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The evaluation of analytical performance of Total PSA and Free PSA tests by using 6-sigma method

Total PSA ve serbest PSA testlerinin analitik performansının 6-sigma yöntemi ile değerlendirilmesi

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ABSTRACT

Objective: Sigma Metrics Methodology is a quality measurement method in order to evaluate and compare the analytical performance of laboratories. Six Sigma can be used as a clinical analytical phase assessment methodology to form an internal quality control (IQC) strategy and plan its frequency. In this study, we aimed to evaluate the analytical performance of the tumor markers total and free prostate specific antigen (PSA) by calculating process sigma values.

Methods: Sigma levels for both tests were analyzed by using IQC values retrieved from laboratory information system for consecutive 3 months. Bias and coefficient of variation (CV) were calculated. Biological variation databases were used for Total Allowable Error (TEa). The calculated sigma values were classified as follows: ">5", "4-5", "3-4" and " <3" as very good, good, minimum and unacceptable respectively.

Results: The sigma values of total PSA and free PSA tests according to the 3-month data of two IQC levels were found to > 5 and 4-5 for total PSA,while those for free PSA were <3 for both levels.

Conclusion: Our results showed that performance of total PSA is good while performance of free PSA is unacceptable. It is possible to determine a test with

ÖZET

Amaç: Sigma metrik yöntem, laboratuvarların analitik performansını değerlendirmede ve karşılaştırmada kullanılan bir kalite ölçüm yöntemidir. 6 sigma yöntemi ile klinik analitik faz değerlendirilebilir ve iç kalite kontrol (İKK) stratejisi ve sıklığı planlanabilir. Bu çalışmada tümör belirteçleri olan total PSA ve serbest PSA testlerinin sigma değerlerinin hesaplanarak analitik performanslarının değerlendirilmesi amaçlandı.

Yöntem: Her iki test için de ardışık 3 aylık iç kalite kontrol sonuçları değerlendirilerek sigma seviyeleri hesaplandı. Bias ve varyasyon katsayısı (CV) hesaplandı. Toplam izin verilebilir hata (TEa) değeri için biyolojik varyasyon veritabanları kullanıldı. Hesaplanan sigma değerleri sırasıyla şöyle değerlendirildi: ">5", "4-5", "3-4" ve "<3" çok iyi, iyi, minimum ve kabul edilemez.

Bulgular: 3-aylık iki seviye iç kalite ile değerlendirilen sigma değerleri total PSA için >5 ve 4-5, serbest PSA için ise her iki seviyede <3 olarak bulunmuştur.

Sonuç: Bulgularımız, total PSA analitik performansının iyi, serbest PSA'nın ise kabul edilemez derecede olduğunu göstermiştir. Yüksek hata oranına sahip bir testi sigma değerlendirmesi

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high error probability by evaluating the fine sigma levels and the tests that must be quarded by more stringent quality control applications. Our study has shown that we need to apply a more stringent IQC regime for free PSA test.

Key Words: Total PSA, free PSA, 6 sigma

ile saptamak mümkündür ve bu test daha sıkı kalite kontrol uygulamaları ile kontrol altında tutulabilir. Bizim çalışmamız, kendi laboratuvarımız için serbest PSA testine daha katı bir İKK protokolü uygulamamız gerektiğini ortaya koymuştur.

Anahtar Kelimeler: Total PSA, serbest PSA, 6 sigma

INTRODUCTION

Clinical laboratory processes are simply divided into three major steps: preanalytical, analytical and postanalytical. Laboratory errors in each of these phases affect test results and therefore the aim is to detect and minimize the sources of error at every step (1,2). When the sources of laboratory errors are evaluated, it is found that most of the errors occur in the pre-analytical phase followed by post-analytical phase. Analytical phase errors seem to be the least source of error in clinical laboratories (3). Although the impact of analytical phase errors on total laboratory errors is low, it is important to manage laboratory processes correct to improve the quality of health service provided to patients. Standardization of the analytical phase, increased evaluation of internal and external quality control programs and technological improvements have increased the reliability of laboratory results to a great extent (4).

Two types of analytical errors are random errors and systematic errors which are expressed as inaccuracy and imprecision respectively (5). The expression of inaccuracy is bias and the expression of imprecision is coefficient of variation (CV), which are used in combination to detect total error (TE) as Bias + 1.65 CV (6).

