Effects of dietary style on adult enterovirus infection risk factors: a retrospective analysis

Linghua Yu (yu.lh70s@gmail.com)
Jiaxing University https://orcid.org/0000-0001-7622-6127
Linlin Wang
Zhejiang University
Huixing Yi
First Affiliated Hospital Of Gannan Medical College
Xiaojun Wu
Jiaxing University
Fei Sun
Jiaxing University

Research Article

Keywords: Diet, adult, enterovirus infection

Posted Date: September 1st, 2020

DOI: https://doi.org/10.21203/rs.3.rs-64911/v1

Abstract

Background: Gut microbiota serves as a defense against enteric pathogens, whereas dietary intake influences the composition and function of gut microbiota. We aimed to examine the impact of diet on the enteroviral infection in adult patients of hand, foot, and mouth disease (HFMD).

Methods: A total of 266 adult patients of HFMD were recruited in this study, with 80 healthyvolunteers served as the control. Swab samples and clinical characteristics were collected. Enteroviral genotype was further assessed by PCR testing. Social-demographic data and dietary records were obtained through follow-up phone calls. Dietary patterns were derived with PCA analysis. Correlation between dietary patterns and clinical characteristics, enterovirus genotype, and HFMD risk factors were evaluated.

Results: Three distinct dietary patterns were identified in the participants, which were modern, "atypical south", and "traditional north", respectively. This study found the dietary pattern of adult HFMD significantly differed from that of the controls. A vast majority of controls followed the modern pattern, which was a healthy diet. In contrast, the result showed unhealthy dietary patterns ('atypical south' and 'traditional north') were risk factors for adult HFMD. Besides, the dining place was a leading contributor to the dietary pattern. Our data showed eating at a food stall, or take-out is a risk factor of adult HFMD, whereas eating at the dining room is a protective factor.

Conclusions: Our study indicated dietary pattern was associated with the incidence of adult HMFD. Improving the dietetic habit might contribute to HFMD prevention.

Background

Hand, foot, and mouth disease (HFMD) is a highly contagious disease that has overwhelmed the Asia-Pacific region in recent years [1–3]. HFMD usually occurs in infants and children under 5-years-old, but it can also affect adults [4, 5]. HFMD is caused by human enteroviruses, members of the Picornavirus family, which are primarily transmitted via the fecal-oral route or close contact with the infected individuals [6, 7]. The major causative agents of HFMD are Enterovirus 71 (EV-A71) and Coxsackievirus A16 (CV-A16), followed by Coxsackievirus A6 (CV-A6) and Coxsackievirus A10 (CV-A10) [8–11]. Among which, EV-A71 might cause severe neurological and cardiovascular complications and have a high fatality rate [12]. Besides, CV-A6-associated HFMD is becoming more prevalent in recent years [13].

Enterovirus infect human from the gastrointestinal (GI) tract via the fecal-oral route. The virus replicated in the GI tract first and then transmitted to other sites [14, 15]. The human GI tract is the key defender against insult from the outside environment [16, 17]. An ensemble of microorganisms collectively referred to as the gut microbiota resides in the human GI tract. Gut microbiota has a direct impact on human's metabolism, immunity, and response to infection [18–20]. The commensal bacteria promotes the resistance to colonization of pathogenic species by competing for nutrients that are important for pathogens' establishment in the gut [21–23]. Numerous studies reported dietary intake influences the composition, activity, and function of gut microbiota [24–26]. A study by Martens EC showed eating a

low-fiber diet have a higher risk of bowel infection by degrading the colonic mucus barrier, which serves as a defense against enteric pathogens [27]. Other studies proved that probiotics enhancing intestinal epithelial barrier function and protecting the host from enteric virus infection [28, 29]. However, there's no study revealing the impact of diet on the enteroviral infection to date.

This study systematically analyzed the epidemiological characteristics and dietary records of the participates. A total of 266 adult patients of HFMD were recruited in this study, with 80 healthy volunteers served as the control. Virus genotype was confirmed by laboratory testing. Social-demographic data and dietary records were obtained through follow-up phone calls. Dietary patterns were derived with PCA analysis. Correlation between dietary patterns and clinical characteristics, HFMD risk factors were evaluated. Results of the study suggested diet was correlated with adult HFMD pathogenesis and progression.

Results

Demographics of the study projects

A total of 299 adult patients were screened from 47,383 reported HFMD cases from August 2014 to June 2018. Results of PCR testing showed 266 patients were positive for the enterovirus. Finally, these 266 cases were recruited according to the inclusion criteria.

The ages of the 266 patients were ranging from 16 to 70 years old (27.5 ± 7.8) (Table 1). Females outnumbered males with a male-to-female ratio of 0.89. The most common occupation was house-hold (62/266, 23.3%), followed by the worker (57/266, 21.4%) and intellectual (54/266, 20.3%). Regarding the dining place, most of the adult patients preferred take-out or food stall (168/266, 63.2%), few of them ate at the dining room or restaurant (6/266, 2.3%). A vast majority of them didn't smoke (224/266, 84.2%), lived in the rural area (151/266, 56.8%), and had low income (167/266, 62.8%).

