THE EFFECT OF CERTAIN BACTERIAL INHIBITORS ON SHELF-LIFE OF FRESH POULTRY MEAT

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Robert James McVicker

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THE EFFECT OF CERTAIN BACTERIAL INHIBITORS ON SHELF-LIFE OF FRESH POULTRY MEAT

By

Robert James McVicker

AN ABSTRACT

Submitted to the College of Agriculture
Michigan State University of Agriculture and
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Approved L. E. Dawson

During recent years the poultry industry of the United States has grown from an industry of relative minor importance to a major source of farm income. This growth has been characterized by many notable achievements which have resulted in making higher quality poultry meat more readily available to the consumer at lower prices.

One of the most recent achievements which has had an effect on the poultry industry has been the use of antibiotics and other bacterial inhibitors for extending shelf-life of fresh poultry meat. Two antibiotics which have been approved by the Food and Drug Administration for use in and on uncooked foods in specified amounts are chlortetracycline and oxytetracycline.

A total of four studies were carried out, (1) to determine the effectiveness of chlortetracycline, oxytetracycline, and chlorine in extending the shelf-life of fresh poultry meat treated under laboratory processing conditions, (2) to determine the effectiveness of chlortetracycline in extending shelf-life of fresh poultry meat treated in a commercial processing plant, (3) to measure the difference, if any, in shelf-life between packaged and unpackaged chicken meat, and (4) to determine if people can distinguish flavor differences in cooked chicken meat with high and low numbers of bacteria.

Two of the four studies were replications involving birds slaughtered in the Poultry Department processing laboratory and treated with chlortetracycline, oxytetracycline, and chlorine. Both raw odor and flavor determinations were made. Bacteria counts were determined in the Department of Microbiology and Public Health. Members of the Foods and Nutrition Department cooked the birds and administered the taste panels.

The last two studies involved purchasing birds from a commercial processing plant. In the first commercial study four treatments were evaluated. They were (1) split, unpackaged control (2) cut-up, packaged control (3) split, unpackaged, chlortetracycline-treated, and (4) cut-up, packaged chlortetracycline-treated.

The second commercial study involved only controls and chlortetracycline-treated birds. The controls were divided into two groups, one frozen and the other unfrozen. The same division was made for the chlortetracycline treatments.

Raw odor determinations were made for the first of these two studies and taste panels were conducted for both commercial studies. Bacterial counts were determined in the Department of Microbiology and Public Health.

The results of these studies showed that under laboratory processing conditions chlortetracycline was more effective in extending shelf-life of fresh poultry meat than oxytetracycline or chlorine. Fresh poultry meat, treated commercially with chlortetracycline, deteriorated more rapidly than poultry receiving the same treatment and processed under laboratory processing conditions.

Chlortetracycline-treated commercial birds were considered unacceptable prior to cooking from 7 to 8 days sooner than similarly treated laboratory birds. According to raw odor determinations, fresh poultry meat was unacceptable when bacterial contamination reached a population of, or near 1×10^6 organisms per square inch of skin surface.

Bacteria counts on chlortetracycline-treated commercial birds reached a bacteria population of 1×10^6 from 7 to 8 days sooner than similarly treated laboratory birds.

No apparent relationship exists between bacterial contamination and cooked flavor of breast and thigh chicken meat (without skin), as determined by taste panels for the length of time involved.

Results of a poultry processing plant sanitation survey showed that bacterial inhibitors are no substitute for good plant sanitation. Fresh chicken meat treated commercially with chlortetracycline in a single processing plant did not remain acceptable longer than untreated birds from the same plant.

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INTRODUCTION

During recent years the poultry industry of the United States has grown from an industry of relative minor importance to a major source of farm income. This growth has been characterized by many notable achievements which have placed the poultry industry among the leading agricultural industries. These achievements have resulted in (1) a laying hen which can produce 250-300 eggs per year at a lower body weight and on less feed per dozen eggs, (2) a broiler which can be grown to a body weight of 4.5 pounds in ten weeks on 2.5 pounds of feed per pound of body weight, (3) fewer, but larger and more efficient hatcheries and feed mills, (4) poultry rations designed for better efficiency by increasing the protein and energy levels and lowering the fiber content, (5) efficient and modern poultry processing plants which prepare poultry ready-to-cook rather than New York dressed, and (6) chicken on the family menu during every day of the week throughout the entire year.

Through improved breeding, feeding, management, marketing, merchandising, and disease control, higher quality poultry is more readily available to the consumer at lower prices. This, in part, accounts for the reason why per capit

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capita consumption of ready-to-cook chicken meat has increased from 13.2 pounds during the period of 1935-39 to 22.7 pounds per person in 1955.[1]

Today most of the poultry meat is prepared in processing plants in ready-to-cook style. Also, most of the chicken meat distributed in retail stores is sold as fresh rather than frozen, live, or New York dressed. It is therefore necessary for fresh chicken meat to move rapidly through the various marketing channels and be held at favorable low temperatures during this movement if spoilage is to be deterred.

Spoilage is greater in ready-to-cook birds than in dressed poultry according to Baker et al (1956).

Since ready-to-cook poultry meat is preferred over dressed poultry by the consumer, the trend has been for an increasing amount of poultry meat to be marketed in this form.

In the process of preparing poultry in ready-to-cook form, it is necessary for the poultry to be handled several times, thus increasing the possibility of higher bacterial contamination. Mundt, Stokes, and Goff (1954) reported that whenever the skin of dressed poultry is cut or torn, there is a marked increase in bacterial contamination. Therefore, bacterial contamination of ready-to-cook poultry

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[1] Statistical Abstract of the United States (1956).

could present a serious problem toward maintaining high acceptability by the consumer for fresh poultry meat.

One of the most recent achievements which has had an effect on the poultry industry has been the use of antibiotics and other bacterial inhibitors for extending shelf-life of fresh poultry meat.

On November 30, 1955 the Food and Drug Administration[1] approved the antibiotic Aureomycin chlortetracycline for use on certain uncooked foods. In the case of poultry, the Food and Drug Administration states, "A tolerance of seven parts per million is established for residues of chlortetracycline in or on uncooked poultry. This tolerance level shall not be exceeded in any part of the poultry."

Since the approval of Aureomycin chlortetracycline (referred to commercially as Acronize) another antibiotic has been approved by the Food and Drug Administration. The name of this antibiotic is Terramycin oxytetracycline, more commonly known by the commercial name of Biostat.

Both of these products are in powder form and can be dissolved in the chill water, which presently is the most common method used in processing plants for cooling poultry.

Some of the possible benefits which could be derived from treating fresh poultry meat with antibiotics are: (1)

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[1] Federal Register, 20 F. R. 8776 (Nov. 30) 1955.

it may improve the economy of the over-all poultry marketing process by allowing fresh poultry meat to be cut-up and packaged in the poultry processing plant, (2) it may be possible to scald birds at 140° F. and still hold bacteria counts relatively low. More of the skin is removed at 140° F., thereby setting up greater possible bacterial contamination, (3) extending the distribution range of fresh poultry meat from a given poultry processing plant, (4) reducing frequency of deliveries to retail outlets, (5) may provide greater protection against loss for the retailer, especially, in carrying poultry over a weekend period, and (6) may provide greater assurance to the housewife that she is getting a cleaner, more wholesome product.

Due to the many problems involved in retaining freshness in poultry meat, it was felt that further studies would be desirable in order to explore new possibilities and substantiate or reject findings by other workers in both commercial and educational institutions.

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LITERATURE REVIEW

Studies have been carried on in various parts of the United States and Canada to determine the effectiveness of different bacterial inhibitors on extending the shelf-life of flesh foods. Miller (1955) reported that Dr. Hugh Tarr and associates of the Canadian Pacific Fisheries Experimental Station in Canada began studies involving the use of penicillin to extend the shelf-life of flesh foods as early as 1944. Since this early work with penicillin several other wide spectrum antibiotics have been introduced for use in preserving flesh foods.

Use of Bacterial Inhibitors on Poultry Meat, Fish and Beef

Early studies by Tarr, Southcott and Bissett (1952) involving preservation of fish and meat with streptomycin, penicillin, subtilin, polymixin B, circulin, neomycin, bacitracin, gramicidin, methyl gramicidin, tyrothricin, rimocidin, terramycin, chloromycin, aureomycin, and one unnamed antibiotic showed favorable results for the use of antibiotics in the field of food preservation. Aureomycin, terramycin, and chloromycetin, in order named, proved the most effective inhibitors of growth of the mu

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natural mixed bacterial flora at temperatures between 0°C. and 21°C., while rimocidin inhibited yeast growth. Aureomycin caused marked inhibition of spoilage in 0.5 to 2.0 micrograms per gram concentration when incorporated in minced flesh. Immersing steaks in solutions containing the antibiotic in 5 or 10 micrograms per gram concentration either exerted a less intense bacteriostatic action or were without effect.

Further studies by Goldberg, Beatherage and Weiser (1953) to determine which of the wide spectrum antibiotics would be more useful in the field of food preservation, were carried out by adding 0, 0.5, 1.0, 1.5, and 2.0 parts per million of penicillin, bacitracin, streptomycin, chloramphenicol, aureomycin, and terramycin to ground beef. The samples were stored at 10°C. Only the last three delayed spoilage. This work confirmed completely the work of Tarr, et al. (1952).

In order to decide which of the antibiotics to use, Lepovetsky et al. (1953) screened aureomycin, terramycin, and chloramphenicol against 93 strains of organisms from 492 isolates from meat. These organisms represented 12 genera. Aureomycin inhibited growth in 81 strains, terramycin in 77, and chloramphenicol in 74. Nine strains were unaffected by all three antibiotics.

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Ziegler and Stadelman (1955) compared untreated, packaged chicken meat with packaged, aureomycin treatments at various concentration levels. Concentrations of 10, 20, and 40 parts per million were used. Daily examination of the treated birds showed the microflora was composed mostly of yeasts having the shape and budding characteristics of the family Saccharomycetaceae. On the controls the predominant organisms were gram negative organisms. Placing eviscerated chicken fryers in the various concentrations significantly increased their shelf life. These results are similar to those obtained by Kohler, Broquist, and Miller (1954), Tarr, Southcott and Bissett (1952) and Goldberg, Weiser, and Deatherage (1953) who were studying the effect of antibiotics in the preservation of poultry, fish, and beef, respectively. Spencer, Ziegler, and Stadelman (1954) reported that aureomycin was one of the most effective bacteriostatic agents for poultry meats. Their work showed that aureomycin extended shelf-life of fresh poultry meat as follows: controls, 11.5 days; 10 parts per million, 17.7 days; 20 parts per million, 18.3 days and for those treated with 40 parts per million, 19.0 days. They also reported the difference between chlorine dipped broiler halves and their control halves was very small, in fact, less than one day for either a chlorine concentration of 10 or 20 parts per million.

Kohler, Miller and Broquist (1955) found that subtilin, bacitracin, gramicidin and streptomycin were only partially successful in deterring spoilage of canned foods. Their results showed aureomycin was more effective in controlling poultry spoilage than were nine other antibiotics tested.

Increased shelf-life of chlortetracycline treated poultry meat was significantly higher than controls at storage temperatures of 32, 37, 42, and 60° F. according to Shannon and Stadelman (1957). Spencer, Ziegler, and Stadelman (1954) stated that birds stored at 31° F. had a shelf-life of 16 days compared to 10 days for those stored at 36° F. Ice storage had the same effect as a 31° F. refrigeration temperature.

According to Lockhead and Landerkin (1935) spoilage of chicken held at refrigerator temperatures above freezing is due primarily to the growth of slime-forming bacteria on the exposed surfaces. These observations have been confirmed by Gunderson, Rose and Henn (1947) and Ayres, Ogilvy and Stewart (1950). Spoilage of fresh packaged poultry meat stored at various temperatures (31, 34, and 36° F.) was detected from one to three days earlier by drawing smear samples from the pectoral feather tract immediately beneath the wing by means of a wire loop and spreading these uniformly on a glass slide as compared to a sensory panel as reported by Ziegler, Spencer and

Stadelman (1954).

Goldberg, Weatherage and Weiser (1954) explored the possibilities of treating ground meat with antibiotics and infusing antibiotics into whole animals prior to dressing. Weiser, Kunkle, and Weatherage (1954) found in their tests that by using aureomycin in combination with a physiological saline solution the cut appearance was normal, but without the saline solution the cut appearance was moist. The moistness was attributed to coagulated blood which had been flushed under pressure by the infused antibiotic solution. Cahill, Kunkle, Goldberg, Weiser, and Weatherage (1952) found that rounds treated with aureomycin and held at a temperature of 70-80° F. for 24-72 hours did not sour, but control rounds did. In further studies involving beef, Goldberg, Weiser and Weatherage (1953) found that chloromycetin, terramycin and aureomycin successfully lengthened keeping quality of the beef and also showed a high activity against pure cultures of microflora isolated from deep tissues of beef. Assay for aureomycin residue in stored beef showed no detectable antibiotic after 72 hours. Weiser, Goldberg, Cahill, Kunkle, and Weatherage (1953) have suggested that the infusion of aureomycin may prevent deep spoilage in carcasses where refrigeration is delayed. There are indications that spoilage may arise from organisms present in the lymph nodes.

These workers also found that aureomycin is more rapidly destroyed by beef tissue than by water alone in a relatively short time. Beef infused with aureomycin showed slightly altered appearance, but from the standpoint of eating quality was indistinguishable from normal beef.

No adverse effect was noted when beef infused with aureomycin was consumed. Processing of beef by infusion before dressing out shows some promise, but some modification of certain packing house and inspection procedures would be required according to these workers.

Tarr, Boyd, and Bissett (1954) showed that spoilage of whole eviscerated fish was retarded markedly by ices containing 1 to 4 parts per million chlortetracycline by holding 6 days at -1° C. in sea water containing 2 parts per million, or by one minute immersion in solutions containing 50 or 100 parts per million of the antibiotic prior to icing. It was observed that fish iced with ordinary ice attained a state of obvious staleness about four or five days earlier than fish stored in the chlortetracycline-containing ices. These organoleptic improvements in quality were even more obvious with fish stored in sea water containing chlortetracycline, or briefly immersed in the stronger chlortetracycline solutions. No extensive tests were made, but treated fish were generally accepted in the few tests that were made. Boyd, Brumwell and Tarr (1953) also reported that aureomycin at the level of 2 parts per million effectively retarded bacterial growth in fresh eviscerated fish as compared to controls.

Farber (1954), in a study designed to determine preservative action of antibiotics at minimal concentrations, found that at 2 parts per million chlortetracycline and hydroxytetracycline had a definite preservative effect on the fish fillets in the order named. Neomycin was not effective and none of the antibiotics tested were effective in preserving shrimp.

Bacterial Contamination of Dressed Versus Ready-to-Cook Poultry

It has been reported by several workers that the primary source of bacterial contamination of poultry meat comes from outside the carcass. Gundt, Stokes and Goff (1954) studied the source of bacterial contamination of poultry meat by examining possible sources of infestation both interior and exterior to the bird. In one case fifty milliliters of 13 percent saline solution was poured into the body cavity of fresh killed birds and samples of the contaminated solution were plated to determine the extent of bacterial infestation. Counts were very low and in many cases the sample of solution was sterile. Cores of flesh removed from under the skin were found to be sterile in almost every case.

These investigators stated that bacteria could be carried through the skin layer by damage to the skin during scalding, especially at high temperatures, by mechanical damage during plucking and washing, and by the temperature gradient established in the chilling tank, in which contraction due to cooling could serve to lend mechanical assistance to the penetration of both water and bacteria.

The authors stated that failure to recover intestinal bacteria from the cavities of birds generally indicates that passage through the intestinal tract signifies that contamination of the flesh from this source is not an important factor. Factors other than penetration during processing are indicated as being responsible for contamination with bacteria and the subsequent deterioration of poultry meat.

Newell, Gwin and Jull (1948) concluded that chickens stored in ice keep an acceptable appearance longer, but developed off-odors sooner than birds refrigerated in air cooling units. They also observed that eviscerated birds developed off-odors sooner than New York dressed poultry.

Goresline et al (1951) reported that most bacterial contamination in dressed poultry is found on the surface

of the body.

Baker et al (1956) found that bacterial counts on ready-to-cook poultry were higher to start with and increased much more rapidly than on dressed poultry. Counts were highest for poultry stored at 45°F., intermediate for those stored at 35°F., and lowest for those in ice. They also stated that bacterial counts of the caecal contents were high at the beginning of the experiment and did not change significantly during the storage period. Dressed carcasses showed more hydrolytic rancidity of the fatty tissue than the ready-to-cook birds.

Poultry Processing Plant Sanitation And Other Factors Affecting Bacterial Contamination

Walker and Ayres (1956) investigated sanitary conditions in a poultry processing plant by swabbing known areas of the skin surface of birds being processed or by removing known amounts of scald or chill tank water. Samples taken at various stations on the processing line indicated that the numbers of organisms per square centimeter of skin decreased after operations in which washing was involved. Scald water contained an average of 1×10^4 organisms per milliliter; chill water 1×10^5 organisms per milliliter. Increases in the microbial flora on the birds resulted as a consequence of handling and immersing in water. Initial bacterial populations on birds obtained from different sources ranged from 5×10^3 to 1×10^6 organisms per square centimeter. Yeasts were present initially at average levels of 1×10^3 per square centimeter and increased upon storage to levels which seldom exceeded 1×10^6. Organisms typical of Pseudomonas, Alcaligenes and Achromobacter made up the majority of the population and were associated with the off-odors and sliminess typical of poultry spoilage.

Another report by Gunderson, Schwartz, and Rose (1955) shows that another potential source of bacterial contamination in the processing plant may be the chill tanks. They found that in one plant the bacterial count in a chill tank was 5,400 organisms per milliliter of water but in seven hours increased to 65,000,000 organisms per milliliter. On another day initial count was 6,000,000 and increased to 292,000,000 organisms per milliliter. This difference can be accounted for by the difference in cleanliness of the tanks. The researchers suggested using an overflow sufficient to remove all water every 30 minutes or spray-wash all poultry to cut-down on bacterial contamination.

