Mimosine Toxicity in Broiler Chicks Fed
Leucaena leucocephala Seed Powder

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Abstract Leucaena leucocephala has been utilized with limit as livestock feeds due to the existence of a toxic mimosine. So, it is necessary to induce mimosine toxicity to the experimental animals to study its cure and elucidation. The studies on inducing mimosine toxicity by feeding L. leucocephala seed powder to chicks were carried out. Chicks were fed normal diets with 0, 10, 15 and 20% of L. leucocephala seed powder, and 1% crude mimosine for 12 days. Chicks fed toxic diets had decreased food intake and body weight gain, leg weakling symptom and enlargement of kidney. Mimosine was detected in the tissues of chicks fed the poisonous diets. Chicks orally administered crude mimosine were found to have alleviated the decrease of food intake and body weight gain. Chicks fed 15% seed powder diet for 12 days at 1–5 weeks of age, respectively, had decreased food intake and body weight gain regardless of growing stages, but mimosine content in the tissues decreased with growing age. After feeding 15% seed powder diet for 12 days, chicks were fed a normal diet for 20 days. Food intake of the chicks increased from the 1st day when the diet was changed to normal diet. Mimosine considerably existed in the feather and thyroid gland on the 20th day, but disappeared in the kidney, serum, liver and excretion. So, male broiler chicks were found to be rapidly and easily induced to mimosine toxicity by feeding 15% L. leucocephala seed powder diet for 12 days, and stored mimosines in the tissues rapidly reduced by feeding a normal diet.

Key words: Mimosine, Toxicity, Broiler chicks, Leucaena leucocephala seeds

Leucaena leucocephala, tropical and subtropical legume family, has been used as livestock feeds, owing to excellent growth effect and have their high contents of protein, carotenoids, vitamin K, xanthophylls and minerals. The leaves and seeds of the plant, however, contain a toxic substance, mimosine, \( \beta-(N-(3\text{-}hydroxy-4\text{-}oxo-1\text{-}4(1H)-pyridyl)})-\alpha\text{-}aminopropionic acid, \) so that feeding large quantities of the plant to livestock leads to toxicity, such as alopecia, growth inhibition and enlargement of thyroid gland.

L. leucocephala has been restrictively utilized for this reason. In ruminants, however, no such toxicity appear because of the degradation of mimosine by ruminal bacteria. It has been reported that this detoxification is attributed to a bacterial enzyme, cleaving mimosine to 3-hydroxy-4(1H)-pyridone (3, 4-DHP), pyruvic acid and ammonia further 3, 4-DHP to other compounds. On the other hand, it has been generally assumed that mimosine is responsible for the protection of L. leucocephala from the attack of noxious insects. Noxious insects called Leucaena psyllids, however, has suddenly grown in large
numbers in recent years\(^{12,16}\). The *Leucaena* psyllids suck a sap in the plant, and kill the plant\(^{12,16}\). It has been reported that the *Leucaena* psyllids also enzymatically degrade mimosine to 3, 4-DHP, pyruvic acid and ammonia\(^{11}\).

In livestock except ruminants, this special toxicity still appear by feeding large quantity of the plant. Curing or elucidating of this toxicity has been very important problem and immediate study has been desired. Therefore, it is first necessary to induce mimosine toxicity to experimental animals. The purpose of this study, thus, is to establish experimental animal for curing or elucidating this toxicity when *L. leucocephala* seeds and leaves is used as livestock feeds. Generally speaking, mimosine should be administered to induce this toxicity, but preparation of mimosine takes time. So, studies on speedy and simple inducement of mimosine toxicity to 7 days old male broiler chicks fed commercial diet with *L. leucocephala* seed powder including higher mimosine and metabolism of mimosine in the tissues were carried out.

**Materials and Methods**

1. Material

1) Preparation of *L. leucocephala* seed powder

Brown ripe seeds were collected from bushes of *L. leucocephala* on the campus of the Faculty of Agriculture, University of The Ryukyus, from 1994 to 1995. The seeds were crushed by feeds smasher (Ikemoto Physico-chemical Industry), and *L. leucocephala* seed powder were prepared by passing crushed seeds through 20 mesh sieve to remove most outside shells of seed.