Table 1
Social-demographic characters of the participants in the study

Social-demographic characters	Control (n = 80)	HFMD (n = 266)	χ^2	p-value	OR (95% CI)
Age	28.53 ± 5.99	26.97 ± 7.57		0.07	
Gender			0.60	0.44	
Male	33	125			
Female	47	141			
Dietary type				**< 0.01**	
Modern	73	37			
Atypical south	2	132			130.22 (30.51 ~ 555.85)**
Traditional north	5	97			38.28 (14.34 ~ 102.19)**
Dining place				**< 0.01**	
at home	60	92			
Diningroom or restaurant	11	6			0.32 (0.12 ~ 0.92)**
Food-stall or take-out	9	168			13.70 (6.28 ~ 29.88)**
Smoking			0.51	0.48	
Yes	16	42			
No	64	224			
Residence			3.68	**0.05**	1.69 (1.02 ~ 2.79)*
Urban	45	115			
Rural	35	151			
Occupation				0.30	
Intellectual	17	54			
Worker	19	57			
Service	7	24			

*: p-value < 0.05, **: p-value < 0.01

Social-demographic characters	Control (n = 80)	HFMD (n = 266)	χ^2	p-value	OR (95% CI)
Farmer	4	36			
Student	8	33			
House-hold	25	62			
Income			5.33	**0.02**	1.86 (1.13 ~ 3.09)*
High	42	99			
Low	38	167			
*: p-value < 0.05, **: p-value < 0.01					

This study found several epidemiological characteristics of adult patients of HFMD that were significantly differ from those of the controls, including dietary pattern (p-value < 0.01), dining place (p-value < 0.01), residence (p-value = 0.05) and income (p-value = 0.02) (Table 1).

Clinical Characteristics Of The Adult Patients With HFMD

Few of the adult patients of HFMD (59/266, 22.2%) in this study presented with fever (37.4 ± 0.54℃), and none had severe complications (Table 2). A majority of the patients had an oral ulcer (205/266, 77.1%) and vesicular rash on the hands, feet, or buttocks (219/266, 82.3%). Patients tended to have normal white blood cells (WBC), creatine kinase-MB (CK-MB), lactate dehydrogenase (LDH), alanine amiontransferase (ALT), aspartate aminotransferase (AST), and Glucose (GLU). Remarkably, most of the patients presented with high C-reactive protein (CRP) comparing with the reference value (243/266, 91.4%). No severe case was found in this study, and the prognosis of adult patients of HFMD was generally good.

Table 2
The clinical characteristics of adult patients of HFMD

Clinical characteristics	Adult HFMD	Reference
WBC (10^9/L)	6.52 ± 1.09	3.50–9.50
CK-MB (IU/L)	18.92 ± 5.96	0–24
LDH (U/L)	193.27 ± 32.32	120–250
ALT (IU/L)	35.12 ± 5.98	7–40
AST (IU/L)	26.04 ± 3.76	15–40
CRP (mg/L)	**28.12 ± 6.08**	0–8.2
GLU (mmol/L)	4.99 ± 0.60	3.90–6.10
Temperature (℃)	**37.43 ± 0.54**	36.3–37.2
Fever (%)	**22.2**	-
Rash (%)	**82.3**	-
Oral ulcer (%)	**77.1**	-

Enterovirus Strains

In consistence with previous reports [30], the most prevalent enterovirus strain was CV-A16 (101/266, 37.97%), followed by EV-A71 (61/266, 22.93%), pan-enterovirus (61/266, 22.93%), CV-A6 (37/266, 13.91%), and CV-A10 (6/266, 2.26%) (Fig. 2A).

Aligned with the prior reports [30], EV-A71-associated adult HFMD cases decreased in recent years (Fig. 2B). Although sporadic cases of CV-A6 occurred since the beginning of this study, the epidemic of CV-A6-associated HFMD started in October 2016. Meanwhile, no pan-enterovirus-associated HFMD case was reported from August 2015 to August 2017. Because a multitude of the pan-enterovirus-associated HFMD cases could be confirmed positive for CV-A6. CV-A10-associated cases were rare in this study, it was first reported in the year 2016.

Risk Factors For Adult HFMD

This study analyzed the social-demographic characteristics of participants with a single-factor analysis ($\alpha = 0.1$) to determine the risk factors of adult HFMD (Table 1). Results showed that eating at food stall or take-out (odds ratio (OR): 13.70, 95% confidence interval (CI): 6.28–29.88), living at rural (OR: 1.02, 95% CI: 1.02–2.79), and low income (OR: 1.86, 95% CI: 1.13–3.09) were risk factors of adult HFMD. Interestingly, this study indicated eating at dining room or restaurant (OR: 0.32, 95% CI: 0.12–0.92) are protective factors of adult HFMD.

Dietary Pattern Of The Participants

Principal components analysis (PCA) revealed three distinct dietary patterns: modern, "atypical south",and "traditional north".The three principal components (PC1, PC2, and PC3) accounted for 26% (13.3%, 7.0%, and 5.7%, respectively) of the variance in total food group intake (Fig. 3A/B). The loadings of food groups of each dietary pattern were shown in Table 3. The modern dietary pattern (PC1) was highly correlated with the intake of fruit (0.321), poultry (0.285), aquatic products (0.277), milk (0.273), fruit juice or vegetable juice (0.267), fungi or algae (0.257), cereals (0.234), and tea (0.209), while negatively correlated with alcoholic beverages (-0.318) and carbonated drink (-0.277). The "atypical south" pattern (PC2) was characterized by high intakes of rice (0.566) and rarely intake of wheat (-0.581), legumes (-0.291), and vegetables (-0.086). The "traditional north" pattern(PC3) was positively correlated with the intake of wheat (0.217), tubers (0.397), and edible herbs (0.411), whereas negatively associated with the intake of rice (-0.221), vegetables (-0.220) and legumes (-0.205).

