Title : A Cost-Effective and Efficient egg windowing method to teach early embryonic development in chick (*Gallus gallus domesticus*) to under-graduate students

Dr. P.Jayaraj, Assistant Professor, Department of Zoology

Work : Embryology and developmental biology teachings rely mostly on online resources as animations or time-lapse videos. Fantasying the embryological stages falls far behind in giving an exact picture of the events to undergraduate students. This study uses chicken embryo biological models for observing naked eye development stages. These model systems not only prove to be a realistic approach to the subject of embryology but also provide an ideal experimental medium for research.

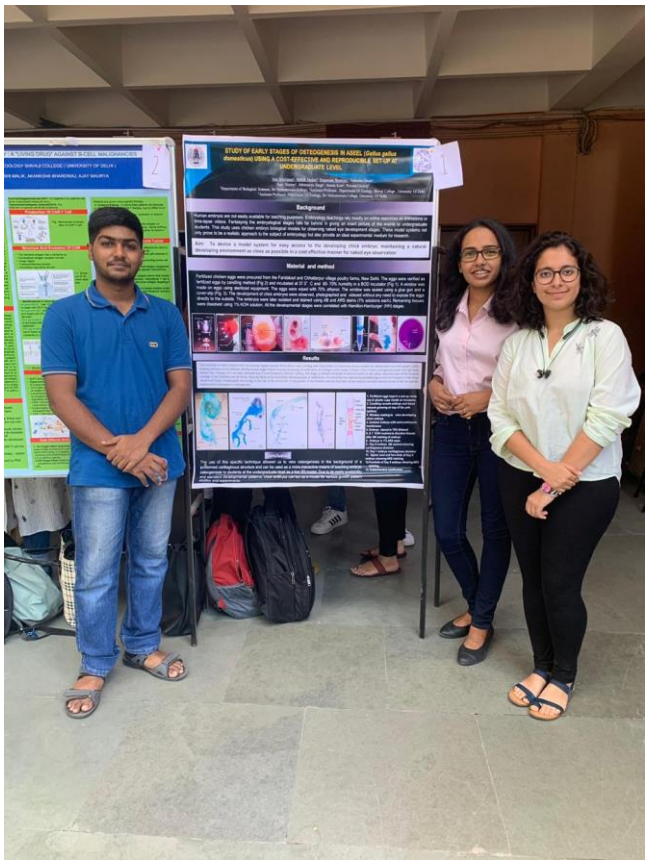
Chick embryo were cultured and observed by novel window making method – Cup in cup and candling methods, all the practical were performed within college premises using tool and instruments available in the department

Participating students: sixty

Diversity of students : Students of B.Sc (H) Biological Science and B.Sc (H) Zoology were benefitted

Outcome and Value addition: Studying the real-time developmental process has always remained a challenge, This embryological stages observed in this model system can be good specimens for teaching embryology in anatomy museums. This naked eye observation system is accurate, cost effective and easily reproducible for undergraduate research and academic purposes.

In Addition students were able do additional research based on the chick embryo development and presented their work in national conference and symposia



First prize for best poster presentation entitled " study of early stages of osteogenesis in Aseel using a cost effective and reproductive set-up" **at National seminar on recent trends in biological research and career prospects"** held on 27th September 2019 at Shivaji college, University of Delhi. (authors: Vani Srinivasan, Mehak Madan, Deepanjan Banerjee,**Perumal Jayaraj)**

Study of Early stages of Osteogenesis in Aseel (*Gallus gallus domesticus*) using a Cost-Effective and Reproducible set-up at undergraduate and post graduate level

Nabanita Ghosh (hghosh161@gmail.com), Sejal Sharma (sejal.sharmasejal08@gmail.com),
Vani Srinivasan, Mehak Madan, Deepanjan Banerjee, Abhimanyu Singh, Ms. Nimita Kant[2], Perumal Jayraj[3]
[1]Department of Biological Sciences, Sri Venkateswara College[2]Assistant Professor ,Department Of Zoology, Shivaji College , University Of Delhi
[3]Assistant Professor ,Department Of Zoology, Sri Venkateswara College, University Of Delhi., Bsc Biological Sciences, Sri Venkateswara College