Further evidence of processing plant contamination was pointed out by Clark (1954) when she reported that researchers found the average bacterial count on birds eviscerated at the following times to be: (1) fresh killed, warm birds--4,888 per square centimeter, (2) frozen then defrosted 37,200, and (3) unfrozen, chilled--60,600 per square centimeter. This study also suggested that the use of chill tanks is a poor practice. High counts (98,000 organisms per cubic centimeter) were found in chill tanks and water at the bottom of the tanks showed 44,000,000 organisms per cubic centimeter. Counts as high as 30,000,000 per milliliter were found in drippings from the trough water at the eviscerating table.

Clark (1954) also reported that an in-plant chlorination program using 10 and 20 parts per million of chlorine successfully lowered both viable and coliform bacterial counts. In-plant chlorination reduced bacterial contamination by as much as 90 percent on equipment and working surfaces.

The effects of in-plant chlorination (20 parts per million of available chlorine) in poultry processing plants were reported by Goresline, Howe, Baush and Gunderson (1951) to have increased shelf-life, lowered bacteria counts on the poultry, reduced odors in the plant, and reduced slime on equipment.

According to Ziegler and Stadelman (1955) chlorine used in the cooling water significantly increased shelf-life as determined by off-odor or appearance of slime. The use of a chlorine dip gave a statistically significant decrease in appearance of slime. This difference was of such small magnitude to be of no practical importance. They also stated that the use of chlorine in the cooling water had considerably more effect on shelf-life of birds processed at 128°F. than on those processed at 140°F.

Spencer, Ziegler, and Stadelman (1954) studied factors affecting the shelf-life of chicken meat and found scalding temperature has very little effect on shelf-life. Their test compared 128°F. with 140°F. with birds scalded at the lower temperature reportedly having a shelf-life of one extra day over those scalded at 140°F. They also observed that packaging has no effect on shelf-life, but does have an effect on weight loss and discoloration. Thin cellophane was found to be undesirable as a protective wrap. Further studies by these investigators showed no difference from a bacterial standpoint between eviscerating birds before or after cooling.

Another possible reason for bacterial counts on ready-to-cook poultry being higher than on dressed poultry was pointed out by Bailey, Stewart, and Lowe (1948). They found that New York dressed birds absorbed 1.5 percent water, eviscerated whole carcasses 7.4 percent, and cut up carcasses 9.4 percent moisture during 135 minutes of chilling in ice slush at 32°F. Cut-up chicken absorbed more water when chilled at 32°F. than when chilled at 70°F.

Organoleptic Tests

Baker et al (1956) reported that bacterial counts within storage conditions did not seem to materially influence personal opinion flavor ratings of cooked broilers. The ready-to-cook birds which had the highest bacterial counts were preferred in flavor to the dressed birds. Off flavor first appeared in the liver, gizzard, thigh, and "oyster." Loss of good flavor was appreciably slower in the breast and wings.

Ziegler and Stadelman (1955) stated that the undesirable odor of the treated birds was different from that of the control birds. The average number of days before off-odor occurred were 11.6, 17.6, 18.41 and 18.9 for controls and groups treated with 10, 20, and 40 parts per million of chlortetracycline, respectively.

Miller (1955) stated that a person would have to eat 145 pounds of raw meat in a day to receive 0.5 grams of Aureomycin chlortetracycline. This is the amount which has been given to geriatrics as a prophylactic dose for as long as four years with only some beneficial results.

OBJECTIVES

Four separate studies were undertaken to determine the effectiveness of certain bacterial inhibitors on shelf-life of poultry meat.

In Studies I and II the primary objective was to determine the effectiveness of chlortetracycline, oxytetracycline, and chlorine in extending the shelf-life of fresh poultry meat treated under laboratory processing conditions.

The objectives of Study III were (1) to determine the effectiveness of chlortetracycline in extending shelf-life of fresh poultry meat treated in a commercial processing plant and (2) to measure the difference, if any, in shelf-life between packaged and unpackaged chicken meat.

There were two basic objectives for undertaking Study IV. They were (1) to check the findings of Study III for unpackaged birds and (2) to determine if people can distinguish flavor differences between chicken meat with high bacteria contamination as compared to that with low bacterial counts after the meat has been cooked.

PROCEDURE

Study I was completed before the author started graduate study at Michigan State University. However, the procedure for this study is practically identical to that of Study II which is considered to be a replica of Study I. Therefore, the procedure discussed will be that of Study II.

Sixty White Plymouth Rock broilers were purchased from a commercial broiler grower the evening before the birds were slaughtered. Feed was withheld from the birds for approximately 12 hours, after which the birds were slaughtered in the Poultry Department Processing Laboratory.

The broilers were slaughtered with a standard sticking knife by severing the jugular veins. After thorough bleeding the birds were placed in a Rotomatic scalding tank for approximately one-minute and twenty-five seconds at a temperature of 130°F. Most of the feathers were then removed on a Greenbrier automatic picker. Most of the "bloom" remained on the birds after removal of the feathers.

All birds were immediately eviscerated and split into halves after picking. Before the birds were placed into the various chill tanks, they were thoroughly rinsed with

cold water. Split broiler halves were tagged through the wing web with identification cards which bore the treatment and the number of each broiler half as it was removed from the processing line.

Immediately after tagging and rinsing, thirty halves were placed individually into each of the chill tanks in consecutive order. An equal amount of chipped ice had previously been placed in each chill tank. The order and treatment within the chill tanks was as follows: tank A--control, tank B--10 parts per million of chlortetracycline, tank C--10 parts per million of oxytetracycline, and D--20 parts per million of chlorine. All chill tanks were thoroughly scrubbed prior to processing although no disinfectant was used. The birds were allowed to remain in the chill tanks for from three to four hours. Time in the tanks was dependent upon when each bird was slaughtered.

After the birds were thoroughly chilled, they were removed from the tanks and packed in chipped ice in drainable stainless steel trays. The iced birds were then removed from the processing laboratory to adjacent refrigerated coolers. All trays were carefully covered with three layers of waxed paper before being removed from the processing laboratory. The refrigerated room temperature was approximately 45°F. and the temperature within the iced trays measured 34°F.

The day following slaughter one broiler half from each treatment was removed from the trays by touching only a small portion of the hock joint. Each half was then placed in a polyethelene bag and carefully removed to another room where a raw odor determination test was made by a panel of four experienced people.

The procedure for the raw odor panel consisted of the author removing each broiler half from the bag by the tip of the hock joint and letting each panel member smell the surface area. Then the birds would be returned to the bag individually after each determination was made. The polyethelene packaged birds were then carefully wrapped in two layers of waxed paper and taken to the Department of Microbiology and Public Health where bacterial determinations were made.

The bacteriological procedure consisted of cutting the polyethelene bags with sterile scissors and swabbing a four square inch area of each bird. The area swabbed was the thigh. The contaminated swab was then placed in a test tube which contained 10 milliliters of saline solution. This tube was shaken for two minutes.

Before shaking the test tubes containing the swabs, however, the swabbed birds were each placed in individual one-gallon jars which contained 200 milliliters of distilled water. The jars were then sealed and placed on a small platform where they were securely held and shaken, automatically, at a rate of 200 oscillations per minute for two minutes. After shaking, one milliliter of the rinse water from each jar was added to a small bottle containing 99 milliliters of saline solution.

After the shaking process for both swab and rinse tests, suitable dilutions were made from the respective solutions to facilitate counting the colonies of organisms after the incubation period.

The organism-containing solutions were then plated on two types of media with the exception of the solution obtained from the swab test. The only determination made for the swab test was on tryptone-glucose-extract agar which was incubated for three days at 20°C. For the rinse test, three determinations were made. One was the same as that described for the swab test, but the others consisted of plating a portion of the solution on phytone-naccarol agar and incubating at 20°C. for three days while the third technique was to plate some of the solution on tryptone-glucose-extract agar and incubate the plates for seven days at 4.5°C.

The tryptone-glucose-extract agar gave the total bacteria counts, while the phytone-naccarol agar was used for measuring only gram-negative organisms.

Following this bacteriological procedure, the same birds were taken to the Department of Foods and Nutrition where raw odor tests were again made and all birds were cooked and ranked according to cooked flavor. The raw odor and flavor score card had a range of from one to seven with a word description of each score. A score of one meant "very poor" and seven denoted "excellent."

This same procedure was continued on each of the test days until the study was completed. The raw odor panel, which assembled in the Poultry Department, met at 7:45 a.m., the birds were delivered to the bacteriologists at 10:00 a.m., and finally to the Foods and Nutrition Department by 11:00 a.m. on each test day.

Study III

The author obtained the birds at a commercial processing plant for this study already treated and packaged.

The treatments were: (A) 30 split, unpackaged controls (B) 30 cut-up, packaged controls (C) 30 split, unpackaged, and treated with chlortetracycline and (D) 30 cut-up, packaged, and treated with chlortetracycline. All birds were to have been split into halves, but the packaged birds were prepared in cut-up style.

No attempt was made to determine the source of breeding, but the birds were representative of commercially prepared chickens ready for distribution. Some minor changes in procedure were made as compared to Studies I and II. Raw odor determinations, however, were continued as described for Studies I and III.

The bacteriologists decided it would be desirable to measure bacterial contamination every other day during the study and thus eliminate the uncertainty of determining bacterial growth over the weekends. Therefore, birds were taken to be swabbed and rinsed immediately after raw odor determinations were made as described for Studies I and III. One change was made in the bacteriological procedure. Since the birds used in Study II were considerably larger than those used in Study I, the amount of distilled rinse water was increased from 200 to 400 milliliters.

A change in procedure was required for the flavor panel determinations due to the change in dates of bacterial contamination tests. The Foods and Nutrition Department had made previous arrangements for their taste panel to meet every Monday, Wednesday, and Friday at noon. Since bacterial counts were obtained every other day, irrespective of week-ends, it was necessary to furnish the taste panel with birds which had not been subjected to swabbing and rinsing part of the time. During the weeks when swabbing and rinsing were done on Tuesday, Thursday, and Saturday, unswabbed and unrinsed birds were supplied to the Foods and Nutrition Department directly from the iced trays.

Bacterial counts and raw odor determinations were made for a period of 17 days after slaughter. However, flavor determinations were discontinued after the 15th day of storage.

Study IV

Due to the results of Study III, it was felt that additional investigations should be continued dealing with fresh chicken meat processed in a commercial plant.

At the beginning of this study Dr. L. E. Dawson of the Poultry Department and three members of the Bacteriology Department traveled to the commercial processing plant to obtain birds for Study IV and to check the sanitary conditions of the plant.

The sanitation check consisted of either swabbing known surfaces or obtaining solutions from known containers. The positions in the processing plant were checked as follows: (1) a bird on the processing line after being singed and washed (2) a bird after viscera was removed and before second wash (3) a bird after neck was removed (4) a bird after second wash (5) a bird on weighing pan (6) a solution of chlortetracycline in chill tank #1 (7) a solution of chlortetracycline in chill tank #2 (8) a solution of chlortetracycline in chill tank #3 (9) a solution of chlortetracycline in chill tank #4

(10) chlortetracycline treated pieces of cut-up chicken meat after being placed in boat-shaped, cardboard packaging cartons and (11) a swab of the table top where chilled poultry was cut-up for packaging.

The treatments used in this study were (A) 30 unfrozen control birds (B) 30 frozen control birds (C) 30 chlortetracycline-treated, unfrozen birds and (D) 30 chlortetracycline-treated, frozen birds. All birds were halved and loosely wrapped in polyethelene bags.

The frozen birds were placed in a freezer at -40°F. and the unfrozen birds were refrigerated in a 35°F. cooler. This was the only study in which birds were either frozen or refrigerated in air.

No official raw odor determinations were made with the exception of determining rejection dates,

Each evening, prior to check days, the frozen birds were removed from the freezer and placed in a 60°F. room for thawing.

The procedure for bacteria counts was identical to that of Study III with all determinations being made at 9 a.m.

The primary change in procedure involved the taste panel. Birds were cooked in the Poultry Department meat technology laboratory and a ten-member panel instead of a five-member panel which had been used in Studies I, II, and III was used. No attempt was made to have the same ten people taste the cooked meat during each day of the trial. Only flavor was considered by this panel. The panel met every other day in accordance with the dates of bacterial counts. All birds subjected to cooking had been examined for bacterial contamination.

RESULTS AND DISCUSSION

Studies I and II

Bacteriological Data:

Studies I and II involving chlortetracycline, oxytetracycline, and chlorine have shown there are differences among bacterial inhibitors in deterring bacterial growth.

Figure 1 and Table I show that the initial bacteria counts taken from birds of the three treatments were below that of the control group, although none of the counts were high. The counts after one day of storage ranged from 3.00×10^2 for the chlorine treated birds to 1.57×10^4 for the control birds. After six days of storage, bacteria counts of controls began to rise quite rapidly, reaching a population of 3.97×10^6 after ten days of storage and rising to a peak of 2.45×10^8 after 17 days of storage. No noticeable increase in organisms for birds treated with chlortetracycline was noted until after approximately 11 days of storage. The controls reached a population of 1.00×10^6 about seven days sooner than did the chlortetracycline treated birds.

While the counts were slightly higher for the controls on the first day after storage, the initial effects of bacterial contamination were observed later. The oxytetracycline-treated birds began to show rapid increases in organisms after eight days of storage. Both the control and oxytetracycline-treated birds reached an organism population of 1.00 x 10^6 or more ten days after storage. After 14 days of storage the bacterial counts were generally higher for the oxytetracycline-treated birds than for the controls. In comparing the actual counts of bacterial contamination listed in Table I, it should be noted that the actual number of organisms present on these groups (oxytetracycline and control) are quite similar. Therefore, these studies showed that chlortetracycline was superior to no treatment or oxytetracycline in controlling bacterial counts.

The chlorine-treated birds did not show a bacteria population of 1.00 x 10^6 until about 13 days after storage or two days later than the controls and birds treated with oxytetracycline. A close analysis of Table I will reveal that there was very little difference in bacteria counts for the controls, oxytetracycline-treated birds, or chlorine-treated birds.

These results show that chlortetracycline was more effective in inhibiting the growth of bacteria organisms than all other treatments listed.

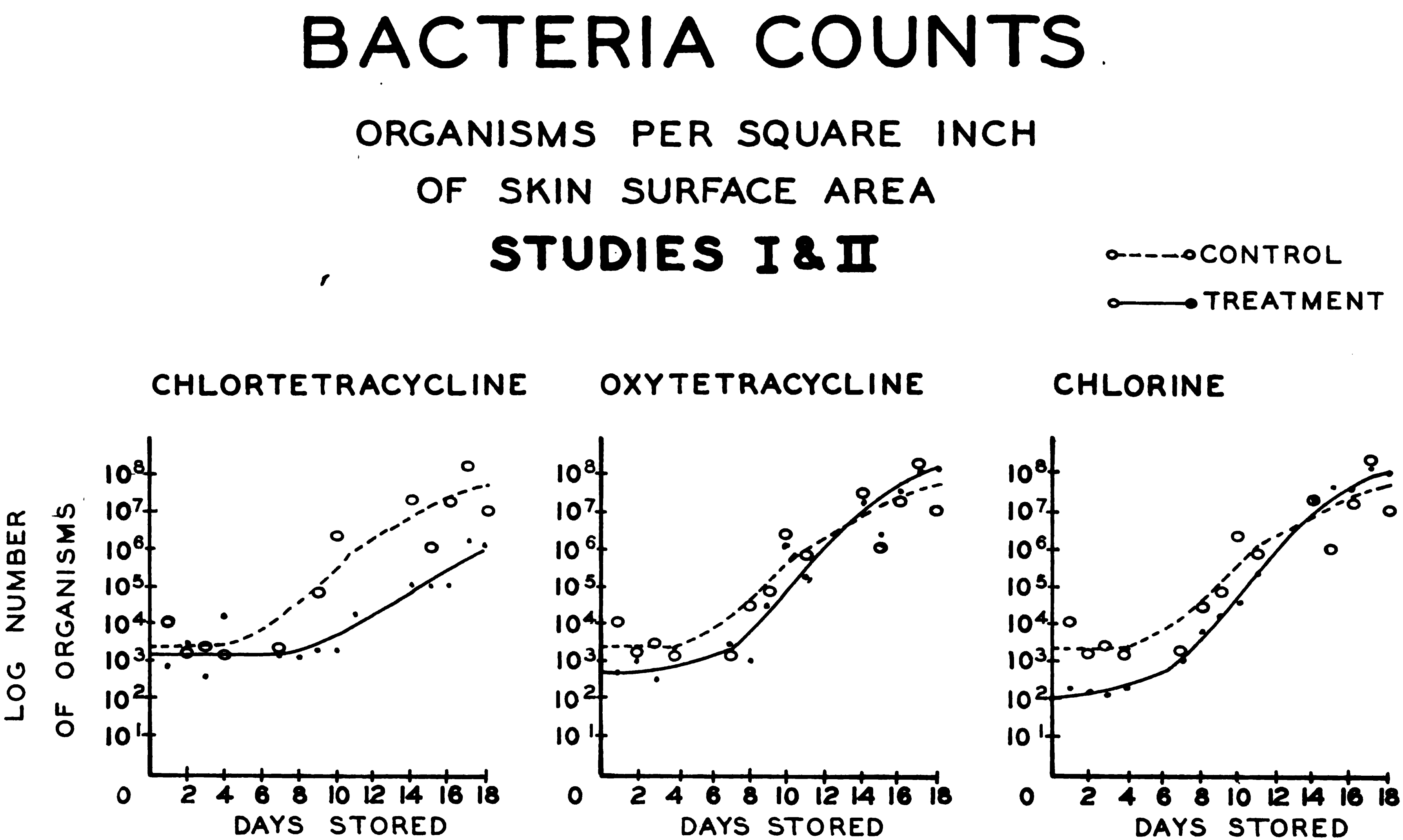


Figure 1. The effect of treatments on total bacteria counts. Counts of organisms were obtained by the swab method. Plates were incubated for three days at 20°C. on tryptone-glucose-extract agar.