Table 3
Dietary type of the participants in the study

Food Items	Modern	Atypical south	Traditional north
Rice	-0.001	**0.566**	**-0.221**
Wheat	-0.016	**-0.581**	**0.217**
Other cereals	**0.234**	0.079	-0.196
Tubers	0.132	0.113	**0.397**
Legumes	0.169	**-0.291**	**-0.205**
Vegetables	0.121	-0.086	**-0.220**
Fungi & algae	**0.257**	-0.004	0.139
Fruit	**0.321**	0.105	**0.211**
Nuts	0.196	-0.047	-0.175
Red meat	0.114	0.195	-0.106
Organ meat	-0.192	0.145	0.181
Poultry	**0.285**	-0.196	-0.084
Milks	**0.273**	0.132	0.024
Eggs	0.018	**0.203**	-0.190
Aquatic products	**0.277**	-0.079	-0.065
Western fast food	-0.103	-0.026	-0.187
Chinese fast food	-0.193	-0.048	0.009
Carbonated drink	**-0.277**	-0.028	-0.066
Alcoholic beverages	**-0.318**	-0.110	**-0.262**
Fruit & vegetable juice	**0.267**	-0.115	-0.113
Tea	**0.209**	0.037	-0.044
Sweets & Candies	-0.156	-0.077	**-0.353**
Ethnic foods	-0.134	-0.053	0.124
Edible herbs	-0.106	0.131	**0.411**

Association Of Dietary Type And Social-demographics

The dietary pattern of each participant was determined by the factor score. Results showed a vast majority of the controls belonged to modern pattern (73/80, 91.3%), whereas the predominated dietary pattern of adult HFMD patients was "atypical south" (132/266, 49.6%), followed by "traditional north" (97/266, 36.5%) and modern (37/266, 13.9%) (Table 1). Comparing with the modern pattern, dietary pattern "atypical south" (OR: 130.22, 95% CI: 30.51–55.85) and "traditional north" (OR: 38.28, 95% CI: 14.34–102.19) were risk factors of adult HFMD.

Association of various dietary patterns with social-demographic characteristics were further investigated (Table 4). Comparing with the control, there was a significant difference in the dining place (eating at home, dining room, or take-out) for all three dietary patterns (p-value < 0.01 for all three patterns). In addition, "traditional north" pattern presented a significant difference in smoking (p-value < 0.05).

Table 4
The distribution of social-demographic characters related to the dietary type [1]

Social-demographic characters	Modern	Atypical south	Traditional north
Gender	0.07	0.57	0.84
Smoking	0.83	0.05	**3.79***
Dining place	**27.07****	**21.47****	**9.64****
Residence	2.61	0.60	0.19
Occupation	3.16	7.04	3.79
Income	1.81	0.88	0.66
1: χ^2			
*: p-value < 0.05, **: p-value < 0.01			

Spatial Distribution Of The Dietary Pattern

For a better understanding of the association between dietary patterns and adult HFMD morbidity, the geographic distribution of both was further analyzed. Results showed there is statistical significance in the geographic distribution of adult HFMD patients and the control (p-value < 0.01), whereas no statistical difference was found between various dietary patterns in the geographic distribution (p-value = 0.3). A heat map was used to represent the density of the participates. No similarity was found between the adult HFMD morbidity (Fig. 4A) and various dietary patterns (Fig. 4B/C/D). Most of the adult HFMD cases were clustered in several spots, whereas the dietary patterns were distributed evenly all over the map.

Association Of Dietary Pattern And HFMD Risk Factors

A multivariate model was used to assess the associations between dietary patterns and the aforementioned risk factors of adult HFMD. Results indicated "eating at home" (β coefficients: -0.61 ± 0.20, p-value < 0.01) and "eating at dining room or restaurant" (β coefficients: -0.63 ± 0.13, p-value < 0.01) were negatively associated with the modern pattern (Table 5). Similarly, "eating at home" (β coefficients: 0.53 ± 0.19, p-value < 0.01) and "eating at food stall or take-out" (β coefficients: 0.31 ± 0.12, p-value < 0.01) positively contributed to the "atypical south" pattern. Besides, this study found "eating at food stall or take-out" (β coefficients: 0.26 ± 0.12, p-value = 0.03) was positively associated with the "traditional north" pattern.

Table 5

Associations between dietary patterns and HFMD risk factors [1]

HFMD risk factors	Modern	Atypical south	Traditional north
Eating at home	-0.61 ± 0.20**	0.53 ± 0.19**	0.03 ± 0.19
Eating at dining room or restaurant	0.08 ± 0.13	-0.08 ± 0.12	0.02 ± 0.13
Eating at food stall or take-out	-0.63 ± 0.13**	0.31 ± 0.12**	0.26 ± 0.12*
Residence	-0.32 ± 0.28	-0.09 ± 0.25	0.35 ± 0.26
Income	0.37 ± 0.27	0.04 ± 0.25	-0.50 ± 0.27
1: βcoefficients ± standard error			
*:p-value < 0.05, **: p-value < 0.01			

Correlation Between Dietary Type And Clinical Characteristics

This study examined the association between clinical characteristics of adult patients and the dietary patterns. Correlation analysis showed there was no statistical difference between the clinical characteristics (WBC, CK-MB, LDH, ALT, AST, CRP GLU, peak temperature, fever, rash, and oral ulcer) and various dietary patterns (modern, atypical south, traditional north) (Table 6).

