

Greiner Bio-One VACUETTE® EDTA K3 Evacuated Blood Collection Tube Evaluation for Immunohematology

Device Names

Greiner VACUETTE® EDTA K3, 6.0mL, 13x100mm tube, Product Listing #456099

Greiner VACUETTE® EDTA K3, 3.0mL, 13x75mm tube, Product Listing #454217

Comparator Devices

Becton Dickinson Vacutainer™ Glass K₃EDTA, 7.0mL, 13x100mm tube, Product Listing #366450

Becton Dickinson Vacutainer™ PLUS K₂EDTA, 3.0mL, 13x75mm tube, Product Listing #367856

Intended Use

The Greiner VACUETTE® EDTA K3 tube provides a means of collecting and transporting an undiluted plasma specimen in a closed evacuated system. The tube contains spray-dried EDTA, yielding a ratio of 1.8mg/mL of blood when the evacuated tube is filled correctly to its fill volume. The EDTA binds calcium ions which block the coagulation cascade.^{1,2}

Specimen Collection

Blood specimens were obtained using each site's standard phlebotomy techniques referencing Standard Operating Procedures and OSHA's safety requirements for blood collection. The order of draw was randomized.

The following two tubes were drawn from each donor at two Donor Centers: 1) one Greiner VACUETTE® EDTA K3, 6.0mL, 13x100mm tube, and 2) one Becton Dickinson Vacutainer™ Glass K₃EDTA, 7.0mL, 13x100 tube. In addition, one Greiner VACUETTE® EDTA K3, half evacuated to simulate half draw, 6.0mL, 13x100mm tube was collected from each of the 10 known red cell antibody positive donors at Donor Center #1. The following two tubes were drawn from each patient at the University Hospital: 1) one Greiner VACUETTE® EDTA K3, 3.0mL, 13x75mm tube and 2) one Becton Dickinson Vacutainer™ PLUS K₂EDTA, 3.0mL, 13x75mm tube.

A. Donor Center - Site #1:

- 1) 50 apparently healthy donors (full draw tubes)
- 2) Subset: 10 apparently healthy donors for antigen phenotyping
- 3) Subset: 10 apparently healthy donors for delayed antigen phenotyping (0, 15 or 19 days)
- 4) 15 known red cell antibody positive blood donors (full draw tubes)
- 5) Subset: 10 known antibody positive donors (half draw/half-evacuated tubes)

- 6) Subset: 10 known antibody positive individuals (full and half draw/half-evacuated tubes) for delayed testing

B. Donor Center - Site #2:

The following donors were drawn:

- 1) 52 apparently healthy donors
- 2) Subset: 10 apparently healthy donors for antigen phenotyping
- 3) 10 known red cell antibody positive donors

C. University Hospital - Site #3:

Fifty patients, as follows:

- 1) Multi-transfused [Hb SS (2), thalassemia (1) and others with antibodies (2)] (5)
- 2) Cardiology (5)
- 3) Leukemia (5)
- 4) Bone Marrow Transplant (5)
- 5) Liver Disease (5)
- 6) General Surgery (10)
- 7) General Medicine (15)

Handling Techniques

The tubes were gently mixed using eight complete inversions immediately following blood collection. Tubes were centrifuged using the laboratory's standard procedure, to separate cellular elements completely from the plasma. All but three samples were tested at each of the Donor Centers within 24 hours. At Donor Center #1, testing was delayed for two days for two positive antibody samples and three days for another positive antibody sample. In addition, ninety-five percent of the samples at the University Hospital were tested within eight hours of collection, while the remaining five percent were tested within 16 hours of collection.

Study Design

The study design was based on recommendations made by reviewers from the FDA Center for Biologics Evaluation and Research, Division of Blood Applications (CBER).

Instrumentation and Tests

A. Donor Center - Site #1:

Olympus® PK7200™ Automated Microplate System: ABO, Rh

Manual Method: DAT, Antibody Screening and Identifications, Antigen Phenotyping

Sample Stability Study/Delay in Testing: 1) Antibody Positive Samples: ABO, Rh, Antibody Screening and Identification using full and half draw/half-evacuated tubes, and 2) Antigen Phenotyping Samples: Antigen Phenotyping using full draw tubes.

