

SUMMARY

**PUBLIC HEALTH
INVESTIGATION INTO THE
LEGIONELLA OUTBREAKS
IN SYDNEY CBD**

MARCH AND MAY 2016



Health

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Summary

This report summarises the findings of public health investigations into *Legionella* outbreaks in Sydney CBD, March and May 2016. The report outlines public health actions, case identification, environmental investigations, testing, results and actions taken. The investigation involved the Ministry of Health, Health Protection NSW, several NSW public health units, the City of Sydney, Forensic and Analytic Science Services, and the Centre for Infectious Diseases Microbiology. For the first time in NSW whole genome sequencing was used in a *Legionella* investigation. Epidemiological, environmental and laboratory investigations determined that 13 people and two cooling towers shared a common strain of *Legionella*, although the actual source of the human cases cannot be proven.

Background

The Sydney CBD is approximately 12.3km² and on any one day there are an estimated 610,000 visitors to the area on top of a resident population of approximately 205,000 people ([City of Sydney](#)). The City of Sydney Council has approximately 1200 registered cooling towers within this area.

Legionnaires' disease is an infection caused by one of a number of bacteria of the genus *Legionella* characterised by fever, pneumonia, and muscle aches. It is notifiable by laboratories under the NSW Public Health Act 2010. Symptoms develop between 2 - 10 days after exposure to the bacteria. People with significant underlying health conditions and people who smoke are more likely to develop Legionnaires' disease after exposure to the bacteria.

The most common cause of Legionnaires' disease notified in NSW is *Legionella pneumophila* serogroup 1 (LP1). LP1 is commonly found in water, and can be spread when contaminated water is aerosolised, and then inhaled by people. This disease is not transmitted from human to human.

The Public Health Act and Public Health Regulation 2012 set out the regulatory requirements for "regulated systems", which are the systems that are most susceptible to contamination by *L. pneumophila*. Regulated systems include air-handling systems, hot water systems, humidifying systems, warm water systems and water cooling systems. A water cooling system includes a water cooling tower. Each local government authority maintains a register of regulated systems within its local area.

However, *L. pneumophila* can also colonise other water systems subject to aerosolisation such as decorative fountains. These are not subject to regulation under the Public Health Act as they are only rarely identified as a cause of Legionnaires' disease, however in the context of an outbreak all potential water sources are investigated.

Following the notification of four cases of Legionnaires' disease on March 7 and 8, NSW Health initiated a comprehensive investigation and response. Here we report on the cases identified in this outbreak, the response, and the subsequent outbreak in May 2016.

Methods

March outbreak

Response coordination

- On 9 March 2016, the public health emergency operations centre (PHEOC) and the Incident Control System (ICS) was activated, and an incident management team was established.
- A field team consisting of approximately 20 environmental health officers from City of Sydney, Health Protection NSW and NSW public health units were deployed to inspect and sample all cooling towers as well as other potential *Legionella* sources in the investigation area.
- Coordination of human and environmental laboratory testing between primary public and private laboratories, the Centre for Infectious Diseases & Microbiology Laboratory Services (CIDMLS), at the Institute of Clinical Pathology and Medical Research (ICPMR), Pathology West and the *Legionella* Reference Laboratory at NSW Forensic and Analytical Science Service (FASS) was activated.
- Daily incident management team teleconferences were held at 9am and 4pm
- Daily media statements were issued from the Ministry of Health
- SITREPs were produced and disseminated daily (a daily SITREP is a report which summarises cases, laboratory findings, exposure site inspections and planned actions).
- Correspondence was issued to all cooling tower operators in the CBD requesting they inspect and maintain their plants.

Epidemiological investigation

Exposures of all Legionnaires' cases notified from late December 2015 to March 2016 were reviewed and cases reinterviewed if required. All public health units were notified and asked to keep in daily contact with their laboratories to ensure timely identification of new cases. Active case surveillance was carried out by the relevant public health units and any new notifications were interviewed as a matter of priority.

Clinicians, emergency departments, intensive care units, general practitioners and pathology laboratories were issued alerts about the outbreak and requested that appropriate testing be undertaken for suspected cases. Alerts to other jurisdictions were disseminated due to the possibility of interstate and international visitors. The NSW Health Public Health Real-Time Emergency Department Surveillance System (PHREDSS) was utilised to monitor emergency department presentations and admissions retrospectively and prospectively for people presenting with pneumonia-like illness.

Environmental investigation

The environmental investigations involved physical inspections of building cooling towers as well as unregulated sources, such as water fountains within defined areas of the Sydney CBD in order to identify and sample possible sources of *L. pneumophila*. Other regulated systems, such as warm water systems were not inspected as exposures related to such systems occur inside buildings, and there was no indication that any single building had been visited by more than one case. Hence the investigation focussed on *Legionella* sources that could contaminate the outdoor environment. The investigation areas were determined on the basis of reviewing the movements of the cases and defining an area of common outdoor exposure between these cases. The investigations were jointly conducted by the City of Sydney Council and NSW Health (including local health district public health units).

The City of Sydney Council maintains the register of regulated systems within the Sydney CBD and this was used to identify known regulated systems within the investigation areas. Water treatment companies who undertake the regular maintenance and testing of cooling towers were also contacted and asked to carry out rapid cleaning and disinfection of those cooling towers they were contracted to manage. Concurrently, historical information held on the City of Sydney register was also reviewed to ascertain if there were any problematic towers or systems that needed to be prioritised for inspection during the environmental investigation.

