Comparison of VACUETTE® K_2EDTA and VACUETTE® K_3EDTA Tubes

Background:

The VACUETTE® evacuated blood collection tubes are used for testing parameters in haematology. The tubes are available with interior coated spray-dried K_2EDTA (dipotassium ethylenediaminetetraacetic acid) or K_3EDTA (tripotassium ethylenediaminetetraacetic acid).

Both EDTA salts inhibit the coagulation of the blood specimen by binding Calcium (Ca^{2+}), thus preserving the blood cells for test analyses.[1]

The K_2EDTA and K_3EDTA additives preserve erythrocytes, leucocytes and thrombocytes up to 24 hours. The differential should be analyzed and the peripheral smear be made within 3 hours of specimen collection.[2]

Study Objective:

A clinical evaluation was carried out to compare the performance of the VACUETTE® K_2EDTA tube to the VACUETTE® K_3EDTA tube.

Study design:

The following tube types were used in this study:

Sample ID	Description
A	VACUETTE® K_2EDTA 4 ml, spray dried (item No.: 454023)
B	VACUETTE® K_3EDTA 4 ml, spray dried (item No.: 454021)

Blood was collected from forty-six normal and abnormal donors. The instructions for use[2,] was followed. The order of draw was also randomized.

All salts of EDTA are hyperosmolar, which causes water to leave the cells and results in cell shrinkage. The higher the concentration of EDTA, the greater the osmotic withdrawal of water from the cells. It was therefore ensured that the tubes are filled-completely.

In addition, under-filling of the tubes also decreases the blood to additive ratio, resulting in cell shrinkage (reduction of the Mean Corpuscular Volume and an increase of the Mean Corpuscular Haemoglobin Concentration[3]). The K_3EDTA Tubes may be slightly more affected, because of the presence of the higher potassium-ion concentration.

Blood specimens were obtained using the institution's standard phlebotomy techniques. Immediately following blood collection, the tubes were gently inverted 8 to 10 times to ensure proper mixing of the blood and additive in the specimens.

Determination of the most common parameters (listed below) in hematology was performed using the Sysmex XE2100 Hematology Analyzer with accompanying reagents.

Specimens were analyzed as follows:

1) Seven donors were analyzed within 15 minutes after blood collection.

2) Fifteen donors were analyzed between 15-30 minutes after blood collection.

3) Twenty-four donors were analyzed between 30 minutes and 3 hours after blood collection.

4) All samples were reanalyzed 24 hours after blood collection

The tubes were stored at room temperature.

The results were checked for correctness. If an outlier was observed, an investigation was conducted and the test was repeated.

The evaluation of the results included directly measured and calculated parameters. [4,5,6]

Directly measured parameters included Leukocytes, Erythrocytes, Haemoglobin, Hematocrit and Thrombocytes.

The calculated parameters included Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH) and Mean Corpuscular Haemoglobin Concentration (MCHC).

The differential parameters included Neutrophile Granulocytes, Lymphocytes, Monocytes, Eosinophile Granulocytes and Basophile Granulocytes.

Conclusion:

The VACUETTE® K_3EDTA tube demonstrated substantially equivalent performance to the VACUETTE® K_2EDTA tube. No clinically significant differences were observed.

References:

(1) NCCLS. *Tubes and Additives for Venous Blood Specimen Collection; Approved Standard—Fifth Edition*. NCCLS document H1- A5 (ISBN 1-56238-519-4). NCCLS, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087-1898 USA, 2003.

(2) Gruber, H., *Product Manual: VACUETTE® Evacuated Blood Collection System- For In Vitro Diagnostic Use*. Rev.06. 2005, Kremsmünster, Austria: Greiner Bio-One GmbH.

(3) Chen BH, Fong JF, Chiang CH, *Effect of different anticoagulant, underfilling of blood sample and storage stability on selected hemogram*. The Kaohsiung journal of medical sciences, 1999. 15(2): p. 87-93.

(4) *Sysmex XE-2100 Operator´s Manual*. 1999, 2003: Scientific Center Sysmex Corporation.

(5) NCCLS. *Method Comparison and Bias Estimation Using Patient Samples; Approved Guideline—SecondEdition*. NCCLS document EP9-A2 (ISBN 1-56238-472-4). NCCLS, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087-1898 USA, 2002.

(6) *Richtlinie der Bundesärztekammer zur Qualitätssicherung quantitativer laboratoriumsmedizinischer Untersuchungen*, Bundesärztekammer (Arbeitsgemeinschaft der deutschen Ärztekammer). 2001, Rev. 2003.

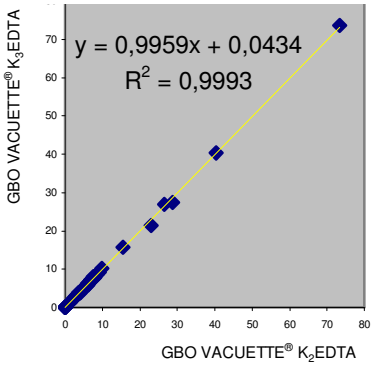
Results in detail:

Leucocytes (WBC)

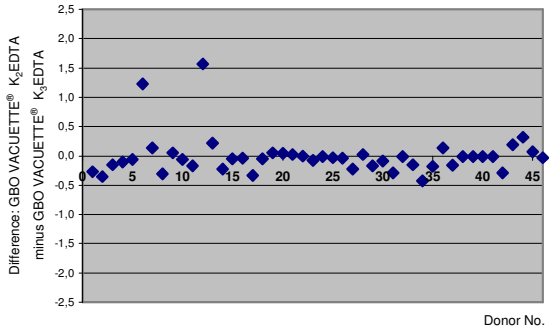
Normal range: 4,0 – 9,0 [10³/µL]

Measurement 0-3h after blood collection:

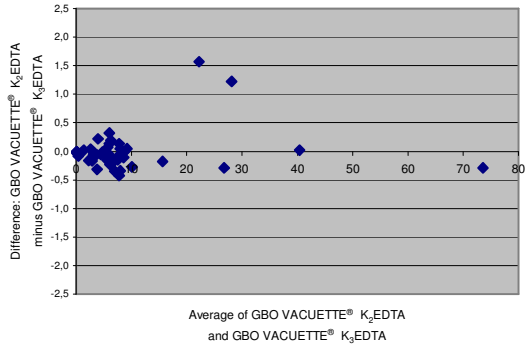
Regression WBC [10³/µL]



Deviation plot, y-axis WBC [10³/µL] :



Bland-Altman plot WBC [10³/µL]:

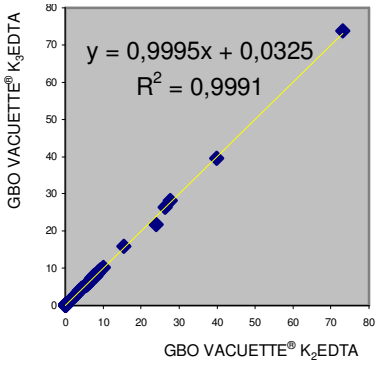


Result paired two tailed t-test at a confidence level of 95%:

