Angiogenesis in congenital vascular malformations: a dynamic view on a static lesion

Jorna, L.B.

Citation for published version (APA):

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Chapter 2

Benign vascular lesions of skin and soft tissue
- classification
Benign vascular anomalies are the most common lesions in children and adolescents. They represent a highly heterogeneous group in terms of clinical appearance and also biological behavior, and in practice, it can be difficult to determine whether a specific lesion is in fact a true tumor, a malformation or otherwise a reactive process, which has implications for clinical management. A proper terminology of vascular anomalies is not always used by clinicians, and certainly not by pathologists, who often use the same name for (biologically) different entities. It is obvious that such a situation can lead to great confusion.

**Classification**

In 1982, 30 years ago, Mulliken and Glowacki\(^1\) came up with a biological classification system of vascular anomalies, while they noticed already at that time a bewildering nomenclature of cutaneous vascular lesions. ‘Hemangioma’ was used in the broadest sense to describe vascular lesions with a totally different etiology and natural history. In their classification system, vascular anomalies were categorized as either hemangiomas or malformations on the basis of difference in growth behavior and endothelial cell (EC) turnover in cell cultures studies of both types of lesions (Table 1). Mulliken et al suggested that the term ‘hemangioma’ should be reserved for lesions with an increase in cellular turnover. This concept was later confirmed in various studies on the presence of biomolecular markers for endothelial cell proliferation and vessel growth on these distinct types of vascular lesions.\(^2-4\) However, at the same time such molecular studies associated with the biological behavior of lesions, revealed that this strict dichotomy cannot be applied to all lesions and therefore may lead to oversimplification. Examples are the spindle cell hemangioma, which has histological features of both proliferative tumors and vascular malformations (Figure 1), and also the verrucous type of venous malformation with its recently described expression of GLUT1 protein, basically a distinctive feature of infantile hemangiomas.\(^3,5\) Moreover, new entities are still reported, of which the biological behavior, at least on the long term, is not always very clear.\(^6\)

Pathologists involved in the diagnostic pathology of soft tissue tumors often use the classification of Enzinger & Weiss (Table 2).\(^7\) In this classification the term ‘hemangioma’ is used in the widest sense as a benign, nonreactive process, composed of an increase in normal or abnormal vessels. In other words, no distinction between tumors and malformations has been made, leading to the misunderstanding that lesions such as arteriovenous hemangioma, cavernous hemangioma and venous hemangioma are tumors. This is also mentioned in the same book (Enzinger & Weiss), where the authors recognize that “many of these lesions represent tissue **malformations** rather than **true tumors**”.  

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Figure 1. A. Macroscopy of an excised spindle cell hemangioma. Note the multifocal growth of the blood filled spaces (hemorrhagic areas- arrows) of the lesion, like that of a vascular malformation. B. Histopathology shows a vasoproliferative pattern composed of solid areas of spindled cells (hematoxylin and eosin, H&E stain). C. Anti-CD31 immunostain highlights the numerous vessels with slit-like vascular spaces. The spindled cells are negative (anti-CD31).

Table 1. Classification of vascular lesions in infants and children with cellular and clinical features by Mulliken en Glowacki.

<table>
<thead>
<tr>
<th>Hemangiomas</th>
<th>Malformations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Proliferating phase</td>
<td>Capillary</td>
</tr>
<tr>
<td>Involuting phase</td>
<td>Venous</td>
</tr>
<tr>
<td></td>
<td>Arterial</td>
</tr>
<tr>
<td></td>
<td>Lymphatic</td>
</tr>
<tr>
<td></td>
<td>Fistulae</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Endothelial cell proliferation</th>
<th>Normal endothelial cell cycle</th>
</tr>
</thead>
<tbody>
<tr>
<td>40% present at birth (small red mark)</td>
<td>90% recognized at birth</td>
</tr>
<tr>
<td>Rapid postnatal growth and slow involution</td>
<td>Grow commensurately with child</td>
</tr>
<tr>
<td>F:M = 5:1</td>
<td>F:M = 1:1</td>
</tr>
</tbody>
</table>

F, female; M, male
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Table 2. Classification of benign tumors and tumor-like lesions of blood vessels by Enzinger and Weiss.*

