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Review

The application of inductively coupled plasma mass spectrometry in pharmaceutical and biomedical analysis

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Abstract

With the development of life science, pharmaceutical and biomedical analysis becomes more and more important in medical science. Further studies will be hopefully established if it is possible to use inorganic elemental standards or small organic compounds in the quantitative determination of all kinds of drugs, nucleotides and sulfur or phosphorus containing peptides and proteins at appropriate concentration with an acceptable accuracy. Since 1980, inductively coupled plasma mass spectrometry (ICP-MS) has emerged as a new and powerful analytical technique which is suitable for element and isotope analysis. It offers extremely wide detection range of element and co-analysis of most elements in the periodic table. Also, it can be applied to perform qualitative, semiquantitative, and quantitative analysis and isotopic ratios through mass-to-electric charge ratio. With the help of ICP-MS, the struggle of searching for an excellent quantification technique in, e.g. drugs and proteomics has come appreciably close to an end. This review mainly focuses on the introduction of application of ICP-MS in pharmaceutical and biomedical analysis. Some problems in application and the handling strategies are simply presented at the end.

Keywords: Inductively coupled plasma mass spectrometry (ICP-MS); Pharmaceutical analysis; Biomedical analysis

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1. Introduction

Since 1980, inductively coupled plasma mass spectrometry (ICP-MS) which is composed of plasma, as the high temperature (8000 K) ionization source, quadrupole mass spectrometer (MS) analyzer, as the sensitive rapid scanning detector and a distinctive interface has emerged as a new and powerful technique for element and isotope analysis [1]. In approximately 10 years, ICP-MS has progressed from a laboratory experiment to commercial development and widespread analytical applications [2–9]. This growth is primarily due to the fact that ICP-MS offers extremely wide detection range of element and co-analysis of most elements in the periodic table, for example, a wide range of elements in concentration levels from ppt to ppm level can be measured in a single analysis. It can perform qualitative, semiquantitative, and quantitative analysis through mass-to-charge ratio. It can also measure isotopic ratios, since employed a mass analyzer. And it is so versatile that the technique can substitute almost all traditional inorganic analytical technique, such as ICP-AES, GF-AAS, F-AAS, etc., in analytical capability. ICP-MS has been coupled to all forms of sample introduction or separation techniques for special analyses, including laser-assisted sample introduction [10,11], low pressure chromatography, high-performance liquid chromatography [12], gas chromatography [13], capillary electrophoresis [14,15], and so on. As one of the most significant developments in analytical science nowadays, ICP-MS has been widely used in many industries. The application which was introduced into geological science research at first has rapidly enlarged to other fields including semiconductor [16], environmental [17], nuclear [18], chemical, clinical [19], and research laboratories after1984 when the first commercial instrument emerged.

As the ionization source of MS, the advantage of this technique is that the ICP can solve two problems delicately in design of this sort of ionization source. For instance, it supplies the controllable, non-polluting and high-temperature environment which are suitable for specimen stimulation and sampling condition; otherwise, it provides an environment where the retention time is enough for all expected procedures and which is fit for rapid and complete sample introduction. Compared with the traditional inorganic analytical technique, ICP-MS offers wider linear range, lesser interference, higher analytical precision, shorter analytical time and lower detection limit which ranges from sub part per billion (ppb) to sub part per trillion (ppt) for most elements, besides providing precise isotope information. However, the biggest advantage of ICP-MS is that all spectrograms are extremely simple because the peaks mainly come from single charge ion, no matter how complicated the matrix is.

For many sample types and substances, reference or standards are lacking. This problem is often encountered in environmental, biological and clinical analyses as well as in drug development. In this paper, we will describe the construction, principle and some selected analytical application in pharmaceutical and biomedical analysis of ICP-MS.

2. The principle and construction of ICP-MS

The principle and construction of modern ICP-MS instrument are basically identical on the whole. The ICP-MS instrument employs plasma (ICP) as the ionization source and a mass spectrometer (MS) analyzer to detect the ions produced. The mainly used plasma gas is argon, since it can simultaneously excite and ionize most of the elements in periodic system efficiently, which makes multi-element analysis possible.

