Effects of origin, genotype, harvest year and their interactions on stable isotope, multi-element and near-infrared fingerprints in wheat

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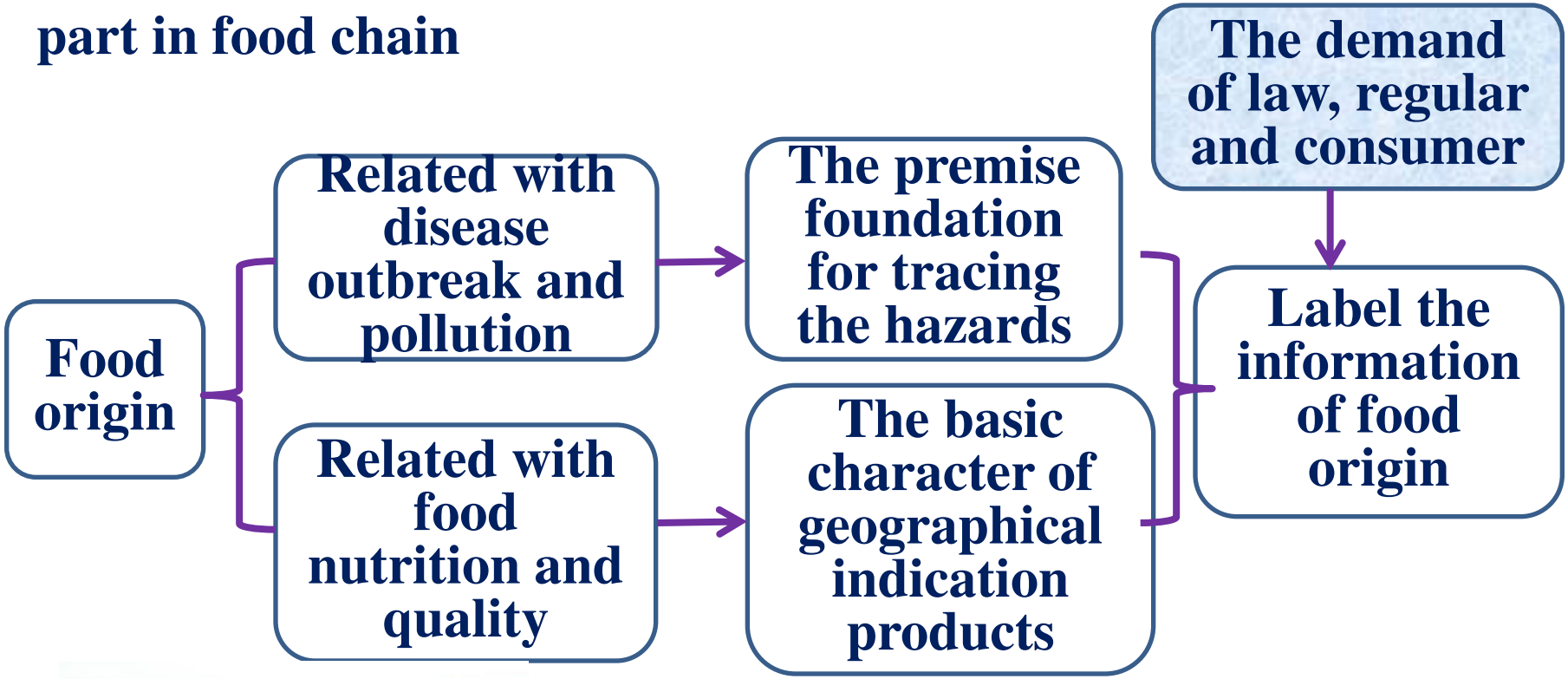
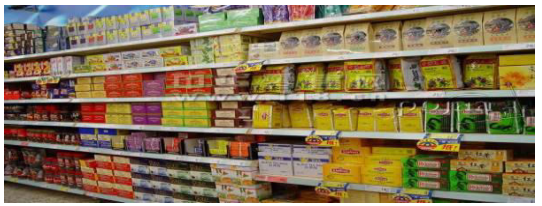
1. Introduction

1.1 Food geographical origin traceability is an important part in food chain





1. Introduction (continued)

1.2 The independent and scientific technology is needed to preclude and supervise the adulterated and fraud food

Protected Designation of Origin（PDO）

Protected Geographical Indication （PGI）

Traditional Speciality Guaranteed（TSG）

Agro-product Geographical Indication（AGI）

Techniques used in food geographical origin traceability

Agro-Products

Meat(Beef and Lamb)

Tea

Fruit (Kiwi fruit, Pear)

Cereal(Wheat and Millet)

