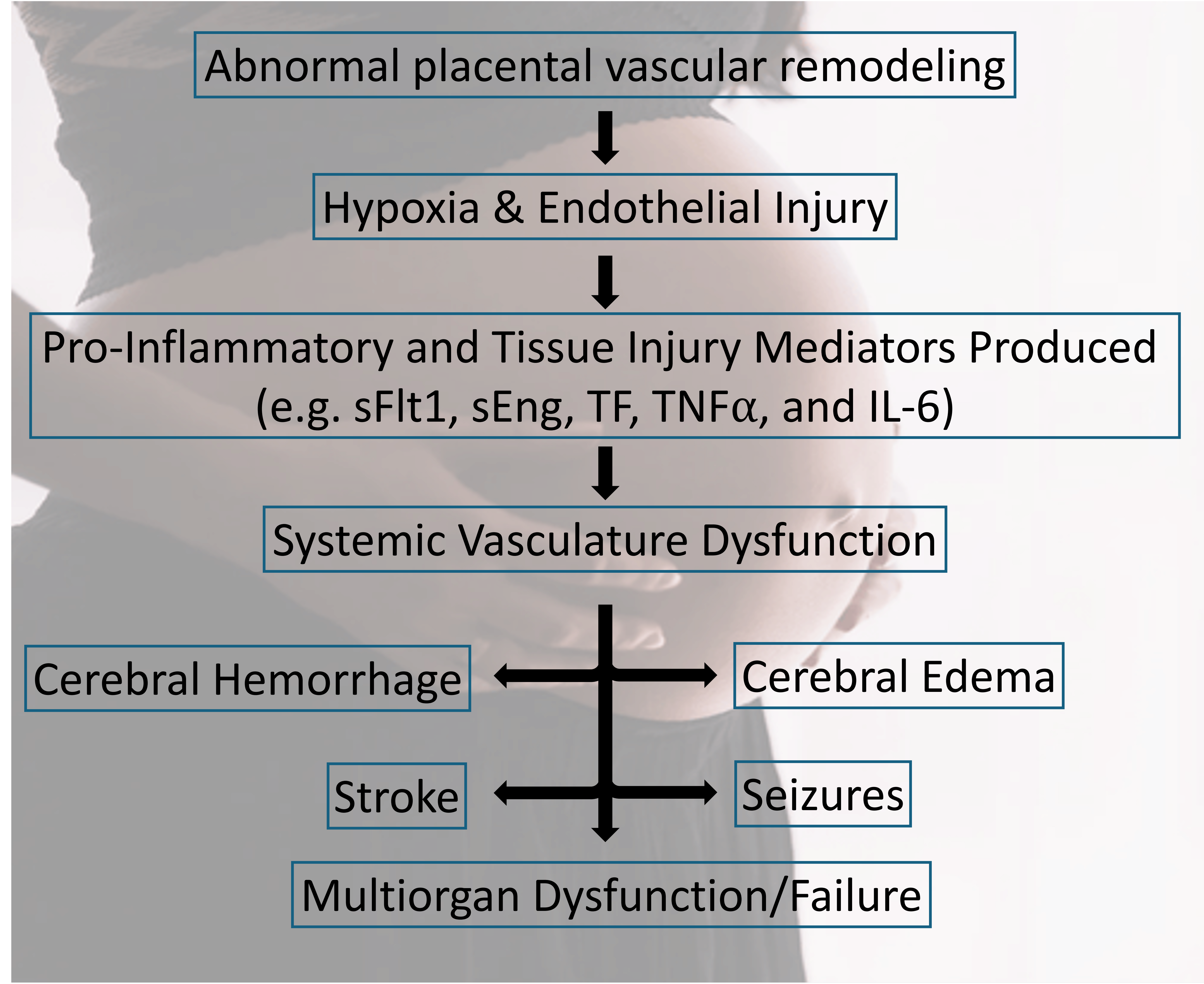
