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Original Article

A Survey on the Effects of Gold-Nanoparticles in Allergic Asthma

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Abstract

Background and Aim: Asthma is an inflammatory airway disease and allergies are the most important cause of asthma. Different types of drugs have been developed to control asthma, and the use of carrier systems to transfer drugs to the airways is an effective method. Gold nanoparticles (AuNP) is a subject of substantial research that can be easily synthesized and, in this study, the effect of gold nanoparticles on allergic asthma was evaluated.
Methods: There are 4 groups of mice, including: the control group, the control group receiving AuNPs, the asthmatic group, and the asthmatic group receiving AuNPs. An animal model of asthma was produced using ovalbumin (OVA). The negative control group was sensitized and challenged with PBS. Broncho-alveolar lavage fluid (BALF) and lung tissue were collected, then quantitative real-time PCR for the four target genes (IL-4, IL-5, IL-13, and MUC5ac) and histopathological study of lung tissue was done.
Results: In the OVA group, the mRNA expression of targeted genes had no significant differences ($P>0.05$). Mucus hypersecretion, goblet cell hyperplasia, peribronchial and perivascular inflammation had no significant difference between AuNPs receiving groups with non-treated groups ($P>0.05$).
Conclusion: In this study, it was observed that AuNP did not affect asthma and control mice. These nanoparticles did not elicit any immune or allergic responses and can be easily used for therapeutic or diagnostic purposes.

Keywords: Gold Nanoparticles; Nanoparticles; Asthma; Allergic Asthma; Biomedicine, Drug Delivery Systems; Carrier.

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Introduction

Asthma is an inflammatory respiratory tract disease of the airways with a prevalence of 7 to 35% and allergies are the most important cause of asthma. Inflammation and asthma-induced spasm in the bronchi, obstruct the airways in the lungs. Different types of drugs have been developed to treat and control asthma, and today it is trying to use nanomaterials for the therapeutic approach of asthma to have the higher possible effect on the target tissue with the lower dosage of the drug. Therefore, the use of carrier systems to transfer the desired material to the airways is an effective and practical method. The most important of these materials are designed for drug delivery nanoparticles (1, 2).

AuNPs are a subject of substantial research because of their potential applications in a wide area variety including electronics, nanotechnology, and biomedicine. It can be easily synthesized using various techniques and also, can be simply attached with several molecules, such as nucleic acid, protein, carbohydrate, and lipid (3).

The difference in the size of AuNP leads to different optical properties that can increase the wide use in modern biomedical applications (1, 4). Functionalized AuNPs have recently emerged as an attractive delivery carrier into targeted tissues. AuNPs can be conjugated to some specific molecules such as antibodies and aptamers for dual-use, as diagnostic and therapeutic functionalized nanoparticles and prospectively can be used alone. Therefore, after the synthesis of considered AuNP,

the effect of this nanoparticles on allergic asthma was evaluated.

Methods

Animals and experiment

Female BALB/c mice were purchased at 6 weeks of age (from Pasteur Institute of Iran) and kept under standard laboratory conditions (temperature 24±2°C; humidity 60±5%; and 12 h light-dark cycles). In the tests, 4 groups of mice of the same gender, same breed, and the same age were considered (each group consisted of 10 mice, that 5 mice were used to obtain lung tissue and the other 5 mice in each group were used to a sampling of BALF). Prior to the experiments, the mice were kept for two weeks to adapt to the new environment**.**

Animal treatment protocol

The 4 groups including: control group, the control group receiving AuNPs (the average diameter: 100-200 nm, Zeta potent Z-average: 137.9 d.nm) on day 23, 25, 27, and 29 (inhalation for 30 min per day), asthmatic group, the asthmatic group receiving AuNPs on day 23, 25, 27 and 29 (inhalation for 30 min per day). An animal model of asthma was produced using OVA according to Athari et al., 2016 (1). In brief, the mice were sensitized by 20 µg of OVA with 50 µL Alum adjuvant via intraperitoneal injection (IP) on days 1 and 14. Then challenged by 1% OVA solution aerosolized by an ultrasonic nebulizer via inhalation (IT) for 30 min per day on days 24, 26, 28, and 30 (Figure 1). The negative control group was sensitized and challenged with PBS. BALF and lung tissue were collected 1 day after the last challenge (day 31).

In all procedures we followed ethical guidelines. Animals were killed humanely using CO_2 gas and then sampling was done.

Quantitative Real-Time PCR

Total RNA was isolated from BALF cells according to the manufacturer's instructions (1). Extracted RNA was reverse transcribed to cDNA using a cDNA synthesis kit (Maxima First Strand cDNA Synthesis Kit, Thermo Scientific, USA). Quantitative PCR analysis was performed. Primers for the four target genes (IL-4, IL-5, IL-13, and MUC5ac) and one primer-pair for GAPDH are shown in Table 1.