BACTERIA COUNTS

ORGANISMS PER SQUARE INCH
OF SKIN SURFACE AREA

STUDIES I & II

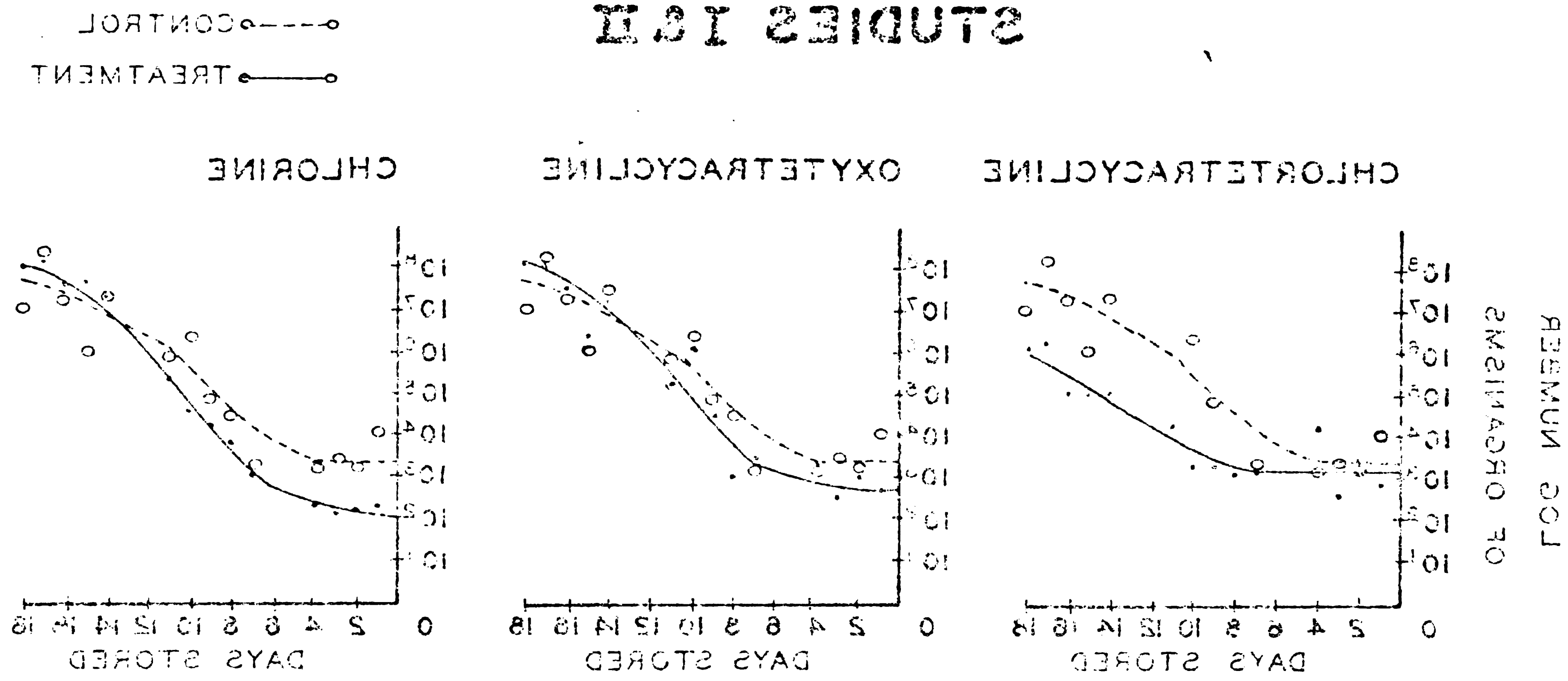


TABLE I

A COMPARISON OF TOTAL BACTERIAL COUNTS[1]
BETWEEN TREATMENTS

Studies I & II

Storage Time (Days)	TREATMENTS[2] Control	Chlortetracycline	Oxytetracycline	Chlorine
	Bacterial Counts			
1	1.57×10^4	9.65×10^2	7.27×10^2	3.06×10^2
2	2.92×10^3	4.10×10^3	1.19×10^3	2.31×10^2
3	4.65×10^3	6.46×10^2	5.70×10^2	1.71×10^2
4	2.15×10^3	1.24×10^4	3.05×10^3	3.31×10^2
7	3.47×10^3	2.31×10^3	4.95×10^3	1.12×10^3
8	5.60×10^4	1.60×10^3	1.39×10^3	8.10×10^3
9	8.44×10^4	3.01×10^3	4.02×10^4	2.77×10^4
10	3.97×10^6	3.00×10^3	1.69×10^6	6.19×10^4
11	9.76×10^5	2.19×10^4	3.17×10^5	3.50×10^5
14	3.66×10^7	1.25×10^5	2.71×10^7	3.37×10^7
15	1.05×10^6	1.28×10^5	4.12×10^6	6.72×10^7
16	3.82×10^7	1.27×10^5	5.00×10^7	6.15×10^7
17	2.45×10^8	2.12×10^6	1.23×10^8	1.95×10^8
18	1.29×10^7	1.50×10^6	1.67×10^8	1.29×10^8

[1] Total bacterial counts are expressed in log number of organisms per square inch of skin surface area.

[2] All birds in these studies were unpackaged and stored in crushed ice.

Raw Odor Panel Data:

The one sense which is frequently used as a measure of acceptability for uncooked foods is smell. For this reason a raw odor panel was used to judge odor scores and determine when birds were unacceptable. Figure 2 and Table II show how the panel rated the various treatments.

One interesting fact to note is that chlortetracycline-treated birds were initially ranked below the control group. After 14 days of storage this panel began detecting noticeable differences in odor between control birds and those treated with chlortetracycline. This is in accordance with the bacteria counts found in Table I in which bacteria population data indicated that the controls reached the stage of 3.97×10^6 at 10 days which apparently indicates a point of noticeable difference in odor. This study was discontinued when the control group reached a point of obvious unacceptability. If the study had been extended, it is quite possible the differences between the controls and the treatment would have been further apart. Table II shows a score of 3.25 for control birds after 18 days of storage compared to 4.75 for the chlortetracycline-treated birds. When treated statistically this difference was not significant although there was a noticeable difference as determined by the panel. A t-value of .67 was obtained for 0-18 days of storage and .76 for 11-18 days. To be significant at the 5 percent level a t-value in excess of 2.18 was

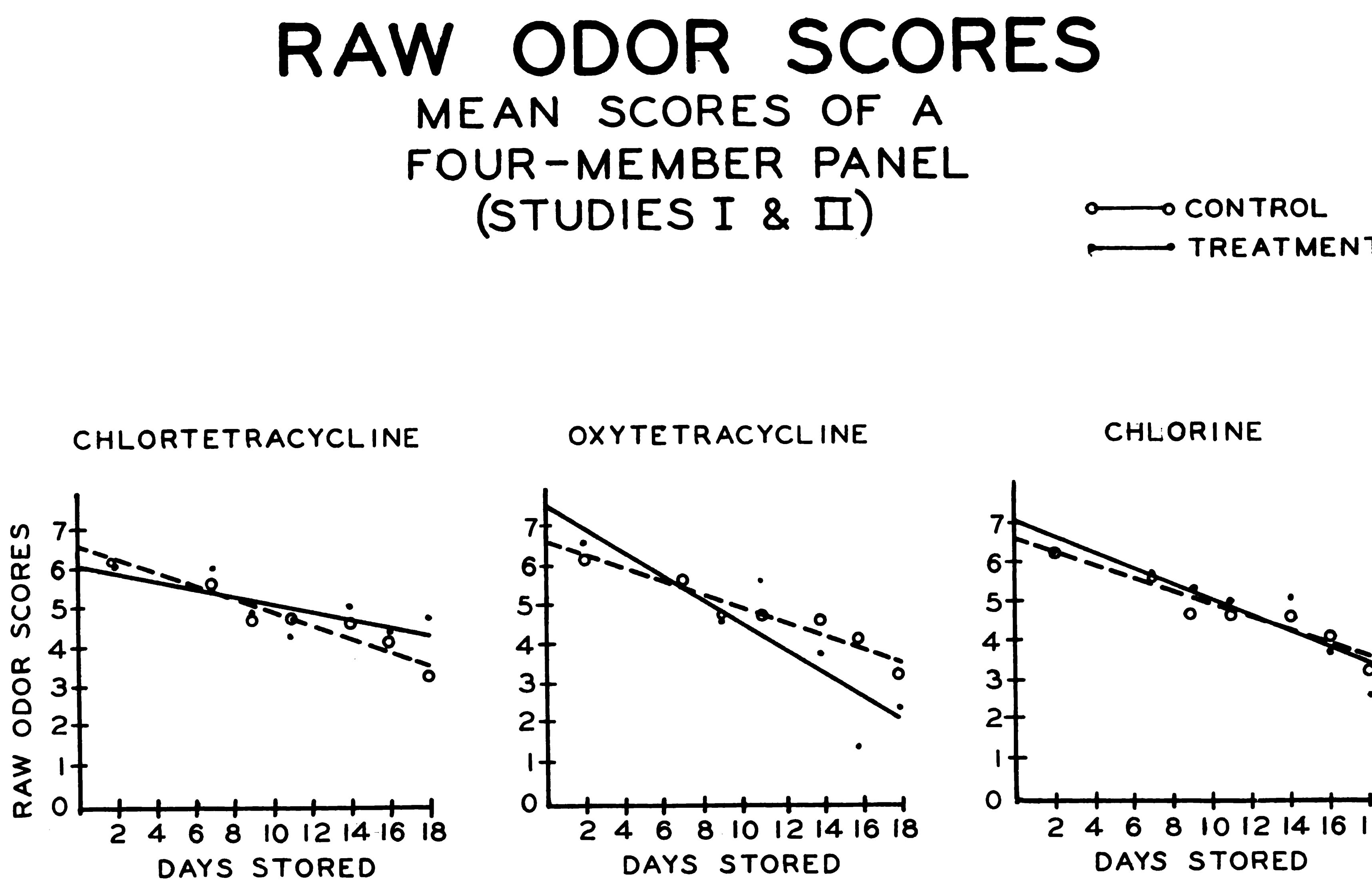


Figure 2. The effect of treatments on raw odor scores. A scoring range of 1-7 was used, with 1 meaning very poor and 7 meaning excellent.

RAW ODOR SCORES

MEAN SCORES OF A
FOUR-MEMBER PANEL
(STUDIES I & II)

CONTROL o——o
TREATMENT •- - -•

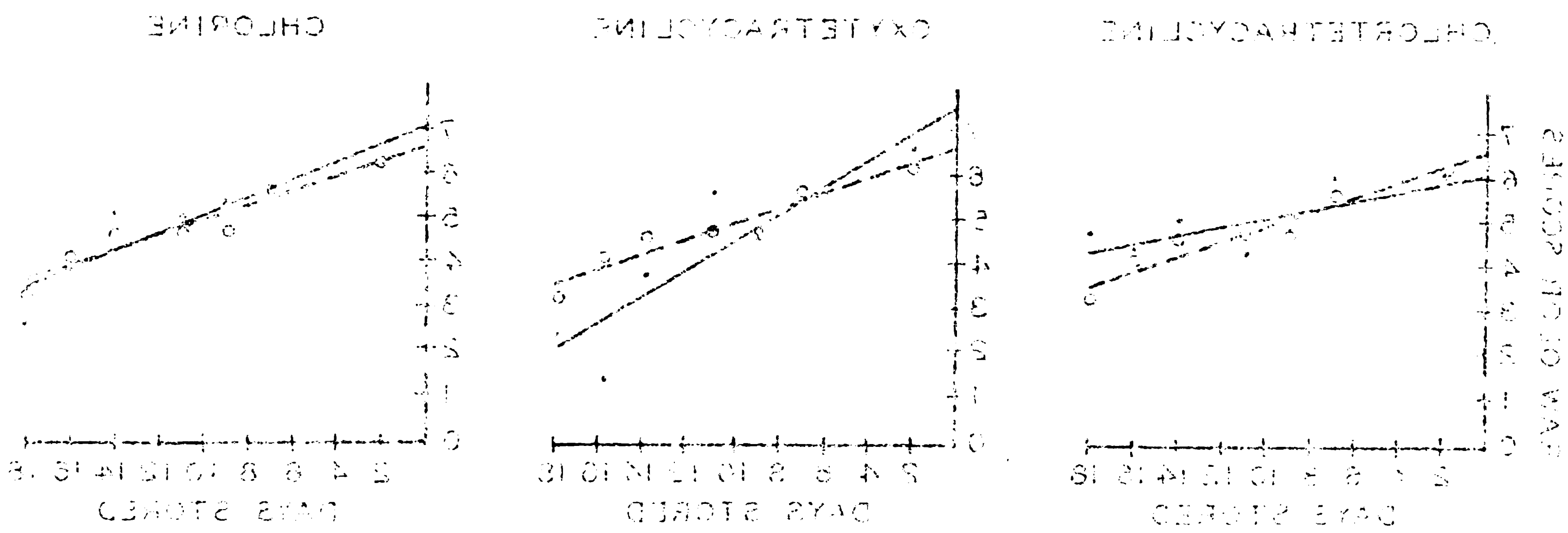


TABLE II

A COMPARISON OF RAW ODOR SCORES
BETWEEN TREATMENTS

Mean Scores of A
Four-Member Panel

Studies I & II

Storage Time Days	TREATMENTS			
	Control	Chlortetracycline	Oxytetracycline	Chlorine
	Mean Panel Scores For The Various Treatments [1]			
2	6.25	6.12	6.62	6.25
7	5.62	6.00	5.50	5.75
9	4.75	4.87	4.62	5.37
11	4.75	4.25	5.50	5.00
14	4.62	5.00	3.75	5.12
16	4.12	4.37	1.37	3.75
18	3.25	4.75	2.37	2.62

[1] A scoring range of 1-7 was used with 1 meaning very poor and 7 meaning excellent.

needed for 12 degrees of freedom.

Beginning the 14th day after storage, birds treated with oxytetracycline received scores lower than controls on the same day. The raw odor panel ranked oxytetracycline-treated birds below the controls the last three test days according to data in Table II. After 6-8 days of storage, control birds were better accepted than were oxytetracycline-treated birds.

No differences were found between birds treated with chlorine and the untreated controls. Differences in raw odor scores were small. Chlortetracycline treated birds were acceptable for a longer period of time by the raw odor panel than were either oxytetracycline or chlorine treated birds.

Correlations between raw odor quality and time were fairly high ranging from -0.59 for chlortetracycline to -0.71 for controls. When all treatments and the controls were grouped, a correlation coefficient of -0.67 was obtained. Statistical treatment of the data showed no significant differences in comparing the various groups for the entire period of study.

The raw odor scores of Study II are shown separately in Table III. Birds receiving a score of 4.50 or below were considered unacceptable. After 11 days of storage two of the four panel members rejected the bird treated with chlortetracycline. However, it has been observed that there are wide

TABLE III

THE RELATIONSHIP BETWEEN RAW ODOR SCORES AND ACCEPTANCE

Mean Scores Of A Four-Member Panel

Study II

Storage Time--Days	TREATMENTS			
	Control	Chlortetracycline	Oxytetracycline	Chlorine
	Mean Panel Scores For The Various Treatments [1]			
2	6.50	6.25	6.75	6.50
7	5.75	6.00	5.50	6.00
9	5.00	3.75	5.25	5.25
11	5.00	4.50[2]	5.00	4.50
14	4.75	5.00	2.00[3]	4.25[2]
16	2.25[3]	3.75	1.25[3]	2.50[3]
18	2.00[3]	4.00[3]	1.25[3]	1.25[3]

[1] A scoring range of 1-7 was used, with 1 meaning very poor and 7 meaning excellent.

[2] Rejected for acceptability by at least two panel members.

[3] Rejected for acceptability by at least three panel members.

differences between individual birds within the same treatment and also differences in panel member's opinions and sensory keenness from day to day. This can be exemplified by noting the panel score for the chlortetracycline-treated bird on the 16th day after storage. The score on the 16th day was 3.75, but the bird was accepted by three of the four members.

After 14 days of storage, three panel members rejected the oxytetracycline-treated bird. The only bird considered acceptable after 16 days of storage was the chlortetracycline treated bird. After 18 days of storage all birds were rejected.

In comparing the bacteria counts found in Table I with rejection dates, in terms of storage, found in Table III it can be noted that date of rejection corresponds to a degree with a bacterial count of approximately 1.00×10^8. However there are exceptions to this observation.

Taste Panel Data:

An experienced five-member taste panel sampled portions of the breast and thigh meat of the same birds for which bacteria counts and raw odor scores are listed in Tables I and II, respectively.

