S-3 Domains Determining FSH/LH Specificity in the Three Dimensional Conformation of the LH β Subunit Molecule: A Comparative Approach from Receptor Binding Specifi-city of Chicken LH

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We recently found that chicken LH strangely bound specifically to the rat testicular FSH receptor with a higher affinity than chicken FSH(Miya and Ishii, 1994). Rat LH, of course, hardly binds to this receptor. To explain this strange fact, we carefully examined the primary structure of the chicken LH β-subunit molecule which we previously estimated from the nucleotide sequence of cDNA encoding the LH β-subunit molecule precursor molecule(Noce *et al*, 1989). The primary structure of the chicken LH β-subunit molecule was also compared with those of LH β-subunit molecule has common amino acid residues in a number of positions with LH β-subunit molecules of the other vertebrate species, mostly mammals, but most amino acid residues in certain regions of the molecule, position 40 to 50 and 101 to 106, are not common with the corresponding residues of LH but common with those of FSH of various species as shown in the figure given below(the FSH common residues are underlined).

		40 48	100 110
LH	bovine	S M K R V L P V I	C G G P R T Q P L A C
LH	sheep	S M K R V L P V I	C G P G R T E P L A C
LH	pig	S M V R V L P A A	C G G P R A Q P L A C
LH	rat	S M V R V L P A A	C G G P R T Q P M T C
LH	horse	S M V R V M P V I	C G V F R D Q P L A C
LH	human	T M M R V L Q A V	C G G P K D H P L T C
LH	chicken	T R E P V Y R S P	C T V Q G L G P A F C
FSH	bovine	T R D L V Y R D P	C T V R G L G P S Y C
FSH	sheep	T R D L V Y K D P	C T V R G L G P S Y C
FSH	pig	T R D L V Y K D P	C T V R G L G P S Y C
FSH	rat	T R D L V Y K D P	C T V R G L G P S Y C
FSH	human	T R D L V Y K D P	C T V R G L G P S Y C
FSH	bulfrog	T K D A N L M Y P	C T V R A L G P T V C

We postulated from these results that two regions are the sites for recognition of FSH in the molecule. Dias *et al.*(1994) reported in their study using cimeric human FSH that human FSH β residues, ^{95}TVRGLG^{100}, which correspond to the residues ^{101}TVQGLG^{106} in the chicken LH β, are important for FSH binding specificity. Our conclusion completely coincides with their conclusion. This region locates just next to the determinant loop which is known to be important in the

receptor interaction. The other region, position 50-60, is included in the large loop which was also reported to participate in the receptor interaction.

Recently, we were interested in predicting the three dimensional conformation of the β-subunit molecule from the primary structure and constructed a three dimensional model of the molecule by using the method developed by Wako and Blundel(1994).

After we submitted this paper to a journal in 1994, two papers on the determination of the conformation of the hCG molecule were published(Lapthorn *et al.*, 1994; Wu *et al.*, 1994). Their results well coincided with ours(Wako and Ishii, 1995).

We further allocated the two regions which we consider important for the FSH specificity in the three dimensional model of the gonadotropin/thyrotropin molecule. All these two regions as well as regions in α and β subunit molecules of gonadotropins reported to be important for the receptor interactions locatd in a place in close contact.

We are now attempting to predict the three dimensional conformation of the extra-cellular receptor binding domain in the gonadotropin receptor molecule. By combining both conformation results, we may visualize the hormone-receptor interaction.