MIGRATION OF BACTERIOPHAGE T_4 IN PERCOLATING WATER THROUGH SELECTED OAHU SOILS

by

Richard M. Tanimoto
Nathan C. Burbank, Jr.
Reginald H. F. Young
L. Stephen Lau

Technical Report No. 20

June 1968

Partial Technical Completion Report

for

POLLUTION EFFECTS ON GROUND WATER RECHARGE IN HAWAII

OWRR Project No. A-001-HI, Grant Agreement No. 14-01-0001-1079

Principal Investigators: L. Stephen Lau & Nathan C. Burbank, Jr.

Project Period: July 1, 1967 to June 30, 1968

The programs and activities described herein were supported in part by funds provided by the United States Department of the Interior as authorized under the Water Resources Act of 1964, Public Law 88-379.

ABSTRACT

A laboratory study was made of the ability of three Oahu soils, Wahiawa, Lahaina, (both Low Humic Latosols), and Tantalus cinder (of the ashy cindery isothermic family of Typic Dystrandepts) to remove the coliphage T_4Br^{II} mutant (a vegetative virus) from percolating waters.

The soils were selected on the basis of their occurrence on the island of Oahu in areas where percolating water may enter directly and in quantity into the ground-water body that provides the principal portion of the island's domestic water supply. On Oahu, 95 percent of the total agricultural product is cultivated in Low Humic Latosols.

Two of the soils proved to be effective in retaining the T_4Br^{II} mutant virus but only at thicknesses greater than 2½ inches.

The Wahiawa and Lahaina soils were effective in the removal or adsorption of the coliphage T_4 from percolating waters at the applied concentration of 2.5 x 10^6 per ml of feed solution through soil thicknesses of 6 inches and 2½ inches. Adsorption at these thicknesses was 100 percent.

Breakthrough of the viruses occurred in both Wahiawa and Lahaina soils at soil thickness of 1½ inches at applied concentration of 1.5 x 10^6 per ml of feed solution. The breakthrough began slowly, but increased rapidly with time and the rate of breakthrough varied with the soil. A higher rate was observed for Lahaina soil during the first two days of sampling.

The Tantalus cinder proved ineffective in withholding the viruses at the applied concentration of 1.5 x 10^6 per ml of feed solution. Breakthrough of concentrations of 10^5 of the phage (plaques) were recorded through soil thicknesses of 15, 12, and 6 inches.

CONTENTS

LIST OF FIGURES

LIST OF TABLES

INTRODUCTION

Within the past two decades, the study of the underground movement of pollutants has broadened in scope to include viruses. This was brought about by the problems arising from increasing water consumption and waste water disposal for a burgeoning population in the United States and by several epidemics and numerous lesser outbreaks of viral diseases that have been attributed to water-borne viruses (1, 2, 3). Most of these epidemics and outbreaks have involved small well supplies contaminated by subsurface sewage disposal systems. Thus, in order that measures be established to prevent and guard against such outbreaks, there is a great need to know and understand the factors which govern the movement of viruses through subsurface aquifers.

Very limited recent research has been published in the literature regarding viral contaminants and there is no record of such work on Oahu. However, published laboratory analyses of the Board of Water Supply of the City and County of Honolulu show that Oahu's ground-water supply maintains its excellent quality according to the United States Public Health Service Drinking Water Standards (4).

The abundance of fresh water in the subsurface and deep soils of weathered basaltic lava are two of the major natural resources which have led to the economic growth of Oahu.

The island of Oahu is composed of permeable volcanic piles of fractured and solidified lava flows and is transected by impermeable intrusive dike barriers which impeded water movement through the island mass. These volcanic piles constitute the ground-water aquifers.

Precipitation in the Hawaiian Islands is high and highly variable both with time and place. The island-transecting Koolau range on Oahu coincides with the highest rainfall belt. It receives orographic rainfall as the moisture-laden tradewinds sweep in from the sea and up the mountain range, and as the rising clouds are cooled, condensation occurs, and rain drops on the uplands. Additionally, cold-front storms and local convection contribute intense rainfall to the entire island mass during the six months from October to April. Rainfall in the wettest watersheds averages almost an inch a day well spread throughout the year.

The portion of rain which percolates eventually enters the basal

ground-water body either directly or by way of the compartments formed by the dikes. Below the basal water table in the crevices and voids of the rock formation is accumulated a vast body of fresh water (Ghyben-Herzberg lens) that floats on the denser sea water.

This huge basal-water lens has been the primary source of water supply for Oahu for over half a century, supplying 90 percent of the total water consumed by a population of almost half a million. It also serves the water requirements of intensive armed forces activity and large agricultural and industrial enterprises.

This unique underground basal water is of such purity that over 90 percent of the domestic water supply receives no chlorination, and the remaining supply receives a nominal chlorination dosage of 0.3 parts per million (ppm) before it is piped into the distribution system (7). Few places in the world have water comparable in quality to Oahu's basal water.

In order to provide adequate recharge to the basal ground water, some 123,000 acres of land on Oahu, primarily in mountainous areas where heavy rainfall occurs, have been set aside as Forest Reserve Areas and entry to and use of the land are governed by stringent regulations (8). Sugar cane and pineapple cultivation account for 34,900 and 22,050 acres of land respectively, while pasture land takes up another 58,300 acres. These four land uses comprise about 61.5 percent of the total land area of Oahu. Most of the land in these classifications are located in areas where there are no impervious strata and percolating waters may pass through the surface soil and enter directly into the underground basal-water body (9). The land use areas of Oahu are shown in Figure 1.

The soils and subsoils of Oahu are derived from volcanic lavas, cinders, and ash, and to a limited extent from coral reefs. The parent materials of the soils are dominantly basic igneous rocks--basalts or andesites, or their pyroclastic equivalents (10). Climate and vegetation are the active factors in soil formation, accounting for the major differences among broad groups of soil on Oahu. The humid, subtropical climate causes rapid chemical weathering of the parent materials.

The amount of silica in Hawaiian lavas in considered to be relatively low, and with laterization dominating the different soil-forming processes, silica is further leached from the soil. On the other hand, the amount of

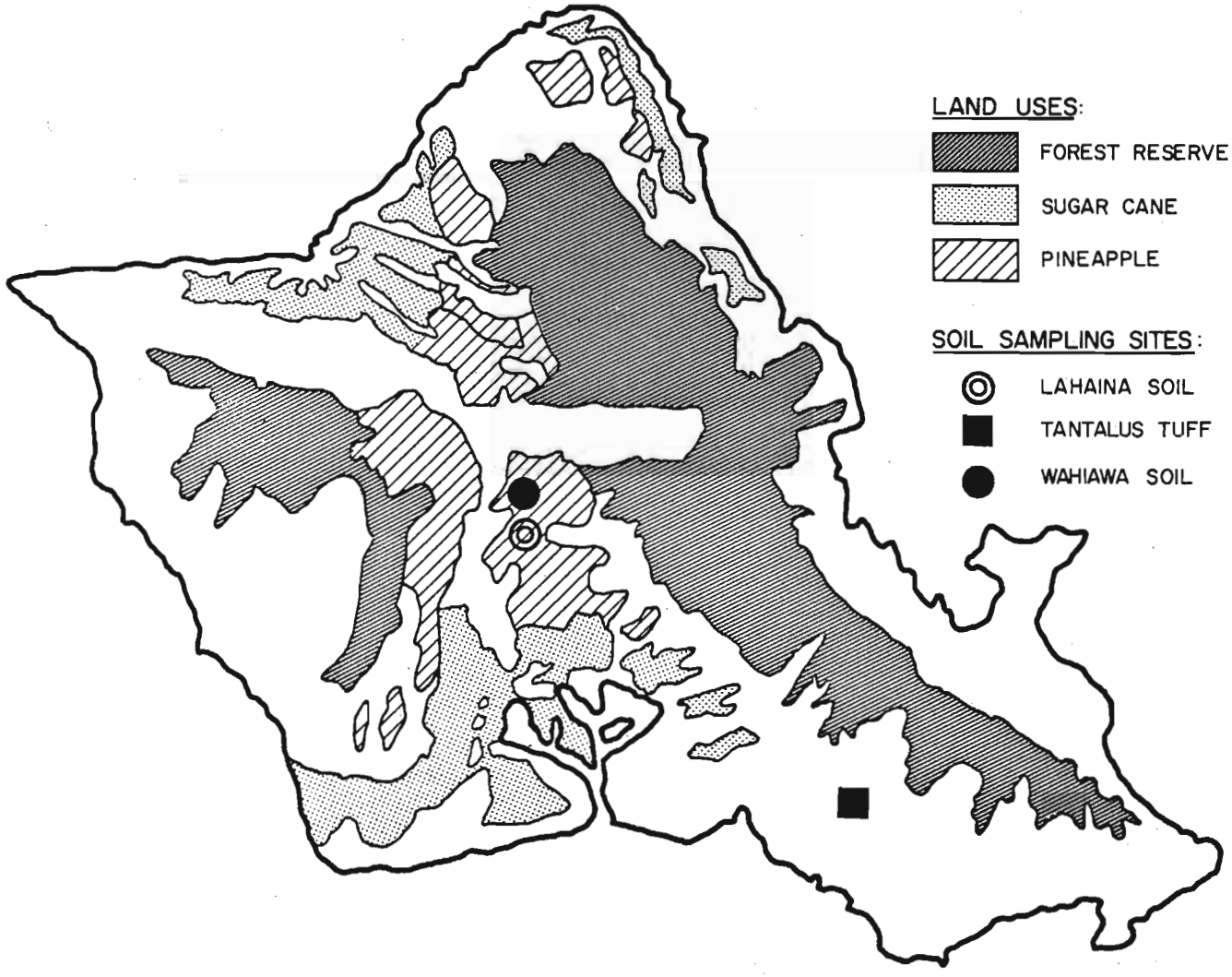


FIGURE 1: MAP OF OAHU SHOWING LAND USES AND SOIL SAMPLING SITES.

iron, aluminum, and titanium is considered high by temperate-region standards (11).

Most soils found on Oahu are considered clays by mechanical analysis classification. In the fields, however, these soils exhibit properties similar to silty clay loams. Sherman and Alexander (12) found all stages of clay mineral breakdown in Hawaiian soils. Among the Great Soil Groups occurring on the island of Oahu are the Low Humic Latosols and the Humic Latosols (10).

On Oahu, 95 percent of the total agricultural crops are cultivated in Low Humic Latosols (personal communication, P. C. Ekern). The Low Humic Latosols occur in dry to moderately humid areas where the accumulation of organic matter is small compared to that accumulated in more humid and more heavily vegetated areas. This Great Soil Group has a very weak to moderately strong A horizon over a red or reddish-brown alluvial B horizon that grades to parent material. Iron and aluminum are concentrated throughout the solum, mainly because silica and bases have been lost through weathering of the primary minerals, thus reducing the soil materials mainly to clay. Another distinct characteristic is the concentration of manganese dioxide in the upper part of the profile (10). Low Humic Latosols are subdivided into soil families, two of which are Wahiawa[1] and Lahaina.[2] These two soils are suited to cultivation and are among the best in the Hawaiian Islands for pineapple and sugar cane culture.

The Humic Latosols, of which the Tantalus[3] soil series is a member, is differentiated from the Low Humic Latosols on the basis of the degree of expression of the A horizon and other characteristics associated with high rainfall (10). The Tantalus soil is mainly present in forests and, to a limited degree, as grazing lands and homesites.

Land use projection for Oahu during the period 1960 through 1980, as given in the General Plan for the State of Hawaii, shows an increase in urban area from 28,000 to 58,000 acres, while decreases are forecasted in plantation agriculture lands from 70,000 to 66,000 acres and open space lands (primarily forest reserves and grazing land) from 206,000 to 187,000

---

[1]See Appendix A.
[2]See Appendix B.
[3]See Appendix C.

acres (13). Thus, the expansion of suburban developments on Oahu will mean diverting of lands once set aside for forest areas and plantation use. Furthermore, this will reduce the natural recharge of the basal water while creating a higher demand for domestic water.

Although there is no accurate record of the number and locations of existing cesspools, an estimate of 35,000 cesspools spread out over the entire island of Oahu has been established. Additionally, some new urbanized areas on Oahu rely on cesspools as temporary disposal of domestic waste (14).

Fortunately, there has been no report of contamination of Hawaiian ground water by enteric viruses, a group that includes the Polioviruses, the Coxsackie viruses, the ECHO (enteric cytopathogenic human orphan) viruses, the adenoviruses, and the viruses of infectious hepatitis. In a 1960 survey by the American Water Works Association, Hawaii was one of only three of the 50 states that could report no health incidents due to water contamination (15). However, this is by no means an indication that this condition will remain unchanged.

PURPOSE AND SCOPE

This laboratory study examined the ability of selected Oahu soils of given thicknesses to remove or adsorb bacteriophage T_4 from percolating water. Columns containing three Oahu soils, Wahiawa, Lahaina, and Tantalus were subjected to intermittently percolating water containing a known concentration of the phage.

The Wahiawa and Lahaina soil series are two very common soils found on the island of Oahu. They occur throughout the central Wahiawa plain where the predominant land use is for the cultivation of pineapple and sugar cane. The Tantalus soil is mainly found in the Tantalus spur of the mountains behind Honolulu, a typical watershed area. Soil selections for this study were based on their location in areas where impermeable strata to hinder surface waters from penetrating the soil cover and entering the underground basal-water body were sparse.

LITERATURE SURVEY

Bacteriophages--the Coliphage T_4

The name "bacteriophage" was given by F. d'Herelle (16) to a bacteriolytic substance that he isolated from feces. The name, usually shortened to "phage," means "eater of bacteria" and refers to the remarkable ability of bacteriophages to bring lysis to growing bacterial cultures. Today, phages are universally recognizes as a group of bacteria-specific viruses, that is, ultramicrobes of diverse character exhibiting all the signs of a long history of manifold variation, adaptation, and specialization (17, 18).

