

# Histological Study of Chick Embryo Retina Exposed To Radiofrequency Radiation Emitted From 2G Cell Phone

Mary Hydrina D'Silva<sup>1</sup>, Rijied Thompson Swer<sup>2</sup>, J. Anbalagan<sup>3</sup>

<sup>1</sup>Assistant Professor, Department of Anatomy, Mahatma Gandhi Medical College & Research Institute, Pillaiyarkuppam, Puducherry, India, Pin Code - 607402

<sup>2</sup>Associate Professor, Department of Anatomy, Mahatma Gandhi Medical College & Research Institute, Pillaiyarkuppam, Puducherry, India, Pin Code - 607402

<sup>3</sup>Professor, Department of Anatomy, Mahatma Gandhi Medical College & Research Institute, Pillaiyarkuppam, Puducherry, India, Pin Code - 607402

**Abstract:** *The mobile phone technology has become an integral part in everyone's life. It emits a pulsed radiofrequency electromagnetic field which is absorbed into the user's body particularly the head region. The present study is undertaken to evaluate the possible tissue damage in developing retina of chick embryo following chronic exposure of radiofrequency radiation emitted from 2G cell phone. Fertilized chick embryos were incubated in three groups (Group A-sham control, Group B-experimental and Group C-control) in a standard egg incubator. Group B was exposed to radiation emitted from a 2G cell phone. On completion of scheduled duration, the embryos were collected and gross morphological features were noted. The three group embryos were processed for routine histological studies. The thickness of each layer of the retina was measured using oculometer and then compared statistically. Our study concludes that the exposure of chick embryos to radiation emitted from 2G cell phone increased the thicknesses of internal nuclear layer, internal plexiform layer, ganglion layer, optic nerve fibre layer and total retinal thickness. Exposure also caused structural changes in the form of increased spaces between the cells and the disintegration of optic nerve fibres.*

**Keywords:** Radiofrequency radiation, retinal pigment epithelium, neural retina, melanogenesis.

## 1. Introduction

The cell phones are the most important source of radiofrequency radiation. At present, about 6 billion Global System Mobile communication (GSM /2G) cellular phones functioning in the frequency range of 900- 1800 MHz are in use throughout the world. The number is increasing day by day as the cell phone technology has become an integral part in everyone's life. A cell phone in operation emits a pulsed radiofrequency electromagnetic wave and the human body acting like an antenna absorbs this into the user's body particularly the head region. There are numerous contradictory scientific reports on the health effects of UHF/RFR on biological tissues in both animals and humans. Frequent use of cell phone is associated with depressive symptoms, head ache, dizziness, memory changes, tremors and sleep disturbances [1-3]. Exposure of chick embryo to RFR emitted from a cell phone increased the mortality rate significantly [4-7], caused congenital anomalies [8, 9] and damage to the developing kidneys [10].

Fatima Al-Qudsi and Solafa Azzouz[9] reported an increased body growth and eye development of chick embryo till the 10th day of incubation when exposed to electromagnetic radiation of 900- 1800MHz and further radiation resulted in brain malformations with reduced eye and body growth. RFR emitted from GSM mobile phone caused retarded retinal differentiation of chick embryos of 10 days and enhanced retinal growth and pigmentation of embryos of 15 days [11] and increased retinal thickness in rat exposed to electromagnetic waves for 4 weeks [12].

RFR/UHF emitted from cell phone is non- ionizing radiation. They cause both thermal and non-thermal effects. Non-thermal effects are reported to cause more damage to biological tissues by producing oxidative stress. The production of heat shock proteins (HSP-27&HSP-70) on prolonged exposure resulted severe pathological changes in biological tissues. Studies on human and animal models have shown an increase in HSP-70 and HSP-27 protein expression in lens epithelial cells [13, 14] due to radiation exposure. Oxidative stress is considered as the leading cause for cataract. Exposure of lens of both human & animal models to RFR caused structural damage in lens epithelial cells that affected its transparency leading to cataract formation [14-19]. Hydrina et al [20] reported microstructural changes in the form of multilayered lens epithelium, formation of cystic cells and spaces and distorted arrangement of lens fibres in the chick embryo lens exposed to 2G cell phone radiation.

