Elemental Impurities – Efficient Preparation of Diverse Samples

Efficient Digestions Enable Elemental Impurities Control

Recently regulations for the testing of elemental impurities have been tightened across a variety of industries. In this regard microwave closed vessel digestion plays a significant role in order to achieve reliable results. Especially for samples representing a wide range of matrices using a pressurized digestion cavity provides major advantages compared to conventional pressure vessels.

Elemental impurities not only constitute a toxicological risk for the consumer, for products containing active ingredients i.e. medication or nutritional supplements it may affect their quality and efficacy. Traditionally in particular in the pharmaceutical industry heavy metal analysis was performed by sulfidic precipitation according to more than hundred years old procedures which were non-specific and did not allow the determination of concrete quantitative results. Finally after an intensive period of development the outdated methods based on wet chemical analysis have been replaced step by step by modern instrumental analysis documented in relevant regulations. Thereby the microwave closed vessel digestion has a decisive influence on the quality of analytical results.

Elemental Impurities in a Variety of Products and Regulatory Background

Pharmaceutical Products

For prescription medicines, vaccines, vitamins and minerals the Therapeutic Goods Administration (TGA) in Australia, refers to default standards to be followed. 'Default standard' means any of the British Pharmacopoeia, European Pharmacopoeia, and United States Pharmacopoeia-National Formulary. (Therapeutic Goods Act 1989 (Chapter 1, section 3)). Due to profound changes in each of the relevant pharmaceutical regulations the consideration of elemental impurities is mandatory for all pharmaceutical products since January 2018.

For elemental impurities like lead, arsenic, cadmium and mercury, compliance to either the United States Pharmacopoeia USP <232> and USP <233> or the International Conference on Harmonisation ICH Q3D are acceptable for pharmaceutical products. For dietary supplements USP<2232> is applicable. All these documents provide guidance on methods for establishing maximum values for elemental impurities based on the permitted daily exposure (PDE) of each element.

(*https://www.tga.gov.au/book-page/minimum-quality-standards* 1 April 2019)

Dairy & Infant Formula Exports to China

The Australian Department of Agriculture advises that since 1 May 2012, exporters of dairy products to China are required to provide test results that demonstrate that the products comply with China's food safety standards (GB standards). Especially stringent are the requirements for Baby Infant Formula, which needs to be tested for Elemental Impurities such as Iron, Copper, Zinc and Lead amongst other substances. (https://micor.agriculture.gov.au/ 17 Aug. 2018)

The Chinese National Food and Safety Standard: GB5009.12-2017 for the Determination of Lead in Foods list a microwave digestion system as a required apparatus. (https://www.chinesestandard.net/PDF.aspx/GB5009.12-2017)

Medicinal Cannabis

After legislation in Australia and New Zealand producing Medicinal Cannabis has developed into a growing industry with both governments issuing licenses to growers who comply with a range of stringent requirements. The quality and safety of the product is to be tested as outlined in the Therapeutic Goods Standard for Medicinal Cannabis (TGO 93) Order 2017 which also provides limits for heavy metals such as arsenic, cadmium, lead and mercury amongst other impurities.

(https://www.legislation.gov.au/Details/F2019C00328 31 March 2019)

To test for the presence of these elemental impurities samples need to be digested, here a microwave system offers an efficient method.

Elemental Impurities – Efficient Preparation of Diverse Samples

Importance of Reliable Sample Preparation

The recent regulatory requirements are highly demanding concerning the specifications of analytical instruments. In this regard particular attention is paid to the performance characteristics specificity, limit of determination, precision and accuracy. As an important premise to obtain suitable data any kind of sample material has to be transferred into a measurable liquid form whilst avoiding the introduction of possible interferences to the measuring system. Not only USP <233> but also Ph. Eur. 2.4.20. refers to closed vessel digestion to ensure appropriate digestion conditions. In order to digest different and sometimes very complex matrices of pharmaceutical products high temperatures are required for a total digestion and subsequently interference-free measurement.

Drawbacks of Common Microwave Digestion Systems

The key parameter of the digestion quality is the temperature: With increasing temperature the residual carbon content and subsequently the related measuring interferences as well as the process times are decreasing. In conventional microwave digestion systems reaching a high temperature (higher than the boiling point of the acids) is enabled by employing closed pressure vessels placed inside a rotor which is heated up in a large microwave cavity. The pressure inside a vessel correlates based on the vapor pressure of the used acids with the temperature and the pressure caused by the formation of gaseous reaction products. To be able to reach required temperatures of 240 °C the vessels have to withstand pressure levels of at least 40 bar. High pressure stability usually is ensured by more complexly designed vessels and related handling steps. In order to not exceed the pressure or temperature limit of those pressure vessels and on the other hand to ensure to reach the temperature levels being necessary for complete digestions, individual methods have to be developed for different samples resulting in a huge validation effort when working with different kinds of samples. During the daily routing, sample clustering is necessary.

