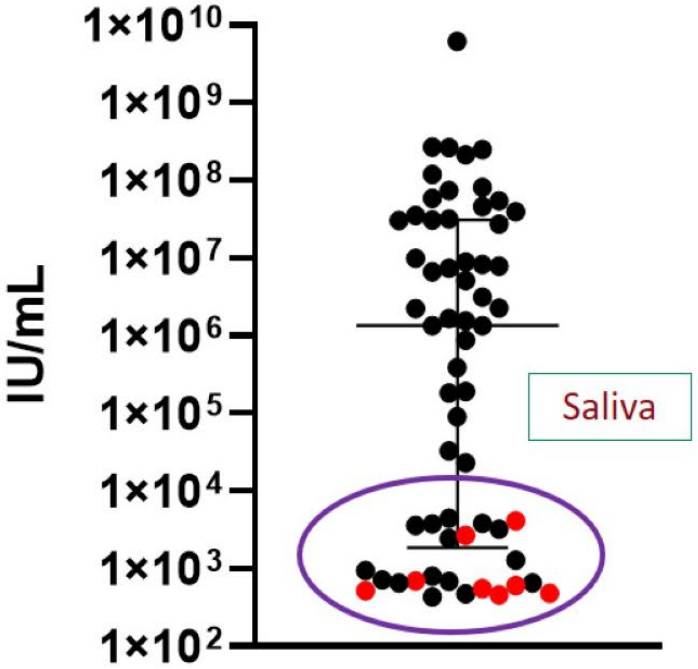
Sensitivity of Dried Blood Spot Testing for Detection of Congenital Cytomegalovirus Infection

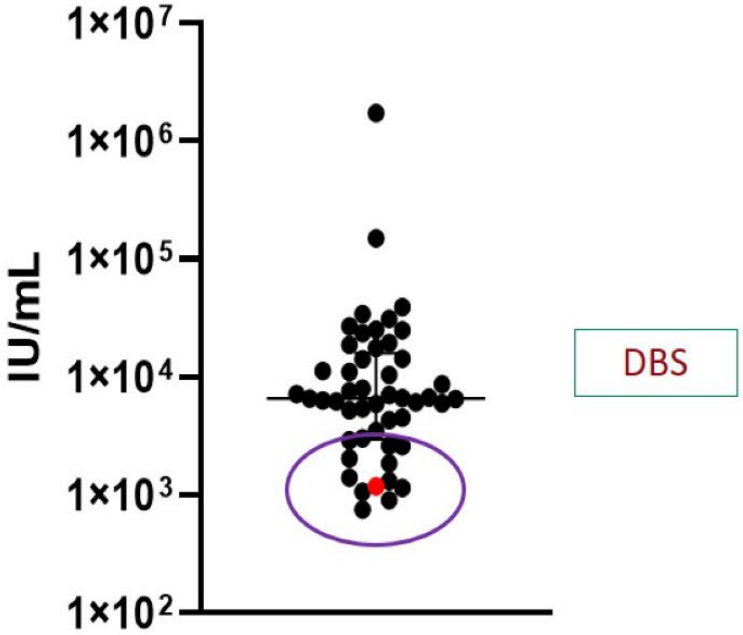
Sheila C. Dollard, PhD; Maggie Dreon, MS; Nelmary Hernandez-Alvarado, MS; Minal M. Amin, MPH; Phili Wong, MS; Tatiana M. Lanzieri, MD, MPH; Erin A. Osterholm, MD; Abbey Sidebottom, PhD; Sondra Rosendahl, MS; Mark T. McCann, BA; Mark R. Schleiss, MD

Table 2. Performance of DBS and Saliva Polymerase Chain Reaction Testing for Identifying Newborns with Congenital CMV Infection (N = 12 554)

