DESENSITIZATION IN THE TREATMENT OF TUBERCULOUS GUINEA PIGS. J. Durward Thayer, University of Washington, Seattle.

In the present paper, several antigens have been tested for their desensitizing power, and the two which proved most satisfactory have been given more extensive study.

Results. I. (1) Two bacillary antigens, one killed by ether and chloroform and the other by nitrous acid, are found to accomplish more effective desensitization when given in daily decreasing doses than when administered in weekly constant doses.

(2) Bacillary antigens are unsuitable agents for desensitization because of the massive adhesions and sterile abscesses they cause upon injection.

II. An antigen, prepared by extracting tubercle bacilli with glycerol is highly specific and brings about good desensitization.

III. Tuberculin (S.M.T.) prepared by the "Sensitized Vaccine" method of Besredka, renders large doses of tuberculin non-toxic for the sensitive tuberculous guinea pig, but, at the same time, the greater part of the tuberculin desensitizing power is lost.

IV. Tuberculin (S.M.T.) treated with nitrous acid, is apparently so denatured that it no longer serves as a satisfactory desensitizing agent.

V. (1) The tuberculin, T. P. T., is a slightly better antigen than Old Tuberculin for the production of desensitization; smaller amounts being required to bring about an equal degree of desensitization.

(2) Desensitization treatment employing the tuberculins T.P.T. and P.P.D. greatly reduces the skin sensitivity and the general systemic allergy of tuberculous guinea pigs.

(3) The gross and microscopic lesions of guinea pigs treated daily with tuberculin are less extensive than those of the control animals.

THE RAPID PRODUCTION OF TUBERCULIN ON AN INEXPENSIVE SYNTHETIC MEDIUM. Sam Wong, The Alice McDermott Foundation, University of Washington, Seattle.

A comparative study was made to correlate the weight of organisms to the yield of tuberculin in 4 synthetic media at the end of 6 weeks of growth, using Mycobacterium tuberculosis, H 37. It was found that the bulk of organisms was not so essential in the rapid production of tuberculin as was a slightly alkaline terminal pH. There was presented a glycerol-free medium which at the end of 6 weeks contained about half of the weight of organisms yielded by Long and Seibert's medium but yielded 3 times as much tuberculin.
Furthermore, the yield of tuberculin at the end of 6 weeks on this medium was about the same as on Long and Seibert's at 12 weeks. Evidence was presented to show that the yield of tuberculin on the modified Long and Seibert's medium can be increased two-fold merely by readjusting the terminal acid pH of 5.8 to a slightly alkaline pH of 7.2 by means of sterile 10 percent sodium carbonate solution and incubating for 2 more weeks. Glucose was a satisfactory substitute for glycerol in growing H37 under the conditions specified in the report. It was better as a carbon source than glycerol in the production of synthetic medium tuberculin. The tuberculoprotein obtained from the glycerol-free medium was apparently identical with those obtained from other glycerol media; comparative skin tests and systemic tests made on sensitive tuberculous guinea pigs showed no difference in reaction.

**Antigenic Relationships of Some Tubercle Bacilli as Determined by Serological Reactions with Their Natural Tuberculins.** Russell S. Weiser, University of Washington, Seattle.

Natural tuberculins, which are filtered saline extracts of tubercle bacilli obtained by the use of liquid air in the repeated alternate freezing and thawing of tubercle bacilli, were injected intraperitoneally into young incubator-hatched Leghorn pullets. The extracts were derived from virulent avian, human and bovine strains of tubercle bacilli. One week after a series of 11 weekly injections of antigen the birds were bled and the serum tests made for precipitin, cross precipitin and precipitin absorption reactions to all of the natural tuberculins employed. The minimum absorbing dose was determined for each serum and used for absorption studies.

The avian strain could be differentiated from the human and bovine strains by cross precipitin and precipitin absorption reactions, whereas the bovine and human strains could be differentiated from each other only by precipitin absorption reactions with their homologous tuberculins.

The results indicate that tubercle bacilli of human and bovine strains are antigenically similar although not identical and that they differ markedly in this respect from the avian strain.

