Update of the Opinion of the Scientific Panel on Additives and Products or Substances used in Animal Feed on a request from the European Commission related to the safety and efficacy of “Kokcisan 120G”

(Question N° EFSA-Q-2006-006)

Adopted on 11 July 2006

SUMMARY

The European Food Safety Authority received a request from the European Commission to issue an opinion on the safety of Kokcisan 120G for chickens for fattening based on the new data provided by the applicant in the supplementary dossier.

Data submitted by the applicant on Kokcisan 120G were previously reviewed by the FEEDAP Panel in May 2004 and found to be insufficient for conclusions to be reached on efficacy and safety. In response to this review the applicant has conducted further studies to address the inadequacies previously identified and the additional studies are reviewed in the following opinion.

From the supplementary metabolic studies, it has been concluded that unchanged salinomycin represents less than 10% of the total radioactivity in the excreta regardless of gender. The results presented identify more than 20 metabolites in chicken excreta, each representing less than 10% of the total radioactivity. The metabolic profiles in chicken and rat excreta and tissues are found to be qualitatively similar. Kinetics study of total residues and SAL-Na residues in chicken tissues from animals dosed at maximum dose proposed was provided and Salinomycin sodium could be considered as a marker residue.

Based on the revised study report provided by the applicant of a 13-week study in dogs previously considered, the NOEL derived from the study in question has been reviewed and changed to 0.5 mg SAL Na kg⁻¹ bw day⁻¹. A reproduction and teratology study in rabbits provided a NOEL for maternal toxicity of 0.5 mg SAL-Na kg⁻¹ bw day⁻¹. Pharmacology studies by the oral route in dogs indicated a NOEL for pharmacological effects (heart rate increase) of 0.625 mg kg⁻¹ bw.

A chronic toxicity study in rats was reviewed and concluded overall to be poorly conducted, yielding insufficient numbers of animals for scheduled necropsy to comply with current standards and regulatory guidelines. The study is compromised by high mortality, which may have served to select the least sensitive animals for the latter stages of the study. The changes to dose and route of administration leave some uncertainty as to the exact dose which each treatment group received, although this problem alone would not have been insurmountable. The number of animals which died during this study caused concerns for animal welfare. Bringing all of the above factors together, it is not possible for the FEEDAP Panel to reach any conclusion on the chronic safety of Kokcisan 120G in rats from the study provided.

Since no adequate chronic study is available for this product it is not possible for the FEEDAP Panel to derive an ADI or MRL from the submitted data. Setting an MRL is further impeded by the lack of consistent data for the marker residue in the relevant tissues and the lack of data for the proposed target tissue skin/fat.

It cannot be excluded that the use of Kokcisan 120G at the recommended dose range poses a risk for the terrestrial compartment.
In the previous opinion on this product, the FEEDAP Panel criticized the lack of an up-to-date floor pen studies. The applicant submitted a new study in August 2005 and the FEEDAP Panel has assessed this study and included the results in Appendix, as this issue is not specifically addressed in the question from the European Commission. This study has shown that Kokcisan 120G at a level of 60 mg salinomycin sodium kg$^{-1}$ diet is effective in supporting the growth of chicken for fattening under the conditions of an artificial *Eimeria* infection.

**Key words:** Coccidiostat, salinomycin sodium, Kokcisan 120G, chickens for fattening, metabolism, residues, consumer safety, environmental safety, efficacy.
BACKGROUND

Council Directive 70/524/EEC\(^1\) lays down rules governing the Community authorisation of additives for animal nutrition and, in particular, defines the conditions that substance/product should meet to be granted authorisation.

The Commission received a supplementary dossier from the applicant company, KRKA, to complete the missing data on the product “Kokcisan 120G”, based on Salinomycin sodium, when it is used as a feed additive for chickens for fattening.

TERMS OF REFERENCE

The Commission requests the European Food Safety Authority to issue an opinion on the safety of the product with the trade name “Kokcisan 120G”, salinomycin sodium, for chickens for fattening, based on the new data provided by the supplementary dossier.

