# Plasminogen Activity Assay

Activity is measured by the level of chromogenic substrate cleaved by plasmin or urokinase-activated plasminogen

### Materials

1. Versa Max microtiter plate reader (Molecular Devices, CA)
2. 96-well microtiter plate
3. Citrated plasma samples
4. Normal mouse plasma (NMP)
5. 100 mM Tris, pH 8.5 with 8.3 mM EACA (Tris/EACA)
6. HN/BSA
7. 2500 Ploug U/mL urokinase
8. 1.2 mM chromogenic substrate specific for plasmin and urokinase-activated plasminogen in HN buffer
9. 20% acetic acid

## Procedure

1. Plasma samples may be diluted to 1:50 in Tris/EACA
2. Log-log standard curve is prepared with each plate by diluting NMP 1:10 to 1:320 in Tris/EACA, analyzed simultaneously on the sample plate
3. Plate wells are loaded as follows:

| | | | | |
|---|---|---|---|---|
| **Sample dilutions, in duplicate** | **60uL** | **37℃** | **90sec** | |
| **Urokinase** | **20uL** | | **60sec** | |
| **Chromogenic substrate in HN** | **100uL** | | **10min** | **Color development** |
| **Acetic acid** | **100uL** | | | **Stop reaction** |

4. Plate is read at 405 nm
5. Absorbance are converted to %NMP plasminogen using the standard curve