Techniques

Stable isotopic fingerprint

Multi element fingerprint

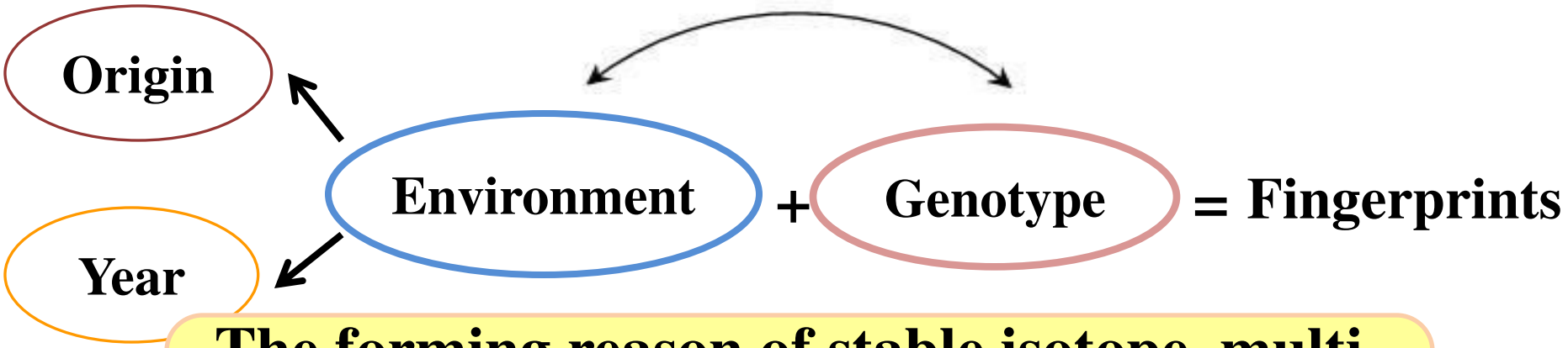
NIR fingerprint







1. Introduction (continued)



The forming reason of stable isotope, multi-element and near-infrared fingerprints of agricultural products

Total variance of fingerprints

= Origin + Year + Genotype + Interaction

1. Introduction (continued)

Wheat——Model plant

Wide distribution, Diversiform genotypes, Strong adaptability

2. Materials and Methods

Experimental Design

Wheat variety: Han 6172, Heng 5229, Hengguan 35, Xinong 889, Xinong 979, Xiaoyan 22, Xinmai 18, Zhengmai 366, Zhoumai 16 and Zhoumai 18.

Field experiment: Three regions for three years. Randomized complete blocks for a total of 10 plots. The locally recommended agricultural practices.

2. Materials and Methods (continued)

Stable isotope ratio Analysis

$\delta^{13}C$、$\delta^{15}N$ and δD were determined by IRMS.

Multi elemental Analysis

The concentrations of 55 elements (Li, Na, Mg, Al, etc.) were determined by HR-ICP-MS.

