Applicability of Two Separation Methods for Elemental Analysis of Typical Agricultural Biomass in China

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China still lacks standardized methods for element compositional analysis of agricultural biomass, in particular crop straw and livestock manure, which severely restricts the efficiency of the comprehensive utilization of agricultural biomass. Two separation methods, namely adsorbed–desorbed separation (AS) and chromatographic separation (CS), were applied to simultaneously analyze the carbon, hydrogen, nitrogen, and sulfur elements in major agricultural biomass. The optimal sizes of 1.00 mm for crop straws and 0.50 mm for livestock manure were obtained through optimization experiments. Afterwards, the sample mass was considered on two different elemental analysis instruments. From the metrological characteristics and variance analysis, a sample mass of 40 mg of the AS method was applicable for all the agricultural biomass, even with a content of 0.5%. On the other hand, 5 mg of the CS method was suitable for agricultural biomass with contents greater than 1.5%. It is recommended that samples should be kept free of impurities and completely homogeneous, especially for livestock manure. The results provide significant data for establishing a national standard system in the near future.

Keywords: Applicability; Elemental analysis; Separation methods; Agricultural biomass

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INTRODUCTION

China is rich in biomass resources. Taking major agricultural biomass into account, the annual yield of livestock manure is more than 4 billion tons (Shen *et al*. 2015), and that of crop straws is approximately 0.8 billion tons (Wang *et al.* 2010). Biomass contains a number of chemical elements, such as C, H, O, N, and S, and elemental compositional analysis is important to evaluate and predict their recycling potential (Parikh *et al.* 2007; Vassilev *et al.* 2010). Agricultural biomass comes in a wide range of types, forms, compositions, and structures in China (Vassilev *et al*. 2013). Inconsistency in research and analytical methods has led to an inconsistency in results, so that basic data on biomass elemental characteristics are not uniform and are sometimes unscientific, reducing their adaptability, reliability, and authoritativeness. This has become a bottleneck in evaluating the scientific, technological, safety, and industrial factors, restricting the comprehensive utilization of agricultural biomass in China.

Common international methods of elemental detection include chemical and instrumental analyses. The Liebig Method (a three-stage furnace method), for example, is used to determine C and H (ASTM E777-08 2008). Chemical analysis uses sorbents to absorb elements, and weight gains are calculated after combustion. The element N can be determined using digestion analysis, *i.e.* the Kjeldahl method (ISO 5983-1 2009). This method requires the use of chemical reagents, which can be risky, and the digestion time is long. The Askal method is suitable for determining S (ASTM E775-87 2008; ISO 334 2013), which is precipitated in the form of barium sulfate after mixed combustion. Instrumental analysis is based on the principle that absorption is reduced after combustion, and the elements C and H (SN EN 15104 2011; ASTM D5373-14 2014) can be analyzed with a detector. This combustion method may also be used to determine the elements N (AOAC 2001.11 2001; ISO 16634-1 2008) and S (BS EN 15289 2011) in solid biomass fuels.

At present, the Liebig, Kjeldahl, and Askal methods (GB/T 21923 2008; GB/T 28734 2012; GB/T 28732 2012.) are generally used to determine the elements C, H, N, and S in solid biomass fuels in China. The instrumental method can simultaneously analyze the carbon, hydrogen, nitrogen, and sulfur elements, so that these methods are quick and easy. However, different elemental analysis instruments and variability of different biomass make the measurements non-uniform and sometimes unscientific. Some scholars have studied the impact of different analysis instruments on experimental results (Sieper *et al*. 2006; Gazulla *et al*. 2012), as well as the impact of pre-processing methods for various feedstock (Ryba and Burgess 2002). The potential of instrumental methods to determine CHNS was also reported by Eksperiandova *et al*. (2011). The current studies showed that the instrumental methods have great potential, while the applicability of this method to agricultural biomass should be explored.

The aforementioned methods are not specifically applicable for agricultural biomass, especially in China. Inconsistency in research and analytical methods have led to an inconsistency in results. Currently, China still lacks methods for measuring the elemental composition of agricultural biomass, especially for straw and livestock manure. Nowadays, the widespread instrumental analysis is distinguished by two separation methods, specifically, adsorbed–desorbed separation (AS) and chromatographic separation (CS). The normal sample mass recommended for AS can be dozens of milligrams, while only a few milligrams are sufficient for CS. Thus, the required sample mass and the measured results differ greatly on the basis of the different separation methods. Because of the diversity and complexity of biomass materials, discussion of the applicability and accuracy of different instrumental methods is necessary.

Representative samples were selected from China's Agricultural Biomass Database funded by the state. The purpose was to determine optimal conditions of particle size and sample mass for element compositional analysis of the main agricultural biomasses in China. Authorized by the Ministry of Agriculture of China, this study aims to estimate the applicability of instrumental analysis with different separation methods (AS and CS) and to estimate the metrological characteristics for the main agricultural biomasses in China to provide reliable data and methodology for the establishment of elemental analysis criteria.

EXPERIMENTAL

Preparation of Samples

Samples were selected from the National Agricultural Biomass Samples Base, including five crop straw samples, originating from wheat, corn, rice, cotton, and rape, respectively; and five livestock manure samples, originating from pig, beef cattle, dairy cattle, broiler chicken, and layer chicken, respectively. The samples are all typical and major species in China to ensure the applicability and representativeness of the method.