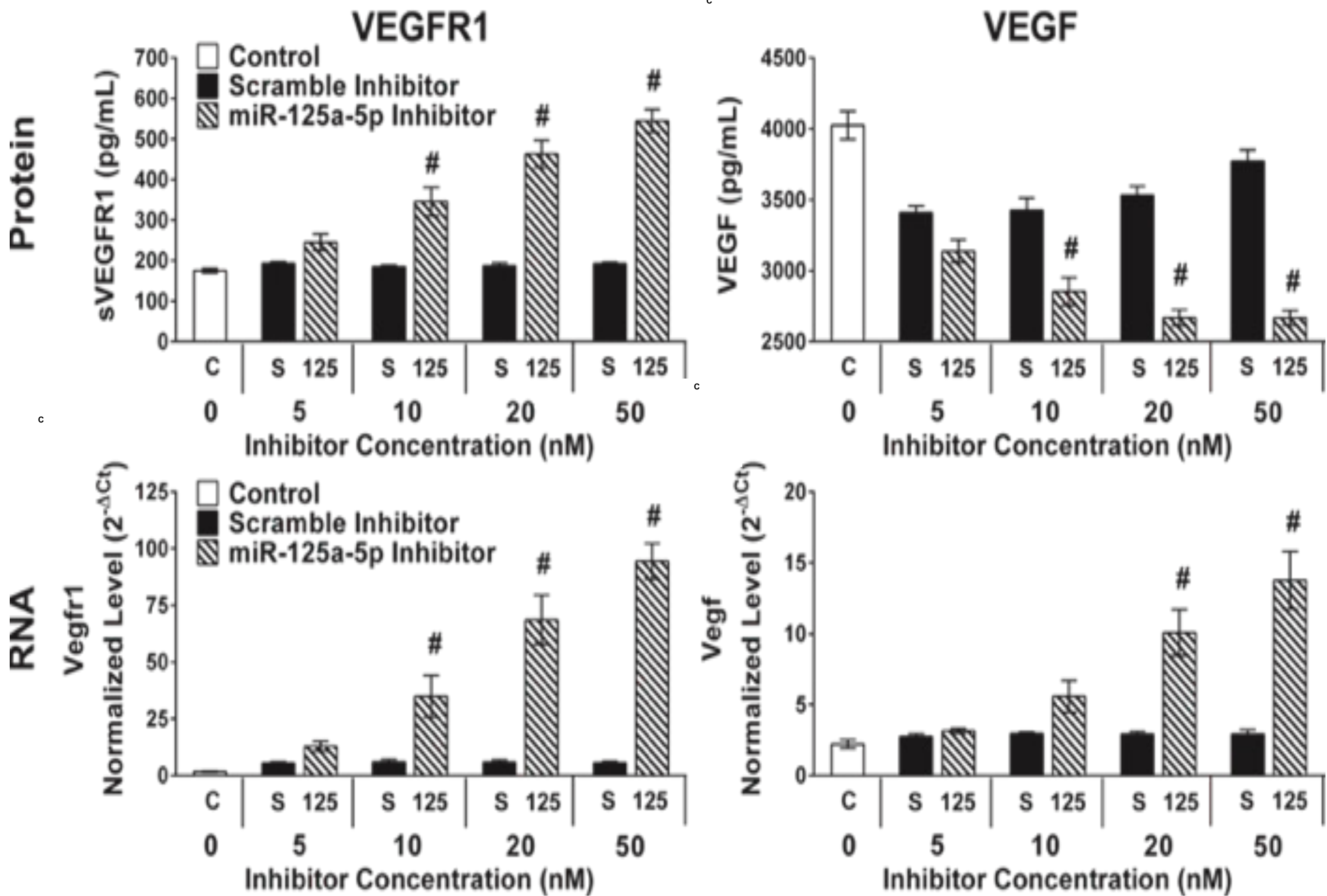


