

MORPHOMETRICAL ANALYSIS OF MAJOR LYMPHOID ORGANS OF CHEMOTHERAPY TREATED CHICKENS

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ABSTRACT

The study was carried out to know the gross morphological and morphometrical changes of major lymphoid organs of chemotherapy treated chickens during the period from July to September 2004 in the Department of Anatomy and Histology, Faculty of Veterinary Science, Bangladesh Agricultural University, Mymensingh. The 12-day-old 16 chickens (Vencobb broiler) were divided into four groups viz, group A, group B, and group C that were treated with cyclophosphamide @ 3 mg / kg body weight intramuscularly, vincristine @ 0.07 mg / kg body weight intramuscularly and the combination of the former two drugs using same dose and route for 3 consecutive days respectively and group D was kept as untreated control. After 7 days of last chemotherapy all birds were weighed and killed one by one through cervical subluxation. The body weights of the chemotherapy treated groups were decreased in comparison to control but more decreased in cyclophosphamide treated chickens. The major lymphoid organs (thymus, bursa of Fabricius, spleen and cecal tonsils) were exposed through ventral neck and abdominal dissection and grossly atrophy of the major lymphoid organs were observed in the chemotherapy treated chicken than the control chicken. After collection, the major lymphoid organs were weighed with the help of electric balance one by one. The weights of major lymphoid organs reduced in the chemotherapy treated chickens than the control chickens but the weight of lymphoid organs more reduced in the cyclophosphamide treated chickens. The result of the present study revealed that the size and weight of the major lymphoid organs of the chemotherapy treated chicken decreased in comparison to control chicken. These results indicated that the reduction of weight of major lymphoid organs was due to the action of drug in the lymphoid organs of chemotherapy treated chickens.

Key words: Lymphoid organs, morphology, chemotherapy, chickens

INTRODUCTION

Lymphoid system is a complex and highly sophisticated defense system in the body (Cortan *et al.*, 1989). The lymphoid system of fowl is consisting of unique organs and divided into two morphologically and functionally distinct components (Cooper *et al.*, 1965, 1966). The thymus-dependent component is represented by the smaller lymphocytes and is responsible for cell mediated immunity (CMI), whilst the bursa-dependent component is represented by the larger lymphocytes which transformed into plasma cell in the tissue and plays an important role in humoral immunity (HI). The chicken has central (thymus and bursa of Fabricius) and peripheral (spleen and all mucosa associated lymphoid tissues, MALT) lymphoid tissues (Getty, 1975). Chickens have been used as experimental animals for studies of immune system because T and B cell of the chicken mature in the thymus and bursa of Fabricius respectively and these organs are easily manipulated. The development, differentiation, histological distribution of lymphocytes in these tissues, including caecal tonsil and spleen of chickens, have been investigated by using surgical thymectomy, bursectomy, X-ray irradiation, colloidal carbon uptake method (Hoshi and Mori, 1973; Sugimura and Hashimoto, 1980; Befus *et al.*, 1980; Graetzer *et al.*, 1963; Papermaster and Good, 1962).

Therefore, immunosuppressive drugs (cyclophosphamide, vincristine and their combination) were used to study the morphometry of major lymphoid organs of chickens, which will be helpful to the immunologist and veterinarians for better understanding of morphometric changes in immunosuppressive drugs treated bird, man and animals in case of treatment lymphoproliferative diseases.

MATERIALS AND METHODS

The present study was conducted in the Department of Anatomy and Histology, Faculty of Veterinary Science, Bangladesh Agricultural University, Mymensingh from July to September 2004 on the major lymphoid organs of 16 chemotherapy treated chickens. The 12-day-old 16 chickens (Vencobb broiler) were purchased from the Al-Aarafa Poultry Complex, Mymensingh. The chickens were reared in the departmental poultry house with commercial feed and fresh drinking water *ad libitum* and with sufficient air and light. The chickens were divided into four groups (groups A, B, C and D) and each group consisted of 4 chickens.

At 15 days old, the chickens of groups A and B were injected with cyclophosphamide (Endoxan® 200 mg vial, German Remedies Limited) @ 3 mg / kg and vincristine (Vincristin® 1 ml vial, Medipex) @ 0.07mg / kg body weight intramuscularly for three consecutive days respectively. The chickens group C were treated with the combination of the former two using same dose and route for three consecutive days and chickens of group D received no injection and served as control. The chemicals were dissolved and diluted in distilled water (200mg cyclophosphamide in 10 ml and 1 ml vincristine in 5 ml distilled water) and injected through intramuscular route using tuberculin syringe once a day for 3 consecutive days.

