

PAPER • OPEN ACCESS

Cleaning efficiency of feed production lines after production of feedstuffs with coccidiostats

To cite this article: S Jankovic *et al* 2021 *IOP Conf. Ser.: Earth Environ. Sci.* **854** 012037

View the [article online](#) for updates and enhancements.

You may also like

- [Utilization of local feed to support new entrepreneur in poultry business](#)
Adrizal Adrizal, James Hellyward and Dwi Yuzaria
- [The feed consumption of Deer \(*Axis axis*\) fed three difference type of feedstuffs](#)
Ismartoyo, A Asriany, Syahriany et al.
- [Estimation and validation of total digestible nutrient values of forage and concentrate feedstuffs](#)
Anuraga Jayanegara, Muhammad Ridla, Nahrowi et al.



The Electrochemical Society
Advancing solid state & electrochemical science & technology

241st ECS Meeting

May 29 – June 2, 2022 Vancouver • BC • Canada

Extended abstract submission deadline: Dec 17, 2021

Connect. Engage. Champion. Empower. Accelerate.
Move science forward



Submit your abstract



Cleaning efficiency of feed production lines after production of feedstuffs with coccidiostats

S Jankovic¹, T Radicevic¹, S Stefanovic¹, S Tankovic², J Djinovic-Stojanovic¹, D Spiric¹ and D Nikolic¹

¹ Institute of Meat Hygiene and Technology, Kacanskog 13, 11000 Belgrade, Republic of Serbia

² Veterinary Office of Bosnia and Herzegovina, Ministry of Foreign Trade and Economic Relations, Marsala Tita 9a, Sarajevo, Bosnia and Herzegovina

E-mail: sasa.jankovic@inmes.rs

Abstract. Intensive broiler production implies regular use of coccidiostats approved as feed additives. However, due to their chemical properties, coccidiostats can stay behind in the production line, and consequently, unavoidable cross-contamination of non-medicated feedstuffs can result in the exposure of non-target animal species and in the potential for coccidiostat residues in foods, such as chicken meat and eggs, derived from these species. In this way, coccidiostats enter the human food chain and can pose a health problem. The aim of this study was to determine the success of line cleaning after the application of salinomycin and maduramicin in feed. We tested the cleaning matrix (wheat groats) in order to demonstrate how many cleaning replicates are needed to safely produce coccidiostat-free feedstuffs. After the application of salinomycin, it is recommended that, for safety reasons, the line be cleaned with at least five batches of wheat groats of 480 kg each. In the case of maduramicin, it is recommended the line be cleaned with a minimum of eight batches, considering the relatively low permissible level of maduramicin as a contaminant in medication-free feed.

1. Introduction

Coccidiosis is a parasitic disease affecting livestock, especially poultry. The disease is caused by protozoan parasites of the genus *Eimeria*. In the case of a mass infection on farms, deaths and large material losses can occur [1]. Today, intensive broiler production implies regular use of coccidiostats approved as feed additives. The use of coccidiostats in the EU is regulated by Commission Regulation 2003/1831/EC [2]. This Regulation classifies feed additives into five categories: technological, sensory, nutritional, zootechnical and coccidiostats or histomonostats. Currently, the eleven following coccidiostats are authorized as feed additives according to 2003/1831/EC [2] – decoquinate, diclazuril, halofuginone, lasalocid, maduramicin, monensin, narazin, nicarbazin, robenidine, salinomycin and semduramicin.

Feed business operators can produce, in one facility, a wide range of animal feeds using one or several production lines. Due to their specific chemical properties, coccidiostats bind to processing equipment, which results in contamination of subsequently produced feed (cross-contamination). This is especially important for production of feed for those species and/or categories of animals for which coccidiostats are not allowed. This unavoidable carry-over or cross-contamination could result in the



exposure of non-target animal species, with potential health risks for animals, as well as the potential for residues in foods, such as meat and eggs, derived from these species. In this way, coccidiostats enter the human food chain and can pose a health problem [3]. Therefore, the testing of coccidiostats in animal feed is included in Serbia's National Monitoring Residue Control Program [4].

In order to avoid cross-contamination and to reduce the level of coccidiostats below the limits prescribed by the valid regulation, feed business operators carry out different cleaning procedures. These procedures include cleaning the lines with corn and /or wheat groats. For the purpose of more efficient cleaning, more rational business and greater product safety, it is necessary to determine which type of cleaning procedure is the most efficient, and in which time period and what amount of cleaning material should be applied in order for the lines to be cleaned.

Maximum levels of unavoidable carry-over of specific coccidiostats is regulated by Commission Directive 2002/32/EC on undesirable substances in animal feed [5]. A carry-over rate of 1% should be considered for feed used during the period before slaughter (withdrawal feed), for other feed to which no coccidiostats are added and for non-target feed for continuous feeding to food producing animals.

