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Review

Structural biology of glycoprotein hormones and their receptors: Insights to signaling ☆

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ABSTRACT

This article reviews the progress made in the field of glycoprotein hormones (GPH) and their receptors (GPHR) by several groups of structural biologists including ourselves aiming to gain insight into GPH signaling mechanisms. The GPH family consists of four members, with follicle-stimulating hormone (FSH) being the prototypic member. GPH members belong to the cystine-knot growth factor superfamily, and their receptors (GPHR), possessing unusually large N-terminal ectodomains, belong to the G-protein coupled receptor Family A. GPHR ectodomains can be divided into two subdomains: a high-affinity hormone binding subdomain primarily centered on the N-terminus, and a second subdomain that is located on the C-terminal region of the ectodomain that is involved in signal specificity. The two subdomains unexpectedly form an integral structure comprised of leucine-rich repeats (LRRs). Following the structure determination of hCG in 1994, the field of FSH structural biology has progressively advanced. Initially, the FSH structure was determined in partially glycosylated free form in 2001, followed by a structure of FSH bound to a truncated FSHR ectodomain in 2005, and the structure of FSH bound to the entire ectodomain in 2012. Comparisons of the structures in three forms led a proposal of a two-step monomeric receptor activation mechanism. First, binding of FSH to the FSHR high-affinity hormone-binding subdomain induces a conformational change in the hormone to form a binding pocket that is specific for a sulfated-tyrosine found as sTyr 335 in FSHR. Subsequently, the sTyr is drawn into the newly formed binding pocket, producing a lever effect on a helical pivot whereby the docking sTyr provides as the 'pull & lift' force. The pivot helix is flanked by rigid LRRs and locked by two disulfide bonds on both sides: the hormone-binding subdomain on one side and the last short loop before the first transmembrane helix on the other side. The lift of the sTyr loop frees the tethered extracellular loops of the 7TM domain, thereby releasing a putative inhibitory influence of the ectodomain, ultimately leading to the activating conformation of the 7TM domain. Moreover, the data lead us to propose that FSHR exists as a trimer and to present an FSHR activation mechanism consistent with the observed trimeric crystal form. A trimeric receptor provides resolution of the enigmatic, but important, biological roles played by GPH residues that are removed from the primary FSH-binding site, as well as several important GPCR phenomena, including negative cooperativity and asymmetric activation. Further reflection pursuant to this review process revealed additional novel structural characteristics such as the identification of a 'seat' sequence in GPH. Together with the 'seatbelt', the 'seat' enables a common heteodimeric mode of association of the common α subunit non-covalently and non-specifically with each of the three different β subunits. Moreover, it was possible to establish a dimensional order that can be used to estimate LRR curvatures. A potential binding pocket for small molecular allosteric modulators in the FSHR 7TM domain has also been identified.

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1. Introduction

The glycoprotein hormone (GPH) family consists of the three gonadotropins, luteinizing hormone (LH), follicle-stimulating hormone (FSH) and chorionic gonadotropin (CG), and a fourth non-gonadotropin member, thyroid-stimulating hormone (TSH). All four members are important pharmaceutical drugs (PDR, 2013). FSH is clinically used for controlled ovarian stimulation in women undergoing assisted reproduction, most commonly involving in vitro fertilization of retrieved oocytes. It is also used to treat anovulatory infertility in women and hypogonadotropic hypogonadism in men, while LH is used to support FSH therapy. CG is used to induce ovulation in women and to increase sperm count in men, as well as to treat young boys when their testicles do not normally descend into the scrotum. TSH, in combination with ^{131}I, is administered to post-surgery thyroid cancer patients to suppress and ablate remnant cancerous tissues. Despite decades of successful clinical use and multi-billion-dollar annual sales, it remains poorly understood how glycoprotein hormones activate their receptors in host cells at an atomic level. In this article, we review the progress made by several groups, including ourselves, in the field of the structural biology of glycoprotein hormones and their receptors in an attempt to provide an insightful picture which portrays how FSH binding leads to FSHR activation at the atomic level.

GPHs belong to the superfamily of cystine-knot growth factors (CKGF). FSH, LH and TSH are all secreted from anterior pituitary gland as heteodimeric (two dissimilar subunits) glycoproteins of ~30 kDa. Each is composed of a common α-subunit with the same amino acid sequence and a hormone-specific β subunit. Their secretion is controlled by releasing hormones from the hypothalamus. Specifically, gonadotropin-releasing hormone (GnRH) controls the secretion of FSH and LH, and thyroid-releasing hormone (TRH) controls TSH (Simoni et al., 1997; Szkudlinski et al., 2002; Tao and Segaloff, 2009; Ulloa-Aguirre et al., 2007; Vassart and Costagliola, 2011). Acting to control thyroid functions, TSH induces production of thyroxine (T4) and triiodothyronine (T3), two molecules that are required for metabolism in almost every tissue in the human body (Porter, 2011). FSH and LH act synergistically to regulate follicular growth and ovulation, respectively, in ovaries and maintenance of normal sperm quality and quantity in testis. Another glycoprotein hormone, human chorionic gonadotropin (hCG) is secreted by human placenta during early pregnancy, and acts on the corpus luteum of pregnancy inducing progesterone production, which plays a critical role in maintaining pregnancy (Pierce and Parsons, 1981). CG and LH are directly related in evolutionary origin, as the CG beta subunit gene evolves from the LH beta-subunit gene by duplication and subsequent reading through into the 3′-untranslated region in the same chromosome location (19q13.2 for human) (Fiddes and Goodman, 1980; Talmadge et al., 1984). The characteristics in sequence and function shared by the four members indicates their common evolutionary origin (Uchida et al., 2010).

GPHs exercise their biological function upon interacting with their cognate receptors. Like the hormones, their receptors are also closely related. LH and CG share the same receptor, LHR; FSH binds to FSHR and TSH to TSHR (Combarnous, 1992; Dias, 1992; Nagayama and Rapoport, 1992; Segaloff and Ascoli, 1993). These receptors belong to the leucine-rich-repeat-containing G-protein coupled receptor (LGR) subfamily (Hsu et al., 1998, 2000). The LGR subfamily, in turn, belongs to Family A of the G-protein cou-