Figure 3 and Table IV show how the panel ranked birds of various treatments for flavor. No skin was included in the flavor tests. The results are somewhat in agreement

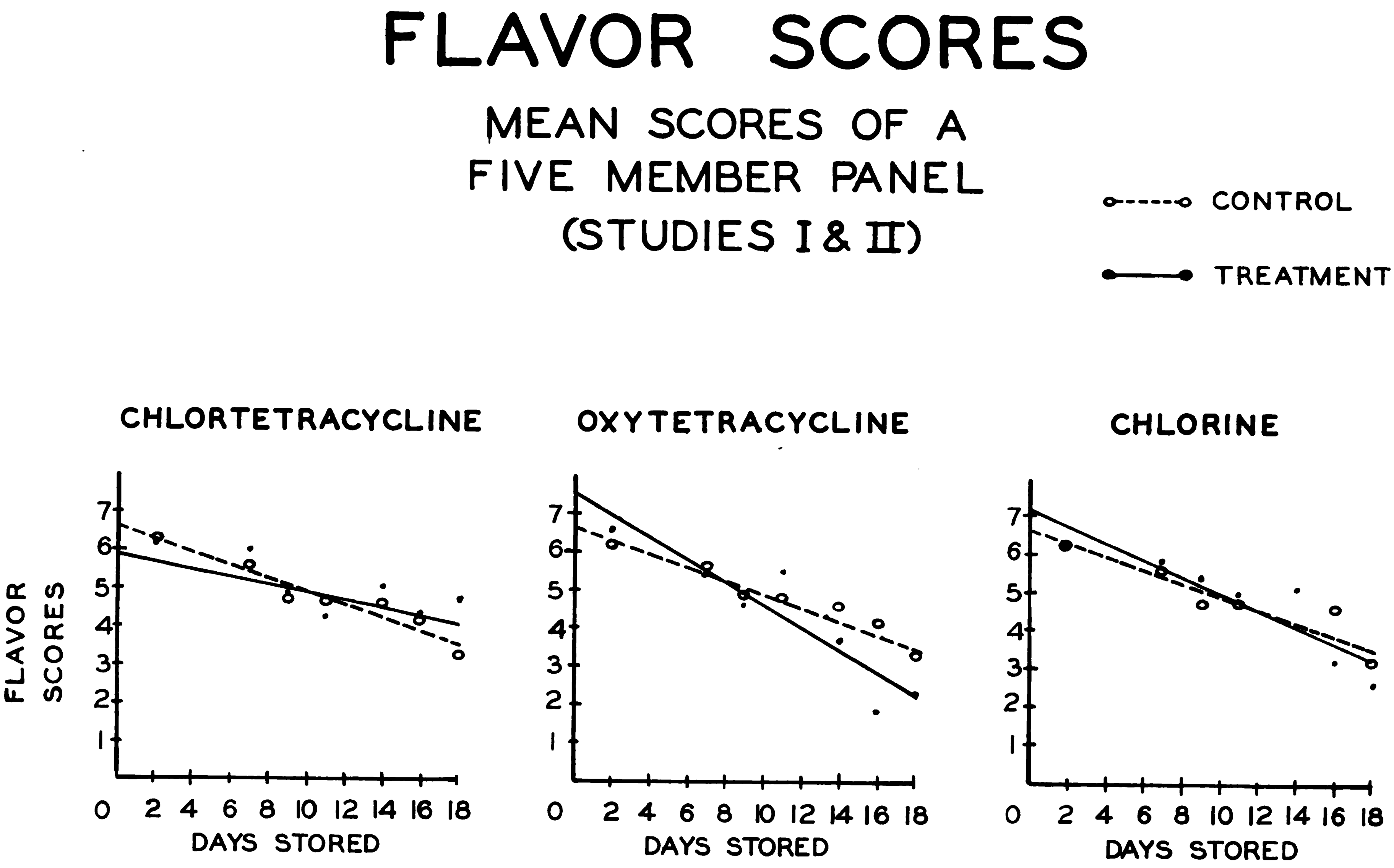


Figure 3. The effect of treatments on flavor scores. A scoring range of 1-7 was used with 1 meaning very poor and 7 meaning excellent.

FLAVOR SCORES

MEAN SCORES OF A
FIVE MEMBER PANEL
(STUDIES I & II)

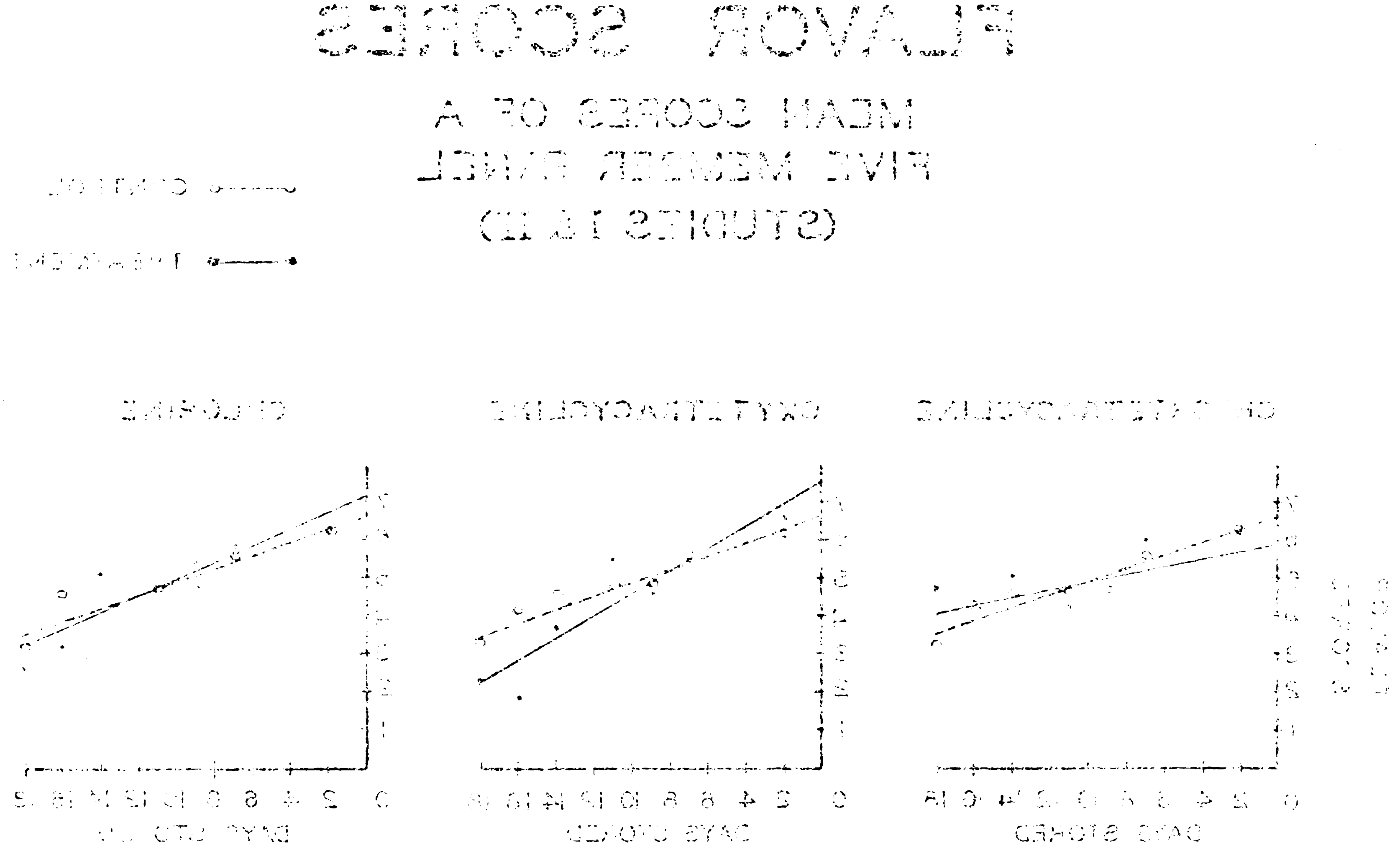


TABLE IV

A COMPARISON OF FLAVOR SCORES BETWEEN TREATMENTS

Mean Scores Of A Five-Member Panel

Studies I & II

Storage Time (Days)	TREATMENTS			
	Control	Chlortetracycline	Oxytetracycline	Chlorine
	Mean Panel Scores For The Various Treatments[1]			
2	6.25	6.10	6.60	6.25
7	5.60	6.00	5.50	5.75
9	4.75	4.85	4.60	5.35
11	4.75	4.25	5.50	5.00
14	4.60	5.00	3.75	5.10
16	4.10	4.35	1.85	3.25
18	3.25	4.75	2.35	2.60

[1] A scoring range of 1-7 was used, with 1 meaning very poor and 7 meaning excellent.

with the corresponding bacteria and raw odor results with the flavor panel ranking oxytetracycline-treated birds lowest of all groups. The taste panel began lowering the scores for this group after 14 days of storage which is in close agreement with the results obtained by the raw odor panel. Table IV shows an average score of 3.75 was obtained after 14 days of storage.

There is no appreciable difference in flavor scores when controls are compared with either chlortetracycline-treated or chlorine-treated birds. Table IV shows that chlortetracycline-treated birds received consistently higher scores than birds from the other treatments although the differences were small and not significant.

Initially, the taste panel ranked the chlortetracycline-treated below all other groups which is consistent with the raw odor scores. Again the differences were small and not significant although there has been a consistent trend to place the chlortetracycline-treated birds slightly lower than the other groups on the first check day.

Study III

Bacteriological Data:

This study was designed to measure the effectiveness of chlortetracycline under commercial conditions.

Figure 4 and Table V reveal that initial bacteria counts

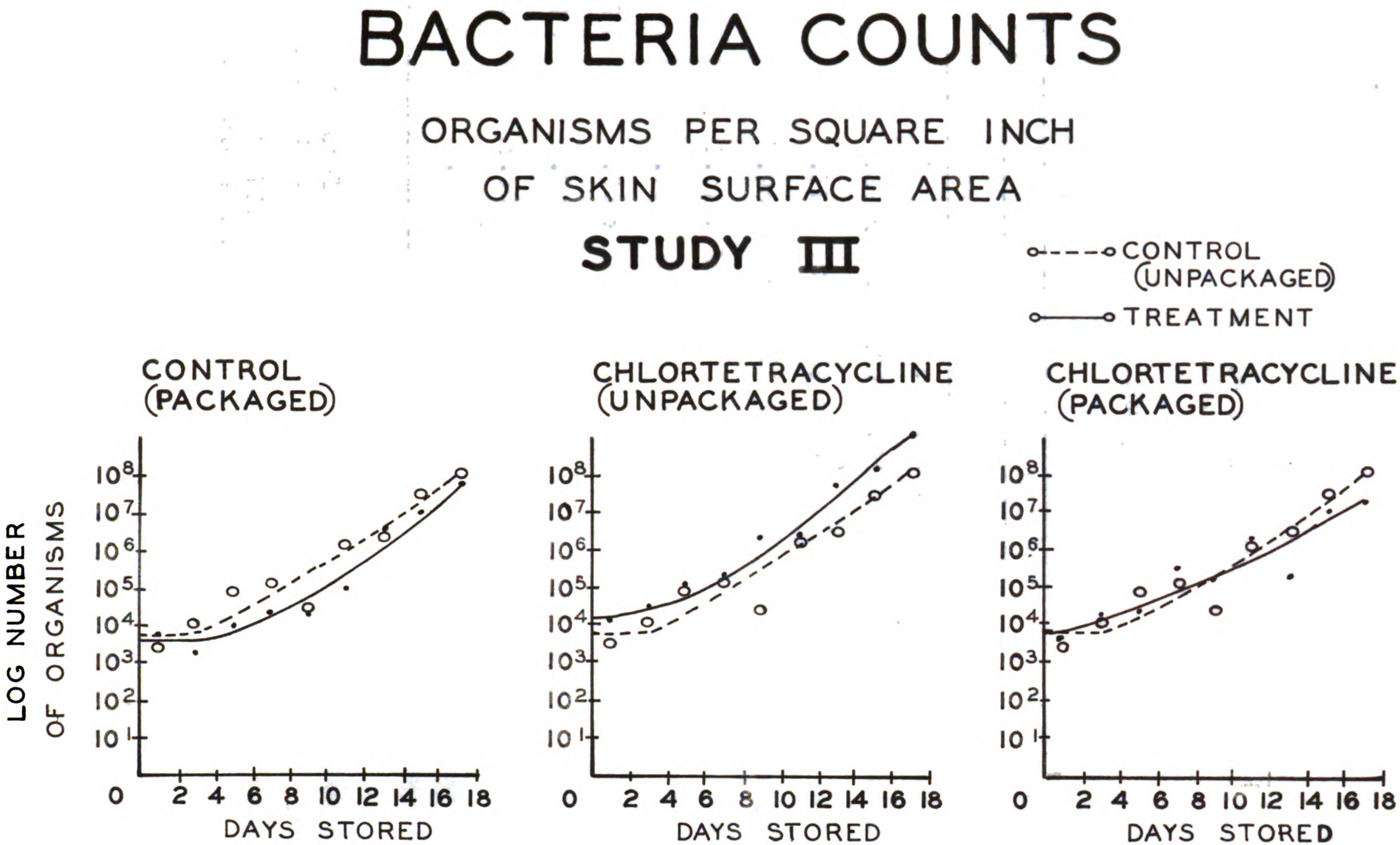


Figure 4. The effect of treatments on total bacteria counts. Counts of organisms were obtained by the swab method. Plates were incubated for three days at 20°C. on tryptone-glucose-extract agar.

BACTERIA COUNTS

ORGANISMS PER SQUARE INCH

OF SKIN SURFACE AREA

STUDY III

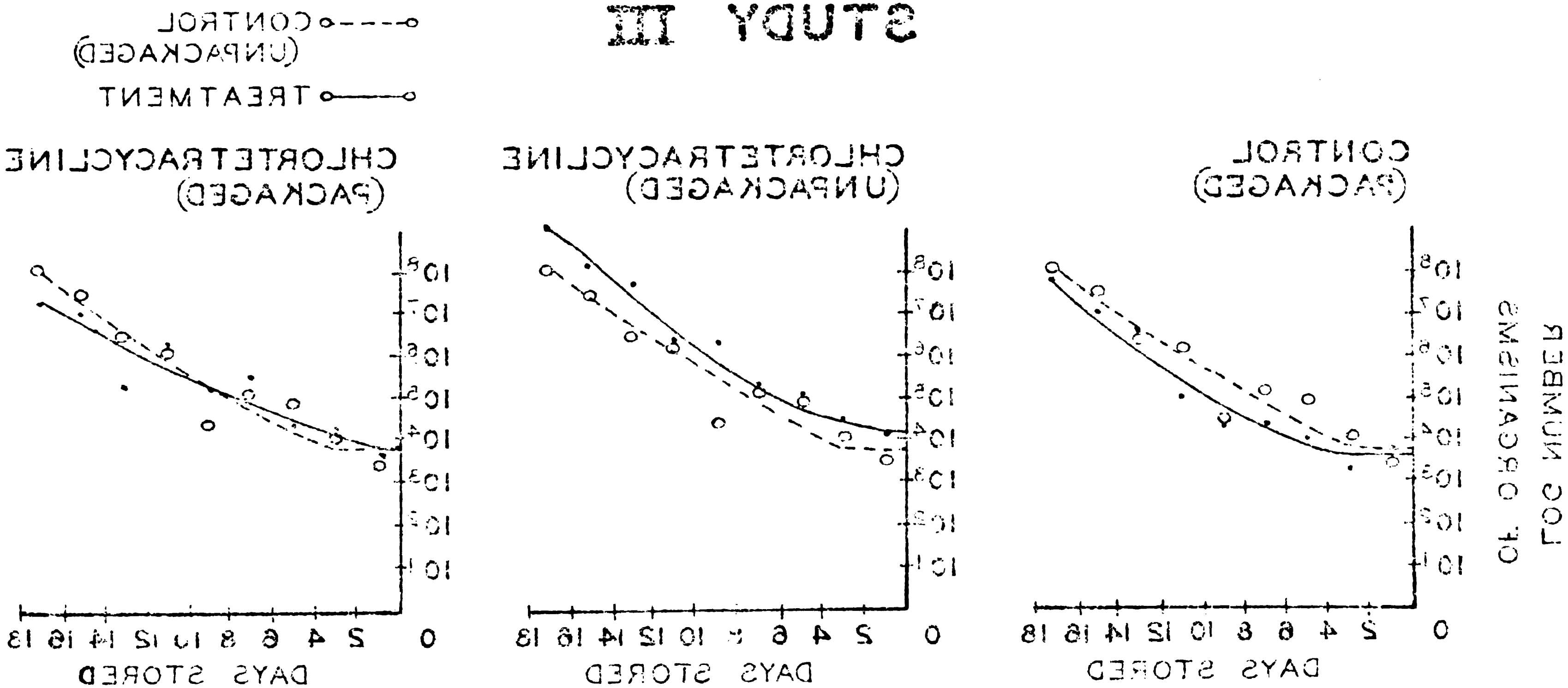


TABLE V

A COMPARISON OF TOTAL BACTERIA COUNTS[1]
BETWEEN TREATMENTS

Study III

Storage Time (Days)	TREATMENTS[2]			
	Control		Chlortetracycline	
	(Unpackaged)	(Packaged)	(Unpackaged)	(Packaged)
	Bacterial Counts			
1	5.83×10^3	8.72×10^3	1.81×10^4	6.18×10^3
3	1.11×10^4	3.59×10^3	4.68×10^4	2.88×10^4
5	9.96×10^4	1.29×10^4	1.06×10^5	3.00×10^4
7	1.61×10^5	3.06×10^4	2.99×10^5	5.56×10^5
9	4.70×10^4	3.00×10^4	3.60×10^6	2.20×10^5
11	1.96×10^6	1.00×10^5	3.37×10^6	2.73×10^6
13	4.70×10^6	5.30×10^6	7.26×10^7	3.00×10^5
15	5.50×10^7	1.30×10^7	1.88×10^8	1.20×10^7
17	1.08×10^8	8.33×10^7	1.17×10^9	2.13×10^7

[1] Total bacteria counts are expressed in log number of organisms per square inch of skin surface area.

[2] All birds were stored in crushed ice. Heat-sealed polyethelene was used for the packaging material.

were higher on commercially treated birds than on laboratory treated birds.

Bacteria counts for unpackaged controls and packaged controls increased at approximately the same rate although the unpackaged birds began to show higher bacterial contamination from one to two days sooner. The unpackaged controls generally had higher bacteria counts than did the packaged controls, and reached a bacteria population of 1.00×10^6 or more approximately two days earlier.

Figure 4 shows an unexpected trend in bacteria counts. Chlortetracycline-treated, unpackaged birds, had higher bacteria counts than the corresponding control birds during every day of this study. The chlortetracycline-treated, unpackaged birds reached a population of 1.00×10^6 or more after nine days of storage or two days sooner than the comparable control group. Table V shows that bacteria counts for the chlortetracycline-treated, unpackaged birds had higher initial bacterial counts and the increase of the organisms was more rapid than for any other group in this study.

These results indicate that the chlortetracycline-treated, unpackaged birds were probably contaminated either in the tank or after removal from the tanks The higher initial organism counts for the commercial birds of this study as compared to the laboratory birds of Studies I and II indicates that some improvements in sanitation could be employed by the commercial processing plant.