Dasdag et al [21] observed alterations on apoptosis of glial cells and antioxidant capacity and catalase changes in rat brain due to 900MHz radiation. Dogan M et al [22] reported no significant change in catalase and glutathione peroxidase enzyme activities due to 3G mobile phone exposure.

RF exposure can cause physiological changes in cells and tissues even at the level of DNA. It is reported to produce single and double stranded DNA breaks, marked suppression of DNA synthesis and mitosis of lens epithelial cells, inhibition of DNA synthesis and cell arrest at G0/G1 phase [23, 24]. Exposure of chick embryos to 2G cell phone radiation resulted in increased double stranded breaks in the

eye [20]. Lixia et al [13] examined the effects of microwaves on DNA damage in human LEC and reported as repairable DNA damage. DNA breaks could easily accumulate, especially in neurons as they have a low capacity for DNA repair that could lead or accelerate the development of neurodegenerative diseases like Alzheimer's, Huntington's and Parkinson's disease. Also glial cells can become cancerous [25]. The WHO has classified mobile phone radiation on the IARC scale as group 2B- possibly carcinogenic.

The advancement in the mobile phone technology with multi-functional features attracts more users without any concern on the possible health hazards on the long term effects of radiofrequency radiation from the cell phone on the common man. The present study is undertaken to evaluate the possible effects of chronic exposure to RFR emitted from 2G/GSM mobile phones in developing chick embryo retina.

## 2. Materials and Methods

This study was approved by the Institutional Animal Ethical Committee (IAEC) and was carried out as per their recommendations. Fertile hen eggs (*Gallus domesticus*) were procured from the Rajiv Gandhi college of Veterinary and Animal sciences, Puducherry. The eggs were incubated in 9 batches of 12 eggs each (108 eggs) in a standard egg incubator at  $37\pm 0.5^{\circ}\text{C}$  and 50-55% of humidity and ventilation. The eggs were rotated manually 4 times a day and checked with a Candler for the viability of embryos.

The first batch (12 eggs) was treated as control (Group -C) and they were incubated without any external factors interfering with their developmental process. Next 4 batches (48 eggs) were treated as sham exposed group (Group-A). They were incubated along with a popular brand 2G cell phone with a frequency range of 900 – 1800MHz and SAR of 0.310 watts/kilogram hung from above with 5 cm distance separating the egg and kept in null status (switched off). Morphological features and structure of retina of both these groups were similar. So we have considered the sham exposed group as a control group for the present study.

The remaining 4 batches of (Group B) experimental group were exposed to RFR from the same cell phone that was used for sham exposed group. The eggs were incubated in a similar manner with the cell phone kept in silent mode (switched on) with headphone plugged in. This arrangement ensured that the cell phone gets switched on automatically each time it receives a call (Fig: 1). For exposure, the cell phone was rung from another cell phone for a duration of 3 minutes each, every half an hour, with the first exposure delivered at 12th hour of incubation (4.30am-4.30pm). The total exposure for a 12 hour period was 72 minutes, followed by 12 hours of exposure-free period. This was repeated daily.

Six embryos per day were sacrificed from 5<sup>th</sup> day to 12<sup>th</sup> day for Group A and B. Their weight and gross morphological features were recorded. The embryos were fixed in 10% formalin and then processed for routine histological studies. 5 micron thick sections were cut in sagittal, coronal and transverse planes and stained with H&E. The thicknesses of each layer of retina in control and exposed group were measured using calibrated oculometer and the values obtained were statistically analyzed using student t-test and Man Whitney test using SPSS 22 version software.