Teams of environmental health officers (EHOs) undertook inspections of the cooling towers, including obtaining samples and reviewing maintenance records where available. In conjunction with the City of Sydney a review of satellite imagery was also undertaken in an attempt to identify potential unregistered cooling towers and other potential sources from unregulated sites. One team was re-deployed to identify and inspect potential unregistered towers, as well as unregulated sources, such as decorative water fountains. Visibly unclean cooling towers were issued with immediate orders to clean and disinfect their systems. As the laboratory results became available appropriate notices and prohibition orders were issued to owners of cooling towers whose systems tested positive (Appendix 3.1).

The environmental investigation began on 9 March 2016 and focused on a 1.56km² investigation area which was subsequently expanded by a further 1.32km² on the basis of case exposures (Figure 1). The environmental response teams undertook risk-based inspection and sampling of air conditioning system cooling towers starting from Market and George Streets bounded by the area Clarence, King, Park and Pitt Streets (Figure 1). Initially the City of Sydney identified 17 high risk, 28 medium risk and 20 low risk regulated systems within this area. Risk is determined by a system's past testing history and whether or not it is located in an area of high pedestrian traffic density. Further to this, 49 potential unregistered systems were identified by satellite imagery and were inspected (and tested if verified to be an operating cooling tower) although all but a few were found to be no longer in use.

On 11 March 2016 through additional examination of cases' movements in and around the city it was decided to extend the search area to the east, up to and including Elizabeth Street. Therefore the area under investigation was bound by Clarence, King, Park and Elizabeth Streets. A further 60 registered towers and no unregistered towers were identified in this area.

On 14 March 2016 with addition of information from new cases it was decided to extend the search area south to include premises up to and including Bathurst Street. The area was now bound by Kent, King, Elizabeth and Bathurst Streets (Figure 1). A further 23 registered systems and no unregistered towers were identified in this area.

As of 15 March 2016 the environmental response team completed all testing within the area under investigation. In total 199 samples from registered and unregistered systems were sent for analysis at the FASS laboratory.

Laboratory investigation - patients

Legionnaires' disease is difficult to distinguish from other types of pneumonia on the basis of patient symptoms alone. Chest X-rays help to diagnose pneumonia but the diagnosis of Legionnaires' disease requires special laboratory tests.

To confirm cases of *L. pneumophila* during the outbreaks required:

- 1) a positive urinary antigen test for LP1 which is available in local laboratories, or
- 2) a positive *Legionella* culture from sputum or bronchial washings.

To centralise testing for the outbreaks, laboratories were requested in an alert email to refer all sputum and bronchial washing samples from patients suspected to have Legionnaires' disease to CIDMLS, ICPMR for *Legionella* culture and serogrouping.

At the time of the outbreaks CIDMLS, ICPMR were evaluating a new multiplex polymerase chain reaction (PCR) test for *Legionella* spp., LP1 and other *Legionella* which is highly sensitive and specific, has a more rapid turnaround time than culture methods, and is particularly useful as *Legionella* is difficult to isolate in culture. All respiratory specimens received by CIDMLS, ICPMR for *Legionella* culture were also tested by the PCR.

All *Legionella* culture isolates obtained from patients during the outbreaks and identified as LP1 were characterised by whole genome sequencing (WGS), and compared with the environmental isolates, to determine the relatedness of the organisms as an indicator of a possible common source.

Laboratory investigation – environmental

All environmental water samples were referred to the Legionella Reference Laboratory at NSW Forensic and Analytical Science Service (FASS) for testing. The samples were analysed using microbiological cultural techniques. Negative results are reported as <10 colony forming units per millilitre (cfu/mL) (limit of detection of the method). Results \geq 10 and <100cfu/mL occur not infrequently and may represent ineffective maintenance practices.

Although there is little evidence at what level a system may become a risk to public health, levels greater than or equal to 100 cfu/mL are considered potentially sources of infection. Presumptive results were available 1-2 days prior to final results, allowing earlier intervention.

All environmental isolates of LP1 were forwarded to CIDMLS, ICPMR for whole genome sequencing (WGS) analysis.

A timeline of the March outbreak is in Appendix 1.1.

May outbreak

Response Coordination

- A field team consisting of approximately 20 environmental health officers from City of Sydney, Health Protection NSW and NSW public health units were deployed to inspect and sample all cooling towers as well as other potential *Legionella* sources in the investigation area.
- Coordination of human and environmental laboratory testing between primary public and private laboratories, the Centre for Infectious Diseases & Microbiology Laboratory Services (CIDMLS), at the Institute of Clinical Pathology and Medical Research (ICPMR), Pathology West and the Legionella Reference Laboratory at NSW Forensic and Analytical Science Service (FASS) was reactivated.
- Incident management team teleconferences were held daily with minutes and response actions recorded.
- Daily media statements were issued from the Ministry of Health
- Correspondence was issued to all cooling tower operators in the CBD requesting they inspect and maintain their plants.

Epidemiological investigation

All public health units were notified and asked to keep in daily contact with their laboratories to ensure timely identification of new cases. Active case surveillance was carried out by the relevant public health units and any patients newly notified with LP1 were interviewed as a matter of priority. Clinicians, emergency departments, intensive care units and pathology laboratories were issued alerts about the outbreak and requested that appropriate testing be undertaken for suspected cases.