2) Preparation of crude mimosine

Crushed seeds (350 g) were boiled for 1 h in about 2400 ml of distilled water to extract mimosine. Equivalent ethanol was added to the solution to precipitate impurities before the solution cooled, and then impurities in the solution were filtered by using suction filter. Other impurities in the filtrate were precipitated by storing in a freezer for 8 h, and were removed by centrifuging (5,000 rpm, 20 min). The supernatant solution was concentrated to about 100–150 ml by a rotary vacuum evaporator. Crude mimosine in the solution was precipitated by storing in a refrigerator. About 5 g of crude mimosine was prepared by centrifuging (5,000 rpm, 20 min) the precipitate.

3) Experimental animals

Hatched Cobb strain male broiler chicks were obtained from Ishikawa Hatchery (Okinawan Animal Development Corporation), and previously kept for 1 week on commercial broiler feed (ME 3100 kcal/kg, CP 23.5%) from Ryukyu Kyodo Feed MFG. CO., Ltd.

2. Method

At first, necessary amount of *L. leucocephala* seed powder to induce mimosine toxicity in broiler chicks was decided. Seven days old chicks were fed *ad libitum* commercial diets with 10, 15 and 20% (mimosine content 0.65, 0.98 and 1.30%, respectively) of *L. leucocephala* seed powder including higher mimosine and metabolism of mimosine in the tissues were carried out.
collected on the last day. Mimosine in the serum, liver, kidney, testis, thyroid gland, pectoral muscle, biceps femoris muscle, skin, feather and crown of slaughtered chicks were extracted, and its contents were determined by HPLC.

Secondly, relationship between growing stages and inducement of mimosine toxicity was examined. Chicks were fed 15% seed powder diets for 12 days from 1, 2, 3, 4 and 5 weeks of age, respectively, and measurements of food intake, body weight and mimosine content in the tissues were performed (Experiment IV).

Finally, after feeding 15% seed powder diet for 12 days to induce mimosine toxicity, chicks were fed a commercial diet for 20 days. The four chicks fed the commercial diet were slaughtered every four days, and mimosine content in the tissues was measured (Experiment V).

Ryukyu University’s guide for the care and use of laboratory animals was followed in this study, so chicks were slaughtered by exhausting blood under the ether anesthesia. Chicks in all experiments were housed individually in controlled room temperature (25±2°C) and lighting cycle for 12h, and were fed each diet by means of vessel for powder diet.

3. Determination of mimosine content in the tissues

One gram of each tissue except serum was homogenized in 6 ml of 0.1 N HCl solution, and 4.8 ml of 15% trichloroacetic acid (TCA) solution were added. The solution was allowed to stand for 20 min, and followed by centrifugation at 12,000 rpm for 20 min to remove the precipitates. The supernatant solution was filtered by filter for HPLC, and mimosine content was analyzed by injecting the filtrate (5–15 μl) into HPLC. To 1.0 ml of serum, 0.8 ml of 15% TCA solution was directly added, and subsequent assay was applied as mentioned above. A Shimpack CLC-ODS (150×60 mm) column was used in HPLC (Shimadzu LC-6 A) with a UV (250 nm) detector as previously described7).  

4. Statistical Analysis

T-test analysis was applied throughout all experiments. The marks (*) in figures mean significant difference for control group at p<0.01.

Results

1. Mimosine content in *L. leucocephala* leaves and seeds

Mimosine content in leaves of *L. leucocephala* was 0.56% FM, but the content in seeds was 3.46% FM, showing that seeds contained about six times as much mimosine as leaves. The mimosine content in germs completely removed seed shells was 11.07% FM, which was three times higher than that in seeds. The mimosine content in the *L. leucocephala* seed powder removed most of seed shells by passing crushed seeds through 20 mesh sieve was 6.55% FM, which was twice as high as that of seeds. Therefore, seed powder containing relatively higher mimosine was used in this experiment. Purified rate of the crude mimosine was 87.2% FM (Table I).

