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Risk Factors for Contamination of Domestic Hot Water Systems by Legionellae

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To assess risk factors associated with the contamination of the domestic environment by legionellae, 211 houses in the Quebec City area were randomly selected and water samples were collected from the hot water tank, the shower heads, and the most frequently used faucet. After centrifugation, concentrated samples were seeded in triplicate on BCYE and GPV media. Data on the characteristics of the hot water system and plumbing in the house and on the personal habits of the occupants were collected for each house. Among these 211 houses, hot water was provided by either an oil or gas heater in 33 and by an electric heater in 178. Legionellae were isolated from none of the samples from houses with oil or gas heaters and from 39% (69 of 178) of those with electric water heaters ($P < 0.0001$). This association remained highly significant after control for water temperature and other variables in a stratified analysis. In the 178 houses with an electric heater, 12% of the faucets, 15% of the shower heads, and 37% of the water heaters were contaminated. *Legionella pneumophila* serogroups 2 and 4 were the most frequently isolated strains. Logistic regression showed that factors associated with electric water heater contamination were (i) location of the house in older districts of the city ($P < 0.0001$), (ii) old age of the water heater ($P = 0.003$), and (iii) low water temperature ($P = 0.05$). Contamination of the water heater was the only factor significantly associated with the contamination of peripheral outlets ($P < 0.0001$). This study shows that the presence of an electric heater is strongly associated with contamination of domestic hot water systems by Legionellae. The public health importance of this contamination is still unknown.

Bacteria of the family *Legionellaceae* can be found in both natural and man-made environments. Legionellae of all types have been isolated from natural environments in different areas of the world (12, 14, 26, 31, 32, 34). These studies suggest that the distribution of legionellae is worldwide and that their concentration is directly related to water temperature (14, 19). Despite this relationship with temperature, legionellae have been recovered in areas with cold climates where rivers may be frozen for as long as 6 months (26).

Man-made environments are thought to act as either amplifiers or disseminators of legionellae, and the study of their contamination is thus of extreme importance in understanding the epidemiology of Legionnaires disease (15). Cooling towers were the first man-made environments from which legionellae were isolated (11). Although cooling towers are strongly suspected as a factor in the Philadelphia outbreak (16), the relationship between contaminated cooling towers and disease has been demonstrated only in a few instances. Epidemiologic evidence of such a relationship has been reported (11), and *Legionella pneumophila* organisms of similar subtypes were identified in epidemic cases of legionellosis as well as in the incriminated cooling tower (28).

The first evidence that potable water might be associated with legionellosis was reported by Tobin et al., who obtained two *L. pneumophila* serogroup 6 isolates from patients in a renal graft unit and isolated similar strains from shower-bath mixers in the same unit (45). Nosocomial legionellosis has been frequently related to contaminated potable water supplies, especially for the different serogroups of *L. pneumophila* (8, 9, 20, 36, 40, 42, 43). In these outbreaks, the hot water system was the most frequently involved. It is currently believed that hot water storage devices are the breeding ground for legionellae (1, 38, 44, 46, 49), although dead-end loops and stagnation in the plumbing system have also been suggested as potential reservoirs and amplifiers for these bacteria (5).

Legionellae are by no means restricted to the hospital potable water supply. For instance, workplace contamination has been linked to at least two outbreaks of Pontiac fever (17, 21), and hotels have also been clearly linked to Legionnaires disease in a few instances (16, 37, 44). Finally, contamination of the domestic environment by *L. pneumophila* has frequently been observed (10, 10a, 27) and has been associated with some cases of Legionnaires disease (2, 4, 41).

Factors that might be associated with contamination of the domestic environment have not been extensively studied. In two previous studies, legionellae were isolated in water samples from electric water heaters but not from fossil fuel heaters (27, 50). The objectives of the present study were to estimate the frequency of contamination by legionellae of different domestic hot water devices (as assessed by culturing samples) and to examine the possible relationship of environmental factors (especially the type of water heater) to contamination.

MATERIALS AND METHODS

Selection of houses. All the houses included in this study were privately owned, occupied by their owner, and located within the boundaries of a suburban municipality located in the Quebec City area. This municipality was selected because it is supplied by one water treatment plant and is large

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enough to provide an adequate sampling scheme. In this municipality, the water is treated by flocculation, chlorination, and ozonation.

