American Journal of Biomedical Science & Research

www.biomedgrid.com

ISSN: 2642-1747

Mini Review

A Double Life of Freeze-Dried *Enterococcus Faecium* in Pet Food Industry

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***To Cite This Article:** : Jie Xu.A Double Life of Freeze-Dried Enterococcus Faecium in Pet Food Industry. Am J Biomed Sci & Res. 2019 - 5(6). AJBSR. MS.ID.000972. DOI:* 10.34297/AJBSR.2019.05.000972.

Received: October 17, 2019; **Published:** October 23, 2019

Abstract

Freeze-dried *Enterococcus faecium* may able to lead a double life in pet food industry. Freeze-dried *Enterococcus faecium* can be identified both as a substitute for *Salmonella* in thermal processing validations and a special form of probiotics in formulations of pet foods. For the first life, freeze-dried *Enterococcus faecium* can be used as an indicator since its high thermal tolerance during processing, which is able to validate the inactivation efficiency of thermal processing in pet food industry, such as extrusion cooking. For a second life, freeze-dried *Enterococcus faecium* can be used as a special form of probiotics and added at the beginning of the process maintaining considerable number of variable cells, or also, can be directly added into pet foods without increasing the moisture of final products. It can be asserted that the double life of freeze-dried *Enterococcus faecium* satisfied both quality and nutrition aspects concerned in pet food industry.

Keywords: *Enterococcus Faecium*; Freeze-Dried; Validation; Probiotic; Pet Foods

Introduction

Enterococcus faecium shows ability to withstand extreme environmental conditions such as heat and salinity levels and antibiotic resistant. Reports have shown that *Enterococcus faecium* can be used as a surrogate for Salmonella during thermal processing, such as almond process validation [1], radio-frequency pasteurization [2-4], which can help to validate their inactivation efficiency to improve the safety condition of thermally processed foods.

Enterococcus faecium is a species of bacteria that naturally constitutes a major population in the gut as part of the normal gastrointestinal microbial flora in animals and humans. *Enterococcus faecium* has been used as a probiotic in some pet foods, which provides as a digestive aid and health benefits by increasing the number of healthy bacteria in the stomach [5-8]. In the Dog and Cat Food Ingredient Center database, at least 26 dry dog foods list *Enterococcus faecium* on their ingredient decks, including products from Orijen, Acana and Taste of the Wild [9].

Freeze-dried *Enterococcus faecium* has high thermal resistance, low-moisture content, long shelf-life, which has the potential to become both as a surrogate for process validation and probiotics addictive for pet foods. In this paper, a description of the double life of freeze-dried *Enterococcus faecium* in pet food industry is present.

Freeze-Drying Technology

Freeze-drying is currently an industrial standard for preservation of microorganisms [10,11]. However, freeze-drying can cause cells death and injury because of the changes in the physical state of the membrane lipids and protein damage or protein denaturation. However, injured cells retain their viability and upon resuscitation under a nutritionally adequate environment [12]. Factors associated with freeze-drying process (freezing speed, temperature, protectants, etc.), rehydration process (rehydration medium, temperature, PH, etc.), and storage conditions (chances to touch moisture, oxygen, and storage temperature) can influence the survival of bacteria after freeze-drying [13,14].

Protective agents during freeze-drying can be used to reduce the number of dead cells, which can protect bacterial cells during the freeze-drying process and further storage. Many studies have found that protectants can prevent fusion and thermotropic phase transitions of the cell membrane caused by freezing via electrostatic repulsion or steric hindrance [15]. Substances such as sugars,

polymers, polyols and amino acids have been investigated for their protective effect during freeze-drying (Abadias, et al. 2001). [16] found that skim milk serves as a good protectant for *Enterococcus faecium* during the freeze-drying process. When the bacteria were freeze-dried with skim milk, it survived and remained heat resistant longer than bacteria protected by a buffer specifically meant for freeze-drying.

Freeze-Dried *Enterococcus faecium* as a surrogate

About 95% of dry pet foods are produced by extrusion cooking technology [17]. There are multiple numbers of outbreaks of salmonellosis have been associated with extruded pet foods [18,19]. In this case, the inactivation efficiency of extrusion technology is needed to be validated by using a nonpathogenic, heat resistance, surrogate microorganism, such as *Enterococcus faecium.* Several studies have reported the ability of using *Enterococcus faecium* as a surrogate of *Salmonella* to validate extrusion processing [20-22]. found that *Enterococcus faecium* was inactivated at higher temperatures than Salmonella, indicating that its use as a surrogate would provide an appropriate margin of error in extrusion processes designed to eliminate this pathogen. The ability of using freeze-dried *Enterococcus faecium* as a surrogate for validation was firstly reported by Xu et al. Xu found that freeze-dried *Enterococcus faecium* has a comparable thermal resistance with its liquid form and can be used as an indicator for radio-frequency pasteurization validation of wheat flour [23].

Freeze-Dried *Enterococcus faecium* as probiotics

Freeze-dried *Enterococcus faecium* can survive better during extrusion process since its high thermal tolerance. Extrusion cooking is a process with thermomechanical treatment involving high-temperature conditions as high as 135 °C [24]. If *Enterococcus faecium* is included in the pet food or treat recipe before extrusion, the probiotic could be killed by the heat, or reduced in effectiveness [9]. Freeze-dried *Enterococcus faecium* with protectant showed a fairly good resistance to heat, which provided a more heat resistant way of including the bacteria could improve the probiotic quality of the final products [9,23].

Freeze-dried *Enterococcus faecium* can be used as a special form of probiotics without introducing extra moisture to the final products. The introduction of liquid inoculants can change the physical characteristics of foods and requires extra waiting while the product returns to its intended moisture level. By using dry-form probiotic, the changes in food properties can be avoided and the need of extra time for re-equilibration can be eliminated.

Freeze-dried *Enterococcus faecium* is storage stable and remain viable during long time storage [23]. The cell counts for many probiotic microorganisms may fall away completely within one or two days, resulting a big issue incorporating probiotic microorganisms into foodstuffs [25]. Developed a ready-to-eat cereal product includes a coating or filling containing a probiotic micro-organism. The probiotic microorganism was spray-dried with a suitable carrier substrate and has remain viable during long time storage. Compared with coating method, freeze-dried microbes are shelf stable, and faster and easier to prepare.

Conclusions

Freeze-dried *Enterococcus faecium*, with a high tolerant to thermal treatment, can be used as a surrogate of Salmonella for processing validation in pet industry, or, as a special form probiotics of pet foods added at the beginning of the process or directly added into final products. However, the functionality of *Enterococcus faecium* after freeze-drying has to be further confirmed. Further systematic studies are needed for its application in pet food area.

Acknowledgements

We acknowledge the support from Dr. Juming Tang, regent professor of Department of Biological Systems Engineering in Washington State University, for providing all the facilities and financial supports on author Jie Xu's studies on freeze-dried *Enterococcus faecium*. Special appreciation for Tim Wall, a reporter from PetfoodIndustry.com is also acknowledged.

Conflict of interest

There is no conflict of interest among Authors and Co-authors and have no conflict of interest to declare.

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