The original samples of crop straws were dried in a DHG-9246A oven (Shanghai Jinghong Laboratory Instrument Co., Ltd, Shanghai, China) at 45 ± 3 °C to a constant weight, crushed and pulverized by SF130 crushers (Zhejiang Rui'an Dongyuan Medical Device Machinery Plant, Zhejiang, China) into 1-mm particles, and bagged and sealed for tests in reference to ASTM E1757-01 (2007) and NREL/TP-510-42620 (2008). The livestock manure samples were dried in an oven at 70 ± 5 °C to a constant weight, ground using a ZM200 cyclone mill (German Retsch GmbH, Haan, Germany) into 0.5-mm particles, and bagged and sealed for tests with reference to TMECC05.07-A (2000), TMECC 03.09-A (2000) and TMECC02.02 (2000).

Experimental Design and Methods

Sample particle size and sample mass are the two most important impact factors in determining and analyzing the elemental composition of biomass samples. First, the sample size was optimized by consideration of 1.00-, 0.50-, and 0.25-mm sample sizes for crop straw and 0.50-, 0.25-, and 0.125-mm sample sizes for livestock manure. The sample sizes were chosen according to the sample preparation methods. Samples for tests were placed in tin capsules, weighed, and packed carefully. The prepared samples were analyzed by a Vario Macro Elemental Analyzer (German Elemental GmbH, Frankfurt, Germany) equipped with AS equipment.

After the optimization of sample size, the sample mass was considered on two different elemental analysis instruments. The AS method was performed by the Vario Macro elemental analyzer using four sample masses (5, 10, 20, and 40 mg), denoted AS5, AS10, AS20, and AS40. Another elemental analyzer applying the CS method, the Thermo Scientific Flash 2000 (CE Elantech, Inc, Lakewood, NJ), was utilized with sample masses of 2.5, 5, and 10 mg (denoted CS2.5, CS5, and CS10, respectively). It was recommended that each sample was subject to three parallel elemental analyses to ensure the reliability of the test results (Oasmaa and Meier 2005).

Data Processing and Analysis

The variance analysis was performed using SPSS 17.0 (IBM Corporation, Armonk, NY) to determine the approximate elemental composition content of crop straw and livestock manure under various operation conditions. Diagraph analysis was performed using the software Origin 8.5 (OriginLab, Northampton, MA).

RESULTS AND DISCUSSION

Influence of Sample Particle Size

With uniform sample sizes, the measurement could be more scientific, reliable, and authoritative. Since the variability among different species are significant, sample size optimization experiments were carried out. Elemental composition was measured and analyzed in five crop straw samples of three different particle sizes (1.00, 0.50, and 0.25 mm) and five livestock manure samples of three different sizes (0.50, 0.25 mm, and 0.125 mm). Each sample had a mass of 40 mg and was subject to a gas flow rate of 100 $mL \cdot min^{-1}$ and an oxygenation time of 90 s.

The discussion of degree of the influence was based on the variance analysis and Duncan comparison method. The same letters denote that the factor in the same column is not significant ($p > 0.05$). Different letters denote a significant difference of the factor in the same column ($p < 0.05$). As shown in Table 1, the crop straw particle size has no influence on the analysis of H ($p > 0.05$), only a minor influence on that of S, and some influence on that of C and N ($p < 0.05$), but there is not a significant trend. This should be because of a difference in the contents of C and H at different positions in the straw sampled, rather than the particle size.

As shown in Table 1, particle size had no significant effect on the elemental analysis results of livestock manure samples, except for some influence without a trend on layer manure. This may be because of the unevenness in the strength of this manure that would affect its crushing, which is caused by the addition of a calcium-containing substance (*e.g.*, lime) in layer feeds or forage to facilitate the generation of egg shells.

Table 1. Influence of Particle Size on Elemental Analysis of Five Straw Samples and Five Livestock Manure Samples