Table V also shows that the bacteria counts of the unpackaged, control group and the chlortetracycline-treated, packaged group were similar. Both groups reached a bacteria population level of 1.00 x 10^6 after 11 days of storage.

After nine days of storage the controls began to rise above the treated birds in bacteria counts although the differences were small.

Raw Odor Panel Data:

A comparison of Figures 4 and 5 shows a close relationship between bacterial counts and raw odor on both control groups. Although the changes in raw odor scores approximated the changes in bacteria populations, the packaged controls received scores slightly below the unpackaged controls.

The differences in raw odor scores between the two controls are not significant as shown in Table VI and Figure 5. Neither of the control groups were rejected until after 14 days of storage although both groups had reached a bacteria population of at least 1.00 x 10^6 after 13 days of storage and the unpackaged controls reached a population level of 1.96 x 10^6 after 11 days of storage. This should illustrate the fact that there are odor differences among birds within the same group and/or treatment, and birds may

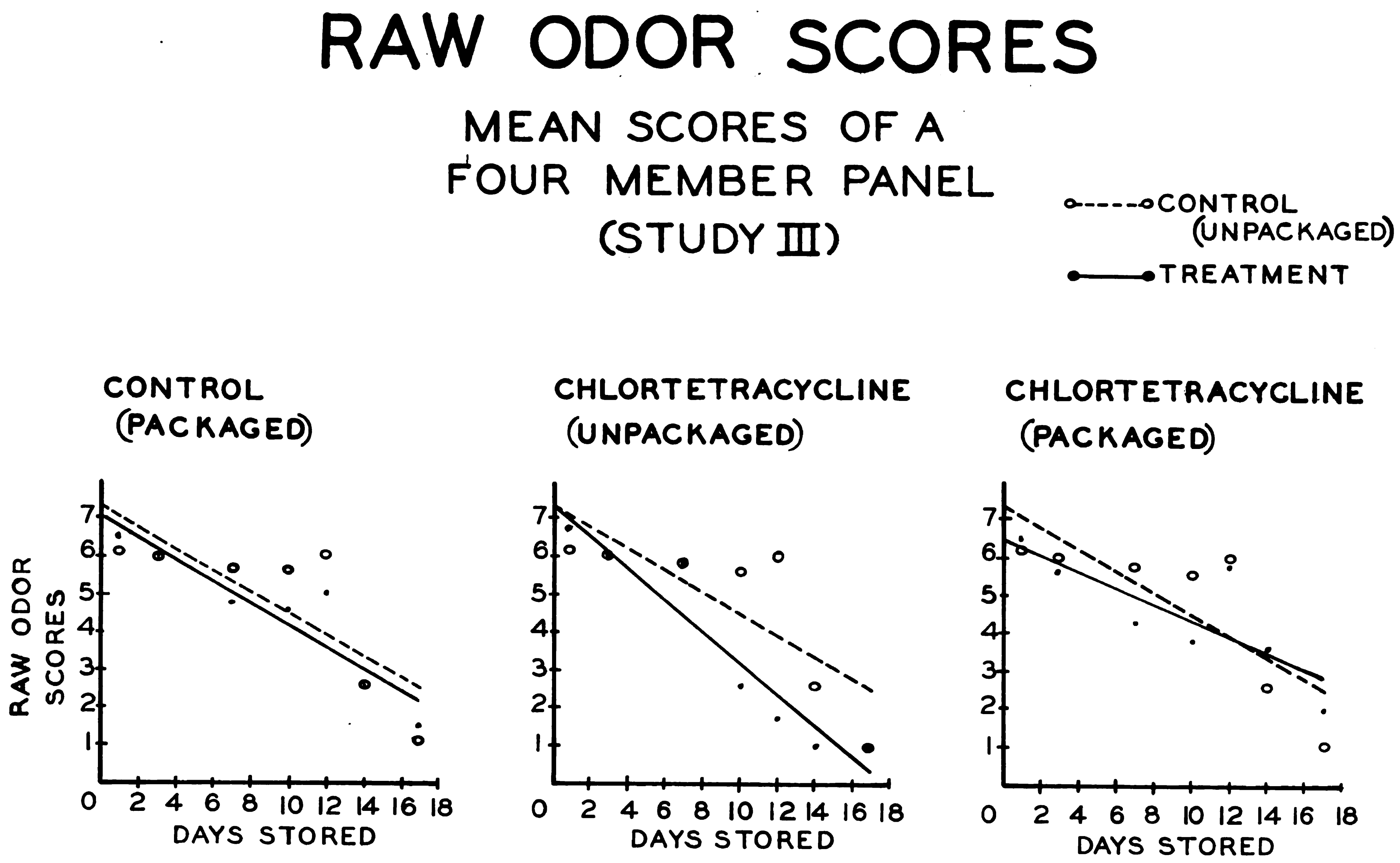


Figure 5. The effect of treatments on raw odor scores. A scoring range of 1-7 was used, with 1 meaning very poor and 7 meaning excellent.

RAW ODOR SCORES

MEAN SCORES OF A
FOUR MEMBER PANEL
(STUDY II)

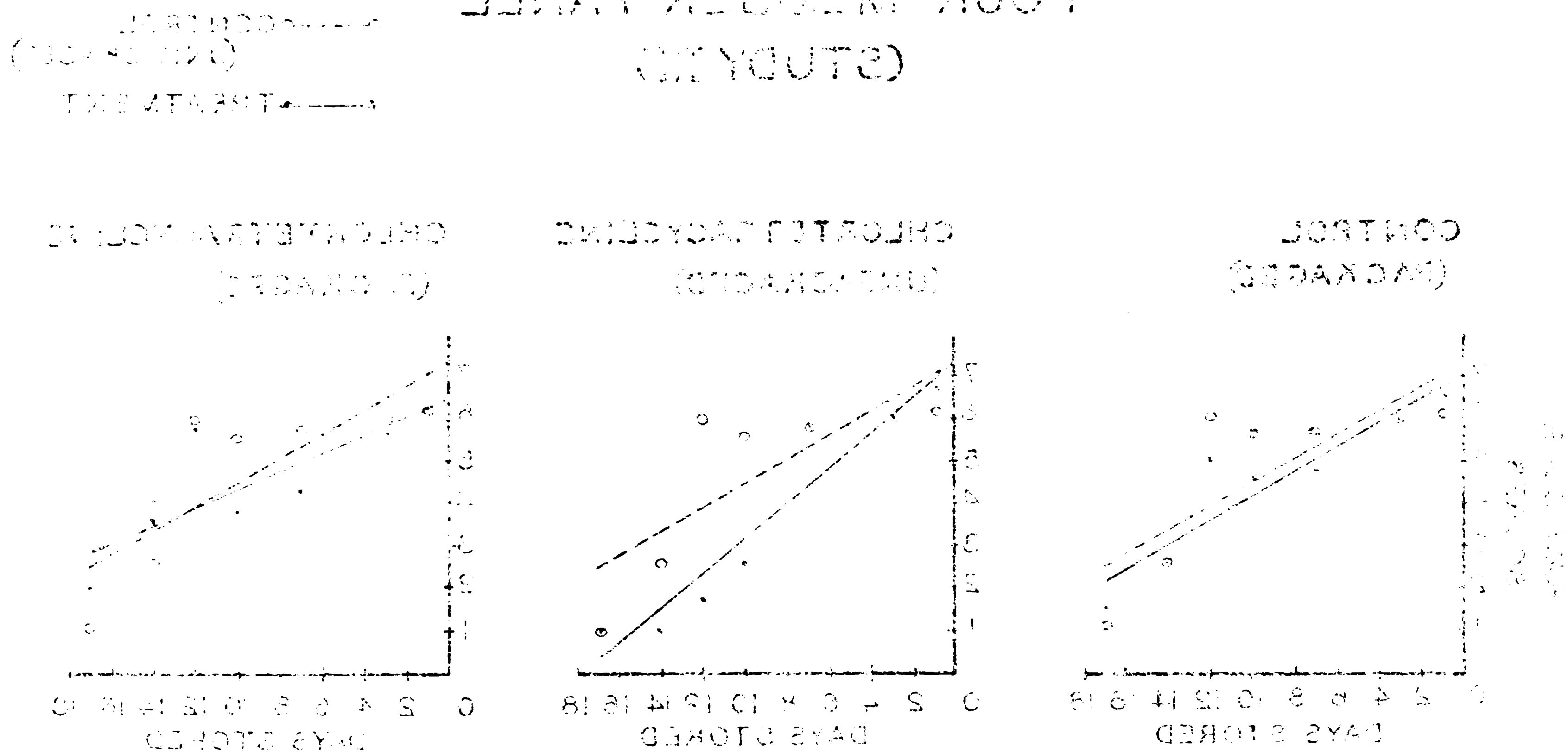


TABLE VI

THE RELATIONSHIP BETWEEN RAW ODOR SCORES AND ACCEPTANCE

Mean Scores Of A Four-Member Panel

Study III

Storage Time (Days)	TREATMENTS			
	Control		Chlortetracycline	
	Unpackaged	Packaged	Unpackaged	Packaged
	Mean Panel Scores For The Various Treatments[1]			
1	6.25	6.50	6.75	6.50
3	6.00	6.00	6.00	5.66
7	5.75	4.75	5.75	4.25
10	5.60	4.60	2.60[2]	3.80
12	6.00	5.00	1.75[2]	5.75
14	2.60[2]	2.60[2]	1.00[2]	3.60
17	1.00[2]	1.50[2]	1.00[2]	2.00[2]

[1] A scoring range of 1-7 was used, with 1 meaning very poor and 7 meaning excellent.

[2] Rejected for acceptability by at least three panel members.

be rejected at organism levels below 1.00×10^6 and accepted at levels above 1.00×10^6. Further work with raw odor detection for acceptability is needed. It appears, however, that the level of positive rejection according to raw odor occurs when the bacteria contamination reaches about 1.00×10^6 organisms per square inch of skin surface.

Figure 5 and Table VI show a very rapid decline in odor score for chlortetracycline-treated birds as compared to the comparable control group. This difference was significant at the 0.5% level. The chlortetracycline-treated, unpackaged birds were rejected by the raw odor panel at ten days of storage and the panel rejected the birds in this group every day thereafter.

In Studies I and II it was observed that the raw odor panel would begin rejecting birds with a score of 4.50 or lower. Table VI shows that chlortetracycline-treated, unpackaged birds received scores of 2.60 and lower after 10 days of storage, and all such birds were rejected. The chlortetracycline-treated packaged bird on the 10th day of storage received a mean score of 3.80 and on the 14th day of storage received a score of 3.60, but in both cases the birds were considered acceptable by the panel. Table V shows the bacteria counts for these days were near or above the level of 1.00×10^6.

Figure 5 again portrays a peculiarity which has been observed with chlortetracycline-treated birds. The raw odor panel usually scores the treated birds below the untreated controls during the first few days. Figure 5 substantiates this observation, but also shows that the chlortetracycline-treated packaged birds had a higher score after 17 days of storage.

Table V shows that bacteria counts for the chlortetracycline-treated, packaged bird after 15 days of storage was 1.20 x 10^7. This bird was accepted by the raw odor panel.

During the course of this study it was noted that the packaged birds did not exhibit off odors as soon as the unpackaged lots, but this could possibly be due to the fact that the unpackaged birds were halved and the packaged birds were cut-up. In order to avoid unnecessary contamination of the birds during the process of determining raw odor scores, small openings were cut in the packages which allowed a minimum of meat surface area to be exposed. The entire surface area of the unpackaged, split birds was exposed. The body cavity of the split birds was exposed for raw odor determinations, but not for the cut-up groups. This probably had some effect on the scores of the unpackaged birds as compared to those which were packaged.

Taste Panel Data:

Figure 6 and Table VII show the flavor scores for the control and treated groups. There were no apparent differences

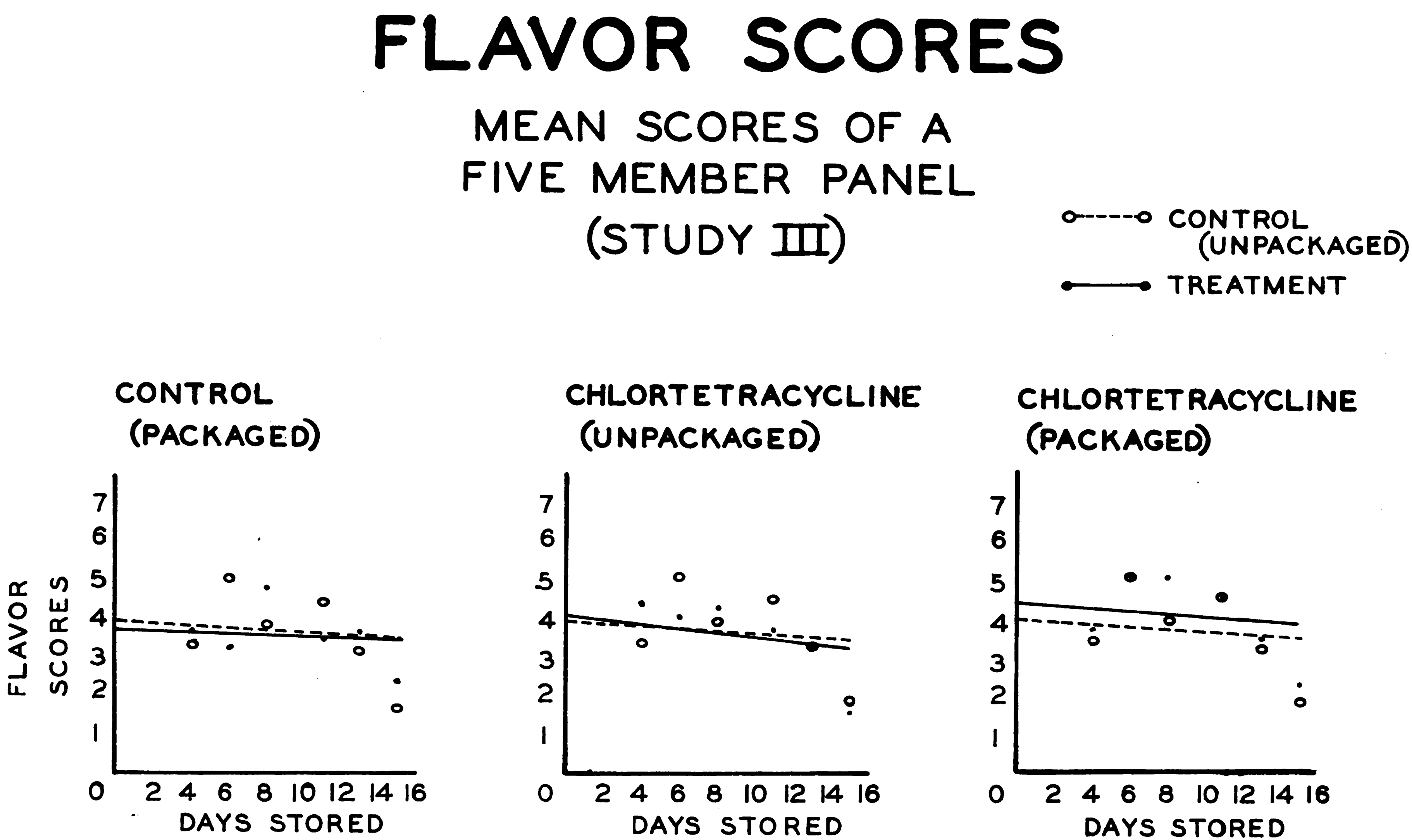


Figure 6. The effect of treatments on flavor scores. A scoring range of 1-7 was used with 1 meaning very poor and 7 meaning excellent.

FLAVOR SCORES

MEAN SCORES OF A FIVE MEMBER PANEL (STUDY III)

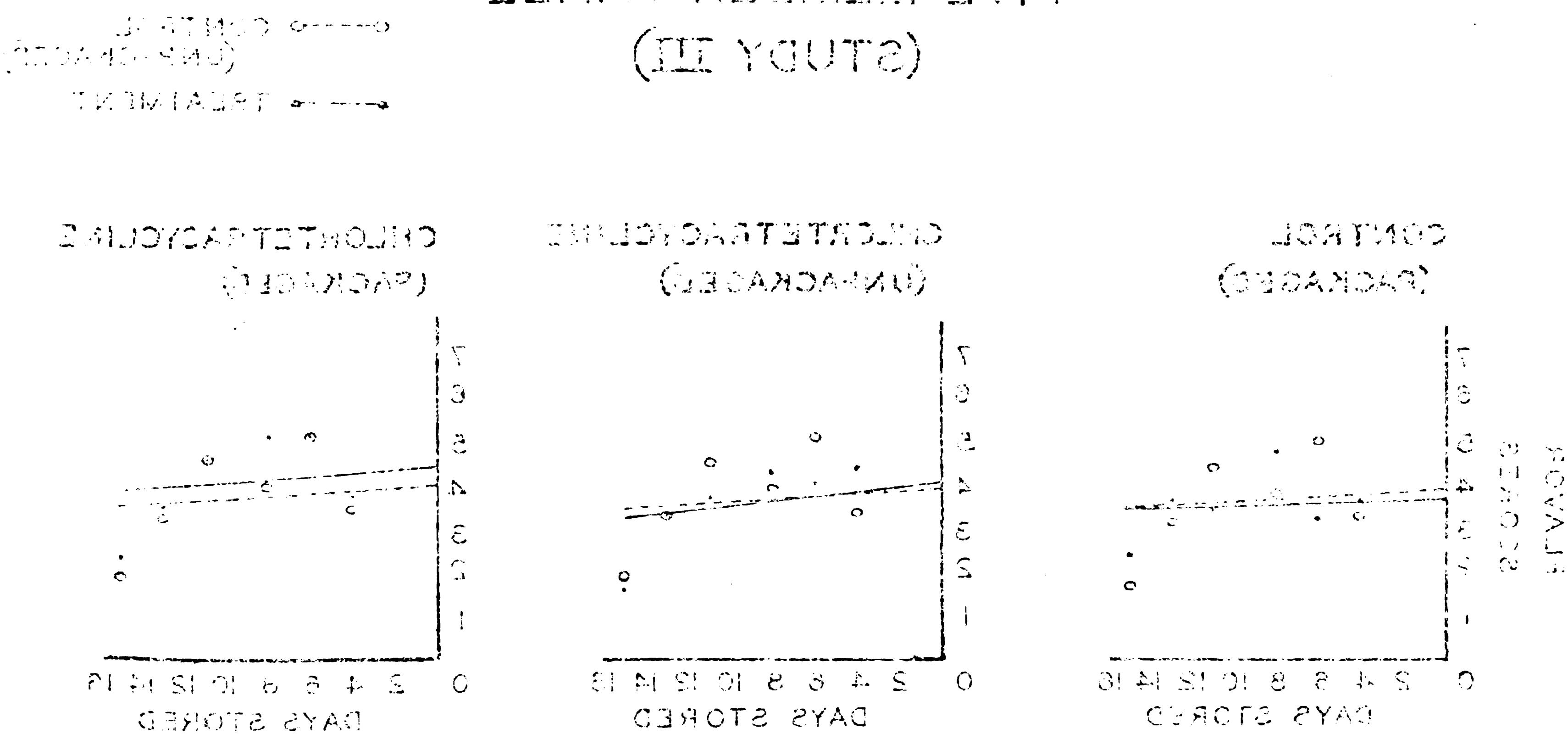


TABLE VII

A COMPARISON OF FLAVOR SCORES
BETWEEN TREATMENTS

Mean Scores of A
Five-Member Panel

Study III

Storage Time (days)	TREATMENTS			
	Control		Chlortetracycline	
	Unpackaged	Packaged	Unpackaged	Packaged
	Mean Panel Scores For The Various Treatments[1]			
4	3.50	3.80	4.50	3.80
6	5.20	3.40	4.20	5.20
8	4.00	5.00	4.40	5.20
11	4.60	3.60	3.80	4.60
13	3.30	3.80	3.30	3.50
15	1.80	2.50	1.50	2.30

[1] A scoring range of 1-7 was used, with 1 meaning very poor and 7 meaning excellent.

in flavor between the packaged and the unpackaged birds. Flavor scores for the chlortetracycline-treated unpackaged birds were practically identical with the unpackaged controls.