From the municipal tax list, a systematic random sample was compiled. The first house was selected by using a random-number table. Thereafter, each fifth house that satisfied the above-mentioned criteria was eligible for the study. The name, address, and telephone number of the owner of each selected house were noted. These persons were then contacted by a letter briefly informing them of the nature of the study. Within 1 week a research nurse visited them and solicited their participation in the study. At the time of this visit, in addition to water sampling, a questionnaire was completed by the nurse with the owner or spouse. The questionnaire was used to gather information about the water heater (brand name and type [electric, oil, or gas], year of installation, year of last cleanup, capacity of the reservoir (imperial gallons), power of the upper and lower elements for electric heaters, and power of the oil and gas heaters), the house (age of the house, age of the plumbing, type of plumbing [copper, ferrous metal, etc.], diameter of the inlet and outlet water piping, distance between the water heater and municipal main water line, presence of aerators at the sampled faucet, and location of the house within the city [district]), and the inhabitants of the house (number, ages, and hot water consumption habits [number of showers, baths, dishwasher cycles, etc.]).

Sampling within the domestic environment. In each house, water samples were taken from the water heater, the shower heads, and the most frequently used faucet. For the water heater, the first 500 ml of water obtained from the drainage valve was used. The temperature of an aliquot of this sample was measured. Similar samples (including measurement of temperature) were taken from all shower heads and from the selected faucet. In addition, scrapings of the inner parts of shower heads were taken. The temperature was also measured after allowing the hot water alone to flow from the selected faucet for 3 min.

Sterile bottles were used throughout the study, and, to avoid cross-contamination within and between houses, an additional aliquot of water was collected when the temperature was measured. In all instances, these additional water samples were discarded. Water samples were transported to the laboratory and stored at 4°C for a maximum of 12 h prior to culture.

Culture, isolation, and quantitation of legionellae. The two media that were used in this study were buffered charcoal-yeast extract medium supplemented with α-ketoglutarate (BCYE medium [33]) and BCYE medium supplemented with 3 mg of glycine per ml, 100 U of polymyxin B per ml, and 5 μg of vancomycin per ml, GPV medium [48]). Each lot of culture medium was tested for sterility prior to use and was also evaluated for its ability to support the growth of a wild-type strain of *L. pneumophila*.

Each of the water samples was concentrated 100-fold by centrifugation for 30 min at 5,000 × *g*. Thereafter, 100 μl of the concentrated sample and 5 10-fold dilutions were inoculated in triplicate on each of the two culture media. Plates were incubated at 35 ± 2°C for 7 days and were examined on days 1, 5, and 7. Bacteria (legionellae and nonlegionellae) were counted on day 7. Colonies with a morphology similar to that of the family *Legionellaceae* were subcultured on BCYE medium and on blood agar plates; those that grew on the former but not on the latter were further characterized and tested by direct immunofluorescent-antibody techniques as previously described (26). Finally, bacteria that were not stained with these antisera were tested with a commercial monoclonal antibody (Genetic Systems Co., Seattle, Wash.) that is specific for all serogroups of *L. pneumophila* (18).

Scrapings of the interior of the shower heads were transported to the laboratory in 50-ml conical tubes that contained 40 ml of water obtained from the sampled shower. These were processed as above except that the final concentration obtained was 16-fold. Bacterial counts were adjusted accordingly.

Statistical analysis. All of the following analyses were performed for contamination of at least one device (heater or peripheral), of the water heater, of at least one peripheral device, of the most frequently used water outlet, and of at least one shower head.

The two-tailed Fisher exact test and chi-square test were used to compare proportions of contamination over categorized variables. Student's *t* test and the Wilcoxon rank sum test were used to compare mean values of continuous variables according to the presence of legionellae in the different devices. Stratified analysis, using the technique described by Mantel and Haenszel (30), or logistic regression (22) was used to assess the independent contribution of each studied variable to contamination. Because many of these variables were correlated and because of the inherent multicolinearity problem in the multivariate analysis, a careful examination of the joint distribution of these variables was done first. When two or more variables were found to be highly correlated, only one of them was kept for multivariate analysis. Statistical interactions were also evaluated. The goodness of fit of the final logistic regression models was assessed by using the Hosmer-Lemeshow test (22).