	Particle Size (mm)	C (%)	H (%)	N (%)	S (%)
Wheat straw	1.00	42.94±0.15^{b}	5.67±0.03^{a}	0.60±0.01^{a}	0.43±0.15^{a}
	0.50	39.77±0.10^{a}	5.60±0.02^{a}	0.86±0.02^{c}	0.33±0.10^{a}
	0.25	42.60±0.11^{b}	5.76±0.22^{a}	0.70±0.04^{b}	0.48±0.11^{b}
Corn straw	1.00	43.64±0.10^{a}	5.83±0.01^{a}	0.82±0.03^{ab}	0.25±0.00^{a}
	0.50	43.77±0.07^{a}	5.83±0.01^{a}	0.79±0.02^{a}	0.25±0.00^{a}
	0.25	43.62±0.08^{a}	5.82±0.06^{a}	0.86±0.01^{b}	0.30±0.04^{a}
Rice straw	1.00	45.32±0.05^{a}	5.89±0.03^{a}	0.85±0.02^{b}	0.30±0.01^{a}
	0.50	45.59±0.13^{b}	5.92±0.02^{a}	0.81±0.01^{a}	0.29±0.00^{a}
	0.25	45.53±0.11^{b}	5.96±0.02^{a}	0.89±0.02^{b}	0.30±0.00^{a}
Cotton straw	1.00	39.66±0.04^{a}	5.60±0.01^{a}	0.88±0.01^{a}	0.31±0.00^{a}
	0.50	39.77±0.10^{a}	5.60±0.02^{a}	0.87±0.02^{a}	0.33±0.01^{a}
	0.25	40.12±0.00^{b}	5.63±0.01^{a}	0.92±0.00^{b}	0.35±0.04^{a}
Rape straw	1.00	43.32±0.09^{a}	5.74±0.00^{a}	0.64±0.02^{a}	0.44±0.00^{a}
	0.50	43.68±0.05^{c}	5.77±0.01^{a}	0.62±0.02^{a}	0.45±0.02^{ab}
	0.25	43.49±0.06^{b}	5.76±0.01^{a}	0.66±0.02^{a}	0.47±0.00^{b}
Pig manure	0.50	37.58±0.36^{a}	5.54±0.05^{a}	2.54±0.03^{a}	0.54±0.01^{a}
	0.25	37.43±0.12^{a}	5.50±0.04^{a}	2.52±0.01^{a}	0.55±0.01^{a}
	0.125	37.99±0.04^{a}	5.59±0.01^{a}	2.52±0.00^{a}	0.63±0.05^{b}
Beef manure	0.50	37.86±0.09^{a}	5.18±0.04^{ab}	2.43±0.02^{a}	0.83±0.06^{a}
	0.25	38.02±0.13^{ab}	5.15±0.04^{a}	2.40±0.01^{a}	0.80±0.02^{a}
	0.125	38.18±0.09^{b}	5.25±0.04^{b}	2.45±0.03^{a}	0.84±0.01^{a}
Dairy manure	0.50	34.07±0.24^{a}	4.81±0.05^{a}	1.75±0.03^{a}	0.50±0.01^{a}
	0.25	34.25±0.24^{a}	4.82±0.05^{a}	1.72±0.02^{a}	0.51±0.00^{a}
	0.125	34.42±0.04^{a}	4.83±0.03^{a}	1.76±0.01^{a}	0.55±0.02^{b}
Broiler manure	0.50	34.51±0.11^{a}	4.89±0.04^{a}	4.08±0.05^{a}	1.44±0.02^{a}
	0.25	34.99±0.05^{b}	4.94±0.05^{a}	4.06±0.00^{a}	1.44±0.01^{a}
	0.125	35.07±0.03^{b}	4.97±0.02^{a}	4.10±0.01^{a}	1.50±0.03^{b}
Layer manure	0.50	34.15±0.22^{a}	4.72±0.04^{a}	3.07±0.03^{b}	0.71±0.02^{b}
	0.25	34.31±0.11^{a}	4.74±0.02^{a}	3.01±0.01^{a}	0.62±0.01^{a}
	0.125	34.65±0.07^{b}	4.84±0.01^{b}	3.07±0.01^{b}	0.76±0.02^{c}

Notes: The same letters denote that particle size in the same column is not significant ($p > 0.05$). Different letters denote a significant difference in particle size in the same column ($p < 0.05$).

The particle size was generally 1.00 mm for the prepared straw samples and 0.50 mm for the prepared sample of livestock manure. With decreasing particle size, the mixing of straw improved. However, considering energy consumption, screening, complicated operation of the crusher, and other conditions with practical applications, it is recommended that the crushed particle sizes of the straw and livestock manure should be 1.00 mm and 0.50 mm, respectively. This result agrees well with the recommended size of less than 1 mm mentioned by BSEN 15104 (2011).

Influence of Sample Mass on Crop Straws

The impact of AS and CS methods on elemental analysis was compared. Measurement of C, H, N, and S compositions was performed separately on five straw samples.

Table 2. Elemental Analysis of Straw Biomass based on the Principle of Absorption and Desorption Separation

	Mass (mg)	C (%)	H (%)	N (%)	S (%)
Wheat straw	5	42.61±0.32^{b}	7.79±0.82^{c}	0.60±0.05^{ab}	1.68±0.27^{b}
	10	40.02±1.52^{a}	9.05±0.48^{d}	0.66±0.07^{b}	0.77±0.05^{a}
	20	41.42±0.15^{ab}	6.88±0.13^{b}	0.53±0.04^{a}	0.65±0.01^{a}
	40	41.73±0.12^{b}	5.88±0.06^{a}	0.57±0.01^{ab}	0.53±0.00^{a}
Corn straw	5	44.07±0.32^{a}	9.14±0.37^{b}	0.80±0.24^{a}	1.20±0.06^{c}
	10	41.37±5.27^{a}	9.45±1.18^{b}	0.75±0.13^{a}	0.62±0.10^{b}
	20	42.22±0.26^{a}	7.20±0.19^{a}	0.85±0.04^{a}	0.53±0.02^{ab}
	40	43.13±0.30^{a}	6.26±0.04^{a}	0.84±.004^{a}	0.43±0.03^{a}
Rice straw	5	40.75±0.53^{b}	8.20±0.85^{c}	0.82±0.20^{a}	1.15±0.04^{c}
	10	35.70±1.81^{a}	7.68±1.04^{bc}	0.75±0.05^{a}	0.62±0.09^{b}
	20	39.28±0.29^{b}	6.80±0.11^{ab}	0.87±0.07^{a}	0.54±0.02^{b}
	40	40.23±0.46^{b}	5.96±0.14^{a}	0.88±0.04^{a}	0.43±0.01^{a}
Cotton straw	5	44.91±0.59^{a}	9.07±0.21^{b}	0.88±0.17^{a}	1.06±0.11^{d}
	10	43.16±3.26^{a}	8.72±1.90^{b}	0.90±0.15^{a}	0.90±0.08^{c}
	20	44.63±0.90^{a}	7.59±0.21^{ab}	0.92±0.11^{a}	0.55±0.01^{b}
	40	43.76±0.74^{a}	6.11±0.08^{a}	0.91±0.04^{a}	0.41±0.03^{a}
Rape straw	5	42.39±0.81^{b}	9.34±1.04^{b}	1.10±0.01^{a}	1.23±0.03^{a}
	10	38.47±0.72^{a}	8.61±0.78^{b}	1.13±0.23^{a}	1.11±0.30^{a}
	20	41.64±1.01^{b}	6.64±0.42^{a}	1.17±0.02^{a}	1.01±0.30^{a}
	40	41.70±0.12^{b}	5.88±0.19^{a}	1.20±0.05^{a}	0.71±0.02^{a}