Opinion on Kokcisan 120G for chickens for fattening

ASSESSMENT

1. Introduction

Data submitted by the applicant on Kokcisan 120G were previously reviewed by the FEEDAP Panel (EFSA, 2004) and found to be insufficient for any conclusion to be reached on efficacy and consumer safety. In response to this review, the applicant has conducted further studies to address the inadequacies previously identified and the additional studies are reviewed in the following opinion. The question asked by the Commission relates solely to the issue of safety which is addressed below. The concerns of the FEEDAP Panel that a recent floor-pen study in chickens had not been submitted have also been addressed by the applicant. Since review of these data was not required by the Commission, the review of this study is included in Appendix.

2. Safety – studies on target species

2.1. Metabolism

The former assessment of the metabolism of salinomycin sodium from Kokcisan 120G (EFSA, 2004) emphasized the following shortcomings:

i. The inconsistency of the results concerning the proportion of unchanged SAL-Na in chicken excreta;

ii. The lack of identification of SAL-Na metabolites in chicken excreta and tissues and subsequently the unavailability to determine the marker residue;

iii. The impossibility of comparing the metabolic profiles of chicken and rat due to the use of different analytical conditions.

The applicant has submitted two new GLP-studies concerning the metabolic fate of salinomycin sodium in chicken and rat.

The metabolic fate of \([^{14}C]\)-salinomycin sodium (generally labelled, without further indication on the position of the labelling) was studied in 21-day old chickens, dosed twice a day and for five consecutive days with capsules containing labelled salinomycin sodium at a dose equivalent to the exposure resulting from the consumption of feed supplemented with 70 mg kg\(^{-1}\). The animals (three males and three females per timepoint) were slaughtered 0.25, 1, 3, 5 and 7 days after the last administration, the last two capsules (day 7) being administered simultaneously and tissues sampled. The seven-day cumulated excreta of the one-day withdrawal group of animals were collected. Tentative identification of metabolites in excreta and tissues was performed using LC-MS/MS analysis.

The metabolic fate of \([^{14}C]\)-salinomycin was studied in rats administered by gavage twice daily for five consecutive days a dose equivalent to the exposure resulting from the consumption of feed supplemented with 70 mg kg\(^{-1}\). The animals were killed four hours and seven days after the last dose. Metabolic profiling of excreta and tissues was performed using the same LC-MS/MS technique as that used for chicken.

The results presented complete the data available and identify more than 20 metabolites in chicken excreta that represent each less than 10% of the total radioactivity. These correspond to monohydroxy, diketo-monohydroxy, dihydroxy, diketo-dihydroxy, trihydroxy and diketo-trihydroxy-salinomycin. The exact structure (hydroxylation positions) of these

\(^2\) Supplementary Dossier. November 2005. 10694-10782
\(^3\) Supplementary Dossier. November 2005. 10784-10855
compounds is not described. Similar qualitative profile is obtained in tissues, but the proportion of the different metabolites is different, monohydroxy compounds being higher in the liver and trihydroxy metabolites higher in the excreta. Each of these metabolites represents less than 10% of the total radioactivity in tissues.

The FEEDAP Panel concludes that:
   i. Unchanged salinomycin sodium represents less than 10% (estimated by the FEEDAP Panel from the metabolic profile) of the total radioactivity in the excreta regardless of gender;
   ii. The metabolic profiles in chicken and rat excreta and tissues are qualitatively similar.

2.2. Residues

The former assessment of residues of SAL-Na from Kokcisan 120G (EFSA, 2004) identified the absence of depletion of the total residues in the liver, kidney and muscle over the 0.5, 1, 2 and 5-day withdrawal period and, despite erratic values measured in the fat, an increase in this tissue. No relevant explanation was found based on the available data and a flaw in the experiment was suggested.

The new study carried out on chicken provides another set of data on the total residual radioactivity and unchanged salinomycin sodium level in tissues, measured by LC/MS/MS method with a LOQ of 0.001 mg SAL-Na kg\(^{-1}\). The extractable radioactivity measured in the liver of the 0.25-day withdrawal chickens amounted to 82% and 100% for the male and female, respectively. No value for other tissues or longer withdrawal periods was given. The results are given in Table 1.