2. Mimosine toxicity in chicks fed *L. leucocephala* seed powder diet

In Experiment I, food intake and body weight gain smoothly increased daily in the control group, but decreased from the first day in the seed powder diet groups, and never increased. Therefore, extreme loss of appetite and growth depression appeared. Compared with the control group, food intake and body

<table>
<thead>
<tr>
<th>Table 1. Mimosine content of young leaf, seed, germ, seed powder, and crude mimosine of <em>L. leucocephala</em> (% FM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mimosine</td>
</tr>
<tr>
<td>Young leaf</td>
</tr>
<tr>
<td>Seed</td>
</tr>
<tr>
<td>Germ</td>
</tr>
<tr>
<td>Seed powder</td>
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<tr>
<td>Crude mimosine</td>
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</tbody>
</table>
weight gain in the 10% seed powder diet group significantly decreased \((p<0.01)\) from the second day and the fourth day, respectively, and in both the 15 and 20% seed powder diet groups decreased from the first and third day, respectively. The 15 and 20% seed powder diet groups had remarkable loss of appetite (Fig. 1, 2). Specific leg weakling symptom, sitting down and cramping, appeared from the fourth day in all the seed powder diet groups (Fig. 3). Mortality of the 15 and 20% seed powder diet groups was 20 and 40%, respectively. Besides, enlargement of kidney was observed in the seed powder diet groups (Fig. 4).

Mimosine was detected in all the tissues tested. Majority of the mimosine was found in the excretion. Rate of mimosine taken in the body was calculated to be about 56.1% from the amount of the daily excretion and food intake. The feather, kidney, skin, crown and testis had higher mimosine concentration in all the seed powder diet groups. Mimosine content in the tissues except the kidney and thyroid gland in the 15% seed powder diet group was higher than those in the 10% seed powder diet group (Table 2).

In Experiment II, food intake and body weight gain smoothly increased daily in the control group, but decreased from the first day in the 15% seed powder diet group and 1%

Fig. 1. Food intake of chicks fed commercial diets with 0, 10, 15 and 20% *L. leucocephala* seed powder.

Fig. 2. Body weight of chicks fed commercial diets with 0, 10, 15 and 20% *L. leucocephala* seed powder.

Fig. 3. Leg weakling symptoms of a chick induced by feeding 15% *L. leucocephala* seed powder diet.

Fig. 4. Kidneys of chicks fed 15% *L. leucocephala* seed powder diet (A) and commercial diet (B).
Mimosine Toxicity in Broiler Chicks

Table 2. Mimosine content in tissues of chicks fed commercial diet with 10, 15 and 20% of L. leucocephala seed powder (µg/g)

<table>
<thead>
<tr>
<th>Tissues</th>
<th>10% seed</th>
<th>15% seed</th>
<th>20% seed</th>
</tr>
</thead>
<tbody>
<tr>
<td>Feather</td>
<td>213.6±54.74</td>
<td>377.5±238.80</td>
<td>303.4±40.05</td>
</tr>
<tr>
<td>Skin</td>
<td>119.9±51.40</td>
<td>241.0±65.13</td>
<td>117.3±36.47</td>
</tr>
<tr>
<td>Kidney</td>
<td>154.4±130.65</td>
<td>142.1±99.66</td>
<td>73.3±44.95</td>
</tr>
<tr>
<td>Biceps femoris muscle</td>
<td>31.1±7.80</td>
<td>54.5±19.28</td>
<td>46.2±36.72</td>
</tr>
<tr>
<td>Pectoral muscle</td>
<td>35.7±17.71</td>
<td>75.5±25.07</td>
<td>48.9±36.60</td>
</tr>
<tr>
<td>Serum</td>
<td>27.2±15.56</td>
<td>76.6±36.96</td>
<td>64.8±18.24</td>
</tr>
<tr>
<td>Crown</td>
<td>149.8±71.60</td>
<td>289.6±277.65</td>
<td>361.9±272.44</td>
</tr>
<tr>
<td>Testis</td>
<td>143.2±54.20</td>
<td>243.8±69.79</td>
<td>221.3±57.69</td>
</tr>
<tr>
<td>Thyroid gland</td>
<td>80.0±19.07</td>
<td>74.2±32.88</td>
<td>130.9±117.50</td>
</tr>
<tr>
<td>Liver</td>
<td>20.2±19.66</td>
<td>41.1±14.90</td>
<td>48.5±5.55</td>
</tr>
<tr>
<td>Excretion</td>
<td>24661.9±3660.02</td>
<td>24369.6±2008.18</td>
<td>23653.3±4008.73</td>
</tr>
</tbody>
</table>