Notes: The same letters denote that particle mass in the same column is not significant ($p > 0.05$). Different letters denote a significant difference in particle mass in the same column ($p < 0.05$).

Biomass elements can be classified into major elements (> 1%), minor elements (0.1% to 1.0%), and trace elements (< 0.1%) (Vassilev *et al.* 2010). As shown in Tables 2 and 3, sample mass had a significant influence on the analysis of the five crop straws' C, H, N, and S, but the trend was not significant. Significant differences were presented for condition AS10, which means the measured results of AS10 are suspect and not recommended for C detection of crop straw. Results of analysis-of-variance and Duncan comparisons are shown in Tables 2 and 3. Notably, the results of C from CS were basically lower than the measured results from AS, except for AS10. For C analysis, the different sample masses of AS made no differences ($p > 0.05$) except 10 mg; meanwhile, hardly any difference was found among the different CS sample masses ($p > 0.05$). The relative standard deviation (RSD) was estimated as a metrological characteristic, and the results are demonstrated in Fig. 1. The variation degree of C in Fig. 1 reveals stable measuring accuracy under all conditions, with none exceeding 5% except AS10. The results indicate that different separation methods and various sample masses had little effect on the elemental C measurement of crop straw, unless there was an instrumental error.

Table 3. Elemental Analysis of Straw Biomass based on the Principle of Chromatographic Separation

	Mass (mg)	C (%)	H (%)	N (%)	S (%)
Wheat straw	2.5	40.86 ± 0.35^{a}	5.20 ± 0.11^{b}	0.71 ± 0.02^{b}	0.74 ± 0.04^{b}
	5	40.70 ± 0.37^{a}	5.44 ± 0.02^{c}	1.01 ± 0.13^{c}	0.69 ± 0.02^{b}
	10	40.36 ± 0.08^{a}	4.96 ± 0.15^{a}	0.42 ± 0.03^{a}	0.13 ± 0.00^{a}
Corn straw	2.5	41.77 ± 0.05^{b}	5.38 ± 0.01^{b}	1.18 ± 0.10^{b}	0.71 ± 0.06^{c}
	5	41.03 ± 0.35^{a}	5.55 ± 0.08^{b}	1.31 ± 0.05^{b}	0.63 ± 0.04^{b}
	10	41.25 ± 0.05^{a}	4.93 ± 0.21^{a}	0.70 ± 0.05^{a}	0.15 ± 0.01^{a}
Rice straw	2.5	38.97 ± 0.03^{a}	5.20 ± 0.06^{b}	0.94 ± 0.08^{b}	0.64 ± 0.06^{b}
	5	38.49 ± 0.52^{a}	5.42 ± 0.14^{b}	1.21 ± 0.06^{c}	0.56 ± 0.03^{b}
	10	38.57 ± 0.44^{a}	4.82 ± 0.16^{a}	0.75 ± 0.09^{a}	0.15 ± 0.01^{a}
Cotton straw	2.5	43.58 ± 0.12^{b}	5.20 ± 0.35^{a}	1.15 ± 0.11^{b}	0.66 ± 0.04^{b}
	5	42.15 ± 0.84^{a}	5.63 ± 0.10^{b}	1.45 ± 0.05^{c}	0.61 ± 0.04^{b}
	10	42.63 ± 0.03^{ab}	4.98 ± 0.02^{a}	0.80 ± 0.01^{a}	0.16 ± 0.01^{a}
Rape straw	2.5	40.73 ± 0.57^{a}	5.17 ± 0.05^{a}	1.26 ± 0.08^{a}	0.75 ± 0.08^{b}
	5	40.02 ± 0.16^{a}	5.45 ± 0.07^{b}	1.46 ± 0.14^{b}	0.68 ± 0.07^{b}
	10	40.34 ± 0.47^{a}	5.05 ± 0.09^{a}	1.07 ± 0.06^{a}	0.27 ± 0.01^{a}

Notes: The same letters denote that particle mass in the same column is not significant ($p > 0.05$). Different letters denote a significant difference in particle mass in the same column ($p < 0.05$).

Similarly, the results from CS were also lower compared with those of AS in the measurement of elemental H, as shown in Tables 2 and 3. As the sample mass decreased, the AS measured results became larger. While the trend was different for CS, for which the maximum results were obtained at CS5. From Table 2, all four different sample masses had significant differences with respect to wheat straw, whereas the AS5 and AS10 were at the same class for the other four straws, and the same as AS20 and AS40. The measurement accuracy depicted in Fig. 1 shows that the RSD of all straws at AS40, CS5, and CS10 were within 5%. Given the results, a sample mass of 40 mg for AS instruments, and 5 or 10 mg for CS instruments, are recommended for the measurement of crop straws' elemental H.

