## Supplementary Table 1. Local Anesthetic Effects on Cells and Tissues.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Author/Year | Injectate | Target Cells/Tissue | Local Anesthetic | Study Design | Results/Conclusion/Summary |
| Dregella et al 2014 (14) | Human mesenchyma-l stem cells (hMSCs) | NA | Lidocaine  Bupivacaine  Ropivacaine  Mepivacaine | Experimental study measuring cell viability after exposing cultured hMSCs to different equipotent concentrations of lidocaine, bupivacaine, ropivicaine and mepivacaine for 40mins, 120mins, 360mins and 24hrs. | Significant reduction in cell viability at 120mins, 360 mins, and 24hrs.  At equipotent doses, cell death was greatest with bupivacaine > lidocaine > mepivacaine > ropivacaine.  Ultimately, amide-type local anesthetics causes hMSC death in a time- and dose- dependent manner that may harm clinical outcomes. |
| Gugerell et al 2014 (23) | Human adipose derived stem cell (ASC) | Fibroblasts | Lidocaine | In vitro study; ASC and fibroblasts were incubated in lidocaine 1% with saline as control. Cell viability and proliferation were measured on day 1 and day 3. | Lidocaine demonstrated time- dependent reduction in cell viability and proliferation of ASCs and fibroblasts. |
| Rahnama et al 2013 (24) | hMSCs | NA | Lidocaine  Bupivacaine  Ropivacaine | hMSCS were exposed to varying concentrations of lidocaine, bupivacaine, and ropivicaine for 60minutes. Cell viability was measured at 24hrs. | Lidocaine resulted in significantly more reduction in cell viability when compared with bupivacaine and ropivacaine.  Ropivacaine and bupivacaine exhibited no differences in toxicity when compared with control. |
| Girard et al 2013 (25) | ASC | NA | Lidocaine | ASCs harvested from human lipoaspirates were exposed to clinically relevant concentrations of lidocaine for 1-24hrs. | Lidocaine decreased cell viability after 24hrs, even when cells were only exposed to 1 or 2hrs. |
| Tayton et al 2012 (26) | Skeletal stem cells | NA | Lidocaine  Bupivacaine  Levobupivacaine | Skeletal stem cells from 3 patients were exposed to varying clinical concentrations of lidocaine, bupivacaine, and levobupivacaine for 2hrs. Cell viability was measured at 1 and 7 days. | Significant time and dose dependent reduction in stem cell viability with lidocaine>bupivacaine>levobupivacaine |
| Gray et al 2015 (27) | hMSCs | NA | Lidocaine  Bupivacaine  Ropivacaine  Procaine | In vitro study; Cultured hMSCS incubated with increasing concentrations of lidocaine, ropivacaine, procaine, and bupivacaine.  Cell viability was measured after 48hrs. | Neither lidocaine nor procaine had major effect on unstimulated hMSCs.  Both ropivacaine and bupivacaine had significant and dose dependent effect on cell viability. |
| Haasters et al 2011 (28) | Stem/progenitor cells | NA | Bupivacaine  Ropivacaine | Experimental study measuring cell viability after exposure to different local anesthetics for 0.5, 2, and 6hrs. | Bupivacaine and ropivacaine exhibited significant time dependent cell death.  Bupivacaine>ropivacaine. |
| Zhang et al 2016 (10) | hMSCS | Fibroblasts and tenocytes. | Bupivacaine  Ropivacaine | Experimental study measuring cell viability after incubation with different concentrations of local anesthetics for 30mins.  Assays were measured at 1hr, 24hrs, and 7 days. | Bupivacaine demonstrated significant dose-dependent reduction in cell viability with fibroblasts and tenocytes.  Ropivacaine demonstrated no significant effect.  No statistically significant effect on hMSCs viability. |
| Breu et al 2013 (17) | hMSCs | NA | Bupivacaine  Ropivacaine  Mepivacaine | In vitro study; hMSCs were exposed to varying concentrations of bupivacaine, ropivacaine, and mepivacaine for 1hr. Viability assay measured at 24hrs and 96hrs. | Significant time and concentration dependent reduction in hMSCs viability.  Comparing equipotent concentrations of local anesthetics, viability rates were higher 24 hours after treatment with 0.75% ropivacaine than with 0.5% bupivacaine and 2% mepivacaine. After 96 hours, ropivacaine was less cytotoxic compared with bupivacaine and mepivacaine. |
| Keck et al 2010 (29) | ASCs | NA | Bupivacaine  Mepivacaine  Ropivacaine | In vitro study; ASCs cultured from 15 patients were treated with equipotent concentrations of local anesthetics for 30mins. Viability was determined immediately afterwards. | All local anesthetics resulted in impairment of ASC differentiation with lidocaine exerting the most significant effect. |
| Augereau et al 2004 (30) | Platelets | NA | Lidocaine  Dibucaine  Tetracaine | Isolated blood platelets were incubated with different local anesthetics for 1hr. Mitochondrial transmembrane potentials in platelets were evaluated using fluorescence imaging. | Dibucaine and tetracaine induced mitochondrial apoptotic-like cascade in platelets.  Less hydrophobic lidocaine produced no effect. |
| Bausset et al 2014 (31) | Platelet rich plasma (PRP) | NA | Lidocaine  Ropivacaine | PRP from 9 healthy volunteers was treated with lidocaine and ropivacaine. Platelet function was assessed by measuring aggregometry and growth factor release. | Significant decrease in platelet functionality especially platelet aggregation with no effect on growth factor release.  Concomitant use of LA has the potential to compromise the therapeutic effect of PRP. |
| Pinto et al 2004 (32) | PRP | NA | Lidocaine  Bupivacaine  Benzocaine | PRP was treated with different LA. Platelet function was assessed by platelet aggregation. | Significant decrease in platelet function and aggregation. |
