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UNCERTAINTY OF MEASUREMENT IN DRUGS QUALITY CONTROL

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Abstract:

For a product proves effective should be performed quality control tests to certify their suitability for purpose; some of these tests are: determining the assay and uniformity content. Besides, obteined data must be evaluated by the measurement uncertainty to the final result to be considered reliable. One way is to calculate this uncertainty by the Gage Repeatability & Reproductibility (R&R) study, which aims to evaluate the dispersion of the data by means of the accuracy of a method. The main purpose of this study was to investigate the total variation of a measurement system.

Keyswords: Uncertainty of measure, gage R&R study, content uniformity, assay, drug quality control.

Introduction:

The quality control of drugs aims to control the production process from raw materials to final product, which is performed by tests that together assess if their use is appropriate. Thus, the need arises strict criteria to ensure that the quality of measurements in order to verify the homogeneity of each lot, as well, given the patterns of activity, purity and efficacy. Some of these criteria are based on the uncertainty of the measurement results.
As the Guide to the expression of Uncertainty in Measurement - GUM, uncertainty defines the term: "It is a parameter associated with the result of a measurement that characterizes the dispersion of values that could reasonably be attributed to the analyst." Measurement uncertainty comprises many components. Some of these components may be evaluated from the statistical distribution of the results of series of measurements and can be characterized by standard deviations [1]. According Leito et al [2], calculate the uncertainty is one of the most important characteristics of quality and has become a standard requirement in chemical analysis to determine the accuracy of the result.
In estimating the overall uncertainty, it may be necessary to inform each source of uncertainty and treat it separately for the contribution of this source. Each of the separate contributions uncertainty is referred to as an uncertainty component. When the standard deviation expressed as an

uncertainty component is known as a standard uncertainty. If there is a correlation between the components, then it must be taken into account by determining the covariance. However, it is possible to evaluate the combined effect of several components. This application can alleviate all involved in mainly the evaluated components whose contribution is correlated together, there may be no further need to have this correlation [3].

The Gage R & R study is an analysis technique that uses a random-effects model analysis of variance to assess the suitability of a measurement system. Every action taken has some error associated with it, and if this error is large compared with the range of values, also called tolerance range, the meter can often accept the bad parts and reject the good [4].

There are two important aspects evaluated in the study Gage R & R: repeatability and reproducibility. Repeatability is the variation in measurements taken by a single analyst or instrument, in the same replicate, under the same conditions. Intermediate precision is induced when different operators or instruments measure the same repetition rate. Therefore, the study Gage R & R discusses only the accuracy of a measuring system.

It is common to examine the precision / tolerance (P/T) ratio which is the ratio between the precision of a measurement system on the tolerance of the overall manufacturing process. If the "P/T" ratio is low, the impact on product quality variation due to the measurement system is small. If the "P/T" ratio is high, it means that the measurement system increases a large fraction of tolerance, and that the components involved are not enough to declare as acceptable by the measurement system [5] tolerance.

The objective of this study is to investigate the total variation of the measurement system where the components are related to the precision of a test.

MATERIALS AND METHODS

Materials

Two high performance liquid chromatographs (Agilent Technologies 1200 series - equipped with a quaternary pump, an auto-sampler and UV detector) and two analytical columns C8 (75 x 4,6 mm ; 3,5 μm) were employed. An analytical balance (Mettler Toledo, AG204) was used to measure the weight of reference standard and the sample. Volumetric flasks (10mL, 25 mL, 50 mL, 100 mL e 200 mL) and pipettes (1, 2, 3, 5 e 8mL) were used in the preparation of standard and samples.

Methods

Sample and Standard solutions

The reference standards were diluted in ultra-pure water in order to obtain a concentration of 0.10 mg/mL. For assay test, the sample solutions were prepared from the pool of the five capsules; dilutions were made to obtain solutions with concentrations at 0.1 mg/mL. For the uniformity content, the samples solutions were prepared similarly as specified above, however, was used the content of one capsule for the preparation of each solution.

