

BACTERIA COLONIZING POINT-OF-ENTRY
GRANULAR ACTIVATED CARBON FILTERS AND
THEIR RELATIONSHIP TO HUMAN HEALTH

BY

Rebecca L. Calderon*
Department of Epidemiology and Public Health
Yale School of Medicine
New Haven, Connecticut 06510

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Project Officer

Alfred P. Dufour
Environmental Monitoring and Surveillance Laboratory
United States Environmental Protection Agency
Cincinnati, Ohio 45268

* Current address: Office of Health Research (RD-683)
USEPA
401 M. St, S.W.
Washington, D.C. 20401

ABSTRACT

An epidemiological study on the health effects associated with high densities of heterotrophic bacteria in drinking water was conducted by the Department of Epidemiology and Public Health, Yale School of Medicine. The study consisted of Yale families that lived in houses that received their water from the South Central Connecticut Regional Water Authority. The households were divided into two groups. One group had granular activated charcoal filters installed where the water line entered the home. Hence, all water in the home was filtered through a GAC filter. The second control group had no filter. Water samples and health diaries were collected during monthly visits to both control and filter homes. All water samples were analyzed for heterotrophic bacteria using R2A media. Within a month the filters had colonized with bacteria from the distribution system and were discharging significantly higher numbers of bacteria (1000 cfu/ml) into the household plumbing than the control houses. Information on upper respiratory infections, lower respiratory infections and gastrointestinal infections were collected. Health data collected showed no difference in acute symptomatology between the filter households and the control households. Bacterial diagnoses made by health care professionals were not related to bacteria in drinking water.

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SECTION 1

INTRODUCTION

The recent environmental awareness of the population has led to an increase in the use of home water treatment devices. These devices can be used to treat a specific problem such as water hardening or removal of iron or be used to improve the aesthetics of the water such as taste, odor and color. The most commonly used devices contain granular activated carbon (GAC). Filters with GAC remove common tastes and odors, some turbidity, chlorine, and many organic contaminants. The large surface area of activated carbon provides an excellent adsorbent for a wide range of chemicals and provides an excellent surface for biomass attachment and growth of bacteria. Of special concern is the health hazard that may be created if pathogenic bacteria adhere to and grow on carbon surfaces. While the microbial quality of the influent water meets current microbial standards, many of water's autochthonous bacteria have been implicated in nosocomial infections. Studies by Geldrich and coworkers (1985) reported densities of heterotrophic bacteria exceeding 1000 per ml in the effluent of GAC water filters. While little may be known about infective doses for the autochthonous bacteria, their diverse nature and ability to colonize GAC still raises health concerns.

A previous study (Calderon and Mood, 1988), examined the possible health effects associated with bacteria colonizing point-of-use (POU) GAC filters. Since these filters are installed in the kitchen and are used to provide water for drinking and cooking, the health outcome examined was acute gastrointestinal symptomatology. In that study, no significant health effect was associated with people drinking water from GAC-POU filters. The filters had average bacterial densities of 1000 per ml in their filter effluent. In this study, conducted by the Department of Epidemiology and Public Health, Yale School of Medicine, whole house GAC filters were studied to look at the effect of total body exposure to higher heterotrophic bacterial densities in filter effluents. This study was funded by a cooperative contract with the Environmental Monitoring, Surveillance Laboratory, U.S. Environmental Protection Agency and by a grant from the Water Research Council.

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SECTION 2
CONCLUSIONS

1. Whole house granular activated carbon filters are colonized by heterotrophic bacteria from the water supply distribution system. Once colonized, the filters discharge significantly higher densities of heterotrophic bacteria than unfiltered tap water.
2. Households with filters discharging higher numbers of bacteria in their effluent did not report a higher incidence of acute symptomatology. Subjects in the study were asked to report on upper respiratory, lower respiratory, gastrointestinal and other (rash, infected wounds, body aches, etc.).
3. Of the bacterial infections reported to health care providers, none were related to the bacteria colonizing the GAC filters.

SECTION 3
MATERIALS AND METHODS

A. SCIENCE ADVISORY COMMITTEE

A science advisory board was formed to cover the various disciplines needed to carry out this project. The SAB contained members from the Yale study team, USEPA's Office of Research and Development as well as the Office of Drinking Water, members from the Water Quality Association, the Water Research Council and the local Regional Water Authority. The members of the SAB are listed in Appendix A.

