

Development of sprayer to the control of poultry
viral respiratory disease

Development of multi-purpose sprayer to vaccination

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Evaluation of protection effect and immunization of
sprayer to viral respiratory disease

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Evaluation of protection effect to viral respiratory
disease in poultry flocks

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가 (3) 가 .
Module-T 가 1.2

가 가 .
1 480
1.3 . 1 10
58 μm - 70 μm 가 가 . 46 μm
1.4 가 .
가
가

Table 1.1. General specification of Module-T type sprayer for vaccination

parts	feature	
spray apparatus	material of nozzle	ceramic
	rotation of nozzle	360 ⁰
	number of spray nozzle	3ea
	spray volume	75ml/min
mechanic apparatus	spray motor	7.5HP/ 100A

Table 1.2. Characteristics of spray particles of Module-T type sprayer for vaccination

No of test	average particle size μ m (min - max)	SD	50- 100 μ m/total observation (%)	spray volume (ml/min)
1	189(55.0- 452.06)	68.50	22/ 250(8.8)	75
2	150(42.00- 370.50)	52.70	54/ 295(18)	75
3	140.20(32.00- 412.34)	54.25	58/ 340(17)	75
4	192(48.24- 432.28)	76.46	34/ 242(14)	75

Table 1.3. Comparison of antibody titers after Newcastle disease virus vaccination using Module-T type sprayer

days	particle size		
	46 μ m	58 μ m	70 μ m
14	34.50	95.30	80.28
21	35.20	72.42	14.28
28	27.34	32.24	112.224

Table 1.4. Distribution of antibody titers in unvaccinated control group

days	antibody titers
	20.20
14	18.42
21	119.06
28	32.24

2.

가

가

가

가

50 μ m, <100 μ m,

>100 μ m

ND, IB

가

100 μ m

(coarse spray)

가

가

가

가

가

100 μ m

(coarse spray)

가

100 μ m - 400 μ m

가

가

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1

2⁵, 3

2³, 5

2^{2.5}

100 μ m

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3

2⁴, 5

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1

500

coarse spray

ND AVINew

IBH20

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 가 .
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 가
 가 (9003) ELISA (7000) 49
 IB 가 49 2^{4.7} 49
 2^{3.7} 가 IB
 가 ELISA 49 가
 (28383) (8600)
 ND (6 log₁₀EID₅₀/dose) IB
 (3 log₁₀EID₅₀/dose) 30 가
 ND IB ELISA H 가 가 가
 30
) 가 가
 500 (23) 100 (23)
 t-test 1.81 ±0.137Kg
 1.79 ±0.164Kg (0.05 <P)
 19 41
 (0.05<P)
 가 500 (6) 100
 (6) t-test
 1.08 ±0.127Kg 1.09 ±0.104Kg
 (0.05 <P) 가

가

(spray)

가

가

가

가

SUMMARY

As the major portal of entry for disease, invasion of the respiratory tract by microbial agents and abnormal physical conditions in the house will lead to interference of the respiratory system, affecting the health and performance of the flock. High temperature, humidity, dust load, ammonia concentration and poor cross ventilation can lead to a high incidence of respiratory disease complex (RDC). This is the condition or syndrome in which primary multiple etiological agents are involved, affecting the respiratory system.

Causal agents and factors

The primary causal agents of RDC are Newcastle Disease, Infectious Bronchitis, Infectious laryngotracheitis and *Mycoplasma gallisepticum*. RDC is also affected by environmental factors such as the ambient temperature, humidity, ammonia and other gases in the house. Stocking density, ventilation etc. and some intrinsic factors such as genetic make up, immune response, nutritional status etc., are all predisposing factors and result in a more severe respiratory disease than the condition caused by primary agent alone. The third important factor in considering the epidemiology of RDC is disease transmission, which involves the spread of the primary agent from one host to another, and also includes secondary microbial factors. Transmission occurs by either direct or indirect means. The spread of the disease is influenced by the availability of a

susceptible population, the route of entry and exit, rate of multiplication and degree and duration of the disease, duration of excretion of the agent, and survival of the agent in the environment.

The simultaneous infection of pathogenic *E. coli* and *Mg* results in a more severe form of the disease condition. Combinations of either *ND* or *IB* virus along with *Mg* and/or pathogenic *E. coli* also cause more severe disease with mortality. The severity is further increased by predisposing environmental factors.

In hatcheries, aspergillosis (a fungal infection) is a major cause of respiratory infection in chicks. Avian influenza, diphtheric fowl pox, fowl cholera and infectious coryza are also involved in respiratory conditions. RDCs which involve other organs in the body include turkey rhinotracheitis (TRT) and ornitho rhinotracheitis (ORT).

Threats to respiratory immunity

The avian immune system consists of the primary lymphoid organs (bursa, thymus) and the secondary lymphoid organs (spleen, harderian gland, caecal tonsils etc). The lymphoid cells aggregated in the respiratory system are the primary defence mechanism in controlling the spread of infection. When the pathogens are inhaled, the first immune cells to become exposed are those of the respiratory tract. These cells mount a local immune response, stimulating first an immune response in other lymphoid organs and then a systemic immune response to control the disease situation. The harderian gland, the secondary lymphoid organ of the respiratory tract, plays an important role in controlling the disease agent as well as generating a local vaccine response.

There are a number of stress factors and infectious agents which compromise immune competence in poultry. Generalised immunosuppression also affects respiratory tract immunity, which involves both antibody and cellular immunity.

The most important immunosuppressive agent is IB viral infection which results in a reduced local immune response in the respiratory tract.

MD infection also leads to T cell immunosuppressive conditions. Aflatoxins and some other viral infections also lead to this condition. Poor ventilation, high ammonia gas levels, over-crowding, high humidity and substandard nutritional conditions all compromise respiratory functions.