For convenience, phages are described as typhoid phages, staphylococcal phages, or coliphages, meaning phages attacking the indicated types of bacteria. In addition, phages have been given further individual designations, such as T_4, P_8, or lambda, which serve to identify a particular phage. These symbols are only used for indicating the origin of the phage from a particular collection. Coliphages T_1 through T_7 have in common only the ability to infect a strain of *Escherichia coli* known as B (19). The coliphages are morphologically identical and serologically related but not identical.

The taxonomy and nomenclature of bacteriophages is in an unsatisfactory state at the present time for a number of reasons. There are no generally applicable taxonomic criteria available for defining the limits of the species and genus of bacteriophages. Because of the lack of information about the phylogeny of phages, it is not possible to consider taxonomic categories above the genus level. The relationships among different groups of phages, of phages to other viruses, and phages to their host cells are not definitive at the present. Adams (18) emphasized that much of the phage literature is worthless for taxonomic purposes because of the impossibility of identifying the phage strains that were used.

The infective process of bacteriophage can be divided into four arbitrary stages, as discovered by d'Herelle in 1926 (20): (1) Adsorption of the phage particle to the host bacterium cell, (2) penetration of the phage particle into the bacterium, (3) the intracellular multiplication of the virus, and (4) the lysis of the host cell and release of about

100 phage progeny to complete one growth cycle. The 100 phage progeny from the first cycle will then infect 100 bacteria to initiate the second growth cycle. The progeny of the second cycle will initiate a third cycle and so the process will continue at an exponential rate until all susceptible bacteria are lysed.

Similarly, if a phage particle is added to a film of susceptible bacteria growing on an agar surface, it will adsorb to a bacterium and initiate the first lytic cycle. The phage progeny from the first cycle will infect neighboring bacteria to initiate the second cycle, thus providing a spreading lesion in the bacterial film. Eventually this process will result in a readily visible area in the film of growing bacteria, the bacteria-free clear circular space is known as a plaque. Under ideal conditions each infective phage will produce a plaque, so that plaque counts give a simple and reproducible measure of the number of phage particles present in a sample innoculated in the assay plate.

Microscopic measurement of the size of virus particles is complicated by shrinkage and distortion on drying, increase in size by shadowing with metals, and difficulties in calibration. Some very careful measurements have been published as to the size of the phage T_4. In electron micrographs, the head of the phage was measured to be 65 to 95 millimicrons, and the tail, 25 to 100 millimicrons (21). By a diffusion method, Polson (22) found the diameter of the phage to be 55 millimicrons. Watson (23) found the diameter to be 50 millimicrons by using X-rays.

Bacteriophages are usually stable over the pH range of 5 to 8. At low temperatures, the range can often be extended from pH 4 to pH 9 or 10 (18). Adams (24) found that at a temperature range of 65° to 75° coliphage T_4 was inactivated. The effect of detergents vary on different phages.

In the main, antibiotics prevent phage formation only to the extent of their antibacterial properties (18). Dyes such as crystal violet (25) and malachite green (26, 27, 28) have been shown to differentially inhibit phage production without affecting the activity of the free phage.

The r mutations of phage T_4 occur at many different genetic loci (29, 30) and are responsible for the high over-all frequency of the mutations. The expression of the character depends on the bacterium; certain mutants are rapid lysers on *E. coli* B, but produce lysis inhibition on *E. coli* K12 and other bacteria (31). Different r mutants of the same phage vary greatly

in physiological properties (32, 31). Cohen (33) suggested that the r mutants contain a larger amount of glucose as a constituent of their nucleic acid than their parental phages.

The r mutation was probably first seen by Sertic (34) and Demerec and Fano (19). Almost any mutation in a phage is likely to alter the appearance of its plaques. Small plaque mutants in coliphage T_4 were described by Doermann and Hill (30). The r mutants from characteristic plaques, which explain their importance in early genetic studies (18).

The Movement of Viruses Through Soils and Related Studies

In the past twenty-five years, very limited studies have been published in the literature on the movement of viruses through soils. Relatively speaking, only a few studies have been undertaken to investigate the removal of viruses by water and waste treatment processes (35, 36, 37, 38, 39). Four of these studies involved various sands, while one included a garden soil (38). Based on the results of these limited studies on virus movement, it appears that adsorption is the removal mechanism and that the removal efficiency is a function of the rate of water movement. Indeed, little is known about the virus removal capacity of soils on the effects of environmental conditions (chemical and physical properties of the soil and ground.water) that might influence virus movement in ground water.

Investigators in the waste-water reclamation project at Whittier Narrows (40) attempted to isolate enteric viruses from test basin percolate after two feet of travel through the soil. Samples were obtained before and during mass community feeding of Sabin poliovirus vaccines. No enteric viruses were isolated from the percolate.

Gilcreas and Kelly (38) did an experimental study on the effect that percolation through soil has on viruses in a water solution and stated:

> Seepage of water through soil has a dual role as far as pollution problems are concerned. In the drainage of surface waters, seepage through soil amy provide purification. Seepage may also, however, serve as a channel from a highly polluted source (such as a privy or cesspool) to an unpolluted one.... The comparative behavior of enteric viruses and bacteria during seepage of water through soil is of importance in determining the validity of the use of the coliform index under such conditions. To compare the penetration of viruses and bacteria through soil, suspensions of Coxsackie virus and

> *E. coli* in water were allowed to percolate through garden soil (moisture content, 4%) contained in glass tubes 36 mm in diameter. A mixture of the agents in 300 ml of spring water was added to 6, 18, and 36 inch columns of soil and the percolates were collected. Percolation through at least 3 feet of soil was necessary for the reduction of either viruses or coliform organisms, and even that distance of travel, however, was insufficient to remove them completely.

A histogram given by Gilcreas and Kelly shows about 50 percent recovery of Coxsackie virus, 30 percent recovery of *E. coli*, and 75 percent recovery of bacteriophage following percolation through three feet of soil. Thus, 50 percent reduction of Coxsackie virus titre was demonstrated after percolation through soil of 4 percent moisture content. Neither the percolation distances required for effective removal of the virus, the effect of rate of flow, nor the effect of different types of soil were investigated.

Epidemiological studies on the ability of infectious hepatitis virus to pass through soils have been made by Hallgren (41) in 1942. He investigated the water supply of a Swedish sanatorium where an epidemic of infectious hepatitis had occurred and found the source of the infection to be a concrete water tank. The tank was sunk through the moraine gravel down to the underlying primaeval rock and lay 12 feet, slightly downhill, from the main sewer of the sanatorium. Investigation revealed a break in the sewer near this point. The ground was frozen at the time of the contamination of the water in the tank.

Neefe and Stokes (42) in 1946 investigated sanitary facilities at a summer camp in the Pocono mountains where numerous cases of infectious hepatitis had been traced to a contaminated well water. Back flow from toilets, direct leaks, and surface contamination were ruled out and it was surmised that the well had become contaminated by seepage from adjacent cesspools several of which lay within 200 feet of the well. The authors reported the following:

> These cesspools were 6 to 8 feet in depth and had open bottoms, stone walls, and cement tops.... None of the cesspools had overflowed prior to the start of the epidemic. In fact, although in use for a number of years, drainage of the cesspools had never been necessary, indicating that the contents diffused into the ground. The ground layer of the area, varying in depths from a few inches to approximately 4 to 6 feet, consisted of ordinary top soil overlying a layer of 'hardpan'. The exact nature of the underlying rock was not determined but it was said to consist of red shale and limestone. In certain places where

> the rock layer protruded through the soil and could be inspected, many cracks and fissures could be detected.

Farquhar, *et al.*, (43) found well water to be the cause of a small outbreak of infectious hepatitis in Pennsylvania. The water appeared to have become contaminated from a cesspool which lay in slightly higher ground about 50 feet away. The creviced shale in the area was overlaid by a thin layer of top soil. Evidently the passage of the virus was facilitated by extremely wet soil in a season of heavy rainfall.

Tucker, *et al.*, (44) in describing conditions prevailing at a camp where 102 cases of hepatitis were contracted stated the following:

> The camp ... is located in a narrow valley on the western side of the Highland Rim section of the State. This section is typical of a great part of middle Tennessee in that the top soil is underlaid with limestone with frequent outcroppings.... It was discovered that although the main sewer line was constructed of metal sewer pipe with lead sealed joints, the sewer lines extending from the cottages to the main sewer were laid with terra cotta pipe

Sodium fluorescein dye was placed in the toilet of the nearest cottage which was located about 50 feet from the spring supplying water to the camp. The dye appeared in the spring after 40 minutes.

Peczenik, *et al.*, (45) in 1956 traced an epidemic of infectious hepatitis in an Austrian hotel to contamination of water from the septic tank of an adjacent hotel where there had been one case of the disease. The septic tank was approximately 20 feet above and 122 feet distant from the water reservoir. The area was described as having a fairly steep gradient and an abundance of ground water. The terrain was composed of fissured limestone overlain with a layer of humus and fine and medium gravel.

An epidemic of hepatitis was investigated by Mosley and Smither (46) in Utica, Kentucky. It was believed that half of the cases became infected via contamination from private wells. These authors stated that the six households in which infections occurred in a number of visitors obtained their water from drilled wells located on their property or on an adjacent lot. The depth of these wells varied from 80 to 212 feet, but reliable information concerning depths of casing, type of casing used, or adequacy of sealing could not be obtained.

Studies relating to the removal of viruses above the ground-water table by filtration and absorption properties of the soil and also by

purification of surface water prior to seepage through the soil were conducted by a number of researchers, dating back to at least 1942. Filtration depends on the size of the viruses, which are known to pass through much smaller pore diameters than bacteria. Chemicals, particularly the breakdown products of sewage, have been shown to penetrate through the soil about twice as far as bacteria (47, 48). On the basis of size, the distance traveled by viruses should be somewhere between chemicals and bacteria. Preliminary work by Gilcreas and Kelly (38) in 1955 is in agreement with this hypothesis.

Earlier studies by Carlson, *et al.*, (36) showed that sand filtration did not remove polio virus from water unless the filter was blocked with large amounts of aluminum floc, producing a very low flow rate. Evidently virus was removed by adsorption rather than filtration, as might be expected.

Adsorption of an agent by the soil would be dependent upon surface area and chemical nature. The small size of enteric viruses, having approximate dimensions of 25 to 30 millimicrons (49), should favor their adsorption by the soil. However, studies of virus adsorption by soil have not yet been made.

A number of experiments in water purification have been carried out with both polio and Coxsackie viruses using the aluminum floc technique of coagulation and sedimentation (36, 50, 51). All of these studies indicated that the viruses were less effectively removed by this treatment than were bacteria. Evidently, those viruses which were removed remained active while adsorbed to the floc (36).

In related studies, the survival of viruses in water show that both polio and Coxsackie viruses remain active in distilled, tap, or river water for many months (52, 38). Gilcreas and Kelly (38) also showed that the presence of phosphates prolonged the survival of Coxsackie viruses. It is quite possible that other salts, which might be present in soils--perhaps occasionally in rather high concentrations could exert either a protective of a destructive effect on the viruses. Synthetic detergents, present in sewage and occasionally in contaminated ground water, were used under certain laboratory conditions to disrupt viruses into their constituent macromolecules (53).

In a study relating to superchlorination of water supplies, it was concluded that enteric vegetative bacteria are most easily destroyed by

chlorine, but cyst forms of intestinal protozoa and the spore-forming bacteria are most resistant to chlorine. Relatively little is known about viruses and their resistance to chlorine except that their resistance probably falls between that of vegetative bacteria and the cyst forms of intestinal protozoa (54).

It has been calculated that the relative density of enteric viruses to coliforms in sewage is about 1 to 65,000 (55), a ratio which makes detection of coliforms considerably easier than virus. Thus, viruses at the present cannot be used as a measure of water quality because it is possible to have fecal pollution with associated enteric pathogenic bacteria in the absence of viruses. However, it is also possible to have viral agents present in the absence of bacterial indicators of pollution, because viruses and bacteria react differently to environmental changes (56).

Bacteriophages, a Possible Alternative to Pathogenic Virus Assays

The assay of phages that occur regularly in sewage is comparable in expense and time to that of bacteria. However, coliphages, like *E. coli* itself, may differ considerably in their removal properties from the pathogenic enteroviruses. Bacteriophages are on the order of 50 to 95 millimicrons, somewhat larger than enteroviruses which normally measure 25 to 30 millimicrons.

Experimental studies (57, 38, 51) indicate that behavior of coliphages is more like behavior of enteroviruses than that of bacteria, but in some cases they are more easily removed from water than enteroviruses. In this regard, for example, adoption of coliphage content as a standard of water quality would perhaps be somewhat of an improvement over the coliform index, but results must still be interpreted "with discretion."

Sinsheimer (58) in 1959 described a coliphage comparable in size to enteroviruses. Survival behavior of this coliphage may be expected to be more similar to that of enteroviruses than the larger well-known T-coliphages. It is possible that this coliphage may provide a very desirable test organism for soil studies.

METHODS AND PROCEDURES

The Coliphage T_4

The coliphage T_4Br^{II} mutant, effective against *Escherichia coli* strains Bb and HS/6 strep^r (streptomycin resistant mutant), was utilized in this laboratory study. A culture of *E. coli* strain Bb was used to prepare high titre phage stocks and a culture of *E. coli* strain HS/6 strep^r was used to assay the phage T_4, using an agar layer (plaque counting) technique.

Since bacterial viruses and animal viruses are believed to be sufficiently similar, possessing many closely identical physical, chemical, and biological properties, *i.e.*, size, net electric charge, protein coating, etc. (18), extrapolation of the coliphage T_4 data may, to varying degrees, be useful in evaluating the significant parameters that influence the general movement of viruses through soils. Furthermore, because there is much more known about the composition, properties, and behavior of bacteriophages than is known about animal viruses, a more complete account can be made of their behavioral movement in soils.

