

High Accuracy of Nitrogen, Carbon and Sulfur Analysis for Agronomy Applications using the Thermo Scientific FlashSmart Elemental Analyzer

Dr. Liliana Krotz, Dr. Francesco Leone
and Dr. Guido Giazzi
Thermo Fisher Scientific, Milan, Italy

Key Words

Accuracy, Automated and unattended analysis, Carbon, C/N ratio, Flash combustion, Nitrogen, Plants, Soils, Sulfur, Total Organic Carbon (TOC)

Goal

To demonstrate the performance of the Thermo Scientific FlashSmart Elemental Analyzer in agronomy applications.

Introduction

Nitrogen and carbon determination in soils is important



Figure 1. Thermo Scientific FlashSmart Elemental Analyzer.



for the evaluation of organic matter and for the determination of fertilizers needed. They also provide significant information on the nutritional elements of plants which are key factors for their future growth. The determination of nitrogen content provides insight on the quality of different types of crop, used for feeding and processing, as well as for N-cycle and N-fixation monitoring in agricultural and environmental research. The differentiation of Total Carbon (TC) and Total Organic Carbon (TOC) is also useful to evaluate the quality of soils while sulfur is an essential component of living matter. The deficiency of sulfur has a negative influence in the growth of vegetables and in the quality of proteins. Also, the carbon/nitrogen ratio of the organic material added to the soil influences the rate of decomposition of organic matter and this results in the release or immobilization

of soil nitrogen. If the added organic material contains more nitrogen in proportion to the carbon, then nitrogen is released into the soil from the decomposing organic material, while if the organic material has less nitrogen in relation to carbon, then the microorganisms will utilize the soil nitrogen for further decomposition and the soil nitrogen will be immobilized and will not be bioavailable. Therefore, cellular carbon and nitrogen metabolism must be tightly coordinated to sustain optimal growth and development for plants.

Soil and plant testing by traditional methods are no longer suitable for routine analysis due to their time consuming preparation and the use of environmentally hazardous reagents. Therefore, the use of accurate instrument analytical techniques is required. As the demand for improved sample throughput, reduction of operational costs and minimization of human errors has increased, a simple and automated technique that allows fast analysis with excellent reproducibility is essential for reliable nitrogen and carbon determination.

The Thermo Scientific™ FlashSmart™ NC Soils Analyzer (Figure 1), based on the dynamic flash combustion of the sample, copes effortlessly with the wide array of laboratory requirements such as accuracy, day to day reproducibility and high sample throughput. Nitrogen and carbon determination in soil and plant samples is fully automated in the Analyzer. The same analytical conditions can be used for Total Carbon and Total Organic Carbon determination, after acid treatment of the sample.

Through its flexibility, the Analyzer allows the simultaneous NCS analysis while for trace sulfur determination, the Analyzer can be easily coupled with a Flame Photometric Detector (FPD). This method combines the advantages of the elemental analyzer with the sensitivity, selectivity and robustness of the FPD.

Methods

The FlashSmart Analyzer operates according to the dynamic flash combustion technique. Samples are weighed in tin containers and introduced into the combustion reactor from the Thermo Scientific™ MAS Plus Autosampler together with a proper amount of oxygen. In NC configuration, after combustion, the resultant gases are carried by a helium flow to a second reactor filled with copper, then through a water trap, a GC column and finally, detected by the Thermal Conductivity Detector (TCD) (Figure 2).

In S (single determination) or simultaneous NCS configuration, after combustion of the sample, the resulting gases are carried by a helium flow to a layer filled with copper, then through a water trap, a GC column and finally, detected by the TCD (Figure 3). For trace sulfur analysis, after the water trap, the gases are carried by a helium flow through a short GC column and finally, detected by a FPD, (Figure 4).

A complete report is automatically generated by the Thermo Scientific™ EagerSmart™ Data Handling Software and displayed at the end of the analysis.

