patients is restricted. This study was designed to determine the in vitro sensitivity to LMWH of different reagents by sonoclot analyser, and to determine whether the ACT can be used to monitor LMWH.

Methods This study was performed in vitro. ACT was measured with different reagents (glass beads, celite, and kaolin) on volunteer (n=30) blood samples spiked with increasing concentrations of LMWH (dalteparin, 0.2–1.8 IU/ml). Linear regression analysis was performed to establish a regression equation from different concentration of dalteparin and corresponding ACT values.

Results Analysis of dose-response curves obtained in vitro, an excellent linear relationship was observed between the ACT and dalteparin concentrations for all three reagents (p<0.01). Differences in slope of the regression curves of ACT were observed with all the reagents tested (glass beads 249.7 s/IU, celite 77.7 s/IU, and kaolin 59.5 s/IU, p<0.01). Reagents vary widely in their in-vitro sensitivity to dalteparin. In the concentration range of 0.2–1.8 IU/ml, the gaolin reagent was too insensitive to dalteparin, and glass beads was the most suitable reagent for monitoring the anticoagulant effect of dalteparin.

Conclusions Using sonoclot analyser, there was an excellent linear relationship between the ACT and dalteparin concentrations for all the three reagents (glass beads, celite, and kaolin) in vitro. Glass beads may be a suitable reagent of ACT test for monitoring the anticoagulant effect of LMWH.

Objective Low molecular weight heparin (LMWH) is currently the most commonly used intravenous anticoagulant drugs, but the lack of point of care testing (POCT) limit its applications in patients with severe renal dysfunction and others. The purpose of this study was to explore the sensitivity of new ACT test reagents for laboratory monitoring of LMWH.

Methods Blood samples collected from 30 healthy volunteers. After taking blood samples, different doses of low molecular weight heparin (dalteparin) were added and the anti-Xa level of taking blood samples, different doses of low molecular weight heparin (dalteparin) were added and the anti-Xa level of samples was 0.1–1.8 IU/ml. ACT and clot rate (CR) were measured with traditional reagent kaolin and new reagent magbar, Linear regression analysis was performed and a regression equation was established between different anti-factor Xa levels and the corresponding ACT, CR values.

Results With dalteparin concentration increased, the ACT values were gradually extended and the CR values were gradually reduced with both two reagents (kaolin and magbar). Analysis of dose-response curves obtained in vitro, an excellent linear relationship was observed between the ACT and dalteparin concentrations for all two reagents (p<0.01), and an exponential relationship was observed between the CR and dalteparin concentrations (p<0.01). Differences in slope of the regression curves of ACT were observed with the reagents tested (magbar 1097.6 s/IU vs kaolin 59.5 s/IU, p<0.01).

Conclusions This in vitro study has shown that the sensitivity of traditional ACT test reagent (kaolin) for laboratory monitoring of dalteparin was poor, and the sensitivity of new ACT test reagents (magbar) for laboratory monitoring of dalteparin increased significantly. The new reagents magbar may be used for bedside monitoring of anticoagulant activity of LMWH.
Conclusions Direct adherent and modicum medium changing method is the best one for MSC isolation and culture. 11% is the most suitable serum concentration for MSC growth.

**e0174** EFFECT OF HIF1A ON PROLIFERATION AND DIFFERENTIATION OF MSC UNDER HYPOXIA CONDITION IN VITRO

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Aims To investigate the effect of HIF-1α on MSC under hypoxia condition.

Materials and methods We transfected HIF-1α into MSC of P3 generation through liposome 2000, and observed the expression of green fluorescence protein in order to assess transfecting efficiency. G418 was used to screen stable transfected cells, and limited dilution method used for monoclonal culture of screened cells. We identified the stable HIF-1α transfected MSC through the cell surface antigen testing. We compared the growth state among stable transfected MSC with HIF-1α, vacant plasmid transfected MSC and untransfected MSC under hypoxia condition, and the expression of HIF-1α mRNA, VEGF mRNA, HIF-1α protein and VEGF protein was tested.

Results pcDNA3.0-HIF-1α-eGFP can be successfully transfected into MSC mediated by liposome 2000, with efficiency of 21%. Stable monoclonal of transfected MSC can be obtained by G418 screening and limited dilution method. Stable transfected MSCs still reserve the ability of differentiating to chondrocyte and lipocyte. MSCs transfected with pcDNA3.0-HIF-1α-eGFP had lower apoptosis (p<0.05), greater proliferation (p<0.05), and more expression of HIF-1α mRNA, VEGF mRNA, HIF-1α protein, VEGF protein than MSCs transfected with vacant plasmid pcDNA3.0-eGFP and untransfected ones under hypoxia condition.

Conclusions Stable transfected MSC with HIF-1α has a significant high expression of HIF-1α protein, HIF-1α mRNA, VEGF protein and VEGF mRNA under hypoxia condition. HIF-1α could reduce MSC apoptosis and enhance its proliferation under hypoxia condition.

