基于Kjeldahl与Dumas方法的农作物秸秆总氮含量分析

王鑫磊，袁湘汝，张潇潇，解茹越，肖卫华，韩鲁佳※

（中国农业大学工学院，北京 100083）

摘 要：凯氏定氮法（Kjeldahl）与杜马斯燃烧法（Dumas）是测定农业生物质总氮含量的主要检测手段，但二者的测定结果数值存在差异。该研究获取农作物秸秆样本（水稻、小麦、玉米、油菜和棉花）共计1 179个，分别采用Kjeldahl和Dumas方法测定总氮（TKN和TCN，total Kjeldahl nitrogen and total combustion nitroyen）含量，通过多种统计与分析方法，系统分析比较了不同农作物秸秆总氮含量及其分布的异同和相关关系。结果表明：不同农作物秸秆氮含量分布均呈非正态分布，建议采用中位数统计；5种秸秆总体的TKN质量分数为（7.12 ± 1.87）g/kg，TCN质量分数为（8.00 ± 2.13）g/kg，TKN含量显著小于TCN含量；小麦和棉花秸秆的TKN含量和TCN含量与其他秸秆间均存在显著差异（$P < 0.05$）；不同生物质TKN含量与TCN含量关系不同，建议采用最小中位数二乘法进行拟合分析。研究结果可为农作物秸秆科学利用提供数据及方法互通性支撑。

关键词：秸秆；氮；模型；凯氏定氮法；杜马斯燃烧法；分布分析；中位数；线性相关关系

doi: 10.11975/j.issn.1002-6819.2020.06.024

中图分类号：X71；S11，S548 **文献标志码**：A **文章编号**：1002-6819(2020)-06-0206-09

王鑫磊，袁湘汝，张潇潇，解茹越，肖卫华，韩鲁佳. 基于Kjeldahl与Dumas方法的农作物秸秆总氮含量分析[J]. 农业工程学报，2020，36(6)：206－214. doi: 10.11975/j.issn.1002-6819.2020.06.024 http://www.tcsae.org

Wang Xinlei, Yuan Xiangru, Zhang Xiaoxiao, Xie Ruyue, Xiao Weihua, Han Lujia. Analysis of the total nitrogen content of crop residues determined by using Kjeldahl and Dumas methods[J]. Transactions of the Chinese Society of Agricultural Engineering (Transactions of the CSAE), 2020, 36(6): 206－214. (in Chinese with English abstract)

doi: 10.11975/j.issn.1002-6819.2020.06.024 http://www.tcsae.org

0 引 言

氮作为生命元素之一，广泛存在于自然界中，在物质代谢与能量转化过程中有着不可替代的作用。在大气污染[1－2]、温室气体[3]、水体富营养、土壤肥力[4－5]、植物生长[6]、食品营养[7]、饲料蛋白[8]等领域，氮均扮演着十分重要的角色[9]。

环境污染、资源短缺的今天，资源储量巨大、可再生的生物质资源已引起人们的广泛关注。农作物秸秆等农业生物质因富含氮、碳、磷、钾等营养元素，众多学者对其不同的基础特性[10-12]及其饲料化、肥料化、能源化等利用技术[13-16]进行了深入的探索与研究。其中，物料总氮的含量是指导农作物秸秆等农业生物质科学利用的重要参数。

目前，氮含量测定最为普遍的方法为凯氏定氮法和杜马斯燃烧法。凯氏定氮法的工作原理是利用浓硫酸消解样品，将氮素转换为NH_4^+，后通过酸碱滴定测定NH_4^+含量，并据此推算出样品氮含量，对样品均匀性要求小[9]，在医学、植物学、食品等学科中应用普遍。杜马斯燃烧法的工作原理是将样品在900 ~ 1 200 ℃下燃烧后，通过将混合气体中氮氧化物还原为氮气，测定含量并计算氮素含量，具有检测速度快、环境污染少、检测误差小等特点[17-21]，现已成为国际通用的检测手段[18]。由于测试原理的不同，凯氏定氮法与杜马斯燃烧法的测定结果存在显著差异。针对蔬菜叶[20]、油料[22]、乳制品[23]、豆制品[24]、鲜花[25]、谷物[26]、肉制品[27]等不同物料的研究结果显示，2种方法所得出的氮含量普遍存在线性相关关系。但是由于被测物料特性不同，这种线性相关关系也存在明显的差别。

因此，本研究以产出量巨大、分布十分广泛的水稻、小麦、玉米、棉花和油菜秸秆为研究对象，分别采用凯氏定氮法与杜马斯燃烧法测定不同农作物秸秆总氮含量，分析其含量变化及其数值分布，并深入比较上述不同测定方法所得氮含量的异同及其相关关系。以期为农作物秸秆科学利用提供数据及方法互通性支撑。

1 材料与方法

1.1 样本及制备

在中国大陆地区11个省级行政区（北京、天津、河北、山东、山西、陕西、甘肃、宁夏、青海、西藏和新疆）获取代表性农作物秸秆样本共1 179个，其中水稻秸秆134个、小麦秸秆526个、玉米秸秆342个、油菜秸秆72个、棉花秸秆105个，去除谷物与根，仅保留秸秆中间的部分，样本采集数量的区域分布如图1所示。每个样本的收到基质量不少于2 kg。将收集到的样本切短后置于烘箱中在45 ℃的条件下烘干36 ~ 48 h，然后用粉碎机粉碎至粒径1 mm，过1 mm筛后封存作为干基样本备用。

收稿日期：2020-01-08 修订日期：2020-02-18

基金项目：现代农业（奶牛）产业技术体系建设专项资金（CARS－36）；国家重点研发计划（2016YFE0112800）；公益性行业（农业）科研专项资金（201003063）

