Effects of Non-ionizing Radio Waves on the Development of a Duck Embryo

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Abstract - There exists increasing concerns on the effects of mobile phones on users brought by the increase of smartphone usage. However, research regarding the effects of radiation on human tissues remains limited. Studies on the effects of non-ionizing radio waves on tissues are plenty. However, the results often show discrepancies. In this paper, the researchers investigated the relationship between the growth rate of duck embryos and the duration of exposure to radiation emitted by a smartphone. Qualitative observations were recorded on the developing tissue and body parts of the embryo. The quantitative results served as evaluation of the growth of embryonic development by the three fetal growth parameters: biparietal diameter, occipitofrontal diameter, and abdominal circumference. The parameters were evaluated through computer analyses of images taken in a fixed position. Growth rate was calculated using data values from day 14 and 18. The results were clear: there was an inversely proportional relationship between the growth rate of the embryos and the period of irradiation. The most prominent data which showed the difference in growth rate was the abdominal circumference where the controlled embryos had a growth rate of 198.50% (3.99 cm to 11.91 cm). Studies concerned with the sustainability impacts and health hazards of the emission of radio waves imply there was an increased risk of tumors called malignant schwannomas of the heart in rats exposed to non-ionizing radiation. Additionally, smartphone production has depleted a massive 968 terawatt hours of electricity worldwide.

Keywords - Duck Embryos, Non-ionizing radio waves, Radiation, SERVQUAL, Smartphone

INTRODUCTION

An online German statistics portal, Statista estimates that the average amount of time a person spent with mobile non-voice media in the United States during the year 2018 is 203 minutes [1]. A clear trend in the increased usage of mobile devices can be determined when it is compared to 88 minutes in 2012. Much of this time is spent calling on the phone. While being used, mobile devices emit radio waves, which can be tracked to their antennas [2]. The electromagnetic waves emitted are classified as a form of non-ionizing radio frequency radiation [3]. These sources may have thermal effects, in which the RF (radio frequency) radiation heats tissues in the body [4]. The human body is capable of holding its temperature. However, once exposure exceeds the threshold of four watts per kilogram, it may cause permanent damage [5]. The mobile phones used commonly today emit low levels of RF radiation and cause non-thermal effects to the user [6]. Despite the technological advances in limiting the specific absorption rate of the device, there arises a public concern that radio frequency energy produced by cellular devices may impact the tissue in the brain and nervous system as the device is often placed near the head [7]. Scientific studies compiled by The Institute for Community Development and Quality of Life discuss the negative effects of non-ionizing radiation on the human body by methodizing the dependent component as the time of exposure, thus elucidating the adverse effects of excessive exposure to cellular devices [8]. This study is used as a literature reference by setting the independent variable as the time of exposure. Due to the close biological resemblance between human and duck embryos, this paper will test the non-thermal effects of cell phone radiation on the duck embryonic development [9].

Laboratory studies warn of the effects of lengthy exposure periods to radiofrequency radiation to the human body. Although not as severe as ionizing radiation, low levels of RF radiation can disrupt the circadian rhythm of the body and its immune system [10]. A study conducted in 2018 by The National Toxicology Program reported the high association of cancer in male rats with exposure to RF radiation from electronic devices such as cell phones [11]. Clear evidence of tumors in the adrenal gland and brain of the exposed rats was displayed in the experiment while exhibiting the prominent physical deviation from the control laboratory rats. However, the considerable limitation to the objective of the experiment, which hypothesizes the association of the physical impact of non-ionizing radiation on rat tissue and that of human tissue, is the difference of the area exposed to the radiation [12]. John Bucher, Ph.D., NTP senior scientist commented on the study highlighting that the whole body of the tested rats was exposed to radiation, whereas for humans, concentrated exposure is only received in specific local tissues near where the phone is held frequently such as the head or ear. Exposure to electromagnetic or RF fields can also lead to radiation-induced genetic modification in blood lymphocytes [13]. Despite the findings, the definite biological effects are unclear, as further experimentation has failed to recreate these effects [14]. Therefore, this paper clarifies the direct biological effect by utilizing growth parameters including biparietal diameter, occipitofrontal diameter and the abdominal circumference of the wet duck embryo as standardized points of evaluating the rate of development. To investigate the effect of radio waves on duck embryos, the present study compares the development of the embryos receiving varying dosage amounts of non-ionizing radiation. Growth rate values are then computed for each group, to qualitatively characterize the progressive stages of growth and quantitatively evaluate the extent of embryonic development.

