ChinaRxiv 中X

AI translation · View original & related papers at
chinarxiv.org/items/chinaxiv-201812.00412

Effects of Yeast Extract Powder on Triglyceride Content and Uric Acid Metabolism in Silkworm Hemolymph (Postprint)

Authors: Li Na, Jin Huijie, Jia Manli, Huang Lu, Xie Yan, Xia Aihua, Yang Guiming, Li Jisheng

Date: 2018-12-24T00:00:00+00:00

Abstract

This experiment aimed to investigate the effects of yeast extract powder on triglyceride content and uric acid metabolism in the hemolymph of silkworms. Using the silkworm variety 'Jingsong × Haoyue' as experimental material, 600 silkworms of uniform size and consistent development were selected and randomly divided into 4 groups (3 replicates per group, 50 silkworms per replicate, with equal numbers of males and females). Starting from the 5th instar, three experimental groups (JM1, JM2, and JM3) were fed fresh mulberry leaves coated with 1%, 2%, and 4% yeast extract powder solution, respectively, while the control group was fed leaves coated with distilled water instead of yeast extract powder solution. The uric acid content, triglyceride content, and xanthine oxidase (XOD) activity in silkworm hemolymph were measured after 3, 4, and 5 days of feeding with yeast extract powder. The results showed that after 5 days of feeding with different concentrations of yeast extract powder, the weight gain rate of silkworms was significantly higher than that of the control group ($P<0.05$), but there was no significant difference among the experimental groups ($P>0.05$). Moreover, the weight gain rate showed a decreasing trend with increasing yeast extract powder concentration. After 5 days of feeding with different concentrations of yeast extract powder, the triglyceride content in silkworm hemolymph increased by 42%–98% compared with the control group, with the differences between JM1 and JM2 groups and the control group reaching significant levels ($P<0.05$). With the extension of feeding time, the uric acid content in silkworm hemolymph of all groups showed a decreasing trend, with significant differences between day 5 and day 3 ($P<0.05$). As the concentration of yeast extract powder increased, the uric acid content in silkworm hemolymph at each time point showed an increasing trend, but the differences between the experimental groups and the control group were not significant ($P>0.05$). The XOD activity in silkworm hemolymph of the three experimental groups was

significantly higher than that of the control group at all time points (P<0.05), but on days 3 and 4, the XOD activity in silkworm hemolymph showed a trend of first increasing and then decreasing with increasing yeast extract powder concentration. There was a significant or extremely significant positive correlation between uric acid content and XOD activity in silkworm hemolymph (P<0.05 or P<0.01), with correlation coefficients ranging from 0.652 to 0.902. It can be concluded that feeding with a certain concentration of yeast extract powder can lead to increased uric acid content, triglyceride content, and XOD activity in silkworm hemolymph, thereby affecting uric acid metabolism in silkworms.

Full Text

Effects of Yeast Extract Powder on Triglyceride Content and Uric Acid Metabolism in Silkworm Hemolymph

Li Na[1,2], Jin Huijie[3], Jia Manli[1,2], Huang Lu[1,2], Xie Yan[1,2], Xia Aihua[1,2], Yang Guiming[1,2], Li Jisheng[1,2*]

[1]Hebei Universities R&D Center for Sericulture and Specialty Enabling Technologies, Chengde 067000, China
[2]Institute of Sericulture, Chengde Medical University, Chengde 067000, China
[3]Zhengding Agro-Tech Extension Station of Hebei, Shijiazhuang 050800, China