The Media

Tryptone broth, containing 13 gm Bacto tryptone, 8 gm sodium chloride, and 1 gm Bacto dextrose per liter of distilled water (59), was prepared for all routine assaying dilutions and growing broth cultures of the *E. coli* strains. Tryptone hard agar plates and soft agar were prepared according to directions by Chase and Doermann (60). Twelve grams of Bacto agar were added to the tryptone broth for the plate agar. The bottom layer of hard agar was overlaid with a thin layer of the more dilute soft agar, made by adding 6 gm Bacto agar to the tryptone broth which contained the susceptible bacterial host, *E. coli* HS/6 strep^r, and the phage to be assayed.

Preparation of Stock Phage

During the course of this research, two 200-ml quantities of tryptone broth containing newly prepared phage T_4 stock, having concentrations of $2.5 \times 10^8 \pm 20\%$ and $1.5 \times 10^{10} \pm 20\%$ phage per ml of solution, were utilized.

The bacteria *E. coli* Bb were grown in freshly prepared tryptone broth.

Two-hundred fifty milliliters of the broth were prepared and autoclaved for 25 minutes at 15 pounds per square inch at 250°C, and cooled in a tap-water bath. The *E. coli* was transferred with a transfer loop from a refrigerated 18-hour growth stock culture on a tryptone agar slant and inoculated into the sterile broth. The flask containing the transferred bacterial strain was then shaken in a Gyrotory Shaker Model RW-150 for about 9 to 10 hours in the water bath maintained at a constant temperature of 37°C until the broth appeared turbid with bacterial growth. The *E. coli* bacteria were, at this point, in the exponential or log phase of growth.

A new phage T_4 stock was prepared by utilizing an old refrigerated high-titre phage stock of unknown concentration. About 0.5 ml of this old stock was innoculated into the flask containing the tryptone broth and the 9 to 10-hour growth of the *E. coli*. Again, the flask was shaken at 37°C but for 17 to 18 hours until the turbid broth appeared favorably clear. This clear appearance indicated that the majority of the *E. coli* cells had been lysed by the rapidly multiplying bacterial virus. Several drops of $CHCl_3$ (chloroform) were added to this cleared broth in an attempt to lyse any remaining viable *E. coli* cells.

The 250-ml broth of the virus was then centrifuged in a Sorvall Angle Centrifuge Model NSE for twenty minutes at approximately 5000 RPM to remove the lysed bacterial cells and debris. The supernatant, sterilized with several milliliters of chloroform, was collected in a flask with a screw-on cap and stored under refrigeration.

A schematic diagram of the procedure involved in preparing the stock phage is shown in Figure 2.

Assaying the New Phage Stock

A culture of plating bacteria *E. coli* HS/6 strep^r was prepared for assaying purposes from a refrigerated 18-hour growth stock on tryptone agar slant by transferring this strain into a flask containing 250 ml of freshly prepared sterile tryptone broth. The flask was shaken at 37°C for 8 to 10 hours until the broth appeared turbid. The new stock of plating bacteria was then stored under refrigeration.

Sufficient quantities of tryptone plate agar and soft agar were prepared and autoclaved for 25 minutes at 15 psi and at 250°C. Thirty to 35 ml of the liquefied plate agar, cooled down to 55 to 60°C, were poured

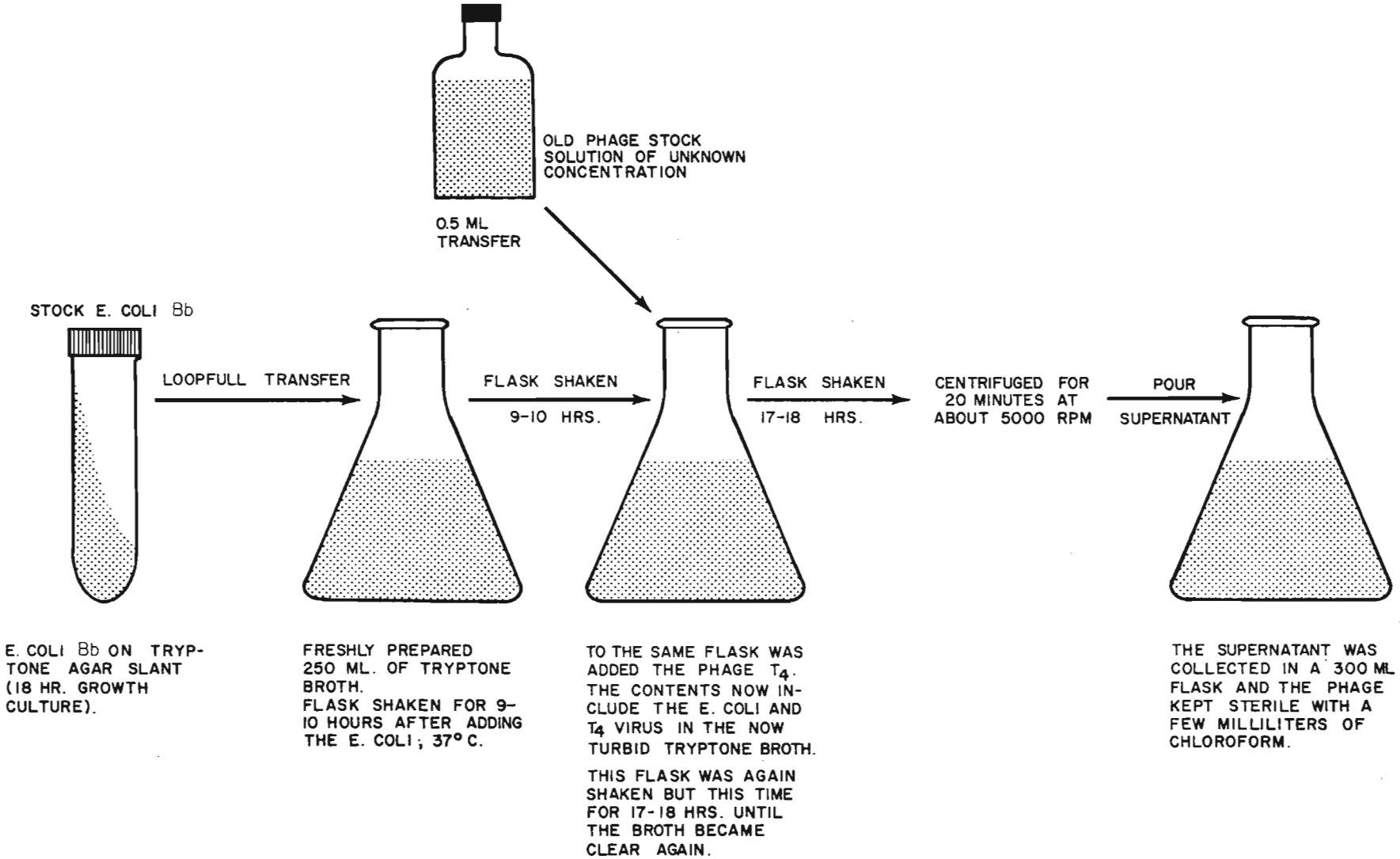


FIGURE 2: SCHEMATIC DIAGRAM OF THE PROCEDURE USED IN PREPARING THE STOCK PHAGE.

into each sterile disposable pertri plate. These plates were then allowed to stand until the agar solidified. They were incubated overnight at 37 to 38°C and were stored under refrigeration the following morning. After being cooled down to 55 to 60°C, the soft agar was poured into test tubes with screw-on caps in 3-ml quantities and autoclaved as mentioned before. Soft agar was kept liquefied in a hot-water bath maintained at 48 to 55°C.

Test tubes containing 5 ml of sterile tryptone broth were required for serial dilutions from the new phage T_4 stock solution. Serial dilutions were made according to the procedure outlined in Figure 3. A sterile pipette was used for each transfer.

From a dropper bottle containing plating bacteria, five to six drops were added to the test tubes containing 3 ml of soft agar in a hot-water bath. Then, 1 and 0.1-ml samples of the diluted virus solution, having a dilution factor of 10^{-5} and a 1-ml sample from the solution having a dilution factor of 10^{-7}, were transferred into test tubes containing soft agar and plating bacteria. This resulted in dilution factors of 10^{-5}, 10^{-6}, and 10^{-7}, respectively, from the new phage stock solution.

The 3 ml of soft agar, plating bacteria, and diluted phage in the test tubes were shaken to obtain uniform dispersion and were poured onto hard agar plates appropriately marked as to dilution factors. Duplicate plates were made for each dilution and the top layer of agar was allowed to solidify. The petri plates were then inverted and incubated at 37°C for a period of 14 to 16 hours.

Counting the Phage

The standard plaque test was used to enumerate virus particles. The petri dishes were removed from the incubator for plaque counting. If no viruses were present, the bacteria grew uniformly over the entire surface of the agar. If viruses were present, clear holes (plaques) were found distributed in the growth of bacteria (Fig. 4). Each plaque represents one virus particle from the sample tested. The plaques of the coliphage T_4 are about 1 to 2 mm in diameter (average size about 1.5 mm in diameter) and can be counted with or without the use of a conventional bacterial colony counter.

If plaques were numerous making counting difficult, the petri plates were divided into quarters or eighths and the plaques in each section counted

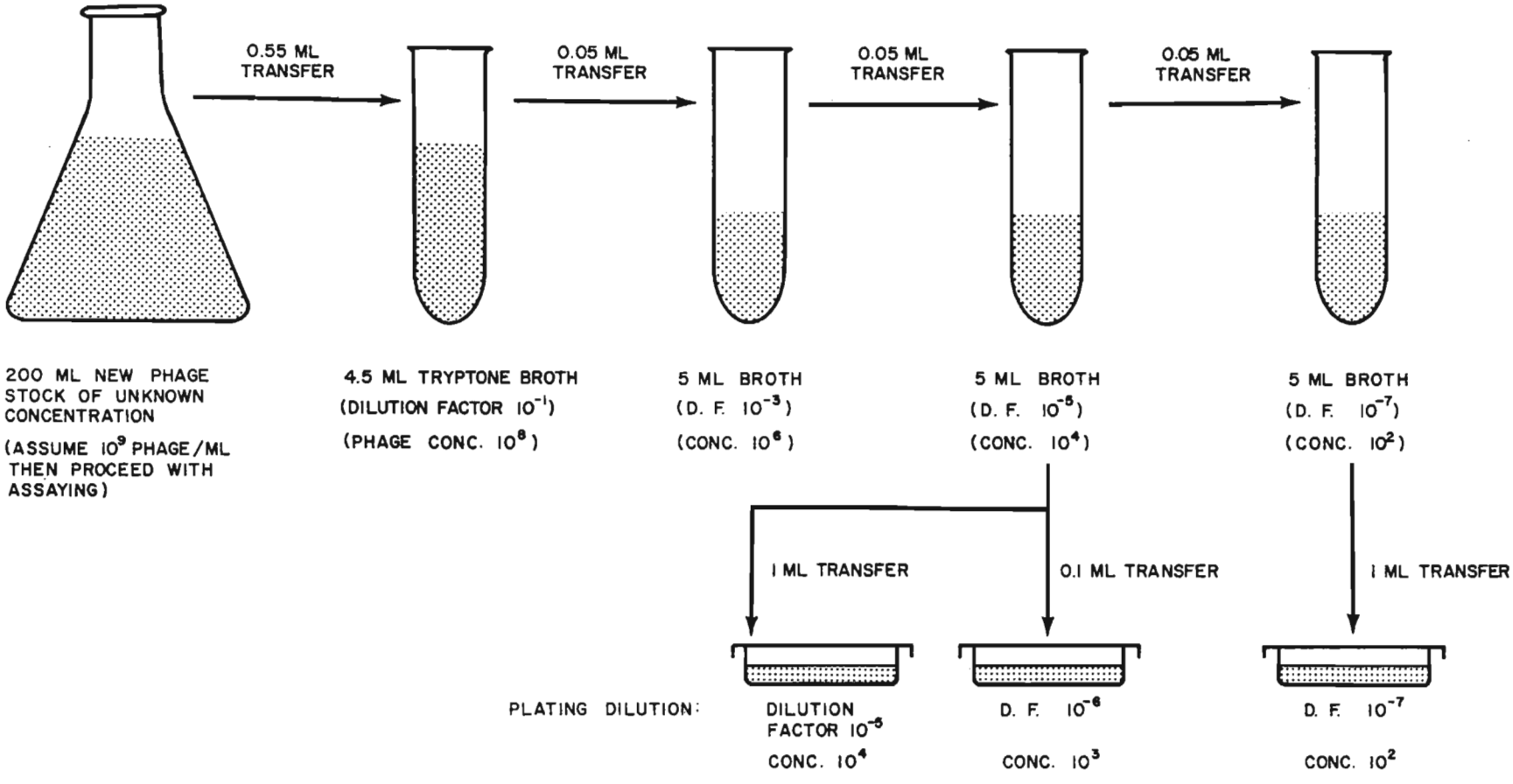


FIGURE 3: SCHEMATIC DIAGRAM OF THE PROCEDURE USED IN ASSAYING THE NEW PHAGE STOCK.