作者简介：王鑫磊，博士生，主要从事生物质数据挖掘研究。

Email: wangxinlei917@ cau.edu.cn。

※通信作者：韩鲁佳，教授，博士，博士生导师，长江学者特聘教授，主要从事农业生物质工程研究。Email: hanlj@cau.edu.cn。

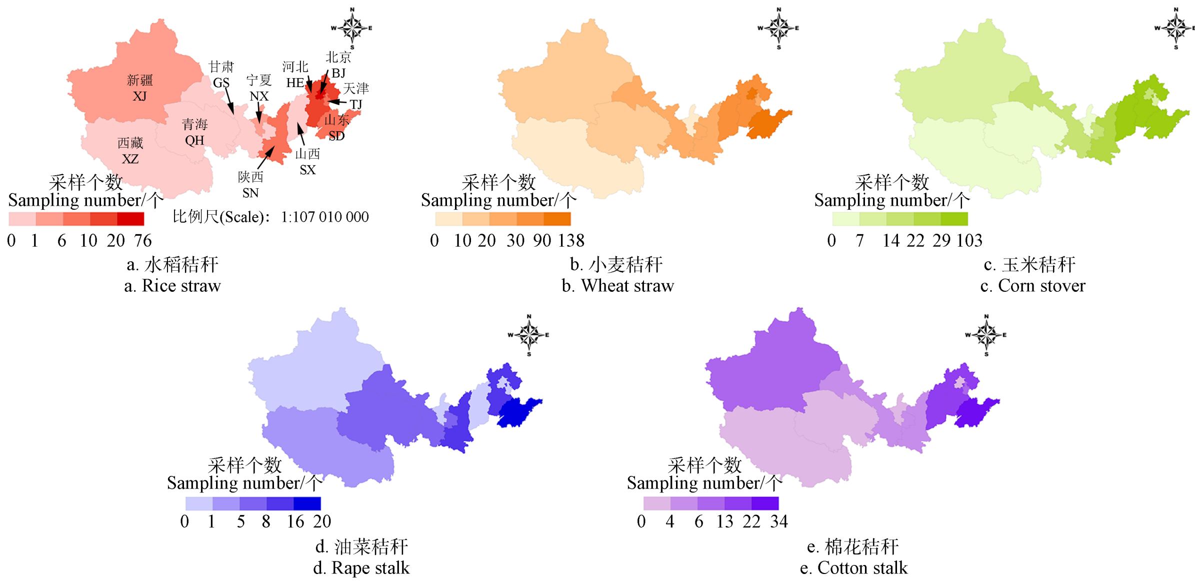


图1　农作物秸秆样本采集数量区域分布图

Fig.1　Distributions of sample collection quantity for different crop residues

1.2　农作物秸秆氮含量的测定

1.2.1　凯氏定氮法

采用AOAC方法[28]。测定时，称量5 g左右样本，加入7.8 g混合催化剂（7 g K_2SO_4 和0.8 g $CuSO_4 \cdot 5H_2O$），并加入12 mL浓硫酸，在200℃下消化1 h后升温到420 ℃消化至溶液澄清。凯氏定氮仪（FOSS 2300）设定参数为：水70 mL、40%氢氧化钠60 mL、1%硼酸30 mL。凯氏定氮法测定的氮含量记为TKN。

1.2.2　杜马斯燃烧法

采用AOAC方法[29]。称量约40 mg样品并用锡箔包被，设定元素分析仪（Elementar, vario MACRO）参数为：燃烧管1 150 ℃、还原管850 ℃、通氧速率100 mL/min、通氧时间90 s、吹扫气（He）速率500 mL/min，并进行自动检测。杜马斯燃烧法测定的氮含量记为TCN。

1.3　数据处理与统计分析

线性拟合可以分析2参数之间的相关关系。普通最小二乘（OLS）和正交回归（ordinary least square, Orth）线性拟合采用Minitab软件（版本：19.1）进行分析，最小中位数二乘（least median of square regression, LMS）线性拟合采用MASS程序包（版本：7.3-51.4，R版本：3.6.1）进行分析。OLS拟合方法仅假设因变量 y 存在误差，由于本研究中作为自变量 x 的TCN同样存在误差，因此同时采用Orth方法对TKN与TCN的相关关系进行拟合。此外，OLS拟合方法适用于数据不存在异常值点的情况下。数据中异常值点的存在会干扰OLS的拟合结果，使得结果难以反映拟合关系的真实信息，因此采用LMS方法进行对比。研究通过AOAC方法中凯氏定氮法与杜马斯燃烧法给出的 RSD_r 计算得出正交回归中误差方差比（EDR）。

$$EDR = (mean(RSD_{r,\ TKN}) / mean(RSD_{r,\ TCN}))^2 \qquad (1)$$

式中 $mean(RSD_{r,\ TKN})$ 表示凯氏定氮法所给出实验室内部标准偏差 RSD_r 的均值，$mean(RSD_{r,\ TCN})$ 表示杜马斯燃烧法所给出 RSD_r 的值。

数据分布分析及图片绘制采用Origin软件（版本：95C），其中分布分析使用常见的Normal、Lognormal、Gamma、Weibull、Exponential、Laplace和Lorentz分布进行对比。利用psych程序包（版本：1.8.12，R版本：3.6.1）进行数据统计分析。利用SPSS（版本：23.0）软件对数据进行两两比较和多重比较，两两比较方法采用Student’s t 检验多重比较方法采用Game - Howell检验，以适应样品总量不相等且方差非齐次的多重比较[11]，同时采用Mann - Whitney U检验和Kruskal – Wallis检验进行非参数两两比较和多重比较。地图底板按照2012年中国政区图绘制。

2　结果与分析

2.1　不同农作物秸秆氮含量及其分布

2.1.1　数据分布

不同农作物秸秆TKN及TCN数据分布如图2、图3所示。图2结果表明，5种农作物秸秆TKN含量均不呈正态分布，其中水稻秸秆、油菜秸秆和棉花秸秆近似服从Lognormal分布，而小麦秸秆和玉米秸秆近似服从Gamma分布。图3结果表明，5种农作物秸秆TCN含量同样均不呈正态分布，其中水稻秸秆和油菜秸秆近似服从Lognormal分布，小麦秸秆近似服从Gamma分布，玉米秸秆和棉花秸秆近似服从Weibull分布。5种农作物秸秆的TKN和TCN总体均近似服从Lognormal分布。对比图2和图3可以发现，水稻秸秆、小麦秸秆、油菜秸秆和秸秆总体的TKN和TCN含量的分布相同，而玉米秸秆和棉花秸秆的TKN和TCN含量分布存在差异。因此，在分析不同农作物秸秆TKN和TCN的差异及关系的同时，也应考虑到数据非正态分布对分析结果的影响。

