

Performance of the Anemia Panel Assays (Ferritin, Folate and Vitamin B₁₂) on the Abbott AxSYM[®] Instrument

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Introduction

The full panel of anemia assays, ferritin, folate and Vitamin B₁₂, is now available on the AxSYM[®] instrument worldwide. These assays are part of a broader evaluation used to diagnose anemia. The diagnostic evaluation may include a medical history, a physical exam and other blood tests such as a complete blood count (CBC). The objective of the studies described here was to verify the performance of each of the anemia panel assays in terms of precision, sensitivity, expected values and method comparison.

Background: Anemia

Anemia is a common blood disorder, defined as a decrease in hemoglobin or hematocrit below normal levels. The individual causes of anemia are numerous, but most can be grouped within three major mechanisms: excessive blood loss, inadequate production of red blood cells, and excessive destruction of red blood cells.

There are three primary forms of anemia:

• Iron Deficiency Anemia (IDA)

One of the most common causes of anemia, IDA results from low or depleted stores of iron, which is needed to produce red blood cells. The definitive test for IDA is the measurement of serum ferritin.

• Vitamin Deficiency Anemia

Deficiency of either Vitamin B₁₂ **or** folic acid (Folate) causes megaloblastic anemia (MA). In MA, the bone marrow produces red cells that are large and abnormal. Measurement of serum Vitamin B₁₂ and serum folate is part of the evaluation of MA.

• Pernicious Anemia (PA)

PA is the most common disorder resulting in Vitamin B₁₂ deficiency. Due to a lack of a substance called intrinsic factor, Vitamin B₁₂ can't be absorbed from the digestive tract.

Methods

AxSYM Ferritin: The AxSYM Ferritin assay is a one-step, microparticle enzyme immunoassay (MEIA) for the quantitative determination of ferritin in human serum or plasma. It has a dynamic range of 1 – 1000 ng/mL with automated dilution and throughput of 122 tests/hour.

AxSYM Folate: The AxSYM Folate assay uses ion capture technology in a two-step format for the quantitative determination of folate in human serum, plasma or red blood cells. It has a dynamic range of 0.9 – 20 ng/mL and throughput of 47 tests/hour.

AxSYM B₁₂: The AxSYM B₁₂ assay is a two-step MEIA for the quantitative determination of vitamin B₁₂ in human serum or plasma. It has a dynamic range of 60 – 1200 pg/mL and throughput of 55 tests/hour.

Representative testing results are shown in the next panels.

Results

Precision

Panel	FERRITIN		FOLATE		VITAMIN B ₁₂	
	Panel Concentration Range (ng/mL)	Percent CV Total (range)	Panel Concentration Range (ng/mL)	Percent CV Total (range)	Panel Concentration Range (pg/mL)	Percent CV Total (range)
Low	18.44 – 21.22	4.2 – 8.4	2.3 – 2.7	6.1 – 13.1	193.6 – 212.5	6.8 – 10.2
Mid	142.01 – 158.00	4.1 – 6.6	6.7 – 7.2	3.5 – 7.4	396.7 – 405.9	3.0 – 5.1
High	366.96 – 417.60	4.1 – 8.7	16.0 – 16.7	4.3 – 7.2	832.5 – 879.5	2.9 – 4.9
	1 reagent lot, 4 instruments (n = 320 per panel)		1 reagent lot, 4 instruments (n = 320 per panel)		2 reagent lots, 2 instruments (n = 320 per panel)	

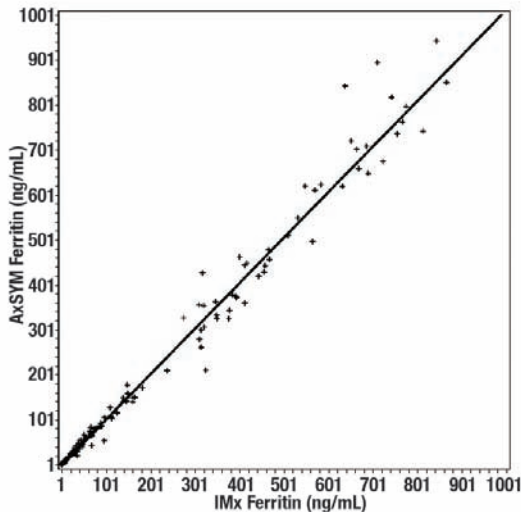
Precision studies were conducted using 2 reps of sample per run, 2 runs per day, over 20 days for a total of 80 sample reps per reagent lot and instrument combination. The number of reagent lots and instruments used in each study are indicated above.

Analytical Sensitivity

	FERRITIN	FOLATE	VITAMIN B ₁₂
Sensitivity	1.0 ng/mL	0.9 ng/mL	58 pg/mL
# Runs	61	36	27

Analytical sensitivity is defined as the concentration at 2 standard deviations above the zero calibrator and represents the lowest measurable concentration of analyte that can be distinguished from zero. The analytical sensitivity is calculated at the 95th percentile for all runs.