Means±S.D.

Fig. 5. Food intake of chicks fed commercial diets with 0, 1% of crude mimosine and 15% L. leucocephala seed powder.

In Experiment III, food intake and body weight gain significantly decreased (p<0.01), in both the groups, from the third and sixth days, respectively (Fig. 5). Chicks (7 days of age) fed 1% crude mimosine diet showed the same toxicity and symptoms, such as extreme loss of appetite, growth depression and leg weakening, with chicks fed 15% seed powder diet, and the chicks had the same mimosine content in all the tissues except crown with chicks fed 15% seed powder diet.

3. Mimosine toxicity in oral crude mimosine administration

In Experiment III, food intake and body weight gain smoothly increased daily in the control group, but decreased in the orally mimosine administered group. The decreases, however, were moderate, compared with the 15% seed powder group. Food intake and body weight gain significantly decreased (p<0.01) from the third and fifth day, respectively in the 15% seed powder diet group, and from the third and tenth day, respectively in the orally mimosine administered group (Fig. 6). Mimosine contents in the skin, liver, kidney...
and thyroid gland in the orally mimosine administered group were almost the same with those in the 15% seed powder diet group.

4. Relationship between growing stages and inducement of mimosine toxicity

In Experiment IV, food intake decreased from the second day in all weeks of age, and significantly decreased (p<0.01) from the fourth, sixth, third, second and third day from 1, 2, 3, 4 and 5 weeks of age, respectively. Body weight gain decreased in all weeks of age with decrease of food intake, and significantly decreased (p<0.01) from the fifth, seventh, fourth, sixth and seventh day at 1, 2, 3, 4 and 5 week days of age, respectively. Therefore, extreme loss of appetite and growth depression appeared at all weeks of age regardless of growing stages. Also, leg weakening symptom and enlargement of kidney were observed.

Mimosine concentration was constant in serum regardless of growth stages, but decreased in the feather, skin, thyroid gland, testis, crown, kidney, liver, pectoral muscle and biceps femoris muscle with growing stages. The ratios of mimosine concentrations at 5 weeks of age to those at 1 week of age were 2.0, 3.7, 4.7, 12.6, 13.5, 17.5 and 19.3% in the thyroid gland, crown, testis, biceps femoris muscle, skin, liver and pectoral muscle, respectively.

5. Metabolism of mimosine in the chicks inducing mimosine toxicity

In Experiment V, food intake and body weight gain of chicks fed 15% seed powder diet for 12 days decreased from the second day, and significantly decreased (p<0.01) from the fifth and sixth day, respectively, compared with control group, and leg weakening symptom appeared. Food intake of the chicks rapidly increased from the first day when the diet was changed to commercial diet, and had no significant difference from that of the seventeenth day, compared with control group (Fig. 7). Body weight of the chicks had significant difference at all days fed commercial diet (p<0.01), but gradually increased with the increase of food intake, and the amount of the increase was the same with that of the control group from the tenth day when the diet was changed to commercial diet (Fig. 8). Mimosine content in tissues suddenly decreased by feeding commercial diet, but still fairly large quantities of mimosine was found in the feather and thyroid gland even on the twentieth day, compared with other tissues. Relative mimosine values

![Fig. 7](image1.png)

**Fig. 7.** Food intake of chicks fed commercial diet for 20 days after feeding commercial diet with 15% *L. leucocephala* seed powder for 12 days.