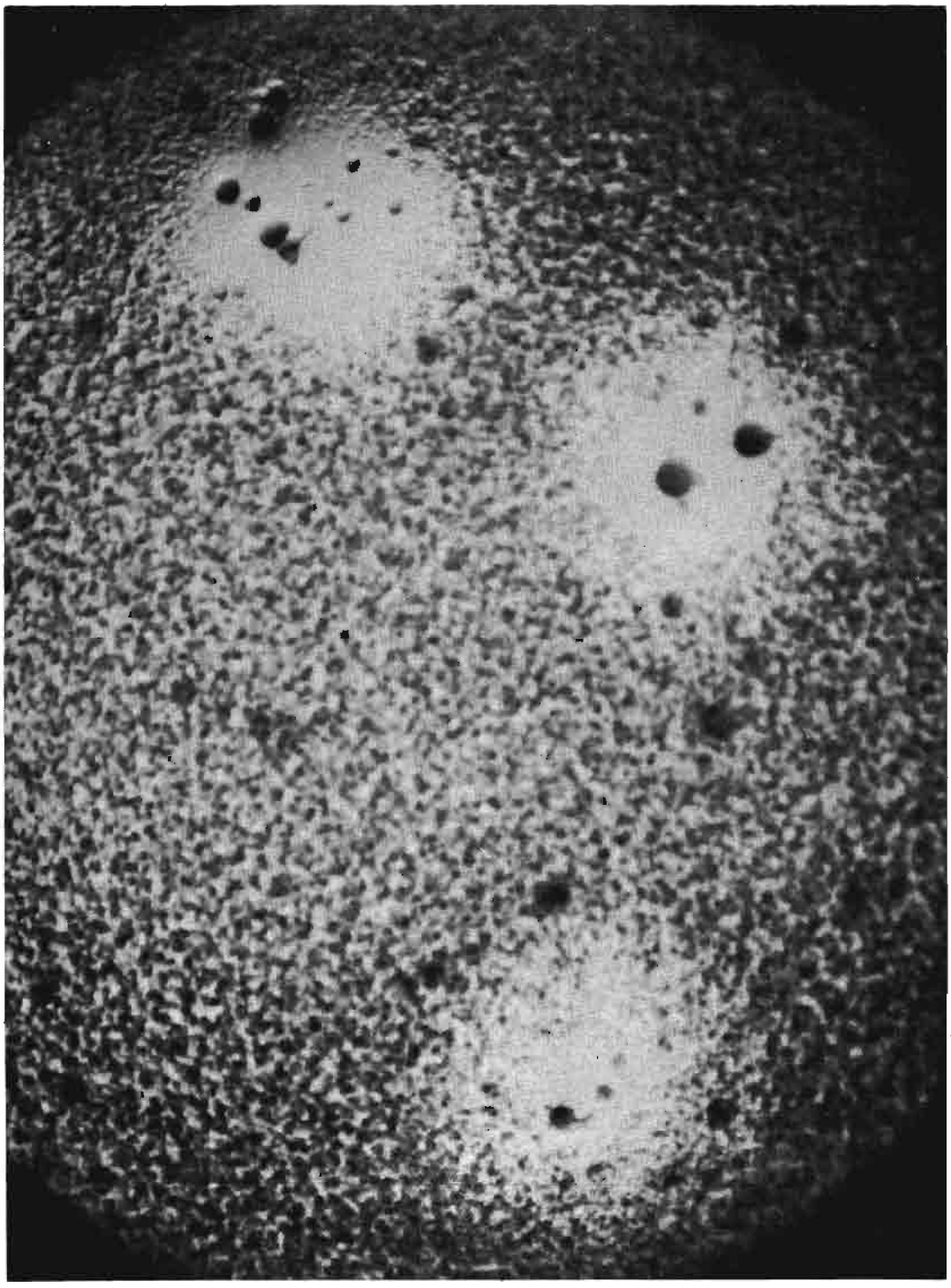


FIGURE 4: PHOTOMICROGRAPH OF PLAQUES OF COLIPHAGE (MAGNIFIED: 40 X).

and totaled. If plaques overlapped to a great extent and large clear spaces peppered with remaining bacterial colonies developed throughout the surface of the agar, confluent lyses occur resulting in too many virus particles to be counted. Figure 5 illustrates a form of confluent lyses. The plaque test is considered the most accurate available method for bacterial virus enumeration at the present.

Sterile media and aseptic techniques were employed in all operations of growing and assaying the coliphage T_4 and *E. coli* strains.

Preparation of Soils and Soil Columns

The two silty clay soils utilized in this study, the Wahiawa and Lahaina series, were collected from sites located close to sampling pits selected by the U. S. Soil Conservation Service in their preparation of a new soil map of Oahu. The third soil type, the Tantalus series, is the gravel-sized cindery material collected along the roadside on Sugar Loaf near Mt. Tantalus. (See Appendices A, B, and C for the current up-dated descriptions and nomenclature.)

Soil samples of the Wahiawa and Lahaina series were obtained from depths of 3 to 12 inches in the A horizon and transferring them to the laboratory. The Tantalus soil samples were obtained from depths of 29 inches in the C horizon.

The Wahiawa and Lahaina soils were screened through a #10 sieve to remove larger lumps of soil, root fragments, and other debris. The screened soils were then placed in 2-liter beakers, sealed, and autoclaved for 40 minutes at 15 psi and 250°C. (For the purpose of this study autoclaving was essential in destroying all biological life that may have been present in the soils.) The Tantalus cinder was autoclaved in the same manner as above but was not passed through a #10 sieve.

The pH of the three sterile soils was determined with a Corning pH Meter, Model 10, by analyzing a paste formed from a mixture of 10 ml of sterile distilled water (pH 6.2 to 6.35) and 10 grams of the soil passing a #10 sieve.

Plastic 18 x 1-3/4-inch cylindrical columns were utilized in this study. They were covered at the base by a #16 mesh screen, sterilized by washings with detergent, distilled water, and 95-percent ethanol and then allowed to dry. Each column was packed with sterile glass wool to a height

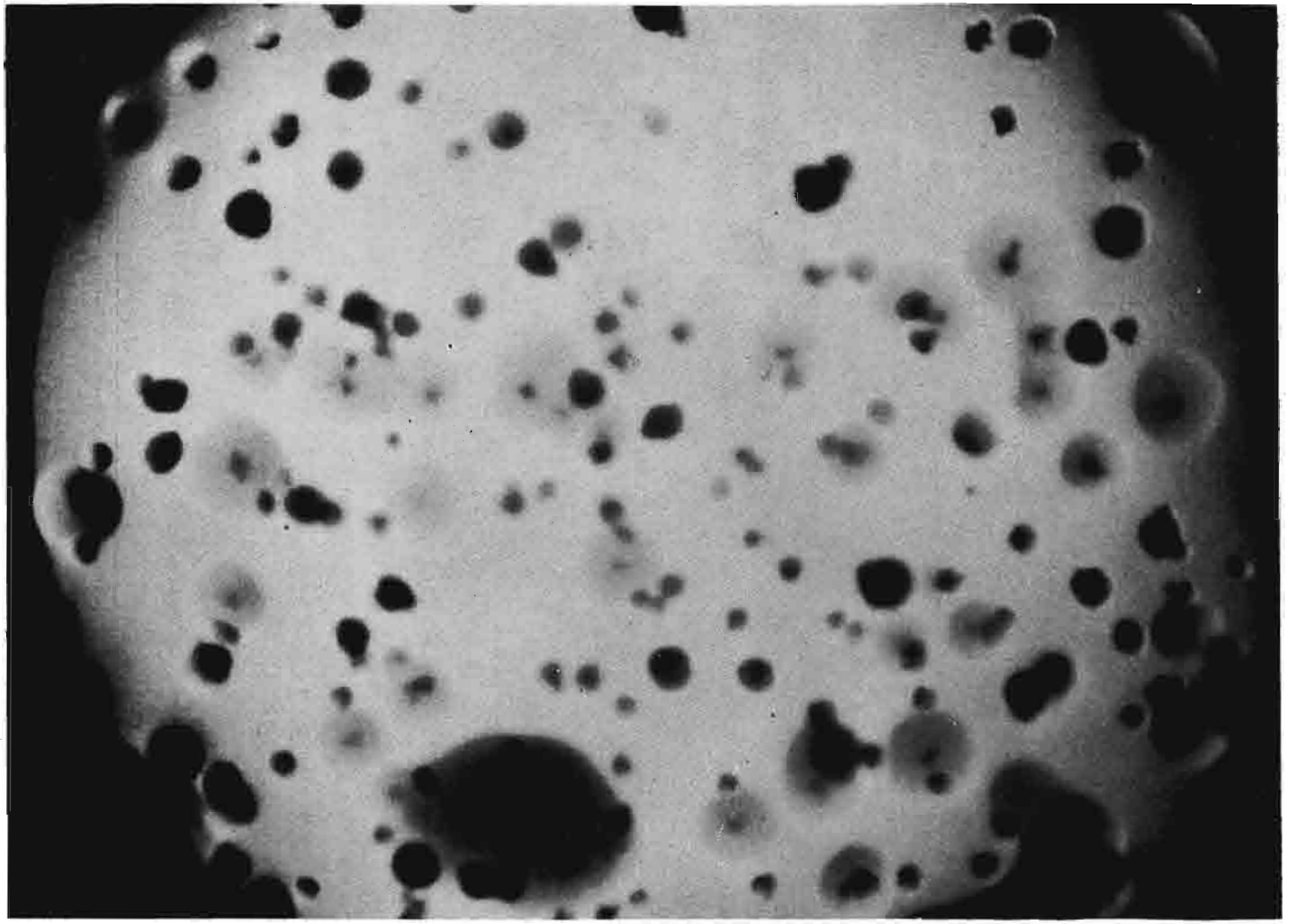


FIGURE 5: PHOTOMICROGRAPH OF CONFLUENT LYSIS OF E. COLI BY THE COLIPHAGE T_4 (MAGNIFIED 40 X).

of 2½ inches and filled with soil to specified thicknesses. The soil columns were conditioned with sterile distilled water for two days prior to the experimentation at a rate of 10 inches per day. The conditioning consisted of five two-inch pondings of the water on the soil surface. The water was allowed to drain through the soil by gravity flow. This was done in an attempt to restore the soil to its natural structure and to soak the soils in preparation for the percolation tests.

For the major part of the study, twelve columns were utilized. Six were packed with Wahiawa soil and six with Lahaina soil. In each soil type, three columns were used for virus tests and three were controls. The column arrangements for both soils are shown in Figures 6 and 7.

When experimentation on the Wahiawa and Lahaina soils were concluded, seven of the columns were washed, sterilized, dried, and packed with Tantalus cinder. Six were used for virus tests and one for control. Percolation tests for the Tantalus cinder were carried on for only one day.

During the course of the experiment, the test columns were dosed with phage T_4 in diluted concentrations of 10^6 (2.5×10^6 and 1.5×10^6) per ml with sterile distilled water, which had been previously autoclaved for 20 minutes at 15 psi and 250°C. The control columns were dosed with sterile distilled water.

The water was applied to the columns by carefully pouring it on the soil surface and allowing the liquid to pond up to half an inch. After the initial dose passed into the soil, the second half-inch ponding was applied. Thus, one inch of water was percolated through the soil at each sampling run per column. Six 1-inch applications, each 1½ hours apart, were run per day for five consecutive days. Hence, 30 inches of water was applied to each test column and control column. This application was carried out for the Wahiawa and Lahaina soils. This intermittent dosing attempted to simulate the loading of a cesspool or a septic tank leaching field.

Since Tantalus cinder is characterized by rapid drainage, the ponding application was not utilized. A 250-ml dosage of the liquid (equivalent of a 0.62-inch depth of liquid in the column) was applied to the cindery substratum per sampling run per column. Six sampling runs, one hour apart, were made for each of the five days of tests.

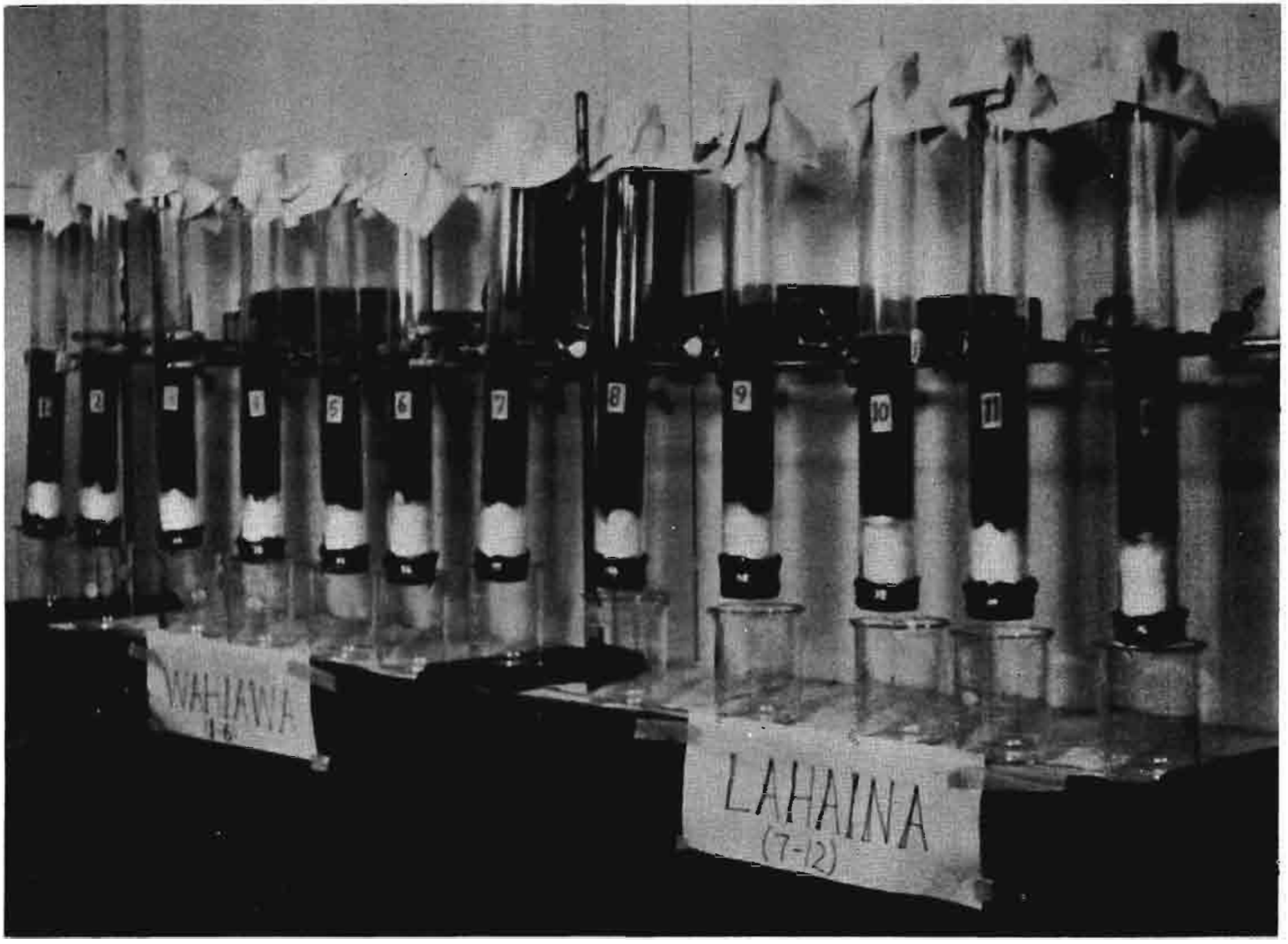


FIGURE 6: PHOTOGRAPH OF COLUMNS FOR PERCOLATION TEST - WAHIAWA AND LAHAINA SOILS.