Loss of EV-miR-125a-5p in CSF May Permit Local sFLT1-Mediated Neurovascular Injury in Severe Pre-eclampsia

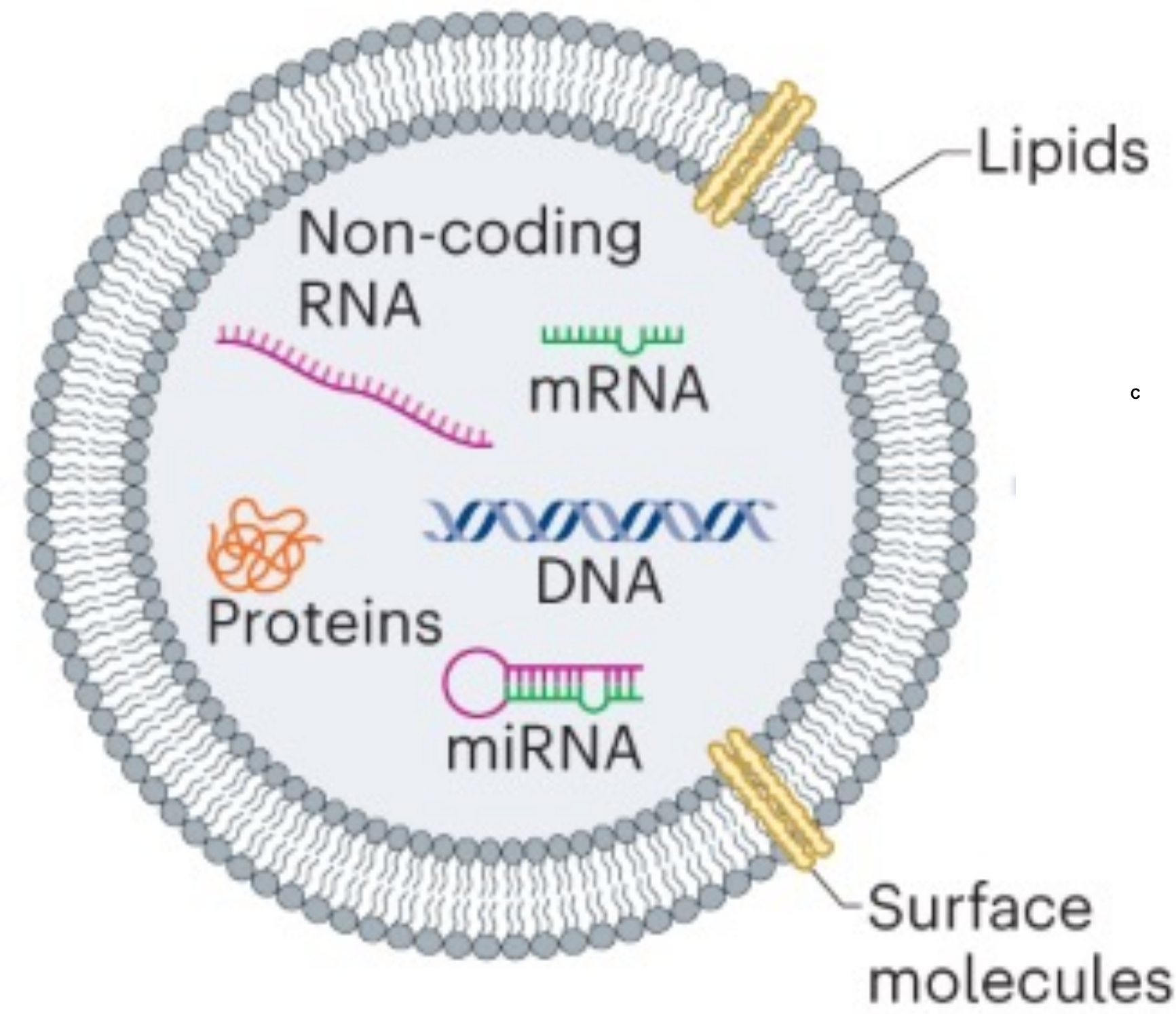


miR-125a-5p Inhibition *In Vitro* Mouse Macrophages sFlt-1 and VEGF Protein/RNA Levels



miR-125a-5p modulate trophoblast cell proliferation, migration, and invasion by regulating VEGFA, likely via sFlt-1 in a human trophoblast cell line

Extracellular Vesicles (EVs)



Multiple EV-miRNA studies in the plasma demonstrated expression patterns distinct in preeclamptic patients including miR-155, -210, -520a, and -125a-5p

Hypothesis: CSF EV-miRNA/proteins will demonstrate compartment-specific patterns that provide value in prognosticating the risk of severe neurological sequela and provide insight into dysregulated pathways in patients with preeclampsia.