The mobile phase was made by the mixture of methanol and buffer phosphate, in the ratio of 60:40 (v/v), respectively. The solvent used in all dilution was the mobile phase.

The chromatographic conditions were oven temperature at 45ºC; 1.5 mL / min flow; detection at 230 nm and injection volume 10 μL.

Experimental Design

For each tests – assay and content uniformity – were perfomed the precision and precision intermediate according to the current official codes - ICH[6], USP 36[7] and FB 5th ed[8]. The results from a trial were evaluated to determine if they were suitable purpose of the test. In the intention to evaluate a possible influence by analysts in the preparation, was proposed an experimental design for two analyst in each different chromatographic systems (CS) and on different days of analysis, as shown in the flowchart below (Figure 1):

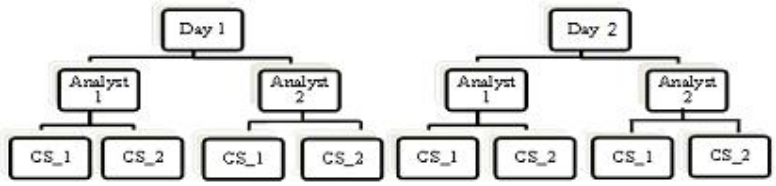


Figure 1. Flowchart of experimental design / Chormatographic System (CS)

2. 3 Calculation

The set of results obtained was applied the Gage R&R study which uses the analysis of variance to demostrate the random effects of a measurement system.

RESULTS AND DISCUSSION

An important aspect of a statistical control is to ensure adequate capability of the measuring system. In any process involving measurements part of variability is due to variability of the product itself and the other part come from measurement error or variability of the measurement system.

Capability of measurement system may be evaluated using "P/T" ratio, which is calculated by the 6σ of measurement system divided by the tolerance range (Upper Specification Limit - Lower Specification Limit), as shown below:

$$P / T = (6 \times \sigma) / (UCL - LCL) \quad (1)$$

The lower the "P/T" ratio the better is the capability of the measuring system. A P/T ratio of 10% is recommended, although "P/T" of 30% is considered enough for its application.

According to official codes, the tolerance range of an analysis of content uniformity is in the range of 85 to 115% of the labeled value. Therefore "P/T" ratio for the content uniformity will be equal to: (6x 0,856) / (115-85) = 0.17 or 17%. And the tolerance range for the analysis of assay is in the range 90-110 % of the labeled value. Thus, the "P/T" ratio for assay is equal to: (6x 1,098) / (110-90) = 0.32 or 32%. A better "P/T ratio may be obtained increasing the number of determination of the tests. For example, using 3 determinations in the test of assay, the "P/T" ratio will be 18%.

In order to assess if the results of two analysts are within the standards established by the laboratory an test by content uniformity of a drug was performed to record the measurement precision of both. Ten solutions were prepared by analysts representing each batch of the product to be tested. With the results of this test calculations that estimated the percentage change of the components of the measurement system (Table 1) were performed.

The calculated values can also be observed in Graphic 1 that greater variability is related to the Part Number components, pointing approximately 88.5% of the total variation.

The other part is related to the total measurement system (total Gage R & R). This portion showed 11.5% of the total variation, where the repeatability contributed a percentage of 7.58% against 3.87% regarding intermediate precision. Intermediate precision is composed of individual items evaluated in the trial: analyst chromatographic system (HPLC / colunm) and the day of analysis, which would be the environmental conditions. Analysts represent 2.98% of this variation, the day of analysis is 0.38% and 0.51% chromatographic system.

