SANTA CLARA COUNTY PUBLIC HEALTH LABORATORY

3/25/2025

Retail Raw Milk Surveillance

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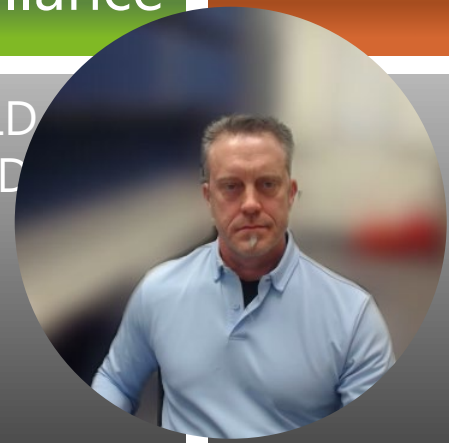
Laboratory D



Santa Clara County PUBLIC HEALTH

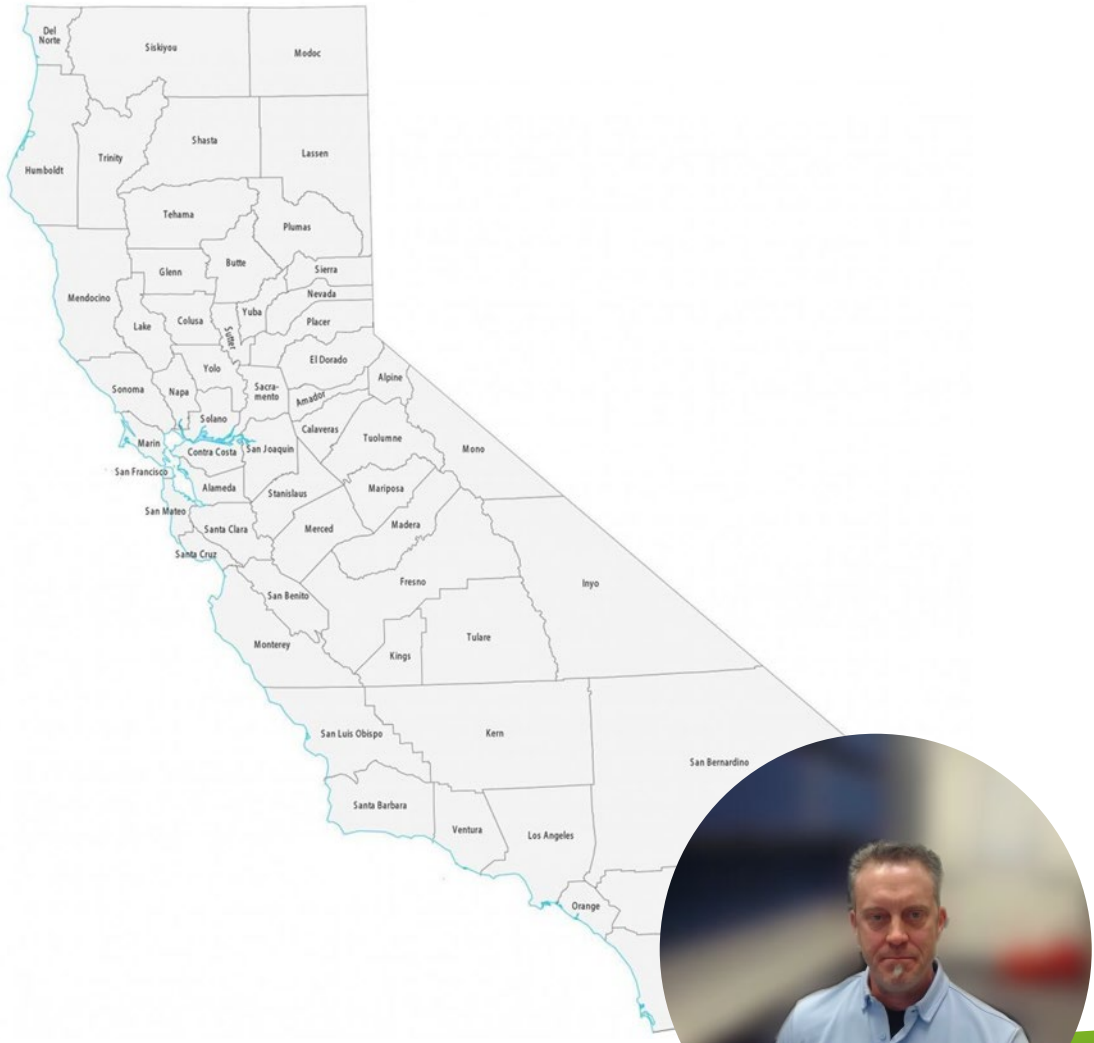
Disclaimer

- I have no financial incentives to disclose outside my role as laboratory director for the County of Santa Clara Public Health Department.
- The views and opinions expressed in this presentation are my own and are not intended to represent the County of Santa Clara.