* Means the first day of diet change to commercial diet.

![Fig. 8](image2.png)

**Fig. 8.** Body weight of chicks fed commercial diet for 20 days after feeding commercial diet with 15% *L. leucocephala* seed powder for 12 days.

↓ Means the first day of diet change to commercial diet.
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Table 3. Mimosine content in tissues of chicks fed commercial diet for 20 days after feeding commercial diet with 15% L. leucocephala seed powder for 12 days (μg/g)

<table>
<thead>
<tr>
<th>Tissues</th>
<th>0 day</th>
<th>4 days</th>
<th>8 days</th>
<th>12 days</th>
<th>16 days</th>
<th>20 days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Feather</td>
<td>698.5±400.7</td>
<td>434.5±99.0</td>
<td>499.0±220.4</td>
<td>198.5±66.9</td>
<td>251.1±45.0</td>
<td>81.4±45.5</td>
</tr>
<tr>
<td>Skin</td>
<td>944.7±323.6</td>
<td>89.9±68.9</td>
<td>41.0±27.0</td>
<td>18.6±16.5</td>
<td>4.1±3.3</td>
<td>10.9±4.1</td>
</tr>
<tr>
<td>Kidney</td>
<td>91.2±62.9</td>
<td>N.D.</td>
<td>N.D.</td>
<td>N.D.</td>
<td>N.D.</td>
<td>N.D.</td>
</tr>
<tr>
<td>Biceps femoris muscle</td>
<td>81.9±33.4</td>
<td>19.4±3.4</td>
<td>17.2±4.4</td>
<td>13.4±8.8</td>
<td>3.7±1.5</td>
<td>0.2±0.1</td>
</tr>
<tr>
<td>Pectoral muscle</td>
<td>92.4±37.6</td>
<td>11.0±1.0</td>
<td>10.0±1.1</td>
<td>7.0±3.0</td>
<td>5.3±2.0</td>
<td>6.2±1.8</td>
</tr>
<tr>
<td>Serum</td>
<td>69.6±53.3</td>
<td>N.D.</td>
<td>N.D.</td>
<td>N.D.</td>
<td>N.D.</td>
<td>N.D.</td>
</tr>
<tr>
<td>Crown</td>
<td>215.9±20.7</td>
<td>41.4±11.0</td>
<td>54.0±25.1</td>
<td>13.6±5.2</td>
<td>10.4±4.2</td>
<td>6.4±0.4</td>
</tr>
<tr>
<td>Testis</td>
<td>191.0±66.0</td>
<td>43.9±17.7</td>
<td>49.2±24.9</td>
<td>15.4±15.2</td>
<td>9.4±6.5</td>
<td>12.8±10.1</td>
</tr>
<tr>
<td>Thyroid gland</td>
<td>101.4±58.1</td>
<td>45.6±8.6</td>
<td>80.5±96.1</td>
<td>47.3±27.7</td>
<td>24.9±20.0</td>
<td>58.9±34.0</td>
</tr>
<tr>
<td>Liver</td>
<td>43.1±31.0</td>
<td>N.D.</td>
<td>N.D.</td>
<td>N.D.</td>
<td>N.D.</td>
<td>N.D.</td>
</tr>
<tr>
<td>Excretion</td>
<td>26182.6±4419.9</td>
<td>386.0±309.0</td>
<td>N.D.</td>
<td>N.D.</td>
<td>N.D.</td>
<td>N.D.</td>
</tr>
</tbody>
</table>

Means±S.D.

of twentieth day to those of zero day before the changed to commercial diet in the feather and thyroid gland reduced to about 13 and 50%, respectively, but mimosine was not found in the kidney, serum and liver from the fourth day, and in excretions from the eighth day.