B. MICROBIOLOGY

Sample Collection

Four water samples were collected monthly from each filter household and two were collected from each control household. A cold water tap sample from the kitchen sink was collected first. The cold water was allowed to run 10 seconds and then a 100 milliliter sample was collected in a sterile bottle containing sodium thiosulfate. The second sample was a hot water sample from the kitchen sink. The hot water tap was turned on and allowed to run until the maximum hot temperature was reached (indicated by steam rising or by touch). A cold and hot water sample were collected from filter and control households.

A filter effluent sample and a filter influent sample were collected from filter households. The filter effluent sample was collected first by allowing 200 mls to discharge and then a 100 ml sample was collected into a sterile bottle containing sodium thiosulfate. Finally the filter influent sample was collected by allowing 200 mls to discharge and then collecting 100 mls into a sterile bottle containing sodium thiosulfate.

Bacterial Analysis

The total heterotrophic plate count was measured in all water samples using R2A (Reasoner, 1985). All samples collected before 7:00 pm were plated within two hours of collection. Occasionally samples collected after 7:00 pm were refrigerated and plated the next morning. Originally 0.1 and 0.5 ml aliquots of samples were plated in triplicate onto predried (35° C for two hours) R2A. As the study progressed 10 fold dilutions in sodium phosphate buffer (pH 7.2) were performed as indicated.

The cold water tap, filter influent and filter effluent

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samples were incubated at room temperature (22°C) for one week. The hot water tap samples were incubated at 35°C for one week. A previous laboratory experiment indicated slightly better recovery of bacteria in hot water samples at the elevated temperature of incubation. The extended time of incubation was used as Reasoner and workers (1985) reported increased recoveries of bacteria with extended incubation.

Chemical Analysis

The primary chemical analysis done by our study group was free and total chlorine determination. A 25 ml sample was collected from the filter effluent and influent taps in glass screw cap tubes with teflon caps. A Hach DPD test kit was used to determine the free and total chlorine concentrations of each test. After three months of testing the total chlorine test was dropped and only the free was done. One sample from each day of collection was repeated to verify that days collections.

Approximately seven households with filters were chosen for extended organic and inorganic chemical testing. The samples were collected by the Yale researchers. The analysis was done by the South Central Connecticut Regional Water Authority (RWA). All sampling and analysis procedures were done according to Standard Methods (1985). The RWA is a state certified laboratory for the analysis of drinking water.

C. FILTERS

Design

The filter was designed by representatives from the Water Research Council and the Water Quality Association. The design was pressure tested by the Water Quality Association. The design chosen was generic in that it was a single tank containing two cubic feet of carbon (Figure 1). The two spigots, before and after, were added for sampling, the pressure gauges (before and after) to monitor reduced flow, and a water meter to measure volume passed through the filter. The carbon specified was 12 by 40 mesh with an iodine number of 1000. The filter specifications are listed in Appendix B.

A competitive bid process was set up using a list of GAC filter manufacturers provided by the Water Quality Association. A brief description of the study and the filter design was sent to the manufacturers requesting a bid for filters, their installation, maintenance and potential removal at the end of the study. A manufacturer was chosen on the quality of the services offered rather than the

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price.

Installation and Maintenance

The GAC filters were installed in homes immediately after the Regional Water Authority's water meter. The installation was carried out by a Connecticut licensed plumber. The initial cold and hot kitchen tap water samples were taken prior to the filter being installed. After the filter had been installed and the filter flushed until the water no longer had carbon fines, a filter influent and effluent sample was taken.

The maintenance of the filters originally consisted of monitoring the before and after water pressures to be sure there would be no loss of flow due to particulate blocking of the filter. Due to problems following installation chlorine removal was tested on a monthly basis. Repairs were performed by a licensed plumber and consisted of repairing parts stressed by high pressure of the Regional Water Authority's distribution system.

D. STUDY SITE AND POPULATION

South Central Connecticut Regional Water Supply

The South Central Connecticut Regional Water Authority (RWA) has both ground and surface water supplies. To have a homogenous supply as possible, the study participants were limited to those that lived only in the areas supplied by three surface supplies that comixed in the distribution system. The study area was also chosen as the RWA has a known coliform problem that occurs occasionally within the distribution system.

