Doping abuse or meat contamination?
Clenbuterol enantiomer ratio determination

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**Introduction**

In recent years clenbuterol was detected during routine doping controls of several athletes (Thevis et al, 2013). The majority of them claimed it came into their body via consumption of clenbuterol contaminated meat. Clenbuterol is a chiral molecule allowing for two enantiomers, R(−)- and S(+) -clenbuterol, which consist of a racemic mixture in human and veterinarian preparations. Previous work shows that this ratio changes in meat when clenbuterol is administered to animals. A human study was performed to determine if this change in ratio is also visible in human urine after consuming contaminated meat. 14 volunteers were administered clenbuterol by eating contaminated meat and liver or by ingesting clenbuterol preparations. Urine samples were collected and measured using a chiral SFC-MS/MS method according Parr et al (2013).

**Methods**

![Figure 1. Enantiomers of clenbuterol](image1.png)

- 1 gram meat
- Extraction with acetonitrile
- SPE with Stata-AC
- Reconstitute in MeOH
- LC-MS/MS Chiral separation
- 3 ml urine
- Extraction 2x5 ml TBME
- Reconstitute in MeOH
- SFC-MS/MS Chiral separation

Figure 2. Scheme of meat and urine sample preparation

**Results**

![Figure 3. Excretion curve of a volunteer who consumed Spiropent®](image3.png)

Subject 2 (Spirolin®)
The concentration of clenbuterol as well as the proportion S(+) -clenbuterol is plotted against the time after administration in hours

![Figure 4. Excretion curve of a volunteer who consumed clenbuterol enriched liver](image4.png)

Subject 3 (liver)
The concentration of clenbuterol as well as the proportion S(+) -clenbuterol is plotted against the time after administration in hours

![Figure 5. Proportion of S(+) -clenbuterol in the urine samples of the subjects (1-12), pure reference standards (13-14), the contaminated muscle tissue and the contaminated liver](image5.png)

Conclusions

- Clenbuterol treated calves show enriched S-clenbuterol liver, but no enriched S-clenbuterol meat.
- Clenbuterol is detected in urine of volunteers after consumption of clenbuterol enriched meat or liver and intake of clenbuterol containing preparations.
- Enantiomeric composition (proportion S-clenbuterol) of liver volunteers deviates significantly from preparation volunteers.
- Enantiomeric composition not dependant of concentration or time.

**References**


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