Generally, the measurement of minor elements should be more critical. Due to the low content and resulting variability of measurement results (Eksperiandova *et al.* 2011), it's definitely demanding for stable conditions. As can be seen from Tables 2 and 3, sample mass had an obvious effect on the AS and CS measurement of N. The AS results showed an irregular trend, whereas a regular influence emerged from CS, *i.e.*, the maximum value was determined from CS5, followed by CS2.5 and CS10, accordingly. The detailed significance analysis of AS in Table 2 shows that sample mass had no significant effect on crop straws ($p > 0.05$), apart from wheat straw. However, significant differences were observed in the analysis of CS data of elemental N in Table 3 ($p < 0.05$). As shown in Fig. 1, the RSD results for elemental N from AS5, AS10, and AS20 are distributed in a wide range. Therefore, AS40 is recommended for the measurement of all crop straws. On the other hand, if CS is applied, the proposed masses for measurement of N are 2.5 mg for wheat and corn straw, 5 mg for rice straw, and 10 mg for cotton and rape straw.

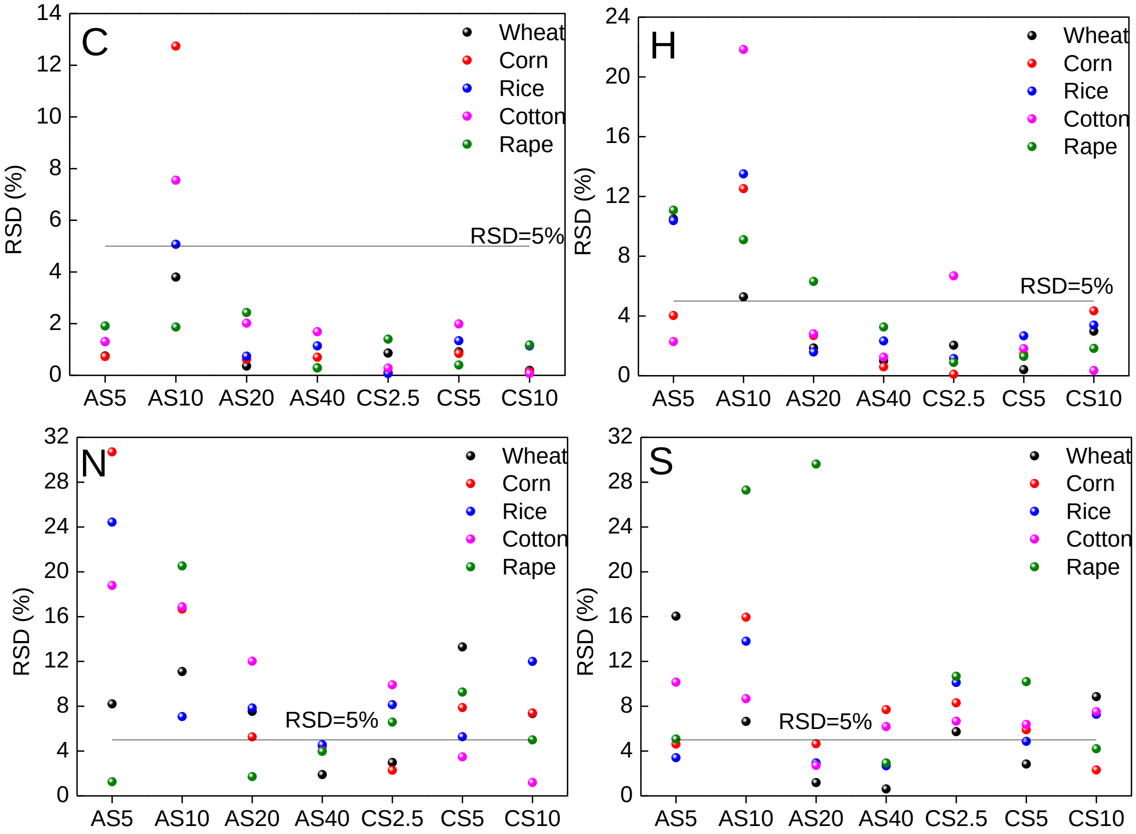

Fig. 1. The corresponding RSD concentration of five crop straws

As shown in Tables 2 and 3, sample mass also had a certain effect on AS and CS measurements of S. With decreasing sample mass, the measured values for AS and CS gradually decreased, and the trend was similar to that of elemental H. From the significance analysis in Table 2, significant differences were observed in crop straws, except rape straw. On the other hand, there was no obvious difference between CS2.5 and CS5, except for corn straw. However, the RSD results changed relatively greatly comparing with 5%, as illustrated in Fig. 1. For the minor element S, the results indicate that elemental measurement on the basis of AS showed better accuracy and precision. The minimum results for measured S could be as low as 0.5%, as deduced from Tables 2 and 3.

Given these results, different elemental instruments and various sample masses had different effects on the measurement of crop straws. Overall, a sample mass of 40 mg for AS and 5 mg of CS are recommended for C and H measurement in crop straws, whereas, for determination of the minor elements N and S, applying a sample mass of 40 mg for AS is recommended, which may be related to the absolute content of N and S elements. The recommended masses were lower than the scope of exceeding 50 mg (Gazulla *et al.* 2012).

Influence of Sample Mass on Livestock Manure

Measurement of C, H, N, and S compositions was performed separately on five livestock manure samples. The impact of AS and CS methods on elemental analysis was compared as shown in Table 4 and Table 5.