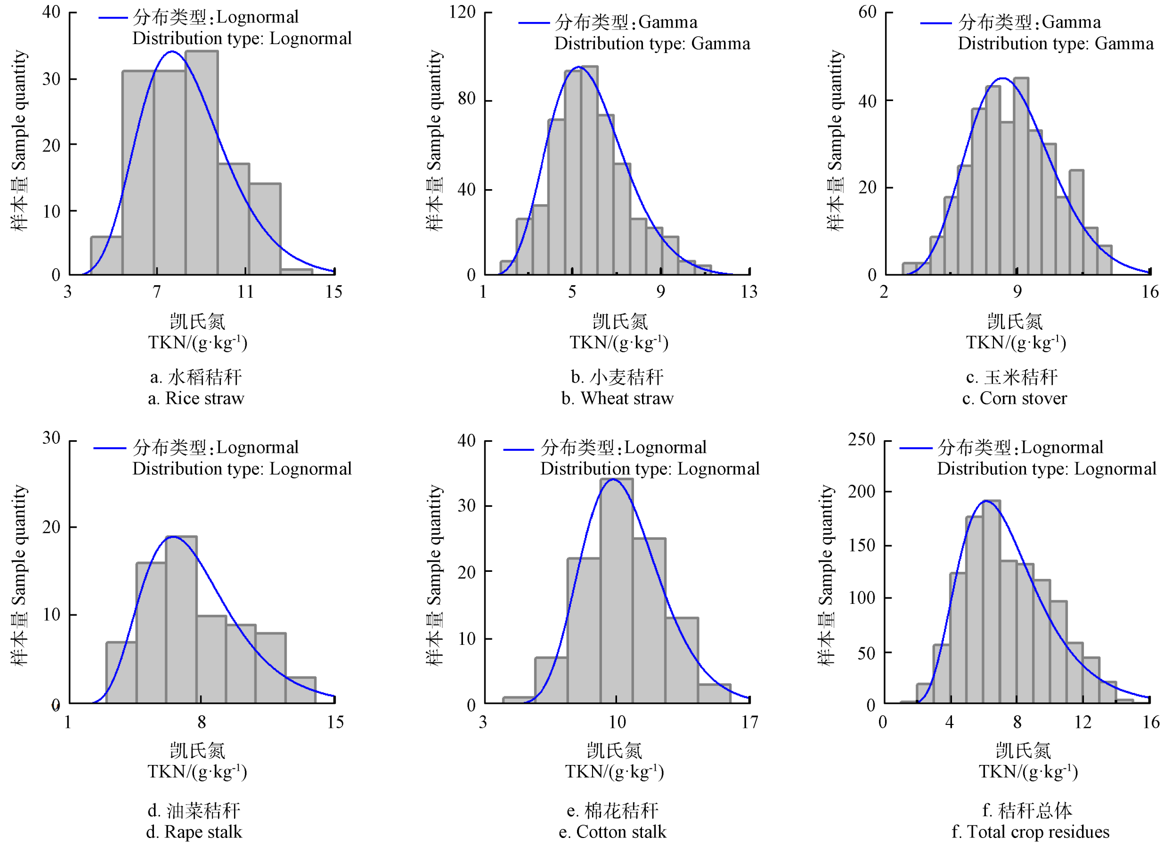


图2　不同农作物秸秆凯氏氮分布直方图

Fig.2　Histograms of TKN distribution in different crop residues

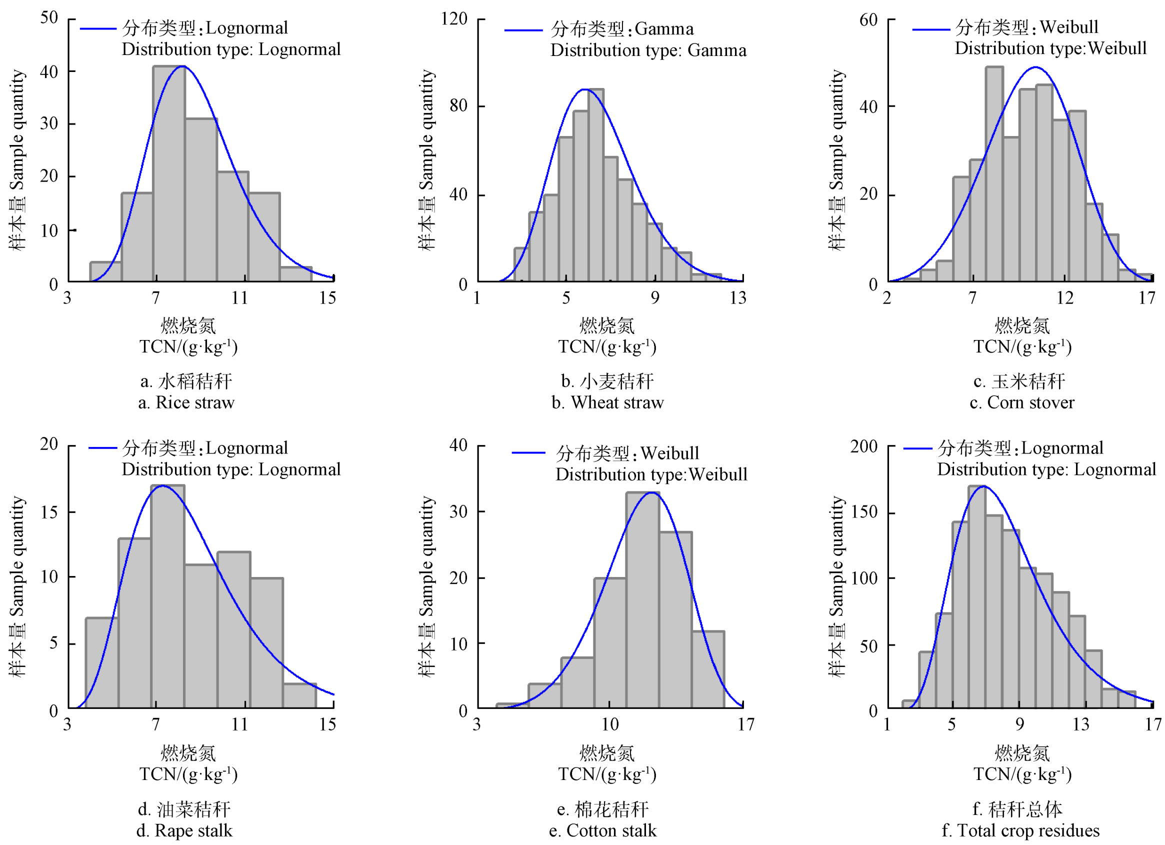


图3　不同农作物秸秆燃烧氮分布直方图

Fig.3　Histograms of TCN distribution in different crop residues

2.1.2　统计分析

不同农作物秸秆TKN和TCN含量的统计结果如表1所示，氮含量的箱型分布如图4所示。由表1可见，5种农作物秸秆的TKN均值在5.82～10.43 g/kg之间，TCN均值在

6.39～11.64 g/kg之间。除水稻秸秆外，各种农作物秸秆的TKN和TCN均存在显著差异（$P < 0.05$）。其中，TKN整体分布低于TCN（图4），可能是常规的凯氏定氮法难以将杂环化合物中的有机氮，以及N－N和N－O键中的氮元素进行还原，例如核酸、硝态氮与亚硝态氮[30－31]；而杜马斯燃烧法因在高温条件下可将所有氮素转化为氮氧化物，进而转化为氮气进行检测[32－33]，不仅可以检测到样本中有机态氮和铵态氮，而且也可以检测出硝态氮和亚硝态氮[21]。

