Heparin-binding growth factors and their receptors

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Abstract

Heparin-binding growth factors modulate diverse biological activities including cellular proliferation, cellular differentiation, morphogenesis, and angiogenesis. Biochemical characterization for two members of the heparin-binding growth factor family, acidic and basic fibroblast growth factors, is extensive, while characterization of the remaining five members is forthcoming. Cell surface receptors have been identified for acidic and basic fibroblast growth factors, but little is known concerning their sites of action in vivo or the mechanisms involved in transducing the energy of growth factor binding to a biological response. An understanding of the biological basis for the diversity of the heparin binding growth factor family and the in vivo actions of these factors will prove a major challenge to future research efforts.

Introduction

Elucidation of the mechanisms that control cellular growth and differentiation is a major focus of biomedical research. The observations that oncogenes can be either altered forms of growth factors or their cognate receptors has underscored the importance of such factors in maintenance of normal cellular growth controls (reviewed in Bishop, 1983; Deuel, 1987; Yarden and Ullrich, 1988). Because most growth factors are potent mediators of cellular growth, they are often present at very low concentrations making biochemical characterization inherently difficult. In 1984, a growth factor from chondrosarcoma cells was shown to bind avidly to heparin affinity columns, thereby facilitating its purification (Shing et al., 1984). Shortly after, acidic and basic growth factors isolated from bovine brain were also shown to bind avidly to heparin (Bohlen et al., 1984; Lobb and Fett, 1984; Thomas et al., 1984). Until 1986, the heparin-binding growth factor family consisted of two mitogens known as heparin binding growth factor I (HBGF I) or acidic FGF (aFGF), an anionic factor, and heparin-binding growth factor II (HBGF II) or basic FGF (bFGF), a cationic growth factor (reviewed in Baird et al., 1986; Folkman and Klagsbrun, 1987; Gospodarowicz et al., 1987; Burgess and Maciag, 1989). Many partially characterized growth factor activities were subsequently found to fall into one of these two classes of HBGFs. These factors possess a broad spectrum of biological activities and affect cellular proliferation, cellular
differentiation and morphogenesis (reviewed in Burgess and Maciag, 1989). Within the last two years, five additional members of the HBGF family have been identified. The five new members of the HBGF family increased the already diverse spectrum of biological activities reported for aFGF and bFGF. Identifying the sites of action and elucidating the molecular mechanisms involved in HBGF signal transduction will undoubtedly be the focus of future research.

Identification and structure of the HBGF family

Acidic FGF and basic FGF were first purified to homogeneity by classical biochemical methods and shortly thereafter were found to bind avidly to heparin affinity columns, which greatly simplified their purification (Bohlen et al., 1984; Lobb and Fett, 1984; Lobb et al., 1986; Thomas et al., 1984). Sequencing of the proteins led to isolation of cDNAs encoding both factors and clearly demonstrated they are closely related but distinct gene products, exhibiting 53% sequence identity (Abraham et al., 1986; Jaye et al., 1986). Purified preparations of aFGF and bFGF appear as microheterogeneous proteins differing in their N-terminal amino acid sequences. For aFGF and bFGF, acid-activated proteases may be involved in the generation of N-terminally deleted proteins (Klagsbrun et al., 1987). The biological activity of N-terminally deleted FGFs is identical to the parental factor. Whether the N-terminal deletions are artifacts of purification or in vivo processing events remains unresolved. Alternate translation start sites at CUG codons occurring 5' to the methionine AUG codon are responsible for the generation of at least four distinct bFGF proteins (Florkiewicz and Sommer, 1989; Prats et al., 1989). The biological function of alternative translational initiation is not apparent, since all forms of bFGF appear to have similar biological potencies. Similar high molecular weight forms of aFGF have not yet been detected.

Both aFGF and bFGF mediate their biological responses via cell surface receptors, however, neither factor possesses a recognizable signal peptide sequence (Abraham et al., 1986; Jaye et al., 1986) and thus the mechanisms involved in FGF secretion are not understood. Constitutive expression of a bFGF gene, in which a signal secretory peptide sequence was fused to bFGF coding sequences, in cells results in the secretion of biologically active bFGF that induces cellular transformation in culture (Blam et al., 1988; Rogelj et al., 1989) and tumorigenesis in vivo (Rogelj et al., 1989). Expression of bFGF genes lacking secretory signal peptide has minimal effects, but can induce some phenotypic changes characteristic of cellular transformation (Blam et al., 1988; Neufeld et al., 1988; Rogelj et al., 1989; Sasada et al., 1988). If aFGF and bFGF are secreted by as yet unidentified mechanisms, it is likely that the secretion of these potent factors is under strict regulatory controls.

Since 1986, five additional members of the heparin-binding growth factor family have been identified. The int-2 (Dickson and Peters, 1987), hst (Tiara et al., 1987; Yoshida et al., 1987) or k-FGF (Delli-Bovi et al., 1987), and FGF-5 (Zhan et al., 1988) oncogenes exhibit 40 to 55% sequence identity with aFGF and bFGF (Table 1), while two other factors FGF-6 (Marics et al., 1989), isolated by screening murine cDNA libraries with hst cDNA, and KGF, isolated as a keratinocyte cell growth factor (Rubin et al., 1989), do not yet have identified oncogenic activities. In contrast to aFGF and bFGF, the primary translation products of k-FGF, hst, and FGF-5 all contain a hydrophobic leader sequence that is likely to serve as a signal peptide and promote secretion (Delli-Bovi et al., 1987; Yoshida et al., 1987; Zhan et al., 1988). This prediction has been confirmed for K-FGF, which is secreted from NIH 3T3 and COS-1 cells as a processed glycoprotein that appears smaller than in vitro translation products (Delli-Bovi et al., 1987). Both K-FGF and FGF-5 are stabilized by the addition of heparin to conditioned medium, identifying these factors as structural and functional members of the heparin-binding growth factor family (Delli-Bovi et al., 1987; Zhan et al., 1988).