Welcome to
California
ENTERING PACIFIC TIME

Quick facts about California-the Golden State

- 58 counties
- Decentralized
- 28 local Public Health Labs
- 39.4 million residents in 2024
- Nearly 900 miles from North to South



Milk production in California

Santa Clara County

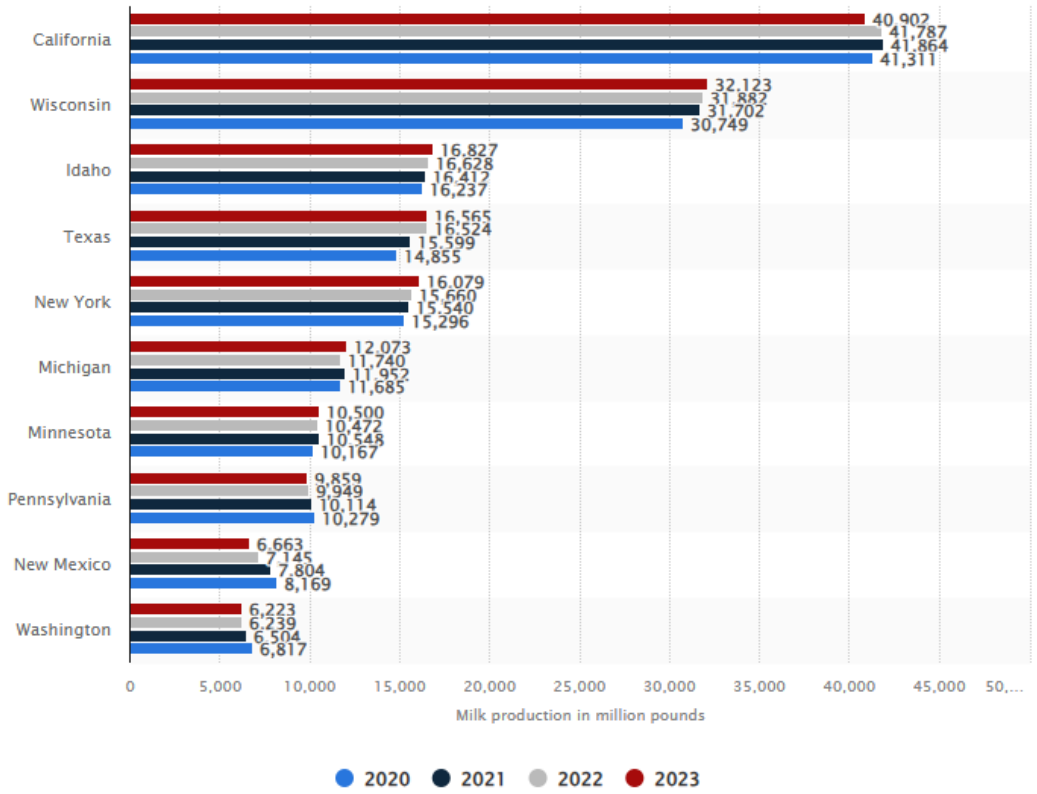
8 major milk producing counties in California

Data produced by Statista

Agriculture › Farming

Top 10 milk producing U.S. states in 2020 and 2023

(in million pounds)



Decision to test retail raw milk for H5

- May 2024
  - Met with Public Health Leadership to discuss ongoing transmission of H5 in dairy cattle and the possibility of testing retail raw milk sold in our jurisdiction.
  - Health Officer deemed this testing important; laboratory recognizes some unknowns and possible limitations.
  - PHL performed a series of proof-of-concept tests to determine if CDC assay would work with raw milk.

Initial barriers and concerns for implementing this test

- FDA assay was not released yet to PHLs
- No positive milk to use for assay development
- CDC assay isn't "approved" for milk
- Unknown limit of detection, ie are negatives really negative?
- No system in place to purchase retail raw milk
- Biosafety concerns-needed a separate risk assessment BSL2 or BSL3?