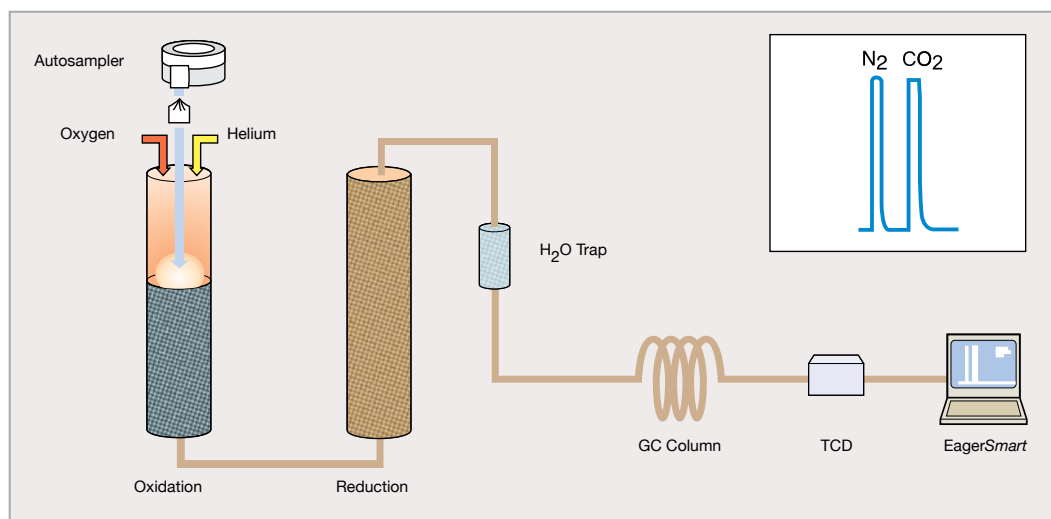


Figure 2. FlashSmart NC configuration.

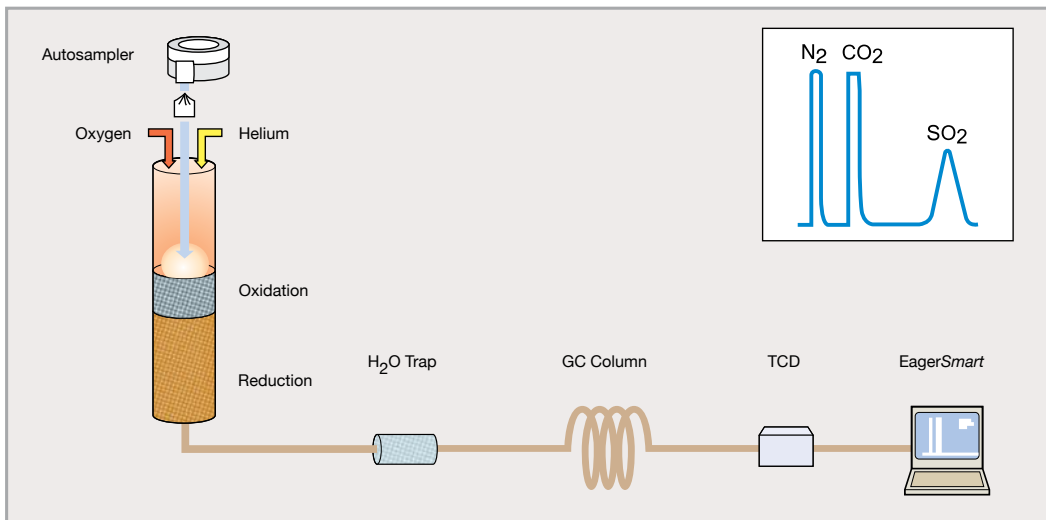


Figure 3. FlashSmart NCS configuration.

