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June 17, 2008

Ms. Valerie Namba, Senior Environmental Planner
California Department of General Services, Real Estate Services Division
Professional Services Branch, Environmental Services Section
707 Third Street, Third Floor, MS 509
West Sacramento, CA 95605-9052

Dear Ms. Namba:

Re: Health Risk from Pathogenic Organisms in Copper and PEX Water Pipe

On behalf of the California Pipe Trades Council, Veritox has been requested to respond to your Draft Environmental Impact Report ("DEIR") entitled "Adoption of Statewide Regulations Allowing the Use of PEX Tubing." This response addresses specifically the biofilm discussion found on pages 4.2-6 and 4.2-7 of the DEIR.

In preparing this response, Michael Krause, MSPH, CIH, of Veritox, has researched the literature addressing the potentially pathogenic bacterial growth in biofilms in various pipe materials, focusing on copper and cross-linked polyethylene (PEX). The materials in the attached bibliography were reviewed, along with related materials. The articles and publications were found through a Medline library search and Internet searches using appropriate key words.

Bacterial Growth in Piping Systems

The booklet entitled "Biofilm" provides a good overview of biofilms in piping systems (Dreeszen 2003). It states that 99 percent of the bacteria in water systems are likely found in biofilms attached to internal surfaces. Biofilms can be a source of bacteria which can cause infection and disease (pathogens). Biofilms form as follows: A clean pipe surface comes into contact with water and an organic layer deposits quickly, forming a "conditioning layer." Free floating or "planktonic" bacteria are carried in the flowing water. "Pioneer" bacteria attach, adhere to the surface, and become "sessile." Biofilm bacteria divide and excrete "sticky polymers, which hold the biofilm together and cement it to the pipe wall. In addition, these polymer strands trap scarce nutrients and protect bacteria from biocides." Secondary colonizers may join the biofilm, forming a "complex, metabolically cooperative community." The biofilm may grow and spread downstream.



Most of the volume of a mature biofilm is the loose polymer matrix (75-95%) and the rest is bacterial cells (5-25%). Anaerobic colonies may prosper next to the pipe surface and aerobic bacteria thrive above them in the water stream.

Health Risks from Bacteria in Water Pipes

Biofilms can harbor a variety of pathogenic bacteria, including *E. coli*, *Pseudomonas*, *Mycobacter*, *Campylobacter*, *Klebsiella*, *Aeromonas*, *Legionella*, *Heliobacter pylori*, and *Salmonella typhimurium* (Wingender 2004). Exposure can occur by drinking water, getting organisms into wounds, or breathing aerosolized water. *Pseudomonas* is a principal pioneering organism and is common in biofilms (Rogers 1994). It is an opportunistic pathogen, attacking immune compromised humans and experimental animals (Dreeszen 2003).

Infection with *Legionella* bacteria can cause Legionnaires' disease (Legionellosis). About 90% of disease cases have been caused by the species *Legionella pneumophila* (Flannigan 2001). General control strategies for domestic water systems should prevent stagnation of water; promote use of materials that do not provide nutrients for growth of legionellae, such as neoprene or other synthetic gaskets rather than natural rubber; and promote copper rather than plastic piping, which promotes *Legionella* colonization.

Guideline 12-2000 from the American Society of Heating Refrigerating and Air Conditioning Engineers states that natural rubbers, wood, and some plastics have been shown to support the amplification of *Legionella*, while other materials such as copper inhibit their growth (ASHRAE 2000).

Measurement of Bacteria

Bacterial levels can be measured or estimated in water and in biofilms. The heterotrophic plate count (HPC) is common in the literature. The bacteria in water samples collected from pipes are plated on agar media and cultured. Colonies grow and they are counted and speciated. The results are reported as CFU/ml (colony forming units per milliliter of water) and can be converted to CFU/cm² (CFU per square centimeter) if the pipe surface area involved is known. Special plating techniques are used to culture and count *Legionella*. The shortfall of plating methods is that the sessile bacteria in biofilms may not be accurately measured (van der Kooij 2005).

Another technique utilizes measurement of ATP (adenosine triphosphate), a chemical compound found in all living cells. Measurement of picograms of ATP per liter of water have been found to correlate with a given amount of biomass in pipes (van der Kooij 2005). These measurements can be converted to ATP/cm² of pipe surface. Other techniques are used as surrogates of the levels of total bacteria, biomass, and viruses (Lehtola 2004).



Biofilm Growth in Copper, PEX, and Non-PEX Pipe

Using plate count techniques, researchers found that copper pipe inhibited both biofilm growth and *L. pneumophila* at various temperatures in a laboratory experiment over three weeks (Rogers 1994). PEX pipe was not studied. Other types of plastic pipe supported growth. The study concluded that “The use of copper as a plumbing material may help to minimize the risk of Legionnaires’ disease.”

