

The tubes which were held at 55°C. had an original pH of 6.8 and a bacterial count of 4,000,000 per gm. At the end of 15 days, the count was reduced to 100 per gm. with a pH of 4.8. Further incubation at the same temperature for 4 or more days killed all of the organisms with practically no change in pH. A similar experiment was carried out with No. 2 cans of corn. These cans were proven to be free from viable spoilage organisms before their use. They were then opened aseptically and inoculated with spores of organism No. 1518. Before inoculation, the cans were thoroughly heated in the Arnold and a small opening made, through which the spore suspension was introduced. There were 3,000,000 viable spores per cubic centimeter in the suspension. The cans were then incubated at 55°C., one can being opened each week over a considerable period of time. No viable organisms could be demonstrated in the contents of the cans after 28 days, when the hydrogen ion concentration had reached 4.8. The rest of the cans in the series were then opened and similar results secured. It was found that the reaction of the inoculated incubated corn became acid very rapidly, and reached its maximum at the end of 2 weeks. In another experiment, viable cells were absent after 9 days. The pH was as reported above.

A few experiments with an organism producing putrefactive spoilage of corn did not indicate that autosterilization occurred. The optimum temperature of this organism was 37°C.

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### Penetration of Intestinal Wall of Rabbit by Yeasts.

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Casagrandi<sup>1</sup> early concluded that blastomycetes seldom penetrate the mucous layers of the intestinal wall. Positive results were explained on the basis of existing injuries which escaped detection. Fisher,<sup>2</sup> however, recently found that yeast fed fasting dogs is absorbed like bacteria into the portal circulation. Cells injected into the portal vein also proved that yeasts can invade all the organs, the distribution varying with the amount injected.

<sup>1</sup> Casagrandi, O., *Annali d'Igiene sperimentale*, 8. Reviewed in *Cent. Bakt.*, 1908, 1, Ref. 24, 758.

<sup>2</sup> Fisher, Virginia, *PROC. SOC. EXP. BIOL. AND MED.*, 1931, 28, 948.

In this study the blood of 30 rabbits, daily fed large doses of viable yeast cells, was frequently cultured. The heart's blood, bile, the spinal cord, and portions of certain of the vital organs including the liver, lungs, spleen and kidneys were cultured upon autopsy. Blood sera of the animals were also periodically examined for the presence of agglutinins.

Before feeding the blood of experimental and control animals was cultured and found to be free from yeasts. The yeasts isolated from the blood and organ cultures were compared with the strain which each animal received.

The following varieties of yeast were used: *Saccharomyces carlsbergensis*, *Saccharomyces cerevisiae*, *Saccharomyces glutinis*, *Saccharomyces turbidans*, *Endomyces albicans*, *Monilia tropicalis*, *Torula cremoris*, Hunter's yeast isolated from oysters, a commercial pressed yeast, *Saccharomyces ellipsoidens*, *Willia saturnus*, *Zygosaccharomyces mandshuicus*, *Monilia albicans*, *Monilia candida*, *Mycoderma cerevisiae*, *Oidium albicans*, *Torula hansen*, *Torulopsis sanguinia*, and yeast isolated from a sore throat. The first 9 species listed were recovered from the blood after feeding. All, with the exception of a commercial pressed yeast were taken from stock cultures in the laboratory. Suspensions of the commercial yeast were prepared by dissolving a 15 gm. cake in 50 cc. saline, 4 to 6 cc. of which were given certain of the rabbits daily. The remaining animals were fed saline suspensions of yeasts grown on dextrose agar slants at room temperature. No attempt was made to feed exactly the same number of cells or cells of the same age each day, the primary purpose being to keep the alimentary tract of each animal heavily seeded with yeasts.

The animals were bled regularly from the marginal vein of the ear. Bleeding from the ear was found to be much more satisfactory than from the heart.

Macroscopic agglutinations were made with sera dilutions of 1-10, 1-20, 1-40, 1-80, 1-160, 1-320, and 1-640. Suspensions of the cells of 48 hr. cultures (room temperature) in 10 cc. of saline were used as antigen. The tubes were observed frequently after being held an hour at 37°C. and were reported as positive when showing clumps and flakes suspended in the serum and saline. It was found that if all tubes were completely inverted before reading positive results were more easily detected. The precipitate in the control would spread uniformly through the tube while the clumps in positive ones would remain suspended in the liquid. Seven out of 21 animals showed the presence of agglutinins, at the end of

the first month, 8 out of 17 at the end of the second month, and 11 out of 16 fed for 3 months. Maximum agglutination (1-320) was ordinarily demonstrated within 3 or 4 hours although reactions could be read when tubes were held overnight.

Portions of the serum obtained before and during feeding were inoculated with the various yeasts fed and held at body and at room temperatures. In this study blood serum was never found to exert noticeable effect upon the yeast fed when held at room temperature. Some yeasts, however, failed to develop as well as others upon transplanting to dextrose agar slants after being held in fresh blood serum at 37°C. for a period of 48 hours. That was not due entirely to any direct action of the serum itself, nevertheless, as the same yeasts also failed to develop readily when inoculated on dextrose agar slants and held at that temperature.

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**The pH and the CO<sub>2</sub> Content of Cerebrospinal Fluid in Epilepsy.\***

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Although considerable work has been done on the acid-base relationships in epilepsy, the evidence presented thus far has been conflicting and indecisive. Because of the importance of this phase of the problem, we have determined the pH and CO<sub>2</sub> content of the cerebrospinal fluid in 125 epileptics and 30 non-epileptic control subjects by procedures which eliminate errors due to loss of CO<sub>2</sub>.<sup>1, 2</sup>

The colorimetric method of McQuarrie and Shohl<sup>1</sup> used for determining the pH may be briefly described as follows: The clear fluid is drawn into a special glass sampling burette over clean mercury after all air in the connecting tubes has been eliminated by means of a 3-way stop-cock. In preparation for taking the sample a number of tenths of a cc. of 0.0075% phenol red equal to the number of cc. of fluid to be taken is measured into the appara-

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<sup>1</sup> McQuarrie, Irvine, and Shohl, A. T., *J. Biol. Chem.*, 1925, **66**, 367.

<sup>2</sup> Van Slyke, D. D., and Neill, J. M., *J. Biol. Chem.*, 1924, **61**, 523.