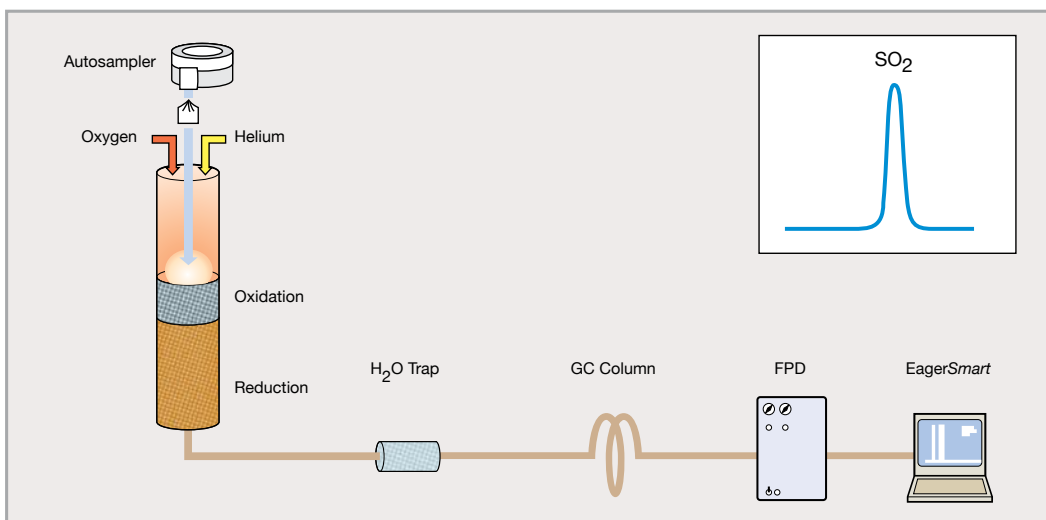


Figure 4. FlashSmart Sulfur Configuration by FPD detector.

The differentiation of Total Carbon from Total Organic Carbon requires sample manipulation prior to analysis, following the Official Italian Method on Soils Analytical Chemistry, Method 248 (Gazzetta Ufficiale).

TOC was determined after removing carbonate minerals by acidification of the sample with HCl 1:1 (Figure 5) using the kit shown in Figure 6. The TC and TOC analyses were performed consecutively using the same analytical conditions of the instrument.

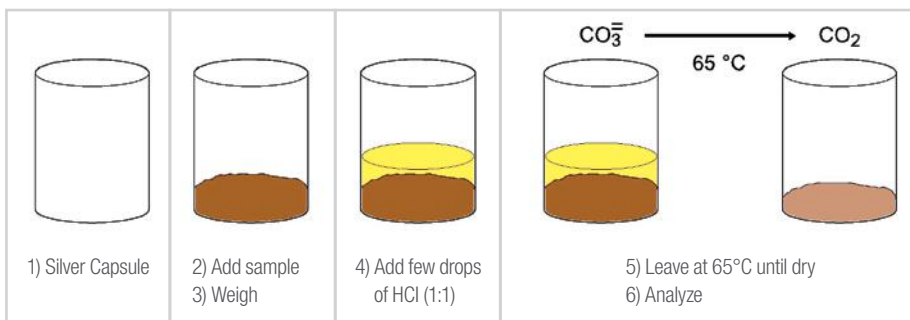


Figure 5. Method for sample manipulation.



Figure 6. Kit for Total Organic Carbon (TOC) for solid samples.

Results

To demonstrate the performance of the FlashSmart Analyzer, five instruments were chosen to perform a typical analytical test using the NC configuration. The calibration was performed with Aspartic acid standard using K factor as the calibration method. Following, three analyses of about 100 mg Thermo Scientific Soil Reference Material were performed as unknown.

Table 1 shows the correlation between the certified NC percentages of the Soil Reference Material, the accepted range according to the technical specification of the system and the experimental data obtained in triplicate analyses for each instrument. All data are acceptable and comparable confirming the repeatability and reproducibility of the FlashSmart Analyzer.

Table 1. NC data of Thermo Scientific Soil Reference Material.

Technical Specification			FlashSmart Analyzers									
Elements	Theoretical	Range	1		2		3		4		5	
	%	(±)	%	RSD%	%	RSD%	%	RSD%	%	RSD%	%	RSD%
N	0.186	0.02	0.185	0.31	0.186	0.31	0.183	0.31	0.184	0.54	0.185	0.31
			0.185		0.186		0.184		0.185			
			0.184		0.185		0.184		0.186			
C	1.697	0.02	1.693	0.51	1.688	0.51	1.706	0.36	1.697	0.2	1.701	0.56
			1.705		1.704		1.695		1.691			
			1.688		1.702		1.696		1.689			

Table 2 shows the C/N ratio for each NC determination from five instruments. The calculated values are comparable confirming the excellent reproducibility of the FlashSmart Analyzer which is calculated automatically by

the dedicated EagerSmart Software. The values obtained are comparable confirming the excellent reproducibility of the FlashSmart Analyzer.

Table 2. C/N ratio of Thermo Scientific Soil Reference Material.