In Serbia, the use of eleven coccidiostats as feed additives and unavoidable carry-over in non-target feed is regulated by the Rulebook on the Quality of Feed [6]. Maximum levels of unavoidable carry-over of salinomycin and maduramicin in non-target feed in Serbia are presented in Table 1.

Table 1. Maximum levels of unavoidable carry-over of coccidiostats in non-target feed

Coccidiostats	Maximum levels of unavoidable carry-over, mg /kg feed for laying hens and fattening chickens in the pre-slaughter period in which the use of coccidiostats is prohibited
Salinomycin	0.7
Maduramicin	0.05

The aim of this study was to determine the efficiency of a feed production line cleaning procedure after feed containing salinomycin and maduramicin was produced.

2. Materials and Methods

The experimental study was based on the examination of the line after the production of medicated feed with coccidiostats – salinomycin (50 mg/kg) and maduramicin (5 mg/kg), at different cleaning stages, as well as examination of feed that came first off the production line after the applied cleaning procedure.

- Test samples (12) of wheat groats after salinomycin application - check of cleaning efficiency. Each sample represented one of 12 stages of the cleaning procedure (480 kg wheat groats per stage).
- Test samples (12) of wheat groats after maduramicin application - check of cleaning efficiency. Each sample represented one of 12 stages of the cleaning procedure (480 kg wheat groats per stage).
- Test samples of laying hen feed (6) - verification of the process.

Salinomycin and maduramicin were both purchased from Sigma-Aldrich (St. Louis, USA). Water, methanol, acetonitrile and N,N-dimethylformamide were all HPLC grade and purchased from Sigma-Aldrich (St. Louis, USA). Formic acid LC grade was from Merck (Merck KGaA, Darmstadt, Germany). Individual stock solutions, concentration 1.0 mg/mL, were prepared in methanol and stored at -20 °C. Working standard solution (mixture of analytes) was prepared in acetonitrile by diluting stock solutions into range that equated to the carry-over levels in feed and stored at 4 °C.

Portions of ground feed (5 g) were weighed individually into 50 mL polypropylene tubes with caps. Acetonitrile 25 mL was added and the tubes were shaken on a horizontal shaker IKA Yellow line (IKA Werke, Germany) for 60 minutes. The extracted samples were filtered through nylon 0.22 μm syringe filters into HPLC vials. Quantification was carried out using matrix extracted calibrations curves at four levels. Blank feed samples were fortified at four different levels with mixed working standard solution and submitted to the full extraction procedure.

Coccidiostats were analysed with SHIMADZU LCMS 8040 (Shimadzu, Kyoto, Japan). The instrument was controlled by LabSolutions software. The analytical column used for separation was Kinetex 100 x 2.1 mm 2.6 $\mu\text{C}18$ 100A with UltraGuard cartridge (Phenomenex, Toren, CA, USA). The oven temperature was set at 45°C. The chromatographic separation was achieved in gradient mode using water acidified with 0.1% formic acid (mobile phase A) and acetonitrile acidified with 0.1% formic acid (mobile phase B) at a flow rate of 0.3 mL/min. Electrospray ionization (ESI) was used in positive mode, with the following parameters: capillary voltage 3.5 kV, cone voltages 30 V, desolvation temperature 400 °C. Argon was used as collision gas. The precursor and product ions for analytes are presented in Table 2.

Table 2. MS/MS parameters

Compound	Precursor ion (m/z)	Product ions (m/z)
Salinomycin	773.50	265.10
		431.10
Maduramicin	934.8	629.50
		647.50

3. Results and Discussion

Salinomycin and maduramicin levels in analysed batches of wheat groats used in 12 cleaning stages after production of medicated feed are shown in Table 3.

Table 3. Levels of salinomycin and maduramicin in wheat groat batches used in 12 cleaning stages

Cleaning wheat groat batch	Salinomycin (mg/kg)	Maduramicin (mg/kg)	Cleaning wheat groat batch	Salinomycin (mg/kg)	Maduramicin (mg/kg)
1	2.872	0.328	7	0.254	0.018
2	1.182	0.190	8	0.250	0.020
3	0.906	0.106	9	0.170	0.013
4	0.524	0.096	10	0.071	0.011
5	0.520	0.078	11	0.066	0.022
6	0.312	0.028	12	0.046	0.018

The level of salinomycin, which had been used to produce medicated broiler feed, decreased from the initial value of 50 mg/kg (in the medicated feed) to 2.872 mg/kg in the first batch of groats used for line cleaning, which represents 5.7% of the initial value. The results indicate cleaning efficiency of almost 95% after the first cleaning batch. However, the absolute value of salinomycin was four times higher than the maximum allowed. Salinomycin at a level below the regulation limit (0.7 mg/kg) was achieved after the 4th cleaning stage. The following cleaning batches effected a gradual decrease of salinomycin to 0.046 mg/kg in the 12th batch. The decrease of salinomycin content in all the analysed batches of cleaning wheat groats is presented in Figure 1. The level of salinomycin decreased rapidly after the first three batches, while from batches 4 to 12, the trend continued but at a significantly slower rate.

