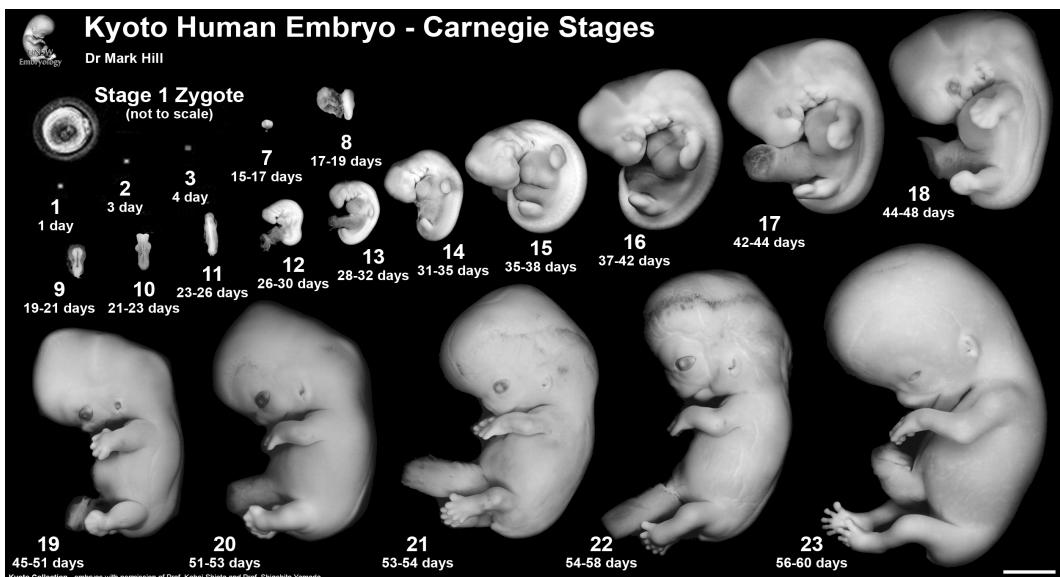


# ANAT 2341

## Embryology



The first 8 weeks of human embryological development.

## COURSE OUTLINE

Trimester 3 - 2019

## **GLOSSARY**

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### **ONLINE TIMETABLE**

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Please read this manual/outline in conjunction with the following pages on the [School of Medical Sciences website:](#)

- [Advice for Students](#)
- [Learning Resources](#)

(or see "STUDENTS" tab at [medicalsciences.med.unsw.edu.au](http://medicalsciences.med.unsw.edu.au) )

# **ANAT2341 Course Information**

## **OBJECTIVES OF THE COURSE**

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The lecture content of this course will provide students with a robust understanding of early human embryogenesis, and the anatomy and development of the major organs and organ systems of the body. Students will also acquire a basic understanding of how major birth abnormalities arise.

In the practical classes, students will actively apply the lecture content by completion of online modules, through modelling of embryonic development using playdough, by digital embryo dissections using online resources, by working with animal models of development in laboratory classes, and in a journal club. Furthermore, students will be exposed to cutting-edge developmental and stem cell biology research presented by experts in the field.

### How the course relates to the Medical Sciences Program

The embryology course is appropriate for a Medical Sciences pathway that includes anatomy, cell biology, histology, and pathology courses, and it prepares for an Honours project in a developmental biology, stem cell or cancer research lab.

### Applications of Embryology in Future Careers

The embryology course prepares graduates for a wide range of careers. Graduates can apply their knowledge of anatomy and developmental biology directly, such as by choosing a career in the biomedical sphere. Some of these include biomedical research scientist, science educator, policy advisor, IVF scientist and forensic scientist. Alternatively, graduates can use the general skills and knowledge acquired to pursue careers in other areas.

## **COURSE CO-ORDINATOR AND STUDENT ADMIN SUPPORT**

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Course Coordinator / Lecturer

- Dr Annemiek Beverdam [A.Beverdam@unsw.edu.au](mailto:A.Beverdam@unsw.edu.au)

Room 234, Wallace Wurth East                    T: +61 2 9385 0019

Students wishing to see the course coordinator should make an appointment *via* email as offices are not readily accessible. We will organize to meet you in a convenient location elsewhere in the building.

For SOMS students administrative matters, please submit enquiries online via UNSW Student Portal Web Forms: <http://unsw.to/webforms>.

Student policy/resource information:

<https://medicalsciences.med.unsw.edu.au/students/undergraduate/advice-students>

For Special Consideration Inquiries: <https://student.unsw.edu.au/special-consideration>

## **COURSE STRUCTURE and TEACHING STRATEGIES**

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Learning activities occur on the following days, times and locations:

- Lectures 1 and 2: Trimester 3, Wednesdays 9-11 am: [Mathews 227](#)
- Practicals: Trimester 3, Wednesdays 11 am – 1 pm: Anatomy Labs, Biosciences Building D26, Level 1 (except in Weeks 4 and 7: teaching labs, Wallace Wurth 120)

Students are expected to attend all scheduled activities for their full duration (2 hours of face-to-face lectures per week, and 2 hours of practical class).

Students are reminded that UNSW recommends that a 6 units-of-credit course should involve about 150 hours of study and learning activities. The formal learning activities are approximately 60 hours throughout the semester and students are expected (and strongly recommended) to do at least the same number of hours of additional study.

### **Lectures:**

Lectures will provide students with the concepts and theory essential for a robust understanding of embryology. Students are encouraged to ask questions or for clarification during the lectures. But also Sli.do events will be created so that students can ask questions anonymously during the lectures and afterwards.

### **Practical classes:**

Practical classes and collaborative learning sessions will help students to revise and consolidate lecture content, to develop their insights in developmental processes in 3D and 4D, and to assist them in their development of research and analytical skills. These classes will allow students to engage in a more interactive form of learning than is possible in the lectures. The skills students will learn in practical classes are relevant to their development as professional scientists.

The 2-hour practical classes will consist of various elements:

1. Each lab will start with a 10-minute quiz to assess knowledge of the previous week's lecture and lab content (part of students' ongoing individual assessment).
2. Each lab, we will take time to revisit the current week's lecture content using questions asked in class, and posted on Sli.do and on Moodle.
3. We will further develop your insight in developmental processes in 3D and 4D using playdough activities, completing SmartSparrow modules and online, by performing virtual embryo dissections using the 3DEmbryo atlas, and by using the online Virtual Human Embryo resource and UNSW Virtual Slides.
4. In some of the labs, guest lecturers will present their developmental biology research on a topic relevant to preceding lecture content.
5. We will have two wet lab practical classes where students will dissect and investigate life chicken embryos, determine the developmental stages, and annotate the structures.

