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IMMUNOPHYSIOLOGICAL STUDIES ON THE EFFECT OF FOWL POX VACCINE ON THE BLOOD PICTURE

(With 2 Tables)

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دراسات فسيولوجيه مناعيه على تأثير لقاح جدري الطيور على صورة الدم

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تمت دراسة التغير في صورة الدم في دجاج الهيرد قبل وبعد التحصين بلقاح جدري الطيور - هذا وقد شملت الدرسة عدد كرات الدم الحمراء والبيضاء في المليمتر المكعب من الدم علاوة على تقدير العدد المتفرق لكرات الدم البيضاء ونسبة الهيموجلوبين في الدم بالاضافه الى تعيين البروتين الكلى في سيرم الدم. وقد اوضحت النتائج إختلافا كبيرا في صورة الدم بين الطيور في المجموعه المحصنه والطيور في المجموعه الضابطه غير المحصنه نتيجة تأثير فيروس اللقاح. كما أجرى إختبار التعادل المصلى على سيرم الدم لقياس مستوى الأجسام المضاده، وأوضحت النتائج أن المناعه المكتسبه قد وصلت إلى أعلى مستوى خلال الأسبوع الثانى وأستمرت إلى الأسبوع السادس بعد التحصين.

SUMMARY

The blood picture of Habbard chickens was studied before and after vaccination with fowl pox vaccine. Total erythrocytes and leucocytes counts (in mm^3 blood), differential leucocytic count (in %), haemoglobin content (in Gr%) were calculated in the whole blood. In addition total serum protein content was calculated in the blood serum. The obtained results indicated a great variation between vaccinated and nonvaccinated birds. The serum samples were tested for serum neutralizing antibody level and showed that the acquired immunity reached to its highest level within the second week post vaccination and persisted to the 6th week.

Key Words: Fowl Pox Vaccine-Blood Picture.

INTRODUCTION

Changes in the blood picture may be aid in the diagnosis of different diseases caused by bacteria or viruses (Schilling 1929). The blood picture of the chicken vaccinated with fowl pox vaccine have not adequately studied, therefore the main object of this study is to record the haematological changes of Hubbard chickens vaccinated with fowl vaccine in comparison with the immune response.

MATERIAL and METHODS

1. Chickens:

One hundred, 45 days old apparently healthy Hubbard chickens were divided into two groups, the first group was kept without vaccination (control), while the second one was used for vaccination.

2. Embryonated chicken eggs:

Five Hundred fertile chicken eggs (ECE), 9-11 days old obtained from El-Motaheda poultry company, Cairo. These ECE were used in the neutralization test.

3. Viruses:

a) Vaccinal strain:

Egg adapted Beaudette strain of fowl pox vaccine, produced in Veterinary Serum and Vaccine Research Institute Pox Department, Abbassia, Cairo. It was administered to the chickens by the stick wing web (w.w) method in the right wing.

b) Virulent strain:

Egg adapted Egyptian virulent strain of fowl pox, isolated and identified by Sabban (1954) was used for challenge the vaccinated and non vaccinated control chicken one month after vaccination in the left wing by w.w. method.

4. Chemicals:

a) Sodium salt of ethylene diamine tetra acetic acid (EDTA) was used as anticoagulant for haematological studies.

b) Giemsa stain, was used for staining of the blood films for differential leucocytic count.

5. Haematological procedure:

Whole blood samples were obtained from each chicken of the two groups: 7, 10, 12, 15, 21, 30 and 45 days post vaccination in clean dry sterile bottles containing EDTA at a concentration 3 mg/ml of blood, which were used for estimation of:

a) Haemoglobin content (Hb) which was estimated as gm/100ml of blood (gm%) by sahli's haemometer.

b) Total Erythrocyte (RBCs) and Leucocyte (WBCs) counts per mm^3 blood were counted using the haemocytometer according to Thoma's methods. The diluent used in this study was Hayem's solution for red cells and Nutt and Herrick solution (1952) for white cells.

c) Differential leucocytic count was done by the standard methods of Battelment described by Schalm *et al.* (1973) which was calculated after staining the blood film taken from each chicken with Giemsa stain.