B. Donor Center - Site #2:

Ortho ID-Micro Typing System™ (ID-MTS) Gel Test™: ABO, Rh, DAT, Antibody Screening and Identifications, Antigen Phenotyping

Ortho Selectogen® Reagent Red Blood Cells Two Cell Panel: Antibody Screening

Ortho's Resolve® Panel A: Antibody Panels and Identifications

C. University Hospital - Site #3:

Immucor® ABS2000: ABO, Rh, Antibody Screening

Manual Method: ABO, Rh, DAT

Standard LISS Tube Method: Antibody Screening and Identification

Discussion

ABO/Rh Typing

ABO/Rh typing was performed on matching tubes of blood from 102 apparently healthy blood donors, 25 known antibody positive donors and 50 patients. The testing was performed using an Olympus® PK7200™, an Immucor® ABS2000 and the ID-MTS Gel Test™, according to each manufacturer's recommended procedure. The positive antibody donors at Donor Center #1 had the ABO and Rh typing performed manually. In addition, ABO/Rh typing was manually performed on the fifty patients, in parallel with the Immucor® ABS2000 testing. There were no inaccurately reported results with the Greiner VACUETTE® EDTA K3 tubes when compared to the BD Vacutainer™ EDTA tubes.

Antigen Phenotyping

Antigen phenotyping was performed on matching tubes of blood from 20 apparently healthy blood donors. The samples were screened for the most common antigens of the Rh (C, E, c, e), Kell (K), Duffy (Fy^a, Fy^b), Kidd (Jk^a, Jk^b), and MNS (M, N, S, s) blood group systems. The distribution of results is summarized in Table #1.

Table #1		
	Donor Center - Site #1 (#Pos/#Neg)	Donor Center - Site #2 (#Pos/#Neg)
C	6/4	5/5
E	1/9	2/8
c	8/2	NT
e	9/1	NT
K	0/10	1/9
k	NT	10/0
Fy ^a	7/3	6/4
Fy ^b	9/1	5/5
Jk ^a	8/2	9/1
Jk ^b	5/5	7/3
S	7/3	4/6
s	10/0	10/0
M	7/3	NT
N	8/2	NT

* NT = Not Tested

Antibody Screening and Identification Full Draw Tube

Antibody screening was performed on 102 apparently healthy blood donors, 25 known positive blood donors, and 50 patients using the full draw Greiner VACUETTE® EDTA K3 tube and the BD Vacutainer™ EDTA tubes. The testing was performed using the Immucor® ABS2000, the ID-MTS Gel Test™, or a manual system according to the manufacturer's recommended procedures. Antibody screening was manually performed on the 50 patients in parallel with the Immucor® ABS2000 testing. All positive antibody screening samples were followed up with antibody identification.

Concordant results were obtained between the Greiner VACUETTE® EDTA K3 tubes when compared to the BD Vacutainer™ EDTA tubes. However, in some of the comparisons, there was a 1+ difference in reaction grade, but none of these results demonstrated a change to a negative reading. This variation is within the expected reproducibility of a subjective grading system.

Half-Draw Tube

In addition, ABO/Rh typing, antibody screening and antibody identification were performed on a subset of 10 of the known antibody positive blood donors using half draw/half-evacuated Greiner VACUETTE® EDTA K3 tubes and full draw BD Vacutainer™ Glass K₃EDTA tubes. The testing was performed manually, according to the Donor Center's established procedure.

Concordant results were obtained between the half draw/half-evacuated Greiner VACUETTE® EDTA K3 tubes and the full draw BD Vacutainer™ Glass K₃EDTA tubes. However, in some of the comparisons, there was a 1+ difference in reaction grade, but none of these results demonstrated a change to a negative reading. This variation is within the expected reproducibility of a subjective grading system.

Delay in Testing

Ten of the antigen phenotyping samples and 10 of the known antibody positive blood donor samples (full and half draw/half-evacuated tubes) were stored at 2-8°C following initial testing. Testing was repeated at 15-19 days after collection. The antigen phenotyping samples were only repeated for antigen phenotyping testing. The antibody positive blood donor samples were repeated for ABO/Rh typing and antibody screening and identification.

