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Validation of On-line Total Organic Carbon Analysers for Release Testing Using ICH Q2

Introduction

As the pharmaceutical industry seeks to reduce the number and associated costs of quality control laboratory tests, reliance on results from on-line TOC analysers for product release is becoming more attractive. The International Conference on Harmonisation (ICH) is an expert working group with representation from the United States[2], European[3], and Japanese[4] Pharmacopoeias. In their harmonised tripartite guideline ICH Q2[1], Validation Of Analytical Procedures, they outline characteristics for consideration during the validation of analytical procedures included as part of registration applications submitted within the EC, Japan and USA. This paper discusses how these characteristics may be applied to on-line Total Organic Carbon (TOC) analysers to enable them to be used to provide release test data for Water for Injection (WFI) and Purified Water (PW).

TOC analysers overview

TOC analysers measure one of the four key critical quality attributes mandated by the pharmacopoeias. TOC analysis is a non-specific test, i.e. it is simply a measure of the carbon found in any organic compound in the water, it cannot tell you what type of organic molecule is present. A pharmaceutical-grade TOC analyser uses ultra-violet light (UV) to oxidise the organic molecules to release the carbon atoms present and then measures the difference in water conductivity caused by the resultant carbon dioxide.

TOC is to be calculated by measuring Total Inorganic Carbon (TIC) and Total Carbon (TC) and subtracting one from the other.

$$TC - TIC = TOC$$

Fig. 1 TIC and TC are measured and TOC is calculated

The European Pharmacopoeia chapter on TOC for PW and WFI, EP 2.2.44, calls for complete oxidation of the organic molecule for accurate TOC analysis, i.e. if some of the carbon atoms are not oxidised and remain bound into the organic molecule, then they would not be measured and the TOC analyser would under-report TOC. For this reason it is important that the TOC analyser is capable of detecting when oxidation is complete before reporting TOC levels.

TOC results are reported in Parts Per Billion (ppb) which in this case is the mass (weight) of organic carbon per litre of water. Longer-chain complex organic molecules will contain more carbon atoms than short-chain organic molecules, so equivalent numbers of the long- and short-chain molecules will be reported differently by the TOC analyser, with reported TOC from the long-chain organics delivering higher TOC results.
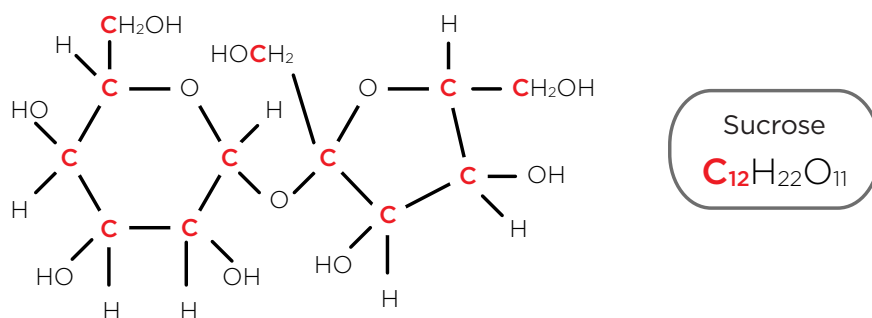
Sucrose
$C_{12}H_{22}O_{11}$

Fig. 2 Organic molecule sucrose contains 12 carbon atoms

Overview of ICH Q2

The ICH Q2 guideline covers three different applications for analysers: Identification, Testing for Impurities and Assay tests. TOC analysers used to test PW and WFI for contamination naturally falls under the Testing for Impurities application. ICH Q2 goes on to differentiate between the validation characteristics required for those analysers used for limit testing of impurities and those used for quantitative analysis of impurities.

- » Accuracy

- » Precision

- » Repeatability

- » Intermediate precision

- » Specificity

- » Detection Limit

- » Quantitation Limit

- » Linearity

- » Range

**Fig.3** ICH Q2 validation characteristics to be considered

On-line TOC analysers have the purpose of determining that the compendial limit of 500ppb is not breached, but they are also used for trending TOC levels and as such they fall under the scope of the validation characteristics applied to both quantitative testing and limit testing analysers.

