WHITEPAPER

BECKMAN COULTER
Life Sciences

European Pharmacopoeia EP 2.2.44 and Total Organic Carbon

Abstract

Chapter 2.2.44 of the European Pharmacopoeia[4] recommends that Total Organic Carbon (TOC) analysers designed for measuring Purified Water (PW) and Water for Injection (WFI) fully oxidise the organic contaminants to carbon dioxide to release the organic carbon for accurate TOC measurement. The new revision of the European Pharmacopoeia chapter on Water for Injection (WFI) now permits the generation of WFI using Reverse Osmosis[1] (RO), paving the way for companies to reduce their energy bills and carbon footprint by moving away from using stills. However, this change came with a warning from the expert workshop[2] that guided the European Directorate on this change that reverse osmosis may not be as robust as a still for manufacturing WFI and they suggested that users should put an increased emphasis on on-line TOC and conductivity measurements as leading edge indicators that water quality may be about to be compromised. This paper discusses the background behind the European Pharmacopoeia's focus on complete oxidation of TOC in the light of the increased importance of on-line Total Organic Carbon (TOC) and conductivity measurement.

Importance of TOC & Conductivity with the new EP chapter on WFI

The Working Party who supported the European Directorate warned that RO water treatment may not be as robust as a still and highlighted the role of TOC and Conductivity analysers as early detectors of potential impending microbial or endotoxin excursion events.

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Nevertheless, by-products or remains from micro-organisms would be of organic nature and would likely impact the parameters currently monitored in the monograph. Should such materials be released in the system it was considered that TOC and conductivity would be impacted.
Therefore, the adjunction of requirements regarding stability of values measured on-line for TOC and conductivity might provide information regarding potential presence of micro-organisms in the system or release of by-products.

Fig. 1 Working group supporting European Directorate emphasises the importance of TOC and Conductivity analysers

An increase in TOC results in a water system that usually has consistent TOC results can be an important indicator that either the quantity of organic molecules present in the water is increasing, or that there is a different, more complex long-chain molecule entering the water system. Such changes in TOC results can be an indicator that the integrity of the water treatment system is being threatened and can be an early warning of a potential impending microbial breech. As such events are often transient in nature, it is important to capture a sample of the water at the time when the TOC increase is detected so that it can be sent to the quality control laboratory for further analysis to try to get to the root cause of the transient.

Clearly TOC and Conductivity analysers must be calibrated correctly and their calibration validated to ensure that they are working correctly in case a change in the water chemistry occurs. Equally, the intensity of the oxidising UV lamp needs to be monitored to ensure that it is strong enough to oxidise organic molecules sufficiently to extract all of the carbon atoms for accurate measurement.

Although TOC analysers cannot determine what type of organic molecules are present in the water, they can be used to detect a change in the organic chemistry in the water as longer-chain organic molecules tend to exhibit different oxidation profiles compared to short-chain organic molecules. If the TOC analyser is able to detect a change in oxidation profile when the water organic chemistry changes, this information combined with a change in TOC results can indicate to the user the need to carry out an investigation to determine if the water system integrity is potentially about to be compromised.

TOC and Conductivity

TOC analysis is a non-specific test, i.e. it is simply a measure of the carbon atoms found in any organic compound in the water, it cannot identify exactly which 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 as carbon dioxide (CO_2) and then measures the difference in water conductivity caused as the carbon dioxide dissolves into the water.

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

$$\text{TC} - \text{TIC} = \text{TOC}$$

Fig. 2 TIC and TC are measured and TOC is calculated

TOC results are reported in Parts Per Billion (ppb) which in this case is the mass (weight) of organic carbon per litre of water reported in mg/Litre. Longer-chain complex organic molecules may 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.