Concordant results were obtained between the full and half draw/half-evacuated Greiner VACUETTE® EDTA K3 tubes and the full draw BD Vacutainer™ Glass K₃EDTA tubes at Day 14. However, in some of the comparisons, there was a 1+ difference in reaction grade. This variation is within the expected reproducibility of a subjective grading system. A decrease in grading results was observed in some samples between Day 0 the last day of testing (Day 15 or 19).³ This is also not unexpected, considering the age of the sample.

DAT

Antibody screening was performed on 102 apparently healthy blood donors, 10 of the known positive blood donors, and 50 patients using the Greiner VACUETTE® EDTA K3 tubes and the BD Vacutainer™ EDTA tubes. There were no DAT positive results among the 112 blood donors and only one positive DAT positive result among the hospitalized patients' samples. Concordant results were obtained with the Greiner VACUETTE® EDTA K3 tubes and the BD Vacutainer™ EDTA tubes.

In addition, a panel of five simulated DAT positive samples were prepared and tested at Donor Center - Site #1 using the Greiner VACUETTE® EDTA K3 tubes and the BD Vacutainer™ Glass K₃EDTA tubes. Preparation of the coated red cells followed the procedure for using red cells coated with Anti-Fy^a described in the FDA Center for Biologics Evaluation and Research Guidance Document, "Recommended Methods for Anti-Human Globulin Evaluation", issued in March 1992.⁴ The dilutions used in this study were selected to represent a range of positive reactivity. The samples were tested on Day 0 (date of preparation) and repeated on Days 7 and 14. Concordant results were obtained between the Greiner VACUETTE® EDTA K3 tubes and the BD Vacutainer™ Glass K₃EDTA tubes on Days 0, 7 and 14. In some of the samples, there was a 1+ difference in reaction grade of the results. This variation is within the expected reproducibility of a subjective grading system.

Conclusion

The Greiner VACUETTE® EDTA K3 tubes (full and half draw/half-evacuated) demonstrated substantial equivalence to the Becton Dickinson Vacutainer™ Glass K₃EDTA and PLUS K₂EDTA tubes with various standard assays using donor and

recipient populations. Antigen and antibody identification did not change over time when samples were stored in the Greiner VACUETTE® EDTA K3 tubes, demonstrating that these proteins were not adsorbed onto the plastic walls of the tubes and interfering substances were not leached from the walls of the tubes.^{5,6,7,8,9,10,11}

References

1. Greiner Bio-One. Evacuated Blood Collection System For In Vitro Diagnostic Use. Product Insert. Kremsmunster, Austria. 2001.
2. Gruber, H. Greiner Bio-One. Product Manual. Kremsmunster, Austria. July 2002.
3. Sandler, G.S., M.D. Personal Communication. Georgetown University Hospital. July 2003.
4. FDA Center for Biologics Evaluation and Research Guidance Document. Recommended Methods for Anti-Human Globulin Evaluation. March 1992.
5. Greiner Bio-One 510(k) Submission. Blood Collection Tube - EDTA K3 Pre-Market Notification Addition of Immunoematology Claim. Monroe, NC. December 2002.
6. Kemper, M. Final Report: Greiner VACUETTE® Evacuated Blood Collection Tubes Blood Bank Study K₂EDTA, K₃EDTA vs. BD K₃EDTA. SMF-Center for Blood Research. Sacramento, California. March 14, 2003.
7. Kemper, M. Personal Communication. SMF-Center for Blood Research. Sacramento, California. March 2003.
8. Korte, L. Final Report: Comparison of Greiner Vacuette Blood Collection Tubes. Gulf Coast Regional Blood Center. Houston, Texas. January 23, 2003.
9. Korte, L. Personal Communication. Gulf Coast Regional Blood Center. Houston, Texas. March 2003.
10. Langeberg, A. Personal Communication. Georgetown University Hospital. March 24, 2003.
11. Sandler, G.S., M.D. Personal Communication. Georgetown University Hospital. March 2003.

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