2. Materials and Methods (continued)

Near Infrared fingerprints

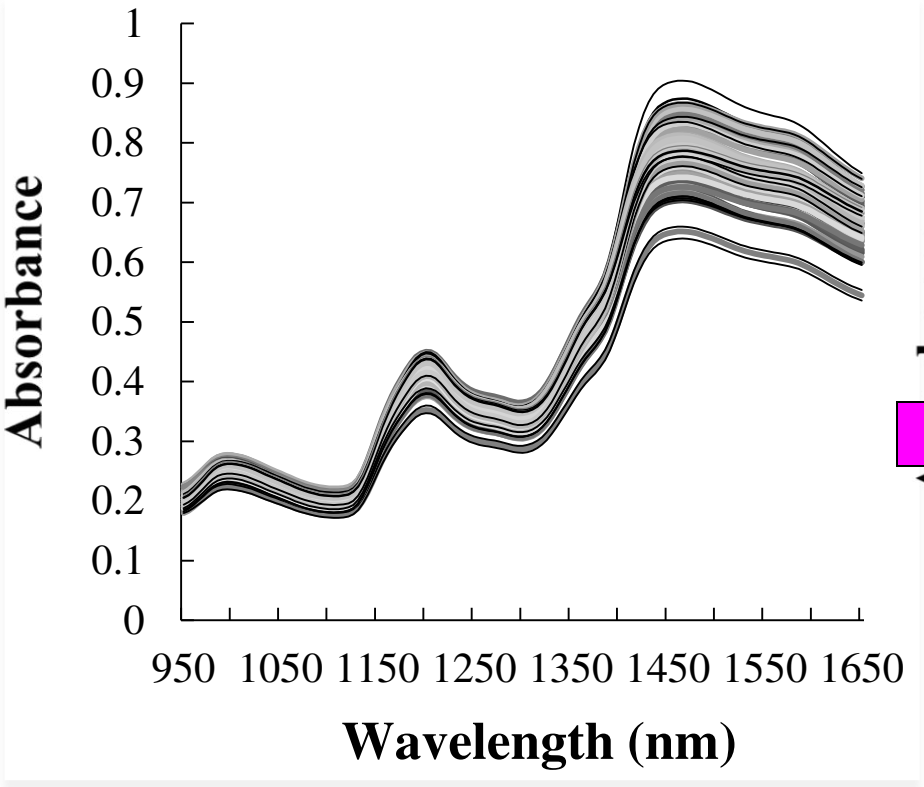


Fig. 1 Raw spectra of whole wheat flours

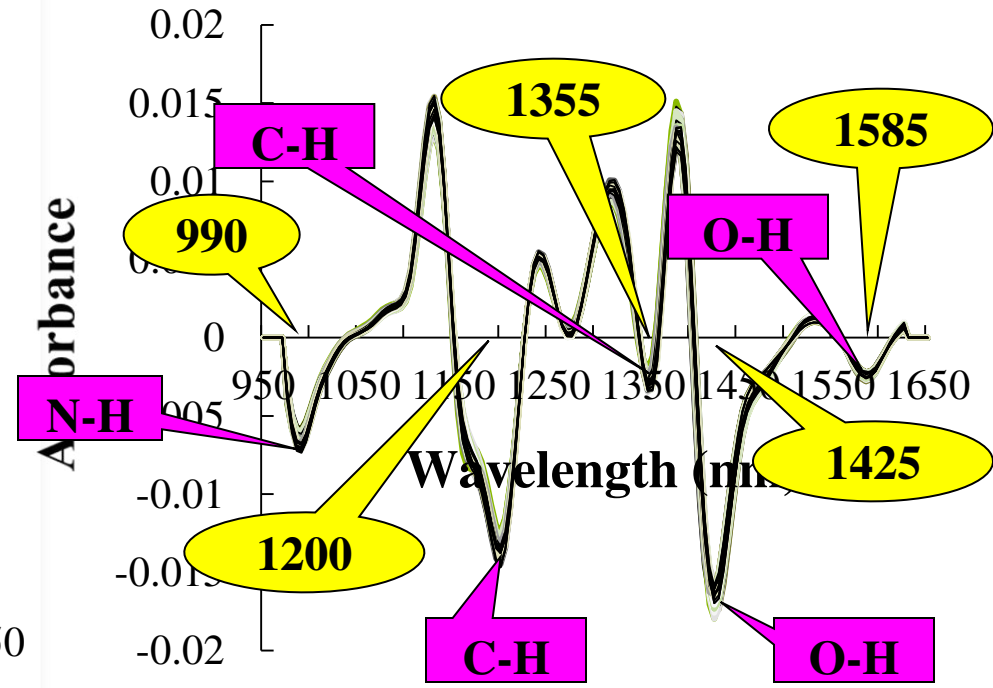


Fig. 2 Spectra after pretreatment

3. Results

The effects on stable isotopic fingerprints

Table 1 The $\delta^{13}C$、$\delta^{15}N$ and δD valuess in wheat samples among three regions of three years

Isotope	Huixian	Yangling	Zhaoxian
$\delta^{13}C$(‰)	$-27.92^{B} \pm 0.41$	$-26.59^{A} \pm 0.96$	$-28.00^{B} \pm 0.55$
$\delta^{15}N$(‰)	$0.95^{A} \pm 1.46$	$-2.76^{C} \pm 1.51$	$0.33^{B} \pm 1.16$
δD(‰)	$-66.42^{B} \pm 6.44$	$-61.18^{A} \pm 7.62$	$-71.58^{C} \pm 6.43$

Note: the different letters in rows represent statistically significantly different at $p < 0.01$.

3. Results(continued)

Table 2 Variance Analysis for the stable carbon ($\delta^{13}C$), nitrogen ($\delta^{15}N$) and hydrogen (δD) of wheat kernels

Source of variation	F value	$\delta^{13}C$ variance	$\delta^{13}C$ Sig.	$\delta^{15}N$ variance	$\delta^{15}N$ Sig.	$\delta^{2}H$ variance	$\delta^{2}H$ Sig.
Region（R）	2	111.782	0.000	713.509	0.000	4867.902	0.000
Genotype （G）	9	31.441	0.000	13.092	0.000	2020.534	0.000
Year（Y）	2	40.122	0.000	5.853	0.006	3608.058	0.000
R×G	18	2.576	0.000	33.802	0.000	395.810	0.804
G×Y	18	2.576	0.000	33.802	0.000	395.810	0.658
R×Y	18	32.897	0.000	295.798	0.000	970.957	0.000
R×G×Y	36	5.119	0.000	33.974	0.015	465.409	0.993

