Elemental Profiling of Whiskey using the Agilent 5100/5110 ICP-OES and MPP Chemometrics Software

Food authenticity

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Introduction

Whiskey production is a lucrative global industry that generates billions of dollars of business every year. There are over 20 whiskey producing countries, with Scotland leading the market with Scotch whisky, followed by the US, Canada, Ireland and Japan. Out of the 200+ countries that have developed a taste for whiskey, India consumes the most – more than three times as much as the US. Unsurprisingly, India is beginning to increase its own production of the spirit [1].

With the value of a whiskey highly dependent on type, brand and heritage, quality, age, and legal product definition, producers are keen to establish analytical methods to help them identify the unique aspects of their product and ways to preserve its authenticity against fraudulent practices.

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Elemental profiling of wines and spirits using an atomic spectroscopy analytical technique is widely used to discriminate between different foods and beverages, as the elements present in the product will vary depending on geography, raw materials, production methods, storage etc.

Due to its high sensitivity and wide multi-element coverage, ICP-MS has been used successfully to profile the elemental composition of other alcoholic beverages such as wine [2] and to differentiate wines due to geographical origin as well as processing site [3, 4]. Microwave Plasma-Atomic Emission Spectroscopy (MP-AES) has also been used to profile wine using a few target elements [5].

In this study, the suitability of the Agilent 5100 Synchronous Vertical Dual View (SVDV) ICP-OES to profile six different whiskey types was investigated. Agilent's Mass Profiler Professional (MPP) software was also utilized to analyze the results. MPP enables the user to display the data in different ways, making it easier to compare and interpret the results. Data provided by Agilent's 5100/5110 ICP-OES series instruments is suitable for use with MPP. This application is also applicable to Agilent's 5110 ICP-OES.

Experimental

Instrumentation

All measurements were carried out using the Agilent 5100 SVDV ICP-OES equipped with a Dichroic Spectral Combiner (DSC). The DSC allows both axial and radial view emissions from the plasma to be measured at the same time, in a single reading, over the entire wavelength range. The 5100 ICP-OES uses a vertically orientated torch and a solid-state RF (SSRF) system operating at 27 MHz to deliver a plasma with the stability and robustness necessary for the analysis of organic samples. To maintain full user flexibility, the 5100 SVDV ICP-OES can also be operated in Vertical Dual View (VDV) mode, dedicated Radial View (RV) and dedicated Axial View (AV). The latter mode was selected in this study as only trace level elements were of interest.

The Agilent 5100 SVDV ICP-OES was fitted with a standard sample introduction system comprising a glass concentric nebulizer, 1.8 mm torch injector and a glass, single-pass cyclonic spray chamber. Sample delivery was via an Agilent SPS 3 autosampler. Instrument operating conditions are listed in Table 1.

Table 1. Agilent 5100 ICP-OES operating parameters

Parameter	Setting
RF power (kW)	1.20
Aux gas flow (L/min)	1.00
Plasma flow (L/min)	12.0
Nebulizer flow (L/min)	0.70
Ar/O_2 addition	none
Pump speed (rpm)	12
Uptake delay (s)	25 (Fast pump ON)
Rinse time (s)	30 (Fast pump ON)
Stabilization time (s)	15
Read time (s)	20
Number of replicates	3
Viewing mode	Axial
Sample pump tubing	Black/Black
Waste pump tubing	Blue/Blue
Background correction	Fitted

Samples and standards

Details of the sixty-nine commercial whiskey products used in this study are listed in Table 2, including 16 Bourbons, 8 Irish, 9 Japanese, 1 Rye, 33 Scotch and 2 Tennessee. All samples were prepared in triplicate, diluted 20-fold in 1% (v/v) nitric acid and 0.5% (v/v) hydrochloric acid to decrease the ethanol level to 2%.

Multi-element calibration standards (SPEX CertiPrep, Metuchen, NJ, USA) were used to prepare six-point calibration curves for all elements listed in Table 3 between 0 and 1000 µg/L. All standards were matrix-matched (1% HNO_3, 0.5% HCl, 2% ethanol, all (v/v)) to account for the sample dilution and matrix interferences. Each element was analyzed in triplicate.

Table 2. The 69 whiskey samples, including code, age (if known), and proof. Products from the same distillery are indicated. Regions indicated in brackets in the Distillery column of the table for the 33 Scotch whiskies.