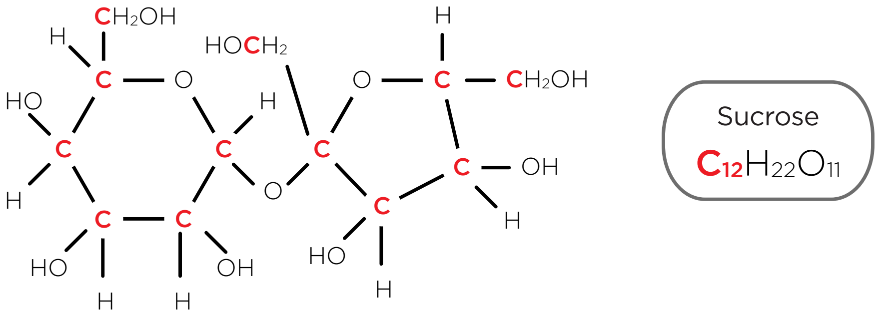


Fig. 3 Organic molecule sucrose contains 12 carbon atoms

TOC analysers that use conductivity to measure the organic carbon can also be used to detect inorganic contamination. However, the pharmacopoeias are very specific regarding calibration of conductivity analysers and the TOC analyser would need to comply fully with their requirements. USP<645> is very clear that the conductivity meter cell constant must be verified against a certified conductivity solution and that the meter electronics must be verified by the use of certified calibration resistor.

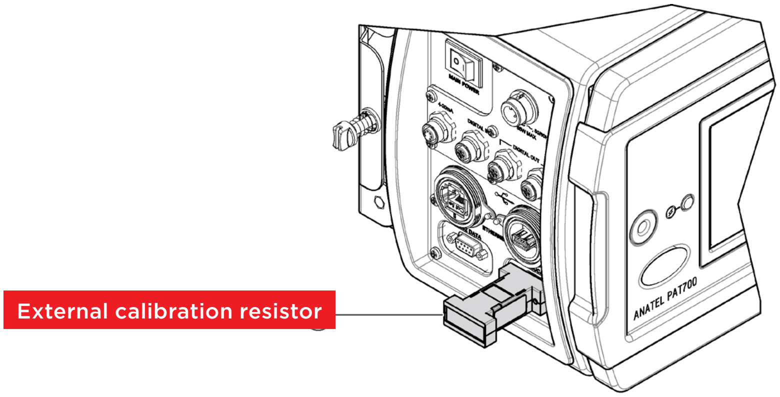
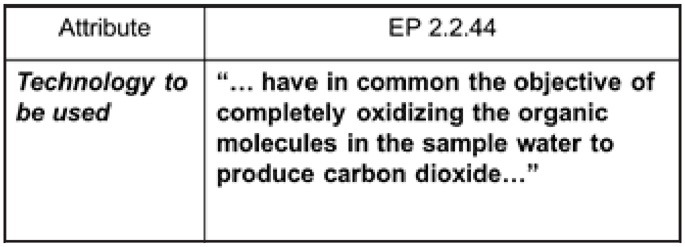


Fig. 4 Beckman Coulter PAT700 direct conductivity analyser meter accuracy can be verified using a certified resistor as per the requirements of USP<645>

Importance of Complete oxidation

The EP 2.2.44 chapter on TOC analysis[4] emphasises the importance of complete oxidation of the organic contamination in order to get an accurate TOC measurement.



Attribute	EP 2.2.44
Technology to be used	**"… have in common the objective of completely oxidizing the organic molecules in the sample water to produce carbon dioxide…"**

Fig. 5 Extract from EP 2.2.44 on TOC where complete oxidation is emphasised

As organic carbon is measured by extracting it from the organic molecules and measuring it as CO_2, either as a gas or in dissolved form using conductivity measurement, it is clear that the carbon present in the molecule must be completely oxidised to CO_2, otherwise it simply won't be measured.

Implications of incomplete oxidation

TOC analysers with short, fixed oxidation times can struggle to report TOC levels accurately. The International Conference on Harmonisation (ICH) is an expert working group with representation from the European, United States and Japanese Pharmacopoeias and in their ICH Q2, Validation Of Analytical Procedures[5], tripartite guideline they recommend a system suitability test to determine the robustness of an analyser's measurement. Should a TOC analyser with fixed oxidation times be calibrated with TOC standards using relatively easy to oxidise sucrose, then it will likely not report accurately if challenged with a relatively difficult to oxidise TOC standard such as SDBS as recommended in the Japanese Pharmacopoeia[6] chapter on TOC. The fixed flow-path and exposure time to the UV light will mean that the SDBS will not be fully oxidised and much of the carbon will remain un-measured.

Some TOC analysers employ persulphate as well as UV light to extend their measuring range. In the presence of UV light persulphate becomes a strong oxidising agent, enabling the TOC analyser to oxidise relatively large amounts of TOC in a short time. However, in designs employing gas-selective membranes as part of the measurement cell this can present challenges. If too much persulphate is added, there is a danger that bubbles may form in the sample and these bubbles can occlude the measurement membranes leading to under-reporting of TOC levels. Equally if too little persulphate is added, then the TOC will not be fully oxidised and, once more, the TOC analyser will under-report the TOC content of the sample. In this paradoxical situation, the analyser user somehow has to know in advance the value of TOC in the sample so that they can configure the persulphate dosing correctly on the analyser before they carry out their TOC measurement.

