Detection of Heterocyclic Amine Content in Different Types of Pork Products

Ting Li[1,3][a], Haibin Li[2][b], Jiayun Kang[1,3][c], Wenhui Liang[1,3][d], Fang Wang[1,3][e], Jinghua Qi[1,3][f] and Meixia Pang[1,3,*][g]

[1]*Beijing Key Laboratory of Agricultural, Product Detection and Control of Spoilage Organisms and Pesticide Residue, Beijing University of Agriculture, Beijing, China*

[2]*Pengcheng food branch of Beijing Shunxin agriculture Co., Ltd, Beijing, China*

[3]*Beijing Innovation Consortium of Swine Research System, Beijing, China*

Keywords: Heterocyclic Amines, Pork Products, LC-MS/MS.

Abstract: There are different levels of heterocyclic amines in pork products processed by different methods, and the total heterocyclic amine content was 7.68-64.18 ng/g. Among them, non-polar heterocyclic amines Harman and Norharman were mostly 1.42-3.69 ng/g and 1.40-40.47 ng/g. The distinction within the content of heterocyclic amines in meat products with different processing methods was obvious. The total amount of heterocyclic amines from high to low were cured meat, sauced meat, dried meat, roast meat, fried meat, canned, sausage and ham.

1 INTRODUCTION

Along with our country residents living standards improve, people produce harmful substances in food processing is more and more attention, and heterocyclic amines in a lot of food and different processing technology produces, has been more and more researchers focus, but bad amine for miscellaneous domestic research is still in its infancy, the related research and less reported, and focused on the detection method to explore and optimize. Pork is the main type of meat consumed by Chinese residents, and there are four main cooking methods in China: sauce, barbecue, pan frying and deep-frying. As can be seen from foreign research reports, hcas are ubiquitous in meat products, and most of the meat products studied are cattle, sheep, fish and poultry, while pork is rarely studied. In addition, the traditional way of cooking in China and abroad have very big different, the kinds of heterocyclic amines and content in the meat products also have bigger difference, at the same time of heterocyclic amines in overseas has for meat formation mechanism, influencing factors, analysis method and suppression measures have a more in-depth study, and domestic research is just getting started in recent years, various aspects are in-depth study, Therefore, this study focuses on the influencing factors of hCAS content in pork products.

According to GB/T 26604-2011(2011), meat products are primarily separated into cured meat, sauced meat, roast meat, dried meat, fried meat, sausage, ham, prepared meat products and other meat products. This study chose 14 sorts of pork items which are well known and loved by consumers within the market. This paper analyzed the influence of

[a] https://orcid.org/0000-0002-5644-5768
[b] https://orcid.org/0000-0003-0899-5068
[c] https://orcid.org/0000-0003-0174-9710
[d] https://orcid.org/0000-0001-8982-7693
[e] https://orcid.org/0000-0002-0438-6786
[f] https://orcid.org/0000-0001-9204-3697
[g] https://orcid.org/0000-0003-3025-5305

Li, T., Li, H., Kang, J., Liang, W., Wang, F., Qi, J. and Pang, M.
Detection of Heterocyclic Amine Content in Different Types of Pork Products.
DOI: 10.5220/0011293600003438
In *Proceedings of the 1st International Conference on Health Big Data and Intelligent Healthcare (ICHIH 2022)*, pages 289-294
ISBN: 978-989-758-596-8

processing methods on the content of heterocyclic amines, and provided data reference for future studies on the population exposure of heterocyclic amines.

2 MATERIALS AND METHODS

2.1 Materials and Reagents

Pork floss, jerky, dried meat crisps, spiced pork, bacon, preserved pork, grilled tenderloin, braised pork head, American ham, starch-free ham, wide-flavored sausage, ham, spam, and toothpick meat were all purchased from the market.

10 HAAs standards (IQ, MeIQ, 8-MeIQx, 4,8-DiMeIQx, 4,7,8-TriMeIQx, PhIP, Norharman, harman, Trp-P-1, AαC), Methanol, Acetonitrile, Ammonium formate, Formic acid (HPLC, TRC Canada); Dichloromethane, Sodium hydroxide, Ammonium acetate, Hydrochloric acid (AR, Sinopharm reagent Co., Ltd.); Diatomite (Extrelut-20NT, Merck, Germany); PRS-SPE column, C18-SPE column (500mg / mL, 300mg / mL, Agilent, USA).

