Huiming Zhou1#, Jinxian Gao2#, Xianghui Zhu¹, Zhentao Zhang3* and Changxi Bai1*

1Key Laboratory of Inner Mongolia Autonomous Regions for Mongolian Medicine, Academy of Mongolian Medicine, Inner Mongolian Medical University, Jinshan Economic Development Zone, Hohhot 010110, Inner Mongolia, China
2Department of Pharmacology, College of Nationality Medicine, Inner Mongolian Medical University, Jinshan Economic Development Zone, Hohhot 010110, Inner Mongolia, China
3Department of Pharmacochemistry, College of Pharmacy, Inner Mongolian Medical University, Jinshan Economic Development Zone, Hohhot 010110, Inner Mongolia, China
*these authors contributed equally to this work

Dates: Received: 23 January, 2017; Accepted: 09 February, 2017; Published: 14 February, 2017

**Corresponding authors:** Changxi Bai, Key Laboratory of Inner Mongolia Autonomous Regions for Mongolian Medicine, Academy of Mongolian Medicine, Inner Mongolian Medical University, Jinshan Economic Development Zone, Hohhot 010110, Inner Mongolia, China Tel: +86-0471-6653161, E-mail: changbai2010@126.com

Zhentao Zhang, Department of Pharmacochemistry, College of Pharmacy, Inner Mongolian Medical University, Jinshan Economic Development Zone, Hohhot 010110, Inner Mongolia, China, E-mail: 857805364@qq.com

Keywords: Regular consumption; Pepper; Allergic Constitution; Harmful actions

https://www.peertechz.com

Introduction

Pepper (chili or hot pepper) (Capsicum annuum L.) is the fruit from plant of Capsicum that belongs to the Solanaceae, one important plant for homology of food and medicine. In the traditional medicine including Mongolian Medicine, Chinese Medicine, Indian Medicine and Tibetan medicine, etc., pepper has pungent flavor and hot-natured, has been used to treatment of cold syndrome, for example cold syndrome of stomach and pain from rheumatism, etc. [1–5]. In the Western Medicine, pepper exhibit a variety of pharmacological effects, including analgesia, antibiosis, anticancer, anti-radiation, reduce weight and treatment of some skin diseases [6], etc. At present, peppers spread all over the world, 1/4 people world worldwide are often taking peppers and become a dietary habit, and growing number of people eating peppers.

However, side or toxic effects of peppers are unclear. A notable problem is growing number of people gating allergic constitution, up to 1/4 people world worldwide are suffer from allergic constitution[6–8]. We observed clinically many people who have allergic constitution are often taking peppers. The relation between often taking peppers and getting allergic constitution are also unclear. Constitution is an important form of human life as well as an integrated, relatively stable trait in the performance of the morphological structure, physiological function based on genetic and acquired. Constitution is closely related to human health and disease [7,9]. Therefore, the research on the relationship between constitution and disease has become an important subject in the Medical Sciences and Life Sciences research. Individual immune function disorder has a crucial role in the formation of allergic constitution. The main characteristics of the immune function disorder of allergic constitution are high level serum immunoglobulin , abnormal in the proportion of Helper T cells 1(Th1) and Helper T cells 2(Th2 ) class two cells and often Th2 cell dominant, high level serum histamines, leukotriene (LT) and interleukin 16 (IL-16), an allergic asthma related chemokine, etc.. Latest research shows that excessive accumulation of free radicals in the body is closely related to formation of allergic constitution and allergic attack [10–12]. In the present study, therefore, we examined the IgE, IgM, IgG, histamine, Th1-associated cytokines such as interleukin 2 (IL-2), γ- interferon (IFN-γ) and...
β- tumor necrosis factor (TNF-β), Th2-associated cytokines such as IL-4, IL-5, IL-6 and IL-10, an allergic asthma related chemokine IL-16, and LT, activity of superoxide dismutase (SOD), contents of malonaldehyde (MDA), bile acid (BA) and immune cells in the serum of rabbit that was administrated by peppers; to test whether often taking peppers changes body constitution to allergic constitution.