Table 6

Associations between dietary type and clinical characteristics [1]

Clinical characteristics	Modern	Atypical south	Traditional north
WBC (10^9/L)	-0.03	-0.08	-0.03
CK-MB (IU/L)	0.12	-0.04	-0.1
LDH (U/L)	0.02	0.05	-0.02
ALT (IU/L)	0.03	0.08	-0.05
AST (IU/L)	-0.09	-0.08	0.03
CRP (mg/L)	0.08	-0.06	-0.05
GLU (mmol/L)	0.07	-0.03	-0.05
Temperature (°C)	-0.04	0.08	0.13
Fever (%)	-0.01	0.01	0.03
Rash (%)	-0.03	0.02	-0.07
Oral ulcer (%)	0.02	0.1	-0.04
1: Spearman correlation coefficients			
*: p-value < 0.05, **: p-value < 0.01			

Correlation Between Dietary Type And Enterovirus Strains

We examined the association between enterovirus strains and the dietary patterns of the participants. Correlation analysis showed there was no statistical difference between the enterovirus strains (EV-A71, CV-A16, CV-A6, CV-A10, and Pan-enterovirus) and various dietary patterns (modern, atypical south, traditional north) (Table 7).

Table 7

Associations between dietary type and enterovirus genotype [1]

Clinical characteristics	Modern	Atypical south	Traditional north
EV-A71	-0.01	0.02	0
CV-A16	-0.02	-0.05	-0.11
CV-A6	0.10	0	0.03
CV-A10	0.05	0.01	0.01
Pan-enterovirus	-0.06	0.03	0.09
1: Spearman correlation coefficients			
*: p-value < 0.05, **: p-value < 0.01			

Materials And Methods

Ethics statement

This study was approved by the Ethical Committee of the Affiliated Hospital of Jiaxing College (reference number: 2014096). All individual-level data were anonymized. Written informed consent was obtained from all participants.

Study Design

This study screened adult patients from reported HFMD cases. Cases were further confirmed by PCR testing. Clinical characteristics data were collected. Social-demographic data and dietary records were obtained through follow-up phone calls. Dietary patterns were derived with PCA analysis. Correlation between dietary patterns and clinical characteristics, HFMD risk factors were evaluated. The purpose of this study was to reveal the association between dietary and HFMD pathogenesis and progression.

Case Definition

Clinical criteria for the diagnosis of HFMD was published by the Chinese Ministry of Health in 2010 [42]. Patients with the following symptoms were defined as having HFMD: fever, oral ulcers, and vesicular rash on the hands, feet, or buttocks.

Study Population

The inclusion criteria for the enrollment of this study were as the following: 1) diagnosed as HFMD case; 2) positive for enterovirus by laboratory testing; 3) adult patients who defined as 16-years-old or older; 4) could be tracked through follow-up phone calls.

A total of 299 adult patients were screened from 47,383reported HFMD cases in Jiaxing from August 2014 to June 2018. Among which, 266 cases were confirmedpositive for enterovirus by laboratory testing. According to the inclusion criteria, this study recruited 266 adult patients of HFMD, with 80 healthy adult subjects served as the controls. All healthy volunteers were negative for enteroviral infection. Socio-demographic data of adult HFMD patients were collected through follow-up phone calls.

Enterovirus Genotype

Throat swab specimens from 299 adult patients of HFMD were collected by trained medical personnel. Samples were preserved at -80℃.RNA was extracted from the specimens by TRIzol (Invitrogen, CA, USA). The cDNA sample was synthesized by using the PrimeScript TM RT kit (Takara, Dalian, China). One-step RT-PCR assays were performed to detect enterovirus RNA, using EV-A71/CV-A16/Pan-enterovirus commercial kits and CV-A6/ CV-A10 commercial kits (Da An Gene Co. Ltd, China).

Dietary Assessment

A semi-quantitative FFQ (food frequency questionnaires) was used to assess the weekly intake of foods. The FFQ contained 24 food groups from the China Food Composition data, with each food group included several typical food items (Supplementary Table 1). Participants were asked during the follow-up phone calls how often they had consumed each food group over the last week (Supplementary Table 2).

The 1-week dietary records were then analyzed with PCA (Principal components analysis) to assess the dietary pattern, which was the uncorrelated linear combinations of food groups extracted by PCA. The factors were rotated by an orthogonal transformation (Varimax rotation) for better interpretability. Dietary pattern score was calculated as a weighted sum of food intake by the factor loading extracted by PCA. A food group was positively correlated with the dietary pattern when the corresponding factor loading had a positive value, whereas a negative association existed with a negative factor loading value. Further, the contribution of the food group to the dietary pattern was represented by the corresponding factor loading value. Food groups with absolute loading greater than 0.2 were retained. For every dietary pattern, a factor score was calculated for each individual, in which intakes of 24 food groups were weighted by their factor loadings and summed, to determine the dietary pattern of the participates.

Statistical analysis

Proportional data were analyzed usingχ^2 tests or Fisher exact test. Continuous data were tested by Student's t-test. Multivariate models were used to assess the linear relationship between dietary patterns and adult HFMD risk factors. Correlation between dietary patterns and clinical characteristics was calculated by Spearman correlation coefficients. Data were preprocessed by Python 3.6 and statistics were performed by R 3.5.1. A difference with p-values below 0.05 was considered to be statistically significant.

Discussion

Adult hand, foot, and mouth disease is used to be seen as a mild self-limiting viral infection, and its hazard was underestimated [33–35]. Our study showed female patients outnumbered males with a male-to-female ratio of 0.89, and most of the patients were housewives, teachers (intellectual), and students. These patients had a high chance of close contact with the children. Since close contact with the infected individuals is a primarily transmitting route of human enterovirus, these adult patients might serve as a potential infectious source of HFMD.

