Eosinophilic pneumonia induced by lettuce: A case report

Reina Sekiya
Kobe University Graduate School of Medicine
Tatsuya Nagano (tnagano@med.kobe-u.ac.jp)
Kobe University Graduate School of Medicine https://orcid.org/0000-0003-0790-5139
Tatsuya Moriyama
Kinki University: Kinki Daigaku
Aki Kawaguchi
Kobe University Graduate School of Medicine
Takafumi Fukui
Kobe University Graduate School of Medicine
Chihiro Mimura
Kobe University Graduate School of Medicine
Yohei Kimura
Hyogo Prefectural Awaji Medical Center: Hyogo Kenritsu Awaji Iryo Center
Hisashi Ohnishi
Akashi Medical Center: Akashi Iryo Center
Yoshikazu Kotani
Hyogo Prefectural Awaji Medical Center: Hyogo Kenritsu Awaji Iryo Center
Yoshihiro Nishiura
Kobe University Graduate School of Medicine

Case report

Keywords: Lettuce, Eosinophilic pneumonia, Allergy

Posted Date: November 15th, 2021

DOI: https://doi.org/10.21203/rs.3.rs-1029130/v1

Abstract

Background: Lettuce (*Lactuca sativa*) belongs to the Composite family and is a vegetable widely consumed globally. Although lettuce is extensively cultivated and consumed, lettuce-associated occupational allergy is rarely reported. Herein, we are reporting a case of eosinophilic pneumonia induced by lettuce for the first time.

Case presentation: A 56-year-old female lettuce farmer was admitted to the hospital with a low-grade fever, worsening cough, and dyspnoea. A blood test revealed eosinophilia and a high serum IgE concentration. A chest X-ray taken on admission showed an infiltrative shadow in the upper lung field. Chest CT revealed patchy ground glass opacity on the upper lung field and thickening of the bronchial wall. The bronchoalveolar lavage fluid contained 8% eosinophils. The IgG-binding proteins that reacted with the patient's sera were identified by immunoblot analysis. She was diagnosed as lettuce induced eosinophilic pneumonia and was treated with prednisolone, and her symptoms and radiological findings improved. Wearing a mask and reducing the amount of the crop improved her symptoms the following year.

Conclusions: This is the first case report about lettuce-induced eosinophilic pneumonia which occurred in a lettuce farmer. The avoidance from antigen is quite useful in this patient.

Background

Lettuce (*Lactuca sativa*) is a widely produced and consumed vegetable worldwide. Several studies have reported on the allergenicity of lettuce in relation to oral allergy syndrome, anaphylaxis and occupational dermatitis [1–6]. We reported that some lettuce farmer had respiratory symptoms during harvesting and packaging lettuce and identified epidermis-specific secreted glycoprotein EP1-like (51 kDa) as new lettuce allergen [7].

Acute eosinophilic pneumonia (AEP) was first reported by Allen et al. in 1989 [8]. AEP has an acute onset and shows respiratory insufficiency, hypoxemia, fever, diffuse pulmonary infiltrates, increased eosinophil count (>25%) on bronchoalveolar lavage (BAL) and no evidence of infection or previous atopic illness. Herein, we report a first case of eosinophilic pneumonia (EP) induced by lettuce in a lettuce farmer.

Case Presentation

A 56-year-old female lettuce farmer was admitted to our department in May with a 2-week complaint of low-grade fever, worsening cough, and dyspnoea on exertion. She was treated with 200 mg of amikacin and 500 mg of azithromycin by a family physician, but the therapy had no effect on her low-grade fever, cough or dyspnoea on exertion. The patient had been followed for thyroid nodule. She had no history of smoking, alcohol consumption, animal breeding, bird breeding or chemical agent exposure. She had negative findings on her family history. She had not experienced episodic wheezing or rhonchi. She had

been working as a lettuce farmer for 36 years. The lettuce harvest time was from December to June, with the largest harvest in April.

Her body temperature was 37.9°C, and her percutaneous oxygen saturation was 91% under ambient air conditions. Clubbing was not noted on her fingers. Physical examination of the head, neck, and abdomen was unremarkable. However, auscultation of her chest revealed bilateral wheezing during inspiration. Her arterial blood gas measurements under ambient air conditions were as follows: pH, 7.431; pO_2, 65.4 mmHg; pCO_2, 36.9 mmHg; HCO_3, 24.2 mmol/L. Her leukocyte count was 7,330/μL. C-reactive protein was positive.