The Difference - Pressurized Digestion Cavity

The core feature of the microwave digestion system Multiwave 7000 is the PDC (Pressurized Digestion Cavity). In contrast to conventionally used microwave digestion systems, the cavity is used as a pressure jacket instead of using pressure vessels. As reaction containers serve simple vials made of quartz, PTFM or borosilicate glass which only have to be closed with plug-on caps. Prior to microwave heating the cavity is pressurised with nitrogen. This pressure-sealing suppresses foaming and boiling during the digestion and thus eliminates cross-contamination or loss of volatile elements. During the digestion the vials are surrounded with a load solution which ensures homogenous heating of all samples and acts as an absorber for exothermic reactions.

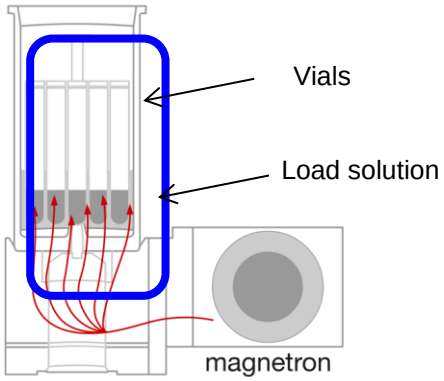

Figure 1: The Pressurized Digestion Cavity together with the microwave introduction: the blue frame shows the pressurised cavity, the red arrows indicate the path the microwaves take from the magnetron to the samples.

Contrary to conventional microwaves, with a PDC there is no need for sample clustering or time-consuming method development and validation efforts, the implemented standard method for pharmaceutical samples just heats up to 250 °C and holds the temperature for 15 minutes. Using this method pills, tablets, capsules and liquids as well as raw materials, sugars and oils can be perfectly digested in the same digestion run.

Proof of Concept: Digesting Different Pharmaceutical Samples with One Method

Four pharmaceutical products were digested with the microwave system Multiwave 7000 by Anton Paar. Following the aim to cover a wide range of different formulations and varying reaction behaviour during the digestion the below listed products were used:

- ***Painkilling Tablet:***
  Containing acetylsalicylic acid as active ingredient and difficult to digest silicon dioxide as excipient
- ***Dietary Supplement Capsule:***
  Containing omega-3 fatty acids and reactive salmon oil
- ***Cold Syrup:***
  Containing a triple active ingredient complex and high amounts of alcohol, sugar and glycerol
- ***Nicotinic Acid:***
  Aromatic API requiring temperatures above 200 °C for total digestion

In accordance with the validation requirements stated in USP <233> and Ph.Eur. 2.4.20. all four drug products were spiked at 50%, 100% and 150% of the so-called target limits. The samples were digested with Multiwave 7000 equipped with a rack for 18 quartz or PTFE vials. After measuring the solutions with ICP-MS the recovery rates were calculated.
Because of the toxicological and analytical relevance (volatile mercury and arsenic compounds) the focus was placed on the "big four" Cd, Pb, As and Hg.