Fig. 3 lists the validation characteristics used to determine the suitability of a TOC analyser. Whilst robustness is not listed, ICH Q2 recommends this aspect of validation during initial qualification of the analyser and at suitable periods thereafter.

Demonstrating Compliance to ICH Q2

Accuracy

ICH Q2 suggests that accuracy may be established by determining the closeness of agreement between the analyser and an accepted reference value. For TOC analysers, this can be achieved by carrying out a calibration validation, i.e. by running certified TOC standards as grab samples and determining if the results provided by the analyser are within accepted/specified performance limits. Naturally the analyser should have had a calibration adjustment before carrying out this test, i.e. the normal calibration adjustment procedure recommended by the manufacturer should have been carried out.
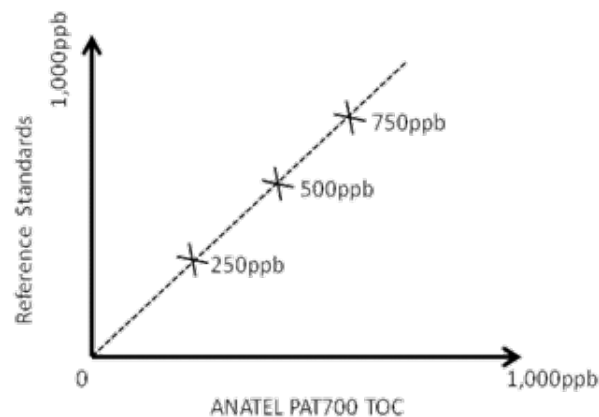
Fig. 4 determining accuracy of the TOC analyser using certified TOC standards

ICH Q2 suggests that accuracy should be demonstrated using 3 replicates of 3 concentrations, as shown in Fig.4.

Precision

The validation of analysers for quantitation of impurities includes an investigation of precision. ICH Q2 recommends the determination of precision of repeatability be validated using 3 replicates each of 3 certified standards at different concentrations. It also recommends that the effect of environmental changes and human error on the intermediate precision of the analyser be validated.

Specificity

This establishes the analyser's ability to measure the analyte of interest in the presence of potentially interfering substances. In the case of a modern PW or WFI system, the challenge TOC analysers face is that the TOC levels are relatively small, often less than 30ppb, whereas the levels of TIC and TC are comparatively high, sometimes between 1,000ppb and 2,000ppb. As stated in Fig. 1, TOC analysers measure TIC and TC and then calculate TOC. Small errors in measuring TC and TIC can lead to big errors in calculated TOC. This problem is most noticeable in TOC analysers that have separate sensors to measure TC and TIC, see example in Fig. 5.

Example:		Results:	
Actual TC	1000ppb	Measured TC	980 to 1020ppb
Actual TIC	990ppb	Measured TIC	970 to 1010ppb
Actual TOC	10ppb	Calculated TOC	-30 to 50ppb
Sensor			
measurement error	+/-2%		

Fig. 5 Error in reported TOC due to interference from TIC

Detection Limit

The pharmacopoeias state that suitable TOC analysers must have a detection limit (LOD) of ≤50ppb. Validating such a low level of TOC is difficult using traceable TOC standards as these are generally not available. Equally, attempting to make your own TOC solution at 50ppb is not practical. ICH Q2 suggests an alternative is to calculate the LOD by running multiple samples from a blank TOC solution (<100ppb TOC according to the pharmacopoeias) and using the standard deviation between the measurements to calculate the LOD, see Fig. 6.

$$LOD = \frac{3.3\,\sigma}{S}$$

where $\quad$ LOD = limit of detection
σ = the standard deviation of the blank sample results
S = the slope of the calibration curve

Fig. 6 Calculating the limit of detection

Quantitation limit

This is the lowest level of TOC that the analyser can accurately measure and report TOC values. Like the LOD, this can be difficult to establish using traceable TOC standards as these are generally not available. ICH Q2 again suggests an acceptable method to validate this is characteristic is by running multiple analyses of blank samples and using the standard deviation to calculate the quantitation limit, see Fig. 7.

$$QL = \frac{10\,\sigma}{S}$$

where   QL = quantitation limit
           σ = the standard deviation of the blank sample results
           S = the slope of the calibration curve

Fig. 7 Calculating the limit of quantitation

As can be seen from the formulas in Fig. 6 and Fig. 7, the limit of quantitation is approximately three times the detection limit.