Taking the Agilent 7500 as an example, liquid samples are generally introduced by a peristaltic pump, to the nebulizer where the sample aerosol is formed. A double-pass spray chamber ensures that a consistent aerosol is introduced into the plasma. Argon (Ar) gas is introduced through a series of concentric quartz tubes which form the ICP. The torch is located in the center of a RF coil, through which RF energy is passed. The intense RF field causes collisions between the Ar atoms, generating high-energy plasma. The sample aerosol is instantaneously decomposed in the plasma (plasma temperature is in the order of 6000–10,000 K) to form analyte atoms which are simultaneously ionized. The ions produced are extracted from the plasma into the mass spectrometer region which is held at high vacuum (typically 10^{-4} Pa), which is maintained by differential pumping: the analyte ions are extracted through a pair of orifices, known as the sampling and skimmer cones. The analyte ions are then focused by a series of ion lenses into a quadrupole mass analyzer, which separates the ions based on their mass-to-charge ratio. Finally, the ions are measured using an electron multiplier, and collected by a counter for each mass number.

3. The application of ICP-MS in pharmaceutical analysis

Nowadays, pharmaceutical analysis not only refers to static routine control, but also includes dynamic analysis and has been used for monitoring in the reaction mechanism, metabolic pathway in vivo and full-scale estimation with the assistance of modern analytical method and technique. According to the international drug standard and rapid developments of instrumental analysis, analysts have to promote their ability unceasingly and master the application of modern instrument in pharmaceutical analysis.

Present means of analyses mainly face to organic synthetic drug, and determination of inorganic drug is accomplished by volumetric analysis directly or converted to organic analysis indirectly. However, with the development of modern inorganic analytical technique, the analytical task of inorganic drug can be

completed directly; meanwhile we can also change the analyses of organic drugs to simple inorganic analyses. As an inorganic analytical technique, ICP-MS has played an important role in many fields and been introduced into research and analysis of pharmaceutical science, including quantitative analysis of drug and its metabolites, biopharmaceutical analysis, limit tests of impurity and evaluation and quantity control of the Traditional Chinese Medicine.

3.1. Quantitative analysis of medicine and its metabolites

Quantitative analysis of drugs which can be enforced under diverse assay methods is one of the most important subjects in pharmaceutical analysis and also the primary means in quality evaluation of drugs. Complicated pretreatment which is not only time-consuming and cumbersome, but also specimen-wasting and unacceptable for biological specimen is needed and usually involves in heating or ashing step before analysis of metal- or halogen-containing organic drugs. Although identification of drug metabolites can be executed through LC–MS, quantitative analysis of metabolites will be very difficult if there is not proper reference (usually there is no reference). Synthesis with radioactive labeling is another way for this problem, but it is money-costing and time-consuming, and not a good choice unless combining NMR (^{1}H, ^{19}F) for quantitative analysis.

ICP-MS not only offers high temperature (8000 K) ionization source for specimen provocation, but also overcomes the limitation of RI, UV or MS for accurate quantification which is concerned with the molecular structure of specimen. Even if the chemical structure or elemental composition is known, the response from these detectors is difficult to predict with any accuracy. In ICP-MS, compounds are atomised and ionised irrespective of the chemical structure incorporating the element of interest. Therefore, it is not necessary to choose the same or analogous molecular structure of reference as the specimen and one reference is enough when quantitative analysis is carried out with ICP-MS. Axelsson et al. [20] applied ICP-MS coupling with LC in generic detection for structurally non-correlated organic pharmaceutical compounds with common elements like phosphorus and iodine. They found that detection of selected elements gave a better quantification of tested 'unknowns' than UV and organic mass spectrometric detection and did not introduce any measurable dead volume and preserves the separation efficiency of the system.