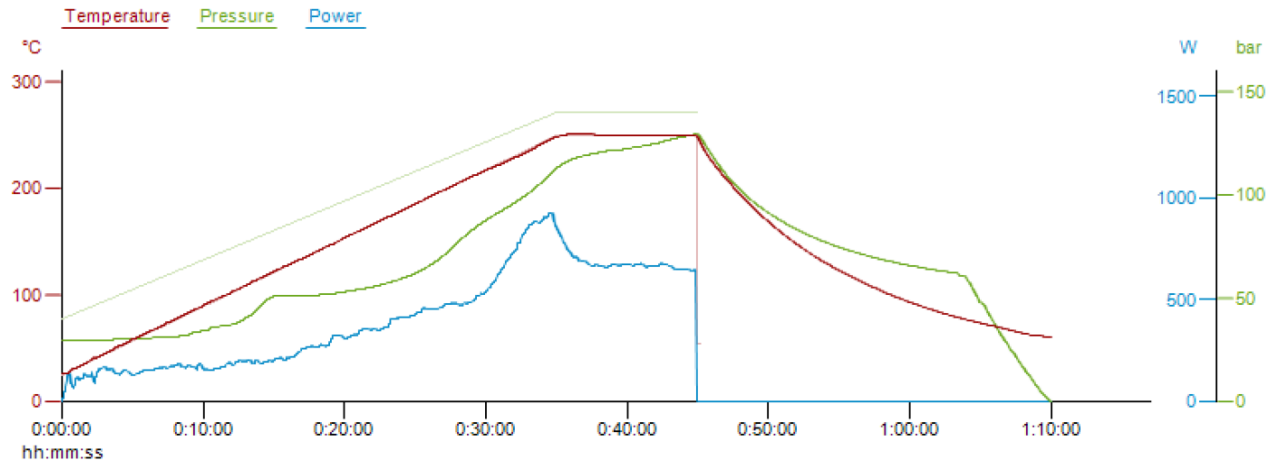

Figure 2: Run Data

Elemental Analysis in Accordance with Regulatory Requirements

Not only in relation to the limits stated under procedure validation in USP <233> and Ph.Eur. 2.4.20. (70 % - 150 % for spike recovery and not more than 20 % for the relative standard deviation - RSD) but also concerning the absolute figures itself, excellent results were achieved: On average the recovery rates for all spike levels and products reached 101 % for Cd, 94 % for Pb, 102 % for As and 100 % for Hg (see. Figure 3). The relative standard deviation (RSD, n = 3) in any case is not higher than 5 %.

It could be shown that microwave closed vessel digestion with Multiwave 7000 represents an efficient sample preparation technique for the elemental analysis of a huge variety of pharmaceutical products in compliance to all current regulatory requirements. Highly reproducible and comprehensibly documented results can be achieved by means of temperature controlled digestion conditions in combination with easy

handling. For the applications in the pharmaceutical industry, the specific qualification documentation according to 21 CFR Part 11, GAMP 5 and USP <1058> is available, which allows a GMP compliant operational readiness of the device within one to two days.

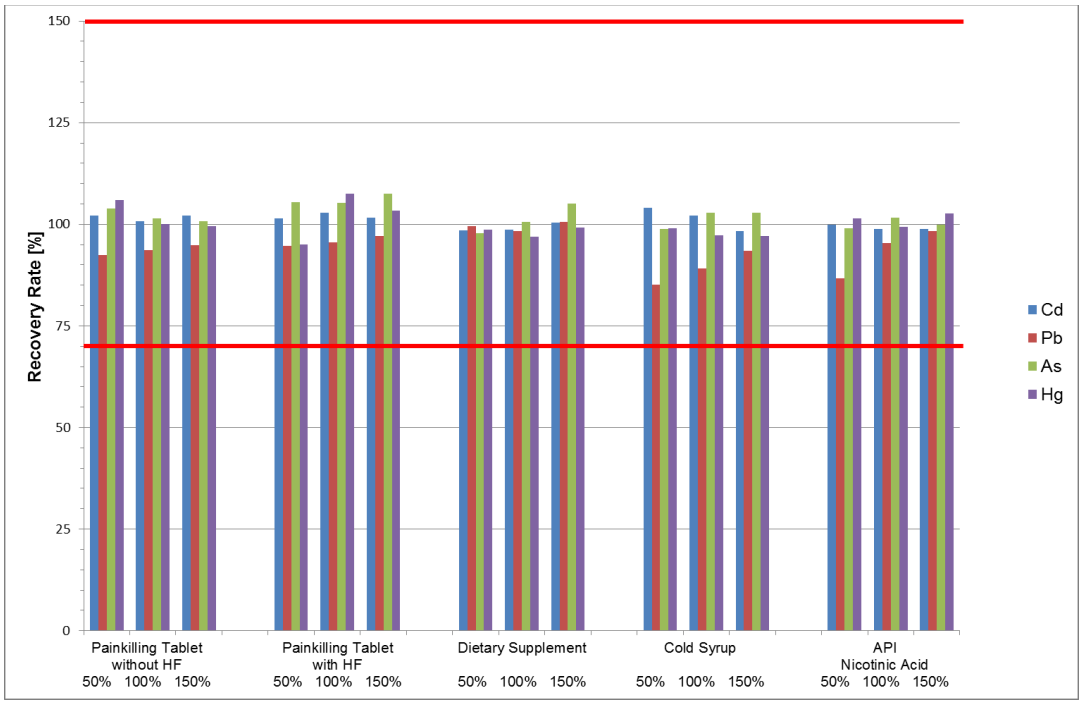

Fig. 3: Recovery Rates for Cd, Pb, As and Hg

Fig. 3: All products were digested with microwave-assisted digestion system Multiwave 7000

Fig. 4: The digestions were performed with Pressure Sealed Vials in Rack 18

Conclusion

The Multiwave 7000 from Anton Paar offers an efficient way to digest a variety of samples simultaneously with one method without cross contamination, thus guaranteeing a very high recovery rate of commonly present elemental impurities. Application notes are available for digestion of challenging matrices such as petroleum samples, polymers, food and many more.