قسم طب الحيوان
كلية الطب البيطرى ـ جامعة القاهرة
رئيس القسم : ا.د / أحمد عبد الحميد بسيونى

# أنتقال فيروسات النيوكاسل والرعاش الوبائى من خلال البيض وطرق مقاومته

أحمد الملا ، عزت الغزاوى ، رؤوف رزق
مصطفى بدر طامى ، فتحى سعد

١) تأكد من الدراسة انتقال فيروسات النيوكاسل والرعاش الوبائى من خلال البيض ولكن يحتاج لأثباته الى اعاده عزل هذه الفيروسات من البيض المعدى .

٢ ) وضح من الدراسة أن فيروس النيوكاسل يتأثر نسبيا بالمطهرات الآتية

١٠ ٪ ايزوبروبيل الكحول

١ ٪ حمض الفينول

وكذلك فيروس الرعاش الوبائى يتأثر بالمطهرات الأتية

١٠ ٪ ايزوبروبيل الكحول

٢ ٪ فورماليين

٣ ) زيادة نسبة تركيز هذه المطهرات لا يزيد بالمثل من كفاءتها ولكن نقصان التركيز يؤدى الى نقص فى التأثير على هذه الفيروسات .

*Dept. of Animal Medicine,*
*Faculty of Vet. Med., Cairo University,*
*Head of Dept. Prof. Dr. A.H. Bassioni.*

# *TRANSMISSION OF NEWCASTLE DISEASE AND AVIAN ENCEPHALOMYELITIS VIRUSES THROUCH EGG SHELL, AND ITS CONTROL.*

*(With 3 Tables)*

*By*
***A. ELMOLLA; E.F. EL GHAZZAWI; T.H. MAHMOUD*; R.E. RIZK***
***M.A. BASTAMI** and F.E. SAAD****

*(Received at 9/1/1985)*

## *SUMMARY*

Transmission of Newcastle disease, and avian encephalomyelitis viruses were evident and confirmed, but it requires virus reisolation. Relatively 10% isopropyl alchol and 1% phenolic acid (for Newcastle disease virus), and 10% isopropyl alcohol and 2% formalin (for avian encephalomyelitis virus) were considered the most effective disinfectants on the Newcastle disease, and avian encephalomyelitis viruses.

Increasing the concentration of these disinfectants not increase their effeciancy, but decreasing this concentration decrease their effeciency.

## *INTRODUCTION*

Newcastle disease virus isolated from the yolk sac of chicken embryos, this emphasizes the posibility of transmission of the disease from breeding hens to their offsprings (DELAY, 1947). HOFSTAD (1949) isolated Newcastle disease virus from dead embryos and infertile eggs collected during decline in egg production following an outbreak of Newcastle disease. AHMED and EL-SISI (1964) showed the possibility of shedding virus vaccine strain in eggs for at least 31 days following vaccination. Formalin (BRANDLY et al. 1946), beta propiolactone and phenol (MACK and CHOTISEN, 1955) had been used to destroy the infectivity of the virus of Newcastle disease. Avian encephalomyelitis virus was evident in some flocks in Egypt (AHMED et al. 1975).

The aim of this work was to study the possibility of Newcastle disease, and avian encephalomyelitis viruses transmission through egg shell, effect of certain virucidal disinfectants and fumigation with formaldhyde on viability of the viruses.

## *MATERIAL and METHODS*

1) Embryonated chicken eggs free from haemagglutination inhibition antibodies against Newcastle disease virus and neutralizing antibodies against avian encephalomyelitis virus.
2) Virus strains: A velogenic viscerotropic strain of Newcastle disease virus obtained from the Animal Health Research Institue, Cairo (SHEBLE and REDA, 1976).

---

* Animal Production Research Institute.
** Faculty of Vet. Med., Cairo University.

3) An egg adapted standard avian encephalomyelitis virus strain was obatined from Institute für Geflugel, Hannover, West Germany.
4) Disinfectants: formalin, phenolic acid, acriflavin, isopropyl - alcohol, iodine plus potassium iodide, and chloramin T, were used in this study.

## *EXPERIMENTS*

**1) Experiment (1):**

Was designed to study the possibility of Newcastle disease, and avian encephalomyelitis viruses transsmission through shell. Seventy two eggs were dipped in different dilutions of Newcastle disease virus ($EID_{50}$ $10^3$, $10^6$, and $10^7$) for 4 minutes at 4°C, the eggs were subjected to isolation of the virus at 24, 48, 72, 120 and 144 hours, after infection. Thirty eggs were dipped in suspension of $10^3$ $EID_{50}$ of avian encephalomyelitis virus for 4 minutes at 4°C. The eggs were subjected to virus isolation by reinoculation in embryonated chicken eggs.