	Saliva		DBS combined		DBS UMN		DBS CDC	
Congenital CMV infection[a]	Yes	No	Yes	No	Yes	No	Yes	No
Positive screen, No. (%)	52 (0.4)	8 (0.1)	48 (0.4)	1 (0)	41 (0.3)	0 (0)	43 (0.3)	1 (0)
Negative screen, No. (%)	4 (0)	12 490 (99.5)	8 (0.1)	12 497 (99.5)	15 (0.1)	12 498 (99.6)	13 (0.1)	12 497 (99.5)
Parameter, % (95% CI)	Saliva		DBS combined		DBS UMN		DBS CDC	
Sensitivity	92.9 (83.0-97.2)		85.7 (74.3-92.6)		73.2 (60.4-83.0)		76.8 (64.2-85.9)	
False negative	7.1 (2.8-17.0)		14.3 (7.4-25.7)		26.8 (17.0-39.6)		23.2 (14.1-35.8)	
Specificity	99.9 (99.9-100)		100.0 (100-100)		100.0 (100-100)		100.0 (100-100)	
PPV	86.7 (75.8-93.1)		98.0 (89.3-99.6)		100.0 (91.4-100)		97.7 (88.2-99.6)	
False positive	13.3 (6.9-24.2)		2.0 (0.4-10.7)		0.0 (0.0-8.6)		2.3 (0.4-11.8)	
NPV	100 (99.9-100)		99.9 (99.9-100)		99.9 (99.8-99.9)		99.9 (99.8-99.9)	

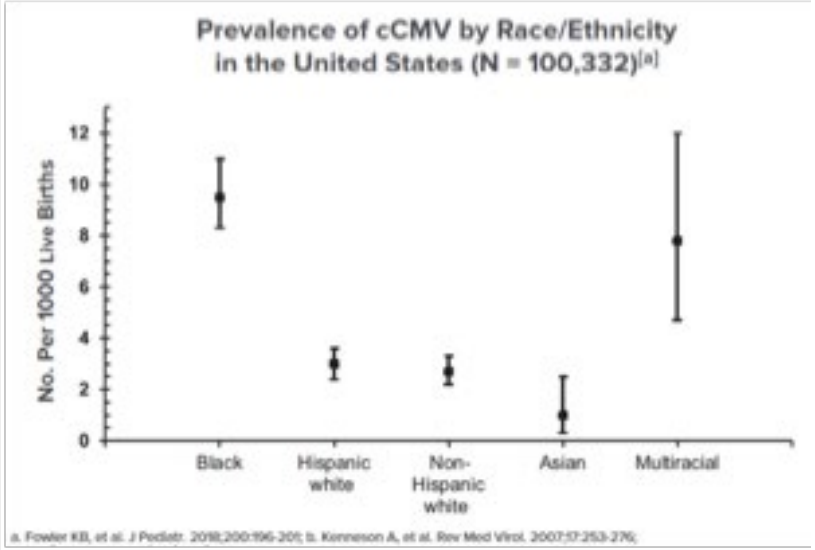
Viral Load Distribution in Saliva and DBS Positives





cCMV prevalence by nursery type and demographics

Characteristic	No. of newborns screened (%)	No. of newborns with cCMV (%)	cCMV prevalence per 1000 (95% CI)	Prevalence ratio (95% CI)
Hospital nursery				
Well baby	18 669 (94)	71 (93)	3.8 (3.0-4.8)	1 [Reference]
Neonatal intensive care	1107 (6)	5 (7)	4.5 (1.8-10.8)	1.2 (0.5-2.9)
Mother's age group, y				
≤24	1846 (9)	11 (14)	6.0 (3.3-10.7)	1.9 (0.9-4.2)
25-29	4432 (22)	14 (18)	3.2 (1.9-5.3)	1 [Reference]
30-34	8383 (42)	33 (43)	3.9 (2.8-5.5)	1.2 (0.7-2.3)
≥35	5255 (26)	18 (24)	3.4 (2.1-5.4)	1.1 (0.5-2.2)
Mother's race or ethnicity				
Hispanic[c]	1678 (9)	1 (1)	0.6 (0.1-4.2)	0.1 (0.02-0.9)
Non-Hispanic				
Black	1764 (9)	9 (12)	5.1 (2.7-9.8)	1 [Reference]
White	13 600 (73)	62 (82)	4.6 (3.6-5.8)	0.9 (0.4-1.8)
Other	1676 (9)	4 (5)	2.4 (0.9-6.4)	[illegible]
Birth order				
First	8447 (46)	27 (36)	3.2 (2.2-4.7)	1 [Reference]
Second	6316 (34)	38 (50)	6.0 (4.4-8.3)	1.9 (1.2-3.1)
Third or higher	3755 (20)	11 (14)	2.9 (1.6-5.3)	0.9 (0.5-1.8)



a. Fowler KB, et al. J Pediatr. 2018;200:196-201; b. Kenneson A, et al. Rev Med Virol. 2007;17:253-276;

