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Pharmaceutical

Analysis of elemental impurities in pharmaceutical products in accordance with USP General Chapters <232> and <233>

Authors

Bhagyesh Surekar, Daniel Kutscher

Thermo Fisher Scientific,
Bremen, Germany

Keywords

USP 232/233, impurity analysis, ICP-MS, ICH Q3-D, KED, system suitability, risk assessment

Goal

To develop an analytical method using single quadrupole ICP-MS for the determination of 24 analytes in pharmaceutical products and assess the performance of the method in accordance with the requirements of USP General Chapters <232> and <233> and ICH Guideline Q3-D (R1)

Introduction

Analysis of pharmaceutical products for elemental impurities is critical due to their toxicological effects on human health. The presence of certain impurities could also have adverse effects on the stability and shelf-life of pharmaceutical products. Therefore, monitoring and control of elemental impurities at every stage of drug product manufacturing is very important. The analysis of elemental impurities is governed by the United States Pharmacopeia (USP) General Chapters <232> and <233>, first introduced on January 1, 2018. The new chapters replaced USP General Chapter <231>, originally introduced in 1905, as severe limitations regarding accuracy and reproducibility became obvious. With the introduction of USP <233>, USP advises use of modern and advanced analytical instrumentation such as inductively coupled plasma – optical emission spectroscopy (ICP-OES) or inductively coupled plasma – mass spectrometry (ICP-MS) instead of the non-specific colorimetric method suggested by USP <231>.

Considering the significantly lower limit concentration requirements for critically important analytes, such as Pb, As, Cd, and Hg, and mandatory testing of these impurities in every drug product, ICP-MS is the best suited and widely preferred technique due to its superior sensitivity at trace levels (µg·kg^{-1} and lower). ICP-MS also typically offers a wide dynamic range and overcomes the need to use specific accessories, such as hydride generation, for the analysis of critical contaminants like arsenic or mercury, among others.

In practice, USP <232> provides guidelines on the limits of elemental contaminants, whereas USP <233> suggests analytical procedures for their determination, as well as a set of parameters for method validation. In order to harmonize the determination of elemental impurities globally, the International Council of Harmonization (ICH) introduced a new guideline (ICH Q3-D) to provide guidelines for the entire process including risk assessment and limits.

USP <233> suggests two options to assess the level of impurities in a given product. The final product can be analyzed as a whole, or each of its components can be analyzed, and the overall level of impurities is calculated based on the formulation. Whereas the first option also covers impurities accumulated during the manufacturing process, the latter option is a more flexible approach, as the results of some components can be used for multiple formulations. This necessary control measure leads to analysis of different components of finished pharmaceutical products, such as active pharmaceutical ingredients (API), raw materials, intermediates, and the finished product itself for elemental impurities.

Apart from the analytical requirements described in the USP chapters (including those listed in General Chapter <730> for plasma spectrochemistry), laboratory managers and operators need to ensure that all data created is genuine and authentic, as per the requirements of the US Food and Drug Administration's (FDA) 21 CFR Part 11 regulations regarding electronic records and validation of electronic signatures. To assess traceability and authenticity of generated analytical data, control software should have effective tools including user authentication, user access management, provision for automatic data back-up, time and date stamps for all electronic records, audit trail of every user action, and capability to sign analytical data electronically.

This note describes an analytical method and overall workflow with the Thermo Scientific™ iCAP™ RQ ICP-MS for accurate, precise, and sensitive quantification of elemental impurities in pharmaceutical products. The method was developed to quantify 24 elements in accordance with the USP chapters in a single analytical measurement.

The Thermo Scientific™ Qtegra™ Intelligent Scientific Data Solution™ (ISDS) Software was used to control the ICP-MS instrument and to generate, process, and report analytical data following an entire workflow that meets the requirements described in 21 CFR Part 11.

Experimental

Instrument parameters and experimental conditions

An iCAP RQ ICP-MS was used in this study. To allow for unattended operation, the system was operated in conjunction with a Teledyne CETAC ASX-560 autosampler (Teledyne CETAC Technologies, Omaha, NE, USA). The sample introduction system was configured for the analysis of aqueous solutions and consisted of a glass concentric nebulizer (400 µL·min^{-1}), cyclonic spray chamber, 2.5 mm quartz injector, Ni-tipped sample and skimmer cones, and a quartz torch. The iCAP RQ ICP-MS was operated in KED mode, using pure helium as the only collision cell gas to remove potential polyatomic interferences on various analytes. Details of the components and typical instrument parameters used during this study are summarized in Table 1.

