



# e-Network Forum

## CALIFORNIA BLOOD BANK SOCIETY

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### ***Warm autoantibody-adsorption procedures with PEG***

**A blood banker in Spain** has begun to do warm autoantibody-adsorption procedures with PEG. According to the written protocol provided to him, the harvested serum-PEG mixture must be tested in **tube** methods. However, in Spain, **gel** methods are widely used, and young technicians and doctors, such as those who work in the inquiring blood banker's laboratory, are uncomfortable with tube methods. The inquiring Spanish blood banker wants to know if there is **a modified IAT-gel test that can be used with a serum-PEG mixture?**

Before sharing the above question with the full e-network, two internationally recognized immunohematologists were consulted for their opinions, which follow.

**Expert #1** (who is a member of the CBBS!) is one of the co-authors of the original paper on the use of PEG as a potentiator. He wrote the following: "We have no experience of PEG + gel procedures and I know of only 2 abstracts on using the gel test for PEG adsorptions (Clarke et al, Trans Med 1998;830 and Hernandez-Jodra et al, Trans Med 1998;8278). The abstracts suggest that this may be an acceptable approach, but as these were only abstracts, I would need more data before I would be happy to use such an approach. I would suggest that the blood banker in Spain carry out parallel studies in his/her own laboratory, taking care to show that there is no weakening of alloantibody activity. It would be inadvisable to use the procedure without such studies. Several investigators (e.g., Judd et al, Immunohematology 2001;1782, and Combs et al, Transfusion 2001;4130S) have found that PEG adsorptions can lead to weakening of residual alloantibody; others did not find this (see Leger RM, Garratty G. Transfusion 1999;3911-16, and Cheng et al, Transfusion 2001;4113). I respect the other investigators' findings and believe that the differences in results are due to different test procedures used by the different laboratories. The differences in technique were sometimes minor, e.g., between our approach and that of Judd's group. These findings emphasize the importance of always carrying out studies in your own lab if you are going to deviate even in a minor way from a reported method or package insert!"

**Expert #2** (who lives in the Midwestern USA) wrote "There are two issues to address in response to your question. First, I do not recommend the PEG adsorption method. Second, there are technical difficulties associated with using PEG by the gel technique. We were unable to validate the PEG-adsorption method as being equivalent to the ZZAP method. We found that substantial loss of antibody occurs when adsorption tests are done with equal volumes of PEG (Gamma or 20% in-house PEG), serum and antigen negative RBCs. This loss of antibody is undoubtedly due to the well known fact that PEG precipitates proteins. This is readily observed by incubating equal volumes of serum and PEG (no RBCs) at 37 C for 15 minutes, centrifuging the precipitate and measuring the protein/IgG level of the supernate. IgG levels are about half that of serum+saline controls. Our data, which are consistent with those of Champagne K, Moulds M Transfusion 1996;36384, have been both presented orally at an AABB meeting, then published in Immunohematology Judd WJ, Dake L. PEG adsorption of autoantibodies results in loss of concomitant alloantibodies. Transfusion 2000;40(S)28. Judd WJ, Dake L. PEG adsorption of autoantibodies causes loss of concomitant alloantibodies. Immunohematology 2001;1782-5. I know of only one report concerning use of PEG in gel tests Combs MR, Issitt PD Transfusion 1997;37(S)31. The authors set tests up in two different ways. In one experiment they added 25 mL of 2% RBCs, 12.5 mL serum and 25 mL 20% PEG to the gel card incubation chamber, incubated the cards at 37 C for 15 minutes, added 200 mL PBS, mixed the contents in the chamber and centrifuged the card. In the other experiment they incubated 2 drops of serum, 2 drops of 3-4% RBCs and one drop of 34% PEG at 37C for 15 minutes, added 10 drops of PBS and mixed. They then added 150 mL of this mixture to IgG gel cards prior to centrifugation. The first method offered no advantages over a standard gel method, and the second method missed 41% of PEG-only (by tube) antibodies, In talking to Harry Malyska from ID-MTS, who assisted in designing both sets of experiments, false-positive tests occur if PEG concentrations/volumes are too high. Unwanted negatives occur when the PEG content is reduced to a level at which false positives can be avoided."

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The following comment has been received.

**ADDENDA** Jan. 9, 2005

1. **Editor's comments:** The discussion [Use of the EDTA-Glycine-Acid Method for Red Cell autoadsorption and phenotyping in patients with autoantibodies](#) to be of interest to this dialogue.

Please submit comments to the [e-Network Forum](#).



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**Posted:** June 13, 2002

**Addenda:** Jan. 9, 2005