Study Design and Methods

- Cohort 1:** Gestational age-matched controls (without evidence of preeclampsia) - **Control**
- Cohort 2:** Patients with preeclampsia without evidence of severe neurological features – **PreE**
- Cohort 3:** Patients with preeclampsia with at least one severe neurological feature – **PreE w/SNF**

Inclusion Criteria

- Pregnant women 18-45 years old
- Gestational age greater than 24 weeks
- Admitted to the labor and delivery floor at BIDMC
- Receiving neuraxial anesthesia alone or as a component in their anesthetic plan.
- Diagnosed with preeclampsia/eclampsia (up to 6 weeks postpartum) with strict ACOG criteria.²⁵

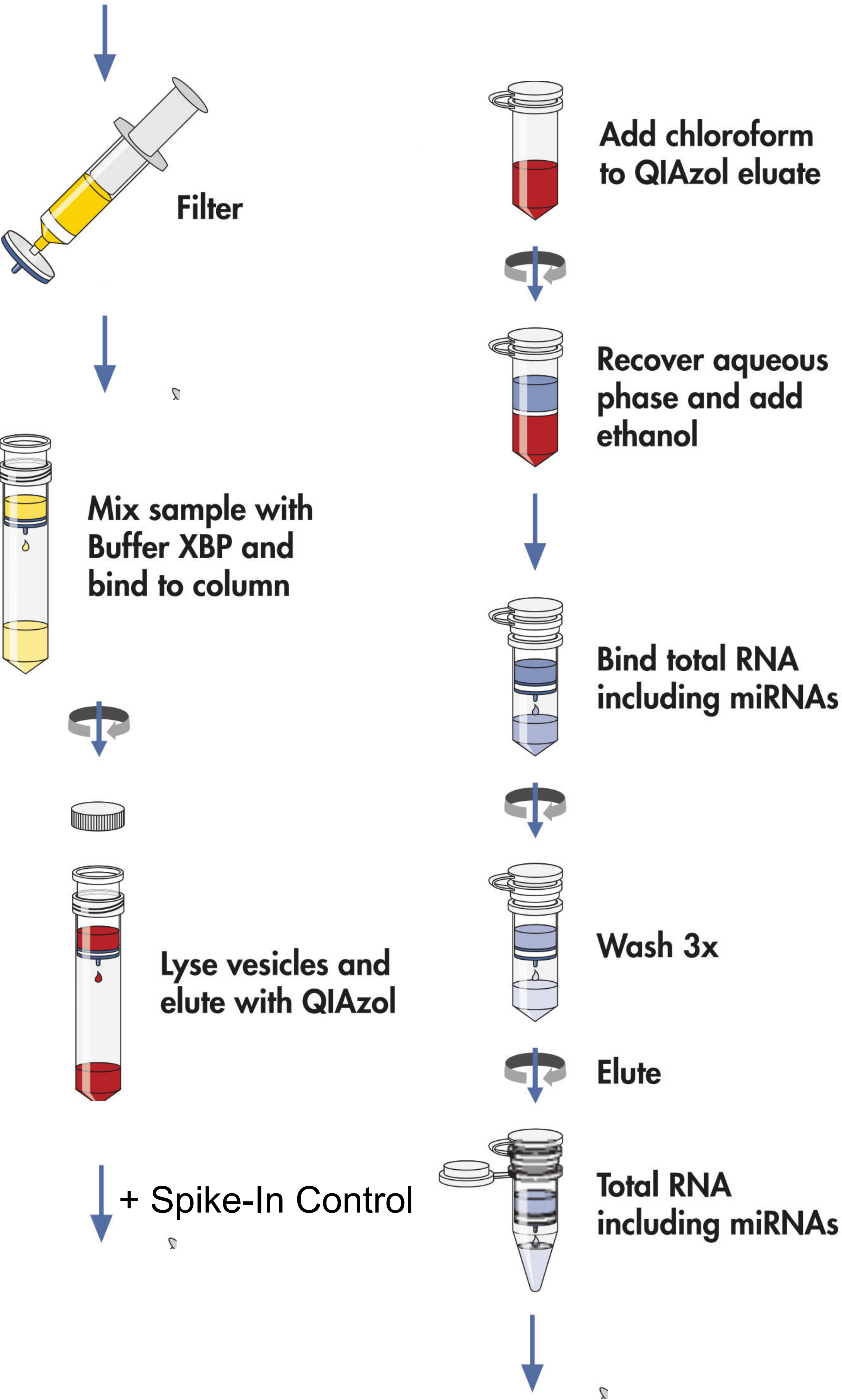
Exclusion Criteria

- Emergency case
- Unable to provide consent for self
- Contraindicated neuraxial anesthesia (including refusal)
- Any preexisting neurological diagnosis, excluding headache
- Personal history or diagnosis of cancer
- Family history of autoimmune disease
- Current or recent use of psychoactive or neuroactive medications
- Current or recent use of immune-modulating medications
- Diagnosis of chorioamnionitis and pre-existing systemic infectious or inflammatory conditions