After 7 days of last chemotherapy all the chickens including control were weighed and killed one by one by cervical subluxation. The major lymphoid organs (thymus, spleen, bursa of Fabricius and cecal tonsil) were exposed through the ventral neck and abdominal dissection and gross morphology observed including their color and presence of any gross lesions and/or deformities. The major lymphoid organs then collected carefully by using forceps and scalpel and weighed with the help of electric balance. The level of significance of weight of major lymphoid organs between chemotherapy treated and control chicken were determined by Student's 't'-test (Zar, 1974).

RESULTS AND DISCUSSION

The total body weights of the chemotherapy treated groups (groups A, B and C) were reduced in comparison to the control group chickens (group D) (Table 1). It was due to loss of appetite and action of chemotherapy.

Table 1. The total body weight and the mean weight of major lymphoid organs of different chemotherapy treated and control groups of chickens [Mean \pm SE (g)]

S/N	Groups (n = 4)	Treatment	Body weight	Bursa of Fabricius	Thymus	Spleen	Cecal tonsil
1	A	Cyclophosphamide treated chickens ^a	446.25 \pm 51.61	0.7525 \pm 0.2101**	1.2850 \pm 0.3905**	0.4550 \pm 0.0843*	0.3925 \pm 0.0675*
2	B	Vincristine treated chickens ^b	540.00 \pm 25.50	0.8375 \pm 0.15992**	1.5125 \pm 0.6098*	0.7375 \pm 0.1841*	0.4500 \pm 0.0289**
3	C	Cyclophosphamide and vincristine treated chickens ^c	467.50 \pm 37.50	0.7600 \pm 0.08134*	1.5325 \pm 0.3277**	0.6525 \pm 0.0579**	0.5125 \pm 0.0343*
4	D	Control chickens	710.25 \pm 8.52	1.0425 \pm 0.13344	2.7152 \pm 0.4293	0.9250 \pm 0.1127	0.5375 \pm 0.0125

^adose rate 3 mg / kg body weight intramuscularly; ^bdose rate 0.07 mg / kg body weight intramuscularly; ^cdose rate and route are same as groups A and B; n = Total number of birds; SE = Standard error; *Significant at ($p < 0.05$); **Significant at ($p < 0.01$).

Thymus

The thymus of the chicken of control group was a paired lobulated gland, one half of which was located either side of neck in subdermal location. Each half consisted of six to eight, flattened pale white lobes of varying size of lymphoid tissue lying in the subdermal connective tissue of the neck (Fig. 1). The shape and color of the thymus were similar to that report of King (1975) and Hodges (1974) in the hybrid chickens. The upper 1/5th of the neck was devoid of thymus and the gland was closely associated with the jugular vein and vagus nerve and surrounded by considerable connective tissue and adipose tissue. The finding was similar to the report of Bacha (1978). The size of the thymus reduced in all chemotherapy treated chickens (Fig. 3). The mean weight of the thymus in treated chickens were 1.2850 \pm 0.3905 g (Cyclophosphamide treated chickens), 1.5125 \pm 0.6098 g (Vincristine treated chickens), 1.5325 \pm 0.3277 g (combined treated chickens) and in control group of chickens it was 2.7125 \pm 0.4293 g (Table 1). The weight of thymus in all the groups of chemotherapy treated chickens was reduced in comparison to control but more reduced in cyclophosphamide treated chickens (group A). It may be due to the action of drug in the thymus.

Bursa of Fabricius

The bursa of Fabricius of the chickens of the control group was a single lymphoepithelial organ appeared as a dorsal median diverticulum of the proctodeum, being smooth and globular in shape and yellowish white in color and lack of any gross lesion (Fig. 2). The location, shape and color were similar to the report of Hodges (1974) in hybrid chickens. Grossly, atrophy of the bursa was observed in all of the chemotherapy treated chickens (Fig. 4).

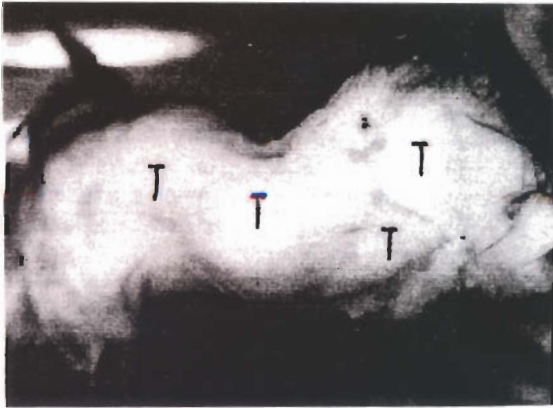


Fig. 1. Gross photographs of the of thymus (T) in the sub-dermal location of control chicken.

