burning in this area can be significant sources of carbon that contribute to the atmospheric particulate carbon burden.

The type and concentration of carbon particles in sediments not only may provide historical records of natural and anthropogenic burning but also may act as indicators of other substances such as metals and organics mobilized to the environment during the combustions. Finally, examination of the types, dimensions, and amounts of carbon particles deposited over the past decades will indicate how effective are the pollution control devices that have been installed on plants producing energy from fossil fuels.

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End-Capillary Loops in the Heart: An Explanation for Discrete Myocardial Infarctions Without Border Zones

Abstract. *Separate perfusions of canine coronary arteries with colored silicone-rubber compound reveal that in the region where two microcirculations abut, capillaries derived from individual large vessels are discrete, with no interconnections. Terminal homologous capillaries form loops rather than anastomosing with heterologous capillaries. This anatomic arrangement may account for discrete myocardial infarctions without ischemic border zones.*

Studies from our laboratory have shown that the amount of myocardium susceptible to necrosis is determined by the volume perfused by the vessel supplying that tissue. These investigations revealed a homogeneous necrotic zone, with uniform depletion of creatine kinase from the central to the lateral edge of the infarction (*1*). No lateral border zone of intermediate depletion could be demonstrated when tissue supplied by the nonoccluded vessel was excluded from analysis. Histologically, a serial-section study of acute canine myocardial infarctions demonstrated that the boundary between necrotic and normal tissue at the lateral edge of an infarction was composed of a remarkably complex interdigitation of discrete tissue, with no identifiable border zone of ischemic but viable myocardium (*2*). More recently, we extended the serial-section analysis by doubly injecting the occluded left anterior descending coronary artery (LAD) and the nonoccluded main left coronary artery (or nonoccluded LAD branch) with white and red silicone rubber (Microfil), respectively. We were able to show that all necrotic myocardium was supplied by the LAD; the surrounding normal myocardium was supplied by the main left or nonoccluded LAD branch (*3*).

Although the concept that the myocardium at risk is equal to the volume of tissue perfused by the occluded vessel appears obvious, it conflicts with the commonly accepted "bull's-eye" view of myocardial infarction: an area of central necrosis, surrounded by a border zone of ischemic but viable tissue, which blends imperceptibly into normal myocardium (*4*). The ischemic zone has been inferred from electrophysiological and biochemical data demonstrating intermediate levels of ST-segment elevation and depletion of creatine kinase in this region when compared to normal and necrotic tissue (*5*). We interpret this intermediate zone as an artifact derived from the summation of discrete normal and infarcted tissue peninsulas in the measured samples of myocardium (*1*, *2*). These peninsulas are so interdigitated along the lateral boundary of an infarction that manual or visual separation of tissue is impossible.

Proponents of the bull's-eye view of myocardial infarction have not addressed the issue of what anatomic arrangement of the microvasculature could account for an ischemic border zone. Two arrangements of the vessels are conceivable. In one, complex alternation of capillaries derived from both the occluded and nonoccluded coronary arteries would allow individual cells to be perfused by both circulations. In the other, a network of existing precapillary and capillary anastomoses between vessels derived from occluded and nonoccluded coronary arteries could provide sufficient blood flow at the border to maintain viability of ischemic tissue. Although extensive preformed collateral networks between larger vessels exist in many species (*6*–*8*), anastomoses of small caliber vessels in the mammalian heart have rarely been mentioned (*9*). Most studies of the coronary vasculature utilized technology that precluded analysis of vessels below arteriolar size (*6*, *8*). The purpose of the study reported here was to characterize the anatomic arrangement of the normal microcirculation in the region where two vascular networks supplied by separate coronary arteries or branches of a single coronary artery interdigitate. This area would be equivalent to the lateral border zone if myocardial infarction were present.