Study Population

All families studied were recruited from the members of the Yale Community. This population was chosen because 1) most lived in the specified water supply area; 2) they had access to free medical care and; 3) this is a relatively non-transitory population and therefore would minimize participants dropping out of the study. Approximately 1000 study participants were randomly selected from the Yale Directory. A initial letter of invitation was sent, briefly describing the study, what their participation would entail and a return form that asked them to answer a few questions (Appendix C). From the pool of returned questionnaires, people were selected for the study if 1) they indicated they wished to participate 2) the source of their household water

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supply was the RWA and; 3) there were more than two people in the household. From this group only those that owned their own homes were eligible for the filter group. Those that did not own their own home were immediately assigned status as a control household. The remainder that owned their own home were broken into groups according to how many people were in their household. Each group of households containing two through six people were randomly assigned to either a control or filter study group. The designated filter households were then contacted and asked to participate in the study. This initial conversation described that filter and asked if such could be installed in the house. Some households had no basement or accessible point to install the water filter. These people were then asked if they were willing to be controls and were replaced from the control list.

Questionnaires and Human Investigation Committee

The first questionnaire was a short screening questionnaire on how residents of the house felt about taste, odor and color of their water. Additional questions were asked about number of people in the household, source of the household's water supply, ownership of house and source of medical care. This questionnaire was accompanied by an information letter stating the purpose and requirements of the study (Appendix C).

At the first visit all subjects, filter and control, were asked to read and sign a consent form (Appendix C). They were given a form to fill out on the age, sex and allergy status of all participating members of the household. Each member of the household was assigned a number and a copy of the form given back to the household. In this study health was measured in terms of acute symptomatology. The symptomatology was measured using a revised diary based on those used previously in EPA studies and a standardized questionnaire used to assess pulmonary health. The diary method is a variation on self-administered questionnaires. Self-administered questionnaires were preferred for many reasons. The first is that this avoids the problems of interviewer error. Secondly, since information on all household members was collected, the self-administered questionnaires have been shown to collect information more accurately (Cannel and Fowler, 1963). The diary consisted of a list of respiratory, gastrointestinal and dermatological symptoms. The severity of the symptoms was measured in terms of effect on subjects daily living such as staying home, staying in bed and seeking medical care. If they sought medical care they were asked to call us immediately. The subjects were requested to ask their health care pro-

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viders to take cultures when the cause was thought to be bacterial.

This diary was exchanged for a new one at the monthly visit and the symptoms, if any, were reviewed by the researcher with an adult member of the house. This supplemental interviewing is recommended for self-administered questionnaires (Moser and Kalton, 1972). The researcher confirmed the doctor's visit and diagnoses.

Water Use Survey

To help quantify the various exposure routes to drinking water in the home, a water use questionnaire was given to a subset of filter and control households (Appendix C). The questionnaire was given twice during the course of the study. The first questionnaire was given during January and February to cover winter months. The second questionnaire was given to the same group in June and July. Approximately fifteen households in the control group and fifteen households in the filter group were issued two questionnaires per household. The households were instructed that at least one of the two people filling out the questionnaire had to be an adult. The subjects were asked to fill the questionnaire out for two different days. Subjects were asked about type of activity and the length of time the water ran during that activity.

Human Investigation Committee

All questionnaires, health diaries, consent forms, information letters, went to the Yale University Human Investigation Committee (HIC) for clearance and approval. No questionnaire was used until an HIC approval was received.

E. MEDICAL CLINIC VISITS AND CLINICAL LABORATORY PROCEDURES

The health survey approach to establishing an association between the microbial quality of GAC filtered water and illness in water consumers was supplemented by examining the direct relationship between bacteria isolated from patients visiting a health care provider and bacteria colonizing a GAC filter used by the study participant. The participants were requested at the beginning of the study to notify the study coordinator if they consulted a physician so that a follow-up investigation could be conducted to determine how the illness was diagnosed and if a clinical specimen was obtained. In the event that a bacterial isolate was obtained from a clinical specimen, it was to be characterized biochemically and identified to the species

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level if possible. Subsequent to the identification of the clinical bacterial isolate, the GAC in the filter serving the patient would be removed from the filter housing and examined for the presence of a bacterial isolate identical to the patient isolate. Although the finding of a bacterial strain in a GAC filter that is similar or identical to one isolated from a patient using the filter is not unequivocal evidence of cause and effect it does present a fairly strong indication that the filter may have been the source of the causative organism.