Code[1]	Age[2]	Proof	Distillery	Code[1]	Age[2]	Proof	Distillery	Code[1]	Age	Proof	Distillery[3]	Code[1]	Age	Proof	Distillery[3]
B1	7	107	D1	R1	12	80	D16	S1	10	86	D17 (A)	S19	12	86	D28 (C)
B2	8	80	D1	I1	N.A.	80	D9	S2	10	92	D18 (B)	S20	12	86	D29 (D)
B3	N.A.	90	D2	I2	N.A.	80	D10	S3	12	80	D19 (C)	S21	10	80	D30 (D)
B4	N.A.	100	D2	I3	N.A.	80	D11	S4	18	86	D19 (C)	S22	12	80	D31 (A)
B5	10	90	D2	I4	8	80	D12	S5	27	116	D19 (C)	S23	18	86	D31 (A)
B6	N.A.	86.6	D2	I5	N.A.	80	D12	S6	12	92.6	D20 (B)	S24	10	80	D32 (A)
B7	N.A.	100	D3	I6	12	115	D13	S7	12	86	D21 (B)	S25	16	80	D32 (A)
B8	12	86	D1	I7	15	92	D13	S8	N.A.	88	D22 (D)	S26	16	86	D33 (B)
B9	N.A.	101	D4	I8	12	92	D13	S9	10	80	D23 (D)	S27	10	80	D34 (B)
B10	N.A.	90	D5	J1	12	86	D14	S10	10	80	D23 (D)	S28	15	86	D34 (B)
B11	9	100	D1	J2	12	86	D14	S11	12	80	D24(E)	S29	12	80	D35 (E)
B12	N.A.	114	D4	J3	N.A.	96	D15	S12	15	92	D24 (E)	S30	12	80	D36 (D)
B13	N.A.	90.2	D6	J4	N.A.	110	D15	S13	12	80	D25 (E)	S31	16	80	D37 (A)
B14	N.A.	90.4	D7	J5	10	90	D16	S14	10	80	D26 (E)	S32	15	92	D38 (F)
B15	12	90	D2	J6	12	90	D16	S15	15	92	D26 (E)	S33	10	86	D30 (D)
B16	N.A.	113	D8	J7	N.A.	80	D16	S16	21	86	D26 (E)	T1	N.A.	90	D39
				J8	17	86	D16	S17	12	80	D27 (E)	T2	N.A.	80	D40
				J9	12	80	D16	S18	15	80	D27 (E)				

Results and discussion

Calibration linearity

All elements showed excellent linearity of their calibration curves with correlation coefficients between 0.999 and 1.000. Representative calibration curves are presented in Figure 1. The plots for Cu, Mg and Zn show excellent linearity across the calibrated range, with correlation coefficients of 0.99999, 0.99995 and 1.00000, respectively.

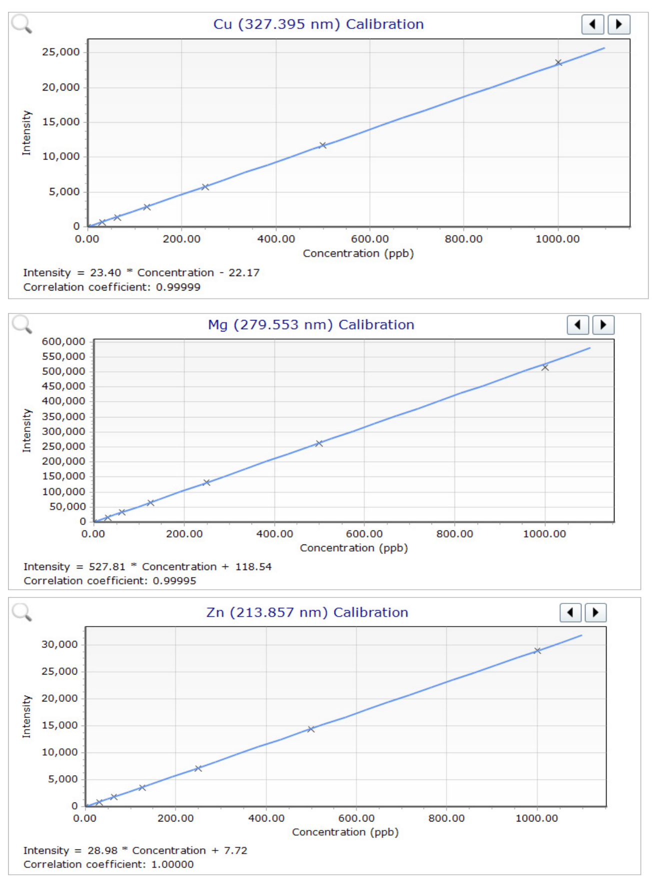

Figure 1. Calibration curves for Cu, Mg and Zn using ICP-OES

[1]Whiskeys are coded by type: B (Bourbon), I (Irish), J (Japanese), R (Rye), S (Scotch), T (Tennessee). [2]N.A =Not available. [3]Scotch regions: A (Island); B (Islay); C (Lowland); D (Highland); E (Speyside); F (Campbeltown).

