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In vitro and *in vivo* angiogenic assays

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The formation of new vessels in a tumour requires a number of steps including chemotaxis, migration, proliferation and tubular formation of the endothelial cells. Thus, in vitro assays, which dissect out each of these steps or a combination of these, can be designed. For example, tubular formation assays can be conducted using collagen gels as matrix and angiogenic factors such as VEGF and bFGF as stimuli. In addition to studying these functions of the endothelial cells under various conditions, these assays can also be used to test the importance of different molecules including molecules with a potential inhibitory effect on tumour angiogenesis. The next question that arises is which endothelial cells to use. HUVEC (human umbilical vein endothelial cells) can be obtained from commercial sources and these cells can grow for approximately 15 to 20 passages. Another possibility is to establish primary cultures of endothelial cells. We have used this technique to establish primary lung endothelial cell cultures from wild-type mice and from knock-out mice. The resulting cell lines can then be compared using some of the above-mentioned in vitro assays. A large number of in vivo angiogenesis assays have been described. The more common ones are implantation of bFGF or VEGF pellets either just subcutaneously, in a dorsal air sack or embedded in Matrigel. Other models include wound healing, retina damage etc. However, caution should be taken regarding extrapolating results from assays including non-malignant conditions to tumour angiogenesis. The mediators of tumour angiogenesis may very well be different from those mediating angiogenesis in non-tumour conditions.