3. Results(continued)

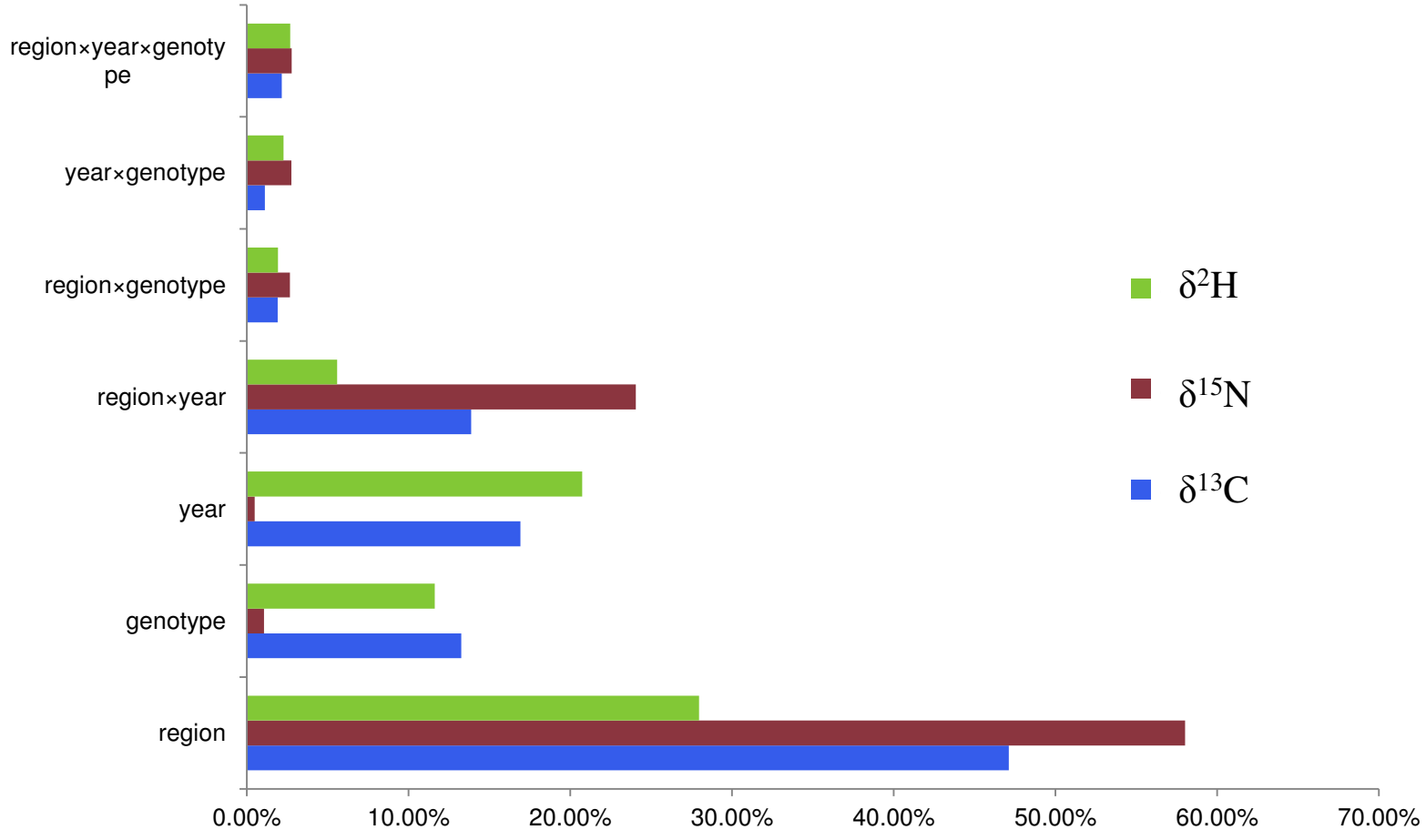


Fig.3 the percentage of total square variance of each factor for $\delta^{13}C$, $\delta^{15}N$ and δD in wheat kernels

3. Results(continued)

The effects on multi-elemental fingerprints

- The contents of 20 elements (Be, Ge, Nb, In, Te, etc.) were lower than LOD.

- The contents of 22 elements (Se, Hg, etc) could not be determined precisely by ICP-MS.

- 13 elements (Mg, Al, Ca, Mn, Fe, Cu, Zn, As, Sr, Mo, Cd, Ba, Pb) were analyzed.

3. Results(continued)

Table 3 The element contents of wheat kernels from different regions（μg·g^{-1}）

Element	Huixian	Yangling	Zhaoxian
Mg*	1432±252 ^{b}	1490±240 ^{b}	1658±253 ^{a}
Al*	5±3 ^{b}	8±6 ^{a}	5±3 ^{b}
Ca*	478±96 ^{b}	535±81 ^{a}	519±140^{b}
Mn*	32.6±3.6 ^{b}	39.3±4.3 ^{a}	33.3±4.1^{b}
Fe*	38.6±13.5 ^{b}	46.5±18.8 ^{a}	38.11±14.3^{b}
Cu	4.63±1.21 ^{a}	4.78±1.08 ^{a}	4.61±0.96^{a}
Zn*	37.2±8.7 ^{a}	25.0±5.8^{c}	30.9±6.2 ^{b}
As*	0.028±0.014^{b}	0.024±0.010^{b}	0.038±0.016^{a}
Sr*	4.5±1.3^{c}	5.4±1.9^{b}	9.1±1.6 ^{a}
Mo*	0.438±0.087 ^{b}	0.463±0.110 ^{b}	0.646±0.107 ^{a}
Cd*	0.140±0.041 ^{a}	0.016±0.004 ^{b}	0.016±0.004 ^{b}
Ba*	3.63±0.99 ^{a}	2.76±0.92 ^{b}	3.73±1.28 ^{a}
Pb	0.158±0.129 ^{a}	0.178±0.347 ^{a}	0.167±0.161 ^{a}