Newcastle disease(ND)

Newcastle disease is caused by the parainfluenza virus group. The viruses in this group have varying degrees of virulence: velogenic (highly virulent), mesogenic (moderately virulent) and lentogenic (avirulent), which lead to disease conditions ranging from high mortality to a very mild or even asymptomatic infection. Based on the tropism (response to external stimulus to grow in certain directions) some are neurotropic, pneumotropic, or viscerotropic. The velogenic forms are based on tropism and the organs involved are termed as very virulent viscerotropic, very virulent neurotropic or pneumotropic forms. But this is not absolute. The ND live vaccine strains are mainly derived from mesogenic and lentogenic types of virus strains.

The disease and environmental conditions with involvement of MG, E. coli and IB will increase the severity of disease, including the level of mortality and drops in egg production. The diagnosis and isolation of the primary disease agent and secondary pathogens is important in preventing and controlling the disease situation, as well as for the application of an effective vaccination programme. Periodical serological monitoring of the flock will provide information about a vaccination response and exposure to the disease.

Infectious bronchitis(IB)

This infection is caused by the corona groups of viruses. IB is a rapidly

spreading contagious disease with a very short incubation period. The involvement of antigenetically different serotypes is characteristic of this disease. Isolation, serotype identification and application of a serotype specific vaccination programme is the most important factor in controlling this disease. The respiratory system reproduction system and the kidneys are involved in this disease. The respiratory disease is usually very mild, rarely giving rise to mortality. However a very virulent strain has recently emerged.

IB infection at an early age may permanently damage the oviduct, and in the adult stage affected hens appear as false layers. Infection at point of lay or later causes a reduction in egg quality and drop in egg production which may never return to normal. In the renal form which generally affects young stock, there is marked nephritis and uraemia leading to visceral gout with some mortality in the affected flock.

Secondary complications with the involvement of *Mg* and pathogenic coli increases the severity of IB infection. A simultaneous infection of IB and ND can increase the severity of both the disease. The combined involvement of IB and ILT exacerbates both conditions. The synergetic effect of dual infection of coryza and IB is also documented.

Various stress factors including bad ventilation, high ammonia levels, humidity and other stressors increase the severity of the condition. Diagnosis of the disease, isolation of the primary causal agent and typing for an effective vaccination programme is important to control IB.

In most countries of Europe and Korea the control of Newcastle disease and Infectious bronchitis is a major respiratory disease in poultry industry. The control of these disease is based primarily on veterinary administrative measures. In some cases these measures may be supported by vaccinations subject to special authorization. Of vaccines produced from lentogenic viruses strain

Bl and Lasota are most widely used. These strains are administered mainly via the drinking water, by eyedrop or intranasal instillation or more recently, in spray form. Spray vaccination provides the best protection; however sometimes it can cause respiratory signs and activate mycoplasmosis. In an effort to avoid this, recently vaccines prepared from apathogenic strains have come to the fore.

1. Development of multi-purpose sprayer to vaccination

The technical development has been focused on testing the interaction of atomizing and separating system. This resulted in the equipment capable of producing effective aerosol 75ml/min, 94 to 95% of above aerosol consists of droplets with a size 100 μm . The equipment works on electricity. The aerosol generator can be used for not only the vaccination but also the disinfection of closed space as well as for disinfection. Spray vaccination with 58 - 70 μm particle size induced higher serological response than other particle size.

2. Evaluation of protection effect and immunization of sprayer to viral respiratory disease

One-day old chicks from 5 groups were immunized by different methods of vaccination: 1) spray: 2) eyedrops 3) unvaccinated. The flocks were vaccinated by one of the following methods: 1) spray vaccination with developed sprayer with 50 μm , <100 μm and >100 μm particle size with Newcastle disease virus and IB virus 2) spray vaccination with botanical sprayer 3) eye drops vaccination. Blood samples were taken on weeks 1, 3 and 5 for HI and ELISA test. Spray vaccination with <100 μm particle showed higher HI titer 2^8 by 1 week and 2^3 by 5 weeks age.

3. Evaluation of protection effect to viral respiratory disease in poultry flocks

One-day old chicks from 2 groups were immunized by different methods of vaccination: 1) spray and drinking water, eyedrops 2) unvaccinated. The flocks were vaccinated by one of the following methods: 1) spray vaccination with developed sprayer from 50 μm <100 μm >100 μm particle size with Newcastle disease virus and IB virus. The immune response of commercial layer chickens against Newcastle disease and Infectious bronchitis were compared among different methods. A total of 1000 day-old chickens were divided into 2 groups of 500 birds each. At two and 4 weeks after each time of vaccination, 50 birds from each group were challenged with virulent ND virus at the dose of $6 \log_{10}$ EID₅₀ and IB virus at the dose of $3 \log_{10}$ EID₅₀ per bird to examine the protection rate. 10 to 20 birds from each group were bled at two weeks interval to determine HI and ELISA titer. Spray vaccinated group showed higher ELISA titer (28383) compared to eye drops (8600) on 49 days. When birds were challenged death was not occurred in vaccinated group. To compare the production performance, body weight and egg production was compared in chickens vaccinated or control group. The average of body weight of vaccinated group was 1.08 ± 0.127 and that of body weight of control group in 1.09 ± 0.104 in control group. There was not significant difference in production performance between vaccinated and control group.

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 가 (3) 가 .

