Respiratory and cardiovascular responses to oxygen and carbon dioxide levels in internally pipped chicken embryos

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Lab Goals

- To understand the development of human respiratory and cardiovascular systems using avian models
  - To characterize the ductus arteriosi in chicken embryos
Mammalian Ductus Arteriosus

- Fetal Circulatory System
- Vessel that connects pulmonary artery with aorta
- Shunts blood away from lungs to body
- Closes upon birth

Randell et al., 2002: Circulation In Womb
Avian Ductus Arteriosi

- Notice:
  - Aorta branches left, not right.
  - Two longer ductus arteriosi
Lab Goals

- To understand the development of human respiratory and cardiovascular systems using avian models
  - To characterize the ductus arteriosi in chicken embryos
Focus

- How does O$_2$ uptake at the CAM and interact during internal pipping?
- How does CO$_2$ uptake at the CAM and lungs interact during internal pipping?
Pre-pipped

CAM

Internally Pipped

CAM + Lungs

12% $\text{O}_2$

5% $\text{CO}_2$
Focus

- How does $O_2$ uptake at the CAM and interact during internal pipping?
- How does $CO_2$ uptake at the CAM and lungs interact during internal pipping?
Lung and CAM Respiration

Air Cell Gas Mixture
- 12% O$_2$, 5% CO$_2$, 83% N$_2$

Egg
- Air

Measure lung and CAM oxygen consumption separately
- Open flow respirometry
## O$_2$ Level of Exposure

<table>
<thead>
<tr>
<th>CONTROL</th>
<th>TESTING</th>
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<tbody>
<tr>
<td><strong>Egg O$_2$</strong></td>
<td><strong>Air Cell O$_2$</strong></td>
</tr>
<tr>
<td>21%</td>
<td>12%</td>
</tr>
<tr>
<td>21%</td>
<td>21%</td>
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<td>30%</td>
<td>5%</td>
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</table>
Lung metabolic rate vs. total metabolic rate at normoxia
Results

- There was a positive correlation between $\text{Vo}_2$ lungs and $\text{Vo}_2$ total.
Lung metabolic rate vs. total metabolic rate
Results

- There was a positive correlation between \( \text{Vo}_2 \) lungs and \( \text{Vo}_2 \) total.

- Different stages of internal pipping
Changes in Lung MR vs. changes in CAM MR

Δ Δ \( \Delta V_{O_2}^{\text{lung}} \) (ml \( O_2 \) min\(^{-1}\))

Δ \( \Delta V_{O_2}^{\text{CAM}} \) (ml \( O_2 \) min\(^{-1}\))

-0.3 -0.2 -0.1 0.0 0.1 0.2 0.3

-0.3 -0.2 -0.1 0.0 0.1 0.2 0.3

-egg 21% : AC 5%
-egg 21% : AC 21%
-egg 15% : AC 12%
-egg 30% : AC 12%
-egg 15% : AC 5%
-egg 30% : AC 21%
-egg 30% : AC 5%
-egg 15% : AC 21%
Changes in Lung MR vs. changes in CAM MR

\[ \Delta \Delta V_{O_2}^{\text{lung}} \text{ (ml O}_2\text{ min}^{-1}) \]

\[ \Delta V_{O_2}^{\text{CAM}} \text{ (ml O}_2\text{ min}^{-1}) \]

-0.3 -0.2 -0.1 0.0 0.1 0.2 0.3

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-egg 30% : AC 12%
-egg 21% : AC 21%
-egg 15% : AC 5%
-egg 30% : AC 21%
-egg 30% : AC 5%
-egg 15% : AC 21%
Results

- Embryos have the ability to use the CAM or lungs to compensate for each other.

