Interaction of resident microbiome and *Listeria* on pears during cold storage



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Summary

Although there have been no listeriosis outbreaks associated with fresh whole pears, recent recalls and outbreaks associated with other tree fruits due to potential *Listeria monocytogenes* contamination have prompted the pear industry to implement validated interventions and proactive measures to minimize post-harvest risks. The microbiome of pear surfaces is thought to play a role in the persistence of foodborne pathogens. However, there is a general lack of knowledge on the fate of *Listeria* on pears during storage, as well as the dynamic changes of the pear microbiome over long-term cold storage, and the impact of such changes on the survival of *Listeria* on pears. The overall objective is to evaluate the temporal changes of the microbiome and its interactions with *Listeria* on pears.

Objectives

- 1. Examine the fate of Listeria and temporal changes of the resident microbiome on pears and their interactions during long-term cold storage.
- 2. Evaluate the impacts of organic practices on the microbiome community and persistence of *Listeria* on pears of the selected varieties during long-term storage.

Methods

Unwaxed pears of the selected varieties at commercial maturity will be inoculated with *L. innocua*, a non-pathogenic surrogate of *L. monocytogenes*, and both inoculated and uninoculated pears will be stored in commercial cold storage facilities for up to 9 months, following the pear industry's storage practices. Pears will be sampled at 0, 3, 6,12, 24, and 36 weeks of storage to examine temporal changes of the microbiome using metagenomic sequencing. The die-off of *Listeria* on pears and the interactions between *L. innocua* and the resident microbiome will also be examined. Total resident bacterial counts, yeast/mold counts, and the selected resident microbes will be further enumerated at each sampling point. Fruit quality attributes will be assessed during storage and correlated with microbial data.

Results to Date

The research team has developed a standardized protocol for inoculation, establishment, and enumeration of *Listeria* on pears to ensure a consistent initial inoculation level of three selected pear varieties using a 3-strain *Listeria* cocktail (**Figures 1** and **2**). To further investigate the behavior of *Listeria* on pear varieties, a 12-week cold storage study was conducted in the BSL2 lab, which revealed that *Listeria* counts dropped rapidly during the first 2 weeks of storage, resulting in reductions of $1.4-1.8 \log_{10} \text{CFU/pear}$ (**Figure 3**). Subsequently, the *Listeria* populations on pears remained relatively stable, resulting in reductions of $2.4 \log_{10} \text{CFU/pear}$ at 12 weeks of storage. Additionally, similar *Listeria* survival rates were observed among the pear varieties evaluated (**Figure 3**).

Benefits to the Industry

Successful completion of this project will provide the fresh pear industry with a better understanding of *Listeria* survival on pear surfaces, and interactions with indigenous microbiota on pear surfaces, offering vital information to optimize agricultural practices and storage conditions to ensure microbiological safety.

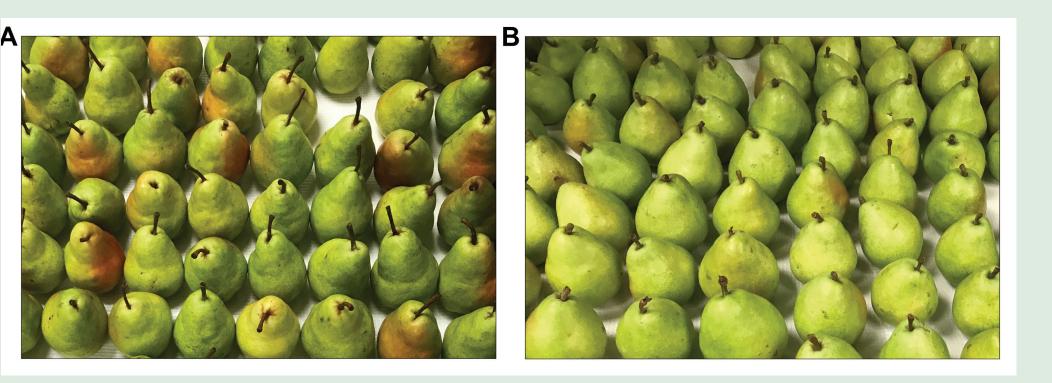


Figure 1. Representative images of pears inoculated with *L. monocytogenes*. A: Bartlett pear; B: D'Anjou pear.

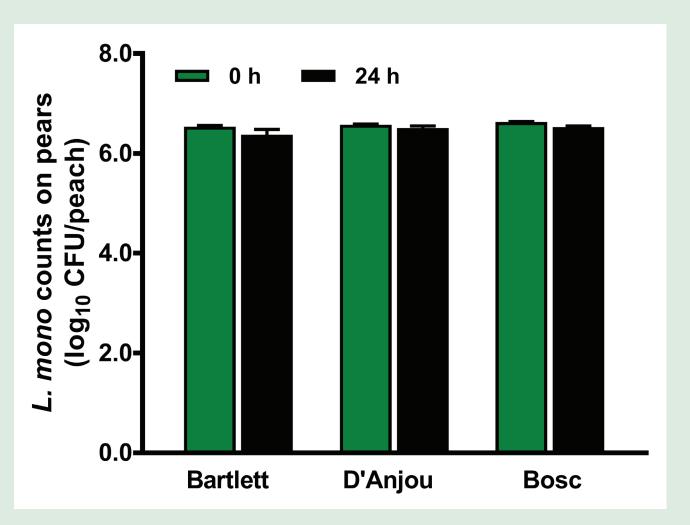


Figure 2. Initial inoculation level of *L. monocytogenes* on pears of selected varieties. Mean ± SEM, n=12. Oh: right after inoculation; 24h: 24h after inoculation. Three-strain *L. monocytogenes* cocktail was used for inoculation

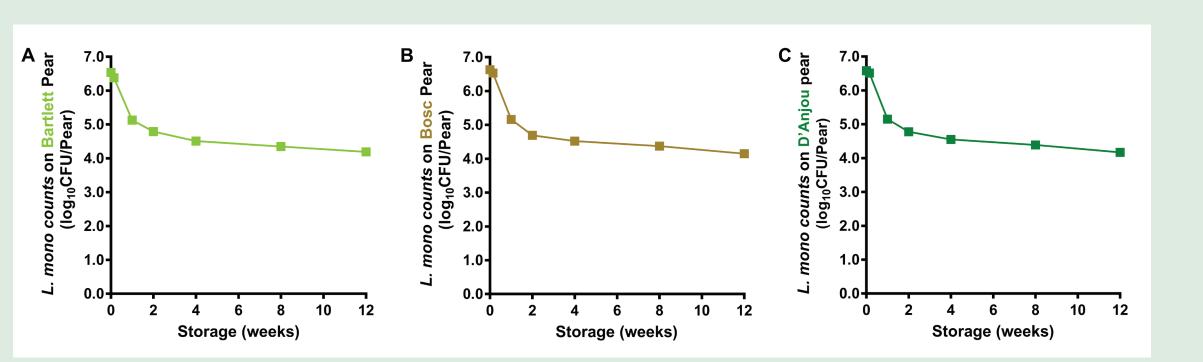


Figure 3. Survival of *L. monocytogenes* on pears during 12 weeks of cold storage. A: Bartlett pear; B. Bosc pear; C: D'Anjou pear. Mean \pm SEM, n = 12. The selected varieties were inoculated with *L. monocytogenes* at ~6.5 \log_{10} CFU/apple, then subjected to cold storage 24 hours after inoculation.