The wider linear range, lesser interference, higher analytical precision, shorter analytical time and lower detection limits of ICP-MS provide enormous convenience for quantitative analysis of drug and its metabolites. One of these reports employing this approach was carried out by Nicholson et al. [21], which described the profiling and quantification of metabolites of 4-bromoaniline in rat urine. This was followed by a similar study [22] that provided the simultaneous detection of the metabolites of 2-bromo-4-trifluoromethyl-[^{13}C]-acetanilide in rat urine by ICP-MS. The metabolites present in the sample were separated by reversed-phase LC and introduced into ICP-MS instrument where bromine-containing metabolites were detected and quantified by ICP-MS.

ICP-MS was also investigated to determine the ^{99}Tc/^{99m}Tc ratios in sodium pertechnetate by Hill et al. [23]. It has been recognized for over 20 years that the presence of increased levels of the long-lived radioisotope ^{99}Tc in solutions of ^{99m}Tc can adversely affect labeling efficiencies of a number of sensitive reagent kits used in nuclear medicine. Some government authorities impose strict regulatory requirements on the manufacturers of ^{99}Mo/^{99m}Tc generators to ensure that high quality pertechnetate is available to the nuclear medical community. Trace levels of ^{99}Tc in pertechnetate samples have been quantitatively determined using a number of analytical techniques such as HPLC [24]. Each of these analytical methods suffered a sensitivity problem in the low ng mL^{-1} concentration range. However, the ICP-MS analysis had a sensitivity of approximately 50 pg mL^{-1} for technetium in an isotonic saline matrix and technetium concentrations were measured down to 200 pg mL^{-1} (200 ppt).

The ICP-MS is a multi-element detector with high sensitivity, less interference and therefore is a useful tool for investigating metal species behavior. Cisplatinum and carboplatinum complexes are widely used in the treatment of solid tumor. Falter and Wilken [25] adopted ICP-MS coupling with RP-HPLC for investigating the species behavior of the two platinum anti-tumor drugs in aqueous phases. The detection limits were found to be 80 pg for cisplatinum and 130 pg for carboplatinum. The standard deviation was 5% for both compounds.

There are some significant elements for quantitative analysis in the application of ICP-MS: (1) base metal element (Li, Na, K, Rb) and alkali metal element (Mg, Ca, Sr, Ba); (2) transition element which is associated with enzyme, including Cr, Fe, Cu, Zn; (3) Pt, which is used for anti-tumor drugs; (4) the hetero-atoms P, S, Cl, Br, I, which are usually the ingredients of organics. And element-specific detection via quadrupole ICP-MS of chlorine and bromine-containing drugs has been successfully applied [21,22,26]. Accordingly, ICP-MS has been used in analysis of protein phosphorylation by monitoring the phosphorus [27]; (5) other elements, such as Hg, As, and so on. On the whole, all specimen containing elements mentioned above or not can be analyzed rapidly by ICP-MS. In addition, radioelement can be also assayed by ICP-MS.

Vitamin B_{12}, the only metal-containing vitamin, is an essential nutrient for all cells. It acts as a co-enzyme for normal DNA synthesis and promotes normal fat and carbohydrate metabolism. The most means of detection such as UV/Vis, AAS and AEC has limited sensitivity and/or are non-selective, making them unsuitable for determining low levels of cobalamins in complex matrices. Baker and Miller-Ihliet [28] tried to optimize the combination of CE and ICP-MS to determine the suitability of the approach for cobalamin speciation measurements in pharmaceutical preparations and food samples. The purpose of their work was to investigate CE-ICP-MS for the analysis of cobalamins and the potentially harmful corrinoid analogue, cobinamide dicyanide. It is suggested that the technique could be used for the rapid screening of samples for CN-Cb1 in a quality control setting, since most pharmaceutical preparations and fortified products utilize this form. And they believed that ICP-MS is much less time-consuming and would be proved useful for routine determinations.