## 3. Observations

Histological examination of retina of 5 days control group showed mild pigmentation of pigment retina and neural retina showed closely packed cells without spaces between them. 3 layers were distinguishable; the layers being pigment layer, germinative or proliferative layer and inner marginal layer (putative optic nerve fibre) (Fig.2). Experimental group showed thin pigment retinal layer with mild pigmentation and neural retina showed 2 layers – germinative or proliferative layer showing spaces between the cells and an inner marginal layer (Fig.3). The thicknesses of all the 3 layers of the experimental group were significantly more when compared with control group ( $P = 0.01507, 0.00013, 0.028$  respectively) (Table1)

The retinal features of 6 days control group showed similar features of 5 day control. The experimental group retina showed 3 distinct layers. The pigment layer showed mild pigmentation, germinative or proliferative layer with cleft like spaces between cells and some of embryo showed disintegration in the inner marginal layer. The mean thickness of the pigment layer of both groups was same. The thickness of the germinative or proliferative layer was significantly more in the experimental group ( $P = 0.000004$ ) and inner marginal layer showed no significant difference in both groups. However, the total retinal thickness of the experimental group was significantly more when compared with control group ( $P = 0.000001$ ) (Table1).

Retina of 7 days old control group showed mild to moderate pigmentation with similar 3 layers. The experimental group showed similar 3 layers with pigment layer showing moderate - intense pigmentation and thickness of neural retina was more compared to control group. The germinative or proliferative layer also showed spaces between cells & inner marginal layer showed disintegrated optic nerve fibres. The thickness of pigment cell layer of both group showed same thickness. The mean thickness of germinative layer & inner marginal layer of experimental group was significantly more when compared with the control group ( $P = 0.000014, 0.000001$  respectively). Total retinal thickness of experimental group showed increased thickness which was highly significant ( $P = 0.00004$ ) (Table1)

**Table 1:** Mean Thickness of Each Layer of Retina in Both Groups

Age (Days)	Pigment layer (mm)		Germinative layer (mm)		Optic fibre layer (mm)		Total thickness (mm)	
	Control	Exp.	Control	Exp.	Control	Exp.	Control	Exp.
5	0.0030	0.0039*	0.475	0.0553**	0.0052	0.0067*	0.0558	0.066**
6	0.0049	0.0049	0.0558	0.0882**	0.0054	0.0061	0.0660	0.0793**
7	0.0049	0.005	0.0716	0.0825**	0.0059	0.0075**	0.0824	0.0951**

(\* P value ≤ 0.05, \*\* P value ≤ 0)

Retina of 8<sup>th</sup> day old control embryo showed moderate pigmentation and few spaces were observed between the cells. Most of the control retina showed only 3 layers – pigment layer, germinative layer and inner marginal layer (Fig.4). In two embryos (33.2%) of control group 5 layered retina was observed. The layers were pigment layer, outer neuroblastic layer, inner neuroblastic layer and a layer of tangled cell processes demarcating them (transient layer of chievitz) and inner marginal layer. The entire experiment group embryo showed 5 layers of the retina. The pigment layer showed mainly moderate pigmentation of the retina. They also showed increased spaces between the cells in inner neuroblastic layer and disintegrated optic nerve fibre (Fig.5). On comparing the thickness of all the layers, the pigment cell layer had same thickness in control and experimental group. The thickness of outer neuroblastic layer of the experimental group was significantly more than control group (P=0.029). Inner plexiform layer, inner neuroblastic layer and optic nerve fibre layer showed no significant change in both groups. However, the total retinal thickness of the experimental group was significantly more when compared with the control group (P=0.029)(Table 2).