2.2 HAAs Analysis

According to the method of Shen(Shen 2017), the sample was purified by PRS column and C18 solid phase extraction, then detected and analyzed by LC-MS/MS.

2.3 Statistical Analysis

All data was organized using Microsoft Excel, and Origin 8.0 was used for graphing, and IBM SPSS Statistics 20 statistical software was used for data analysis.

3 RESULTS AND DISCUSSION

The samples of pork products collected in this experiment are all bagged products sold in the market, except that toothpick meat is currently unpackaged. Bacon, preserved pork and grilled tenderloinare all raw meat products, after heating mature meat products, and then tested.

The contents of polar heterocyclic amines of all meat products are shown in Table 1, and the contents of non-polar heterocyclic amines are shown in Table 2.

Table 1: The content of polar HAAs in different meat products.

Meat products	IQ	MeIQ	8-MeIQx	4,8-DiMeIQx	PhIP	Total
Pork floss	0.97±0.05^{e}	0.66±0.02^{d}	0.07±0.02^{d}	0.05±0.01^{d}	0.04±0.03^{e}	1.78±0.06^{fg}
jerky	1.18±0.02^{e}	0.66±0.01^{d}	0.06±0.02^{d}	0.03±0.02^{d}	0.10±0.03^{d}	2.03±0.17^{fg}
dried meat crisps	1.24±0.01^{e}	0.64±0.01^{d}	0.15±0.05^{c}	0.08±0.01^{d}	0.12±0.04^{d}	2.23±1.01^{f}
Spiced Pork	1.84±0.07^{e}	1.01±0.12^{bc}	0.41±0.48^{b}	0.32±0.17^{b}	0.12±0.04^{d}	3.70±1.230^{e}
bacon	6.19±0.32^{a}	0.83±0.03^{c}	0.40±0.02^{b}	nd	0.13±0.02^{d}	7.55±0.51^{b}
preserved pork	5.42±0.12^{b}	0.81±0.04^{c}	0.76±0.34^{a}	0.17±0.07^{c}	1.95±0.16^{a}	9.10±2.23^{a}
grilled tenderloin	nd	0.75±0.02^{cd}	0.15±0.04^{c}	0.17±0.07^{c}	0.15±0.01^{d}	1.23±0.15^{g}
braised pork head	nd	0.91±0.02^{c}	0.12±0.04^{cd}	0.18±0.03^{c}	0.20±0.06^{d}	1.41±0.16^{fg}
American ham	nd	1.08±0.02^{bc}	0.18±0.09^{c}	0.27±0.09^{b}	0.30±0.07^{cd}	1.83±0.37f^{g}
starch-free ham	4.17±0.24^{c}	1.82±0.11^{b}	0.12±0.01^{cd}	0.13±0.02^{c}	0.16±0.01^{d}	6.41±2.12^{cd}
wide-flavored sausage	2.69±0.10^{d}	3.56±0.11^{a}	0.26±0.02^{c}	0.29±0.02^{b}	0.33±0.02^{c}	7.12±2.43^{bc}
ham	1.37±0.17^{e}	1.92±0.45^{b}	0.18±0.01^{cd}	0.26±0.02^{b}	0.33±0.01^{c}	3.42±1.20^{e}
spam	nd	2.51±0.12^{b}	0.39±0.08^{b}	0.50±0.12^{a}	0.46±0.16^{c}	3.86±0.58^{e}
toothpick meat	1.69±0.11^{e}	1.95±0.24^{b}	0.81±0.10^{a}	0.51±0.35^{a}	1.22±0.10^{b}	6.16±2.11^{d}

nd=not detected.

Each value is represented as mean± standard deviation (n= 3). Means with different superscript letters within the same column are significantly different at $P<0.05$.

