α2-Antiplasmin Activity Assay

Antiplasmin activity is measured by level of inhibition of plasmin

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. TBS with 120 mM methylamine chloride
6. 10 µg/mL plasmin
7. 1.5 mM chromogenic substrate specific for plasmin
8. 20% acetic acid

Procedure

1. Plasma samples may be diluted to 1:50 in TBS/120 mM methylamine chloride
2. Standard curve is prepared with each plate by diluting NMP 1:10 to 1:1280 in TBS/120 mM methylamine chloride, analyzed simultaneously on the sample plate
3. Plate wells are loaded as follows:

Sample dilutions, in duplicate	**50uL**	**37℃C**	**15min**	
Plasmin	**50uL**		**90sec**	
Chromogenic substrate	**50uL**		**10min**	**Color development**
Acetic acid	**50uL**			**Stop reaction**

4. Plate is read at 405 nm at desired time points as color develops and after the reaction is stopped
5. Absorbance are converted to %NMP α2-antiplasmin using the standard curve