Sulfasalazine reduces soluble Flt-1 and soluble endoglin and quenches endothelial dysfunction in primary human tissues:

A novel therapeutic candidate

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Introduction
Preeclampsia – A serious complication of pregnancy
• Preeclampsia complicates 5-8% of pregnancies
• Caused by the placental release of sFlt-1 and soluble endoglin (sENG) into the maternal bloodstream
• The only treatment is delivery
• At preterm gestations this leads to significant neonatal disability and death
• A therapeutic would be a major clinical advance

Sulfasalazine – A possible therapeutic
• Sulfasalazine is an anti-inflammatory and immune modulating drug
• It is safe in pregnancy (Category B)

Aim
• To assess the effect of sulfasalazine on sFlt-1 and sENG production
• To explore whether sulfasalazine is mediating an effect on sFlt-1 through HO-1 or NFκB
• To see whether sulfasalazine rescues endothelial dysfunction

Methods
We performed functional experiments in primary human tissues. We added sulfasalazine at increasing doses
• For HUVECs 0, 200, 500, 1000μM
• For preterm (<=34 weeks gestation) preeclamptic placental explants and trophoblasts 0, 500, 750, 1000μM

We silenced HO-1 and NFκB to explore mechanisms

Outputs:
1. sFlt-1 and sENG production from placenta using ELISA
2. e15a, MMP14, HO-1, NFκB, VCAM-1 PCR
3. HO-1 western blot
4. xCELLigence for endothelial dysfunction - HUVEC migration

Results

Sulfasalazine decreases sFlt-1 production

A) HUVECs

B) Trophoblast

C) Preeclamptic explants

Fig. 1. Sulfasalazine decreases sFlt-1 production from A) HUVECs B) trophoblasts and C) preeclamptic placental explants. Sulfasalazine reduces D) sFlt-1 e15a expression in preeclamptic placental explants.

Sulfasalazine decreases soluble endoglin production

A) HUVECs

B) Preeclamptic explants

Fig. 2. Sulfasalazine reduces soluble endoglin secretion in A) HUVECs and B) preeclamptic placental explants. It reduces MMP14, the protease that produces soluble endoglin, in C) HUVECs and D) preeclamptic placental explants.

Sulfasalazine upregulates heme oxygenase 1

A) Preeclamptic explants

B) Trophoblast

C) Trophoblast

Fig. 3. A) Sulfasalazine upregulates heme oxygenase expression in preeclamptic placental explants. B) Silencing HO-1 did not change the effect of sulfasalazine on sFlt-1 secretion in trophoblasts

Sulfasalazine may reduce sFlt-1 by inhibiting NFκB

A) Preeclamptic explants

B) Trophoblast

Fig. 4. Sulfasalazine reduces NFκB in preeclamptic explants. Silencing NFκB in trophoblasts reduces sFlt-1 secretion

Conclusion

Sulfasalazine reduces sFlt-1 and sENG production and decreases endothelial dysfunction.

It potentially acts by inhibiting NFκB

It shows promise as a novel therapeutic for preeclampsia

All figures: * p<0.05, **p<0.01, ***p<0.001, ****p<0.0001. Data mean ± SEM, n=3-4 experiments