Table 1. Tissue residue kinetics of \([^{14}C]\)-salinomycin sodium in 21-day old chickens (three males and three females) following oral administration (capsule) over five days of a dose equivalent to the exposure to feed supplemented with 70 mg kg\(^{-1}\) and application of a withdrawal period

<table>
<thead>
<tr>
<th>Withdrawal (days)</th>
<th>Liver</th>
<th>Kidney</th>
<th>Muscle</th>
<th>Fat</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>TRR(^1)</td>
<td>SAL(^2)</td>
<td>TRR</td>
<td>SAL</td>
</tr>
<tr>
<td>0.25</td>
<td>1.45 ± 0.33</td>
<td>0.07</td>
<td>0.46 ± 0.39</td>
<td>nd(^4)</td>
</tr>
<tr>
<td>1</td>
<td>0.45 ± 0.22</td>
<td>0.002</td>
<td>0.13 ± 0.05</td>
<td>&lt;LOQ(^3)</td>
</tr>
<tr>
<td>3</td>
<td>0.48 ± 0.33</td>
<td>&lt;LOQ(^3)</td>
<td>0.09 ± 0.03</td>
<td>&lt;LOQ</td>
</tr>
<tr>
<td>5</td>
<td>0.19 ± 0.09</td>
<td>&lt;LOQ</td>
<td>0.04 ± 0.004</td>
<td>&lt;LOQ</td>
</tr>
<tr>
<td>7</td>
<td>0.19 ± 0.08</td>
<td>&lt;LOQ</td>
<td>0.03 ± 0.004</td>
<td>&lt;LOQ</td>
</tr>
</tbody>
</table>

Note: Values represent means ± SD

\(^1\) TRR = total residual radioactivity expressed as mg equivalent SAL-Na kg\(^{-1}\) wet tissue

\(^2\) SAL = SAL-Na contents mg kg\(^{-1}\) wet tissues

\(^3\) LOQ = limit of quantification < 0.001 mg kg\(^{-1}\)

\(^4\) nd = not determined due to insufficient samples

Semi-log plotting of these values indicated a two stage exponential decrease of the radioactivity: fast during the first 24 hours, especially in the liver and kidneys, then much slower, with a half-life of 3.0 to 3.25 days similar for the liver, kidney, muscle and fat. The liver appears to be the target tissue up to 3-day withdrawal, then the fat for longer withdrawal times. In view of the delayed depletion of residues in fat; skin/fat is proposed as the target tissue for control purposes. Salinomycin sodium was measured at very low concentrations in the liver and muscle at 0.25-day withdrawal but was no longer detectable in these tissues at withdrawal times greater than one day. Due to insufficient samples, no

\(^4\) Supplementary Dossier. November 2005. 10694-10782
data were available for kidney and fat. The ratio SAL-Na vs total residues could be established only for the 0.25-day withdrawal in two tissues.

The data presented in this dossier does not comply with Directive 2001/79/EC⁵ according to which the appropriate control tissue for fat in chicken is skin together with fat.

However, for practical purposes, salinomycin sodium could be retained as marker residue.

3. Studies concerning the safety for the consumer – Studies on Laboratory animals.

3.1. Revision to 90-day study in dogs

The FEEDAP Panel stated in its former opinion on Kokcisan 120G (EFSA, 2004) that the no observed effect level for SAL-Na fermentation product established by a 13-week subchronic study was 0.5 mg fermentation product kg⁻¹ bw day⁻¹. This is equal to a NOEL of 0.117 mg SAL-Na kg⁻¹ bw day⁻¹.

The FEEDAP Panel consequently concluded (EFSA, 2004) that the overall NOEL for SAL-Na from Kokcisan 120G was about 0.12 mg kg⁻¹ day⁻¹ (0.117 mg kg⁻¹ bw day⁻¹), based on the results of a 90-day dog study. Serious adverse effects were seen in one of four male dogs in the top dose group. The possibility that the adverse effects were related to the treatment with SAL-Na could not be excluded.

In a new submission, the applicant provided an amendment to the previous report, based on a statement of the study director. The letter states:

“At the original request of the Sponsor a correction factor of 4.27 was used to correct for the purity of the test item when formulating/calculating the dose levels administered to each animal at (name of contract laboratory) in both studies. Although this was stated in the report for the oral (capsule) Maximum tolerated Dose Study in Dogs, it was not clearly stated in the report for the 13 Week Oral (Capsule) Toxicity Study in Dogs.

