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Sporolactobacillus in Food: Literature Discussing Prevalence and Control

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Members of the genus *Sporolactobacillus* (of which approximately 10 species have been identified) are anaerobic, Gram-positive, rod-shaped, motile, and unusual in that they are lactic acid-producing bacteria that form spores (Doores, 2014; Bozkurt et al., 2016). *Sporolactobacillus* spp. bacteria were first discovered in chicken feed in the 1960s (Kitahara and Suzuki, 1963). Although they are typically found in soil and plants, *Sporolactobacillus* spp. are occasionally found in spoiled or fermented foods (Nakayama and Yanoshi, 1967; Yanagida et al., 1997), including fermented fish/shrimp paste (Surono and Hosono, 1994), Japanese non-salted pickles (Karki et al., 1983), green olives (Lucena-Padros et al., 2014), fermented sweet flour paste (Shen et al., 2013), spoiled orange juice (Fujita et al., 2010), spoiled jelly (Lan et al., 2016), silage (Li et al., 2019), wine grapes (Doores, 2014), fermented soybean paste (Ling et al., 2022), and fermented Chinese beverages and soups (Yan et al., 2022; Zhou et al., 2022; Xu et al., 2023). *Sporolactobacillus inulinus* was found in sugar beet “thick juice”, an intermediate in sugar cane processing (Juste et al., 2008); the researchers speculated the organism came from the air and suggested that an air filter at the top of the thick juice tank might reduce this contamination (Juste et al., 2008). More recently, *Sporolactobacillus nakayamae* was isolated from a food model containing cooked mashed potatoes with fresh cleaned scallions added post-cooling (Bozkurt et al., 2016).

A comprehensive study of 699 food (mostly produce and chicken) and environmental samples found the bacteria in 2 of 38 soil samples but in none of the food samples (including 32 mushroom samples) tested (Doores and Westhoff, 1983). Another study isolated 260 spore-forming bacteria from different types of retail beers in Brazil; no *Sporolactobacillus* spp. were isolated (Munford et al., 2017). Similarly, a study of 75 craft beers in Argentina did not identify *Sporolactobacillus* contamination in any of the beers tested (Latorre et al., 2023).

The ability to sporulate make *Sporolactobacillus* spp. more heat resistant than other lactic acid-producing bacteria. Several early studies reported on the thermal tolerance of *Sporolactobacillus* (Kitahara and Suzuki, 1963; Doores and Westhoff, 1981), and this work is summarized in the 2014 review (Doores, 2014). Heat resistance for *Sporolactobacillus* spp. varies depending on the media in which it is measured. In one study, the average D values for *Sporolactobacillus inulinus* spores were 53.2 min at 75°C, 19.5 min at 80°C, 6.8 min at 85°C and 5.1 min at 90°C, with an average z value of 13.0°C (Doores and Westhoff, 1981). However, in other studies with different media, the D value for *S. inulinus* spores ranged from 1.9 to 1.4 minutes at 80°C and ~1 min at 90°C (Bozkurt et al., 2016). For *Sporolactobacillus nakayamae* spores, the D values in various media were found to be 10.95 to 25.24 min at 70°C; 3.30 to 9.25 min at 75°C, and 1.53 to 3.45 min at 80°C (Bozkurt et al., 2016).

Growth of *Sporolactobacillus* from endospores can occur at relatively high sodium nitrite concentrations (up to 2 mg/mL) and at higher potassium sorbate (up to 4 mg/mL) concentrations than other endospore-forming bacilli (Botha and Holzapfel, 1987). Complete inhibition of vegetative growth of *Sporolactobacillus* was found to occur between 5 and 7 mg/mL potassium sorbate (Botha and Holzapfel, 1987). Most *Sporolactobacillus* strains are resistant to low pH (pH

3) (Hyronimus et al., 2000). The organism can also survive at low water activity (<0.86, in sugar thick juice) but cannot grow vegetatively from spores at water activities lower than 0.955 when salt is used to reduce water activity. Unusually, however, when the water activity is reduced with glycerol, vegetative cell growth can occur at low water activities, with such growth completely inhibited at A~~w~~=0.905 and growth from spores inhibited at A~~w~~=0.880 (Botha and Holzapfel, 1988a; Juste et al., 2008). Vegetative *Sporolactobacillus* cells show similar resistance to gamma radiation when compared to vegetative cells of *Bacillus* and *Clostridium* species, with D_{10} values ranging from 0.350 to 0.525 kGy, while the average D_{10} value for spores is 2.5 kGy (Botha and Holzapfel, 1988b).

Sporolactobacillus grows in sugar solutions up to 70% and can convert sugar to D-lactate better than other lactic acid bacteria (Doores, 2014). *Sporolactobacillus* spores have been suggested to serve as a spoilage indicator for refrigerated foods: if the finished product is temperature abused, the spores will germinate, produce acids and make the product obviously spoiled to the consumer (Mossel and Struijk, 1991). In recent years, the ability of *Sporolactobacillus* to produce commercially important D-lactic acid from inexpensive materials as carbon sources (including waste materials) has been demonstrated (Bai et al., 2016; Michalczyk et al., 2021).

The presence of *Sporolactobacillus* has been associated with the biosynthesis of volatile flavor compounds in fermented soybean paste (Ling et al., 2022), in the Chinese fermented beverages Baijiu (Xu et al., 2023) and Huangjiu (Yan et al., 2022), and in Chinese red sour soup (Zhou et al., 2022). Together with the lactic acid produced by the organism, the unintentional presence of *Sporolactobacillus* could affect the organoleptic properties of other foods. However, control of the *Sporolactobacillus* to prevent spoilage in food products has not been addressed in many studies in the literature, although a model of the thermal inactivation kinetics of S. nakayamae has been published (Bozkurt et al., 2016).

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