



NTM in Hospital Water Supply

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A dark grey arrow points to the right from the left edge of the slide. Below it, several thin, curved lines in shades of blue and grey sweep across the left side of the slide.

Background

- ▶ Four patients while housed in the ICU at a single Medical Center had sputum specimens which grew *Mycobacterium farcinogenes/senegalense*
 - ▶ Initial ID by MALDI-TOF at a reference laboratory
- ▶ Specimens collected between February 26, 2015 and March 15, 2015
- ▶ No patients appeared to have clinical disease due to this mycobacterium species
- ▶ Two of the patients housed in the same room on separate days
- ▶ One patient housed in the adjacent room
- ▶ One patient housed in a non-adjacent room



Background

- ▶ No report of recent construction or other engineering problems
- ▶ All four patients underwent induced sputum collected for AFB
- ▶ Three of the four patients had received nebulizer treatments with inhalation solution as well
 - ▶ No solutions with the same lot number as used on these patients remained for testing
- ▶ No complaints from other customers to the manufacturers of the solutions
- ▶ Lab had not seen any similar cultures of this organism in the rest of the network
- ▶ No other trends in the ICU were seen

Maybe the Lab contaminated it?

- ▶ Lab was asked to look into their processing
 - ▶ Isolate #1
 - ▶ Three other specimens processed in the same run. All were no growth.
 - ▶ Patient had no record of previous AFB isolates.
 - ▶ Isolate #2
 - ▶ Two other specimens processed in the same run. One was no growth and the other is isolate #3.
 - ▶ Isolate #3
 - ▶ Two other specimens processed in the same run. One was no growth and the other is isolate #2.
 - ▶ Patient with isolate #2 and #3 had been previously positive for *M. xenopi*. Also had two recent positive AFB cultures with *M. xenopi*
 - ▶ Isolate #4
 - ▶ Three other specimens processed in the same run. All were no growth.
 - ▶ Patient had no record of previous AFB isolates.
- ▶ None of these patients had previous *M. fortuitum complex* isolated from any AFB cultures.
- ▶ Felt at this time that this was not a lab contaminant



What about the Saline?

- ▶ Manufacturer of saline used for the induced sputum stated that their sterility testing had been negative and they had no other customer complaints
- ▶ Lab helped with investigation by trying to culture the saline and grow on chocolate agar
 - ▶ Nothing grew from the Lab's saline cultures
- ▶ With additional investigation of the saline, it was determined that the inhalation solution was an unlikely source.



More Patients and Another Hospital

- ▶ Since the initial investigation, any additional isolates were tracked.
- ▶ A second facility within the same region as the initial Medical Center also started to have positive cultures
 - ▶ Medical Centers located 11 miles apart within same urban metropolitan area
- ▶ The initial cluster under investigation consisted of:
 - ▶ 14 critical care patients at two Medical Centers
 - ▶ Isolates from respiratory specimens obtained between 2/26/15 and 09/27/15
 - ▶ All specimens processed at Milwaukee Medical Center
 - ▶ Milwaukee is able to perform probes in house
 - ▶ Perform MTB complex, MAC, gordonae, and kansasii
 - ▶ Additional identification performed by an outside reference lab.
 - ▶ Specimens identified as *M. senegalense/farcinogenes* by MALDI
 - ▶ Specimens sent to reference lab for PFGE testing



Results from PFGE testing

- ▶ All ten isolates from Medical Center #1 had the same pattern
 - ▶ Identified as *M. senegalense*
 - ▶ PFGE Pattern Xba I: A1 and Ase I: B1
- ▶ All six isolates from Medical Center #2 had the same pattern
 - ▶ Identified as *M. senegalense*
 - ▶ PFGE Pattern Xba I: A2 and Ase I: B2



Possible Outbreak Investigation

- ▶ November 2015
- ▶ Meeting with Public Health Surveillance and Research, CDC, Infectious Disease, and Lab to determine if was a potential outbreak.
- ▶ Plans from meeting:
 - ▶ Explore possibility of a water source
 - ▶ Send specimens to reference lab for 16S sequencing, ITS, *hsd*, and *rpoB* gene sequencing
 - ▶ Noted that Milwaukee's reference lab changed testing methodology from HPLC to MALDI-TOF in early 2015
 - ▶ Previously these would have been reported as *M. fortuitum complex*
 - ▶ Suggestion that maybe this is not a new problem
 - ▶ Milwaukee had *M. fortuitum complex* isolates dating back to 2012
 - ▶ Sampling of these isolates also sent to reference lab
 - ▶ Determine if this organism type is seen at other Medical Centers outside of the Network

