II

(Acts whose publication is not obligatory)

COMMISSION

ELEVENTH COMMISSION DIRECTIVE 93/70/EEC
of 28 July 1993
establishing Community analysis methods for official control of feedingstuffs

THE COMMISSION OF THE EUROPEAN COMMUNITIES,

Having regard to the Treaty establishing the European Economic Community,

Having regard to Council Directive 70/373/EEC of 20 July 1970 on the introduction of Community methods of sampling and analysis for the official control of feedingstuffs (1), as last amended by Regulation (EEC) No 3768/85 (2), and in particular Article 2 thereof,

Whereas Directive 70/373/EEC requires official controls on feedingstuffs for the purpose of checking compliance with requirements arising under quality and composition provisions laid down by law, regulation or administrative action, to be carried out using Community sampling and analysis methods;

Whereas a Community analysis method for the additive halofuginone should be established for use in checking compliance with the conditions for its use in animal nutrition;

Whereas the measures provided for in this Directive are in accordance with the opinion of the Standing Committee for Feedingstuffs,

HAS ADOPTED THIS DIRECTIVE:

Article 1

Member States shall require that analyses for halofuginone content for the purposes of official checks on feedingstuffs be made using the method described in the Annex hereto.

Article 2

Member States shall bring into force the laws, regulations and administrative provisions necessary to comply with this Directive by 30 June 1994. They shall immediately inform the Commission thereof.

When Member States adopt these provisions, these shall contain a reference to this Directive or shall be accompanied by such reference at the time of their official publication. The procedure for such reference shall be adopted by Member States.

Article 3

This Directive is addressed to the Member States.

Done at Brussels, 28 July 1993.

For the Commission
René STEICHEN
Member of the Commission

(1) OJ No L 170, 3. 8. 1970, p. 2.
(2) OJ No L 362, 31. 12. 1985, p. 8.

ANNEX

DETERMINATION OF HALOFUGINONE

DL-trans-7-bromo-6-chloro-3-[3-(3-hydroxy-2-piperidyl)acetonyl]-quinazolin-4-(3H)-one hydrobromide

1. **Purpose and scope**

The method is for the determination of halofuginone in feedingstuffs. The lower limit of determination is 1 mg/kg.

2. **Principle**

After treatment with hot water, halofuginone is extracted as the free base into ethyl acetate and subsequently partitioned as the hydrochloride into an aqueous acid solution. The extract is purified by ion-exchange chromatography. The content of halofuginone is determined by reserved-phase high-performance liquid chromatography (HPLC) using an UV detector.

3. **Reagents**

3.1. Acetonitrile, HPLC grade

3.2. Amberlite XAD-2 resin

3.3. Ammonium acetate

3.4. Ethyl acetate

3.5. Acetic acid, glacial

3.6. Halofuginone standard substance (DL-trans-7-brome-6-chloro-3-[3-hydroxy-2-piperidyl)acetonyl] quinazoline-4-(3H)-one hydrobromide, E 764)

3.6.1. Halofuginone stock standard solution, 100 µg/ml

Weight to the nearest 0,1 mg, 50 mg of halofuginone (3.6) in a 500 ml graduated flask, dissolve in ammonium acetate buffer solution (3.18), make up to the mark with the buffer solution and mix. This solution is stable for three weeks at 5 °C if stored in the dark.

3.6.2. Calibration solutions

Into a series of 100 ml graduated flasks transfer 1,0, 2,0, 3,0, 4,0 and 6,0 ml of the stock standard solution (3.6.1). Make up to the mark with mobile phase (3.21) and mix. These solutions have concentrations of 1,0, 2,0, 3,0, 4,0 and 6,0 µg/ml of halofuginone respectively. These solutions must be freshly prepared before use.

3.7. Hydrochloric acid (ρ_{20} approximately 1,16 g/ml).

3.8. Methanol

3.9. Silver nitrate

3.10. Sodium ascorbate

3.11. Sodium carbonate

3.12. Sodium chloride

3.13. EDTA (ehtylenediaminetetraacetic acid, disodium salt)

3.14. Water, HPLC grade

3.15. Sodium carbonate solution, θ = 10 g/100 ml

3.16. Sodium chloride-saturated sodium carbonate solution, θ = 5 g/100 ml

Dissolve 50 g of sodium carbonate (3.11) in water, dilute to 1 l and add sodium chloride (3.12) until the solution is saturated.

3.17. Hydrochloric acid, approximately 0,1 mol/l

Dilute 10 ml of HCl (3.7) with water to 1 l.

3.18. Ammonium acetate buffer solution, approximately 0,25

Dissolve 19,3 g of ammonium acetate (3.3) and 30 ml of acetic acid (3.5) in water (3.14) and dilute to 1 l.