A CASE HISTORY OF DISINFECTION OF WATER IN RURAL AREAS OF MEXICO

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ABSTRACT

Effective use rainwater as a source of water supply in rural and urban areas of the world, requires storage and containment in surface reservoirs, cisterns or tanks. While the purity of rainwater can be high when initially collected, especially in rural areas away from industrial air pollution, storage of water for long periods of time can subject this valuable resource to contamination from disease vectors, blowing dust and dirt, bacterial contamination from collection surfaces and direct contamination.

The purpose of this paper is to provide a case history of an alternative disinfection method whereby water storage vessels may be rendered virtually free of pathogenic and nuisance miroorganisms for long periods of time, thus providing a reliable disinfected water

supply.

A silver based colloidal suspension of silver in a protein carrier has been used successfully throughout Mexico to disinfect rural, urban and institutional water tanks for up to one year by coating the inside surfaces. When dry the coating becomes hard, but slowly redissolves and disinfects the water over a long period of time. Recent laboratory research confirms the efficacy of this disinfection method.

INTRODUCTION

This documents a case history of a protein based colloidal silver compound used as a disinfectant for municipal water supplies over a thirty year period, and recent laboratory efficacy tests demonstrate the principal advantages and limitations of this disinfection method.

The silver disinfectant, sold under the trade name of Microdyn, is a colloidal suspension of ionic and molecular silver in soluble and insoluble fractions, finely dispersed in a protein carrier. This material has unique properties that make it ideally suited for disinfecting water for domestic, municipal, institutional and industrial use where more conventional disinfection techniques are difficult, impractical or overly expensive.

Historical Background

Microdyn was developed in 1957 in Mexico City based upon work carried out in Czechoslovakia, Rensselear Polytechnic Institute, New York and Princeton University in the late 1930's. Since introduction as a commercial water disinfectant in 1957, the product has been used extensively throughout Mexico and is presently sold and distributed throughout thirty Mexican States.

For industrial use a concentrated form of 3.2 percent by weight of silver is available, while 0.32 percent solution is utilized for personal or domestic use. The commercial grade is utilized for disinfecting reservoirs, cisterns and water storage tanks for municipalities, hospitals, clinics, stores, cities and industrial

concerns.

Method Of Application

Apart from the fact that it is tasteless and odorless, one of the principal reasons for the appeal and wide use of Microdyn in Mexico is in its method of application. For disinfecting large reservoirs Microdyn is applied to the walls of the tanks, a process known as "activation". The 3.2 percent Microdyn is applied at rate of about 50 to 60 cc per square meter or alternatively 1 liter per 20 square meters of surface. The applied material is allowed to dry into a hard shellac like appearance. Over a period of one year or more the Microdyn slowly dissolves into the water, thus not only is the water disinfected, but microbiological growth on the walls of the container is prevented for an entire year. This precludes the growth of algae, slimes, molds and other material that may be detrimental to the purity of the drinking water. Because of the constituents in Microdyn, some of the material dissolves into solution, while the remaining part forms an insoluble suspension. Because of the finely divided nature of the ionic and molecular silver atom, and further, because of the strong affinity of silver atoms for sulphur groups, Microdyn is effective in interrupting enzymatic functions and in destroying protein materials in microorganisms.

In rural areas, more particularly in undeveloped regions, storage of rainwater is essential, since treatment and distribution of potable water is rarely available. Moreover, continuous disinfection with alternative disinfectants is not always suitable

because of the need for constant attention and the acquisition, use, and maintenance of metering equipment.

Contamination of stored rainwater supplies originates from rainwater collection surfaces, atmospheric pollution, dirt, dust and sediment and other disease vectors. The overall simplicity of applying Microdyn once annually to the walls, floor and ceiling of a water cistern or pot is simple, direct and finds favor with regulatory agencies since the cistern must be cleaned of dirt and debris annually before activation.