Materials and methods

The investigation was conducted in accordance with the rules and regulations of the Institutional Animal Care and Use Committee of Inner Mongolia Medical University.

Experimental animal

18 Healthy male Japanese white rabbits (2.0±0.2 kg body weight) were purchased from Xing Long Experimental Animal Central (Peking, China). Animals were housed in a temperature-controlled environment (22°C) with alternating 12-hour light-dark cycles. The 18 rabbits were randomly divided into control group (C), low dose peppers group (LDP) and high dose peppers group (HDP), 6 rabbits in each group.

Materials

Reagents were purchased from Sigma Chemical Co. (St. Louis, MO) unless otherwise indicated. Peppers (chilli) were purchased from Sichuan Honglin Foods Co; Ltd.in China. Commercially available Kits (ELISA) that using for measuring indexes such as IgE, IgM, IgG, histamine, IL-2, IFN-γ, TNF-β, IL-4, IL-5, IL-6, IL-10, LT, SOD, MDA, and BA in the serum of rabbit were purchased from Peking Huan Ya Tai Biomedical Technology Co., Ltd in China.

Preparing peppers powder and peppers solution

Made of 120 fine powder of peppers by a pulverizer special equipment. 10 min before intragastric administration, make 20% peppers solution (20g peppers: 100 ml double distilled water) for administration of low dose peppers and 40% peppers solution (40g peppers: 100 ml double distilled water) for administration of high dose peppers.

Intragastric administration of peppers or distilled water

Rabbits in the low and high dose peppers group were administrated by peppers by means of intragastric administration q.d. using a stomach pump for 3 months. The low dose peppers group rabbits were administrated by peppers 1.443 g /kg/rabbit (Equivalent to 5g of adult 70kg body weight, q.d.), the high dose peppers group were administrated by peppers 4.329 g /kg/rabbit (equal to 3 times the low dose). Rabbits in the control group were administrated by only double distilled water with the same volume and stomach pump as the low and high dose peppers group.

Preparation of serum samples and assessment of the indexes

The end of the experiment (after 3 months administrated peppers), all of rabbits were fasted 12 hours, take 10 ml of venous blood and placed it at room temperature after coagulation, and then Centrifugal the coagulated blood for 10 minutes and take the serum. The indexes (such as IgE, IgM, IgG, histamine, IL-2, IFN-γ, TNF-β, IL-4, IL-5, IL-6, IL-10, LT, SOD, MDA, BA and immune cells) are determined in strict accordance with the kits (ELISA) and the relevant instructions and procedures.

Statistical analysis

Origin Pro 8.5.1 analysis software was used for Statistical analysis. All data are expressed as mean ± SE. The differences between groups were analyzed by Student’s t test. Statistical significance was set at P <0.05, very significant difference was set at P <0.01.

Results

Effects of pepper on the serum level of IgE, IgM, IgG and histamine

Rabbits were administrated by peppers for 3 months, compared with the control group, the contents of IgM in the serum of rabbits were increased at all of the testing points (end of the first month, second month and third month administrated peppers) in the both of low dose peppers group and high dose peppers group (P <0.05) and more significant in the low dose peppers group, as shown in the Figure 1A; the contents of both IgE and IgG were slightly decreased in the both of low dose peppers group and high dose peppers group, but no significant (P > 0.05); the concentrations of histamine
in the serum were not significantly changed in both of the low dose peppers group and high dose peppers group (P > 0.05).

Effects of pepper on the serum level of LTC4, LTD4 and LTE4

As shown in the Figure 1B, compared with the control group, the contents of LTD4 in the serum of rabbits were increased at all of the testing points (end of the first month, second month and third month administrated peppers) in the both of low dose peppers group and high dose peppers group and more significant in the low dose peppers group, (P <0.05) ; whereas the contents of LTC4 and LTE4 in the serum were not significantly changed in the both of low dose peppers group and high dose peppers group (P > 0.05).