The corrected report contains consequently the following conclusion:

The no observable effect level (NOEL) for Salinomycin Sodium established by this study is therefore considered to be 0.5 mg kg⁻¹ day⁻¹ (administered as Salinomycin Sodium Fermentation Product).”

The FEEDAP Panel has no reason to question this amendment and revises consequently its conclusion on the NOEL in the 13-week dog study. The NOEL is now considered to be 0.5 mg SAL-Na kg⁻¹ bw day⁻¹.

3.2. Chronic toxicity

Following a dose-ranging study, groups of 60 rats of each sex were initially given a salinomycin sodium fermentation product by gavage in suspension in 0.5% carboxymethylcellulose to provide doses of 0, 2.5, 5.0 or 7.5 mg SAL-Na kg⁻¹ bw day⁻¹. Severe adverse effects were seen in the first two weeks of treatment in females which caused the doses to be reviewed and changed from day 14 to 0, 2.0, 4.0 or 6.0 mg SAL-Na kg⁻¹ bw day⁻¹. For males, the highest dose was reduced to 6.0 mg SAL-Na kg⁻¹ bw day⁻¹ at the same time. This change did not entirely remedy the adverse effects. At week 37,
gavage was discontinued and the administration of test substance switched to diet, using
concentrations designed to provide 0, 2.0, 4.0 or 6.0 mg SAL-Na kg−1 bw day−1 for females
and 0, 2.5, 4.0 or 6.0 mg SAL-Na kg−1 bw day−1 for males. Blood samples were taken for
haematology and clinical chemistry before the start of the study at day 30, 60, 90, 180,
and after nine and 12 months of treatment. Food intake and body weight were monitored
throughout the study and a full range of observations were made at intervals. Six animals
of each sex and from each treatment group were scheduled for necropsy at 3, 6 or 12
months. A further six animals of each sex and from each group were returned to control
diets after 52 weeks of treatment and brought to necropsy four weeks later. Histopathology
was confined to the animals scheduled for necropsy and grossly abnormal tissues from
those which died.

Mortality during the study occurred only during the gavage phase of the study and was 25,
43, 55 and 57% for control, low, mid and high dose males, respectively. Corresponding
mortality for females was 10, 45, 38 and 53%. The majority of these deaths seem to be
related to consequences of the gavage route of administration. However, some deaths,
where cardiovascular lesions were seen, are considered as related to treatment. The
incidence of treatment-related mortality is concluded by the study pathologist to be one
female from the low-dose group, eight from the mid-dose group and 16 from the high-dose
group; for males, the treatment-related mortality was considered to be 19 in the mid-dose
and 15 in the high-dose group. This conclusion does not appear to reflect all of the
increased mortality seen in all treated groups compared with controls. Examination of the
animals which died during the study seems to have been mainly confined to a gross
examination. Therefore, treatment-related effects at the microscopic level cannot be
excluded.

Body weight was adversely affected by treatment at the highest dose in both sexes. This is
associated with a dose-related decrease in food consumption. No effect of treatment was
seen on CNS functional observations or ophthalmoscopy. Haematological investigations
revealed reduced neutrophils, RBC, Hb and Haematocrit and increased lymphocytes, MCV
and MCHb in the highest dose, particularly in females. Clinical chemistry showed a range of
consistent changes at all doses of salinomycin sodium, including: increased alkaline
phosphatase in both sexes, increased LDH and aspartate aminotransferase in females and
disturbed serum electrolytes in all groups of both sexes.

Gross examination at scheduled necropsy showed cardiac effects in animals from both
sexes at the middle and high dose after three and six months of treatment, with no similar
changes at the lowest dose. At the 12-month necropsy, similar effects were not seen.
Caecal enlargement was consistently associated with treatment at all stages of the study.
Microscopic examination identified cardiac dilatation at mid and high dose at six months,
but not at 12 months. Other cardiac findings were of similar incidence in control and
treated groups. Organ weights showed only a few consistent trends including increased
liver, spleen and thymus weights at the highest dose.

Although this is not a carcinogenicity study and the number of animals surviving to 12
months is very low, there is no indication of any difference between the groups in the
incidence of tumours.