F. PRIMARY DATA HANDLING, PROCESSING AND ANALYSIS

Two classes of data were generated in this study, one from the field (health diaries and water use questionnaires) and laboratory data (bacterial analysis and chlorine determinations). A system of data verification and cross checking was instituted for all forms and data. All data (field and laboratory) were entered into computers for data analysis. A system of data entry error checking was instituted by duplicating entry on a subset of the data and by comparing various computer listings of data with original records.

For each individual water sample raw laboratory bacterial data were converted to cfu per ml. The triplicate values were then entered into a computer and converted to logarithms to generate geometric means. Chlorine data were handled similarly.

Two software programs were used to analyze the data. The majority of data entry and error checking were done with STATPAC GOLD (Walnick, 1985). The remainder of the analysis and data management was done with SYSTAT ().

SECTION 4

RESULTS

A. CHEMICAL ANALYSIS

At the second visit to filter households (visit after installation), a subset of those households began to chronically complain about chlorine taste and odor. After four months of data collection, there was a clear dichotomy in bacterial colonization of filters. Some filters clearly were colonized in that the filter effluent had bacterial densities 100 to 1000 times higher than the filter influent. The other filters showed little difference if any between the filter influent and effluent bacterial densities. Routine testing of water samples from filter influent and effluent for chlorine residual was instituted. At the recommendation of the Science Advisory Committee, a filter was considered to be not working if it did not remove 85% of the chlorine in water. Based on that number it was found that 46 of the filters did not work four months after installation. Bacterial densities in working filters were significantly higher than non-working filters effluents as well as in kitchen hot and cold water tap samples (Table 1). The non-working filters were rebuilt and for the remainder of the study period, filter efficiency was checked by monthly chlorine determinations on filter influent and effluent.

Extended chemical analyses on a subset of filters were performed by the Regional Water Authority. The results of those analyses are in Appendix D. For the majority of inorganic measurements there was very little difference between filter influent and effluent. Except for filters that were not working most filters removed total organic carbon.

B. MICROBIOLOGY

Approximately 6000 water samples were analyzed during the study period. Since 44 filters did not initially work, that data were treated as a separate group. Table 2 shows the distribution of samples collected for the three study groups. An initial set of samples was taken after the filters had been installed. Those installation samples are summarized in Table 3. There were no significant differences between installation samples in the working filter group and the non-working group.

The overall geometric means and standard deviations for the three groups are in Table 4. The geometric means for working filter cold and hot water tap samples are significantly higher than the non-working filters and controls. The filter effluent bacterial densities of the working filters are significantly higher than the non-working filters. The geometric means of bacterial densities for the three groups are plotted by month. The graph of the cold water taps (Figure 2) shows that the

bacterial densities for the controls and the non-working filters are very similar. The filter cold water bacterial densities are approximately one to two logs higher. The hot water tap samples (Figure 3) are similar. Again the control and non-working samples look very similar to each other and the working filter water samples have higher bacterial densities. The effluent samples are compared to the influent samples in Figure 4. The working filter effluents were one to two logs higher than the non-working filter effluents. The working filter effluents were highly correlated with water temperature as measured in the influent (Figure 5).

The influent samples showed a steady increase in bacterial density over the course of the study (Figure 6). This may have been related to water temperature but was probably due to a steady decline of the free chlorine residual in the distribution system over the course of the study. Informal discussion with the RWA indicated that they had slowly been lowering the chlorine residual to approximately 1 ppm at the water treatment plant.

C. DEMOGRAPHICS

The numbers of households, average number of people per household and average length in study are summarized in Table 5. A few extra households were solicited for the control group as it was anticipated that the drop out rate would be slightly higher. An especially important overall characteristic of both groups was their willingness to continue in the study and, hence, their relative stability as a subject population. The vast majority stayed until completion of the agreed upon 12 month period of enrollment. The study had originally planned to go for only 13 months but was extended to 21 months. These extra months were to cover for the months several of the filters were not working. There was no difference between the average number of people per households.

The distributions of all the members of the study by age and gender are in Figure 7. The distribution between males and females was very similar between the filter and control group. The distribution in age was quite different between the two groups. The filter group had more subjects under the age of 10 years. The control group had more subjects over 21, particularly over 55 years of age.

