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Research Note

Does Relapse Occur in Chickens Recovered from Infection with *Leucocytozoon caulleryi*?

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Leucocytozoon caulleryi, the causative agent of chicken leucocytozoonosis, was first described by Mathis and Leger (1909), and in Japan by Akiba et al. (1958). Leucocytozoonosis in chickens has been recognized in various Asian countries and affects the productivity of chickens through a reduction in egg production, weight loss and sometimes death. In Japan, the prevalence of the disease has been observed mostly in the summer season. No information is available on the latent state of L. caullervi infection in such periods as late autumn and early winter when transmission by Culicoides arakawae ceases in Japan. It is speculated that chickens recovered from the infection may preserve the protozoa up to the next season. The recurrence of gametocytemia, which is known as a spring relapse (Alverson and Noblet; 1977; Desser et al., 1968), occurred in various avian hosts recovered from the infection of some Leucocytozoon species. This investigation was conducted to determine whether relapse would occur in chickens recovered from infection with L. caullervi.

The strain of *L. caulleryi* was derived from a naturally infected chicken in Fukui Prefec-

ture, Japan, in August, 1976. It had been maintained at the authors' laboratory by cyclic transmission in specific-pathogen-free (SPF) chickens and colonized C. arakawae. The procedure of breeding, rearing and feeding of C. arakawae for infection with L. caullervi, and preparation of the sporozoite suspension for injection into chickens were the same as described previously (Isobe et al., 1984). SPF chickens (Furuta et al., 1980) were reared in the chicken house isolated from C. arakawae. Twenty-two chickens of 3 to 30 weeks old were inoculated intravenously with 1 to 10 sporozoites for each host. The infection was confirmed by the detection of merozoites and gametocytes in the peripheral blood on the 14th to 24th day after sporozoite inoculation. After recovery from the infection: i.e., 35 weeks after sporozoite inoculation, these chickens were divided into 6 groups and each injected intramuscularly with one of six drugs (Table 1). Blood smears were prepared every two or three days for one month to detect the protozoa. Two chickens in each group were killed 31 days after injection and the lung. heart, liver, spleen and kidney were taken out. These materials were fixed in 10% buffered formalin solution, dehydrated, embedded in paraffin, cut into sections, stained with hematoxylin and eosin, and examined with a light microscope for detection of the protozoa.

No protozoa, such as merozoites, gametocytes and schizonts, were observed in blood

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Group	Drug	Dose of drug*	Period of injection	No. of chicken tested	Relapse of protozoa [†]
1	Cyclophasphamide	100 mg/day	5 days	4	
2	Epinephrine	2.5 mg/day	10 days	4	_
3	Corticosterone	10 mg/day	8 days	3	_
4	Gonadotropin	200 IU/day	10 days	4	-
5	Progesteron	25 mg/day	3 days	4	_
6	Estradiol	1.25 mg/day	5 days	3	_

Table 1 Relapse of Leucocytozoon caulleryi in infected chickens using some drugs

*Injected intramuscularly.

[†]Detected by blood smears and tissue sections.

smears and sections of the tissue, even when the hormonal stimulation or immunosuppression to the host were conducted (Table 1). For example, cyclophosphamide has been reported to cause immunosuppression in chickens (Lerman, S. P. et al., 1970; Glick B., 1971). In the present study, the antibody titer (geometric mean) decreased from 1:16 to 1:10 after injection with cyclophosphamide in the gel diffusion test (Morii, T., 1972). But we could not detect any protozoa in blood smears or any schizont in tissue sections. Hashimoto et al. (1980) and Fujisaki et al. (1982) have detected the schizonts in the chickens recovered from infection with L. caulleryi. They speculated that those schizonts might serve as a source of new epizootics. Kitani et al. (1983) reported the detection of gametocytes in chickens recovered from natural infection with L. caullervi. However, it was not clear whether the chickens were involved in reinfection or relapse. As yet, no one has recognized the recurrence of parasitemia, such as L. smithi (Alverson and Noblet, 1977) and L. simondi (Desser et al., 1968), in chickens recovered from an infection with L. caulleryi. From these results, we suppose that the chances of relapse in chickens recovered from infection with L. caulleryi are quite few.

References

 Akiba, K., Kawashima, H., Inui, S. and Ishii, S. (1958): Studies on *Leucocytozoon* of chickens in Japan. I. Natural infection of *L. caulleryi*. Bull. Natl. Inst. Anim. Health, 34, 163–180.

- Alverson, D. R. and Noblet, R. (1977): Spring relapse of *Leucocytozoon smithi* (Sporozoa: Leucocytozoidae) in turkeys. J. Med. Entomol., 14, 132-133.
- Desser, S. S., Fallis, A. M. and Garnham, P. C. C. (1968): Relapse in ducks chronically infected with *Leucocytozoon simondi* and *Parahaemo*proteus nettionis. Can. J. Zool., 46, 281-285.
- 4) Fujisaki, K., Kitaoka, S., Kamio, T. and Takamatsu, H. (1982): Detection of schizonts in a chicken recovered from experimental infection with *Leucocytozoon caulleryi*. Natl. Inst. Anim. Health Q., 22, 144-145.
- Furuta, K., Ohashi, H., Obana, J. and Sato, S. (1980): Performance of 3 successive generations of specified-pathogen-free chickens maintained as a closed flock. Lab. Anim., 14, 107-112.
- Glick, B. (1971): Morphological changes and humoral immunity in cyclophosphamide-treated chicks. Transplantation, 11, 433–439.
- 7) Hashimoto, H., Tagata, H., Akiba, K., Takaya, G., Fukunaga, S., Fujiyama, S., Fukushima, K., Hamada, K. and Isobe, T. (1980): Detection of schizonts in winter from chickens surviving after natural *Leucocytozoon caulleryi* infection in the previous summer. J. Jap. Vet. Med. Assoc., 35, 528-531.
- Isobe, T., Yamaguchi, S., Imada, T., Kawakita, F. and Akiba, K. (1984): Development of second generation merozoites of *Leucocytozoon caulleryi* in vitro. J. Protozool., 31, 186–187.
- Kitani, T., Isobe, T., Akiba, K., Nakashima, Y. and Ebisawa, S. (1983): Detection of gametocytes in chickens recovered from natural infection with *Leucocytozoon caulleryi*. Natl. Inst. Anim. Health Q., 23, 108-110.
- 10) Lerman, S. P. and Weidanz, W. P. (1970): The

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effect of cyclophosphamide on the ontogeny of the humoral immune in chickens. J. Immunol., 105, 614-619.

 Mathis, C. and Leger, M. (1909): Leucocytozoon de la poule. C. R. Soc. Biol., Paris 67, 470–472.

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Morii, T. (1972): Presence of antigens and antibodies in the sera of chickens infected with *Akiba caulleryi*. Natl. Inst. Anim. Health Q., 12, 161-167.

Leucocytozoon caulleryi 感染耐過鶏での再発?

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Leucocytozoon caulleryi 感染耐過鶏に,エスト ラジオール,ゴナドトロピン等のホルモン剤および サイクロフォスファマイド,コルチコステロン等の 免疫抑制剤を各々投与し,末梢血液中の原虫の再出 現を調べた.その結果,いずれの鶏においても原虫 は検出されなかった.また残存シゾントの検索を行ったが、いずれの群の鶏にも検出されなかった.こ れらのことから、Leucocytozoon caulleryi 感染耐 過鶏において再発の可能性は非常に少ないと考えら れる.