Molecular Immunology 58 (2014) 139–149

Contents lists available at ScienceDirect

Molecular Immunology

journal homepage: www.elsevier.com/locate/molimm

Therapeutic potential of anti-IL-1β IgY in guinea pigs with allergic asthma induced by ovalbumin

Hu Wei-xu[a,1], Xiang Qin[b], Wen Zhu[c], Chen Yuan-yi[b], Zeng Li-feng[b], Liu Zhi-yong[d], He Dan[b], Wu Xiao-mu[b], Hu Guo-zhu[b,*,1]

[a] Department of Radiation Oncology, Zhongshan Hospital, Fudan University, Shanghai 200032, China
[b] Institute of Clinical Medical Sciences, Jiangxi Province People's Hospital, Nanchang 330006, China
[c] Department of Hematology, Jiangxi Academy of Medical Science, Nanchang 330006, China
[d] Department of Experimental Animal, Institute of Occupational Medicine of Jiangxi, Nanchang 330006, China

ARTICLE INFO

Article history:
Received 4 May 2013
Received in revised form 8 November 2013
Accepted 9 November 2013

Keywords:
Allergic asthma
Cytokines
Guinea pigs
Immunoglobulin yolk
Interleukin-1β
OVA

ABSTRACT

Background: Interleukin-1 beta (IL-1β) plays pivotal roles in the progression of allergic airway inflammation. This study aims to determine whether the blockade of IL-1β can inhibit airway inflammation in guinea pigs with allergic asthma induced by the inhalation of aerosolized ovalbumin (OVA).
Methods: Healthy guinea pigs treated with saline were used as normal controls (group C). The guinea pigs with allergic asthma induced by the inhalation of aerosolized OVA were randomly divided into three groups: (1) the M group containing negative control animals treated with saline; (2) the Z_1 group containing animals treated by the inhalation of atomized 0.1% anti-IL-1β immunoglobulin yolk (IgY); and (3) the Z_2 group containing positive control animals that were treated with budesonide. The inflammatory cells in the peripheral blood (PB) and bronchoalveolar lavage fluid (BALF) were evaluated using methylene blue and eosin staining. Cytokine concentrations were measured using an enzyme-linked immunosorbent assay. Pulmonary sections were examined using hematoxylin-eosin staining.
Results: Allergic inflammation and damage to the pulmonary tissues were decreased in the Z_1 group compared to the M group. Eosinophils and neutrophils in the PB and BALF were significantly decreased in the Z_1 group compared to the M group ($P < 0.05$). Treatment with anti-IL-1β IgY significantly reduced the levels of IL-1β, IL-4, IL-8, IL-13, TNF-α, TGF-β_1 and IgE in the BALF ($P < 0.05$).
Conclusion: The inhalation of aerosolized anti-IL-1β IgY inhibits pathological responses in the pulmonary tissues of guinea pigs with allergic asthma. The inhibitory activity may be due to the decrease in the numbers of eosinophils and neutrophils and the reduced levels of inflammatory cytokines and IgE in the PB and BALF.

© 2013 Elsevier Ltd. All rights reserved.

1. Introduction

Allergic asthma is an inflammatory disease of the airways. T helper type 2 (Th2) cells and proinflammatory cytokines play an important role in the development and progression of airway hyper-responsiveness (AHR), mucus production, infiltration of eosinophils, neutrophils and lymphocytes and airway remodeling. It has been shown that the levels of eosinophils, neutrophils, lymphocytes and cytokines, such as IL-4, IL-5, IL-13, TNF-α and TGF-β1, and IgE, are increased in the peripheral blood (PB), bronchoalveolar lavage fluid (BALF) and bronchi and pulmonary tissues in patients with allergic asthma (Holgate et al., 2009; Brown et al., 2012; Manuyakorn et al., 2008) as well as ovalbumin (OVA)-sensitized guinea pigs (Franova et al., 2013; Toward and Broadley, 2004; Mahajan and Mehta, 2011; Chang et al., 2011), rats and mice (Kang et al., 2010; Yadav et al., 2011).