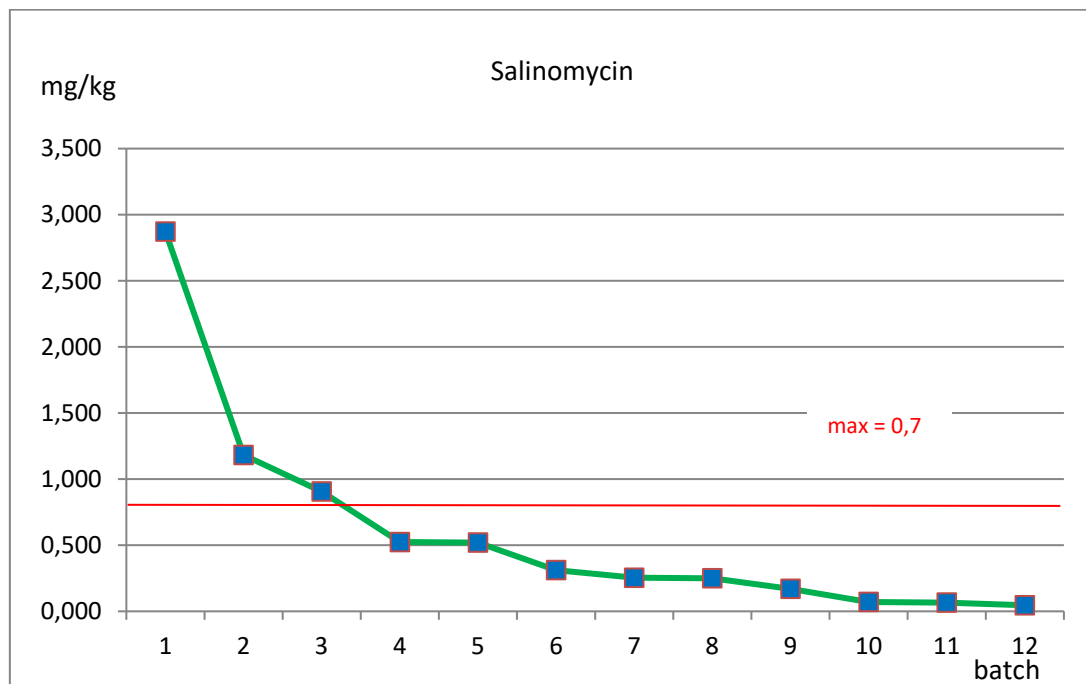


Figure 1. Salinomycin levels in the 12 batches of cleaning wheat groats

The initial level of maduramicin in the medicated broiler feed was 5 mg/kg. After production of medicated feed with maduramicin, levels in cleaning batches of wheat groats decreased gradually from 0.328 mg/kg in the first to 0.018 mg/kg in the last batch (Figure 2). The most rapid decrease in the level of maduramicin in feed was observed in the first three cleaning batches, while the decrease in the following batches was significantly slower. However, a maduramicin level below the maximum allowed limit (0.050 mg/kg) was achieved after the 6th cleaning stage.

