The taxonomic positions of *Weissella kimchii* and *Weissella cibaria* were examined using phenotypic tests, 16S rRNA gene sequencing and DNA–DNA hybridization. The two species displayed very similar sugar-fermentation patterns and produced the D-form of lactic acid. Phylogenetic analysis showed that *W. cibaria* is the closest neighbour of *W. kimchii*, sharing about 99.7% similarity in the 16S rRNA gene sequence. DNA–DNA hybridization experiments further reinforced these results, demonstrating a 98.2% reassocation level between *W. cibaria* LMG 17699T and *W. kimchii* DSM 14295T. On the other hand, strain RO5, an isolate from paddy rice silage, exhibited 99.4% 16S rRNA gene sequence similarity and a DNA reassocation level of more than 97% with both type strains. Therefore, it is proposed that the species *W. kimchii* should be reclassified as *W. cibaria*.

In a recent study, while investigating lactic acid bacteria inhabiting paddy rice silage in Japan, we reported the presence of isolates belonging to the genus *Weissella* (Ennahar et al., 2003). On the basis of an analysis of the 16S rDNA sequences, strain RO5 appeared to be very closely related to the type strains of *W. kimchii* and *W. cibaria*, suggesting a close phylogenetic affiliation between these species. *W. kimchii* was isolated from a traditional fermented vegetable in Korea (Choi et al., 2002), whereas *W. cibaria* originated from a traditional unfermented food ingredient in Malaysia (Björkroth et al., 2002). Although separate studies clearly distinguished the two species from other members of the genus *Weissella* on the basis of their phenotypic and phylogenetic characteristics (Björkroth et al., 2002; Choi et al., 2002), a comparative investigation including both species has yet to be carried out.

The present study set out to identify strain RO5, isolated from paddy rice silage, and to investigate whether *W. cibaria* and *W. kimchii* are phylogenetically different. Comparative 16S rDNA sequence and DNA–DNA hybridization analyses as well as a physiological analysis were carried out. Since *Weissella* spp. are relatively common in Japanese rice silage (Cai et al., 1994, 1998; Ennahar et al., 2003), our study is also interesting from an ecological point of view.

The *Weissella* type strains used in the present study are shown in Fig. 1. Strain RO5 was isolated previously from paddy rice silage (Ennahar et al., 2003). Bacterial cultures were grown on MRS agar (Difco) at 30°C for 24 h, before being transferred to nutrient broth (Difco) with 10% glycerol and stored as stock cultures at −80°C for further analysis.

Strain RO5 was compared with type strains of *Weissella* species by using carbohydrate assimilation/fermentation on API 50 CH strips (bioMérieux), as described previously (Ennahar et al., 2003). RO5 displayed fermentation patterns matching those of *W. cibaria* LMG 17699T and *W. kimchii* DSM 14295T. The three strains produced acid from D-fructose, D-glucose, D-mannose, N-acetylglucosamine, amygdalin, arbutin, salicin, cellobiose, maltose and sucrose (see Table 1, supplementary data), and hydrolysed aesculin. Weak acid production was observed with β-gentiobiose and gluconate. Mannitol, methyl α-D-glucoside, melibiose,
W. kimchii DSM 14265T (AF312874), W. cibaria DSM 17699T (AJ299089), Strain RO5 (This study) (AF515221), W. confusa JCM 1093T (M20368), W. paramesenteroides NCDO 803T (AB023238), W. thaladiensis JCM 10695T (AY023838), W. helenica NCIMB 2973T (X95981), W. minor DSM 20014T (M20309), W. viridescens DSM 20410T (M20400), W. halotolerans DSM 20190T (M20307), W. kandieri DSM 20593T (M20308), W. koreensis JCM 11263T (AY035891), W. soli DSM 14420T (AY028260), Leuconostoc mesenteroides DSM 20343T (M20305).

DNA base content was determined by the method of Tamaoka & Komagata (1984) using HPLC, as described previously (Ennahar et al., 2003). The mean G+C content of the DNA of strain RO5 was 48.5 mol%, which is comparable to the corresponding published values for W. kimchii and W. cibaria, i.e. 48.2 and 44–45 mol%, respectively (Björkroth et al., 2002; Choi et al., 2002).

As described above, phenotypic characterization, 16S rDNA sequence analysis, DNA–DNA hybridization and G+C-content determination showed that strain RO5, isolated from paddy rice silage, should be classified within the genus Weissella, with W. cibaria and W. kimchii as its nearest phylogenetic relatives. More importantly, on the basis of the detailed phenotypic and genetic characteristics, we could find no significant differences to justify separation of the type strains W. kimchii DSM 14295T and W. cibaria DSM 17699T. Therefore, given the earlier publication of the W. cibaria description, we propose that W. kimchii (Choi et al., 2002) is a later heterotypic synonym of W. cibaria (Björkroth et al., 2002) and consequently should be renamed.

**Fig. 1.** Phylogenetic tree showing the relative positions of strain RO5 and Weissella species as inferred by the neighbour-joining method for 16S rDNA sequences. Bootstrap values for a total of 100 replicates are shown at the nodes of the tree. References of the type strains used for comparison are given, as well as the accession numbers for all 16S rDNA sequences (in parentheses). Leuconostoc mesenteroides was used as an outgroup. Bar, 1% sequence divergence. A phylogenetic tree constructed using a larger data set can be found in ISEM Online.

**References**


Weissella kimchii is a synonym of *Weissella cibaria*