Although Th2 cytokines have been shown to be the main drivers of allergic asthma, treatment with anti-IL-4, anti-IL-13 or combined IL-4/IL-13 antagonism (Antoniu, 2009) has been found to have limited efficacy in clinical studies and in animal models of allergy-related asthma. Recent studies have shown that the progression of airway inflammation is significantly improved in patients with allergic asthma and in OVA-sensitized/challenged mice when anti-TNF-α antibody (infliximab), soluble TNF-α receptor (sTNFR) and TNF-α antagonists were administered intravenously and

Abbreviations: AHR, airway hyperresponsiveness; BALF, bronchoalveolar lavage fluid; HE, hematoxylin-eosin; IgE, immunoglobulin E; IgY, immunoglobulin yolk; IL-1β, interleukin-1beta; IL-1ra, IL-1 receptor antagonist; IL-4, interleukin-4; IL-8, interleukin-8; IL-13, interleukin-13; Infliximab, anti-TNF-α antibody; Omalizumab, anti-IgE antibody; OVA, ovalbumin; PB, peripheral blood; sRaw, specific airway resistance; sTNFR, soluble TNF-α receptor; TGF-β_1, transforming growth factor-beta 1; Th2, T helper type 2; TNF-α, tumor necrosis factor-alpha.

* Corresponding author. Tel.: +86 0791 86896246.
E-mail address: hgz56@126.com (H. Guo-zhu).

[1] These authors contributed equally to the work.

0161-5890/$ – see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.molimm.2013.11.006

intraperitoneally (Deveci et al., 2008; Nam et al., 2009; Berry et al., 2006). However, anti-TNF-α therapies in asthma might be limited by the unfavorable risk/benefit ratio associated with their long-term use (Antoniu, 2009). The proinflammatory cytokine IL-1β is an important inducer of the pathological progression of allergic asthma. Mature IL-1β creates a proinflammatory milieu that produces airway inflammatory responses (Besnard et al., 2012; Lappalainen et al., 2005). In OVA-sensitized rats, the intratracheal administration of IL-1β has been shown to significantly increase AHR, bradykinin and neutrophil counts in the BALF (Tsukagoshi et al., 1994). However, Li et al. demonstrated that the asthmatic symptoms in guinea pigs were obviously attenuated after administration of rhIL-1ra solution via ultrasonic spraying (Li et al., 2006).

In the current study, we used the inhalation of aerosolized anti-IL-1β IgY to treat allergic asthma in guinea pigs induced by aerosolized OVA inhalation. We have investigated the curative effect of anti-IL-1β IgY and explored the potential mechanisms. Our studies provide strong experimental evidence that supports a novel therapeutic strategy against allergic asthma, which may avoid the adverse reactions associated with systemic therapy and alleviate the pathological inflammatory response.

2. Materials and methods

2.1. Animals

Hartley guinea pigs (male, 7 weeks old, 230 g ± 40 g) were purchased from Changsha City Kaifu District Dongchuang Animal Science and Technology Service (Production license SCXK (Xiang) 2009-0012, Changsha, China). The experimental studies in guinea pigs were performed in accordance with animal experiment guidelines established by The Ministry of Science and Technology of the People's Republic of China. The animal experimental procedures have been approved by the Jiangxi Province People's Hospital Ethics Committee.

2.2. Establishment of a guinea pig model of allergic asthma and experimental groups

After adaptation for 7 days, the guinea pigs were divided into a healthy control (group C) ($n = 15$) and allergic asthma groups. The guinea pig model of allergic asthma was established with 1.0% OVA in saline (Grade II, Sigma, USA), which was atomized by ultrasonic atomization every other day (days 1, 3, 5 and 7) for a total of four times. Each administration delivered 20 mL over 10 min. In group C, the guinea pigs were administered only atomized saline. After a 1-week rest period, the guinea pigs were administered atomized 1.5% OVA in saline (20 mL over 10 min) every day from day 15 to day 25 for a total of eleven times in the allergic asthma groups. In group C, the guinea pigs were administered only atomized saline. A total of 48 guinea pigs with allergic asthma were randomly divided into three groups. (1) A negative control group (the allergic asthma model group, group M) ($n = 16$) was treated with 0.9% saline. (2) The 0.1% anti-IL-1β IgY treatment group (group Z_1) ($n = 16$) was treated with 0.1% anti-IL-1β IgY (prepared in the laboratory, purity 85%, valence combined recombinant human IL-1β: 1:3200 mg^{-1} protein). Laying hens were inoculated with recombinant human IL-1β. Anti-IL-1β IgY was extracted from the yolk using the water dilution method and purified by salt fractionation. The purity of the IgY was determined by nonreducing SDS-polyacrylamide gel electrophoresis. The valence of anti-IL-1β IgY was determined by an enzyme-linked immunosorbent assay (ELISA) using 1 mg of purified IgY. (3) The positive control group (the budesonide suspension treatment group, group Z_2) ($n = 16$) was treated with a budesonide suspension (1 mg/2 mL, AstraZeneca Pty Ltd.). Beginning on the 21st day after challenge (on the seventh day to the eleventh day in the challenge phase), the asthmatic guinea pigs in the allergic asthma model group (group M) received atomized 0.9% saline, those in the 0.1% anti-IL-1β IgY treatment group (group Z_1) received 0.1% anti-IL-1β IgY and those in the budesonide suspension treatment group (group Z_2) received budesonide for 5 days. Each group received 20 mL over 10 min.