Table 1. Instrument configuration and typical operating parameters

Parameter	Value
Nebulizer	Glass concentric nebulizer, 400 µL·min^{-1}
Spray chamber	Cyclonic quartz
Torch	Quartz
Injector	Quartz, 2.5 mm i.d.
RF power	1,550 W
Cool gas flow	14 L·min^{-1}
Auxiliary gas flow	0.8 L·min^{-1}
Interface cones	Ni-tipped sample and skimmer
Skimmer cone insert	High matrix
Nebulizer flow	1.16 L·min^{-1}
KED flow rate (Helium)	4.5 mL·min^{-1}
Number of replicates	3
Number of sweeps	10
Dwell time	0.05 s

Prior to analysis, instrument performance was verified using the automated performance check available within the Qtegra ISDS Software. In this test, the sensitivity across the mass range is checked for ^{7}Li, ^{59}Co, ^{115}In, and ^{238}U. Other plasma-related performance parameters, such as oxide formation and doubly charged ion formation rates, were also checked using the $^{140}Ce^{16}O^{+}/^{140}Ce^{+}$ and $^{137}Ba^{++}/^{137}Ba^{+}$ ratios, respectively.

Standard and sample preparation

The sample used in this study is an active pharmaceutical ingredient (API) that is used in a drug product that is administered orally. To determine the concentration limits for all 24 elements, the maximum permitted daily exposures (PDEs) of elemental impurities for oral drugs given in USP <232> were used. The concentration limits were then calculated based on the PDEs and maximum daily dose of 10 g as suggested by the USP guideline using the equation given below.

$$\text{Limit concentration (µg/g)} = \frac{\text{Permitted daily exposure (µg/day)}}{\text{Max daily dose (g/day)}}$$

The sample was prepared for analysis by simple dissolution of an accurately weighed 0.1 g aliquot in 20 mL of 1% (*v/v*) nitric acid, corresponding to a total dilution factor of 200 during sample preparation (or 0.5% *w/v* sample concentration). To monitor and compensate for physical interferences during analysis, all solutions including blanks, standards, and samples were spiked with an internal standard solution containing Be, Sc, Y, Tb, and Bi. Three different batches of the same API were prepared and analyzed in the experiment.

The linearity for all analytes was determined in the range of 5 to 200% of the previously established limit concentration (J) in the prepared sample solutions. The corresponding J value for each analyte was calculated using the following equation:

$$\text{J (µg/kg)} = \frac{\text{(Limit concentration (µg/g))}}{\text{(Total dilution factor)}} \times 1{,}000$$

Six individual calibration solutions were prepared by gravimetric dilution from an intermediate stock standard solution (with a concentration of 25J), covering the range of 0.05J to 2J.

Details of the linearity standards and concentrations of the different analytes are given in Table 2.

As it is a common requirement in the industry to generate a report of the concentrations used for the determination of linearity, the reporting tool inside the Qtegra ISDS Software allows generation of a comprehensive and easily accessible report of the concentrations used. Although USP <233> suggests that linearity establishment in the range of 0.5J and 1.5J for each analyte is generally sufficient, method performance was assessed under more demanding conditions. For the analysis of a series of individual components, potentially being used in multiple formulations, a sensitive method with an extended linear range is generally more suitable. This analytical method offers flexibility to perform reliable analysis of pharmaceutical products with a maximum daily dose of 10 g or more per day and it also provides a comprehensive tool for risk assessment of process related components such as water.

