

Torulaspora nypae sp. nov., a novel yeast species isolated from nipa (*Nypa fruticans* Wurmb.) inflorescence sap in southern Thailand

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Abstract

Two strains (YSP-384 and YSP-399), representing a novel *Torulaspora* species, were isolated from two nipa inflorescence sap samples collected in Trang province in the southern part of Thailand. The two strains had identical sequences of the D1/D2 domains of the large subunit (LSU) rRNA gene and the internal transcribed spacer (ITS) regions. The two strains were closest to *Torulaspora maleeae* CBS 10694^T, but with 1.1% nucleotide substitutions in the D1/D2 domains of the LSU rRNA gene and 5.2% nucleotide substitutions in the ITS regions. Phylogenetic analysis based on the concatenated sequences of the ITS regions and the D1/D2 domains of the LSU rRNA gene supported that the two strains represented a distinct species in the genus *Torulaspora*. Some phenotypic characteristics of the two strains differed from *T. maleeae* including the two strains have ability to assimilate D-xylose, D-glucono- δ -lactone and melizitose, and inability to ferment maltose and raffinose, whereas *T. maleeae* has opposite results. Therefore, the two strains are described as representing a novel species, for which the name *Torulaspora nypae* sp. nov. was proposed.

The genus *Torulaspora* was first established by Lindner in 1904 and *Saccharomyces delbrueckii* was transferred to this genus as *Torulaspora delbrueckii* [1]. Later, *Torulaspora* was merged into the genus *Saccharomyces* together with the genus *Zygosaccharomyces* by Lodder and Kreger van Rij [2]. However, in 1975, the genera *Torulaspora* and *Zygosaccharomyces* were redefined by van der Walt and Johanssen [3]. In 2003, the family *Saccharomycetaceae* was resolved by phylogenetic analysis using a multigene sequence consisting of an rRNA gene repeat, single-copy nuclear genes and mitochondrially encoded genes into 11 well-supported clades, including *Kazachstania*, *Kluyveromyces*, *Lachancea*, *Nakaseomyces*, *Naumovia*, *Saccharomyces*, *Tetrapisispora*, *Torulaspora*, *Vanderwaltozyma* and *Zygotoruspora* [4, 5]. On the basis of the multigene phylogenetic analysis, five species, *Torulaspora delbrueckii*, *Torulaspora globosa*, *Torulaspora franciscae*, *Torulaspora microellipsoides* and *Torulaspora pretoriensis*, have been accepted in the genus *Torulaspora* [5]. In 2008, *Torulaspora maleeae* was proposed based on strains

isolated from moss, leaves, soil and sediment in mangrove in Thailand [6]. After that, *Torulaspora quercuum* was described from strains isolated from oral cavities of healthy human and a leaf of an oak tree [7]. Lastly, *Torulaspora indica* was proposed on the basis of strains isolated from coal mine soil samples [8]. Therefore, at present the genus *Torulaspora* consists of eight species. The strains of *Torulaspora* species can be found in various habitats; for example, strains of *T. delbrueckii*, an ubiquitous yeast species, were reported to be found in soil, fermenting grape and other berry juices, agave juice and tea-beer [9], stem and rhizosphere of sugarcane [10], tree bark [11], phylloplane [12], spontaneous coffee bean fermentation [13], and wastewater treatment systems [14]. *T. globosa* was isolated from soil [9], sugarcane juice [15] and insect frass [16]. Strains of *T. pretoriensis* were found in soil, natural fermentations of cocoa and phylloplane [17]. *T. microellipsoides* was found in apple juice, tea-beer, sandalwood exudates and lemonade [9]. *T. franciscae* was reported from soil [9] and mud [18].

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Abbreviations: PDA, potato dextrose agar; YM, yeast extract–malt extract.

The GenBank/EMBL/DDBJ accession numbers for the sequences of the D1/D2 domains of the LSU rRNA gene of strains YSP-384 and YSP-399 are LC486359 and LC486360, and of the ITS regions are LC486365 and LC486366, respectively. The MycoBank number for *Torulaspora nypae* sp. nov. is MB832324.