Severe Neurological Features - Headache (Dull, throbbing, migraine-like headache not accounted for by other diagnoses and unrelieved by acetaminophen therapy), vision changes (Blurry vision, flashing lights, light sensitivity, or vision loss), altered mental status, or seizure activity.

IRB: 2024P000156

Collect & Store: EDTA Plasma, Urine, and Cerebrospinal Fluid



RT-qPCR of Candidate miRNAs:

miR-125a-5p, -155-5p, -210-3p, -520a-5p, -125b-5p, -190a-5p, and -297

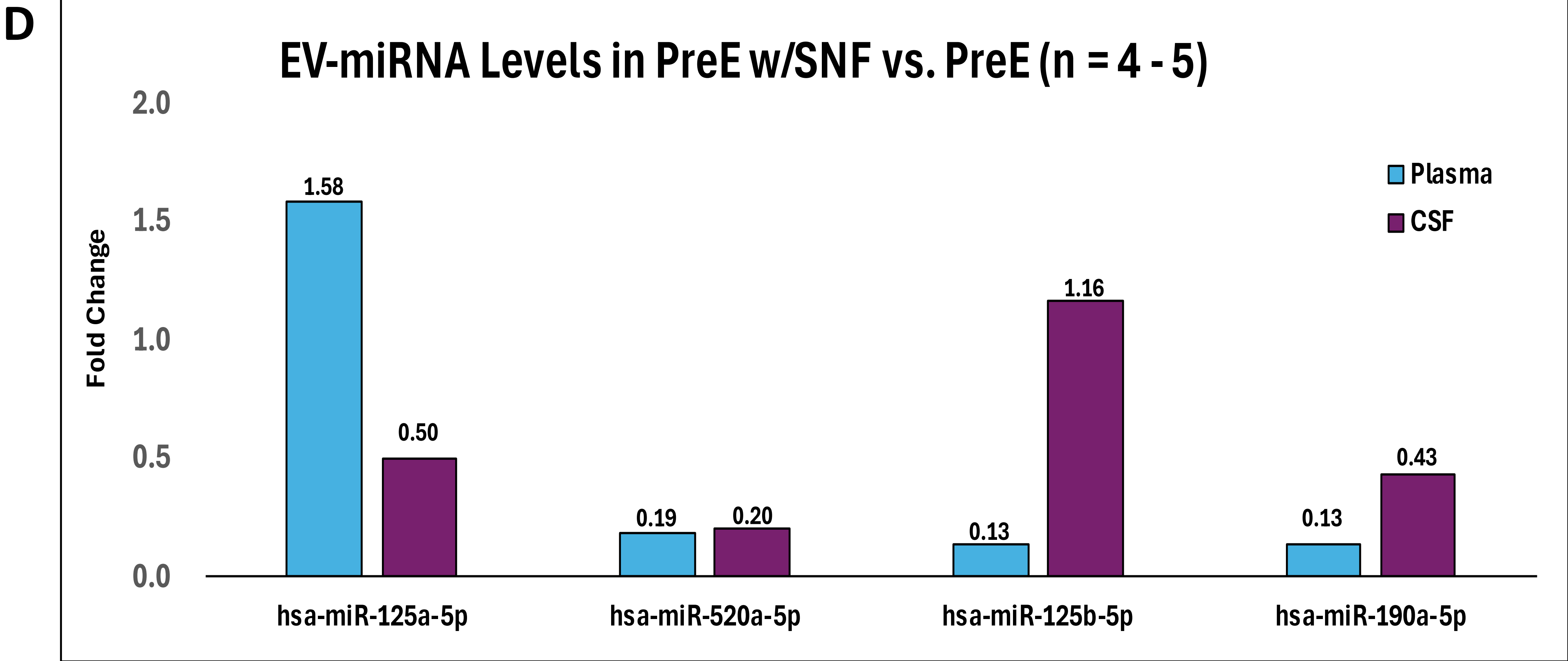
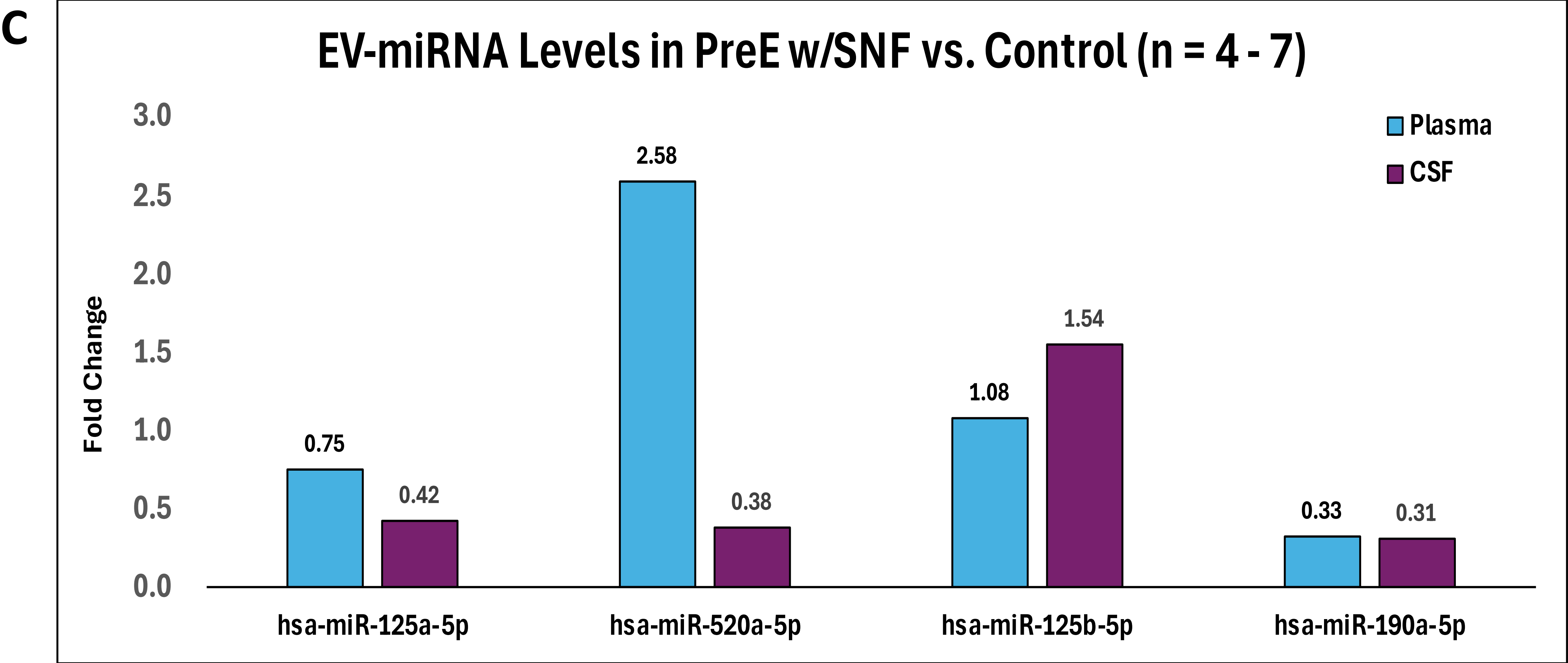
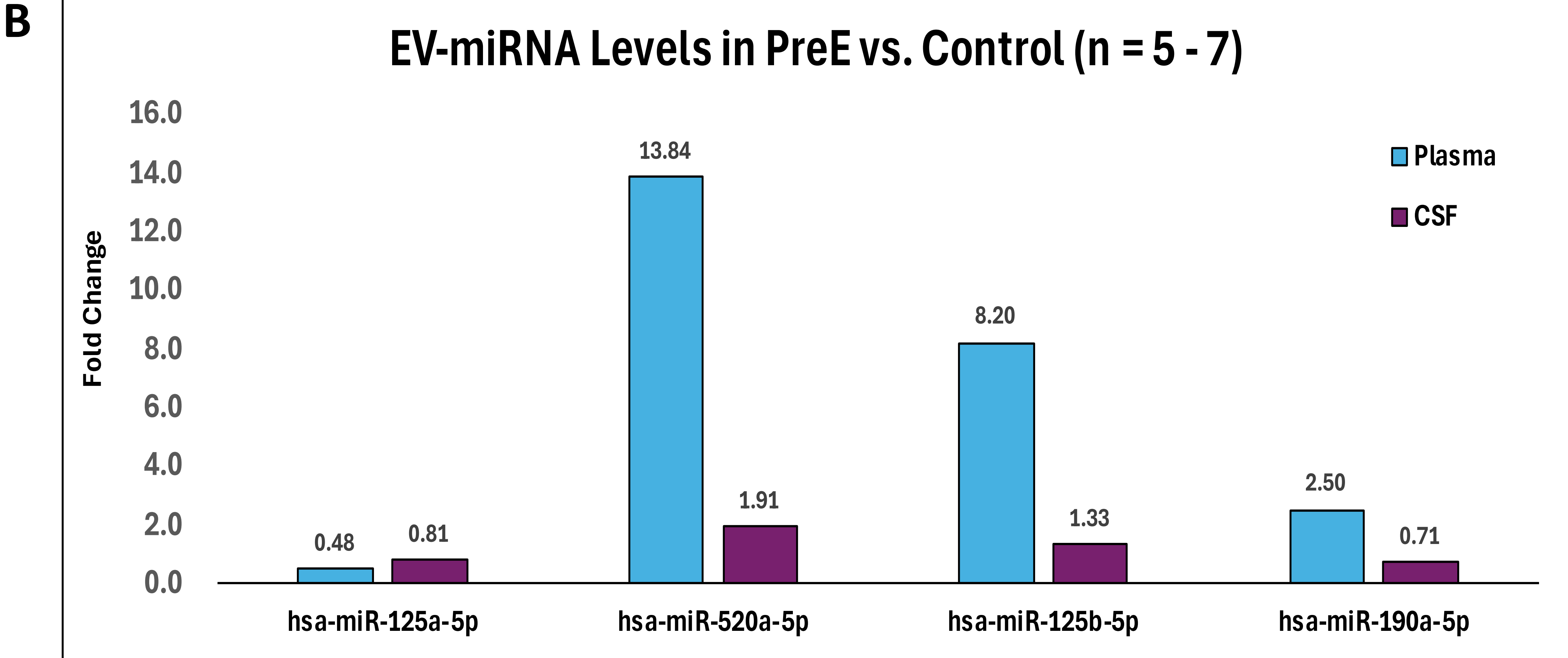
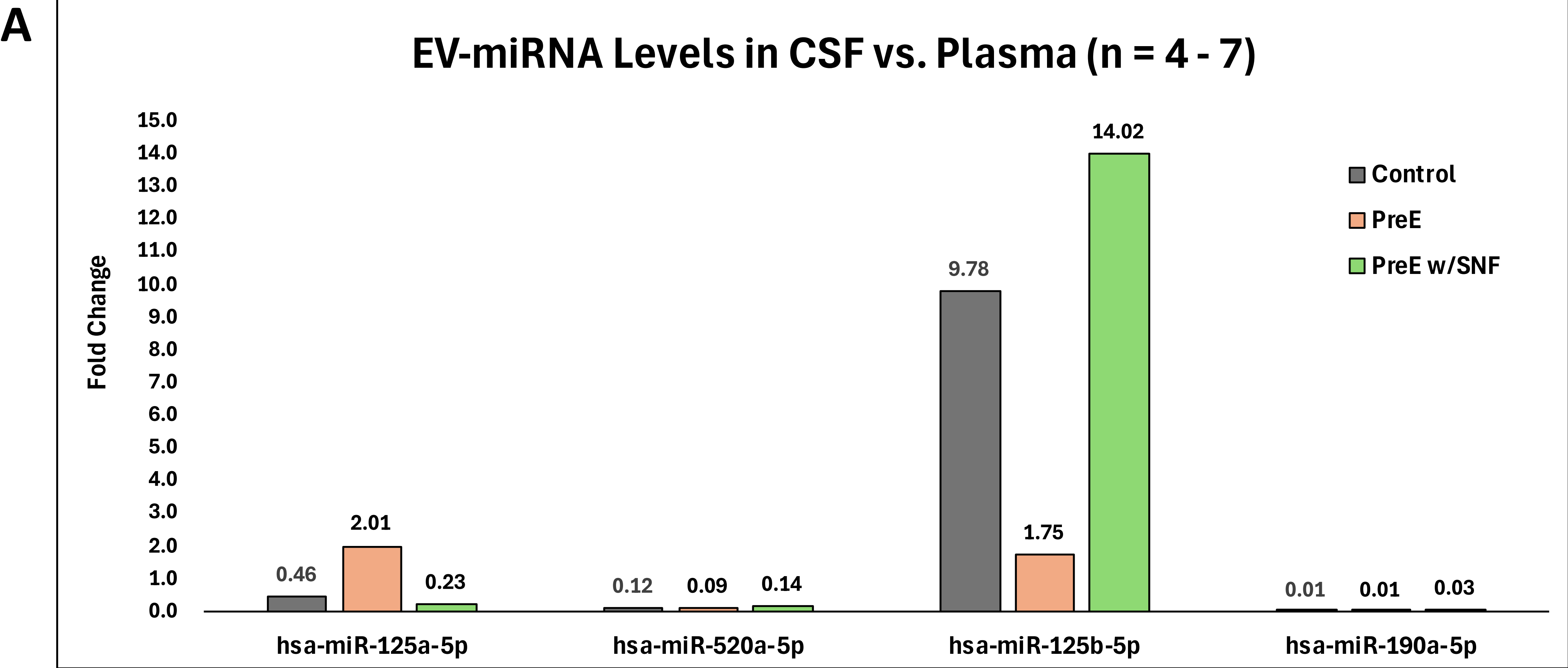
Pilot Results