Water Testing

- ▶ Under the CDC's instruction, samples were obtained from Medical Center #1
 - ▶ Ten one liter samples were obtained from the ICU and sent to reference lab
 - ▶ Included sinks and ice machines
 - ▶ Reference lab vacuum filtered the samples
 - ▶ Filters were placed on Middlebrook 7H11 agar plates and incubated at 37°C
 - ▶ Colony growth was analyzed by MALDI-TOF and 16S gene sequencing if MALDI-TOF did not provide an identification
 - ▶ All 10 water samples had visible growth on agar plates
 - ▶ In 7 of 10 water samples an identification was obtained
 - ▶ The identification *Acidovorax temperans*
 - ▶ In 3 of the 10 water samples there was bacteria growing
 - ▶ Unable to obtain a reliable identification
- ▶ No initial water samples had any growth of mycobacterium



Water Testing

- ▶ Under the CDC's instruction, samples were obtained from Medical Center #2
 - ▶ Sinks and ice machines were tested
 - ▶ The endemic strain was not seen from the water cultures
 - ▶ An ice machine grew *M. mucogenicum*



Further Investigation

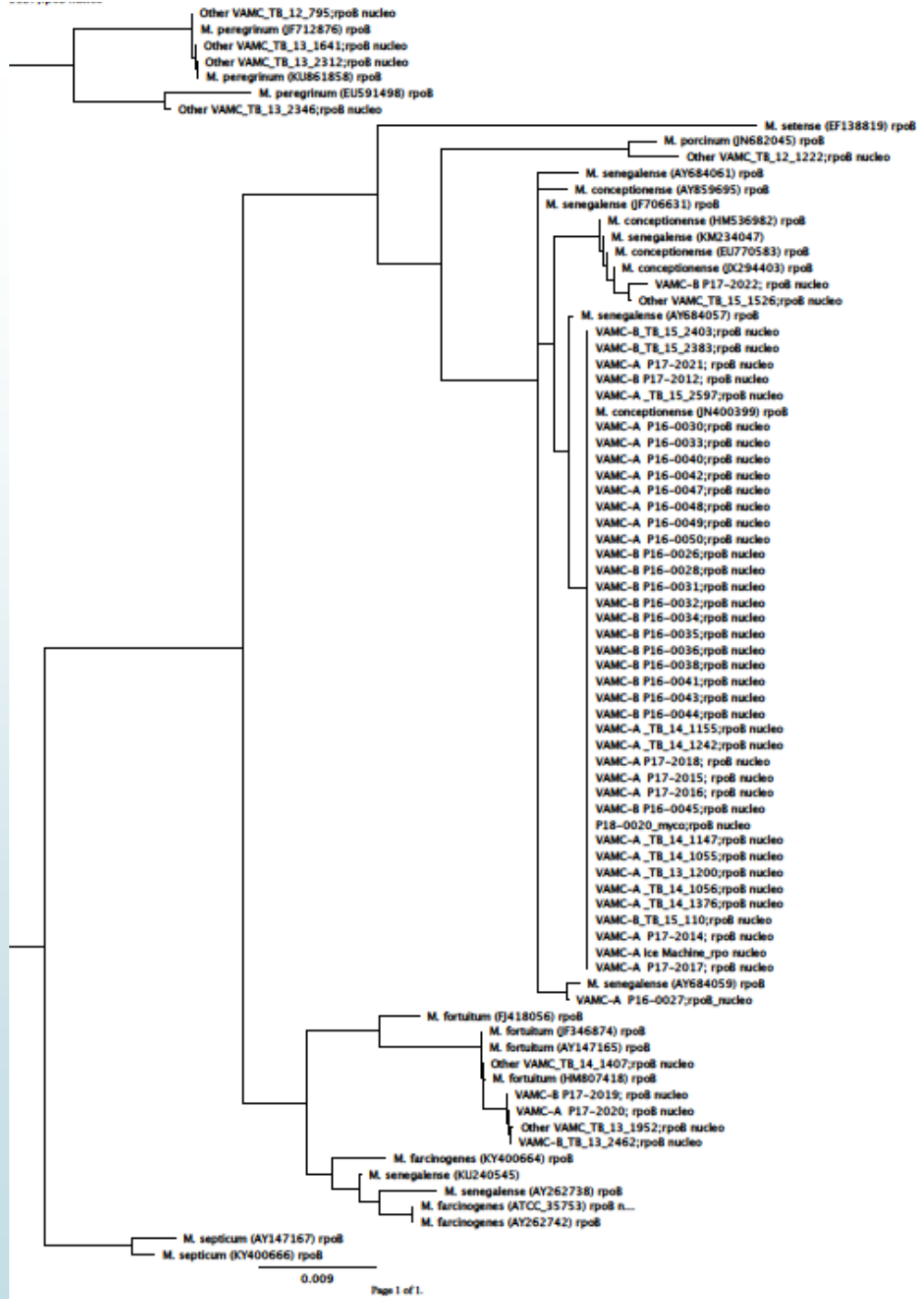
- ▶ Found that a private hospital within the same urban area as Medical Center #1 and #2 had two cultures with *M. senegalense* in November/December
- ▶ Lab processing was reviewed for the eight additional specimens from 04/22/15 to 09/27/2015
 - ▶ All specimens processed on the same date as the isolate containing *M. farcinogenes/senegalense* either had no growth or contained *MAC*, *M. gorondae*, *M. xenopi*, or *M. kansasii*.
 - ▶ Again, no indication of a contamination issue from Lab processing
- ▶ Additional water sent for testing