In the pharmaceutical industry, the detection and identification of impurities/metabolites structurally related to the drug substance are of utmost importance. Conventional methods of analysis to detect, track, quantify, and identify drug substances and related impurities use molecular mass spectrometry via atmospheric pressure ionization (API-MS) or, in the case of drug metabolism and pharmacokinetics, use radiolabeling. However, a significant number of drug substances containing heteroatoms can be detected using element-specific detectors such as ICP-MS. In the work of Evans et al. [29], structurally related impurities well below the 0.1% mass fraction level relative to the main drug substance in the sulfur-containing drug substance cimetidine could easily be detected with liquid chromatography coupled to sector field inductively coupled plasma mass spectrometry (SF-ICP-MS). The structure of most of the impurities was confirmed by electrospray mass spectrometry (ESI-MS), and thus, the complementarity of the two techniques for drug analysis is shown. The limit of detection by SF-ICP-MS for cimetidine in solution was 4–20 ng g^{-1}. In the analysis of sulfur-containing cimetidine drug substance, the main isotope of sulfur (^{32}S, abundance 95.018%) suffers from a serious interference as a result of the polyatomic ion $^{16}O^{16}O^{+}$ at nominal *m/e* 32. However, Evans et al. resolved this problem through regulating the resolution, allowing the sensitive and selective determination of sulfur.

3.2. Biopharmaceutical analysis

Compared to the routine analyses, there are many distinctions in selectivity, sensitivity and analytes in biopharmaceutical analysis. The more complex biological specimen, trace drugs distributing into massive body fluid and the interference from considerable endogenous interferent and metabolites make the separation and analysis more difficult. Because of the small amount of biological specimen which is not easy to re-obtain, it is more important to develop an analytical method of high sensitivity and well selectivity. ICP-MS will become an excellent and ideal choice if the analytes in biological specimen include any element that is proper for detecting by this analytical tool of high sensitivity and good selectivity.

Research indicates that the tumor is influenced by As in various aspects with the result of cytodifferentiation and apoptosis or re-duplication inhibition of tumor cells. Therefore, it is significant to monitor the concentration of As exactly in serum due to the serious toxicity and extremely narrow therapeutic window. Wang et al. [30] determine the concentration of As in blood samples from healthy volunteers in the study of pharmacokinetics of the Compound Realgar Natural Indigo Tablets (CRNIT) with ICP-MS. This method had a low detection limit of 1 ppm, RSD of 1.45% and recover of 95.03–98.68% and eliminated all sorts of drawbacks (such as large requirement of blood sample, low sensitivity, unreliable technology and obtaining a higher concentration of As in blank specimen than drug-taking sample) of other routine methods such as fluorometric method and AES.

Bismuth compounds have long been used in the treatment of a variety of gastrointestinal disorders, including diarrhea, gastritis and ulcers. Compounds containing radioactive bismuth isotopes (^{212}Bi, ^{213}Bi) have also been used as targeted radiotherapeutic agents for cancer therapy. Sun and Szeto [31] presented a study on interaction of bismuth anti-ulcer agents with albumin. The competitive binding of bismuth to albumin and transferring was investigated by HPLC and followed by the measurement of Bi^{3+} contents via ICP-MS. They also provided a basis on the pharmacology of Bi^{3+} drugs.

Zhao et al. [32] had described a sensitive method for measuring cisplatin and some possible metabolites. The separation and detection for cisplatin hydrolysis products and the reaction products of cisplatin with methionine, cysteine, and glutathione were investigated with their method combining reversed-phase ion-pairing LC with ICP-MS. The detection limit for cisplatin was found to be 0.1 ng. Three methods, AAS, ICP-AES and ICP-MS, for tissue platinum measurement were attempted by Minami et al. [33]. At last they had a conclusion that ICP-MS is the most suitable and sensitive method because of its low detection limit for determination of tissue platinum. These researches had demonstrated the usefulness of ICP-MS for studies of platinum-containing anti-tumor drugs.

