Angiogenesis occurs by vessel elongation in proliferative phase human endometrium

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BACKGROUND: Angiogenesis occurs by at least three mechanisms: sprouting, intussusception and elongation. Studies to date have failed to identify the mechanisms or timing of endometrial angiogenesis during the menstrual cycle. The aim of this study was to determine if vessel elongation plays a role in human endometrial angiogenesis.

METHODS: Forty-nine full thickness endometrial sections from 27 hysterectomy samples were immunostained for CD34 to identify blood vessels, and analysed using an interactive computerized stereological program. Based on counts from 9746 individual microscope fields, blood vessel length density (Lv), branch point density (Nv) and mean vessel length per branch point (Lv/Nv) were calculated for three endometrial zones during five phases of the menstrual cycle.

RESULTS: There was an increase in Lv/Nv in the mid–late proliferative compared with early proliferative, early–mid secretory and late secretory phases of the menstrual cycle in the functionalis (mean/SD: 174.5 ± 20.1 versus 76.6 ± 8.4, 118.6 ± 9.4 and 104.2 ± 8.4 µm respectively, P < 0.001) and between the mid–late proliferative and the menstrual phases in the basalis (158.0 ± 18.2 versus 95.4 ± 10.0 µm, P = 0.025). An increase in Lv occurred in the subepithelial capillary plexus in the mid–late proliferative and early–mid secretory phases compared with the early proliferative phase (316.7 ± 32.4 and 338.8 ± 45.3 versus 178.5 ± 8.9 mm/mm3, P = 0.027).

CONCLUSIONS: These data are the first evidence that vessel elongation is a major angiogenic mechanism in mid–late proliferative phase human endometrium.

Key words: angiogenesis/elongation/endometrium/human/stereology

Introduction

Angiogenesis, defined as the development of microvessels from pre-existing blood vessels, can occur by at least three and possibly four different mechanisms. These are sprouting, intussusception, vessel elongation and incorporation of circulating endothelial progenitor cells into growing vessels (Burri and Tarek, 1990; Asahara et al., 1997; Risau, 1997). Sprouting angiogenesis can be divided into a number of steps that include activation of endothelial cells (EC), local degradation of the vascular basement membrane and extracellular matrix, EC proliferation and migration to form a sprout, lumen formation and finally stabilization of the newly formed vessel by recruitment of pericytes and vascular smooth muscle cells. Intussusceptive angiogenesis has been shown to occur in the rat pulmonary circulation (Burri and Tarek, 1990). This type of blood vessel growth has four main phases: creation of an area of contact between opposite endothelial walls, reorganization of the endothelial junctions, formation of the interstitial core of the endothelial post or septum, and remodelling of the newly divided vessel into two discrete capillaries. Elongation or vessel widening is thought to occur as existing vessels restructure in response to the metabolic demands of surrounding cells. This process has also been described as remodelling or pruning (Risau, 1997). Another mechanism that has been described recently is the incorporation of circulating EC (Asahara et al., 1997). The recruitment of circulating cells may contribute to angiogenesis occurring through sprouting, intussusception, or elongation.

Angiogenesis rarely occurs in the adult under normal circumstances, with the exception of the female reproductive organs, where it occurs in the ovary during folliculogenesis and corpus luteum formation, and in the endometrium during the menstrual cycle and pregnancy (Gordon et al., 1995). There are at least three different stages during the menstrual cycle when angiogenesis probably occurs (Rogers and Gargett, 1998). The first is at menstruation when there is repair of ruptured blood vessels. The second is in the proliferative phase, when there is rapid growth of endometrial tissue. The third is during the...
secretory phase when there is development and coiling of the spiral arterioles and growth of the subepithelial capillary plexus. Despite the fact that angiogenesis must occur in the human endometrium during the menstrual cycle, there is only limited evidence as to the mechanisms involved, the exact location and the stages of the menstrual cycle during which angiogenesis is occurring. Immunohistochemical studies have shown that proliferating endometrial EC exist at all stages of the cycle, and within existing endometrial blood vessels rather than associated with vascular sprouts (Gooodger and Rogers, 1994). These observations support, but do not prove, the hypothesis that growth of blood vessels in the endometrium does not occur through sprouting, but possibly through other mechanisms such as elongation and intussusception.