Table 2. List of target elements and their concentrations in linearity standards (µg·kg^{-1})

Analyte	0.05J	0.1J	0.2J	0.5J	1J	1.5J	2J
Li	13.75	27.5	55	137.5	275	412.5	550
V	2.5	5	10	25	50	75	100
Cr	275	550	1,100	2,750	5,500	8,250	11,000
Co	1.25	2.5	5	12.5	25	37.5	50
Ni	5	10	20	50	100	150	200
Cu	75	150	300	750	1,500	2,250	3,000
As	0.375	0.75	1.5	3.75	7.5	11.25	15
Se	3.75	7.5	15	37.5	75	112.5	150
Mo	75	150	300	750	1,500	2,250	3,000
Ru	2.5	5	10	25	50	75	100
Rh	2.5	5	10	25	50	75	100
Pd	2.5	5	10	25	50	75	100
Ag	3.75	7.5	15	37.5	75	112.5	150
Cd	0.125	0.25	0.5	1.25	2.5	3.75	5
Sn	150	300	600	1,500	3,000	4,500	6,000
Sb	30	60	120	300	600	900	1,200
Ba	35	70	140	350	700	1,050	1,400
Os	2.5	5	10	25	50	75	100
Ir	2.5	5	10	25	50	75	100
Pt	2.5	5	10	25	50	75	100
Au	2.5	5	10	25	50	75	100
Hg	0.75	1.5	3	7.5	15	22.5	30
Tl	0.2	0.4	0.8	2	4	6	8
Pb	0.125	0.25	0.5	1.25	2.5	3.75	5

System suitability – correlation coefficient and instrument detection limits

The correlation coefficient is an important figure of merit and often considered as a system suitability criterion in many regulated methods. As per the guideline given in USP <730> on plasma spectrochemistry, the correlation coefficient (R) should not be below 0.99, which needs to be confirmed and recorded appropriately before proceeding with sample analysis. Details of analytes, *m/z* (i.e., ion mass) ratio used, obtained correlation coefficient, and achieved instrument detection limits are summarized in Table 3. The obtained correlation coefficients for all analytes are well above 0.998 for the concentration range investigated in this study, indicating fulfillment of this system suitability criterion.

Table 3. List of analytes, *m/z*, correlation coefficients, and instrumental detection limits (all results expressed as µg·kg^{-1})

Element	*m/z*	R	IDL
Li	7	>0.9999	0.0702
V	51	>0.9999	0.0005
Cr	52	0.9998	0.0044
Co	59	>0.9999	0.0003
Ni	60	0.9998	0.0042
Cu	63	0.9994	0.0122
As	75	0.9998	0.0007
Se	77	0.9986	0.7793
Mo	95	0.9999	0.0080
Ru	101	>0.9999	0.0045
Rh	103	0.9997	0.0002
Pd	105	0.9999	0.0036
Ag	107	0.9997	0.0003
Cd	111	0.9998	0.0006
Sn	118	0.9998	0.0832
Sb	121	0.9997	0.0027
Ba	137	0.9999	0.0081
Os	189	0.9987	0.0040
Ir	193	0.9999	0.0001
Pt	195	0.9997	0.0005
Au	197	0.9996	0.0017
Hg	202	0.9999	0.0053
Tl	205	>0.9999	0.0007
Pb	208	>0.9999	0.0006

Sample analysis

The analysis of three different batches of API was performed after confirmation of the system suitability criteria as mentioned above. The determined levels of impurities in all analyzed samples were found to be below the method quantification limit (MQL) for all analytes. Method quantification limits were calculated based on the targeted instrumental limit of quantification (0.1J) and total dilution factor used in the sample preparation.

In this study, the lowest concentration tested to demonstrate the accuracy was 0.1J, or 10% of the concentration limit for each analyte. Though the instrument detection limits (Table 3) achieved for all analytes are significantly lower, suggesting that significantly lower analyte concentrations can be determined with the required degree of accuracy and precision, method quantification limits presented here are based on the targeted limit of quantification (LOQ) of 0.1J. Method quantification limits (MQL) are calculated following the equation below:

Method quantification limit = Instrument quantification limit (0.1J) × total dilution factor

Results of the analysis obtained for all three samples are presented in Table 4. The apparent concentrations of all analytes in all analyzed samples were found to be below the method quantification limit of the respective analytes.