Mimosine content in other tissues considerably decreased at the fourth day, but remained constant on the twentieth day. The ratios of mimosine concentrations at the fourth and twentieth day to those at zero day before the changed to commercial diet were 10.0, 12.0, 19.0, 24.0 and 45.0% on the fourth day, and 1.2, 6.7, 3.0, 0.2 and 6.7% on the twentieth day in the skin, pectoral muscle, crown, biceps femoris muscle and testis, respectively (Table 3).

Discussion

Chicks fed L. leucocephala seed powder diet had extreme loss of appetite, growth depression, specific leg weakening symptom, sitting down and cramping, and an accumulated mimosine in various tissues. Chicks fed 1% crude mimosine diet showed the same extent of toxicity with chicks fed 15% seed powder diet. Therefore, it was found that the toxicity was caused by mimosine. Mimosine content in the tissues and mortality decrease corresponded with growing stages, so it seems that the chick's resistance against mimosine toxicity gets gradually high with chronological age.

In chicks fed ad libitum L. leucocephala seed powder diet or crude mimosine diet, mimosine was detected in the serum, liver, kidney, testis, thyroid gland, pectoral muscle, biceps femoris muscle, skin, feather and crown. Especially, feather, skin and kidney had higher mimosine concentration. Sahlu et al.14) reported that mimosine was found in the serum, liver and kidney of goats performed the intravenous infusion of mimosine, but not found in the spleen, heart, lung and muscle. In the present experiment, however, mimosine was detected in the muscle of chicks fed seed powder diet. Food intake of chicks fed ad libitum 1% mimosine diet or 15% seed powder diet was 15-20g/day, so the amount of ingested mimosine was calculated at about 200mg/day, and the measured dry excretion (3.3g/day) included mimosine (80mg/day). The absorbed mimosine content, therefore, was 120mg/day. Besides, the average of body weight was 0.123kg, so the amount of ingested mimosine per metabolic body size was calculated at 576.9mg·kg BW⁻⁰.⁷⁵/day. In the report of Sahlu et al.14), however, goats were given 200mg·kg BW⁻¹/day of mimosine by intravenous infusion. Therefore, the difference in results may be caused by the different
amount of mimosine given to the animals. On the other hand, Sahlu et al.\textsuperscript{14)} used goats as experimental animals, but the authors used broiler chicks, so the difference in results may be brought by the difference in animal species. On the other hand, Sahlu et al.\textsuperscript{14)} reported that mimosine content (57.2 µmol/g) in the kidney was higher than mimosine content (25.6 µmol/g) in the liver. In this experiment, mimosine content (91.2 µg/g) in the kidney of chicks fed 15% seed powder diet was higher than that (43.1 µg/g) in the liver as shown in Table 3. At this point, our results agreed with their results.

Chicks inducing mimosine toxicity were fed a commercial diet for 20 days, and the disappearance of mimosine in the tissues was detected. Consequently, mimosine in the feather and thyroid gland was detected even on the twentieth day but not in other tissues or if detected, in an extremely small quantity. It seemed that mimosine may not be accumulated, and may be degraded or excreted in the tissues except in the feather and thyroid gland. A large quantity of mimosine in chicks inducing mimosine toxicity were found in the excretion.

When seed powder diet was changed to commercial diet, mimosine was not detected in the excretions from the eighth day, and mimosine in the skin, biceps femoris muscle, pectoral muscle, crown and testis slowly decreased even from the eighth day. Therefore, mimosine in the tissues may not be excreted but metabolized. Mimosine in the feather may be excreted by alopecia because alopecia sometimes appeared. Mimosine concentration in the same tissues varied in different experiments, so various capability of degradation or excretion of mimosine in the tissues may cause different mimosine concentration in the tissues.