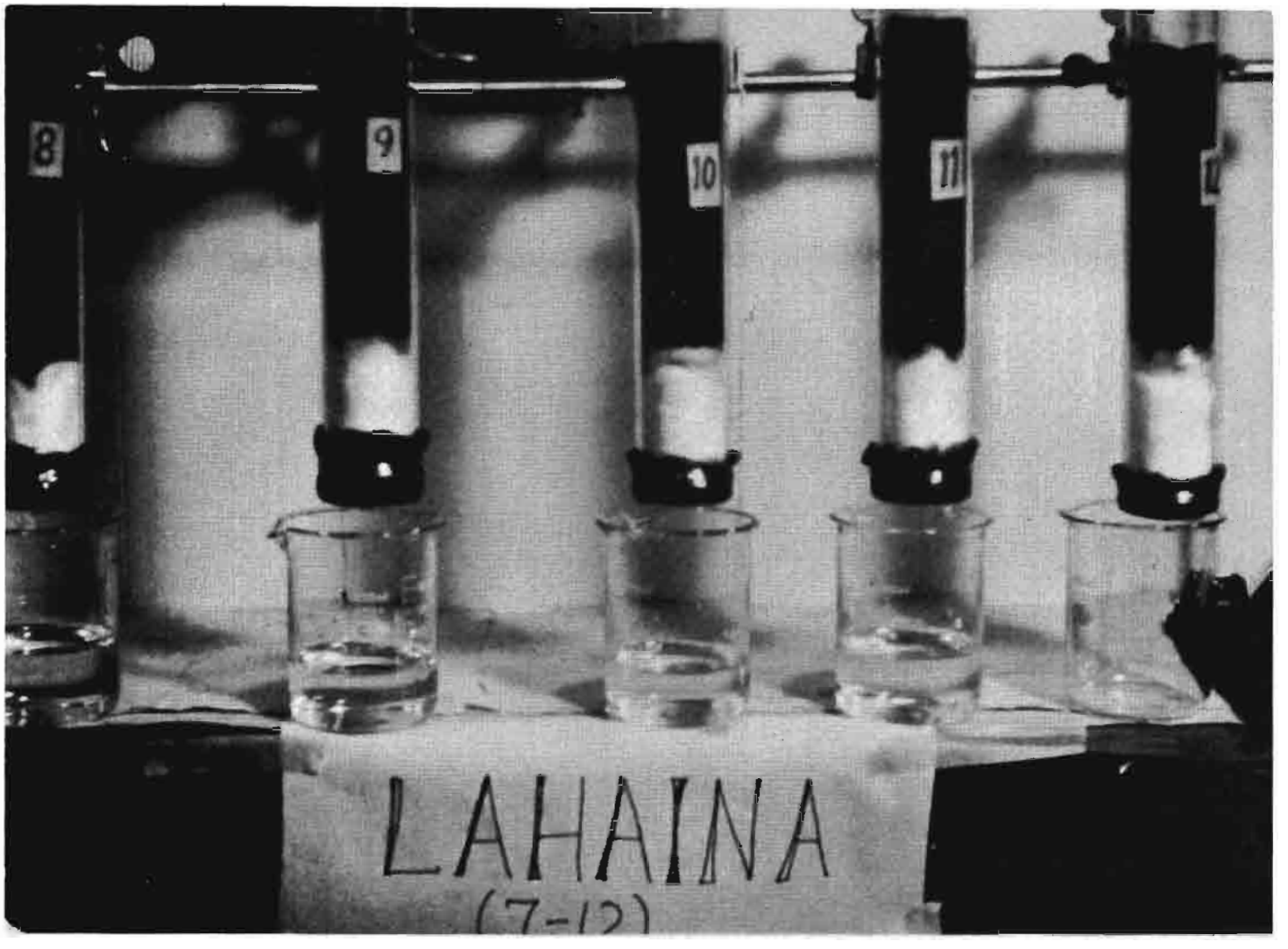


FIGURE 7: PHOTOGRAPH OF CLOSEUP OF COLUMNS OF WAHIAWA AND LAHAINA SOILS.

Percolation Tests

Percolation tests on soil thicknesses of 6, 2½, and 1½ inches were carried out on Wahiawa and Lahaina soils. On the Tantalus cinder, a single percolation test was conducted on six test columns divided into 3 pairs at 15, 12, and 6-inch soil thicknesses, and on the control column, at a soil thickness of 12 inches. The materials in all the columns were essentially in an unsaturated condition prior to and during the percolation tests.

Assaying the Percolant

The percolant of the test and control columns was collected in a sterile beaker. One millimeter of the percolant was transferred by a pipette into a marked test tube. Assaying for the T_4 virus was carried out using serial dilutions of the percolant and agar layer plaque-counting techniques as previously described. Daily control plates were made to reassure the viability and concentration of the feed solution.

Soil Permeability Tests

Soil permeability tests were conducted by measuring the amount of percolant collected over a period of 10 minutes while maintaining a constant hydraulic head of two inches of water above the soil surface. The computations for laboratory coefficient of permeability (hydraulic conductivity) of the Wahiawa and Lahaina soils are outlined in Appendix D. The data has been corrected to $60^{\circ}F$.

All aspects of this study were performed in a laboratory where the temperature range was relatively constant between 22 to $22.5^{\circ}C$.

DISCUSSION OF THE RESULTS

Soil pH

Lahaina soil was more acid than Wahiawa soil, having a pH of 4.9 compared to the latter's 6.2. Tantalus cinder was found to have an alkaline pH of 7.7. The pH values recorded for Wahiawa and Lahaina soils were col-

lected from the A horizon (depths of 3 to 12 inches) and for Tantalus cinder from the C horizon (depths below 29 inches).

Some pH values obtained by other researchers, the U.S. Soil Conservation Service (61), Young (62), and Eto (63), are presented in Table 1.

TABLE 1. A SUMMARY OF SOIL pH

SOIL SERIES	CLASSIFICATION*	TANIMOTO	S.C.S.***	YOUNG	ETO
WAHIAWA	TROPEPTIC EUTHRORTHOXS	6.2	5.6	5.5	5.7
LAHAINA	TROPEPTIC HAPLUSTOX	4.9	5.2	6.3	5.9
TANTALUS	TYPIC DYSTRANDEPTS	7.7	--		

*Denotes classification as of July, 1966.
***U.S. Soil Conservation Service.

Permeability

Data was obtained on Wahiawa and Lahaina soils so that permeability could be computed. The average flow rate for Wahiawa and Lahaina soils was 102.4 ml/600 sec and 117.9 ml/600 sec, respectively, with a constant head of 5.08 cm (2 inches) on the soil surface. Lahaina soil is slightly more permeable than Wahiawa soil. Flow rates reduced to coefficients of permeability were 142 gal/day/ft^2 (24.2 cm/hr) and 137 gal/day/ft^2 (23.3 cm/hr). (See Appendix E.)

Young (62) reported coefficients of permeability of 38 gal/day/ft^2 (6.5 cm/hr) for Lahaina soil and 64 gal/day/ft^2 (11 cm/hr) for Wahiawa soil. There are other evidences of laboratory permeability for Wahiawa soil in the above orders (personal communication, B. G. Cagauan). It is recognized that permeability varies with packing and properties of the test fluid.

Permeability of Tantalus cinder was not determined in this study; however, Ishizaki and Young reported a value of 35,200 gal/day/ft^2 (6000 cm/hr).*

*Ishizaki, Kenneth and Reginald Young, WRRC Technical Memorandum Report No. 11, 1967.

Percolation Test Results of Wahiawa and Lahaina Soils

The percolation tests with the 6 and 2½-inch soil columns showed no breakthrough of the virus over a period of five consecutive days of runs with intermittent irrigation of 30 inches of water for each of the soil thicknesses. Assaying by the agar-layer plaque-counting technique indicated that no virus was found in the percolant collected from all sample runs of all test and control columns. Daily check platings from the feed solution proved that the phage was viable at the given concentration.

These results indicate that Wahiawa and Lahaina soils possess a high retention capacity for this virus. Virus removal was 100 percent.

The 2½-inch soil columns were separated for analysis of bacterial virus concentrations at various depths in the soil. All three test soil columns of both Wahiawa and Lahaina soils were separated into four layers as follows: 1 in, 2 in, 2¼ in, and 2½ in, and in the glass wool in the bottom of the column. A volume of 100 ml of sterile distilled water was added to each of the soil-layered specimens and the contents were shaken and stirred thoroughly and allowed to settle. One ml of the supernatant was assayed by the agar-layer technique. Table 2 shows the average numbers (plaques) of the phage detected in each soil thickness for both soils. The results gave some indication that the glass wool has some removal (adsorption) properties. Most of the virus probably accumulated in the layer of sediment that collected in the top surface of the glass wool.

Breakthrough occurred in the 1½-in soil columns of the Wahiawa and Lahaina soils under the same intermittent flow conditions and dosing that was used with the 6 and 2½-in soil columns. All of the 1½-in columns recorded initial breakthroughs at the same sampling time and with the same throughput volumes on the percolation system. However, the breakthrough numbers (plaques) of viruses were much less in Wahiawa soil than in Lahaina soil during the first two to three sampling days, as indicated in Table 3.

Breakthrough curves for the 1½-in soil columns of the Wahiawa and Lahaina soils are shown in Figures 8 and 9.

TABLE 2. CONCENTRATIONS OF THE COLIPHAGE IN THE SEPARATED SOIL THICKNESS LEVELS OF THE 2 1/2-IN TEST SOIL COLUMNS OF WAHIAWA AND LAHAINA SOILS

DEPTH FROM TOP OF SOIL SURFACE	WAHIAWA	LAHAINA
1"	TNTC⁂	TNTC
1"	TNTC	TNTC
1/4"	1.85×10^3 - TNTC	1.785×10^3 - TNTC
1/4"	1.247×10^3 - TNTC	8.45×10^2 - TNTC
2 1/2" GLASS WOOL	3.72×10^2	1.07×10^2

⁂ *Too numerous to count* - Confluent lysis occurred on whole agar surface.

TABLE 3. AVERAGE BREAKTHROUGH NUMBERS (PLAQUES) OF COLIPHAGE T_4 FROM 1 1/2-IN TEST SOIL COLUMNS OF BOTH LAHAINA AND WAHIAWA SOILS (1.5×10^6/ml APPLIED)

		COLIPHAGE T_4 PLAQUES PER ML			
		LAHAINA		WAHIAWA	
SAMPLING DAY	SAMPLE RUNS	EXP.	CONTROL	EXP.	CONTROL
FIRST DAY	#1	169	0	12	0
	2	237	0	14	0
	3	257	0	8	0
	4	243	0	17	0
	5	325	0	29	0
	6	311	0	44	0
SECOND DAY	#1	188	0	26	0
	2	575	0	47	0
	3	738	0	82	0
	4	1242	0	208	0
	5	2365	0	933	0
	6	TNTC⁂	0	1445	0
THIRD DAY	#1	828	0	725	0
	2	1396	0	1282	0
	3	1475-TNTC	0	957	0
	4	1860-TNTC	0	TNTC	0
	5	TNTC	0	TNTC	0
	6	TNTC	0	TNTC	0
FOURTH DAY	#1	TNTC	0	1785-TNTC	0
	2	TNTC	0	TNTC	0
	3	TNTC	0	TNTC	0
	4	TNTC	0	TNTC	0
	5	TNTC	0	TNTC	0
	6	TNTC	0	TNTC	0
FIFTH DAY	#1	TNTC	0	TNTC	0
	2	TNTC	0	TNTC	0
	3	TNTC	0	TNTC	0
	4	TNTC	0	TNTC	0
	5	TNTC	0	TNTC	0
	6	TNTC	0	TNTC	0

⁂ *Too numerous to count* - Confluent lysis occurred on whole agar surface.