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Module-T 가 1.2
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 . 가 가 .
 1 480
 1.3 . 1 10
 58 μm - 70 μm 가 가 . 46 μm
 1.4 가 .
 가
 가

Table 1.1. General specification of Module-T type sprayer for vaccination

parts	feature	
spray apparatus	material of nozzle	ceramic
	rotation of nozzle	360 ⁰
	number of spray nozzle	3ea
	spray volume	75ml.min
mechanic apparatus	spray motor	7.5HP/ 100A

Table 1.2. Characteristics of spray particles of Module-T type sprayer for vaccination

No of test	average particle size μ m (min - max)	SD	50- 100 μ m/total observation (%)	spray volume (ml/ min)
1	189(55.0- 452.06)	68.50	22/ 250(8.8)	75
2	150(42.00- 370.50)	52.70	54/ 295(18)	75
3	140.20(32.00- 412.34)	54.25	58/ 340(17)	75
4	192(48.24- 432.28)	76.46	34/ 242(14)	75

Table 1.3. Comparison of antibody titers after Newcastle disease virus vaccination using Module-T type sprayer

days	particle size		
	46 μ m	58 μ m	70 μ m
14	34.50	95.30	80.28
21	35.20	72.42	14.28
28	27.34	32.24	112.224

Table 1.4. Distribution of antibody titers in unvaccinated control group

days	antibody titers
	20.20
14	18.42
21	119.06
28	32.24

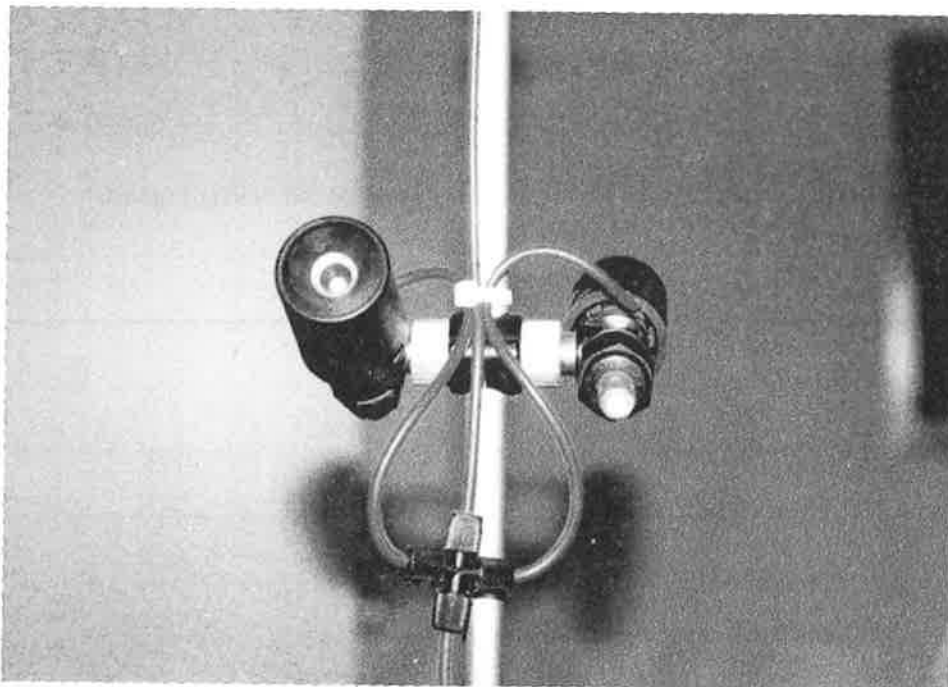
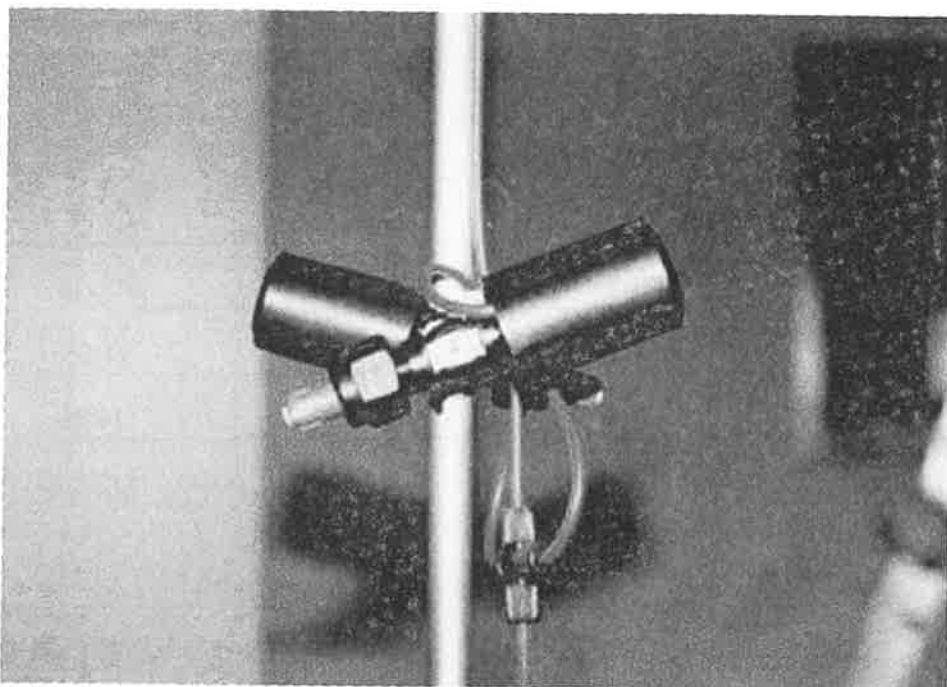


Fig1. Feature of ceramic nozzle of Module- T sprayer for vaccination.

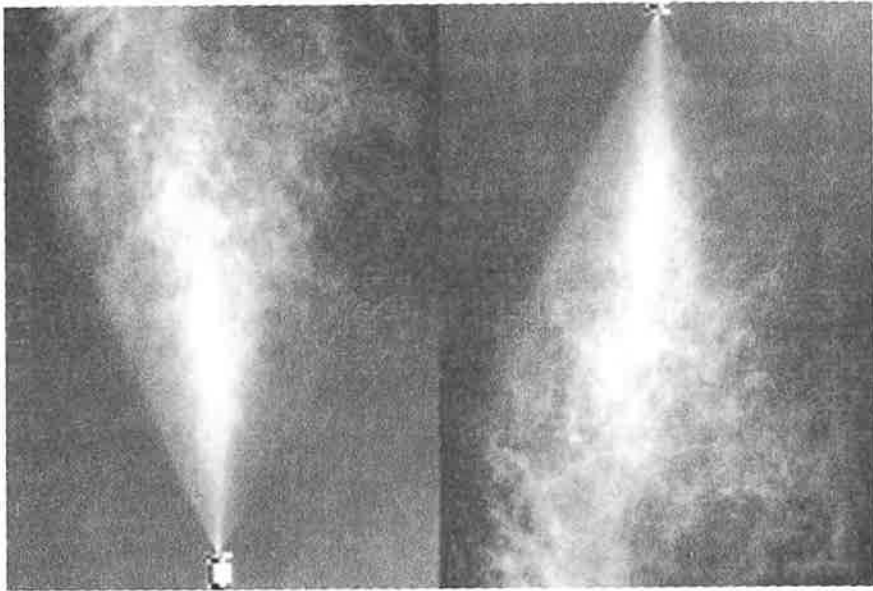


Fig2. Feature of spray nozzle which sprays coarse particle size.