Table 4. List of analytes, apparent concentrations, and MQL (all results expressed as mg·kg^{-1})

Element	Batch 1	Batch 2	Batch 3	MQL
Li	< MQL	< MQL	< MQL	5.5
V	< MQL	< MQL	< MQL	1.0
Cr	< MQL	< MQL	< MQL	110
Co	< MQL	< MQL	< MQL	0.5
Ni	< MQL	< MQL	< MQL	2
Cu	< MQL	< MQL	< MQL	30
As	< MQL	< MQL	< MQL	0.15
Se	< MQL	< MQL	< MQL	1.5
Mo	< MQL	< MQL	< MQL	30
Ru	< MQL	< MQL	< MQL	1
Rh	< MQL	< MQL	< MQL	1
Pd	< MQL	< MQL	< MQL	1
Ag	< MQL	< MQL	< MQL	1.5
Cd	< MQL	< MQL	< MQL	0.05
Sn	< MQL	< MQL	< MQL	60
Sb	< MQL	< MQL	< MQL	12
Ba	< MQL	< MQL	< MQL	14
Os	< MQL	< MQL	< MQL	1
Ir	< MQL	< MQL	< MQL	1
Pt	< MQL	< MQL	< MQL	1
Au	< MQL	< MQL	< MQL	1
Hg	< MQL	< MQL	< MQL	0.3
Tl	< MQL	< MQL	< MQL	0.08
Pb	< MQL	< MQL	< MQL	0.05

Method accuracy

As mentioned previously, despite a full validation, a test for the accuracy of the method may be repeated in each analysis to ensure that the developed method is suitable for its intended purpose of impurity analysis in a specific sample material. To assess the method performance in terms of accuracy, sample 'Batch 1' was spiked with an appropriate volume of stock standard solution before solubilization. The sample was spiked at three different concentration levels, equating to 0.1J, 1J, and 2J, using the same standard stock solution used for the preparation of the working standards. Samples were prepared in triplicate for each level of spiked concentrations. Accuracy of each measured sample was then calculated based on the spiked concentration and concentrations observed in spiked and unspiked samples.

Accuracy data obtained for all spiked samples were found to be in the range of 90 to 110%, which is well within the acceptance criteria of 70–150% specified in USP <233>.

Qtegra ISDS Software provides comprehensive calculation algorithms and reporting functionality for automatic calculation and reporting of results in the format required by the user, reducing manual efforts for data export and potential further processing. It also helps minimize potential systematic errors that could occur and affect data quality in the process. Figures 1, 2, and 3 represent the results obtained while testing the accuracy for spiked levels of 0.1J, 1J, and 2J, respectively. Reported accuracy values correspond to the mean calculated from three independently prepared spiked samples at each concentration level.