Consistent with the previous reports, our study demonstrated CV-A16 is the most prevalent causative agents of adult HFMD, followed by EV-A71, Pan-enterovirus, CV-A6, and CV-A10 [8–11]. Interestingly, this study found the incidence rate of CV-A6-associated adult HFMD increased since the year 2016. This finding was in accordance with the prior reports that CV-A6-associated HFMD is becoming more prevalent in recent years and having high morbidity in adults [13]. Notably, our study demonstrated the morbidity of adult HFMD dropped in the year 2017 and elevated dramatically in the year 2018. Taken together, these findings reminded us of the tough situation in HFMD prevention.

Numerous studies reported diet alters the composition and activity of gut microbiota, and in turn influences gut's resistance to colonization of pathogens [21–23]. Our study revealed three dietary patterns (modern, atypical south, and traditional north) of the participates. The modern pattern presented as a healthy diet, which characterized by the main intake of fruit, poultry, aquatic products, milk, vegetable juice, fungi or algae, and cereals. Whereas the "atypical south" and "traditional north" could be classified as unhealthy diet, which characterized by low-fiber (rarely eating of vegetable and legumes) and high-fat (red meat, organ meat) food groups. A low-fiber dietor high-fat diet would impactthe host's health. A study indicated the low-fiber diet would degrade the colonic mucus barrier which serves as a defender against enteric pathogens [27]. Other studies proved a high-fat diet promoted pathobiont expansion and caused colitis or even intestinal carcinogenesis [36, 37].Consistent with these reports, our study showed dietary patterns "atypical south" and "traditional north" are risk factors of adult HMFD. As mentioned by prior studies, the long-term of low-fiber/high-fat diet ("atypical south" and "traditional north" patterns) might affect the composition and function of gut microbiota, and weaken the protective effect of gut against the colonization andreplication of enterovirus. Proceeding studies reported probiotics could enhance intestinal epithelial barrier [38, 39]. Commensal bacteria, such as lactobacillus, can protect human intestinal epithelial from enteric virus infection [28, 29]. Consistently, our study indicated a vast majority of controls followed the modern diet, which is rich in indigestible fibers. The fermentation of indigestible fibers produces short-chain fatty acids, which is essential in anti-inflammation and intestinal homeostasis [40, 41]. The impact of microbiome on the enterovirus infection required further studies.

Besides what's to eat, where to eat also matters. Our study revealed the dining place is a significant contributor to the dietary pattern. Eating at food stall or take-out was a risk factor of adult HFMD, whereas eating at the dining room was a protective factor. Multivariate linear regression analysis showed "eating at home" and "eating at food stall or take-out" were negatively correlated with the modern dietary pattern,

whereas these dining places positively contributed to the unhealthy dietary patterns "atypical south" and "traditional north". Most of the food stall or take-out faced poor sanitary condition, especially the "gutter oil" problem. Besides, the best selling on the food-stall or take-out was always low-fiber/high-fat food like organ meat and fast food. Notably, "eating at home" did not necessarily mean eating healthy. Imbalanced diet and dietary preference might be the problem of "eating at home".

Conclusions

In conclusion, our study analyzed the dietary pattern and its association with the epidemiological and clinical characteristics of adult HFMD. To the best of our knowledge, this is the first study focused on the impact of diet on the morbidity of HFMD. However, the dietary records collected in this study can't reflect this seasonal pattern. Food records in the different season should be analyzed in the future study.

Abbreviations

HFMD; hand, foot, and mouth disease; EV-A71: Enterovirus 71; CV-A16: Coxsackievirus A16; CV-A10: Coxsackievirus A10

Declarations

Acknowledgments

We thank the Centers for Disease Control OF Jiaxing.

Authors' contributions

LHY participated in the design, data analysis and interpretation, and drafted the manuscript. XJW and FS performed the survey. LLW and HXY participated in the interpretation of data and helped to finalize the manuscript. All the authors have read and approved the final manuscript.

Funding

Supported by grants from the Zhejiang Provincial Natural Science Fund(LY16H030016, LY17H030012), the Zhejiang Science and Technology Public Welfare Project(2015C33279). Center for gastroenterology and hepatology connecting with Shanghai. Key medical discipline in jiaxing –Gastroenterology(2019-ZC-08)

Availability of data and materials

The dataset used in the study are available from the corresponding author

on reasonable request.

Ethics approval and consent to participate

This study was approved by the Ethical Committee of the Affiliated Hospital of Jiaxing College (reference number: 2014096). All individual-level data were anonymized. Written informed consent was obtained from all participants.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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Supplementary Tables

Supplementary table 1. Food groups

Food groups	Foods included in the group
Rice	Rice and rice products
Wheat	Wheat bun, wheat noodles
Other cereals	Corn, barley, millet,
Tubers	Tubers, starches, potato
Legumes	Soybean, mung bean, adzuki bean, and products
Vegetables	Leafy vegetable, root vegetable, leguminous vegetable and aquatic vegetable
Fungi & algae	Mushroom, laver, kelp, and others
Fruit	Apple, peach, pear, orange, berry, melons, and others
Nuts	Nuts, seeds
Red meat	Pig, beef, lamb, and others
Organ meat	Organ meat
Poultry	Chicken, duck, goose, and others
Milks	Liquid milk, milk powder, yogurt, cheese, butter, and others
Eggs	Eggs
Aquatic products	Fish, shrimp, crab, shellfish, and others
Western fast food	Hamburger, pizza, hotdog, sandwich, French fries, and others
Chinese fast food	Convenience food, take-out
Carbonated drink	Coke, Sprite, and others
Alcoholic beverages	Liquor, beer, yellow rice wine, wine
Fruit& vegetable juice	Fruit juice, vegetable juice
Tea	Green tea, red tea, and others
Sweets & Candies	Sugars, honey, candies,preserves, confectionary
Ethnic foods	Ethnic food, cake, cookies, snack
Edible herbs	Mint, liquorice, carthamus, and others