No alert was sent to general practitioners as it was felt that they were still acting on the previous alert sent in March. An alert to other jurisdictions was disseminated due to the possibility of interstate and international visitors.

Environmental investigation

The reported exposures of the cases indicated a search area of 1.58km² to the immediate north of the investigation area from the March outbreak.

On the morning of 4 May 2016 the environmental response teams undertook risk-based inspection and sampling of air conditioning system cooling towers bounded by the area Margaret, George, King and Kent Streets (Figure 2). Based on additional information from case movements, it was decided to extend this area to include the area bound by Pitt Street, George Street, King Street and Curtin Place (which is in line with Margaret Street) on May 5.

Samples were taken from 87 registered cooling towers in the area of interest. Additionally a York Street tower was retested as this tower returned a positive result from the March testing and another two cooling systems located in George Street were tested as a result of recent positive results for *Legionella* (not LP1).

Teams of EHOs collected 130 environmental samples over a period of five days.

Laboratory investigations – patient and environmental

Laboratory methods were as described for the March outbreak.

A timeline of the May outbreak is in Appendix 1.2.

Results

March outbreak

Epidemiological investigation – see Appendix 2.1

On 7 March the Northern Sydney Local Health District Public Health Unit (NSLHD PHU) was notified about a male in his 60's with confirmed LP1 (Case 1). The next afternoon (8 March 2016) the South Eastern Sydney Local Health District Public Health Unit (SESLHD PHU) received two notifications of LP1 in a male resident in his 30's (Case 2) and a male resident in his 60's (Case 3). On the same day Queensland Health (QLD Health) notified the NSW Health Protection Communicable Diseases Branch (CDB) of a confirmed LP1 infection in a male tourist in his 60's who had stayed in the Sydney CBD during his incubation period and then travelled to Queensland (Case 4).

Information obtained by the PHUs and QLD Health revealed all four cases had spent considerable time in the Sydney CBD around Town Hall, Market/George/Druitt/Sussex Streets during their incubation periods.

On 9 March 2016 as a result of a review of the cases notified previously to NSW Health, cases 5 and 6 were identified. Case 5, a male resident in his 40's, had visited several CBD locations including Liverpool Street, Criterion Hotel (Pitt and Park streets), Town Hall Station/Square and had walked down York Street in his incubation period in February. Additionally a case notified in late December 2015, a male resident in his 80's, had exposures around Town Hall station and the City Tattersalls Club in Pitt Street (Case 6).

On 11 March 2016 cases 7 and 8 were reported by the Western Sydney Local Health District Public Health Unit (WSLHD PHU): case 7, a male in his 50's reported exposures between Chinatown to Town Hall and also walked from Central Station to Goulburn Street. Case 8, a male resident in his 40's caught a train at Central Station.

On 13 March 2016 a ninth and final case was reported by NSLHD PHU: a male resident in his 80's, whose family reported him visiting George Street and Castlereagh Street as well as Pitt Street Sydney. This person died in March 2016.

The pneumonia presentations and admissions reported through PHREDSS identified 61 possible cases in people aged 35-64 years for the week ending 8 March 2016. All of these people were followed up by the local PHU and were either one of the known cases (2), had a negative result for LP1 (56), or had a positive result for LP1 (3 cases) but had not gone into the CBD.

Environmental investigation – see Appendix 3.1.

Samples from the regulated systems of seven premises (ten samples) had 100 cfu/mL or greater of LP1 grow on culture, including one with an excessively high count of 2500 cfu/mL. Samples from two other positive towers had 10 cfu/mL growth of LP1.

As a result of inspections and laboratory findings, the following notices and orders were issued under the Public Health Act:

- One Prohibition Order – for elevated cfu/mL levels
- Three Section 33 Notices – for visibly unclean towers
- Three Improvement Notices – for elevated cfu/mL levels
- Seven warning letters – Issued by City of Sydney for minor non-conformances

Laboratory investigation

Six patient samples were positive for *Legionella pneumophila* serogroup 1 (LP1) by PCR. Of these, culture was successful for samples from three patients, cases 2, 8 and 9.

Whole genome sequencing results were available on April 6 and showed that isolates from two patients (cases 2 and 9) were closely genetically related, and that the isolate from the third patient (case 8) was distinctly different from the other two.

This patient had only been to Central Station, and thus epidemiologically and genomically was a sporadic case not linked to the outbreak.

Whole genome sequencing on the environmental isolates found that all were distinct from the patient samples, that is, none were likely to be responsible for the human cases.

May outbreak

Epidemiological investigation - see Appendix 2.2

On 28 April 2016, NSLHD PHU received notification of a male resident in his 80's with confirmed LP1 (case 1). This man reported North Sydney exposures and also daily visits to Clarence Street in the CBD.

On 3 May 2016, two cases of LP1 were notified to the SESLHD PHU and WSLHD PHU. Case 2, a female resident in her 30's reported that she worked in the CBD and would walk from Martin Place down Erskine Street and along Shelley Street each business day during her incubation period. Case 3 a female resident in her 30's, worked in the CBD and walked from Wynyard Station along George Street each day.