Effects of pepper on the serum level of the Th1-associated cytokines

Compared with the control group, the contents of IFN-γ in the serum of rabbits were increased at all of the testing points (end of the first month, second month and third month administrated peppers) in the high dose peppers group (P <0.05), as shown in Figure 2A, no significant change in the low dose peppers group (P > 0.05). The contents of IL-2 was significantly increased at end of the first month administrated peppers (P <0.05), than gradually restored at second and third month administrated peppers in both of the low dose peppers group and high dose peppers group, as shown in Figure 2B, whereas the contents of TNF-αβ were not significantly changed at all of the testing points (end of the first month, second month and third month administrated peppers) in both of low dose peppers group and high dose peppers group (P > 0.05).

Effects of pepper on the serum level of the Th2-associated cytokines

As shown in Figure 3A, compared with the control group, the contents of IL-5 in the serum of rabbits were increased at all of the testing points (end of the first month, second month and third month administrated peppers) in the low dose peppers group (P <0.05) and no significant change in the high dose peppers group (P > 0.05). As shown in Figure 3B, the contents of IL-10 in the serum of rabbits were increased at all of the testing points (end of the first month, second month and third month administrated peppers) in the high dose peppers group (P <0.05), no significant change in the low dose peppers group (P > 0.05). There are no significant change in the contents of IL-4 and IL-6 in the serum at all of the testing points (end of the first month, second month and third month administrated peppers) in both of the low dose peppers group and high dose peppers group (P > 0.05).

Effects of pepper on SOD activities and concentrations of MDA in the serum

Compared with the control group, the SOD activities were slightly decreased at the end of first month and significantly decreased at the end of the third month after administrated peppers in both of the low dose peppers group and high dose peppers group (P <0.05) and more significant in the low dose peppers group (P <0.01); whereas the concentrations of MDA in the serum were slightly increased at the end of first month and significantly increased at the end of the third month after administrated peppers in both of the low dose peppers group and high dose peppers group (P <0.05) and more significant changed in the low dose peppers group (P <0.01), as shown in Figure 4A and Figure 4B.

Effects of pepper on the serum level of the allergic asthma related chemokine IL-16

Compared with the control group, the contents of IL-16 in the serum of rabbits were increased at all of the testing points (end of the first month, second month and third month administrated peppers) in both of the low dose peppers group and high dose peppers group (P <0.05) and more significantly in the both of low dose peppers group and high dose peppers group (P <0.05) and more significant changed in the low dose peppers group (P <0.01), as shown in Figure 5A.

Effects of pepper on concentrations of BA in the serum

Compared with the control group, the concentrations of BA in the serum of rabbits were significantly increased at all of the testing points (end of the first month, second month and third month administrated peppers) in the both of low dose peppers group and high dose peppers group (P <0.05) and more significant in the low dose peppers group; (P <0.01), as shown in Figure 5B.
Effects of pepper on immune cells in the serum

As shown in Table 1 and Table 2, compared with the control group, in the serum of rabbits, the white blood cells (WBC), especially the eosinophile granulocyte (EO) were very significantly increased with beyond normal range at all of the testing points (end of the first month, second month and third month administrated peppers) in the both of low dose peppers group and high dose peppers group (P <0.01); the neutrophile granulocytes (NE) were very significantly increased with beyond normal range at the end of the first month (P <0.01), than shows a trends to gradually restore at the end of second and third month administrated peppers in the both of low dose peppers group and high dose peppers group; whereas the lymphocytes (LY) were more significantly increased with beyond normal range at the end of the third month (P <0.01) than at the end of the first month (P <0.05) in the both of low dose peppers group and high dose peppers group; enhancement of monocytes (MO) in the both of low dose peppers group and high dose peppers group has similar trends to LY. Those effects are more significant in the low dose peppers group. There are no significant change in basophile granulocyte (BA) at all of the testing points (end of the first month, second month and third month administrated peppers) in both of the low dose peppers group and high dose peppers group (P >0.05).