6. Total serum protein:

5 ml of blood samples were taken from each chicken by heart puncture methods in sterile dry vials for serum separation which were used for estimation of the total serum protein by Weichselbaum method (1946).

7. Serum neutralization test:

Was applied according to the methods of Boulter (1957) using serial two fold dilution of serum mixed with fowl pox antigen suspension ($100 \text{ EID}_{50}/0.1\text{ml}$) in equal volumes. The mixtures were kept at 37°C for one hour, then 0.1ml of virus serum mixtures was inoculated on the chorioallantoic membrane (CAM) the 9-11 days old ECE. Virus control (positive), were prepared by inoculating fowl pox virus on the VAM of five ECE. Negative control for each serum samples were made by inoculating the samples and saline mixture by the same manner. Inoculated eggs were sealed by milted wax and incubated at 37°C for five days with daily candling. Results were interpreted as pock lesions recorded and calculated according to the formula of Reed and Muench (1938).

RESULTS and DISCUSSION

Fowl pox is a common infectious disease of poultry in Egypt (Sabban 1954); and there are no available data on its effect on the blood picture of infected or vaccinated birds. Our results indicated that there are no effect of fowl pox virus on the total red blood cell count per mm^3 , blood which ranged from 4-4.5 million/ mm^3 blood (table 1) when compared with the normal RBCs count of habbard chicken (meat producing fowl) which is usually high, reaching up to 5.1 million/ mm^3 blood (Cook and Dearstyne 1934, Tanaka and Rosenberg 1954, Kodriatsov *et al.* 1969 and Zietsov 1971).

The haemoglobine percentage was slight less than normal 9.3gm% (the normal percentage in meat producing fowl is 11.2-12.8 gm).

The total serum protein was increased from the 2nd to the 6th week post vaccination which indicated the presence of an increase in the antibody formation and in the immunological capacity especially if we put in consideration that the pox virus is one of the immune stiulating agents (Mayr *et al.* 1985). this results agree with (Boguth 1954; Sadakova *et al.* 1967; Venzil 1967 and Zietsoy 1971) who reported that the blood protein play an important role in supplying immune bodies and other physiological function as synthesis of anzymes and hormones, transportation of blood constituents and maintenance of suitable osmotic pressure.

Total leucocytic count was increased and reached to its peak 12-30 days after vaccination which also indicated the presence of an improvement in the immunological function of the vaccinated chickens at this time (Babsky *et al.* 1970).

The neutrophil is the first line of defense in the blood against the different pathological aagent (Rouse *et al.* 1980) Therefore the slight occurrence of neutrophilia at first and then decreasing of it to sub normal level is the results of the use of these neutrophils against the viral agent. On the contrary the presence of high lymphocytes percentage especially one month post vaccination, indicated the presence of the disease or vaccination (Talaat 1955).

The slight increase in the eosinophils may be attributed to the effect of the pox virus on the skin or due to antigen-antibody reaction.

Monocytes and basophiles nearly persisted constant. This indicated that they have no role in the protection against fowl pox virus.

The results of the serum neutralization test confirmed the former obtained results, in which the neutralizing antibodies, against pox virus were performed and increased from 2nd to the 6th week post vaccination (Table 2).

Our findings demonstrated that there are changes in the blood picture after vaccination or challenge with fowl pox viruses which help in the diagnosis of infection and in the judgment on vaccination in field.

Further studies are necessary to investigate the relationship between the biochemical components of the blood and fowl pox viruses.

