Impact of different cryoprotectants on the survival of freeze-dried *Lactobacillus rhamnosus* and *Lactobacillus casei/paracasei* during long-term storage

Beneficial Microbes (2015) 6, 381-386

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Abstract

The production of long shelf-life highly concentrated dried probiotic/starter cultures is of paramount importance for the food industry. The aim of the present study was to evaluate the protective effect of glucose, lactose, trehalose, and skim milk applied alone or combined upon the survival of potentially probiotic Lactobacillus rhamnosus CTC1679, Lactobacillus casei/paracasei CTC1677 and L. casei/paracasei CTC1678 during freeze-drying and after 39 weeks of storage at 4 and 22 °C. Immediately after freeze-drying, the percentage of survivors was very high (\geq 94%) and only slight differences were observed among strains and cryoprotectants. In contrast, during storage, survival in the dried state depended on the cryoprotectant, temperature and strain. For all the protectants assayed, the stability of the cultures was remarkably higher when stored under refrigeration (4 degrees C). Under these conditions, skim milk alone or supplemented with trehalose or lactose showed the best performance (reductions $\leq 0.9 \log$ units after 39 weeks of storage). The lowest survival was observed during non-refrigerated storage and with glucose and glucose plus milk; no viable cells left at the end of the storage period. Thus, freeze-drying in the presence of appropriate cryoprotectants allows the production of long shelf-life highly concentrated dried cultures ready for incorporation in high numbers into food products as starter/potential probiotic cultures.