The Comparison of Glass EDTA Versus Plastic EDTA Blood-Drawing Tubes for Complete Blood Count

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Abstract: Blood-drawing tubes made from plastic containing K2EDTA as an anticoagulant are gaining widespread use in clinical hematology. In this study it is compared complete blood count parameters, white blood cell differentials and flagging rates obtained with Becton Dickinson Vacutainer K3EDTA glass tubes and Vacutainer K2EDTA Plus plastic tubes and found only slight discrepancies in the results obtained with the two tube types. Although some parameter values obtained with K3EDTA glass tubes were significantly lower than those obtained with K2EDTA plastic tubes, many of these differences could be explained by the known effects of the liquid K3EDTA anticoagulant. Flagging rates on an automated cell counter were identical for the two tube types. Finally, the conclusion of this study defined that the differences between results obtained with K3EDTA glass tubes versus K2EDTA plastic tubes are minimal and unlikely to be of any clinical significance.

Key words: Cell counters % Glass tubes % Plastic tubes % Hematology % EDTA

INTRODUCTION

For the last 50 years, blood-drawing tubes made from glass have been the standard devices for obtaining blood from patients for clinical laboratory testing [1]. Recently, concerns for the safety of laboratory employees and for biological waste disposal have led to the development of plastic tubes. Plastic tubes have several advantages over glass tubes: increased shock resistance, tolerance of higher centrifugation speeds and reduced solid waste after incineration [2]. In addition, the slight flexibility of plastic tubes makes them more suitable for use in an automated laboratory with robotics-based sample handling [1]. A 1.8 mg/mL (15%) solution of K3EDTA has been widely used as an anticoagulant in blood collection tubes. However, recent studies have shown that dry K2EDTA may be better suited as an anticoagulating agent, because its use avoids specimen dilution and it has less influence on mean corpuscular volume (MCV) than does K3EDTA [3, 4]. The International Council for Standardization in Hematology has recommended the use of K2EDTA as the anticoagulant of choice in specimen collection for blood cell counting and sizing [5]. Because of the advantages of plastic tubes and K2EDTA as an

anticoagulant, blood-drawing tubes made from plastic containing K2EDTA are rapidly gaining widespread use. In spite of the theoretical advantages of these tubes, it is important to ascertain that the interaction of blood specimens with the tube material does not change laboratory results. Several reports have compared results obtained with glass and plastic tubes for parameters measured in clinical chemistry [1, 2, 6], endocrinology [7], molecular testing [8], serology [9-11] and coagulation [12- 14]. Manufacturers of blood-drawing tubes have provided product monographs and white papers showing an excellent correlation of results for clinical hematology analytes drawn into glass and plastic tubes and with K3EDTA or K2EDTA as the anticoagulant [15]. Therefore this study compared the results obtained with glass and plastic tubes for these basic hematologic parameters.

MATERIALS AND METHODS

Blood-drawing Tubes: Blood samples were drawn into either BD Vacutainer K3EDTA glass tubes or into BD Vacutainer K2EDTA Plus plastic tubes (Becton Dickinson, Franklin Lakes, NJ, USA). Vacutainer Plus tubes are clear, shatter-resistant plastic blood collection

tubes made of polyethylene terephthalate. The glass tubes contained liquid K3EDTA as the anticoagulant and the plastic tubes were spray-coated with dry K2EDTA.

Paired Samples from Healthy Donors: Paired blood samples from 50 healthy blood donors were collected by trained technologists into BD Vacutainer K3EDTA glass tubes and into BD Vacutainer K2EDTA Plus plastic tubes. Both samples were obtained from a single puncture and the order of the tubes (plastic or glass) was alternated randomly. Samples were processed in parallel and analyzed in adjacent positions on the cell counter. All samples were processed within 4 h of draw.

Complete Blood Count and White Cell Differentials: CBC and WBC differentials were performed on an Beckman Coulter AcT 5 Diff Hematology Analyzer (Beckman Coulter, Minnesota, USA). This analyzer uses a cyanmethemoglobin method for the measurement of hemoglobin, isovolumetric sphering and light scatter for all other red cell and platelet parameters and Absorbance cytochemistry and Volume (AcV) technology. Monocyte, neutrophil and eosinophil populations are identified, using the absorbance patterns produced by differential cytochemical staining of their granules versus volume. Lymphocytes remain unstained and the basophil population is analyzed on a separate channel using volume gating and selective lysis. The CBC parameters included WBC counts, hematocrit, hemoglobin, red blood cell count, platelets, MCV, mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC) and red cell distribution width (RDW). The WBC

differential consisted of the percentages of neutrophils, lymphocytes, monocytes, eosinophils and basophils.

Statistical Analysis: Statistical analysis was performed with Microsoft Excel software. A 2-tailed paired *t* test was used to evaluate differences between tube types and the Pearson correlation coefficient was used to assess correlation.