Four mongrel dogs had white Microfil injected into the LAD; simultaneously, and under equal pressure, red Microfil was injected into an artery perfusing an adjacent region: either the main left or a proximal branch of the LAD (two dogs each). In seven other dogs white Microfil was injected into the LAD, red into a proximal branch, and yellow into the LAD proximal to the branch. In one dog white Microfil was injected only into the LAD. The entire heart was cleared with organic solvents as described by Schaper (*8*). After several weeks, the hearts were semitranslucent and amber, with colored zones corresponding to the filled capillary networks. Portions of tissue 1 to 2 mm thick were removed freehand from the boundary of the two circulations. Sections oblique to the longitudinal axis of the vessels as visualized under a dissecting microscope were taken. The tissue slices were examined with epi-illumination under the $\times 2.5$ and $\times 6.3$ objectives of a standard photomicroscope to permit color characterization of the vessels.

The entire depth of the tissue slices was visualized with appropriate focusing, and individual vessels could be followed for relatively long distances. The capillary beds were remarkably discrete

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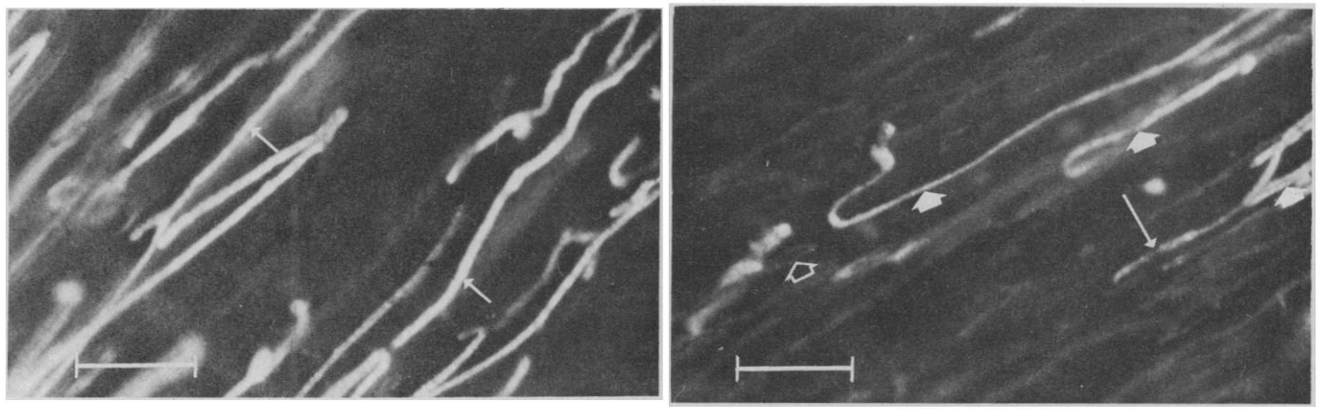

Fig. 1 (left). Multiple capillaries filled with white Microfil (hence derived from the left anterior descending coronary artery) approach a zone perfused by the main left coronary artery (out of the field, to the upper right) and loop back on themselves, forming sharp hairpin turns. Several loops appear incomplete because they extend out of the plane of focus. No anastomoses with capillaries perfused with red Microfil are noted, nor is there a complex alternation of red- and white-filled capillaries in this border region. Two straight extensions from the apex of capillary loops are noted (arrows); both subsequently form additional loops. Epi-illuminated cleared 2-mm section, ×230; scale bar, 100 μm. Fig. 2 (right). Several capillaries perfused with white Microfil derived from the left anterior descending coronary artery (solid short arrows) extend into this zone, which is predominantly supplied by red-filled main left capillaries. Rather than anastomosing with heterologous vessels, the capillaries originating from separate large coronary arteries form loops at their terminus. In the upper portion of this field, a white loop approaches within 17 μm of a red loop (open arrow), with no evidence of a connection between the two vessels. The internal radius of this obliquely oriented white loop is 13 μm, consistent with the size of a single myocardial cell. The white capillary on the lower right has a straight extension (long arrow) at the apex of its loop, which appears to end abruptly. Epi-illuminated cleared 2-mm section, ×230; scale bar, 100 μm.

where they abutted (Fig. 1). No complex alternation of capillaries filled with differently colored Microfil was observed. Characteristically, capillaries arising from one artery approached the capillaries in the adjacent vascular bed and looped back on themselves without anastomotic connection (Fig. 2). These loops have a narrow hairpin-turn radius sufficient to envelop one or two myocardial cells. Straight extensions from the apex of the loop were observed frequently. These distal branches either extended out of the plane of section, ended abruptly (possibly because of inadequate filling), or gave off additional loops. Arcades and loops filled with white (or yellow) Microfil came within several micrometers of red-filled capillaries with no evident interconnection. Moreover, no capillary-size vessels filled with two colors of Microfil were observed.