Laboratory Efficacy Tests

Laboratory tests have been conducted by the University of Arizona Microbiology Department to demonstrate the bactericidal efficacy of Microdyn. Bacteria selected for study included Salmonella, Shigella, Klebsiella, Legionella and Pseudomonas; viruses studied included Poliovirus, Rotavirus, and Herpes viruses - IBR and PRV. Other tests, carried out by Pemex in Mexico on cooling towers also indicate that algae, Tribonema and Spirogyra are inhibited by Microdyn.

Overnight cultures of Salmonella typhosa, Klebsiella pneumoniae, and Pseudomonas aeruginosa were prepared on tryptic soy agar slants (TSA) (Gibco Laboratories, Madison, WI) and rinsed with sterile distilled water and filtered through #2 Whatman filter paper (Whatman Limited, England). Bacterial cultures were obtained from the American Type Culture Collection (ATCC) (Rockville, MD).

Suspensions of bacteria or poliovirus (type 1, LSC, also obtained from the ATCC) were added to sterilized (autoclaved) Tucson

tapwater (Table 1 shows water composition) and incubated at room temperature on stir plates (Bell-Stir, Bellco Glass, Inc., Vineland, NJ).

TABLE 1

Composition Of Tucson Tapwater

Chemical Parameter	mg/l
Calcium	48.0
Magnesium	5.2
Sodium	42.0
Bicarbonate	150.0
Chloride	30.0
Nitrate	8.0
Sulfate	48.0
Total Dissolved Solids	298.0
Total Hardness	141.0
Organic Carbon	< 1.0
pH (after autoclaving)	7.2

A working solution of Microdyn, a colloidal silver solution, 3200 µg/l was prepared daily by diluting in sterile distilled water to yield a final concentration of 32 µg/l.

A neutralizer solution (100x) which neutralizes the effect of the Microdyn solution was prepared daily by dissolving ten grams thioglycolic acid (Sigma Chemical Company, St. Louis, MO) with 14.6 gm sodium thiosulfate (Fisher Scientific, Fair Lawn, NJ) in 100 ml distilled water and filtering through a 0.2 micron filter (Nalgene, sterilization filter unit, Type S, Nalge Corp., Rochester, NY).

Studies using different concentrations of Microdyn were conducted by adding different quantities of the Microdyn working

solution to sterile Tucson tapwater and sampling at predetermined time points. Neutralizer solution (100x) was added to each sample (at one-hundredth the sample volume) at time of collection.

Bacterial assays were performed by membrane filtration as described in the Compendium of Methods for the Microbiological Examination of foods (1984) (Gelman 3-47 mm filter holder, using GN-6 membrane filters, Gelman Instrument Comp., Ann Arbor, Mich.). Filter housings were disinfected between samples by rinsing with 0.5% bleach (Georgia-Pacific, Los Angeles, CA) followed by rinsing with 5% sodium thiosulfate and exposed to ultraviolet light (BioGARD Hood, The Baker Comp., Inc., Sanford, MA) until use. Bacterial samples were plated on TSA plates and incubated at 37 degrees centigrade overnight. Poliovirus samples were assayed on Buffalo Green Monkey cells as described by Melnick (1979).

Survival Curves:

Table 2 shows test parameters for average and worst case water quality conditions as a measure of disinfectant efficacy. Worst case water quality conditions are designed to stimulate situations which would adversly effect the performance of most commonly used disinfections such as halogens, and the worst likely water quality to be used for drinking water.