Method detection limits (MDLs)

MDLs were calculated as 3 times the standard deviation of ten replicate measurements of the calibration blank using the 5100 operating in axial view mode.

The MDLs and minimum/maximum range analyzed for each element are shown in Table 3.

Table 3. Method detection limits (MDL) and Min-Max concentration (µg/L) for the 69 whiskey samples tested.

Element & wavelength (nm)	MDL (µg/L)	Min-Max (µg/L)	Element & wavelength (nm)	MDL (µg/L)	Min-Max (µg/L)
Ag 328.068	0.17	<MDL	Mn 257.610	0.04	0.74 - 203.0
Al 396.152	0.55	<DL - 1066	Mo 202.032	1.81	< MDL
As 193.696	5.37	< MDL	Na 588.995	2.29	440.09 - 25625
B 249.772	0.23	17.63 - 501.5	Ni 231.604	2.02	<DL - 0.00
Ba 455.403	0.11	1.13 - 159	Pb 220.353	1.96	<DL - 0.00
Be 313.042	0.04	< MDL	Rb 421.552	0.03	1.45 - 57.62
Ca 396.847	0.03	246.54 - 9292	Se 196.026	8.86	< MDL
Cd 214.439	0.14	19.54 - 19.54	Si 251.611	2.09	189.02 - 19253
Co 238.892	0.99	< MDL	Sr 407.771	0.02	1.85 - 56.98
Cr 267.716	0.26	3.63 - 49.21	Ti 336.122	1.31	<MDL
Cu 327.395	0.23	20.58 - 2448	Tl 190.794	3.49	<MDL
Fe 238.204	0.20	3.61 - 753.9	V 292.401	0.46	<MDL
K 766.491	4.02	3524 - 47154	Zn 213.857	0.22	6.94 - 820.4
Mg 279.553	0.02	12.58 - 5016			

QC spike recoveries

A Quality Control (QC) sample was prepared by spiking 3 whiskey samples (S27, J4 and S2), each in triplicate, with the 5 µg/L calibration standard. To check the validity of the method throughout the analytical cycle, a CCB and CCV (200 ppb) sample was analyzed every 10 samples. All mean recoveries were within ± 10% of the expected CCV value. The results are given in Table 4.

Table 4. Mean spike recoveries of 5 µg/L spiked QC sample in whiskey samples (n=3).

Element & wavelength (nm)	Spike (µg/L)	Mean recovery + 1σ (%) (n=3)	Recovery range (%)	Element & wavelength (nm)	Spike (µg/L)	Mean recovery + 1σ (%) (n=3)	Recovery range (%)
Al 396.152	5	101 ± 1	100-103	Mo 202.032	5	100 ± 2	98-101
As 193.696	5	108 ± 2	106-109	Na 588.995	5	105 ± 10	98-112
B 249.772	5	99 ± 2	98-101	Ni 231.604	5	99 ± 1	98-100
Ba 455.403	5	100 ± 1	99-102	Pb 220.353	5	98 ± 2	96-100
Be 313.042	5	108 ± 1	107-110	Rb 421.552	5	101 ± 1	100-102
Ca 396.847	5	97 ± 5	93-102	Se 196.026	5	105 ± 0	105-105
Cd 214.439	5	99 ± 1	98-100	Si 251.611	5	95 ± 5	90-101
Co 238.892	5	98 ± 1	97-99	Sr 407.771	5	100 ± 1	99-101
Cr 267.716	5	98 ± 1	97-100	Ti 336.122	5	100 ± 1	99-101
Cu 327.395	5	100 ± 2	99-103	Tl 190.794	5	92 ± 9	99-101
Fe 238.204	5	99 ± 1	97-100	V 292.401	5	101 ± 1	100-102
Mg 279.553	5	98 ± 6	91-104	Zn 213.857	5	98 ± 1	97-99
Mn 257.610	5	105 ± 0	105-105				

Data analysis using Mass Profiler Professional

All of the 69 whiskeys were analyzed and the exploratory data analysis was completed using Agilent's Mass Profiler Professional (MPP) software. The resultant box-whisker plots based on the full data set are given in Figure 2.