<table>
<thead>
<tr>
<th>Benign vascular tumors</th>
</tr>
</thead>
<tbody>
<tr>
<td>Papillary endothelial hyperplasia</td>
</tr>
<tr>
<td>Hemangioma</td>
</tr>
<tr>
<td>• Capillary hemangioma</td>
</tr>
<tr>
<td>• Cavernous hemangioma</td>
</tr>
<tr>
<td>• Venous hemangioma</td>
</tr>
<tr>
<td>• Arteriovenous hemangioma</td>
</tr>
<tr>
<td>• Pyogenic granuloma</td>
</tr>
<tr>
<td>• Acquired tufted angioma</td>
</tr>
<tr>
<td>• Hobnail hemangioma</td>
</tr>
<tr>
<td>• Spindle cell hemangioma</td>
</tr>
<tr>
<td>Lymphangioma</td>
</tr>
<tr>
<td>Lymphphiomycymoma/lymphangiomyomatosus</td>
</tr>
<tr>
<td>Angiomatosis</td>
</tr>
<tr>
<td>Lymphangiomatosis</td>
</tr>
</tbody>
</table>

*Adapted from Enzinger and Weiss’s Soft tissue tumors (2008, 5th edition, Ch. 1, pg. 5).

In other frequently used pathology books on soft tissue tumors, like the Soft Tissue and Bone issue of the WHO Classification of Pathology and Genetics series⁸ and Miettinen’s Modern Soft Tissue Pathology –Tumors and non-neoplastic conditions⁹, the preferred terminology is also hemangioma. Miettinen states that “the distinction of vascular malformation versus hemangioma by the previous criteria requires strong clinical correlation and cannot generally be made by morphology. Therefore, the term hemangioma is used for all lesions here”. But at the same time this author mentions that “there is probably more agreement among specialists that large vessel-type hemangiomas (venous hemangiomas) are mostly vascular malformations rather than true neoplasms”. This creates a serious problem for clinicians, who claim that a clear distinction between malformations and hemangiomas should be made, since biological behavior and also management strategies are different.

For these reasons, the historical distinction between vascular tumors and malformations of Mulliken and Glowacki has been adopted in the classification of the ‘International Society for the Study of Vascular Anomalies’ (ISSVA) in 1996, mainly because of its simplicity and clinical relevance.¹⁰ In this ISSVA classification other types of vascular tumors are added, because their importance for the differential diagnosis and management (Table 3). The classification is often used by clinicians, and also recently validated by pathologists in a series of 144 consecutive cases of cutaneous vascular lesions sent to a pediatric pathology unit.¹¹ They have recognized it as a useful framework for histopathologists, albeit dependent on the adequacy of clinical information and the use of GLUT1 immunostaining (see later). Knowledge is usually derived from small case series, but recently
Greene et al\textsuperscript{12} characterized 5621 referral cases of the Vascular Anomalies Center (Children’s Hospital Boston) with the use of the ISSVA classification. Only 53\% of the submitted cases had an initial correct diagnosis. Tumors were correctly labeled in 70.4\% of cases, and vascular malformations in only 45.6\%, showing that especially the identification of vascular malformations is problematic. A literature research on vascular anomalies by Hassanein et al\textsuperscript{13} revealed that an incorrect diagnosis resulted in incorrect management in up to 20\% of cases.

**Table 3. Updated ISSVA classification of vascular anomalies.**

<table>
<thead>
<tr>
<th>Vascular tumors</th>
<th>Vascular Malformations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infantile hemangioma</td>
<td>Simple</td>
</tr>
<tr>
<td>Congenital hemangiomas (RICH and NICH)</td>
<td>Capillary, AVF, AVM</td>
</tr>
<tr>
<td>Tufted angioma</td>
<td>Lymphatic, CVM, CLVM</td>
</tr>
<tr>
<td>Kaposiform hemangioendothelioma</td>
<td>Venous, LVM, CAVM</td>
</tr>
<tr>
<td>Hemangiopericytoma</td>
<td>Arterial, CLAVM</td>
</tr>
<tr>
<td>Pyogenic granuloma</td>
<td></td>
</tr>
<tr>
<td>Spindle cell hemangioendothelioma</td>
<td></td>
</tr>
</tbody>
</table>

RICH, rapidly involuting congenital hemangioma; NICH, noninvoluting congenital hemangioma; AVF, arteriovenous fistula; AVM, arteriovenous malformation; CVM, capillary venous malformation; CLVM, capillary-lymphatic venous malformation; LVM, lymphatic venous malformation; CAVM, capillary AVM; CLAVM, capillary-lymphatic AVM

