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 59.3 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.

e0171 THE SENSITIVITY OF NEW REAGENTS FOR LABORATORY MONITORING OF LOW MOLECULAR WEIGHT HEPARIN: AN IN VITRO STUDY

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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 activated clotting time (ACT) 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 final blood 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 59.3 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.

Basic Science: Experiment Research

e0172 EFFECTS OF EPO ON PROLIFERATION OF RABBIT BONE MARROW MESENCHYMAL STEM CELLS

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Objective To explore effects of erythropoietin (EPO) on proliferation, activity and cell cycle of rabbit bone marrow mesenchymal stem cells (BMSCs).

Methods A total of four healthy male New Zealand white rabbits aged 6 weeks were supplied by the Experimental Animal Center of Nachang University. Rabbit BMSCs were in vitro isolated by the density gradient centrifugation. Cells were assigned into normal, 2 u/ml, 4 u/ml, 8 u/ml, 16 u/ml groups. In the normal group, cells received normal culture, without additional EPO stimulation. Following EPO intervention, cell morphology and growth were observed. Cell proliferation was detected by MTT assay. Cell cycle changes were measured by flow cytometry. Cell supernatant was collected to detect mass concentration of matrix metalloproteinase-2 (MMP-2).

Results EPO to intervene BMSCs 3d, cells had no abnormal changes, showing colony growth. With increase EPO concentration, cell absorbance gradually increased, especially in the 16 u/ml group. ($F=28.029$, $p=0.008$). Compared with the normal group, cell cycle changed in the 2 u/ml, 4 u/ml, 8 u/ml, 16 u/ml groups, and proliferation index significantly increased ($p<0.05$ or 0.01). Mass concentration of MMP-2 in supernatant was the lowest in the 16 u/ml group, and the highest in the normal group. Significant differences were detected among groups ($t=213.21$, $p<0.001$).

Conclusion EPO can increase the proliferative ability of BMSCs.

e0173 ISOLATION CULTURE AND IDENTIFICATION OF RAT MSC

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Aims To investigate different ways of isolating and culturing rat MSC and different serum concentrations of medium for the best selection.

Materials and methods Direct adherence and density gradient centrifugation methods are used in MSC isolation, routine and modicum medium change methods are used in MSC culture. We compared the growth state, cell quantity and population doubling time of MSC under different culturing ways and different serum concentration medium such as 10%, 11% and 15%. We identified cultured MSC in logarithmic growth phase (P3 generation) by cell surface antigen and its inducing differentiation function.

Results 4 methods, which are direct adherent and routinely changing of medium method, direct adherent and modicum medium changing method, density gradient centrifugation and routinely changing of medium method and density gradient centrifugation and modicum medium changing method, are used during MSC isolating and culturing respectively. The cellular average doubling time is 36.0±0.9 h, 23.5±1.1 h, 49.8±12 h and 48.0±0.8 h respectively There are cellular colonies forming 3 to 10 days after isolation, shaping like whirlpool. From the serum concentration screening experiment, we find that 11% is the most suitable one for MSC growth. The result of cell surface antigen identification of MSC through immunol histochemistry is CD45 (−), CD90 (+), and CD45 0.38%, CD90 98.4% for positive expression of MSC through flow cytometry. MSC can be successfully induced to differentiate to chondrocyte and lipocyte.