### **Devices:**

Students are expected to bring their own devices (Laptop, MacBook, and/or smart phone with Moodle) to all the practical classes. There are limited loan iPads available in case students do not have their own device. Please contact [Annemiek](#) asap in case you require a loan iPad during the practical classes.

**3D Atlas of Human Development:**

In the practical classes we will be performing digital dissections of human embryos using the [3D Atlas of Human Development](#). This atlas consists of 14 3D-PDF files representing Carnegie stages 7 through to 23. The compressed files are freely available through [this link](#). Please download this 84Mb file at home before the practical classes.

**The Virtual Human Embryo:**

In practical classes we will use the online [Virtual Human Embryo](#) resource. The Virtual Human Embryo Project generated nearly 34 gigabytes of embryonic imagery encompassing all 23 stages of the human embryo. This \$3.2 million, 11-year initiative tapped the world's largest collection of human embryos to identify, digitize, and catalogue some of the best serial sections of normal human embryos ever seen. These images were then reviewed and labeled by one of the leading embryologists of the last half century, and are now available to researchers and educators everywhere.

**Protective Gear in Wet Lab Classes:**

Students should bring lab coats, safety glasses and fully enclosed shoes to the two fertile chicken egg practical classes.

**Revision opportunities:**

The embryology course has significant theory content. Therefore, time has been set apart in the practical classes for course content revision. It is highly recommended that students take advantage of these revision opportunities by asking questions in the lectures, in the practical classes, and on Sli.do and Moodle, and per email.

## COURSE PROGRAM (draft program, subject to changes)

[https://embryology.med.unsw.edu.au/embryology/index.php/ANAT2341\\_Course\\_Timetable\\_2019](https://embryology.med.unsw.edu.au/embryology/index.php/ANAT2341_Course_Timetable_2019)

| Week | Lecture 1<br>Wednesday 9-10 am<br><u>Mathews 227</u> | Lecture 2<br>Wednesday 10-11 am<br><u>Mathews 227</u> | Lab<br>Wednesday 11-1 pm<br><u>AGSM LG06</u> (unless advised otherwise)   |
|------|--|---|---|
| 1    | Introduction   | Fertilization   | No practical class in week 1  |
| 2    | Week 1/2   | Week 3  | Quiz,<br>Smart sparrow (fertilization to implantation)<br>Playdough embryo reconstruction activities<br>The Virtual Human Embryo  |
| 3    | Ectoderm and neural development                      | Mesoderm  | Quiz<br>3DEmbryo dissections: implantation, cavitation, gastrulation, early embryogenesis<br>Playdough embryo reconstruction activities<br>The Virtual Human Embryo<br><br>Guest lecture: Rob Gilchrist |
| 4    | Endoderm, GI, respiratory tracts                     | Research technologies                                 | Location: <a href="#">Wallace Wurth 120 (K-C27-120)</a><br>Quiz<br><br>Fertile egg prac: early embryogenesis<br>Back up activity: Smart sparrow (GI tract)  |
| 5    | Neural crest   | Head  | Midterm exam<br><br>Fabien Delerue – Transgenic technologies  |
| 6    | Heart<br>Lecturer: Nalini Pather                     | musculoskeletal                                       | Quiz<br>3DEmbryo dissections: heart, musculoskeletal<br>The Virtual Human Embryo<br>Smart sparrow (GI tract and Head)<br><br>Guest lecture: Sally Dunwoodie   |
| 7    | placental  | endocrine   | Location: <a href="#">Wallace Wurth 120 (K-C27-120)</a><br>Quiz<br><br>Fertile egg prac: organogenesis  |
| 8    | genital  | renal   | Quiz<br>3DEmbryo dissections: urogenital system<br>Smart Sparrow (sexual differentiation)<br>The Virtual Human Embryo<br><br>Guest lecture: Kirsty Walters  |
| 9    | integumentary  | sensory   | Quiz<br>Smart sparrow exercises (implantation to 8 weeks)<br>The Virtual Human Embryo<br><br>Guest lecture: Stuart Fraser   |
| 10   | Fetal and birth<br>Lecturer: Nalini Pather           | Stem cells  | Quiz<br>Stem cell journal club  |

## **APPROACH TO LEARNING AND TEACHING**

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The learning and teaching philosophy underpinning this course is centred on student learning and aims to create an environment which interests and challenges students. The teaching is designed to be engaging and relevant to prepare students for future careers.

Although the primary source of information for this course is the lecture material, effective learning can be enhanced through self-directed use of other resources such as textbooks and Web based sources, and attendance of the practical classes. It is up to you to ensure you perform well in each part of the course; preparing for classes; completing assignments; studying for exams and seeking assistance to clarify your understanding.

## **TEXTBOOKS AND OTHER RESOURCES**

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These resources will take the form of textbooks, journal articles or web-based resources. Links to resources will be provided in the online Wiki and Moodle.

### **Textbooks:**

There are two embryology textbooks, either of which can be used for this course, both are online accessible through UNSW Library.

1. Moore, K.L., Persaud, T.V.N. & Torchia, M.G. (2015). *The developing human: clinically oriented embryology* (10th ed.). Philadelphia: Saunders.
2. Schoenwolf, G.C., Bleyl, S.B., Brauer, P.R., Francis-West, P.H. & Philippa H. (2015). *Larsen's human embryology* (5th ed.). New York; Edinburgh: Churchill Livingstone.

### **UNSW Embryology wiki pages:**

Content of each of the lectures is summarized on the [online embryology wiki pages](#). This is examinable material together with Annemiek's lecture slides, which are uploaded on the relevant lecture wiki page and on Moodle before each lecture. Please note that only the content of the main lecture page is examinable. Material available through links on the lecture page is provided for interest and will not be examined.

### **3D Atlas of Human Development:**

In the practical classes we will be performing digital dissections of human embryos using the [3D Atlas of Human Development](#). This atlas consists of 14 3D-PDF files representing Carnegie stages 7 through to 23. The compressed files are freely available through [this link](#). Please download this 84Mb file at home before the practical classes.

### **The Virtual Human Embryo:**

In practical classes we will use the online [Virtual Human Embryo](#) resource. The Virtual Human Embryo Project generated nearly 34 gigabytes of embryonic imagery encompassing all 23 stages of the human embryo. This \$3.2 million, 11-year initiative tapped the world's largest collection of human embryos to identify, digitize, and catalogue some of the best serial sections of normal human embryos ever seen. These images were then reviewed and labeled by one of the leading embryologists of the last half century, and are now available to researchers and educators everywhere.