Longer chain, more complex organic molecules may create organic acids as intermediate species as they are being oxidised down to CO_2. Organic acids may contribute more strongly to the conductivity measurement than the final CO_2 from the completely oxidised organic, so failure to ensure complete oxidisation of the organic may result in an incorrect TOC value. Equally, failure to completely oxidise the organic may leave carbon atoms trapped in what remains of the organic molecule structure, leading to under-reported TOC results.

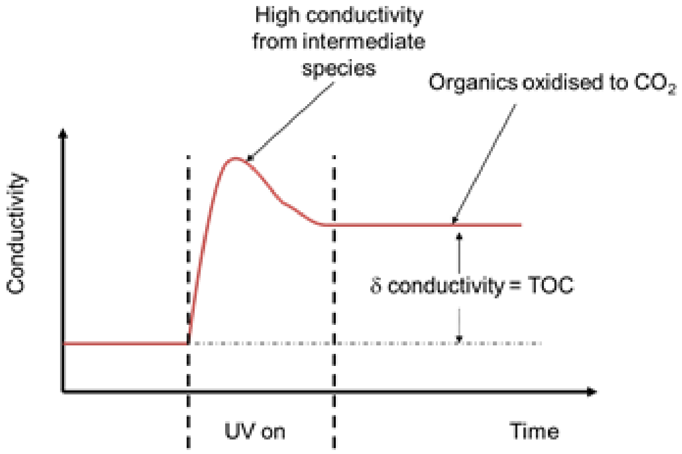


Fig. 6 intermediate species formed as the organic is oxidised may contribute more strongly to the sample conductivity than the final oxidised carbon

The PAT700 TOC analyser from Beckman Coulter

Designed specifically for TOC analysis on PW and WFI, the PAT700 TOC analyser from Beckman Coulter employs variable oxidation time combined with dynamic end-point detection to determine when the sample oxidation is complete. An aliquot of the water being sampled is trapped in the measurement cell. A single conductivity sensor measures the conductivity of this sample which is representative of the TIC in the sample. A UV light is then turned on to oxidise the organics in the sample. As the molecules are oxidised, the released organic carbon in the sample turns to carbon dioxide, which dissolves and increases the conductivity of the trapped aliquot. The PAT700 monitors the change in conductivity of the sample and when the conductivity stops changing, the PAT700 knows that the analysis end point has been reached and the sample has been completely oxidised. Now the sample contains both TIC and TOC, i.e. TC. The TC conductivity is measured using the same conductivity sensor used to measure TIC and one subtracted from the other to calculate TOC.

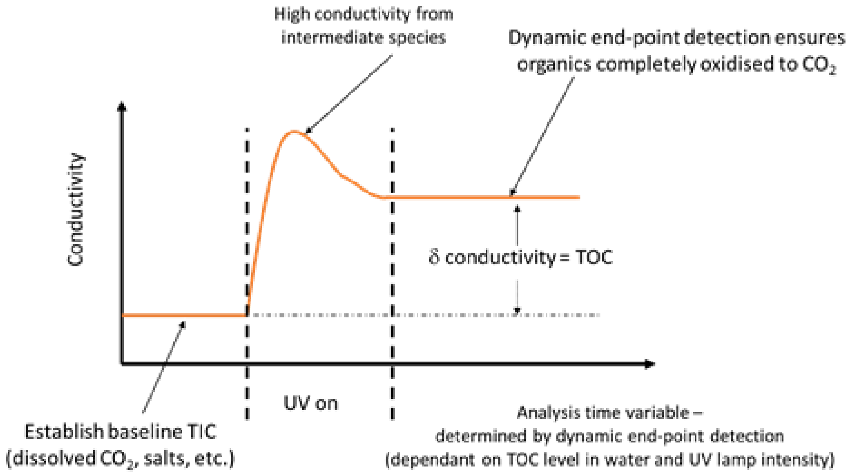


Fig. 7 Beckman Coulter PAT700 uses dynamic end-point detection to ensure complete oxidation for accurate TOC analysis, even when UV lamp intensity decreases

By using a single sensor to measure both TIC and TC, the effects of any slight conductivity sensor drift over time on the accuracy of the TOC calculation is eliminated as the conductivity sensor drift error would apply equally to both TIC and TC measurements and thus cancel out.