**2) Experiment (2):**

Was designed to study the effect of different disinfectants on viability of Newcastle diseas, and avian encephalomyelitis viruses inside the eggs, and disinfectants toxicity to the embryos. The viruses were serially diluted in isopropyl alcohol, phenolic acid, iodine and potassium iodide, chloramin T, formalin, and acriflavin seperatly and titrated. The viruses were titrated in sterile normal saline as well as control. The minimum $EID_{50}$ for the tested virus was the best effective disinfectant.

**3) Experiment (3):**

Was designed to check the suitable concentration of effective disinfectants on Newcastle, and avian encephalomyelitis viruses. Ten fold filutions of the viruses were made in different concentrations of each disinfectant and inoculated into five, 9 days old embryonated chicken egg into the allantoic sac and then the $EID_{50}$ was calculated according to the method of READ and MUENCH (1938).

The minimum EID was the suitable concentration of each disinfectant.

## *RESULTS*

**Table (1): Results of Newcastle disease, and avian encephalomyelits viruses transmission through egg shell**

| Virus conc. | No. of dipped eggs | No. of dead embryos had Path. changes | No. of+ve HA eggs | No. of+ve eggs after reisolat. | percent. of virus trans. |
|---|---|---|---|---|---|
| $10^3$ ND virus | 18 | - | - | 3 | 16.66 |
| $10^6$ ND virus | 18 | - | 3 | 9 | 66.6 |
| $10^7$ ND virus | 18 | - | 2 | 10 | 66.6 |
| $10^3$ AE virus | 30 | 3 | - | - | 10 |
| Control (non infected) | 30 | - | - | - | - |

Transmission was evident, but its detection requires reisolation

**Table (2): Effect of disinfectants on ND and AE viruses**

| Disinfectant | Titer of treated N.D virus with disinfectent | Titer of A.E. virus with disinfectent |
|---|---|---|
| Phenolic acid | $10^{2}$ | $10^{2.5}$ |
| Isopropyl alcohol | $10^{3}$ | $10^{1.5}$ |
| Acriflavine | $10^{6.3}$ | $10^{3.5}$ |
| Iodine and Pot. Iodide | $10^{6.4}$ | $10^{4}$ |
| chloramine T. | $10^{6.5}$ | $10^{4}$ |
| formalin | $10^{7.5}$ | $10^{2}$ |
| control | $10^{8}$ | $10^{4}$ |

None of the above chemicels had complete effect on the viruses.

**Table (3): Suitable concentration of effective diainfectents on N.D and A.E viruses**

| Virus treated | Effective disinfectents | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | Isopropyl alcohol | | | Phenolic acid | | | Formalin | | |
| | 5% | 10% | 15% | 0.5% | 1% | 2% | 0.5% | 1% | 2% |
| 1) Titer of N.D. Virus after treat. with | $10^{5.5}$ | $10^{3}$ | $10^{3}$ | $10^{4.5}$ | $10^{2.5}$ | $10^{2.5}$ | - | - | - |
| 2) Titer of A.E Virus after treat. with | $10^{2}$ | $10^{1.5}$ | $10^{1.5}$ | $10^{3.5}$ | $10^{2.5}$ | $10^{2.5}$ | $10^{3}$ | $10^{2}$ | $10^{2}$ |

10% isopropyl alcohol and 1% phenolic acid were the most effective concentrations against ND and A.E. viruses.

## *DISCUSSION*

Newcastle disease virus transmission, through egg shell was evident, not by embryonic deaths but by virus reisolation and + ve hemagglutination activity. WALKER and POWELL (1950) showd that no evidence that it transmilttes to the eggs, but VAN ROCKEL (1946), ASPLIN (1949), and ZARGER and POMEROY (1950) reisolate Newcastle disease virus from eggs. Eggs were dipped in either 10% isopropyl alcohol or 1% phenolic acid, although incubated with infected eggs were protected from the virus, the disinfectants used inactivated the virus withen a certain limit, however no complete inactivation occurred. BREAMER et al. (1949), and SCHMITTE and MONSFIELD (1950), reported that fumigation with formaldhyde inactivated the virus on egg shell. MANHELL and HERLYN (1976) showed that chloramin T and Iodine were the most effective disinfectants on the virus, but REDA and AHMED (1967) showed that Newcastle disease virus was not inactivated by hydroxylamine.