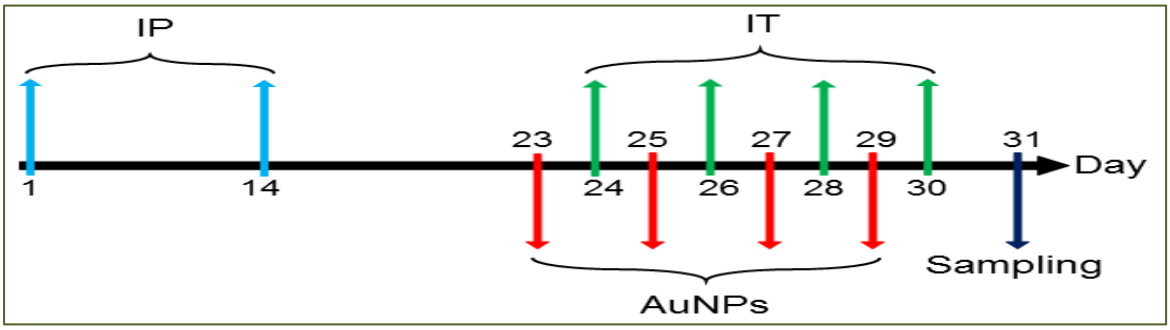

Figure 1. Sensitization, challenge, and treatment program in the studied groups. IP of OVA with alum adjuvant was performed on days 1 and 14 and also, OVA via IT was administered on days 24, 26, 28, and 30. Cases were treated on days 23, 25, 27, and 29**.**

Table 1. Used primers sequences

Gene	Primer	Sequence (5′ to 3′)
GAPDH	Sense	TGTTCCTACCCCCAATGTGT
	Antisense	GGTCCTCAGTGTAGCCCAAG
IL-4	Sense	AGATCATCGGCATTTTGAACG
	Antisense	TTTGGCACATCCATCTCCG
IL-5	Sense	ACATTGACCGCCAAAAAGAG
	Antisense	ATCCAGGAACTGCCTCGTC
IL-13	Sense	GGTCCACACAGGGCAACT
	Antisense	AATAAGATCAAGAAGAAATGTGCTCAA
Muc5a	Sense	CAGGACTCTCTGAAATCGTACCA
	Antisense	AAGGCTCGTACCACAGGGA

Histopathological study

Mice lungs were isolated and tissue sections were stained with hematoxylin and eosin (H&E) stain. The ratio of mucus production, the goblet cell hyperplasia, peribronchial and perivascular inflammation was studied (1).

Statistical analysis

Experimental results are shown as means ± S.D. Results were compared in all pairs of groups or two-tailed, student's t-test. Results were considered statistically significant when $p < 0.05$.

Results

Effects of gold nanoparticles on cytokines and mucin mRNA

In the AuNPs receiving OVA group, the mRNA expression of IL-4 (7.01±1.44) and IL-5 (2.69±0.19) was decreased compared non-treated OVA group (IL-4: 8.11±1.37 and IL-5: 3.12± 0.22), but these decreasing were not significant ($p >0.05$). there is no significant difference between the control group and nanoparticles receiving control group for gene expression of IL-4, IL-5, IL-13, and mucin and also, between the OVA group and nanoparticles receiving OVA group for mucin gene expression ($p >0.05$) (Figure 2). IL-13 gene expression increased significantly in nanoparticles receiving OVA group (2.97±0.22) compared to OVA group ($p <0.05$) (2.01±0.13).

Effects of gold nanoparticles on lung histopathology

Mucus hypersecretion, goblet cell hyperplasia, and peribronchial inflammation had no significant difference between AuNPs receiving control group and control group, and also between, nanoparticles receiving OVA group and OVA group ($p>0.05$) (Figure 3). Perivascular inflammation was increased in the airway of OVA-challenged mice that received AuNPs (3.70±0.20), compared with the OVA group (3.40±0.10) and also, inflammation in the perivascular was increased in control mice that received gold nanoparticles (0.60±0.40), compared with the non-treated control group (0.50±0.10) (Fig. 3), but these increases were not statistically significant ($p >0.05$).