Abstract: This experiment investigated the effects of yeast extract powder (YEP) on triglyceride content and uric acid (UA) metabolism in silkworm hemolymph. The silkworm variety "Jingsong × Haoyue" was used as experimental material. Six hundred silkworms of uniform size and developmental stage were randomly divided into four groups (three replicates per group, 50 silkworms per replicate, half male and half female). Starting from the fifth instar, three experimental groups (JM1, JM2, and JM3) were fed fresh mulberry leaves coated with 1%, 2%, and 4% YEP solution, respectively, while the control group received leaves coated with distilled water. After 3, 4, and 5 days of YEP supplementation, uric acid content, triglyceride content, and xanthine oxidase (XOD) activity in silkworm hemolymph were measured. The results showed that body weight growth rate increased significantly in all YEP-treated groups after 5 days compared with the control (P<0.05), though no significant differences were observed among the experimental groups (P>0.05). Notably, body weight growth rate tended to decrease as YEP concentration increased. Triglyceride content in hemolymph after 5 days of YEP supplementation was 1.42 to 1.98 times higher than the control, with JM1 and JM2 groups showing significant differences (P<0.05). As supplementation time extended, uric acid content in hemolymph decreased across all groups, with significant differences between day 5 and day 3 (P<0.05). Although uric acid content tended to increase with higher YEP concentrations at each time point, differences between experimental and control groups were not statistically significant (P>0.05). XOD activity in hemolymph was significantly higher in all three experimental groups than in the control at each time point (P<0.05), but showed a trend of initial increase followed by decrease with rising YEP concentration on days 3 and 4. A significant or extremely significant positive correlation existed between uric acid content and XOD activity in silkworm hemolymph ($P<0.05$ or $P<0.01$), with correlation coefficients ranging from 0.652 to 0.902. These findings demonstrate that supplementation with appropriate concentrations of YEP can elevate uric acid content, triglyceride content, and XOD activity in silkworm hemolymph, thereby affecting uric acid metabolism.

Keywords: silkworms; yeast extract powder; triglyceride; uric acid metabolism

---

Uric acid represents the end product of purine metabolism in humans. Approximately 80% of uric acid originates from endogenous sources through the catabolism of nucleoproteins, while the remaining 20% derives from exogenous dietary purines. Excretion occurs primarily through the intestines (one-third) and kidneys (two-thirds). Hyperuricemia is diagnosed when blood uric acid levels exceed 488 mol/L in males or 387 mol/L in females, resulting from abnormal production or excretion. High-protein diets can exacerbate purine metabolic pressure and induce metabolic disorders. Numerous medical studies have demonstrated that excessive consumption of fats and proteins significantly increases the risk of hyperuricemia and gout, which is caused by urate crystal deposition in joints and soft tissues. Additionally, uric acid promotes platelet aggregation and thrombosis formation. Consequently, gout has emerged as a severe metabolic disease comparable to diabetes in its impact on human health.

Yeast extract powder (YEP) is rich in proteins, nucleotides, and B vitamins, which upon complete hydrolysis produce nitrogenous organic bases (including purine and pyrimidine bases) and phosphates. High doses of yeast can disrupt normal purine metabolism, manifested by elevated XOD activity and accelerated uric acid production. In medical research, mice and rats are commonly used to establish hyperuricemia models through dietary supplementation with yeast paste or YEP, which effectively induces the condition.

The silkworm (*Bombyx mori*) serves as the second most important model organism among Lepidoptera and represents a valuable resource for studying animal physiology, biochemistry, genetics, and genomics. With abundant bioinformatics resources and a well-established research platform, silkworms exhibit numerous physiological characteristics similar to humans, providing a solid foundation for their use as an excellent model animal for studying human physiology, diseases, genetics, and novel drug development. Regarding uric acid metabolism, Tamura et al. found that injecting xanthine dehydrogenase (XDH) increased uric acid content in the excrement of translucent silkworm mutants, gradually changing their integument from translucent to opaque white. This visible color change in the silkworm integument offers a unique advantage for studying uric acid metabolism using this model. Zhang Xiaoli and Wang Changchun demonstrated that oral administration of gout therapeutic agents allopurinol or febuxostat effectively reduced XOD activity and uric acid content in silkworm hemolymph and fat body, with effects consistent with clinical outcomes. Both studies concluded that silkworms could serve as an animal model alternative to mice for screening and evaluating anti-gout drug efficacy. However, these investigations focused primarily on existing drug effects, while the impact of YEP on silkworm uric acid metabolism and the methodology for inducing hyperuricemia in silkworms remain underexplored.