Table 4. Elemental Analysis of Manure Biomass based on the Principle of Absorption and Desorption Separation

	Mass (mg)	C (%)	H (%)	N (%)	S (%)
Pig manure	5	38.16±0.68^{a}	8.35±0.54^{c}	2.46±0.09^{a}	0.93±0.03^{b}
	10	42.80±0.74^{c}	8.14±0.09^{c}	2.81±0.06^{c}	0.98±0.07^{b}
	20	39.52±0.33^{b}	6.69±0.12^{b}	2.65±0.02^{b}	0.72±0.01^{a}
	40	40.03±0.10^{b}	6.11±0.02^{a}	2.59±0.02^{b}	0.74±0.01^{a}
Beef manure	5	30.55±2.24^{a}	2.61±0.92^{a}	1.48±0.13^{a}	0.55±0.08^{ab}
	10	32.85±2.93^{a}	3.64±0.75^{ab}	1.57±0.15^{a}	0.71±0.09^{b}
	20	31.16±0.22^{a}	3.84±0.34^{b}	1.5±0.01^{a}	0.61±0.13^{ab}
	40	32.69±0.38^{a}	4.40±0.10^{b}	1.54±0.05^{a}	0.52±0.01^{a}
Dairy manure	5	35.01±1.36^{a}	6.25±0.44^{b}	1.73±0.01^{a}	0.68±0.03^{b}
	10	37.16±0.83^{b}	6.10±0.11^{b}	1.9±0.05^{b}	0.74±0.04^{c}
	20	34.63±1.04^{a}	4.99±0.14^{a}	1.77±0.02^{a}	0.57±0.01^{a}
	40	34.61±0.24^{a}	4.83±0.02^{a}	1.76±0.03^{a}	0.56±0.02^{a}
Broiler manure	5	38.06±0.66^{a}	6.86±0.44^{b}	5.39±0.12^{a}	1.03±0.02^{a}
	10	40.39±1.39^{b}	6.70±0.11^{b}	5.86±0.16^{b}	1.21±0.08^{b}
	20	38.48±0.68^{a}	5.98±0.08^{a}	5.56±0.15^{a}	0.97±0.02^{a}
	40	38.67±0.15^{a}	5.67±0.07^{a}	5.41±0.08^{a}	0.96±0.01^{a}
Layer manure	5	32.75±1.47^{a}	4.87±0.57^{a}	2.49±0.1^{a}	0.78±0.04^{a}
	10	36.06±1.60^{b}	5.56±0.69^{a}	2.73±0.09^{b}	0.97±0.11^{b}
	20	33.73±0.26^{a}	4.78±0.10^{a}	2.60±0.01^{a}	0.80±0^{a}
	40	34.26±0.65^{ab}	4.77±0.13^{a}	2.58±0.02^{a}	0.81±0.02^{a}

Notes: The same letters denote that particle mass in the same column is not significant ($p > 0.05$). Different letters denote a significant difference in particle mass in the same column ($p < 0.05$).

Compared with the results of crop straws, more complex trends are shown in Tables 4 and 5. All four sample masses had an effect on the AS measurement of livestock manure's C, H, N, and S based on Table 4. In addition, there was no significant difference between 20 mg and 40 mg for all livestock manures, except pig manure's H. However, different sample masses for CS measurement of C had almost no impact on pig manure, broiler manure, and layer manure, and little influence on beef manure and dairy manure, from Table 5. For elements N and S, the significance of different sample masses was noteworthy. From the view of various livestock species, considerable variation was generated from the measured results of the four elements in dairy and beef cattle manure, as shown in Fig. 2, which was less precise than the results of other livestock manures. To ensure the stability of measured results during analysis, the uniformity of sample is critical and an assurance of the complete uniformity of sample is essential.

Table 5. Elemental Analysis of Manure Biomass based on the Principle of Chromatographic Separation

	Mass (mg)	C (%)	H (%)	N (%)	S (%)
Pig manure	2.5	39.60±1.00^{a}	5.73±0.03^{a}	2.57±0.12^{a}	0.80±0.05^{c}
	5	37.76±0.70^{a}	5.73±0.15^{a}	2.92±0.06^{b}	0.61±0.03^{b}
	10	38.64±1.23^{a}	5.45±0.23^{a}	2.68±0.12^{a}	0.39±0.01^{a}
Beef manure	2.5	31.09±0.96^{a}	4.26±0.01^{b}	1.80±0.14^{b}	0.68±0.08^{b}
	5	32.32±0.35^{b}	4.47±0.12^{c}	1.85±0.08^{b}	0.59±0.03^{b}
	10	30.58±0.46^{a}	4.03±0.09^{a}	1.33±0.03^{a}	0.22±0.01^{a}
Dairy manure	2.5	31.12±1.71^{a}	4.04±0.26^{a}	1.72±0.02^{a}	0.74±0.02^{c}
	5	34.10±0.88^{b}	4.87±0.30^{b}	2.00±0.04^{b}	0.61±0.04^{b}
	10	34.01±0.84^{b}	4.30±0.17^{a}	1.67±0.01^{a}	0.25±0.01^{a}
Broiler manure	2.5	37.16±0.04^{a}	5.20±0.02^{b}	5.32±0.16^{a}	0.71±0.05^{b}
	5	36.79±0.48^{a}	5.52±0.10^{c}	5.83±0.14^{b}	0.59±0.04^{ab}
	10	36.55±0.17^{a}	5.05±0.05^{a}	5.65±0.09^{b}	0.62±0.02^{a}
Layer manure	2.5	33.13±0.61^{a}	4.43±0.12^{a}	2.64±0.19^{a}	1.02±0.17^{c}
	5	32.68±0.26^{a}	4.12±0.70^{a}	2.58±0.39^{a}	0.78±0.03^{b}
	10	34.48±1.85^{a}	4.70±0.26^{a}	2.62±0.21^{a}	0.52±0.01^{a}

Notes: The same letters denote that particle mass in the same column is not significant ($p > 0.05$). Different letters denote a significant difference in particle mass in the same column ($p < 0.05$).