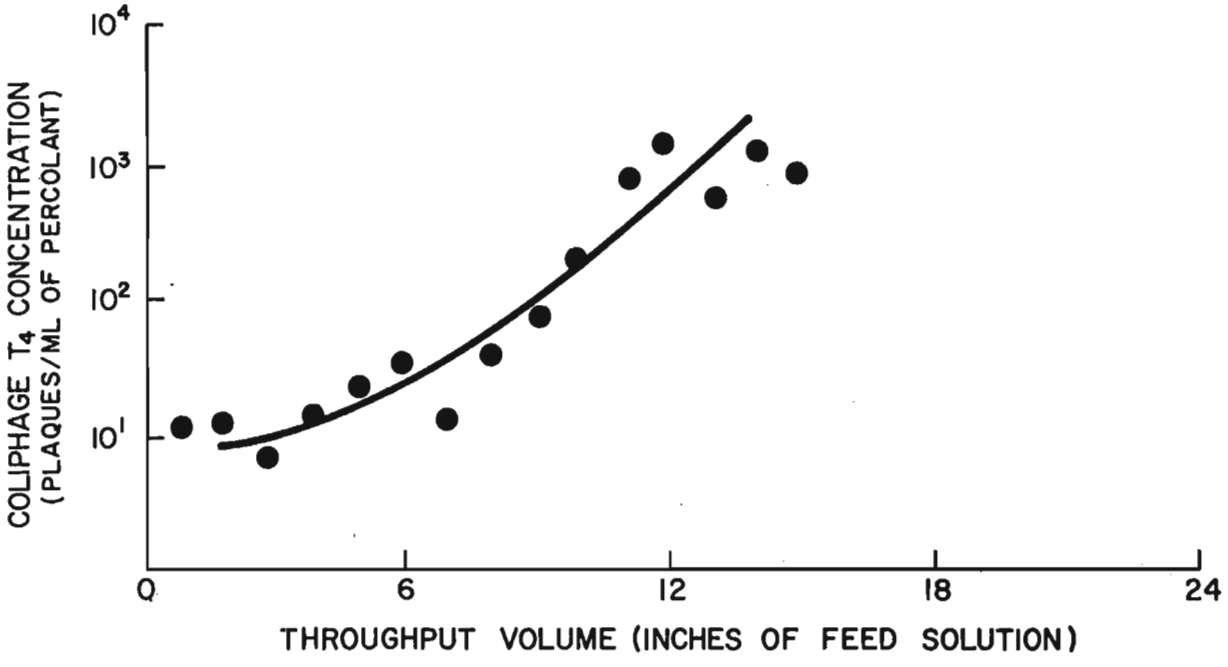


FIGURE 8: BREAKTHROUGH CURVE OF COLIPHAGE THROUGH 1 1/2 IN. TEST SOIL COLUMNS OF WAHIAWA SOIL.

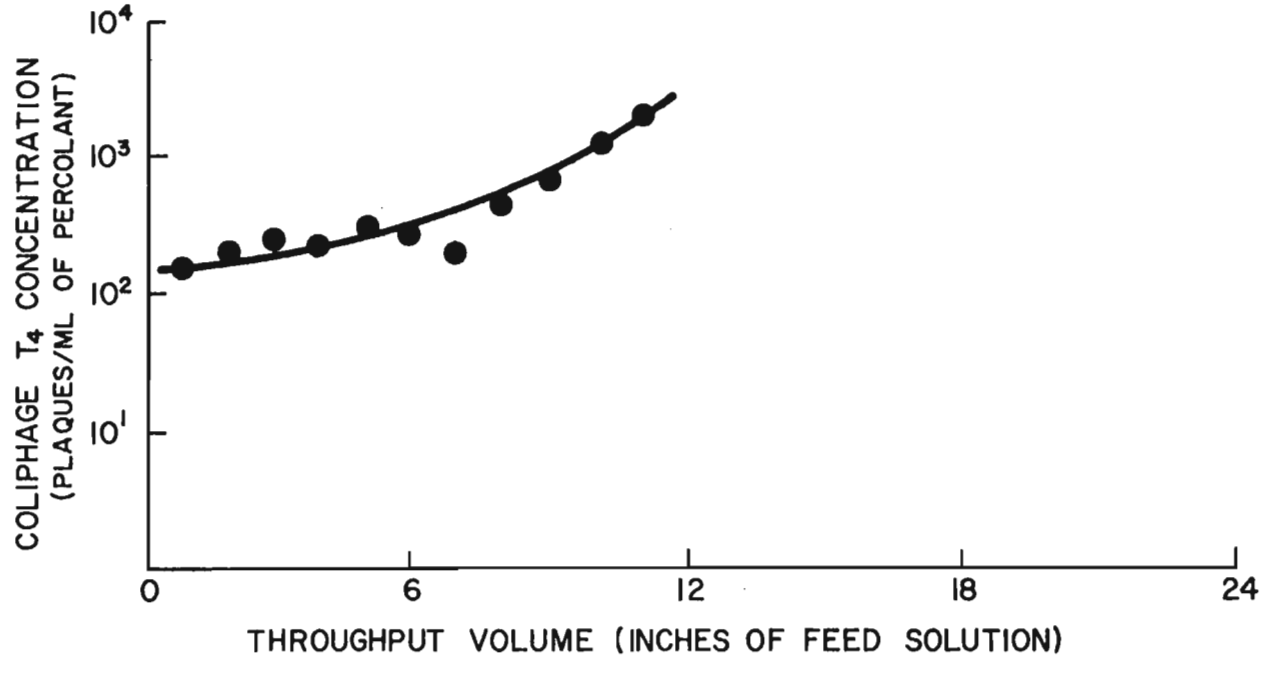


FIGURE 9: BREAKTHROUGH CURVE OF COLIPHAGE THROUGH 1 1/2 IN. TEST COLUMNS OF LAHAINA SOIL.

Percolation Test Results of Tantalus Cinder

Percolation tests of the cindery substratum of the Tantalus soil series dosed with 1.5 x 10^6 phage per ml of feed solution indicated an immediate breakthrough of the phage particles in all 15-in, 12-in, and 6-in soil columns in concentrations of 10^5 viruses per ml of percolant.

Breakthrough numbers of the T_4 viruses were from as low as 3.24 x 10^5 viruses (plaques) per ml of percolant assayed in the first sampling run to as high as 7.25 x 10^5 in the last sampling run, a percentage range of virus breakthrough of 21.6% to 48.3% (Table 4).

TABLE 4. AVERAGE BREAKTHROUGH NUMBERS (PLAQUES) OF COLIPHAGE T_4 THROUGH VARIOUS SOIL THICKNESSES OF TANTALUS CINDER (1.5 X 10^6/ml APPLIED)

	AVERAGE BREAKTHROUGH—NOS. PER ML			
SAMPLING RUNS	15" TEST COLUMNS	12" TEST COLUMNS	6" TEST COLUMNS	12" CONTROL COLUMNS
#1	3.96 X 10^5	3.86 X 10^5	3.24 X 10^5	0
2	5.44 X 10^5	4.72 X 10^5	4.98 X 10^5	0
3	4.62 X 10^5	5.42 X 10^5	5.49 X 10^5	0
4	5.54 X 10^5	5.10 X 10^5	5.16 X 10^5	0
5	4.66 X 10^5	5.81 X 10^5	5.75 X 10^5	0
6	5.29 X 10^5	7.25 X 10^5	6.76 X 10^5	0

General Discussion

This laboratory study has shown the relative effectiveness of Wahiawa and Lahaina soil series and Tantalus cinder in the removal of coliphage T_4Br^{II} from percolating waters. However, no attempt was made to investigate the mechanism of removal of the viruses at the different soil thicknesses by increasing or decreasing the pH or the concentration of the bacterial virus in the feed solution or applying saturated flow conditions to the soil columns.

Several properties possessed by soils may serve as possible removal mechanisms of bacteriophage T_4. These are the effects of surface straining (or clogging), cation exchange potentials, and physical adsorption. But in turn, these soil properties are influenced by such variables as the size of soil particles, the pH, temperature, and permeability of the soil; the concentration of the bacteriophage in the feed solution; and biological

activity (the latter if the feed solution and/or soil is not freed of microorganisms and other contaminants by autoclaving or some other method of sterilization).

Surface straining (or clogging) is operative only when size of the particles are greater than the void spaces and pore openings of the soil surface and the uppermost layers. Continued dosing and available head pressure might well clog the surface layer, causing particles to be forced farther and farther into the percolation system until the concentration of virus particles in the effluent is as great as the influent concentration. But because straining mechanisms usually are operative only if coarse particles are present in the feed solution, this possibility was ruled out.

Cation exchange potential is simply the ability of one cation to be displaced by another. While ion exchange has been an important process for removal of certain cations and anions from water and waste water, it probably will not be an operative mechanism in removal of viruses in a soil system. A study of the cation exchange properties of the Hawaiian Great Soil Groups by Kanehiro and Chang (11) in 1956 showed a very great range in cation exchange capacity, but it is low in the Low Humic Latosol group. The low value is associated with the high kaolinite content found in this group.

Physical adsorption is probably by far the most effective mechanism for virus removal. Such adsorption occurs as a result of nonspecific forces of attraction (generally referred to as van der Waal's forces, for example, forces that are electrostatic in nature as dipole-dipole forces) between the adsorbate (the coliphage T_4) and adsorbent (the soil particles), and the feed solution being the transporting medium or liquid phase. Physical adsorption differs from chemisorption in the specific degree of chemical reaction between the adsorbate and adsorbent; *i.e.*, bonds similar to covalent bonds may be formed. The small size of the enteric virus particles should favor their adsorption by the soil, since the smaller particles expose a greater surface area to be adsorbed to the soil particles.

Although the reports of the Honolulu Board of Water Supply record the continuing excellent quality of basal water, the presence or absence of pathogenic viruses are untested.

It is recommended that further investigation be done on the coliphage T_4 or related bacteriophage to more fully understand their means of migration and adsorption by Oahu soils.

SUMMARY AND CONCLUSIONS

A laboratory study to determine the ability of three Oahu soils to remove the coliphage T_4Br^{II} mutant from percolating water at different thicknesses showed that two of the soils proved to be effective in retaining the viruses but only at thicknesses greater than 2½ inches. This study did not investigate the removal mechanism or other mechanisms that play an important role in preventing breakthrough of the viruses, thus leaving a rich field for further research.

The major findings of this research are as follows:

1. For assaying breakthrough numbers of the bacterial viruses, the agar-layer plaque-counting technique employed was very efficient and offered the following advantages: (a) phage samples up to 1 ml in volume were transferred from the collected percolant and plated in petri dishes in less than 30 seconds, (b) the phage T_4 were generally 1 to 2 mm in size and clear, thus enabling easy counting, and (c) high efficiency of plating, *i.e.*, the ability of the phage particles to readily become adsorbed to a suitable host cell and the ability of infected cells to reliberate the phage to initiate a chain reaction resulting in a plaque. Although the plaque count is an excellent relative assay method, it will nevertheless not give the absolute number of phage particles present in the percolant sample; this will occur only under ideal conditions according to Adams (18).

2. The Wahiawa and Lahaina soils effected a 100% adsorption of the coliphage T_4 from percolating waters at the applied concentration of 2.5 x 10^6 per ml of feed solution through soil thicknesses of 6 inches and 2½ inches.

3. Breakthrough of the viruses occurred in both Wahiawa and Lahaina soils at a soil thickness of 1½ inches at applied concentration of 1.5 x 10^6 per ml of feed solution. The rate of breakthrough which varied for the two soils occurred slowly at first and increased rapidly with time. A higher flow rate was observed for Lahaina soil during the first two sampling days.

4. Tantalus cinder proved very ineffective in withholding viruses at the applied concentration of 1.5 x 10^6 per ml of feed solution. Breakthrough of concentrations of 10^5 of phage (plaques) were recorded through soil thicknesses of 15, 12, and 6 inches.

ACKNOWLEDGEMENTS

Advice and assistance from Dr. C. E. Folsome, Professor of Microbiology, in the preparation of high titre phage T_4 stocks and the supply of the bacteriophage and effective *Escherichia coli* strains obtained from the Department of Microbiology for conducting this research is gratefully acknowledged.

Thanks are due to Mr. Sakuichi Nakamura of the Soil Conservation Service for the up-to-date soil descriptions and information of the soil sampling sites and to Robert Parker for his help in taking the photographs and photomicrographs included in this report.

This report is based on a master's thesis by the senior author.

BIBLIOGRAPHY

1. Bankroft, P., Englehard, N. and C. Evans. Poliomyelitis in Huskerville, Nebraska. *J. Amer. Med. Assoc.* (164:836), 1957.

2. Hayward, M. L. Epidemiological Study of Outbreak of Infectious Hepatitis. *Gastroenterology* (6:504), 1946.

3. Little, G. M. Poliomyelitis and Water Supply. *Can. J. Public Health* (45:100), 1954.

4. Board of Water Supply. *Supplement to Annual Report for 1962 Fiscal Year.* Board of Water Supply, City and County of Honolulu, 1962, 113 p.

5. *The Water Sources of Oahu.* The Honolulu Board of Water Supply, August 15, 1966.

6. *Board of Water Supply Annual Report, 1964.* Board of Water Supply, City and County of Honolulu, 1964.

7. Yuen, G. A. L. *Safeguarding Honolulu's Water Supply.* Address to the Reconvened Session of the WPCF, October 1963.

8. *Statistical Abstract of Hawaii, 1962.* State of Hawaii, Department of Planning and Research, May 1962.

9. Nelson, L. A. *Detailed Land Classification--Island of Oahu.* Land Study Bureau, University of Hawaii, L. S. B. Bulletin No. 3, January 1963,

10. Cline, M. G. *Soil Survey of the Territory of Hawaii.* Soil Survey Series 1939, No. 25, U. S. Dept. of Agric., Soil Conservation Service, 1955.

11. Kanehiro, Y. and A. Chang. *Cation Exchange Properties of the Hawaiian Great Soil Groups.* Hawaii Agric. Experiment Station, Univ. of Hawaii, Tech. Bull. No. 31, June 1956.

12. Sherman, G. D. and L. T. Alexander. Characteristics and Genesis of Low Humic Latosols. *Soil Science of Amer., Proc.* (23:2), March-April 1959, pp. 168-170.

13. *The General Plan of the State of Hawaii.* State of Hawaii, State Planning Office, January 1961.

14. Anonymous. *Water Resources Activities in the United States: Water Resources of Hawaii.* Selected Committee on National Water Resources. U.S. Senate, 86th Congress, January 1960.

15. Survey of Ground Water Contamination of Waste Disposal Practices. *Amer. Water Works Assoc. J.* Task Group 2450 K, LII, 1211-1219 pp., September 1960.

16. d'Herelle, F. *The Bacteriophage.* Baltimore: Williams and Wilkins, 1922.

17. Hershey, A. D., Kalmanson, G. and J. Bronfenbrenner. Quantitative Methods in the Study of the Phage-Antiphage Reaction: Unity and Homogeneity of the Reactants. *J. Immunol.* (46:281-299), 1943.