On 6 May 2016, case 4 was notified to Sydney Local Health District Public Health Unit (SLHD PHU) and case 5 to South Western Sydney Local Health District Public Health Unit (SWSLHD PHU). Case 4, a male resident in his 80's travelled by train to Circular Quay and took a ferry on 24 April 2016. This person died in May 2016 and was later excluded from the outbreak by whole genome sequencing results. Case 5, a male resident in his 50's attended the Anzac Day march and had walked extensively through the CBD on April 24 and 25.

On 9 May 2016, Communicable Diseases Branch received a notification from the Victorian Health Department of a male resident in his 40's who attended a course and stayed in York Street from 25 April to 1 May 2016 (case 6). This case took a morning walk around the CBD each day of his stay.

Environmental investigation - see Appendix 3.2

Samples from the regulated systems of two premises had 100 cfu/mL or greater of LP1 grow on culture, including one with an excessively high count of 9800 cfu/mL. Samples from the regulated systems of two other premises had between 10-100 cfu/mL of LP1.

As a result of inspections and laboratory findings, the following notices and orders were issued under the Public Health Act:

- One Prohibition Order - elevated cfu/mL count
- One Improvement Notice: [Issued by City of Sydney] - elevated cfu/mL
- Two Improvement Notices: [Issued by City of Sydney] - no maintenance/operations manual
- Two warning letters - Issued by City of Sydney for minor non-conformances

Laboratory investigation

Five patient samples were positive for *Legionella pneumophila* serogroup 1 (LP1) by PCR. Of these, culture was successful for samples from four patients, cases 1, 3, 4 and 5.

Whole genome sequencing showed that cases 1, 3 and 5 were closely related however case 4 had a separate and distinct sequence from all other cases. As this case's only CBD exposure was a brief visit to Circular Quay, based on epidemiological and genetic evidence, it is likely they obtained their infection from another exposure source (this case was subsequently linked to the Burwood cluster investigated by Sydney Local Health District, also in May 2016).

Whole genome sequences from all isolates obtained from patients and environmental sources in March and May were compared. Two patient specimens from March (cases 2 and 9), three patient specimens from May (cases 1, 3 and 5) and isolates from two cooling towers from May (Clarence Street site 2 and York Street site 4) were genetically indistinguishable. (Figure 2)

Discussion

In March and May 2016, 15 people who had visited the Sydney CBD during their incubation period were identified with Legionnaires' disease due to *Legionella pneumophila* serogroup 1. Two of these people were excluded from the outbreak following genetic sequencing of available clinical and environmental isolates, leaving 13 people linked to the outbreak. A thorough investigation of possible sources related to reported case movements took place. *L. pneumophila* SG1 was isolated from several cooling towers including two cooling towers with excessively high colony counts of greater than 1000 cfu/mL. The use of whole genome sequencing provided strong evidence that at least five cases from the March and May outbreaks were linked to a common source, or separate sources contaminated by the same microorganism.

Two cooling towers were found to be contaminated with *L. pneumophila* SG1 genetically indistinguishable from the five available patient samples suggesting that either or both of the towers could have been responsible for the patient infections. Of these two towers, further regulatory action is being considered in respect of the regulated system at the Clarence Street site 2 premises. The York Street site 4 tower was followed up by the City of Sydney with a warning letter as this count was less than 100 cfu/mL. Alternatively an unknown source contaminated with the same microorganism could have been responsible for the patient infections and contamination of these two towers, but was either not sampled or, more probably, cleaned prior to environmental sampling.

There was a period of six weeks where no cases were detected between March and May. It is possible that any contaminated tower(s) were cleaned and disinfected in response to the initial March outbreak reducing the *Legionella* count but not completely removing the contamination and therefore after a six week period it was able to re-grow to unsafe levels.

Other methods were explored to try and better define the most likely source of infection, including review of meteorological records and review of case movements. However due to the uncertainties around wind tunnel effects in the Sydney CBD, highly variable wind direction at different times of the day, overlapping patient exposures, and also some uncertainty with some patient movements, any further resolution of the source was not feasible.

It is often very difficult to pinpoint the source of a Legionnaires' disease outbreak with accuracy. The main aim of a public health response is to stop the outbreak by ensuring that cooling towers and other possible sources of infection in a location suspected to be the cause of the outbreak are controlled as quickly as possible. To achieve this, building owners are warned to maintain cooling towers to ensure they are free of contamination, and environmental health officers carry out door to door inspections of cooling towers.

A cooling tower that is the source of an outbreak may not be identified despite careful investigations. This is because a cooling tower may be only transiently contaminated by a *Legionella* contaminated aerosol, and water vapour from that cooling tower may infect people walking by, as well as contaminate other nearby cooling towers. However that cooling tower's continuous disinfection and regular cleaning processes may decontaminate it, even before infected patients are diagnosed. Should this cooling tower be tested, it may therefore test negative for *Legionella* (because it's been automatically or manually disinfected), even though it's the real source of patients' infection, while the nearby cooling towers (contaminated by the first cooling tower, but not yet disinfected) may test positive, even if they have not caused any infections in patients. Further, patients may have been infected from a tower from which we did not obtain an isolate due to factors including: it was not tested, water system sampling technique, failure of microorganism to grow on culture, and cleaning of the tower before sampling.

Despite extensive awareness of the outbreaks in the community and medical practitioners, it is possible that not all cases were identified, thus limiting the information available to focus the environmental investigation. Patients who present early in the course of their illness and receive an appropriate antibiotic, or who have a mild form of illness, may recover and not undergo investigation to identify Legionnaires' disease.