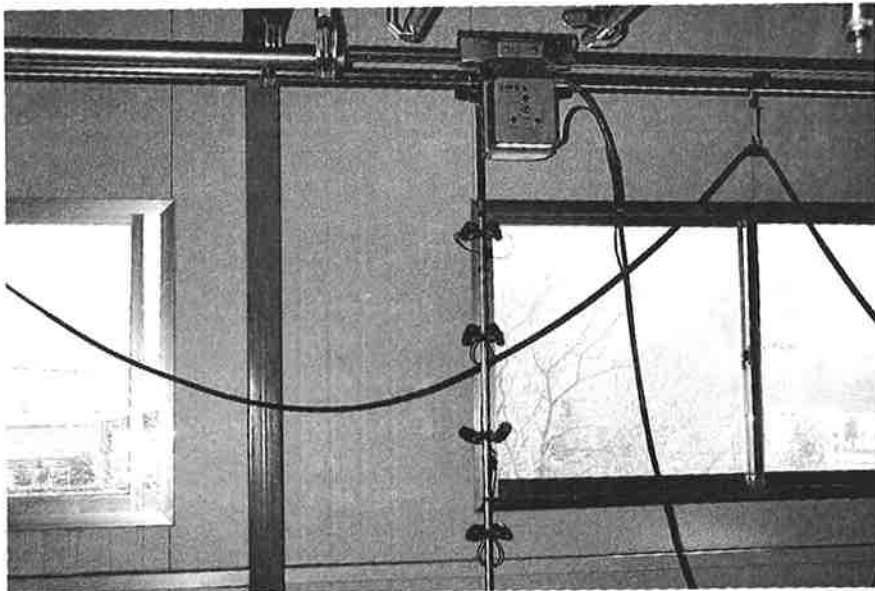


Fig3. Feature of automatic Module- T type sprayer system.

개발된 분무기의 백신접종용으로의 사용은 분무된 입자의 균일도에 의해서 결정된다. 기존의 양계농장에서 백신 접종용으로 사용되고 있는 분무기는 주로 원예용 분무기를 사용하여 왔으며 이러한 분무기가 갖는 분무 입자의 불규칙, 압력의 불균형에 의한 입자 크기의 불균질은 효과적인 백신 접종의 기대를 갖을 수 없었다.

그러나 본 연구에서 확인된 평균 입자 크기 $100 - 150 \mu\text{m}$ 은 바이러스 입자를 상부기관지에 도달 시키는데 적절하다고 생각된다. 바이러스의 상부 기관지 정착은 효과적인 국소면역 반응을 유도 할 수 있을 뿐 아니라 모체 이행항체의 간섭현상을 극복할 수 있어 1일령의 병아리에게 사용할 수 있는 효과적인 백신접종 방법으로 선택할 수 있다.

2.

가

1

가

가

가

2

ND IB 1 A, B, C
 D 20 A
 50 μ m B 가 100 μ m
 , C D 100 μ m E
 ND IB
 ND IB H 가 ELISA 가
 HI test β method 가 16
 2¹, 2², 2³, 2⁴ log₂
 1, 2, 3, 4 NDV IB 가 ELISA 가
 Indirect method ND IB coating ELISA plate 가
 enzyme labelling
 ELISA KPL 가

3

					50 μ m, <100 μ m, . 50 μ m	
>100 μ m	ND, IB		가			
	5	ND	2 ^{3.5}	가		
100 μ m	(coarse spray)		6	2 ⁴	가	
	가	가				
	가	.			가	
가						
(7) . 100 μ m		(coarse spray)			가	
		100 μ m - 400 μ m			가	
		가				
			가	1	2 ⁵ , 3	
2 ³ , 5	2 ^{2.5}	100 μ m		가	1	2 ⁸ ,
3	2 ⁴ , 5	2 ³			가	(
2.4).		1		ND	IB	100 μ
m	1	6	2 ⁴ - 2 ⁸		가	
				ELISA	가	
3000 - 6000	가	.				1
	ND	IB				
	가					

Table2.1. HI titers against ND after vaccination with AVINew or with AVINew + IBH120 spray with 50um particle size

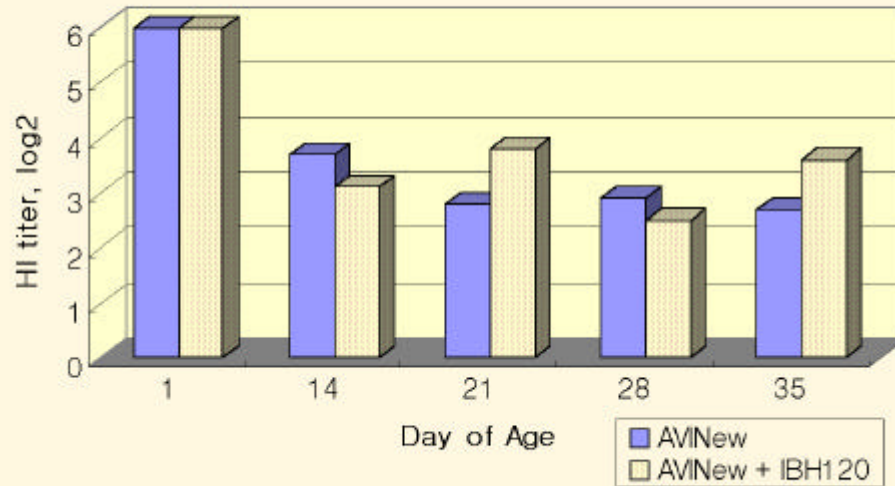


Table2.2. HI titers against ND after AVINew or AVINew + IBH120 spray with < 100um particle

