Introduction to the renal histopathology
The glomerular evaluation by routine optic microscopy

Ana Calado¹²
¹. Faculty of Medicine, University of Coimbra, Portugal
². Escola Universitária Vasco da Gama

Introduction
The glomerular corpuscle is a spherical structure in the renal cortex. Human glomeruli are all similar in size, but the rat juxtamedullary glomeruli are larger than glomeruli of the superficial cortex. The glomerular corpuscle generates an ultrafiltrate of plasma composed by water, electrolytes and small proteins. The glomerular elements are a capillary network lined by a thin layer of endothelial cells, a central region of mesangial cells and surrounding mesangial matrix, the visceral epithelial cells or podocytes and the parietal epithelial cells of Bowman’s capsule and associates basement membrane. The two epithelial layers are separated by a narrow cavity which receives the primary ultrafiltrate identified as Bowman’s space or urinary space. Two poles are defined: the vascular pole, where the afferent arteriole enters and efferent arteriole exits the glomerular tuft, and the urinary pole where the parietal epithelium continues with the proximal tubule (Figure 1). The glomerular tuft is composed of lobules. Within each lobe there is an axis with a small number of mesangial cells and mesangial matrix (Figure 2). The precise limits of lobules are difficult to determine in normal glomerulus.

The blood filtration
The glomerular capillaries, derived from the glomerular afferent arteriole, are perfused at high pressure that favors the production of a glomerular filtrate. The filtration barrier is composed by the fenestrated endothelium negatively charged, the associated glomerular basement membrane (GBM) and the epithelial slit diaphragm connecting podocyte projections or pedicels that wrap the capillaries. The filtration end result is a great permeability to water and small molecules and a near impermeability to positively charged molecules like albumine or larger (3,6nm and 70,000D). In many diseases associated with proteinuria, the podocytes are replaced by a continuous cytoplasmic band along the GBM that thickens the hyaline perivascular capillaries of the glomerulus. The mesangial cells, mesenchymal in origin, are important to support the glomerular elements. They have contractile properties and produce vasoactive prostaglandins able to influence filtration. Under injury conditions they proliferate and produce mesangial matrix around GBM with filtration impairment. The thickness of Bowman’s capsule decreases markedly at the vascular and urinary poles. The Bowman’s capsule or parietal epithelium is delimited by squamous podocytes sensitive to testosterone. Male mice have sexual dimorphism as Bowman’s capsule has cuboidal epithelial cells. In these animals this finding has no pathology relevance (Figure 3). Proliferation of parietal epithelial cells occurs in certain pathologic conditions, such as rapidly progressive glomerulonephritis, and con-
tributes to the formation of crescents (see above). The juxtaglomerular apparatus is a crucial part of all nephrons, although on microscopy evaluation few are observed. The reason is that a renal histopathology slide has many round glomeruli randomly cut noticed as round small to large structures and only a few of them are at the juxtaglomerular apparatus level. The juxtaglomerular apparatus is situated at the vascular pole of the glomerulus, where a portion of the distal tubule comes into contact with its ascendant glomerulus (Figure 2). At this point, the cuboidal tubular cells became columnar making the macula densa. Macula densa contacts with both afferent and efferent arterioles and with granular (renin granules) and agranular extraglomerular mesangial cells. The macula densa cells are sensible to sodium and chloride concentration inside the tubular lumen. When the tubular sodium and chloride content is reduced macula densa cells send signals to the glomerular arterioles in order to control glomerular filtration rate and to the granular cells in order to release renin and subsequent angiotensin system. Renin synthesis and secretion are additionally controlled by neurotransmitters of the sympathetic nervous system, by glomerular perfusion pressure and by mediators in the macula densa. The end result of renin release is an increase in blood volume and blood sodium content. Together these actions increase the blood pressure.

Glomerular diseases

Glomerular lesions are very unspecific since many glomerular injuries stimulate cellular proliferation, leucocyte infiltration or basal membrane thickness. The pathology term applied to any of these occurrences is glomerulitis or glomerulonephritis (GN). The immune-mediated lesions cause a thickness of glomerular basal membrane and glomerular cellular proliferation. The immune complex deposition is chemotactic to complement system and to leucocytes with severe effects on the integrity of the GBM and disruption of filtration selectivity resulting on proteinuria.

Glomerular critical points to consider

The popular hematoxylin and eosin (HE) method stains nucleic acids of blue (acidophil) and non-specific proteins in pink (basophil or eosinophil). During the chromatic method the alcohol dissolves lipids and their spaces appear in white. Also in white are: water, gases, glycogen and mucopolysaccharids. Special stainings can be used to differentiate lipids, acid from neutral mucopolysaccharids and glycogen.

Normal glomerulus and glomerular lesions urge to be recognized. The rat has a unique feature in what concerns to glomerular size, when the glomerulus of the superficial cortex are smaller than the juxtamedullary glomerulus. Normal glomerulus have a narrow Bowman’s space observed as a thin white line that becomes larger on glomerular atrophy (Figure 4).