TABLE 2

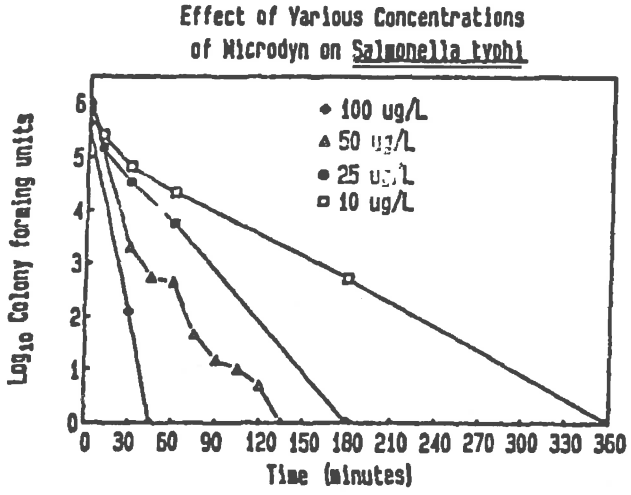
Water Quality And Test Conditions

Parameter	Average Water Quality Conditions	Worst Case Water Quality Conditions
pH	7.2	9.0
Organic Carbon (mg/l)	< 1.0	10.0
Temperature (degrees C.)	25	4
Turbidity (NTU)	< 0.1	32
TDS	298	1300

Average water quality conditions Microdyn and neutralizer solutions were prepared and used as described above. Assays were performed by spread plate technique as described in the Compendium of Methods for the Microbiological Examination of Foods (1984) (Fig. 1).

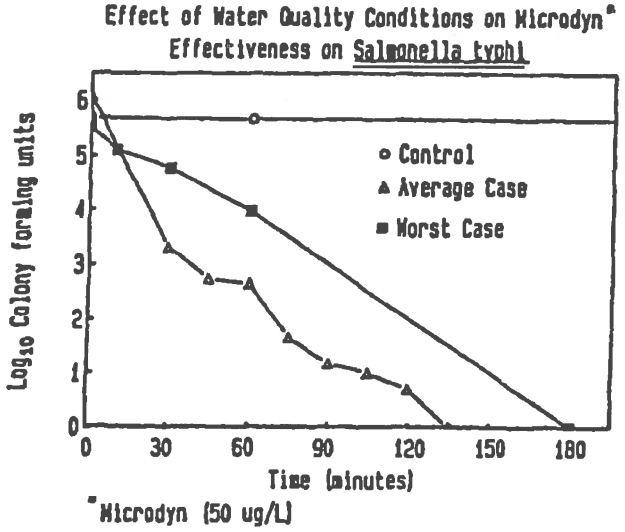
Worst case water quality conditions In order to test Microdyn under adverse conditions, sterilized Tucson tapwater was treated with 10 mg/l humic acid (Aldrich Chem. Comp., Inc., Milwaukee, WI), 150 mg/l AC Spark plug, air cleaner test dust (General Motors, Flint, MI) to yield a turbidity of 32 nephelometric turbidity units (model 2100A turbidimeter, Hach, Ames, Iowa), adjusted to pH 9 using 5N NaOH (MCB Manuf. Chem., Inc., Cincinnati, Ohio, using Beckman model 70 pH meter, Beckman, Irvine, CA) and incubated at four degrees Centigrade. Microdyn and neutralizer solutions were prepared and used as described above. Spread plate assays were conducted as described above.

Figure 1 presents die-off curves of Salmonella under average



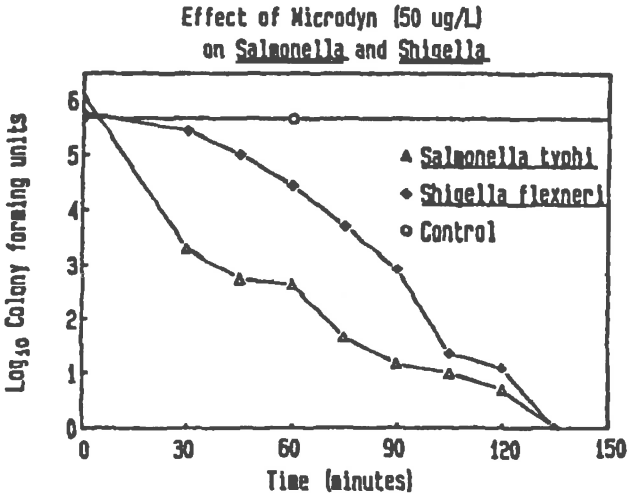
(A)

SALMONELLA, AVERAGE CONDITIONS



(B)

SALMONELLA, WORST CASE COMPARISON



(C)