Geometrical means (computed with $\log_{10}$) of the number of CFU per milliliter of the original water samples were computed. Multiple linear regression was used to evaluate the importance of independent variables in relation to bacterial counts of legionellae and other bacteria.

RESULTS

Culture results for samples from the houses. A total of 316 households were randomly selected. From these, 211 (66.8%) householders agreed to participate in the study and 970 water samples were collected. One sample from the water heater and one from the most frequently used faucet were taken in each house, whereas a total of 274 water samples and a similar number of scrapings were obtained from showers located in 209 houses (2 of the 211 did not have any shower, 57 had two showers, and 4 had three showers).

A total of 69 houses (32.7%) had at least one sample that was positive by culture for the family *Legionellaceae*. Of these 69 contaminated houses, 36 (52.2%) had a contaminated hot water reservoir only, 30 (43.5%) had both a contaminated hot water reservoir and a contaminated peripheral outlet (shower or faucet), and 3 (4.3%) had a contaminated peripheral outlet only. In 18 of the 30 properties (60.0%) where legionellae were found by culture in at least one outlet and the reservoir, the same species or serogroups were found in both instances. In seven houses (23.3%), similar species or serogroups were found in both sites, but additional strains were found in one or both locations. In three houses (10.0%), there was no correlation between the species or serogroups identified from different sites. Finally, two houses (6.7%) were excluded from this correlation analysis because the bacterial strains were lost prior to species identification.

If the same serogroups of *L. pneumophila* identified in a

TABLE 1. Distribution of *Legionella* species and serogroups isolated in the studied houses[a]

Species and serogroups	No. (%) isolated
L. pneumophila serogroup 1	10 (9.5)
L. pneumophila serogroup 2	22 (21.0)
L. pneumophila serogroup 3	11 (10.5)
L. pneumophila serogroup 4	26 (24.8)
L. pneumophila serogroup 5	4 (3.8)
L. pneumophila serogroup 6	11 (10.5)
L. pneumophila serogroup 7	0 (0.0)
L. pneumophila serogroup 8	14 (13.3)
L. pneumophila other serogroup[b]	3 (2.9)
L. longbeachae	1 (1.0)
L. micdadei	1 (1.0)
Unknown *Legionella* spp.[c]	2 (1.9)
Total	105 (100.0)

[a] When two or more samples within a given house contained the same serogroups or species, the isolates were considered identical strains.
[b] *L. pneumophila* that did not react with antibodies against serogroups 1 to 8 but with a species-specific monoclonal antibody (Genetic Systems Co., Seattle, Wash.).
[c] Strain lost prior to identification.

given house are considered to be a single strain, 105 different strains were isolated from the 69 contaminated houses (39 houses with one strain, 25 houses with two strains, 4 houses with three strains, and 1 house with four strains). *L. pneumophila* serogroups 2 and 4 were by far the most frequently isolated species (Table 1).

Correlation between contamination by legionellae and type of water heater. Of the 211 houses studied, 33 (15.6%) had an oil or gas water heater and 178 (84.4%) had an electric water heater. Results of cultures showed that none of the houses with an oil or gas water heater harbored legionellae. In the crude analysis, contamination at any studied site was significantly associated with the presence of an electric heater (Table 2).

Additional variables were associated with the contamination of water heaters by these bacteria. Houses located in districts urbanized in the last 20 years were less likely to be contaminated than those in older districts. Indeed, whereas 47.9% of households located in the older districts were contaminated, only 17.9% of those in the newer districts were colonized (chi-square 1 df = 21.71; $P = 0.000003$). A low temperature of the water at the bottom of water heater, a low temperature of the water at the faucet, and old plumbing were also associated with contamination (P = 0.0001, 0.0001, and 0.009, respectively, by Student's t test).

TABLE 3. Variables associated with the type of water heater[a]

Variable	Value for type of water heater: Electric	Value for type of water heater: Fossil fuel	P^b
Mean age of plumbing (yr) ± SE	20.3 ± 0.9	23.3 ± 1.1	0.032
Mean temp at faucet (°C) ± SE	56.6 ± 0.4	61.5 ± 1.1	0.0002
Mean temp at bottom of heater (°C) ± SE	30.3 ± 0.6	49.2 ± 1.2	0.0001

[a] There were 178 houses with an electric heater and 33 with a fossil fuel heater.
[b] Student's t test. Similar results were obtained with the Wilcoxon rank sum test.