H5 testing in Santa Clara County

- First sample tested June 17, 2024
- Used CDC assay as FDA/USDA method had not been released yet.
- One sample per week.
- 3 samples per week starting July 2024.
- Currently 5 samples per week.

First detection November 2024

- Detected Flu A/H5 in retail raw milk November 20, 2024, CT 25.6
- We screen for Flu A first then reflex to subtyping to ensure no seasonal contamination, then reflex to H5.
- November 21, 2024. 2 50mL conical vials of sample were shuttled to California Animal Health & Food Safety Laboratory (CAHFS) in the evening for testing Friday morning November 22, 2024.

CAHFS testing

- 2 aliqouts were tested by CAHFS
  - Both aliqouts were positive for Flu A
  - Both positive for Avian Influenza H5
  - Both positive for H5 Gs_GD subtype
  - All CT values <27

Biotechnology

Avian Influenza virus H5 gene qRT PCR

Animal/Source	Specimen	Specimen Type	CT Result	Final Result
1	(1) 24SCPH035146-1	Milk, Raw	25.29	Virus Detected
1	(2) 24SCPH035146-2	Milk, Raw	24.9	Virus Detected

Avian Influenza virus H7 gene qRT PCR

Animal/Source	Specimen	Specimen Type	CT Result	Final Result

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CAHFS Final Version 1 Accession # B2402552 November 25, 2024

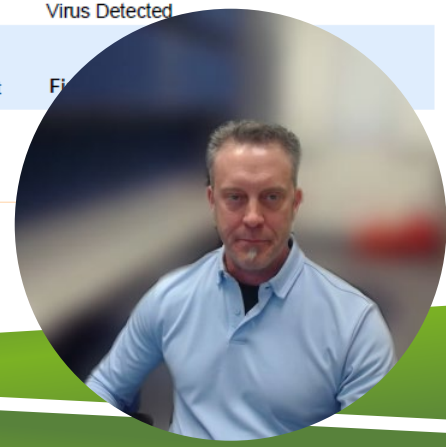
1	(1) 24SCPH035146-1	Milk, Raw	0	Not Detected
1	(2) 24SCPH035146-2	Milk, Raw	0	Not Detected

H5 Gs_GD influenza subtype qRT PCR

Animal/Source	Specimen	Specimen Type	CT Result	Final Result
1	(1) 24SCPH035146-1	Milk, Raw	26.32	Virus Detected
1	(2) 24SCPH035146-2	Milk, Raw	26.30	Virus Detected

Influenza A PCR with Internal Control

Animal/Source	Specimen	Specimen Type	CT Result	Fi
1	(1) 24SCPH035146-1	Milk, Raw	25.62	
1	(2) 24SCPH035146-2	Milk, Raw	25.64	



CAHFS samples sent to NVSL

- Both positive samples sent to National Veterinary Services Laboratory for further testing
  - NVSL confirms both as PCR positive for H5
  - Confirms one of the two viable virus isolation

Sample: 24SCPH035146-1 **Animal ID:** B2402552-01.0001 **Specimen Type:** Milk **Species:** Cattle, Domestic Dairy (No breed specified)
Text ID: 24-035262-001

Virus Isolation	*RRTPCR IAV*	*RRT-PCR EA H5 HPAI*	*RRT-PCR IAV N1*
Negative	Detected (Ct 29.5)	Detected (Ct 26.6)	Detected (Ct 28.6)
RRT-PCR H5 2.3.4.4	***Virus Type***	***Strain ID***	
Detected (Ct 28.0)	HPAI	EA/AM 2.3.4.4b H5N1 (B3.13)	

Sample: 24SCPH035146-2 **Animal ID:** B2402552-01.0002 **Specimen Type:** Milk **Species:** Cattle, Domestic Dairy (No breed specified)
Text ID: 24-035262-002

Virus Isolation	*RRTPCR IAV*	*RRT-PCR EA H5 HPAI*	*RRT-PCR IAV N1*
Positive	Detected (Ct 29.8)	Detected (Ct 27.2)	Detected (Ct 28.9)
RRT-PCR H5 2.3.4.4	***Virus Type***	***Strain ID***	
Detected (Ct 28.6)	HPAI	EA/AM 2.3.4.4b H5N1 (B3.13)	

Samples were tested for influenza A by molecular assays including a real-time RT PCR targeting Eurasian lineage goose/ Guangdong H5 clade 2.3.4.4b as well as an assay that targets the cleavage site of highly pathogenic avian influenza (RRT-PCR EA H5 HPAI). Molecular testing reported on 11/27/2024.