**Bacteriological Problems of Canning Industry.** J. R. Esty, National Canners Research Laboratory, San Francisco.

**The Thermal Death Time of Clostridium Botulinum Spores at Temperatures and pH Values Commonly Encountered in Home Canning.** W. V. Halversen and Glen L. Hayes, Bacteriology Department, University of Idaho, Moscow, Idaho.

This study presents data showing a comparison of the thermal death time at temperatures corresponding to the boiling point of water at altitudes commonly encountered in Idaho and also at temperatures encountered in the pressure cooker. These data emphasize the extreme exposures necessary to kill the spores at temperatures corresponding to the boiling point of water. It appears obvious that a steam pressure cooker would be necessary for processing vegetables at higher altitudes. In this experiment, at 90°C, which would correspond to the boiling point of water at 9000 feet altitude, the processing time for hominy would be 10 hours, corn 9½ hours, spinach and bean 8½ hours, pears 2½ hours, and prunes 1 hour. These exposures are...
much greater than those recorded in
the commercial time tables where the
rule is to increase the processing time
10 per cent for each additional 500 feet
above the first 1000 feet altitude. We
would therefore conclude that the
recommended time is inadequate.

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<tr>
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Since several outbreaks of botulism have been traced to pears, it seems
logical to recommend that, at altitudes
above sea level pears should be pro-
cessed in the steam pressure cooker.

**The Water Content of Bacterial Spores.** B. S. Henry and C. A. Friedman, University of Washington, Seattle.

The water content of the spores and
vegetative cells of *Bacillus subtilis*,
*Bacillus megatherium* and *Bacillus mycoides* was determined. Using air
dried cells as a basis the moisture con-
tents of the two types of cells were
found to be almost identical. Moist
weight is considered unsatisfactory as
a basis for comparisons of the two
types. It was concluded that differ-
ences in water content in the two types
of cells is not the true explanation of
the differences in heat resistance of
the two forms. The possibility of a differ-
ence in the ratio of free and bound
water in the two cell types is suggested.

**Effect of a Starvation Medium on Bacterial Variations.** Victor Burke, Washington State College.

Water containing 200 mgm. of potas-
sium permanganate and 750 mgm.
potassium hydroxide per liter was dis-
tilled from a pyrex flask. The water
was immediately sterilized. Agar agar
was thoroughly washed in distilled
water and dried. A medium consisting
of 1.5 per cent agar agar was made by
the addition of the distilled water
titrated to pH 7.2 and sterilised. This
starvation medium was inoculated with
washed cells of *Sarcina lutea*,
*Staphylococcus albus*, *Staphylococcus aureus*, *Rhodococcus roseus*, *Escheri-
chia coli* and *Bacillus megatherium*.

With *Sarcina lutea* about one-third
of the plates showed growth in 3–5
weeks. The morphology of the colonies
varied greatly—none were typical
except on two plates where all that
developed were typical. Nearly all
extremes in colony types were noted,
rough, spreading, discrete, G colonies,
daughter colonies, etc. Only two pig-
ments showed up, a lemon yellow char-
acteristic of the stock culture and a
deep orange. Pigments did not appear
on the plates, only on the cultures on
slants taken from the plate. The
colony changes appeared temporary,
the pigment permanent. On all but
two plates the morphology of the cells
and staining reaction varied. Some
colonies consisted of Gram-negative
cocci and Gram-negative bacilli.
Other colonies consisted of Gram-
negative bacilli. In general when
colonies were typical the cells were
typical. When colonies were atypical
the cells were atypical. Fermentation
tests indicated some variations from
normal.

Attempts to bring about reversion
have so far failed. Starting with a
colony consisting wholly of bacilli, no cocci ever appeared on successive slants. However, reversion may occur in some other medium or following some other treatment. If reversion never occurs the bacillus represents a mutation, and if the Gram-negative cocci never revert to the Gram-positive possibly they also represent a mutation.

Comparable results with minor differences were obtained with Staphylococcus aureus. A Gram-negative yellow-pigment-producing bacillus that did not revert to the original type was produced at will. The colonies were all on the surface. Some had a faint lavender shade.