Linearity

Linearity, when applied to TOC analysers, is the ability to draw a straight line through three or more points on the TOC analyser calibration curve. This may be ascertained by running three or more certified TOC standards on the analyser. As long as the linearity correlation coefficient is >0.99, then the analyser can be said to have a linear response. The correlation coefficient can be calculated during calibration if three or more calibration standards are used.

Range

Pharmaceutical-grade TOC analysers are designed to demonstrate that the 500ppb limit defined in the pharmacopoeia has not been exceeded. The acceptable range of a pharmaceutical TOC analyser is established by confirming that the analytical procedure provides an acceptable degree of linearity, accuracy and precision when applied to samples containing amounts of TOC within or at the extremes of the specified range of the analytical procedure. As the specified maximum is 500ppb, then the linearity, accuracy and precision must be demonstrated around this maximum. ICH Q2 recommends testing with certified TOC standards of at least +/-20% of the maximum. It is common practice to use 250, 500 and 750ppb calibration standards to determine the linearity and accuracy of the TOC analyser around the 500ppb maximum range as this exceeds the +/-20% required, giving +/-50% of the maximum.

Robustness

The robustness characteristic is used to demonstrate the reliability of an analyser with respect to deliberate variations in method parameters. For TOC analysers, the United States and European pharmacopoeias suggest that this is established using the System Suitability Test (SST), where the analyser response to TOC standards in deliberately varied organic materials is tested to ensure there is no large variation in results. Solutions of easily oxidised sucrose and difficult to oxidise benzoquinone, both containing 500ppb of carbon, are analysed on the TOC analyser. To demonstrate that the analyser is robust the reported results from both SST solutions must be within +/-15% of each other.

How the PAT700 on-line TOC analyser from Beckman Coulter demonstrates compliance to ICH Q2

The PAT700 from Beckman Coulter is designed to support compliance to ICH Q2 using the following:

ICH Q2 Characteristic	How the PAT700 Demonstrates Compliance to ICH Q2
Accuracy	Calibration validation using tripple replicates of three certified TOC standards.
Precision	
» Repeatability	Calibration validation using tripple replicates of three certified TOC standards.
» Intermediate precision	Automated, pre-programmed SOPs combined with no manual data entry and automatic pass/fail calculations removes opportunities for human error.
Specificity	Single measurement sensor for TIC and TC means PAT700 is not affected by the presence of large amounts of TIC, unlike TOC analysers that use separate sensors to measure TIC and TC and calcualte TOC.
Detection Limit	Run multiple samples of a blank and calculate using ICH Q2formula.
Quantitation Limit	Run multiple samples of a blank and calculate using ICH Q2formula.
Linearity	Use built-in calibration SOP, which automatically calculates and reports the linearity correlation coefficient and only allow a 'pass' if this is >0.990.
Range	250, 500 and 750ppb calibration standards to determine the linearity and accuracy of the TOC analyser around the 500ppb test limit.
Robustness	Use System Suitability test to demonstrate that analyser is reliable and not succeptable to errors when deliberate variations in method parameters are made.

References

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Author Biography

Tony Harrison is a Compliance and Applications Specialist for Beckman Coulter Life Sciences. Tony has spent the last twelve years in applied metrology in the pharmaceutical and healthcare manufacturing industries. Prior to that, he worked for companies providing process control automation solutions for manufacturing industries.

Tony was joint-editor of the ISPE Guide to Ozone Sanitization of Pharmaceutical Water Systems and was also chief editor of the PHSS Best Practice Guide for Cleanroom Monitoring.

Tony is a well-known international speaker and has provided educational seminars on pharmaceutical water quality, parenteral particle testing, ozone sanitization for water systems and cleanroom monitoring in UK, France, Italy, India, Germany, Malaysia, China, USA, Scandinavia, Ireland, Hungary, Switzerland, Indonesia, Belgium, Greece, Switzerland, Turkey, Egypt and Denmark.

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