18. Adams, M. H. *Bacteriophages.* New York: Interscience Publishers, Inc., 1959.

19. Demerec, M. and U. Fano. Bacteriophage-resistant Mutants in *Escherichia coli. Genetics* (30:119), 1945.

20. d'Herelle, F. *The Bacteriophage and Its Behavior.* Baltimore: Williams and Wilkins, 1926.

21. Williams, R. C. and D. Fraser. Morphology of the Seven T-Bacteriophages. *J. Bacteriol.* (66:458), 1953.

22. Polson, A. Diffusion Constants of the *E. coli* Bacteriophages. *Proc. Soc. Exptl. Med.* (67:294), 1948.

23. Watson, J. D. The Properties of X-ray inactivated Bacteriophage: I. Inactivation by Direct Effect. *J. Bacteriol.* (60:697), 1950.

24. Adams, M. H. The Stability of Bacterial Viruses in Solutions of Salts. *J. Gen. Physiol.* (32:579), 1949.

25. Graham, D. M. and F. E. Nelson. Inhibition of Lactic Streptococcus, Bacteriophage by Crystal Violet and Other Agents. *J. Gen. Physiol.* (37:121), 1954.

26. Wolff, L. K. and J. W. Janzen. Action de Divers Antiseptiques sur le Bacteriophage de d'Herelle. *Compt. Rend. Soc. Biol.* (87:1087), 1922. (As quoted by M. H. Adams in *Bacteriophages,* 286 p.)

27. Fitzgerald, R. J. and D. Babbit. Studies on Bacterial Viruses: I. The Effect of Certain Compounds on the Lysis of *Escherichia coli* by Bacteriophage. *J. Immunol.* (52:127), 1946.

28. Czekalowski, J. W. Studies on the Reproduction of Bacterial Viruses: II. Effect of Enzyme-inhibitors on the Multiplication of the Coliphage T_2. *Brit. J. Exptl. Pathol.* (33:57), 1952. (As quoted in M. H. Adams' *Bacteriophages,* 286 p.)

29. Hershey, A. D. and R. Rotman. Linkage among Genes Controlling Inhibition of Lysis in a Bacterial Virus. *Proc. Nat'l. Acad. Sci. U. S.* (34:89), 1948.

30. Doermann, A. H. and M. B. Hill. Genetic Structure of Bacteriophage T_4 as Described by Recombination Studies on Factors Influencing Plaque Morphology. *Genetics* (38:79), 1953.

31. Benzer, S. The Elementary Units of Heredity. In W. D. McElroy and B Glass, eds., *The Chemical Basis of Heredity.* Baltimore: John Hopkins Press, 1957, 70 p.

32. Hershey, A. D. Spontaneous Mutations in Bacterial Viruses. *Cold Spring*

Harbor Symposia Quant. Biol. (11:67), 1946.

33. Cohen, S. S. Molecular Basis of Parasitism of Some Bacterial Viruses. *Science* (123:653), 1956.

34. Sertic, V. Origine de la Lysine D'une Race du Bacteriophage. *Compt. Rend. Soc. Biol.* (100:614), 1929. (As quoted by Adams in *Bacteriophages*, 304 p.)

35. Clark, N. A. and S. L. Chang. Enteric Viruses in Water. *J. A. W. W. A.* (51:1299), 1959.

36. Carlson, H. J., *et al.* Efficiency of Standard Purification Methods in Removing Poliomyelitis Virus from Water. *Amer. J. Public Health* (32: 1256), 1942.

37. Dieterich, B. H. *A Study of the Adsorption Phenomenon in the Removal of Bacterial Viruses by Sand Filtration.* Unpub. Masters Thesis, Harvard, 1953.

38. Gilcreas, F. W. and S. M. Kelly. Relation of Coliform-Organism Test to Enteric-Virus Pollution. *J. A. W. W. A.* (47:683), 1955.

39. Robeck, G. G., *et al.* Effectiveness of Water Treatment Plant Processes in Virus Removal. *J. A. W. W. A.* (54:1275), 1962.

40. McMichael, F. C. and J. E. McKee. *Report of Research on Waste Water Reclamation at Whittier Narrows.* California Water Quality Control Board. Final Report. W. M. Kech Laboratory of Environmental Health Engineering, California Institute of Technology, September 1965.

41. Hallgren, R. Epidemic Hepatitis in the County of Vasterbotten in Northern Sweden--An Epidemiological and Clinical Study. *Act. Med. Scand. Suppl.* (140), 1942.

42. Neefe, J. R. and J. Stokes. An Epidemic of Infectious Hepatitis Apparently due to a Water-Borne Agent. *J. A. M. A.* (128:1063), 1945.

43. Farquhar, J. D., Stokes, J. and W. D. Schrack. Epidemic of Viral Hepatitis Apparently Spread by Drinking Water and by Contact. *J. A. M. A.* (149), 1952.

44. Tucker, C. B., Owen, W. H. and R. P. Farrell. An Outbreak of Infectious Hepatitis Apparently Transmitted Through Water. *Southern Med. J.* (47:732), 1954.

45. Peczenik, A., Duttweler, D. W. and R. H. Moser. An Apparently Water-Borne Outbreak of Infectious Hepatitis. *Amer. J. Public Health* (46:1009), 1956.

46. Mosley, J. W. and W. W. Smither. Infectious Hepatitis Report of an Outbreak Probably Caused by Drinking Water. *N. E. J. Med.* (257:590), 1957.

47. Stiles, C. W., Crohurst, H. R. and G. E. Thompson. Experimental Bacterial and Chemical Pollution of Wells, Via Ground Water, and the Factors Involved. *U. S. P. H. S. Hygenic Laboratory Bulletin No. 147*, 1927.

48. Fournelle, H. J., Day, E. K. and W. B. Page. Experimental Ground Water Pollution at Anchorage, Alaska. *Publ. Health Rpts. No. 72*, 1957, 248 p.

49. Burnet, F. M. and W. M. Stanley. *The Viruses*. New York: Academic Press, Vol. I, 1959.

50. Kempf, J. E., Wilson, M. G., Pierce, M. E. and M. H. Soule. Effect of Aluminum Hydroxide Sedimentation, Sand Filtration and Chlorination on the Virus of Poliomyelitis. *Amer. J. Publ. Health* (32:1366), 1942.

51. Chang, S. L., Stevenson, R. E., Bryant, A. R., Woodward, R. L. and P. W. Kabler. Removal of Coxsackie and Bacterial Viruses in Water by Flocculation. II. Removal of Coxsackie and Bacterial Viruses and the Native Bacteria in Raw Ohio River Water by Flocculation with Aluminum Sulfate and Ferric Chloride. *Amer. J. Publ. Health* (48:159), 1958.

52. Rhodes, A. J., Clarke, E. M., Knowles, D. S., Goodfellow, A. M. and W. L. Donohue. Prolonged Survival of Human Poliomyelitis Virus in Experimentally Infected River Water. *Can. J. Publ. Health* (41:146), 1950.

53. Rushizky, G. W. and C. A. Knight. Ribonuclease-sensitive Infectious Units from Tomato Bushy Stunt Virus. *Virology* (8:448), 1959.

54. Baumann, E. R. Should Small Water Supplies be Superchlorinated? Part I. *Water and Sewage Works*. Reference Number, November 30, 1962, 161-163 pp.

55. Melnick, J. L., Emmons, J., Opton, E. M. and J. H. Coffey. Coxsackie Viruses from Sewage: Methodology, Including an Evaluation of the Grab Sample and Gauze Pad Collection Procedures. *Amer. J. Hyg.* (59: 185), 1954.

56. Kelly, S. M., Clark, M. E. and M. B. Coleman. Demonstration of Infectious Agents in Sewage. *Amer. J. Publ. Health* (45:1438), 1955.

57. Gilcreas, F. W. and S. M. Kelly. Significance of the Coliform Test in Relation to Intestinal Virus Pollution of Water. *J. N. E. Water Works Assoc.* (68:255), 1954.

58. Sinsheimer, R. L. Purification and Properties of Bacteriophage X-174. *J. Molecular Biol.* (1:37), 1959.

59. Folsome, C. E. Specificity of Induction of T_4Br^{II} Mutants by Ultraviolet Irradiation of Extracellular Phages. *Genetics* (47:611-622), May 1962.

60. Chase, M. and A. H. Doermann. High Negative Interference over Short

Segments of the Genetic Structure of Bacteriophage T_4. *Genetics* (43: 332-353), 1958.

61. Womack, D. W. *Tentative Description Legend of the Island of Oahu.* U. S. Soil Conservation Service, Hawaii, 1959, (unpublished report).

62. Young, R. H. F., Lau, L. S. and N. C. Burbank, Jr. *Travel of ABS and Ammonia Nitrogen With Percolating Water Through Saturated Oahu Soils.* Water Resources Research Center, University of Hawaii, Technical Report No. 1, January 1967.

63. Eto, M. A., Burbank, N. C., Jr., Klemmer, H. W. and L. S. Lau. *Behavior of Selected Pesticides With Percolating Water in Oahu Soils.* Water Resources Research Center, University of Hawaii, Technical Report No. 9, August 1967.

64. Todd, D. K. *Ground Water Hydrology.* New York: John Wiley & Sons, Inc., 1960, 53 p.

APPENDICES

APPENDIX A. WAHIAWA SERIES

The Wahiawa series is a member of a clayey, halloysitic, isothermic family of Tropeptic Eutrorthoxs. Typically, these soils have a very dusky red or dusky red granular A horizon over a compact B horizon with moderate or strong structure. Manganese is common throughout the solum, giving a purplish cast when moist.

Typifying Pedon: Wahiawa silty clay - pineapple
(Colors are for moist soil unless otherwise noted.)
(All textures are "apparent field textures.")

Ap1 -- 0-6" -- Very dusky red (2.5YR 2/2) silty clay, dusky red (2.5YR 3/2) dry; moderate medium, fine and very fine granular structure; very hard, friable, sticky and plastic; abundant roots; many medium, fine and very fine interstitial pores; many black concretions (1/8 - 1/4 inch in diameter); violent effervescence with hydrogen peroxide; medium acid (pH 5.6); abrupt smooth boundary. 2 to 6 inches thick.

Ap2 -- 6-12" -- Dusky red (2.5YR 3/2) moist and dry silty clay; common dark reddish brown (2.5YR 3/4) material forms the B horizon mixed by cultivation; moderate coarse subangular blocky structure; hard, firm, sticky and plastic; abundant roots; few fine and very fine tubular pores; compact in place; many black concretions; violent effervescence with hydrogen peroxide; medium acid (pH 5.8); abrupt wavy boundary. 5 to 8 inches thick.

B21 -- 12-16" -- Dark reddish brown (2.5YR 2/4) silty clay, dark reddish brown (2.5YR 3/4) dry; moderate fine and very fine subangular blocky structure; hard, firm, sticky and plastic; plentiful roots; common fine and very fine, few coarse tubular pores; many black concretions; strong effervescence with hydrogen peroxide; medium acid (pH 5.6); gradual wavy boundary. 4 to 8 inches thick.

B22 -- 16-33" -- Dark reddish brown (2.5YR 2/4) silty clay, dark reddish brown (2.5YR 3/4) dry; moderate and strong fine and very fine subangular blocky structure; hard, friable, sticky and plastic; few roots; common fine and very fine tubular pores; nearly continuous pressure cutans; many fine distinct black stains; few black concretions; strong effervescence with hydrogen peroxide; slightly acid (pH 6.5); diffuse wavy boundary. 14 to 20 inches thick.

B23 -- 33-45" -- Dark reddish brown (2.5YR 2/4) silty clay, dark reddish brown (2.5YR 3/4) dry; moderate and strong very fine subangular blocky structure; hard, friable, sticky and plastic; common fine and very fine tubular pores; nearly continuous pressure cutans; many fine distinct black stains; few black concretions; moderate effervescence with hydrogen peroxide; neutral (pH 7.1); diffuse wavy boundary. 10 to 14 inches thick.

B24 -- 45-60"+ -- Dark reddish brown (2.5YR 2/4) silty clay, dark reddish brown (2.5YR 3/4) dry; moderate and strong very fine subangular blocky structure; hard, friable, sticky and plastic; common fine and very fine tubular pores; few fine black stains; thin patchy clay films; continuous pressure cutans; many distinct slickensides up to 2 inches long; very few black concretions; slight effervescence with hydrogen peroxide; neutral (pH 6.9).

Type Location: Honolulu County, Hawaii. Junction of Kamehameha Highway and entrance road to Mililani Cemetery. East 0.7 mile then 400 feet north in Dole Corporation field No. 4101, block 30. Photo No. DACE 1-25.

Range in Characteristics: Black concretions occur on the surface and to depths of 4 to more than 5 feet. Depth to highly weathered basalt varies from 5 to more than 10 feet; a few boulder cores may occur in the lower solum. Mean annual soil temperature is about 71° F.

The A horizon has dry or moist values of 2 or 3 and chromas of 2 to 4 when dry or moist. The B horizon ranges in hue from 2.5YR to 10R with dry and moist values of 2 or 3, and chromas of 3 to 6 when dry and 3 to 5 when moist.

Competing Series and Their Differentiae: These include the Helemano, Kemoo, Kunia, Lahaina, Leilehua and Manana series. The Helemano soils lack clay films in the lower B horizon and do not effervesce with hydrogen peroxide. Kemoo soils have an argillic horizon with thin to moderately thick continuous clay films. The Kunia soils lack clay films and slickensides in the lower B horizon. The Lahaina soils have weak structure in the A and upper B horizons and have no clay films in the B horizon. The Leilehua soils have an argillic horizon, weak structure in the upper B and are extremely acid throughout the solum. The Manana soils have an argillic horizon and a thin panlike layer at depths of 15 to 50 inches.