It can be seen from Table 1 that the content of IQ was nd-6.19 ng/g, and the average content was 1.91 ng/g. The highest content was Bacon at 6.19 ng/g. Among which the highest content is bacon 6.19 ng/g, which has a significant difference compared with other meat products (P <0.05), but it is not detected in grilled tenderloin, braised pork head, American ham and spam. The IQ content of most meat products was 1-3 ng/g.It should be noted that bacon is sealed packaging of raw bacon, bought by the canteen after fried and then tested, because when fried meat and heat source direct contact, The various precursors formed by heterocyclic amines in meat constantly exude from the meat and react on the contact surface to form more IQ-type heterocyclic amines. The content of MeIQ ranged from 0.64-3.56ng/g, and the average content was 1.37 ng/g. The content of the sausage was the highest. There was no significant difference in most samples (p> 0.05). Wide-flavored sausages are processed through pickling, enema, airing, and baking. The auxiliary materials contain a relatively high proportion of sugar(Huang 2019). Sugar is the precursor of polar heterocyclic amines. The high content of MeIQ may be due to the high sugar content in Wide-flavored sausages. The measured 8-MeIQx in samples was 0.06-0.81ng/g, and the average content was 0.29 ng/g. The content of toothpick meat was the highest. This heterocyclic amine may be easily formed due to the higher frying temperature. The content of PhIP ranged from 0.04-1.95ng/g, the average content was 0.40 ng/g, the content was low, and most samples were not significantly different (p>0.05). The total content of the five polar heterocyclic amines measured in the surveyed meat samples was 0.96-7.28ng/g. The highest content of polar heterocyclic amines was preserved pork, and the lowest content was roasted tenderloin.

Table 2 The content of non-polar HAAs in different meat products

Meat products	Norharman	Harman	Trp-P-1	AaC	Total
Pork floss	3.72±0.21^{e}	18.74±3.08^{b}	nd	nd	22.46±1.2^{c}
jerky	4.06±0.95^{e}	14.25±0.62^{b}	nd	nd	18.31±0.56^{d}
dried meat crisps	10.35±0.14^{c}	5.98±0.20^{c}	nd	nd	16.33±0.78^{d}
Spiced Pork	21.02±0.64^{b}	32.69±0.34^{a}	nd	nd	53.71±1.39^{a}
bacon	8.85±0.46^{cd}	3.49±0.13^{cd}	nd	nd	12.34±0.65^{ef}
preserved pork	40.47±0.95^{a}	11.58±0.65^{b}	nd	nd	52.05±2.45^{a}
grilled tenderloin	4.69±0.13^{e}	4.89±0.19^{c}	nd	0.02±0.01^{c}	9.60±0.34^{g}
braised pork head	9.37±0.19^{c}	26.00±0.69^{a}	nd	0.04±0.01^{bc}	35.41±0.89^{b}
American ham	2.35±0.91^{f}	2.86±0.43^{d}	nd	0.02±0.01^{c}	5.24±1.12^{h}
starch-free ham	2.17±0.15^{f}	4.79±0.15^{c}	nd	0.02±0.01^{c}	6.98±0.91^{h}
wide-flavored sausage	1.40±0.16^{g}	1.42±0.15^{e}	0.26±0.01^{a}	0.01±0.01^{c}	3.08±0.89^{i}
ham	3.73±0.01^{e}	5.79±0.64^{c}	0.14±0.03^{b}	0.02±0.01^{c}	9.68±0.81^{g}
spam	5.53±1.00^{d}	4.62±0.93^{c}	nd	0.11±0.02^{a}	10.26±0.65^{fg}
toothpick meat	5.53±0.26^{d}	8.10±1.18^{c}	nd	0.07±0.01^{b}	13.70±1.11^{e}

nd=not detected.
Each value is represented as mean± standard deviation (n= 3). Means with different superscript letters within the same column are significantly different at P<0.05.

The non-polar heterocyclic amine content and total heterocyclic amine content are shown in Table 2. The content of Norharman ranges from 1.40-40.47 ng/g, with an average content of 8.80 ng/g. The highest content was Bacon. The bacon is made by curing, air-drying, smoking and roasting. Although the temperature of the whole process is low, because the smoke contains heterocyclic amines, it adheres to the surface of meat products during smoking. After heating treatment before eating, Heterocyclic amines are immersed in the meat from the surface, resulting in an increase in the content of heterocyclic amines.

It should be noted that bacon is a sealed package of cured bacon, bought from the supermarket and cooked in water before being tested.