Supplementary table 2. Food frequency questionnaires

Participants No: Date:

Participants Type: ☐ Control ☐ HFMD ☐ Measles ☐ Drug eruption

Gender: ☐ Male ☐ Female

Are you a local resident? ☐ Yes ☐ No

Where do you live? ☐ Urban ☐ Rural

Profession:

☐ intellectual ☐ blue-collar worker ☐ clerk ☐ farmer ☐ unemployed
☐ retired ☐ house-hold

Toxic substance contact history:

☐ working in a chemical factory ☐ working in dyeing mill
☐ living in newly decorated house ☐ none

Drink alcohol: ☐ never ☐ occasionally ☐ frequently

Smoking: ☐ never ☐ occasionally ☐ frequently

FriedFood:

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

SaltedFood:

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Smoked Food:

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Rice (rice and rice products):

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Wheat (wheat bun, wheat noodles):

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Cereals (corn, barley, millet):

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Tubers(tubers, starches, potato):

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Legumes(soybean, mung bean, adzuki bean, and legumes products):

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Vegetables (leafy vegetable, root vegetable):

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Fungi and algae (mushroom, laver, kelp):

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Fruit (apple, peach, pear, orange, berry, melon):

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Nuts (nuts, seeds):

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Red meat (pig, beef, lamb):

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Organ meat:

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Poultry (chicken, duck, goose):

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Milk (liquid milk, milk powder, yogurt, cheese, butter):

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Eggs:

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Sweets & Candies (sugars, honey, candies, preserves, confectionary):

☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Carbonated Drink (Coke, Sprite):
☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Aquatic products (fish, shrimp, crab, shellfish):
☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Western Fast Food (hamburger, pizza, hotdog, sandwich, French fries):
☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Chinese Fast Food (convenience food, take-out):
☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Alcoholic beverages (liquor, beer, yellow rice wine, wine):
☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Fruit and vegetable juice (fruit juice, vegetable juice):
☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Tea (green tea, red tea):
☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Ethnic foods (ethnic food, cake, cookies, snack):
☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Edible herbs (mint, liquorice, carthamus, and others):
☐ never ☐ 1~3 x per week ☐ 4~6 x per week
☐ 1 x per day ☐ 2 x per day ☐ 3 x per day or more

