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Efficacy of the feed additive consisting of amprolium hydrochloride (COXAM®) for use in chickens for fattening and chickens reared for laying (Huvepharma N.V.)

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Abstract

Following a request from the European Commission, the Panel on Additives and Products or Substances used in Animal Feed (FEEDAP) was asked to deliver a scientific opinion on the efficacy of amprolium hydrochloride (COXAM®) for chickens for fattening and chickens reared for laying. COXAM® is a coccidiostat intended to be used to control coccidiosis in chickens for fattening and chickens reared for laying up to 12 weeks of age at a dose of 125 mg amprolium hydrochloride (HCl)/kg complete feed. In its former opinion, the FEEDAP Panel was not in the position to conclude on the efficacy of COXAM® for chickens for fattening and chickens reared for laying due to insufficient number of positive and significant effects on relevant parameters in one of the anticoccidial sensitivity tests (ASTs) evaluated. In the present submission, an additional AST has been provided by the applicant. In this study, challenge by an *Eimeria* inoculum of low pathogenicity indicated the coccidiostatic potential of amprolium HCl based on the reduction of intestinal lesions. Considering the results of three floor pen trials and two ASTs described and assessed in a previous EFSA opinion and taking into account the newly submitted AST, the FEEDAP Panel concludes that COXAM® is efficacious in controlling coccidiosis in chickens for fattening at a dose of 125 mg amprolium HCl/kg complete feed. This conclusion is extended to chickens reared for laying.

Keywords: Coccidiostat, COXAM®, amprolium hydrochloride, efficacy, chickens for fattening, chickens reared for laying

Requestor: European Commission

Question number: EFSA-Q-2020-00535

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1. Introduction

1.1. Background and Terms of Reference as provided by the requestor

Regulation (EC) No 1831/2003 establishes rules governing the Community authorisation of additives for animal nutrition and, in particular, Article 9 defines the terms of the authorisation by the Commission.

The applicant, HuvePharma NV, is seeking a Community authorisation of amprolium hydrochloride as a feed additive to be used as a coccidiostat for chickens for fattening and chickens reared for laying. (Table 1).

Table 1: Description of the substances

Category of additive	Coccidiostats and histomonostats
Functional group of additive	
Description	Amprolium hydrochloride
Target animal category	Chickens for fattening and chickens reared for laying
Applicant	HuvePharma NV
Type of request	New opinion

On 13 June 2018, the Panel on Additives and Products or Substances used in Animal Feed of the European Food Safety Authority ("Authority") in its opinion on the safety and efficacy of the product, could not conclude on the efficacy of amprolium hydrochloride in chickens for fattening under EU farming conditions. Consequently, a conclusion on the efficacy for chickens reared for laying was also not possible. After the discussion with the Member States on the last Standing Committee, it was suggested to check for the possibility to demonstrate the efficacy.

The Commission gave the possibility to the applicant to submit complementary information in order to complete the assessment and to allow a revision of Authority's opinion. The new data have been received on 4 December 2019.

In view of the above, the Commission asks the Authority to deliver a new opinion on amprolium hydrochloride as a feed additive for chickens for fattening and chickens reared for laying based on the additional data submitted by the applicant.

2. Data and methodologies

2.1. Data

The present assessment is based on data submitted by the applicant in the form of additional information[1] to a previous application of the same product.[2]

2.2. Methodologies

The approach followed by the FEEDAP Panel to assess the efficacy of COXAM® (amprolium hydrochloride) is in line with the principles laid down in Regulation (EC) No 429/2008[3] and the relevant guidance document: Guidance on the assessment of the efficacy of feed additives (EFSA FEEDAP Panel, 2018a).

3. Assessment

The additive COXAM®, containing as active substance amprolium hydrochloride (amprolium HCl), a synthetic coccidiostat, is intended to be used as a feed additive for the prevention of coccidiosis in chickens for fattening and chickens reared for laying up to 12 weeks of age at a dose of 125 mg amprolium HCl/kg complete feed.