Currently there are no readily accepted methods that can be used to detect the angiogenic mechanisms of elongation or intussusception in tissue sections. The application of stereological methods can, however, provide useful quantitative information about blood vessel formation (Batra et al., 1995).

Previous stereological approaches to estimate capillary length have usually assumed that they are isotropic lineal features. However, capillaries often exhibit marked anisotropy. A structural feature is anisotropic if the statistical averages of a geometric property (such as length) of the structure are not identical in all directions (Wreford, 1995). Anisotropic lineal features can be measured by using isotropic sections or by using fixed orientation vertical sections which are isotropic around a vertical axis. A stereological method for estimating the length of anisotropic features, such as blood vessels, was developed (Gokhale, 1990). This method is unbiased, efficient and also convenient. It involves using light microscopy, vertical sections and intersection counting with an orientated cycloid test system.

By using a stereological approach it is possible to determine both the vessel length density (length per volume or Lv) of endometrial blood vessels and the numerical density (number per volume or Nv) of blood vessel junctions or branches. From these measurements it is possible to calculate the average segment length of the endometrial capillaries (Lv/Nv) in each zone of the endometrium at different stages of the menstrual cycle. If sprouting or intussusception are contributing to endometrial blood vessel growth then this will lead to a greater number of blood vessel junctions and a shorter average microvessel segment length. However, if elongation is occurring there will be an increase in average microvessel segment length with fewer blood vessel junctions per unit length of the vessel.

The aim of the present study was to determine the average length of microvessels, defined as the average distance between vessel branch points, for the subepithelial capillary plexus, functionalis and basalis zones of the endometrium at the different stages of the menstrual cycle. Our working hypothesis was that endometrial blood vessel segment length would increase during proliferative phase endometrial growth, providing evidence for vessel elongation as a mechanism of endometrial angiogenesis.

Materials and methods

Collection and preparation of tissue sections

Endometrial blood vessels were identified by routine immunohistochemistry using a mouse monoclonal anti-human antibody directed against the CD34 antigen (clone QBEND/10; Serotec, Oxford, UK) (Gargett et al., 2001). Briefly, sections were dewaxed in histolene, hydrated and endogenous peroxidase blocked with 3% H2O2 in methanol for 10 min at room temperature. Sections were incubated with 100 µl Maxitags protein blocking agent (Shandon Immunon, Pittsburgh, PA, USA) for 10 min and incubated with mouse anti-human CD34 primary antibody (0.2 µg/ml) for 1 h at 37°C in 1% bovine serum albumin/phosphate-buffered saline. CD34 binds to a surface glycoprotein on the luminal surface of endothelial cells. The secondary antibody and horse-radish–streptavidin (Dako LSAB+ Peroxidase kit; Dako, CA, USA) were used according to instructions from the supplier. Sections were incubated for 10 min with AEC Chromagen (Zymed, CA, USA) at room temperature, then covered-slipped with Faramount aqueous mounting medium (Dako).

Endometrial sections were examined under a light microscope (Olympus BX50) fitted with the stereological image analysis system, Olympus Computer Assisted Stereological Toolbox (C.A.S.T. Grid; Olympus DK A/S, Herstedvester J 29-29, 2620 Albertslund, Denmark). A total of 49 tissue blocks were available from the 27 hysterectomy specimens, with between one and four blocks from each patient. One section was cut from each block. For statistical analysis, the intersection and branch point counts from all sections from each subject were combined. The three zones of the endometrium: subepithelial capillary plexus, functionalis and basalis were mapped using the delineate two-dimensional area function and the ×10 objective. The subepithelial zone, containing the subepithelial capillary plexus, was defined as being ≤200 µm below the surface epithelium. The basalis was defined as being ≤300 µm above the myometrium. The functionalis was selected from the area in between the subepithelial capillary plexus and basalis areas marked. Using the meander sampling function of C.A.S.T. Grid, the x-step length and y-step length were set for each individual to divide the whole section
into contiguous frames. Following this, all the intersections and branch points were counted in each zone of the endometrium throughout the total available section area. On average, 68, 91 and 76 frames were counted on each section for the subepithelial capillary plexus, functionalis and basalis respectively.

