

A Probable Structure of the Bifidus Factor in Carrot Root

In the previous communication,¹⁾ we reported the purification of a new growth promoting factor of *Lactobacillus bifidus* from roots of *Daucus carota* L.

Owing to the strong acidic character of the factor, numerous kinds of nucleotides and their related compounds were bioassayed with a strain of *L. bifidus*²⁾. Among these, only one possessing a high growth promoting activity was coenzyme A, although its chromatographic behavior was quite different from that of the factor in carrot. Consequently 4'-phosphopantetheine and pantethine, precursors of coenzyme A, were examined. Both of them showed very high activity on *L. bifidus*, and the former's behavior on papers was quite similar to the factor. These results are summarized in the Table I.

TABLE I.

	Specific activity carrot unit/mg.	Relative mobility in	
		PEP ^{a)}	PC ^{b)}
Purified carrot (still impure)	40,000	1.00	0.91
Coenzyme A	40,000	1.02	0.09
4'-Phosphopantetheine	230,000	1.03	0.88
Pantethine	250,000	—	—
Pantothenic acid	0	—	—

a) Paper electrophoresis with 0.1M HCOOH, 17 volt/cm., 1 hr., taking the mobility of uridine monophosphate as 1.00 and caffeine as a reference.

b) Paper chromatography with solvent system of isopropanol, 28% ammonia and water (7:1:2), taking Rf value of uridine-3',5'-cyclic phosphate as 1.00.

Hydrolysis of the highly purified factor with *N* HCl produced gradually phosphoric acid, but not sulfuric acid. The fact indicates that the strong acidic character of the factor is due to a phosphate group in its molecule. After hydrolysis with 6*N* HCl, β -alanine was detected on a thin-layer chromatogram as its dimethylaminonaphthalene sulfonyl derivative.³⁾

From these experimental results, it is most probable that the factor is identical with 4'-phosphopantetheine or its closely related compound. This conclusion coincides with the finding of György who demonstrated the growth promoting activity of pantethine on var. *penn.* and other strains of *L. Bifidus*,⁴⁾ indicating that they have no enzyme to convert pantothenic acid to pantethine in the course of biosyntheses of coenzyme A.

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Faculty of Pharmaceutical Sciences,
University of Tokyo, Hongo, Tokyo

Zenzo Tamura (田村善藏)
Terumi Nakajima (中島輝躬)
Keijiro Samejima (鮫島啓二郎)

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