Figures

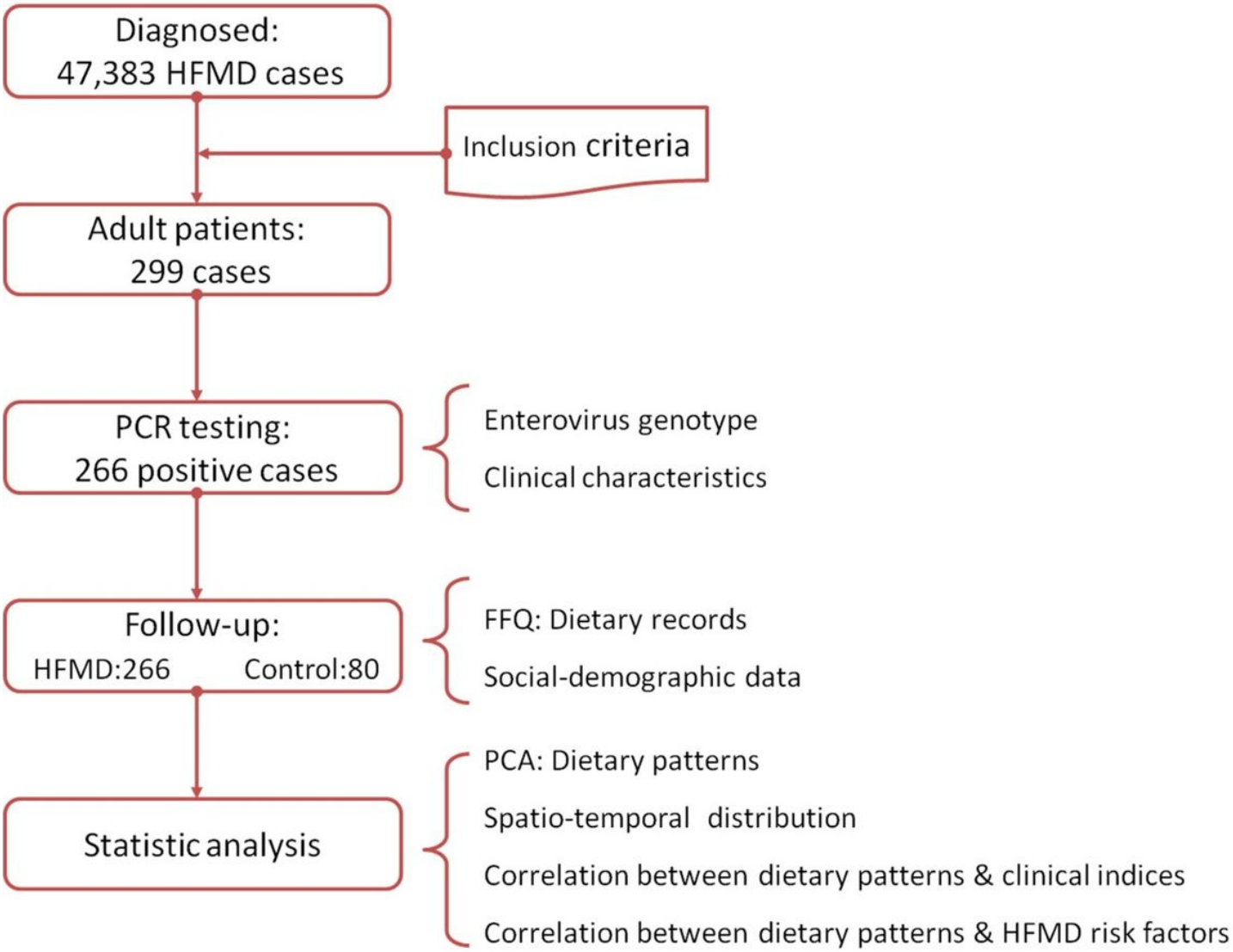

Figure 1

Study design.

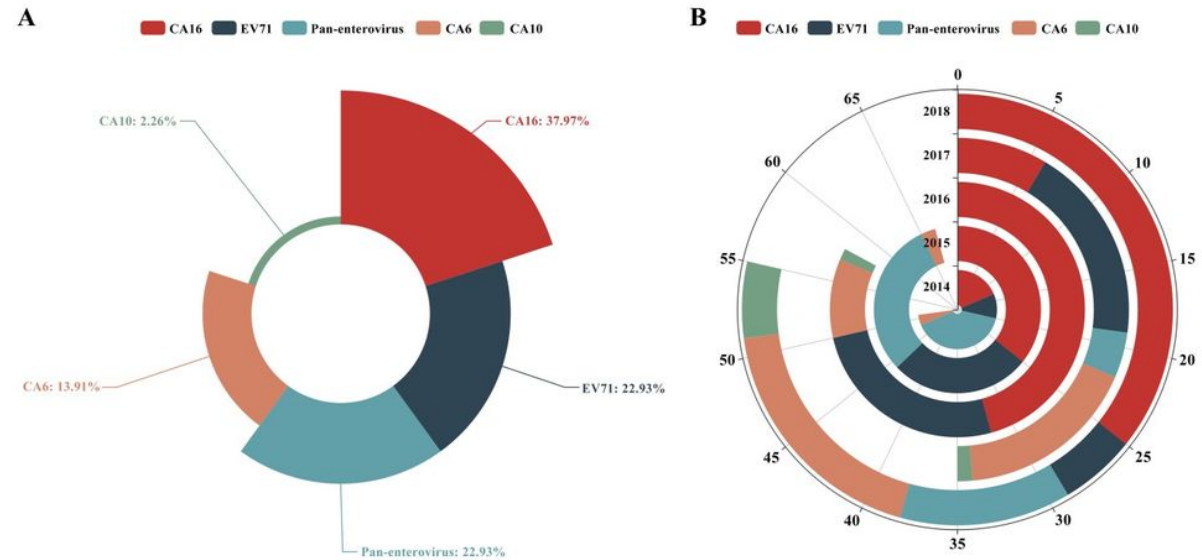

Figure 2

Distribution of enterovirus genotype.