Allowable total error (TEa) is a simple comparative quality concept which is used to define acceptable analytical performance and can be used for (1) assessment of an individual instrument's analytical performance, which is of benefit if one uses this information during instrument selection or assessment of in-clinic instrument performance, (2) Quality Control validation, and (3) as a measure of agreement or comparability of results from different laboratories (eg, between the in-clinic analyzer and the reference laboratory) (7).

The six-sigma technology, which was first used in evaluation of errors in the industrial field, has been widely used recently also in evaluation of laboratory errors. Six-sigma metrics combines bias, imprecision and TEa. Being a statistical method, six-sigma methodology includes 5 steps known as define, measure, analyze, improve, control (DMAIC). The sigma model provides an objective evaluation of the performance of a method and therefore for laboratories it is valuable measure for self-control in the laboratories. The sigma value of a test enables to determine targets for improving the quality of the test in laboratory, or to accept the current quality of the test if the quality is adequate (8,9).

Sigma Metric is calculated by using the formula of sigma (s)=(TEa-bias)/CV (5). High sigma values means low analytical errors and acceptable test results (6). Low sigma metric value is accepted as an error or a defect. The defect value is measured in defects per million (DPM). The Six-Sigma is focused to control a process in 6 standard deviations (SD) and it is equal

to 3.4 DPM. The success with Six Sigma Quality is accepted as the perfection standard. A performance at the 3-sigma level is considered as the minimum quality for manufacturing process (2, 5).

Total PSA and free PSA are the two most commonly used diagnostic tools in clinical practice for prostate cancer screening accepted worldwide. The analytical phase in total and free PSA measurement is of critical importance as their ratio gives discriminative diagnosis between prostate cancer and benign prostate hyperplasia (10,11).

In the present study, based on these facts, we aimed to evaluate the analytical phase by using sigmametrics and also to reveal the quality control (QC) strategy of these two critical tests.

MATERIAL and METHOD

The present study was conducted in the Clinical Biochemistry Laboratory of University of Health Sciences Gülhane Training and Research Hospital. Internal quality control (IQC) data of the two analytes were analyzed retrospectively over a period of 3 months from August 2019 to October 2019 using DxI 800 analyzer (Beckman Coulter Inc, USA). Both tests were immunoassay methods, all reagents were obtained from Beckman Coulter Inc and used according to the manufacturer's directions.

Two level controls; normal (Seronorm-1), and pathological (Seronorm-2,) levels of QC materials were assayed before analysing of patient samples every day during the study period. The lot numbers of the QC material used were the same: 1804832 for normal and 1805833 for pathological controls respectively. The instruments were calibrated on a regular basis. IQC data were obtained from Laboratory Information System (FONET LIS). Faulty values arising from false control samples were excluded.

The values given in the insert provided by Beckman Coulter Inc. for IQC target values were used as reference values. % Bias values were calculated for each test for every month during the 3 month study period. TEa, is determined by biological variation and the performance of the analytical method. TEa values given in Westgard biological varition database were used to calculate the sigmametric performance characteristics (7).

Following determination of mean and SD values, %CV, bias and sigma values were calculated according to the following formulations. Coefficient of variation (CV%):

Coefficient of variation (% CV) is the expression of imprecision. It is the percent ratio of standard deviation (SD) to the mean ($\bar{x}$) for a given data set. It was determined from the calculated mean and Standard deviation evaluated from IQC data.

CV(%)=(SD/ Mean of IQC data) × 100.

Bias:

Bias was calculated as the percentage difference of the average of observed results for each analyte from the target values provided in the Beckman Coulter's control material inserts. %Bias values of each test were calculated by getting the mean values between August, September and October 2019.

%Bias= [(IQC data mean of our laboratory-target mean of IQC data)/target mean of IQC data]×100

Allowable total error (TEa):

Detected for total PSA and free PSA using the Desirable Biological Variation Database and The Royal College of Pathologists of Australasia and the Australasian Clinical Biochemist association Quality Assurance Program (RCPA) respectively. These sources are regularly updated and can be freely accessed through http://www.westgard.com and https://www.westgard.com/rcpa-biochemistry.htm.

Sigma (σ) value:

Sigma value was calculated by using CV (obtained from IQC data), %Bias (obtained from target values of IQC data) and TEa values. Sigma value calculated

using the standard equation;

σ metric= (%TEa-%Bias) / %CV

Sigma values were calculated to determine the analytical performance characteristics of each analyte. Sigma values ">5", "4-5", "3- 4" ve "<3" were categorized as " very good", "good", "minimum" and "unacceptable" respectively (8).

RESULTS

For each level of free PSA and total PSA tests, the target mean given by manufacturer, labaratory mean and SD values are presented in Table 1.

