1.0. Aims

1. To develop an understanding of acute inflammation as a major pathological process involved in the response to injury and infection.
2. To understand how the cells involved in innate and adaptive immunity originate in the bone marrow, travel through the blood and migrate into the tissues.
3. To recognise the neutrophil as the key cell of acute inflammation.
4. To develop the skills of description and interpretation of pathological changes in tissue sections, with emphasis on the identification of pathological processes.

2.0. Introduction

Nature has evolved a variety of sophisticated and subtle processes by which danger to the survival of an organism is recognised, challenged and, hopefully, overcome. These processes constitute two overlapping sets of reactions and their effects and are called innate and adaptive immunity (immunity means protection). Innate immunity is an immediate, albeit relatively non-specific response to danger and is elicited by tissue damage, e.g. at a site of injury or at a focus of infection. The reaction is termed inflammation.

2.1. Acute Inflammation

Acute inflammation is the local response elicited by tissue damage. A characteristic feature is leakage of blood proteins into the tissue (predominantly fibrinogen, which is quickly converted to fibrin) and the recruitment of leukocytes from the blood. Together these constitute an inflammatory exudate. The first leukocytes to be recruited are neutrophils (NPLs), also called polymorpho-nuclear leukocytes (PMNLs) and these are characteristic cellular markers of acute inflammation.

The central aim of today's practical is to recognise the characteristic histological features (footprint) of acute inflammation by the identification of neutrophils and fibrinous exudate in pneumonia (acute inflammation in the lung).

As time passes following acute injury other leukocytes are recruited into the tissue, e.g. lymphocytes and monocytes and these give rise to the characteristic pattern of chronic inflammation. These features, along with the processes of organisation of dead tissue and of repair will be dealt with in later classes.

It should be remembered that all of the leukocytes that enter tissues in any form of inflammation are derived from the pool of circulating cells in the blood. The predominant leukocyte in normal blood is the neutrophil. It is readily recognisable by a poly-lobated nucleus. When it enters the tissue this characteristic morphology is still recognisable, but, as the cells begin to die, the poly-lobated structure of the nucleus is less obvious (why might this be?).

Neutrophils in Tissue: Image Map: N_CS_NU_09
3.0. Cells of the haemopoietic system:

Although most of the classes that deal with the appearance in disease are based around tissue sections, cells can also be visualised in films or smears of biological fluids. Seen this way, whole cells rather than a section through them are represented and it is often easier to recognise details of cellular structures such as the nucleus. Before studying neutrophils in a tissue section, therefore, it is useful to identify them in a blood film (also look at the demonstration boards which detail the origins of these cells and their development).

**Look at the thin end of the film where the cells are well spread out.**

Try to identify:

(1) Red blood cells (RBCs; erythrocytes) which are round, anucleate, and have a pale biconcave centre

(2) Leukocytes, the predominant cell being the neutrophil.

**Q1** What are the other cell types present in the blood?
4.0. Acute Inflammation of the lung: Pneumonia

You are provided with sections of normal lung for reference:

**NDP Image: Normal Lung - CR8: 70.16A**
**Glass slide: CR8: 70.16A**
**Image Map: N_CR_LU_02**

and two cases of acute bronchopneumonia. Look for evidence of an acute inflammatory exudate, noting the large number of neutrophils and also the presence of fibrin.

**Glass Slide: 2.1: 80.226**
**Image Map: A_AI_BP_LU_01**

This tissue section was taken from a 30 year-old woman who died after an attack of acute bronchopneumonia.

Note the following features (in comparison to the healthy lung):

- In the centre of this section, viewed with the naked eye, there is a dark, Y-shaped structure - it is a longitudinal section through a bronchus at a branch-point. Is this the normal appearance of a bronchus? Which cells fill the bronchus?

- Note the patchy appearance of the alveolar spaces. Which cells fill these spaces? What are their key features?

- Large areas look red because of vasodilatation and haemorrhage. Are the red blood cells contained within vessels (thin walls)? What causes haemorrhage?