3. Results(continued)

Table 4 The percentages of total square variance of each effect on each element

Element	Region (R)	Year (Y)	Genotype (G)	R × Y	R × G	G × Y	R × G × Y	error
Mg	9.6%	33.7%	16.5%	3.7%	4.8%	5.6%	6.6%	15.1%
Al	9.7%	29.7%	1.5%	9.6%	5.4%	4.7%	3.4%	26.6%
Ca	5.1%	66.3%	3.6%	4.8%	1.4%	3.4%	1.5%	6.2%
Mn	34.2%	5.1%	22.7%	2.2%	3.4%	6.2%	5.6%	14.0%
Fe	6.0%	56.9%	1.7%	2.0%	1.2%	1.2%	2.1%	4.1%
Cu	0.4%	56.8%	20.1%	2.3%	2.8%	2.0%	2.6%	6.6%
Zn	34.3%	36.6%	6.8%	4.6%	3.9%	2.4%	3.8%	5.3%
As	12.5%	51.5%	8.4%	3.7%	1.8%	3.9%	3.3%	7.1%
Sr	39.6%	11.8%	4.5%	8.5%	2.8%	1.0%	2.3%	6.0%
Mo	35.0%	5.6%	34.9%	2.1%	3.5%	1.7%	4.4%	61.0%
Cd	78.8%	2.9%	1.3%	5.5%	1.8%	0.3%	0.5%	0.7%
Ba	11.6%	2.1%	27.3%	8.7%	9.7%	7.9%	10.4%	14.9%
Pb	0.1%	32.8%	2.1%	1.1%	4.2%	4.7%	8.4%	43.1%

The influence of each factor on the content of every element in wheat kernels

wheat origin

Region had significant influence on the elements of **Mn (manganese), Sr (strontium), Mo (molybdenum)** and **Cd (cadmium)**

Harvest year

Harvest year had significant influence on the elements of **Mg (magnesium), Al (aluminum), Ca(calcium), Fe(ferəm), Cu (copper), Zn(zinc), As(Arsenic), Pb(lead)**

Genotype

Genotype had significant influence on the contents of **Ba(barium)** ($p < 0.01$)

PCA and LDA for the samples from different regions with the elements related to geographical origin