Research Letter | Pediatrics

September 2, 2022

Assessment of Congenital Cytomegalovirus Prevalence Among Newborns in Minnesota During the COVID-19 Pandemic

Mark R. Schleiss, MD[1]; Sondra Rosendahl, MS[2]; Mark McCann, BA[2]; et al

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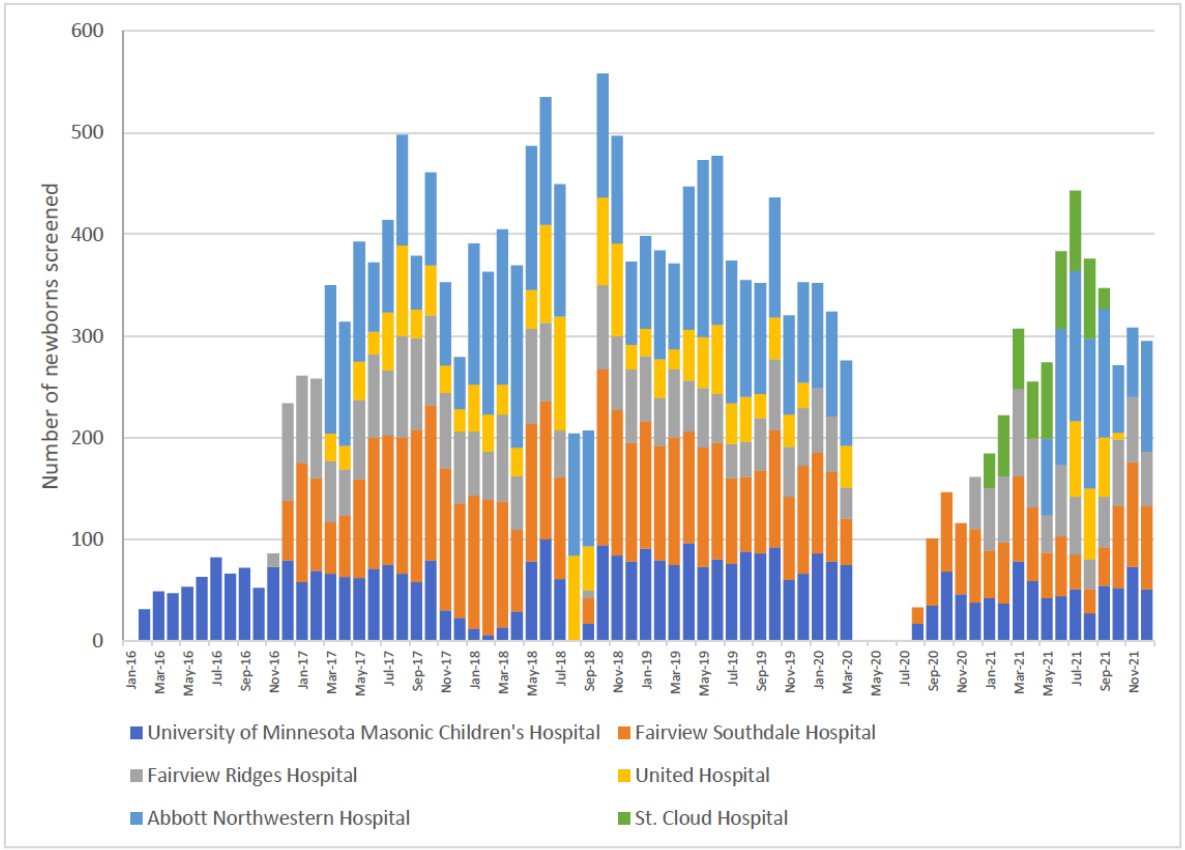
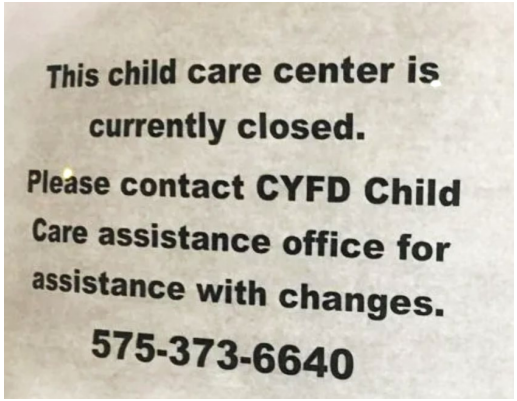
JAMA Netw Open. 2022;5(9):e2230020. doi:10.1001/jamanetworkopen.2022.30020





https://jamanetwork.com/journals/jamanetworkopen/fullarticle/2795877

Characteristic	No. of newborns screened (%)	No. of newborns with cCMV (%)	cCMV prevalence per 1000 (95% CI)	Prevalence ratio (95% CI)
Overall	19 919 (100)	76 (100)	3.8 (3.0-4.8)	NA
Study period[b]				
Prepandemic	15 697 (79)	70 (92)	4.5 (3.5-5.6)	1 [Reference]
Pandemic	4222 (21)	6 (8)	1.4 (0.6-3.2)	0.3 (0.1-0.7)