**Tissue markers for differential diagnosis**

All benign vascular lesions are lined with endothelial cells that are reactive with the pan-endothelial cell antibodies von Willibrand factor (vWF), CD34 and CD31, as such showing the same reaction pattern as the endothelial cell lining of normal vessels, be it arteries, veins or microvessels.\textsuperscript{14} Moreover, in all benign vascular lesions, there is a subendothelial lining of pericytes and one or more layers of smooth muscle that are reactive with SMA-1 antibody (recognizing the smooth muscle specific \(\alpha_1\) isoform of actin). This pattern can be noticed even in the immature stages of lesions with a biphasic growth pattern such as infantile hemangiomas, in which vessel growth can appear as morphologically infiltrating solid sheets. Also in these cases SMA-1 immunostaining clearly outlines the inconspicuous microvessels. Such a staining pattern clearly contrasts to that of true angiosarcomas, in which the SMA-1 staining pattern is discontinuous or even absent.\textsuperscript{15}

Differential diagnosis between lesions composed of either lymphatic- or blood vessels can be difficult, but at present this has been enormously facilitated by the identification of antibodies that specifically recognize lymphatic endothelium, such as LYVE-1, VEGFR-3, D2-40 and Prox1.\textsuperscript{16-19} In diagnostic pathology D2-40 antibody has proven to be a very reliable tool for this purpose (Figure 2).

Highly specific and therefore helpful in the differential diagnosis of infantile
hemangiomas (IHs) is the expression of GLUT1 (Figure 3). GLUT1, one of the isoforms of the sodium independent glucose transporter gene family, is expressed by the endothelial cells of tissues which have a barrier function like the blood-brain barrier, retina, and placental tissue. North et al found strong immunohistochemical expression of GLUT1 on endothelial cells in all phases (proliferating and involuting) of infantile hemangiomas. GLUT1 is absent on endothelial lining of vessels in vascular malformations and other vascular anomalies (Table 4). Other immunohistochemical studies by North et al revealed that infantile hemangiomas can be differentiated from other benign vascular lesions not only by GLUT1 expression but also with the use of merosin-, IDO-, Lewis Y antigen-, CD15-, and FcgRII- antibodies.

An exception, reported only very recently, is the verrucous type of venous malformation, on the basis of its growth pattern clearly identified as a true malformation, but which shows focal expression of GLUT1 on endothelial cells (Figure 3G and H).

Another tissue marker that recently appeared to be of great interest for differential diagnostic purposes is the Wilms tumor protein-1 (WT1). Lawley et al performed an immunohistochemical study in which she compared WT1 expression in hemangiomas
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and vascular malformations. WT1, initially isolated in hereditary Wilms tumors, is a transcription factor that plays a role in the development of the urogenital system and is also involved in angiogenesis. Lawley et al found strong cytoplasmatic staining of endothelial cells in hemangiomas and absence of staining in capillary, venous and lymphatic malformations and suggested a role for WT1 gene in the differentiation between hemangiomas and malformations. Arteriovenous malformations were not included in her series. In a large series of Al Dhaybi et al23 and Trindade et al24 expression of WT1 was found in vascular tumors and AVMs. Other types of vascular malformations, including the verrucous type of venous malformations were negative. Therefore immunoexpression of WT1 is a helpful tool in the differentiation of vascular tumors and most vascular malformations.

Table 4. GLUT1 and WT1 expression in benign vascular anomalies.

<table>
<thead>
<tr>
<th>Vascular anomaly</th>
<th>GLUT1</th>
<th>WT1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infantile hemangioma</td>
<td>positive</td>
<td>positive</td>
</tr>
<tr>
<td>RICH</td>
<td>usually negative</td>
<td>positive</td>
</tr>
<tr>
<td>NICH</td>
<td>negative</td>
<td>positive</td>
</tr>
<tr>
<td>Kaposiform hemangioendothelioma</td>
<td>negative</td>
<td>*</td>
</tr>
<tr>
<td>Tufted angioma</td>
<td>negative</td>
<td>positive</td>
</tr>
<tr>
<td>Pyogenic granuloma</td>
<td>negative</td>
<td>positive</td>
</tr>
<tr>
<td>AVM</td>
<td>negative</td>
<td>negative</td>
</tr>
<tr>
<td>CM, VM, LM</td>
<td>negative</td>
<td>negative</td>
</tr>
<tr>
<td>Verrucous type of VM</td>
<td>focal positive</td>
<td>negative</td>
</tr>
</tbody>
</table>

*not known
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Vascular malformations

Vascular malformations are rare (0.3 to 0.5% of the population) congenital anomalies which are present at birth, but in some cases they may turn into clinically visible lesions only later in life. The growth is commensurate with the life of the child and malformations never regress. The lesions are stable; however, episodes of sudden expansive growth do occur after trauma, sepsis or hormonal influences like pregnancy or puberty. Clinically, they are divided in low/slow flow or high/fast flow lesions, based on their flow characteristics. High flow results from usually several direct communications between arteries and veins of the malformations. The flow can be evaluated by physical examination or it can be documented by Doppler ultrasonography, angiography, magnetic resonance imaging (MRI) scan and/or computed tomography (CT) scan.

