Evaluation of the Sigma Quality level for Serum Iron determination by two colorimetric methods, Ferrozine and Ferene S

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Introduction

Iron plays important functions in the body such as the formation and functioning of hemoglobin and it's disorders are among the most common diseases of human[1]. It is essential to ensure that its levels determination through laboratory tests are accurate and precise. The participation of laboratories in the External Quality Assessment (EQA) allows the increases of the quality level of the laboratory results and improvement of its performance.[2]This study was developed in the Portuguese Nacional EQA Program (PNAEQ) concerning the laboratories results from the Clinical Chemistry Scheme.

Objective

The main objective of this study was to evaluate and improve the sigma quality level regarding the Iron parameter and reduce the variability of the laboratories results participating in the EQA program of Clinical Chemistry of the Nacional External Quality Assessment Program (PNAEQ).

Material and Methods

The present study uses data from EQA results of the Iron parameter, obtained from the analyzed serum samples control during 2018, from the 38 participant laboratories that used the Colorimetrics Ferrozine and Ferene S methods. The results from 12 control samples with different concentration levels were evaluated. The DMAIC cycle methodology (Define, Measure, Analyze, Improve, Control), Figure 1, was applied, integrating several techniques and quality tools. Six Sigma was used as a methodology and metrics to evaluate the performance of laboratories. The Six Sigma metric was calculated by the inaccuracy (bias) associated to the result obtained by each laboratory through the calculous of DPMO (Defects Per Million of Opportunities) and tables that convert DPMO to Sigma quality level (Formula 1 and 2). Outlier's treatment was applied, and the Normality was studied using the Kolmogorov-Smirnov test. When necessary, the Box-Cox transformation was practiced ensuring the data normality. The two-way ANOVA (Variance Analysis) with α = 0,05 was utilized to verify if the different colorimetric methods and the different control sample concentrations produced a significantly different bias. It was verified a need to identify the potential causes of the variability and inaccuracy in serum Iron determination by the use of the Pareto Diagram, which evaluates the questionnaire sent to the participants and the AHP (Analytic Hierarchy Process) method to hierarchize the identified potential causes. In order to verify the efficiency of the actions taken to improve the sample control reconstitution procedure, a Pilot Test (PT) was applied to 35 participant laboratories in the second Clinical Chemistry 2019 survey using two control samples with different concentration levels to reevaluate the Sigma quality level.

Figure 1 – DMAIC cycle

Formula 1

$$\text{DPMO} = P\left(Z \geq \frac{X_{Bias\ Desirable\ specification} - \hat{\mu}_{Bias}}{\hat{\sigma}_{Bias}}\right) \times 10^6$$

Formula 2

$$Bias_{\%} = \frac{|Laboratory\ result_i - Target\ Value|}{Target\ Value} \times 100$$

$P(Z \geq a) - Normal\ Distribuition\ Probability$

$\hat{\mu}_{Bias}$ $- mean\ of\ Bias$

$\hat{\sigma}_{Bias} - standard\ deviation\ of\ Bias$

$X_{Bias\ Desirable\ specification}$ $- Bias\ desirable\ specification\ value$

Results

Through ANOVA it was verified that the results of 38 laboratories from 12 control samples produced a significantly different bias (p-value =0.00000). The analyzed detection methods, Ferrozine (n=24 laboratories; mean bias=4.5%) and Ferene S (n=14 laboratories; mean bias = 5.8%) produced a significantly different bias (p-value =0.00048) (Table 1).

The mean Sigma quality levels were 3.2 (ranging 1.64 to 4.16) and 2.7 (ranging 1.52 to 4.46) for Ferrozine and Ferene S methods respectively, regarding the 12 control samples (A1 to A12). The mean Sigma quality levels for the 12 control samples was 2.9 (ranging from 1.82 to 4.06) (Table 2 and Figure 2).

Regarding the Pilot Test it was verified through ANOVA that the samples and the analyzed Colorimetrics methods did not produced a significantly different bias (p-value > 0.05) (Table 3). The Sigma quality level improved to 3.04 (Figure 2).

Table 1 – Two-away ANOVA for the 12 control samples and the 2 colometrics methods.

Source of Variation	SS	df	MS	F_0	p-value
Samples	0,19273	11	0,017521	12,286	**0,00000**
Methods	0,01769	1	0,017697	12,406	**0,00048**
Samples*Methods	0,03286	11	0,002988	2,095	0,01998
Error	0,52051	365	0,001426		
Total	0,76215	388			

Table 2 – Sigma quality levels for the 12 control samples and the 2 colometrics methods.