## **STUDENT LEARNING OUTCOMES**

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1. Be able to describe human development, stem cell biology, regenerative medicine and organoid biology, and how major congenital abnormalities arise.
2. Be able to apply basic practical laboratory skills and work with embryo and regeneration models annotate embryonic structures, and define developmental stages.
3. Be able to communicate the contents of primary research articles in the field of stem cell research effectively and appropriately to an audience.
4. Be able to work effectively within a small team to complete academic tasks.
5. Demonstrate critical thinking and problem-solving skills in diverse contexts.
6. Be able to self-manage and work independently with an ability to take responsibility for their own learning, and an appreciation of the value of learning.

## **ASSESSMENT PROCEDURES**

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Assessments:

|  |            |
|--|------------|
| • Individual assessment: weekly quizzes                      | <b>20%</b> |
| • Group project assessment: stem cell journal club (week 10) | <b>10%</b> |
| • Mid-term exam (1-hour duration, week 5)                    | <b>30%</b> |
| • End of session examination (2 hours duration)              | <b>40%</b> |

Exam dates for Term 3, 2019:

- Mid-term exam: Week 5 T3: 16 October 2019
- Final Exam: 29 November – 14 December 2019
- Supplementary exam: 13 – 17 January 2020

## **COURSE EVALUATION AND DEVELOPMENT**

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Each year constructive feedback is sought from students about the course and continual improvements are made based on this feedback.

The “[myExperience](#)” process of UNSW linked through Moodle or student email is the way in which student feedback is evaluated and significant changes to the course will be communicated to subsequent cohorts of students.

Furthermore, at the end of the course, we will also ask your feedback on specific aspects of the course in an independent and anonymous Survey Monkey survey.

Students are also welcomed to provide constructive feedback at any time in person or by email.

Course content, structure and assessment are continuously updated and revised based on the students’ feedback.

## **LECTURE and PRACTICAL OUTLINES**

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The [course timetable](#) is available online and shows references to the relevant textbook chapters for each lecture. Both textbooks are available online through the UNSW Library or as hardcopies.

Practical classes are linked from the online timetable and relate to either the weekly lecture content, specialised research topics or student assessment work.

## **PRACTICAL CLASSES**

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The wet lab practical classes are an opportunity for students to work with and observe live chicken embryos, which allows them to actively apply the knowledge obtained in the lectures. Safety information will be provided at the start of these classes and in the relevant practical class manuals.

Students must take due care with biological and hazardous material. They should wear lab coats, safety glasses and fully enclosed shoes. They have to make sure all equipment is left clean and functional. In the interests of safety, special attention should be paid to any precautionary measures recommended in the notes. If any accidents or incidents occur, they should be reported immediately to the demonstrator in charge of the class who will record the incident and recommend what further action is required.

For more details see [Advice for Students-Practical Classes](#)

## Practical class 1: Preimplantation, early implantation and gastrulation lab

In this practical class, we will do 4 activities that will allow you to apply the knowledge you obtained during the lectures. Annemiek will be available for help, and you can also consult your lecture notes and the internet.

### Activity 1: Preimplantation

Make the following embryonic stages using playdough. Use different colours to represent the different blastomere types/structures:

1. Zygote:
2. Morula:
3. Compacted morula:
4. Blastocyst:

Please complete ‘Activity 3: week 1’ of the online SmartSparrow module accessible via the link for this practical class provided on Moodle.

### Activity 2: Early Implantation and embryonic cavities

Please study the 3D reconstructions and serial H&E stained tissue sections through a [Carnegie stage 3 human embryo](#), [Carnegie stage 4 human embryo](#) [Carnegie stage 5 human embryo](#) and [Carnegie stage 7 human embryo](#).

Please perform digital dissection on a Carnegie stage 7 embryo using a 3D PDF available [here](#).

Please complete ‘Activity 3: week 2’ of the online SmartSparrow module accessible via the link for this practical class provided on Moodle.

### **Activity 3: Gastrulation:**

Make playdough models of the following three developmental stages that illustrate the transformation of the bilaminar embryonic disc into a trilaminar embryonic disc (please only model the embryonic disc, and not the cavities and surrounding membranes). Use different colours to represent the different structures:

1. Bilaminar embryo, excluding amniotic cavity and yolk sac (not the chorionic cavity, etc)
2. Gastrulating embryonic disc
3. Trilaminar embryo

Ensure that the following structures are represented in these models:

- Epiblast
- Hypoblast
- Node
- Primitive streak
- Ectoderm
- Mesoderm
- Endoderm
- Connecting stalk

What embryonic tissues are formed at gastrulation?

Please study the 3D reconstructions and serial H&E stained tissue sections through an implanting [Carnegie stage 7 human embryo](#), [Carnegie stage 9 human embryo](#).

Please perform digital dissection on a Carnegie stage 8 embryo using a 3D PDF available [here \(stage 8\)](#).

Please complete ‘Activity 3: week 3’ of the online SmartSparrow module accessible via the link for this practical class provided on Moodle.

## Practical class 2: Early embryogenesis, ectoderm and mesoderm

In this practical class, we will apply newly acquired knowledge from the lecture content in various activities.

In the second hour, we will listen to a guest lecture by [Professor Robert Gilchrist](#), who is a famous researcher in the School of Women's and Children's Health. He is an expert in IVF and reproductive technology.

### **Activity 1: Virtual human embryo dissections:**

Please open the 3D-PDF files representing Carnegie stages 7 to 16 of the [3D Atlas of Human Development](#). Identify the following features in these files:

#### Stage 7:

Placental lacunae  
Extraembryonic mesoderm  
Chorionic cavity  
Connecting stalk  
Yolk sac  
Hypoblast  
Epiblast, amniotic cavity  
Connecting stalk  
Node

#### Stage 8:

Trilaminar embryo  
Yolk sac  
Connecting stalk  
Allantois  
Ectoderm  
Mesoderm  
Endoderm  
Notochordal process,  
Primitive streak  
Node

#### Stage 9:

Yolk sac  
Embryo  
Amnion  
Neural folds and groove  
Lateral ectoderm  
Mesoderm  
Somites  
Lateral plate mesoderm  
Intermediate mesoderm

## Practical class 2: Early embryogenesis, ectoderm and mesoderm

### Stage 10:

Neural fold fusion  
Neuropores (anterior and posterior)  
Surface epithelium/epidermis covering the neural tube