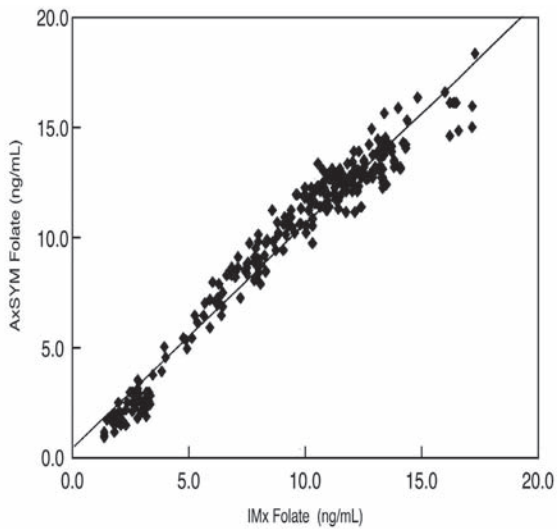
AxSYM Ferritin Correlation With IMx Ferritin Serum Specimens



n = 198
 correlation coefficient = 0.992
 slope = 1.01
 y-intercept = 0.36
 Passing-Bablok analysis (a linear regression method with no special assumptions regarding the distribution of the samples and the measurement of errors)

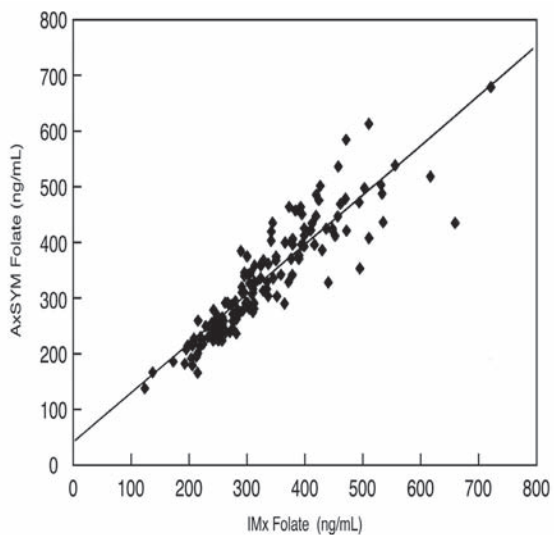
Results (cont.)

AxSYM Folate Correlation With IMx Folate Serum Specimens



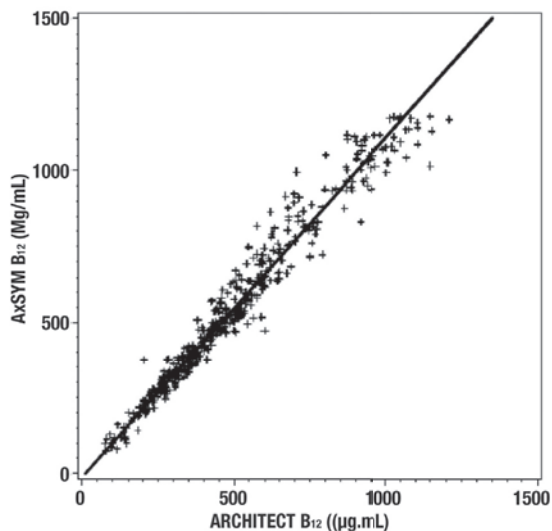
n = 261
correlation coefficient = 0.979
slope = 0.991
y-intercept = 0.5
Least squares analysis

AxSYM Folate Correlation With IMx Folate Whole Blood Specimens



n = 145
correlation coefficient = 0.906
slope = 0.885
y-intercept = 39.7
Least squares analysis

AxSYM B₁₂ Correlation With ARCHITECT B₁₂



n = 441
correlation coefficient = 0.98
slope = 1.12
y-intercept = -13.9
Passing-Bablok analysis
(a linear regression method with no special assumptions regarding the distribution of the samples and the measurement of errors)

Results (cont.)

Expected Values

AxSYM Ferritin		
	Number of Subjects	Range* (ng/mL)
Adult Males (18 – 30 years of age)	53	18.7 – 323.0
Adult Males (31 – 60 years of age)	46	16.4 – 293.9
Adult Females (Premenopausal)	97	6.9 – 282.5
Adult Females (Postmenopausal)	99	14.0 – 233.1

* Non-parametric estimate of the 95% confidence interval.

These individuals were determined to be normal based on test results for blood count, transaminases, gamma GT or erythrocyte sedimentation rate. Pregnant women were excluded from the premenopausal group. It is recommended that each laboratory establish its own normal range appropriate to the population it serves.

Ferritin levels below 10 ng/mL have been reported as indicative of iron deficiency anemia.

AxSYM B ₁₂				
Specimen Type	Number	Median Concentration (pg/mL)	Range (pg/mL)	Central 95% Range (pg/mL)
Normal Serum*	258	464.7	98.5 – >1200	208.0 – 963.5

* Serum specimens with normal MCV, homocysteine and folate results.

Further testing is required for symptomatic patients with the following serum B₁₂ levels:

Hematological Abnormalities: 100 – 300 pg/mL

Neurological Abnormalities: 100 – 400 pg/mL

AxSYM Folate: United States				
Specimen Type	Number	Median Concentration (ng/mL)	Range (ng/mL)	Central 95% Range (ng/mL)
Fasting Serum	124	13.0	5.6 – 19.8	7.2 – 15.4
Red Blood Cell	145	473.4	218.4 – 1010.8	252.6 – 813.7

The nutritional status of the donors was unknown; however, all specimens tested were from apparently healthy adults.

AxSYM Folate: Europe*			
Specimen Type	Number	Central 95% Range (ng/mL)	* Data from the Laboratoire de Biochimie Generale et Nutritionnelle, France, and General Army Hospital of Athens, Greece.
Serum	158	2.9 – 15.3	
Red Blood Cell	105	156.2 – 1150.6	

Folate deficiency is typically associated with serum levels less than 3 ng/mL or RBC values less than 150 ng/mL.

Conclusion

The AxSYM Anemia Panel assays provide sensitive and precise performance with confirmed accuracy. The AxSYM platform has proven reliability with a broad menu of available assays, allowing laboratory testing consolidation and significant workflow improvements.