Methods	Samples												Mean Sigma Quality Level
	A1	A2	A3	A4	A5	A6	A7	A8	A9	A10	A11	A12	
Ferene S	1,64	2,11	2,47	3,76	2,07	3,01	3,66	4,16	2,18	2,03	3,09	2,19	**2,70**
Ferrozine	2,56	1,52	4,31	4,05	3,10	4,29	4,46	3,59	2,95	1,85	3,53	1,85	**3,17**
Mean Sigma Quality Level	2,10	1,82	3,39	3,91	2,58	3,65	4,06	3,87	2,57	1,94	3,31	2,02	2,94

Table 3 – Two-away ANOVA for the 2 control samples (PT) and the 2 colometrics methods,

Source of Variation	SS	df	MS	F_0	p-value
Samples	0,00282	1	0,00282	1,653	**0,20301**
Methods	0,00085	1	0,00085	0,503	**0,48077**
Samples*Methods	0,00252	1	0,00252	1,480	0,22803
Error	0,11274	66	0,00170		
Total	0,11806	69			

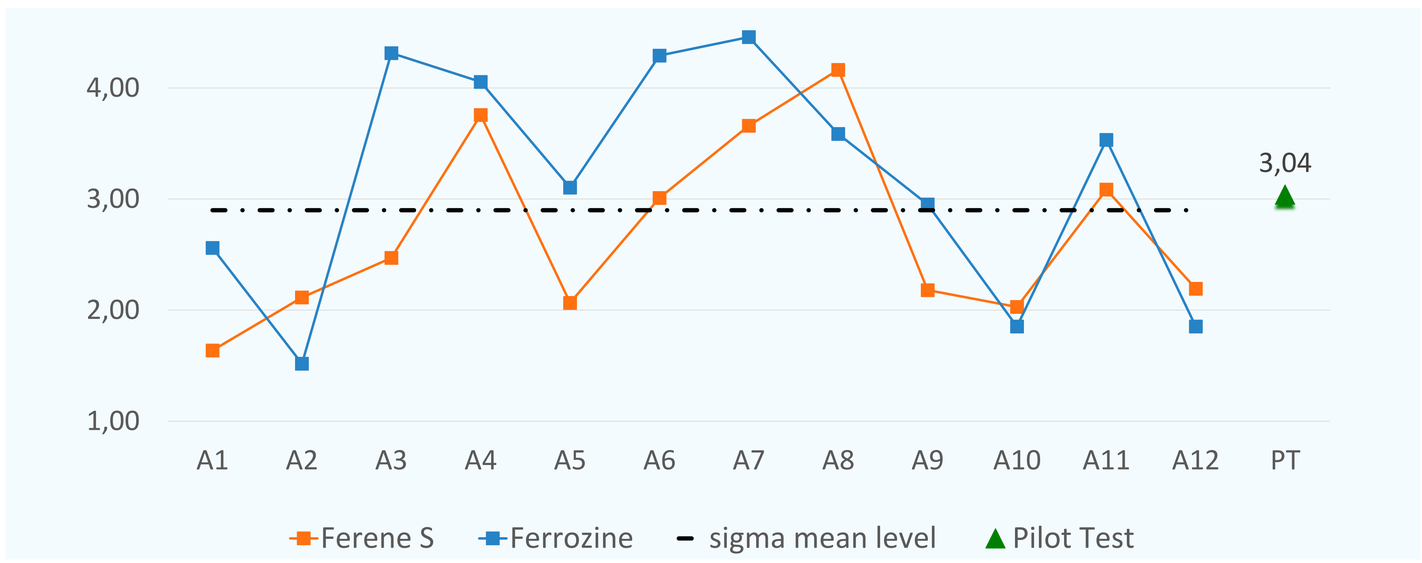

Figure 2 – Graphic of the sigma quality levels of the 12 control samples and of the Pilot Test for the 2 colometrics methodos.

Conclusion

The mean Sigma quality level indicated that the Ferrozine method had a better performance compared with Ferene S method. Half of the control samples had a sigma quality level higher than 3.0, which is set as the minimum acceptable quality.[3] Despite of the improved of the Sigma quality level in the Pilot Test, the results demonstrated a need to improved the analytical process performance and to identified more potential causes and implement new improvement actions. It becomes necessary to raise awareness with the laboratories, improving the Pilot Test participation frequency, resulting in a recurrent and current assessment of the laboratory activity performance. Developing Six Sigma projects on a periodic basis is important for continuously and progressively increasing the level of Sigma quality in laboratory examinations. The main advantage of quality assessment on the sigma scale is providing evidence of overall laboratory performance, taking into account random and systematic errors.

References:

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