### Stage 11:

Embryonic epidermis  
Neural tube  
Neuropores  
Somites  
Notochord  
Lateral Plate Mesoderm

### Stage 12:

Embryonic epidermis  
Primary brain vesicles: proencephalon, mesencephalon, rhombencephalon  
Notochord  
Somites  
Mesonephros  
Mesonephric duct

### Stage 15 and 16:

Epidermis  
Secondary brain vesicles: telencephalon, diencephalon, mesencephalon, metencephalon, myelencephalon.  
Developing sclerotomes, mesonephros and mesonephric duct

### **Activity 2: Playdough activity:**

1. Show the process of primary neurulation in 3 developmental steps: neural plate, neural fold development, neural tube closing. Please ensure you show the following features: lateral ectoderm, midplate ectoderm, neural plate, neural fold, neural groove, neuropores.
2. Prepare play dough models displaying the embryonic nervous systems at 3/4 weeks, and at 5 weeks of gestation. Name the structures: proencephalon, telencephalon, diencephalon, rhombencephalon, mesencephalon, metencephalon, myelencephalon, spinal cord, and all the flexures.
3. Prepare a play dough model of early mesoderm development, and show the following features: endoderm, neural tube, surface epithelium, notochord, paraxial mesoderm, intermediate mesoderm, lateral plate mesoderm.

### **Activity 3: The Virtual Human Embryo:**

Investigate the 3D reconstructions and H&E sections of the Carnegie stage 7-16 embryos relevant to this practical class at [The Virtual Human Embryo](#) online resource.

## Practical class 3: Gastrulation, early somitogenesis and neurulation lab



The chicken (*Gallus gallus*) embryo is an excellent model for the study of early vertebrate embryogenesis and later organogenesis. The embryo is encased within a hardened eggshell which provides a natural incubator or culture dish. Through a hole in the eggshell, the embryo can be visualized and easily manipulated with microsurgical tools or gene constructs, then allowed to continue development *in ovo* to determine the consequence of the experimental manipulation.

Fertilized chicken eggs are readily available anywhere in the world and the equipment needed is minimal – a humidified incubator (39°C, no CO<sub>2</sub> required), a dissecting microscope, microsurgical tools that can be prepared in the lab or purchased, and either a hand-held mouth pipette or a manufactured micromanipulator and picospritzer.

Fertilized eggs can be held at between 13-16°C for up to 1 week prior to incubation. They are incubated at 38°C-39°C to the desired stage in a humidified incubator with the eggs placed on their side (horizontal). For long-term post-operative survival, it is best that the eggs be left in the incubator until experimental manipulation. However, eggs can be removed from the incubator and held at room temperature to slow development.

For staging of chicken embryos:

[https://embryology.med.unsw.edu.au/embryology/index.php/Hamburger\\_Hamilton\\_Stages](https://embryology.med.unsw.edu.au/embryology/index.php/Hamburger_Hamilton_Stages)

### SAFE WORKING PROCEDURES AND ANIMAL ETHICS

#### Risks Associated with Practical

Eggs have the potential to be contaminated with the bacteria *Salmonella*. Wear gloves throughout and students should always wash their hands before leaving the lab.

Dissection implements are sharp, so students should take care not to cut themselves or other students.

Students should wear a lab coat, gloves and enclosed shoes to protect themselves from egg splatter.

#### Animal Ethics Compliance

The procedures used in this practicum are in compliance with the UNSW Animal Care and Ethics Committee and the National Health and Medical Research Council ‘Australian code of practice (8<sup>th</sup> edition, 2013).

## Practical class 3: Gastrulation, early somitogenesis and neurulation lab

### EXPERIMENTAL PROCEDURES

#### Opening of the egg

1. Eggs were incubated for several days at 38.5°C to generate embryos that undergo organogenesis.
2. Each student pair should have one egg and an egg holder, a pair of blunt forceps and a pair of scissors, 3 petridishes, PBS, disposable pipet, syringe and 23G needle, Indian ink solution, dissection microscope.
3. Put the egg into the holder with the blunt end up (pointed end down).
4. Use the pointy end of the scissors to tap a hole in the top of the egg into the air chamber, be very careful not to push the scissors too far into the egg.
5. Use the forceps to pick bits of shell out. Do not remove egg shell beyond the air chamber. You will see the air chamber and the vitelline membrane.
6. Carefully remove the vitelline membrane at the top.
7. If you do not see the embryo (a ring of blood vessels should be visible) then gently swirl the egg so that it floats up to the top of the yolk. If this doesn't work you will need to take another egg if available.
8. Use the forceps to break off pieces of shell down to the yolk so that the embryo (visible as a ring of blood vessels) is exposed and the rim of the shell is just above the surface of the egg white or albumen.
9. You may be able to see the heart beating without magnification. If not, then put the egg under the dissection microscope.

#### Early stage somitogenesis embryos

1. Draw Indian Ink solution (if available) up into a 1 ml syringe fitted with a 23G syringe needle.
2. Open an egg as described above. Once the shell has been removed down to the level of the yolk and the vitelline membrane has been removed, slide the syringe needle under the
3. embryo. It is easiest to insert the syringe needle vertically at the edge of the egg initially and then rotate the needle until it is almost horizontal using the edge of the egg shell as a support. The tip of the needle should end up just below the embryo in the centre of the egg.
4. Slowly inject the Indian ink solution to reveal the embryo and view under a dissecting microscope.

## Practical class 3: Gastrulation, early somitogenesis and neurulation lab

5. Identify the HH stage and the diverse embryonic structures. See last page or link:  
[https://embryology.med.unsw.edu.au/embryology/index.php/Hamburger\\_Hamilton\\_Stages](https://embryology.med.unsw.edu.au/embryology/index.php/Hamburger_Hamilton_Stages)
6. Draw your embryo, indicate the Hamburger & Hamilton (HH) developmental stages using the guide on course manual pages 22-24, and annotate the embryonic structures that you have identified.
7. Hand your drawing in at the end of the prac.

### Observation of developmental stages

Move around the class to study the embryonic chicken stages of your colleagues. Identify the following structures:

- Amniotic sac
- Hensen's node
- Neural groove and folds
- Head ectoderm
- Somites
- Brain vesicles
- Cardiogenic mesoderm
- Heart and vasculature

### Tidying up after the prac

- Please discard egg waste and disposable consumables into the yellow bins (not the paper bins).
- Dispose of Syringes and needles in sharps bins
- Wash forceps and scissors with water, and wipe with ethanol, place in provided containers on the sink.
- Clean surfaces (microscope stages, benches) with water and ethanol, and wipe dry.