These three variables were also highly associated with the type of water heater (Table 3).

Modeling with logistic regression was impossible because oil and gas heaters were never contaminated with legionellae. Stratified analysis was therefore used, but because of the small sample size, it was impossible to stratify simultaneously on numerous variables. By stratifying individually for each of the other variables that were collected (quartiles were use for stratification on continuous variables), we examined the influence of the type of water heater on contamination by legionellae. In all cases, the association between the presence of an electric water heater and contamination of any device remained significant after stratification for any other collected variable ($P < 0.05$ in all cases).

Special attention was given to variables that were highly associated with contamination or that were simultaneously associated with contamination and type of heater. Two additional stratified analyses were therefore performed: simultaneous stratification for district (old versus new) and quartiles of temperature at the bottom of the heater as well as simultaneous stratification for district and quartiles of temperature at the faucet (both analyses were performed over eight strata). The association between the type of heater and its contamination remained highly significant in these two analyses (Mantel-Haenszel chi-square with 1 df of, respectively, 6.96 [$P = 0.008$] and 15.27 [$P = 0.00009$]). Similar results were found for the contamination of peripheral outlets (Mantel-Haenszel chi-square with 1 df of, respectively, 5.32 [$P = 0.021$] and 5.95 [$P = 0.015$]).

Since the plumbing systems were older in contaminated houses but newer in those with an electric heater, no additional analysis was done to control for age of plumbing in the correlation between type of heater and contamination.

TABLE 2. Distribution of culture-positive samples according to the type of water heater[a]

Culture	No. of culture-positive samples in: Water heaters		Faucets		Shower heads		Houses	
	Oil or gas heater	Electric heater	Oil or gas heater	Electric heater	Oil or gas heater	Electric heater	Oil or gas heater	Electric heater
Positive	0	66	0	22	0	26[b]	0	69
Negative	33	112	33	156	33	150	33	109
Total	33	178	33	178	33	176[c]	33	178

[a] By the two-tailed Fisher exact test, $P < 0.001$ for heaters, 0.020 for faucets, 0.018 for shower heads, and <0.001 for houses.
[b] Scrapings and water samples from shower heads were contaminated in 6 and 25 houses, respectively. Thus, scrapings were positive and water was negative in only one house.
[c] Two houses with an electric heater did not have any shower.

TABLE 4. Categorical variables associated with the contamination of houses with an electric water heater by legionellae

Variable	Value for water heater		Value for peripheral outlet	
	% (no.) contaminated	P^a	% (no.) contaminated	P^a
District				
Old	58.4% (77)		31.2% (77)	
New	19.8% (101)	<0.0001	8.9% (101)	0.0002
Power of upper heating element (kW)[b]				
3.0	50.0% (66)		21.2% (66)	
3.8 or 4.5	29.7% (111)	0.007	17.1% (111)	0.50
Power of lower heating element (kW)[b]				
1.5 or 3.0	49.2% (65)		21.5% (65)	
3.8 or 4.5	30.3% (109)	0.012	16.5% (109)	0.41
Vol of heater (gal)				
40	49.2% (65)		21.5% (65)	
60	30.1% (113)	0.011	16.8% (113)	0.43

[a] Chi-square with 1 df.
[b] Treated as a categorical variable because these powers were either 1.5, 3.0, 3.8, or 4.5 kW. Categories were collapsed because there were only two heaters with a 3.8-kW element and one with a 1.5-kW element. Numbers for these variables do not add up to 178 because of missing values.

Correlation of contamination by legionellae with other factors. The influence of different factors on the contamination of any heater was examined mainly to assess confounding of the association of contamination with the type of heater. However, since only houses with an electric water heater were contaminated by legionellae, the specific analysis of the influence of these other factors on contamination was restricted to these 178 houses.