- The bronchial epithelium has become detached in places. Why?

- Note the pale pink homogeneous material that is fluid exudate containing protein (particularly fibrin) in some alveoli. Why might this impair breathing?

The patchy pattern of inflammation involves the bronchi and the adjacent lung parenchyma; it is therefore called ‘Bronchopneumonia’

[Note: black granules of inhaled carbon, often around the bronchi and within the lung macrophages which is normal]

4.2. Lobar pneumonia – NDP Image: 2.2: 65.140; 62.38
**Glass Slide: 2.2: 65.140; 62.38**

This tissue section was taken from a 53 year old homeless person who died, having been breathless for a few days, and who had not sought any medical attention.

Note the features in comparison to the case of bronchopneumonia (4.1):

- Diffuse infiltration contrasting with the patchiness of bronchopneumonia (notice that although all the alveoli contain an inflammatory exudate, the alveolar walls are intact)
- Fibrin within the alveoli appears as delicate threads
• Fibrin over the pleura becomes compacted as a result of respiratory movements (fibrinous pleurisy)

**Lobar pneumonia** refers to a rapidly spreading inflammation which can develop as a result of infections by bacteria, particularly those which have thick capsules, e.g. pneumococci, which have polysaccharide capsules. The lung parenchyma quickly fills with proteinaceous fluid and leukocytes and when the infection reaches the pleura, this in turn becomes inflamed and fibrinogen leaks from blood vessels onto the surface.

**Museum Specimens**

**Lung: Lobar Pneumonia - 21.53**
The lower and middle lobes are affected with a fibrinous membrane coating the pleura. No clinical details available.

**Lung: Lobar Pneumonia - 28.205**
The upper and part of the lower lobe are affected and there is extensive pleurisy. A case of pneumococcal pneumonia and septicaemia which developed in a 34 year old woman following childbirth.

4.2. **Lobar pneumonia – special stain (Trichrome)**

NDP Image: 2.3: **62.38 & 56.36**
Glass Image: 2.3: 62.38
This special stain is a mixture of three dyes and is used to stain **fibrin scarlet**. (Look for the red network in the air spaces). **Connective tissue** and other elements are **blue**; red blood cells vary in colour, from orange to yellow.

5.0. Questions

Discuss with your colleagues, and eventually with a demonstrator, the answers to the following:

Q2 Where in the body are neutrophils formed? What route did the neutrophils take to reach the lumen of the bronchus?

Q3 How long do leukocytes remain in the blood? What happens to them eventually?

Q4 What would their future have been, had the tissue not been harvested?

Q5 Are there deleterious as well as beneficial consequences of this degree of neutrophil extravasation and activation?

Q6 How do you suppose a viral infection (such as influenza) of the bronchial epithelium may increase susceptibility to bacterial bronchopneumonia?

Now build up a picture of how tissue reactions to injury are initiated and how they produce effects that are recognised as symptoms (i.e. the things that the patient identifies) and the signs (i.e. the things that can be demonstrated objectively by clinical examination or various imaging methods). Fill in the empty boxes in the following flow chart, indicating the factors responsible for the progression from one stage to the next:
viral infection

(1)

bacterial infection

(2)

acute inflammation

(3) blood-stained sputum

(4) purulent sputum

(5) fever

(6) rapid shallow respiration
6.0. Writing a report: Interpretation of pathological changes in tissues and identification of pathological processes.

The aim of this section of the class is to learn how to interpret a histological section to reveal the pathological processes and how to write a concise report describing the condition. In future classes you will be provided with a series of slides of unknown conditions and asked to write a report along the following lines. You are briefly reminded of the principles of how to look at tissue sections, which were given in the last class and then given a guide to writing a report.

6.1. Looking at tissue sections

1. Look with the **naked eye** first for distinct areas or shapes.
2. Use the **low power objective** to scan the section getting an overall impression of the different areas.
3. Using a **higher power** objective (x10, then x40) to home in on representative areas of normal or abnormal looking regions. **Identify cellular and other details**. Return to low power to see the whole picture.
4. Remind yourself about what the **normal tissue** should look like, at the **edge** of the section.