**Stereological method**

The stereological method used to measure the length of vessels in three dimensions was based on a published method (Gokhale, 1990) using the cycloid test system with minor modifications as detailed. Values for two parameters were estimated, length density of blood vessels (length per unit volume):

\[ L_v = \frac{2[I_{LC}]_{prj}}{\Delta} \]

and numerical density of branch points (number per unit volume):

\[ N_v = \frac{Q^*}{\Sigma P \cdot a(p) \cdot \Delta} \]

where: \( \Delta \) is section thickness, \([I_{LC}]_{prj} \) is the number of intersections between the projected image of the endometrial capillaries and the cycloid test lines whose minor axis is perpendicular to the vertical, \( Q^* \) is the number of blood vessel branch points counted, \( a(p) \) is the area of reference space associated with each cycloid test line (\( \mu \text{m}^2 \)), and \( \Sigma P \) is the sum of cycloid end points on the reference space multiplied by the length of the cycloid associated with each point (40.5 \( \mu \text{m} \)) and thus provides the total length of cycloid test line on the reference space. Three parameters were recorded in each counting frame: (i) the number of intersections (I) between the cycloids and the projected blood vessels; (ii) the number of cycloid end points located in the relevant zone (p) with each cycloid having two end points; and (iii) the number of blood vessel branch points (Q'), where a branch point was defined as any vessel segment within the section that had three or more distinct lumens or cut ends. Any capillary segment that appeared ambiguous for this criterion was not counted as a branching vessel. The average length of capillary per branch point was determined by dividing the length density of the capillaries by the numerical density of the branch points \((L_v/N_v)\). Note that in this computation the section thickness appears in both the numerator and the denominator and thus cancels out.

**Statistical analysis**

Group results are given as mean ± SEM. Multiple comparisons were made using one-way analysis of variance (ANOVA) in conjunction with post-hoc Tukey’s test (Sigma Stat version 2.0). The level of significance was taken as \( P < 0.05 \).

**Results**

Mean vessel length per branch point \((L_v/N_v)\) in all three endometrial zones (subepithelial capillary plexus, functionalis and basalis) showed a consistent pattern of increasing from the early proliferative to the mid–late proliferative phases of the menstrual cycle, and then reducing from the mid–late proliferative to the early–mid secretory phases of the cycle (Figure 1). By ANOVA, differences in mean vessel length per branch point (vessel segment length) occurred in both the functionalis \((P < 0.001)\) and basalis \((P = 0.025)\). Post-hoc testing demonstrated that in the functionalis mean vessel segment length was significantly greater in the mid–late proliferative phase compared with the early proliferative, early–mi secretory and late secretory phases \((174.5 \pm 20.1 \text{ versus } 76.6 \pm 8.4, 118.6 \pm 9.4 \text{ and } 104.2 \pm 4.1 \mu \text{m respectively})\), while in the basalis, the mid–late proliferative vessel segment length was significantly greater than during the menstrual phase \((158.0 \pm 18.2 \text{ versus } 95.4 \pm 10.0 \mu \text{m})\) (Figure 1).

Length density \((L_v)\) data showed a consistent pattern across the different phases of the menstrual cycle for all three zones of the endometrium (Figure 1), with an increase from early proliferative through to early–mid secretory phases before reducing in the late secretory phase. Despite an almost identical pattern of change in each of the three endometrial zones, these changes were only statistically different in the subepithelial capillary plexus when tested by ANOVA \((P = 0.027)\). Post-hoc testing demonstrated that in the subepithelial capillary plexus, mean vessel length density was significantly greater in the mid–late proliferative and early–mid secretory phases than during the early proliferative phase \((316.7 \pm 32.4 \text{ and } 338.8 \pm 45.3 \text{ versus } 178.5 \pm 8.9 \text{ mm/mm}^3)\).

The numerical density of vessel branch points \(N_v\) was relatively similar across the different stages of the menstrual cycle in all three zones of the endometrium, apart from being consistently higher in the early–mid secretory phase, although this was not statistically significant by ANOVA (Figure 1).

There was no evidence for different blood vessel growth patterns between the three different zones of the endometrium during the different phases of the menstrual cycle (Figure 2). Overall, the subepithelial capillary plexus always had a greater vessel length per branch point than the functionalis or basalis. Of the three endometrial zones, the basalis showed the least variation in vessel length per branch point across the different phases of the menstrual cycle, and the subepithelial capillary plexus the most. The data for \(L_v\) and \(N_v\) showed no consistent pattern of difference between the three endometrial zones (Figure 2).