- Significance:
  - CAM hyperoxic, lungs hypoxic $\rightarrow$ increased VO$_2$total
  - Both CAM and lungs are hypoxic $\rightarrow$ both decrease

- Could response be due to changes in ventilation patterns?
Changes in ventilation after changing air cell $O_2$ and $CO_2$

Data presented as % of control

<table>
<thead>
<tr>
<th></th>
<th>$AC$ 5% $O_2$</th>
<th>$AC$ 21% $O_2$</th>
<th>$AC$ 2.5% $CO_2$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tidal volume</td>
<td>103 ± 6</td>
<td>96 ± 7</td>
<td>55 ± 6*</td>
</tr>
<tr>
<td>Frequency</td>
<td>103 ± 3</td>
<td>105 ± 6</td>
<td>96 ± 7</td>
</tr>
<tr>
<td>Pulmonary ventilation</td>
<td>107 ± 8</td>
<td>101 ± 9</td>
<td>51 ± 6*</td>
</tr>
</tbody>
</table>
At internal pipping:
- Ventilatory oxygen chemosensitivity has little role in controlling \( \text{O}_2 \) exchange
- Develops during external pipping
- \( \text{CO}_2 \) chemosensitivity is functioning

\( \Delta \) in ventilation plays no role in controlling gas exchange in response to hypoxia or hypoeroxia
Blood Flow

- Dr. Dzialowski, Upward Bound Students
- Microspheres Technique
Hypoxia

Repeated Measures ANOVA
CAM $p = 0.013$
Brain $p = 0.001$

5% $O_2$ into air cell

Organ Blood Flow
Lung Blood Flow

Time (min) 0 10 20 30 40 50 60

* CAM
* Brain

$\text{5\% } O_2 \text{ into air cell}$
### Hyperoxia

![Graph showing organ blood flow over time with repeated measures ANOVA results: CAM p = 0.024, Brain p = 0.26.]

**Graph Details:**
- **X-axis:** Time (min) ranging from 0 to 60.
- **Y-axis:** Blood Flow with scales for Organ and Lung Blood Flow.
- **Data Points:**
  - Red circles represent CAM with a significant decrease in blood flow.
  - Blue triangles represent Brain with a decrease in blood flow.
- **Significance:**
  - CAM p = 0.024
  - Brain p = 0.26
- **Note:** 21% O2 into air cell
Results

- Lung hypoxia $\rightarrow$ increased blood flow through the ductus arteriosi
- Lung hyperoxia $\rightarrow$ decreased blood flow through the ductus arteriosi

- Achieved by altering blood flow to the lungs and through the ductus arteriosi and the interatrial foramina
Focus

- How does $O_2$ uptake at the CAM and interact during internal pipping?
- How does $CO_2$ uptake at the CAM and lungs interact during internal pipping?
# CO₂ Level of Exposure

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<tbody>
<tr>
<td>Egg CO₂</td>
<td>Air Cell CO₂</td>
</tr>
<tr>
<td>0%</td>
<td>5%</td>
</tr>
<tr>
<td>0%</td>
<td>2.5%</td>
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<tr>
<td>0%</td>
<td>5%</td>
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</table>
Changes in Lung MR vs. changes in CAM MR

- Egg 0% : AC 0%
- Egg 0% : AC 2.5%
- Egg 0% : AC 10%
- Egg 5% : AC 5%

$\Delta V_{O2}^{\text{Lung}} (\text{ml O}_2 \text{ min}^{-1})$

$\Delta V_{O2}^{\text{CAM}} (\text{ml O}_2 \text{ min}^{-1})$
Results

- Lung hypercapnia $\rightarrow$ no change $\text{VO}_2\text{total}$
- Lung hypocapnia $\rightarrow$ decreasing $\text{VO}_2\text{total}$
- Egg hypercapnia $\rightarrow$ increase both $\text{VO}_2\text{total}$

- Brain senses $\text{CO}_2$ levels (carbon dioxide chemosensitivity)
Final Conclusions

- During internal pipping, chicken embryos have two sites of respiration: CAM and air cell.

- When there is a change in oxygen level at one respiration site, the other site compensates.
  - Blood flow is the compensation mechanism for $O_2$ changes.

- $O_2$ chemosensitivity in the IP chicken embryo is weak and developing, while $CO_2$ chemosensitivity is functional.
Future Research

- Measure ventilation and blood flow patterns during hypercapnia and hypocapnia
QUESTIONS?
Acknowledgments

- Dr. Dzialowski, Faculty Mentor
- Dr. Eve and Dr. Cox, Honors College
- Upward Bound Students:
  - Miguel Cavazos
  - Felicia Guerrero
  - Heather Morgan