

In Ovo Sexing of Chicken Eggs by Virus Spectroscopy

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Abstract

Recently, some new methods for sexing of chicken eggs by fluorescence and Raman spectroscopy through the shell membrane have been proposed. On the other hand, in another investigation, a new virus medical imaging has been suggested. In this research, summing over these considerations, a new technique for sexing of chicken eggs by virus spectroscopy through the shell membrane is proposed. It is shown that viruses outside the shell of egg can communicate with materials inside it and determine the gender of chick embryo and it's evolutions.

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Introduction

Determination of the sex of birds is very important subject in science. Because, usually, birds with the gender of male have no commercial value and are culled immediately after hatching. Normal experimental techniques for in ovo sexing require taking samples and are applicable after embryos' sexual differentiation. Until now, scientists have proposed some models for biometric evolutions of chick embryo in ovo by using MRI. For example, a model was proposed to characterize embryonic

development during incubation in ovo and also to analyze the putative influence of repetitive ultrahigh-field MRI (UHF-MRI) measurements on this development [1]. In another research, scientists have used of MR microscopy. They have shown that MRM allows in vivo assessment of embryonic development of the chicken in ovo without affecting normal development. The method provided anatomical information supplemented by quantitative evaluation of lens development using DWI. With increasing availability of ultrahigh-field MR systems, this technique provided a noninvasive complementary tool in the field of experimental ophthalmology [2]. On the other hand, some other authors have demonstrated

that Raman spectroscopy enables contactless in ovo sex determination of the domestic chicken already at day 3.5 of egg incubation. A sexing accuracy of 90/100 have been obtained by analyzing the spectra of blood circulating in the extraembryonic vessels [3]. In another attempt, fluorescence spectroscopy has been applied to determine nondestructively in ovo the sex of early embryos of the domestic chicken. In this method, Sex-related differences in the fluorescence spectrum have been found at day 3.5, and principal component (PC) analysis showed that the blood of males was characterized by a specific fluorescence band located at ~ 910 nm [4].

In ovo Raman and fluorescence spectroscopy of blood of eggs incubated until day 3.5 enables correct sexing rates over 90/100 barely affecting the hatching rate. Full automatization of the processes to guarantee high sexing speed and fulfill industrial demands is needed to permit transferring the technology inside the hatcheries in the next future [5].

In parallel, there are some other techniques that used of differences between electromagnetic signals for male and females for determining gender. In one of these researches, authors have argued that the type of packing of DNA in chromosomes of men and women are different. This causes radiated waves from DNAs of men and women to have opposite signs and cancel the effect of each other in a pair. Using this property, authors have suggested another mechanism to cancel the effect of extra waves, which are produced by DNAs in cancer cells of a male or a female, by extra waves which are produced by DNAs in similar cells of a female or a male and prevent the progression of the disease [6].

In another investigation, authors have proposed a new virus medical imaging technique. In this technique, viruses can communicate with cells, interior of human's body via two ways: 1) Viruses can form a wire that pass the skin and achieve to a special cell; and 2) Viruses can communicate with viruses interior of body in the wireless form and send some signals for controlling evolutions of cells interior of human's body [7].

Motivated by these researches, we suggest a new a new method for determining the gender of chicken eggs by virus spectroscopy through the shell membrane. In our method, virus outside the shell acts like the receiver of signals of cells inside the shell and give us this opportunity to analyze evolutions of chick embryo.

The outline of the paper is as follows. In section II, we consider materials and methods in this consideration. In section III, we will consider differences between radiated signals from viruses around chick embryos. The last section is devoted to conclusions.

Material and Methods

Chicken Eggs

All of the fertilized eggs used in this consideration were Dekalb Brown eggs, which were obtained from a village.

Culture Vessels

We have two types of culture vessel. In one type, we use of in ovo system (Shell culture system). In another type, we use of ex-ovo system (Shell-less culture system).

Our culture vessels for ex-ovo method are the same used in Tahara [8] however type of incubating, temperature and rotation were different. Similar to Tahara [8], a 450 ml polystyrene plastic cup was applied as the pod for the culture vessel. A 1-1.5 cm diameter hole was made in the side of the cup approximately 2 cm from the bottom, and the hole was plugged with a cotton pledget as a filter. A 2 mm diameter plastic tube was inserted through the space between the pledget and the hole to provide an oxygen supply. An aqueous solution (40 ml) of benzalkonium chloride was then added to the cup. A polymethylpentene film was formed into a concave shape, carefully avoiding wrinkles and installed as an artificial culture vessel in the pod. A polystyrene plastic cover was placed on top of the culture vessel.