In its former opinion (EFSA FEEDAP Panel, 2018b), the FEEDAP Panel concluded that amprolium HCl from COXAM® was effective as a coccidiostat in three floor pen studies and in two anticoccidial

[1] FEED dossier reference: FAD-2019-0075.
[2] FEED dossier reference: FAD-2016-0017.
[3] Commission Regulation (EC) No 429/2008 of 25 April 2008 on detailed rules for the implementation of Regulation (EC) No 1831/2003 of the European Parliament and of the Council as regards the preparation and the presentation of applications and the assessment and the authorisation of feed additives. OJ L 133, 22.5.2008, p. 1.

sensitivity tests (ASTs). Since three ASTs showing positive effects of the treatment with the coccidiostat under application are required, the FEEDAP Panel was not in the position to conclude on the efficacy of COXAM® for chickens for fattening under EU farming conditions. Consequently, a conclusion on the efficacy of COXAM® for chickens reared for laying was also not possible.

In the present submission, the applicant submitted a new AST.

3.1. Efficacy

The newly submitted AST was conducted in July 2019.[4] Three treatment groups were included: an uninfected untreated control (UUC), an infected untreated control (IUC) and an infected treated (IT) groups. The feed for the IT group was supplemented with amprolium HCl from COXAM® starting from day 14 of age at an intended concentration of 125 mg/kg feed (116 mg/kg analysed value). The birds (Ross 308, males) were randomly allocated to the groups on day 14 of age representing 10 replicates of 5 birds per treatment group for UUC and IT and 14 replicates for IUC (13 with 5 birds and one with 4 birds). At the age of 16 days, all birds of the IUC and IT treatment groups were orally inoculated with 1 mL of inoculum of sporulated oocysts from recent field isolates (Italy, July 2019).[5] Birds of the UUC group were inoculated with a sham inoculum. The birds were under study until day 21–23 of age. Feed intake was measured throughout the study and body weight was measured at the beginning and the end of the study, feed to gain ratio was calculated. Animal health and mortality were monitored daily. The day before inoculation (day 15), excreta samples were examined to ensure animals were not shedding relevant amounts of oocysts. Oocyst excretion was analysed at days 21, 22 and 23, corresponding to 5–7 days post-inoculation (PI). Intestinal lesions were scored following the method of Johnson and Reid (1970) (0 = no lesion, 1 = very mild, 2 = mild, 3 = moderate and 4 = severe) on days 21, 22 and 23.

The data were analysed using linear regression models (mixed or not) considering the treatment as the fixed effect, and the cage was the statistical unit. Group means were compared to the IUC group as reference (independent comparisons). Statistical significance was assessed at $p \leq 0.05$.

During the entire study period, none of the birds showed clinical symptoms in relation to coccidiosis and no birds died indicating low pathogenicity of the inoculum.

The results of the parameters measured are summarised in Table 2. Intestinal lesion scores (ILS) due to *Eimeria tenella* and *Eimeria brunetti* were significantly reduced in the IT group compared to IUC. Oocyst count per gram of excreta (OPG) on day 22 and 23 (6 and 7 days PI) was also lower in the IT group compared to IUC, but not significantly. However, excretion of *E. brunetti* was significantly lower on these days (OPG 6,888 vs 56,593 on day 6 PI and 57,167 vs 178,792 on day 7 PI). The performance parameters (weight gain, feed intake) were generally not influenced by the additive.

Table 2: Main results of the anticoccidial sensitivity test

Group	Lesion scores		Total OPG			Daily feed intake (g)	Body weight (g)	Daily weight gain (g)	Feed to gain ratio
	Eimeria tenella	*Eimeria brunetti*							
Days	**21–23**	**21–23**	**21**	**22**	**23**	**23**	**23**	**14–23**	**14–23**
UUC(1)	0.10*	0.08*	27*	109*	3,518*	93	928*	66*	1.50*
IUC(2)	1.52	1.27	2,880	66,478	314,995	98	792	50	1.68
IT(1)	0.54*	0.58*	5,068	12,242	60,850	93	866	60	1.49*

Means within a column with * are significantly different to IUC group ($p \leq 0.05$).
(1): On days 21, 22 and 23, three, four and three cages, respectively, with 5 birds each.
(2): On days 21, 22 and 23, four, six and four cages, respectively, with 5 birds each (except one cage with 4 birds).