HPAI: highly pathogenic avian influenza per cleavage site analysis
EA 2.3.4.4b: H5 goose/Guangdong clade; this lineage is highly pathogenic for poultry
EA/AM: reassortant of H5 goose/Guangdong and North American wild bird lineage (genotype in table above)

Santa Clara County response to detected H5 in raw milk

- PHL detects first positive on Nov 20, 2024.
- Public Health Department Operations Center (PHDOC) activated Nov 21, 2024.
- CAHFS confirms samples positive for H5 on Nov 23, 2024
- Nov 24, 2024 CDPH and CDFA secured a voluntary recall (by the producer).
  - SCCPHD recommended local retailers remove raw milk from shelves
  - PIO implemented public information and warning campaign
- Nov 26, 2024 SCCPHL detected a second H5 positive in raw milk from same producer with different lot
- Dec 3, 2024 CAHFS confirmed H5 positive in producer bulk milk tank test
  - Broader recall of milk issued
  - PHDOC continued to monitor for human cases
- Dec 11, 2024 PHDOC deactivated

California Department of Public Health gets involved

California Acts to Protect Public Health: CDPH Warns Against Drinking Single Lot of Raw Milk Following Bird Flu Detection; Secures Voluntary Recall

November 24, 2024
NR24-039

No illnesses have been reported; Pasteurized milk is safe to drink

What You Need to Know: CDPH warns consumers not to drink one batch of cream top, whole raw milk from Raw Farm, LLC of Fresno County due to a detection of bird flu virus. Raw Farm, LLC issued a voluntary recall at the state's request of the affected lot code 20241109 with a Best By date of 11/27/2024. Consumers should immediately return any remaining product to the store where it was purchased. Pasteurized milk remains safe to drink

Sacramento – The California Department of Public Health (CDPH) today is warning the public to avoid consuming one batch of cream top, whole raw milk produced and packaged by Raw Farm, LLC of Fresno County due to a detection of bird flu virus in a retail sample. At the state's request, the company has issued a voluntary recall of the affected raw milk with lot code 20241109 and Best By date of 11/27/2024 printed on the packaging.

No illnesses associated with this lot of raw milk have been reported. Out of an abundance of caution, and due to the ongoing spread of bird flu in dairy cows, poultry, and sporadic human cases, consumers should not consume any of the affected raw milk. Customers should immediately return any remaining product to the retail point of purchase. CDPH is also notifying retailers to remove the affected raw milk from their shelves.

The County of Santa Clara Public Health Laboratory has been testing raw milk products from retail stores as a second line of consumer protection. The county identified bird flu in one sample of raw milk purchased at a retail outlet. The testing was then verified by the California Animal Health and Food Safety Laboratory System.

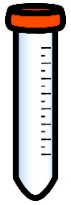
H5 detection in raw milk in California

Implementing the FDA assay

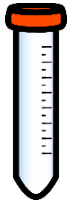
- Initially delayed weeks due to purchasing/shipping delays on some of the required reagents.
- Decided to compare CDC assay and Qiagen manual spin column extraction to FDA assay and Qiagen manual spin column extraction.
- Goal of comparison was to compare both extraction methods and both PCR assays.
- Used archived confirmed positive raw milk as validati samples

Comparison study

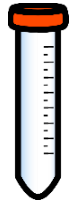
10mL + #1



10mL + #2



10mL - #1



- Each 10mL sample spiked with 100uL MHV and HSC
- Side by side extractions using DSP Viral RNA kit
- 8 replicates from each 10mL sample using both extraction methods
- 24 samples for CDC extraction and 24 samples for FDA extraction = 48 total samples extracted
- 48 samples tested on both PCR assays
- Also did reproducibility and precision testing

Average CT values	
CDC extraction + FDA PCR	30.5
FDA extraction + FDA PCR	31.1
CDC extraction + CDC PCR	24.6
FDA extraction + CDC PCR	24.4

Takeaways from comparison study and retail raw milk testing

- No major difference in the extraction protocols compared with both PCR assays
- ~6 CT value lower result for the CDC assay compared to the FDA assay
- FDA is more time consuming assay for both extraction and PCR
- FDA assay not quite as user friendly as the CDC assay

Thank you

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Santa Clara County

PUBLIC HEALTH

BETTER HEALTH FOR ALL