PROJECT REVIEW

"Clenbuterol in meat: a source for a positive doping control? Search for analytical strategy to distinguish abuse from meat contamination"

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Clenbuterol is a well known beta-agonist which is being used in animal husbandry and sports for growth promoting purposes. Lately adverse analytical findings in athletes were claimed to be due to contaminated meat. This project is aimed at finding a strategy and marker to distinguish Clenbuterol in urine from athletes due to meat consumption or illegal use of Clenbuterol containing supplement or preparations.

The hypothesis is that Clenbuterol, a racemic (1:1) mixture of stereoisomers as a pure compound, is present in another composition, 3:1, in meat due to differences in pharmacokinetics and pharmacodynamics. Ingestion of this different ratio from meat can lead to a different ratio in athletes urine. If this is the case then it is possible to discriminate consumption of contaminated meat from illegal administration of Clenbuterol (supplements). For this, methods of analysis have to be developed to separate the stereoisomers in supplements, meat and urine.

Extraction of Clenbuterol from meat must be optimised because the incidence of positive Clenbuterol findings in residue control is very low. And several poisoning cases and positive doping cases have been found. So there is a possible underestimation of Clenbuterol residues in meat due to possible bound residues or incompete extraction.

Information on levels of the different isomers in meats has to be collected. As also quantification of stereoisomers in preparations and urine samples from controlled studies or from inadverted use from eg. tourists.

After that a proposal, workflow, how to use these tools in doping control must be developed.

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Results and Conclusion

WADA funded project 11A18SS was written to show a proof of principle that it is possible to discriminate adverse analytical findings by clenbuterol illegal use from consumption of contaminated meat.

Tools for this proof of principle were developed.

Sensitive and selective analytical methods were developed using UHPLC-MS/MS and SFC-MS/MS for meat, bovine and human urine and preparations.

Incurred meat with clenbuterol was produced.

The tools were used on real samples provided by anti-doping laboratories.

It was shown that meat from an animal treated with clenbuterol contained a different S/R ratio for clenbuterol. The meat was R-enantiomer enriched.

Pharmaceutical preparations showed S/R ratio around 1.

From the small number of urine samples from Mexico, presumed to be contaminated via meat the S/R clenbuterol ratio were lower than the S/R clenbuterol ratio from humans having consumed clenbuterol in an administration trial.

Publications and presentations

- Lecture at Manfred-Donike Workshop, Cologne 2013, 26.02.2013, title "Enantiomeric separation of clenbuterol as analytical strategy to distinguish abuse from meat contamination"
- Poster presentation HPLC 2013, 16-20 June, Amsterdam, Netherlands titled: "Separation of clenbuterol enantiomers – A comparison between HPLC and SFC-MS/MS".
- Poster presentation at RAFA 2013, 5-8 November 2013, Prague, Czech republic titled: "Enantiomeric separation of clenbuterol as analytical strategy to distinguish abuse from meat contamination"
- Article: Parr MK, Blokland MH, Liebetrau F, Schmidt AH, Schänzer W, Sterk SS. Enantiomeric separation of clenbuterol as analytical strategy to distinguish abuse from meat contamination. In: Schänzer W, Geyer H, Gotzmann A, Mareck U (eds). Recent advances in doping analysis (21).

Sportverlag Strauß, Köln (2013) in press.
- Parr MK, Blokland MH, Schmidt AH, Liebetrau F, Stanic M, Witte P, Sterk SS. Supercritical fluid chromatography versus high performance liquid chromatography for determination of Clenbuterol enantiomers – a comparative trial, Journal of Analytical methods, submitted, March 2014.
- Sterk SS, Liebetrau F, Meijer T, Schmidt AH, Blokland MH, Parr MK. Clenbuterol in meat: A source for a positive doping control? Search for analytical strategy to distinguish abuse from meat contamination, Food Additives and Contaminants, in preparation.
- Newspaper article: Matthias Thiele. Gedopt oder Steak gegessen? Newspaper report in „Der Tagesspiegel" June 08, 2013, B3.