Results

- ▶ Initially twenty-six isolates from 23 different patients were tested
 - ▶ Collection dates ranged from 03/02/2012 to 10/23/15
 - ▶ 16S results indicate that isolates were aligned with the *M. fortuitum* group
 - ▶ ITS, *hsd*, and *rpoB* gene testing suggested that the isolates were most closely aligned to *M. conceptionense*
 - ▶ Phylogenic analyses further indicated that all isolates (except possibly one) were the same strain.
 - ▶ The three patients with two isolates, and one patient with isolates collected 15 months apart also had identical strains

Results

- ▶ Additional isolates were sent for testing, this also included *M. fortuitum complex* isolates from the Milwaukee area.
- ▶ There were 46 isolates (25 from Medical Center #1, and 21 from Medical Center#2) tested
 - ▶ 37 isolates were identified via rpoB gene sequencing to be an endemic strain of *M. conceptionense*
 - ▶ The strain was also isolated from an ice machine located in the ICU from Medical Center #1
 - ▶ Two isolates (1 from each Medical Center) were a strain of *M. conceptionense* differing by 6 nucleotide sequences out of a total of 727 from the endemic strain
- ▶ Of 9 *M. fortuitum complex* isolates from other Medical Centers, there was one *M. conceptionense* strain that differed by 6 nucleotide sequences out of a total of 727 from the endemic strain
 - ▶ Other specimens tested were not closely related



0.009



Conclusions

- ▶ A single strain of *M. conceptionense* was isolated from multiple patients at two Medical Centers between 12/21/2012 and 11/04/2016
- ▶ Of patients who had the *M. conceptionense* endemic strain isolated, 89% were located in the critical care units of the Medical Center
- ▶ The ice machine from Medical Center #1 that was found to be positive with the endemic strain was removed from service on 11/29/2016. No further *M. conceptionense* isolates have occurred at that Medical Center since its removal
- ▶ No definitive water source was found at Medical Center #2
 - ▶ A common municipal water source cannot be ruled out
- ▶ Possible mechanisms for colonization for direct or indirect patient-to-patient transmission remain unknown



M. conceptionense

- ▶ Part of the *M. fortuitum* complex
 - ▶ Others in complex include *M. fortuitum*, *M. houstonense*, *M. farcingenes*, *M. peregrinum*, *M. porcinum*, *M. senegalense*, and *M. septicum*
 - ▶ Species differentiation is difficult
- ▶ Nonpigmented rapid grower
- ▶ First identified in 2006 from a patient with post-traumatic osteitis
- ▶ Emerging opportunistic pathogen
- ▶ Water is a common source of this group
- ▶ Usually cause skin and soft tissue infection
- ▶ If found in respiratory isolates, normally representative of colonization or transient infection



Questions?