The second critical point to observe is the number of cellular nucleus. A normal glomerulus has very few extravascular or mesangial cells. When numerous nuclei (i.e. cells) are identified they suggest mesangial proliferation or extravascular leucocyte diapedesis into mesangial matrix and GBM.

The third critical point to observe is the GBM thickness. A normal glomerulus has an almost imper-
50% of the cortex glomeruli are involved in the lesion it is described as diffuse (Figure 5). The extension of glomerular lesion is termed global when the entire glomerulus is involved and segmental when the glomerulus is partially affected by a certain lesion (Figure 6).

2. The type of inflammatory infiltrate on glomeruli depends on the etiopathogenesis. In the glomerular tuft, inflammatory cells tend to adopt an endocapillary pattern. The acute inflammation is an exudative stage characterized by inflammatory infiltrate of polymorphonuclear cells like neutrophils, congestion, edema and fibrin - fibrillar acidophil extracellular protein. Chronic inflammation is recognized by inflammatory infiltrate with mononuclear cells of exogenous origin like macrophages, lymphocytes and plasma cells or of endogenous origin like glomerular proliferation – proliferative stage (Table 1). Chronic inflammation hasn’t prominent congestion or edema.

Hyalinosis is the presence of amorphous acidophil protein (hyaline) deposition, commonly a sequel of chronic inflammation associated with immune complex deposition positive on PAS (Periodic Acid Schiff) stain. Kidneys with hyalinosis, usually have diffuse and global hyaline glomeruli with low cellularity. Other hyaline origins should be identified with special stains for amyloid (Congo red positive) and fibrosis (PAS negative).

3. Localization of the abnormalities: inflammatory cells, immune complexes or sclerosis.

A normal glomerulus has a thin Bowman’s capsule and a narrow Bowman’s space. The glomerular tuft has an axis of mesangial matrix and mesangial cells that support capillary loops surrounded by a thin imperceptible GBM and podocytes. When a glomerulus has normal capillary vessels and only the mesangial component is affected, the term

![Figure 5. Schematic classification of affected glomeruli in terms of cortical renal extension on a histopathological slide. A) Mild when involved glomeruli is under 20%; B) Focal when altered glomeruli is over 20% and under 50%; C) Diffuse when more than 50% of the glomeruli are affected.](image)

![Figure 6. Classification of affected glomerulus in terms of individual extension - schematic illustration and practical examples. A) Scheme: a pathological process that alters a glomerulus in a broad spectrum is called global. Image of global membranoproliferative glomerulitis. H&E, 100x.. B) Scheme: a pathological process that results on a lesion affecting a part on a glomerulus is called segmental. The microscopic image has a segmental mesangiolysis. Original image H&E, 400x.](image)
illary thickening of the basal membrane caused by subentothelium deposits of immune complexes. The term mesangiocapillary is used almost interchangeably with membranoproliferative glomerulonephritis. In this pattern there is deposition of immune complexes and a reactive duplication of the GBM giving a “double contour” or “tram track” appearance on silver or PAS stains. Mesangial proliferation is usually present (Figure 9). The excess production of fibrous tissue with collagen type I or IV is termed sclerosis or fibrosis. Formation of a sclerotic lesion can be initiated by a primary insult to podocytes with resulting denuded areas of GBM. An adhesion point is formed between the injured site and the Bowman’s capsule by activated parietal epithelial cells or by direct fusion of the basement membranes. The activated mesangial and parietal cells synthesize evident collagen deposits into the endocapillary compartment on a segmental pattern. Advanced sclerosis is denoted when capillary loops are covered exclusively by parietal epithelial cells and collagen (Figure 10).

Conclusion
The glomerular response to injury is a wide chapter of kidney pathology that should be understood on each specific case regarding the etiology. This approach aims to be a starting point for novels with pathology demands.

Suggested Readings
Figure 9. The reactions of glomerular basement membrane - schematic illustration and practical examples. A) Scheme: hylalinosis or membranous reaction appears as a simple thickening surrounding the capillary loops. On the micrograph the GMB is easily identified – membranous glomerulitis. H&E, 400x. B) Mesangiocapillary or membranoproliferative reaction with a double contour appearance of the GBM and increased number of mononuclear cells. The micrograph shows a membrano-proliferative glomerulitis because there are simultaneously endocapilar hyper cellularity and GBM hyalinization. Original image H&E, 400x.

Figure 10. Glomerulosclerosis formation - schematic illustration and practical examples. A) Scheme: Podocyte lesion is the starting event leaving a denuded GBM (red spot); B) Scheme: an adhesion (red) of the affected capillary to Bowman’s capsule is formed. The micrograph is an example of initial fibrosis (blue circle) H&E, 400x. C) Scheme: parietal activated cells invade the affected segment of the glomerular tuft and deposit collagen matrix. Within the endocapillary compartment, mesangial sclerosis (red) develops within the affected segment. The micrograph puts in evidence (blue line) the fibrous tissue invading the glomerulus. H&E, 400x D) Scheme: advanced sclerosis, at the site of adhesion, a continuous collagen bridge (red) has formed between Bowman’s capsule and the glomerular tuft. The micrograph puts in evidence (red line) the fibrocellular tissue into glomerulus. Original image H&E, 400x.