Her chest X-ray revealed infiltration in the right upper lobe (Fig. 1A). Plain computed tomography (CT) revealed mild infiltration, ground-grass opacities and reticulo-nodular shadows in both of the upper lobes, indicating interstitial pneumonia (Fig. 1B).

Pulmonary function test (PFT) revealed mixed disorder of both obstructive and restrictive type (Fig. 2A), although PFT revealed normal finding during non-harvesting season (Fig. 2B). Intriguingly, the level of fractional exhaled nitric oxide (FeNO) was 42 ppb, suggesting that eosinophilic inflammation was present in airway. Skin prick test was positive for lettuce. Specific IgE test showed multiple allergies for timothy grass, orchard grass, cedar pollen, hinoki cypress pollen, Japanese white birch, cat, wheat, soybeans, peanut, latex, sesame, crab, kiwi fruit, peach and tomato, although specific IgE test for lettuce has not been commercially available.

Bronchoalveolar lavage fluid (BALF) obtained from the right upper anterior lobe (40/100 ml) showed elevated total cell counts (19.7×10^2 /μl), with an increased percentage of eosinophils (8.5%). The ratio of CD4/CD8 lymphocytes was not detected because the percentage of lymphocytes was low. The BALF culture was negative for both bacteria and fungi.

The laboratory test results are shown in table 1. The patient's Krebs von den Lungen-6 (KL-6) level was 226 U/mL (normal range < 500 U/ml). Her serum total immunoglobulin-E (IgE) level was greatly increased (3038.9 IU/ml), and the D-glucan value was decreased to <11 pg/mL. After admission to our hospital, her clinical symptoms gradually improved within a week with 0.5 mg/kg prednisolone (PSL). Even after a gradual decrease in the PSL, her dyspnoea did not flare up. However, she had general malaise and a mild dry cough during the lettuce harvesting season without abnormal shadow on Chest X-ray. The 3-year follow-up showed that her total IgE level increased in December, peaked in May, and suddenly decreased in August. This result was consistent with the lettuce harvest season. The leaf harvest period started in December, peaked in May and ended in June.

Immunoblot Analysis

A fresh lettuce centre core was cut using a sharp scalpel, and the white juice was spread on a section and collected. This lettuce centre core juice was dissolved in distilled water to prevent hardening (i.e., 20 μL of white juice in 500 μL of distilled water).

The lettuce centre core juice protein (approximately 15 µg of protein) was separated through sodium dodecyl sulphate-polyacrylamide gel electrophoresis (SDS-PAGE). Proteins on the gel were stained with Coomassie Brilliant Blue R-350 (GE Healthcare, Chicago, USA) to detect the total protein patterns (Fig. 3A).

Immunoblot analysis was conducted by transferring the SDS-PAGE gel onto an Immobilon-P^{TM} PVDF membrane (Merck Millipore, Burlington, MA) by using a semi-dry blotting method [9]. The membrane was incubated in 10 mM PBS-T (pH 7.5) and 5% skim milk for blocking. The membrane was then incubated overnight at 4^{o}C in diluted serum (20-fold) in the same blocking buffer. After washing the membranes 4 times with PBS-T for 10 min, the bound primary antibodies were detected by using 5000-fold HRP-conjugated goat anti-human IgG mouse-monoclonal antibody (Jackson Immunoesearch Laboratory, West Grove, PA) and an ECL western blotting kit (GE Healthcare, Boston, MA). After washing the membranes 4 times with PBS-T for 10 min, the resultant chemiluminescent signals were detected on X-ray film (Hyperfilm MP, GE Healthcare). The sera from non-atopic healthy volunteers were used as negative controls.

Fig. 3B shows the binding band of the specific IgG from the sera of the patient and a non-atopic healthy volunteer. The 37 kDa band was recognized in only the patient's serum. Control immunoblot assays with sera from non-atopic patients did not show any IgG-binding bands.

Discussion And Conclusions

a

Present case was difficult to distinguish from hypersensitivity pneumonitis (HP). The following criteria are considered to be significant for the diagnosis of HP: respiratory compatible symptoms (dry cough, dyspnoea and bilateral fine crackles), presence of alveolar eosinophilia in the BAL fluid, and radiologic elements (ground-glass opacities and linear atelectasis) [10]. However, BAL lymphocytosis was not present in this case. The diagnosis of acute eosinophilic pneumonia was not applicable to this case because the patient presented 25% > eosinophils. But the absent of lymphocyte in BALF make it hard to diagnosis as HP. Although HP generally reveals diffuse distribution in the lung, EP may have different BAL findings from place to place. In addition, eosinophilia in blood, increased eosinophils in BALF, elevated FeNO level and increased total IgE suggests that patient are matched with EP probably complicated with asthma induced by lettuce, rather than HP. The lacks of specific provocation test and biopsy were limitation of present case.

