

Lacticaseibacillus zeae subsp. silagei,
isolated from corn silages

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INTRODUCTION

In a screening project at 45°C, two rod-shaped, non-motile, non-spore-forming, facultative anaerobic, Gram-stain positive lactic acid bacteria, designated as EB0058^T and SCR0080, were isolated from corn silages. They were characterised using a polyphasic approach and identified as novel subspecies of *Lacticaseibacillus zeae* [1].

MATERIAL & METHODS

- Chun, Oren et al. proposed the computation of the overall genome-related index (OGRI) [2]. They suggest the species boundary to be an average nucleotide identity (ANI) value between 95-96% or a digital DNA-DNA-hybridisation (dDDH) value of 70%. The Genome-to-Genome Distance Calculator calculated the dDDH value with recommended formula 2 [3,4]. The ANI values were calculated using the JSpecies Web Server with BLAST+ mode [5] (Figure 1).
- Growth tests were conducted in MRS broth with a range of parameters, including temperature (5 to 55°C), pH (3.0 to 10.5), NaCl salt concentrations (4% w/v to 10% w/v) and KCl salt concentrations (6% w/v, 8.4% w/v and 10.8% w/v). Using the commercially available test kit API 50 CH system (bioMérieux), the capacity for carbohydrate metabolism was measured (Table 1).

RESULTS & DISCUSSION

Figure 1. ANI and dDDH (formula 2) values for strains EB0058^T, SCR0080 and related species.

	Group A				Group B								
	<i>L. zeae</i> subsp. <i>silagei</i>				<i>L. zeae</i> subsp. <i>zeae</i>								
	1	2	3	4	5	6	7	8	10	11	12	13	
[1] <i>L. zeae</i> subsp. <i>silagei</i> EB0058 ^T	-	99.9	99.9	99.9	96.3	96.3	96.3	96.4	93.7	93.6	93.7	94.1	ANI ↑
[2] <i>L. zeae</i> subsp. <i>silagei</i> SCR0080	99.6	-	99.9	99.9	96.3	96.3	96.2	96.3	93.7	93.6	93.8	93.8	
[3] <i>L. zeae</i> subsp. <i>silagei</i> UD2202	99.8	99.9	-	99.9	96.3	96.2	96.2	96.3	93.7	93.6	93.7	93.7	
[4] <i>L. zeae</i> subsp. <i>silagei</i> CECT9104	99.8	99.9	99.3	-	96.3	96.3	96.2	96.3	93.7	93.6	93.8	93.7	
[5] <i>L. zeae</i> subsp. <i>zeae</i> DSM 20178 ^T	70.8	70.7	70.6	70.7	-	100	99.3	99.5	93.6	93.5	93.5	94.0	
[6] <i>L. zeae</i> subsp. <i>zeae</i> KCTC 3804 ^T	70.8	70.7	70.5	70.7	100	-	99.2	99.5	93.6	93.5	93.5	93.9	
[7] <i>L. zeae</i> subsp. <i>zeae</i> CRBIP24.58	70.5	70.2	70.3	70.4	95.6	95.5	-	99.5	93.6	93.5	93.5	93.7	
[8] <i>L. zeae</i> subsp. <i>zeae</i> FBL8	71.5	71.5	71.3	71.5	95.9	95.7	97.3	-	93.6	93.5	93.6	93.9	
[10] <i>L. parahuelsenbergensis</i> LD0937 ^T	54.2	54.1	53.8	54.2	53.9	54.1	54.3	54.4	-	95.9	95.7	95.2	
[11] <i>L. styriensis</i> SCR0063 ^T	54.8	54.7	54.5	54.8	54.6	54.8	54.4	55.0	67.6	-	95.9	95.2	
[12] <i>L. huelsenbergensis</i> DSM 115425 ^T	54.3	54.2	53.9	54.2	53.4	53.5	53.4	54.2	66.5	65.9	-	95.4	
[13] <i>L. casei</i> DSM 20011 ^T	57.1	56.9	56.5	56.9	57.3	57.4	56.0	57.4	64.1	64.9	63.9	-	
	dDDH <-												

Due to whole-genome sequence-based characterization, EB0058^T and SCR0080, together with two other strains available at NCBI GenBank, were separated into a distinct clade from *Lacticaseibacillus zeae* DSM 20178^T.

In contrast, comparing the new subgroup to all publicly available genomic sequences of *L. zeae* strains including the type strain DSM 20178^T showed dDDH values between 70.2% - 72.5% and ANI values between 96.2% - 96.3% (Figure 1).

Table 1. Differential phenotypic and growth characteristics of the two isolates and their phylogenetically closest related type strain *L. zeae* DSM 20178^T.

Strain	Growth at:				Acid production from:			
	5°C	pH 10	9% NaCl	8.4% KCl	L-Arabinose	Sorbitol	Maltose	D-Arabitol
EB0058 ^T	+	-	+	+	+	+	+	+
SCR0080	+	-	+	+	+	+	+	+
<i>L. zeae</i> DSM20178 ^T	-	+	-	W	-	-	-	-

CONCLUSION

Based on their chemotaxonomic, phenotypic, and phylogenetic characteristics, EB0058^T and SCR0080 represent a new subspecies of *L. zeae*. The name *Lacticaseibacillus zeae* subsp. *silagei* is proposed with the type strain EB0058^T (=DSM 116376 =NCIMB 15474).

References:

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