Given the similar pattern of change in growth between the blood vessels in each of the three endometrial zones across the different phases of the menstrual cycle (Figure 1), and the absence of any statistical difference between the data for blood vessels in each zone (Figure 2), the raw results from each of the zones were combined into a single data group for each patient (Figure 3), giving 27 sets of data on which statistical analyses were performed.

Statistically, the results for the combined blood vessel data are not dissimilar to those for the three individual endometrial zones; however, the overall pattern of change is easier to distinguish (Figure 3). For the whole endometrium, \(L_v/N_v\) was highest in the mid–late proliferative phase of the menstrual cycle and lowest in the early proliferative phase. ANOVA gave a result of \(P < 0.001\), with post-hoc testing demonstrating that mean vessel segment length was significantly greater in the mid–late proliferative phase than at any other stage of the cycle apart from the early–mid secretory. From the combined data, \(L_v\) altered significantly across the menstrual cycle (ANOVA result \(P = 0.033\)), with length density being highest in the mid–late proliferative and early–mid secretory phases. By post-hoc testing, a significant difference was only seen between the early–mid secretory and early proliferative phases. The \(N_v\) data did not show a significant change by ANOVA across the different phases of the menstrual cycle, and was

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Figure 1. Comparison of average vessel length per branch point \( (L_v/N_v) \), vessel length density \( (L_v) \) and vessel branch point density \( (N_v) \) for each endometrial zone. By analysis of variance, average vessel segment length \( (L_v/N_v) \) altered significantly across the different phases of the menstrual cycle in both the functionalis \( (P < 0.001) \) and basalis \( (P = 0.025) \). Comparisons that are significantly different by post-hoc Tukey’s test are shown by \( \diamond \). For \( L_v \), significant changes were seen in the subepithelial capillary plexus across the different phases of the menstrual cycle \( (P = 0.027) \). M = menstrual; EP = early proliferative; M-LP = mid–late proliferative; E-MS = early–mid secretory; LS = late secretory. Values are mean ± SEM.

Discussion

The stereological data obtained in this study provide the first evidence that different mechanisms of angiogenesis are occurring in human endometrium across the different stages of the menstrual cycle. We have made three major findings, each of which has significant implications for understanding how endometrial angiogenesis is regulated. Our primary finding is that vessel elongation is the major mechanism by which endometrial angiogenesis occurs between the early and the mid/late proliferative phases of the menstrual cycle. Our second finding is that blood vessel length density \( (L_v) \) is highest at the mid–late proliferative and early–mid secretory phases of the cycle, demonstrating that between the early proliferative and mid–late proliferative phases new vessel growth, measured on a length per unit volume basis, occurs more rapidly than surrounding tissue growth. Finally, our results show that the patterns of vascular growth at each stage of the menstrual cycle are similar in the subepithelial capillary plexus, functionalis and basalis.

Taken together, the results from this study suggest the following sequence of endometrial angiogenic events during the menstrual cycle. The first major changes in endometrial vascular growth occur between the early and mid–late proliferative phases. At this time the endometrium is growing rapidly under the influence of circulating estrogen, with ultrasound studies reporting an approximate trebling in thickness (Randall et al., 1989; Bakos et al., 1993, 1994). Concomitantly with endometrial growth, our results demonstrate an increase in vessel length density, brought about by a major increase in average vessel segment length. Moving into the early–mid secretory phase, vessel length density remains elevated, but there is a decrease in vessel segment length that occurs due to an increase in vessel junction number \( (N_v) \). There is limited or no change in endometrial thickness at this time (Randall et al., 1989; Bakos et al., 1993, 1994), indicating that the reduction in average vessel segment length must be occurring due to angiogenic remodelling resulting in more vessel junctions, rather than vascular regression. By the late secretory phase, vascular length density has reduced to menstrual/early proliferative levels, suggesting that vascular regression is occurring prior to menstruation.