Avian encephalomyelitis virus transmission through egg shell was evident and easily could be detected by embryonic deaths and presence of specifie lesions, although no eggs from the control group seemed to be contaminated when incubated with infected eggs. JONES (1934), and BOTROFF et al. (1936) showed that no evidence that avian encephalomyelitis virus is transmitted to the eggs, but TAYLOR et al. (1955), and SCHOAF and LAMOREUX (1955) showed that the infection was egg borne. Dipping eggs in disinfectants protect clean eggs from contaminated ones, although set in the same incubator. The disinfectant used inactivated avian encephalomyelitis virus with a certain limit, however no complete inactivation occured, this agreed with CALNEK et al. (1960), MIYAMAE (1974), and ITAKURA and GOTO (1975).

## *REFERENCES*

Ahmed, A.A.S. and M.A. El-Sisi (1964): Isolation of the Haifa - Komarov Newcastle disease vaccinal strain virus from eggs of vaccinated hens, Vet. Med. J., 10: 219.

Ahmed, A.A.S.; F.E. Saad, K.H. Abbassi, and M.A. El-Sisi (1975): A preliminary note on infectionus Avian Encephalomyelitis in Egypt. Egypt. J. Vet. Sci., 12: 111.

Asplin, F.D. (1949): Observations on the viability of Newcastle disease virus. Vet. Res., 61: 159.

Beamer, P.D., A.K. Sutherland and S.C. Schmittle (1949): studies on Newcastle disease. II- The resistance of Newcastle disease virus to formalodehyde fumigation. Am. J. Vet. Res., 10: 384.

Bottroff, C.A., A.E. Tepper, C.L. Martin., T.B. charles, F.D. Reed, T.C. Phillips and S.R. Shrimer (1936): Epidemic tremors (Trembling chick disease) Proc. Sixth World's Poultry Cong., ii, pp. 99-103.

Brandly, C.A.; H.E. Moses; E.E. Jones and El. Jungherr (1946): Epizootology of Newcastle disease of poultry, Am. J. Vet. Res., 7: 243.

Calnek, B.W.; J. Patrichia; J. Taylor and M. Sevoian (1960): Studies on Avian Encepholomyelitis. Epizootology. Avian Dis, 4: 325.

Delay, P.D. (1947): Isolation of Avian pneumoencephalitis (New-castle disease) virus from the yolk sac of four day- old chicks, embryos, and infertile eggs. Science, 5: 545.

Deshmukh, D.R.; C.T. Larsen, T.A. Rude and B.S. Pomeroy (1973): Evaluation of live virus vaccine against Avian encephalomyelitis in Turkey breeder hens. Am. J. Vet. Res., 34: 863.

Hofstad, M.S. (1949): A study on egizootology of Newcastle disease (prneumoencephalitis). Poultry Sci., 28: 530.

Itakura, C. and M. Goto (1975): Avian Encepholomyelitis in embryos and abnormal chicks on the day of hatching. Neurohisto-pathological observation. Jap. J. Vet. Sci., 37: 21.

Jonses, E.E. (1934): Epidemic tremor, an Encephalomyelitis effecting young chickens. Jour. Exper. Med. 59: 781.

Mack, W.N. and A. Chotisen (1955): Beta - propiolatone as a virus altering agent for a Newcastle disease vaccine Poultry Sci., 34: 1010.

Mahnel, H. and M. Herlyn (1976): Stability of Teschen, Hcc, ND and vaccinia viruses against five disinfectants. Zentrablatt fur Veterinar. Medizin, 23 B: 403.

Miyamae, T. (1974): Ecological survey by the immunoflourescent method of virus in enzootics of Avian Encephalomyelitis. Avian Dis., 18: 369.

Reda, I.M. and A.A.S. Ahmed (1967): The response of strains of Newcastle disease virus of differing pathogenicity to treatment with ether and hydrozylamine. Zentbl. Vet. Med., 14 B: 657.

Schoaf, K. and W.F. Lamoreux (1955): Control of Avian encephalomylitis by vaccination. Am. J. Vet. Res., 16: 627.

Schmittle, S.C. and M.E. Mansfield (1950): Studies on Newcastle disease VI. A field test on hatchery fumigation with formalde-hyde against Newcastle disease virus. Cornell Vet., 40: 90.

Taylor, L.W.; D.C. Lowry and L.G. Raggi (1955): Effect of an outbreak of avian Encephalomyelitis (epidemic Tremor) in a breeding flock. Poultry Sci., 34: 1036.

Van Roekel, H. (1946): Proc. Conf. Newcastle disease, U.S. Dept. Agr. P. 17.

Walker, R.V.L. and E.P.B. Powell (1950): Newcastle disease, a study of the carrier problem. Canad. Jour. Comp. Med., 14: 61.

Zarger, S.L. and B.S. Pomeroy (1950): The effects of commercial living Newcastle disease virus vaccines. Am. J. Vet. Res. 11: 272.