### Back up activity: Smart Sparrow exercises

Please complete the online SmartSparrow exercises on GI tract development via the link for this practical class provided on Moodle.

## Practical class 4: Virtual dissections of organogenesis embryos

In the first hour of this practical class, we will complete the mid-term exam.

In the second hour, we will listen to a guest lecture by [Fabien Delerue](#), who is the Head of the Genome Editing at Macquarie (GEM), a transgenic core developed in joint venture by the Dementia Research Centre (DRC) and the Macquarie Animal Research Services (MARS). His research focuses on the production and characterization of new transgenic mouse models of human genetic disorders, using cutting-edge genome engineering techniques such as CRISPR/Cas9. He will discuss recent advances in transgenic technology in his guest lecture today.

## Practical class 5: Virtual dissections of organogenesis embryos

In this practical class, we will perform virtual embryo dissections to witness development of the musculoskeletal system and of the heart.

In the second hour we will listen to a guest lecture by Professor Sally Dunwoodie.

### **Virtual human embryo dissections:**

Please open the 3D-PDF files representing Carnegie stages 8 to 23 of the [3D Atlas of Human Development](#) that are freely available through [this link](#). Please download this 84Mb file at home before the practical classes.

Identify the following features in these files and track how they develop over time:

#### Heart development:

- Fusing primary heart tubes
- Looping of the heart tube
- Atria
- Ventricles
- Outflow tract
- Ductus venosus
- Ductus arteriosus
- Dorsal aorta: note how these fuse over time

#### Musculoskeletal system

- Mesoderm in the trilaminar embryo
- Somites: note an increase in somites over time
- Notochord: what role does it play in somite development?
- Sclerotome differentiation (note that cranial somites are ahead in development)
- Development of the axial skeleton
- Development of the appendicular skeleton
- Development of the skull
- Intervertebral disks

Please compare these 3D-PDFs with the 3D reconstructions available on [The Virtual Human Embryo](#) resource.

### **Smart Sparrow exercises**

Please complete the online SmartSparrow exercises on cranial development via the link for this practical class provided on Moodle.

## Practical class 6: Organogenesis lab

### SAFE WORKING PROCEDURES AND ANIMAL ETHICS

#### Risks Associated with Practical

Eggs have the potential to be contaminated with Salmonella. Wear gloves throughout and students should wash their hands before leaving the lab.

Dissection implements are sharp, so students should take care not to cut themselves or other students.

Students should wear a labcoat, gloves and enclosed shoes to protect themselves from egg splatter.

#### Animal Ethics Compliance

The procedures used in this practicum are in compliance with the UNSW Animal Care and Ethics Committee and the National Health and Medical Research Council “Australian code of practice” (8<sup>th</sup> edition, 2013).

### EXPERIMENTAL PROCEDURES

#### Opening of the egg

8. Eggs were incubated for several days at 38.5°C to generate embryos that undergo organogenesis.
9. Each student pair should have one egg and an egg holder, a pair of blunt forceps and a pair of scissors, 3 petri dishes, PBS, disposable pipet, syringe and 23G needle, Indian ink solution, dissection microscope.
10. Put the egg into the holder with the blunt end up (pointed end down).
11. Use the pointy end of the scissors to tap a hole in the top of the egg into the air chamber, be very careful not to push the scissors too far into the egg.
12. Use the forceps to pick bits of shell out. Do not remove egg shell beyond the air chamber. You will see the air chamber and the vitelline membrane.
13. Carefully remove the vitelline membrane at the top.
14. If you do not see the embryo (a ring of blood vessels should be visible) then gently swirl the egg so that it floats up to the top of the yolk. If this doesn't work you will need to take another egg if available.

## Practical class 6: Organogenesis lab

15. Use the forceps to break off pieces of shell down to the yolk so that the embryo (visible as a ring of blood vessels) is exposed and the rim of the shell is just above the surface of the egg white or albumen.
16. You may be able to see the heart beating without magnification. If not, then put the egg under the dissection microscope.

### Organogenesis embryos

1. Carefully tear a hole in the vitelline membrane.
2. Very carefully tilt egg contents in a large petri dish, holding the egg very close to the dish.
3. Find and dissect the embryo, transfer it to a clean petri dish and cover it with PBS.
4. Study embryo under a dissection microscope, determine Hamburger & Hamilton (HH) developmental stages using the guide on course manual pages 22-24, and identify the embryonic structures.
5. Draw your embryo, indicate the developmental stage (HH, pages 22-24), and annotate the embryonic structures that you have identified.
6. Hand your drawing in at the end of the prac

### Observation of developmental stages

Move around the class to study the embryonic chicken stages of your colleagues. Identify the following structures:

- Somites
- Brain vesicles
- Heart and vasculature
- Optic vesicles
- Branchial arches
- Nasal placodes
- Nasal pits and nasal processes
- Otic vesicle
- Limb buds
- Autopod, stylopod and zeugopod
- Handplate and fingers

## Practical class 6: Organogenesis lab

### E. Mouse and chicken embryogenesis

There will be one demonstration station with dissection and light microscopes where you can investigate mouse embryos from E7.5 (late gastrula) until late organogenesis (E15.5), skeletal preparations of mouse foetuses, and mounted and stained early stage chicken embryos.

Identify the following structures:

- Amniotic sac
- Hensen's node
- Neural groove and folds
- Head ectoderm
- Somites
- Brain vesicles
- Cardiogenic mesoderm
- Heart and vasculature
- Optic vesicles
- Branchial arches
- Nasal placodes
- Nasal pits and nasal processes
- Otic vesicle
- Limb buds
- Autopod, stylopod and zeugopod
- Handplate and fingers

Note that mouse foetuses mainly consist of cartilage (blue), and only some skeletal elements are ossified (red) including the calvaria.

### F. Placentation, Head and GI Tract Exercises

If there is time you may want to try the Placentation, Head, and/or GI tract modules on Smart Sparrow via the links provided on Moodle.

### G. Tidying up after the prac

- Please discard egg waste and disposable consumables into the yellow bins (not the paper bins).
- Dispose of Syringes and needles in sharps bins
- Wash forceps and scissors with water, and wipe with ethanol, place in provided containers on the sink.
- Clean surfaces (microscope stages, benches) with water and ethanol, and wipe dry.