表1　五种典型农作物秸秆凯氏氮与燃烧氮含量及其比较

Table 1　Content and comparison of TKN and TCN in five typical crop residues

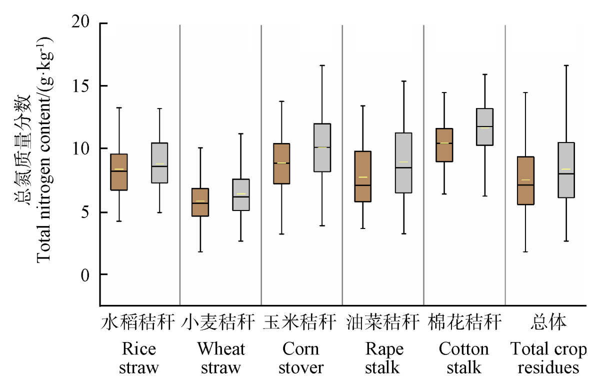
种类 Types	样本量 Sample quantity	凯氏氮 TKN /(g·kg^{-1})				燃烧氮 TCN/(g·kg^{-1})			
		均值±标准差 Mean ± SD	中位数±绝对中位差 Median ± MAD	取值范围 Range	四分位距 IQR	均值±标准差 Mean ± SD	中位数±绝对中位差 Median ± MAD	取值范围 Range	四分位距 IQR
水稻秸秆 Rice straw	134	8.37 ± 2.00Abc	8.20 ± 1.42Abc	4.24 ~ 13.24	2.82	8.78 ± 1.95Ab	8.59 ± 1.45Ab	4.93 ~ 13.18	3.14
小麦秸秆 Wheat straw	526	5.82 ± 1.76Aa	5.66 ± 1.07Aa	1.81 ~ 11.19	2.19	6.39 ± 1.85Ba	6.17 ± 1.17Ba	2.66 ~ 11.54	2.45
玉米秸秆 Corn stover	342	8.84 ± 2.23Ac	8.82 ± 1.59Ac	3.23 ~ 13.77	3.18	10.06 ± 2.50Bc	10.10 ± 1.91Bc	3.90 ~ 16.60	3.81
油菜秸秆 Rape stalk	72	7.72 ± 2.60Ab	7.10 ± 1.87Ab	3.68 ~ 13.41	3.89	8.93 ± 3.00Bb	8.50 ± 2.45Bb	3.26 ~ 15.38	4.69
棉花秸秆 Cotton stalk	105	10.43 ± 2.03Ad	10.42 ± 1.45Ad	4.55 ~ 15.83	2.63	11.64 ± 2.23Bd	11.75 ± 1.48Bd	4.50 ~ 15.92	2.93
秸秆总体 Total	1 179	7.51 ± 2.58A	7.12 ± 1.87A	1.81 ~ 15.83	3.76	8.35 ± 2.88B	8.00 ± 2.13B	2.66 ~ 16.60	4.40

注：MAD：绝对中位差，IQR：四分位距；A, B代表同一种类农作物秸秆两种氮素间的差异显著性（$P < 0.05$），a, b, c, d, e代表不同种类的农作物秸秆间差异显著性（$P < 0.05$）；“Mean ± SD”采用Student’s t和Games－Howell方法进行氮素间显著性差异和秸秆间多重比较的显著性，“Median ± MAD”采用Mann－Whitney U和Kruskal－Wallis方法进行氮素间两两比较和秸秆间多重比较的显著性。

Note: SD: standard deviation, MAD median absolute deviation, IQR: interquartile range; A, B represents the significant difference between two kinds of nitrogen content in the same crop residues ($P < 0.05$). a, b, c, d, e represents the significant difference between different types of crop residues ($P < 0.05$). “Mean ± SD” used Student’s t test and Games－Howell test to compare the significant difference between two kinds of nitrogen content, and the multiple comparison between different types of crop residues. “Median ± MAD” used Mann－Whitney U test and Kruskal－Wallis test to compare the significant difference between two kinds of nitrogen content, and the multiple comparison between different types of crop residues.

此外，部分种类秸秆间的氮含量也存在显著的差异（$P < 0.05$）。就TKN而言，小麦秸秆的氮含量最低，水稻秸秆与油菜秸秆、玉米秸秆的氮含量差异不显著，棉花秸秆的氮含量最高，不同的秸秆中氮含量由小到大排列依次为小麦秸秆＜油菜秸秆＜水稻秸秆＜玉米秸秆＜棉花秸秆；而就TCN而言，氮含量由小到大依次为小麦秸秆＜水稻秸秆＜油菜秸秆＜玉米秸秆＜棉花秸秆，油菜秸秆的氮含量显著高于水稻秸秆，这与TKN的结果不同。与之前的研究对比，凯氏定氮法与杜马斯燃烧法对秸秆氮元素的测定结果与Chen等[34]和Niu等[10]的研究结果相似。

中位数统计结果与均值统计的显著性结果相同，但是由于农作物秸秆的TKN与TCN含量数据不服从正态分布，因此中位数与均值存在差值（表1，图4）。此外，中位数统计结果显示不同秸秆的TKN与TCN含量排序相同，与TKN均值排列结果相同。与均值相比，在非正态分布下中位数统计结果受到极端值影响较少，建议采用中位数形式对农作物秸秆的TKN与TCN含量进行统计分析。



注：左侧为TKN,右侧为TCN。

Note: TKN on the left and TCN on the right.

图4　不同农作物秸秆凯氏氮与燃烧氮箱型图

Fig.4　Box plots of TKN and TCN in different crop residues

2.2　不同农作物秸秆TKN与TCN的相关关系及比较分析

TKN与TCN的相关关系分别采用OLS、LMS和Orth方法对2参数进行分析，线性拟合结果如表2、图5所示，其中Orth的EDR为3.70。可以看出，不同农作物秸秆TKN与TCN含量存在良好的线性相关关系，但不同农作物秸秆TKN与TCN含量的回归方程拟合结果存

在差异（斜率和截距均不同）。5种农作物秸秆的OLS回归方程斜率介于0.72～0.91之间，Orth回归方程斜率均高于对应OLS斜率，取值在0.77～0.96之间。除水稻秸秆与棉花秸秆外，各种农作物秸秆的LMS斜率均高于OLS和Orth，取值在0.79～0.98之间。这3种方法所得的R^2相等，拟合效果相同。但观察图5可以发现，相比于OLS和Orth，LMS所得结果穿越了样本点密度更高的区域，而OLS则向相对样本点相对稀疏的地方偏移，Orth介于二者之间。样本密度越高，表明测量结果分布在该区域的概率越高，而穿过这些区域的拟合结果受到异常点的影响越小。因此建议采用LMS方法对TKN和TCN进行相关关系分析。

