periment. The fact that the temperature of virus preparations containing much extraneous matter rose about 12° C. higher than the temperature of purified virus preparations indicates that the former absorb much more energy.

Immediately after each experiment the irradiated preparations and the corresponding control preparations were tested on half-leaves of *Nicotiana glutinosa* and *Phaseolus vulgaris* L. Dilutions of infectious juice were made with distilled water and dilutions of purified virus were made with 0.1 M phosphate at pH 7. The average of the actual number of lesions per half-leaf obtained on the two species of test plants at three different dilutions of the various preparations of irradiated virus is given in Table I. Another number which represents the average of the lesions obtained with an irradiated virus sample, expressed as a percentage of the average number of lesions obtained with the corresponding control, is also given for each preparation at each dilution. This number indicates the amount of virus present.

It may be seen from Table I that virus prepared as described by Takahashi and Christensen is almost completely inactivated at atmospheric pressure. This is in complete accord with their results. Purified virus diluted with 9 parts of untreated juice from healthy tobacco plants is also almost completely inactivated. However, if infectious juice is sealed under a high vacuum in order to prevent cavitation and then irradiated there is but slight inactivation. Purified virus at atmospheric pressure is inactivated only to such an extent that it gives about 60 per cent. as many lesions as the untreated control, while there is practically no inactivation when purified virus is irradiated under a high vacuum. No reactivation of virus which had been inactivated by supersonic radiation at atmospheric pressure was found. Materials toxic to virus or to test plant are not produced during the irradiation since the addition of such inactivated virus preparation to fresh virus has no effect on the infectivity of the active virus. In general, the results are similar to those of previous workers[7] who have found that cells or particles affected by supersonic waves at atmospheric pressure were unaffected under a high vacuum.

The results indicate that inactivation of virus by supersonic radiation is associated with cavitation of dissolved gas and with the presence of extraneous matter found in untreated juice, since high frequency sound waves of great intensity have practically no effect on purified virus under a high vacuum.

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[7] *Ibid.*

GROWTH OF YEAST BELOW ZERO

THOUGH the growth of "false" yeast, as Torula species, below 0° C. has been recorded,[1,2] probably most bacteriologists incline to the belief that the minimum temperature requirements of "true" yeasts, or forms which sporulate and reproduce by budding, are above the freezing point of water. That this is not true of all representatives of the genus Saccharomyces has been proved in this laboratory in connection with the storage of cider at low temperatures. On two occasions about half the number of 500 cc portions of fresh cider in sealed containers held at −2.2° C. (28° F.) developed pressures of 15 or more pounds per square inch after 2 months storage. Such samples contained many millions of yeast cells per cc and were unmistakably alcoholic.

Of several strains of yeasts isolated from the fermented cider, one in particular has shown ability to increase at −2.2° C. Beer wort, pH 4.8, has been used as the medium, and the rise in number of cells has been followed in the haemocytometer from small samples of the well-agitated culture aseptically withdrawn at about 5-day intervals. Counts at 20, 40, 60 and 70 days are given in Table 1.

TABLE 1

GROWTH OF COLD-TOLERANT YEAST IN WORT AT −2.2° C.

Cells per cu cm				
At inoculation	After 20 days	After 40 days	After 60 days	After 70 days
1,800,000	3,840,000	17,600,000	48,400,000	49,000,000

The viable cell count on wort agar on the 66th day was 32,500,000 per cu cm.

Reproduction was most active from about the 20th to the 50th day, and as shown in the table largely ceased after the 60th day. In a portion of the culture transferred from −2.2° C. to 21° C. on the 66th day, however, reproduction was resumed, the haemocytometer count rising to 83,200,000 per cu cm in 42 hours.

The yeast in question ferments dextrose, levulose and sucrose, but not maltose or lactose. Ascospores are present in 2- to 4-week old thin wort agar plate cultures. Despite its ability to grow at −2.2° C. the yeast is not "psychrophilic" or cold-loving, as the 60-day count at the temperature mentioned is equaled in 23 hours at an incubation temperature of 21° C.

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[1] R. B. Haines, Report of the Food Investigation Board (Gt. Brit.) for the year 1931. 46–51.

[2] J. A. Berry, "How Freezing Affects Microbial Growth," *Food Industries*, 4: 6, 205, 1932.