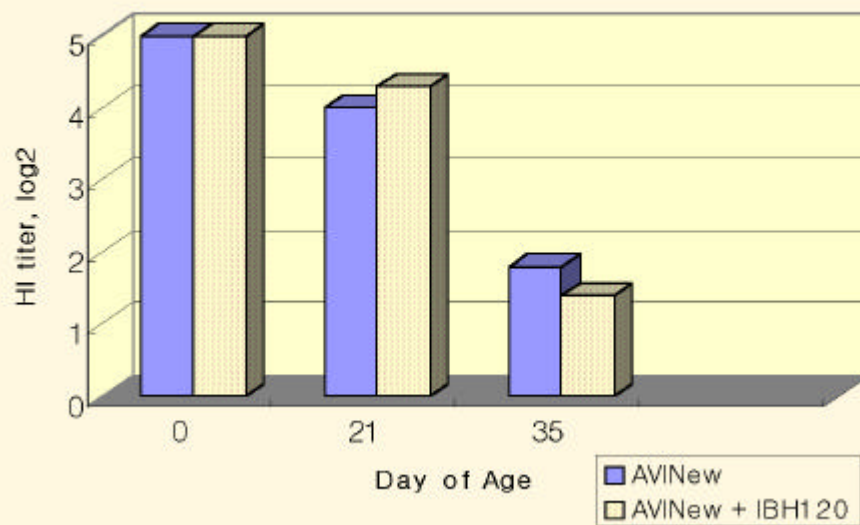


Table2.3. HI titers against ND after immunization with AVINew or with AVINew + IBH120 spray with >100um particle size

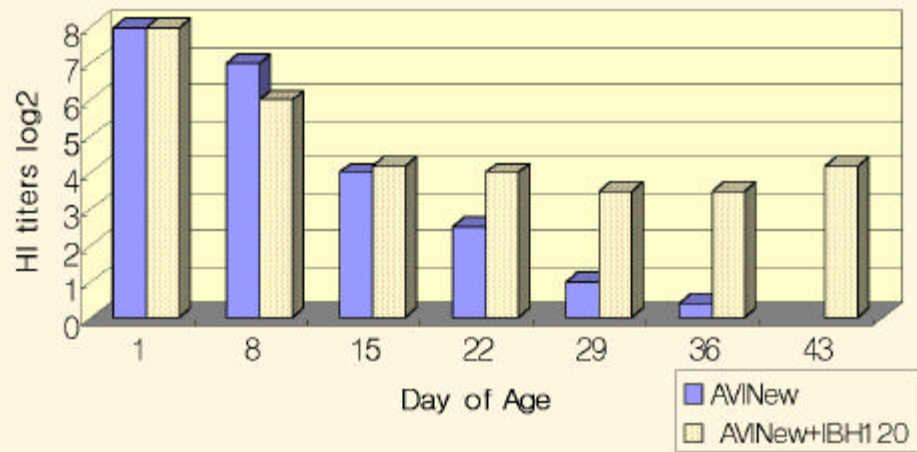


Table2.4. HI titers of non-immunized chickens and immunized intraconjunctivally with AviNew of different titer at day old

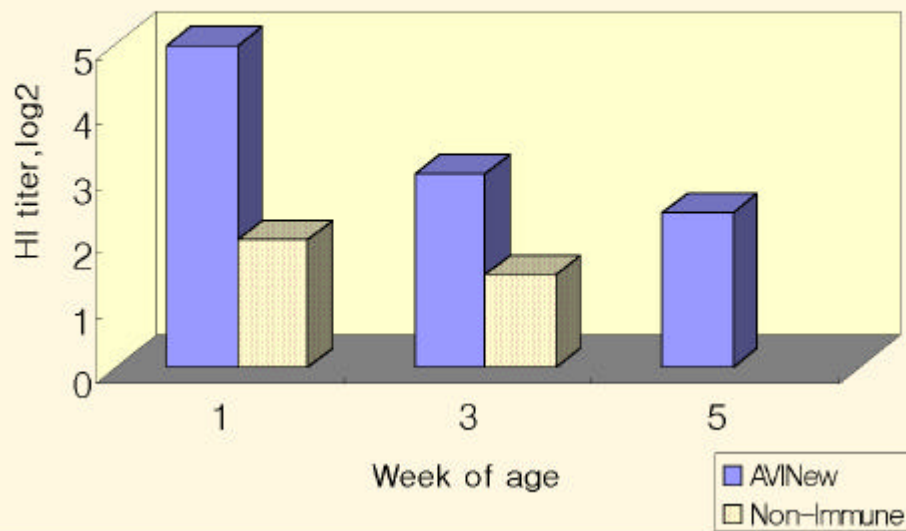


Table2.5. HI titers against IB after immunization with AVINew+IBH120 spray

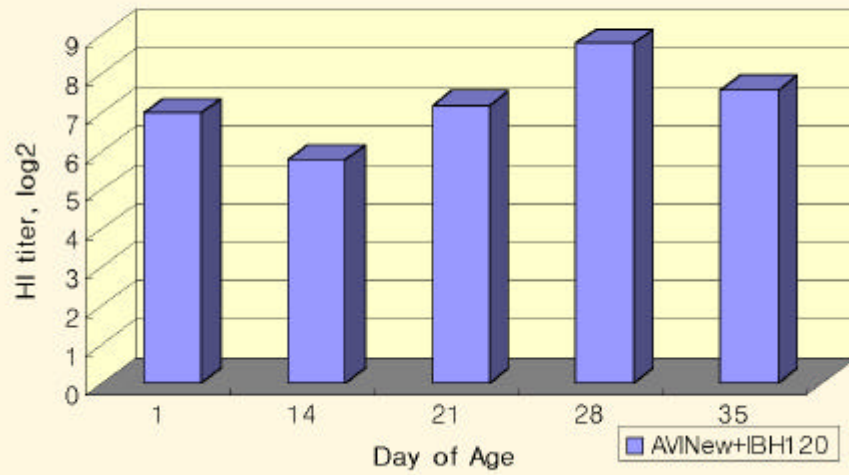


Table2.6. ELISA titers against ND after immunization with AVINew or with AVINew + IBH 120 spray with > 100um particle size