Elongation has not been identified previously as an
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angiogenic mechanism in endometrium, although other studies have demonstrated that proliferating endothelial cells located within existing vessel walls are associated with vessel elongation (Ausprunk et al., 1974). There is little information available on how the angiogenic mechanism of vessel elongation is regulated. With vascular sprouts it is generally assumed that endothelial cells migrate along a diffusion gradient towards an angiogenic stimulus outside the vessel. It seems unlikely that a similar process would be effective in initiating or regulating elongation of existing vessels. Possible alternatives to an extravascular angiogenesis factor include a direct physical stretch stimulus on the endothelial cells from the rapidly growing tissues that surround the blood vessels, or an angiogenic source within the blood vessels. One such intra-vascular angiogenic source could be intravascular neutrophils. It has been demonstrated that circulating leukocytes contain vascular endothelial growth factor (VEGF) in specific granules, which are released when these cells become activated (Gaudry et al., 1997), and that intravascular neutrophils stain strongly for VEGF in human mucosal chronic inflammatory lesions (Taichman et al., 1997). More recently it has been shown that the presence of VEGF-positive neutrophils inside endometrial blood vessels correlates with endothelial cell proliferation (Gargett et al., 2001). The factors that regulate the recruitment of these VEGF-laden neutrophils are currently unknown; however, it is plausible that rising levels of circulating estrogen, or physical stimuli such as stretching of the vessel by growth of the surrounding tissues, could provoke endothelial cell up-regulation of surface adhesion molecules.

The two angiogenic mechanisms by which an increase in vessel junctions can occur between the mid/late proliferative and the early/mid secretory phases of the cycle are either sprouting or intussusception. We have previously speculated that sprouting is not a major endometrial angiogenic mechanism (Gargett and Rogers, 2001), based on three lines of evidence. Firstly, it has not been possible to identify structures that might be sprouts, although recently limited evidence for some endometrial vascular sprouts has been published (Ono and Shiina, 2001). Secondly, proliferating endothelial cells always appear within existing vessel profiles rather than associated with sprouts (Rogers and Gargett, 1998), and finally, αβ

Figure 2. Comparison between the three different endometrial zones (SCP = subepithelial capillary plexus; Funct = functionalis; and Basalis) for average vessel length per branch point (Lv/Nv), vessel length density (Lv) and vessel branch point density (Nv) for different phases of the menstrual cycle. Statistical analysis showed no differences between any of the groups. Values are mean ± SEM.
seems likely that intussusception is the main angiogenic mechanism by which new vessel junctions arise in the early–mid secretory phase. Based on ultrastructural data, endometrial endothelial cells undergo cytoplasmic growth during the secretory phase and some enlarge to almost fill the capillary lumen (Roberts et al., 1992). This description is consistent with vessels undergoing intussusception. Whatever the angiogenic mechanism at this time, there is probably also generalized vascular remodelling as the endometrium differentiates and the glands increase in volume. The observation that endothelial cell proliferation continues at approximately similar levels throughout the menstrual cycle, with no obvious times of increase or decrease (Goodger and Rogers, 1994; Rogers et al., 1998), is consistent with the concept of secretory phase intussusception and vascular remodelling, without any net increase in total endometrial blood vessel length density occurring.

There have been a large number of published studies investigating endometrial expression of angiogenic factors and their receptors in human endometrium (Rogers and Gargett, 1998; Smith, 2001). Molecules most recently investigated include (but are not limited to) VEGF-A, VEGF-B, VEGF-C, VEGF-D, the VEGF receptors VEGFR1, VEGFR2, VEGFR3, basic and acidic fibroblast growth factor (FGF), FGF receptor-1 and -2, epidermal growth factor (EGF), EGF receptor, angiopoietin-1 (ang-1), ang-2 and the ang receptors Tie-1 and Tie-2 (Li et al., 2001; Möller et al., 2001; Sandberg et al., 2001). Despite extensive categorization of the expression patterns of these factors and their receptors throughout the menstrual cycle, no pattern has emerged that is consistent with the presumed times of endometrial vascular growth. Given that the present study demonstrates major angiogenesis by vessel elongation in the mid–late proliferative phase, and vascular remodelling to increase vessel junction number in the early–mid secretory phase, the challenge now is to identify which factors and receptors play central roles in regulating endometrial vascular growth.

The current study found no differences in vessel segment length between the subepithelial capillary plexus, the functionalis and the basalis. We had hypothesized that angiogenesis might be regulated differently in these three areas of the endometrium, based on functional differences between the zones and preliminary data from an earlier study (Dockery et al., 2000). What is perhaps most surprising is the finding that basalis blood vessels increase in segment length and length density to a similar extent to those in the functionalis. It is generally assumed that the functionalis regrows from the basalis during the early proliferative phase, with very few data being available on the growth dynamics of the basalis itself.

Limited data indicate that endothelial cell proliferation rates are significantly lower in the basalis than functionalis (Ferenczy et al., 1979; Rogers et al., 1998). The present results demonstrate that the basalis, defined as the 300 µm of endometrium closest to the myometrium, undergoes a growth pattern similar to the functionalis, at least as far as vessel elongation is concerned.