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TABLE (I)
HAEMATOLOGICAL EFFECT OF FOWL POX

MEAN VALUE	CONTROL*	B.V	DAYS POST VACCINATION							
			7	10	12	15	21	30	45**	
RED CELL COUNT 10 ⁶ /mm ³	4.1	4.0	4.0	4.1	4.0	4.2	4.1	4.2	4.2	4.2
HAEMOGLOBIN (gm%)	9.1	9.15	9.2	9.1	8.8	8.9	8.9	8.9	9.2	9.3
TOTAL SERUM PROTEIN (gm/%)	5.1	5.3	5.5	5.8	5.8	6.1	6.2	6.2	6.2	6.0
TOTAL LEUCOCYTIC COUNT (10 ³ /mm ³)	40	40.5	41.0	43.0	42.6	45.0	46.0	46.0	46.0	44.0
NEUTROPHIL %	31.1	30.5	33.5	35.0	36.0	33.0	28.5	23.0	21.2	21.2
LYMPHOCYTE %	60.1	61.0	59.0	56.3	55.0	57.7	62.0	67.6	69.0	69.0
MONOCYTE %	4.9	4.8	4.9	4.9	5.0	5.0	5.1	5.2	5.2	5.2
BASOPHIL %	0.9	0.7	0.7	0.8	0.9	0.8	0.8	0.7	0.8	0.8
EOSINOPHIL %	3.0	3.0	2.9	3.0	3.1	3.5	3.6	3.5	3.5	3.8

* Control group had slight changes at different time that was of no significant imprtance.

** = 15 days post challenge

B.V = Before vaccination

Total serum protien was increased from the 10th to the 45th days post vaccination.
Neutrophile percent was increased at first then decreased from the 3rd week after vaccination and on the contrary the lymphocytes % was increased.

TABLE (2)
SERUM NEUTRALIZATION TEST

TIME OF SAMPLING	NEUTRALIZING ANTIBODY TITER (Geometric mean)	
	G I	G II
B.V	0	0
7 DPV	1.93	0
14 DPV	3.10	0
21 DPV	4.96	0
28 DPV	4.66	0
7 DPC	3.96	1.7
14 DPC	4.83	3.13
21 DPC	4.70	4.72

- B.V. = Before vaccination.
DPV = Days post vaccination
DPC = Days post challenge
G I = vaccinated and challenged group
G II = Non vaccinated and challenged group

Year	1900	1901	1902	1903	1904	1905	1906	1907	1908	1909	1910	1911	1912	1913	1914	1915	1916	1917	1918	1919	1920	1921	1922	1923	1924	1925	1926	1927	1928	1929	1930	1931	1932	1933	1934	1935	1936	1937	1938	1939	1940	1941	1942	1943	1944	1945	1946	1947	1948	1949	1950	1951	1952	1953	1954	1955	1956	1957	1958	1959	1960	1961	1962	1963	1964	1965	1966	1967	1968	1969	1970	1971	1972	1973	1974	1975	1976	1977	1978	1979	1980	1981	1982	1983	1984	1985	1986	1987	1988	1989	1990	1991	1992	1993	1994	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021	2022	2023	2024																																																								
Population	100	105	110	115	120	125	130	135	140	145	150	155	160	165	170	175	180	185	190	195	200	205	210	215	220	225	230	235	240	245	250	255	260	265	270	275	280	285	290	295	300	305	310	315	320	325	330	335	340	345	350	355	360	365	370	375	380	385	390	395	400	405	410	415	420	425	430	435	440	445	450	455	460	465	470	475	480	485	490	495	500	505	510	515	520	525	530	535	540	545	550	555	560	565	570	575	580	585	590	595	600	605	610	615	620	625	630	635	640	645	650	655	660	665	670	675	680	685	690	695	700	705	710	715	720	725	730	735	740	745	750	755	760	765	770	775	780	785	790	795	800	805	810	815	820	825	830	835	840	845	850	855	860	865	870	875	880	885	890	895	900	905	910	915	920	925	930	935	940	945	950	955	960	965	970	975	980	985	990	995	1000

The following table shows the population of the United States from 1900 to 2024. The population has increased steadily over the period, with a significant acceleration in growth starting around 1950. The population reached approximately 330 million in 1950 and is projected to reach approximately 400 million by 2024.