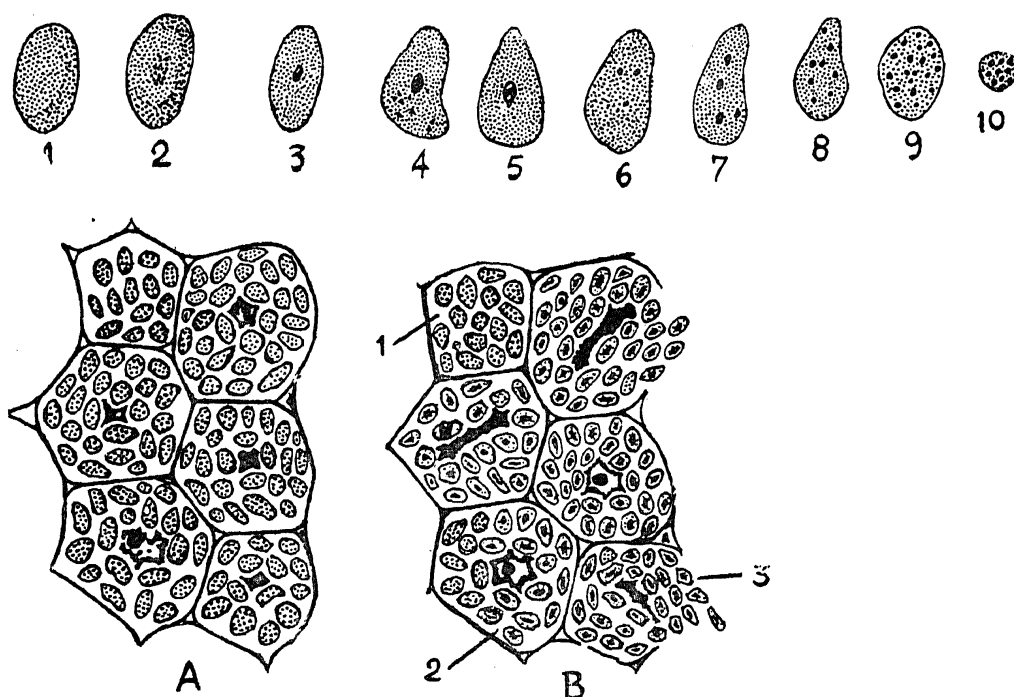


himself is more diffident and designates the cell contents merely as symbiotes. In Sulc's¹ Fig. 9 (2), which is illustrated in part as Fig. B here, he gives the picture of the tumour in a more advanced stage. In Fig. B (tumour cell No. 1) the yeasts are identical with those seen in Fig. A; in both, the germs are immature. In Fig. B (tumour cell No. 2) some yeasts are immature while the rest are nucleated. The yeasts in Fig. B are relatively more developed—particularly those seen as cell inclusions in tumour cell No. 3 are fully mature and on their way to infect future eggs. To a critical observer Figs. A and B would strike as similar. They would hardly appear to have been drawn from a histological section or with a camera lucida and are more or less diagrammatic in representation. Such an idealisation appears to have

tures. What is interesting is to find in Nolte's Fig. 12, m., an identical stage with that of Sulc's Fig. 10 (10), or Fig. 10 here, although Nolte considers his germ a bacterium and Sulc, an yeast. In representing such life-cycles, white blood cells of insects, secretion granules and bacterial metabolic products have been confused; and the results, although based on correct observations, have to be very critically interpreted. For these reasons, a physiological test, like the production of a pigment *in vitro* by the culture of the symbiotic germ, conclusively proves the correct isolation of the symbiote. Neither the insect nor the symbiote produces any colour and hence the only finding that has to be emphasised is the existence of a bacterium in the smears from Sulc's Margarodes.



been applied also to the detailed study of the symbiote whose yeast-like nature he proposed to establish.

On p. 17 in Figs. 1 to 10, Sulc¹ gives the life-cycle of the symbiote. His illustrations are reproduced with his numerical indications. Fig. 1 is the youngest stage, then comes Fig. 2 and thirdly No. 3; all these forms are seen while the yeasts are in the tumour cell. The development occurs mainly in the formation of a nucleus-like central body which has been merely illustrated without any explanation. The symbiote, when it leaves the tumour, enters the lumen of the oviduct and shows more and more clearly the chromatin body as in Figs. 4 and 5. By the time it reaches the follicle it shows the development represented in Figs. 6 to 9. The yeast, finally ready for infecting the egg, is reduced in size but is rich in chromatin bodies resembling chromosomes, being the stage shown in Fig. 10—the end of a mysterious life-cycle of an unknown germ.

Nolte³ has carefully studied symbiosis in the beetle, *Erythrapin miniatum*. The symbiote is a bacterium, but with a complicated life-cycle, as in many other cases of symbiotes which have been studied only in sections and never in cul-

The work was undertaken, at Brünn, in the Laboratory of Prof. Sulc, to whom it is a pleasant duty to thank here again.

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1. Sulc, *Pub. bio. Ecole. Haut. Etud. veter.*, Brünn, 1923, 12, 16. 2. Buchner, *Erg. d. Biol.*, 1928, 4, 73. 3. Nolte, *Zeit. f. Morph. u. Okol.*, 1937, 33, 179.

RATE OF GROWTH OF DIPLOID AND TETRAPLOID YEASTS

THE effect of duplication of chromosomes has been known to result in a change in the norm of reaction. In higher plants investigations show that the geographical distribution of the autotetraploids differ in many cases from those of the diploids. In fact, autotetraploids have been known from localities which are considered unfavourable to the diploids.