6.2. Describing and interpreting pathological changes in tissues

An essential preliminary to interpreting pathological changes in tissues is to observe and describe the topography of the tissue, i.e. the pattern of cellular abnormalities and the specific cellular appearance. It is helpful, therefore, to draw a simple schematic diagram and to point out, by labels, the areas of interest, which you wish to describe. Having described the key pathological features you can go on to offer an interpretation of the pathological processes and give an opinion of the nature of the disease and its possible aetiology.

(1) Draw a **sketch diagram** of the whole section and where applicable a more detailed diagram of a representative abnormal area to show the cell types present etc. As with everywhere else, all diagrams must have **headings** and clear **labels** with no ambiguous unlabelled structures.

(2) Give a **description**. Referring to the above diagram, describe the distribution of the abnormality and whether there is any remaining normal tissue. Within the area of abnormality, describe the architecture of the tissue, which cell types are present and then whether these cells appear normal.

(3) **Give an interpretation, identifying the pathological process.** Interpret the features you have described in terms of the likely processes occurring e.g., acute or chronic inflammation, granuloma formation, thrombosis, infarction, neoplasia. You may not be able to give a single overall diagnosis. However, do so if you can. The most important part is the **identification of the pathological process** (e.g. acute inflammation).
6.3. Cerebrum: unknown condition

NDP Image: 2.4: 66.315
Glass Slide: 2.4: 66.315

Description:

Slide 66.315 is a section of cerebrum in which the sub-arachnoid space is expanded by a dense infiltrate of cells, mostly neutrophils, but also a few large mononuclear cells, probably macrophages. The cells are within a meshwork of fine threads, which are probably fibrin. (This could be confirmed by using the trichrome stain which specifically stains fibrin scarlet). The blood vessels are dilated. The underlying cerebrum appears normal.

Museum Specimens

Brain: Acute purulent meningitis - P63.377
A 66 year old man developed pneumonia, septicaemia (bacterial infection of the blood) and meningitis after a partial gastrectomy, an operation to remove a diseased part of the stomach. Why do you think each of these infections came about?

Brain: Pneumococcal meningitis - 27.28
From a 13 year old girl who had been ill for 4 days with fever, headache and vomiting. Pneumococci were cultured from cerebro-spinal fluid. Pale, purulent exudate fills the sulci and there is generalised vasodilatation. The ventricular system is not affected by the meningitis.
### 7.0. Some words used today

<table>
<thead>
<tr>
<th>Leukos</th>
<th>White</th>
<th>Leukocyte</th>
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<tbody>
<tr>
<td>Cytos</td>
<td>Cell</td>
<td></td>
</tr>
<tr>
<td>Phagein</td>
<td>To eat</td>
<td>phagocyte</td>
</tr>
<tr>
<td>Philos</td>
<td>Loving</td>
<td>Eosinophil (avid for Eosin, an acid stain)</td>
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<tr>
<td></td>
<td></td>
<td>basophil (avid for basic stains)</td>
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<tr>
<td>Lympha</td>
<td>Clear water</td>
<td>lymphocyte (originally cell of the lymph)</td>
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<tr>
<td>Mono</td>
<td>One</td>
<td>Monocyte, mononuclear</td>
</tr>
<tr>
<td>Poly</td>
<td>Many, excess</td>
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<tr>
<td>Morphe</td>
<td>Form</td>
<td></td>
</tr>
<tr>
<td>Nucleus</td>
<td>Small nut, (nux=nut)</td>
<td>Polymorphonuclear</td>
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</tbody>
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*Please make sure the desktop is switched to Pathology Pt1B folder on the PC.*
*Dim and switch off your microscope light.*
*Return the wooden block, if used.*
*Cover the microscope.*
*Push your stool under the bench.*

*Thank you!*