Other limitations associated with this investigation were the accuracy and completeness of case recall about possible exposure sites, as well for some cases, their exposure histories could only be obtained from family or friends as the case was too unwell to be interviewed.

Cases identified through review of earlier notifications did not have isolates available to compare with cases identified during the outbreak, as the additional measures necessary to obtain isolates are not routinely employed in non-outbreak settings. Thus cases 5 and 6 did not have isolates available for characterisation, so cannot be conclusively linked to the March outbreak. Similarly isolates were not able to be obtained from six of the cases notified during March and May as either these cases were not able to provide a sputum sample or a sputum sample was obtained but *Legionella* was not able to be grown on culture, so these remain linked to the outbreak only by exposure time and place.

In addition, conclusions reached by genome sequencing analysis are based on a limited number of LP1 isolates. Whole genome sequencing remains a relatively new approach to characterising *Legionella*. It is possible that with the gradual uptake of genome sequencing and corresponding increase in the number of LP1 genomes in public databases internationally, our confidence in inferring or excluding epidemiological links from genomic similarity will also increase.

Conclusions and Outcomes

An extensive epidemiological, environmental and laboratory investigation was undertaken in response to two clusters of Legionnaires' disease associated with the Sydney Central Business District in March and May 2016. Fifteen people were notified with Legionnaires' disease due to *Legionella pneumophila* serogroup 1. Of these, two were shown to have unique and different strains, and were not strongly linked epidemiologically to the other cases. Five people were shown to have the same strain by whole genome sequencing and that strain was matched to the strain identified in two cooling towers. Epidemiological investigations suggest that the remaining eight were also linked to the outbreak. While the actual source of the human cases cannot be proven, the response by NSW Health, City of Sydney and building owners controlled the risk through identifying and cleaning contaminated water cooling systems.

In May 2016 the NSW Chief Health Officer convened an expert panel to develop advice on additional measures that could be adopted to reduce the risk of *Legionella* outbreaks in NSW in the future.

Appendix

Appendix 1.1. Time line of events CBD 1 March 2016

DATE	EVENT	COMMENTS
7 March	Case 1 notified by NS PHU	
8 March	1400 Cases 2 & 3 notified by SES PHU 1630 Case 4 notified QLD Health	
9 March	0900 PHEOC operational	Daily teleconferences begin
	1000 ICPMR & FASS alerted	FASS received 31 samples
	1030 Jurisdiction/CDNA alerted	
	NSW Public Health Units alerted	
	1100 Town Hall operations centre opened	EHO command centre
	1200 Alerts sent to GPs EDs Clinicians	
	Case 5 identified	Through look back previous notifications
	1430 LHD CE's alerted	
	Integra report positive LP1 results from Pitt Street tower from the 25 Feb 2016	
	1700 Environmental testing complete Day 1. Area bound by Clarence, King, Park and Pitt Streets	
	1830 NSW Laboratory Network alerted	
10 March	Environmental testing day 2	62/64 registered and 43/49 unregistered systems sampled FASS received 53 samples
11 March	Cases 6 & 7 notified by PHU	
	Case 8 identified	Further look back of cases and exposures
	Environmental investigation expanded to include Clarence, King, Park and Elizabeth Streets	40/60 additional systems sampled FASS received 80 samples
12 March	Possible case notified	Case excluded due to alternative diagnosis
13 March	Case 9 notified	
14 March	Case 9 passed away	
	Environmental investigation expanded to include Kent, King, Elizabeth and Bathurst Streets	FASS received 31 samples
15 March	Environmental sampling complete	FASS received 4 samples (Total 199 samples sent to FASS)

Appendix 1.2. Time line of events CBD 2 May 2016

DATE	EVENT	COMMENTS
28 April	Case 1 notified by NS PHU	
3 May	Cases 2 & 3 notified by SES PHU and WS PHU	
4 May	1000 ICPMR & FASS alerted	Daily teleconferences begin
	Jurisdictions/CDNA alerted	
	NSW Public Health Units alerted	
	Alerts sent to ED Clinicians	
	LHD CE's alerted	
	Environmental testing complete Day 1. Area bound by Margaret, George, King and Kent Streets	57 environmental samples collected and sent to FASS
	NSW Laboratory Network alerted	
	1430 Environmental testing area expanded to be bound by Pitt Street, George Street, King Street and Curtin Place (which is in line with Margaret Street).	
5 May		FASS received 35 samples
6 May	Case 4 & 5 notified by Sydney PHU and SWS PHU	
9 May	Case 6 notified by Victoria Health	FASS received 6 samples
10 May		FASS received 28 samples
11 May	Environmental testing completed	FASS received 4 samples (total 130 samples to FASS)

Appendix 2.1. Line list of confirmed LP1 cases – CBD March 2016

CASE	ONSET DATE	SEX	AGE	STATUS	LUAG	PCR	CULTURE	EXPOSURES
1	02/03/16	Male	60's	Alive	Positive	Detected	Not grown	All across Sydney
2	03/03/16	Male	30's	Alive	Positive	Detected	LPI isolated	Cycles Oxford Pitt and, Kent streets
3	27/02/16	Male	60's	Alive	Positive	Detected	Not grown	State Theatre, Market St, Town Hall, catches bus from Prince Alfred Park
4	01/03/16	Male	60's	Alive	Positive	Detected	Not grown	Tourist Hop on /off bus
5	08/02/16	Male	40's	Alive	Positive	Not done	Not grown	Works Liverpool Street, Visits World Square, Town Hall Pitt and Park Streets, Myers
6	24/12/15	Male	80's	Alive	Positive	Not done	Not grown	City Tattersalls Club catches train from Town Hall 6/7 days
7	04/03/16	Male	50's	Alive	Positive	N/A	Not grown	Works Goulburn Street, Walked Dixon street to Town Hall 25/2
8	02/03/16	Male	40's	Alive	Positive	Detected	LPI isolated	Central Station 27/2
9	01/03/16*	Male	80's	Deceased	Positive	Detected	LPI isolated	George Street and Castlereagh Street on 23/2. Also visited Pitt Street, Castlereagh St on 25/2.

