1. **Principle**

Chloroquine diphosphate reagent may be used to remove IgG antibody, which is bound to a patient’s cells in order to allow for adsorption of an autoantibody from the plasma. This allows for the detection of alloantibodies, which may be masked by the presence of the autoantibody.

1. **Scope and Related Policies**
	1. This procedure must not be used for recently transfused (< 3 months) patients as alloantibodies could be adsorbed by the circulating donor red cells.
	2. If the antigen to the suspected antibody is destroyed by ZZAP (Kell, Lutheran) or enzymes (MNS) chlorquine treated cells should be used for adsorption.
	3. This procedure should be considered for the elucidation of problems involving:
		* autoantibodies
		* multiple antibodies
		* antibodies to high/low frequency antigens
		* "Like" antibodies or weakly reactive antibodies

1. **Specimen**

EDTA anticoagulated whole blood preferably less than 72 hours old.

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

 Anti-IgG

Chloroquine treated autologous red cells

1. **Quality Control – N/A**
2. **Procedure**

|  |
| --- |
| * 1. For method for cell treatment refer to Procedure SP.011- Dissociation of IgG by Chloroquine Diphosphate.
 |
| * 1. Three aliquots of chloroquine treated patient cells should be prepared. Incubate the first aliquot with the patient’s plasma at 37°C for 30 minutes. Gently mix every 10 minutes. After incubation, centrifuge the tube to obtain clear plasma/cell separation (3 minutes at 3000 rpm). Harvest the adsorbed plasma and add it to the second aliquot of chloroquine treated red cells, mix well. Repeat this step for the third aliquot of treated cells.
 |
| * 1. After the final adsorption, remove the absorbed plasma and place it in a tube labeled with the patient’s name and “Autoadsorbed plasma x 3 with chloroquine”. The plasma may now be used to test for the presence of alloantibodies.
 |

1. **Reporting – N/A**
2. **Procedural Notes – N/A**

1. **References**
	1. GAMMA BIOLOGICALS, INC Serologic Techniques – Rev November 1996.
2. **Revision History**

|  |  |
| --- | --- |
| **Revision Date** | **Summary of Revision** |
| September 1, 2014 | * Revised name of manual
* Changed “red cells” to “whole blood” in section 3.0
* Changed “normal saline” to “0.9% saline” in section 4.0- *Reagents*
* Changed reference from RP.007 to SP.011 in section 6.1
* Revised wording to include “gently mix every 10 minutes” to section 6.2
* Changed the centrifugation time from 5 minutes to 3 minutes in section 6.2
 |