The content of Harman is in the range of 1.42-32.69 ng/g, with an average content of 10.37 ng/g, and the highest content was Sauced meat. Although the processing method of sauced meat was relatively mild, the heating temperature was not high at 100℃, but the cooking time was more than 1h, and the resulting Harman content was indeed high. The Norharman and Harman content of wide-flavored sausage was the lowest. Both Norharman and Harman belong to β-carboline heterocyclic amines. Regarding the formation of β-carboline, it is generally believed that it is directly generated by pyrolysis of a single amino acid or protein, and the formation temperature is generally higher than 300 ℃ (MLC 2004), But later studies have shown that β-carboline can also be formed at temperatures below 100°C (Liu 2020). Because Norharman and Harman can enhance the mutagenicity of other heterocyclic amines in the Ames/Salmonella experiment, they are called auxiliary mutagens (Kanithaporn 2011). At the same time, studies have shown that it is related to neurotoxins and enzyme inhibitors.

The content of Trp-p-1 was nd-0.26 ng/g, and the average content was 0.03 ng/g. Most samples were not detected, only wide-flavored sausage and common ham detected low levels of this heterocyclic amine, which were 0.26ng/g and 0.14ng/g, respectively.

The content of AaC was nd-0.11ng/g, and the average content was 0.02 ng/g. The content was very low and almost undetectable.

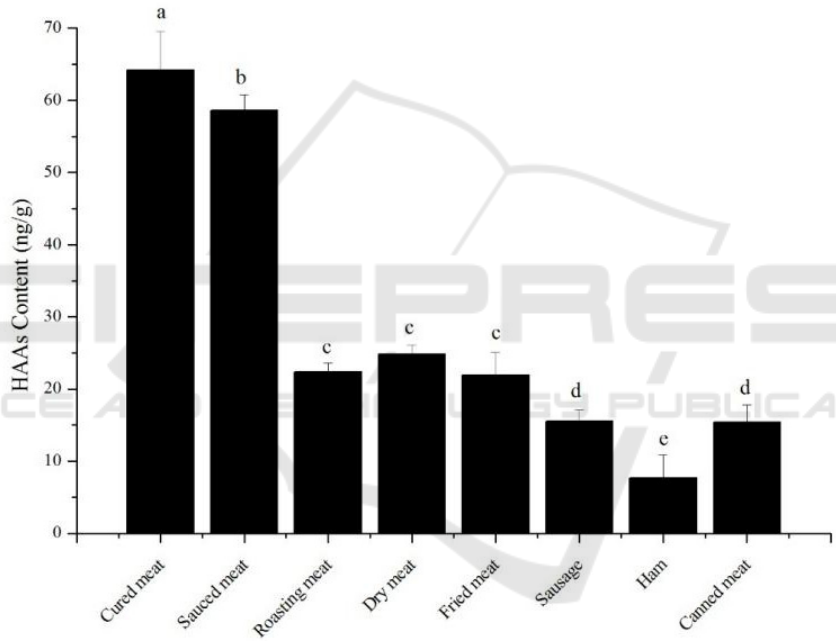

Figure 1: The total HAAs content of pork products in different processing methods.

In order to reflect the influence of different processing methods on the content of heterocyclic amines in pork products, the meat products with the highest content of heterocyclic amines among various types were selected to compare 8 types of meat products. It can be seen from Figure1 that the total amount of heterocyclic amines in descending order were Cured meat, Sauced meat, Dried meat, Roast meat, Fried meat, Canned meat, Sausage and Ham. The total amount of heterocyclic amines in cured meat products and braised meat products was much higher than other meat products, at 64.18ng/g and 58.64ng/g, which were almost 2-3 times the content of other meat products.