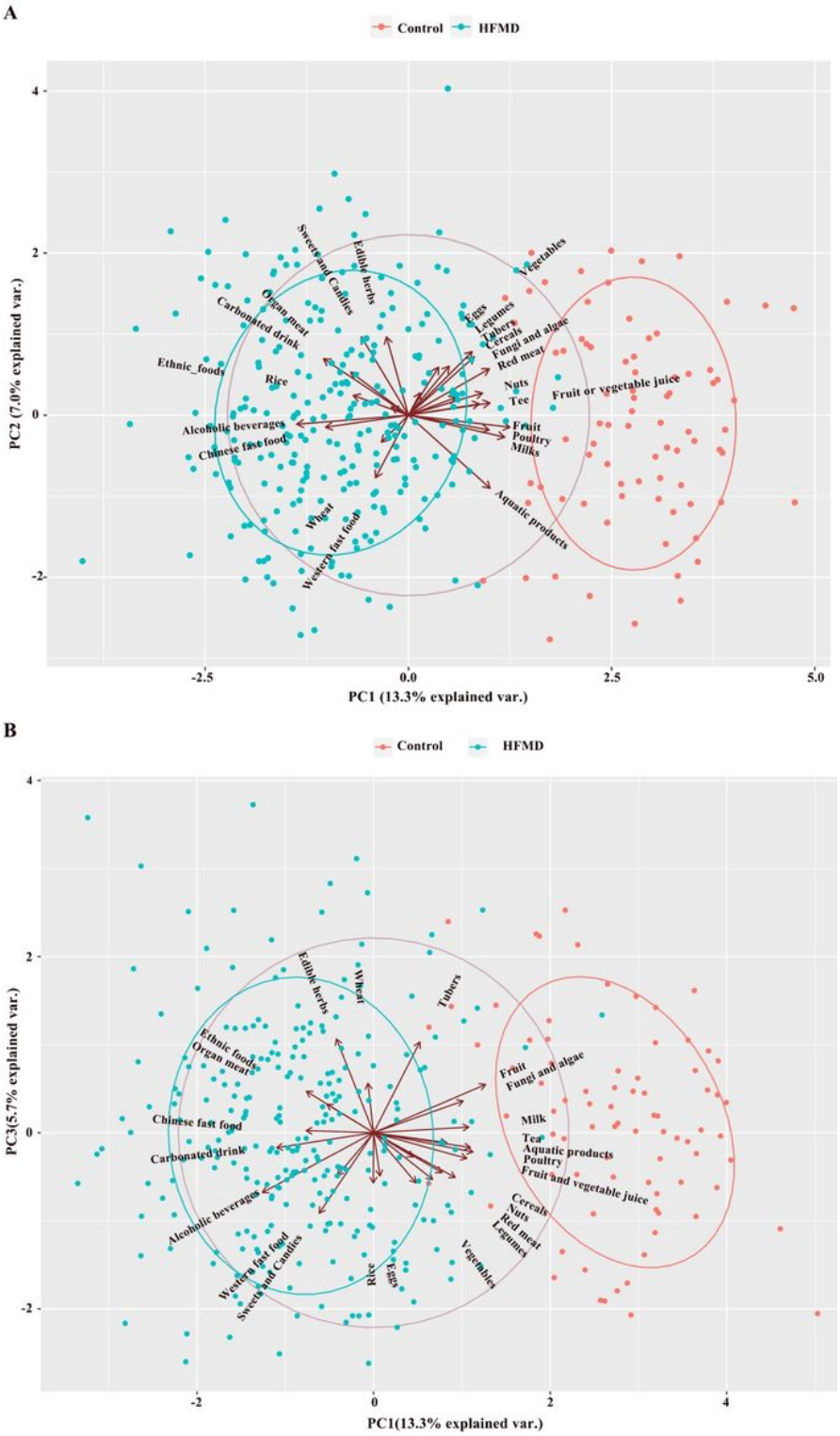

Figure 3

Dietary patterns of adult HFMD patients.

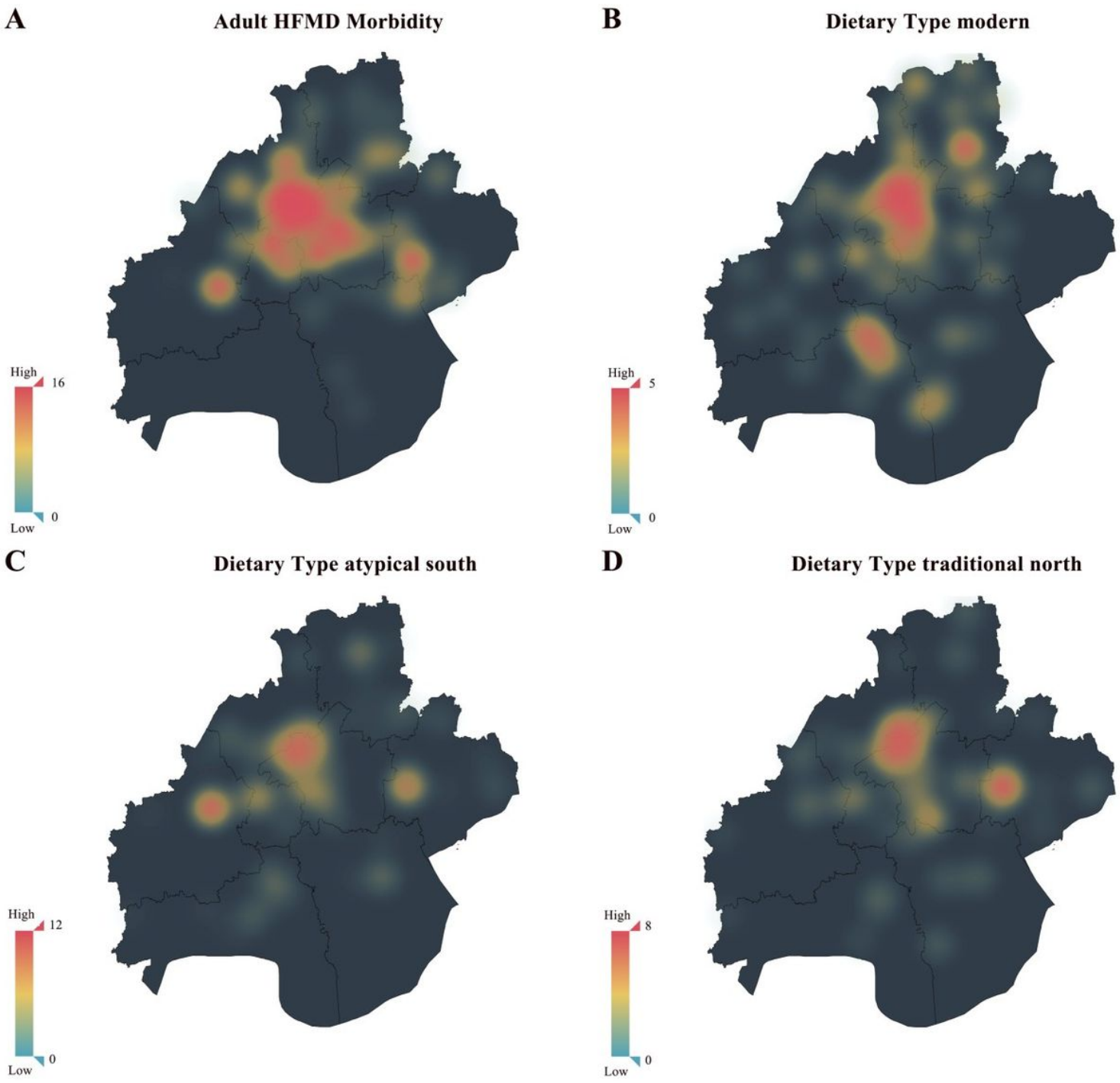

Figure 4

Spatial distribution of dietary patterns.