Blood uric acid and triglyceride levels are routine biochemical parameters in clinical testing. Abnormal triglyceride metabolism represents a key manifestation of metabolic syndrome, which frequently co-occurs with hyperuricemia. These conditions demonstrate significant correlation (correlation coefficient of 0.64). Therefore, this study evaluated the effects of YEP on silkworm uric acid metabolism by supplementing mulberry leaves with different YEP concentrations and measuring body weight growth rate, hemolymph uric acid content, XOD activity, and triglyceride content, aiming to provide scientific basis for establishing a silkworm hyperuricemia model.

1.1 Experimental Silkworms and Reagents

The experimental silkworm variety "Jingsong × Haoyue" was selected and maintained by the Silkworm Research Laboratory of the Institute of Sericulture, Chengde Medical University. Uric acid assay kits, XOD assay kits, and triglyceride assay kits were purchased from Nanjing Jiancheng Bioengineering Institute. Yeast extract powder was obtained from Tianjin Zhiyuan Chemical Reagent Co., Ltd.

1.2 Experimental Design and Feeding Method

Fresh mulberry leaves were used to rear silkworms according to standard protocols until the fifth instar. Six hundred silkworms of uniform size and developmental stage were randomly divided into four groups: control, JM1, JM2, and JM3. Each group comprised three replicates of 50 silkworms (half male and half female). Based on preliminary experiments indicating silkworm aversion to YEP, appropriate amounts of YEP were dissolved in distilled water to prepare 1% (JM1), 2% (JM2), and 4% (JM3) solutions for leaf coating. The control group received distilled water only. Each group received 20 mL of solution uniformly applied to 60 g of mulberry leaves daily. After consuming the coated leaves, silkworms were provided with unlimited fresh mulberry leaves.

1.3.1 Sample Collection

On days 3, 4, and 5 of YEP supplementation, six silkworms were selected from each replicate (half male and half female). Hemolymph was collected by clipping the caudal horn and gently applying abdominal pressure into ice-cooled centrifuge tubes containing a small amount of phenylthiourea, then stored at -80°C until analysis.

1.3.2 Parameter Determination and Methods

Body weight growth rate was determined using a 0.001 g precision balance to measure total group weight at the start and on day 5 of the experiment, from which average initial and final body weights were calculated. Body weight growth rate was computed as: 100 × (average final weight - average initial weight) / average initial weight. Hemolymph triglyceride content was measured using a BioTek microplate reader at 510 nm according to the kit instructions. Hemolymph uric acid content and XOD activity were determined using a T6 UV-Vis spectrophotometer at 690 nm and 530 nm, respectively, following kit protocols.

1.4 Data Processing

Data were analyzed using SPSS 18.1 software. Inter-group differences were evaluated using Duncan's multiple comparison test, and Pearson correlation analysis was performed. Significance was set at $P < 0.05$ and extreme significance at $P < 0.01$.

2.1 Effects of YEP on Silkworm Body Weight Growth Rate

[Figure 1: see original paper] illustrates changes in body weight growth rate after 5 days of YEP supplementation at different concentrations. The results demonstrate that YEP supplementation affected silkworm growth to varying degrees. Compared with the control group, body weight growth rate increased significantly in groups fed 1%, 2%, and 4% YEP ($P < 0.05$), though no significant differences were observed among the experimental groups ($P > 0.05$). Notably, body weight growth rate tended to decline as YEP concentration increased.

Data columns with different letters indicate significant differences ($P < 0.05$), while the same letters indicate no significant difference ($P > 0.05$). Error bars represent standard error of the mean. This applies to all figures.

2.2 Effects of YEP on Triglyceride Content in Silkworm Hemolymph

[Figure 2: see original paper] shows changes in hemolymph triglyceride content after 5 days of YEP supplementation. Triglyceride content decreased with increasing YEP concentration, yet all experimental groups exhibited higher levels than the control (1.42 to 1.98 times the control value). Specifically, triglyceride content in the 1% and 2% YEP groups was significantly higher than the control ($P < 0.05$), whereas the 4% YEP group showed no significant difference from the control ($P > 0.05$).

