Combined boiling and irradiation treatment on the shelf life and safety of Ready-To-Eat bovine tripe

By

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Declaration

I declare that the dissertation herewith submitted for the degree MSc Food Science at the University of Pretoria, has not previously been submitted by me for a degree at any other university or institution of higher education.

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Dedication

I dedicate this dissertation to my parents, Hector and Irene Parry-Hanson, and to my siblings, Freda, Michael and Jocelyn Parry-Hanson.
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ABSTRACT

COMBINED BOILING AND IRRADIATION TREATMENT ON THE SHELF LIFE AND SAFETY OF READY-TO-EAT BOVINE TRIPE

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Bovine tripe is not optimally used in South Africa because it is highly perishable, it is not easily accessible and it requires long cooking time. For these reasons, ready-to-eat (RTE) technology was used to process tripe. This study was thus undertaken to determine the effect of vacuum packaging and boiling in combination with gamma irradiation at a target dose of 9 kGy on the microbiological safety, with respect to inoculated Clostridium perfringens ATCC 13124 spores, and microbiological quality, with respect to aerobic plate counts (APC) and aerobic spore counts (ASC), of RTE bovine tripe during storage at 5 and 15 °C for 14 days. Irradiation dosage of 9 kGy was chosen as an appropriate dosage to eliminate bacterial spores of C. perfringens and aerobic spores that were present on inoculated tripe. Also, transmission electron microscopy (TEM) was conducted on C. perfringens ATCC 13124 spores to determine whether boiling and gamma irradiation has a synergistic effect on C. perfringens spore structure.

In order to maintain sensory properties of tripe, mild preservation treatments were used in the processing of RTE tripe. For this reason, the following hurdles were employed in the processing of RTE tripe: boiling, vacuum packaging, gamma irradiation and chilled storage. Prior to boiling, rough washed tripe was tenderized with papain to reduce the cooking time of raw bovine tripe.
In Phase 1, the fresh tripe was processed as a *sous-vide* RTE product. The washed and papain treated tripe was inoculated in vacuum bags, sealed, boiled in the vacuum bags and gamma irradiated at a target dose of 9 kGy (10±1 ºC). Despite vacuum packaging the raw tripe prior to boiling and irradiation, aerobic conditions prevailed due to the presence of residual oxygen in the RTE tripe packs. This resulted in inhibition of *C. perfringens* after boiling. The fresh tripe had a high microbiological load of 8.6 \( \log_{10} \text{cfu/g} \) for both APC and ASC and 4.5 \( \log_{10} \text{cfu/g} \) CC due to the high levels of microorganisms naturally present in the ruminant stomach. Although boiling significantly reduced APC and ASC, their levels (6.3 \( \log_{10} \text{cfu/g} \) for APC and 6.1 \( \log_{10} \text{cfu/g} \) for ASC) remained high after boiling probably due to the presence of heat resistant spores. Although irradiation significantly reduced APC and ASC on *sous-vide* RTE tripe, aerobic bacteria and aerobic spores showed high resistance to gamma irradiation, with *ca* 4 \( \log_{10} \text{cfu/g} \) bacteria surviving on irradiated RTE tripe. Storage at 5 ºC inhibited increase of APC and ASC on both irradiated and control RTE tripe samples, thus extending the shelf life of RTE tripe to at least 14 days. However, rapid growth occurred in 0 kGy RTE tripe stored at 15 ºC. By day 7, APC and ASC on 0 kGy samples stored at 15 ºC had exceeded 7 \( \log_{10} \text{cfu/g} \) and was thus considered spoiled. Low APC and ASC were maintained in irradiated samples stored at 15 ºC throughout 14 days of storage. Therefore, in an aerobic environment, irradiation of RTE tripe and storage at 5 ºC is required to reduce microbiological counts and extend the shelf life of RTE tripe to at least 14 days.

In Phase 2, tripe was boiled prior to inoculation with 7 \( \log_{10} \text{cfu/g} \) *C. perfringens* ATCC 13124 spores, vacuum packaged, irradiated at a target dose of 9 kGy (10±1 ºC) and stored for 7 days at 5 and 15 ºC. The change in processing of RTE tripe in Phase 2 was done to eliminate the residual oxygen in Phase 1 to create appropriate growth conditions for inoculated *C. perfringens* spores. Gamma irradiation significantly reduced *C. perfringens* and APC below detection (detection limit of 1 \( \log_{10} \text{cfu/g} \)) throughout 7 days of storage at 5 and 15 ºC. However, aerobic bacteria re-emerged on irradiated samples during storage at 5 and 15 ºC due to repair of irradiation injury.
TEM of boiled and irradiated *C. perfringens* ATCC 13124 spores showed that boiling alone caused reduction of spore material possibly due to initiation of germination. Gamma irradiation alone caused elongation of *C. perfringens* spores indicating that although germination occurred, outgrowth was inhibited. Boiling and gamma irradiation had a synergistic effect on *C. perfringens* spores as indicated by complete loss *C. perfringens* spore material.

However, due to the high levels of aerobic spores and aerobic bacteria that persisted on irradiated RTE tripe in Phase 1, *sous-vide* processing of RTE tripe is not recommended because it might be unsafe for consumption when pathogenic *Bacillus cereus* spores are present. Irradiated RTE bovine tripe in an anaerobic environment (Phase 2), proved to be safe for consumption throughout 7 days of storage regardless of the storage temperature.
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1.3 PROBLEM STATEMENT

Bovine tripe is not optimally utilized in South Africa for a number of different reasons. It is not easily accessible to consumers since it is only available at abattoirs and selected retailers; it requires long cooking times (Flynn and Fox, 1981) to tenderize the collagen and elastic fibers; and raw tripe is highly perishable due to the presence of gut microorganisms and autolytic enzymes (van den Hurven, 1977), therefore causing economic losses to suppliers when demand is low. Since tripe is still enjoyed by all age groups especially among the African populations in South Africa, it is anticipated that if made available as a chilled Ready-To-Eat (RTE) product, it will be well patronized