Overall, this study has been poorly conducted by present day standards, yielding insufficient
numbers of animals for scheduled necropsy to comply with current standards and
regulatory guidelines. The study is compromised by high mortality, which may have served
to select the least sensitive animals for the latter stages of the study. The changes to dose
and route of administration leave some uncertainty as to the exact dose which each
treatment group received, although this problem alone would not have been
insurmountable. The number of animals which died during this study raises serious
concerns that animal welfare was insufficiently considered both during the design (decision
to use gavage) and the conduct of the study. Bringing all of the above factors together, it is not possible to reach any conclusion on the chronic safety of Kokcisan 120G in rats from the study provided.

3.3. Reproduction toxicity, including teratogenicity

Following a range-finding study, a GLP-compliant teratogenicity study was conducted in groups of 23 time-mated female rabbits receiving diets containing salinomycin sodium fermentation product to provide dietary concentrations of 0, 5.0, 15.0 or 45.0 mg SAL-Na kg\(^{-1}\) diet. Diets were given from day 6 of gestation until sacrifice on day 29.

At necropsy, a full range of observations were made on the ovaries, and uterine contents and foetuses were examined both for soft part and skeletal abnormalities. Body weight gain of all the treated groups, from the start of treatment, was lower than control in a dose-related manner, but was only significantly different at the highest dose. This was associated with reduced food intake. Some rabbits did not eat at all during the test period and are excluded from subsequent analyses. Data for the study are therefore based on groups of 19, 17, 18 and 18 for control, low-, mid- and high-dose groups, respectively.

The intake of salinomycin sodium is calculated to be 0.2, 0.5 and 1.1 mg kg\(^{-1}\) bw day\(^{-1}\) between day 6 and day 24 of gestation. The fecundity and gestation indexes were similar in all groups. The number of small foetuses (<60% of mean control foetus weight) was increased in the mid- and high-dose groups. In the mid-dose groups, the difference reflects the larger number of foetuses per litter. Therefore, it is not considered to be treatment-related. The difference seen at the highest dose has no such explanation and is thus considered to be an effect of treatment. Soft part examination of foetuses revealed no differences which could be attributed to treatment. Delayed ossification was detected in the high-dose group skeletal examinations. However, this is considered to be related to the poorer maternal nutritional status of this group and hence secondary to maternal toxicity.

The mid-dose group is concluded to show no adverse effects and thus a NOEL is established for maternal toxicity at 0.5 mg salinomycin kg\(^{-1}\) bw day\(^{-1}\).

3.4. Other specific toxicological and pharmacological studies

Groups of two dogs were given gelatine capsules containing salinomycin sodium fermentation product to provide oral doses of 1.25, 0.625 or 0.313 mg kg\(^{-1}\) bw. General condition, ECG and heart rate were recorded for each dog just before treatment and 1, 2, 4, 6, 8 and 24 hours after treatment. This experimental design followed the dosing of one dog at 2.5 mg kg\(^{-1}\) bw, which resulted in significant adverse effects.

One dog given 1.25 mg kg\(^{-1}\) bw showed signs of leg weakness for the first eight hours after treatment. No other effects on condition were recorded. Heart rate increases were recorded for dogs given highest doses but not in those receiving the lowest two doses of 0.625 or 0.313 mg kg\(^{-1}\) bw. The pharmacological NOEL following oral administration of salinomycin sodium is thus concluded to be 0.625 mg kg\(^{-1}\) bw.

3.5. Determination of a NOEL

Since the chronic study provided is considered inadequate, there is no possibility to derive a chronic NOEL at this time.

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9 Supplementary Dossier. October 2005. 10209-10577
4. **Safety Evaluation for the Human Consumer**

4.1. **Proposal for an ADI**

The data provided in this supplementary submission completes, in principle, the required information for assessing consumer safety. However, the chronic study provided has severe shortcomings and is insufficient to provide a NOEL as the basis for an ADI.

4.2. **Proposal for a MRL**

Since it is not possible to derive an ADI for this product from the data provided, there is no possibility of calculating a MRL, even though much of the necessary residue data is available.