AVERAGE CONDITIONS, SALMONELLA AND SHIGELLA

FIGURE 1
DIE OFF CURVES

and worst case conditions with concentrations of silver from 10 to 100 micrograms per liter. With concentrations of 50 micrograms per liter to 200 micrograms per liter the disinfection capability increases dramatically. Tests with concentrations of 200 micrograms per liter provided a six log kill within 20 minutes. Assuming that a detention of more than 6 hours is maintained in a cistern, six log kill may be obtained with Microdyn silver concentrations below 10 micrograms per liter. The results in Figure 1(b) demonstrates that adverse water quality conditions (i.e. high turbidity, organic matter, and dissolved solids) do not have any significant effect on the performance of Microdyn against Salmonella typhi. Figure 1(c) demonstrates that Microdyn is effective against both Salmonella and Shigella at concentrations of 50 micrograms per liter. Other efficacy tests, not yet complete, indicate that Microdyn is effective against 1) Pseudomonas and Legionella at 200 micrograms per liter, 2) algae and fungi, 3) Trophozoites parasitic forms, 4) lipid viruses such as the Herpes viruses IBR and PRV. Microdyn is not effective against non-lipid viruses at concentrations less than 200 micrograms per liter. However, these are not expected to be present in stored rainwater cisterns. Because of the low silver concentrations required to disinfect water supplies the slow dissolution of Microdyn from the walls of the cistern is adequate to provide sufficient disinfection for up to 12 months.

Cistern Water Quality

Most rainwater from roofs and cisterns that we have examined contains coliform and fecal coliform bacterial numbers above that

recommended for drinking water (Table 3). The origin of these organisims may result from animal droppings in the collection areas (e.e. rooftops, runoff from the land surface, dust, soil and seepage).

TABLE 3

Microbial Water Quality Of Cistern Water In Tucson, Arizona*

Standard Plate Count(/ml)	$10^{6.4}$
Coliforms(/100 ml)	$10^{4.4}$
Fecal Colifroms(/100 ml)	$10^{3.9}$
Turbidity (NTU)	8.1

* Average of eight samples from cistern water in Tucson, Arizona.

Another application of Microdyn is for domestic use in which Microdyn is applied directly to the water by droplet form from small 30 ml bottles of 0.32 percent solution. In this application one drop per liter is sufficient to disinfect water in 20 to 30 minutes by providing a concentration of approximately 100 migrograms per liter of silver.

Toxicology

Under Environmental Protection Agency guidelines for hazard evaluation for humans and domestic animals, the following toxicology was carried out for Microdyn: acute oral LD50, acute dermal LD50, primary eye irritation and primary skin irritation studies. These tests were carried out by an independent testing laboratory in New Jersey.

In the test for LD50, adult albino rats were dosed with full strength 3.2 percent Microdyn solution by the oral route. None of

the ten animals died over the 14 day test period and all gained an average of over 50 percent in weight. Autopsies performed after the test showed that liver, spleen, kidney, lung, stomach and small intestine all appeared normal. The LD50 was calculated at greater than 5 grams per kilogram of body weight.

For allergic contact dermatitis (McGuire Test) guinea pigs were utilized in which the full strength 3.2 percent Microdyn solution was applied to a previously sensitized area of skin at time period 0, 2, 4 and 7 days. No allergic contact dermatitis was observed.

During acute dermal LD50 in rabbits, Microdyn 3.2 percent formulation was applied topically at a rate of 1.67 mils per kilogram of body weight. All animals survived the 14 day test period gaining in weight; the LD50 dose was computed at greater than 2 grams per kilogram of body weight, and therefore not toxic to adult rabbits when applied topically.

The acute toxicity by eye irritation included injecting 0.1 mils of 3.2 percent into the eye of an albino adult rabbit with and without rinsing; observations extending over a period of 21 days. All irritation in both groups disappeared by day 4 with the conclusion that Microdyn had no affect on the cornea of animals.

