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

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Hypertrophy in facial capillary malformations: clinical and pathological findings in 11 patients

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Hypertrophy in facial capillary malformations

Abstract

Hypertrophy in capillary malformations (CMs) may be present at birth or manifest itself later in life. To gain insight into the pathology of hypertrophic CMs we investigated a series of eleven excisional biopsies of hypertrophic lips. All biopsies showed dilated thin walled microvessels in the superficial dermis without a neural component. However, large multinodular conglomerates of thick walled vessels with a substantial increase in nerve fibers were found in the deeper parts of the lesions. These veins extended deep into the facial musculature. Hypertrophy in CMs is caused by a venous malformation underlying the capillary malformation. These CMs associated with generalized hypertrophy should be considered as capillary-venous malformations.
Introduction

Capillary malformations also referred to as port-wine stain (PWS) or nevus flammeus are the most common type of vascular malformations. Capillary malformations (CMs) have a reported incidence of 0.3-2.1% in newborns, with an equal sex distribution. Most CMs are isolated findings however a CM may occur in association with more extensive vascular malformations, i.e. Klippel-Trenaunay syndrome and Sturge-Weber syndrome (SWS). Clinically the lesions appear as a red or purple stain, in which hypertrophy can arise over time. Hypertrophy on the skin surface can cause irregularities for which the term ‘cobblestone’ is generally applied (Figure 1).

In other patients a bulging mass may be seen (Figure 2). The reported incidence ranges from: 11% till 23% which may be influenced by the lack of consensus regarding the definition of hypertrophy. Microscopically, CMs are composed of dilated thin walled capillary like vessels lined by a single layer of flattened endothelial cells, located in the papillary and upper reticular dermis. The diameter of these ectatic vessels slowly increases with aging. It has been suggested in the literature that hypertrophy may be related to an absence of autonomic nerves. However the exact nature of the hypertrophy is unclear and detailed histopathological descriptions are lacking. The aim of this study was to find out if capillary malformations associated with generalized hypertrophy is only limited to the dermis or whether it also involves the subcutis and underlying muscles and to identify the nature of the vessels.
Materials and methods

Setting
The department of Plastic and Reconstructive Surgery at the Academic Medical Center in Amsterdam, the Netherlands, a tertiary referral center for vascular anomalies.

Population
Patients with capillary malformations in the 2nd or 3rd branch of the trigeminal nerve, who underwent surgical reduction of their hypertrophic lips. Patients with a solitary venous malformation were excluded from this series. The indication for surgery was facial disfigurement, difficulty with eating or articulating. The lip excision included all layers of the lip including skin, vermilion, mucosa and the underlying orbicularis oris muscle.

Table 1. Clinical characteristics of patients with facial capillary malformations and hypertrophy of the lip.

<table>
<thead>
<tr>
<th></th>
<th>Lower lip</th>
<th>Upper lip</th>
<th>Both lips</th>
<th>Yes</th>
<th>No</th>
<th>Unknown</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex (M:F)</td>
<td>3:8</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age ¹ (mean, years)</td>
<td>23</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CM in isolation</td>
<td>10 (91%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hypertrophy lip</td>
<td>9 (82%)</td>
<td>0</td>
<td>2 (18%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Present since birth</td>
<td></td>
<td></td>
<td></td>
<td>6 (55%)</td>
<td>2 (18%)</td>
<td>3 (27%)</td>
</tr>
<tr>
<td>Progression</td>
<td></td>
<td></td>
<td></td>
<td>8 (73%)</td>
<td>0</td>
<td>3 (27%)</td>
</tr>
</tbody>
</table>

M, male; F, female. ¹ Age of patient at the time of surgical excision.

Data collected included
Gender, age of onset and rate of hypertrophy progression, age at the time of surgery, and diagnostic imaging, as well as medical photographs were available.

Histopathology and immunohistochemistry
Paraffin embedded material sections were stained with hematoxylin and eosin (H&E) and Elastic van Gieson (EvG) stains for histopathological observations. Specifically, the type of vessels, the topographic localization of the vessels and the morphologic features of the extracellular matrix were evaluated as well as evidence of possible complications such as chronic inflammation, hemorrhage and thrombosis. Serial sections were mounted for additional immunohistochemical staining with anti-smooth muscle actin-1 (anti-SMA-1) to determine the thickness of the smooth muscle layer of intralesional vessels, anti-D2-40 antibody to investigate the presence of lymphatic vessels, glucose transporter-1 protein (anti-GLUT1) antibody to exclude infantile hemangiomas, and anti-S100 antibody to demonstrate the presence of nerve bundles.
Results

Clinical features
Eleven patients were included in this study; their clinical characteristics are presented in Table 1. Hypertrophy of the lip and surrounding skin occurred without the appearance of a cobblestone pattern of the rest of the CM (Figure 2). In six patients, the hypertrophy was present at birth. In one patient hypertrophy developed between the ages of one and five, and one patient noticed lip hypertrophy around the age of 30. Eight patients noticed a progression of hypertrophy over time, in one patient this was related to pregnancy. Diagnostic magnetic resonance imaging (MRI) prior to surgery was required only once. The MRI scan showed no pathological signal intensities and no evidence of venous malformation in the subcutis, muscles or bones.