SDS-PAGE and IgG immunoblotting showed that the patient's serum contained lettuce-specific IgGs compared to the negative control sera. The specific IgG-binding bands were identified in the white lettuce juice from the centre core at approximately 37 kDa. No case of lettuce-induced EP or HP has been reported in the past.

EP is classified into few type, and one of them is tropical eosinophilia [11]. IgE were significantly higher in cases with tropical eosinophilia [12], and the same is true of present case.

In cases of respiratory symptoms in mushroom worker, the spores from the mushrooms induced pollinosis, rhinitis, asthma and HP [13,14]. Similar to mushroom-related respiratory symptoms, the avoidance from antigen is quite useful in this patient. Although complete avoidance from antigens is very important, it is difficult to stop lettuce cultivation for economic reasons in some cases.

In conclusion, we demonstrated a case of lettuce-associating EP for the first time. Wearing a mask and reducing the amount of the crop improved her symptoms the following year.

Abbreviations

AEP, acute eosinophilic pneumonia; BAL, bronchoalveolar lavage; EP, eosinophilic pneumonia; HP, hypersensitivity pneumonitis

Declarations

Ethics approval and consent to participate

Ethics approval was obtained from the institutional review board of the hospital (permission number #280007) and written informed consent was obtained from patient.

Consent for publication

Written informed consent was obtained from patient.

Availability of data and materials

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

Competing interests

The authors declare that they have no conflicts of interest.

Funding

None declared.

Authors' contributions

RS, TN and TM designed the study. RS, TN, AK, TF, CM, and YK wrote the draft. HO, YK and YN performed the critical review of the manuscript. All authors reviewed and approved the final version.

Acknowledgements

The authors thank patient and investigators who participated in this study.

References

1. Muñoz-García E, Luengo-Sánchez O, Haroun-Díaz E, Maroto AS, Palacín A, Díaz-Perales A, et al. Identification of thaumatin-like protein and aspartyl protease as new major allergens in lettuce (Lactuca sativa). Molecular nutrition & food research. 2013;57.

2. Hartz C, del Mar San Miguel-Moncín M, Cisteró-Bahíma A, Fötisch K, Metzner KJ, Fortunato D, et al. Molecular characterisation of Lac s 1, the major allergen from lettuce (Lactuca sativa). Molecular Immunology. 2007;44.

3. San Miguel-Moncin M, Krail M, Scheurer S, Enrique E, Alonso R, Conti A, et al. Lettuce anaphylaxis: identification of a lipid transfer protein as the major allergen. Allergy. 2003;58.

4. Bascones O, Rodríguez-Pérez R, Juste S, Moneo I, Caballero ML. Lettuce-induced anaphylaxis. Identification of the allergen involved. Journal of investigational allergology & clinical immunology. 2009;19.

5. Vila, SÁnchez, Sanz, DIÉguez, MartÍnez, Palacios, et al. Study of a case of hypersensitivity to lettuce (*Lactuca sativa*). Clinical & Experimental Allergy. 1998;28.

6. Alonso MD, Martin JA, Cuevas M, Parra F, Lezaun A, Conde Salazar L, et al. Occupational protein contact dermatitis from lettuce. Contact Dermatitis. 1993;29.

7. Sekiya R, Nagano T, Moriyama T, Kishi T, Shinke H, Yano E, et al. Occupational respiratory allergy to lettuce in lettuce farmers. Clinical & Experimental Allergy. 2020;50.

8. Allen JN, Pacht ER, Gadek JE, Davis WB. Acute Eosinophilic Pneumonia as a Reversible Cause of Noninfectious Respiratory Failure. New England Journal of Medicine. 1989;321.

9. Kyhse-Andersen J. Electroblotting of multiple gels: a simple apparatus without buffer tank for rapid transfer of proteins from polycrylamide to nitrocellulose. Journal of Biochemical and Biophysical Methods. 1984;10.

10. Lacasse Y, Selman M, Costabel U, Dalphin J-C, Ando M, Morell F, et al. Clinical Diagnosis of Hypersensitivity Pneumonitis. American Journal of Respiratory and Critical Care Medicine. 2003;168.