The pathogenesis of vascular malformations is still not well understood. They probably arise as a result of dysmorphogenesis during vascular development in embryonic life. From only a few inherited vascular malformations is known that they are a result of genetic abnormalities. These are mentioned in the respective sections on different types of malformations.

In histopathology, the lesions are subdivided by the predominant vessel type of which they are composed, that is capillary, venous, lymphatic, and arterial or a combination of these vessel types. CM, VM and LM are always slow flow lesions; AVM or combined lesions can be either slow or fast flow.

Although most vascular malformations are sporadic lesions, they can also be part of a more complex often dysmorphic syndrome. Particularly when a patient with a special type of malformation has multiple vascular anomalies or an underlying systemic disease, an associated syndrome has to be considered. Examples are Blue rubber bleb naevus syndrome (VM), Sturge-Weber syndrome (CM), Parkes-Weber syndrome (AVM), Klippel-Trenaunay syndrome (CLVM), Proteus syndrome (LVM) and Maffucci syndrome (VM; hemangioendothelioma). Careful examination (physical examination and imaging techniques) is necessary to obtain a proper diagnosis for further management of the patient. Their recognition, including proper characterization of the specific types of malformations involved, is of importance since several of these syndromes have a genetic background.

Capillary malformation

A capillary malformation (CM), also called port-wine stain, is the most common type of vascular malformation. Capillary malformations are usually present at birth and grow in size commensurately with the child, without involution of the lesion. The lesions are composed of single or multiple flat and reddish maculae, mostly involving the head and neck area. With increasing age, they can become...
thicker and darker in color. In approximately 65% of facial CM the surface may become thickened with a cobblestone appearance. Because CMs show a decrease or absence of nerve density within the malformation, an abnormal neural regulation is suggested as part of their pathogenesis. A few families show an autosomal dominant inherited trait of CMs, located on chromosome 5q. Other inherited forms have a arteriovenous component (CM-AVM). In these CM-AVM families germline RASA1 mutations were found.

In histopathology capillary malformations show an increase of ectatic capillaries and postcapillary venules in the upper dermis, with increased vessel density (Figure 3E and F). The vessels are lined by flat endothelium. A decrease in nerve density, noticed in anti-S100 immunostain, has been reported, but is to our opinion not very helpful in the diagnosis of such lesions. Later in life there is marked increase of the vessel walls of these ectatic microvessels. Sporadically, pyogenic granulomas or tufted angioma are formed in CMs.

**Venous malformation**

Venous malformations (VMs) are rare congenital vascular malformations with an overall incidence of 1 in 10,000. They present as soft, bluish to deep purple masses, which can cause disfigurement or functional impairment. Large VMs can be present in underlying tissues, and depending on their localization and seize, VMs can cause symptoms of pain. In venous malformations with large dilated vessels thrombus formation is a frequent complication due to low flow/stasis of blood. Eventually thrombi may calcify and can be easily recognized on radiography as “phleboliths” (Figure 4A). Elevated levels of D-dimer, a fibrin degradation product, can be detected in patients with VMs, and in these situations it is associated with spontaneous thrombosis and thrombolysis in patients. This phenomenon, termed localized intravascular coagulopathy (LIC), can aggravate into disseminated

![Figure 4. A. Radiographic findings of a venous malformation showing small calcifications or “phleboliths” (arrows). B. Surgical resection of another venous malformation. The skin and subcutis are opened. A part of the malformation is lifted up to demonstrate the complex of large thin walled vessels.](image)
intravascular coagulopathy (DIC) in patients with extensive VMs, particularly in those located in the extremities.

Like other vascular malformations, VMs are a result of aberrant vascular development in embryonic life. Most VMs are sporadic. Rare inherited cases of VMs can be classified as cutaneous and mucosal venous malformations (VMCMs). VMCMs are more superficially located, often asymptomatic and are visible as multifocal, small, hemispherical bluish lesions. Besides located in skin and oral mucosa they can also involve the gastro-intestinal tract, lungs and brain. Autosomal dominant inherited cutaneous and mucosal venous malformations are known to be a result of a mutated gene (VMCM-1) on chromosome 9 (locus 9p21) which encodes the receptor tyrosine kinase Tie2.41,42

The histopathology of a venous malformation reveals a convolute of tortuous dilated veins (clinical picture of a convolute shown in Figure 4B) lined with flat endothelium. Thrombus formation is frequently seen in veins with different stadia in organization of the thrombus, indicating occurrence of recurrent thrombotic events in the lesion (Figure 5); also formation of papillary endothelial hyperplasia (pseudo-Masson tumor, a peculiar type of thrombus organization) can be seen relatively often in the cystically dilated vessels of the VM.