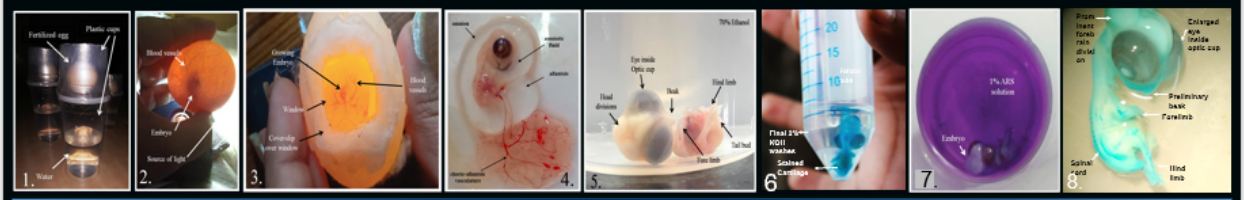
Background

Human embryos are not easily available for teaching purposes. Embryology teachings rely mostly on online resources as animations or time-lapse videos. Fantasizing the embryological stages falls far behind in giving an exact picture of the events to undergraduate students. This study uses chicken embryo biological models for observing development stages using windowed eggs. These model systems not only prove to be a realistic approach to the subject of embryology but also provide an ideal experimental medium for research.

Aim: To device a model system for easy access to the developing chick embryo, maintaining a natural developing environment as close as possible in a cost effective manner for naked eye observation

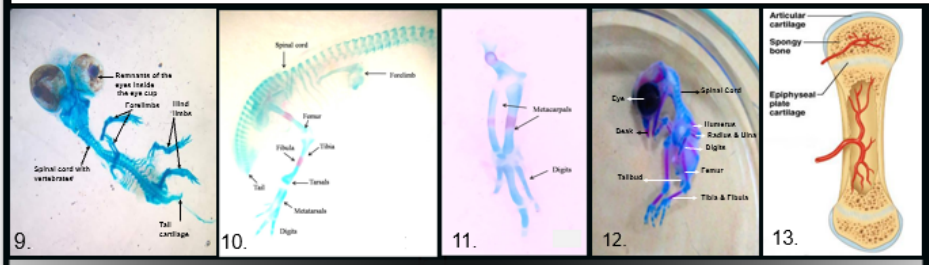
Material and method

Fertilized chicken eggs were procured from the Faridabad and Chhattarpur village poultry farms, New Delhi. The eggs were verified as fertilized eggs by candling method (Fig 2) and incubated at 37.5° C and 60- 70% humidity in a BOD incubator (Fig 1). A window was made on eggs using sterilized equipment. The eggs were wiped with 70% ethanol. The window was sealed using a glue gun and a cover-slip (Fig. 3). The development of chick embryos was observed, photographed and videoed without any need to expose the eggs directly to the outside. The embryos were later isolated and stained using AB and ARS stains (1% solutions each). Remaining tissues were dissolved using 1% KOH solution. All the developmental stages were correlated with Hamilton-Hamburger (HH) stages.



Results

The treatment of chick embryos with the staining reagents namely Alcian Blue to stain cartilage and Alizarin Red S to stain bone tissues revealed the skeletal structures with greater clarity. Staining embryos of two different developmental stages helped to study the process of ossification of cartilage to form bones. Embryo 1 (Day 7) had a cartilaginous spinal cord and limbs stained blue. Absence of a red stain indicated lack of bone formation. Embryo 2 (Day 9, HH stage 34) showed red bands at identical points on the radius, ulna and a part of the humerus cartilage of the forelimbs and the femur, tibia and fibula of the hind limbs indicating onset of ossification. Embryo 3 (Day 13, HH stage 39) showed red colour along the length of the limb while blue color was seen at the joints, digits, spinal cords etc. We concluded that ossification probably begins with the formation of the limb bones, subsequently continuing to the rest of the structures. A comparison of the forelimb and the hind limb of the embryos provides absolute proof of the site-specific endochondral mechanism of osteogenesis.



1. Fertilized eggs kept in a set-up made out of plastic cups inside an incubator.
2. Candling reveals embryo and blood vessels growing on top of the yolk (yellow)
3. Window making ton view developing chick embryo
4. Isolated embryo with extra-embryonic membranes
5. Embryo 2 stored in 70% Ethanol
6. KOH washes to dissolve tissues after AB staining of embryo
7. Embryo in 1% ARS stain
8. Day 9 embryo AB stained showing cartilaginous skeleton
9. Day 7 embryo cartilaginous skeleton
10. Spinal cord and fore-limb of Day 9 embryo showing ARS staining
11. Forelimb of Day 9 embryo showing ARS staining.
12. Day 13 embryo AB stained and ARS stained showing cartilages and bones
13. Endochondral ossification

Conclusion

The use of this specific technique allowed us to view osteogenesis in the background of a preformed cartilaginous structure and can be used as a more interactive means of teaching embryo osteogenesis to students at the undergraduate level as a live 3D model. Due to its ready availability and standard developmental patterns, chick embryos can act as a model for various growth pattern studies and experiments.

Award winning Poster: " presented at National seminar on recent trends in biological research and career prospects" held on 27th September 2019 at Shivaji college, University of Delhi