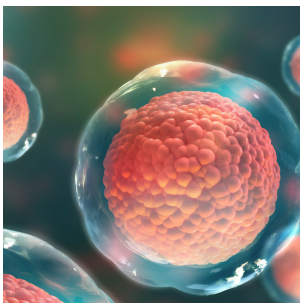
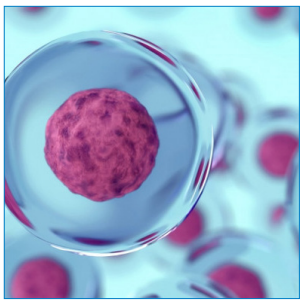
Accepted Abstracts

Diabetes 2022

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An autofluorescence-based isolation of Leydig cells for testosterone deficiency treatment

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Statement of the Problem: Testosterone deficiency (TD) occurs when the serum testosterone levels are insufficient and can cause a series of clinical symptoms, including sexual dysfunction, obesity, muscle weakness and osteoporosis. Leydig cells (LCs) produce more than 95% of serum testosterone and the in-depth study of biological characteristics and regulatory mechanisms of LCs on testosterone production is helpful to elucidate the pathogenesis of TD. However, the density gradient centrifugation, as the currently main method for LC isolation, remain challenging. The purpose of this study is to identify the testicular autofluorescent cells and describe a simple and effective autofluorescence-based method for isolating LCs. Methodology: Testicular autofluorescent cells were isolated by FACS, and identified by qRT-PCR analysis and immunofluorescence staining. Then, the autofluorescent cells were further divided into two subpopulations by the combination of two fluorescence channels in FACS. The immunofluorescence staining, Live/Dead assay, cell counting, substrate utilization assay and LH stimulation assay were respectively used to evaluated the purity, viability, quantity and function of obtained LCs. Finally, the isolated LCs were subcutaneously implanted into castrated mice to evaluate the therapeutic potential of LCs *in vivo*. Findings: Testicular autofluorescent cells were composed of macrophages and LCs. Our autofluorescence-based method by the combination of two fluorescence channels successfully purified LCs from macrophages. Of note, the isolated LCs had high purity (>98%), viability (>98%) and quantity (approximately 4 × 105 cells per mouse) and maintained intact biochemical function. Moreover, subcutaneous transplantation of isolated LCs could relieve the symptoms of TD in castrated mice. Conclusion & Significance: we established a simple autofluorescence-based method that allows efficient isolation of well-functioning LCs with high purity, viability and quantity. This method can be used in detailed biological studies of LCs and will promote further advances in LC replacement therapies for TD.

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Magnesium sulfate improves insulin resistance in type 2 diabetic parents and their offspring in both animal model and diabetic patients

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The role of magnesium sulfate ($MgSO_4$) in attenuates insulin resistance (IR) in type 2 diabetic (T2D) patients and the reduction of the risk of IR in liver and muscle of their offspring. T2D was induced by high fat diet and 35 mg/kg of streptozotocin. The male and female diabetic rats were then divided into three groups: CD, Mg, and insulin. NDC group received a normal diet. All the animals were studied for a six-month. Their offspring were just fed with normal diet for four months. Blood glucose was measured weekly in patients and their offspring. Intraperitoneal glucose tolerance test (IPGTT), urine volume, and water consumption in both patients and their offspring were performed monthly. The hyperinsulinemic euglycemic clamp in both patients and their offspring was done and blood sample collected to measure hemoglobin A1c (HbA1c) and plasma lipid profile. IRS1, Akt and GLUT4 gene expressions in muscle were evaluated in all the groups. FOXO1 and phosphoenolpyruvate carboxykinase gene expressions also measured in all groups. Plasma and liver lipid profile and liver glycogen level were measured in all groups. $MgSO_4$ or insulin therapy decreased blood glucose, IPGTT, plasma lipid level and HbA1c in patients and their offspring compared to DC group. They also increased glucose infusion rate in patients and their offspring. IRS1, Akt and GLUT4 gene expressions improved in both patients in comparison with DC group. $MgSO_4$ could decreased gluconeogenesis pathway in both parents and their offspring. $MgSO_4$ exerts beneficial effects on IRS1 and Akt gene expressions in Mg treated offspring. $MgSO_4$ therapy improved insulin resistance in diabetic patients by increasing the expression of GLUT4 in the muscle and reduce gluconeogenesis pathway in the liver it also could improve plasma lipid profile in both parents and their offspring. $MgSO_4$ is also indirectly able to reduce insulin resistance in their offspring possibly through the increased gene expressions of IRS1 and Akt and gluconeogenesis pathway in the liver.

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