**Table 2:** Mean Thickness of Each Layer of Retina in Both Groups

Age (Days)	Layers (mm)					
	Pigment	Outer neuroblastic	Transient (chievitz)	Inner neuroblastic	Optic fibre	Total thickness
8 (CON)	0.005	0.075	0.0044	0.0106	0.0099	0.1044
8 (EXP)	0.005	0.0868*	0.0068	0.0088	0.0131	0.1206*

(\* P value ≤ 0.05)

9 days control embryo showed well-formed 8 layers. The external plexiform layer was clearly seen from 9<sup>th</sup> day onwards separating external and internal nuclear layer. Pigment layer showed moderate pigmentation. All layers were well-formed with little space between cells. 9 days experimental embryo showed intense pigmentation of the retina with well differentiated 8 layers and spaces were visible between the cells in external nuclear layer, internal nuclear layer and ganglion cell layer. The thickness of all the layers were found to be almost same for both control and experimental group except internal nuclear layer which was found to be more in thickness in the experimental group (P=0.005). The total thickness of the experimental group also showed a significant increase in thickness (P=0.0473) (Table 3).

10 days old control showed moderate pigmentation of the retina with other normal features. All the embryos of experimental group showed intense pigmentation and increased space between cells of internal nuclear layers, ganglion cell layer and optic nerve fibre. Internal plexiform layer was well developed in comparison with control group. Moreover, internal limiting membrane was also visible. The thickness of pigment layer, layer of rods and cones, external nuclear layer & external plexiform layer of both groups showed same thickness. The thickness of internal nuclear layer, internal plexiform layer, ganglion cell layer & optic nerve fibre layer of experimental group were more when compared with control group and was highly significant (P=0.000009, 0.000008, 0.000001, 0.000021 respectively). The total thickness of experimental group also showed significant increase in thickness (P=0.0000001) (Table 3).

11 days old control embryos showed moderate to intense pigmentation of the retina and normal histological features. The experimental group showed intense pigmentation of retina with spaces in inner nuclear and ganglionic cell layer. Optic nerve fibres were intact in some places, but some areas showed disintegration. Internal plexiform layer was well formed in the experimental group. The thickness of pigment layer, layer of rods and cones, external nuclear layer, external plexiform layer & internal nuclear layer of both groups showed no significant change in thickness. The thickness of internal plexiform layer & optic nerve fibre layer were significantly more in the experimental group (P=0.00001, 0.02291 respectively). However, the thickness of ganglion cell layer was significantly more in control group (P=0.00007). The total retinal thickness showed no significant change in both groups (Table 3).

12 day old control embryo showed normal retina with moderate pigmentation (Fig 6). Experimental group showed intense pigmentation of retina with spaces in inner nuclear layer and disintegrated optic nerve fibre (Fig 7). The thickness of pigment layer, layer of rods and cones, external nuclear layer & ganglion cell layer of both groups showed same thickness. External plexiform layer, internal nuclear layer & optic nerve fibre layer of experimental group showed increased thickness than control group (P=0.0085, 0.00001, 0.0000001 respectively). The thickness of internal plexiform layer of control group embryos was significantly more than experimental group (P=0.0000003). The total retinal thickness of experimental group was significantly more (P= 0.0007) (Table 3).

**Table 3:** Mean Thickness of Each Layer of Retina in Both Groups

Thickness of layers (mm)	9 <sup>th</sup> day		10 <sup>th</sup> day		11 <sup>th</sup> day		12 <sup>th</sup> day	
	control	exposed	control	exposed	control	Exposed	control	exposed
Pigment	0.005	0.005	0.005	0.005	0.005	0.005	0.005	0.005
Rods& cones	0.0025	0.0025	0.0025	0.0025	0.0025	0.0025	0.0025	0.0025
External nuclear	0.0103	0.0109	0.0098	0.0098	0.01	0.01	0.0120	0.0118
External plexiform	0.0033	0.0038	0.0044	0.0043	0.0038	0.0039	0.0029	0.0037**
Inner nuclear	0.0588	0.0694**	0.0666	0.0761**	0.0801	0.0789	0.0809	0.0932**
Inner plexiform	0.0068	0.0067	0.0046	0.0066**	0.0073	0.0091**	0.0128**	0.0094
Ganglionic	0.0164	0.0156	0.0160	0.0226**	0.0263*	0.0225	0.0240	0.0234
Optic layer	0.0095	0.009	0.0116	0.0173**	0.0108	0.0154*	0.0155	0.022**
Total retinal Thickness	0.1126	0.1231*	0.1205	0.1436**	0.1461	0.1477	0.1558	0.1712**