Embryo culture, incubating, temperature and rotation

For in ovo method (Shell culture method), fertilized chicken eggs were incubated at 38°C and rotated with 120° clockwise twice a day. After 48 h, in most of eggs, chick embryos begin to grow.

For ex-ovo mechanism (Shell-less culture method), fertilized chicken eggs were not incubated before transferring to the culture vessels. Their eggshell was wiped and cracked and the whole egg contents were transferred to the culture vessel without pre-incubating period. The culture vessels were maintained at 38°C and rotated with 120° clockwise twice a day [8]. After 54 h, in most of vessels, chick embryo is emerged (See figure 1).



Figure 1: Formation of chick embryo in shell-less culture vessel (In-Ovo) less than 48 h after incubating at 38°C

Two systems for Taking signals of DNAs in a chick embryo by viruses

Previously, it has been shown that viruses can communicate with DNAs. They can act like the receiver or sender in electronic devices (See Figure 2) [7].

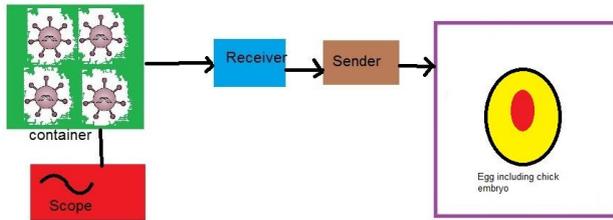


Figure 2: Viruses act like the receiver or sender of electromagnetic waves

Also, type of waves which exchange between viruses and DNAs of male is different respect to type of waves which exchange between viruses and DNAs of females (see Figure 3) [7]. We use of these properties for determining the gender of chick embryos in ovo and ex ovo.

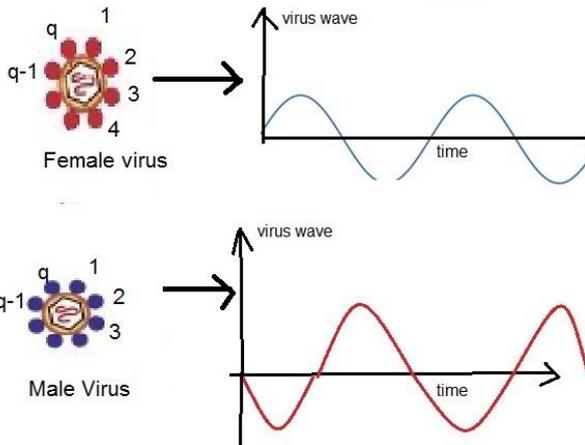


Figure 3: Viruses exchange two different waves with males and females

We form two systems for considering signals of chick embryos.

In first system, we take water around chick embryos in shell less culture systems (ex-ovo) and pour them in a tube of viruses. We use of viruses of influenza. We connect this tube to a computer or laptop with Radio-SkyPipe. This software helps us to consider evolutions of waves interior of tube. We also use of an Amperemeter for measuring currents (See Figure 4).

In second system, we put a tube of viruses and water around chick embryos in egg culture systems (in-ovo). We use of viruses of influenza. We connect this tube to a computer or laptop with Radio-SkyPipe. This software helps us to consider

evolutions of waves interior of tube.

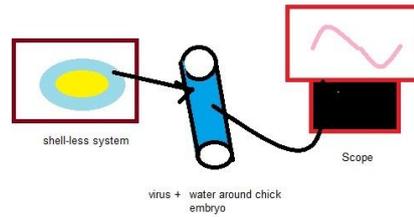


Figure 4: Viruses take signal of DNAs interior of shell-less culture system and send them to Radio-SkyPipe

We also use of an Amperemeter for measuring currents (See Figure 5). If signals couldn't be observed clearly, we can make a coil in a tube of viruses around egg and measure differences between output and input currents.

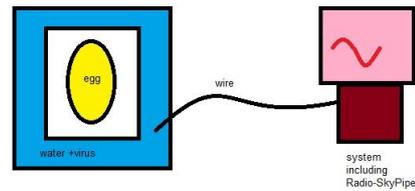


Figure 5: Viruses take signal of DNAs interior of egg system and send them to Radio-SkyPipe

Results

Considering differences between radiated signals of viruses around chick embryos

In figure 6, we present probability for producing each current which is taken from water + virus around a shell-less culture system by some scopes. We used of Radio-Skypipe software and converter for converting current to electrical signal. This probability is emerged by counting number of times which each current is emerged and dividing it to total current.