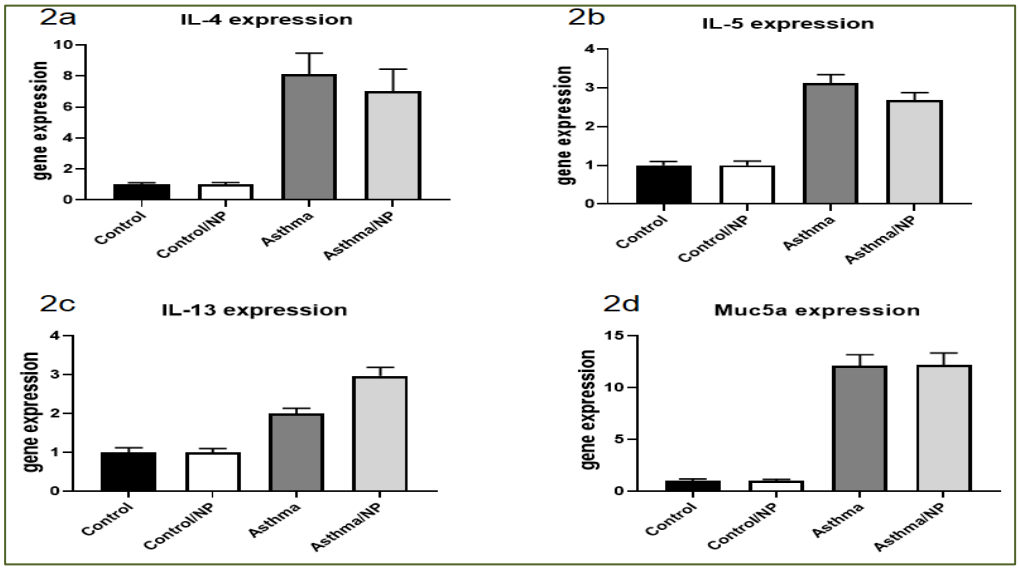

Figure 2. The gene expression of cytokines, IL-4 (2a), IL-5 (2b), IL-13 (2c), and mucin [MUC5ac (2d)] were shown.

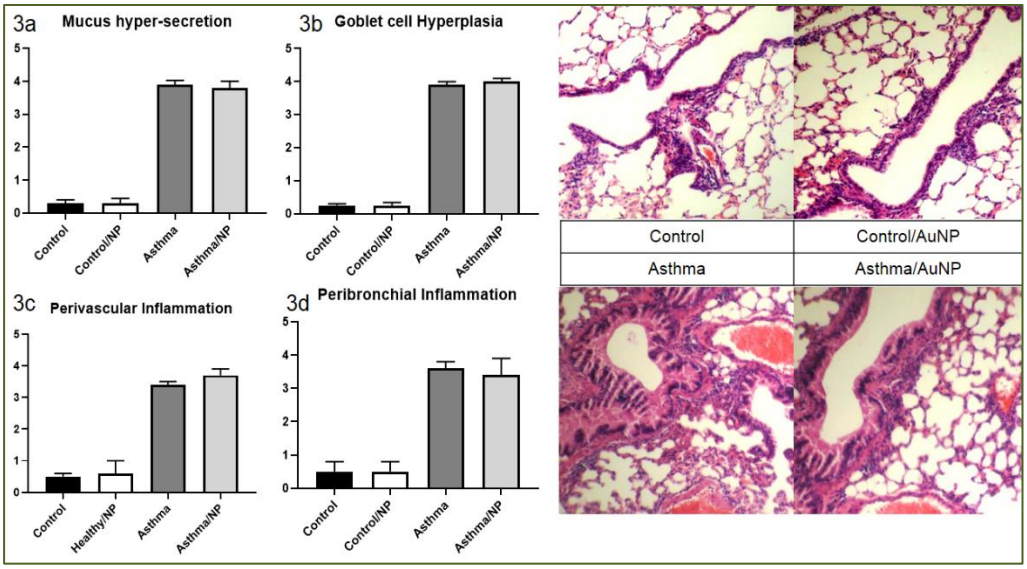

Figure 3. Lung histopathological sections in all groups were shown and study of mucus hyper-secretion (3a), goblet cell hyperplasia (3b), perivascular inflammation (3c), and peribronchial inflammation (3d) were presented.

Discussion

In this study, it was observed that AuNPs with described characteristics do not affect allergic asthma and control mice. It would be a beneficial factor for any studies that AuNPs will use as a drug delivery system for control and treatment of asthma without any changes in results of targeted drug effect.

However, small-sized AuNPs (under 50 nm) have shown considerable antimicrobial activity. The antibacterial activity of the AuNPs can be attributed to the reactive oxygen species (ROS) generation that intensifies the oxidative stress of microbial cells (5, 6). But in asthma, ROS is a dangerous factor in the pathophysiology of asthma (7) and increased levels of ROS would lead to an increased asthma attack. Therefore, these produced AuNPs are not suitable to use as drug delivery or carrier for control and treatment of asthma. Because this component can increase asthma severity and reduce the effect of used drugs or treatments.