RESULTS

Paired blood samples from 50 healthy volunteers were collected into glass K3EDTA and plastic K2EDTA blooddraw tubes and analyzed on an automated cell counter. Basic CBC parameters and 5-part WBC differentials were obtained from all 50 samples. Mean results and standard deviations for all parameters are shown in Table 1. The mean WBC, red blood cell, platelet and reticulocyte counts and hemoglobin, hematocrit, MCV, MCHC and RDW values were significantly lower statistically for specimens collected into glass tubes than for samples collected into plastic tubes (Table 1). However, these differences were minimal (approximately 2.1%) and not likely to be of any clinical relevance. There were no statistically significant differences in the percentages of neutrophils, lymphocytes, eosinophils, or basophils between samples collected into glass or plastic tubes. A slightly lower percentage of monocytes were observed for samples drawn into plastic tubes than for the specimens collected in glass containers. Again, the difference (5.2%), although statistically significant, was not likely to be of clinical relevance. The manual

Table 1: Basic complete blood count parameters, reticulocyte counts and white blood cell differentials in glass and plastic tubes analyzed within 4 h of the blood draw*

	WBC, ×10 ⁹ /L		RBC, ×10 ¹² /L		Hgb (g/dl)		Hematocrit (%)		Platelets, ×10 ⁹ /L	
	Glass	Plastic	Glass	Plastic	Glass	Plastic	Glass	Plasti	c Glass	Plastic
Mean	6.56	6.86	4.34	4.82	13.0	14.2	40	45	261	289
SD	2.62	2.73	0.39	0.41	0.97	1.01	39	42	52	54
P value	0.0004		< 0.0001		0.0003		< 0.0001		0.005	
	MCV (fL)		MCH (pg)			MCHC (g			RDW (%)	
	Glass	Plastic	Glass		Plastic	Glass	Pla	astic	Glass	Plastic
Mean	80.5	85.2	27.8		28.2	33.5	31	.5	14.0	13.8
SD	4.16	3.99	1.52		2.0	1.3	1.2	28	0.09	0.089
P value	< 0.0001		0.41			0.002			0.12	
	Neutrophils (k/μl)		Lymphocytes (k/µl)		Monocytes (k/µl)		Eosinophils (k/µl)		Basophils (k/µl)	
	Glass	Plastic	Glass	Plastic	Glass	Plastic	Glass	Plastic	Glass	Plastic
Mean	5.32	5.81	3.15	2.85	0.81	0.71	0.30	0.28	0.1	0.1
SD	0.71	0.72	0.65	0.61	0.23	0.26	0.12	0.14	0.04	0.04
P value	0.12		0.25		0.65		0.066		0.42	

assessment of WBC counts and differentials that have been rejected by an automated analyzer is one of the most time-consuming tasks in routine hematology laboratory analysis [16].

DISCUSSION

In this study it is compared the performance of glass K3EDTA and plastic K2EDTA tubes for complete blood counts, reticulocyte counts automated white cell differentials and found that although statistically significant differences occurred between these sampling systems. The results confirm the data provided by the tubes' manufacturer and are very similar to other studies that have analyzed the effects of tube material on other laboratory parameters. For example, Hill and coworkers compared plastic blood-drawing tubes containing a serum-separating barrier gel for common clinical chemistry analyses and found no significant differences in these tests [1]. Similarly, Reinartz and colleagues reported no effect of plastic tubes on results obtained for endocrinologic analytes [7]. Two studies found no significant effect of plastic tubes on therapeutic drug measurements, except for decreased levels of carbamazepine [2, 6]. The use of plastic tubes in serologic testing has been the subject of several investigations. Three studies found the results with plastic and glass tubes to be comparable in pre-transfusion serologic testing [9, 10, 17] however, Black and Kay reported that plastic tubes reduced the strength and titer of the antiglobulin test and led to weak reactions that were less clearly defined [11]. No significant impact was observed with the use of plastic Vacutainer tubes for the quantitation of human immunodeficiency virus type 1 in blood specimens [8]. Most studies of plastic tube use in hematology have focused on coagulation and erythrocyte sedimentation rate assays. Biron-Andréani and coworkers and van den Besselaar and colleagues have reported that tube type had a significant effect on coagulation parameters [13, 14]. These results followed earlier studies that showed a greater risk of clotting in glass syringes compared with plastic ones [18, 19]. A study by Pollack et al showed no differences in factor VIIa concentrations in plastic versus glass tubes [12]. The effects of tube material on the erythrocyte sedimentation rate are equivocal: one study found identical results for plastic and glass tubes [20], but another investigation reported a significantly higher sedimentation rate in plastic pipettes [21]. Furthermore, other group concludes that there were no significant differences between results obtained with EDTA glass tubes and EDTA Plus plastic tubes, many parameter values were significantly lower statistically in

the glass tubes than in the plastic tubes [22]. Similarly, same observations were detected in this study. These differences are most likely due to the difference in the anticoagulant present in the two types of tubes and not to the material composition of the tubes. K3EDTA, the anticoagulant in the glass tubes, is dispensed as a liquid and is known to lead to 1.5 to 2.5% lower values because of specimen dilution [5]. Similarly, K3EDTA is known to cause lower MCV values because of red blood cell shrinkage [5]. This effect of the type of anticoagulant on the MCV, which has been shown to be influenced by the type of automated cell counter used and by the pH of the blood sample [22, 23] may explain the slightly lower MCV seen in glass tubes than in the plastic tubes in this study. According to the International Council for Standardization in Hematology, K2EDTA is the preferred anticoagulant for hematology measurements [5]. An important limitation of these data is that they were mostly obtained from healthy blood donors. Further studies will be needed to establish the equivalence of plastic K2EDTA and glass K3EDTA in patients with pathologic conditions. Such studies will be especially important in light of the reports in the literature that indicate that the effects of anticoagulants on laboratory parameters are influenced by the pH of the blood sample [23].

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