1. **Principle**

Urine from Sd (a+) individuals or from guinea pigs contains soluble Sda substance which will inhibit or neutralise anti-Sda if present in plasma.

1. **Scope and Related Policies**
	1. A frozen pool of urine will be maintained for this purpose.
	2. For human urine, an early morning sample (after fasting) is most suitable.
	3. Guinea pig urine contains potent Sda substance and is the product of choice.
2. **Specimen**

Patient Plasma (suspected of containing anti-Sda )

1. **Material**

**Equipment:** Cell Washer

 Serological centrifuge

 Block for test tubes

Microscope

Water bath/Heating block at 37°C

**Supplies:** Test tubes – 10 x 75 mm

 Serological pipettes

**Reagents:** 0.9% saline

Urine from guinea pigs (G.P.U.) or

Urine from Sd (a+) individuals.

Phosphate buffered saline PBS - pH 7.4

1. **Quality Control**
	1. A dilution control must be used if human urine is used for the inhibition.
2. Procedure

**Urine:**

|  |
| --- |
| * 1. Centrifuge urine specimen
 |
| * 1. Aliquots of urine can be frozen until needed
 |
| * 1. For use, dilute G.P.U. with an equal volume of pH 7.4 PBS
 |

**Inhibition:**

|  |
| --- |
| * 1. Add one volume of diluted G.P.U. to 10 volumes of plasma, or one volume of human urine to 1 volume of plasma (TEST).
 |
| * 1. If human urine is used, control plasma using 1 volume of saline to 1 volume of plasma is required (CONTROL).
 |
| * 1. If G.P.U. is used, the dilution factor is very minimal: parallel testing of inhibited plasma with neat plasma would be adequate (CONTROL).
 |
| * 1. Incubate both test and control plasma at room temperature for 30 minutes.
 |
| * 1. Test the control and test plasma with appropriate red cell samples at room temperature, 37°C and antiglobulin phases.
 |

1. **Reporting**
	1. Interpretation of results:

|  |  |  |
| --- | --- | --- |
| TEST PLASMA | CONTROL PLASMA | INTERPRETATION |
| No reactivity | No Reactivity | Dilution of antibody has occurredNo conclusion can be made |
| Reactivity | Reactivity | Specificity not Anti-Sda |
| No Reactivity | Reactivity | Anti-Sda present |

1. **Procedural Notes**
	1. G.P.U. is very alkaline (pH 8.8); some antibodies may fail to react at this pH. The G.P.U. can be acidified to approximately pH 7 with HCl.
	2. Human urine may also contain ABO and Lewis group substances depending on the secretor status of the donor(s).9.2
2. **References**
	1. Judd, WJ ed. Judd’s Methods in Immunohematology, 3rd ed., Bethesda, MD; 2008: 323-326.
	2. Roback, JD. ed. AABB Technical Manual, 17th ed. Bethesda, MD: American Association of Blood Banks; 2011:913-914.
3. **Revision History**

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| --- | --- |
| **Revision Date** | **Summary of Revision** |
| September 1, 2014 | * Revised name of manual
* Revised wording of section 1.0
* Added “suspected of containing anti-Sda” to section 3.0
* Replaced “Normal Saline” with “0.9% Saline” in section 4.0- *Reagents*
* Revised and renumbered sections 6.0 and 8.0
* Updated list of references to include most recent editions
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