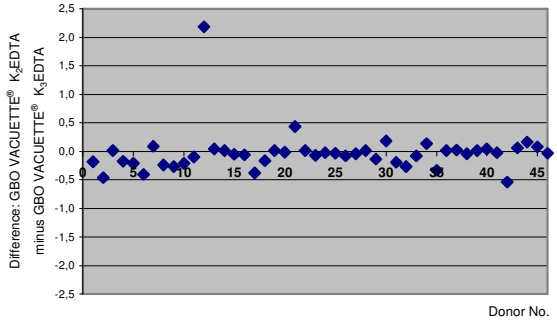
N=46
P-value= 0,871
Critical P= 0,05
No significance

Measurement 24h after blood collection:

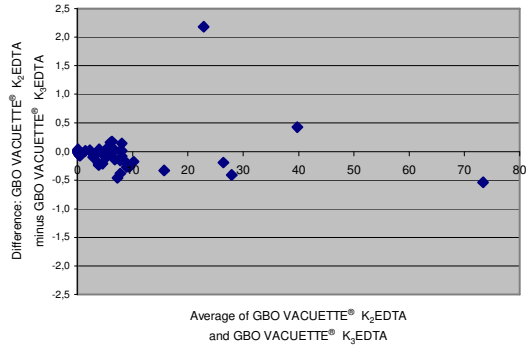
Regression WBC [10³/µL]



Deviation plot, y-axis WBC [10³/µL] :



Bland-Altman plot WBC [10³/µL]:



Result paired two tailed t-test at a confidence level of 95%:

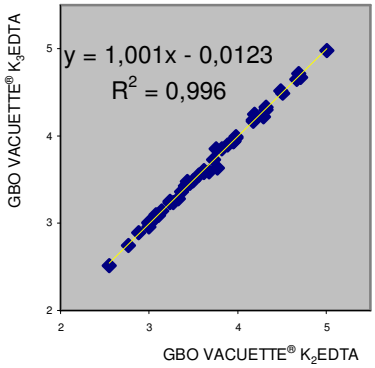
N=46
P-value= 0,614
Critical P= 0,05
No *significance*

Erythrocytes (RBC)

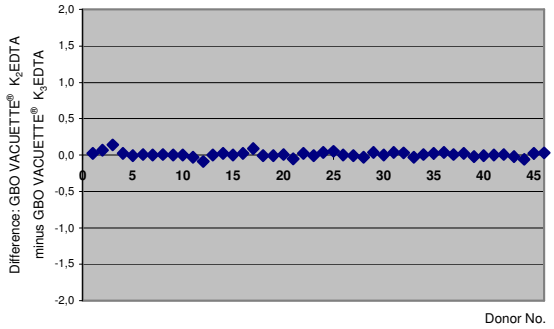
Normal range: 4,5 – 6,0 [10^6/µL] (male); 4,2 – 5,5 [10^6/µL] (female)

Measurement 0-3h after blood collection:

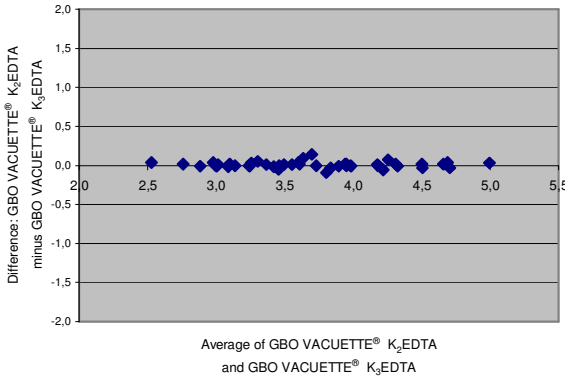
Regression RBC [10^6/µL]:



Deviation plot y-axis RBC [10^6/µL]:



Bland-Altman plot RBC [10^6/µL]:

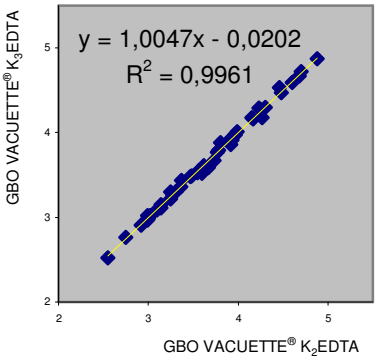


Result paired two tailed t-test at a confidence level of 95%:

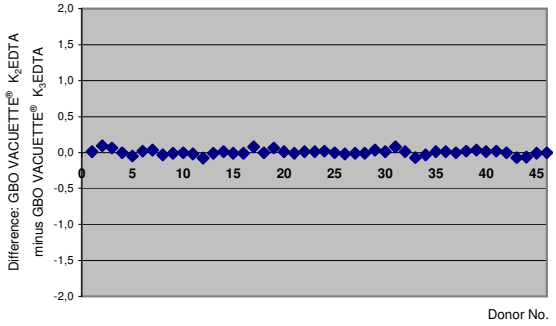
N=46
P-value= 0,119
Critical P= 0,05
No significance

Measurement 24h after blood collection:

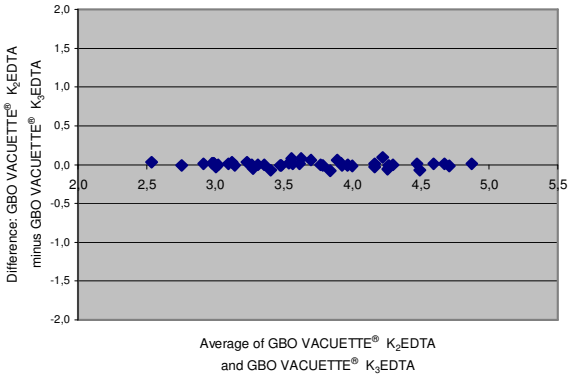
Regression RBC [10^6/µL]:



Deviation plot, y-axis RBC [10^6/µL]:



Bland-Altman plot RBC [10^6/µL]:



Result paired two tailed t-test at a confidence level of 95%:

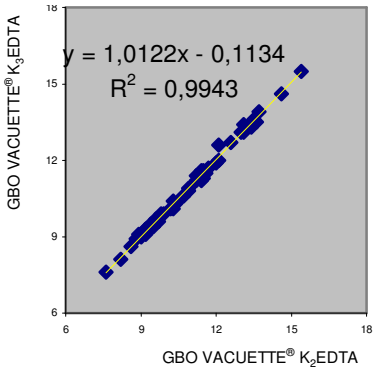
N=46
P-value= 0,598
Critical P= 0,05
No significance

Haemoglobin (HGB)

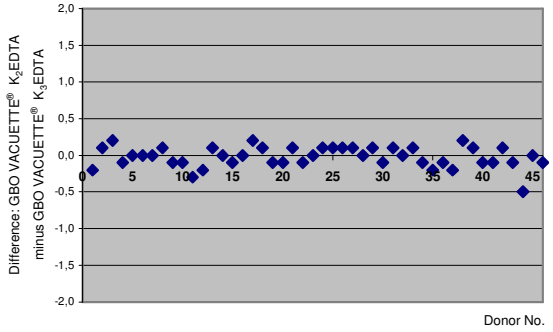
Normal range: 13,5 – 18,0 [g/dL] (male); 12 – 16,5 [g/dL] (female)

Measurement 0-3h after blood collection:

