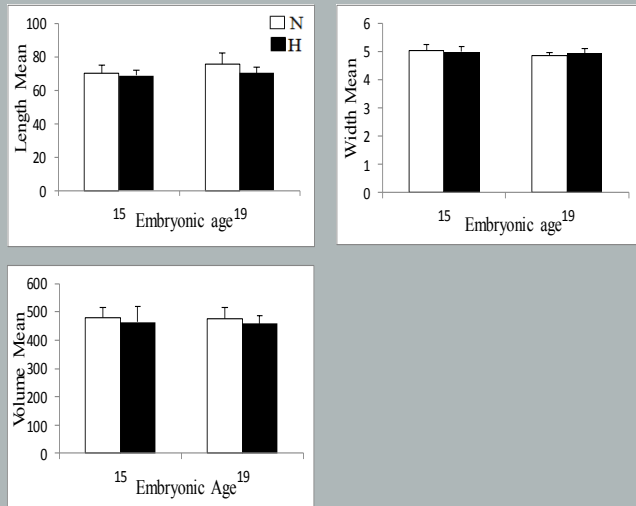


Result 3— Hypoxic exposure did not produce cardiomyocyte hypertrophy in the third stage of incubation



This figure shows the mean measurements of length width and cell volume of chicken cardiomyocyte in 15 and 19 day old embryos.

Conclusions:

1. There is no change in ventricular wall thickness due to chronic hypoxia at both stages of incubation.
2. There is no cardiomyocyte hypertrophy in both 15 and 19 day old chicken embryos due to chronic hypoxia
3. There are no binucleated cells due to hypoxia during the last stage of incubation

Take Home Message:

Chronic hypoxia is not affecting the structure of heart in 15 & 19 day old chicken embryos!!!



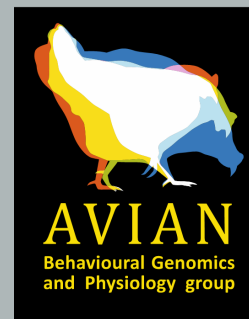
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Chronic hypoxia impairs embryonic growth but did not affect ventricular wall thickness and cardiomyocyte size in the broiler chicken fetus



Thesis

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Background:

Proliferation and differentiation are the two main regulators for the cell growth. During the period of proliferation and maturation, oxygen plays a vital role. The fetus responds to chronic hypoxia by increasing its cardiac output by 50% than the normal. Chronic hypoxic exposure during development can produce impact in both anatomical and physiological ontogeny. When exposed to hypoxic conditions chicken embryos develop relatively larger hearts than under normal conditions.

Previous studies suggest that lower oxygen tension in the fetus is essential for normal heart formation. Studies in chickens to understand the hemodynamic changes (mean arterial pressure, heart rate and peripheral arterial resistance) during fetal development under hypoxic conditions and transition of cardiomyocytes from hyperplasia to hypertrophy during postnatal development have been extensively made (Faqian et al., 1997). The effects of cardiovascular changes during fetal development under hypoxic condition are poorly understood.

Aim of the study:

This study aims at measuring ventricular wall thickness and cardiomyocyte size to understand the physiological changes in structure of the heart due to chronic hypoxic exposure at 15 and 19 days of incubation age.

Experimental design:

Fourteen chronically hypoxic chickens at 15 and 19 days of age each were taken for study; 14 control fetuses were of similar age. The hypoxic incubator is maintained at 14.5% O₂ and normoxic incubator is maintained at 21% O₂

Ventricular wall thickness measurement:

Excised hearts were rinsed in PBS buffer and pinned to silicon plate. Through the two catheters inserted into pulmonary artery and aorta, PFA is flushed into the chambers and then stained with 1% methylene blue in PFA. The heart is flushed with PBS and filled with embedding media (OCT gel). The heart is made frozen at -80°C and sections are made in cryostat. Sliced sections are captured with mediscopes and pictures are analyzed with NIS-AR elements. Measurements are made manually and calibrated.

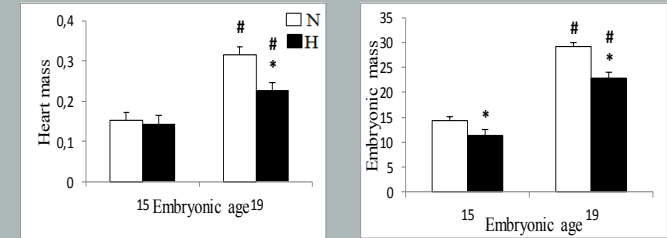
Cardiomyocyte size measurement:

Cardiomyocytes are isolated through retrograde perfusion technique. Myocyte slurry is fixed with PFA and stained with DAPI stain. Processed slides are viewed under fluorescent microscope and cell pictures are captured with the help of QIMC camera. Cells are measured manually with NIS-AR elements software.

With the length and width measurements of cardiomyocyte, the cell volume is calculated.

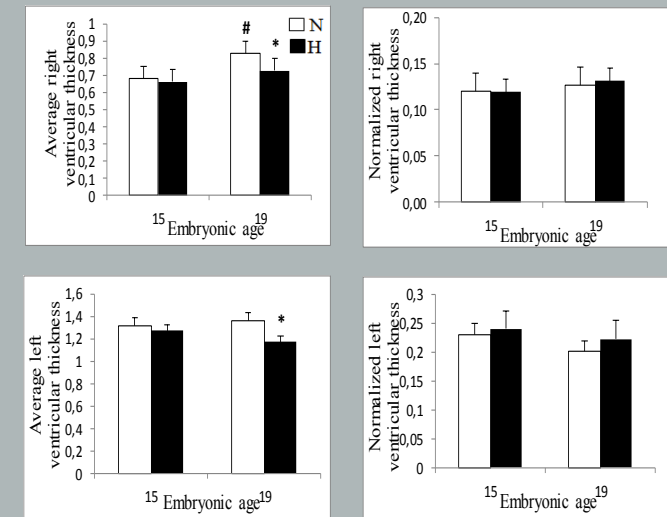
Results:

1. Chronic hypoxia impairs embryonic and heart growth



This figure shows that hypoxic exposure impaired embryonic growth and heart growth compared to the normoxic groups. # and * symbols denote the significant difference with respect to treatment and age respectively.

2. Chronic hypoxia did not affect the thickness of ventricular wall in both the incubation age



This figure shows the average wall thickness measurements and the wall thickness normalized with respect to the mean diameter of each heart sample in both 15 and 19 day old embryos.