A total of 15,697 were screened during the pre-pandemic period (2/2016–3/2020, **70 cCMV cases identified**; 4.5 per 1,000), and 4,222 during the pandemic period (8/2020–12/2021, **6 cCMV cases identified**; 1.4 per 1,000; $p<0.01$).

Mother's age group, y				
≤24	1846 (9)	11 (14)	6.0 (3.3-10.7)	1.9 (0.9-4.2)
25-29	4432 (22)	14 (18)	3.2 (1.9-5.3)	1 [Reference]
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- Higher prevalence in women <24 years of age
- No significant differences by race/ethnicity
- All 45 (62.5%) infants born to multiparous mothers had a sibling in daycare.
- Prevalence was higher among second-born than first-born infants (prevalence ratio: 1.9; 95% CI: 1.2-3.1; p=0.01).


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21

CIDMTR
Center for Infectious Diseases & Microbiology Translational Research

Is Universal Newborn Screening for Congenital CMV Good Public Policy?

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Disclosure Slide

I do support implementation of universal newborn screening for congenital cytomegalovirus infection

- [x] Yes
- [ ] No

Clinical Sensitivity

- Ability of a test result to identify CMV **disease**
- Test characteristics favoring enhanced clinical sensitivity may be relevant to assessment of the value and utility of universal cCMV screening
- Clinical definition of CMV disease was modified from Rawlinson *et al*. to include late-onset SNHL/isolated SNHL at birth (Sidesinger *et al*., this meeting)

- Asymptomatic
- Asymptomatic except for SNHL
- Mildly symptomatic
- Moderately to severely symptomatic

Rawlinson et al., 2017, Lancet http://dx.doi.org/10.1016/S1473-3099(17)30143-3

87 infants identified with cCMV out of 23,644 total screened (prevalence of 3.7 per 1,000)

68 infants classified as asymptomatic

4 infants classified as asymptomatic with isolated SNHL

15 infants classified as symptomatic

9 infants classified as mildly symptomatic

6 infants classified as moderately-to-severly symptomatic

- 2 with delayed hearing loss
- 2 with hearing loss at initial diagnosis

2 Infants with late onset SNHL

- Asymptomatic 78%
- Symptomatic 17%
- Asymptomatic with SNHL 4.6%

In total 10/87 infants to date have demonstrated variable degrees of SNHL (11.5%)

Clinical Sensitivity

- 21 infants with symptomatic CMV disease at birth and/or isolated SNHL

  18 were treated with valganciclovir

- Clinical sensitivity of saliva testing -> 20/21 = 95%
- Clinical sensitivity of DBS testing (UMN) -> 17/21 = 81%
- Clinical sensitivity of DBS testing (CDC) -> 19/21 = 90%

Conclusions

- Universal screening study demonstrated a prevalence of cCMV of 3.7/1000 in a group of selected Minnesota newborn nurseries
- 78% asymptomatic*
- Enhancement in clinical sensitivity of DBS over analytical sensitivity may predict that DBS PCR has particular value in identify children at risk for sequelae

* Two children in this group with late-onset SNHL

https://gvbc.org/sermon/are-we-there-yet-freedom/

Is the DBS PCR for cCMV RUSP-Ready?

- Range of 81-90% clinical sensitivity
- Captured all but one of the infants that received valganciclovir therapy
- However negative DBS tests occurred in two babies in both laboratories
- Clinical sensitivity of saliva testing = 95%

Unresolved Questions

- What does the enhanced clinical sensitivity in symptomatics "really mean"?
  - Is viremia/DNAemia really the issue?
- Are asymptomatics "really asymptomatic"?
- Is there a viral load threshold that we worry about?
  - What's the denominator?
- Our goal is sensitive detection, but does the "compartment" matter?
- What does RUSP need to "hear" – and do they matter?

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CMV 2023 LITA

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