Slight differences in each plot can be noted. This suggests that the element profiles of whiskeys can be used to distinguish samples based on the age, type, and region of the sample. If more elements are analyzed in the future, greater separation between the samples would be apparent.

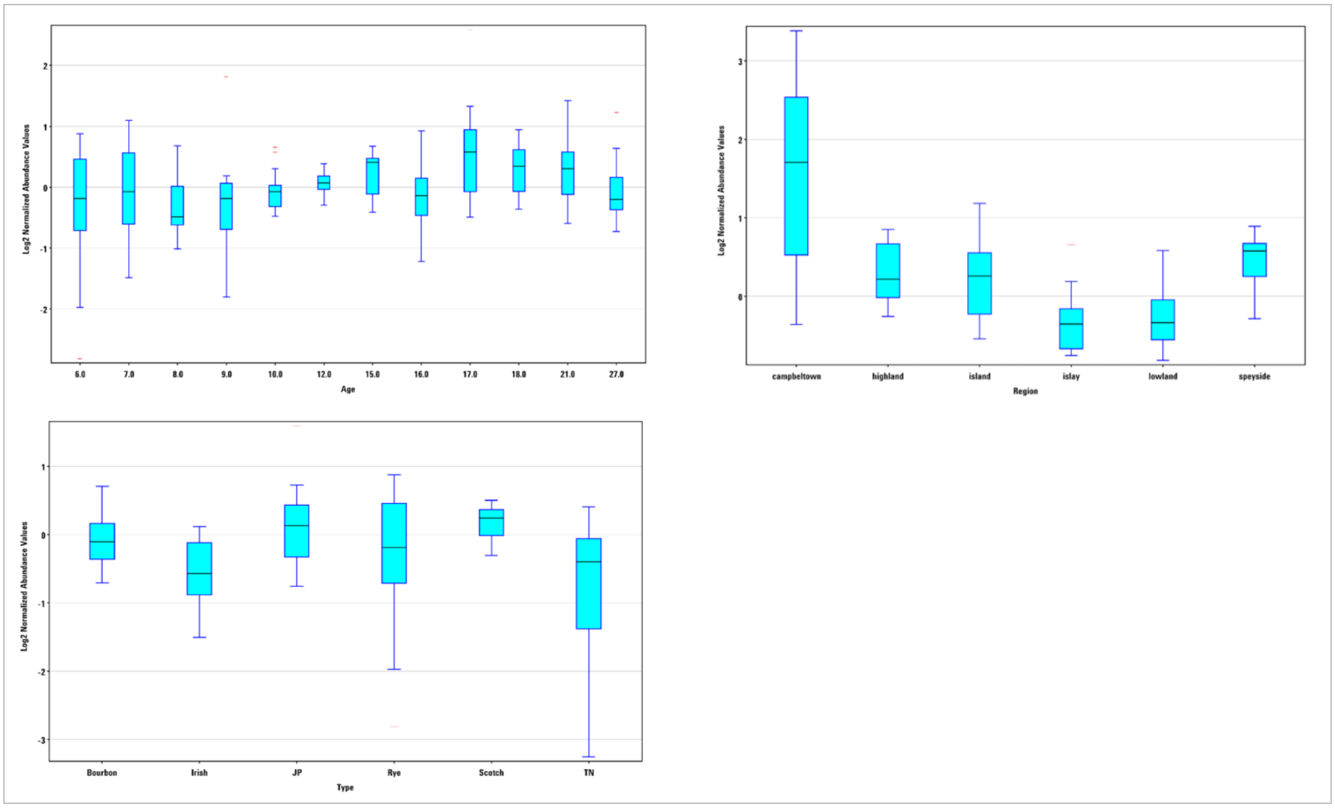

Figure 2. Agilent MPP box-whisker plots using the Agilent 5100 ICP-OES data and organizing by (a) age, (b) type and (c) region (Scotch whiskies).