表2 农作物秸秆凯氏氮和燃烧氮含量线性拟合结果

Table 2　Linear fitting results of TKN and TCN contents in crop residues

种类 Types	拟合方法 Fitting methods	拟合结果 Fitting results	R^2
水稻秸秆 Rice straw	普通最小二乘 OLS	TKN = 0.91TCN + 0.42	0.78
	正交回归 Orth	TKN = 0.96TCN - 0.02	0.78
	最小中位数二乘 LMS	TKN = 0.96TCN + 0.54	0.78
小麦秸秆 Wheat straw	普通最小二乘 OLS	TKN = 0.86TCN + 0.35	0.81
	正交回归 Orth	TKN = 0.89TCN + 0.13	0.81
	最小中位数二乘 LMS	TKN = 0.98TCN - 0.50	0.81
玉米秸秆 Corn stover	普通最小二乘 OLS	TKN = 0.72TCN + 1.64	0.65
	正交回归 Orth	TKN = 0.77TCN + 1.13	0.65
	最小中位数二乘 LMS	TKN = 0.79TCN + 0.98	0.65
油菜秸秆 Rape stalk	普通最小二乘 OLS	TKN = 0.75TCN + 1.06	0.74
	正交回归 Orth	TKN = 0.78TCN + 0.74	0.74
	最小中位数二乘 LMS	TKN = 0.82TCN + 0.97	0.74
棉花秸秆 Cotton stalk	普通最小二乘 OLS	TKN = 0.74TCN + 1.79	0.67
	正交回归 Orth	TKN = 0.79TCN + 1.20	0.67
	最小中位数二乘 LMS	TKN = 0.78TCN + 1.54	0.67
总体 Total	普通最小二乘 OLS	TKN = 0.81TCN + 0.72	0.82
	正交回归 Orth	TKN = 0.84TCN + 0.49	0.82
	最小中位数二乘 LMS	TKN = 0.96TCN - 0.20	0.82

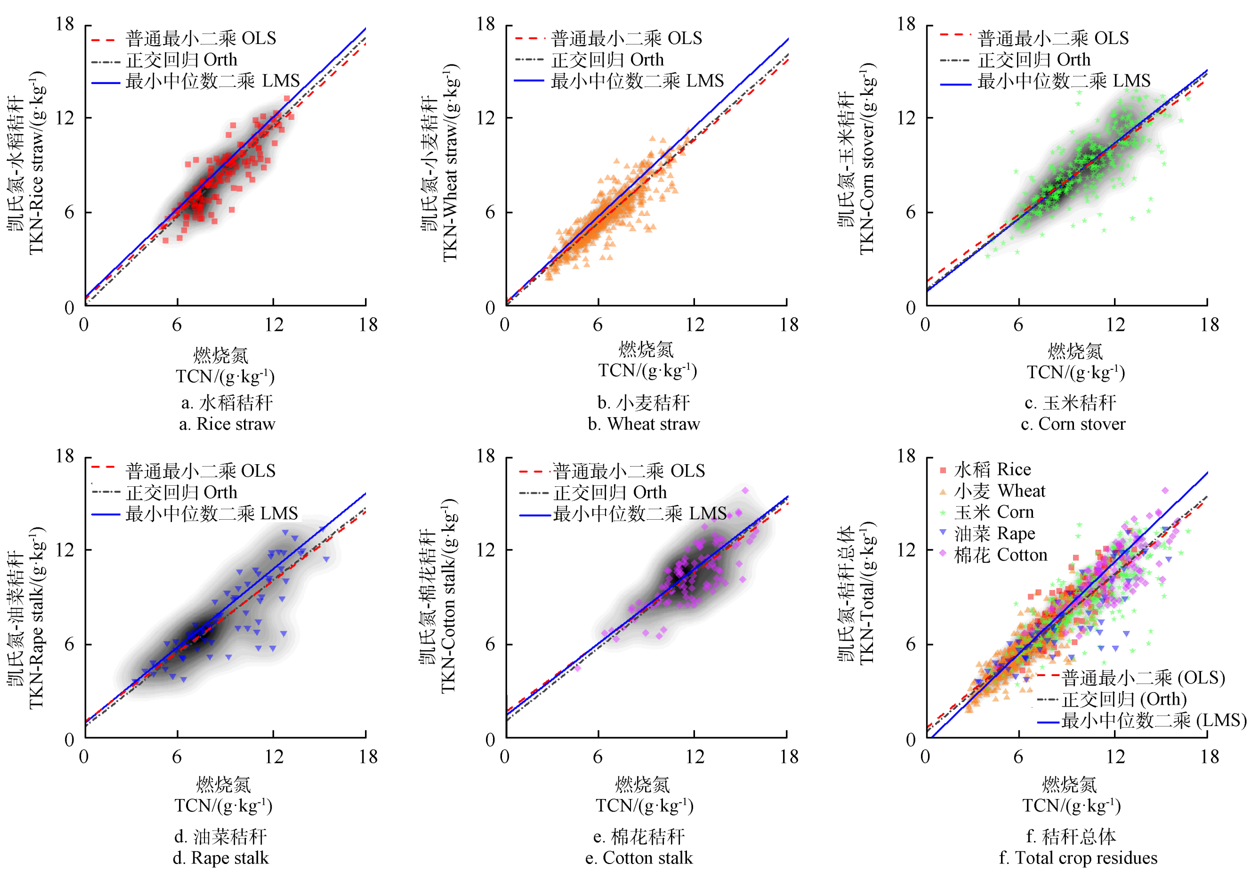


图5　农作物秸秆TKN和TCN线性拟合结果核密度-散点图

Fig.5　Kernel density-scatter plots of linear fitting results for TKN and TCN in crop residues

2.3　不同物质的TKN与TCN的相关关系及比较分析

本研究结果与已有文献研究结果的比较如表3所示。由于大部分研究结果均采用OLS方法进行回归拟合，因此本研究依然基于OLS的拟合结果进行分析讨论。由表3可以看出，针对不同被测物料，TKN与TCN线性回归方程的斜率处于0.37 ~ 1.74之间，差异较大。大部分植物类的原料其线性拟合的斜率在0.68 ~ 0.94之间，本文的研究与之相似。相对其它类的生物质，植物类的生物质拟合斜率较低，可能是植物样本中硝态氮的含量较高，当高于0.70%时，TKN测得的结果与TCN有较为明显的差距[35 - 36]，而施用硝酸根为氮素的肥料会使得蔬菜等作物中含有较高的硝态氮[19]。此外叶片中的TKN与TCN含量线性回归方程的斜率在0.68 ~ 0.86之间，其TCN的含量相对于TKN来说较高，可能是叶片中含有较多的叶绿素等物质，含有较多的杂环态氮及硝态氮。畜禽粪便等生物质的TKN与TCN含量斜率与蔬果等植物类含量相比较高[37]，可能是由于其硝酸盐含量多数低于0.40%[21]，TKN与TCN测得的结果差距不明显。物料中含量过低，接近仪器的检出限，会导致斜率远大于1.00，如酸性洗涤纤维[21]。

表3　不同物料凯氏氮与燃烧氮含量的线性回归拟合结果及比较

Table 3　Linear regression results and comparison of total nitrogen content values determined by Kjeldahl method and combustion method (TKN and TCN) in different materials