Escherichia coli developed atypical colonies and a yellow pigment. The pigment was retained on potato extract agar. The colonies were all surface colonies. Bacillus megatherium grew with atypical colony formation. Other changes have not been determined. Some organisms failed to grow.

Since double-distilled water is toxic we should consider that single-distilled water is slightly toxic even though growth occurs. Therefore, the variations arising may be due to either of two causes, starvation or toxicity of the water. Distillation leads to a reduction of salts as well as of organic matter. The addition of a small amount of potato juice, containing organic matter and salts, causes the toxicity to disappear.

The change in colony formation on the starvation medium is to be expected since growth is very slow. The other changes are more permanent and of greater significance. The biophysics involved in turning a coccus into a bacillus is obscure. Such a change might conceivably result from the weakening of the cell wall. The fact that the change in staining is toward the Gram-negative may offer support to this view. We have experiments under way to determine whether there is a change in the permeability of the cell wall. If the Gram reaction depends on the permeability of the cell wall as I believe then there must be such a change.


Relationship between Nasal and Humoral Antipoliomyelitic Substances. B. F. Howitt, Hooper Foundation, San Francisco, California.

Based on the work of Amoss and Taylor in 1917 dealing with presence of neutralizing substances in the nasal washings of different individuals, a comparison has been made between the humoral and nasal antipoliomyelitic substances in a group of 61 individuals. This group was divided into 2 groups of about 30 each, one half adults over 25 years of age and the other children varying in age from 2 to 15 years. Both adults and children were subdivided into 3 groups of 10 each, those with apparently normal and with abnormal nasal mucosa, and a third series representing those recovered from poliomyelitis after varying periods of time.

In vitro neutralization tests were positive on 36 (59 per cent) of the serums and 14 (22.9 per cent) of the filtered nasal washings from a total of the 61 individuals examined. No particular correlation could be noted between the presence of positive serums and positive nasal filtrates, although more of the latter occurred among so-called normal adults than among the recovered poliomyelitic cases. Nine individuals gave positive tests in both serums and nasal filtrates, 5 showed
antiviral washings and negative serums, while in 27 the serums were positive and the filtrates negative. About an equal number of positive filtrates were found among those with abnormal nasal conditions as among those with normal. Serums of 71.3 per cent of the normal adults were antiviral as compared to 60 per cent from those recovered from poliomyelitis, while serums from 55 per cent of normal children were positive as compared to 40 per cent from the group of recovered poliomyelitic cases.

Concentration of nasal filtrates by the Flosdorf and Mudd lyophile apparatus was of value in securing more positive neutralization tests. No antiviral substances were found in the nasal washings of 5 immunized monkeys, taken at different stages of immunity. Their tissue immunity failed to run parallel with the humoral response, since 3 animals possessed neutralizing antibodies in the serum without developing resistance to intracerebral inoculation of virus.

**Antibody and Immunity in Poliomyelitis.** J. F. Kessel, R. T. Fisk and F. Stimpert, University of Southern California.

**Comparative Study of Two Strains of Rocky Mountain Spotted Fever Virus with Special Reference to the Weil-Felix Reaction.** G. E. Davis, U. S. P. H. S. Rocky Mountain Spotted Fever Laboratory.

**A Study of Several Factors Affecting Alcohol Formation by Saccharomyces Ellipsoideus.** Leonora Hohl and W. V. Cruess, University of California.

The so-called syrumped method of fermentation first described by Cruess, Brown and Flossfeder (Jour. Ind. Eng. Chem., 8, 1124, 1916) was used in securing the data reported. Of the factors studied temperature proved the most important; at 7–20°C. approximately 16.5 per cent alcohol by volume was attained; at 25°C., 13.8 per cent and at 30°C., only 6.4 per cent.

Grape concentrate proved more suitable than other materials used as substrates for pure sugars compared in respect to maximum alcohol attained.

Of the various fruit and vegetable juices compared, tomato and grapefruit juices attained the highest alcohol content.

Various yeast foods and vitamin concentrates were added at the time of syrumping in several experiments; of these di-ammonium phosphate gave the greatest increase in final alcohol content. Vitamin C appeared to have a positive effect also. Several small additions of grape concentrate gave higher final alcohol content than a single large addition.