Tables 4 and 5 show the factors that were associated with the contamination of at least a peripheral outlet and of the water heater. All other factors listed in Materials and Methods were not significantly related to contamination. The only variable that was associated with the contamination of all types of water heater that is not associated with the contamination of electric heaters is the water temperature at the bottom of the heater. It is interesting that although water temperatures at the bottom of the heater and at the faucet were highly correlated in houses with a fossil fuel heater ($r = 0.54$, $P = 0.001$), no significant correlation was observed between these two variables in houses with an electric heater ($r = 0.12$, $P = 0.1$). Other variables that are specific to electric heaters (power of upper and lower elements), as well as the volume of the heater, emerged as significant factors for contamination.

Logistic regression was first used to determine variables that were independently associated with contamination of electric heaters by legionellae. The age of the plumbing and the age of the house were highly correlated continuous variables ($r = 0.75$). In addition, the power of the lower and upper elements was always in the highest category when the volume of heater was 60 imperial gallons (273 liters). Taking into account these strong correlations, the following variables were included in the logistic regression model: district, temperature at the faucet, age of the water heater, age of the plumbing system, volume of the heater, and water temperature at the bottom of the heater. The last variable was included, although it was not associated in the crude analysis, because it was associated with contamination when all heaters were considered. No significant interaction could be found between these variables.

Only three variables remained significantly associated with heater contamination in this model: old district, low temperature at the faucet, and old age of the water heater. The temperature at the bottom of heater was not a significant factor (as in the crude analysis restricted to electric heaters). All other variables included in the model did not remain significant because they were associated with the localization of the house: old plumbing systems and small water heaters were found more often in houses located in old districts. Since coefficients of the three associated factors did not change when other variables were dropped from the model, only district, temperature at the faucet, and age of the heater were kept in the final model (Table 6). This logistic regression model fitted very well to the data when the goodness of fit was assessed by the Hosmer-Lemeshow test (chi-square 8 df = 7.01; $P = 0.54$).

Since only three contaminated houses did not have a contaminated water heater, results of the multivariate analysis for the contamination of at least one device were similar to those presented above.

Finally, the same three factors associated with the contamination of electric heaters were also linked to contamination of peripheral outlets. However, since contamination of peripheral outlets was much more frequent in houses with a contaminated electric heater than in other houses (30/66 = 45.5% versus 3/112 = 2.7%, $P < 0.0001$ by Fisher's exact test), these associations might only be a consequence of this strong association between contamination of the heater and contamination of the outlet. We therefore made an analysis of factors associated with the contamination of outlets restricted to houses with a contaminated heater. Stepwise logistic regression did not select any variable that was associated with peripheral outlet contamination in this restricted sample. However, the mean water temperature at the faucet was lower in houses with a contaminated peripheral outlet (54.8°C) than in others (55.6°C). This difference

TABLE 5. Continuous variables associated with the contamination of houses with an electric water heater by legionellae[a]

Variable	Value for water heater		P^b	Value for peripheral outlet		P^b
	Contaminated	Not contaminated		Contaminated	Not contaminated	
Mean age of the house (yr) ± SE	25.9 ± 1.3	19.9 ± 1.3	0.002			
Mean age of plumbing (yr) ± SE	23.6 ± 1.3	18.2 ± 1.1	0.002			
Mean age of the heater (yr) ± SE	10.1 ± 0.7	6.4 ± 0.5	0.0001	10.3 ± 1.3	7.2 ± 0.5	0.010
Mean temp at the faucet (°C) ± SE	55.3 ± 0.6	57.3 ± 0.5	0.008	54.3 ± 0.8	57.1 ± 0.4	0.002

[a] There were 66 contaminated heaters and 33 contaminated outlets out of 178.
[b] Student's *t* test. Similar results were obtained with the Wilcoxon rank sum test.

TABLE 6. Variables associated with the contamination of electric water heaters in a logistic regression model

Variable	Median value within quartile	n[a]	Crude % of contamination[b]	Adjusted % of contamination[c]
District				
New		101	20.8%	21.4%
Old		77	58.4%	55.4%
β[d]			1.678	1.515
SE[e]			0.245	0.353
P[f]			<0.001	<0.001
Age of heater (yr)				
Quartile 1	2.0 yr	40	15.0%	14.7%
Quartile 2	5.0 yr	39	33.3%	34.1%
Quartile 3	9.5 yr	40	37.5%	32.5%
Quartile 4	14.0 yr	38	60.5%	57.0%
β			0.128	0.111
SE			0.034	0.037
P			<0.001	0.003
Water temp at faucet (°C)				
Quartile 1	51.7°C	47	51.1%	39.9%
Quartile 2	55.5°C	44	52.3%	55.3%
Quartile 3	57.9°C	45	24.4%	28.8%
Quartile 4	61.6°C	42	19.1%	14.6%
β			−0.092	−0.071
SE			0.035	0.036
P			0.008	0.049

[a] Total number of observations within each category. Numbers differ slightly within quartiles of continuous variables because of multiple observations at cutoff values between quartiles. There were 21 missing values for the age of the heater.