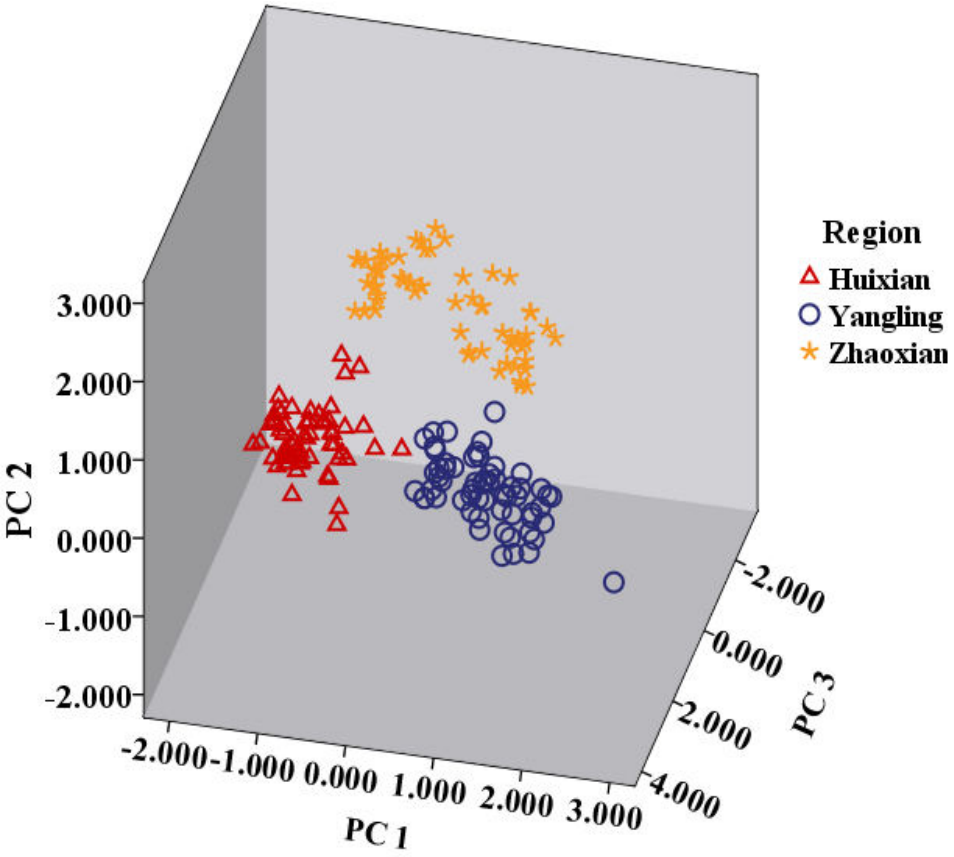


Fig. 5 Scatter plot of PCs 1, 2 and 3

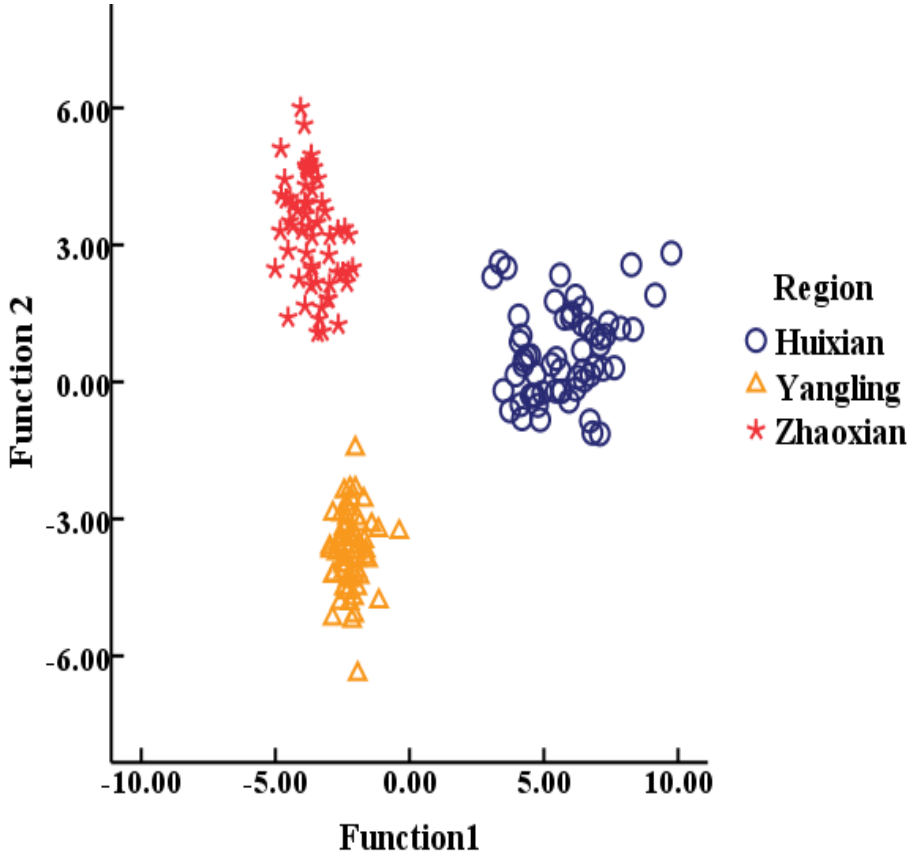


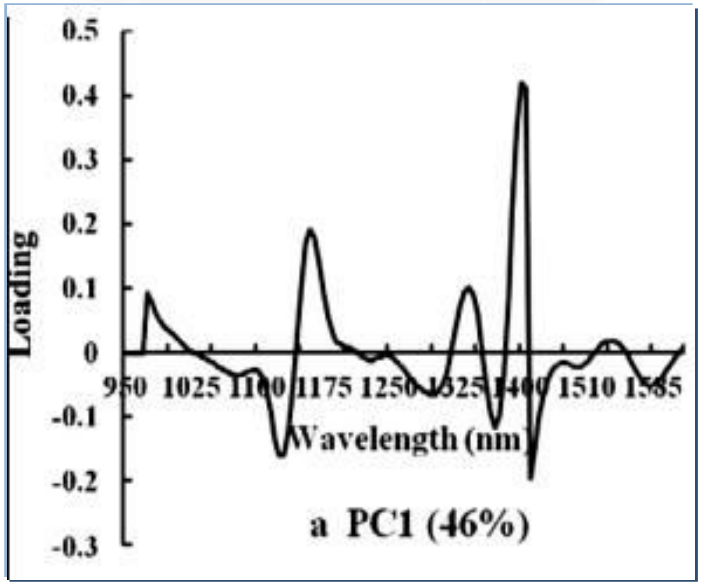
Fig. 6 Scatter plot of functions 1 and 2