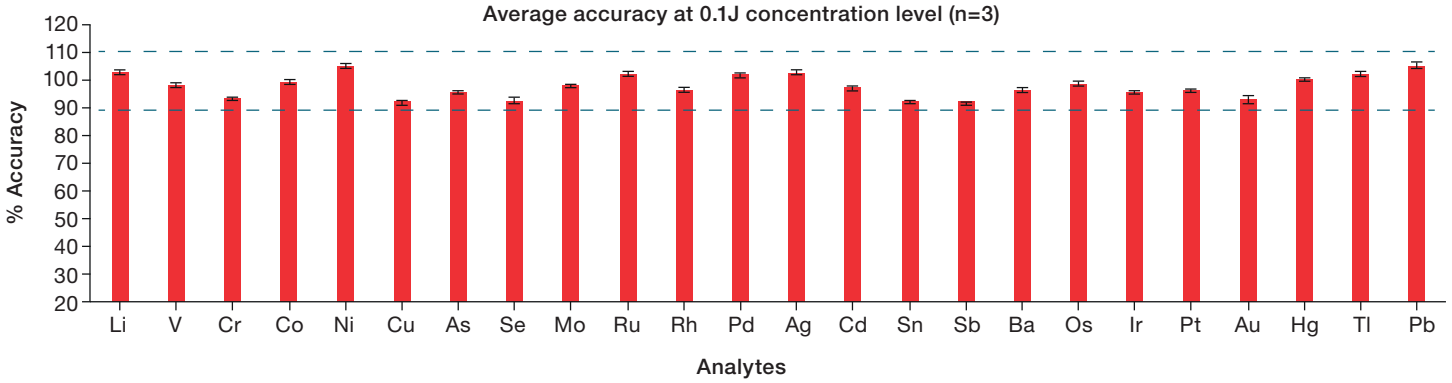

Figure 1. Average percent accuracy at 0.1J spiked concentration

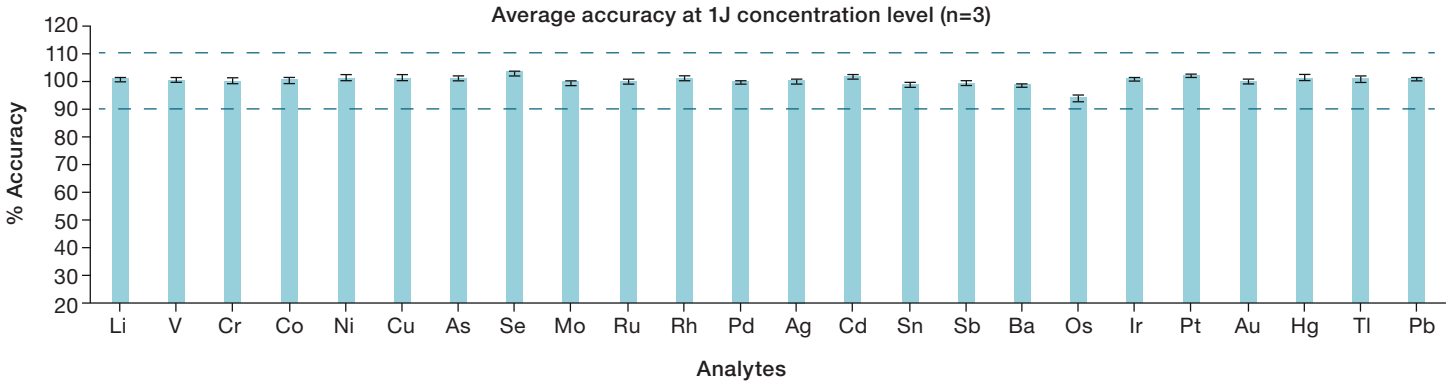

Figure 2. Average percent accuracy at 1J spiked concentration

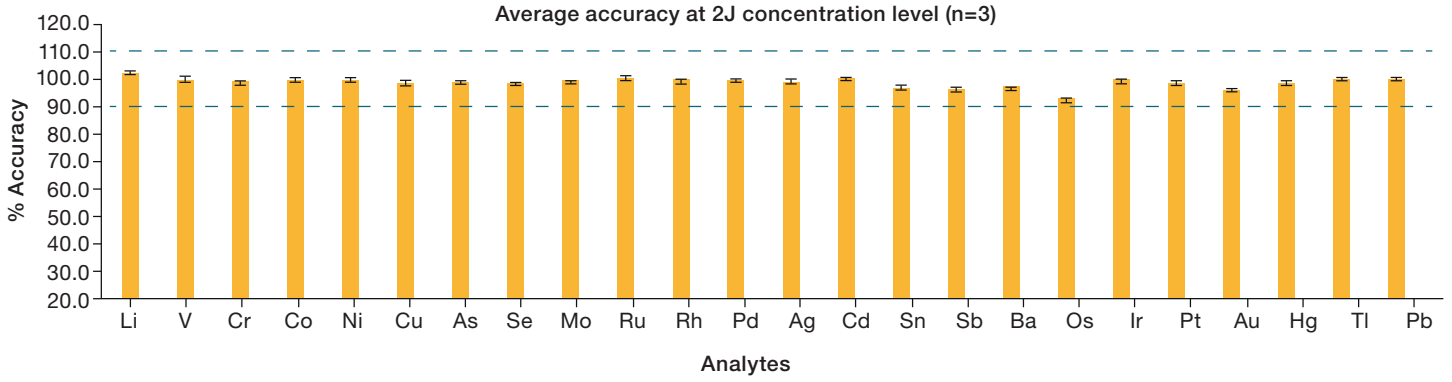

Figure 3. Average percent accuracy at 2J spiked concentration

Figure 4 highlights how the reporting functionality in Qtegra ISDS Software can help to directly assess the results of the accuracy test for a series of samples in a measurement. In this example, results from three independently prepared and measured samples spiked at 2J level are pulled together to report mean values with statistical information, including standard deviation (SD) and percent relative standard deviation (% RSD), for the Class-1 contaminants (arsenic, cadmium, mercury, and lead).