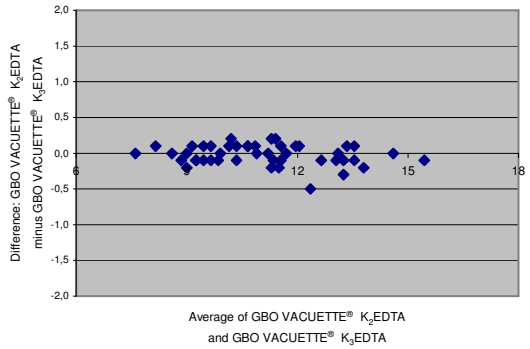
Regression HGB [g/L]:



Deviation plot, y-axis HGB [g/L]:



Bland-Altman plot HGB [g/L]:

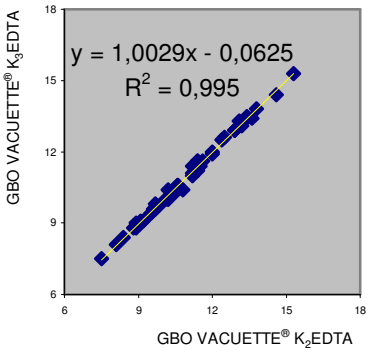


Result paired two tailed t-test at a confidence level of 95%:

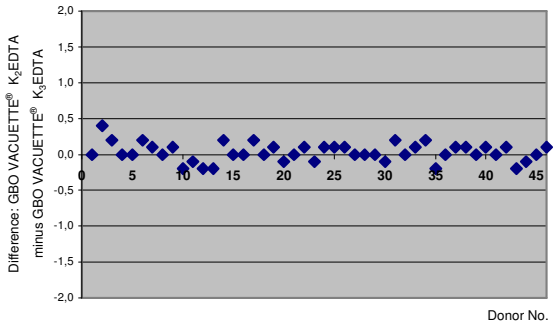
N=46
P-value= 0,297
Critical P= 0,05
No significance

Measurement 24h after blood collection:

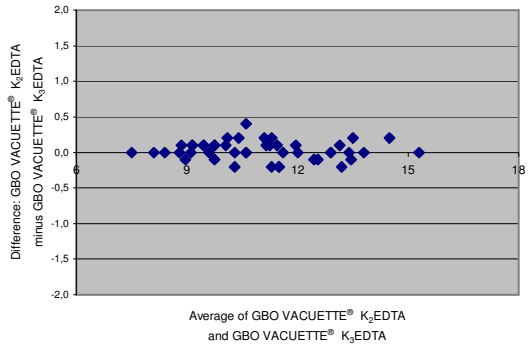
Regression HGB [g/L]:



Deviation plot, y-axis HGB [g/L]:



Bland-Altman plot HGB [g/L]:



Result paired two tailed t-test at a confidence level of 95%:

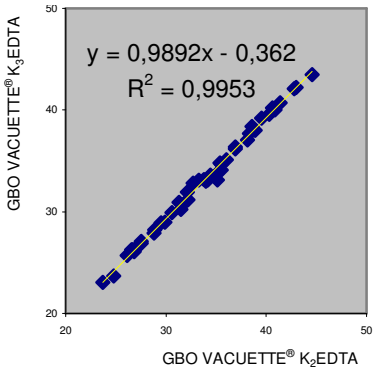
N=46
P-value= 0,114
Critical P= 0,05
No significance

Haematocrit (HCT)

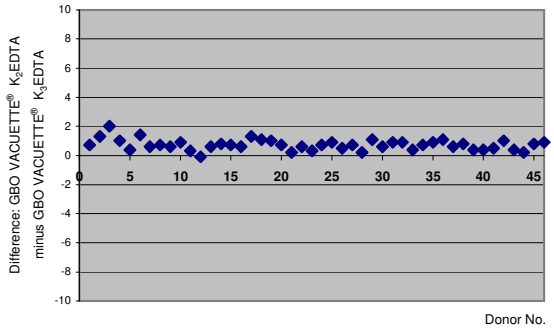
Normal range: 40 – 52 [%] (male), 36 – 48 [%] (female)

Measurement 0-3h after blood collection:

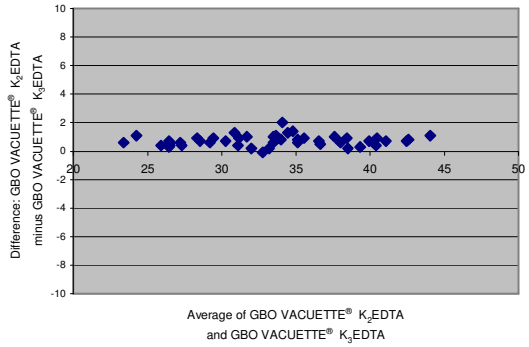
Regression HCT [%]:



Deviation plot y-axis HCT [%]:



Bland-Altman plot HCT [%]:

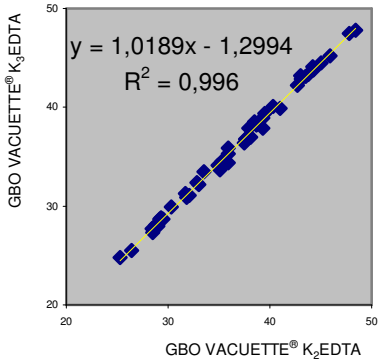


Result paired two tailed t-test at a confidence level of 95%:

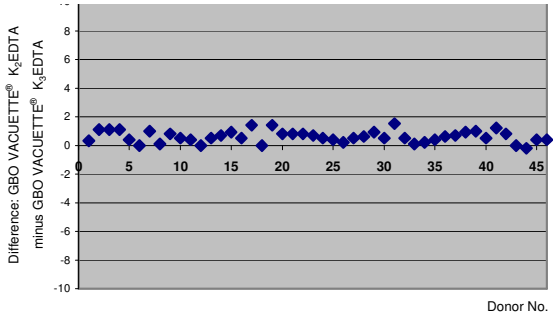
N=46
P-value= 0,000
Critical P= 0,05
Significant

Measurement 24h after blood collection:

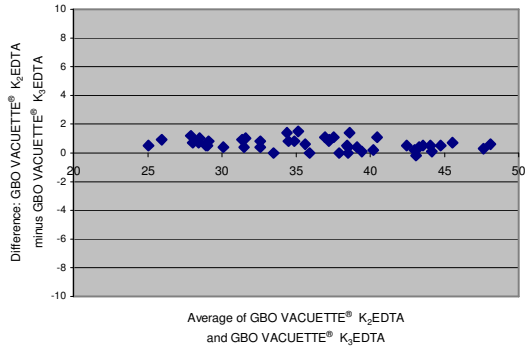
Regression HCT [%]:



Deviation plot, y-axis HCT [%]:



Bland-Altman plot HCT [%]:



Result paired two tailed t-test at a confidence level of 95%:

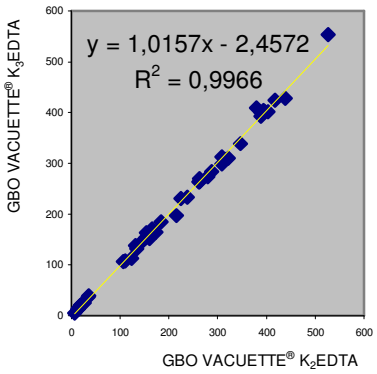
N=46
P-value= 0,000
Critical P= 0,05
Significant