## STAGING CHICK EMBRYOS - HAMBURGER HAMILTON STAGES

| HH Stage   | Time and Features  |
|------------|--|
| 2          | 6-7 hr: Initial primitive streak, 0.3-0.5 mm long  |
| 3          | 12-13 hr: Intermediate primitive streak  |
| 4          | 18-19 hr: Definitive primitive streak, $\pm 1.88$ mm long  |
| 5          | 19-22 hr: Head process (notochord)   |
| 6          | 23-25 hr: Head fold  |
| 7          | 23-26 hr: 1 somite; neural folds   |
| 7 to 8-    | ca. 23-26 hr: 1-3 somites; coelom  |
| 8          | 26-29 hr: 4 somites; blood islands   |
| 9          | 29-33 hr: 7 somites; primary optic vesicles  |
| 9+ to 10-  | ca. 33 hr: 8-9 somites; anterior amniotic fold   |
| 10         | 33-38 hr: 10 somites; 3 primary brain vesicles   |
| 11         | 40-45 hr: 13 somites; 5 neuromeres of hindbrain  |
| 12         | 45-49 hr: 16 somites; telencephalon  |
| 13         | 48-52 hr: 19 somites; atrioventricular canal   |
| 13+ to 14- | ca. 50-52 hr: 20-21 somites; tail bud  |
| 14         | 50-53 hr: 22 somites; trunk flexure; visceral arches I and II, clefts 1 and 2                              |
| 14+ to 15- | ca. 50-54 hr: 23 somites; premandibular head cavities  |
| 15         | 50-55 hr: 24-27 somites; visceral arch III, cleft 3  |
| 16         | 51-56 hr: 26-28 somites; wing bud; posterior amniotic fold   |
| 17         | 52-64 hr: 29-32 somites; leg bud; epiphysis  |
| 18         | 65-69 hr: 30-36 somites extending beyond level of leg bud; allantois                                       |
| 19         | 68-72 hr: 37-40 somites extending into tail; maxillary process   |
| 20         | 70-72 hr: 40-43 somites; rotation completed; eye pigment faint grey  |
| 21         | 3.5 da: 43-44 somites; pharyngeal arch IV, pharyngeal cleft 4; eye pigment faint                           |
| 22         | 3.5 da: Somites extend to tip of tail; eye pigmentation distinct   |
| 23         | 3.5-4 da: Dorsal contour from hindbrain to tail is a curved line   |
| 24         | 4 da: Toe plate in leg-bud distinct; 4th pharyngeal cleft reduced to small pit                             |
| 25         | 4.5 da: Elbow and knee joints; 3rd & 4th pharyngeal clefts reduced to small pits                           |
| 26         | 4.5-5 da: 1st 3 toes; 3rd and 4th pharyngeal clefts no longer visible                                      |
| 27         | 5 da: Beak just barely recognisable  |
| 28         | 5.5 da: 3 digits, 4 toes; beak outgrowth distinct in profile   |
| 29         | 6 da: Rudiment of 5th toe; beak more prominent; no egg tooth   |
| 30         | 6.5 da: Feather germs; scleral papillae; egg tooth   |
| 31         | 7 da: Web between 1st and 2nd digits; feather germs dorsally continuous from brachial to lumbosacral level |
| 32         | 7.5 da: Anterior tip of mandible has reached beak  |
| 33         | 7.5-8.0 da: Web on radial margin of wing and 1st digit   |
| 34         | 8 da: Nictitating membrane   |
| 35         | 8 - 9 da: Phalanges in toes  |

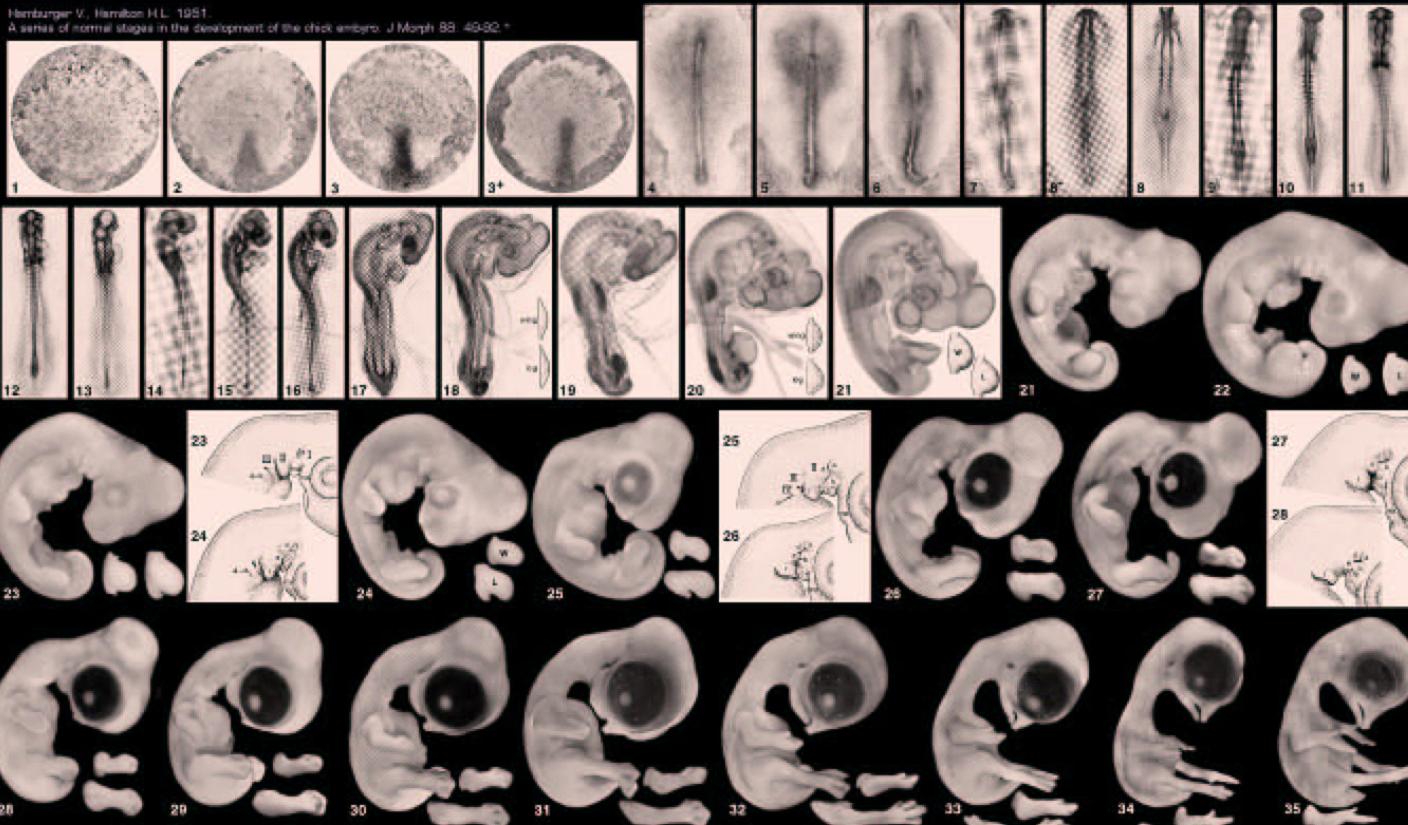
|    |  |
|----|--|
| 36 | 10 da: Length of 3rd toe from tip to middle of metatarsal joint = $5.4\pm0.3$ mm; length of beak from anterior angle of nostril to tip of bill = 2.5mm; primordium of comb; labial groove; uropygial gland |
| 37 | 11 da: Length of 3rd toe = $7.4\pm0.3$ mm; length of beak = 3.0 mm   |
| 38 | 12 da: Length of 3rd toe = $8.4\pm0.3$ mm; length of beak = 3.1 mm   |
| 39 | 13 da: Length of 3rd toe = $9.8\pm0.3$ mm; length of beak = 3.5 mm   |
| 40 | 14 da: Length of beak = 4.0 mm; length of 3rd toe = $12.7\pm0.5$ mm  |
| 41 | 15 da: Length of beak from anterior angle of nostril to tip of upper bill = 4.5 mm; length of 3rd toe = $14.9\pm0.8$ mm  |
| 42 | 16 da: Length of beak = 4.8 mm; length of 3rd toe = $16.7\pm0.8$ mm  |
| 43 | 17 da: Length of beak = 5.0 mm; length of 3rd toe = $18.6\pm0.8$ mm  |
| 44 | 18 da: Length of beak = 5.7 mm; length of 3rd toe = $20.4\pm0.8$ mm  |
| 45 | 19-20 da: Yolk sac half enclosed in body cavity; chorio-allantoic membrane contains less blood and is "sticky" in living embryo  |
| 46 | 20-21 da: Newly-hatched chick  |