Discussion

Causes of allergic constitution are complex and diverse, both genetic factors and acquired factors affect the formation of allergic constitution. Whether in innate and acquired factors, the individual immune dysfunction plays a key role in the formation of allergic constitution [13]. The main characteristics of the immune function disorder of allergic constitution are high level serum immunoglobulin, abnormal in the proportion of Helper T cells 1(Th1) and Helper T cells 2(Th2 ) class two cells and often Th2 cell dominant, high level serum histamines, leukotriene (LT) and interleukin 16 (IL-16) , an allergic asthma related chemokine, etc.. Latest research shows that excessive accumulation of free radicals in the body is closely related to formation of allergic constitution and allergic attack [14,15]. Our data indicate that often taking peppers could change body constitution to allergi constitution through induce individual immune dysfunction such as enhancement of IgM, IL-2, IL-5 , IL-16 , LTD4 and IFN-γ, abnormal in the proportion of Th1 and Th2 class two cells, and increased free radicals, increase of BA and immune cells, especially EO and LY in the serum. These effects of peppers are more predominant in low dose of peppers which the dose often consumed by most people.
Excessive free radical generation by often taking peppers

Our data shows that the activities of SOD were decreased, whereas the concentrations of MDA were increased by administration of peppers. This effect was more obvious in low doses of pepper and also more obvious for long term taking peppers, it may be because of the free radicals are gradually accumulated by peppers. Free radicals that damage to human immune system are the basis for the formation of allergic constitution, and it also directly oxidized human mast cells and basophils, lead to cell membrane

Enhancement of BA in the serum by taking peppers

In the present study, the concentrations of BA were significantly increased by administrated with both low dose and high dose of peppers. This effect was more obvious in low doses of peppers. Elevated BA is common in chronic liver disease, a manifestation of liver cell injury. Excessive BA has multiple toxic effects including cytotoxicity, acute and chronic toxicity, liver injury and systemic multiple organ damage, eventually lead to cirrhosis, liver failure and death [22]. In clinical, excessive BA in the serum appear itching and allergic asthma like symptoms, especially in pregnant women. In addition, in the Traditional Indian Medicine (Ayurvedic medicine), Traditional Mongolian Medicine and Traditional Tibetan medicine, Individuals with “Biliary” type constitution more prone to suffering from allergic diseases and autoimmune diseases. Bile is the physical basis for “Biliary” type constitution, main component of bile is BA, so BA may be considered as the molecular basis for “Biliary” type constitution. Moreover, peppers as drug is used to treat “cold” syndrome due to the increased bile [23]. Therefore, in the present study, the results that taking peppers increased BA in the serum are consistent with the traditional medical theory and theory of physical fitness. More interesting is that BA could be the molecular basis for “Biliary” type constitution as well as mechanism underlying the actions of often taking peppers lead to change body constitution to allergi constitution. This needs further research. In the present study, our results indicate that often taking low dose peppers to make the body function in an over sensitive state such as allergic state whereas often taking large dose peppers to make the body function in a loss sensitive state such as morphine-like anesthesia and production of excessive BA to damage systemic multiple organ, because of large dose pepper produced more endorphins, an endogenous morphine-like substance, through capsacin receptor (vanilloid receptor subtype 1, VR1) elicit morphine-like actions including Pleasure and anesthesia. The production of morphine like substance is the main reason for the addiction of peppers, so more and more people are eating pepper.

Enhancement of immune cells in the serum by taking peppers

Our data shows that WBC and EO were very significantly increased with beyond normal range at all of the testing points (end of the first month, second month and third month administrated peppers) in the both of low dose peppers group and high dose peppers group, indicating that taking peppers

Citation: Zhou H, Gao J, Zhu X, Zhang Z, Bai C (2017) Often Taking Peppers Change Body Constitution to Allergic Constitution. Ann Bone Marrow Res 2(1): 001-007.
could induce inflammatory reaction and allergic reaction, because of elevated EO is common in allergic diseases such as allergic asthma, urticarial, and allergic dermatitis, etc.[25,26]. Enhancement of NE is dominant at end of third month administrated peppers; whereas enhancement of LY is dominant after administrated low dose peppers for 3 months. C: control group; LDP: low dose peppers group; WBC: white blood cell; NE: neutrophile granulocytes; LY: lymphocytes; MO: monocytes; EO: eosinophile granulocyte; BA: basophile granulocyte, data are expressed as means ± SE. n = 6 in each group. *P <0.05 and ** P <0.01 vs control group.

