Microvascular Changes in Human Gastric Carcinomas with Coagulative Necrosis: An Ultrastructural Study

Rosario Alberto Caruso *; Francesco Fedele *; Giuseppe Finocchiaro *; Giandomenico Pizzi *; Mirella Nunnari *; Giuseppina Gitto *; Valerio Fabiano *; Luciana Rigoli *

* Dipartimento di Patologia Umana, Policlinico Universitario, Messina, Italy
† Clinica Chirurgia VI, Policlinico Universitario, Messina, Italy
‡ Dipartimento di Scienze Pediatriche, Mediche e Chirurgiche, Policlinico Universitario, Messina, Italy

Online Publication Date: 01 October 2008

To cite this Article Caruso, Rosario Alberto, Fedele, Francesco, Finocchiaro, Giuseppe, Pizzi, Giandomenico, Nunnari, Mirella, Gitto, Giuseppina, Fabiano, Valerio and Rigoli, Luciana(2008)'Microvascular Changes in Human Gastric Carcinomas with Coagulative Necrosis: An Ultrastructural Study',Ultrastructural Pathology,32:5,184 — 188

To link to this Article DOI: 10.1080/01913120802289231

URL: http://dx.doi.org/10.1080/01913120802289231
Microvascular Changes in Human Gastric Carcinomas with Coagulative Necrosis: An Ultrastructural Study

Rosario Alberto Caruso, MD, Francesco Fedele, MD, Giuseppe Finocchiaro, MD, Giandomenico Pizzi, MD, Mirella Nunnari, BS, and Giuseppina Gitto, BS
Dipartimento di Patologia Umana, Policlinico Universitario, Messina, Italy

Valerio Fabiano, MD
Clinica Chirurgica VI, Policlinico Universitario, Messina, Italy

Luciana Rigoli, MD
Dipartimento di Scienze Pediatriche, Mediche e Chirurgiche, Policlinico Universitario, Messina, Italy

ABSTRACT

Ultrastructural findings in three cases of gastric carcinoma with coagulative necrosis are reviewed with special emphasis on microvascular changes. Intratumoral microvasculature revealed more or less stabilized vessels. Some were characterized by a close association between pericytes and endothelial cells, whereas others showed laminated basement membrane, with a loose association between pericytes and endothelial cells. Some mural cells exhibited ultrastructural signs of regressive changes, including lipofuscin granules, swollen mitochondria, and cytoplasmic lucency. These findings are discussed in relationship to a number of recent studies of the microvascular injury caused by hypoxia and reoxygenation, in humans and animals.

Keywords: coagulative necrosis, electron microscopy, gastric carcinoma, microvasculature

The growth of a solid tumor depends on the development of its own blood supply [1]. This process, called angiogenesis, results in an irregular and chaotic vascular network [1]. Most tumor vessels have an irregular diameter and random branching, and lack the defining structural features of arterioles, capillaries, or venules [2, 3]. The thickness of tumor blood vessel walls does not correlate with the size of the vessel, most wide-caliber tumor vessels having thin walls like those of capillaries [3]. Abnormalities are present in all components of the vessel wall including, endothelium, mural cells (smooth muscle cells and pericytes), and basement membrane. Endothelial cells sprout and proliferate consistent with their dynamic nature [4]. Defects in endothelial cell barrier function, due to abnormal cell-cell junctions and other changes, exagerate vessel leakiness [5]. Pericytes in experimental tumors have multiple abnormalities. Unlike pericytes on normal blood vessels, which cling tightly to endothelial cells and are enveloped by a tightly fitting basement membrane, most pericytes in tumors are loosely associated with endothelial cells [5, 6] and have extra layers of loosely fitting basement membrane [7–11].

The original concept of hypoxia in tumors, put forward by Thomlison and Gray [12] almost 50 years ago, was of hypoxic cells residing at the limits of oxygen diffusion from functional blood vessels and being on the edge of regions of necrosis in the tumor. Such cells were believed to be exposed to prolonged or chronic...
hypoxia. Later, it was recognized that fluctuations in blood flow (perfusion limitations) may play an equally important role in the development of hypoxia in tumors, such that cells in tumors may be exposed to short-term hypoxia, termed acute or fluctuating hypoxia [13]. Thus, it has been established that most human tumors develop a pathophysiologic microenvironment during growth, characterized by an irregular microvascular network and regions of chronically and transiently hypoxic cells [14].

Coagulative necrosis is a common feature of solid tumors, and has been reported as an indicator of a poor prognosis in a number of solid tumors, including breast [15, 16], thyroid [17], renal [18], and non-small cell lung carcinomas [19]. Recently, we had the opportunity to study by light and electron microscopy three cases of gastric carcinoma with extensive coagulative necrosis [20–22]. In this paper, we present our experience with microvascular changes in perinecrotic areas of these tumors.

MATERIALS AND METHODS

In our department, gastric tumors are routinely processed for both light and electron microscopic observations. Briefly, the fragments of fresh tumor tissue were divided into 2 portions with a sharp razor blade. The first member of the pair was processed for routine paraffin embedding together with additional tissues samples taken from the tumor and from surgical borders of the specimen. These sections were stained with hematoxylin–eosin (HE). The second piece of the paired samples was minced into smaller pieces and destined for electron microscopy. Small pieces of the fresh tumor tissue were immediately fixed in 3% phosphate-buffered glutaraldehyde, pH 7.4, and postfixed in 1% osmium tetroxide. Semithin araldite embedded sections were stained with Giemsa’s reagent. Selection of fields was focalized on perinecrotic tumor areas. From the selected blocks, sections of 80-nm thickness were double-stained with uranyl acetate and lead citrate; they were then examined and photographed in a Philips EM 300 electron microscope.