3.3. Heavy metal limit tests

The current limit tests methods for heavy metal were developed before the advent of modern analytical instrumentation and are based on Pharmacopeia, which can be easily transferred from one laboratory to another and do not require expensive instrumentation or highly trained laboratory personnel to perform them. However, the methods rely on a subjective visual examination, require large amounts of sample, provide no qualitative or element-specific information, and usually involve a heating or ashing step, which is known to cause losses of the volatile elements. In addition, the treatment of colored sample, the interference of insoluble sulfuret because of the detected drugs and the presence of other metal elements make the limit test more formidable. In a word, all these factors mentioned above make limit tests methods of Pharmacopeia a difficulty in obtaining reliable and reproducible results.

The advantages of ICP-MS redeem all the shortcoming of conventional methods and it was selected as the basis of the alternative method, making the analysis simple and controllable, since it provides good sensitivity, requires minimal sample size, affords minimal elemental interferences, provides a means to perform rapid and automated multi-elemental analyses and there is no dependence of the various chemical functionalities contained in the sample matrices on the individual element recoveries. Accordingly, Wang et al. [34] and Lewen et al. [35] suggested that ICP-MS can be applied to do limit tests of drug substances, intermediates, and raw materials instead of the routine methods collected in the Pharmacopeia such as USP, BP, EP, and so on.

3.4. Evaluation and quality control of Traditional Chinese Medicine

Evaluation of Traditional Chinese Medicine (TCM) is very important to the quality control of TCM, and can supply instruction for planting and culture, too. The characteristics of trace

elements of TCM, including complex composition, extremely low content and intricate existing form, are the main standards for quality evaluation. So, the high sensitivity and good selectivity should be considered at first in the choice of analytical methods. Before ICP-MS, ICP-AES played an important part in application of this direction. Later ICP-MS was introduced into analysis of trace elements of TCM [36,37], and became a received and effective analytical tool with the microwave-assisted oven systems in this field.

Huang et al. [38] had accurately determined the heavy metals in reference material of radix salvia planted in Zhongjiang, Sichuan Province, China under the good agricultural procedure (GAP) with ICP-MS. The measuring method was validated by running certificated reference materials under the same conditions. The recoveries of the elements mostly ranged from 90 to 110%, and the RSD was within 5%. They also repeated their measurement by different laboratories with ICP-MS and by several time intervals in one year for the stability. The results showed that the concentrations of the heavy metals provided were accurate and the reference material was stable. At last they made a conclusion that the reference material is suitable to be the criterions of heavy metals for radix salvia in the qualities controlling, and is also suitable to be the criterions of poisonous heavy metals of other herbs in the administration of GAP.

4. The application of ICP-MS in biomedical analysis

With the continuing developments of new techniques and applications, inorganic MS has stretched into life science, and combining the organic MS to solve some advancing front topics. It appears that ICP-MS has opened a new door to biomedical research. The emerging interdisciplinary research has the potential to become a new bridge between inorganic MS and the life sciences for improving health. In this part, we shall depict some application of ICP-MS and relative coupling techniques in biomedical analysis.

4.1. Analysis involved in DNA

Initiation of cancer growth has been associated with modification of the common nucleobases in DNA and changes in DNA structure which can be caused by chemical modification such as styrene oxide. To gain deeper insight into the induction of cancer, detection and quantification of DNA adducts formed by carcinogenic substances is essential. Several methods for detection of DNA adducts including immunoassays, mass spectrometry, and ^{32}P-post-labelling assays, based on the synthetic reference materials which are not often available or easily made, have been developed in the last two decades. However, identification of unknown modifications is not possible. It is also questionable whether these techniques really includes all adducts quantitatively accurately because of the different response of DNA adducts, especially with the technique ^{32}P-post-labelling assays. A solution to this problem could be found only if a common feature in all nucleotides regardless of their structure could be used, which would allow a sensitive and quantitative determination based on an internal standard, e.g. LC–ICP-MS was employed to determine quantitatively the content of modified nucleotides in standard solutions based on the signal of phosphorus; phosphoric acid served as an internal standard [39]. Such a feature could be the natural phosphorus in nucleotides with a detector specific for that element. The technique of choice is ICP-MS, since it has structure-independent sensitivity and element speciation-independent in detection capability. This technique also can be applied in the separation of alkylated nucleotides from the large excess of native nucleotides.

