1. INTRODUCTION

Tumor hypoxia, or the condition of low tumor oxygenation, has been a focus of considerable debate in radiation therapy for almost 50 yr since the pioneering work of Gray and colleagues (1) demonstrating an oxygen dependency in the radiosensitivity of cells and tissues. During this period, interest among researchers has waxed and waned as promising new directions emerged from the laboratory, only to fail in clinical trials. However, with the emergence of new concepts and the development of new tools, the prospect of targeting tumor hypoxia and identifying patients who would most benefit from this approach appears more tangible clinically. This chapter discusses the significance of tumor hypoxia in head and neck squamous cell carcinomas (HNSCC) as well as past, present, and future strategies for targeting this microenvironmental factor.

2. TUMOR HYPOXIA AND MALIGNANT PROGRESSION

Poorly oxygenated regions develop within solid tumors because of aberrant blood vessel formation, changes in blood flow from intermittent closure of existing blood vessels, and increasing tumor oxygen demands for growth (2) (Fig. 1). The existence of hypoxia in human tumors was suggested in 1955 by Thomlinson and Gray (3), who showed with histological sections that there was a constant distance (100–150 μm) across tumor tissues between blood vessels and necrosis and that this distance was the oxygen diffusion distance based on capillary oxygen partial pressure and cellular oxygen consumption. They postulated that hypoxic cells existed adjacent to necrotic areas, just beyond the oxygen diffusion distance. This is now known as chronic hypoxia. A second form of hypoxia, known as acute hypoxia, also exists owing to fluctuating flow in tumor blood vessels. This was first postulated by Brown (4) and subsequently demonstrated in transplanted mouse tumors using injections of two different diffusible dyes minutes apart showing that temporary reduction in flow or closure of blood vessels can be observed in solid tumors, resulting in areas of acutely hypoxic cells (5). It is likely that acute and chronic hypoxia are the extremes of a continuum caused by the dynamic nature of tumor blood flow and that both can give rise to tumor cells that are prone to metastasis and resistant to conventional therapy.
Laboratory studies have indicated that tumor hypoxia can play an important role in regulating cell viability and promoting cell metastatic potential. Graeber et al. (6) have shown that hypoxia induced apoptosis in minimally transformed mouse embryo fibroblasts with normal p53 function, but not in cells with mutant p53 proteins in both cell culture and transplantable tumors. These data suggest that hypoxia may exert a physiologic pressure for selection by clonal expansion of mutant p53 tumor cells, which, in HNSCC, have been shown to behave more aggressively than their wild-type counterparts (7). Young et al. (8) observed that cells treated with hypoxia were more likely to invade the lungs of recipient mice than untreated cells. Likewise, acute hypoxia has been shown to enhance the formation of spontaneous nodal metastases in an orthotopic murine model or cervical carcinomas (9). Clinical studies have supported the link between hypoxia and tumor metastasis: studies of soft tissue sarcomas and carcinomas of the cervix have shown that hypoxia is an independent and significant prognostic factor that correlates with metastatic spread (10–12).

At the molecular level, multiple stress-response pathways are turned on when cells are exposed to hypoxia. Changes in the expression of genes and proteins are important for hypoxia-induced cellular adaptation to an anaerobic environment. One of the most well-described oxygen-response pathways is mediated by the hypoxia-inducible factor (HIF-1), which has been shown to play an important role in tumor development (13,14). HIF-1 regulates genes that are involved in metabolism, angiogenesis, invasion, metastasis, and apoptosis, all of which can influence tumor growth and metastasis (Fig. 2). One of the most important HIF-1 targets is vascular endothelial growth factor (VEGF), which is a proangiogenic protein that has been implicated in poor prognosis in head and neck cancer (15,16). Rapidly advancing knowledge of hypoxia-regulated genes and proteins via proteomic and genomic approaches will provide a better understanding of the molecular basis of hypoxia and give rise to novel concepts for exploiting this microenvironmental factor.

**Fig. 1.** Schematic representations of tumor hypoxia and necrosis that resulted from differences in the vasculature between tumor and normal tissues. (Reprinted from ref. 2, with permission from the American Association for Cancer Research.)