FlashSmart Analyzers : C/N ratio				
1	2	3	4	5
23.23	23.23	23.88	23.52	23.33
23.36	23.44	23.66	23.28	23.30
23.30	23.57	23.62	23.14	23.48

In order to evaluate the accuracy of NC data, five soil and three plant reference materials were analyzed. The calibration was performed with Aspartic acid using

K factor as the calibration method. Table 3 shows the certified N% and C% and the relative uncertainty while Table 4 shows the experimental results obtained.

Table 3. Certified NC data from soil and plant reference materials.

Reference Material Information	Specification			
	N %	Uncertainty (±)	C %	Uncertainty (±)
Low Organic Content Soil	0.133	0.023	1.61	0.09
Medium Organic Content Soil	0.27	0.02	3.19	0.07
Loamy Soil	0.27	0.02	2.75	0.12
Chalky Soil	0.35	0.02	5.39	0.09
Sandy Soil	0.07	0.01	0.83	0.05
Birch Leaves	2.12	0.06	48.09	0.51
Orchard Leaves	2.28	0.04	50.40	0.40
Alfalfa	3.01	0.20	n.a.	n.a.

Table 4. Experimental NC data from soil and plant reference materials.

Sample	N %	RSD %	C %	RSD %
Low Organic Content	0.122 0.120 0.123	1.25	1.56 1.55 1.55	0.37
Medium Organic Content	0.27 0.26 0.27	2.16	3.14 3.14 3.16	0.37
Loamy Soil	0.26 0.26 0.27	2.19	2.70 2.70 2.73	0.63
Chalky Soil	0.36 0.36 0.37	1.58	5.33 5.33 5.35	0.22
Sandy Soil	0.0699 0.0712 0.0690	1.57	0.822 0.835 0.815	1.23
Birch Leaves	2.11 2.12 2.10	0.47	48.12 48.22 48.29	0.18
Orchard Leaves	2.28 2.31 2.29	0.66	50.51 50.31 50.36	0.20
Alfalfa	2.93 2.94 2.89	0.90	43.60 43.51 43.65	0.17

To demonstrate the validity of the TOC method, two soil reference materials were analyzed after acid treatment of the sample. Table 5 shows the TOC data obtained, which

are in agreement with the declared values demonstrating the accuracy of the TOC method.

Table 5. TOC data from soil reference materials.

Sample	Declared TOC %	Experimental TOC %	RSD %
Chalky Soil	3.30	3.338 3.332 3.337	0.096
Sandy Soil	0.76	0.757 0.755 0.758	0.202

Table 6 shows the NC and TOC data obtained from five different soil samples analyzed in the NC configuration: the calibration was performed with Aspartic acid using

K factor as the calibration method and the soil sample weight was about 100 mg.

Table 6. NC and TOC data of soil samples.

Sample	N %	RSD %	C %	RSD %	TOC %	RSD %
A	0.147 0.144 0.146 0.146 0.144	0.923	1.495 1.488 1.492 1.481 1.486	0.364	1.330 1.356 1.339 1.365 1.334	1.117
B	0.147 0.147 0.147 0.146 0.148	0.481	4.697 4.763 4.685 4.782 4.686	0.980	3.966 3.917 3.911 3.929 3.962	0.648
C	0.249 0.258 0.250 0.249 0.250	1.526	3.808 3.791 3.781 3.810 3.785	0.350	2.862 2.864 2.844 2.858 2.870	0.340
D	0.321 0.321 0.312 0.311 0.313	1.578	5.159 5.116 5.124 5.149 5.108	0.426	3.697 3.699 3.738 3.705 3.724	0.478
E	0.168 0.165 0.164 0.163 0.165	1.134	2.522 2.552 2.575 2.574 2.523	1.022	1.842 1.844 1.902 1.860 1.881	1.371

Table 7 shows data for simultaneous NCS and TOC determination on soil samples. For NCS, BBOT (2,5-Bis (5-tert-butyl-benzoxazol-2-yl) thiophene) was analyzed as standard using K factor as the calibration method and the soil sample weight was 15-20mg. For TOC determination,

3-4 mg of aspartic acid was used for calibration and 10-12 mg of soil samples. Table 8 shows data for simultaneous NCS determination on plant samples. BBOT was analyzed as standard using K factor as the calibration method and plant sample weight was 3-4 mg.

Table 7. NCS and TOC data of soil samples.