Edler et al. [40] had quantification of the adducts in the DNA sample, with an internal standard bis(4-nitrophenyl)phosphate (BNPP) by means of the phosphorus signal measured at mass $m/z = 31$ with ICP-MS. It is important to realize that it is not necessary at this stage to know the structures of the DNA adducts, since the measurement is based on the atomic signal of phosphorus. The absolute limit of detection of 45 fmol corresponds to the detection of three modified nucleotides among 10^7 native nucleotides.

It is generally accepted that the toxicity of cisplatin is primarily the consequence of its capacity to bind genomic DNA. Nevertheless, Amran et al. [41] have demonstrated that cisplatin causes intracellular oxidation, which may also be important for apoptosis. For these reasons, experiments [41] were carried out to measure DNA platination, as an indication of cisplatin-DNA binding, with FI-ICP-MS.

4.2. Analysis related to protein

The use of ICP-MS in the biomedical arena has grown enormously in recent years and will continue to do so, particularly in the area of metal concentration determinations in protein samples for the proteomics field [42].

A similar simultaneous detection approach was utilized on bradykinin metabolism in human and rat plasma [43,44]. In these studies, the bradykinin was bromine-labelled to enable detection and quantification by ICP-MS. A further enhancement of this approach was afforded by Axelsson et al. [45] who combined ICP-MS with accurate mass measurement of their organic pharmaceutical compounds.

Metals play an important and essential role as cofactors of proteins in biological systems. The absence or a deficit of essential metals (such as Fe, Cu, Se, Zn) in proteins results in deficiency diseases, but these metals can also catalyze cytotoxic reactions. The investigation of metal-containing proteins is a new and challenging task in the proteomics field including the protein identification and the determination of the metal concentration, which requires sensitive analytical techniques and powerful equipment. Apart from the determination of phosphorus, the quantitative determination of zinc, copper and iron in brain proteins is of special interest for studying neurodegenerative diseases, such as Alzheimer's and Parkinson's disease. FI-ICP-MS has been successfully applied to the selective analysis of Alzheimer's plaque core for the first time by Beauchemin and Kisilevsky [46].

LA-ICP-SFMS also represents a powerful tool for the detection of metal-containing proteins in Alzheimer-diseased brain. Becker et al. [47] had done some researches on the metal-

containing proteins (especially with copper, iron and zinc ions) in Alzheimer brain protein sample after separation by 2D gel electrophoresis via tracer experiments with laser ablation ICP-MS (LA-ICP-MS). In their study, the main advantage of the screening procedure by LA-ICP-MS is that the time required for the structure analysis of all proteins can be reduced significantly by a pre-selection of protein spots containing metals of interest. A qualitative survey analysis several protein spots from Alzheimer-diseased brain containing Cu, Zn and Fe were analyzed with respect to $^{54}Fe/^{56}Fe$, $^{65}Cu/^{63}Cu$ and $^{67}Zn/^{64}Zn$ isotope ratios.

The liver is an important organ that performs many metabolic functions. Wang et al. [48] offered a method to identify the molecular weight fractions that contain particular elements of interest in aqueous extracts from liver with SEC-ICP-MS which provided very high sensitivity. They removed polyatomic interferences for some difficult elements like Fe, S, and P through measuring at medium spectral resolution.

At elevated concentrations, trace elements may induce toxic effects, hence there is a need to monitor and assess heavy metal status, particularly in occupationally exposed individuals and for patients receiving metallodrug therapy. Neilsen et al. [49] reported that laser ablation ICP-MS in combination with gel electrophoresis provides a novel route for the identification, quantitation and distributions of metal binding proteins in serum. With CE-ICP-MS, these studies including confirmation of a specific affinity of cisplatin and novel Pt complexes to HSA, measurement of the kinetics of binding reactions, and determination of the number of drug molecules attached to the protein were successfully done by Timerbaev et al. [50].