D. Illnesses observed

For simplicity of reporting, the symptoms were divided into four categories. These categories are summarized in Table 6. Upper respiratory was defined as anything from the neck above including respiratory allergies. Lower respiratory was defined as anything below the neck. The gastrointestinal illnesses were obvious and the remainder was categorized as "other". Many of the illness episodes often had symptoms that were in more than

one category of symptoms. These were reported as combinations. The months for the non-working filter groups were deleted from the analysis.

The reported illness experiences of each study group are summarized in Table 7. The overall frequency of reported illnesses as well as the frequencies of the categories was not significantly different for the two groups. The vast majority of illnesses were principally upper respiratory. The most common combination was gastrointestinal with upper respiratory symptoms. Another interesting combination was upper respiratory and other. A viral disease known as "foot and mouth" disease that consisted of flu like illnesses as well as a rash accounted for most of those.

A highly credible illness was defined as an illness that required a change in the individuals normal routine such as stayed at home, stayed in bed or sought medical care. The categories were reanalyzed and the highly credible symptom rate is in Table 8. Approximately 31% of the filter individuals' illness episodes were highly credible as compared to 32% of the controls. There were no significant differences between the two groups for the highly credible illnesses. The bacterial diagnoses reported were strep throat and one case of Campylobacter enteritis that was traced to a foodborne source. The remainder of the diagnoses were attributed to viruses.

Information concerning fever in relation to illness is summarized in Table 9. The filter group reported a fever with 4.6% of their illness episodes compared with 2.6% in the control group. The higher reported fever rate may have been due to the higher percentage rate of children in the filter group. In general fever was reported evenly across all categories of illness. A higher percentage of highly credible illnesses reported a fever in both filters and controls. The filter group reported a slightly higher rate than did the controls particularly for high fevers.

E. Water Use Survey
(to go here)

SECTION 5.

DISCUSSION

The most common short term disease complex affecting the population of the United States is respiratory illness followed by acute enteric illnesses. The role of potable water in the transmission of enteric illness has been reviewed (ref). The role of drinking water in the transmission of respiratory illnesses has been addressed primarily in the hospital setting where it is a concern regarding immunocompromised patients. The emergence of Legionella (ref) and non-avian Mycobacterium (DuMoulin, 1989) as important nosocomial infections transmitted through water has raised health questions regarding other commonly found heterotrophic bacteria in drinking water and their possible relationship to health effects in normal populations. This has become an important issue with the increase in use of home water treatment devices. The GAC devices do colonize and elute elevated numbers of heterotrophic bacteria. This paper is the second of two reports that looked at elevated levels of heterotrophic bacteria (bacteria colonized GAC filters) in drinking water. The first study examined the gastrointestinal exposure route (point-of-use GAC filters). This second paper addresses whole body exposure to elevated levels of heterotrophic bacteria in drinking water with emphasis on respiratory exposure. The elevated levels of heterotrophic bacteria arising from whole-house GAC filters.

Work on bacterial colonization of whole house GAC filters has not been as thorough as the point-of-use GAC filters. Before this study was done, it was not known what levels of colonization could be expected and what variables would affect that colonization. As this study showed, these filters do colonize and elute significantly higher numbers of heterotrophic bacteria than that found in the distribution system. This level of bacteria was shown to fluctuate up and down with water temperature. If there had been more time and resources the following should have been addressed: 1) Ability of these filters to flush to level of influent; 2) Types of bacteria colonizing the filters and relationship to those found in distribution and; 3) The virulence characteristics of the bacteria that colonize the filters.

Table 1. Six month geometric means of heterotorphic bacterial densities per ml.

Type	S T U D Y		G R O U P
	Working Filters	Non-Working Filters	
Kitchen Cold Tap	416	15	9
Kitchen Hot Tap	134	14	17
Filter Influent	7	10	NA
Filter Effluent	399	4	NA
Total	3567	1738	2318

Table 2. Total number of water samples collected for each sample type.

Type	S T U D Y		G R O U P
	Working Filters	Non-Working Filters	
Kitchen Cold Tap	893	434	1167
Kitchen Hot Tap	890	436	1151
Filter Influent	891	435	NA
Filter Effluent	893	433	NA
Total	3567	1738	2318

Table 3. Geometric mean and standard deviations of heterotrophic bacterial densities from installation water samples.