One of the weaknesses of the PAT700 is that it cannot be used on water samples not compliant with the pharmacopoeias' TOC limit test for PW and WFI because it does not use persulphate to extend its measurement range. It is limited to a measurement range of 2,000ppb. However, the upside of this is, unlike TOC analysers that use persulphate, the user does not need to guess at the TOC levels in the PW and WFI in order to correctly set the dosing level of persulphate.

As the oxidation time of the PAT700 is variable and controlled by dynamic end-point detection, the analysis of harder to oxidise materials such as SDBS recommended in the System Suitability Test in the Japanese Pharmacopoeia does not cause a problem: the PAT700 simply extends its analysis time, continuing to oxidise until it detects that the sample conductivity has stopped changing and the oxidation is complete.

The level of UV light being emitted from the UV lamp is measured to ensure that it is sufficiently powerful to oxidise the sample and, in the event of a lamp failure, a second, standby UV lamp is turned on to ensure complete oxidation.

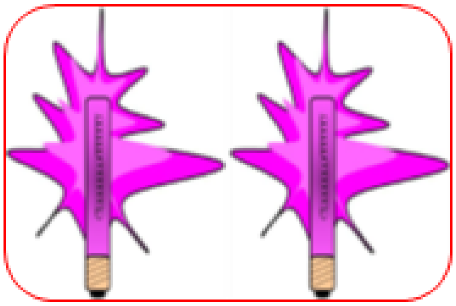
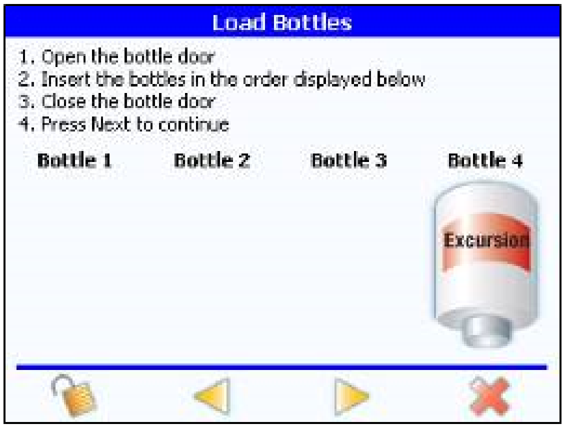


Fig. 8 PAT700 ensures there is always sufficient oxidising UV light by measuring UV output and having auto-switching main and stand-by UV lamps

Capturing TOC transient excursions for root-cause analysis

TOC or conductivity contamination events can be transient in nature. For example, a water system that is experiencing gradual biofilm build up may experience TOC excursions just after sanitisation cycles as biofilm sloughs off the inside of the pipework. This excursion can quickly disappear, either by being diluted as it passes into the large quantity of water in the loop storage tank, or broken down by UV lamps connected to the pipework distribution network put in place to discourage microbial build-up. However, this rapid disappearance may mask the increasing biofilm build up until there is a breakdown of system control and run-away microbial contamination. The PAT700 can be programmed to capture a water sample should a TOC excursion be detected so that the sample can be further analysed to get to root cause.



Load Bottles

1. Open the bottle door
2. Insert the bottles in the order displayed below
3. Close the bottle door
4. Press Next to continue

Fig. 9 Beckman Coulter PAT700 can capture a water sample to support root cause analysis if a TOC excursion is detected

TOC as a lead indicator of water system contamination

One of the additional advantages of dynamic end-point detection for complete oxidation in the PAT700 from Beckman Coulter, is that it can provide an indication of the oxidation curve profile during each TOC analysis. A change in the type of profile, combined with a change in TOC levels may provide insight into a change in the water system organic chemistry suggesting a potential degradation of the RO system integrity, prompting the user to investigate further and consider corrective action to prevent a large-scale contamination event and to put in place preventative measures to avoid the recurrence of the problem in the future.