**Histopathological variants of venous malformations**

- **Verrucous VM**, also (erroneously) named verrucous hemangioma. The lesion is mainly located in the lower extremities, presenting as a warty, purple or bluish black papulonodular plaque which never shows spontaneously regression. They are
Benign vascular lesions of skin and soft tissue are considered as malformations rather than a true hemangiomas, because of their clinical behavior of slowly progressive growth and failure to regress.\(^5,43\) Recent publications reported endothelial GLUT1 reactivity in these lesions, which is in contrast with all other types of vascular malformations, and can lead to confusion in the differential diagnosis with infantile hemangiomas (Figure 6). However, WT1 is absent in verrucous VMs, as has been reported for all types of venous malformations.\(^{43,44}\) In our experience the faint GLUT1 endothelial immunostaining in verrucous type of VMs can still be reliably distinguished from the intense continuous GLUT1 immunostaining that is seen in infantile hemangiomas (Figure 3B).\(^{20}\) Characteristic in histopathology is an overlying verrucous hyperplasia of the epithelium (Figure 6A and B). Abnormal vessels, lined with flat endothelium, are located through the dermis and may extend deep into the subcutis (Figure 6C).

**Figure 6.** A. Superficial part of a vascular lesion with dilated thin walled vessels underneath and partially surrounded by hyperplastic epidermis, thus closely resembling an angiokeratoma (boxed area of B; H&E stain). B and C. Full resection specimen of the same lesion revealing conglomerates of thin walled veins extending into the deep resection planes of the specimen (C) (H&E stains). D. Anti-GLUT1 immunostain showing faint expression in the endothelium of vessels consistent with diagnosis verrucous type of venous malformation (anti-GLUT1).

- **Glomuvenous VM (GVM),** also referred to as glomangioma or multiple glomus tumors, is characterized by large, dilated blood vessels lined with several rows of glomus cells (Figure 7).\(^{38}\) Glomus cells are modified (anti-SMA1 positive) smooth muscle cells, derived from the Sucquet-Hoyer canals, which are involved in cutaneous thermoregulation. The endothelial lining is flat and GLUT1 stains are consistently negative. GVMs are mostly located in the skin and soft tissue of extremities and are multifocal, small bluish to purple lesions with a cobblestone or hyperkeratotic...
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appearance. Multiple GVMs, which occur in a widespread or segmental distribution, are often autosomal inherited. Patients have germline mutations in the glomulin gene (1p21-p22) on chromosome 1.\textsuperscript{45}

Arteriovenous malformation

Arteriovenous malformations are composed of a convolute of arteries and veins, characterized by abnormal connections between arteries and veins without an intervening capillary bed.\textsuperscript{25} For this reason, lesions clinically defined as high/fast flow lesions are always arteriovenous malformations (AVMs), and the flow velocity through the lesion is largely depending on the number and size of fistulas. Forty to sixty percent of AVMs are visible at birth and AVMs are predominantly located in the head and neck area. Physical examination reveals a warm overlying skin with a palpable thrill. Before treatment, determination of the flow of the lesions and/or the exact location and extent of the lesion can be done through Doppler-

Table 5. Schobinger staging of arteriovenous malformations,*

<table>
<thead>
<tr>
<th>Stage</th>
<th>Clinical findings</th>
</tr>
</thead>
<tbody>
<tr>
<td>I (quiescence)</td>
<td>Warm, pink-blue, shunting on Doppler examination</td>
</tr>
<tr>
<td>II (expansion)</td>
<td>Enlargement, pulsation, thrill, bruit, tortuous veins</td>
</tr>
<tr>
<td>III (destruction)</td>
<td>Dystrophic skin changes, ulceration, bleeding, pain</td>
</tr>
<tr>
<td>IV (decompensation)</td>
<td>Cardiac failure</td>
</tr>
</tbody>
</table>

ultrasonography, magnetic resonance imaging (MRI) or angiography. It is known that AVMs expand over time, which is clinically staged according to Schobinger (Table 5).\textsuperscript{46,47} Disproportionate growth can be seen after trauma, infections or hormonal changes like puberty or pregnancy. Complete excision of an AVM (or also a VM; see Figure 8A and B) is difficult or impossible, since the lesions have a diffuse growth pattern with poorly delineated outlines and involve several tissue planes.

