

## QUANTITATIVE DETERMINATION OF SUNFLOWER IN MIXED CONCENTRATE FEEDS BY REAL TIME PCR

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### ABSTRACT

Feeding farm animals such as, high-yielding dairy, poultry, swine and beef cattle at certain periods with a balanced and adequate mixed concentrate feed is a must. Concentrate feed is produced from grain feed materials that goes through various processes and mixed with oilseed meals, food industry by-products and feed additives. The aim of this study was to determine proportional amounts of sunflower in mixed concentrate feeds by DNA-based Real Time Polymerase Chain Reaction (RT-PCR) in order to monitor compliances of mixed feed labels, investigate the appropriateness of the required quality criteria, detect and avoid unintended feed materials that are mixed in imported plant raw materials. For this purpose, concentrate feed samples that were prepared at the laboratory by weight to weight and also the samples with known ingredients from a local feed plant were used to test quantity of sunflower. Genomic DNA (gDNA) extracted using commercial kits and evaluated for quality and quantity prior to use in a PCR assay. For the specific detection of sunflower and taxon specific (for the relative quantification) a fragment of the *helianthinin* and *actin* genes were selected respectively using gene specific primers and probes. The calibration curve was established on the basis of five samples. The average value of the slope of the standard curve was within the range of -3.1 to -3.6, and the R<sup>2</sup> coefficient was  $\geq 0.98$ . This study, first time showed that sunflower in a mixed concentrate feed can be quantified by DNA-based RT-PCR with a high precision.

**Key Words :** Mixed Concentrate Feeds, Sunflower, RT-PCR, Quantitative Determination