On the other hand, AuNPs are used as signal amplifiers to detect some diagnostic factors and hazardous toxins in immunosensor system of flow-type. It can be done by a collaboration of antibodies or other specific molecules such as modified mannose, functionalized aptamer, etc. Also, antibody-conjugated nanoparticles can be used for tissue and cellular imaging using a confocal reflectance microscope. Immune-labeled nanoparticles are used for the monitoring of diseases (8, 9). AuNPs have several advantages for cellular imaging. They scatter light intensity and brightness is much than chemical fluorophores. They do not have to photobleach and can be easily detected in as low as 10^{-16} M concentration. Due to AuNPs can resonantly scatter visible light and also NIR radiations upon excitation of their surface plasmon oscillation. This property can be used for intracellular trafficking of AuNPs as contrast agents in the dark field of optical microscopy (8-10).

The most important criterion for gene delivery system is escaping from the reticuloendothelial clearance system, and not elicits an immune response against the gene (11). AuNPs do not elicit any immune or allergic responses, and can be easily used for therapeutic (carrier of drugs) or diagnostic (nanoprobe) purposes.

Conclusion

In this study, we observed that AuNPs in 100-200 nm size are suitable particles as drug delivery systems to the airway and when it will be necessary to use drug carriers, it may be an effective carrier for the anti-asthma drugs. This can be used in the design and production of new anti-asthma drugs.

Conflict of Interest

The authors declared that they have no conflict of interest.

Acknowledgment

Not declared.

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Ethics

All procedures performed in this study were in accordance with the ethical standards of the institutional and/or national research committee.

This study aapproved in Ethics Committee of Zanjan University of Medical Sciences (IR-ZUMS-REC-1400.156).

References

1. Seyyed Shamsadin Athari, Zahra Pourpak, Gert Folkerts, Johan Garssend, Mostafa Moin, Ian M. Adcock, Masoud Movassaghi, Mehdi Shafiee Ardestani, Seyed Mohammad Moazzeni, Esmaeil Mortaz. Conjugated Alpha-Alumina nanoparticle with vasoactive intestinal peptide as a Nano-drug in treatment of allergic asthma in mice. European Journal of Pharmacology 2016; 791:811–20.

2. SS Athari, SM Athari. The importance of eosinophil, platelet and dendritic cell in asthma. Asian Pacific Journal of Tropical Disease 2014; 4:41-7.

3. D Pissuwan, CH Cortie, SM Valenzuela & MB Cortie. Gold nanosphere-antibody conjugates for hyperthermal therapeutic applications. Gold Bulletin 2007; 40:121-9.

4. Wansen Li, Hong Wan, Shuxun Yan, Zhao Yan, Yalin Chen, Panpan Guo, Thiyagarajan Ramesh, Ying Cui, Lei Ning. Gold nanoparticles synthesized with Poria cocos modulates the anti-obesity parameters in high-fat diet and streptozotocin induced obese diabetes rat model. Arabian Journal of Chemistry 2020; 13(7):5966-77.

5. C. Tao. Antimicrobial activity and toxicity of gold nanoparticles: research progress, challenges and prospects. Letters in Applied Microbiology 2018; 67(6):537-43.

6. Shahzadi Shamaila, Noshin Zafar, Saira Riaz, Rehana Sharif, Jawad Nazir, and Shahzad Naseem. Gold Nanoparticles: An Efficient Antimicrobial Agent against Enteric Bacterial Human Pathogen. Nanomaterials (Basel). 2016; 6(4): 71.

7. You Sook Cho and Hee-Bom Moon. The Role of Oxidative Stress in the Pathogenesis of Asthma. Allergy Asthma Immunol Res. 2010; 2(3):183–7.

8. Afaf El-Ansary and Layla M Faddah. Nanoparticles as biochemical sensors. Nanotechnol Sci Appl. 2010; 3:65–76

9. Alfredode la Escosura-Muñiz, ClaudioParolo, ArbenMerkoçi. Immunosensing using nanoparticles. Materials Today 2010; 13(7-8):24-34.

10. Ling J, Cheng ZH, Yuan FL, et al. Light scattering signals from nanoparticles in biochemical assay, pharmaceutical analysis and biological imaging. Trends Analyt Chem. 2009;(28):4.

Adam Konefał, Wioletta Lniak, Justyna Rostocka, Andrzej Orlef, Maria Sokół, Janusz Kasperczyk, Paulina Jarząbek, Aleksandra Wrońska, Katarzyna Rusiecka. Influence of a shape of gold nanoparticles on the dose enhancement in the wide range of gold mass concentration for high-energy X-ray beams from a medical linac. Reports of Practical Oncology & Radiotherapy 2020; 25(4):579-85.