**Effects of high dose peppers on immune cells in the serum of rabbits**. Represents change in the counts of immune cells in before and after administrated high dose peppers for 3 months. C: control group; HDP: high dose peppers group; WBC: white blood cell; NE: neutrophile granulocytes; LY: lymphocytes; MO: monocytes; EO: eosinophile granulocyte; BA: basophile granulocyte, data are expressed as means ± SE. n = 6 in each group. *P <0.05 and ** P <0.01 vs control group.

### Table 1: Effects of LDP on immune cells in serum of rabbits

| Cell types | C     | LDP   | C     | LDP   |
|------------|-------|-------|-------|-------|
| WBC(10^9/L) | 5.32±0.99 | 5.58±0.69 | 5.46±0.74 | 17.08±1.89** |
| NE(10^9/L) | 2.58±0.93 | 2.63±0.72 | 2.61±0.68 | 7.51±1.08** |
| LY(10^9/L) | 1.73±0.65 | 1.92±0.62 | 1.86±0.70 | 3.03±0.82* |
| MO(10^9/L) | 0.28±0.04 | 0.30±0.07 | 0.26±0.06 | 0.8±0.13* |
| EO(10^9/L) | 0.85±0.08 | 0.89±0.11 | 0.87±0.07 | 3.20±0.16** |
| WA(10^9/L) | 0     | 0     | 0     | 0     |

### Table 2: Effects of HDP on immune cells in serum of rabbits

| Cell types | C     | HDP   | C     | HDP   |
|------------|-------|-------|-------|-------|
| WBC(10^9/L) | 5.32±0.99 | 5.47±0.69 | 5.46±0.74 | 14.18±1.19## |
| NE(10^9/L) | 2.58±0.93 | 2.51±0.58 | 2.61±0.68 | 5.98±1.13## |
| LY(10^9/L) | 1.73±0.65 | 1.83±0.55 | 1.86±0.70 | 3.11±0.96# |
| MO(10^9/L) | 0.28±0.04 | 0.32±0.05 | 0.26±0.06 | 0.53±0.08# |
| EO(10^9/L) | 0.85±0.08 | 0.84±0.09 | 0.87±0.07 | 2.53±0.17# |
| WA(10^9/L) | 0     | 0     | 0.01  | 0.01  |

Acknowledgements

We thank Drs Leonidas Tsiokas for comments on the paper. This study was supported by a grant from National Natural Science Foundation of China (approval number: 81541171) and a Natural Science Foundation of Inner Mongolia Autonomous Regions in China (approval number: 2014MS0844) to Changxi Bai.

References

1. Pepper J (2005) Plant drug database. Foreign medicine and plant medicine, 20: 41.
2. Filomena C, Giancarlo AS, Francesco M (2007) Chemical and biological variability of hot pepper fruits (Capsicum annuum var. acuminatum L.) in relation to maturity stage [J]. Food Chem 102: 1096-1104. [Link: https://goo.gl/EJ4Gfo]
3. Yuqun L (2002) Ayurveda in India traditional medicine, Liaoning Education Publishing House 378. [Link: https://goo.gl/W1uxX1]
4. Yixibalazuri, Hohhot M (2007) four parts of nectar: Inner Mongolia Peoples Publishing House.
5. Gansheng Z, Peking M (2012) Chinese materia medica: China Press of Traditional Chinese Medicine.
6. Jing Z, Sha J, Dong X, Huiping L (2010) Research Progress on pharmacological action of pepper [J].China Pharmacy 21: 663-665.
7. Wang Q, qingfeng L (2004) the concept, formation and regulation of allergic disease - risk factors and predictors [J]. J Pediatr 121:58-63. [Link: https://goo.gl/0as8Yw]
8. Bousquet J, Khaltaev N, Cruz AA, Denburg J, Fokkens WJ et al (2008) Allergic Rhinitis and its Impact on Asthma (ARIA) 2008 update (in collaboration with the World Health Organization, GA (2)LEN and Aller Gen). Allergy 32:8-160. [Link: https://goo.gl/ucpKnw]
9. Croner S (1992) Prediction and detection of allergy development: influence of genetic and environmental factors [J]. J Pediatr 121:58-63. [Link: https://goo.gl/Oas8Yw]
10. Halken S (2003) Early sensitisation and development of allergic air way disease - risk factors and predictors [J]. Paediatr Respir Rev 4:128-134. [Link: https://goo.gl/J5EVTH]
11. Romagnani S (2000) T-cell subsets (Th1 versus Th2) - Annals of Allergy, Asthma & Immunology. [J]. Annals of Allergy Asthma & Immunology, 85:9-18. [Link: https://goo.gl/B7F02z]
12. Yong L, Ling QK, GAO H, Yu IY (2008) Research progress of free
radical and disease [J]. Progress in animal medicine, 29:85-88. Link: https://goo.gl/S84BHt