(\* P value ≤ 0.05, \*\* P value ≤ 0)

On comparing the total thickness of retina of both groups, the experimental group showed significant increase in thickness on all days except on the 11<sup>th</sup> day which showed a non-significant increase in total thickness of retina. (Table-4)

Both 5<sup>th</sup>& 6<sup>th</sup> day control and experimental group showed mild pigmentation of retina. While 7<sup>th</sup>& 8<sup>th</sup> day control embryo showed mild pigmentation, whereas, experimental group showed moderate pigmentation. 9<sup>th</sup> – 12<sup>th</sup> day control embryo showed moderate pigmentation while experimental group showed intense pigmentation. (Table-4)

**Table 4:** Mean Total Retinal thickness & Pigmentation Grade in Both Groups.

Age in days	Mean retinal thickness (mm)		Pigmentation	
	Control	Exposed	Control	Exposed
5	0.0558	0.066**	+	+
6	0.0660	0.0793**	+	+
7	0.0824	0.0951**	+	++
8	0.1044	0.1206*	+	++
9	0.1126	0.1231*	++	+++
10	0.1205	0.1436**	++	+++
11	0.1460	0.1477	++	+++
12	0.1558	0.1712**	++	+++

(\* P ≤ 0.05, \*\* P ≤ 0) + mild, ++ moderate., +++ intense

On comparing the thicknesses of each layer of retina in both control and experimental group the following changes were noticed. The thickness of pigment layer, rods and cones of both control and experimental group for all age group didn't show much difference and remained constant at 0.005&0.0025mm respectively except on 5<sup>th</sup> day where a significant increase in thickness of these layers was observed in exposed group (Table 1). The thicknesses of external nuclear layer and external plexiform layer were of same for control and experimental group. The thicknesses of internal nuclear layer, internal plexiform layer, ganglion cell layer and optic nerve fibre layer showed an increase in thickness in experimental group except on 11<sup>th</sup>& 12<sup>th</sup> day. On 11<sup>th</sup> day the thickness of ganglion cell layer of control group was significantly more than experimental group. On 12<sup>th</sup> day inner plexiform layer of control group was significantly more than experimental group. (Table -3)

#### 4. Discussion

In our study, histological examination of the retina in correlation with control, a gradual increase in retinal thickness was observed in the exposed group. The internal nuclear layer, internal plexiform layer, ganglion cell layer and optic nerve fibre layer were increased in thickness in experimental group and were statistically significant except on the 11<sup>th</sup> day and 12<sup>th</sup> day. On the 11<sup>th</sup> day the exposed group showed decreased thickness of the ganglion cell layer and 12<sup>th</sup> day exposed group showed significant decrease in the inner plexiform layer in comparison with the control group. The differences in the growth parameters of different layers of the retina might be due to different cellular responses to EMF during embryological periods as cells might be trying to rebalance their growth and differentiation rate [8].

Fatima Al Qudsi et al [8] in their study on exposing the chick embryo to 2G cell phone radiation, showed an increased thickness of all the layers of exposed embryo of 7<sup>th</sup> day of incubation which was similar to our findings. The exposed embryo of the 10<sup>th</sup> day of incubation of the same study showed increased thickness of the inner nuclear layer, inner plexiform layer and the ganglion cell layer, disintegrated Optic nerve fibre and spaces in the inner nuclear layer. Similar findings were seen in the present study also. On increasing the exposure up to 14<sup>th</sup> day, the authors found same histological changes similar to that of 10<sup>th</sup> day exposed group and decrease in retinal thickness. In our study these changes in the form of increased spaces between the cells of the inner nuclear layer and ganglion cell layer, disintegrated optic nerve fibre were also found in 11<sup>th</sup>& 12<sup>th</sup> day exposed group. But the total retinal thickness was increased in experimental group which was significant.