Reversible phosphorylation of proteins at Ser, Thr, and Tyr residues is probably the functionally most important covalent modification of proteins. The standard technology for investigating protein phosphorylation is based on incorporation of ^{32}P or ^{33}P from phosphate or an activated phosphate ester such as ATP, respectively. Despite high sensitivity and reliability, this technology has some inherent drawbacks. Wind et al. [51] introduced a new analytical dimension, CapLC-ICP-MS, a new, robust, and specific method in phosphoproteomics, in the analysis of protein phosphorylation. The method was demonstrated for the analysis of a complex mixture of synthetic phosphopeptides and a set of tryptic digests of three phosphoproteins with ^{31}P detection. These include β-casein, activated human MAP kinase ERK1, and protein kinase A catalytic subunit. The detection limit achieved for the CapLC-ICP-MS runs is 0.1 pmol of phosphopeptide injected.

4.3. Analysis of elements related to human health

It is known that while many elements are considered essential to human health, many others can be toxic. However, because the intake, accumulation, transport, storage and interaction of these different metals and metalloids in nature are strongly influenced by their specific elemental form, complete characterization of the element is essential when assessing its benefits and/or risk. Consequently, interest has grown rapidly in determining oxidation state, chemical ligand association, and complex forms of a many different elements.

ICP-MS has become one of the most popular techniques for elemental speciation studies. Fosset et al. [52] used this powerful technique as a tool to measure the uptake of Cu with natural isotopes in HepG2 cells, a liver cell line used extensively to study Cu metabolism. And the $^{63}Cu/^{65}Cu$ ratio can be measured accurately.

Metabolites of many trace elements are present in body fluids and are excreted in urine. These metabolites give clues as to the biological function of the trace elements and may help to assess their toxicity or benefit to human health. Therefore, speciation of these samples has gained interest. ICP-MS analysis of urine, in particular, has become wide spread [53–58].

Sanz-Medel et al. [59] reviewed the metallo-complexes separations, including coordination complexes of metals with larger proteins (e.g. in serum, breast milk, etc.) and metallothioneins (e.g. in cytosols from animals and plants) as well as selenoproteins (e.g. in nutritional supplements), DNA–cisplatin adducts and metal/semimetal binding to carbohydrates, using size-exclusion (SEC), ion-exchange (IE), reverse phase chromatography (RP) and capillary electrophoresis (CE).

Studies on the iron isotopic composition of human blood and liver had been carried out with ICP-MS by Walczyk and Blanckenburg [60]. They evaluated the new data for body tissues which show that blood and muscle tissue have a similar iron isotopic composition while heavier iron isotopes are concentrated in the liver. This phenomenon, known as mass-dependent isotope fractionation, has been known since decades for the lighter elements such as hydrogen, carbon, oxygen, and nitrogen. Bohn et al. [61] and Skulan et al. [62] had also undertaken the same researches to magnesium in the human body and calcium in higher organisms separately.

Trace elements can interact with each other in vivo. This may affect the absorption, metabolism, or utilization of the elements. A better understanding of element interactions could increase the ability to predict susceptibility to trace element toxicity, or to anticipate the development of deficiency/excess. ICP-MS constitutes an advantageous technique, which enables simultaneous multi-element analysis, thus facilitating studies of the relations between element concentrations. Barany et al. [63] presented a study to explore the correlations between different trace elements, both toxic and essential, with ICP-MS in a cohort of adolescents. They found a large number of correlations between 13 trace elements (Co, Cu, Zn, Se, Rb, Rh, Pd, Cd, W, Pt, Hg, Tl, and Pb) in human blood and/or serum, by investigating in 372 Swedish adolescents. Notably, serum Se correlated with blood Pb and blood Hg and Cu and Zn were correlated to each other in both blood and serum.