TEa% for total PSA is taken both from Westgard and RCPA guidelines, while for free PSA, it is only taken from RCPA guideline (12,13). Quality control strategy used to evaluate the paremeters is explained in Table 2.

Table 1. Sigma values for tests according to two different TEa % values obtained from internal quality control results

Assay	Mean		SD		Target mean		CV%		Bias%		TE_a%		Sigma			
	QC1	QC2	QC1	QC2	QC1	QC2	QC1	QC2	QC1	QC2	BV Desirable*	RCPA**	BV Desirable		RCPA	
													QC1	QC2	QC1	QC2
Total PSA	3.53	24.16	0.16	1.34	3.79	25.80	4.58	5.57	6.68	6.34	33.6	15	5.87	4.89	1.81	1.55
Free PSA	1.68	12.97	0.08	0.82	1.80	12.80	5.24	6.37	6.29	1.32	-	15	-	-	1.66	2.14

*Desirable Quality Specifications based on Biological Variation.

**The Royal College of Pathologists of Australasia and the Australasian Clinical Biochemist association Quality Assurance Program

QC: Quality control, SD: Standard deviation

Table 2. Quality control strategy

Sigma metrics	Quality performance	Westgard Rule
<3	Unacceptable	1_{3S} / 2_{2S} / R_{4S} / 4_{1S}
3- <4	Minimum	1_{3S} / 2_{2S} / R_{4S} / 4_{1S}
4-5	Good	$1_{2.5S}$
>5	Very good	1_{3S}

DISCUSSION

PSA is known as the leading tumor marker in evaluation of effectiveness of therapy for prostate cancer patients, assessment of tumor mass and early detection of recurrence. It is also different from other tumor markers as it is very useful in the screening and early diagnosis of prostate cancer. PSA is found free or protein-bound in circulation. Total PSA test has low specificity, especially in patients with total PSA concentration within the "diagnostic gray zone" (total PSA concentration range of 4-10 ng/mL). Free to total PSA ratio (PSA free/PSA total) is the most commonly used diagnostic index for distinguishing between benign prostate hypertrophy and prostate cancer. With the improving immunoassay techniques, it has been proven that the %fPSA is lower in men with prostate cancer. If the free/total ratio is below 8%, the risk of prostate cancer is predicted to be around 80% (14,15). That's why accurate laboratory measurement of these analytes has critical importance.

The practice of using Sigma metrics to improve and design high quality products has been around for several decades. 3-sigma assay is being generally considered as the minimum acceptable performance and a 6-sigma assay performance considered world-class (8). One problem encountered in calculation of sigma values is the differences caused by differences in TEa. It is important to determine which TEa to use as TEa values for many measurands differ greatly, depending on the source. While there is a recommended hierarchy to consider when choosing an appropriate TEa, there is no uniform consensus on which source is most appropriate for a given measurand (16).

For total PSA, sigma values calculated according to Westgard BV guidelines, sigma levels were very good for level 1 and good for level 2. But when the values are evaluated according to RCPA rules, both 1 and level 2 were in the unacceptable range for PSA. These differences point that sigma levels can change depending on the reference we take for TEa values. Since there was no biological variation data in Westgard's site for free PSA, we evaluated fPSA level 1 and level 2 performences solely depending on RCPA values. The results came out as both levels are in the unacceptable range for fPSA.

RCPA gives TEa for both total and fPSA tests as 15%. While Westgard's BV rules give these value as 33.6 % for total PSA and there is no value assigned for fPSA in Westgard's site. These differences in allowable total error values also affect the sigma value calculated for these tests.

In the present study, we evaluated the analytical performances of total and fPSA tests in our laboratory with sigma metric approach. There are very limited studies in the literature evaluating immunoassay tests with sigmametrics. This information can also assist the laboratory in knowing the kind of performance to expect. Another point to consider when using Sigma metrics is that bias and precision influence the Sigma metrics differently, with precision having a greater impact.

Our study is unique as it evaluates the performance of total PSA based on two different BV sources being Westgard and RCPA. Also for sigma metric evaluation of fPSA, this will be the first study.

In conclusion, while defining QC strategies, clinical laboratories should calculate their sigma values according to most recently updated TEa sources and choose their QC rule strategies accordingly. According to our data, we can conclude that we should follow 13S rule for total PSA and 13S / 22S / R4S / 41S rule for fPSA test. It can also be concluded that fPSA test needs a more strict IQC regimen to minimize analytical errors.

ETHICS COMITTEE APPROVAL

* This study does not require Ethics Committee Approval.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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