2.3 Effects of YEP on Uric Acid Content in Silkworm Hemolymph

[Figure 3: see original paper] presents changes in hemolymph uric acid content after 3–5 days of YEP supplementation. Uric acid content varied among groups receiving different YEP concentrations. As supplementation duration extended, uric acid content decreased significantly in all groups, with day 5 showing significant differences from day 3 ($P < 0.05$). Although uric acid content tended to increase systematically with higher YEP concentrations at each time point, no significant differences were observed between experimental and control groups ($P > 0.05$).

2.4 Effects of YEP on XOD Activity in Silkworm Hemolymph

[Figure 4: see original paper] depicts changes in hemolymph XOD activity after 3-5 days of YEP supplementation. XOD activity varied differently among groups over time, though no significant differences were found between time points within each group ($P > 0.05$). XOD activity increased on day 4 compared with day 3 in all groups, but decreased on day 5 in the JM1 and JM2 groups relative to day 4. Comparisons at each time point revealed that XOD activity in all three experimental groups was significantly higher than the control ($P < 0.05$), while showing a trend of initial increase followed by decrease with rising YEP concentration on days 3 and 4.

2.5 Correlations Among Uric Acid Content, XOD Activity, and Triglyceride Content

As shown in , Pearson correlation analysis revealed extremely significant positive correlations between hemolymph uric acid content and XOD activity on days 3 and 4 ($P < 0.01$). On day 5, uric acid content showed significant positive correlations with both XOD activity and triglyceride content ($P < 0.05$).

Table 1 Pearson correlation analysis between uric acid content and XOD activity and triglyceride content in silkworm hemolymph

Item	Day 3	Day 4	Day 5
Uric acid vs. XOD	r = 0.902**	r = 0.858**	r = 0.652*
Uric acid vs. Triglyceride	-	-	r = 0.509*

"**" indicates extremely significant difference ($P < 0.01$), and "*" indicates significant difference ($P < 0.05$).

3.1 Analysis of Factors Influencing YEP-Induced Changes in Hemolymph Uric Acid Content

The present results demonstrate that YEP supplementation increased hemolymph uric acid content and XOD activity in silkworms, though differences from the control group were less pronounced than anticipated. On one hand, varying YEP concentrations elevated uric acid content and XOD activity to different extents, confirming that YEP can alter uric acid metabolism-related parameters in silkworms. On the other hand, considering the body weight growth rate data, although YEP increased hemolymph uric acid content, higher

YEP concentrations simultaneously caused a decline in body weight growth rate. This may be attributed to the silkworm' s nature as an oligophagous insect with sensitive olfactory and gustatory systems. While YEP exhibits good autolysis properties, its inherent odor at high concentrations may reduce feeding appetite. However, the specific mechanisms, such as whether actual food intake decreased, require further investigation.

3.2 Relationship Between Hemolymph Uric Acid Content and XOD Activity

Hypoxanthine and xanthine represent the most direct precursors of uric acid, undergoing stepwise oxidation by XOD—from hypoxanthine to xanthine, and ultimately to uric acid. This metabolic pathway indicates that XOD activity level directly influences the rate of xanthine conversion to uric acid. The present measurements of uric acid content and XOD activity revealed that YEP supplementation simultaneously elevated both parameters in silkworm hemolymph. Pearson correlation coefficients between uric acid content and XOD activity on days 3, 4, and 5 were 0.902, 0.858, and 0.652, respectively, with all P values below 0.05, confirming a positive correlation between hemolymph uric acid content and XOD activity.

3.3 Relationship Between Hemolymph Uric Acid Content and Triglyceride Content

The relationship between uric acid and triglyceride levels constitutes a major focus in medical research. Human studies have demonstrated that blood uric acid content correlates significantly with triglyceride content, independent of alcohol consumption, obesity, and insulin resistance. Pearson correlation analysis of the present results revealed a significant relationship between hemolymph uric acid content and triglyceride content on day 5, with a correlation coefficient of 0.509 and $P < 0.05$. This finding suggests that silkworms and humans share remarkably similar physiological responses in uric acid metabolism.

