Lactobacillus sake Katagiri, Kitahara, and Fukami 1934 Is the Senior Synonym for *Lactobacillus bavaricus* Stetter and Stetter 1980

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The taxonomic relationships of six strains of *Lactobacillus bavaricus*, including type strain 136 (= DSM 20269) and reference strain Tro-8/78, *Lactobacillus curvatus*, and *Lactobacillus sake* were assessed by performing DNA-DNA hybridization experiments. We confirmed that *L. curvatus* and *L. sake* are genotypically distinct species. However, the previously described differentiation of *L. bavaricus* from *L. sake* could not be substantiated. We found that *L. sake* is specifically related to the type strain of *L. bavaricus* and all *L. bavaricus* reference strains except "*L. bavaricus*" Tro-8/78, which is related to *L. curvatus*. Clearly, strains identified as *L. bavaricus* Stetter and Stetter 1980 do not form a separate genotypic group, and the name *L. bavaricus* is a junior synonym of *L. sake* Katagiri, Kitahara, and Fukami 1934.

Bacterial isolates obtained from sauerkraut or fermented cabbage leaves were described previously as members of the new species *Lactobacillus bavaricus* (9). This species was differentiated from the phenotypically very similar species *Lactobacillus sake* and *Lactobacillus curvatus* because it formed only L-(+)-lactic acid and not the racemic product during growth in cabbage leaf juice. Growth experiments (10) revealed that lactic acid racemase activity apparently is not present in *Lactobacillus bavaricus* strains, in contrast to *Lactobacillus curvatus* and *Lactobacillus sake* strains. The separate species status of *Lactobacillus bavaricus* was recently substantiated by Champomier et al. (2), but not by other workers (4). In this paper we present evidence that distinction of *Lactobacillus bavaricus* as a separate species is not justified.

Type and reference strains were obtained from culture collections, as indicated in Table 1. The isolates used in this study were original subcultures used in the study of Stetter and Stetter (9) or extended studies (11) or were obtained from other sources (7). In addition, two apparently mislabelled strains, *Bifidobacterium adolescentis* ATCC 11146 and *Bifidobacterium bifidum* ATCC 11147, which resemble *Lactobacillus sake* and *Lactobacillus curvatus* phenotypically, were also examined to determine their true taxonomic relationships. DNA-DNA hybridization experiments and experiments to determine DNA G+C contents were performed as described previously (5).

As shown in Table 1, the overall DNA G+C contents of the strains belonging to the *Lactobacillus sake*-*Lactobacillus curvatus*-*Lactobacillus bavaricus* group, including the two "*Bifidobacterium*" strains which we studied, ranged from 41 to 44 mol%. Thus, the G+C contents of the "*Bifidobacterium*" strains were clearly outside the range of G+C contents of all known *Bifidobacterium* species (55 to 67 mol%) (8). DNA-DNA hybridization clearly revealed that both "*Bifidobacterium*" strains are closely related to *Lactobacillus sake* (levels of homology, 89 to 98%) (Table 1).

We performed DNA-DNA reassociation experiments and found that the type strain of *Lactobacillus sake*, ATCC 15521, and *Lactobacillus curvatus* reference strain DSM 20010 exhibited about 50% homology with each other but very low levels of homology (around 10%) with strains belonging to other species, including *Lactobacillus casei* DSM 20011^T (T = type strain), *Lactobacillus salivarius* DSM 20492, and *Lactococcus lactis* DSM 20175 (= ATCC 15577). Thus, the separate species status of *Lactobacillus sake* and *Lactobacillus curvatus* was confirmed; our results are consistent with the results of Beck et al. (1) and Dellaglio et al. (3).

As shown in Table 1, *Lactobacillus sake* strains were related at levels of 80 to 100% to *Lactobacillus bavaricus* strains, including *Lactobacillus bavaricus* 136^T (= DSM 20269^T) and all of the *Lactobacillus bavaricus* reference strains except strain Tro-8/78, which was closely related to *Lactobacillus curvatus* instead. Generally, none of the racemase-negative strains, including one isolate obtained from sausage (7), belonged to a separate genotypic group, which could be assigned to *Lactobacillus bavaricus*.

Therefore, we did not confirm the results of Champomier et al. (2). Since these authors used a different subculture of *Lactobacillus bavaricus* 136^T and a different hybridization method, it will be necessary to repeat their study with *Lactobacillus bavaricus* DSM 20269^T.