Setting an MRL is further impeded by the lack of consistent data for the marker residue in the relevant tissues and the lack of data for the proposed target tissue skin/fat.

5. **Safety for the Environment**

Based on the additional information on metabolism, it can be concluded that unchanged salinomycin sodium is excreted at less than 10% of the applied dose. No information is supplied on the ionophoric activity of the remaining metabolites (90%). However, data published by Dimenna et al. (1989) indicate that SAL-Na metabolites in chicken liver retain 20% of the ionophoric activity of salinomycin sodium. As a higher degree of hydroxylation is observed in metabolites from excreta it can be assumed that ionophoric activity is even lower. Consequently, the PEC can be refined based on no more than 30% (0.1 x 100 + 0.9 x 20) of the recommended dose.

The applicant submitted additional information on degradation in pig manure. However, without a demonstration of similarity between pig and chicken manure, the FFEDAP Panel could not consider this newly submitted data.

After refinement the PEC/PNEC ratios are still > 1 (Table 2). Although a withdrawal period and a shuttle program are not taken into account, the provided information is still insufficient to exclude a risk for soil organisms.

<table>
<thead>
<tr>
<th>Location</th>
<th>PEC_{soil} μg kg⁻¹</th>
<th>PNEC μg kg⁻¹</th>
<th>PEC/PNEC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vulnerable areas</td>
<td>100</td>
<td>13</td>
<td>7.5</td>
</tr>
<tr>
<td>Non vulnerable areas</td>
<td>204</td>
<td>13</td>
<td>15.6</td>
</tr>
</tbody>
</table>

5.1. **Risk for groundwater**

In its former opinion, the FEEDAP Panel already concluded that the groundwater trigger of 0.1 μg L⁻¹ is not likely to be exceeded.

5.2. **Risk for aquatic organisms**

If the PECs are refined based on 30% of the applied dose, the PEC/PNEC for both vulnerable and non vulnerable areas are higher than one (Table 3). However, taking into account a withdrawal period, use of shuttle programs and the low probability of single spreading events in non-vulnerable areas, a risk for the aquatic environment is not expected.
Table 3. **Comparison of the PEC and PNEC for the aquatic compartment**

<table>
<thead>
<tr>
<th>Vulnerable areas</th>
<th>PEC surface water μg L⁻¹</th>
<th>PNEC μg L⁻¹</th>
<th>PEC/PNEC</th>
</tr>
</thead>
<tbody>
<tr>
<td>3.0</td>
<td>1.7</td>
<td>1.8</td>
<td></td>
</tr>
<tr>
<td>Non vulnerable areas</td>
<td>6.6</td>
<td>1.7</td>
<td>3.9</td>
</tr>
</tbody>
</table>

5.3. **Conclusion**

It cannot be excluded that the use of Kokcisan 120G at the recommended dose range poses a risk for the terrestrial compartment.

**CONCLUSIONS**

The FEEDAP Panel concluded that:

From the supplementary metabolic studies, it has been concluded that unchanged salinomycin represents less than 10% of the total radioactivity in the excreta regardless of gender. The results presented identify more than 20 metabolites in chicken excreta, each representing less than 10% of the total radioactivity. The metabolic profiles in chicken and rat excreta and tissues are found to be qualitatively similar. Kinetics study of total residues and SAL-Na residues in chicken tissues from animals dosed at maximum dose proposed was provided and Salinomycin sodium could be considered as a marker residue.

Based on the revised study report provided by the applicant of a 13-week study in dogs previously considered, the NOEL derived from the study in question has been reviewed and changed to 0.5 mg SAL-Na kg⁻¹ bw day⁻¹. A reproduction and teratology study in rabbits provided a NOEL for maternal toxicity of 0.5 mg SAL-Na kg⁻¹ bw day⁻¹. Pharmacology studies by the oral route in dogs indicated a NOEL for pharmacological effects (heart rate increase) of 0.625 mg kg⁻¹ bw.

A chronic toxicity study in rats was reviewed and concluded overall to be poorly conducted, yielding insufficient numbers of animals for scheduled necropsy to comply with current standards and regulatory guidelines. The number of animals which died during this study caused concerns for animal welfare. It is not possible for the FEEDAP Panel to reach any conclusion on the chronic safety of Kokcisan 120G in rats from the study provided.