Thrombocytes (PLT)

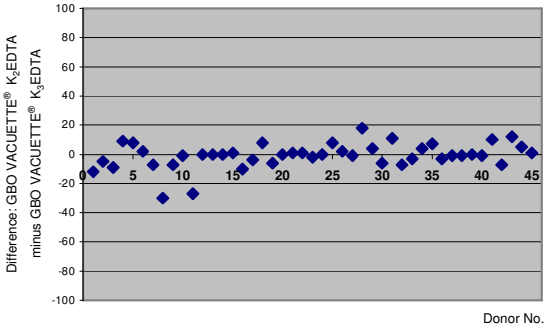
Normal range: 130 – 440 [10^6/µL]

Measurement 0-3h after blood collection:

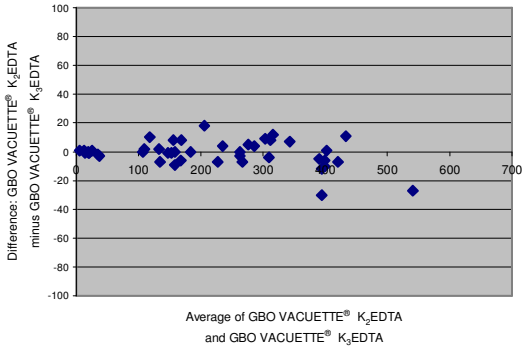
Regression PLT [10^6/µL]:



Deviation plot y-axis PLT [10^6/µL]:



Bland-Altman plot PLT [10^6/µL]:

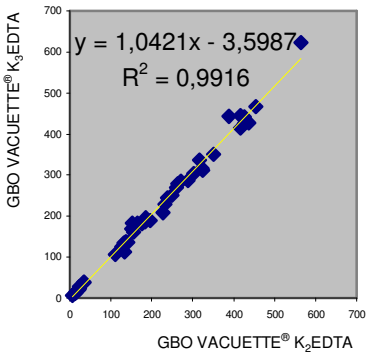


Result paired two tailed t-test at a confidence level of 95%:

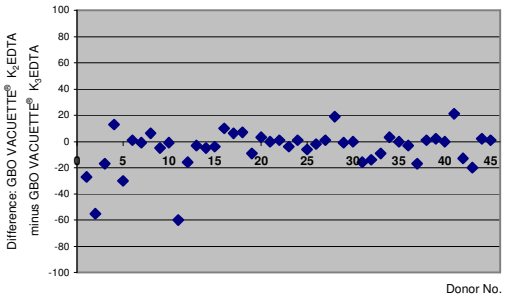
N=45
P-value= 0,307
Critical P= 0,05
No significance

Measurement 24h after blood collection:

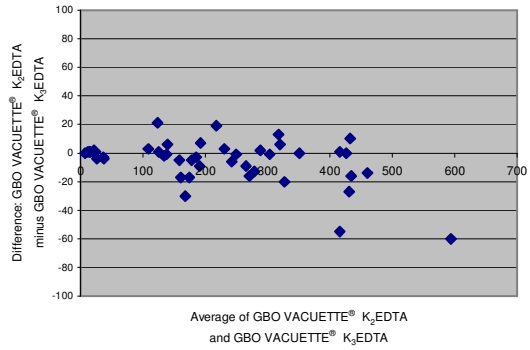
Regression PLT [10^6/µL]:



Deviation plot, y-axis PLT [10^6/µL]:



Bland-Altman plot PLT [10^6/µL]:



Result paired two tailed t-test at a confidence level of 95%:

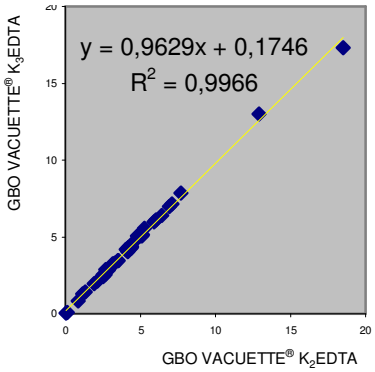
N=45
P-value= 0,024
Critical P= 0,05
Significant

Neutrophile Granulocytes (NEU)

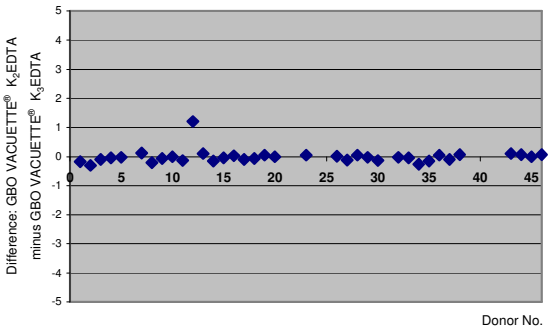
Normal range: 2,2 – 6,2 [10³/µL]

Measurement 0-3h after blood collection:

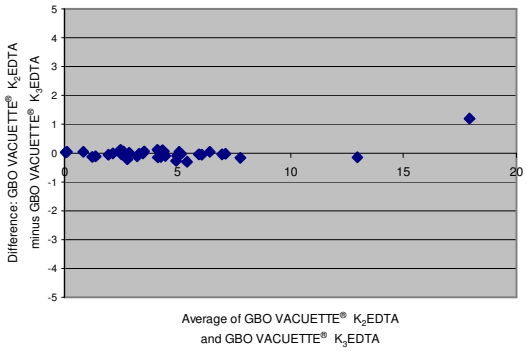
Regression NEU [10^3/µL]:



Deviation plot y-axis NEU [10^3/µL]:



Bland-Altman plot NEU [10^3/µL]:

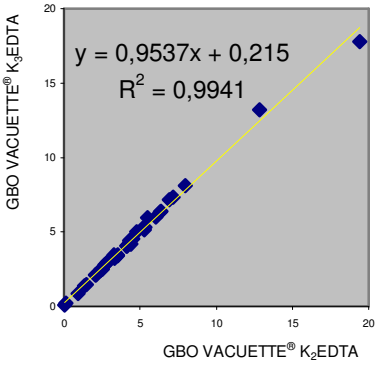


Result paired two tailed t-test at a confidence level of 95%:

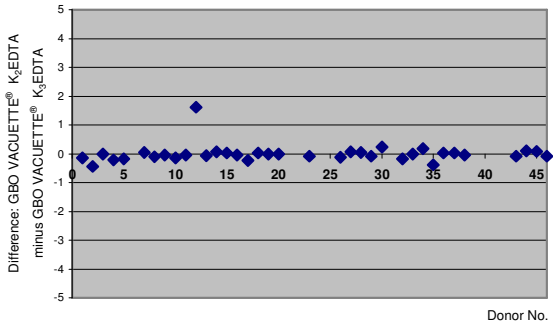
N=36
P-value= 0,786
Critical P= 0,05
No significance

Measurement 24h after blood collection:

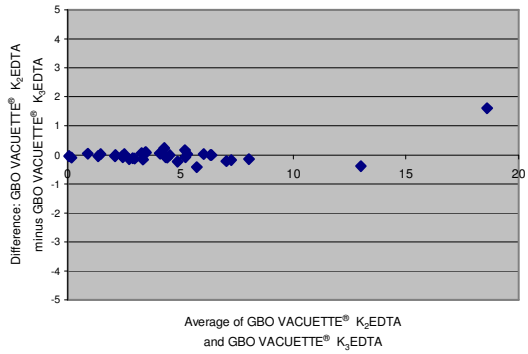
Regression NEU [10^3/µL]:



Deviation plot, y-axis NEU [10^3/µL]:



Bland-Altman plot NEU [10^3/µL]:



Result paired two tailed t-test at a confidence level of 95%:

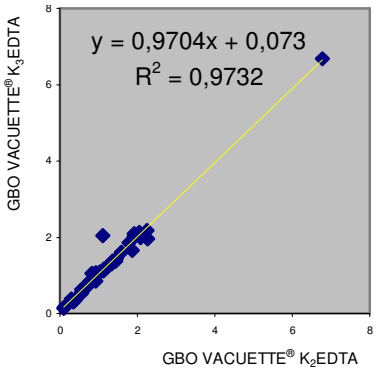
N=36
P-value= 0,906
Critical P= 0,05
No significance

Lymphocytes (LYMPH)

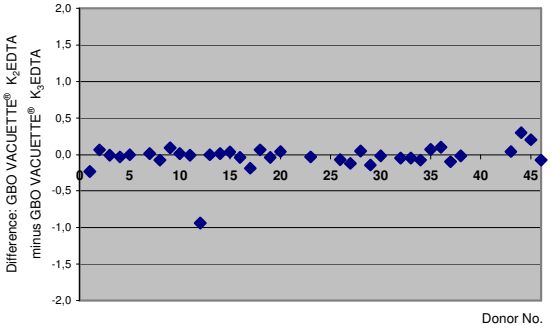
Normal range: 1,0 – 4,0[10^3/µL]

Measurement 0-3h after blood collection:

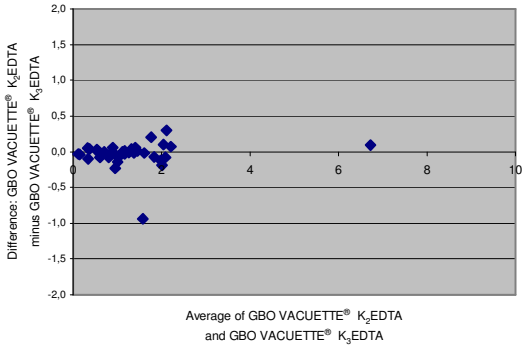
Regression LYMPH [10^3/µL]:



Deviation plot y-axis LYMPH [10^3/µL]:



Bland-Altman plot LYMPH [10^3/µL]:

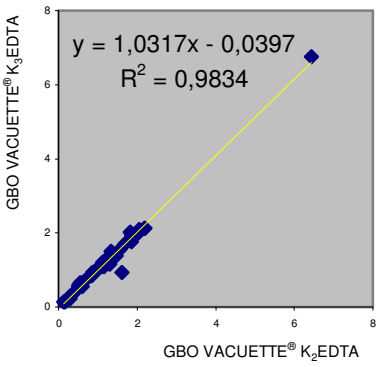


Result paired two tailed t-test at a confidence level of 95%:

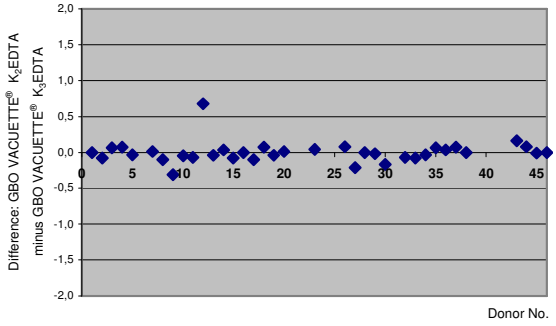
N=36
P-value= 0,259
Critical P= 0,05
No significance

Measurement 24h after blood collection:

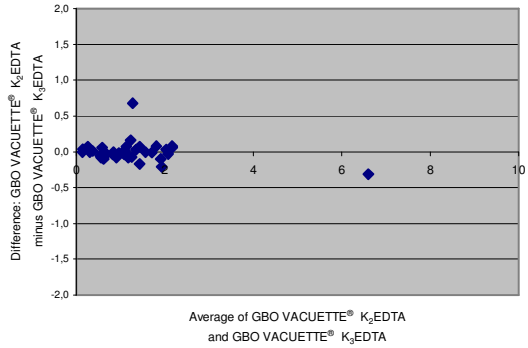
Regression LYMPH [10^3/µL]:



Deviation plot, y-axis LYMPH [10^3/µL]:



Bland-Altman plot LYMPH [10^3/µL]:



Result paired two tailed t-test at a confidence level of 95%:

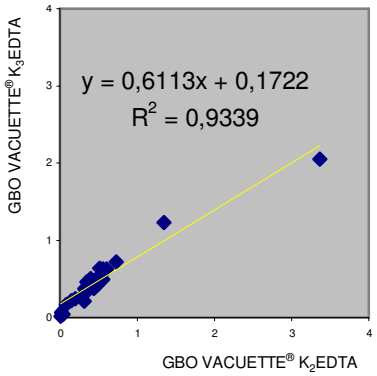
N=36
P-value= 0,946
Critical P= 0,05
No significance

Monocytes (MONO)

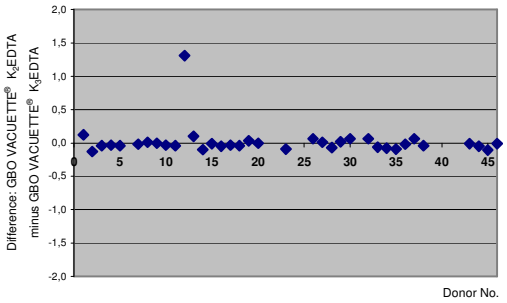
Normal range: 2 – 11 [10^3/µL]

Measurement 0-3h after blood collection:

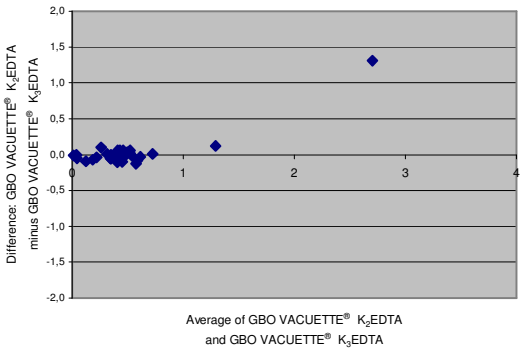
Regression MONO [10^3/µL]:



Deviation plot y-axis MONO [10^3/µL]:



Bland-Altman plot MONO [10^3/µL]:

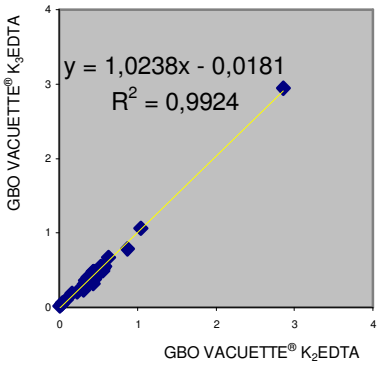


Result paired two tailed t-test at a confidence level of 95%:

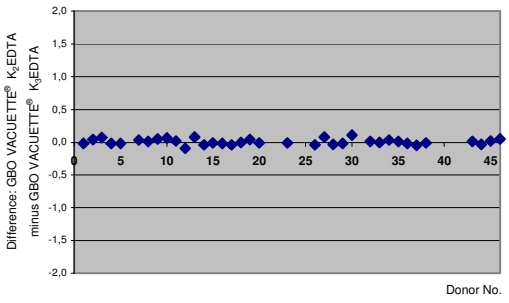
N=36
P-value= 0,639
Critical P= 0,05
No significance

Measurement 24h after blood collection:

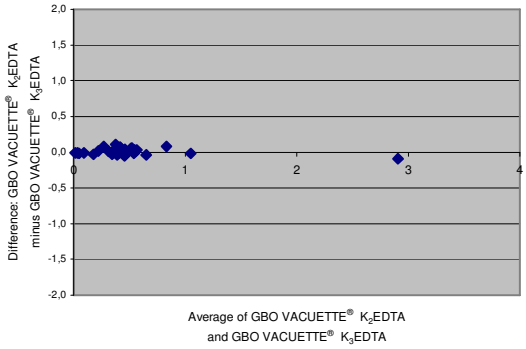
Regression MONO [10^3/µL]:



Deviation plot, y-axis MONO [10^3/µL]:



Bland-Altman plot MONO [10^3/µL]:



Result paired two tailed t-test at a confidence level of 95%:

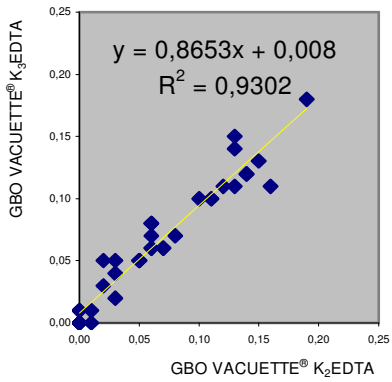
N=36
P-value= 0,352
Critical P= 0,05
No significance

Eosinophile Granulocytes

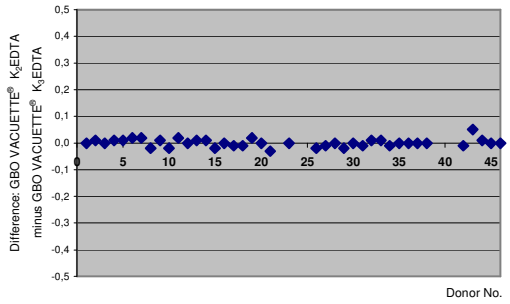
Normal range: 0 – 4 [10³/µL]

Measurement 0-3h after blood collection:

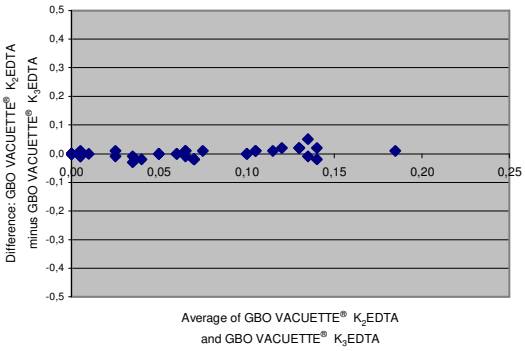
Regression EO [10^3/µL]:



Deviation plot y-axis EO [10^3/µL]:



Bland-Altman plot EO [10^3/µL]:

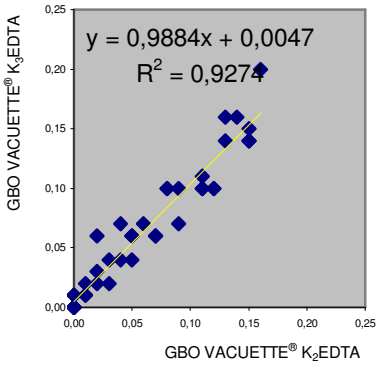


Result paired two tailed t-test at a confidence level of 95%:

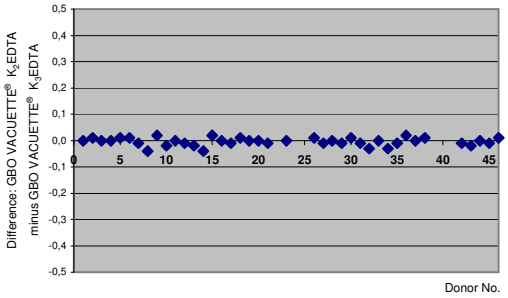
N=40
P-value= 0,749
Critical P= 0,05
No significance

Measurement 24h after blood collection:

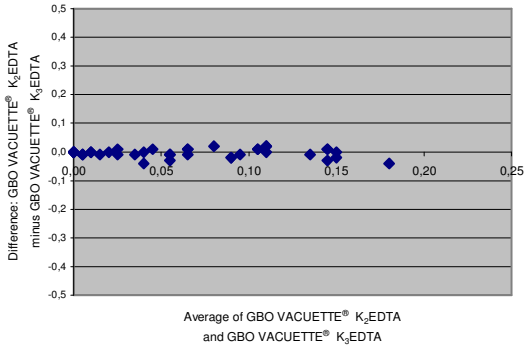
Regression EO [10^3/µL]:



Deviation plot, y-axis EO [10^3/µL]:



Bland-Altman plot EO [10^3/µL]:



Result paired two tailed t-test at a confidence level of 95%:

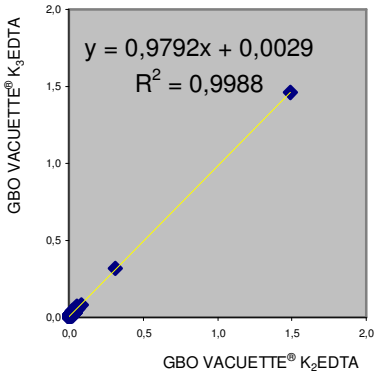
N=40
P-value= 0,096
Critical P= 0,05
No significance

Basophile Granulocytes (BASO)

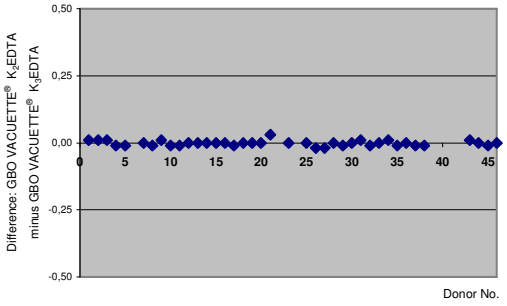
Normal range: 0 – 1 [10³/µL]

Measurement 0-3h after blood collection:

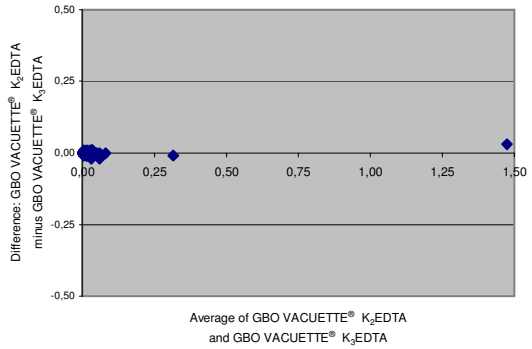
Regression BASO [10^3/µL]:



