

Multiple response optimization of a QuEChERS extraction and HPLC analysis of Coccidiostats compounds in chicken liver

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INTRODUCTION

Coccidiostats are veterinary compounds used for the treatment and prevention of coccidiosis, which threaten poultry farming. These compounds can persist in animal tissues after slaughter and therefore can enter the consumer's organism.

AIM

In order to reduce the volumes and mass of the reagents a multi-residue extraction method based on QuEChERS and HPLC-DAD-FLD were developed for the simultaneous determination of four coccidiostats, **toltrazuril (TOL)**, **decoquinate (DEC)**, **ethopabate (ETH)** and **robenidine (ROB)**.

METHODS

Optimized QuEChERS: 500 µL ACN/formic acid + 200 mg MgSO₄/NaCl + 5 mg PSA + 18mg C18

Conventional extractions

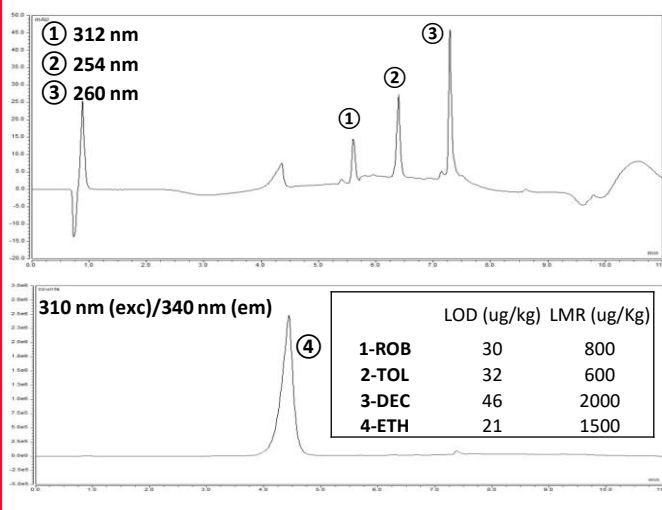
- 1-Non-optimized QuEChERS: 10mL ACN/MeOH/formic acid + 6MgSO₄/sodium citrate + 10mL Hexane.
- 2-Solid-Liquid (S-L): 5 mL ACN + 2mL Hexane.
- 3-Solid Phase extraction (SPE): 10 mL ACN + SPE alumina N + Strata X + 2mL Hexane.



Extractions comparison

%Recovery by HPLC-DAD-FLD + Eco-scale

HPLC-DAD-FLD: H₂O pH 3/ACN:MeOH 7:3 (gradient) – 12min



RESULTS

QuEChERS optimization



Factors:

MgSO₄/ NaCl; Na₂SO₄
Salt amount (200;500 mg)
ACN (500;1000 µL)
Formic acid (5;35 µL)
Mixing time (1;5 min)
PSA (5;50 mg)
C18 (0;10 mg)

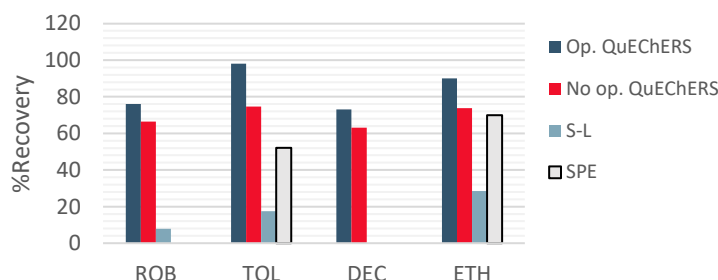
Factors

MgSO₄/NaCl (100-200 mg)
PSA (5-50mg)
C18 (18-30mg)

Simultaneous Desirability = 0,99

Responses: 4 Coccidiostats recoveries

Comparison of extraction methods



Eco-Scale calculation

100 – Penalty Points (Reagents + Instruments + Waste) = Result



Methods	Op. QuEChERS	Non-op. QuEChERS	S-L	SPE
Eco-Scale of Extraction	84	63	79	75

<50 poor; 75-50 acceptable; >75 excellent,

CONCLUSIONS

Using the optimized QuEChERS, in comparison with different extraction techniques, higher analyte recovery and Eco-scale values were obtained.

The analytical figures of merit, and the fact that four coccidiostats can be extracted by a simple technique and analyzed simultaneously by HPLC, prove that this method is suitable to be used in the routine analysis of poultry liver samples

REFERENCES

Silva, J. et. al. Multiple response optimization of a QuEChERS extraction and HPLC analysis of diclazuril, nicarbazin and lasalocid in chicken liver, Food Chemistry 311 (2020)