

4 June 2007

EUROPEAN COMMUNITY COMMENTS ON THE  
**PROPOSED DRAFT**  
**METHODS OF ANALYSIS FOR VETERINARY DRUG RESIDUES**

(CL 2007/04-RVDF)

*Mixed Competence*  
*European Community Vote*

The European Community and its Member States thank the delegation of Canada and the Netherlands for preparing this document.

**General comments**

We would like to re-iterate that the list of methods should be considered as a resource list. It is not intended to be a list of preferred or obligatory methods for Codex purposes. In consequence it has not the status of a Codex Standard, Guidelines or Recommendation but is maintained and updated by the Committee for use by Codex members.

Our comments on specific methods have to remain general not all methods are described in detail. It is difficult to assess the proficiency of a method on the limited description provided in the table attached to the Circular Letter. An effort should be made to specify the detection method used, e.g. LC/FLU (high-performance liquid chromatography with fluorescence detection), LC/DAD (liquid chromatography (LC)-diode array detection), LC/MS (Liquid chromatography - mass spectrometry system) or LC-MS/MS (Liquid chromatography - tandem mass spectrometry system).

We appreciate that an effort has been made to also mention simple detection methods in order to allow less advanced laboratories to identify suitable methods. Some of the methods mentioned for this reason, e.g. the thin-layer chromatography for the detection of sulphonamides can not be recommended without a caveat concerning their reliability. An effort should be made to indicate which methods are providing more precise results and are therefore preferable.

**Comments on particular methods for compounds or compound groups**

**Anthelmintics such as Avermectins: Abamectin, Doramectin, Eprinomectin, Ivermectin and Moxidectin:** Avermectins could be used in animals producing milk for human consumption. The matrix milk should therefore be examined in residue control programmes. The European Community Reference Laboratory in Berlin could provide a description of a validated method for avermectins in milk.

**Benzimidazoles, i.e. Albendazole, Febantel/Fenbendazole:** We suppose that 2-aminobenzimidazole means 2-amino albendazole sulphone. 2-amino-benzimidazole is not a metabolite of albendazole. Albendazole sulfoxide and albendazole sulphone are not taken into account. Although depletion studies and investigations of the metabolism of albendazole show that 2-amino albendazole sulphone is the primary metabolite, this does not apply before day 2 after a single oral dose. In the meantime,

especially after 1 to 2 days, albendazole sulfoxide and albendazole sulphone are the main metabolites.

The European Community Reference Laboratory in Berlin could provide a description of a validated multi-method (LC-MS/MS) which covers the main albendazole metabolites and allows (due to the omission of an oxidation step) detection of several benzimidazoles.

**Flubendazole:** (2-amino-1H-benzimidazole-5-yl)-(4-fluorophenyl) methanone should be added as a further marker residue. The European Community Reference Laboratory in Berlin could provide multi-methods (LC-MS/MS), which cover the main metabolites. For more recent reference see also: *De Ruyck, H., Daeseleire, E. Grjspeerdt, K., De Ridder, H., Van Renterghem, R., Huyghebaert J. Agric. Food Chem. 49 2001 610.*

Methods for the detection of **mebendazole**, **oxibendazole** and **triclabendazole** are not referred to on the list.

**Coccidiostats:** Some of the important compounds, e.g. the ionophores and halofuginone, are not listed. For **diclazuril** methods for all poultry matrices but eggs are provided. The European Community Reference Laboratory in Berlin, Germany could provide a description of a validated method for the determination of anticoccidials in eggs including diclazuril.

**Nicarbazin** should always be investigated together with **narasin** since it is authorised in combination with narasin.

**Beta-agonists:** The MRL suggested by JECFA for **ractopamine** has not been adopted yet. Methods for the detection of **salbutamol** and **zilpaterol** are missing.

The methods presented for **azaperone** (inclusive azaperol) and carazolol are outdated. Newer methods use LC-MSMS. The European Community Reference Laboratory in Bilthoven, The Netherlands could provide a description of a validated method for these compounds..

Concerning trenbolone acetate, it should be read **alpha-trenbolone** in liver and **beta-trenbolone** in muscle (not vice versa).

For **zeranol** more sensible methods are available for cattle muscle and liver using a Resorcylic Acid Lactone<sup>1</sup> profile by LC-MSMS. The European Community Reference Laboratory in Bilthoven, The Netherlands could provide a description of a validated method for this compound.

The LC-UV or LC-FLD methods for the detection of **streptomycine**, **dihydrostreptomycine**, **gentamicine**, **sarafloxacin**, **spiramycine** and **tilmicosin** of the Community Reference Laboratory in Fougères, France (AFSSA-LERMVD) should be deleted from the list in Annex I. They are outdated. On the other hand the use of LC-MS/MS methods for these substances seems to be premature.

We moreover suggest adding the following to the list in Annex I:

Compound	Marker residue	Species	Technique	Tissue (matrix)	LCL/LOQ [µg/kg]
----------	----------------	---------	-----------	-----------------	--------------------

<sup>1</sup> Resorcylic acid lactones (RALs) are mycotoxins produced by a variety of different fungal strains via polyketide biosynthesis. It is the chemical group to which zeranol and taleranol belong

Carbadox	QCA	pig	LC-MS/MS	Liver, Muscle	1.30 (liver)
Olaquinox	MQCA	pig	LC-MS/MS	Muscle	2.20 (muscle)
Chloramphenicol	Chloramphenicol	all species	LC-MS/MS	all Matrices	0.05 (muscle)
Nitrofurans	metabolites				
furazolidone	AOZ	aquaculture, poultry	LC-MS/MS	Muscle	0.10 (in aquaculture)
furaltadone	AMAZ	aquaculture, poultry	LC-MS/MS	Muscle	0.22 (in aquaculture)
nitrofurantoin	AHD	aquaculture, poultry	LC-MS/MS	Muscle	0.12 (in aquaculture)
nitrofurazone	SEM	aquaculture, poultry	LC-MS/MS	Muscle	0.34 (in aquaculture)
Malachite Green	malachite green and leuco-malachite green	fish	LC-MS/MS	Muscle	0.21 and 0.18