Soil Sample	N %	RSD %	C %	RSD %	S %	RSD %	TOC %	RSD %
1	0.0482 0.0481 0.0497	1.84	3.6494 3.7371 3.7084	1.21	0.0147 0.0151 0.0149	1.34	0.4183 0.4229 0.4258	0.89
2	0.2504 0.2484 0.2598	2.41	3.7595 3.7892 3.7112	1.05	0.0674 0.0678 0.0682	0.59	1.8075 1.7955 1.8105	0.44
3	1.0485 1.0468 1.0597	0.67	13.4481 13.2674 13.4497	0.78	0.5253 0.5292 0.5275	0.37	10.0688 10.2617 10.1640	0.95
4	0.0585 0.0582 0.0583	0.26	0.8741 0.8708 0.8702	0.24	0.0103 0.0104 0.0101	1.49	0.7563 0.7304 0.7416	1.75

Table 8. NCS data of plant samples.

Sample	N %	RSD %	C %	RSD %	S %	RSD %
1	2.420 2.457 2.436	0.78	42.060 42.092 42.085	0.04	0.319 0.314 0.310	1.45
2	2.511 2.502 2.520	0.40	41.240 41.132 41.103	0.18	0.117 0.119 0.115	1.72
3	1.065 1.059 1.060	0.30	49.532 49.584 49.526	0.06	0.188 0.183 0.184	1.42

Table 9 shows the trace sulfur data measured using the FPD detector. The calibration was performed with the

Thermo Scientific Soil Reference Material using Quadratic Fit as the calibration method.

Table 9. Trace sulfur data obtained from the FPD detector.

Sample	S ppm	RSD %	Sample	S ppm	RSD %
1	25 22 21	6.79	3	87 83 84	2.43
2	56 51 52	4.90	4	102 99 100	1.51

Conclusions

The all-in-one Thermo Scientific FlashSmart Analyzer is the optimal solution in agronomy for the quantitative determination of nitrogen, carbon and sulfur, and the C/N ratio, in sample matrices with a wide range of concentrations spanning low to high amounts. Specifically, the Analyzer demonstrates excellent repeatability, reproducibility, accuracy and precision.

The nitrogen, carbon and sulfur data obtained by the FlashSmart Analyzer are inside the tolerance declared for the Reference Materials. No memory effect was observed when changing the sample type, indicating the complete conversion and detection of the nitrogen, carbon and sulfur present in the sample.

Thanks to the modularity of the FlashSmart Analyzer, the same hardware, autosamplers and software can be readily used for other configurations such as CHN/O, CHN/S, CHNS/O, CHNS/CHNS, CHN/CHN, NC/S, N-Protein/S and more. This can mainly be realized by changing the consumables as the Analyzer and software are complete, illustrating the all-in-one nature of the Analyzer. The FlashSmart Analyzer can also operate using argon as carrier gas. In addition to the Official Italian Method on Soils Analytical Chemistry, Method 248 (Gazzetta Ufficiale), the Analyzer also fulfills with the requirements of other official methods. The most relevant examples are listed in Table 10.

Table 10. Most relevant Official Methods.

Official Organization	Method
AOAC (Association of Official Analytical Chemists)	Official Method 993.13. Nitrogen (Total) in Fertilizers 2.4.02
EPA (Environmental Protection Agency)	Method 440.0, 1997 Determination of Carbon and Nitrogen in Sediments and Particulates of Estuarine/Coastal Waters Using Elemental Analysis
ISO 10694, 1995	Soil Quality – Determination of organic and total Carbon after dry combustion (elementary analysis)
ISO 13878, 1998	Soil Quality – Determination of total Nitrogen content by dry combustion (elemental analysis)
ISO 13878, 1998	Soil Quality – Determination of total Nitrogen content by dry combustion (elemental analysis)
ISO 15178, 2016	Soil Quality – Determination of Total Sulfur by Dry Combustion
Official Italian Method (Gazzetta Ufficiale)	Method 146, 1998 New regulations for fertilizer's control
Official Italian Method on Soils Analytical Chemistry (Gazzetta Ufficiale)	Method 248, 1999 Nitrogen, Carbon and Organic Carbon and Soils
UNI EN 13654-2	Soil improvers and growing media. Determination of Nitrogen by combustion method.

Find out more at thermofisher.com/OEA