P27 Relationship between genotypes of FSH receptor gene and ovarian response related to the controlled ovarian hyperstimulation protocols

Lee H-S[1], Cha SH[2], Song IO[2], Koong MK[2], Kang IS[2], Jun JH[1]

[1]Laboratory of Reproductive Biology and Infertility, 2Department of OB/GYN, Cheil General Hospital, Sungkyunkwan University School of Medicine, Seoul, Korea

Objectives: The FSH receptor is a main target of controlled ovarian hyperstimulation (COH) for successful outcome of human IVF-ET program. Recently, it have been reported that single nucleotide polymorphism (SNP) in FSH receptor (FSHR) gene could predict patterns of ovarian response by endogenous and exogenous FSH. In this study, we evaluated the association of FSH receptor SNP with ovarian response and clinical outcome of COH cycles in patients undergoing IVF.

Materials and Methods: Genomic DNA was extracted from patients' peripheral blood and $Thr^{307}Ala$ (T/A) and $Asn^{680}Ser$ (N/S) of SNP in FSHR gene were screened by PCR-RFLP. The patients (n=53) were selected for this study who had performed both short and long COH protocols in our hospital. The subjects were grouped into TA/NS (n=28), TT/NN (n=20) and AA/SS (n=5) based on their FSHR-SNP genotypes. Clinical parameters in COH cycles were compared according to the FSHR-SNP genotypes.

Results: Estradiol level of short protocol at the day of hCG administration was higher than that of long protocol in TA/NS genotypes (2,512 ± 1360 vs 1,980 ± 1194 pmol/L, p=0.07), while they were similar between long and short protocols in TT/NN and AA/SS groups. In addition, the incidence of high responder cycles (> 3,000 pmol/L of estradiol) in short protocols (37.1%, 13/35) was higher than that of long protocols (17.5%, 7/40) in TA/NS genotypes (p=0.07). There was no significant difference in number of retrieved oocytes, fertilization rates and pregnancy rate between short and long protocols in all three genotypes examined. Delivery rate per patient with long protocols was also significantly higher than that with short protocol in TA/NS genotypes (35.0% vs 10.0%, p=0.03).

Conclusions: This study showed that the genotypes of FSHR could be related to differential ovarian response by short or long COH protocols in human IVF-ET program. We suggest that the long protocol for TA/NS genotypes could lower the risk of ovarian hyperstimulation syndrome and provide higher delivery rate. This finding should be further evaluated with large sample size and other populations.

Key words: Genotype, FSH receptor, Controlled ovarian hyperstimulation protocols, Ovarian hyperstimulation syndrome, Delivery rate

P28 Prolonged culture duration alters proliferation and differentiation characteristics of human embryonic stem cells

Park YB[1], Kim YY[2], Chung SG[1], Oh SK[2,3], Ku SY[2,3], Kim SH[2,3], Choi YM[2,3], Moon SY[2,3]

[1]Central Research Institute, Sam Jin Pharm. Co. LTD., [2]Institute of Reproductive Medicine and Population, Medical Research Center, [3]Department of Obstetrics and Gynecology, College of Medicine, Seoul National University, Seoul, Korea

Objectives: Despite extensive studies on human embryonic stem cells (hESCs), controversy still exists over changes in proliferation and differentiation characteristics. The authors investigated proliferation rate and differentiation potential of hESCs of various culture periods.

Materials and Methods: In this study, the authors classified hESCs into three groups according to culture periods: early passage (passage up to 40), middle passage (passages up to 90), and late passage (passage up to 130). Ten colonies of each passage were measured the surface area daily and population doubling time was assessed. Proliferation of hESCs was measured by BrdU assay and telomerase activity. Number of cells expressing undifferentiated hESC markers was examined by FACS. Levels of Oct-4 and Nanog were compared using RT-PCR among the three differentiation stage: undifferentiated hESCs, embryoid body (EB), and replating EB. Expression of Oct-4 and Nestin in replating EBs was examined with fluorescence microscope.

Results: Population doubling time of hESCs in early passage was longer than those of middle and late passage. BrdU assay results indicated that proliferation of early and late passage was different. Proliferation activity of hESCs accelerated as the passage number increase. Telomerase activity was not different in early and late passaged hESC. However, it was decreased in differentiated hESCs. Cellular morphology was very different among passages of hESC after attachment culture of EBs, hESCs of late passage had expressed higher levels of undifferentiated markers such as Oct-4 and Nanog than that of earlier passage even if differentiation was induced.

Conclusions: It can be inferred that hESCs show accelerated proliferation rate and decreased differentiation potential as passage number increases. From our results, hESCs of early passage might be considered more adequate than that of middle and late passage in terms of hESC differentiation studies.

Key words: human embryonic stem cell, proliferation, differentiation, prolonged culture

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