Deviation plot y-axis BASO [10^3/µL]:



Bland-Altman plot BASO [10^3/µL]:

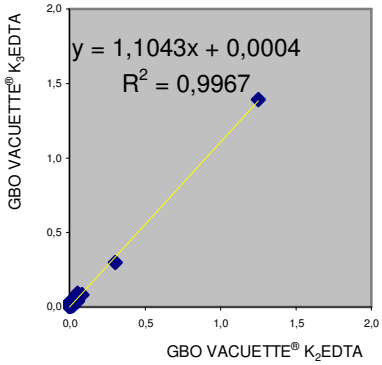


Result paired two tailed t-test at a confidence level of 95%:

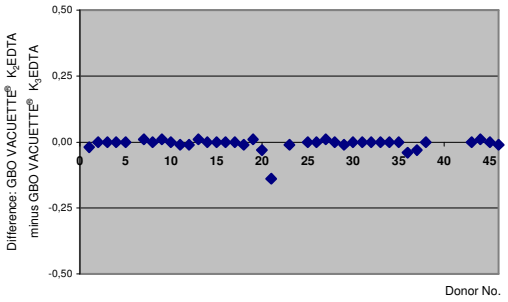
N=39
P-value= 0,324
Critical P= 0,05
No significance

Measurement 24h after blood collection:

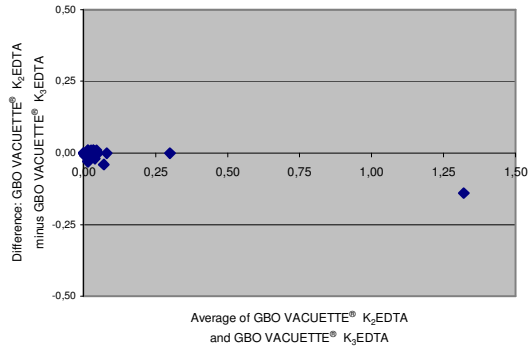
Regression BASO [10^3/µL]:



Deviation plot, y-axis BASO [10^3/µL]:



Bland-Altman plot BASO [10^3/µL]:



Result paired two tailed t-test at a confidence level of 95%:

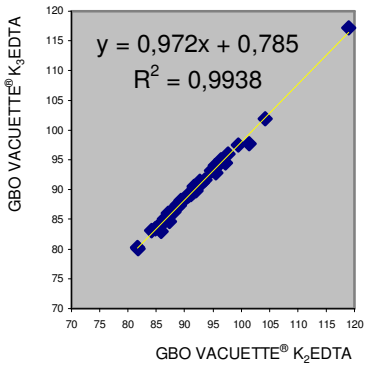
N=39
P-value= 0,098
Critical P= 0,05
No significance

Mean Corpuscular Volume (MCV)

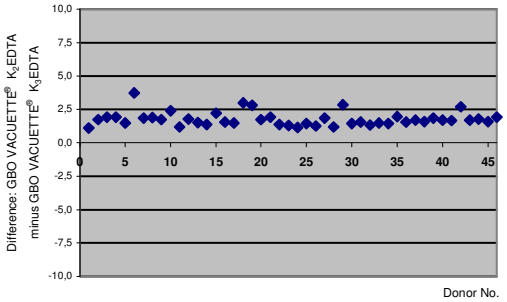
Normal range: 80- 99 [fL]

Measurement 0-3h after blood collection:

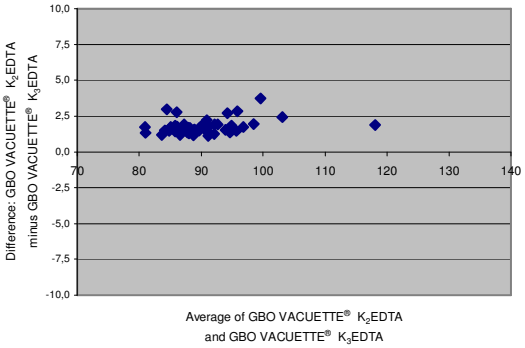
Regression MVC [fL]:



Deviation plot y-axis MCV [fL]:



Bland-Altman plot MCV [fL]:

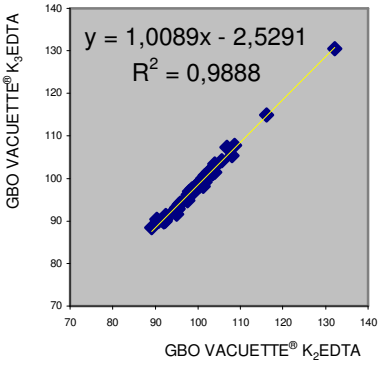


Result paired two tailed t-test at a confidence level of 95%:

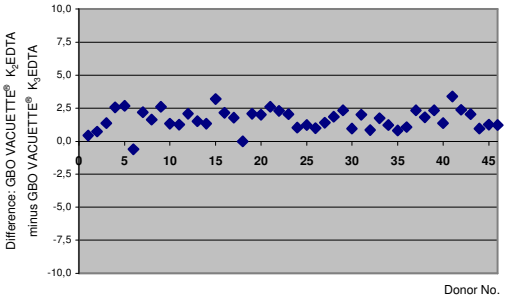
N=46
P-value= 0,000
Critical P= 0,05
Sigificant

Measurement 24h after blood collection:

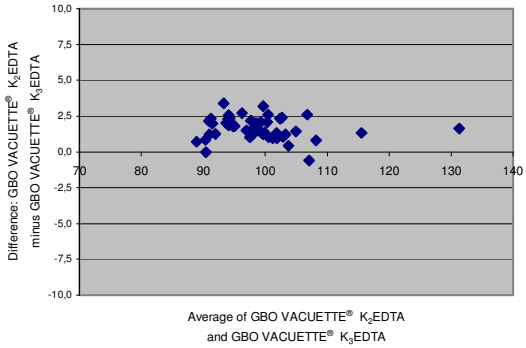
Regression MCV [fL]:



Deviation plot, y-axis MCV [fL]:



Bland-Altman plot MCV [fL]:



Result paired two tailed t-test at a confidence level of 95%:

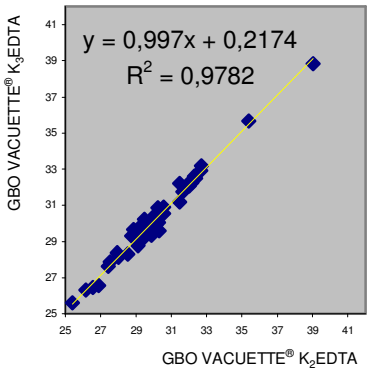
N=46
P-value= 0,000
Critical P= 0,05
Significant

Mean Corpuscular Haemoglobin (MCH)