*Onset date approximate

Appendix 2.2. Line list of confirmed LP1 cases – CBD May 2016

CASE	ONSET DATE	SEX	AGE	STATUS	LUAG	PCR	CULTURE	EXPOSURES
1	25/04/16	Male	80's	Alive	Positive	Detected	LPI isolated	Works Clarence St. Arrives and leaves by taxi.
2	28/04/16	Female	30's	Alive	Positive	Detected	Not grown	Works Shelley St. Walked through Ix Area twice a day.
3	28/04/16	Female	30's	Alive	Positive	Detected	LPI isolated	Works George St. Walks from Wynyard Station
4	29/04/16	Male	80's	Died	Positive	Detected	LPI isolated	Restricted to Circular Quay on one day (24/4).
5	01/05/16	Male	50's	Alive	Positive	Detected	LPI isolated	Stayed in CBD overnight (George St) for Anzac Day march (Elizabeth St, Hyde Park). Walked through Ix Area down to Circular Quay.
6	03/05/16	Male	40's	Alive	Positive	Not done	Not grown	York St. (Stayed in York St Hotel 25-29/4). Walks around the block.

Abbreviations: LUAG = Legionella urinary antigen for *L. pneumophila*. PCR = polymerase chain reaction for *L. pneumophila*.

Appendix 3.1. Line list of cooling towers with positive presumptive result >10 cfu* CBD March 2016

PREMISES	INSPECTION DATE	SAMPLE DATE	PRESUMPTIVE INITIAL RESULT (TOTAL LEGIONELLA CFU/ML)	ACTION TAKEN	RE-INSPECTION DATE	DATE OF ADDITIONAL SAMPLES TAKEN	DATE OF FINAL RESULT	FINAL RESULTS - CFU / ML				
								TOTAL LEGIONELLA	L. PNEUMOPHILA SGI	SG 2-14	OTHER LEG SP.	
York St (site 1)	09/03/2016	09/03/2016	Approx. 180	SESPHU Immediate Response (Verbal Direction issued followed up by S33)	SESPHU 29/03/2016	N/A	16/03/2016	180	180	<10	<10	<10
York St (site 2)	09/03/2016	09/03/2016	Approx. 2500	CoS issue I.N.	SESPHU 16/03/2016	16/03/2016	16/03/2016	2500	2500	<10	<10	<10
Clarence St (Site 1)	09/03/2016	09/03/2016	Approx. 60	CoS issue letter to improve maintenance	CoS responsibility for ongoing follow up	N/A	16/03/2016	70	10	<10	<10	60
York St (site 3)	10/03/2016	10/03/2016	Approx. 350	SESPHU Immediate Response Section 33 followed by CoS Issue I.N.	SESPHU 29/03/2016	N/A	16/03/2016	350	350	<10	<10	<10
			Approx. 410		SESPHU 29/03/2016	N/A	16/03/2016	410	410	<10	<10	<10
			Approx. 450		SESPHU 29/03/2016	N/A	16/03/2016	450	450	<10	<10	<10
Pitt St (site 1)	10/03/2016	10/03/2016	Approx. 20	CoS issue letter to improve maintenance	CoS responsibility for ongoing follow up	N/A	16/03/2016	20	<10	<10	<10	20
George St (site 1)	10/03/2016	10/03/2016	Approx. 100	SESPHU Immediate Response Section 33 followed by CoS Issue I.N.	SESPHU 29/03/2016	29/03/2016	17/03/2016	110	110	<10	<10	<10
			Approx. 140		SESPHU 29/03/2016	N/A	17/03/2016	140	140	<10	<10	<10
Pitt St (site 2)	10/03/2016	10/03/2016	Approx. 670	SESPHU issue P.O. CoS assist to deliver	SESPHU 21/03/and 29/03/2016	21/03/2016	17/03/2016	670	670	<10	<10	<10
Druitt St	10/03/2016	10/03/2016	Approx. 90	CoS issue letter to improve maintenance	CoS responsibility for ongoing follow up	N/A	17/03/2016	100	100	<10	<10	<10
Elizabeth St	11/03/2016	11/03/2016	Approx. 100	CoS issue letter to improve maintenance	CoS responsibility for ongoing follow up	N/A	17/03/2016	<10	<10	<10	<10	<10
Castlereagh St	11/03/2016	11/03/2016	Approx. 100	CoS issue letter to improve maintenance	CoS responsibility for ongoing follow up	N/A	17/03/2016	100	100	<10	<10	<10
Park St	11/03/2016	11/03/2016	Approx. 90	CoS issue letter to improve maintenance	N/A	N/A	17/03/2016	<10	<10	<10	<10	<10
George St (site 2)	09/03/2016	09/03/2016	Approx. 10	CoS issue letter to improve maintenance	CoS responsibility for ongoing follow up	N/A	16/03/2016	10	10	<10	<10	<10