2.3. Plasma and BALF collection

Plasma and BALF were collected according to standard procedures (Nam et al., 2009). In brief, the guinea pigs were anesthetized using 10% chloral hydrate via intraperitoneal injection. Each heart was exposed and heart blood was withdrawn using a 5.0-mL syringe with EDTA. Blood was centrifuged and plasma was stored at −80 °C. The left trachea was exposed and cannulated with silicone tubing attached to a 23-guage needle on a 5.0-mL syringe. After instillation of 2.0 mL of sterile PBS through the trachea into the lung, BALF was withdrawn and centrifuged. Supernatants were stored at −80 °C. Plasma was collected at 2 h (there were three guinea pigs in group C and four guinea pigs each in groups M, Z_1 and Z_2), 4 h (there were four guinea pigs in each group), 8 h (there were four guinea pigs in each group) and 24 h (there were four guinea pigs in each group) in random order. BALF was collected at 2 h (there were three guinea pigs in group C and four guinea pigs each in groups M, Z_1 and Z_2), 4 h (there were three guinea pigs in group C and four guinea pigs each in groups, M, Z_1 and Z_2), 8 h (there were four guinea pigs in each group) and 24 h (there were three guinea pigs in the Z_1 group and four guinea pigs in groups C, M and Z_2) in random order.

2.4. Cell staining

Drops of the blood and precipitated cells in the BALF were placed onto a slide and smeared. After drying at room temperature, methylene blue was dripped onto the slide. The cells were fixed and stained for 1–2 min, and eosin was dripped onto the slide and the cells were stained for an additional 10 min. The slide was then rinsed with distilled water. More than 200 cells were randomly counted in a high power field, and eosinophils, neutrophils, lymphocytes, macrophages, basophils and epithelial cells were counted.

2.5. Pathological examination of lung tissue

Guinea pigs were anesthetized using 10% chloral hydrate via intraperitoneal injection. After the blood and BALF were drawn, air was injected into heart and the guinea pigs were sacrificed. The right lung was exposed and removed. The right lung was removed at the same time that the plasma and BALF were collected. Paraffin section preparation: the lung tissue was fixed in 10% formaldehyde for 4 h, treated with 70% ethanol for 3 h, 85% ethanol twice for 2 h, 95% ethanol twice for 1.5 h, anhydrous alcohol twice for 1 h and dimethylbenzene twice for 1 h. The treated lung tissues were paraffin embedded and cooled, and a 5-μm slice was cut and adhered to a slide treated with polylysine. The hematoxylin-eosin (HE) staining was performed according to the standard procedure (Lillie, 1965). In brief, the slide containing the paraffin section was placed in a slider and then treated as follows: xylene 4 times × 5 min, 100% ethanol 2 times × 5 min, 95% ethanol 2 times × 5 min, 80% ethanol 2 times × 5 min, rinsed for 5 min with tap water, hematoxylin solution for 10 min, rinsed for 5 min with tap water, 1.0% hydrochloric acid alcohol solution for 5–10 s, rinsed for 5 min with tap water, eosin solution for 1–2 min, rinsed for 5 min with tap water, 75% ethanol for 2 min, 85% ethanol for 2 min, 95% ethanol for 2 min,

Download English Version:

https://daneshyari.com/en/article/5916908

Download Persian Version:

https://daneshyari.com/article/5916908

Daneshyari.com