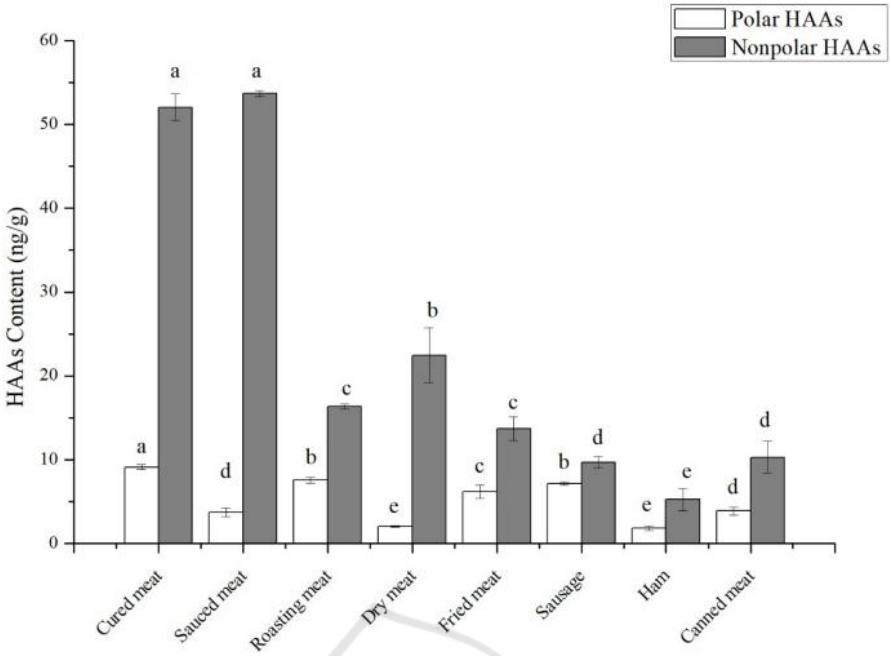

Figure 2: The polar and nonpolar HAAs content of pork products in different processing method**s.**

It can be seen from Figure 2 that the level of polar heterocyclic amines was very low. The content of all meat products surveyed was less than 10ng/g. Except for ham, the content of non-polar heterocyclic amines was almost above 10ng/g, the polar and non-polar heterocyclic amines of cured meat products were the highest, reaching 9.10ng/g and 52.05ng/g respectively. However, only non-polar heterocyclic amines were higher in cured and sauced meat products, while polar heterocyclic amines are lower.

4 CONCLUSIONS

The established LC-MS/MS method was used to detect the content of heterocyclic amines in common commercial pork products. All kinds of meat products have heterocyclic amines to varying degrees, and the total heterocyclic amine content was 7.68-64.18ng/g. Among them, the most common non-polar heterocyclic amines Harman and Norharman were 1.42-32.69 ng/g and 1.40-40.47 ng/g. The content of heterocyclic amines in meat products of different processing methods was significantly different, and the content of heterocyclic amines in cured meat products was the largest. The total amount of heterocyclic amines in descending order were Cured meat products, Sauced meat products, Dried meat products, Smoked and Roasted meat products, Fried products, Canned meat products, sausage and ham. It is inevitable to take in different levels of heterocyclic amines in the daily diet. This study is to investigate and evaluate the safety of pork products, and to further establish the exposure and prevention of heterocyclic amines in meat products.

ACKNOWLEDGEMENTS

This work was supported by Beijing Key Laboratory of Agricultural, Product Detection and Control of Spoilage Organisms and Pesticide. The authors express their thanks to Beijing Innovation Consortium of Swine Research System and Pengcheng food branch of Beijing Shunxin Agricultural Limited Company for providing the primary sample.

REFERENCES

(2011). Meat product classification. GB/T 26604-2011: 7.

Huang Yuanxiang, Zhao Shenglan, Ma Yage, et al (2019). Study on the formulation optimization and texture characteristics of low-calorie, low-sodium, wide-flavor sausage[J]. Meat Industry, (08):16-24

Kanithaporn P, Wannee J, Scott S J.(2011).Inhibitory activity of Asian spices on heterocyclic amines formation in cooked beef patties.[J]. Journal of food

science,76(8).
Liu Tiantian, Yao Yao, Wang Wei, et al.(2020).Formation and inhibition of three heterocyclic amines, PhIP, Norharman, and Harman in the model system[J]. Meat Research,34(03):1-7.
M, L. C. and K. T. H, et al. (2004).Effects of heating time and antioxidants on the formation of heterocyclic amines in marinated foods. Journal of chromatography. B, Analytical technologies in the biomedical and life sciences 802 (1).
Shen X C, Zhang Y L, Cui Y Q, et al.(2017). Simultaneous determination of 9 heterocyclic aromatic amines in pork products by liquid chromatography coupled with triple quadrupole tandem mass spectrometry[J]. IOP Conference Series: Earth and Environmental Science, 77(1).