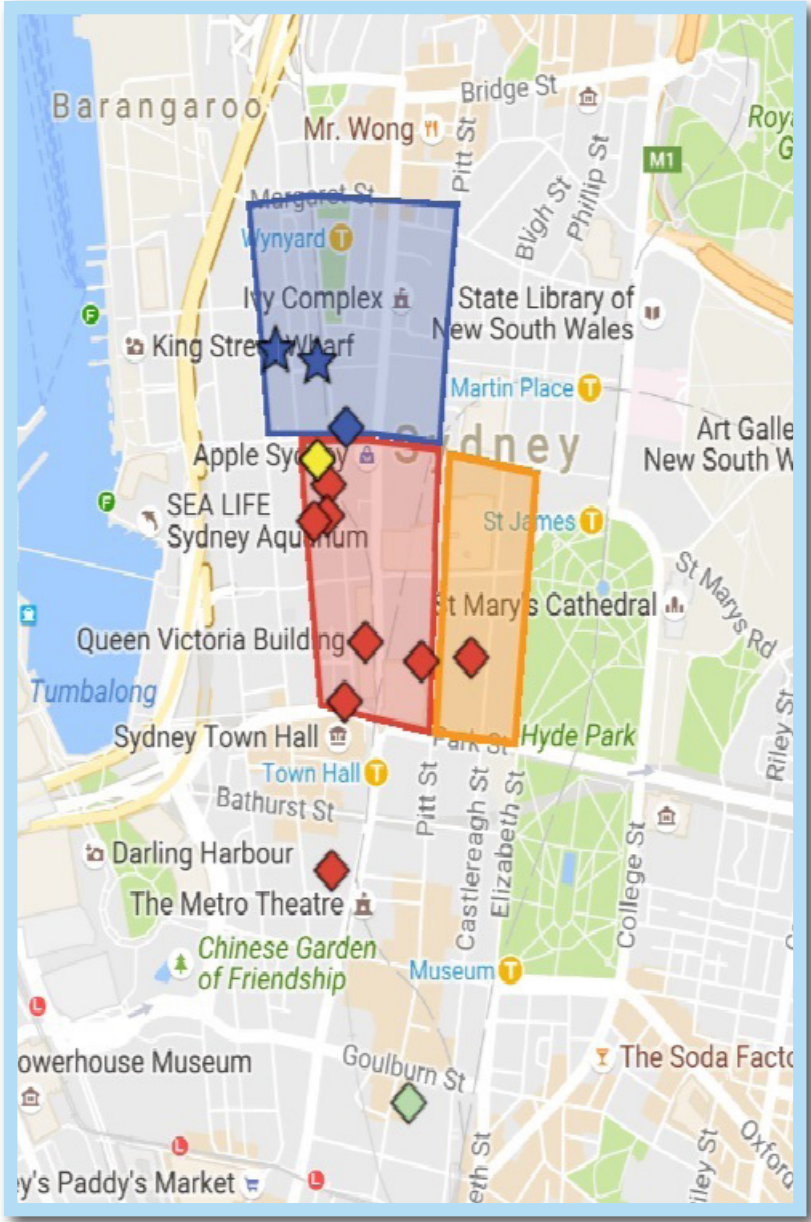
Abbreviations: City of Sydney (CoS); South Eastern Sydney Public Health Unit (SESPHU); Improvement Notice (I.N.); Prohibition Order (P.O.)
 *>10 cfu/mL = more than 10 colony forming units of Legionella species per milliliter of water

Appendix 3.2 Line list of cooling towers with positive presumptive result >10 cfu* CBD May 2016

PREMISES	INSPECTION DATE	SAMPLE DATE	PRESUMPTIVE INITIAL RESULT (TOTAL LEGIONELLA CFU/ML)	ACTION TAKEN	RE-INSPECTION DATE	DATE OF ADDITIONAL SAMPLES TAKEN	DATE OF FINAL RESULT	FINAL RESULTS - CFU / ML			
								TOTAL LEGIONELLA	L. PNEUMOPHILA SGI	OTHER LEG SP. SG 2-14	<10
King St	04/05/16	04/05/16	Approx. 190	Improvement notice issued by CoS	N/A	N/A	11/05/16	190	190	<10	<10
Clarence St (Site 2)	04/05/16	04/05/16	Approx. 9800	Prohibition order issued by SESPHU	16/05/16	16/05/16	11/05/16	9800	9800	<10	<10
York St (Site 3)	04/05/16	04/05/16	Approx. 50	Improvement notice issued by CoS	N/A	N/A	11/05/16	60	60	<10	<10
York St (Site 4)	04/05/16	04/05/16	Approx. 20	Improvement notice issued by CoS	N/A	N/A	11/05/16	20	20	<10	<10
Shelley St	05/05/16	05/05/16	≥10 - <100	CoS issue letter to improve maintenance	N/A	N/A	12/05/16	<10	<10	<10	<10
Alfred St	09/05/16	09/05/16	= 10 - 100 (-20)	CoS issue letter to improve maintenance	N/A	N/A	15/05/16	<10	<10	<10	<10