This loop arrangement at the lateral border of the circulation field of one coronary artery is consistent with the concept of an anatomical end-capillary bed (*10*). No direct anastomoses between capillaries of different coronary arteries were demonstrated. The absence of double filling also weighs against the possibility of significant direct interconnections at the capillary level with only a functional separation of capillary beds by pressure gradients. Although the density of vessels in the injected tissue precluded a systematic study of intercoronary arteriolar anastomoses, indirect evidence suggests that they were not present. If anastomoses were common between arteriolar branches of coronary arteries, we would have expected to observe vessels filled with two colors of Microfil. Double filling was not seen except in several larger vessels, which we interpreted as veins.

Different sites of cannulation were chosen for the injection of the Microfil. This suggests that our results represent a general phenomenon that exists between any two arteries in the heart; that is, that all capillary beds are end-capillary beds. Whether this is true at the level of small arteries and arterioles, however, cannot be determined by this method.

This study has shown that the terminal vascular bed derived from one artery remains discrete in its distal ramifications where it interdigitates with similar vessels derived from another artery. Homologous loops at the capillary level are not interconnected with heterologous loops. The absence of intercoronary capillary anastomoses is consistent with our view of acute myocardial infarction: a discrete infarct zone determined by discrete vessels without a lateral border zone of ischemic tissue. Although large proximal intercoronary anastomoses exist, the absence of small vessel interconnections precludes the existence of an ischemic but viable border zone based on the microvascular anatomy. Other mechanisms such as local differences in contractility or in intramyocardial pressure may be invoked to explain a lateral border zone with dimensions of millimeters, and a zone of less than 0.1 mm may be explained by simple diffusion. On the basis of experiments from our laboratory and others (*2*, *11*), we conclude that these secondary factors contribute minimally to the principal feature of the geometric pattern of an infarct (*12*).

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9. Baroldi and Scomazzoni (*7*) include an annotated bibliography of literature on the intracardiac anastomotic circulation. Of particular interest are several early 20th-century articles about capillary anastomoses.
10. Preliminary work from this laboratory has shown the existence of a complex capillary network in canine skeletal muscle. The gracilis

muscle was filled with red and white Microfil through the femoral artery and a muscular branch, respectively. The tissue was processed in the same way as the heart tissue. Many capillaries at the border region between the two colors were double-filled with red and white Microfil. These results demonstrate that numerous capillary anastomoses are present in skeletal muscle, in contrast to the capillary beds in the heart, which are discrete. They further imply that our finding of end-capillary beds in the heart is not a technical artifact, for the same technique was employed for both myocardium and skeletal muscle; if capillary anastomoses existed in the heart they should have been observed with this method.

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Intestinal Absorption of Immune Complexes by Neonatal Rats: A Route of Antigen Transfer from Mother to Young

Abstract. *Horseradish peroxidase (HRP) in the presence of specific immunoglobulin G antibody to HRP is selectively absorbed from the gut lumen and transferred by intestinal epithelial cells to the lamina propria in newborn rats. The HRP is not transferred in detectable amounts in the absence of the antibody. Transport of maternally derived antigen via antigen-antibody complexes may have important influences on the developing immune system in young mammals.*

The transmission of maternal immunoglobulins across fetal and neonatal tissues is essential for the survival of mammalian offspring (*1*). In this way, developing mammals passively acquire immunity until their own immune systems mature. In the neonatal rat, maternal immunoglobulins that occur in the milk are transported across the proximal small intestine during the first 3 weeks after birth. The transport process is highly selective for immunoglobulin G (IgG). Other immunoglobulin classes, as well as other serum and milk proteins, are transferred in much smaller amounts or not at all and instead are digested intracellularly by absorptive cells in the distal jejunum and ileum (*2*). The transport of IgG is a saturable process, can be competitively inhibited, and requires the presence of the Fc (crystallizable fragment) region of the IgG molecule (*2, 3*). This evidence is consistent with the hypothesis that transport of maternal IgG is mediated by cell membrane Fc receptors (*1–4*).