We concluded that the absence of racemase activity in strains belonging to the *Lactobacillus sake*-*Lactobacillus curvatus* group is not a valid criterion for separating taxa. The distinction of *Lactobacillus bavaricus* from *Lactobacillus sake* or *Lactobacillus curvatus*, as proposed by Stetter and Stetter (9), is not justified, and *Lactobacillus sake* Katagiri, Kitahara, and Fukami 1934 is a senior synonym of *Lactobacillus bavaricus*.

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TABLE 1. Sources, genomic characteristics, and genotypic grouping of the strains which we investigated

Source of unlabeled DNA	Source ^a	Reference	G+C content (mol%)	% Relative binding with ³ H-labelled DNA from ^b :									
				<i>Lactobacillus sake</i> ATCC 15521 ^T	<i>Lactobacillus sake</i> 119	<i>Lactobacillus</i> sp. strain R7c	<i>Lactobacillus bavaricus</i> 136 ^T	<i>Lactobacillus bavaricus</i> 225	<i>Lactobacillus curvatus</i> DSM 20010	<i>Lactobacillus curvatus</i> 148	<i>Lactobacillus casei</i> DSM 20011 ^T	<i>Lactobacillus salivarius</i> DSM 20492	<i>Lactococcus lactis</i> DSM 20175
<i>Lactobacillus sake</i> strains													
ATCC 15521 ^T	ATCC		41	100	100	100	101	97	53	ND ^c	9	11	15
117	Sauerkraut	11	42	105	100	ND	100	95	57	ND	ND	10	12
119	Sauerkraut	11	42	96	100	ND	102	90	53	51	ND	11	10
2/1	Sauerkraut	11	41	100	100	ND	99	99	60	ND	ND	12	14
DL 23	Sauerkraut	11	42	100	ND	ND	ND	ND	46	ND	12	ND	7
Tro 11	FCL ^d	11	42	100	ND	ND	ND	ND	39	ND	8	ND	7
<i>Lactobacillus</i> sp. strain R7e ^e	Sausage	7	43	93	ND	100	ND	ND	36	ND	ND	ND	9
<i>Lactobacillus bavaricus</i> strains													
136 ^T (= DSM 20269) ^{T,e}	Sauerkraut	9	42	80	ND	ND	100	85	48	ND	ND	11	12
225 ^e	Sauerkraut	11	41	87	87	ND	96	100	51	ND	7	11	8
L1 ^e	Sauerkraut	11	44	83	ND	ND	ND	83	47	ND	8	ND	7
L3 ^e	Sauerkraut	11	43	92	ND	ND	ND	88	51	46	9	ND	7
L4 ^e	Sauerkraut	11	43	100	ND	ND	ND	99	57	54	10	ND	8
Tro-8/78	FCL	9	42	48	48	ND	50	47	100	96	10	9	6
<i>Bifidobacterium adolescentis</i> ATCC 11146 ^f	ATCC		43	89	ND	ND	94	85	55	ND	ND	13	14
<i>Bifidobacterium bifidum</i> ATCC 11147 ^f	ATCC		43	89	ND	ND	98	89	56	ND	ND	14	16
<i>Lactobacillus curvatus</i> strains													
DSM 20010 ^g	DSM		43	50	50	48	53	52	100	100	12	11	11
148	Sauerkraut	11	44	48	ND	ND	ND	ND	108	100	9	ND	8
DL 6	Sauerkraut	11	44	51	ND	ND	ND	46	100	100	8	ND	6
DL 17	Sauerkraut	11	44	52	ND	ND	ND	43	96	100	9	ND	6
Tro 17	FCL	11	43	42	ND	ND	ND	ND	100	ND	9	ND	7
<i>Lactobacillus casei</i> DSM 20011 ^T (= ATCC 393 ^T)	DSM		49	8	8	ND	ND	9	11	6	100	10	3
<i>Lactobacillus salivarius</i> DSM 20492	DSM		33	8	9	ND	8	10	8	ND	10	100	11
<i>Lactococcus lactis</i> DSM 20175	DSM		38	9	8	9	6	9	9	7	3	7	100

^a ATCC, American Type Culture Collection, Rockville, Md.; DSM, Deutsche Sammlung von Mikroorganismen, Braunschweig, Germany.

^b As determined by DNA-DNA hybridization.

^c ND, not determined.

^d FCL, fermented cabbage leaves.

^e Lactic acid racemase-negative strain.

^f Isolated from infant feces and placed in the genus *Bifidobacterium* by Malyoth and Bauer (6).

^g The level of DNA-DNA relatedness to type strain DSM 20019 (= ATCC 25601) is 100% according to Dellaglio et al. (3).

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