System suitability – drift

Another requirement to be covered in each analysis as per USP <233> is to ensure that the initial calibration obtained before sample analysis is still valid at the end of the sequence and that no drift may have affected the instrument´s response, and hence the results obtained. Therefore, a verification of the instrument´s response to a standard containing all impurities at a level of 1.5J was determined after the calibration of the system and at the end of the analysis. These results should not read back beyond the set limit of 20%. The results obtained in both measurements were compared to determine the percent variation for each analyte and were found to be well within the acceptance criteria of 20%, indicating that this system suitability requirement was fulfilled. Table 5 presents the percent accuracy of the 1.5J standard solution measured before and after analysis for each analyte and the percent drift between the two measurements.

Table 5. List of analytes, percent accuracy observed in the 1.5J standard measured before and after the sample analysis, and percent drift calculated between two measurements

Analyte	% Accuracy against original concentration		% Drift
	Before sample analysis	After sample analysis	
Li	98.9	102.8	3.9
V	98.4	99.2	0.8
Cr	98	98.8	0.8
Co	99.1	99.6	0.5
Ni	99.8	100.6	0.8
Cu	99.4	100.4	1.0
As	101.2	101.8	0.6
Se	103.6	102.9	0.7
Mo	99.3	100.4	1.1
Ru	99.9	101.9	2
Rh	100.7	102.0	1.3
Pd	100.7	103.0	2.3
Ag	101.3	102.7	1.4
Cd	102.9	104.5	1.6
Sn	97.8	96.0	1.8
Sb	97.6	96.4	1.2
Ba	98.2	96.7	1.5
Os	100.6	96.4	4.2
Ir	100.5	101.1	0.6
Pt	101.1	101.8	0.7
Au	98.9	100.8	1.9
Hg	102.4	102.5	0.1
Tl	102.3	102.7	0.4
Pb	101.6	101.8	0.2

Accuracy & Precision at 2J

Index	Label	75As (KED)	111Cd (KED)	202Hg (KED)	208Pb (KED)
23	Accuracy_2J	99.7 %	100.9 %	98.4 %	100.8 %
24	Accuracy_2J	100.0 %	100.7 %	98.6 %	100.2 %
25	Accuracy_2J	99.0 %	100.6 %	99.9 %	101.0 %
	Average Recovery (%)	99.6	100.7	99.0	100.7
	SD	0.5	0.2	0.8	0.4
	RSD (%)	0.5	0.2	0.8	0.4

Figure 4. Specimen report for accuracy and precision from Qtegra ISDS Software. Results are shown for Class-1 elements, data for remaining analytes can be reported in the same or individual tables.

Summary

This study highlights the use of the iCAP RQ ICP-MS for the routine determination of elemental impurities in pharmaceutical products, both finished as well as single components, tested for levels of impurities as part of the quality control process. Although the method used was previously validated, regular checks and system suitability tests need to be performed as part of each analysis of a product or a component. Accurate and precise analysis is easily possible using the proposed instrumental set up. The most important conclusions are summarized below:

- The iCAP RQ ICP-MS enables a single mode KED approach, using helium as the cell gas to analyze the entire set of specified analytes free from all interferences. This approach eliminates the need for method development or mode switching for specific group of elements. It also helps reduce the cost of analysis significantly, while improving productivity of the analytical laboratory.
- The instrument detection limits achieved for all 24 analytes indicate that the iCAP RQ ICP-MS provides the required sensitivity, not only to fulfil current regulatory needs, but also to meet any future requirements.
- The accuracy and precision obtained at different spiked concentration levels indicates that the developed method can be used routinely for accurate, precise, and reliable quantification of trace as well as relatively higher concentrations of analytes.
- The outcome of the system suitability study regarding signal drift evaluation suggests that the iCAP RQ ICP-MS delivers stable and consistent performance, thereby minimizing the need for re-calibration and reanalysis of analytical batches.
- The Qtegra ISDS Software used for instrument control, data acquisition, data processing, and data reporting is equipped with a powerful toolset to ensure reliable instrument operation and simple data handling, fully meeting the requirements given in 21 CFR Part 11.
- The quality control (QC) and reporting functionality of Qtegra ISDS Software provide comprehensive features for automatic and errorless data reporting.

References

1. USP General Chapter <232>
2. USP General Chapter <233>
3. ICH Guideline Q3-D (R1)
4. USP General Chapter <730> Plasma Spectrochemistry

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