In yeasts duplication of chromosomes leads to a change in the characteristics of the giant

colonies.¹ Occasional spontaneous tetraploids could be seen as smooth sectors in giant colonies of the control (Photo 1, T). Since the

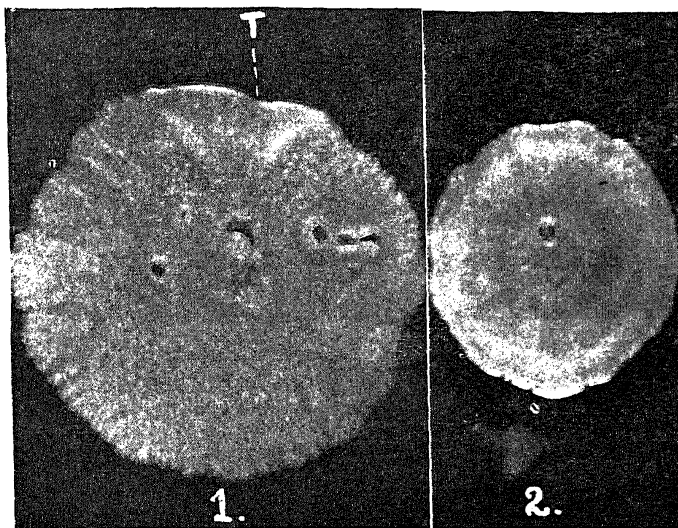
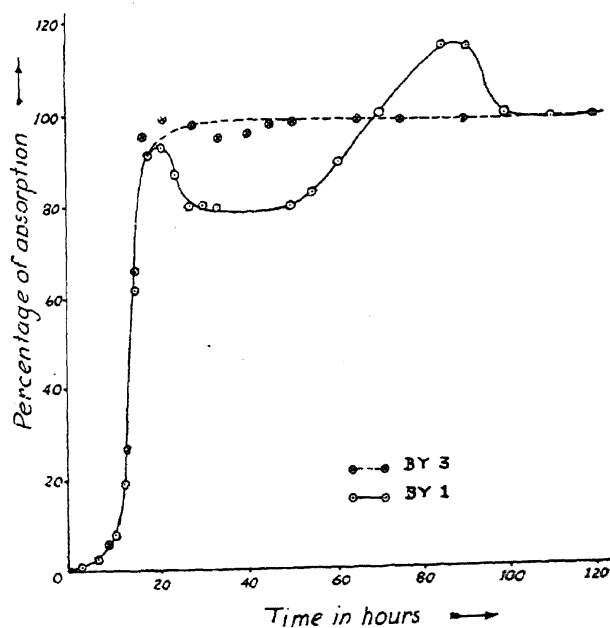


Photo. 1. BY 1. Ten day gr. wth. 2.5 cm. 30-9-1947.
Photo. 2. BY 1 + BY 3. Nine-day growth. 2.2 cm.

surface of the control has folds and striations, it is easy to distinguish between the diploid and the tetraploid. The contour of the sector indicates that its growth is more vigorous than that of the control. It was thought desirable, therefore, to study the structure of a colony developing from an inoculation of a mixture of the control and the tetraploid. Twenty four hour wort cultures of the control, BY 1, and a tetraploid, BY 3,^{2,3} were mixed



together, well shaken and inoculations were made immediately from this mixed culture. Photograph 2 shows the appearance of the colony after a nine-day growth. It has the typical appearance of the tetraploid colony, indicating that the tetraploid cells have completely eliminated the cells of the control strain by their rapid growth.

Investigations⁴ on the nutritional requirements of the diploid, BY 1, and the tetraploid, BY 3, showed no difference. From the data available it could not be judged whether the rate of utilization of the nutrients required for full growth are identical. Since the experiments with giant colonies showed that the rates of growth of the two strains are different, an investigation was carried out on their growth-rates in a standard all-vitamin synthetic medium.⁵ The growth of the yeast inoculated into 5 c.c. of medium in 100 c.c. conical flasks, incubated at 26°C., was measured at various intervals using a Lumetron turbidometer.

The diploid strain shows two cycles of growth. The logarithmic growth phase ends in 21 hours after which there is a fall in the curve which is followed by another steep rise indicating the second cycle of growth referred to by Richards.⁶ On the other hand, the tetraploid shows a quicker growth-rate and the end of the logarithmic phase is reached in 16 hours. The curve afterwards is parallel to the X-axis and does not show a second cycle of growth.

The difference between the diploid and the tetraploid is thus not limited to the changes in the colony characteristics alone. Duplication of chromosomes seems to affect a number of factors. The similarity of the growth curves of the tetraploid and diploid to that recorded by Richards⁶ for the same strain of yeast grown in media with and without colchicine is striking. Without either considering the possibility of polyploidy or a study of the behaviour of the colchicine-treated cells in normal media, Richards claimed not only that colchicine is a food and that it lessens the adverse effects of the increased quantities of toxic waste products released into the medium as a result of growth and fermentation, but also that colchicine fails to reveal mitosis.

From the observations recorded above, it appears that colchicine is neither a food nor is responsible for the disappearance of the second cycle of growth in the case of cultures in media containing the drug. Richards' results, in fact, offer evidence that colchicine has induced polyploidy.

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1. Subramaniam, M. K., and R. Ranganathan, *Proc. Nat. Inst. Sci. (India)*, (in press). 2. Subramaniam, M. K., *Ibid. (India)*, 1946, 12, 143. 3. —, *Ibid. (India)*, 1947, 13, 129. 4. Prema Bai, Miss M., *Curr. Sci.*, 1947, 16, 317. 5. Richards, O. W., *Arch. Protist.*, 1932 78, 263. 6. Richards, O. W., *J. Biol.*, 1938, 36, 187.