# Control of *Staphylococcus aureus* During Biltong Production

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## I. INTRODUCTION

Biltong is a dried meat product native to South Africa. Unlike jerky, biltong is dried at ambient temperatures and relies on salt concentration and acidity to inactivate pathogens. Throughout history, different cultures, countries, and methods have been used to dry meat, many of which are still used today [1]. Drying meat removes water which prevents enzymes and bacteria from reacting with the food [2]. Salt is commonly used to preserve meat as it reduces water activity and contributes flavor [3]. Biltong is made from strips of meat, typically beef. It can be dipped in spices and acidic liquids like vinegar, before drying to add flavor but to also provide antimicrobial activity [3]. Traditional South African dried biltong is prepared by hanging the strips of meat outside for one to two weeks [3] but is made commercially under more controlled conditions.

*Staphylococcus aureus* is commonly found in foods exposed to ambient to higher temperatures along with frequent handling, and it is associated with dried meat products due to its tolerance for low water activity ($a_w$) and high salt concentrations. With the increasing popularity of biltong worldwide [3], it is critical to understand potential microbial risk, and how it may be managed. In the United States, *S. aureus* guidelines for final ready-to-eat meat products state that no more than 2 logs of growth should occur during processing, which includes drying [5]. Due to an acidic marinade and dip, it is essential to consider if acid stress adaption may potentially increase resiliency to treatment. The objective of this study is to 1) evaluate the effectiveness of acid marination and drying of biltong to control *S. aureus*, and 2) determine if prior adaptation to acid stress increased resiliency in *S. aureus*.

## II. MATERIALS AND METHODS

Biltong strips (100 ± 5.0 g; 2 cm x 6 cm x 8 cm) were cut from beef round. Stationary phase *S. aureus* strains (ATCC12600, 13565, and 25923) were used as the inoculum. Each trial contained two pathogen groups, acid stressed *S. aureus* (grown in tryptic soy broth (TSB) with 1% glucose solution) and unstressed *S. aureus* (grown in TSB only). Each biltong strip was inoculated to a target concentration of 8 log cfu/g. After 30 min attachment, each strip was dipped in a 5% lactic acid solution for 5 seconds. Samples were plated before lactic acid dip and immediately after lactic acid dip. After overnight refrigeration, strips were added to a marinade solution of vinegar (2% v/w), salt (2% w/w), black pepper (1% w/w), brown sugar (1.5% w/w), and coriander (1% w/w) and vacuum sealed. Strips were marinated for 24 h at 4°C, then neutralized with 1% w/w sodium bicarbonate for 10 min. Strips were then hung in a Biltong box maintained at 27°C and 55% humidity. From each pathogen group, two strips were taken for enumeration each day for 7 d. Serial dilutions were mixed with molten TSA and overlayed onto Baird Parker agar. Plates were incubated for 20-25 h at 35°C and enumerated. Water activity was measured daily on non-inoculated strips using a water activity meter (Aqua Lab CX-2, Decagon Devices, Inc., Pullman, WA, USA). Microbial inactivation data was fitted with reparametrized Weibull inactivation models using the gnls function in the nlme package in R 4.3.1.

## III. RESULTS AND DISCUSSION

Combined lactic acid dip, marination, and 7 d of drying resulted in 3.5±0.24 log cfu/g reduction ($P<0.0001$) with unstressed *S. aureus.* While reductions were less than the recommended 5 log reduction for *Salmonella*, any reduction is well below processing RTE guidelines for *S. aureus* (<2 logs of growth). A

concern is that acid-adapted *S. aureus* may be more resilient to the biltong process [4]. However, in this study, stress adapted *S. aureus* were much less resilient to treatment (5.1±0.25 log cfu/g reduction, P<0.001) compared to non acid stressed *S. aureus*.

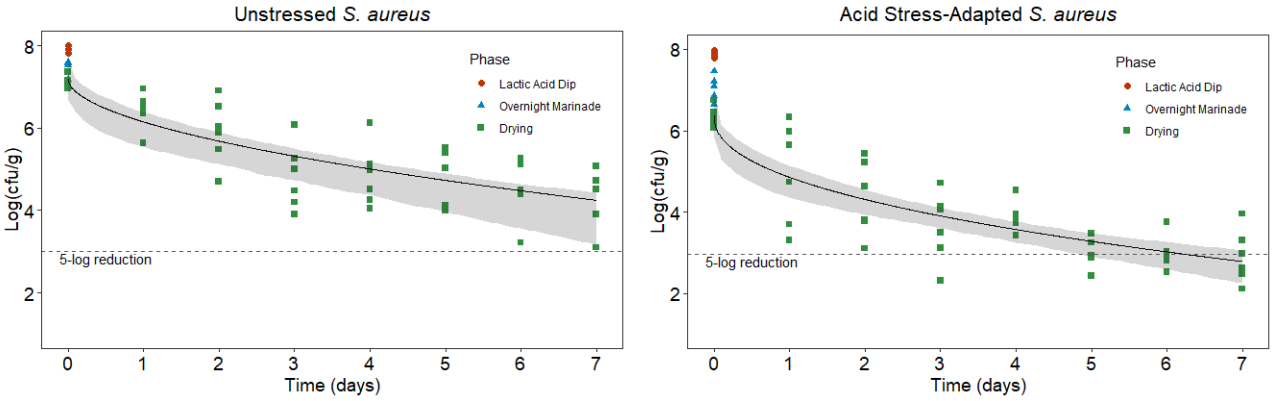


**Figure 1.** *Inactivation of Staphylococcus aureus* after lactic acid dip, acid marination, and 7d drying. Unstressed *S aureus* were 18h stationary phase cultures, whereas acid stress adapted included 1% glucose in the grow up to lower the pH from 7.0 to 4.0. Black line indicates best fit by Weibull model. Shaded area indicates 95% confidence interval.

## IV. CONCLUSION

Risk of *S. aureus* growth and toxin production during biltong manufacture is very unlikely. Acid adaption did not impart any additional resilience compared to stationary phase inoculum suggesting stationary phase cultures are adequate to assess *S. aureus* control in acid-marinated beef products. Future studies should assess the taste of biltong without the neutralization step after marination. Researchers should also explore microbial lethality without sodium bicarbonate neutralization to evaluate potential improvements in food safety.

## REFERENCES


1. Aykın Dinçer, E. (2023). Dried meat products obtained by different methods from past to present. *Food Reviews International*, *39*(5), 2457–2476.
2. United States Dept. of Agriculture. Food Safety and Inspection Service. (2021). *FSIS cooking guideline for meat and poultry products.* (Washington, DC). https://www.fsis.usda.gov/sites/default/files/media_file/2021-12/Appendix-A.pdf
3. Jones, M., Arnaud, E., Gouws, P., & Hoffman, L. C. (2017). Processing of south African biltong—A review. *South African Journal of Animal Science*, *47*(6), 743–757. https://doi.org/10.4314/sajas.v47i6.2
4. Gavai, Kavya, Caitlin Karolenko, and Peter M. Muriana. (2022). "Effect of Biltong Dried Beef Processing on the Reduction of *Listeria monocytogenes*, *E. coli* O157:H7, and *Staphylococcus aureus*, and the Contribution of the Major Marinade Components" *Microorganisms* 10, no. 7: 1308. https://doi.org/10.3390/microorganisms10071308
5. United States Dept. of Agriculture. FSIS Directive - Food Safety and Inspection Service. (2017). *Verification procedures for lethality and stabilization.* (Washington, DC). https://www.fsis.usda.gov/sites/default/files/media_file/2020-07/7111.1.pdf