## STAGING CHICK EMBRYOS - HAMBURGER HAMILTON STAGES

In Memory of **Viktor Hamburger: 1900-2001**

NORMAL STAGES OF CHICK EMBRYONIC DEVELOPMENT

Hamburger V., Hamilton H.L. 1951.  
A series of normal stages in the development of the chick embryo. J Morph 88: 49-92. \*



The stage series of normal chick embryonic development is one of the most frequently cited and enduring biological articles ever published, and set the standard for various species-specific stage series that have followed. The coauthor of this paper, Viktor Hamburger, died in June, 2001, just a few weeks prior to his 100th birthday. In recognition of the tremendous importance of this stage series to countless research and teaching laboratories around the world, and in tribute to Prof. Hamburger's many contributions to developmental biology spanning over decades, the editors and publisher of *Developmental Dynamics* commissioned the poster that accompanies this issue. Stages 1-15 (incubation days through 9) were selected from the original 1951 publication. Embryos have been restored and rescaled for this poster format, and digitally enhanced to better highlight key features of each stage. Schematics of early limb bud and forelimb (pharyngeal) arch development are also included; keys for these are in the original publication.

\*Reprinted in Developmental Dynamics 195: 281-72 (1992).

Poster by Drew M. Noden, Cornell University, sponsored by the American Association of Anatomists; production by Wiley-Liss, Inc.

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## Practical class 7: Development of the Urogenital System

In this practical class, we will perform virtual embryo dissections of the developing urogenital system. Furthermore, we will work through SmartSparrow modules to test your knowledge about urogenital development.

In the second hour we will listen to a guest lecture by [Dr Kirsty Walters](#), who is a Senior Lecturer at Women's and Children's Health, and research leader in the field of female reproduction and ovarian function.

### **Virtual human embryo dissections:**

Please open the 3D-PDF files representing Carnegie stages 8, and 12 through to 23 of the [3D Atlas of Human Development](#) that are freely available through [this link](#). Please download this 84Mb file at home before the practical classes.

Identify the following features in these files and track how they develop over time:

Mesoderm  
Intermediate mesoderm/mesonephros  
Mesonephric duct  
Paramesonephric duct  
Ureteric bud  
Metanephros  
Renal pelvis  
Ureter  
Urethra  
Urinary bladder  
Cloaca and urogenital sinus  
Genital tubercle  
Genital folds  
Genital swellings  
Kidney location

Can you determine the sex of the embryos of stages CS21 and CS23?

Please compare these 3D-PDFs with the 3D reconstructions available on [The Virtual Human Embryo](#) resource.

### **SmartSparrow Exercises**

Please complete the online SmartSparrow exercises on the development of the urogenital system via the link for this practical class provided on Moodle.

## Practical class 8: Revision of Systems Development

This week we will take the opportunity in the first hour to revise all content of systems development, roughly from week 3 until week 8.

In the second hour, [A/Prof Stuart Fraser](#) will present a lecture on blood cell development and on his research into this field.

### **Activity 1: System Development Exercises**

Please complete the online SmartSparrow exercises via the link for this practical class provided on Moodle.

### **Activity 2: Virtual human embryo dissections:**

Please open the 3D-PDF files representing Carnegie stages 10 through to 23 of the [3D Atlas of Human Development](#) that are freely available through [this link](#). Please download this 84Mb file at home before the practical classes.

### **Activity 3: Histology and 3D reconstructions**

Please investigate the histology and 3D reconstructions of human embryos Carnegie stages 10 through to 23 available at the online [Virtual Human Embryo](#) resource.

## Practical class 9: Journal Club Presentation

As part of the assessment for this course, your group will give a 10-15 minutes journal club presentation (including 3 minutes question time). Exact time will be dependent on number of groups, and will be advised closer to the date.

This assignment will allow you to apply the knowledge that you acquired in the stem cell lecture, and it will teach you to search journal databases, to become familiar with the research field of stem cell biology, and to improve your presentation skills.

You will discuss a recent (published after 2014) original research article (not a review!) on stem cell biology or technology.

Please send the PDFs of 2-3 articles to Annemiek ([A.Beverdam@unsw.edu.au](mailto:A.Beverdam@unsw.edu.au)) at least a week before this practical class. Annemiek will judge suitability and select the best article for the journal club.

Please note that the best articles are found in journals with the highest impact factors: Nature, Science, Cell, Cell Stem Cell, Stem Cell Reports, etc). Please contact Annemiek in case you are at a loss, and she will help you find one.