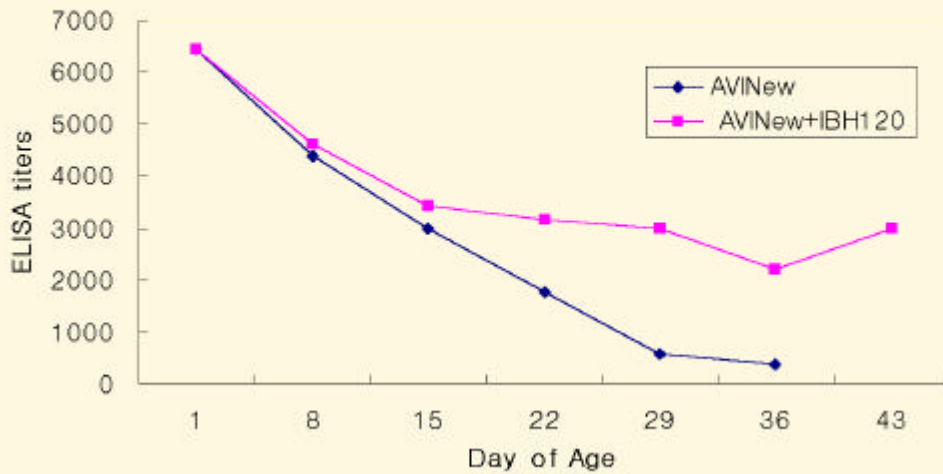
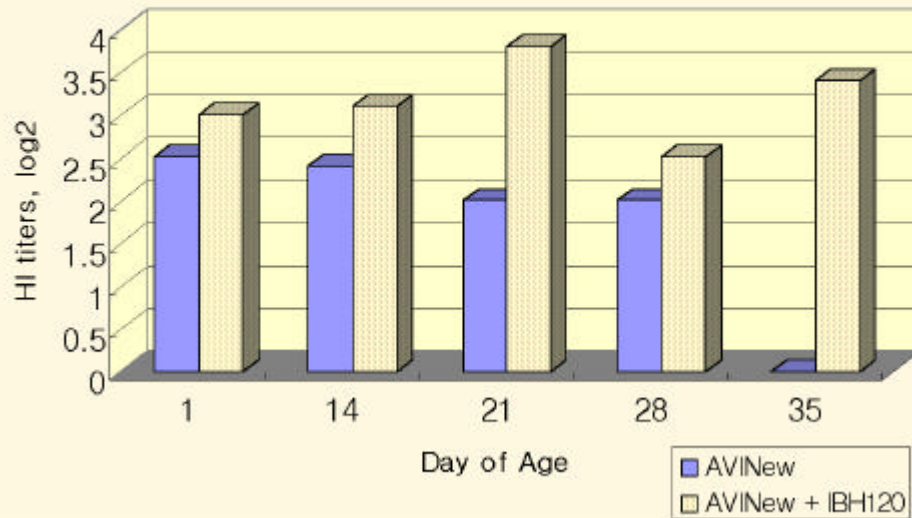


Table 2.7. HI titers against ND after immunization with AVINew or with AVINew + IBH 120 by normal botanic sprayer



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(Ross)

(ISA brown) 1

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coarse spray

ND AM New

IBH20 1

2

가

가

H test β

method

가

16

(3.1, 3.2, 3.3, 3.4).

ND (61 log10EID50/ dose) IB
 (31 log10EID50/ dose) 30 가 .
 ND IB ELISA H 가 가 가
 . 30
 가 가
 가 500 (23) 100
 (23) t-test
 1.81 ±0.137Kg 1.79 ±0.164Kg
 (0.05 <P) 가 (3.6).
 19 41 .
 (0.05<P) 가 500
 (6) 100 (6) t-test
 1.08 ±0.127Kg
 1.09 ±0.104Kg (0.05 <P) 가
 (3.7).