The present study also showed early differentiation of 5 layered of the retina in 8 days old experimental embryos and control embryos showed mainly 3 layers in the same age group. The layers were pigment layer, outer neuroblastic layer, inner neuroblastic layer and a layer of tangled cell processes demarcating them (transient layer of chievitz) and inner marginal layer. Outer neuroblastic layer later differentiates into external nuclear layer, external plexiform layer and internal nuclear layer. Transient layer of Chievitz forms internal plexiform layer and inner neuroblastic layer form ganglion cell layer. Inner marginal layer forms an optic

nerve fiber layer [26]. As age advanced, the thickness of the ganglion cell layer in 11<sup>th</sup> & 12<sup>th</sup> day exposed embryos showed reduced thickness, due to natural cell death or apoptosis that normally happens in ganglion cell layer towards the end of gestation [26]. This probably would have resulted in decreased thickness of inner plexiform layer due to loss of synaptic contact between ganglion cells and cells of inner nuclear layer. These changes exhibit an early onset of maturation of the retina in exposed group than the control group.

Zareen et al [11] on exposing the chick embryos to RF radiation found that the exposure decreased retinal growth and differentiation up to 10<sup>th</sup> day; while prolonged exposure increased the retinal growth up to 15<sup>th</sup> day. The authors reported in their study the role of melanin present in retinal pigment epithelium (RPE) in the differentiation of the neural retina [11, 27]. Dopa, a melanin precursor present in RPE is important for regulating retinal cell mitosis [28].

RF exposure resulted in DNA damage [13, 23, 24] in the form of single strand breaks (SSB) and double strand breaks [20]. This affected melanogenesis which is one of the repair mechanisms [29]. Zareen et al [11] reported that the exposed group up to 10<sup>th</sup> day of incubation showed mild pigmentation of RPE that resulted in retarded growth and differentiation of the neural retina. Whereas, on prolonged exposure it resulted in intense pigmentation of RPE due to increased melanin production that resulted in increased growth of the retina.

In our study, the melanin pigmentation on exposed groups were mild up to 6<sup>th</sup> day followed by moderate pigmentation on 7<sup>th</sup> day & 8<sup>th</sup> day and intense pigmentation from 9<sup>th</sup> – 12<sup>th</sup> day. However, control group showed mild pigmentation upto 8<sup>th</sup> day followed by moderate pigmentation up to 12<sup>th</sup> day.

The increased total retinal thickness and different layers of retina in the exposed group in correlation with control group might be due DNA damage caused by RF radiation on RPE cells. This would have caused increased melanogenesis resulting in increased retinal thickness and differentiation of neural retina.

RF radiations caused irreversible morphological and biochemical changes at the cellular and molecular level, resulting oxidative stress, an increase in the level of heat shock proteins HSP-70, HSP-27 [13, 14]. These proteins protect the body from oxidative stress, but on prolonged exposure results in cell death by compromising the immune system. In the present study the increased space between the cells of various layers of retina in the exposed group might be due to shrinkage of cells or it might be due to cell death caused by chronic exposure of embryos to RF radiation that resulted in oxidative stress rendering the cells vulnerable to the damaging effects of RF radiation.

## 5. Conclusion

The present study shows that the chronic exposure of chick embryos to RF radiation from 2G cell phone resulted in increased retinal thickness and early retinal differentiation

probably due to DNA damage causing increased melanogenesis in RPE. Exposed group also showed structural changes in the form of increased spaces between the cells in the different layers of retina and disintegrated optic nerve fibre layer.