文献 References	样本量 Sample quantity	拟合结果 Fitting results	R^2	样本种类 Sample types
[35]	433	TKN = 0.94 × TCN + 0.60	0.99	花期水稻秸秆，分蘖期水稻秸秆，成熟期水稻和水稻秸秆，杂草和部分豆科植物
[19]	60	TKN = 0.86 × TCN + 0.15	0.98	萝卜叶
[25]	397	TKN = 0.90 × TCN + 0.90	0.93	观赏植物和草坪草种
[20]	134	TKN = 0.68 × TCN + 3.30	0.70	芦笋，甜瓜，青椒，黄瓜，茄子，爱尔兰马铃薯，秋葵，草莓，甜玉米，夏南瓜，南豌豆，甘薯，番茄和西瓜
[32]	-	TKN = 0.78 × TCN + 1.15	0.86	柚子叶
	-	TKN = 0.87 × TCN - 0.38	0.96	柚子水果部分
[21]	89	TKN = 0.97 × TCN - 1.30	0.96	果园杂草，紫花苜蓿，玉米叶，玉米粒，玉米青贮饲料，小麦，生菜，番茄，马铃薯，天竺葵，豆粕，混合饲用谷物，豆类/草青贮饲料，污泥，动物粪便，饲料中的酸性洗涤剂纤维
	37	TKN = 0.87 × TCN + 1.43	0.93	果园杂草，紫花苜蓿，玉米叶，玉米粒，青贮玉米，小麦，生菜，番茄，马铃薯，天竺葵
	6	TKN = 1.74 × TCN - 1.74	0.64	酸性洗涤纤维
	12	TKN = 1.02 × TCN - 0.76	1.00	玉米、小麦和混合饲用谷物
	34	TKN = 1.07 × TCN - 5.96	0.95	污泥和粪便
[17]	-	TKN = 0.92 × TCN + 5.83	0.99	甘蔗，甘蔗与纸浆青贮，海岸十字干草，青贮玉米，提夫顿干草，含羞草，紫荆，青兰，含羞草
	-	TKN = 0.94 × TCN + 3.19	0.72	玉米，柑橘果肉，木薯皮与果肉青贮，木薯废料与果肉青贮，麦粉
	-	TKN = 0.86 × TCN + 32.82	0.97	鱼粉，内脏粉，豆粕，棉籽粕，家禽粪便和纸浆
[42]	14	TKN = 0.92 × TCN - 0.28	0.99	饲料
[43]	10	TKN = 0.97 × TCN + 1.65	-	鱼粉
[23]	41	TKN = 1.00 × TCN - 0.57	1.00	饲料
	20	TKN = 0.98 × TCN + 0.08	0.99	粪便
	3	TKN = 1.14 × TCN - 6.51	0.99	尿液
	8	TKN = 0.37 × TCN + 16.74	0.41	蛋黄
	44	TKN = 0.49 × TCN + 15.15	0.38	动物尸体
	126	TKN = 0.99 × TCN + 0.10	1.00	饲料，粪便，尿液，蛋黄，动物尸体
[18]	9	TKN = 0.92 × TCN + 2.20	0.89	蛋鸡粪便
	4	TKN = 1.05 × TCN - 2.42	1.00	肉鸡粪便
	9	TKN = 0.87 × TCN + 1.96	0.98	生猪粪便
	10	TKN = 0.98 × TCN + 0.21	0.99	奶牛粪便
	7	TKN = 0.87 × TCN + 1.71	0.98	肉牛粪便
	39	TKN = 0.96 × TCN + 0.49	0.99	蛋鸡粪便，肉鸡粪便，生猪粪便，奶牛粪便，肉牛粪便
[26]	240	TKN = 1.01 × TCN + 0.00	0.99	小麦
[37]	101	TKN = 1.00 × TCN - 0.90	0.98	乳制品，饲料，婴儿配方奶粉，谷物，肉类
[23]	9	TKN = 0.81 × TCN + 1.67	0.99	牛奶
[24]	-	TKN = 0.97 × TCN - 0.01	1.00	大豆分离蛋白，大豆浓缩蛋白，脱脂大豆薄片，大豆分离蛋白凝乳，大豆，豆腐，蛋白提取物，豆浆和乳清
[44]	150	TKN = 0.95 × TCN - 0.45	0.99	土壤
本研究 In this research	134	TKN = 0.91 × TCN + 0.42	0.78	水稻秸秆
	526	TKN = 0.86 × TCN + 0.35	0.81	小麦秸秆
	342	TKN = 0.72 × TCN + 1.64	0.65	玉米秸秆
	72	TKN = 0.75 × TCN + 1.06	0.74	油菜秸秆
	105	TKN = 0.74 × TCN + 1.79	0.67	棉花秸秆
	1 179	TKN = 0.81 × TCN + 0.72	0.82	水稻秸秆，小麦秸秆，玉米秸秆，油菜秸秆，棉花秸秆

如前所述，常规的凯氏定氮法可以检测样本中的有机态氮和铵态氮，而硝态氮和亚硝态氮则无法检测出来[6－9, 36－38]。这可能是由于凯氏定氮法在反应过程中无法将硝态氮完全转化为铵态氮检测，而一部分由于硝酸盐由于转化为硝酸，而且自由碳原子会将硝酸根还原为氮气[39]，在凯氏定氮法的反应温度下汽化、分解或转化为氮气挥发出去而无法测量[19]。另有部分学者认为仅硝酸盐还无法完全解释TKN与TCN含量的差异，核酸氮也可能有所不同[20]。此外，高分子化合物阻碍凯氏定氮法流程中的酸碱滴定也是可能致使TKN含量低的原因[40]。不同的物料中氮素的形态、含量各不相同[41]，也会直接导致TKN与TCN线性回归方程斜率和截距也有较大的差距。

3 结 论

本研究所采集的农作物秸秆样本，其凯氏氮TKN与燃烧氮TCN含量分布范围较广，5种农作物秸秆的TKN与TCN含量均呈非正态分布。研究结果显示，除水稻秸秆外，农作物秸秆的TCN分析结果显著大于TKN（$P<0.05$）；小麦秸秆和棉花秸秆与其它秸秆的总氮含量（TKN与TCN）存在显著差异（$P<0.05$）。

采用“中位数±绝对中位差”统计的TKN质量分数由低到高依次为：小麦秸秆（5.66±1.07 g/kg）、油菜秸秆（7.10±1.87 g/kg）、水稻秸秆（8.20±1.42 g/kg）、玉米秸秆（8.82±2.23 g/kg）、棉花秸秆（10.42±1.45 g/kg）；TCN质量分数由低到高依次为：小麦秸秆（6.17±1.17 g/kg）、油菜秸秆（8.50±2.45 g/kg）、水稻秸秆（8.59±1.45 g/kg）、玉米秸秆（10.10±1.91 g/kg）、棉花秸秆（11.75±1.48 g/kg）。

农作物秸秆TKN与TCN间存在显著的线性相关关系且不同农作物秸秆的回归方程不同，建议采用LMS方法进行线性相关关系的拟合。此外，不同种类生物质TKN与TCN的相关关系存在差异。

致 谢

本研究得到现代农业（奶牛）产业技术体系建设专项资金（CARS－36）、国家重点研发计划（2016YFE0112800）和公益性行业（农业）科研专项资金（201003063）的资助。感谢西北农林科技大学郭康权教授在公益性行业（农业）科研专项资金（201003063）项目支持下提供的样本和分析测定数据。

[参 考 文 献]

[1] Cheng Y F, Zheng G J, Wei C, et al. Reactive nitrogen chemistry in aerosol water as a source of sulfate during haze events in China [J]. Science Advances, 2016, 2 (12): e1601530—e1601530.