OBJECTIVES OF THE STUDY

The objective of the study was to examine the effect of radiation on the development of a duck embryo. In order to do so, several growth parameters were predetermined to track embryonic development: Biparietal diameter, occipitofrontal diameter, and the abdominal circumference. These measurements have been obtained in centimeters.

MATERIALS AND METHODS Growth Parameters

Three growth parameters were selected in order to determine and compare the growth rate of the duck embryo. They are the following: Biparietal diameter, Occipitofrontal diameter and Abdominal circumference. Biparietal diameter is defined as the measurement of the greatest transverse diameter across the embryo's skull, from one parietal bone to the other. The Occipitofrontal diameter is the measurement of the greatest length from a point just above the root of the nose to the most prominent portion of the occipital bone. The two diameters of the skull are of many measurements that are obtained during pregnancy ultrasound procedures used to evaluate fetal growth and estimate fetal weight and gestational age; thus, we decided that they could be effective determinants of the embryonic growth rate as well. However, with the thought that there may not be a significant difference in size of the embryo's skull, we added the abdominal circumference as a growth parameter. As the yolk sac begins to be internalized into the body cavity of the embryo, it provides the hatchling with immediate nourishment until exogenous feed is given in the brooder house, constituting 20% of the birth weight of the chick. During the first week of incubation, most of the weight is in the head of the embryo, but as the nutrition is distributed towards the organs, the body of the chick becomes heavier. This led to our assumption that the body, or more specifically the abdomen of the chick would display an increase in size, which would serve to be useful in tracking the development of the embryo. Hence, we concluded our last growth parameter to be the abdominal circumference, the greatest perimeter around the abdomen of the chick.

Preparation for Incubation

Each of the incubators were first adjusted to a temperature of 37.5°C, and a 100 ml of warm water (29°C) was measured and transferred to the humidity controller opening of the incubator. The incubators were then placed in locations that had restricted sunlight flow, in order to achieve the optimal conditions for incubation. Afterwards, the system was left to stabilize in its environment for two days before beginning the incubation process. The humidity of each incubator was maintained at 50%, through daily check-ups on the temperature of the warm water. Additionally, warm water was resupplied when a dearth of it was observed.

Egg Selection Process

Prior to experimentation, each of the eggs were inspected using the method of candling, to ensure that none of the eggs were double yolked, cracked, misshapen, undersized or oversized. Following the filtration of the eggs with abnormalities, the weight of the remaining eggs were measured. After weighing each of the eggs, eight of them that were most similar in terms of weight were selected, to be used in the experiment process. Two eggs pertained to each group (controlled group, experimental group 1, experimental group 2, and experimental group 3). **Incubation Process**

To compare the effect of exposure to nonionizing radio waves on duck embryos, we placed a wooden box over each incubator. A mobile phone was then inserted on the inner surface of the box. This setup was decided upon based on its close resemblance to a phone call in real life. The mobile phone was called each day for the desired amount (10 minutes for experimental group 1, 20 minutes for experimental group 2, 30 minutes for experimental group 3).On days 14 and 18 of incubation, an egg from each group was sacrificed, and observations (both quantitative and qualitative) were recorded accordingly. During the preliminary days of the experiment, candling was performed to ensure growth was present in the embryos. This process was repeated every other day, until day 8 of the experiment, where development was clearly visible in the control group.

Qualitative Observations

On days 14 and 18 of the experiment, an egg was sacrificed from each group. Pictures of the embryo were taken on these days using a phone to allow for clear visibility of its inner characteristics. The following qualitative aspects of the embryo were observed: amniotic fluid, yolk sac, pigmentation, development of tissue, beak, egg tooth, comb, feathers, surface texture, hemorrhagic areas, albumen, and abdominal cavity. Along with qualitative observations, the pictures were integral in obtaining quantitative measurements (explained in the next section).

Virtual Image Analysis

We concluded that the measurements obtained from measuring the physical dimensions of the embryo would be too inaccurate because embryos were too fragile, and that any form of physical contact could distort the measurements. Thus, to acquire numerical data of the embryo regarding the

previously mentioned parameters, we used Logger Pro, a program with a photo analysis feature which scans and provides the measurements of certain parts of an image. The diameter of the bowl was measured to be 16.00 cm, and this measurement was employed as a scale of reference. Relevant measurements of the biparietal diameter. occipitofrontal diameter, abdominal and circumference with the precision of two decimal places were obtained using the program. In order to maximize the accuracy of the photo analysis, the camera was placed on a stand that would be fixed for every trial. This was done to minimize any possibility of having different angled embryonic photos.