Although AVMs are presumed to be congenital lesions they can get manifest much later in life. The specific error is not known. AVMs combined with a capillary malformation can be associated with genetic abnormalities.

Histopathology reveals a conglomerate of disorganized arteries and veins with intimal thickening of arteries, despite the often young age of the patients, and marked tortuosity of the vessels. Veins can show arterialization with disorganization of the media of the vessel wall, which can lead to expansion of the lesion (Figure 8C and D). The expansion can also be due to collateralization, thickening of arteries and/or even aneurysmatically dilatation of vessels. This is due to the high flow forces

**Figure 8.** A. Macroscopy of a resection specimen of a venous malformation (VM) showing a diffuse intramuscular growth pattern with poorly delineated outlines towards one of the marked border (arrow). B. Histopathology of the VM showing the close connection of a malformed vessel and the marked border (arrow) (H&E stain). C. Histopathology of an AVM composed of disorganized malformed arteries and veins surrounded by an abundant extracellular matrix component of adipose tissue, which can be mistaken for an intramuscular lipoma (H&E stain). D. Elastic von Gieson stain showing the elastic internal lamina of arteries (arrow) and arterialized veins (asterisks) (EvG stain).
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in the lesion. The vessels are lined with flat quiescent endothelium. Only very occasionally, the AV-fistula can be seen in the plane of the histopathological sections. Till thus far, cases of vasoproliferation that were associated with AVMs were referred to as acro-angiodermatitis or pseudo-Kaposi sarcoma associated with congenital AVMs. They are small nodules, and considered to be not specifically associated with vascular malformations, since they also arise in association with venous insufficiency, iatrogenic AV-shunts, paralysed limbs and amputation stumps. Although very rare, a few cases of angiosarcoma arising in an AVM have also been described in the literature.

**Lymphatic malformation**

A lymphatic malformation is a sponge-like collection of abnormal and cystically dilated lymph vessels filled with lymph fluid (no erythrocytes). The lesions can be superficial or deep; (multi)focal or diffuse; macrocystic or microcystic. The predilection site is the head and neck region. Lymphatic malformations (LMs) can be diagnosed on prenatal ultrasound. LMs typically grow slowly, however in case of infection or hormonal changes, like in puberty, they can rapidly expand. MRI is recommended for making the diagnosis, extent of the lesion and the outcome (macrocystic LMs have a better prognosis than microcystic). The development and causal factors of LMs are not completely understood. Lymphangiogenic growth factors, like VEGF-C and VEGFR-3 are upregulated in LMs, but the precise role of these factors in the development is not clear.

Histopathology of a LM reveals a convolute of dilated lymph vessels, which are positive in LYVE1, VEGFR-3, and D2-40 immunohistochemistry (Figure 2C). More recently Prospero-related homeobox gene-1 (Prox-1) appeared to be a highly sensitive immunohistochemical marker for lymphatic endothelial cells, especially in the larger lymphatic channels. Lymphoid aggregates are seen in close connection to the lymph vessels of the lesion (Figure 2B).

**Differential diagnosis of vascular malformations**

Most benign vascular lesions show a growth pattern of proliferative microvascular growth followed by vascular maturation. Especially in this latter stage of maturation they are hard to distinguish from vascular malformations in histopathology. The best example is the infantile hemangioma, and especially those cases which show a deep infiltration of the subcutaneous tissues. The next part of this chapter describes vascular tumors which are of interest in the differential diagnosis of vascular malformations.
Infantile Hemangioma

An infantile hemangioma is the most common type of hemangioma in childhood, affecting approximately 10% of children. Premature infants and infants with a low birth weight (<1000g.) have a greater incidence than full term infants. They are 3-5 times more likely to occur in females than in males, and they are more common in Caucasian infants. There is a predilection for the head and neck region (70-80%). An infantile hemangioma often appears as a small red mark or discoloration of the skin at birth or within the first 2 weeks of age. In the first year of life the lesion shows a rapid growth, the proliferating phase. At this phase the lesion can be very painful and/or become infected or may ulcerate. Depending on the size and localisation, infantile hemangiomas (IHs) can lead to visual impairment, deafness or loss of breath due to compression and/or obstruction. Infantile hemangiomas of the face may cause significant facial disfigurement. The maximum size of an IH is usually reached in 9 to 12 months. This is followed by a period of spontaneous involution, the involuting phase. Involution may take one to ten years to complete (the involuted stage). Fifty percent of infantile hemangiomas will show complete involution by the age of 5, and 70% by the age of 7. At 10 to 12 years of age the involution of an IH is always complete, but in 50% of patients residual scaring, fibro-fatty tissue or loosening of the skin with teleangiectasias will remain visible. When an infantile hemangioma is involuted it will never re-grow. Infantile hemangiomas are a result of aberrant and focal proliferation of endothelial cells, however the cause is still not completely understood. Although different hypotheses have been proposed, like chorionic villus sampling, hormonal influence, HHV8 infection and local hypoxia, the exact trigger factor or pathogenesis of IH remains to be elucidated.