[2] Tao G C, Lestander T A, Geladi P, et al. Biomass properties in association with plant species and assortments I: A synthesis based on literature data of energy properties [J]. Renewable & Sustainable Energy Reviews, 2012, 16 (5):3481—3506.

[3] IPCC. The IPCC fifth assessment report "Climate Change 2013: The Physical Science Basis" [R]. Intergovernmental Panel on Climate Change, 2013.

[4] Sinfield J V, Fagerman D, Colic O. Evaluation of sensing technologies for on-the-go detection of macro-nutrients in cultivated soils [J]. Computers and Electronics in Agriculture, 2010, 70 (1):1—18.

[5] Tian X F, Hu H W, Ding Q, et al. Influence of nitrogen fertilization on soil ammonia oxidizer and denitrifier abundance, microbial biomass, and enzyme activities in an alpine meadow [J]. Biology and Fertility of Soils, 2014, 50 (4):703—713.

[6] Munoz-Huerta R F, Guevara-Gonzalez R G, Contreras-Medina L M, et al. A review of methods for sensing the nitrogen status in plants: Advantages, disadvantages and recent advances [J]. Sensors, 2013, 13 (8):10823—10843.

[7] Hall N G, Schonfeldt H C. Total nitrogen vs. amino-acid profile as indicator of protein content of beef [J]. Food Chemistry, 2013, 140 (3): 608—612.

[8] Kamphayae S, Kumagai H, Bureenok S, et al. Effects of graded levels of liquid brewer's yeast on chemical composition and fermentation quality in cassava pulp and rice straw-based total mixed ration silage [J]. Animal Science Journal, 2017, 88 (4): 618—624.

[9] Saez-Plaza P, Michalowski T, Navas M J, et al. An overview of the Kjeldahl method of nitrogen determination. Part I. Early history, chemistry of the procedure, and titrimetric finish [J]. Critical Reviews in Analytical Chemistry, 2013, 43 (4):178—223.

[10] Niu W J, Han L J, Liu X, et al. Twenty-two compositional characterizations and theoretical energy potentials of extensively diversified China's crop residues [J]. Energy, 2016, 100:238—250.

[11] Shen X L, Huang G Q, Yang Z L, et al. Compositional characteristics and energy potential of Chinese animal manure by type and as a whole [J]. Applied Energy, 2015, 160:108—119.

[12] Vassilev S V, Baxter D, Andersen L K, et al. An overview of the chemical composition of biomass [J]. Fuel, 2010, 89 (5): 913—933.

[13] Erickson M C, Liao J, Ma L, et al. Inactivation of Salmonella spp. in cow manure composts formulated to different initial C:N ratios [J]. Bioresource Technology, 2009, 100 (23):5898—5903.

[14] Garcia R, Pizarro C, Lavin A G, et al. Spanish biofuels heating value estimation. Part I: Ultimate analysis data [J]. Fuel, 2014, 117:1130—1138.

[15] Huang G F, Wong J W C, Wu Q T, et al. Effect of C/N on composting of pig manure with sawdust [J]. Waste Management, 2004, 24 (8):805—813.

[16] Zhu N W. Effect of low initial C / N ratio on aerobic composting of swine manure with rice straw [J]. Bioresource Technology, 2007, 98 (1):9—13.

[17] Sader A P O, Oliveira S G, Berchielli T T. Application of Kjeldahl and Dumas combustion methods for nitrogen analysis[J]. Archives of Veterinary Science, 2005, 9 (2): 73—79.

[18] 沈秀丽，杨增玲，薛俊杰，等. 杜马斯燃烧法与凯氏氮法测定畜禽粪便中氮含量的比较[J]. 农业工程学报，2012, 28 (17)：205—209.
Shen Xiuli, Yang Zengling, Xue Junjie, et al. Comparison of Kjeldahl and Dumas combustion methods for determination of nitrogen content in animal manures [J]. Transactions of the Chinese Society of Agricultural Engineering (Transaction of the CSAE), 2012, 28(17): 205 — 209. (in Chinese with

English abstract)

[19] Simonne E H, Mills H A, Jones J B, et al. A comparison of analytical methods for nitrogen analysis in plant-tissues [J]. Communications in Soil Science and Plant Analysis, 1994, 25 (7/8): 943—954.

[20] Simonne E, Harris C, Mills H. Does the nitrate fraction account for differences between dumas - N and Kjeldahl - N values in vegetable leaves? [J]. Journal of Plant Nutrition, 1998, 21 (12): 2527—2534.

[21] Watson M E, Galliher T L. Comparison of Dumas and Kjeldahl methods with automatic analyzers on agricultural samples under routine rapid analysis conditions [J]. Communications in Soil Science and Plant Analysis, 2001, 32 (13/14): 2007—2019.

[22] Daun J K, Declercq D R. Comparison of combustion and Kjeldahl methods for determination of nitrogen in oilseeds [J]. Journal of the American Oil Chemists Society, 1994, 71 (10): 1069—1072.

[23] Etheridge R D, Pesti G M, Foster E H. A comparison of nitrogen values obtained utilizing the Kjeldahl nitrogen and Dumas combustion methodologies (Leco CNS 2000) on samples typical of an animal nutrition analytical laboratory[J]. Animal Feed Science and Technology, 1998, 73 (1/2): 21—28.

[24] Jung S, Rickert D A, Deak N A, et al. Comparison of Kjeldahl and Dumas methods for determining protein contents of soybean products [J]. Journal of the American Oil Chemists Society, 2003, 80 (12): 1169—1173.

[25] Simonne E, Mccrimmon J, Scoggins - Mantero H, et al. Adjustments of sufficiency ranges of selected ornamentals and turfgrasses for assessing nitrogen status with Dumas - N data [J]. Communications in Soil Science Plant Analysis, 1995, 26 (13/14): 2243—2251.

[26] Calvo N I R, Echeverría H E, Rozas H S. Comparison between two plant nitrogen and sulphur determination methods: impact on wheat sulphur diagnostics Comparación de métodos de determinación de nitrógeno *y* azufre en planta: implicancia en el diagnóstico de azufre en trigo [J]. Ciencia Del Suelo, 2008, 26 (2): 161—167.

[27] Kingbrink M, Sebranek J G. Combustion method for determination of crude protein in meat and meat-products - collaborative study [J]. Journal of Aoac International, 1993, 76 (4): 787—793.

[28] AOAC. AOAC Official Method 2001.11 Protein (crude) in Animal Feed, Forage (Plant Tissue), Grain, and Oilseeds [Z]. AOAC, 2000.