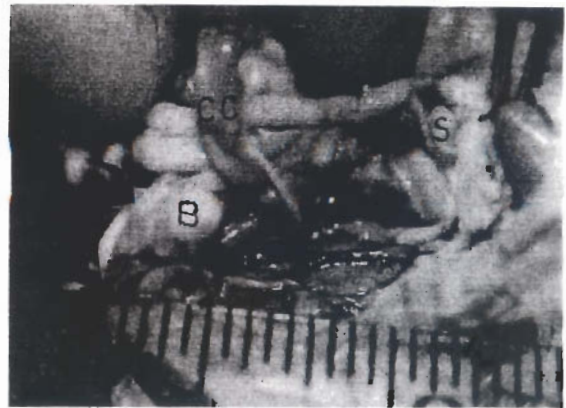


Fig. 2. Gross photographs of the bursa of Fabricius (B), spleen (S) and cecal tonsils (C) of the control chicken.

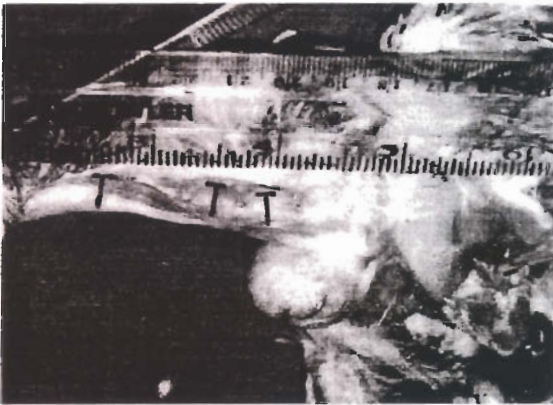


Fig. 3. Gross photographs of the atrophied thymus (T) of the chemotherapy treated chicken.



Fig. 4. Gross photographs of the atrophied bursa of Fabricius (B), spleen (S) and cecal tonsils (C) of the chemotherapy treated chicken.

The mean weight of the bursa of Fabricius was 0.7525 ± 0.2101 g in cyclophosphamide treated chicken, 0.8375 ± 0.1599 g in vincristine treated chicken, 0.7600 ± 0.0813 g in combined treated chickens and 1.0425 ± 0.1334 gm in control chickens (Table 1). It was observed that the weight of bursa of Fabricius in all the groups of chemotherapy treated chickens were decreased in comparison to control chickens. The weight of bursa of Fabricius more decreased in cyclophosphamide treated chickens. The reduction of weights of the bursa was due to action of chemotherapy. The findings were similar to the report of Khan *et al.* (1998) in Dekalb strain chicken of Japan.

Spleen

The color of the spleen of chickens of control group was reddish brown and free from any deformity in shape and size which was similar to the observations made by King (1975) in hybrid chickens. Grossly atrophy of the spleen was observed in all the chemotherapy treated chickens (Fig. 4) in comparison to control chickens. The mean weight of the spleen was 0.4550 ± 0.0843 g in cyclophosphamide treated chicken, 0.7375 ± 0.1841 g in vincristine treated chicken, 0.6525 ± 0.0579 g in combination of the former two drugs treated chickens and 0.9250 ± 0.1127 g in control chickens (Table 1). It was observed that the mean weight of spleen in the all groups of chemotherapy treated chickens were decreased in comparison to control but more decreased in cyclophosphamide treated chickens and it may be due to action of chemotherapy in the spleen. The findings coincide with the result observed by Khan *et al.* (1998) in Dekalb strain chicken of Japan.

Cecal Tonsil

The cecal tonsils of the chickens of the control group were two in number and located bilaterally at the junction of small and large intestines (Fig. 2). This observation similar to the report of Looper and Looper (1929) in the bantam fowl. The cecal tonsil was consisted of mucosa associated lymphoid tissue (MALT) which was found at proximal part or base of the two cecum. The color of two tubular structured cecal tonsils was yellowish white both in chemotherapy treated groups and control group. These findings were similar to that of the cecal tonsils of the hybrid chicken (King, 1975; Hodges, 1974). The weight of the cecal tonsils were 0.3925 ± 0.0675 g in cyclophosphamide treated chicken, 0.4500 ± 0.0289 g in vincristine treated chicken, 0.5125 ± 0.0343 g in combination of the former two drugs treated chickens and 0.5375 ± 0.0125 gm in control chickens. The weight of the cecal tonsils in chemotherapy treated chickens was reduced in comparison to control group. The findings were similar to that of the result reported by Khan *et al.* (1998) in Dekalb strain chicken of Japan.

In conclusion, the present study revealed that the reduction of the size and weight of the major lymphoid organs of chemotherapy treated chickens were due to the action of drug on the lymphoid organs.

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