The measured results of livestock manure showed a larger overall deviation than that of straw biomass except for element N, as presented in Fig. 2, which may be a result of the structural differentiation and the higher content of N (> 1.5%) compared with crop straw. Given the obtained results, 40 mg for AS is recommended for all the elements' measurement for livestock manures. In addition, 5 mg for CS is recommended for C, H, and N measurement, except for dairy and layer manures.

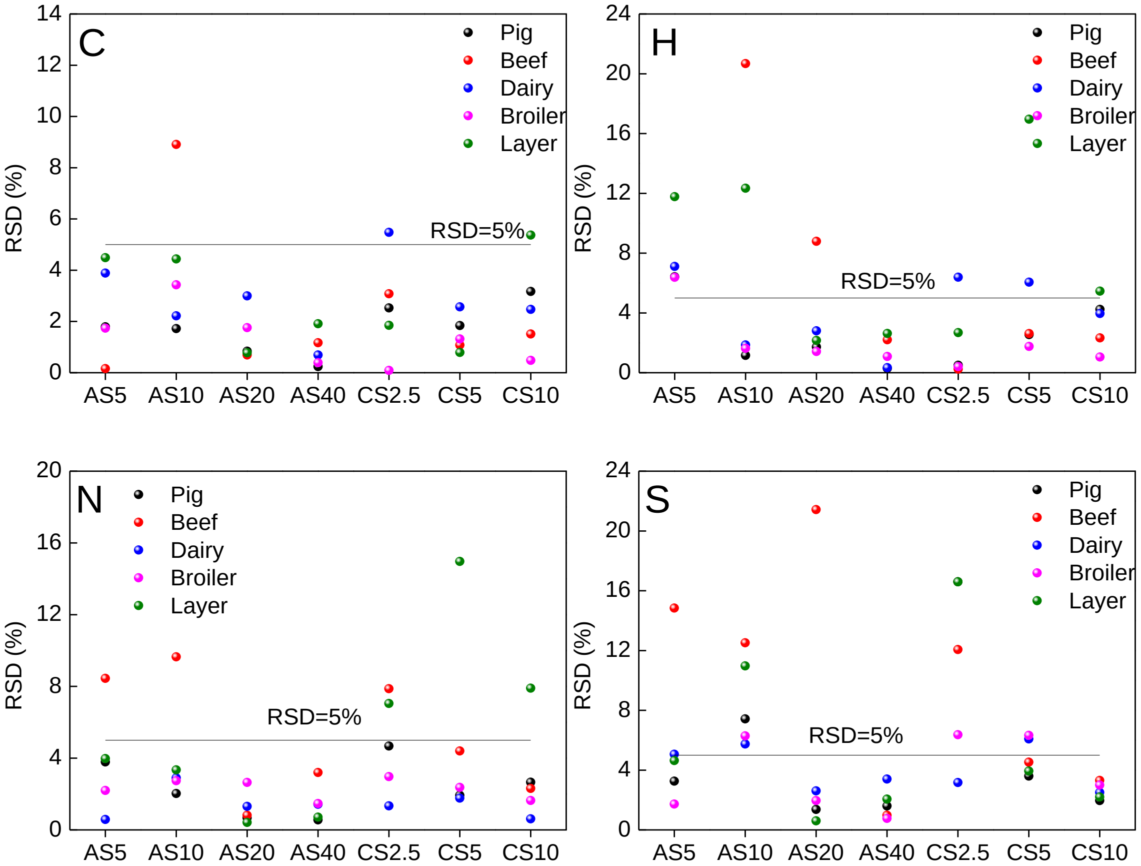

Fig. 2. The corresponding RSD concentration of five livestock manures

Based on the analysis of crop straws and livestock manures, the applicability of different separation analyzers could be matched with the content of elements. Even if the elemental content in agricultural biomass is as low as 0.5%, the AS method with 40 mg is applicable. On the other hand, on the condition of no less than 1.5%, the CS method with only 5 mg could be used for elemental measurement of agricultural biomass with high precision.

CONCLUSIONS

1. Taking complete combustion and sample representativeness into consideration, the optimum parameter combination for instrumental elemental analysis of typical agricultural biomass is as follows: sample particle size of 1.00 mm for crop straw and 0.50 mm for livestock manure.
2. Sample masses of 40 mg for AS and 5 mg for CS are recommended for C, H, and N measurement in crop straw and livestock manure of which the elemental content is above 1.5%. For determination of the minor elements N and S, which are less than 1.5%, it is more suitable to use 40 mg for AS.
3. The measured results of beef and dairy cattle manures were less precise. It is recommended that samples should be kept free of impurities and completely homogeneous, especially for livestock manure.

4. On the basis of correct and precise method for determination of elemental analysis recommended above, the results of elemental analysis regarding agricultural biomass could be reliable and authoritative.