3. Results(continued)

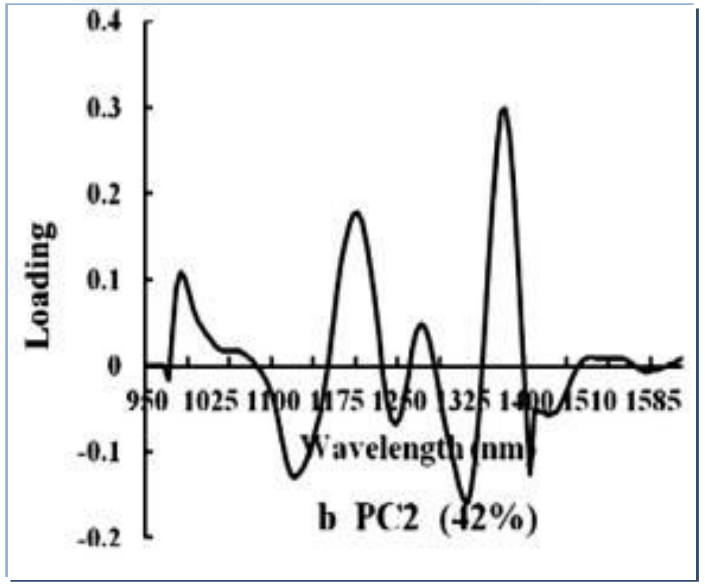
The effects on NIR fingerprints

PCA （Principal Component Analysis）

1135, 1390-1405nm



1200, 1325-1340, 1355-1380 nm

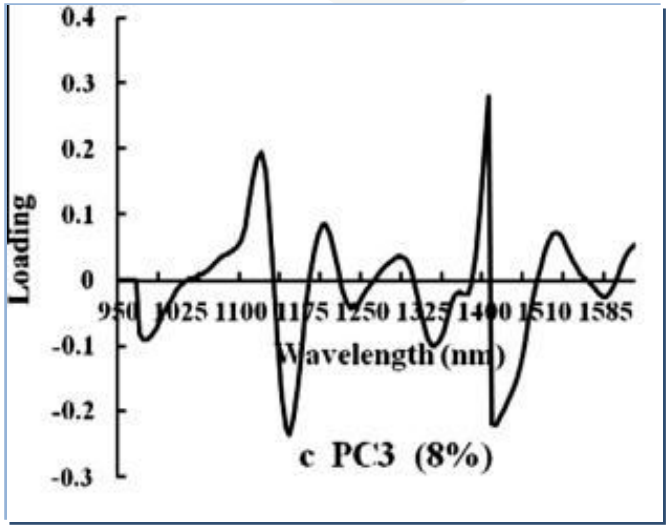


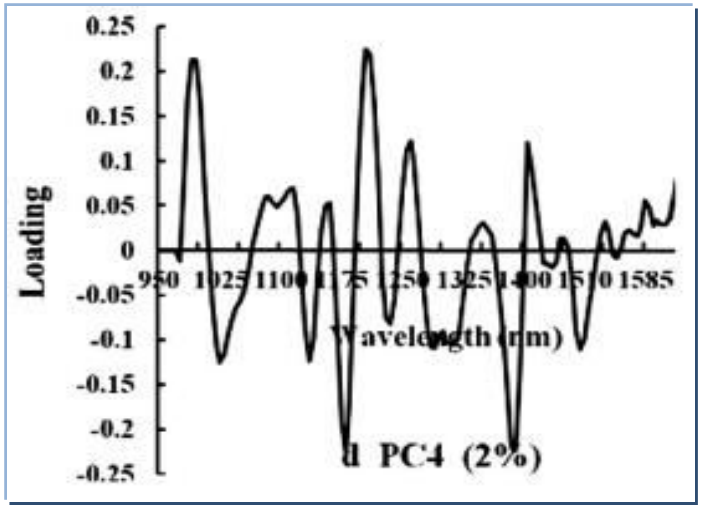
3. Results(continued)

PCA

1110-1130, 1150-1170, 1410, 1445-1480 nm

995-1000, 1020-1030, 1135, 1175-1180, 1205-1210,1255-1260 , 1285–1290, 1385–1390, 1505nm