Potential antigens within the mother are in some instances also transferred to the young and can lead to the suppression or enhancement of the immune response (*5–7*) or, if the antigen is a pathogen, active infection of the young (*8*). However, the mechanisms whereby antigens are passed to the young are poorly understood. By using the electron microscope we have documented the transfer of antigens in the form of antigen-antibody complexes across the proximal small intestine of the suckling rat.

Horseradish peroxidase (HRP), a globular protein (40,000 daltons) that can be readily visualized in the electron microscope, was used as the antigen for our transport experiments. We obtained an antiserum to HRP by immunizing adult rats and rabbits with HRP in complete Freund's adjuvant. Control antiserums were obtained from animals immunized with dinitrophenylated bovine-γ-globulin (DNP-BGG). The IgG fractions of the antiserums were purified by precipitation with ammonium sulfate and then by chromatography with diethylaminoethyl (DEAE) cellulose. The antigenic specificity of each IgG fraction was assessed by immunodiffusion, and the concentration of specific antibody to HRP present in each of the antiserums was estimated by quantitative precipitation. The concentration of total IgG in each preparation was then adjusted to 10 mg/ml in 0.1*M* phosphate buffer, *p*H 6.0. Soluble immune complexes of HRP and antibody to HRP (PAP) were prepared by adding a twofold molar excess of HRP to the antiserum to HRP. An equal amount of HRP was added to the antiserum to DNP-BGG to obtain a control mixture of HRP and IgG (PIg). Neonatal rats (10 days old) were anesthetized with ether, and a 2- to 3-cm segment of proximal jejunum was ligated in situ. A 0.1-ml sample of either PAP or PIg was injected into the lumen of the segment. After 15, 30, 60, or 120 minutes, pieces of ligated jejunal tissue were excised and fixed in 2 percent buffered glutaraldehyde, and then incubated in a mixture of H_2O_2 and diaminobenzidine (DAB) to reveal the location of HRP (*9*). After postfixation in osmium tetroxide, the tissue was dehydrated, embedded, and sectioned for examination in the electron microscope. We followed a similar procedure in assessing HRP transport in the jejunum of 22-day old rats.

Our results reveal strikingly different patterns for the cellular distribution of HRP between tissue exposed to PAP and control tissue incubated with PIg. Within 30 minutes of exposure to PAP, HRP was identified throughout the columnar epithelial cells of the jejunum (Fig. 1a). Reaction product was found within endocytotic pits at the microvillar surface and within small underlying tubules and vesicles. HRP was also apparent near the Golgi region of the cell in small vesicles that have previously been identified as coated vesicles (*2, 3*). Most important, after incubation for 30 minutes or longer, HRP reaction product was seen with progressively increasing concentration in the extracellular spaces between epithelial cells. Coated vesicles containing antigen were frequently observed in close proximity to the lateral border of epithelial cells and at times appeared to be discharging HRP into the extracellular space. Identical results were obtained when immune complexes were prepared with rabbit IgG, a heterologous Ig that is transported across the intestine of the suckling rat (*1*). Thus, the cellular distribution of HRP that results from the exposure of jejunal cells to PAP is essentially the same as the distribution of maternal IgG in the absence of antigen within these same cells (*2–4*).

In the neonatal tissue exposed to PIg, HRP was found only in apical regions of the cell regardless of incubation time (Fig. 1b). The HRP occurred predominantly in large apical vacuoles and multivesicular bodies and, when compared to PAP-exposed cells, was qualitatively much less abundant overall within the cells. After incubation of the cells with PIg HRP was never observed in the more basal regions of the cells, within coated vesicles, or within the extracellular spaces. When jejunal segments of 22-day-old rats were exposed to either PAP or PIg, HRP was not detected within or on the surface of any of the absorptive cells examined.

To investigate whether immune complex transport in newborn rats is mediated by the Fc region of the IgG molecule as has been shown for the transport of uncomplexed IgG (*3*), we prepared immune complexes from $F(ab')_2$ fragments derived from rabbit antibody to HRP di-

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