E-Selectin Inhibitor GMI-1271 Works in Combination with Low-Molecular Weight Heparin to Decrease Venous Thrombosis and Bleeding Risk in a Mouse Model

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Virchow’s Triad Revisited

Circulatory Stasis:
- Immobility
- Pregnancy and post partum
- Previous venous thrombosis, post-thrombotic syndrome
- Disease states (sepsis, heart failure, inflammatory bowel disease)

Endothelial Injury:
- Catheterization
- Trauma
  - Surgery (orthopedic, urology, gynecologic)

Hypercoagulable State:
- Anti-thrombin, Protein C and S deficiencies
- Factor V Leiden
- Oral contraceptives
  - Estrogen replacement therapy
  - Anti-phospholipid antibodies
  - Malignancy
Deep Vein Thrombosis (DVT)

- Consequences of DVT
  - Acute DVT: Pulmonary Embolism
  - Chronic DVT: Post-thrombotic syndrome (PTS)

- Epidemiology of Venous Thromboembolism (VTE):
  - Approximately 900,000 people per year

- Current treatments for DVT
  - Anticoagulants: risk of adverse bleeding events
Inhibition of E-selectin can decrease thrombus formation and associated inflammation.

E-selectin (CD-62E) is a cell adhesion molecule that is expressed on activated endothelial cells.
  • Leukocyte recruitment to the site of vascular injury.

GMI-1271 is designed to mimic the bioactive conformation of the sialyl-Lex carbohydrate binding domain of E-selectin.
  • Specific E-selectin inhibitor.
- Inactivates anti-thrombin III (AT)

- Inactivation of factor Xa
  - (Anti-Xa activity)

- Subcutaneous (SQ) Dosing

- Less risk for heparin-induced thrombocytopenia
Mouse 2 Day Data

Anti-Xa Activity

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LMWH IU/mL

Non-VT Control

Surgery 2D + LMWH

* p<.05

** p<.01

*** p<.001
Mouse 2 Day Data

LMWH
Vein Wall + Thrombus Weight

Weight in grams

CTR-No Tx  3 mg/kg  4 mg/kg  5 mg/kg  6 mg/kg

*P<0.01

** P<0.0065
Translatable therapeutic that can treat DVT in combination with lower, safer levels, of low molecular weight heparin (LMWH).
E-selectin inhibition combined with LMWH will permit lower doses of therapeutic LMWH for the treatment of VT without increasing adverse bleeding events.
Electrolytic IVC Model

Diaz JA et al., Thromb Haemost 104:366-375, 2010
Experimental Design

Day 0:
EIM: Thrombus

Day 2:
Sample Collection

MOUSE GROUPS

Non-treated controls (CTR-No Tx)

LMWH [3-6 mg/kg, SQ, once daily (qd)]

GMI-1271 20mg/kg intraperitoneal (IP) twice daily (BID)

Combination of GMI-1271 + LMWH varying doses
Methods

➢ Treatment: First dose immediately following thrombus induction and through day 2.

➢ Animals euthanized 2 days post-thrombosis.

➢ Evaluations:
  • thrombus weight (grams)
  • anti-Xa testing
  • tail vein bleeding time (seconds)
  • immunopathology
GMI-1271 Works in Combination with LMWH to Decrease Venous Thrombosis

CTR-No Tx vs. 4 mg/kg + GMI 20 mg/kg, **P<0.0055
CTR-No Tx vs. 3 mg/kg + GMI 20 mg/kg, **P<0.0047
GMI-1271 Does not Increase Bleeding Potential

TAIL VEIN BLEEDING TIME

2 DAYS

GMI-1271 Does not Increase Bleeding Potential

CTR-No Tx vs. 5 mg/kg LMWH, *P=0.04
5 mg/kg LMWH vs. 4 mg/kg LMWH + GMI, *P=0.034
5 mg/kg LMWH vs. 3 mg/kg LMWH + GMI, *P=0.032
FIBRIN SCORING
DAY 2

%Fibrin (PTAH Staining)

CTR-No Tx
Sham 2D
2D GMI-1271
LMWH 3mg/kg
GMI+LMWH 3 mg/kg

**P<0.01
*P<0.05

2D LMWH 3mg/kg

Phosphotungstic Acid
Hamatoxylin
(PTAH)

Fibrin Stain

10X Mag.
- **E-Selectin Inhibition and DVT**
  - Mice gene deleted for E-Selectin significantly decreases VT

- **E-Selectin Decreases Fibrin Deposition**
  - Decreased fibrin content correlated with reduced thrombosis
Possible Mechanisms of Combination Therapy

- **E selection inhibition** may work at the level of the leukocyte affecting “immunothrombosis”.
  - Triggering an intravascular immune response involving neutrophils, monocytes, and platelets.

- **LMWH** is working at the level of the coagulation factor inhibiting factor Xa.
Discussion

**Heparin, LMWH**

GMI-1271

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**Chase SD, Magnani JL, Simon SI.** Annals of Biomedical Engineering, 2012.


Nonhuman Primate Model of Iliac Vein Thrombosis

• **Semi-upright animals**
  – cardiovascular and lower extremity physiology homologous to humans.

• **Immune system resemblance to humans**

• **Genetic similarity: ~96% homology at DNA level to humans**
Conclusions

- **GMI-1271** works in combination with LMWH to significantly reduce acute VT.
  - Without increasing bleeding times.

- **GMI-1271** decreases intra-thrombus fibrin content.

- These preliminary studies suggest that E-selectin inhibition with **GMI-1271** may be used to treat VT alone.
  - Or in combination with lower and safer levels of LMWH anticoagulation.