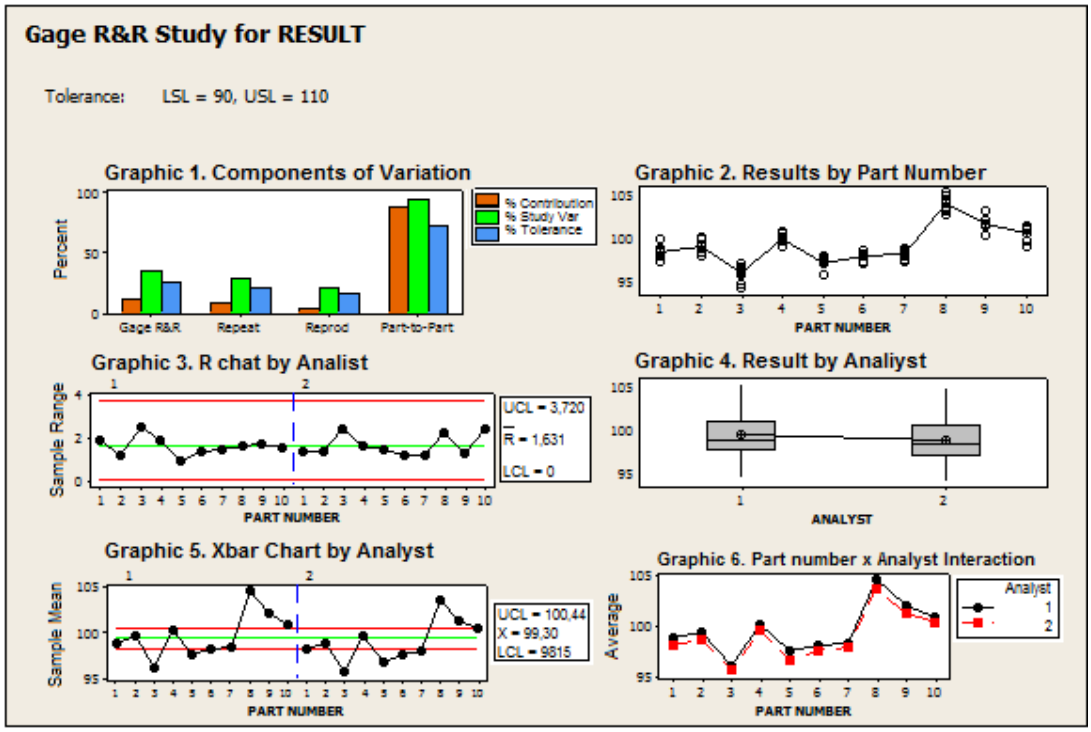
In Graphic 2 it can be seen that the variation between the results obtained vary for each part number is coming from a lot different from the product analyzed. In Graphic 3, its shows the results little scatter in the interval between the highest and lowest value found, indicating that analysts have consistency in their results. With a tolerance range between 90% and 110%, the average was 99%. At result by analyst (Graphic 4), the variation among analysts have measures approximately the same variation. The Graphic 5 is where the Gage R & R compares the part of the variation of the components of repeatability. A large dispersion shows that over 50% of the results is outside the tolerance range. Clearly there is little difference between the average values between each result analyst in Graphic 6. Lines vary, but it is clear that the analyst 2 got the lowest averages.

Table 1 Variance of Components of the Uniformity of Content.

Source	Components of Variation	% Contribuition of Components
Total Gage R&R	0,73355	11,45
Repeatability	0,48543	7,58
Reprodutibility	0,24811	3,87
Analyst	0,19123	2,98
Day	0,02425	0,38
Chromatographic System	0,03264	0,51
Part-to-part	5,67430	88,55
Part Number	5,67430	88,55
Total Variation	6,40785	100,00

Source: Minitab 16

Figure 2 Graphics of Gage R&R for Uniformity Content analisys. Source: Minitab 16



Otherwise, in the results obtained from the test of assay, was used the same calculations to estimate the percentage variation of the components (Table 2). It is showed in Graphic 7, that upper variability is related to the part number components, indicating about 74.2% of the total variation. The other part is related to the total measurement system (total gage R & R).

This portion present 25.78% of the total variation, where the repeatability contributed a percentage of 10.16% against 15.62% regarding intermediate precision. Intermediate precision is composed of individual items evaluated in the trial: analyst chromatographic system (HPLC / colunm) and the day of analysis, which would be the environmental conditions. Analysts represent 0.76% of this variation, the day of analysis is 14.57% and 0.28% chromatographic system.