Comparisons Made:

- Plasma vs. CSF (Figure A)
- PreE vs. Control (Figure B)
- PreE w/SNF vs. Control (Figure 1C)
- PreE w/SNF vs. PreE (Figure 1D)

miR-155-5p, 210-3p, and -297 was not detected in the CSF of any patient

For interpretation, a threshold of at least 2-fold change was considered as differentially expressed
(reported as ≤ 0.5 on this exponential scale for those with > 2 -fold decrease)



Conclusion & Discussion

- miR-520a-5p and -190a-5p demonstrated lower abundance in CSF vs Plasma (Figure A).
- miR-125a-5p in the PreE group & miR-125b-5p in the Control and PreE w/SNF groups demonstrated higher CSF vs. Plasma levels (Figure A).
- miR-125b-5p demonstrated a higher level in the plasma of PreE vs. Control (Figure B) but was suppressed in the plasma of PreE w/SNF vs. PreE (Figure D).
- miR-125a-5p exhibited a lower level (≤ 0.5) in the CSF of PreE w/SNF vs. Control and vs. PreE (Figures C & D).
- miR-520a-5p had a higher level in the plasma of PreE (Figure B) and PreE w/SNF (Figure C) vs. Control, but was less in the plasma of PreE w/SNF vs. PreE (Figure D). It was also suppressed in the CSF of PreE w/SNF vs. Control (Figure C) and vs. PreE (Figure D).
- miR-190a-5p was elevated in the plasma of PreE vs. Control (Figure B) but lower in the plasma of PreE w/SNF vs. Control (Figure C) and vs. PreE (Figure D). miR-190a-5p was lower in the CSF of PreE w/SNF vs. Control (Figure C) and vs. PreE (Figure D).
- While one may argue that perhaps many of the observed lower EV-miRNA levels across cohort may simply be due to a decrease in the number of EVs. However, our preliminary data of EV concentration shows no difference in the EV numbers across cohorts in either plasma or CSF (Plasma ($\times 10^{13}$): Control – 0.996, PreE – 1.36, PreEw/SNF – 1.30; CSF ($\times 10^8$): Control – 5.79, PreE – 5.02, PreEw/SNF – 5.15). We again recognize the lower concentration of EVs in the CSF vs. plasma.

Conclusions:

- Clear distinction in EV-miRNA expression between CSF and plasma.
- miR-125a-5p expression suppressed in CSF of PreE w/SNF with possible dysregulation of sFlt-1 and resulting vascular dysfunction.

References

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