The evidence indicates that some substance or substances other than sugar alone also influences the remarkably high alcohol yields observed in this method of fermentation.

**Hemolytic Streptococci in Market Milk.** J. E. Simmons, Oregon State College.

One hundred and one cultures, which produced definite evidences of hemolysis, were isolated from samples of market milk. Forty-one of these proved to be streptococci occurring in chains having more than ten cells. None of these cultures fermented inulin or raffinose while all were able to ferment glucose, lactose and sucrose. The ability to ferment salicin and mannitol was variable. Thirteen of the cultures gave complete test tube hemolysis in two hours at 37°C. All cultures produced a pH of 4.6–5.0 in glucose.
broth. All hydrolyzed sodium hippurate and none showed any capsules. Most of the organisms could be placed in two large groups having the general characteristics of Streptococcus mastitidis and Streptococcus infrequens. Serological and bacteriophage studies are being carried out to study further the relationship of these streptococci.


Occurrence and Significance of Lactobacillus thermophilus in Pasteurized Milk. D. B. Charlton, Oregon State College.

Microorganisms and Phytamins in Relation to Plant Growth. W. B. Bollen, Oregon State College.


Anaerobic Bacteria Associated with Experimental Pulmonary Abscess. Charles Weiss, Mount Zion Research Laboratories, San Francisco.

Methods of Chemical Analysis of Staphylococcus aureus. Rachel E. Hofstadt and Wesley M. Clark, University of Washington.

Since the dissociation in this laboratory (1932) of Staphylococcus aureus into eleven rough and one G or gonial strain, three studies have been undertaken in order to elucidate the organisms' antigenic structure:

First, an agglutination study showed that each has a distinct antigenic mosaic. Second, non-antigenic soluble specific substances, polysaccharide in nature, were isolated from the smooth and a rough variant and were found to differ widely with regard to their precipitin reactions both when adsorbed and un-adsorbed on collodion.

Third, fractionation of the proteins of the smooth and a rough strain has been completed. One hundred grams of each organism were fractionated at different pH values. The purified and dried materials are being analyzed (1) chemically to identify the proteins, (2) serologically against the whole organisms and against sera prepared with the smooth and rough polysaccharide substances.

It is hoped to correlate differences found in chemical structure with antigenic and biochemical changes observed during dissociation.


Influence of Peptone Concentration on the Metabolic Activities of Escherichia coli. C. E. Clifton, Department of Bacteriology and Experimental Pathology, Stanford University, California.

The rates of oxygen consumption and of carbon dioxide production were determined in cultures of Escherichia coli and related organisms at different periods of growth in 1.0, 5.0 and 10.0 per cent peptone solutions. Evidence is presented which indicates that the maximum metabolic rate per cell observed late in the lag period of growth is due to increased cell size and to a high concentration gradient of food-stuffs between the cells and their
environment during this period. The observed decrease in the rate of metabolic activity per cell observed as the age of the cultures further increases appears to be primarily controlled by the decreased concentration of food-stuff available per cell, the same factor apparently determining the maximum population developed in a culture.

LONGEVITY OF TYPHOID BACILLI IN THE SOIL. P. J. Beard, Stanford University.

STUDIES ON HODGKINS DISEASE. B. J. Olson, H. Donlop and M. Manson, University of Minnesota.

STUDIES ON THE ANTIBODY RESPONSE IN INFECTIOUS MONONUCLEOSIS. Sidney Raffel and G. Howard Bailey, Johns Hopkins University. (Introduced by P. J. Beard.)

OBSERVATIONS ON MELITENESIS INFECTIONS IN THE SOUTHWEST. K. F. Meyer, George William, Hooper Foundation.

PHYSICAL EFFECTS OF AGARS USED IN CULTURE MEDIA. C. S. Mudge, Division of Dairy Industry, University of California.

THE EFFECTS OF 2-4 DINITROPHENOL ON THE GROWTH AND RATE OF METABOLISM OF YEAST. A. Martin, Stanford University.