Evaluation of VACUETTE® K_3EDTA and K_2EDTA Evacuated Blood Collection Tubes Using the ID-Micro Typing System™ (ID-MTS) Gel Test™

Background:

Greiner Bio-One, Austria has sold plastic evacuated tubes (VACUETTE®) for venous blood collection since 1986.

Greiner VACUETTE® K_3EDTA and K_2EDTA tubes provide a means of collecting and transporting an undiluted plasma specimen in a closed evacuated system. The tubes contain spray-dried EDTA yielding a ratio of 1.8 mg/mL of blood when evacuated tube is filled correctly to its fill volume. EDTA binds calcium ions which blocks the coagulation cascade. (1) (2)

VACUETTE® EDTA tubes are used for testing whole blood in the clinical laboratory and may be used for testing in routine immunohematology i.e. red cell grouping, Rh typing and antibody screens.

Study Objective:

A clinical evaluation was carried out to compare the performance of the Greiner VACUETTE® K_3EDTA and K_2EDTA tubes to the Becton Dickinson Vacutainer® K_3EDTA glass tube using the ID-Micro Typing System™ (ID-MTS) Gel Test™.

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).

The following tube types were used in this study:

Sample No.	Description
1	VACUETTE® K_3EDTA, 6 mL (13x100 mm)
2	VACUETTE® K_2EDTA, 6 mL (13x100 mm)
3	Becton Dickinson Vacutainer® Glass K_3EDTA, 7 mL (13x100 mm) (comparator device)

Blood specimens were obtained using the test 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 donors were drawn:

1) 52 apparently healthy donors

2) Subset: 10 apparently healthy donors for antigen phenotyping

3) 10 apparently healthy donors with known red cell antibodies

The following tubes were drawn from each donor:

1) one Greiner VACUETTE® K_3EDTA, 6 mL, 13x100mm tube

2) one Greiner VACUETTE® K_2EDTA, 6 mL, 13x100mm tube

3) one Becton Dickinson Vacutainer® Glass K_3EDTA, 7 mL, 13x100mm tube

The tubes were gently mixed by 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. Plasma was tested within 24 hours.

ID-Micro Typing System™ (ID-MTS) Gel Test™

The ID-MTS is a gel card test that performs the conventional serum and cell tube reactions in a microtubule reaction chamber. Each microtubule is composed of a dexrean-acrylamide gel, suspended in a buffered saline solution. This gel may be specifically manufactured to contain other additives such as, albumin, bromelin and ABO and Rh antisera. A reagent, red cell suspension or serum is added to a specific microtubule. The ID-MTS Gel Card is then incubated for a defined time and temperature. Upon completion of this incubation, the card is centrifuged at a pre-defined speed and time in an MTS centrifuge. After centrifugation, the cards are removed and the front and back of each microtubule is read macroscopically to determine the presence of positive, negative or other end-point reactions. (3) (4) (5)

Tests Performed

ABO, Rh, DAT and Antibody Screens were performed on 52 donors' three blood samples. Antigen phenotyping was performed an a subset of 10 of the donors. An additional ten known positive donors had positive antibody panels and antibody identifications performed, using Ortho's Resolve® Panel A and the ID-MTS Gel Test™.

Conclusion:

The Greiner VACUETTE® K_3EDTA and K_2EDTA tubes yielded comparable results to the Becton Dickinson Vacutainer® Glass K_3EDTA tube for ABO/Rh, DAT, Antibody Screening and where necessary, Antibody Identification tests, using the ID-Micro Typing System™ with a donor population. (6) (7) (8) (9)

Results/Discussion:

ABO/Rh/DAT Antibody Screening

ABO/Rh/DAT and Antibody Screening were performed on matching tubes of blood from 52 apparently healthy blood donors.

The testing was performed using the ID-MTS Gel Test™, according to the manufacturer's recommended procedures. The Ortho Selectogen® Reagent Red Blood Cells Two Cell Panel was used for antibody screening.(10) The results from each matched group of samples were compared. There were no discordances noted. All matched results showed the same reaction strength, with a few results differing only by one reaction grade. (11)

Antigen Phenotyping

Ten of the donors were also phenotyped using a panel of ten antisera. 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 (see Annex).

The ID-MTS Gel Test™ was used for phenotyping the donor cells after internal validation was performed using the manufacturer's guidelines for IgG gel testing. (12) (13) (14) (15)

Antibody Identification

Ten known positive donors had positive antibody panels and antibody identifications performed, using Ortho's Resolve® Panel A and ID-MTS Gel Test™. (16) The identification results are described in Table #2 (see Annex).

References:

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Annex/Results in detail:

Table #1 Antigen Phenotyping		
Antigen Phenotyping		
Antigen	**K_3EDTA (#Pos/#Neg)**	**K_2EDTA (#Pos/#Neg)**
C	5/5	5/5
E	2/8	2/8
c	NT	NT
E	NT	NT
K	1/9	1/9
k	10/0	10/0
Fy^a	6/4	6/4
Fy^b	5/5	5/5
Jk^a	9/1	9/1
Jk^b	7/3	7/3
S	4/6	4/6
s	10/0	10/0
M	NT	NT
N	NT	NT

*NT = Not tested

Table #2				
Antibody Phenotyping				
Known Ab+ Donors	ABO	Rh	Antibody	Details
1	0	+	Anti-E	*one E heterozygous cell reacted with the Greiner K_3EDTA
2	B	-	Anti-SC1	*SC1 is a high incident antigen with 99% cells positive (Note: donor historically known to have underlying Anti-D)
3	A	-	Anti-D, Anti-C, Anti-E	*one heterozygous C cell negative with all tubes
4	A	-	Anti-D, Anti-C, Anti-Fy^a	
5	A	-	Anti-D	
6	0	-	Anti-D	
7	0	+	Anti-K	
8	0	+	Anti-M	*historical conclusion, weak reactions detected on M homozygous cells only, probably due to the IgM nature of antibody
9	A	-	Anti-D	
10	0	-	Anti-K	