During the presentation, it works best if one student discusses the introduction, a second the results section, and a third the discussion section. Please note that one slide takes about 1 minute to talk through. So do not use more than 15 slides total. Please read through the tips below on how to prepare a good presentation.

Each other group will ask at least one question following a journal club presentation.

Your group will receive the same mark, which will contribute to your individual assessment. This mark will be based on:

- insight and comprehension,
- presentation and slide style,
- engagement in question time and discussion,
- keeping within time.

All students of the group are required to contribute to developing and/or presenting the group presentation. If a student fails to do so, this will result in a penalty of the final mark. Please contact Annemiek per email if you feel that a group member did not contribute. She will deal with this confidentially.

Good luck and have fun!

## Presentation Hints for Student

1. Keep your presentation short and concise. Not every detail of the article needs to be discussed in the presentation, but limit it to the bare minimum that is required to get the main message of the article across. For instance, do not go into too much detail in method sections. Not all nitty-gritty detail of the results needs to be discussed. The less info your audience has to take in, the higher the chance that they will understand your story.

2. Split the presentation up in three parts: introduction, Results and Discussion. Do not talk through the Material and Methods Sections separately!

- **Introduction** equips the audience with the information required to understand the research, and supports the research hypothesis and research questions addressed in the article.

- The **results** section consists of multiple sections. Talk through each of these sections in the following sequence: 1. research question asked, 2. assays and methods used to address this question, 3. experimental outcomes, 4. conclusions. - The **discussion** summarizes and interprets the outcomes, discusses the shortfalls, places the results in the larger context of the research fields, discusses the implications of the data for human disease, and issues raised by the findings and future experiments that may resolve these questions.

3. Use mostly figures and very limited text on your slides. Make sure that you know and understand what you want to get across. Do not use cheat sheets and do not learn your presentation literally by heart. Explain carefully. Use your slides as cheat sheets. Make eye contact with your audience and get a feel for whether they understand your story.

4. Talk your audience carefully through each of the slides and engage with the slides and with your audience to gauge their understanding. Slides are an indispensable part of the presentation. Each item on your slides should be relevant and addressed and highlighted with pointer, fingers, stick. Slide shows are indispensable for a presentation, as is the presenter. They should support and enhance a presentation, they should aid your audience in understanding.

5. Talk your audience through each of the figures on your slides. Figures may be obvious to you, but not to your audience unless you explain them carefully. So explain what experiment has been carried out, and what is displayed in the figure:

on the X and Y-axes  
what the bars represent in diagrams  
the tissues/cell types displayed  
the bands on Western blot, RNA and DNA gels,  
What colors represent colors in immunostainings, etc etc.

6. Please note that you only need to highlight this experimental detail that is necessary to get the main message of the figure across.

7. Annotate the figures in your presentation carefully but sparingly. Label panels, axes, images etc so that figures are self-explanatory.

8. To stay in control the presenter should flick through the slide show. Not another member of the team.

9. If you didn't understand the articles in depth, read a recent review or even go back to text books to acquire the basic knowledge. Also, if you discuss results of a crucial experiment but do not understand the technology. Please go back to the original references or your text books to read up on this technology. You should be on top of everything you say or write up in your slides.

10. Stick to your time. Don't make too many slides. Each slide should take about a minute on average to talk through. Try to avoid acronyms and abbreviation.

## HEALTH & SAFETY GUIDELINES

Generic safety rules for UNSW can be found at: [SAFETY.UNSW.EDU.AU](http://SAFETY.UNSW.EDU.AU) and for the School of Medical Sciences at [MEDICALSCIENCES.MED.UNSW.EDU.AU/STAFF/HEALTH-SAFETY](http://MEDICALSCIENCES.MED.UNSW.EDU.AU/STAFF/HEALTH-SAFETY)

Additional safety information will be provided for classes at other locations.

ScienceTeaching Laboratory

Student Risk Assessment

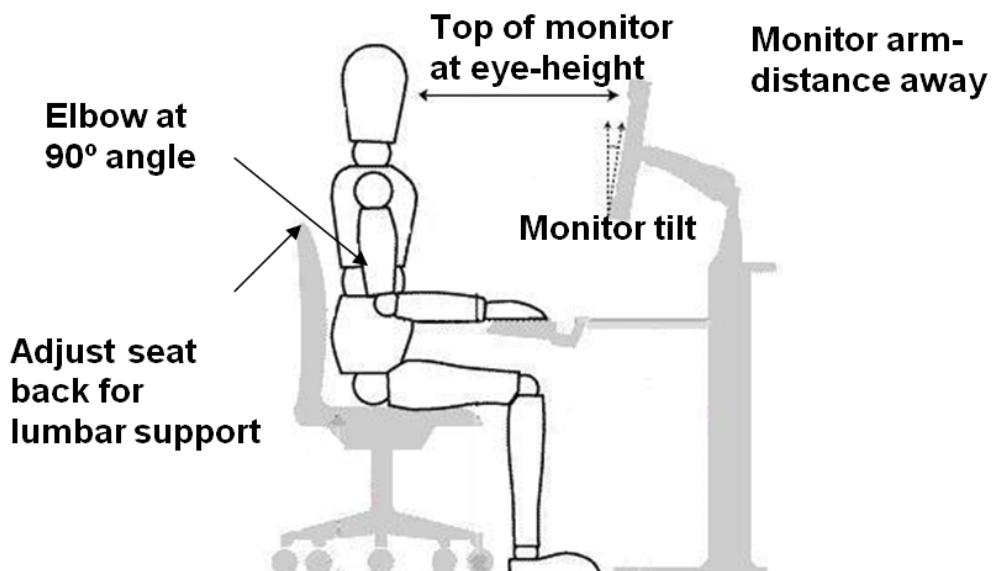


ANAT2341

Wallace Wurth East G6-7  
Semester 2, 2018.

### Workstation set-up

|            |                       |  |
|------------|-----------------------|--|
| Ergonomics | Musculoskeletal pain. | Correct workstation set-up.  |
| Electrical | Shock/fire            | Check electrical equipment in good condition before use. All electrical equipment tested and tagged. |



### Personal Protective Equipment

Not necessary in these practicals. (specimens are fully sealed)

### Emergency Procedures

In the event of an alarm, follow the instructions of the demonstrator. The initial sound is advising you to prepare for evacuation and during this time start packing up your things. The second sound gives instruction to leave. The Wallace Wurth assembly point is the lawn in front of the Chancellery. In the event of an injury, inform the demonstrator. First aiders contact details and Kit locations are on display by the lifts.

### Clean up and waste disposal

No apparatus or chemicals used in these practicals.

### Declaration