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REFERENCES CITED

AOAC 2001.11 (2001). “Protein (crude) in animal feed, combustion method,” Association of Official Analytical Chemists, Arlington, VA.

ASTM D5373-14 (2014). “Standard test methods for measurement of carbon, hydrogen and nitrogen in analysis samples of coal and carbon in analysis samples of coal and coke,” ASTM International, West Conshohocken, PA.

ASTM E775-87 (2008). “Standard test method for total sulfur in the analysis sample of refuse-derived fuel,” ASTM International, West Conshohocken, PA.

ASTM E777-08 (2008). “Standard test method for carbon and hydrogen in the analysis sample of refuse-derived fuel,” ASTM International, West Conshohocken, PA.

ASTM E1757-01 (2007). “Standard practice for preparation of biomass for compositional analysis,” ASTM International, West Conshohocken, PA.

BS EN 15289 (2011). “Solid biofuels - Measurement of total content of sulphur and chlorine,” The European Committee for Standardization, Brussels, Belgium.

Eksperiandova, L. P., Fedorov, O. I., and Stepanenko, N. A. (2011). “Estimation of metrological characteristics of the element analyzer EuroVector EA-3000 and its potential in the single-reactor CHNS mode,” *Microchemical Journal* 99(2), 235-238. DOI: 10.1016/j.microc.2011.05.005

Gazulla, M. F., Rodrigo, M., Orduna, M., and Gomez, C. M. (2012). “Determination of carbon, hydrogen, nitrogen and sulfur in geological materials using elemental analysers,” *Geostandards and Geoanalytical Research* 36(2), 201-217. DOI: 10.1111/j.1751-908X.2011.00140.x

GB/T 21923 (2008). “General testing rules for solid biofuels,” Standardization Administration of China, Beijing, China.

GB/T 28732 (2012). “Determination of total sulfur in solid biofuels,” Standardization Administration of China, Beijing, China.

GB/T 28734 (2012). “Determination of carbon and hydrogen in solid biofuels,” Standardization Administration of China, Beijing, China.

ISO 334 (2013). “Solid mineral fuels. Measurement of total sulphur. Eschka method,” International Organization for Standardization, Geneva, Switzerland.

ISO 5983-1 (2009). “Animal feeding stuffs - Measurement of nitrogen content and calculation of crude protein content - Part 1: Kjeldahl method,” International Organization for Standardization, Geneva, Switzerland.

ISO 16634-1 (2008). “Food products - Measurement of the total nitrogen content by combustion according to the Dumas principle and calculation of the crude protein content,” International Organization for Standardization, Geneva, Switzerland.

NREL/TP-510-42620 (2008). “Preparation of samples for compositional analysis: Laboratory Analytical Procedure,” National Renewable Energy Laboratory, Golden, CO.

Oasmaa, A., and Meier, D. (2005). “Norms and standards for fast pyrolysis liquids—1. Round robin test,” *Journal of Analytical and Applied Pyrolysis* 73(2), 323-334. DOI: 10.1016/j.jaap.2005.03.003

Parikh, J., Channiwala, S. A., and Ghosal, G. K. (2007). “A correlation for calculating elemental composition from proximate analysis of biomass materials,” *Fuel*. 86(12-13), 1710-1719. DOI: 10.1016/j.fuel.2006.12.029

Ryba, S. A., and Burgess, R. M. (2002). “Effects of sample preparation on the measurement of organic carbon, hydrogen, nitrogen, sulfur, and oxygen concentrations in marine sediments,” *Chemosphere* 48(1), 139-147. DOI: 10.1016/S0045-6535(02)00027-9

Shen, X. L., Huang, G. Q., Yang, Z. L., and Han, L. J. (2015). “Compositional characteristics and energy potential of Chinese animal manure by type and as a whole,” *Applied Energy* 160,108-119. DOI: 10.1016/j.apenergy.2015.09.034

Sieper, H. P., Kupka, H. J., Williams, T., Rossmann, A., Rummel, S., Tanz, N., and Schmidt, H. L. (2006). “A measuring system for the fast simultaneous isotope ratio and elemental analysis of carbon, hydrogen, nitrogen and sulfur in food commodities and other biological material,” *Rapid Communications in Mass Spectrometry* 20(17) 2521-2527. DOI: 10.1002/rcm.2619

SN EN 15104 (2011). “Solid biofuels -Measurement of total content of carbon, hydrogen and nitrogen — Instrumental methods,” The European Committee for Standardization, Brussels, Belgium.

TMECC (2000). “Test methods for the examination of composting and compost,” US Composting Council, Bethesda, MD, USA.

Vassilev, S. V., Baxter, D., Andersen, L. K., and Vassileva, C. G. (2010). “An overview of the chemical composition of biomass,” *Fuel* 89(5), 913-933. DOI: 10.1016/j.fuel.2009.10.022

Vassilev, S. V., Baxter, D., Andersen, L. K., and Vassileva, C. G. (2013). “An overview of the composition and application of biomass ash. Part 1. Phase-mineral and chemical composition and classification,” *Fuel* 105, 40-76. DOI: 10.1016/j.fuel.2012.10.001

Wang, Y. J., Bi, Y. Y., and Gao, C. Y. (2010). “The assessment and utilization of straw resources in China,” *Agricultural Sciences in China* 9(12), 1807-1815. DOI: 10.1016/s1671-2927(09)60279-0

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