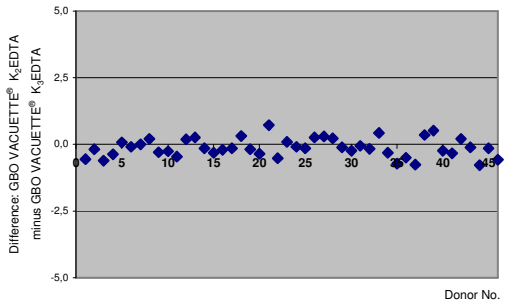
Normal range: 26 – 34 [pg]

Measurement 0-3h after blood collection:

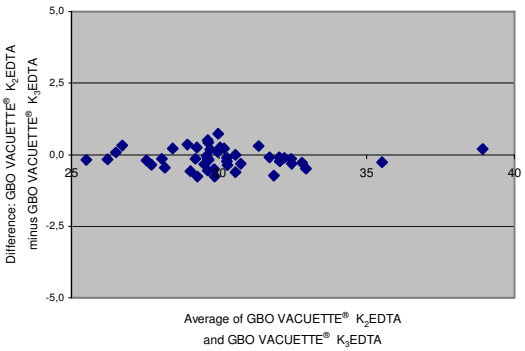
Regression MCH [pg]:



Deviation plot y-axis MCH [pg]:



Bland-Altman plot MCH [pg]:

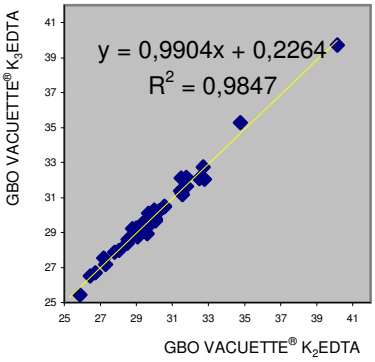


Result paired two tailed t-test at a confidence level of 95%:

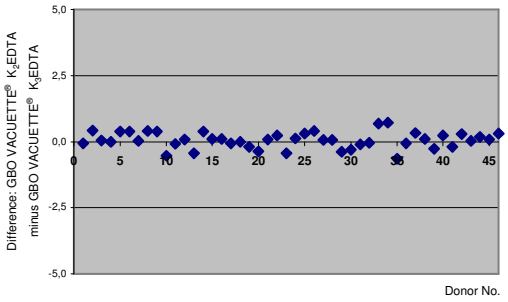
N=46
P-value= 0,017
Critical P= 0,05
Sigificant

Measurement 24h after blood collection:

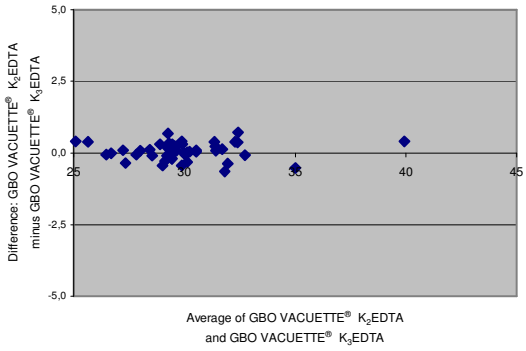
Regression MCH [pg]:



Deviation plot, y-axis MCH [pg]:



Bland-Altman plot MCH [pg]:



Result paired two tailed t-test at a confidence level of 95%:

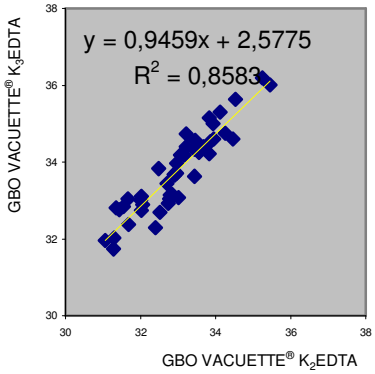
N=46
P-value= 0,186
Critical P= 0,05
No significance

Mean Corpuscular Haemoglobin Concentration (MCHC)

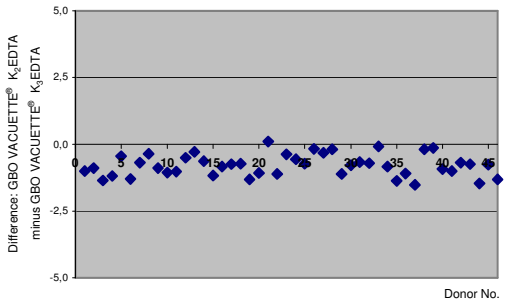
Normal range: 32 – 46 [g/dL]

Measurement 0-3h after blood collection:

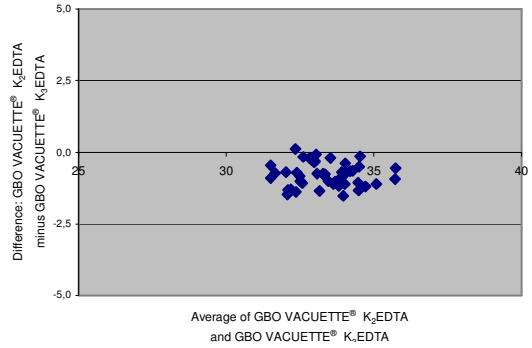
Regression MCHC [g/dL]:



Deviation plot y-ayis MCHC [g/dL]:



Bland-Altman plot MCHC [g/dL]:

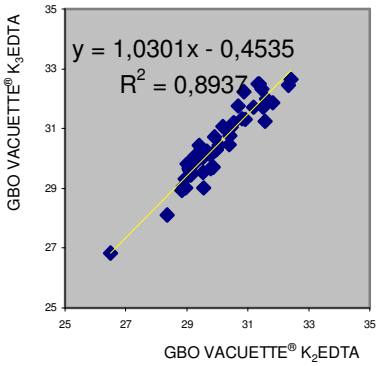


Result paired two tailed t-test at a confidence level of 95%:

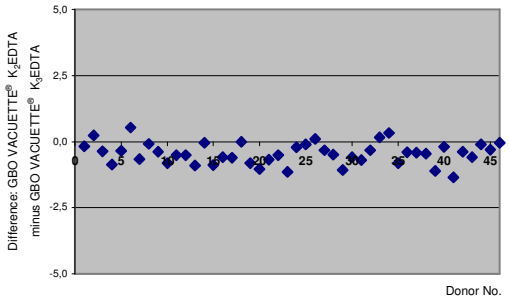
N=46
P-value= 0,000
Critical P= 0,05
Sigificant

Measurement 24h after blood collection:

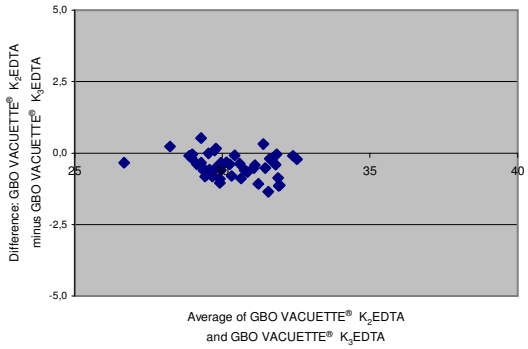
Regression MCHC [g/dL]:



Deviation plot, y-ayis MCHC [g/dL]:



Bland-Altman plot MCHC [g/dL]:



Result paired two tailed t-test at a confidence level of 95%:

N=46
P-value= 0,000
Critical P= 0,05
Significant