Since no adequate chronic study is available for this product it is not possible for the FEEDAP Panel to derive an ADI or MRL from the submitted data. Setting an MRL is further impeded by the lack of consistent data for the marker residue in the relevant tissues and the lack of data for the proposed target tissue skin/fat.

It cannot be excluded that the use of Kokcisan 120G at the recommended dose range poses a risk for the terrestrial compartment.

**DOCUMENTATION PROVIDED TO EFSA**

1. Letter from the European Commission, Health and Consumer Protection Directorate General (DG-SANCO), dated 21 December 2005 from Mrs Paola TESTORI-COGGI requesting a consultation of the scientific Panel on Additives and Products or Substances used in Animal Feed on “Kokcisan 120G”.


5. Submission of the supplementary dossier on safety (studies on laboratory animals) of “Kokcisan 120G”. KRKA. December 2005.


REFERENCES


EFSA (European Food Safety Authority). 2004. Opinion of the Scientific Panel on Additives and Products or Substances used in Animal Feed on the evaluation of coccidiostat Kokcisan120G.

SCIENTIFIC PANEL MEMBERS

APPENDIX

Assessment of a controlled floor pen study

The FEEDAP Panel, in the former opinion on Kokcisan 120G (EFSA, 2004), concluded that:

‘Kokcisan 120G containing 12% salinomycin sodium (SAL-Na) as active substance, added as coccidiostat at a level of 60 mg salinomycin sodium kg\(^{-1}\) feed for chickens for fattening, is as effective as another salinomycin sodium as seen in field trials. However, a full assessment of the efficacy was not possible, as no up-to-date floor pen studies have been carried out.’

One controlled floor pen study\(^{10}\) was completed after August 2005 and was submitted for evaluation by the applicant.

This study compared the efficacy of 60 mg Kokcisan 120G kg\(^{-1}\) with an un-treated control during a 35 growing period of broiler chickens. 1,200 one-day old chickens (600 male and 600 female) were randomly allocated to twenty pens containing 60 chickens. All animals were infected with \(2.5 \times 10^5\) sporulated oocysts of the respective mixture of isolates of *Eimeria* spp. on day 14.

The concentration of salinomycin sodium in feed was confirmed analytically, the maximum deviation being 8%.

Clinical and parasitological signs of coccidiosis developed in both groups. Experimental infection showed to cause moderate to severe coccidiosis. The parameters mortality, litter oocyst counts and lesion scores did not reveal significant treatment related differences (Table A).

The Kokcisan 120G treated broiler chickens showed significantly higher weight gain. Feed conversion ratio did not show significant differences between the groups (Table A).

Table A. Performance of broiler chickens and summary of oocyst counts

<table>
<thead>
<tr>
<th></th>
<th>Kokcisan 120G</th>
<th></th>
<th></th>
<th>Untreated group</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Female</td>
<td>Male</td>
<td>Female</td>
<td>Male</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mortality, due to</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>coccidiosis (%)</td>
<td>18.3</td>
<td>17.0</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Body weight gain (g)</td>
<td>1723(^{a})</td>
<td>1818(^{a})</td>
<td>1660(^{b})</td>
<td>1651(^{b})</td>
<td></td>
<td></td>
</tr>
<tr>
<td>g body weight gain/g feed</td>
<td>0.72</td>
<td>0.69</td>
<td>0.677</td>
<td>0.682</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Litter oocyst counts, (10^3) opg</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Day 7</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Day 14</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Day 21</td>
<td>308</td>
<td>267</td>
<td>476</td>
<td>281</td>
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<tr>
<td>Day 28</td>
<td>118</td>
<td>81</td>
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<td>90</td>
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<td>22</td>
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<td>19</td>
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</tbody>
</table>

\(^{a, b}\): Means in a row with different superscript letters differ significantly (P < 0.05)  
opg: oocyst per gram

Conclusion

This study has shown that Kokcisan 120G at a level of 60 mg salinomycin sodium kg\(^{-1}\) diet is effective in supporting the growth of chicken for fattening under the conditions of an artificial *Eimeria* infection.

\(^{10}\) Supplementary Dossier. October 2005. 10023-10178