[b] Observed proportions of contamination in each category.

[c] Adjusted by using a logistic regression model with the other variables at their mean value. The model used for a given variable was the one with this variable treated as categorical and the other ones treated as continuous (except for the district, which is always dichotomous).

[d] Coefficient of the given variable in the model with the district as dichotomous and the two other variables as continuous. The crude β is that of the univariate logistic regression model, whereas the adjusted β is that of the multivariate model.

[e] Standard error of β.

[f] Wald statistic in the logistic regression model.

was not statistically significant ($P = 0.21$ by the Wilcoxon rank sum test).

Bacterial counts and multiple linear regression of the level of contamination. As expected, contamination by bacteria other than legionellae was more common on BCYE medium than on GPV medium. Indeed, whereas these other bacteria grew on 856 of the 970 BCYE plates (88.2%), they were present on only 331 of the 970 (34.1%) GPV plates. In addition, the average number of CFU was 2.95/ml (range, 0.1 to 14,802) for contaminated GPV media and 17.70/ml (range: 0.1 to 122,965) for contaminated GPV media ($P < 0.0000001$ by Student's t test). Despite this, legionellae were identified as often on BCYE medium (106/970 = 10.9%) as on GPV medium (103/970 = 10.6%), and bacterial counts of legionellae were similar on both media (geometrical mean of 16.2 and 18.3 CFU/ml for GPV medium and BCYE medium respectively; $P = 0.62$ by Student's t test).

Because of their inherent differences, multiple linear regression models had to be made independently for GPV and BCYE media. This further reduced the number of positive samples in each analysis. Therefore, because of the small number of contaminated faucets and shower heads, multiple regression analysis on the level of contamination in these devices was impossible to realize. Only contaminated water heaters were submitted to this type of analysis. No variables (including bacterial counts of other bacteria) could be associated with *Legionella* bacterial counts in contaminated electric water heaters. Similarly, the level of contamination by other bacteria could not be associated with any of the collected variables.

DISCUSSION

This study shows that bacteria of the family *Legionellaceae* are common in domestic water distribution systems. Indeed, nearly one-third of all domestic water heaters were contaminated by these bacteria. In addition, over 15% of the studied houses contained these bacteria in at least one sample taken at the faucet or the shower. Although one-third of selected householders refused to participate in the study, these proportions could probably be generalized to the entire studied city. Indeed, the distribution of factors associated with contamination by legionellae in selected houses whose owners declined to participate was similar to those in the houses that were visited. Hence, the proportion of studied houses equipped with an oil or gas heater (15.6%) was similar to that reported from other sources for all houses located in the same area (15.8% [24]). In addition, the mean age of the selected houses whose owners declined to participate was similar to that of the studied houses. Finally, their distribution over city districts was also similar, as was the age distribution of water heaters (24).

The single most important factor that could explain contamination of the domestic environment was the type of water heater. Indeed, none of the houses with fossil fuel heaters was found to be contaminated by legionellae, whereas nearly 40% of those with electric heaters were contaminated. This finding confirms results found in previous studies (10a, 27) performed in the same city. A similar relationship was also observed in Pittsburgh (29) and in Vermont (50). The most probable explanation for the difference in the contamination of the two types of water heaters is the design of these devices. In oil or gas heaters, the heat source is usually located below the water heater (that was the case for all fossil fuel heaters in our study) and the sediment that accumulates in the bottom of the heater is frequently at an inhospitable temperature for legionellae. In contrast, heating elements are located on the side of electric water heaters (Fig. 1). Because of this, the sediment in this type of device remains almost constantly at a relatively low temperature (30 to 40°C), which is adequate for the growth of legionellae (25, 51). Our data are compatible with these observations; indeed, whereas the water temperature at the bottom of fossil fuel heaters was quite strongly correlated to the temperature at the faucet ($r = 0.54$, $P = 0.001$), this was not the case for electric heaters ($r = 0.12$, $P = 0.1$); in addition, whereas the temperature at the bottom of the water heater was highly predictive of contamination when all heaters were considered, this association disappeared when the analysis was restricted to electric heaters.