[29] AOAC. AOAC official method 990.03, protein (crude) in animal feed, combustion method [Z]. AOAC, 2006.

[30] Bremner J M, Mulvaney C S. Methods of Soil Analysis, Part 2 [M]. Madison, USA: American Society of Agronomy, 1982.

[31] Nelson D W, Sommers L E. Total nitrogen analysis of soil and plant-tissues [J]. Journal of the Association of Official Analytical Chemists, 1980, 63 (4):770—777.

[32] Jaroonchon N, Krisanapook K, Phavaphutanon L. Correlation between pummelo leaf nitrogen concentrations determined by combustion method and Kjeldahl method and their relationship with SPAD values from portable chlorophyll meter[J]. Kasetsart Journal - Natural Science, 2010, 44 (5):800—807.

[33] Lee D, Nguyen V, Littlefield S. Comparison of methods for determination of nitrogen levels in soil, plant and body tissues, and water [J]. Communications in Soil Science and Plant Analysis, 1996, 27 (3/4):783—793.

[34] Chen X L, Zhao G L, Zhang Y, et al. Nitrogen-to-protein conversion factors for crop residues and animal manure common in China [J]. Journal of Agricultural and Food Chemistry, 2017, 65 (42):9186—9190.

[35] Jimenez R R, Ladha J K. Automated elemental analysis: A rapid and reliable but expensive measurement of total carbon and nitrogen in plant and soil samples [J]. Communications in Soil Science and Plant Analysis, 1993, 24 (15):1897—1924.

[36] Mcgeehan S L, Naylor D V. Automated instrumental analysis of carbon and nitrogen in plant and soil samples [J]. Communications in Soil Science and Plant Analysis, 1988, 19 (4):493—505.

[37] Simonne A H, Simonne E H, Eitenmiller R R, et al. Could the Dumas method replace the Kjeldahl digestion for nitrogen and crude protein determinations in foods? [J]. Journal of the Science of Food and Agriculture, 1997, 73 (1):39—45.

[38] Silvertooth J C, Westerman R L. Digestion of plant materials for the determination of total nitrogen to include nitrate [J]. Agronomy Journal, 1988, 80 (5):733—736.

[39] Bradstreet R B. Modifications of Kjeldahl digestion for organic nitrogen [J]. Analytical Chemistry, 1960, 32 (1): 114—117.

[40] Guo W S, Meng Q X. Comparison of Kjeldahl and Dumas combustion methods for determination of nitrogen content in feedstuffs [J]. Acta Veterinaria Et Zootechnica Sinica, 2006, 37 (5):464—468.

[41] Barker A V, Maynard D N, Mills H A. Variations in nitrate accumulation among spinach cultivars [J]. Journal of the American Society for Horticultural Science, 1974, 99 (2): 132—134.

[42] Guo W S, Ren L P, Zhou Z M, et al. Difference of nitrogen contents determined by the combustion and Kjeldahl method in response to nitrate nitrogen in some feedstuffs [J]. Journal of Animal and Feed Sciences, 2007, 16:178—183.

[43] Miller E L, Bimbo A P, Barlow S M, et al. Repeatability and reproducibility of determination of the nitrogen content of fishmeal by the combustion (Dumas) method and comparison with the Kjeldahl method: Interlaboratory study [J]. Journal of Aoac International, 2007, 90 (1):6—20.

[44] Dieckow J, Mielniczuk J, Knicker H, et al. Comparison of carbon and nitrogen determination methods for samples of a paleudult subjected to no-till cropping systems [J]. Scientia Agricola, 2007, 64 (5):532—540.

Analysis of the total nitrogen content of crop residues determined by using Kjeldahl and Dumas methods

Wang Xinlei, Yuan Xiangru, Zhang Xiaoxiao, Xie Ruyue, Xiao Weihua, Han Lujia※
(*College of Engineering*, *China Agricultural University*, *Beijing* 100083, *China*)

Abstract: The Kjeldahl and combustion (Dumas) methods are the main methods used to determine total nitrogen (TKN and TCN) content in agriculture biomass. However, the results obtained using these methods differ because of differences in their underlying principles. Herein, we used these two methods to determine the total nitrogen content in 1 179 crop residues (rice straw, wheat straw, corn stover, rape stalk, cotton stalk) from China, and systematically analyzed and compared in total nitrogen content and their distributions in the collected crop residues by different types with these two methods. Seven common distributions (Normal, Lognormal, Gamma, Weibull, Exponential, Laplace, Lorentz) were used to determine the data distribution types of TKN and TCN in different crop residues. The correlation between the two methods was explored using ordinary least squares regression (OLS), orthogonal regression (Orth), and least median square regression (LMS). Finally, the research reviewed the correlation between different biomass (food, flowers, grass, soil, crop, crop residues, sewage sludge and animal manure, etc.) results of two methods for measuring nitrogen content. The results showed: The distributions of nitrogen content were non-normal distributions in different crop residues. The total TKN and TCN contents were the same distributions in rice straw, wheat straw, rape stalk and total crop residues which approximately followed Lognormal, Gamma, Lognormal and Lognormal respectively. The median method was recommended for data statistics, and results from low to high were: 1) TKN: wheat straw (5.66 ± 1.07 g/kg), rape stalk (7.10 ± 1.87 g/kg), rice straw (8.20 ± 1.42 g/kg), corn stover (8.82 ± 2.23 g/kg), cotton stalk (10.42 ± 1.45 g/kg); 2) TCN: wheat straw (6.17 ± 1.17 g/kg), rape stalk (8.50 ± 2.45 g/kg), rice straw (8.59 ± 1.45 g/kg), corn stover (10.10 ± 1.91 g/kg), cotton stalk (11.75 ± 1.48 g/kg). It was found that TKN was significantly lower than TCN in all types of crop residues ($P < 0.05$). TKN and TCN values were also significantly different among wheat straw, cotton stalk and other crop residues ($P < 0.05$). Although the fitting efficiencies of OLS, Orth and LMS were the same on the determination coefficient (R^2) scale, the fitting results were different. LMS was recommended because it reduced the effect of outliers compared with three methods, observed from kernel density – scatter plots. Five types of crop residues and the total had different fitting result between TKN and TCN. The correlation between TKN and TCN for total crop residues was quantified as the LMS equation. In addition, there was a gap of the linear relationships between TKN and TCN in different types of biomass. The slope of plant biomass was generally lower than that of animal manure, whose potential reason was different forms and contents of nitrogen in different biomass (ammonium nitrogen, nitrate nitrogen, nitrite nitrogen, heterocyclic nitrogen, and nucleic acid nitrogen, etc.). The results may provide extensive and reliable data for reference from large sample size, and methods support for the scientific utilization of nitrogen in crop residues.

Keywords: straw; nitrogen; models; Kjeldahl method; combustion (Dumas) method; distribution analysis; median; linear correlation