In summary, this study demonstrates that supplementation with appropriate YEP concentrations can elevate uric acid content, triglyceride content, and XOD activity in silkworm hemolymph, showing high consistency with physiological responses observed in mouse models. These results indicate that YEP supplementation can induce abnormalities in hemolymph uric acid metabolism-related parameters in silkworms. With increasingly sophisticated physiological, biochemical, and genomic platforms for silkworm research, combined with advantages including abundant natural variation, short life cycle, moderate body size, easy reproduction, convenient organ sampling, and absence of ethical concerns, this study provides scientific basis for further developing silkworms as a model organism for hyperuricemia research.

4 Conclusion

Yeast extract powder supplementation can cause varying degrees of elevation in hemolymph uric acid content, triglyceride content, and XOD activity, thereby affecting uric acid metabolism in silkworms. Comprehensive analysis of the effects of different YEP concentrations on body weight growth rate and hemolymph uric acid content, XOD activity, and triglyceride content suggests that a 1% YEP concentration yields optimal effects.

References

[1] Wei Wenjing, Feng Mali. Research progress on hyperuricemia and non-alcoholic fatty liver disease[J]. *World Latest Medicine Information (Electronic Version)*, 2017, 17(22): 23–24.

[2] Xu Guobin, Zhang Yongqin. Development and standardization of uric acid detection methods[J]. *Chinese Journal of Laboratory Medicine*, 2008, 31(7): 834–836.

[3] Liu Wenhui, Zhang Yuan, Qiu Jian. Research progress on the role of uric acid-related gene polymorphisms in cardiovascular diseases[J]. *Journal of Practical Medicine*, 2017, 33(10): 1710–1712.

[4] Zhang Yanling, Wu Jianyong. Research progress on hereditary renal hypouricemia[J]. *Chinese Journal of Integrated Traditional and Western Nephrology*, 2016, 17(8): 738–740.

[5] Chen Liping, Zhou Haiwei, Zhu Cheng, et al. Comparative analysis of ultrasound imaging and X-ray examination of gout nodules[J]. *Journal of Medical Imaging*, 2010, 20(12): 1842, 1846.

[6] Li Qizhe, Zhou Yukun, Chen Shiping. A case of ankle joint destruction caused by tophi in a 17-year-old patient[J]. *Medical Frontier*, 2017, 7(4): 138–139.

[7] Gu Xiaoye, Zhang Jiong, Zhang Xinju, et al. Relationship between ABCG2 gene rs2231142 and gout/hyperuricemia in Chinese Han population[C]//Proceedings of the 7th National Young and Middle-aged Clinical Laboratory Medicine Academic Conference of Chinese Medical Association. Nanjing: Chinese Medical Association, 2012.

[8] Liu Fang. Research progress on serum uric acid and cardiovascular and cerebrovascular diseases[J]. *Journal of Family Health*, 2017, 11(1): 298.

[9] Chen Guangliang, Xu Shuyun. Research progress on animal models of hyperuricemia[J]. *Chinese Pharmacological Bulletin*, 2004, 20(4): 369–372.

[10] Chen Fengli, Chen Hanyu, Qiu Lianqun. Research progress on the relationship between primary gout, hyperuricemia and constitution[J]. *World Journal of Integrated Traditional and Western Medicine*, 2017, 12(3): 437–439.

[11] Goldsmith M R, Shimada T, Abe H. The genetics and genomics of silkworm, *Bombyx mori*[J]. *Annual Review of Entomology*, 2005, 50: 71-100.

[12] Qin Jian, He Ningjia, Xiang Zhonghuai. Research progress on silkworm modeling[J]. *Acta Sericologica Sinica*, 2010, 36(4): 645-649.

[13] Xia Q Y, Zhou Z Y, Lu C, et al. A draft sequence for the genome of the domesticated silkworm (*Bombyx mori*)[J]. *Science*, 2004, 306(5703): 1937-1940.