Sample	S T U D Y G R O U P			
	Working Filter Mean	Filter Std	Non-working Filter Mean	Filter Std
Kitchen Cold Tap	9	6	8	7
Kitchen Hot Tap	15	7	7	6
Filter Influent	3	3	6	5
Filter Effluent	3	4	3	3

Table 4. Geometric bacterial means and standard deviations for water samples by group and sample type.

Sample Type	Working Filter		Non-working Filter		Controls	
	Mean	Std	Mean	Std	Mean	Std
Cold Water Tap	2080	8	11	9	7	7
Hot Water Tap	212	9	13	1015	13	
Filter Influent	13	13*	13	13*NA		
Filter Effluent	2917	9	25	13	NA	

* pooled samples

Table 5. Number of households, average number of people per household and average length of time (months) in study.

Variable	Filters	Controls
Households	80	87
Average number of people per household	3	3
Average length in study (months)	17	14

Table 6. Categories of symptoms and reported symptoms for each symptom.

Upper Respiratory	Lower Respiratory	Gastrointestinal	Other
Headache	Cough	Vomiting	Body aches
Runny nose	Phlegm	Nausea	Neckpain
Sore throat	Chesttightness	Diarrhea	Rash
Headcold	Chest cold		Infected Wound
Sneezing	Shortness of breath		
Itchy eyes	Bronchitis		
Respiratory Allergies	Bronchiolitis		
	Pneumonia		
	Asthma		

Table 7. Study group symptom illness rate (per person year) by illness category.

Category	S T U D Y G R O U P	
	Filters	Controls
Total illness episodes	1061	962
Rate per person-year	4.4	3.7
Upper Respiratory	3.7	3.0
Lower Respiratory	1.5	1.7
Gastrointestinal	1.2	1.1
Other	0.8	0.9
Combinations		
2 categories*	1.5	1.1
3 categories*	0.4	0.4
4 categories*	<0.1	0.3

*Any combination of symptoms from upper respiratory, lower respiratory, gastrointestinal or other.

Table 8. Highly credible symptom rate (per person year) for each study group by category of symptoms.

Category	S T U D Y G R O U P	
	Filters	Controls
Total illness episodes	327	305
Rate per person-year	1.3	1.2
Upper Respiratory	1.0	1.0
Lower Respiratory	0.5	0.5
Gastrointestinal	0.4	0.6
Other	0.4	0.4
Combinations		
2 categories*	0.6	0.4
3 categories*	0.2	0.2
4 categories*	<0.1	<0.1

*Any combination of symptoms from upper respiratory, lower respiratory, gastrointestinal or other.

Table 9. Percentage of illness categories with fever by study group.

Category	S T U D Y		G R O U P	
	Filters (N)	%	Controls (N)	%
Upper Respiratory Total	(892)	100	(767)	100
Low Fever (< 38.1 °C)	(31)	3.5	(24)	3.1
High Fever (> 38.0 °C)	(84)	9.3	(40)	5.2
Lower Respiratory Total	(365)	100	(325)	100
Low Fever	(21)	3.0	(11)	3.4
High Fever	(40)	11.0	(29)	8.9
Gastrointestinal Total	(281)	100	(278)	100
Low Fever	(8)	2.8	(9)	3.2
High Fever	(30)	10.7	(20)	8.3
Other Total	(206)	100	(227)	100
Low Fever	(10)	4.8	(9)	4.0
High Fever	(27)	13.1	(20)	8.8

Table 10. Percentage of highly credible illness categories with fever by study group.

Category	S T U D Y		G R O U P	
	Filters (N)	%	Controls (N)	%
Upper Respiratory Total	(255)	100	(246)	100
Low Fever (< 38.1 °C)	(14)	5.5	(17)	6.9
High Fever (> 38.0 °C)	(56)	22.0	(36)	14.6
Lower Respiratory Total	(119)	100	(121)	100
Low Fever	(5)	4.2	(6)	5.0
High Fever	(27)	22.7	(22)	18.2
Gastrointestinal Total	(105)	100	(119)	100
Low Fever	(4)	3.8	(5)	1.7
High Fever	(22)	21.0	(22)	18.5
Other Total	(98)	100	(105)	100
Low Fever	(5)	5.1	(7)	6.7
High Fever	(19)	19.4	(19)	18.1