**e0175** THE EFFECT OF GHERLIN ON THE REGRESSION OF ATHEROSCLEROSIS PLAQUE IN APOE-/- MICE AORTA

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Objective To observe the effect of ghrelin on reducing the apoE−/− mice plasma IL-8, MCP-1, TNFα level and the NFκBp65 expression in vascular wall and the regression of atherosclerotic plaque.

Method 8 week ApoE−/− mice were fed with Western style meals, and the same age mice C57BL/6j fed with the same meals as control. In the eighth week, ApoE−/− mice were assigned to ghrelin intraperitoneal injection and saline injection group randomly in the twelfth week. All of the groups had blood drawn from eye sockets, with isolated plasma used to measure IL-8, MCP-1, TNFα by ELISA. Mice were killed for examination with stereomicroscopy and paraffin imbedding for HE and immunohistochemistry, and frozen section for red oil stain.

Result 1. On stereomicroscopy, HE, oil red stain and image analysis equipment measurement demonstrated no plaque at C57BL/6j mice vessels, and both apoE−/− group and apoE−/− + ghrelin groups had atherosclerosis plaque at vessels (22.56 ± 2.2 vs 32.37 ± 2.2 p<0.01). 2. Contrast to C57BL/6j mice, apoE−/− mice has higher plasma TNFα, IL-8, MCP-1 level (28.81 ± 1.8 vs 11.5 ± 0.6, p<0.05; 335 ± 16.7 vs 25.0 ± 2.0, p<0.05; 78.5 ± 6 vs 15.8 ± 2.0, p<0.05), but apoE−/− + ghrelin mice has lower TNFα, IL-8, MCP-1 level than ApoE−/− mice (15.45 ± 0.98 vs 24.5 ± 1.68, p<0.05; 163.32 ± 2.78 vs 335 ± 16.7 p<0.05; 45.5 ± 4.75 vs 78.5 ± 5.6, p<0.05). 3. Contrast to C57BL/6j mice, apoE−/− mice NFκBp65 immunohistochemistry positive cell integral calculus value were increase (1424.23 ± 167.80 vs 659.68 ± 675.34, p<0.01), ghrelin+ ApoE−/− mice NFκBp65 immunohistochemistry positive cell integral calculus value was lower than apoE−/− mice (342.78 ± 321.6 vs 685.96 ± 675.34, p<0.01), ghrelin can decrease the expression of NFκBp65 in apoE−/− mice aorta.

Conclusion Ghrelin can inhibit the inflammatory response to decrease ApoE−/− mice atherosclerosis plaque formation.

**e0176** THE EFFECTS OF ROSUVASTATIN ON THE EXPRESSION OF HOMOCYSTEINE-INDUCED EXPRESSION OF MATRIX METALLOPROTEINASE-2 (MMP-2) AND CELL MIGRATION IN RAT VASCULAR SMOOTH MUSCLE CELLS

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Objective The aim of this study was to investigate the effects of rosuvastatin on the expression of homocysteine-induced expression of matrix metalloproteinase-2 (MMP-2) and cell migration in rat vascular smooth muscle cells (VSMC).

Methods Cultured rat VSMC were incubated with different concentrations of Hcy and rosuvastatin (Hcy 1000 μmol/l) in vitro for 24, 48 and 72 h. The expression of MMP-2 was determined by using the methods of gelatin zymography and western blotting. Cultured rat VSMC was incubated with different concentrations of Hcy and rosuvastatin (Hcy 1000 μmol/l) in transwell for 24, 48 and 72 h. The number of VSMC which transited the membrane represented the aggressivity of VSMC.

Results Hcy (50 ~ 1000 μmol/l) increased the expression and activity of MMP-2 significantly. Incubated with the same concentration of Hcy the expression and activity of MMP-2 of 72 h was higher than that of 24 h and 48 h. Hcy reduced the expression of MMP-2 at the concentration of 5000 μmol/l. Rosuvastatin could inhibit the augmentation of homocysteine-induced expression and activity of MMP-2. Hcy (50 ~ 5000 μmol/l) could stimulate the migration of VSMC. Rosuvastatin could decrease the stimulation of homocysteine-induced migration of VSMC.

Conclusions These data suggested that Hcy can increase the MMP-2 expression/activity and the migration of VSMC. It may be one of the roles in the pathogenesis of atherosclerosis induced by Hcy. Rosuvastatin can inhibit the augmentation of homocysteine-induced MMP-2 expression/activity and migration of VSMC. This may be one of the pleiotropic of rosuvastatin besides lipid-lowering and benefit the therapy of CHD.

**e0177** EXPLORATION NEW METHODS FOR ESTABLISHMENT OF PORCINE MODEL OF ACUTE MYOCARDIAL INFARCTION

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Objective To explore and develop one optimise method that it could establish the porcine model of acute myocardial infarction more safer, quicker, convenient than routine methods. Angioplasty balloon was positioned in the mid-distal of left anterior...