[14] Mita K, Kasahara M, Sasaki S, et al. The genome sequence of silkworm, *Bombyx mori*[J]. *DNA Research*, 2004, 11(1): 27-35.

[15] Chen Min, Song Jiangbo, Li Zhiquan, et al. Research progress and prospects of silkworms as human disease models and for drug screening[J]. *Acta Pharmaceutica Sinica*, 2016(5): 690-697.

[16] Ishii K, Hamamoto H, Kamimura M, et al. Activation of the silkworm cytokine by bacterial and fungal cell wall components via reactive oxygen species-triggered mechanism[J]. *Journal of Biological Chemistry*, 2008, 283(4): 2185-2191.

[17] Hamamoto H, Kurokawa K, Kaito C, et al. Quantitative evaluation of the therapeutic effects of antibiotics using silkworms infected with human pathogenic microorganisms[J]. *Antimicrobial Agents and Chemotherapy*, 2004, 48(3): 774-779.

[18] Orihara Y, Hamamoto H, Kasuga H, et al. A silkworm baculovirus model assessing the therapeutic effects of antiviral compounds: characterization and application to the isolation of antivirals from traditional medicines[J]. *Journal of General Virology*, 2008, 89(1): 188-194.

[19] Lu Zhongyan, Wen Yangli, Zhang Zhen, et al. Effects of high-sugar diet on blood glucose concentration and triglyceride content in silkworm fat body[J]. *Acta Sericologica Sinica*, 2014, 40(5): 933-937.

[20] Wei Guangbing. Study on the mechanism of oxidative damage induced by anti-tuberculosis drug isoniazid (INH) in silkworms[D]. Master' s thesis. Suzhou: Soochow University, 2013.

[21] Tamura T, Sakate S. Granules in the meconium of og-mutant of *Bombyx mori*[J]. *The Journal of Sericultural Science of Japan*, 1975, 44(6): 487-490.

[22] Zhang Xiaoli. JAK/STAT pathway response to BmNPV infection and silkworm as an animal model for gout drug screening[D]. Master's thesis. Suzhou: Soochow University, 2011.

[23] Wang Changchun. Preliminary study on establishing a silkworm model for gout drug screening[D]. Master' s thesis. Chongqing: Southwest University, 2012.

[24] Chen Guangliang, Zhang Qinglin, Ma Xiaoqin, et al. Yeast-induced hyperuricemia mouse model[J]. *Chinese Pharmacological Bulletin*, 2003, 19(4): 467-

469.

[25] Dong Jing, Zhu Ping, Cheng Kangpeng, et al. Exploration of mouse hyperuricemia model[J]. *Chinese Journal of Cardiovascular Medicine*, 2009, 14(3): 237-239.

[26] Dong Zhe, Ding Ning, Cui Jie, et al. Ameliorative effects of sea cucumber cerebrosides and ceramides on hyperuricemia in mice[J]. *Chinese Journal of Marine Drugs*, 2013, 32(6): 65-71.

[27] Xu Huijing, Zhang Hao, Liu Chunhua, et al. Effects of sea cucumber saponins on hyperuricemia in mice[J]. *Chinese Pharmacological Bulletin*, 2011, 27(8): 1064-1067.

[28] Zhao Lanjiang, Zhao Dong. Relationship between abnormal uric acid metabolism and abnormal triglyceride metabolism[J]. *Chinese Journal of Epidemiology*, 2006, 27(4): 362-365.

[29] Berkowitz D. Blood lipid and uric acid interrelationships[J]. *The Journal of the American Medical Association*, 1964, 190: 856-858.

[30] He Yunqing. Theoretical study on the structure and vibrational spectra of hypoxanthine and xanthine[J]. *Guangzhou Chemical Industry*, 2017, 45(8): 19-21.

[31] Zhao Lanjiang, Zhao Dong, Liu Jing, et al. Population study on the relationship between serum uric acid level and triglyceride[J]. *Chinese Journal of Internal Medicine*, 2005, 44(9): 664-667.

Note: Figure translations are in progress. See original paper for figures.

Source: ChinaXiv —Machine translation. Verify with original.