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Maintaining pH of the culture medium in cytogerontological experiments: effect on the cell viability and the shape of the cells' survival curve

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Motivation and Aim: One of the problems arising when modeling stationary phase/chronological aging of mammalian/yeast cells in a non-subcultured culture is the acidification of the growth medium at later stages. Moreover, some researchers believe that the chronological aging of yeast is associated with the accumulation over time of acetic acid in the medium and, as a consequence, acidifying it to pH 4 and lower [1]. The aim of this work was to assess the effect of different methods of maintaining pH on the growth and survival of a non-subcultured culture of mammalian cells.
Methods and Algorithms: Transformed Chinese hamster cells were incubated at 37 °C in Dulbecco's modified Eagle's medium supplemented with 10 % bovine serum. The contribution of various ways of maintaining pH of the growth medium to viability of the culture as well as its growth and survival was analyzed. For this purpose, HEPES buffer solution was added to the medium in hermetically sealed vials to a final concentration of 20 mM, or the cells in non-hermetical vials were cultured at 5 % CO_2.
Results: It was found that the optimal way to maintain pH was to cultivate the cells not at 5 % CO_2, but in hermetically sealed vials with the addition of HEPES buffer to the growth medium. If the buffer was present in the medium from the moment of seeding, the culture quickly reached the "plateau" and its saturating density was greater than in the control group, with the shape of the curve in both groups being the same and all cells' dying out by 52nd day. The groups differed in pH only at the initial stages, after reaching the "plateau" this parameter remained the same in both control and experimental group until the end of the experiment.
Conclusion: Growth medium pH affects the growth rate and saturation density of the culture, but one can not name the acidification of the medium as the main cause of cell death in the stationary phase. Adding a buffer only provides optimal conditions at the initial stages of culture growth. As for works in which the acidification of the medium is called the main cause of the chronological aging of yeast, from such papers' data it is obvious that in the absence of pH-supporting factors, yeasts simply die out "exponentially", i. e. there is no aging [2]. It should be noted that in our experiments on the study of stationary phase aging of mammalian cells in hermetically sealed culture vials, they "grow old according to Gompertz".
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