

## **P.52.- Relationship between rendering process temperatures and DNA degradation in Meat and Bone Meals by Real Time PCR assay**

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Several methods for the detection of Processed Animal Proteins (PAPs), based on the Polymerase Chain Reaction (PCR) technique have been described to implement taxon-specific identification, presently not fully supported by microscopic analysis. Most of PCRs report qualitative data, with performances expressed as detection limits. Nevertheless, their performances could depend on the integrity of the target DNA, influenced by the different rendering processes. This has prompted us to set up a quantitative Real Time PCR to evaluate the target DNA degradation, on a defined MBM, processed at 133°, 137°, 141° and 145°C, fulfilling the present legislation (20 min,  $\geq 3$  bar).

To quantify the target mitochondrial DNA specific for ruminant, we use an exogenous target control. It consists of an exogenous DNA used as internal positive control also for the evaluation of the DNA extraction efficiency. Such approach is mandatory because the absence in MBMs of an endogenous target control. Our results demonstrate that the method could detect ruminant specific DNA also in 145°C treated MBM. The DNA determination indicates that with the increase of the temperatures, the quantity of DNA that can be amplified decreases with an exponential factor. In particular, in the range analysed, we observe a two log decrease of the amplified DNA amount. between 133°C and 145°C treated MBM.

Owing to the above, the assessment of PCR method performances (such as detection limit and quantification) are greatly affected by the kind of MBM tested. Therefore, it must be considered that the quantification of MBM in feeds, by PCR techniques, must be related to the rendering process. For official control purposes, this fact could not allow to use PCR results to discriminate an intentional addition of MBM in feeds from a possible occasional contamination. On the other side, PCR could help within the same rendering plant to monitor the effectiveness of the process, by evaluating the progressive DNA degradation under the present legislative frame

### ***Keywords***

*PCR, DNA degradation, MBM*