Histopathology of infantile hemangiomas in the proliferating phase shows a lobular pattern of immature capillaries with or without lumina, lined with plump endothelium and pericytes (Figure 3A). In this phase, both endothelial cells and pericytes show a higher proliferation index. There is an increase in number of mast cells. In the involuting phase the vessels have a mature appearance, with dilatation of capillaries, thickening of the wall and flattening of the endothelium. The proliferating index is low. Especially in this involuting phase, infantile hemangiomas can be difficult to discriminate from vascular malformations (Figure 9A and B). But here, anti-GLUT1 immunohistochemistry is helpful, since it discriminates infantile hemangiomas in all its developmental stages from all other vascular lesions (Figure 9C and D). Wilms tumor 1 immunoexpression is presently used as an additional discriminator, since it is expressed on all types of vascular tumors, but not on capillary malformations or venous malformations. In the involuted phase the lesion is predominantly composed of fibro-fatty tissue.
Benign vascular lesions of skin and soft tissue

**Congenital hemangioma**

In rare cases, hemangiomas show a rapid prenatal growth (especially in the second trimester) and present fully grown at birth. These types of hemangiomas are called congenital hemangiomas. Due to a different biological behavior they can be divided into: rapidly involuting congenital hemangiomas (RICHs) or non-involuting congenital hemangiomas (NICHs). There is no sex prevalence and the lesions are mainly located in the head and neck area and the limbs. RICHs rapidly regress during the early months in life, with complete resolution in less than a year, sometimes leaving a residual scar. Resection is only indicated when complications such as ulceration, hemorrhage, or visual obstruction occur. In contrast, NICHs never regress, but show a proportional growth during life. Because of this growth pattern a NICH has been considered to be a vascular malformation. But NICHs are more localized and can be excised without recurrence. Clinically the lesions show common features including a violaceous color with prominent telangiectasias, often with a bluish pallor. Both lesions appear to be high flow lesions, detected with Doppler-ultrasonography or MRI-scan. The underlying cause has not yet been established.

Histopathology of congenital hemangiomas shows variable sized lobules of capillaries with moderately plump (hobnailed) endothelial cells and pericytes. The lobules are surrounded by dense fibrous tissue with larger thin-walled central draining channels and may contain hemosiderin, thrombosis, cyst formation, focal calcification, and extramedullary hematopoiesis. In NICH the lobules are somewhat larger and the central vessels appear to be more dilated and larger than those seen in RICH. Small arteries, which are increased in number, are seen in close proximity of the lobules. In both RICH and NICH mitoses are occasionally seen and the number of mast cells is increased. GLUT1 and LeY immunostains are negative (Figure 3D).
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Tufted angioma
Tufted angioma, also known as angioloblastoma of Nakagawa, is a rare vascular lesion.\textsuperscript{63,64} The tumor is rarely present at birth, and most commonly manifest during infancy or early childhood. They are considered to be acquired tumors. The predilection sites of these tumors are the neck and thorax area. Tufted angiomas (TAs) are red to violaceous solitary maculae, papules or nodules. The natural history is not always predictable. They can regress spontaneously, but mostly they expand over time in infiltrating plaques that slowly enlarge. After months to years they become stable without spontaneous involution.\textsuperscript{65} The etiology of tufted angiomas is unknown. Large TAs can be associated with the Kasabach-Merritt phenomenon, a severe life-threatening coagulopathy due to platelet trapping and spontaneous bleeding.\textsuperscript{66}

Histopathology demonstrates a vascular lesion composed of numerous lobules (vascular tufts), spread in a so-called cannonball pattern throughout the dermis. The tufts are composed of tightly packed capillaries, containing narrow cleft like vascular lumina at the periphery. Anti-SMA-1 immunostain gives a pericyte lining around the endothelial cells. Peripheral crescent-shaped vessels stain positive for D2-40 antibody, a lymphatic marker.\textsuperscript{67} Prox1 is expressed in areas of the lesion with a spindle cell morphology.\textsuperscript{68} The mitotic rate is low and there is no cellular atypia. GLUT1 and LeY immunoexpression is absent.\textsuperscript{21}