RESULTS AND DISCUSSION

We utilised three growth parameters to track the growth of the duck embryos: Biparietal diameter (BPD), Occipitofrontal diameter (OFD), and Abdominal circumference (AC). Since we decided that the eggs would be sacrificed for two nonconsecutive days due to the limited number of eggs and incubation slots, we recognized that we would only have two sets of data: one from the 14th day and the other from the 18th. Ultimately we decided to use the difference between the measurements of each growth parameter in order to determine the growth rate. Growth rate was calculated using the formula:

$$\% \, \textit{Growth Rate} \, = \frac{\textit{Final-Initial}}{\textit{Initial}} * \textit{100\%}$$

where the data value for day 18 was considered as the final measurement, and the data value for day 14 was considered as the initial measurement.

As shown in Table 2, the control group showed high values of growth rates (84.43% for the BPD, 136.62% for the OFD, and 198.50% for the AC). Following those growth rates are group 10 embryos, which showed lower values of growth rates (55.65% for BPD, 134.85% for OFD, and 138.17% for AC). Finally, the group 20 embryos showed the lowest growth rate values (52.75% for BPD, 32.46% for OFD, and 63.11% for AC). The embryos in group 30 displayed minimal growth; as a result, the Biparietal diameter, Occipitofrontal diameter and abdominal circumference was unable to be determined.

Furthermore, the weights of the embryos were taken once they were sacrificed. There is an increase in the weight of the duck embryos for all groups from day 14 to day 18 (table 1). This is indicative of the experiment having minimal random error, as anomalies were not present.

Overall, there is a general increase in the wet weights (weight of the embryo including the amniotic fluid) of each embryo between days 14 and 18; This increase can be explained by the development of hard tissue and organs. The data in table 5 show a decrease growth rates for every parameter along the groups, indicating an inversely proportional relationship between growth rate and the period of irradiation for duck embryos. Furthermore, embryos in group 30 were not developed enough to determine accurate measurements. This suggests that 30 minutes of cell phone irradiation may have exceeded the threshold limit for a duck embryo, hence the 100% mortality rate.

Additionally, the wet weights pertaining to each group presented a decreasing trend, with the control group, group 10, group 20, group 30 having wet weight values of 59.6 g, 56.07 g, 53.01 g, 49.61 g, respectively. The observed decreasing trend of wet weight as the irradiation period increased suggested that there is a negative causal relationship between the irradiation period and the wet weight of an embryo. This may suggest that irradiation causes underdevelopment in an embryo, hence the lower weight values.

When inspecting the qualitative aspects of embryos pertaining to the control group, development was slower in comparison to previous duck incubation experiments. The retardation may have been caused down due to the heat emitted from the mobile phone in the closed system. Furthermore, day 14 of the embryo in group 30 presented no growth qualitatively, as there were no visible features (as shown in figure 4).

Figures 5, 6, 7, 8 delineate the differences in embryonic development of species pertaining to each group. Beaks and bodies are visible for the control group and group 10, while group 20 and group 30 present severely underdeveloped embryos. In particular, there were no visible features to measure in the group 30 embryo.

Table 1. Growth Parameters for All Groups					
Day	Embryo group	BPD (cm)	OFD (cm)	AC (cm)	Wet weight (g)
14th	Control	1.22	1.42	3.99	59.6
	Group 10	1.15	1.32	3.93	56.07
	Group 20	0.91	1.14	3.24	53.01
	Group 30	0	0	0	49.61
18th	Control	2.25	3.36	11.91	66.94
	Group 10	1.79	3.1	9.36	65.89
	Group 20	1.39	1.51	5.28	64.09
	Group 30	0	0	0	61.75

The weight of the duck embryo was measured after sacrificing an egg, without including the shell. Images taken of the embryo during days 14 and 18 of the experiment were analyzed; the Biparietal diameter (BPD), Occipitofrontal diameter (OFD), and Abdominal circumference (AC) for each group were determined from the analysis and recorded. Embryos pertaining to group 30 had a mortality rate of 100%; measurements were unable to be taken due to the lack of development in these areas.

Table 2. Growth Parameters for Control Group

Control	14th	18th	Growth rate
BPD	1.22	2.24	84.43%
OFD	1.42	3.36	136.62%
AC	3.99	11.91	198.50%

The Biparietal diameter (BPD), Occipitofrontal diameter (OFD), and Abdominal circumference (AC) for the control group recorded. The growth rate for each growth parameter was acquired using the data value from day 14 and the data value from day 18.

