INACTIVATION OF TOXOPLASMA GONDII IN MEAT SAMPLES

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I. INTRODUCTION

Toxoplasma gondii is one of the most important zoonotic parasites in the world and virtually all warm-blooded animals, including humans, mammals and birds can be infected by this pathogen (Dubey, 2009). Globally, *T. gondii* is ranked fourth out of 24 foodborne parasites (World Health Organization, 2015). *T. gondii* is the causative agent of toxoplasmosis in humans. Pregnant women and severely immune-compromised persons are at risk for serious complications due to congenital transmission or disseminated infection. Consumption of raw and undercooked meat is considered as an important source of *T. gondii* infections. However, most non-heated meat products contain salt and additives, which affect *T. gondii* viability. The mouse bioassay is the standard method for testing of samples on viable *T. gondii* parasites, but ethically undesirable, costly and time consuming. It was our aim to develop an in vitro method to substitute the mouse bioassay for determining the effect of salting on *T. gondii* viability.

II. MATERIALS AND MTHODS

Processing experiments were performed with minced meat from sheep that were experimentally infected with *T. gondii* oocysts (Opsteegh et al. 2024). Portions of 50 g infected sheep meat were supplemented with 0.6 - 2.7 g NaCl/100 g. In addition, a series of 1.2 g NaCl/100g was set up supplemented with 1.4 or 1.8 g sodium lactate/100g and one combination with 1.2 g NaCl/100 g supplemented with 1.4 g sodium lactate and 0.4 g sodium acetate/100g of sheep meat. Besides those samples, untreated sheep meat, *T. gondii* free meat and frozen sheep meat samples were used as control samples. All meat samples were tested twice and in duplicate, some even in quadruplicate. The cell culture method (Opsteegh et al. 2024) and the mouse bioassay were used to determine effect of salting on the viability of *T gondii.*

III. RESULTS AND DISCUSSION

The results with the different salt concentrations showed differences in growth of *T. gondii*. In samples with NaCl concentrations of 1.5 g/100 g or higher, no viable *T. gondii* was detected by cell culture and by mouse bioassay. When sodium lactate (with or without sodium acetate) was added, no viable *T. gondii* was detected at the concentration of 1.2% NaCl or higher. The cell culture method showed the same results as the mouse bioassay (Tables 1 and 2).

Table 1 – Detection of *T. gondii* by cell culture and by mouse bioassay in infected sheep meat supplemented with different salt concentration after 20 h incubation.

	Test week 1		**Test week 2**	
Duplicate samples	Cell culture	Mouse bioassay	Cell culture	Mouse bioassay

Negative	-	-	-	-
Positive	+	+	+	+
0.6 -0.9 g NaCl/100 g	+	+/-	+	+
1.2 g NaCl/100 g	-	-	+	+
1.5 -2.7 g NaCl/100 g	-	-	-	-

+ means growth on both duplicates +/- means growth on one of the duplicates
- means no growth on both duplicates

Table 2 – Detection of *T. gondii* by cell-culture method and mouse bio assay in infected sheep meat supplemented with different concentrations of additives after 20 h incubation.

	Test week 3		**Test week 4**	
quadruplicate samples	Cell culture	Mouse bio assay	Cell culture	Mouse bio assay
Negative	-	-	-	-
Positive	+	+	+	+
1.2 g NaCl/100 g	+	+	+	+
1.2 g NaCl + 1.4 g Na-lac/100 g	-	-	-	-
1.2 g NaCl + 1.8 g Na-lac /100 g	-	-	-	-
1.2 g NaCl + 1.4 g Na-lac + 0.4 g Na-ac /100 g	-	-	-	-

- means no growth on the quadruplicates, + means growth on the quadruplicates

IV. CONCLUSION

The level of NaCl concentration present in meat products affects the viability of *T. gondii* in minced meat. With the addition of Na lactate, negative effects on *T. gondii* were obtained at a lower NaCl concentration. The developed cell culture method can successfully be used to detect viable *T. gondii* in tissues of experimentally infected sheep and replace mouse bioassay in these types of inactivation experiments.

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