Table 5. Averaged data set for the elemental content of the different whiskey types using the Agilent 5100 ICP-OES

Element & wavelength (nm)	Bourbon (n=16) (µg/L).	Irish (n=8) (µg/L).	Japanese (n=9) (µg/L).	Scotch (n=33) (µg/L).	Tennessee (n=2) (µg/L).
Al 396.152	61.65	53.59	77.83	167.4	63.03
B 249.772	119.8	82.85	117.1	145.5	123.4
Ba 455.403	12.60	11.19	3.13	10.88	9.26
Ca 396.847	1213	1040	1055	2085	1320
Cd 214.439	<MDL	<MDL	<MDL	19.54	<MDL
Cr 267.716	<MDL	7.83	<MDL	21.42	<MDL
Cu 327.395	225.06	62.32	793.1	610.9	44.85
Fe 238.204	65.36	63.97	235.9	181.7	61.14
K 766.491	1707	8291	20508	18687	16170
Mg 279.553	530.97	271.92	926.32	1104.40	322.64
Mn 257.610	51.76	20.92	42.21	51.74	22.68
Na 588.995	6048	9305	13245	9532	8233
Rb 421.552	9.43	4.45	6.79	12.27	5.61
Si 251.611	932.2	816.6	6512	1404	1094
Sr 407.771	9.52	4.53	6.99	12.28	5.66
Zn 213.857	175.2	89.49	137.2	44.34	296.0

Many whiskey producers use copper stills for distillation, especially of premium-branded spirits. Producers that use stainless steel stills will add a copper mesh lining or column. The reason Cu is so important in the distillary process is its positive effect on the aroma (and quality) of the whiskey by preventing the formation of potentially-odorous sulfur compounds [6]. The data in Table 5 shows some variation in the average Cu value in the different types of whiskeys. This is most likely the result of different processing equipment rather than raw materials [7].

Principal Component Analysis

Using MPP, all significantly different elements (P value < 0.05) were used in a Principal Component Analysis (PCA) to test whether different whiskey types could be separated based on their elemental profiles. Using the concentration data (Table 5) for the significantly different elements (Al, Ba, Ca, Cu, Fe, K, Mg, Mn, Na, Rb, Si, Sr and Zn), a graphical representation of the sample similarities and dissimilarities was obtained, as shown in Figure 3. Within the first two principal components (PC1 and PC2) over 57% of the total variance was explained, with 43.38% in the first dimension and an additional 13.68% in the second dimension. Although there is overlap between the whiskey samples, separations can also be seen. Looking at the overlaid loading plot with the elements, it becomes apparent which elements drive the separation between the different types in the first and second dimension.

Along PC 1, the most discriminating elements were Ba, Na, Mg, Sr, Rb. Along the second dimension, PC 2, the Bourbon whiskeys were mostly separated from Irish whiskies, with the other whiskey samples in between. The separation is mainly driven by higher concentrations of Na, Al versus K, Mg and Mn. All the other elements are within ±1 of the origin of the y-axis.

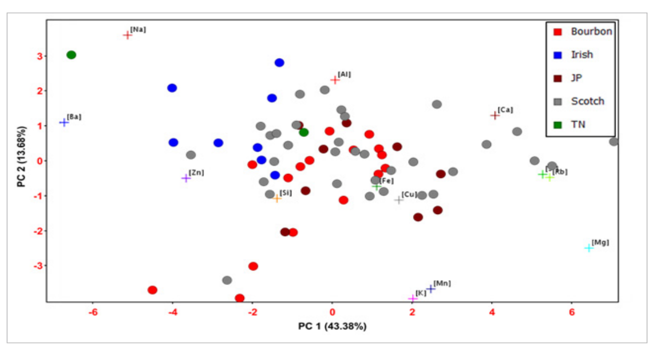

Figure 3. PCA plot showing the separation of different whiskeys by their elemental composition: the PCA score plot and PCA loading plot are overlaid to show the contribution of each element to the separation along PC 1 (43.38%) and PC 2 (13.68%). Each of the 5 different types (averaged), are coded by their type (Green...Tennessee; Blue...Irish; Red...Bourbon; Gray... Scotch; Brown...Japanese).

Conclusions

The Agilent 5100/5110 ICP-OES combined with a powerful data analysis package such as Agilent's Mass Profiler Professional (MPP) is a viable tool for elemental profiling of whiskey. Elemental differences are mainly due to processing equipment and raw materials, such as water. The method showed there is sufficient spread in the data from the analysis of the 69 whiskeys, with differences in several elements to distinguish between 5 types of whiskey.

Cooperation and collaboration with the industry is needed to determine the direction of future research in product differentiation and authentication using elemental fingerprinting techniques.

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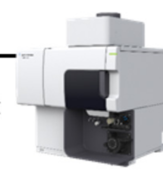

Results presented in this document were obtained using the 5100 instrument, but performance is also verified for the 5110 ICP-OES

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