| Liou et al 2012 (33) | PRP | NA | Levobupivacaine | Whole blood, PRP, and washed platelets were treated with levobupivacaine. Platelet aggregometry was measured. | Negative effect on platelet aggregation. |
| Lo et al 2001 (34) | PRP | NA | Lidocaine  Bupivacaine | PRP collected was treated with different local anesthetics. Level of thromboxane A2 (TXA2) secretion was measured as marker for platelet aggregation. | Local anesthetics demonstrated dose-dependent ability to inhibit TXA2-induced platelet aggregation. |
| Porter el al 2001 (35) | Platelets | NA | Ropivacaine | Whole blood collected from 10 patients was treated with different concentrations of ropivacaine. Platelet function was measured with platelet function analyzer. | Ropivacaine resulted in dose-dependent inhibition of platelet aggregation. |
| Az-ma et al 1995 (36) | PRP | NA | Lidocaine | PRP obtained from healthy volunteered treated with varying concentrations of lidocaine. Platelet aggregation was measured as marker of functionality. | Lidocaine demonstrated dose-dependent antiaggregation effect. |
| Carofino el al 2012 (37) | PRP | Tenocytes | Lidocaine  Bupivacaine | In vitro study; human bicep tendons were incubated with PRP alone or in combination with lidocaine, bupivacaine, and triamcinolone for 5mins, 10mins, and 30mins.   Viability assay measured after 24hrs. | Lidocaine resulted in significant reduction in cell viability.  Exposure to bupivacaine resulted in more cellular apoptosis than lidocaine.  LA+PRP resulted less in tenocyte apoptosis suggesting PRP may offer protective effects against local anesthetics. |
| Scherb et al 2009 (9) | NA | Tenocytes | Bupivacaine | Cultured human tenocytes were incubated with varying concentrations of bupivacaine. | Tenocyte proliferation was significantly lower with bupivacaine vs control group. |
| Grishko et al 2010 (15) | NA | Human chondrocytes cultures | Lidocaine  Bupivacaine  Ropivacaine | Experimental study measuring apoptosis and mitochondrial dysfunction after exposing cultured human chondrocytes to different concentrations lidocaine, bupivacaine, and ropivacaine for 1hr.   Viability assays were measured at 24hrs. | All three local anesthetics demonstrated dose- dependent reduction in cell viability and mitochondrial dysfunction. |
| Iwasaki et al 2013 (11) | NA | Human nucleus pulposus cells | Lidocaine  Bupivacaine | In vitro study; human NP cells were incubated with different local anesthetics. Cell viability was measured after 30mins, 60mins, and 120mins of exposure time. | Lidocaine demonstrated significant dose- and time- dependent reduction in viability.  Exposure to bupivacaine resulted statistically significant more cell death than lidocaine. |
| Rao et al 2013 (18) | NA | Human chondrocytes and synovial cells | Lidocaine  Bupivacaine  Ropivacaine | In vitro study; human chondrocytes and synovial cells were exposed to bupivacaine, ropivacaine, and lidocaine for 90 minutes. Viability assays were measured at 1 3, 5, and 7 after initial exposure. | Lidocaine and bupivacaine, with ropivacaine to a lesser degree, demonstrated time dependent increase in cell apoptosis with peak at 5 days.  Ropivacaine offered the most chondroprotective effect when couple with caspase inhibitor. |
| Jacobs et al 2011 (38) | NA | Human chondrocytes | Lidocaine | In vitro study; cultured human articular chondrocytes were exposed to 1% and 2% lidocaine and 2% lidocaine with epinephrine. Cell viability measured at 15mins, 30mins, and 60mins. | Lidocaine showed a time-dependent cytotoxicity with gradually more dead cells after longer exposure. |
| Dragoo et al 2011 (39) | NA | Human chondrocytes | Lidocaine  Bupivacaine  Ropivacaine | In vitro study; human chondrocytes were exposed to varying anesthetics and normal saline as control. Cell viability measured subsequently at different times. | Single-dose administration of 1% lidocaine resulted in a significant decrease in chondrocyte viability when compared with control cultures.  No significant effect with bupivacaine and ropivacaine. |
| Syed et al 2010 (40) | NA | Human chondrocytes from articular explants | Bupivacaine | Experimental study measuring cell viability after exposing cultured human chondrocytes to 0.25% bupivacaine.  Viability assays was measured at 24hrs. | 0.25% bupivacaine resulted in approximately 30% cell viability at 24hrs of exposure. |
| Piper et al 2008 (16) | NA | Human femoral head articular cartilage explants and cultured chondrocytes | Bupivacaine  Ropivacaine | Experimental study measuring cell viability after incubation with different concentrations of local anesthetics for 30mins. Assays were measured at 24hrs. | Significant reduction of cell viability in both cartilage explants and cultured chondrocytes measured at 78% and 37%, respectively.  While chondrocyte exposure to 0.5% ropivacaine demonstrated decrease in cell viability, it was significant less chondrotoxic compared to bupivacaine. |
| Baker et al 2010 (41) | NA | Human chondrocytes | Bupivacaine  Ropivacaine  Levobupivacaine | In vitro study; chondrocytes were exposed to varying concentrations of levobupivacaine, bupivacaine, and ropivacaine. Viability measured 24hrs after initial exposure. | Significant decrease in cell viability with increasing concentration of bupivacaine, ropivacaine, and levobupivacaine. |
|  |  |  |  |  |  |

Note: Overall, lidocaine and bupivacaine were common anesthetics that had chondrotoxic effects and a decrease in cell viability. Local anesthetic effects were shown on variable cell and tissue types.