One of the limitations of the current study is that the stereological techniques used only show net changes in vessel segment length, and do not give an insight into the possibility that elongation, intussusception and possibly sprouting angiogenesis may all be occurring simultaneously. The net increase in average vessel segment length in the mid/late proliferative phase is clear evidence that elongation is the predominant mechanism by which angiogenesis is occurring at this time, but it is still possible that intussusception is also occurring, but to a much lesser extent. The fact that vessel segment length decreases again in the early/mid proliferative phase suggests that intussusception has become the major mechanism of angiogenesis at this time, but elongation may still be occurring. It is unlikely that the relative contributions of elongation, intussusception and sprouting to overall angiogenesis will be elucidated until satisfactory markers for each mechanism are established.

The estimates of capillary segment length from the current study are comparable with those reported for other tissues. The mean length of the nailfold capillary loop has been reported as 234.9 ± 5.8 µm (Rouen et al., 1972). Another

**Figure 3.** Comparison of average vessel length per branch point (L_v/N_v), vessel length density (L_v) and vessel branch point density (N_v) achieved by combining all the data for each of the three endometrial zones (see results section). By analysis of variance, average vessel segment length altered significantly across the different phases of the menstrual cycle ($P < 0.001$). Comparisons that are significantly different by post-hoc Tukey's test are shown by $\diamond$. For $L_v$, significant changes were also seen across the different phases of the menstrual cycle ($P = 0.033$). M = menstrual; EP = early proliferative; M-LP = mid–late proliferative; E-MS = early–mid secretory; LS = late secretory. Values are mean ± SEM.
study reported the mean length of capillaries on the fourth finger of both hands to be \(~215 \pm 40\) \(\mu m\) (Kabasaki et al., 1996). More recently, the mean length of cutaneous microvessels of the skin from control patients was found to range from 110 to 270 \(\mu m\) (Ohtsuka et al., 1998). It is important to note that in the current study we have measured vessel length from 110 to 270 vessels of the skin from control patients was found to range from 1996). More recently, the mean length of cutaneous microvessels has been up to 33\% (by volume) during \(\mu m\) (Nyengaard, 1993).

It is interesting to speculate on the reasons that underlie the changes in vessel segment length during the menstrual cycle. Presumably, a number of physical and biological factors govern optimal capillary segment length. Physical parameters will include length, diameter and tortuosity of the vessel, all of which influence resistance to flow. Blood pressure differential across the length of the capillary will also strongly affect blood flow rate. Biological factors will include metabolic demand of the tissue, which in turn is influenced by many factors. We have already suggested that the increase in vessel segment length during the proliferative phase may be the most economical solution for the vasculature as it keeps pace with demand of the tissue, which in turn is in

\[ \text{blood flow} \times \text{length} = \text{blood flow rate} \]

Physical parameters will include length, diameter and tortuosity of the vessel, all of which influence resistance to flow. Blood pressure differential across the length of the capillary will also strongly affect blood flow rate. Biological factors will include metabolic demand of the tissue, which in turn is influenced by many factors. We have already suggested that the increase in vessel segment length during the proliferative phase may be the most economical solution for the vasculature as it keeps pace with the rapid growth of the surrounding endometrial tissues. However, once endometrial growth ceases, it is possible that physical factors such as resistance to flow caused by the longer capillary loops initiates vascular remodelling that returns vessel segments to an optimal length for a given tissue. In support of this concept, it has been reported that when glomerular capillary length reaches a certain limit, a new capillary loop is generated so as not to increase the resistance to flow (Nyengaard, 1993).

Very recently, evidence has emerged for organ-specific angiogenesis factors with the identification of endocrine gland vascular endothelial growth factor (LeCouter et al., 2001). Our stereological data demonstrating a specific episode of endometrial angiogenesis by vessel elongation in the mid–late proliferative phase, followed by a reduction in mean vessel segment length in the early–mid secretory phase, lend support to the emerging paradigm of organ-specific angiogenesis. Given the generally disappointing results obtained with systemic administration of angiogenesis inhibitors in different clinical trials to date (Thompson et al., 2000), the emergence of organ specific mechanisms and mitogens opens new possibilities for the effective clinical manipulation of new vessel growth.

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References


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