13. Ballesta F (1998) Genetics and allergy. Allergol Immunopathol (Madr) 26: 83-86. Link: https://goo.gl/S9dCkH

14. Zhixia Yang, Ranran Sun, Yang Tian, Yan Zhang (2004) The correlation study between Traditional Chinese Medicine Constitution and Immunology [J]. Immunology Studies, 2:23-32.

15. Singh VK, Mehrotra S, Agarwal SS (1999) the paradigm of Th1 and Th2 cytokines: its relevance to autoimmunity and allergy [J]. Immunologic Research, 20:147-161. Link: https://goo.gl/1hQndN

16. Lu Dan, Yanbin Wan, Xiaojin Yang, Xiaofeng Li. Study on immunological indexes and complement levels in patients with primary biliary cirrhosis and autoimmune diseases [J]. Laboratory medicine and clinical, 2016, 13(13):1829-1831.

17. Ladi Zhang, Jianan Huang, Gehua Yu, Xueguang Zhang. Measurement and clinical significance of interleukin 2 in peripheral blood of patients with allergic asthma [J]. Clinical meta-analysis, 2003, 18(8):421-423.

18. Han R, Lin AH, Zhu KJ, Cheng H, Wu SD (2009) Detection and significance of serum th1/th2 cytokines and chemokines in patients with allergic disease [J]. Journal of Zhejiang University (Medical Science Edition) 38:352-356. Link: https://goo.gl/CzCN9e

19. Lilan Tan, Zeming Du, Yi Bai. Study on bronchial asthma of blood leukotriene E4 and blood interleukin-4 levels [J]. Jiangxi Med J, 2010, 45(11):1081-1082.

20. Laberge S, Pinsonneault S, Varga EM, Till SJ, Aria KN et al. (2000) Increased expression of IL-16 immunoreactivity in bronchial mucosa after segmental allergen challenge in patients with asthma [J]. Journal of Allergy & Clinical Immunology 106:293-301. Link: https://goo.gl/oCoOgm

21. Mazo VK, Shirina LI (2000) Free radical oxidation and food antioxidants in allergic diseases [J]. Vopr Pitan 69:12-17. Link: https://goo.gl/h189FQ

22. Mäjer F, Sharma R, Mullins C, Keogh L, Phipps S et al. (2014) New highly toxic bile acids derived from deoxycholic acid, chenodeoxycholic acid and lithocholic acid. [J]. Bioorg Med Chem 22:256-268. Link: https://goo.gl/2yGPn8

23. Ba Jigemude (1997). Basic theory of Mongolian Medicine [M]. Hohhot: Inner Mongolia Peoples Publishing House.

24. Chagun Arisileng, Bayin Dalai, Naren Gerile, Changxi Bai. Brief introduction to clinical research of pepper [J]. World Journal of Biotechnology, 2015(8):148-149

25. Valen P,Klion AD, Horny HP, Roufosse F,Gotlib J et al. (2012) Contemporary consensus proposal on criteria and classification of eosinophilic disorders and related syndromes[M]. J Allergy Clin Immunol 130:607-612. Link: https://goo.gl/vNpi2n

26. Simon HU,Rothenberg MB,Bochner BS,Weller PF,Wardlaw AJ et al. (2010) Refining the definition of hypereosinophilic syndrome.[M]. J Allergy Clin Immunol 126:45-49. Link: https://goo.gl/gvOzGm