Table VII shows that the packaged birds were generally favored over the unpackaged birds although the differences were slight. Only very minor changes in flavor were detected by the taste panel throughout the entire study. Table VII shows that there was little consistency in scoring the birds for flavor. For example, after 8 and 11 days of storage the packaged controls received higher flavor scores than during the early stages of storage when bacteria counts were lower. Another interesting observation shows that the chlortetracycline-treated, unpackaged birds with the highest bacteria counts ranked highest in score after four days of storage and retained a high score after eight days of storage when organisms exceeded one million per square inch of skin surface.

The taste panel results of this study indicate that no real differences could be detected between control and treated lots and that during the course of this study bacterial contamination had no detrimental effect on flavor.

Study IV

Bacteriological Data:

This study serves as a partial replica for Study III in determining if the unusually high bacteria counts and early rejection dates would be duplicated. Frozen birds were used in this study to seek further evidence regarding the ability of people to judge the flavor of chicken meat subjected to different treatments and storage time.

The initial bacteria counts of the commercial birds used in this study duplicated those of Study III. This gave further proof that there is apparently a source of contamination in the commercial processing plant which is causing the high initial counts. The lowest initial count for any lot of this study was 6.53×10^3 (Table VIII) and the highest was 1.06×10^4 for the unfrozen controls.

The unfrozen birds in Study IV exhibited similar increases in bacteria to those in Study III. Bacteria counts from the frozen birds remained about the same throughout this trial.

Bacteria counts for the chlortetracycline-treated birds were slightly higher than those for the unfrozen controls throughout the trial. A comparison of actual bacteria counts (Table VIII) indicates little difference between the unfrozen controls and the unfrozen treated birds. This was quite consistent with results obtained in Study III.

The bacteriological results of Study IV were quite consistent with those of Study III indicating a need for further work to determine the source of contamination in the commercial plant.

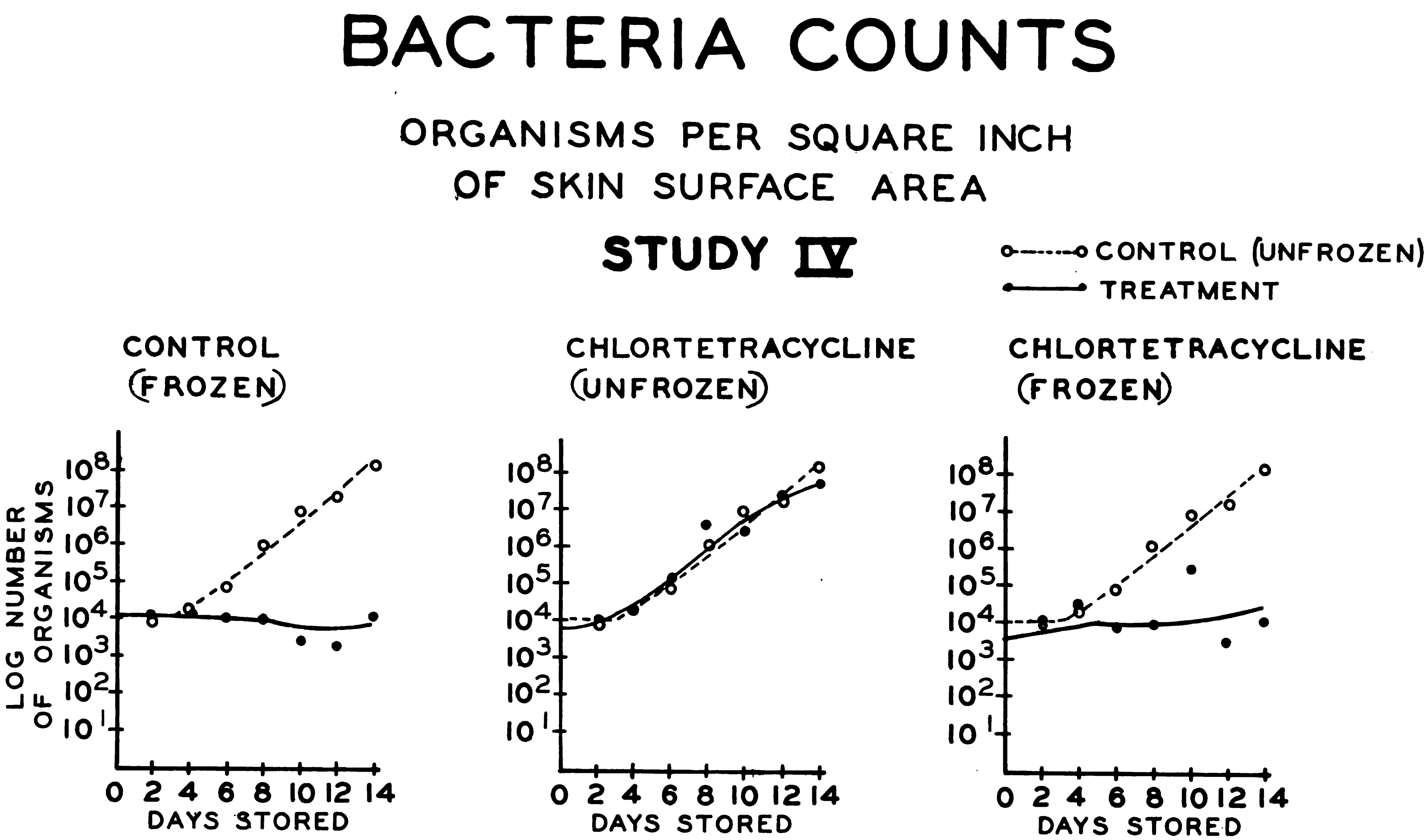


Figure 7. The effect of treatments on total bacteria counts. Counts of organisms were obtained by the swab method. Plates were incubated for three days at 20°C. on tryptone-glucose-extract agar.

BACTERIA COUNTS

ORGANISMS PER SQUARE INCH OF SKIN SURFACE AREA

STUDY III

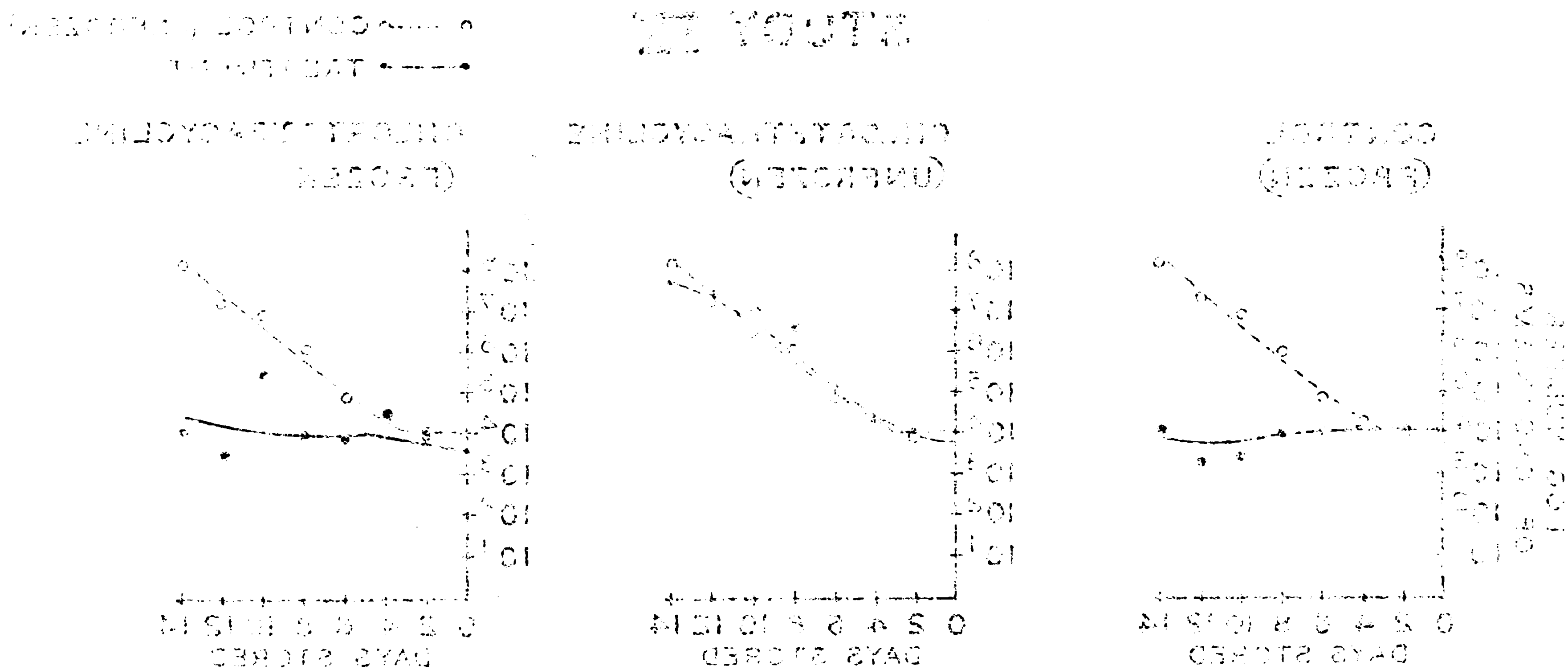


TABLE VIII

A COMPARISON OF TOTAL BACTERIA COUNTS BETWEEN TREATMENTS[1]

Study IV

Storage Time (Days)	TREATMENTS[2] Control (Unfrozen)	Control (Frozen)	Chlortetracycline (Unfrozen)	Chlortetracycline (Frozen)
	Bacterial Counts			
0	1.06×10^4	1.62×10^4	6.77×10^3	6.53×10^3
2	9.17×10^3	1.37×10^4	1.23×10^4	1.10×10^4
4	3.23×10^4	2.27×10^4	2.70×10^4	5.67×10^4
6	8.53×10^4	1.16×10^4	1.82×10^5	9.50×10^3
8	1.36×10^6	1.00×10^4	6.45×10^6	1.00×10^4
10	9.40×10^6	4.30×10^3	4.05×10^6	4.87×10^5
12	3.56×10^7	3.00×10^3	3.96×10^7	5.30×10^3
14	1.98×10^8	1.50×10^4	6.90×10^7	1.30×10^4
16	1.84×10^8	5.30×10^3	5.16×10^7	6.13×10^3

[1] Total bacteria counts are expressed in log number of organisms per square inch of skin surface area.

[2] All birds in this study were packaged in polyethelene bags and air cooled.

Raw Odor Panel Data:

No official raw odor panel was conducted during this study. Only dates of rejection were recorded.

Rejection dates as determined by raw odor were consistent with results obtained in Study III. The only difference was that in Study IV both the control and treated birds were declared unacceptable after nine days of storage. In Study III the treated, unpackaged birds were designated unacceptable four days sooner than were the unpackaged controls.

Taste Panel Data:

A ten member taste panel designed to fully test the accuracy and ability of people to detect flavor differences between cooked chicken meat with high and low bacterial contamination, was used in this study.

The results shown in Figure 8 and Table IX reveal that there was little difference, if any, from the standpoint of flavor whether a bird contains a high or low bacteria population within limits of this study.

Figure 8 shows that there were little differences in flavor between frozen and unfrozen control birds. In fact, after 14 days of storage, when the unfrozen bird was considered putrid, the scores for the controls were the same.

Figure 8 and Table IX also show that the treated unfrozen bird, which was considered unacceptable after nine days of storage, actually improved in flavor as determined by this

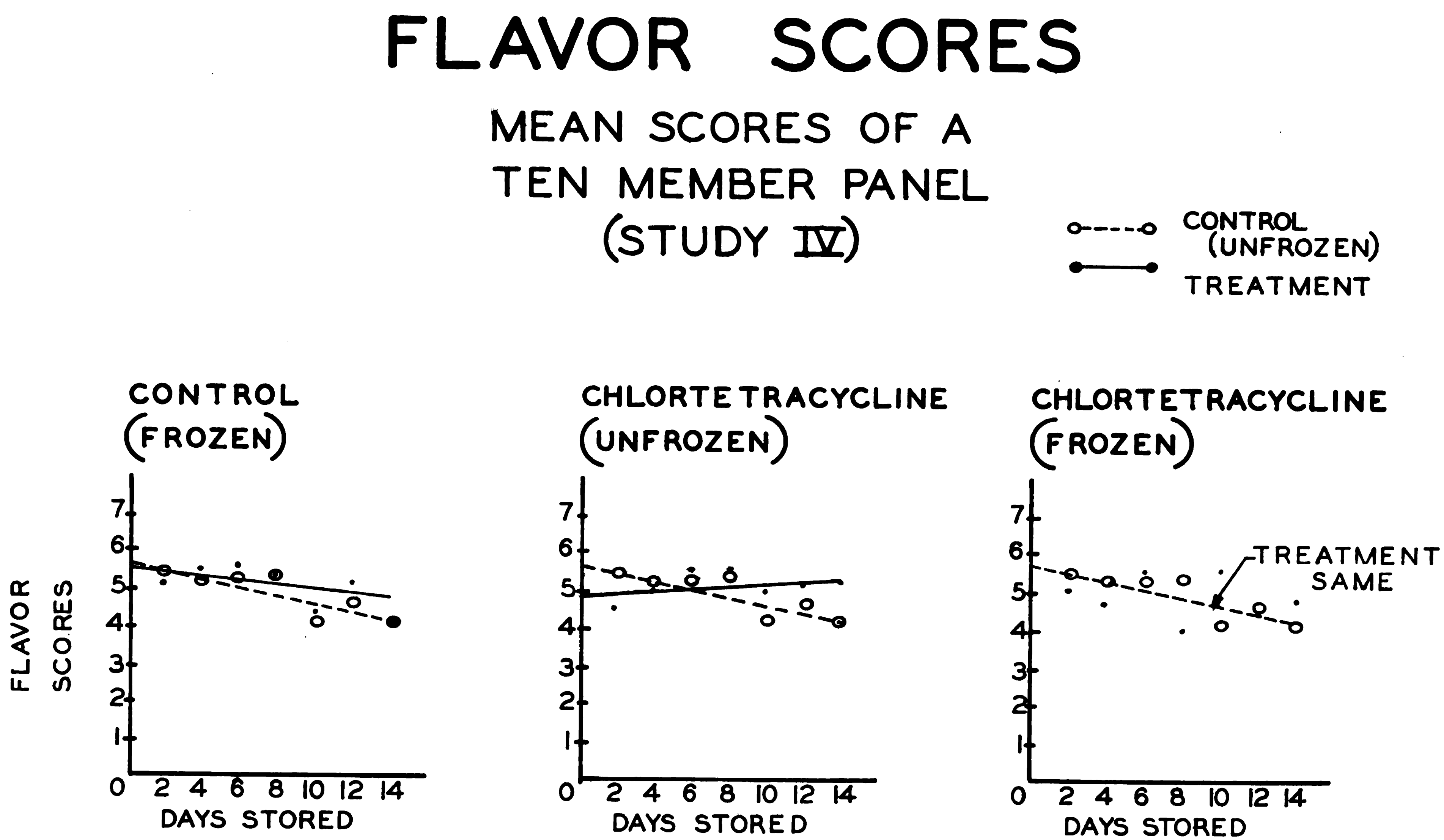


Figure 8. The effect of treatments on flavor scores. A scoring range of 1-7 was used with 1 meaning very poor and 7 meaning excellent.