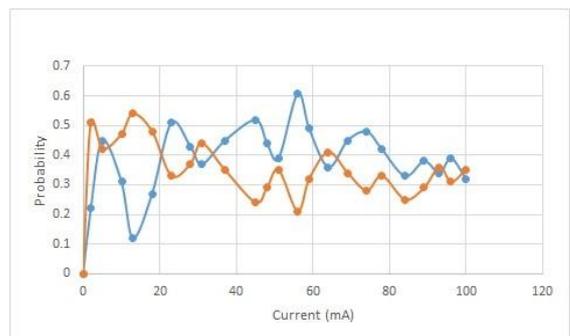


Figure 6: Comparing signals of viruses in a vessel of water which exchange waves with a chick embryo with the gender of female (red color) and male (blue color) interior of shell less culture system (For observing signals, we used of Radio-SkyPipe)

To produce viral system, we mix water with some small amount of milk and involve them with Influenza viruses. In this figure, blue line corresponds to male and red line is related to female. It is observed that male cells send middle currents, while, female cells emit lower currents.

In Figure 7, we show the probability for producing each current which is taken from water + virus around an egg. Egg is located in a container which makes it separated from water. We have added some amount of milk to water for growing viruses and bacteria and involve them with Influenza viruses. It is clear that signals of males (blue color) is different from signals of female (red color). Usually, cells of females produce lower and higher currents, while cells of males produce middle currents. This figure is different from Figure 6 which is due to the existence of shell and its effects on signals.

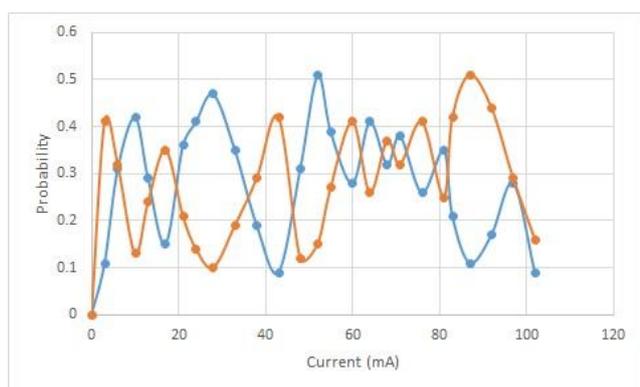


Figure 7: Comparing signals of viruses in water around an egg which exchange waves with a chick embryo with the gender of female (red color) and male (blue color) interior egg (For observing signals, we used of Radio-SkyPipe)

At final stage, using some devices like incubator and cooler, we change temperature and take signals of water and viruses around chick embryos again. In figure 8, we compare radiated signals of chick embryos with the gender of male with chick embryos with the gender of female. It is observed that in both type of embryos, radiated signals grow with temperature.

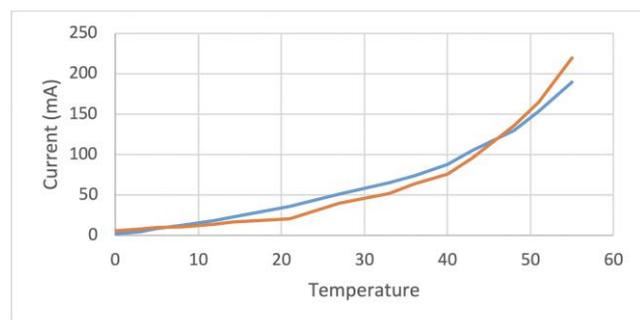


Figure 8: Comparing signals of chick embryo with gender of female (red color) and male with gender of male (blue color) in terms of temperature

This is because that at higher temperature, more viruses and bacteria are born and more signals are emerged. Also, in lower temperature, cells of embryos may be died. Also, for lower and higher temperatures, chick embryos with the gender of female interact more with viruses' respect to males, while for middle temperatures, radiated signals of cells of males are more.

Discussion

Newly, some authors have suggested new techniques for sexing of chicken eggs by fluorescence and Raman spectroscopy through the shell membrane. On the other hand, other investigators have worked on the communications between viruses outside the shell and DNAs inside the shell and proposed a new virus medical imaging. In this investigation, by mixing two techniques, we have proposed a new virus spectroscopy technique for determining the gender of chick embryo inside the shell. We have shown that radiated electromagnetic waves by viruses outside the shell can help us to determine the gender of chick embryo inside the shell and consider its growth rate.

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