Legionellae have been isolated in samples obtained from municipal water treatment plants that adhered to accepted guidelines for water quality (6, 7, 23, 39). Therefore, seeding of the two type of heaters probably occurs at a similar rate.

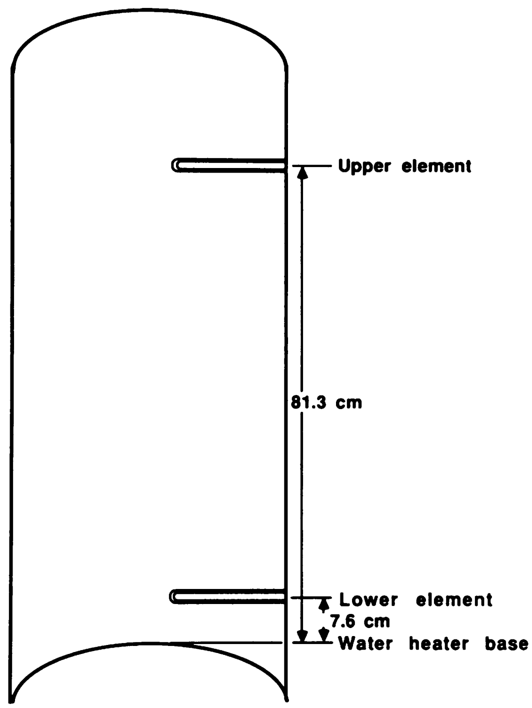

FIG. 1. Drawing of a typical 40-imperial-gallon (182-liter) electric water heater. In 40-gal heaters, the distance between the base and the lower element may vary from 6.4 to 8.3 cm, whereas the distance between the base and the upper element is between 74.9 and 86.4 cm. In 60-gal heaters, these distances are, respectively, 6.4 to 8.9 cm and 90.2 to 105.4 cm. The power of the heating elements was generally 3.0 kW in 40-gallon heaters (in one case, the lower element had a power of 1.5 kW) and 3.8 or 4.5 kW in 60-gal heaters.

However, the adverse conditions in the oil or gas heaters (at least in those heated from below) seem to prevent amplification of the number of legionellae. Not all fossil fuel water heaters are heated from below. Since none of these other types were examined in this study, no inference could be made about their ability to sustain the growth of these bacteria.

The second most important factor in explaining the occurrence of legionellae in the domestic environment is the location of the house. In this study, this variable was transformed into districts. When using this variable, it became obvious that houses located in the older parts of the city were more likely to be contaminated by legionellae. The exact reasons for this association are unknown. One could speculate that the water distribution system in these older areas of the city is more likely to have slime accumulation that might be favorable for local multiplication and dissemination of bacteria. In addition, leaks may be more likely in old water distribution systems, leading to more frequent repairs, which may cause contamination if adequate disinfection procedures are not used. On the other hand, it is also possible that this association reflects the presence of older houses (and thus of older plumbing systems) in old areas of the studied city. Indeed, an association between the age of the house as well as the age of the plumbing and contamination was found in the crude analysis. However, in the logistic regression model, the age of the plumbing was not associated with contamination, whereas the location of the house in an older area of the city was still an important predictor of contamination of the water heater.

The age of the heater was also associated with its contamination. The older the heater, the more likely that legionellae were present. This is of particular interest because although oil or gas heaters were in use in older houses, they were not contaminated by legionellae. In addition, although this difference was not statistically significant, fossil fuel heaters were older than electric water heaters (mean ages of 8.6 and 7.7 years, respectively). Therefore, fossil fuel heaters seemed to have a possible protective effect on the association between the age of the heater and contamination. This observation strengthens the fact that the presence of an electric water heater appears to be the most important factor for the contamination of domestic hot water by legionellae.