FLAVOR SCORES

MEAN SCORES OF A
TEN MEMBER PANEL
(STUDY II)

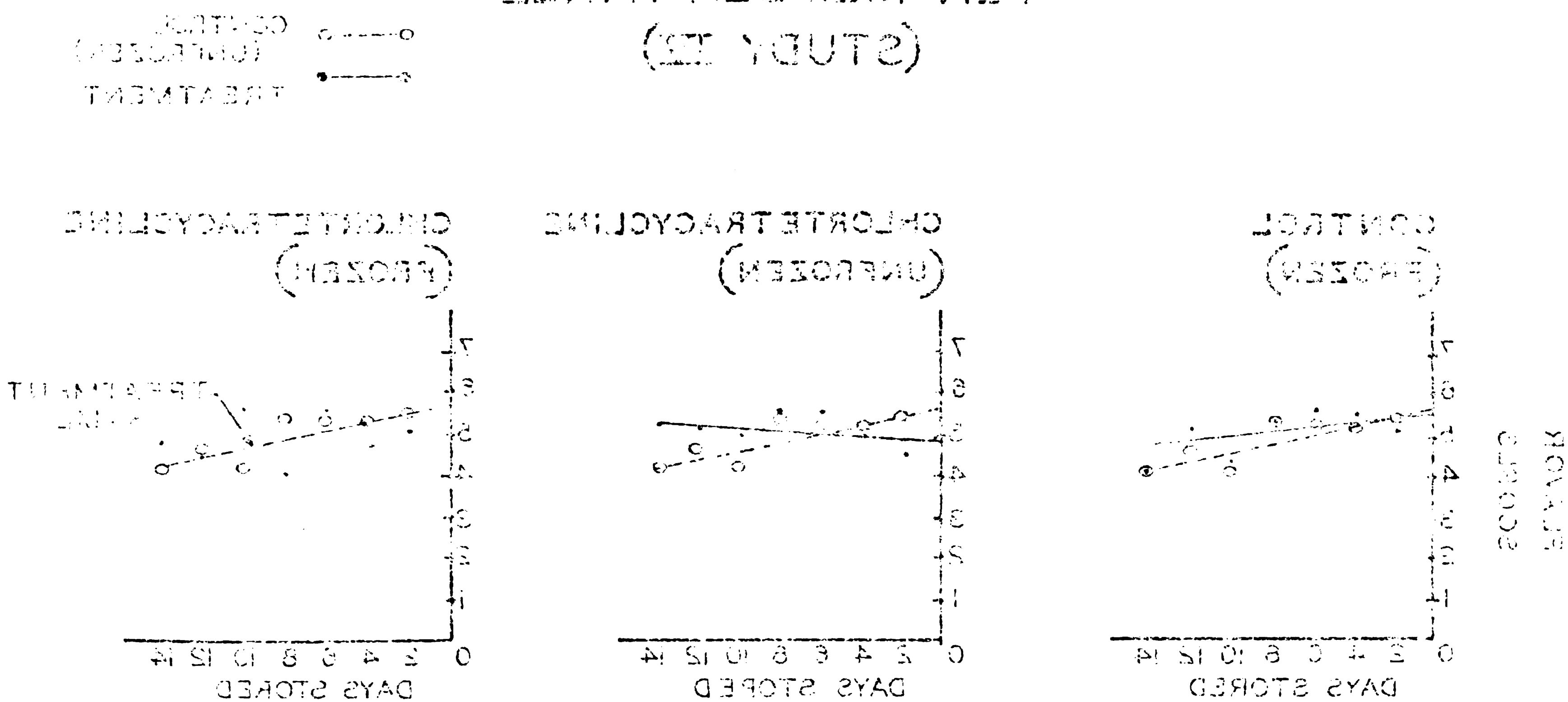


TABLE IX

A COMPARISON OF FLAVOR SCORES BETWEEN TREATMENTS

Mean Scores of a Ten-Member Panel

Study IV

Storage Time Days	TREATMENTS: Control Unfrozen			Control Frozen			Chlortetracycline Unfrozen			Chlortetracycline Frozen		
	Breast	Thigh	Both	Breast	Thigh	Both	Breast	Thigh	Both	Breast	Thigh	Both
	Mean Panel Scores For The Various Treatments[1]											
2	5.50	5.50	5.50	5.30	5.30	5.30	4.60	4.60	4.60	5.10	5.10	5.10
4	5.80	4.80	5.15	5.50	5.50	5.50	5.80	4.30	5.05	5.50	4.00	4.75
6	5.60	5.10	5.35	5.80	5.40	5.60	6.00	5.20	5.60	5.80	5.20	5.50
8	5.20	5.60	5.40	5.20	5.60	5.40	5.50	5.60	5.55	4.20	3.80	4.00
10	4.00	4.40	4.20	5.20	3.60	4.40	4.80	5.20	5.00	6.00	5.20	5.60
12	5.00	4.40	4.70	6.00	4.20	5.10	6.50	4.00	5.25	5.30	4.00	4.65
14	4.00	4.40	4.20	4.60	4.80	4.70	5.00	5.60	5.30	5.00	4.60	4.80
16				5.10	4.50	4.80				4.70	4.60	4.65

[1] A scoring range of 1-7 was used, with 1 meaning very poor and 7 meaning excellent.

heterogeneous taste panel. The flavor score attained by the treated, unfrozen bird after two days of storage was 4.60 compared to 5.30 after 14 days of storage. These birds would have also been considered unacceptable after nine days of storage and considered putrid after 14 days of storage.

Figure 8 also shows that there were only minor flavor differences between birds with high and low bacterial counts. In this case the regression lines for the frozen treated bird and the unfrozen control are identical.

After 16 days of storage it was decided to continue the taste panel, but only supply the panel members with cooked samples of frozen, low bacteria count chicken meat which had been thawed the evening before cooking. On the 16th day after processing the scores were practically identical with those obtained on the 14th day for unfrozen birds. No noticeable differences were detected between thigh and breast meat.

A Comparison Of Laboratory and Commercial Chlortetracycline Treatments

Bacteriological Data:

A comparison of results obtained from chlortetracycline-treated birds prepared in both laboratory and a commercial processing plant revealed the importance of sanitation. Figure 9 reveals that the commercially treated birds had a higher initial bacteria count by almost one log. After

BACTERIA COUNTS

ORGANISMS PER SQUARE INCH
OF SKIN SURFACE AREA

CHLORTETRACYCLINE TREATMENTS

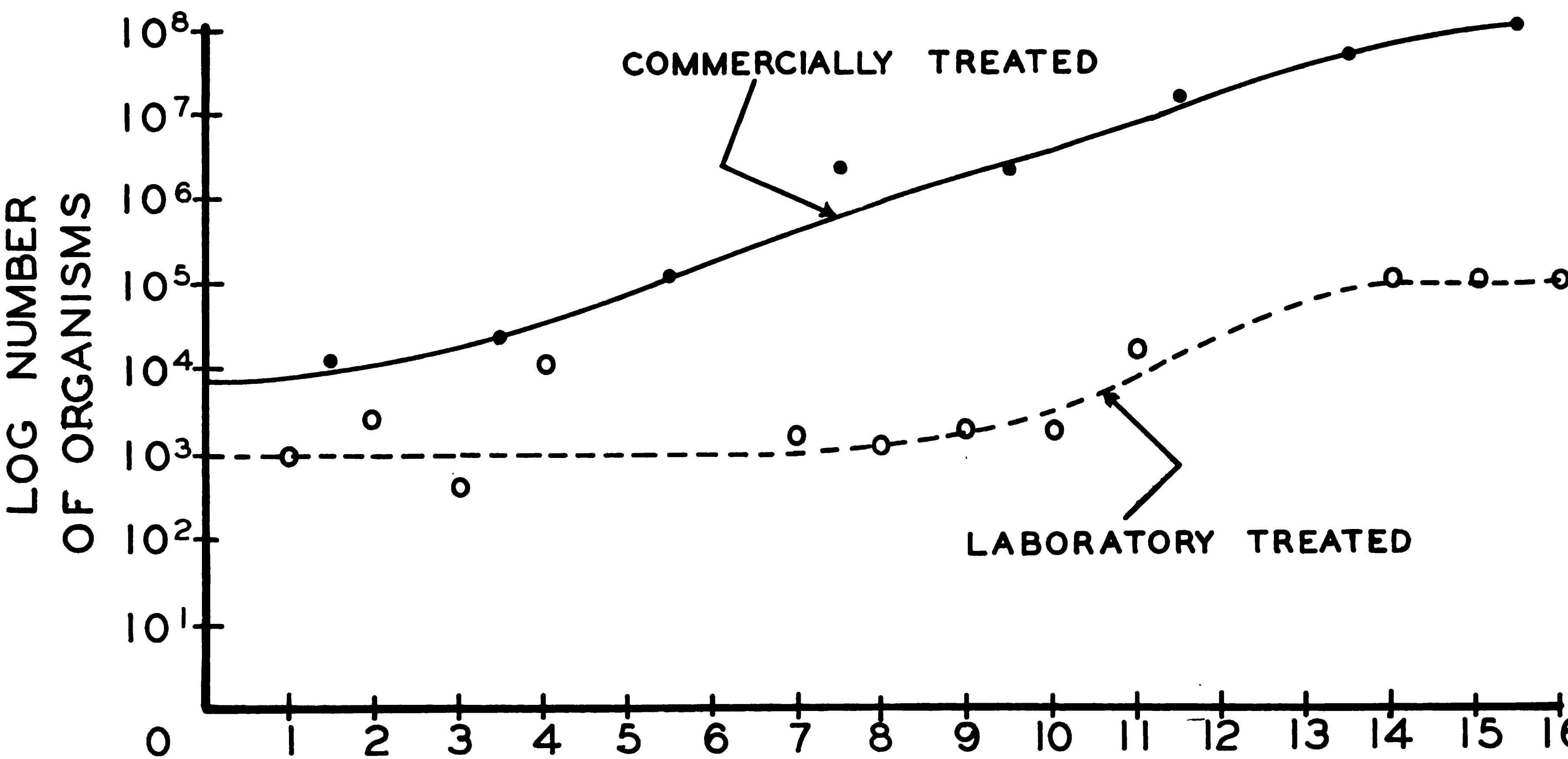
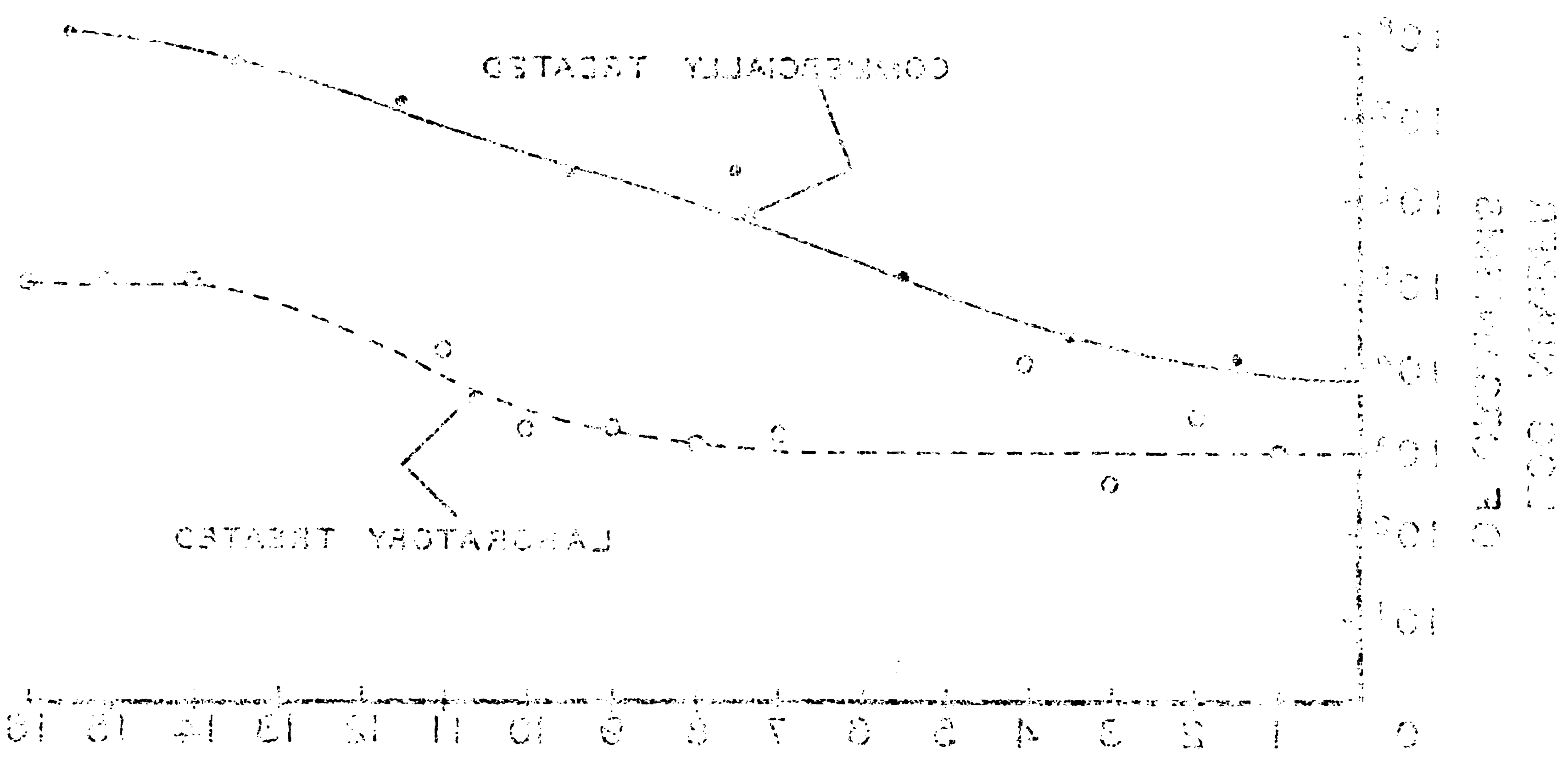


Figure 9

BACTERIA COUNTS

ORGANISMS PER SQUARE INCH
OF SKIN SURFACE AREA
CHLORTETRACYCLINE TREATMENTS



three to four days of storage the bacteria on the commercially treated birds began a steady increase. This is contrasted by the bacteria on laboratory prepared birds which did not increase rapidly until after 10 to 11 days of storage. Commercially treated birds begin to show a marked increase in micro-organisms approximately seven to eight days sooner than did similarly treated laboratory birds.

A comparison of the actual bacteria counts listed in Table X shows that laboratory birds did not reach a bacterial population level of 1.00×10^6 or more until after 16 days of storage. Commercially treated birds reached this level after only eight days of storage.

Due to the higher initial bacterial contamination on the commercially treated birds, it is concluded that this fact is responsible for the faster deterioration and spoilage as compared to laboratory treated birds. Sanitation apparently could be greatly improved in the plant which supplied the birds for this study.

Raw Odor Panel Data:

The commercially prepared, chlortetracycline-treated birds started to show marked increases in microbiological growth after three to four days of storage. This observation was substantiated by the raw odor panel findings. After four days of storage the scores of commercially treated birds dropped below those of birds prepared in the laboratory,

TABLE X

LABORATORY VERSUS COMMERCIAL
TREATMENT OF FRESH KILLED
POULTRY MEAT WITH
CHLORTETRACYCLINE

(Total Bacteria Counts)[1]

Storage Time (Days)	Laboratory Treatment	Storage Time (Days)	Commercial Treatment
1	9.65×10^2	1	
2	4.10×10^3	& 2	1.52×10^4
3	6.46×10^2	3	
4	1.24×10^4	& 4	3.69×10^4
		5	
		& 6	1.44×10^5
7	2.31×10^3	7	
8	1.60×10^3	& 8	3.74×10^6
9	3.01×10^3	9	
10	3.00×10^3	& 10	3.82×10^6
11	2.19×10^4	11	
		& 12	2.14×10^7
		13	
14	1.25×10^5	& 14	7.08×10^7
15	1.27×10^5	15	
16	2.12×10^6	& 16	1.28×10^8

[1] Total bacteria counts are expressed in log number of organisms per square inch of skin surface area.