Histopathological features
All resection specimens consisted of skin, subcutis and segments of underlying skeletal muscle. In all cases dilated thin walled capillary like vessels were present in the superficial parts of the dermis. In addition, large multinodular vascular conglomerates, composed of equally sized thick walled vessels embedded in collagen rich fibrous matrix, were found in the deeper layers of the dermis, and subcutis with extension into the skeletal muscle tissue (Figure 3A, B). In all cases the vessel walls were remarkably uniform, characterized by thick walled veins (no elastic lamellae) with a flat endothelial lining (Figure 3C). Anti-SMA-1 staining showed a media composed of up to five layers of smooth muscle cells (Figure 3D, E). Signs of chronic inflammation, microvascular proliferative reaction, thrombosis or hemorrhage and calcifications were absent in all cases. In the deeper parts of the

Figure 2. Clinical appearance of facial CM with generalized hypertrophy of the lip.

Figure 3. Histopathological features of facial CM with generalized hypertrophy of the lip.
lesions a variable degree of atrophy was seen with fatty replacement and fibrosis of the underlying skeletal muscle. Anti- D2-40 confirmed the absence of a lymphatic component in the malformations. All specimens were GLUT1 negative. Anti-S100 immunostaining demonstrated a remarkable increase of nerve fibers adjacent to the thick walled vessels, in contrast to the superficial parts composed of thin walled, dilated vessels, without any nerve fibers. Occasional small, mature nerve bundles not strictly related to these conglomerates were interpreted as pre-existing nerves of skin and soft tissue (Figure 3F).

**Figure 3.** Histopathologic features of the excision biopsies of the hypertrophied lip. A, B. overview and detail of the vascular lesion composed of thick-walled vessels. (H&E). C. Elastic van Gieson stained section to confirm the presence of veins (absence of internal elastic layer) with a feeding artery (with internal elastic lamina) in the depth of the lesion (EvG). D. Anti-SMA-1 immunostain to highlighting the thick walled vessels of the lesion in contrast to the thin walled capillaries of the pre-existing CM just underneath the surface epithelium. E. Anti-SMA-1 immunostain to confirm a composition up to five layers of smooth muscle cells of the thick walled vessels (anti-SMA). F. Anti-S100 immunostain highlighting nerve fibers in close relation to the thick walled vessels (anti-S100).

**Discussion**

Our findings show that despite the fact that the patients were clinically diagnosed as having only capillary malformations with hypertrophy they in fact had lesions consisting of dermal capillary malformation with venous malformation in the subcutis and muscles. These histopathological observations have never been described before. This provides a new insight in the understanding of hypertrophy in the setting of CMs. Facial CMs can be cleared by dye laser treatment, however hypertrophy, if present, does not respond to this modality. This can be explained by the results of this study. The patients in our series had generalized hypertrophy of the area affected
by the CM. This is contrast with the previously reported distinct papules or nodules that may occur within the boundaries of a CM, and which upon histopathology appear to represent pyogenic granulomas,6,7,12 or in rare instances acral arteriovenous tumors.13 Since our specimen were obtained during lip surgery we were not able to obtain biopsies from unaffected areas. Most of our patients had lip hypertrophy since birth, a finding which is in agreement with other studies.8 They also had a progression of the hypertrophy occurring at a young age (mean age 23 years old). This seems to be in accordance with the findings in venous malformations where puberty and pregnancy are known causes for progression.2,14,15 Localized hypertrophy such as a cobble stone appearance usually presents in the 4th to 5th decade. A capillary malformation is a clinical diagnosis and imaging is seldom required.16 The only patient in our study who underwent MRI imaging prior to surgery had no pathological signal intensities in the affected area, despite the positive clinical and pathological findings, corroborating the observations of Hovius and colleagues.17 Our histopathological examination showed uniform morphological features of superficial thin walled vessels (CM) and deeper thick walled dilated vessels (venous malformation), arranged in conglomerates extending deep in the muscular layer. Our findings of superficial thin walled vessels without nerve fibers correspond with the manifestation of CMs as described previously.4 Barsky described pathologic vessels in CMs as a high number of vessels in the immediate subepidermal area, which diminishes rapidly in the deeper parts with progressive dilation of the vessels (ectasia) with advancing age. He did not mention vessels in the deeper layers probably related to lack of depth when using 3 mm punch biopsies. Substantial increase of nerve fibers in close relation to the thick walled vessels throughout the deeper dermis was a consistent finding, while no nerves were found in relation to superficial thin walled vessels (CM). This is partially in agreement with the recent findings by Meijer-Jorna, who reported an increase in nerve elements in 55% of venous malformations in the studied biopsies.18 Smoller and Selim demonstrated a decrease in the perivascular nerve elements of thin walled vessels of CMs.10,19 Also this study used 3 mm punch biopsies while it is not known if biopsies of hypertrophied lesions were included. Our study differs from other studies in the fact that we did not have superficial samples but full depth of the tissue including the muscle. A potential drawback of this study is the fact that we did not sample other locations than the lips or healthy skin, but that was not possible in the retrospective design of the study and would in fact constitute unnecessary overtreatment. The findings of this study are important for patient education regarding the nature and the prognosis of their hypertrophy as well as the expected results of treatment.
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References