In summary, challenge by an *Eimeria* inoculum of low pathogenicity indicated the coccidiostatic potential of amprolium HCl based on the reduction of intestinal lesions.

[4] Technical dossier/Study Report P19170-CAGE_fully Signed and Supplementary information November 2020.
[5] The inocula used was tested for its virulence in a dose-titration study. The dose selected (18,600 *E. tenella* and 12,800 *E. brunetti* per bird) resulted in lesion scores of 2.2 (*E. brunetti*) at days 6 and 7 post-inoculation (PI), in lesions scores of 2.6 and 2.8 (*E. tenella*) at day 6 and 7 PI, respectively; in a weight gain reduction of 42% at both days and at a 10% of coccidiosis-related mortality on day 7 PI.

4. Conclusions

Considering the results of three floor pen trials and two ASTs described and assessed in a previous EFSA opinion, and taking into account the results of the newly submitted AST, the FEEDAP Panel concludes that COXAM® is efficacious in controlling coccidiosis in chickens for fattening at a dose of 125 mg amprolium HCl/kg complete feed. This conclusion is extended to chickens reared for laying.

5. Documentation as provided to EFSA/Chronology

Date	Event
20/11/2019	Dossier received by EFSA. Additional information on COXAM®. Submitted by Huvepharma N.V.
06/02/2020	Reception mandate from the European Commission
12/08/2020	Application validated by EFSA – Start of the scientific assessment
17/08/2020	A new version of the study report was received by email
06/10/2020	Request of supplementary information to the applicant Article 7(3) of Commission Regulation (EC) No 1304/2003 – Scientific assessment suspended. *Issues: efficacy*
06/11/2020	Reception of supplementary information from the applicant - Scientific assessment re-started
27/01/2021	Opinion adopted by the FEEDAP Panel. End of the Scientific assessment

References

EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), Rychen G, Aquilina G, Azimonti G, Bampidis V, Bastos ML, Bories G, Chesson A, Cocconcelli PS, Flachowsky G, Gropp J, Kolar B, Kouba M, López-Alonso M, López Puente S, Mantovani A, Mayo B, Ramos F, Saarela M, Villa RE, Wallace RJ, Wester P, Anguita M, Galobart J, Innocenti ML and Martino L, 2018a. Guidance on the assessment of the efficacy of feed additives. EFSA Journal 2018;16(5):5274, 25 pp. https://doi.org/10.2903/j.efsa.2018.5274

EFSA FEEDAP Panel (EFSA Panel on Additives and Products or Substances used in Animal Feed), Rychen G, Aquilina G, Azimonti G, Bampidis V, Bastos ML, Bories G, Chesson A, Cocconcelli PS, Flachowsky G, Kolar B, Kouba M, López-Alonso M, López Puente S, Mantovani A, Mayo B, Ramos F, Saarela M, Villa RE, Wallace RJ, Wester P, Brantom P, Halle I, van Beelen P, Holczknecht O, Vettori MV and Gropp J, 2018b. Scientific Opinion on the safety and efficacy of COXAM® (amprolium hydrochloride) for chickens for fattening and chickens reared for laying. EFSA Journal 2018;16(7):5338, 28 pp. https://doi.org/10.2903/j.efsa.2018.5338

Johnson J and Reid WM, 1970. Anticoccidial drugs: lesion scoring techniques in battery and floor pen experiments with chickens. Experimental Parasitology, 28, 30–36.

Abbreviations

AST	anticoccidial sensitivity test
FEEDAP	EFSA Panel on Additives and Products or Substances used in Animal Feed
ILS	intestinal lesion scores
IT	infected treated control
IT	infected treated
IUC	infected untreated control
PI	post-inoculation
OPG	oocyst count per gram of excreta