RAW ODOR SCORES

MEAN SCORES OF A FOUR-MEMBER PANEL CHLORTETRACYCLINE TREATMENTS

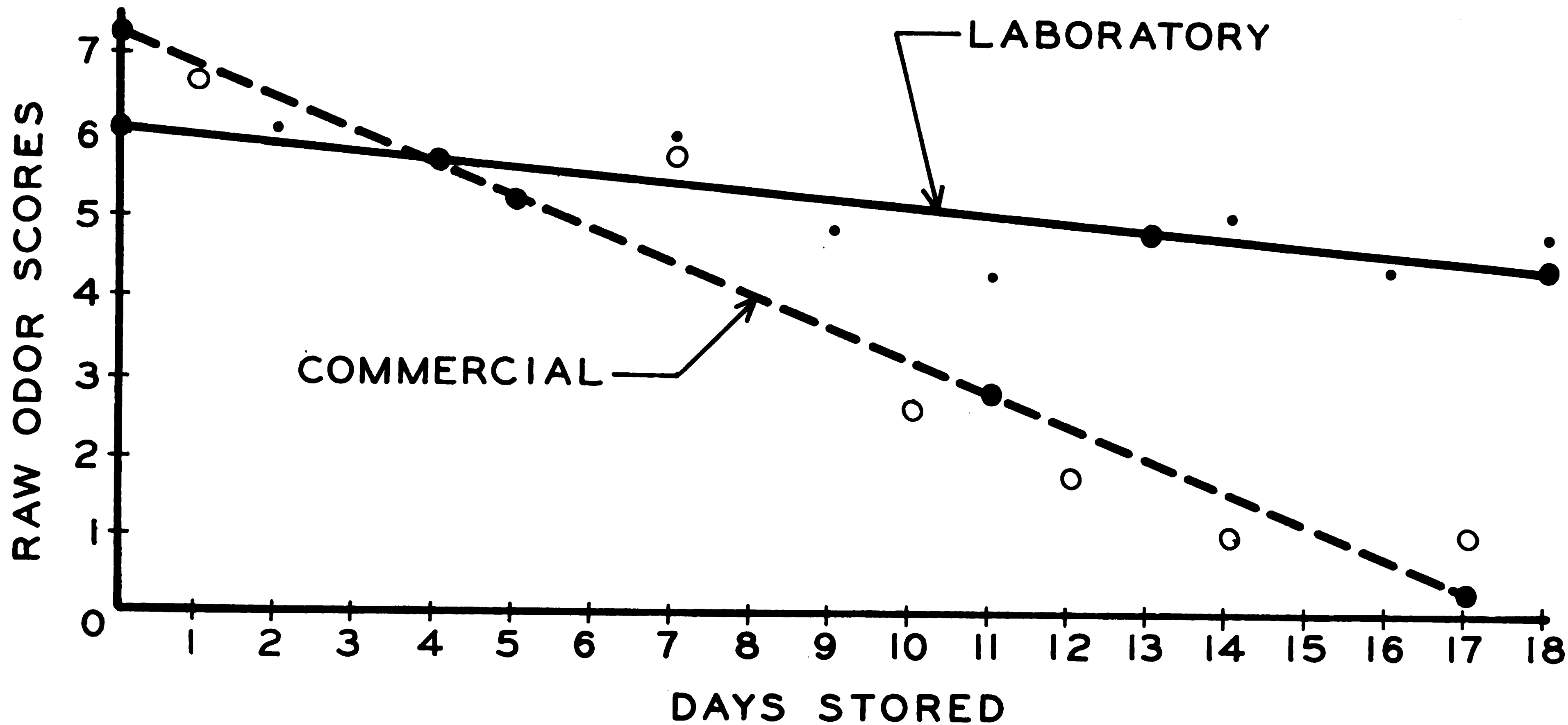


Figure 10

RAW ODOR SCORES

MEAN SCORES OF A
FOUR-MEMBER PANEL
CHLORTETRACYCLINE TREATMENTS

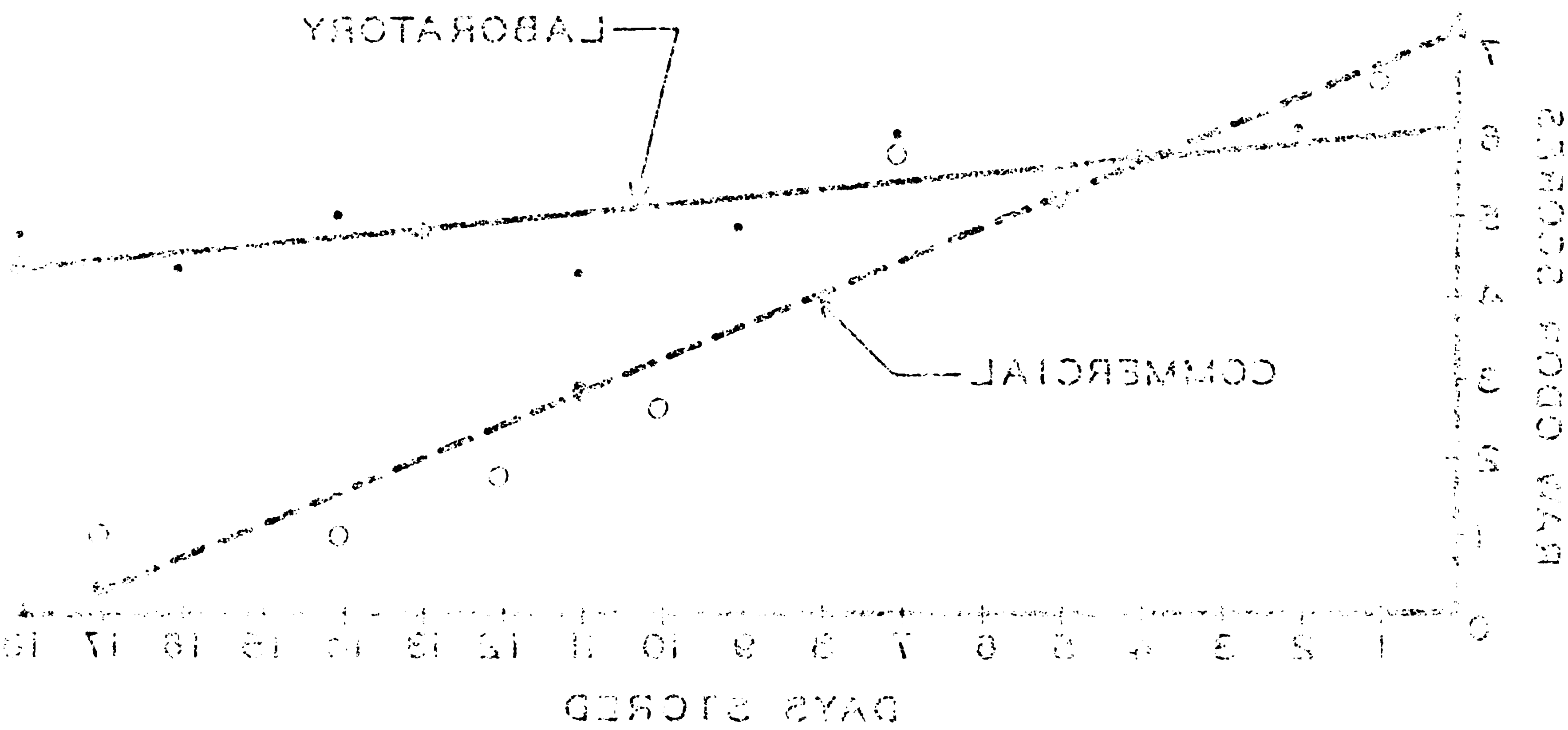


Figure 10. The difference in scores between birds of the two treatments increased with time. This difference is significant at the 0.5 percent level for the entire test period.

Taste Panel Data:

Flavor scores were recorded for each individual study, but the results indicated no basic preference by panel members for either birds with high or low bacteria counts. However, Figure 11 shows a difference between commercial, chlortetracycline-treated birds and similar laboratory treated birds. The differences between the scores are not as important, however, as are the first and last scores for the two groups.

The flavor of laboratory-treated birds was superior to that of commercially treated birds throughout the entire test period. However, the findings of separate studies indicated that people cannot detect noticeable differences between birds with high and low bacteria populations after the meat has been thoroughly cooked.

Commercial Poultry Processing Plant Sanitation Survey

After observing the results of Study III it was decided to visit the commercial processing plant from which the birds were being obtained. Swabs were taken on birds at several

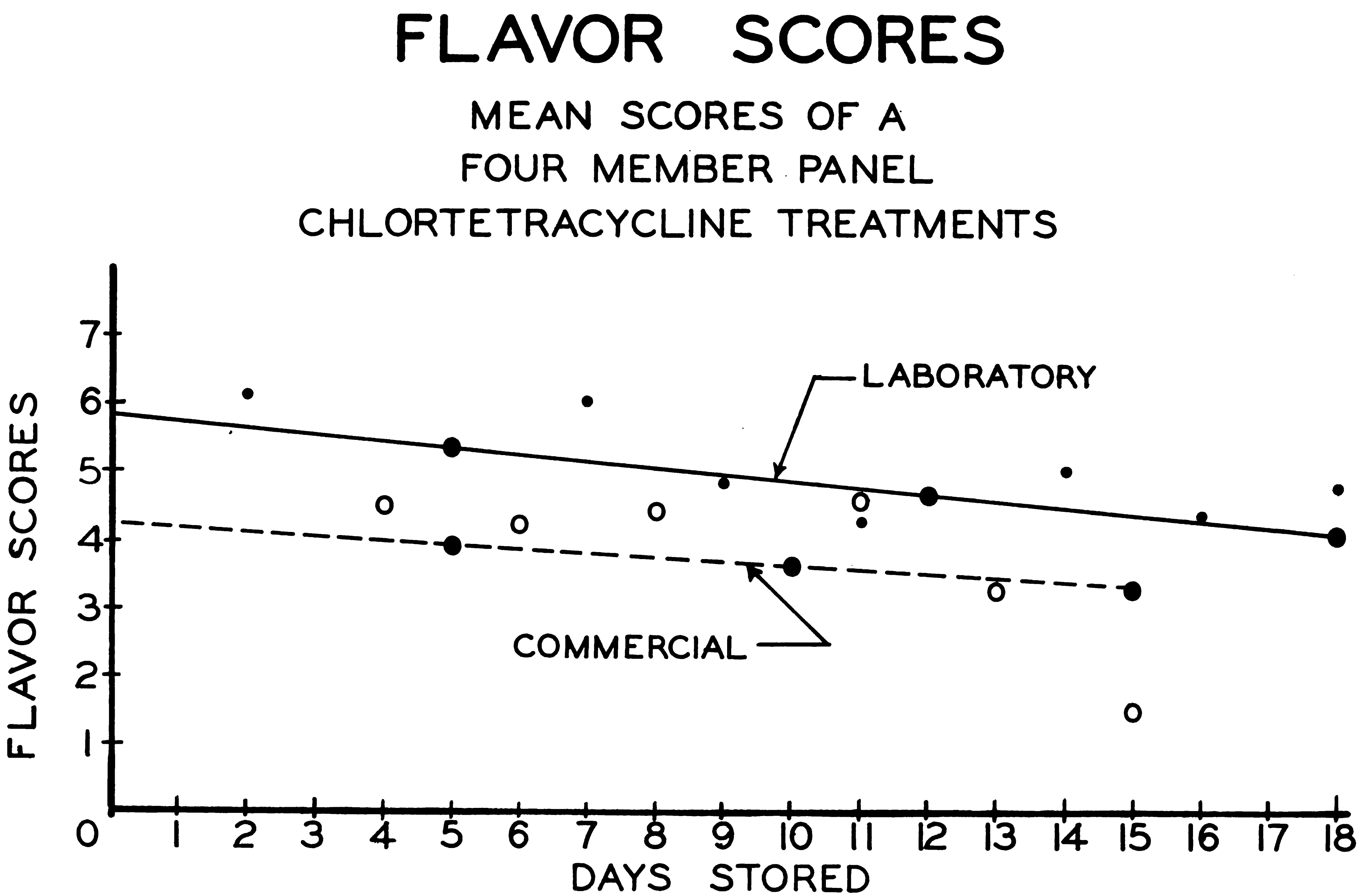
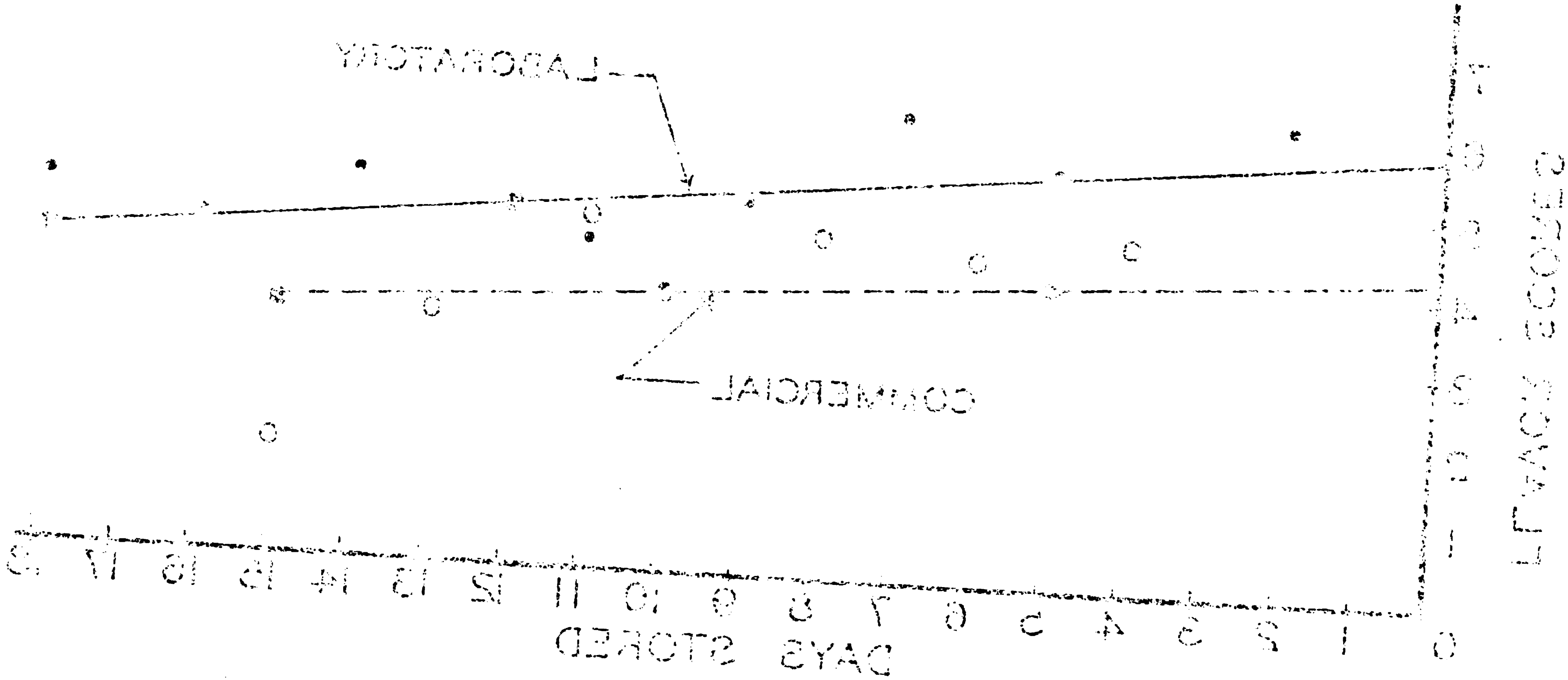

FLAVOR SCORES
MEAN SCORES OF A
FOUR MEMBER PANEL
CHLORTETRACYCLINE TREATMENTS
FLAVOR SCORES
0
1
2
3
4
5
6
7
LABORATORY
COMMERCIAL
0 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18
DAYS STORED

Figure 11

FLAVOR SCORES

MEAN SCORES OF A
FOUR MEMBER PANEL
CHLORTETRACYCLINE TREATMENTS



points on the processing line, and samples of diluted chlortetracycline solutions from the various chill tanks were obtained.

This procedure was followed because it was believed that there was a source of bacterial contamination in this commercial plant which was hindering any useful effects of chlortetracycline.

The results of this bacterial survey illustrated the importance of high sanitary requirements needed in poultry processing plants to insure bacterial control on processed poultry if spoilage of fresh poultry meat is to be deterred. Table XI shows the various locations within the plant which were included in the survey.

The microbiological counts were comparable for birds checked on the processing line and in the chill tanks. A rapid increase in the number of bacterial organisms occurred after the birds were removed from the chill tanks.

The most marked increase in bacterial numbers occurred after the birds had been cut-up and packaged. Therefore, it was probable that this contamination occurred during the cutting-up and packaging process. This hypothesis proved to be at least a partial answer to the contamination problem since the bacterial count on the cut-up table was 1.95 x 10^5 (Table XI).

There were no facilities for running water, hot or cold, on this table and the table was not washed or cleaned

TABLE II

COMMERCIAL POULTRY PROCESSING[1]
PLANT SANITATION SURVEY

Swab Sampling Stations	Bacteria Counts[2] (Organisms per Square Inch of Skin Surface)
No. 1--first wash after singe	4.50×10^4
No. 2--after viscera is removed and before second wash	2.75×10^4
No. 3--after neck is removed	4.25×10^4
No. 4--after second wash	4.75×10^3
No. 5--on weighing pan	2.00×10^4
No. 6--chlortetracycline chill tank #1	2.02×10^4
No. 7--chlortetracycline chill tank #2	1.62×10^4
No. 8--chlortetracycline chill tank #3	2.22×10^4
No. 9--chlortetracycline chill tank #4[3]	5.25×10^3
No.10--chlortetracycline treated pieces of cut-up chicken meat after being placed in boat-shaped packaging boxes	3.00×10^5
No.11--table top where chilled poultry is cut-up for packaging	1.95×10^5

[1] A random selection of birds was obtained and a 4 square inch skin surface area of each bird was swabbed for obtaining bacteria counts.

[2] The plates were incubated at 20°C. for three days.

[3] This tank was only partially filled, and did contain a higher concentration of chlortetracycline at the time of sampling.

TABLE XII

ASSAY OF CHILLING TANK WATER SAMPLES CONTAINING
CHLORTETRACYCLINE OBTAINED AT A
COMMERCIAL POULTRY PROCESSING PLANT

Sample Number	Chlortetracycline Concentration (Parts per Million)	Bacteria Counts (Organisms per milliliter of tank water sample)
1	11.5	4.70×10^4
2	11.5	2.60×10^4
3	7.1	2.90×10^4
4	14.3	1.60×10^4

between different groups of birds which were cut-up and packaged.

Results of an assay to determine the concentration of chlortetracycline in the chill tanks revealed that the concentrations were generally satisfactory (Table XII). When the solution was taken from sample 4, the tank was only half filled with water, thus accounting for the high figure of 14.3 parts per million of chlortetracycline.

The results of this survey are similar to those reported by Clark (1954), Gunderson, Rose and Henn (1947), and Gunderson, Schwartz, and Rose (1955).

Further studies are necessary to fully determine all points of bacterial contamination in this commercial processing plant.

SUMMARY

1. Under laboratory processing conditions chlortetracycline was more effective in extending shelf-life of fresh poultry meat than oxytetracycline or chlorine.
2. Fresh poultry meat, treated commercially with chlortetracycline, deteriorated at a rate significantly higher than poultry receiving the same treatment and processed under laboratory processing conditions.
3. Chlortetracycline-treated commercial birds were considered unacceptable from 7 to 8 days sooner than similarly treated laboratory birds, according to results of a raw odor panel.
4. Fresh poultry meat will generally be rejected for acceptability when bacterial contamination reaches a population of, or near, 1×10^6, according to raw odor determinations.
5. Bacteria counts of chlortetracycline-treated commercial birds reached a bacteria population of 1×10^6 from 7 to 8 days sooner than similarly treated laboratory birds.
6. No apparent relationship exists between bacterial contamination and cooked flavor of breast and thigh chicken meat (without skin), according to results of taste panels.
7. Results of a poultry processing plant sanitation survey showed that bacterial inhibitors are no substitute for

plant sanitation. Unless a rigid sanitation program is employed in this plant, no value can be attained by using chlortetracycline or any other bacterial inhibitor.

8. Fresh chicken meat treated commercially with chlortetracycline in a single processing plant was no more effective in extending shelf-life than untreated birds from the same plant.

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