Abbreviations: City of Sydney (CoS); South Eastern Sydney Public Health Unit (SESPHU);
 *>10 cfu/mL = more than 10 colony forming units of *Legionella* species per milliliter of water

Figure 1. Map of investigation areas and positive cooling towers, March and May 2016 outbreaks



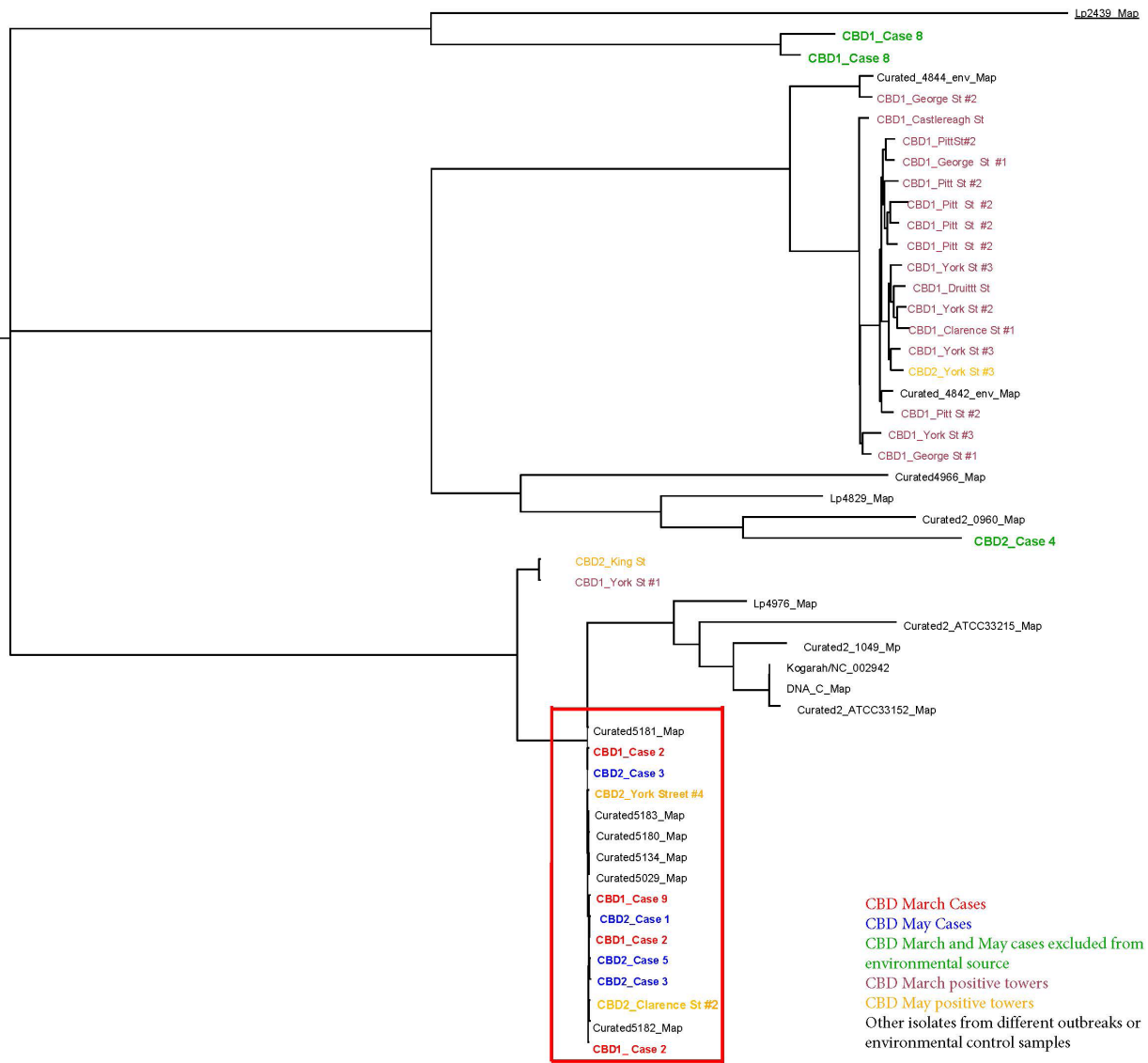
Map data ©2016 Google.

Key

- ◆ Positive CT - March outbreak
- ◆ Positive CT - May outbreak
- ★ Positive CT - May outbreak with outbreak sequence
- ◆ Positive CT - both outbreaks
- ◆ Positive CT - reported by a water treatment company
- March outbreak - expanded investigation area
- March outbreak - original investigation area
- May outbreak - investigation area

Note: In the March outbreak two locations had more than one cooling tower on their site which tested positive.

Figure 2. Whole Genome Sequencing results 2016



A total of 48 LP1 isolates from patients and cooling towers (including isolates not related to the CBD outbreak) were subjected to whole genome sequencing at CIDM- Public Health, ICPMR using the Illumina Nextseq 500 instrument. The main cluster of genomically similar LP1 strains from five cases (two from March and three from May) and two environmental samples from cooling towers is highlighted in red. This cluster also includes samples from one case and three environmental samples not geographically or epidemiologically related to the CBD. Isolates from the clinical cases judged to not be associated with the outbreak are highlighted in green (two isolates from case 7 CBD March and case 4 CBD May). Other CBD environmental isolates clustered together in several groups, but not with any patient samples.

