

BUD POSITION IN *SACCHAROMYCES CEREVISIAE*

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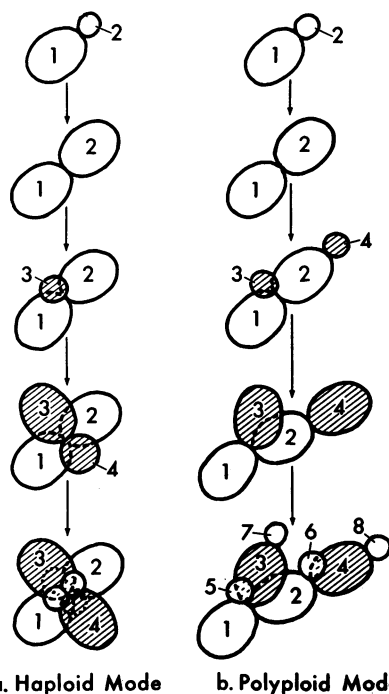
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In a series of radiobiological studies we have observed by time-lapse photography the growth of individual cells of *Saccharomyces cerevisiae*. We found (1) some regularity in the position of the buds produced successively by a single mother cell, and (2) that the position of the first bud of a new cell is invariant but dependent on ploidy.

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MATERIALS AND METHODS

All observations were made on cells actively growing in a liquid medium containing 0.5 per cent yeast extract and 1 per cent glucose. A drop of a growing culture was placed on a small mound of soft agar on a cover slip, which previously had been cemented to a slide with a hole cut through it. A second cover slip was mounted on the opposite side of the slide so it also was in contact with the agar. This sandwich culture provided an air layer between the cover slips which permitted aerobic growth and the agar reduced drifting of the cells by holding them in a thin liquid layer next to the glass. The strains used are listed in table 1. The haploids SC-6 I, II, III, IV, and V were derived from the diploid SC-6 by sporulation. The other strains were from the Carbondale and Berkeley collections. In the discussion the point of contact of a bud with its mother cell is called the birth end; the opposite end is the free end. This defines definite points on the cell surface since the long axis of a bud is al-



a. Haploid Mode b. Polyploid Mode

Figure 1. Schematic diagram of haploid and polyploid modes of budding. Shaded cells are 2nd generation. In both diagrams 3 and 4 have budded from 1 and 2, respectively. In the next generation 5, 6, 7, and 8 are progeny of 1, 2, 3, and 4, in that order. Note that the budding synchrony present in the polyploid mode is absent in the haploid mode (e.g., 3 and 4). In all cases cell 1 had never before budded.

TABLE 1

Ploidy and mating type of the strains of yeast employed

Strain	Ploidy	Mating Type
22042	Haploid	α
8256		a
27127		a
SC-7		Not known
SC-6, I, II, III, IV, V		Not known
22D	Diploid	$\alpha\alpha$
14268 \times 8256		(α) (a)
13844 \times 11296	Triploid	(a) ($\alpha\alpha$)
11294 \times 11296	Tetraploid	(aa) ($\alpha\alpha$)
X323		($\alpha\alpha$) (aa)
X362	Hexaploid	(aaa) ($\alpha\alpha\alpha$)

ways perpendicular to the surface of the mother cell (Nickerson, 1942).

RESULTS AND DISCUSSION

Observation revealed that the first few buds produced by a given mother cell grow from nearly the same point on its surface. This is shown clearly in figure 1*b* in which cells 2, 3, and 5 have budded from cell 1. As cell 3 grows, it pushes 2 aside; similarly cell 5 displaces 3 and 2 as it enlarges. After four or five buds have been produced in this way, further buds arise from other regions of the cell surface. This change may be related to the presumed inability of buds to form in a region containing many bud scars (Mortimer and Johnson, 1959). These later bud sites do not serve as points for repetitive budding, as does the point from which the first bud arises, nor is there a consistent pattern for bud position after the change from the initial bud point. Also, bud position does not seem to be related to interphase position of the nucleus (visualized by acridine orange fluorochroming *in vivo*). Haploid cells differ from those of higher ploidies with respect to the position at which the first bud of a new cell forms. In the haploid mode (figure 1*a*), a cell which has never before budded produces its first bud as near the birth end as possible. For example, cell 2 grew from 1 and budded at its birth end to produce 4. Similarly, further buds came from the birth ends of cells 2, 3, and 4. This mode of budding results in the formation of tightly packed, globular microcolonies.

In the polyploid budding mode, however, the first bud of a fresh cell grows from the free end (figure 1*b*). Cells 3 and 4 are buds of 1 and 2, respectively. Cell 4 clearly grew from the free end of 2. Similarly, cells 7 and 8 have arisen, in turn, from the free end of 3 and 4; consequently, the eight cell stage consists of a planar array of cells rather than the globular configuration of the haploid.

No satisfactory explanation is available for the haploid-polyploid difference. Since different macromolecular components are found in the cell wall of the two mating types of yeast (Brock, 1959), it was thought that the different modes of budding might be related to mating type. But

this is clearly not the case (table 1). The haploid strains 8256 and 22042 are types α and α , respectively, but have the same mode. That the budding mode is not a genetic character is suggested by the fact that all haploid strains derived from SC-6 show the haploid mode. Such abnormal segregation ratios for a genetic trait would be extremely improbable.

The interesting consequence of these observations is that, as long as a cell has not budded more than three or four times, one can predict with certainty where the next bud will arise. This presumptive bud region has been examined prior to budding by phase-contrast, polarization, and interference microscopy but no particular differentiation has been evident. It is suggested that the corresponding electron microscopic examination should be made.

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SUMMARY

The growth of individual yeast cells has been observed by time-lapse photography. The position of the first bud of a new cell is ploidy dependent, being at the birth end in haploid and the opposite end in polyploid cells. The first few buds tend to arise from the same point of the mother cell.

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