Others have tested cold and warm water supply systems and found water supplied by copper pipes to be nearly free of *Legionella* (only 2% of probes were positive), while polyethylene pipe was heavily contaminated (65% positive) (Pongratz 1994). Another study showed that a combination of copper and silver ions were effective in controlling *Legionella* in warm water systems of hospitals (Lin 2002). Copper and silver in combination with low levels of chlorine were found to be especially effective (Landeem 1989).

Some of the most often cited, long-term simulation studies have been done in northern Europe. The DEIR did not address the comparability of copper and PEX pipe studied and used in Europe versus that used in California.

The DEIR relies on basically one source from the Netherlands to support the conclusion that PEX pipe “would not lead to increased risk” (van der Kooij and colleagues). The 1999 report cited was not peer reviewed and published. One statement was from a 2006 symposium. Only the 2005 van der Kooij journal article was peer reviewed. It was obtained and is addressed below.

Using both plate count and ATP methods, researchers in the Netherlands tested copper and PEX pipe in mock-up circulating water systems for over two years (829 days) (van der Kooij et al 2005). The authors reported that PEX “enhanced” biomass production, possibly by the release of organic compounds from the plastic itself (citing Skjevrak). They reported biofilm content for copper pipe was constant over the two years (about 750 pg ATP/cm²) (Fig. 4). Biofilm accumulation increased steadily in PEX pipe over time and appeared to end up about five times higher (about 3,700 pg ATP/cm²). A “Biomass Release Rate” (BRR) was calculated and was estimated to be over twice as high for PEX as for copper pipe. Although the DEIR states that “higher amounts of biofilm could lead to increased risk of human contact with pathogenic bacteria,” the van der Kooij biofilm growth results were not specifically discussed.

Instead, the DEIR reported on the *Legionella* growth results from the van der Kooij study and implied misleadingly that those results were indicative of total biofilm growth over time. The van der Kooij group grew *Legionella* in water in the presence of PEX pieces. The reason for using PEX was not stated. Some of this *Legionella* solution was used to inoculate the copper and PEX water pipe systems under study. *Legionella* concentrations were then measured at various times over two years in both water and biofilm.



The results showed that *Legionella* growth occurred much more slowly in copper pipes than PEX. The median *Legionella* concentrations from water in PEX pipes were apparently over twice as high as in the copper pipes (Table 2). The median *Legionella* level in the biofilm was almost 100 times higher for PEX pipe versus copper pipe (Table 3). However, at the end of the two year period, similar *Legionella* levels were reported in both water and biofilm for PEX and copper pipes.

The DEIR seized on the final result and reported that the *Legionella* levels in PEX and copper pipes were the same under the “more relevant” long term conditions at the end of the experiments. The DEIR implied misleadingly that biofilm formation was also similar at the end of the study. Further, the DEIR inferred that *Legionella* was the only health issue and did not consider other pathogenic bacteria or mycobacteria.

Similar studies from Finland evaluated copper and polyethylene (PE) pipe in a pilot distribution system mimicking that of a home (Lehtola 2004, Lehtola 2005). These researchers found that the formation of biofilm was slower in copper pipes (inhibited by copper ions) than polyethylene pipes, reaching a “steady state” in 37 days and 200 days, respectively. At 200 days, there was no difference in microbial numbers when measured by plating samples from water in the pipes (HPC). However, the levels of ATP in the PE samples were about twice as high as those in copper pipe samples, indicating higher biomass. One postulated cause was leaching of phosphorous compounds from PE pipe, supplying bacteria with a needed nutrient.

Lehtola 2004 reported that there were different microbial communities in the biofilms from PE versus copper pipes. The “biomarker profile” for gram negative bacteria (many of which are reportedly pathogenic) was found to be different within the piping systems. The DEIR did not consider pathogenic bacteria other than *Legionella*.

Lehtola 2004 also measured indicators of viruses. At the end of the experiment (308 days), the biofilms were analyzed for virus-like particles, which were found to be five to ten times more abundant in PE pipe versus copper (Fig. 5). The DEIR did not consider pathogenic viruses.

Conclusions

The Draft Environmental Impact Report does not make a compelling argument for a “less than significant” public health impact from widespread use of PEX water pipe (Impact 4.2-1). Basically, only one research group was relied upon (van der Kooij and colleagues). Their data that indicated increased biomass growth in PEX pipe versus copper pipe was not discussed. Instead, the DEIR misleadingly implied that their finding of similar *Legionella* growth on PEX and copper after two years was fully representative of the biofilm growth over the same time. Further, the DEIR only considered *Legionella* in the discussion of pathogens, disregarding data on other pathogenic bacteria, mycobacteria, viruses, and other organisms. Much stronger support is needed for the DEIR to declare insignificant human exposure and health risks associated with biofilm



growth, pathogens, and plastics leachate as a consequence of use of PEX piping in water supply systems.

Veritox[®], Inc.

Michael Krause, MSPH, CIH
Senior Industrial Hygienist



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