Table3.1. ELISA titers against ND after spray and eyedrop vaccination

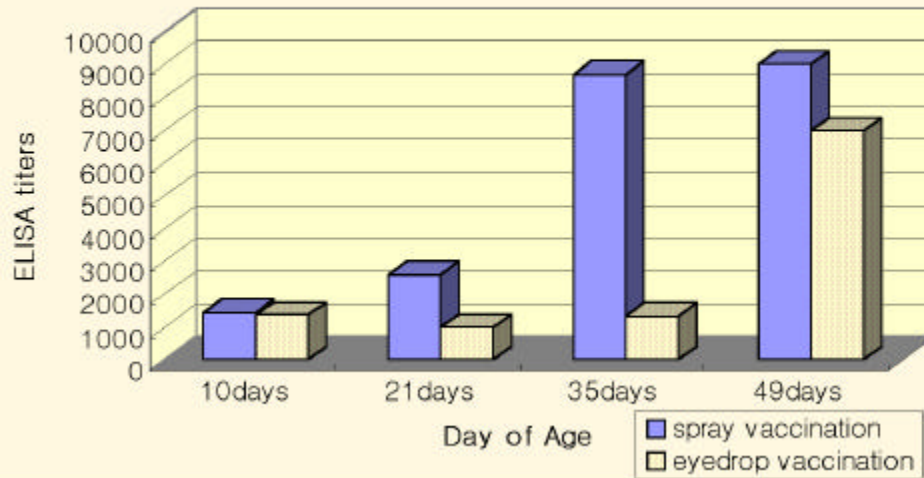


Table3.2 HI titers against ND after spray and eyedrop vaccination

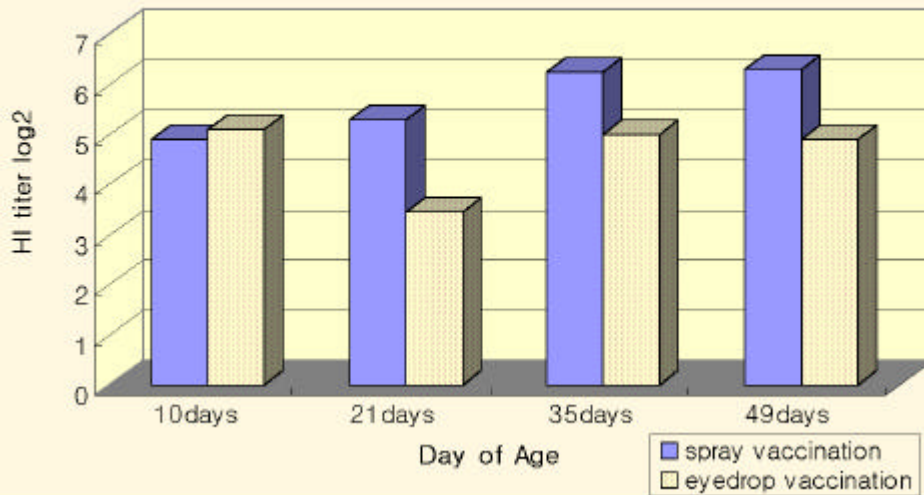


Table3.3. ELISA titers against IB after spray and eyedrop vaccination

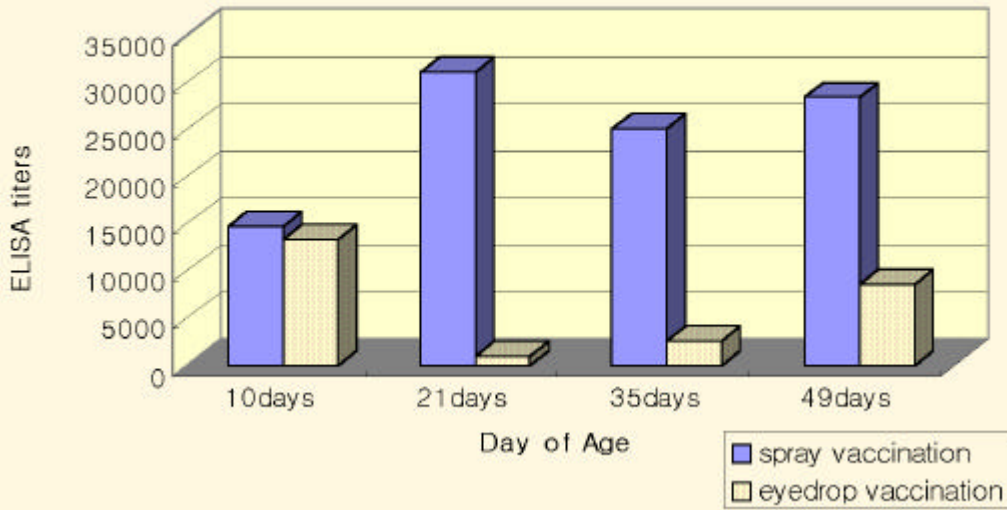


Table3.4. HI titers against IB after spray and eyedrop vaccination

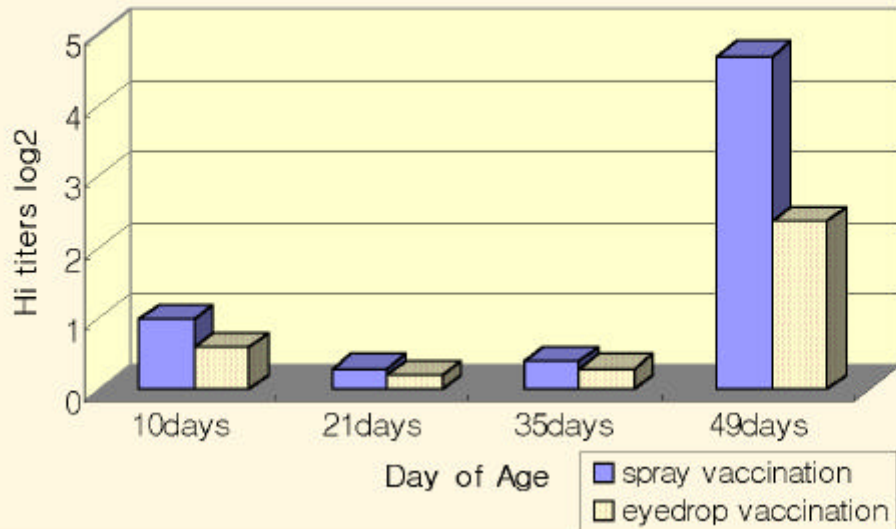


Table3.5. Comparison of egg production in layer after vaccination