RESULTS

The main clinicopathologic characteristics of these gastric tumors have been published elsewhere [20–22]. Histologically, these 3 cases showed as common denominator a solid growth pattern. The first case showed focal neuroendocrine differentiation [20], the second a hepatoid differentiation [21], and the third was an anaplastic carcinoma [22]. Tumor necrosis was of the coagulative type, characterized by homogeneous clusters and sheets of degenerating and dead cells (Figure 1A). Degenerating and thrombosed blood vessels may be found within necrotic areas. Necrosis could follow a peritheliomatous pattern, in which a cord or sheath of viable tumor cells surrounds or clings to a centrally disposed blood vessel (Figure 1B).

Ultrastructural study demonstrated variable association of pericytes with endothelial cells. Close contacts between pericytes and endothelium, and a regular structured basement membrane could be found in some blood vessels (Figure 2). In other blood vessels, pericytes showed an abnormally loose association with endothelial cells and had extra layers of loosely fitting basement membrane (Figure 3). The endothelial cells comprising these microvascular structures varied in thickness and luminal surface contours. Some of these...
endothelial cells exhibited an electron-lucent cytoplasm (Figures 3, 4). Cellular debris was trapped within the layers of basement membrane (Figure 4). Basement membranes were thrown into complex redundant patterns (Figure 5). Microvessels with a regular basement membrane that lacked focal mural cell coverage were observed (Figure 5). Some mural cells showed regressive changes, including lipofuscin granules (Figures 2, 6), cytoplasmic lucency (Figure 4), and swollen mitochondria (Figure 7) with damaged and reduced cristae. Endothelial cytoplasmic sprouting and mitoses were not seen.

DISCUSSION

Tumor necrosis has garnered increased attention over the last few years, in part because a number of studies have now shown that tumor necrotic tissue represents a significant prognostic marker with an independent influence on metastasis-free survival in patients with neoplasia [15–19]. Coagulative tumor necrosis is readily appreciable upon routine histologic review of conventional HE-stained tumor specimens. Dense eosinophilic fibrous tissue or hyalinized connective tissue also represents another common degenerative change in solid tumors that should not be mistaken for necrosis.

Figure 2. A stabilized capillary exhibits endothelial cells and pericytes well integrated. Some pericyte processes contain lipid bodies and profiles of rough endoplasmic reticulum, whereas others exhibit lipofuscin granules. × 12,000.

Figure 3. Pericyte processes within loose, multilayered basement membrane of tumor vessel. Some endothelial cells show cytoplasmic lucency. × 6000.

Figure 4. Multilayered basement membrane contains cell debris. Endothelial cell and pericyte process show focal cytoplasmic lucency. × 6000.

An interpretation of the fine structural changes in the microvasculature of gastric carcinomas must begin by considering the growth pattern and the extensive necrotic areas at the light microscopic level. It is thought that tumor necrosis is caused by chronic ischemia (i.e., hypoxia, low pH, low glucose, high lactate) within tumors, due to vascular collapse, high interstitial pressure and/or rapid tumor growth outstripping its blood supply [23]. The contiguous or sheet-like nature of coagulative necrosis suggested that the cause of death in our cases was likely ischemic injury, affecting a field or group of tumor cells fed or drained by a single vessel. Furthermore, ischemia was also suggested by the presence of peritheliomatous growth pattern, in which a cord or sheath of viable tumor cells surrounded or clung...
to a centrally disposed blood vessel [24]. Therefore, the microvascular changes observed in the present ultrastructural study may be interpreted in large part as a manifestation of chronic ischemic conditions. Swollen endothelial cells, characterized by electron-lucent cytoplasm, have been described after experimentally induced ischemia [25] and in human pituitary tumors [26]. Damage of microvascular cells caused by hypoxia and reoxygenation is accompanied by peroxidation of membrane lipid and morphologically by lipofuscin formation and mitochondrial swelling [27]. Moreover, lipofuscins are believed to be a mitochondrial degradation product [28]. Accordingly, mural cells showed ultrastructural signs of chronic hypoxia, including lipofuscin granules, swollen mitochondria, and cytoplasmic lucency. To our knowledge, this appears to be the first report describing lipofuscins in tumor mural cells.

In this ultrastructural study of solid carcinomas of the stomach, we have described a more or less stabilized microvasculature within the tumor tissue. Stabilized microvasculature was suggested by the presence of continuous endothelial lining, a regular structured basement membrane, and tightly assembled pericyte processes [3]. Our ultrastructural findings of replicating basement membrane are diagnostic of repeated focal injury and repair of pericytes or endothelial cells, because cell debris was found between different layers of the basement membrane of the microvasculature [7, 29]. Consequently, reduced pericyte coverage and laminated basement membrane may limit communication between endothelial cells and pericytes, and thus contribute to their lesser stability [3, 7]. These ultrastructural observations are in good agreement with recent reports on changes in microvascular fine
structure in mouse tumor models [3, 7]. Studies aimed at determining the frequency of tumor necrosis in gastric carcinomas and its influence on prognosis after potentially curative resection are currently underway.

In conclusion, our ultrastructural study confirms the morphologic features of a more or less vascular stabilization described in experimental tumors. Furthermore, it adds descriptive information on the existence of endothelial cell and pericyte regressive changes compatible with chronic hypoxia within tumor tissue in our cases of gastric carcinomas.

Declaration of interest: The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

REFERENCES