In the 1962 U.S. Public Health Service Primary Drinking Water Standards, silver concentration was limited to 50 micrograms per liter to minimize any adverse cosmetic effects. In recent years studies have shown that this in fact cannot be substantiated. Further, the case history of the use of Microdyn and that shown by the laboratory experiments for toxicology indicate that no cosmetic or ill health effects have been observed. In acknowledgment of the

absence of adverse health effects and recognition of the beneficial uses of silver, the United States Environmental Protection Agency early in 1986 proposed removal of silver from the primary drinking water standards.

Case Histories

The follow case histories illustrate specific applications within Mexico where entire communities or institutions receive water disinfected entirely by Microdyn.

The City of Cruz Azul, Lagunas, Oaxaca

This City approximately 800 kilometers southeast from Mexico City centers around a cement works owned and operated by the Cruz Azul Cooperative. This cooperative also operates a second major cement plant in the state of Hildalgo. These cooperative towns have a total population of approximately 10,000 people. Prior to 1977 the water supply was in such poor condition that approximately 20 percent of the entire population died. The water is pumped from an adjacent river to a filtration plant and distributed to reservoirs throughout the city. Contamination from dead animals in the river contributed to the high pollution and presence of pathogenic bacteria.

In 1977 Microdyn treatment was implemented by activation of the open raw water reservoir (upstream from the filters); distribution reservoirs on hills throughout the city; and, private water cisterns in domestic residences. Since initial activation in 1977 the number of cases of gastroenteritis have been substantially reduced.

Ministry of Health applications

Microdyn is widely used by the Mexican government and

administered by the Secretary of Health through the Ministry of Health for Federal Republic. Recently the Ministry of Health purchased large quantities of Microdyn for disinfection of water following the disatrous September 1985 earthquake in Mexico City. The government utilized Microdyn for disinfecting water in schools and hospitals primarily to prevent the outbreak of disease in areas where water supply mains had been broken.

Mexican oil company, Pemex

Pemex, operates oil wells, refineries, distribution and marketing outlets of petroleum products. Microdyn is used at the Tampico refinery near Vera Cruz for disinfection of cooling towers; on drilling rig platforms for disinfection of potable water and on ships distributing fresh water to the platforms.

In the cooling towers, Microdyn is painted directly onto timber splashboards at a rate of 50 cc per square meter. The cooling towers are approximately 8.5 meters high and carry a circulating water flow rate of 3,500 liters per minute. The water temperature entering is approximately 50 degrees Centigrade and 34 degrees Centigrade leaving. Prior to application with Microdyn the cooling towers were heavily fouled with bacterial, algal and fungal growth. Disinfection with other disinfecting agents had proven unsatisfactory and uneconomic. In August of 1985 the towers were cleaned and the timber baffles were painted with Microdyn. Observations were made daily and tests carried out every 45 days to ascertain water quality characteristics and the extent of microorganism fouling on the cooling tower itself. Although this series of tests is presently ongoing, results indicate that none of

the surfaces coated with Microdyn have any microbiological growth attached to them including the more prolific and tenacious algal forms of Tribonema and Spirogyra. The conclusion of this test is anticipated in July of 1986.

Pemex also utilizes Microdyn for disinfection of potable water tanks in ships distributing water to oil platforms throughout the Gulf of Mexico. In this application Microdyn is coated on the insides of the epoxy lined steel tanks.

Summary

Throughout the 30 years of use in Mexico Microdyn has been employed by the armed forces, large industrial concerns, hotels, hospitals and institutions and it is estimated that approximately 1 million people are continuously served with water disinfected with Microdyn. Cistern water usually contains enteric bacteria which should be controlled by proper disinfection. Based on laboratory and field studies Microdyn has been shown effective in control of these organisms and enteric pathogens. Because of the very small amounts required for disinfection of water supplies this material has an important place in improving the sanitation and in protecting water supplies in rural areas or other centers of habitation where water distribution is not available. Microdyn effectively disinfects against pathogenic bacteria, lipid containing viruses and prevents the growth of fungus, algae and virtually sterilizes contact surfaces in which rainwater can be stored. This simple method of disinfection will help to raise public health standards in developing areas.

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