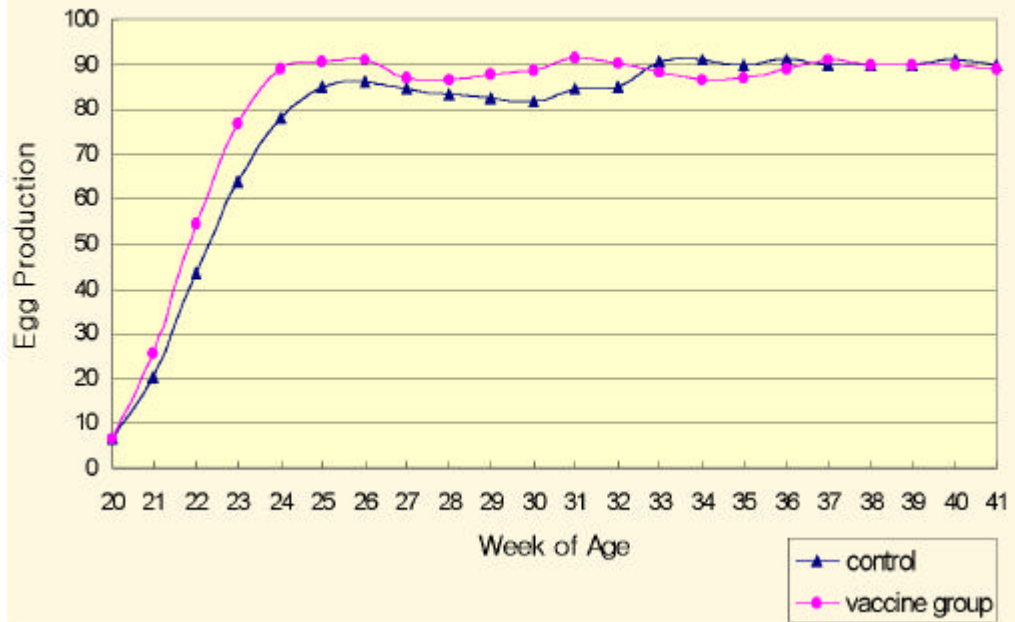
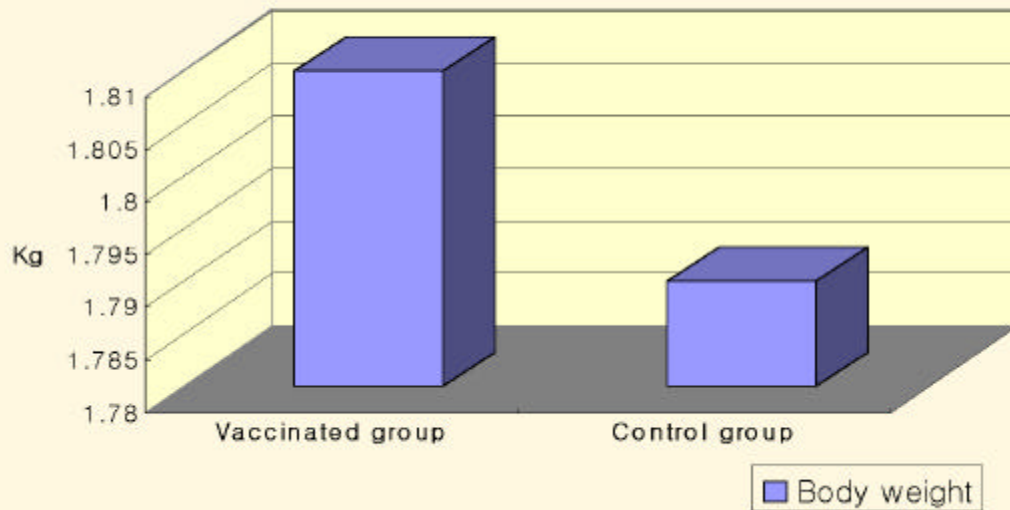


Table3.6. Comparison of body weight in layer



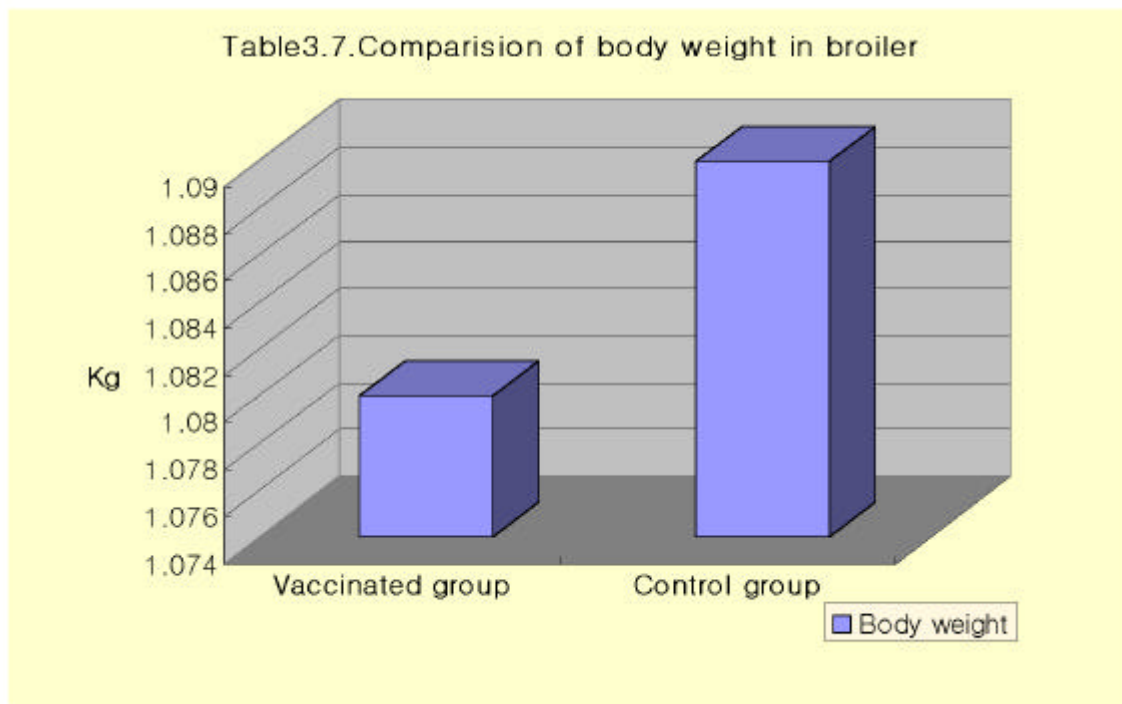


Table 3.8 Efficacy of vaccination against highly pathogenic ND and IB virus

Group	No of dead chicks/total chicks	1st vaccination day-old	2nd vaccination 14days old	challenge 30 days old
Vaccinated	0/50	spray	drinking water	ND: 610g10EID ₅₀ /dose IB: 310g10EID ₅₀ /dose
Control	50/50	-	-	

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