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T. L. Knubovets^a; M. L. Verhovskaya^b; A. N. Belozersky^b

^a Institute of Chemical Physics of the USSR Academy of Sciences, Moscow, USSR ^b Laboratory, Moscow State University, Moscow, USSR

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^{31}P NMR MEASUREMENT OF INTRACELLULAR pH IN BACILLUS FTU

T.L.KNUBOVETS, M.L.VERHOVSKAYA*

Institute of Chemical Physics of the USSR Academy of Sciences, Kosygin Str. 4, Moscow 117977, USSR

*A.N.Belozersky Laboratory, Moscow State University, Moscow 117234, USSR

The ability of alkalotolerant bacterium *Bacillus* FTU to maintain intracellular pH was studied in the wide range of extracellular pH: 6.5-11, by means of ^{31}P NMR. *Bacillus* FTU is an obligatory aerobic microorganism, possessing a high speed of oxygen consumption, that is why it was especially important to provide sufficient oxygen supply during the entire period of measurement. We have worked out a special glass filter insert for the NMR sample tube which allowed for uniform air supply in the cell suspension. The viability of the cells was checked by ATP level analysis since in the case of *Bacillus* FTU ATP content is highly sensitive to the quantity of oxygen. The endogeneous P_i was used as a pH indicator. The level of P_i is extremely high in *Bacillus* FTU - around 70-90 mM, that's why it was possible to use relatively low concentration of cells in the suspension to obtain sufficient NMR sensitivity (approximately 5-7% of intracellular volume). Special procedure was employed to obtain a calibration curve: we used gramicidin in the concentration 10 μM to equilibrate intra- and extracellular pH; the former being measured by ^{31}P NMR and the latter - by pH-meter. *Bacillus* FTU in energized state showed the ability to maintain constant intracellular pH (approximately 8.0-8.5) in the range of extracellular pH from 7.3 till 10.5, which agreed with the pH range of the cell growth. Thus, the obtained data support the idea of intracellular pH being of vital importance for cell metabolism.