It is known that arsenic, similar with Hg, Cd and Se, has different toxicological properties dependent upon both its oxidation state for inorganic compounds, as well as the different toxicity levels exhibited for organic arsenic compounds. The field of arsenic speciation analysis has grown rapidly in recent years, especially with the utilization of ICP-MS, a highly sensitive and robust detector system, which had been applied for speciation analysis of Hg, Cd and Se

coupling with HPLC [64–69]. Dopp et al. [70] had taken a research to investigate the genotoxic effects and the cellular uptake of inorganic arsenic [arsenate, As(V); arsenite, As(III)] and the methylated arsenic species monomethylarsonic acid [MMA(V)], monomethylarsonous acid [MMA(III)], dimethylarsinic acid [DMA(V)], dimethylarsinous acid [DMA(III)], trimethylarsenic oxide [TMAO(V)] in Chinese Hamster ovary (CHO-9) cells. Intracellular arsenic concentrations were determined by ICP-MS. Their results showed that MMA(III) and DMA(III) induced cytotoxic and genotoxic effects to a greater extent than MMA(V) or DMA(V). The uptake of the chemicals was also measured by ICP-MS, and they found that only 0.03% MMA(V) and DMA(V), and 2% MMA(III), As(III) and (V) were taken up by the cells.

5. Brief solutions to the problems in application of ICP-MS

As a new analytical technique, ICP-MS has shown its extraordinary advantages compared with other analytical methods in every aspect. However, the existing problems, such as signal fluctuation, matrix effect, double electric charge ion (because of some elements which have the low ionization energy, but it is a scarce type of interference evoked by these elements), oxides, polyatomic ion (a type of frequent interference), isobar and memory effect, are so widespread in application that every analyst should take it seriously.

It is an effective method to revise the signal fluctuation and matrix effect with the internal standard. The elements with the close mass number will suffer the similar influence of signal fluctuation and matrix effect [71–73]. Thus, it has become a regulation that the mass number of internal standard should be close to the analytes for the selection of internal standard. Accordingly, it will be better to take a double-internal standard rather than single-internal standard in multi-element analysis. Matrix effect which maybe is the most severe interference can also be eliminated with the matching standard of matrix, isotope dilution (ID), or some more simple methods such as specimen dilution and separating the element from the matrix before analysis. Interference from double electric charge ion is so slight that assayers can ignore it unless there are elements with low secondary ionization energy, such as Ba and Ce. The oxides rarely exceed the limit of 5% under normal operating conditions, and this level is acceptable in routine analyses. It is not difficult to control the oxides under the suitable level, including elevating the temperature of plasma, slowing the rate of spraying gas flow and ensuring the area of forming oxides close to the interface. Interference of the polyatomic ion is frequent [74,75]. Regulating the resolution which is mentioned in this review [29] is a common means for resolving this question. Another problem caused by the isobar [76] is inevitable during quantitative analysis via detecting the mass-to-charge ratio. However, since the existence of isotope, the analyte can be determined via detecting another isotope which is not interfered by isobar or eliminating the interference caused by isobar by detecting isotope of isobar. Memory effect [77–81] which can be solved by selecting proper plasma operating conditions such as the rate of spraying gas flow and concentration of matrix is not a significant problem. It will also be decreased by diminishing the concentration of reference to a suitable level, e.g. the same as the analyte.

In fact, the interference will not exceed the acceptable limits under the normal operating conditions, and there will be more accumulated experience which is valuable for the routine analyses for assayers during the application.

6. Conclusions

ICP-MS has the required power to provide ultra-trace elemental detection, which also allows elucidating temporally overlapping chromatographic peaks. With high sensitivities, low detection limits, mass (elemental) selectivity, isotope ratio capabilities, and wide dynamic ranges, ICP-MS is not only the best detector in typical speciation analysis, but also in, for example, drug (especially during the drug development and quantitative analysis of metabolites), protein and gene research at this time. Further studies will be hopefully established if it is possible to use inorganic elemental standards or small organic compounds in the quantitative determination of all kinds of drugs, nucleotides and sulfur or phosphorus containing peptides and proteins at appropriate concentration with an acceptable accuracy. With the help of ICP-MS, the struggle of searching for an excellent quantification technique in, e.g. drugs and proteomics have come appreciably close to an end. In addition, combining the organic MS [29] during the application is another orientation of development for ICP-MS. In a word, ICP-MS will play a more and more important role in pharmaceutical and biomedical analysis.

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