## 6. Future Scope for Study

Whether these reported changes are reversible or not upon removing the radiation source requires further study. Introduction of new generation phones (3G and 4G) widens the scope for future studies to investigate the effect of RF radiation emitted from them on developing tissues and to compare the effects to find out which one has more damaging effect, if any.

## 7. Conflicts of Interest

The authors had no conflicts of interest to declare in relation to this article.

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**Figure 1:** A Photograph showing the experimental set up. The Mobile phone (red arrow) is hung with a distance of 5cms separating it from the fertilized chicken eggs. A RF meter is kept inside the incubator to check the intensity of radiation (yellow arrow).

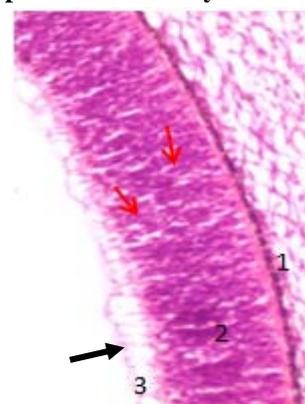
**5 Day Old Control Embryo Retina**



**Figure 2:** Photomicrograph of 5 day old control Embryo retina showing 3 layers –

- 1) Pigmented layer-mild pigmentation
- 2) Germinative layer
- 3) Inner marginal layer (H&E X 400)

**5 Day Old Experimental Embryo Retina**



**Figure 3:** Photomicrograph of 5 day old experimental Embryo retina showing 3 layers –

- 1) Pigmented layer-mild pigmentation
- 2) Germinative layer showing spaces between the cells (red arrow)
- 3) Disintegrated inner marginal layer (black arrow) (H&E X 400)

8 Day Old Control Embryo Retina

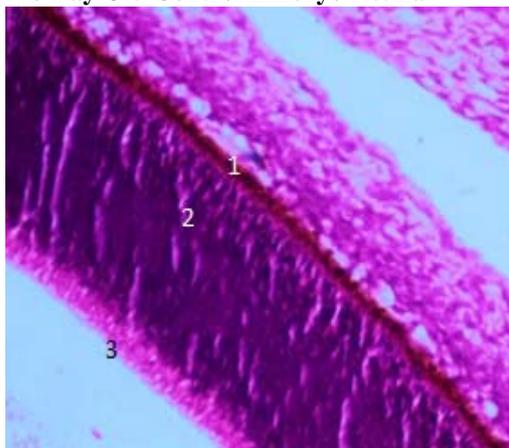


Figure 4: Photomicrograph of 8 day old control Embryo retina showing 3 layers –

- 1) Pigmented layer - moderate pigmentation
- 2) Germinative layer
- 3) Inner marginal layer (H&E X 400)

8 Day Old Experimental Embryo Retina

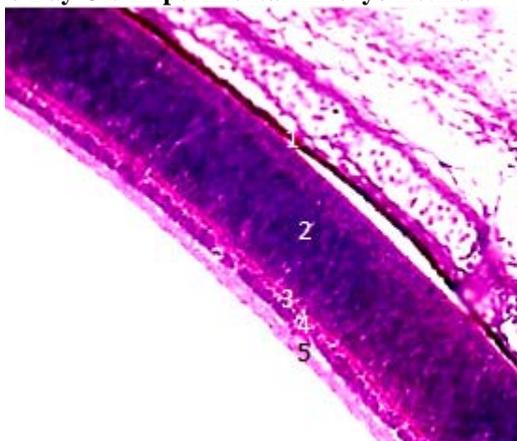


Figure 5: Photomicrograph of 8 day old experiment Embryo retina showing 5 layers –

- 1) Pigmented layer – intense pigmentation
- 2) Outer neuroblastic layer
- 3) Transient layer of Chievitz
- 4) Outer neuroblastic layer
- 5) Inner marginal layer (H&E x 400 )