It is interesting that the date of urbanization and the age of the heater were both independently associated with contamination by legionellae. Indeed, since no major modification has been made to recently manufactured electric heaters to enhance an even heat distribution, these two factors probably reflect complementary changes in the environment of the underground water pipes and the heaters. These changes may be related to the accumulation of sediments or slime that creates an adequate environment for the growth of other bacterial species that have been shown to grow in symbiosis with legionellae (40). The fact that these associations have never been reported before might be explained by the larger size of this study than previous studies (27, 29).

The water temperature at the hot water faucet after 3 min was also independently associated with contamination. This result was expected. Indeed, numerous outbreaks of Legionnaires disease have been linked to the presence of legionellae in hospital potable water supplies and especially hot water (8, 9, 20, 36, 40, 42, 43). Raising the water temperature has been one of the most efficient methods of controlling these outbreaks. Legionellae do not survive at higher temperatures, and this is no different in the domestic environment. It is interesting that the water temperature was significantly higher in oil and gas heaters than in electric heaters. Indeed, in houses with oil or gas heaters the hot water temperature at the faucet was above 60°C in most cases, whereas in houses with electric heaters the temperature was most frequently between 50 and 59°C. Lee et al. found similar differences between the two types of heater (29). However, when they performed a multivariate analysis, they found that the type of heater was not associated with contamination and that the main factor was water temperature. In our study, when stratifying for water temperature, we found that the association of contamination with the presence of an electric heater remained significant ($P < 0.03$ for all studied devices). In some cases, peripheral devices and electric water heaters were contaminated by legionellae although the water temperature at the faucet was above 60°C. This phenomenon might be explained by differences between the design of fossil fuel heaters and that of electric heaters, as described above, and reflects the lack of correlation between the water temperature at the faucet and that at the bottom of electric water heaters. Finally, it is interesting that the water temperature was not the most important factor in explaining contamination of electric heaters by legionellae.

Contamination of the faucet or shower heads was strongly linked to the presence of legionellae in the water heater. Indeed, the presence of these bacteria in the heater was the most important predictor of peripheral outlet contamination. These results do suggest that bacteria initially seed the water heater, where they multiply and eventually disseminate to the different outlets in the houses. Although obstruction to water flow at the faucet level (presence of an aerator) has previously been linked to contamination of peripheral outlets (5), we were not able to demonstrate such an association in this study. However, only 10 studied faucets were not equipped with an aerator, and, consequently, the power of the specific analysis on this factor was extremely weak. A similar lack of power may also explain the lack of significant association between contamination of a peripheral outlet and any collected variable when the analysis was restricted to houses with a contaminated water heater.

It was impossible to find any association between studied factors or contamination by other bacteria and bacterial counts of legionellae in contaminated devices. However, in previous studies, symbiosis between legionellae and other bacterial species has been reported (40). Since no attempts were made in this study to identify any of the other bacterial species that grew on the culture media, it is impossible to exclude a possible relationship between the presence of legionellae and any other bacterial species.

Rowbotham reported in 1980 that *L. pneumophila* was a pathogen for some amoebae (35). Since then, many reports have confirmed the role of amoebae in supporting the growth of legionellae in man-made water environments (3, 13, 47). In this study, no attempt was made to isolate amoebae. This does not modify the importance of the different associations between the studied variables and contamination of hot water devices by legionellae. Indeed, the very adverse conditions for the growth of these bacteria in fossil fuel heaters could also prevent the growth of amoebae. It would, however, be interesting to further investigate the role of amoebae in the contamination of domestic hot water devices where conditions for their growth is appropriate.

The presence of legionellae in domestic water distribution systems is a well-known fact (10, 27, 29). Its public health importance is, however, totally unknown. Anecdotal cases of legionellosis have been linked to the domestic environment (2, 4, 41). Caution must be used in interpreting data from these few case reports. Indeed, to our knowledge, a well-designed case-control study of sporadic community-acquired Legionnaires disease and contamination of the domestic environment has never been performed. Such a study is essential to establish the exact public health importance of legionellae in this environment. Finally, studying risk factors for the presence of legionellae in domestic hot water systems might serve as a model to understand the contamination of larger public buildings (hospitals, hotels, etc.). This is important because contamination within these settings has been clearly linked to the occurrence of outbreaks of Legionnaires disease.

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