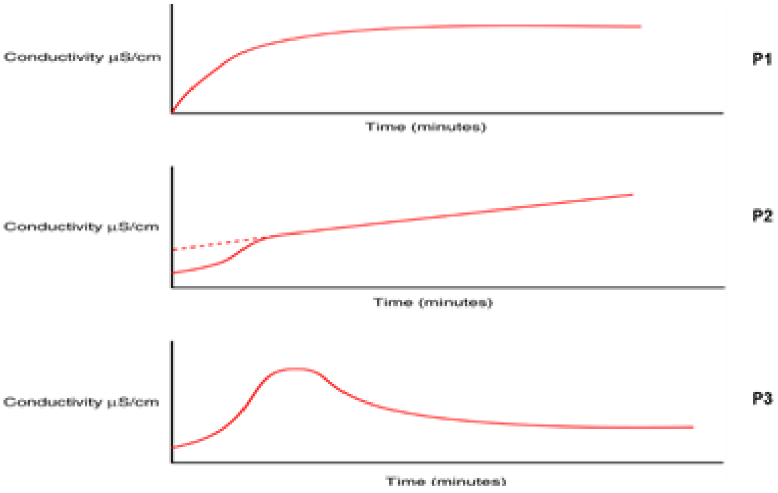


Fig. 10 Changes in TOC oxidation profile curve can indicate potential degradation of water treatment integrity, prompting investigation to prevent a large-scale contamination event

Conclusion

As leading-edge indicators of potential changes in water system health, on-line TOC and conductivity analysers are taking an ever increasing importance in preventing contaminated PW and WFI from mixing with expensive drug product during manufacturing. The ability to report changes in water organic chemistry combined with TOC transient excursion capture provides more support for root-cause investigations before the PW or WFI exceed the pharmacopoeial limits. Complete TOC oxidation to release all of the organic carbon as carbon dioxide, even with long-chain, difficult to oxidise organic molecules as per the European Chapter 2.2.44 is paramount if accurate TOC measurements are to be made.

References

1. European Pharmacopeia (Ph. Eur.) Commission press release 18th March 2016
   https://www.edqm.eu/sites/default/files/press_release_water_for_injections_march_2016_0.pdf

2. EDQM Expert Workshop, 24 March 2011 European Directorate for the Quality of Medicines & Healthcare (EDQM) 7 allée Kastner, CS 30026 F -67081 Strasbourg

3. US Pharmacopeia Convention, United States Pharmacopoeia, Rockville MD, USA and Council of Europe, European Directorate for the Quality of Medicines & Healthcare, European Pharmacopoeia, Strasbourg, France

4. Council of Europe, European Directorate for the Quality of Medicines & Healthcare, European Pharmacopoeia 8.0, 01/2008:20244, Total Organic Carbon in Water for Pharmaceutical Use, Strasbourg, France

5. International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use VALIDATION OF ANALYTICAL PROCEDURES: TEXT AND METHODOLOGY ICH Q2 9, ICH Secretariat, Chemin des Mines, 1202 Geneva, Switzerland
   http://www.ich.org/products/guidelines/quality/article/quality-guidelines.html

6. The Japanese Ministry of Health Labour and Welfare, The Japanese Pharmacopoeia, Seventeenth Edition, Pharmaceuticals and Medical Devices Agency, Shin-Kasumigaseki Building, 3-3-2 Kasumigaseki, Chiyoda-ku, Tokyo 100-0013 Japan **https://www.pmda.go.jp/english/rs-sb-std/standards-development/jp/0019.html**

7. International Society for Pharmaceutical Engineering, The ISPE Good Practice Guide: Ozone Sanitization of Pharmaceutical Water Systems, First edition July 2012
   http://www.ispe.org/ispe-good-practice-guides/ozone-sanitization-pharmaceutical-water-systems
   [14th August 2014]

8. Pharmaceutical and Healthcare Sciences Society, Best Practice for Particle Monitoring in Pharmaceutical Facilities, PHSS Technical Monograph No.16, First Edition 2008, ISBN 978-1-905271-15-3

9. Ministry of Health, Labour and Welfare, The Japanese Pharmacopoeia, 17th Edition, 1-2-2 Kasumigaseki Chiyoda-ku Tokyo, 100-8916 Japan

Author Biography

Tony Harrison is a Compliance and Applications Specialist for Beckman Coulter Life Sciences. An experienced engineer in water system TOC, pH, conductivity and ozone analysis, 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[5] and was also chief editor of the PHSS Best Practice Guide for Cleanroom Monitoring[6].

Tony is a well-known international speaker and has provided educational seminars on TOC, liquid particle counting, 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, Columbia, South Africa and Denmark.

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