12 Day Old Control Embryo Retina

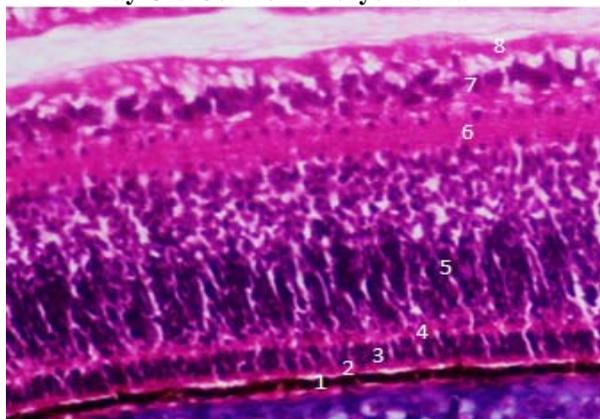


Figure 6: Photomicrograph of 12 day old control embryo retina showing 8 layers –

- 1) Pigment Layer
- 2) Rods & Cones
- 3) External Nuclear Layer
- 4) Ext. Plexiform Layer
- 5) Internal Nuclear Layer
- 6) Well Developed Internal Plexiform Layer
- 7) Ganglion Cell Layer
- 8) Optic Nerve Fibre Layer (H&E X 400 )

12 Day Old Experimental Embryo Retina

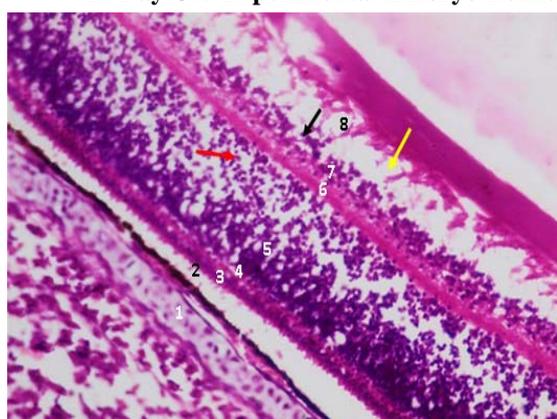


Figure 7: Photomicrograph of 12 day old experiment embryo retina showing 8 layers –

- 1) Pigment layer
- 2) Rods & cones
- 3) External nuclear layer
- 4) External plexiform layer
- 5) Internal nuclear layer showing spaces (red arrow)
- 6) Internal plexiform layer
- 7) Ganglion cell layer showing spaces between cells (black arrow)
- 8) Disintegrated Optic nerve fibre layer (yellow arrow) (H&E x 400)

## Declaration

The Undersigned authors hereby declare that the manuscript “Histological Study of Chick Embryo Retina Exposed to Radiofrequency Radiation Emitted From 2G Cell Phone” has been read and approved and the work has been carried out in the department of Anatomy, MGMC & RI under our supervision. The authors warrant that the article is original and is not under consideration by any other journal and has been previously published and taken responsibility for the context. Furthermore, they warrant that all investigations reported in their publication were conducted in conformity with the recommendation from the declaration of Helsinki and the international guiding principles for biomedical research involving animals.

## Author Profile



**Mary Hydrina D'Silva** has done her MSc Anatomy from Mahatma Gandhi University, Kottayam, Kerala. Currently she is doing her research in cell phone radiation and its effects on developing tissues of chick embryo. At present, she is working as Assistant Professor in Anatomy at MGMC & RI, Pondicherry.



**Rijied Thompson Swer** has done his MD Anatomy from JIPMER, Pondicherry. At present, he is working as Associate Professor in Anatomy at MGMC & RI, Pondicherry.



**J. Anbalagan** has done his MSc Anatomy from JIPMER, Pondicherry and PhD in Anatomy from Mahatma Gandhi institute of Medical Sciences, Sevagram. He has co- authored a text book titled “Histology – Text and Atlas” and has published several research papers in various international and national journals. At present, he is working as Professor of Anatomy at MGMC & RI, Pondicherry.