Table 3. Growth Parameters for Group 10

Group 10	14th	18th	Growth rate
BPD	1.15	1.79	55.65%
OFD	1.32	3.1	134.85%
AC	3.93	9.36	138.17%

The Biparietal diameter (BPD), Occipitofrontal diameter (OFD), and Abdominal circumference (AC) for group 10 recorded. The growth rate for each growth parameter was acquired using the data value from day 14 and the data value from day 18.

Table 4. Growth Parameters for Group 20

Group 20	14th	18th	Growth rate
BPD	0.91	1.39	52.75%
OFD	1.14	1.51	32.46%
AC	3.24	5.28	63.11%

The Biparietal diameter (BPD), Occipitofrontal diameter (OFD), and Abdominal circumference (AC) for group 20 recorded. The growth rate for each growth parameter was acquired using the data value from day 14 and the data value from day 18.

Table 5. Growth Rate Comparison Between All Groups

Parameter (cm)	Control	Group 10	Group 20
BPD	84.43%	55.65%	52.75%
OFD	136.62%	134.85%	32.46%
AC	198.50%	138.17%	63.11%

Comparison of growth rates between the three groups for each parameter - Biparietal diameter (BPD), Occipitofrontal diameter (OFD), and Abdominal circumference (AC)

CONCLUSION AND RECOMMENDATION

There has been an increase in the usage of smartphones in modern society. Due developments in the field of technology, the demand for electronics has increased. The electronics that provide access to wireless communication like the smartphone have become a necessity in our lives. The continuous usage of these electronic devices emit radiation with various frequencies. They normally emit radio waves, which are non-ionizing radio waves in the electromagnetic spectrum. When humans are communicating with one another using a smartphone, the electromagnetic waves can be absorbed by our bodies [15]. The study examines the effect of radio waves on the development of a duck embryo. This study portrays a negative relationship between the duration of exposure to radiation and the growth rate of the duck embryo. The three growth parameters were set to be the Biparietal diameter, Occipitofrontal diameter and Abdominal circumference. The controlled group presented the highest growth rate values, followed by group 10, group 20 and finally group 30.

The paper spreads awareness to the community about the health hazards and damage smartphones may bring to soft tissue (vulnerable to soft skinned animals and infants). By highlighting the relationship between the duration of exposure to electronic devices and the damage or growth retardation, it also discourages excessive usage of electronic devices especially to children of young age as they are susceptible to receive more damage than adults.

Future research in this field of study can take into consideration the data collected in this paper. For example, knowing that 30 minutes of daily exposure to RF radiation is fatal to embryonic duck species, future research can be conducted to investigate smaller time increments of radio wave emissions (between 0 and 30). This process can be utilised in formulating a definite relationship between the development of a duck embryo and irradiation periods. With more data values, a trendline can be obtained, allowing researchers to formulate a numerical relationship between growth rate and length of exposure.

Based on the trends presented in this paper, time increments may be selected between 20 and 30 minutes, to clearly identify the threshold limit for duck embryonic species. However, if one were willing to observe the pure relationship between non-ionizing radio waves and embryonic development, utilising a radio transmitter device instead of a mobile phone would prove to be more realistic. This way, heat transmitted from the mobile phone to the embryo would not be involved during experimentation - a major limitation in the research presented, which was responsible for the lagging development. Furthermore, one must recognize that replicating the procedure of the paper's experiment may not yield the same results, as embryonic development may differ severely based on external factors, such as ambient temperature.

Ultimately, this research attempts to convey a general relationship between duck embryo development and irradiation periods under a relatively controlled system.

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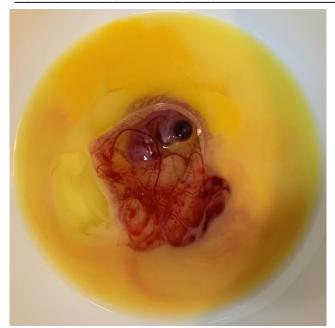


Figure 1. 14 day old embryo from the controlled group



Figure 3. 14 day old embryo from the group 20



Figure 2. Picture of 14 day old embryo from group 10



Figure 4. 14 day old embryo from group 30



Figure 5. 18 day old embryo from the controlled group



Figure 7. 18 day old embryo from the group 20



Figure 6. 18 day old embryo from group 10



Figure 8. 18 day old embryo from group 30