Feeding \textit{ad libitum} 10, 15 and 20% seed powder diets (mimosine content 0.65, 0.98, 1.30%, respectively) or 1% crude mimosine diet for 12 days, chicks (7 days of age) had an extreme decrease in the food intake. Reddy \textit{et al.}\textsuperscript{13)} reported that broiler chicks (10 days of age) fed \textit{ad libitum} 10 and 15% \textit{L. leucocephala} seed diet (mimosine content 0.44 and 0.88%) for 35 days decreased in food intake and body weight gain. Our result considerably agreed with their result.

Hussuian \textit{et al.}\textsuperscript{8)}, however, reported that in broiler chicks (1 day of age) fed \textit{ad libitum} 15% \textit{L. leucocephala} leaf meal diet for 35 days, there were no significant differences in the body weight gain and food intake, compared with the control group. This result was different from our result. We used mimosine seeds, but they used the leaves. Therefore, this result may be brought by the difference of mimosine content because mimosine content in the leaves is different from that in the seeds as shown in Table 1.

Chicks fed \textit{ad libitum} 15% seed powder diet decreased in food intake. On the other hand, when chicks were fed \textit{ad libitum} commercial diet and at the same time 250 mg/day of crude mimosine directly given in the throat by a spoon without touching the sense organs of taste and smell, the decreases in the food intake of the chicks were alleviated. Besides, when chicks induced mimosine toxicity were fed a commercial diet, the food intake of the chicks did not decrease. These results suggested that mimosine is odorless to human beings, but it might have special a taste or smell avoided by chicks.

**Acknowledgments**

We are deeply grateful for the generous gifts of chicks as experimental animals and commercial diet from Okinawan Animal Development Corporation and Ryukyu Kyodo Feed MFG. CO., Ltd., respectively.

**References**

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Mimosine Toxicity in Broiler Chicks


キナデ、オシロ、オク、ホンゴおよびチネン

ギンネム種子粉末を給与したブロイラー鶏
におけるミシオン中毒症

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ギンネムには毒性物質＝ミシオンが存在するため、家畜に多量給与するとミシオン中毒症を引き起こすので、飼料としては制限給与している。このミシオン中毒症の治療、解明を行うためには実験動物にその中毒症を誘発させる必要がある。そこでまずブロイラー鶏にミシオン中毒症を迅速で簡易に誘発させる研究を行い、またその中毒時のミシオンの代謝についても調べた。まず、7日齢のブロイラー雄鶏にギンネム種子を粉碎し20ミリメートルの細粉を用いた種子粉末（ミシオン含量6.55％）を、市販飼料に0、10、15、20％添加し各々12日間自由給与した。その結果、各種子粉末飼料群では食欲不振、体重増加の減少がみられ、更に産卵減少、足をけいれんさせる特異的な脚弱症状、および腎臓の肥大化がみられた。また各組織でミシオンが検出され、特に羽毛、皮膚および腎臓で高い値が得られた。更に1％粗ミシオン飼料を自由給与すると、15％種子粉末飼料給与時と全く同程度の中毒症が認められミシオン中毒症と断定された。次に粗ミシオンを250 mg/日、経口投与しながら市販飼料を給与すると、食欲不振と体重増加の減少は緩和した。しかし、各組織中でのミシオン含量は15％種子粉末飼料給与群同様存在した。次に加齢とミシオン中毒症との関係を調べた、1、2、3、4および5週齢で15％種子粉末飼料を12日間給与すると、食欲不振、体重増加の減少は加齢に関係なく見られ、組織中のミシオン含量は加齢に伴って減少した。最後に、ミシオン中毒症の鶏に市販飼料を20日間給与し4日ごとに処理し、体内でのミシオンの代謝を調査した。その結果、まず市販飼料を給与した初日から食欲が回復し、採食量は市販飼料給与後17日目で対照群と有意差が認められなくなった。それに伴って体重も増加した。また各組織のミシオン濃度は羽毛、甲状腺では20日目でもミシオンが認められた。それに対し腎臓、血清、肝臓は4日目から、総排泄物は8日目から検出されなくなった。皮膚、筋肉、冠、精巣は20日目でも極少量のミシオンが検出された。

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