In Graphic 8, it can be seen that the variation between the results obtained vary for each part number is stemming from a different batch of the product analyzed. As noted in Graphic 9, the results little scatter in the interval between the highest and lowest value found, indicating that analysts have consistency in their results. With a tolerance range between 90% and 110%, the average was 100.2%.

Table 2 Variance of Components of the Assay

Source	Components of Variation	% Contribuition of Components
Total Gage R&R	1,20598	25,78
Repeatability	0,47532	10,16
Reprodutibility	0,73067	15,62
Analyst	0,03561	0,76
Day	0,68181	14,57
Chromatographic System	0,01324	0,28
Part-to-part	3,47255	74,22
Part Number	3,47255	74,22
Total Variation	4,67853	100,00

Source: Minitab 16

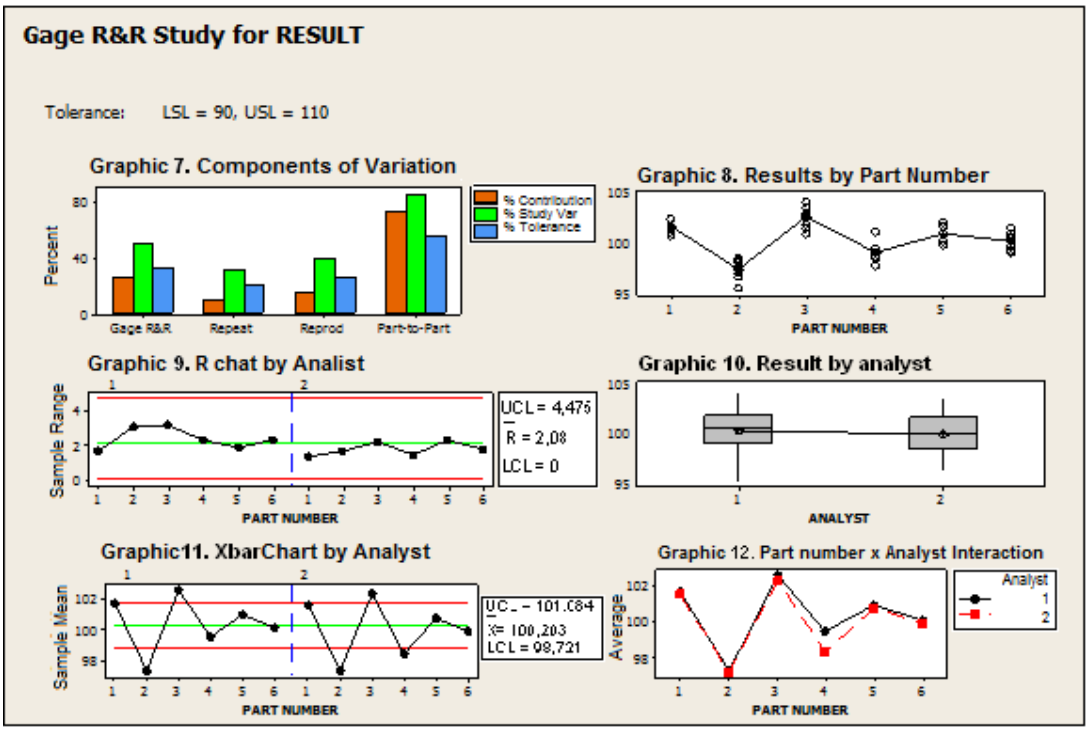


Figure 3 Graphics of Gage R&R for Assay analisys. Source: Minitab 16

In Graphic 10 (Result by Analyst), the variation between measurements analysts have approximately the same variance. The Graphic 11 is where the gage R & R compares the part of the variation of the components of repeatability. A large dispersion shows that over 50% of the results is outside the tolerance range.

There is a little diference between the medium obtained on each analyst results, as shown at Graphic 12, but it is clear that the analyst 2 has obtained the most low average like is possible observed by the variation of the lines of graphic.

Hence it can be concluded that, to the most analysts which have good training on preparing samples and conducting the test safely, also an calibrated equipment, it was possible to say with 99% confidence that the deviations found in the test for content uniformity and assay are stemming from the manufacturing process of the drug.

Aknowledgement

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