Setting: Wahiawa soils are on long, smooth relatively undissected uplands at elevations of 500 to 1,200 feet. Slopes are normally 1 to 8 percent but range from 0 to 25 percent. The soils developed in residuum and alluvium from basalt. Annual rainfall is 40 to 60 inches. Mean annual air temperature is about 71° F.; average January air temperature is 69° F.; average July air temperature is 73° F.

Principal Associated Soils: These are the competing Kunia, Lahaina, Leilehua and Manana series.

Drainage and Permeability: Well drained with slow to medium runoff; moderately rapid permeability.

Use and Vegetation: Used primarily for irrigated sugarcane and dryland pineapple with small areas of pasture. Natural vegetation consists of guava (*Psidium guayava*), koa haole (*Leucaena glauca*), lantana (*Lantana camara*), joee (*Stachytarpheta cayanmensis*), Japanese tea (*Cassia leschenaultiana*), bermudagrass (*Cynodon dactylon*) and honohono (*Commelina diffusa*).

Distribution and Extent: On the medial uplands between the Waianae and Koolau Ranges on the island of Oahu. Approximately 22,250 acres in extent.

Remarks: The Wahiawa series was formerly classified in the Low Humic Latosol great soil group.

National Cooperative Soil Survey, 7-22-66

APPENDIX B. LAHAINA SERIES

The Lahaina series is a member of the clayey, halloysitic, isohyperthermic family of Tropetic Haplustoxs. Typically, these soils have dark reddish brown friable A horizons that have weak granular structure, and dusky red or dark reddish brown B2 horizons that have moderate subangular blocky structure and pressure faces on peds. Effervescence with hydrogen peroxide decreases with depth.

Typifying Pedon: Lahaina silty clay - irrigated sugarcane
(Colors are for moist soil unless otherwise noted.)

Ap1 -- 0-7" -- Dark reddish brown (2.5YR 3/4) silty clay, dark reddish brown (2.5YR 3/4) dry; weak fine, medium and coarse granular structure; hard, friable, very sticky, very plastic; many roots; many fine and very fine pores; many 1 to 3 mm. black concretions that effervesce with hydrogen peroxide; common small earthy lumps that break down on persistent rubbing; strong effervescence with hydrogen peroxide; medium acid (pH 6.0); abrupt wavy boundary. 6 to 9 inches thick.

Ap2 -- 7-15" -- Dark reddish brown (2.5YR 3/4) silty clay, dark reddish brown (2.5YR 3/4) dry; weak medium and coarse subangular blocky structure; hard, friable, very sticky, very plastic; compacted by machinery; many roots; few medium and fine pores; many 1 to 3 mm. black concretions that effervesce with hydrogen peroxide; violent effervescence with hydrogen peroxide in matrix; medium acid (pH 5.7); abrupt wavy boundary. 6 to 9 inches thick.

B1 -- 15-23" -- Dusky red (10R 3/3) silty clay, dark reddish brown (2.5YR 3/4) dry; weak medium and coarse subangular blocky structure; hard, friable, very sticky, very plastic; many roots; many fine and very fine pores; many 1 to 3 mm. black concretions that effervesce with hydrogen peroxide; violent effervescence with hydrogen peroxide in matrix; slightly acid (pH 6.1); gradual wavy boundary. 4 to 10 inches thick.

B21 -- 23-31" -- Dusky red (10R 3/3) silty clay, dusky red (10R 3/4) dry; moderate medium and coarse subangular blocky structure; hard, friable, sticky, plastic; common roots; many fine and very fine pores; nearly continuous pressure faces on peds; many 1 to 3 mm. black concretions that effervesce with hydrogen peroxide; violent effervescence with hydrogen peroxide in matrix; medium acid (pH 6.0); gradual wavy boundary. 6 to 15 inches thick.

B22 -- 31-46" -- Dark reddish brown (2.5YR 3/4) heavy silty clay loam, dark red (2.5YR 3/6) dry; moderate medium subangular blocky structure; breaking to moderate very fine subangular blocky structure when disturbed; hard, friable, sticky, plastic; very compact in place; few roots; many medium and fine pores; nearly continuous pressure faces on peds; few weathered basalt stones and boulders; many 1 to 3 mm. black concretions that effervesce with hydrogen peroxide; slight effervescence with hydrogen peroxide in matrix; slightly acid (pH 6.1); gradual wavy boundary. 10 to 18 inches thick.

B3 -- 46-60" -- Dark reddish brown (2.5YR 3/4) heavy silty clay loam, dark red (2.5YR 3/6) dry; strong medium and coarse subangular blocky structure; hard, friable, sticky, plastic; many fine pores; many small patchy pressure faces on peds; common 1 to 3 mm. black concretions that effervesce with hydrogen peroxide; many strongly weathered 1/4 to 2 mm. basalt particles; common weathered basalt stones; slight effervescence with hydrogen peroxide in matrix; medium acid (pH 6.0).

Type Location: Island of Maui, Maui County, Hawaii; Lahaina Quadrange - 20°55'28" north latitude and 156° 40'27" west longitude; on the Pioneer Mill Company plantation 1.5 mile east of Kaanapali and 0.75 mile southeast of Puukolii Camp, in field No. B-7 about 50 feet south of plantation road.

Range in Characteristics: Thickness of the solum ranges from 36 to more than 60 inches. Mean annual soil temperature is about 72° F. Textures shown are "apparent field textures." Hue of the A horizon ranges from 5YR to 10R, and chroma is 3 or 4 moist and 3 through 6 dry. Hue of the B horizon ranges from 2.5YR to 10R, and chroma is 3 or 4 moist and 3 through 6 dry. Structure is moderate to strong in the lower part of the B horizon.

Competing Series and their Differentiae: These are the Helemano, Hoolehua, Kunia, Molokai, Pamoa, Wahiawa, Waikapu, and Wailuku series. The Helemano soils have moderate structure in the A horizon and do not effervesce with hydrogen peroxide. Hoolehua soils have 5YR or yellower hue in the B horizon. The Kunia soils have mean annual soil temperature of less than 71.6°F., and strong continuous pressure faces on peds in the lower part of the B horizon. The Molokai soils have weak prismatic structure in the upper part of the B horizon and fine-silty controls sections. The Pamoa soils have granular surface mulch and wide cracks when dry and slickensides. Wahiawa soils have moderate structure in the A and B horizons, strong, concentrations of manganese dioxide throughout the sola, and patchy clay films in the lower part of the B horizon. Waikapu soils have silty clay loam texture throughout the B horizon. The Wailuku soils have 5YR hue throughout the sola.

Setting: The Lahaina soils are on nearly level to moderately steep intermediate uplands. Slopes of 2 to 15 percent are the most common. Elevation ranges from 10 to 1,500 feet. The soils formed in residuum and alluvium from basic igneous rock. Average annual rainfall is 20 to 35 inches. Mean annual temperature is about 72° F., average January temperature is 69° F., and average July air temperature is 74° F.

Principal Associated Soils: These are the competing Helemano, Hoolehua, Molokai, Pamoa and Wahiawa series.

Drainage and Permeability: Well drained. Runoff is medium, and permeability is moderate.

Use and Vegetation: This soil is used mainly for growing pineapple and irrigated sugarcane. Natural vegetation is cactus (*Opuntia megacantha*), fingergrass (*Chloris spp.*), kiawe (*Prosopis chilensis*), koa haole (*Leucaena glauca*), and lantana (*Lantana camara*).

Distribution and Extent: Islands of Lanai, Maui, Molokai, and Oahu, Hawaii. The extent is about 21,00 acres.

Remarks: The Lahaina soils were classified as Low Humic Latosols in the 1955 Soil Survey of Hawaii.

APPENDIX C. TANTALUS SOIL SERIES

The Tantalus series is a member of an ashy over cindery, isothermic family of Typic Dystrandepts. Typically, these soils have a thick very dark brown A horizon with moderate fine and very fine subangular blocky structure. The B horizon is a very friable, dark reddish brown, very fine sandy loam that rest on unweathered block fine gravel-size cinders at depths of 15 to 36 inches.

Typifying Pedon: Tantalus silty loam, 40 to 70 percent slopes. (308) This soil comprises 919 acres.

A1 -- 0-18" -- Very dark brown (10YR 2/2) silty loam, dark brown (10YR 3/3) dry; moderate very fine and fine subangular blocky structure; hard, friable, slightly sticky and slightly plastic; abundant very fine and fine roots; common very fine interstitial pores; common fine and very fine sharp cinders; neutral (pH 7.2); gradual smooth boundary. 10 to 18 inches thick.

B2 -- 18-29" -- Dark reddish brown (5YR 3/4) very fine sandy loam, reddish brown (5YR 4/4) dry; massive; soft, very friable, slightly sticky, slightly plastic and weakly smeary; abundant very fine and fine, few medium and coarse roots; many very fine and fine tubular pores; abundant very fine sharp cinders; neutral (pH 6.6); clear wavy boundary. 6 to 12 inches thick.

IIC -- 29"+ -- Black unweathered fine gravel-size cinders.

Location: On Tantalus Mountain northeast of Punchbowl along Tantalus Drive, 3/4 mile northeast of Hawaii Board of Water Supply water tank. Site located 50 feet west of road. Photo No. DACE 3-46.

Range in Characteristics: The solum ranges in thickness from 15 to 36 inches. Texture of the A horizon ranges from silt loam to silty clay loam. Hue in the A horizon ranges from 5YR to 10YR with chromas and values of 2 or 3. Texture of the B horizon may be a very fine sandy loam, silt loam or silty clay loam. Hue of the B horizon ranges from 5YR to 10YR with chromas of 2 to 4.

Permeability is 2.0 to 6.3 inches per hours; internal drainage is rapid; runoff is rapid and erosion hazard is severe; available water holding capacity is 1.8 inches per foot; workability is impractical because of slope; fertility is high; the shrink-swell potential is low. This soil is used for watershed purposes. It is suited to pasture, woodland, wildlife, and recreation.

Included with this soil are small cinder deposits and stony soils within the drainageways. Pasture group 1. Woodland group 3.

Tantalus silt loam, 15 to 40 percent slopes: This soil comprises 264 acres. It is suited to the same crops and is similar to Tantalus silt loam, 40 to 70 percent slopes except runoff is medium to rapid and erosion hazard is moderate to severe; workability is difficult because of slope. This soil is used for watershed purposes. Pasture group 1. Woodland group 1.

Tantalus silty clay loam, 8 to 15 percent slopes: This soil comprises 576 acres. It is similar to Tantalus silt loam, 40 to 70 percent slopes except the texture is a silty clay loam, internal drainage is medium; runoff is slow to medium and erosion hazard is slight to moderate; slopes range from 8 to 15 percent. Workability is difficult because of slope.

This soil is used for urban developments, watersheds and recreation. It is suited to wildlife, woodland, recreation, and pasture.

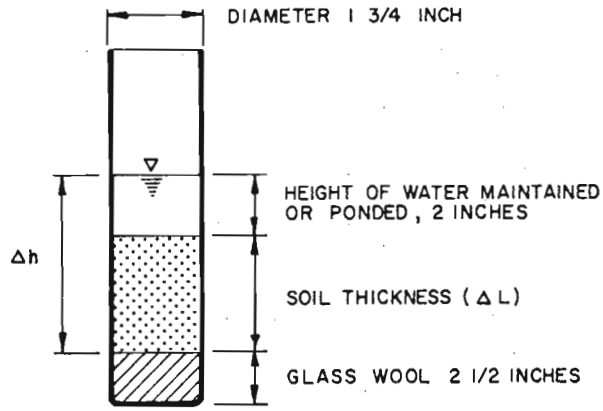
Included with this soil are small areas of stony soils within the drainage ways. Pasture group 1. Woodland group 1.

Tantalus silty clay loam, 15 to 40 percent slopes: This soil comprises 389 acres. It is similar to Tantalus silt loam, 40 to 70 percent slopes except the texture is a silty clay loam, internal drainage is medium; runoff is medium to rapid and erosion hazard is moderate to severe; workability is very difficult because of slope.

This soil is used for urban developments. It is suited to wildlife, woodland, recreation, and pasture. Pasture group 1. Woodland group 1.

Soil Conservation Service
U.S. Dept. of Agriculture

APPENDIX D. COMPUTATIONS ON PERMEABILITY OF LAHAINA AND WAHIAWA SOILS



PERMEABILITY (K)
K as expressed as gal/day/ft^2

$$K = \frac{Q}{A\ (\Delta h/\Delta L)}\ ,$$

Where Q = flow rate in ml/sec

$\Delta h/\Delta L$ = hydraulic gradient

A = area of circle = (πr^2)
= (3.14) (0.785)
= 2.40 in^2 or 15.48 cm^2

Conversions:

1 hr = 86,400 sec

1 gal = 3,785 ml

1 ft^2 = 929 cm^2

SAMPLE CALCULATIONS

PERMEABILITY OF 6-IN SOIL COLUMNS:

Lahaina: $\frac{(76.7/600)\ (929)\ (86400)}{(15.48)\ (20.3/15.2)\ (3785)} = 131\ \frac{gal}{ft^2\ day}$

Wahiawa: $\frac{(116.5/600)\ (929)\ (86400)}{(15.48)\ (20.3/15.2)\ (3785)} = 199\ \frac{gal}{ft^2\ day}$

APPENDIX E. OBSERVED PERMEABILITY OF WAHIAWA AND LAHAINA SOILS AT DIFFERENT THICKNESSES

SOIL SERIES	SOIL THICKNESS IN COLUMN (IN INCHES)	VOLUME COLLECTED ml/600 SEC	K (GAL/DAY/FT^2)	K (cm/HR)
WAHIAWA	6	116.5	199	33.8
	2 1/2	91.6	116	19.7
	1 1/2	99.2	97	16.5
	AVERAGE	102.4	137	23.3
LAHAINA	6	76.7	131	22.3
	2 1/2	89.8	113	19.2
	1 1/2	187.1	182	31.0
	AVERAGE	117.9	142	24.2