Kaposiform hemangioendothelioma
Kaposiform hemangioendothelioma is an uncommon vascular tumor that can be present at birth or develop in children younger than 2 years of age.\textsuperscript{69,70} Sex distribution is equally. A kaposiform hemangioendothelioma (KHE) presents as a warm, firm, indurated, ill-defined purpuric mass. The overlying skin is often tense and shiny. MRI findings of KHEs show a characteristic involvement of multiple tissue planes, with thickening of the skin, subcutaneous stranding, and edema.\textsuperscript{71} KHEs can be locally very extensive and aggressive with involvement of superficial and/or deeper soft tissues, sometimes even leading to destruction of bones. KHEs are often associated with the Kasabach-Merritt phenomenon.\textsuperscript{66,69} The exact cause of KHEs is till thus far not known. Kaposiform hemangioendotheliomas and tufted angiomas are closely related, however TAs are smaller and more superficially (in the dermis) located lesions with a less prominent spindle cell component.\textsuperscript{68} A KHE is classified as hemangioendothelioma, implying an intermediate malignancy rate rather than a benign tumor, because of a mortality risk due to local aggressive growth and/or severe complications like the Kasabach-Merritt phenomenon. And, although distant metastasis do not occur, regional lymph node metastasis have been described occasionally.\textsuperscript{70}
In histopathology this tumor is composed of sheets and irregular lobules of moderately plump spindled cells which form slit-like lumina containing erythrocytes, surrounded by dense fibrous or hyaline stroma (Figure 10A and B). Hemosiderin pigment is frequently seen. The endothelial cells (anti-CD31 positive) are surrounded by pericytes (anti-SMA1 positive). Prox1 is expressed in the spindled cells. Cellular atypia is minimal and the mitotic rate is low. Lymph vessels are seen adjacent to the lobules, which can be highlighted by anti-D2-40 immunoexpression, and can be quite numerous in some cases (Figure 10C). Another typical feature that relates to the Kasabach-Merritt phenomenon is the presence of platelet-rich micro thrombi, which can be highlighted by anti-CD61 immunoexpression. GLUT1 and LeY immunoexpression is absent.

**Pyogenic granuloma**

Pyogenic granuloma, also known as lobular capillary hemangioma, is a common benign skin tumor. It can occur at any age with a predilection for the female gender, but many cases occur in children or pregnant women (epulis gravidarum). In children, pyogenic granulomas (PGs) are most commonly located on the head and neck area, while the gingival mucosa is the predilection site in pregnant women. A PG presents as a rapidly developing red papule, which bleeds easily and may ulcerate and form crusts. PGs are benign vascular proliferations, which are considered reactive lesions, mainly because many cases have a history of recent trauma at the involved site. In other cases such as the epulis gravidarum, it is likely that hormonal influences are involved, although the inciting event is unknown. Interestingly, these pregnancy related PGs can completely regress after delivery. Histopathology shows a lobulated pattern of capillaries separated by edematous, myxoid or fibrous stroma. The capillaries show a variable pattern of maturation from immature capillaries with closed or inconspicuous lumina till mature capillaries with open widened lumina. Inflammation and ulceration are commonly seen. Mitotic
activity can be noted, but the rate is low. Especially the early proliferative stages can be indistinguishable from infantile hemangioma in histopathology, but GLUT1 and LeY immunohistochemistry are invariably negative in PGs.21

Conclusions

• Benign vascular anomalies represent a variety of vessel abnormalities. Inconsequent nosology and a variety of eponyms or descriptive terms that are applied to certain anomalies have resulted in confusion. A proper classification scheme appears to be available in the ISSVA classification system, which has been validated recently in a large referral center for vascular anomalies.12 In our own experience the classification has been proven useful, also for histopathologists, albeit dependent on the adequacy of clinical information and the use of GLUT1 immunostaining and more recently also WT1 immunostaining. A correct classification and diagnosis is not only imperative for clinical management and prognostic consequence, but also for understanding of the mechanisms and pathogenesis of different types of vascular anomalies.
• The identification of an increasing number of genes in which mutations lead to the formation of vascular malformations has increased not only the understanding of molecular pathogenesis of diseases, but also the need for a correct diagnosis of vascular anomalies, in particular malformations, since familial forms follow a dominant inheritance patterns.
References


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