The Metabolism of Phosphoproteins in Lactobacillus casei

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The incorporation of radioactive inorganic phosphate into the phosphoprotein fraction of *L. casei* has been compared with the phosphorus uptake of nucleotide and nucleic acid fractions during the transition from lag

to log phase.

Aliquots of a 90 liter bacteria culture were taken at varying intervals subsequent to inoculation of the 82P-containing medium. Bacteria were prepared from the aliquots as described previously 1. Fractionation was carried out by a modified Schneider procedure. The 10 and 5 % TCA extracts were extracted with ether and run on Dowex 1, formate columns 2, the nucleic acid fraction after partial hydrolysis with 0.1 N KOH, 100°, 2 hours. The alkali-stable fraction from the 5 % TCA extract was hydrolysed enzymatically with desoxyribonuclease and snake venom diesterase and then fractionated on a Dowex 1 column. The "rest protein" was treated as described previously 3. Table 1 shows the uptake of radioactive phosphorus into the major ninhydrin-positive phosphorus-containing, component of the "rest protein" partial acid hydrolysate compared with the number of bacteria and the number of counts per µg dry bacteria.

Table 1.

Time after inoculation in min.	$\begin{array}{c} \textbf{Bacterial} \\ \textbf{count} \\ \textbf{Number/ml} \\ \times \textbf{10}^{\textbf{9}} \end{array}$	Dry bacteria cpm/μg	Component of the rest protein, $cpm/\mu g$ P
0	48		
30	51	96	320
40	52	82	210
50	55	102	700
60	55	147	1 310
70	56	143	1 250
80	74	199	2 350

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Differences between Phosphoproteins of Animal and Bacterial Origin

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The occurrence of phosphoserine and phosphothreonine in partial hydrolysates of the "Schneider protein" fractions of rat tissues has previously been reported ¹. We now have succeeded to isolate these two compounds in crystalline form from sheep liver tissue. The first one has already been isolated from calf liver tissue ². The same method of preparation was used now. The compounds were identified by X-ray powder diagrams. Sufficient amounts of serinephosphate permitted micro chemical analysis. (Found: C 19.53; H 4.52; N 7.73; P 16.75. Calc. for C₃H₂O₆NP: C 19.46; H 4.32; N 7.57; P 16.76).

In earlier communications we also reported the presence of phosphoserine in partial hydrolysates of protein from Lactobacillus casei 3,4. These hydrolysates contain in addition a number of other ninhydrin-positive, phosphorylated compounds, but so far it has not been possible to demonstrate the presence of threeninephosphate. One of these compounds is present in much greater quantities than is phosphoserine. On complete hydrolysis it gives rise to a ninhydrin-positive compound which does not correspond in position to either serine or threonine on paper chromatograms. The possible identity of the compound is discussed. Details of these experiments will be published later.

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