

SACCHAROMYCES ANOMALUS HANSEN(?)

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In his investigations upon spore formation in yeasts Hansen found three typical groups, these three differing either in the form of the spores or in their mode of germination. The third type is furnished by *S. anomalus*, the spores of which are hemispherical in form, having a projecting rim around the flattened surface. The other two types are spherical in form, but differ in their mode of germination. Hansen found this species in an impure Bavarian brewery yeast, and after he had called attention to the peculiar spores, they were also observed by Holm, Lindner and Will,¹ who likewise found them in impure brewery yeast. These investigators did not determine whether the peculiar spores which they observed belonged to Hansen's *S. anomalus* or to allied species. Bay² gives the habitat as impure brewery yeast and grapes, as does Kayser.³

Hansen's⁴ description of the yeast is very brief, as is also the description by Jørgensen⁵ and that given in Saccardo⁶ so that the form of the spores is the most characteristic feature.

A yeast which develops the same form of spores, I found associated with another yeast on the skin of lemon. This is a new habitat for the yeast, and I can find no mention of any such yeast occurring in this country.

The cells are round or oval, occurring either singly or in pairs, though occasionally in a vigorous growth in wort colonies consisting of many cells are found (photograph 1). The cells are very small, measuring 2.4μ in breadth by 3.3μ in length for the small oval ones to 6.6μ in diameter for the cells containing spores (seen in photograph 2).

In wort gelatine plate cultures the colonies are of a dull, grayish white color, round, with rather even outline. They grow rather slowly under the best conditions. The photograph (3) shows a forty-eight-hours' growth. The wort gelatine plates seem to offer better conditions for the

¹ Jørgensen, A. *Micro-Organisms and Fermentation*, 1893, p. 182.

² Bay, J. C. *Am. Naturalist*, Vol. XXVII, 1893, pp. 685-696.

³ Kayser, E. *Les Levures*, p. 104.

⁴ Hansen, E. C. *Central. f. Bakt.*, Bd. XIII, 1893, pp. 101-102.

⁵ L. c., pp. 181-182.

⁶ Saccardo, P. A. *Sylloge Fungorum*, Vol. VIII, pp. 216-228.

formation of spores than any of the other cultures, spores being found in four days at room temperature.

In both wort and beef broth gelatine tube cultures the growth is practically the same. The growth has a dry appearance and forms a dense mass at the needle puncture, without any tendency to spread. The growth tapers gradually from the surface along the needle track until at the bottom of the tube it is just perceptible. It has a characteristic crinkly appearance along its whole length.

Cultures in Pasteur solution, with 5 per cent. sucrose, lactose and dextrose, were made in fermentation tubes, and also in wort which contains maltose. In sucrose no fermentation occurred; there was a heavy growth, however, which caused a strong turbidity of the liquid in the bulb and a heavy sediment; the liquid in the tube remained clear. A film was formed which extended up the sides of the tube. After five days no spores were found either in the film nor in the sediment. Only a very slight growth occurred in the lactose solution, this forming a delicate film and a slight sediment. No gas was formed. The cells in lactose occur singly or in pairs and appear poorly nurtured (photograph 4). In the dextrose solution the growth was vigorous, forming a heavy sediment. Fermentation commenced in four days, and in twenty-four hours 5 cc. of gas were formed. In this, as in the sucrose solution, a strong film was formed. No spores were found in the film nor in the sediment even after seven days' growth. In the wort the fermentation was much more vigorous than in the dextrose, 10 cc. of gas being formed in the time that only 5 cc. were formed in the dextrose solution. Even before fermentation ceased a film formed in spots on the surface. In the wort a delicate ethereal odor is generated, which is very pleasant. Spores were just beginning to form in the film and sediment in eleven days.

Spores formed more readily in a wort gelatine plate culture than even in the regulation manner on a gypsum block. In the plate they formed invariably in four days, not to any great extent, but sufficient to be found in every microscopic examination. The cells in which the spores form are large, and just before the formation the protoplasm becomes very granular and refractive. As the culture ages and more spores are formed they are found free from the cell wall and in groups ranging from two to fourteen in number. Spores can be seen in photographs 5 and 6.

It is not quite certain whether this is the same species as Hansen's S.

anomalus, for the prominent characteristics in the description of that yeast are the peculiar form of the spores and their ready formation in both the film and sediment during fermentation. The shape of the spores in the yeast described is the same as that of Hansen's, but the earliest period at which spores were formed in fermenting wort was eleven days, and then very sparingly. In the other solutions used, no spores were formed even after three weeks' growth. Then no mention is made of their rapid formation in wort gelatine plates, and yet they form there very readily.

Taking into consideration the characteristics which are described by Hansen for *S. anomalus*—spore formation, size and shape of cells, and odor generated during fermentation—and comparing them with those of the yeast described, they agree fairly closely, but the characteristics which are not noticed seem to be sufficiently prominent to not have escaped attention if they existed in *S. anomalus*. Those are lack of fermentation of sucrose and the nonformation of spores in liquids other than wort. To me it seems that the two yeasts are similar, and the failure to note certain characteristics can be accounted for by the brevity of the original description.

EXPLANATION OF ILLUSTRATIONS.

1. Cells grown in beer wort. x700.
2. Eight days' growth in Pasteur solution containing 5 per cent. sucrose. x700.
3. Forty-eight hours' growth on wort gelatine plate. x30.
4. Ten days' growth in Pasteur solution containing 5 per cent. lactose. x320.
5. Cells showing spore development, from wort gelatine plate, ten days' growth. x700.
6. Cells showing spore development, from wort gelatine plate, two weeks old. x700.