11. Kar SK, Mania J. Tropical pulmonary eosinophilia in an Orissa village. The National medical journal of India. 6.

12. Crofton JW, Livingstone JL, Oswald NC, Roberts ATM. Pulmonary Eosinophilia. Thorax. 1952;7.

13. Carneiro-Leão L, Carolino F, Pineda F, Miranda M, Plácido J. A Possible New Mushroom Allergen in a Case of Occupational Asthma. Journal of Investigational Allergology and Clinical Immunology. 2019;29.

14. Stanford CF, Hall G, Chivers A, Martin B, Nicholls DP, Evans J. Farmer's lung in Northern Ireland. Occupational and Environmental Medicine. 1990;47.

Tables

Due to technical limitations, Table 1 is only available as a download in the Supplemental Files section.

Figures

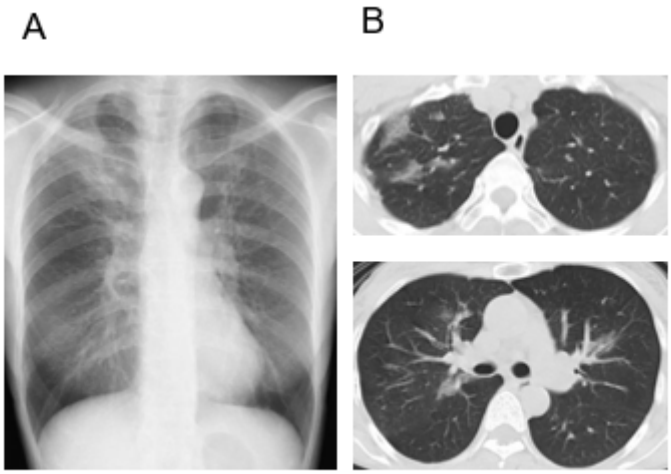

Figure 1

A, Chest X-ray showing infiltration in the right upper lobe. B, Chest plain CT showing mild infiltration, ground-grass opacities and reticulo-nodular shadows in both of the upper lobes, indicating interstitial pneumonia.

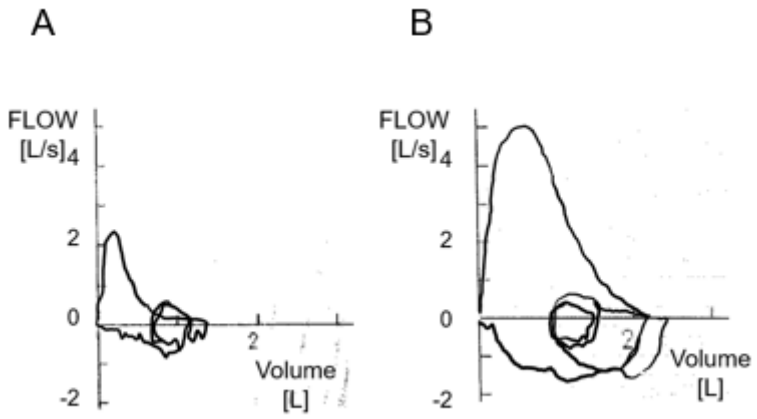

Figure 2

A, Pulmonary function test (PFT) showing mixed disorder during lettuce harvesting seasons. B, PFT showing normal finding during non-harvesting season.

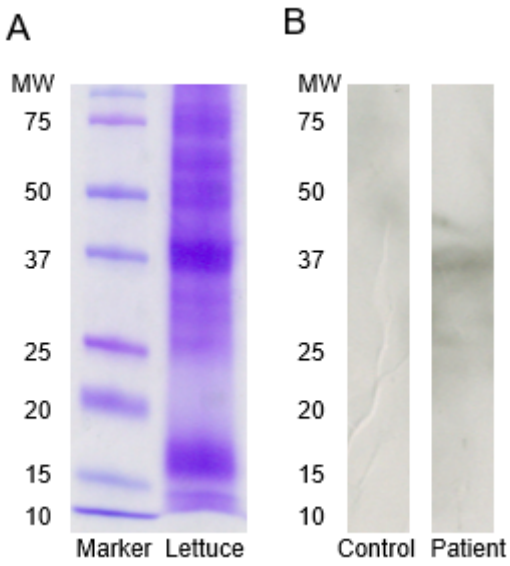

Figure 3

A, Coomassie Brilliant Blue-stained SDS-PAGE gel of the lettuce centre core juice. Marker and molecular weights (MW, kDa) are indicated on the left. B, IgG-binding band of the lettuce centre core juice by immunoblotting is shown. Lane Patient represents the serum from the patient and contains a single protein band at 37 kDa. Lanes Control represents the negative control sera from non-atopic volunteers. MW, molecular weight marker (kDa).

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- renamed038fc.xlsx