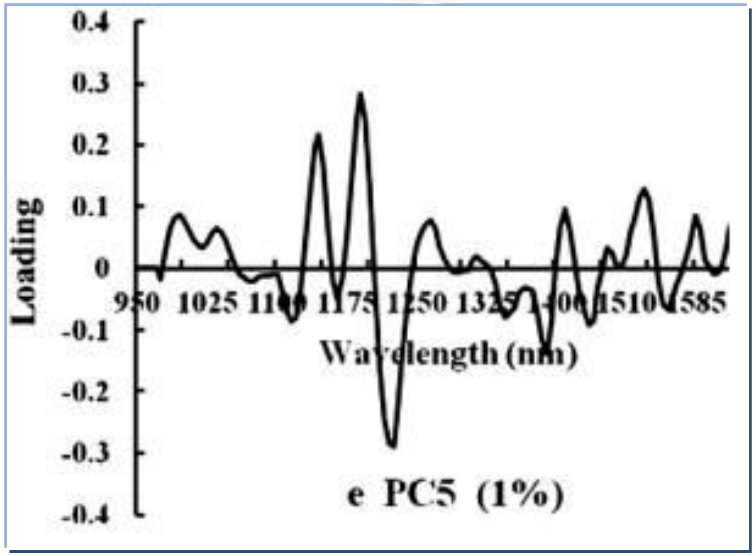


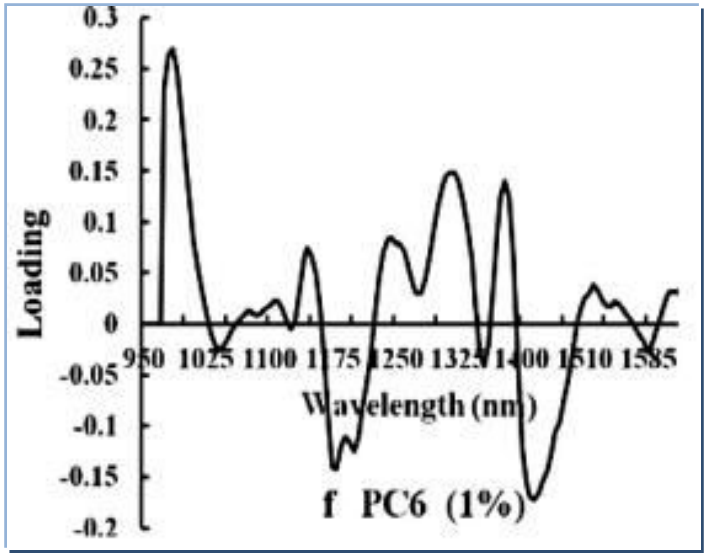
3. Results(continued)

PCA

1135–1145, 1185–1195, 1215–1235, and 1525–1535 nm

975–990, 1005, and 1300–1320 nm





The above 6 PCs explained 98% of the total variance

3. Results(continued)

MANOVA (Multi Analysis of Variance)

PC1 genotype > year > region > region × genotype

PC2 region > genotype > region × year > region × genotype > region × genotype × year

PC3 genotype > region × year > region > year > region × genotype > region × genotype × year > genotype × year

3. Results(continued)

MANOVA

PC4 year > region > genotype > region × year > genotype × year > region × genotype > region × genotype × year.

PC5 year > region > genotype > region × year > genotype × year > region × genotype.

PC6 region > region × year > genotype > year > region × genotype × year > genotype × year > region × genotype

3. Results(continued)

Table 5. Classification analysis for samples from three regions

Analytical wavelengths	Region			
	Zhaoxian	Huixian	Yangling	Total
Wavelength related to region	90.0	80.0	100.0	90.0
Wavelength of absorption bands related to region	90.0	90.0	95.0	91.7

4. Conclusion

The stable isotope ratio of **$\delta^{13}C$、$\delta^{15}N$ and δD ,** the elements of **Mn (manganese), Sr (strontium), Mo (molybdenum)** and **Cd (cadmium)**, and the wavelength ranges **975–990 nm, 1200 nm, 1355–1380 nm** were the suitable traceability indicators to develop robust discriminant model of geographical origin.

Acknowledgement

Funding Sources

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Thanks for your attention

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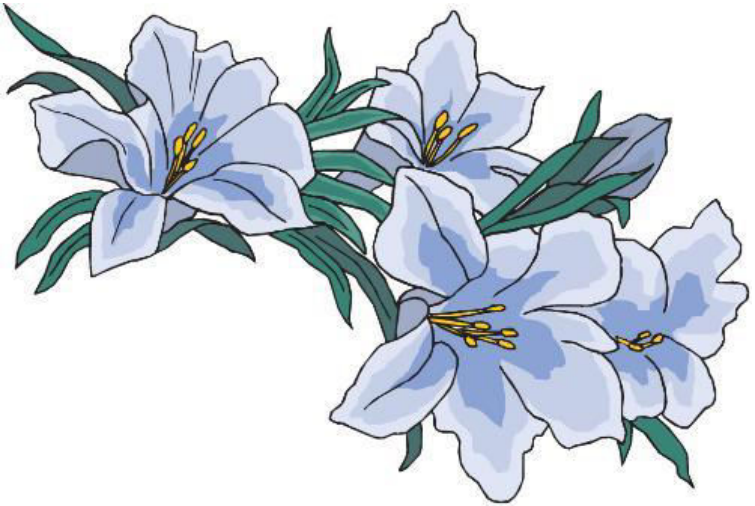

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