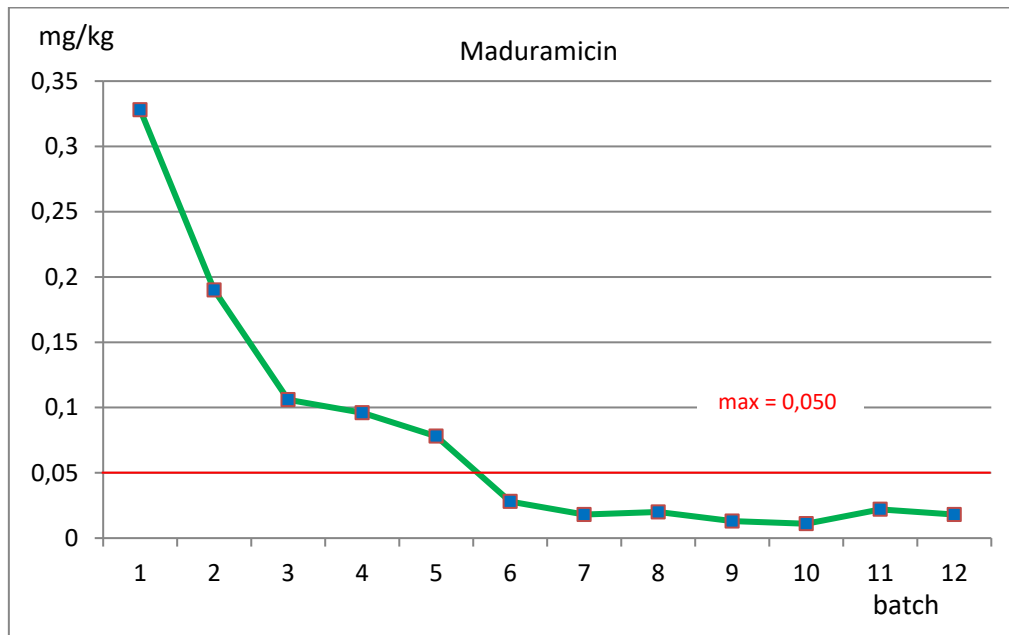


Figure 2. Maduramicin in the 12 batches of cleaning wheat groats

The coccidiostats tested in our study have different chemical structures and properties, and the applied amounts in broiler feed are different – 50 mg/kg for salinomycin and 5 mg/kg for maduramicin. Due to their different chemical structures and the results of this study, these two coccidiostats should be considered separately in terms of cleaning the production lines.

Although the level of salinomycin decreased below the allowed maximum after the 4th cleaning stage, it is recommended that, for safety reasons, the line should be cleaned with at least five batches of wheat groats of 480 kg each. On the other hand, the permissible level of maduramicin is ten times lower than that of salinomycin, which consequently signals the need for more cleaning stages. In the case of maduramicin, it is recommended that the line should be cleaned with a minimum of eight batches of groats.

Dolenc et al. [7] state that the use of feed contaminated with 0.015 mg/kg maduramicin causes residues exceeding 6 µg/kg maduramicin in eggs, while in the case of approximately 0.05 mg/kg in feed, residues exceeding 10 µg/kg were found in eggs [7].

After the cleaning procedure with 12 batches of wheat groats, on the same production line, we produced feedstuff for laying hens without added coccidiostats. On checking the levels of coccidiostats in order to conduct verification of the cleaning procedure, satisfactory levels of salinomycin and maduramicin were measured in this non-medicated feed (Table 4).

Table 4. Salinomycin and maduramicin in laying hen feed

Feedstuff for laying hens (samples taken from one batch)	Salinomycin, mg/kg	Maduramicin, mg/kg
1	0.004	0.006
2	0.005	0.004

3	0.006	0.004
4	0.007	0.003
5	0.015	0.018
6	0.006	0.014

Both coccidiostats were present in the permitted amounts, with a maximum of 0.015 and 0.018 mg/kg for salinomycin and maduramicin, respectively, which indicates a successfully implemented cleaning process in the production line. The coccidiostat levels in non-medicated feed in our study are lower than in a study conducted in Denmark in the period 2004-2007 [8]. The authors stated that of the 111 tested samples of the first batch of non-medical feed, prepared after the production of feed with salinomycin and the cleaning process, 13 samples had noticeable amounts of salinomycin in the range of 0.07-2.95 mg/kg [8].

4. Conclusion

After the production of medicated feed with coccidiostats, it is necessary to clean the production equipment. For salinomycin, the minimum number of cleaning replicates is five while in the case of maduramicin, a minimum of eight batches (480 kg each) of wheat groats are required to ensure the safe, consequent production of non-medicated feed and to avoid the possible harmful effects of coccidiostat carry-over into non-medicated feed.

Acknowledgments

This study was supported by the Ministry of Education, Science and Technological Development of the Republic of Serbia, according to the provisions of the Contract on research financing in 2021 (No 451-03-9/2021-14/200050 dated 05.02.2021).

References

- [1] Mortier L, Daeseleire E and Van Peteghem C 2005 *J. Chromatogr.* **820** 261
- [2] Commission Regulation 2003/1831/EC of The European Parliament and of the Council **L 268** 29
- [3] O'Mahony J, Moloney M, Whelan M and Danaher M 2012 Feed additives and veterinary drugs as contaminants in animal feed – the problem of cross-contamination during feed production. In: *Animal Feed Contamination*, ed Fink-Gremmels J (Cambridge: Woodhead Publishing) pp 385–410
- [4] Spirić A, Batas V, Janković S and Petronijević R 2007 *Meat Technol.* **48** 69–75
- [5] Commission Directive 2002/32/EC of The European Parliament and of the Council **L 140** 10
- [6] *Off. Gaz. Republic Serbia* No. 4/2010, 113/2012, 27/2014, 54/2017
- [7] Dolenc J, Ciglarič R and Sinigoj Gacnik K 2011 *Slovenian Veterinary Research* **48** 129–32
- [8] EFSA 2008 Cross-contamination of non-target feedingstuffs by salinomycin authorised for use as a feed additive, Scientific Opinion of the Panel on Contaminants in the Food Chain *EFSA J.* **591** 1–38