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Original article

Surfing as a risk factor for sensitization to poly(γ -glutamic acid) in fermented soybeans, natto, allergy

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PGA	poly(γ -glutamic acid)
SPT	skin prick test
OR	odds ratio
CI	confidence interval

ABSTRACT

Background: Poly(γ -glutamic acid) (PGA) is an allergen in natto, fermented soybeans, which causes late-onset anaphylaxis. We hypothesized that jellyfish stings sensitize adults to PGA because a surfer had allergies to both natto and jellyfish, whose sting contains PGA. The aim of the study was to identify behavioral factors, such as marine sports, associated with PGA sensitization.

Methods: Outpatients diagnosed with food allergies based on relevant clinical history, positive skin test and/or food challenge test answered a questionnaire during a regular visit in 2016.

Results: Questionnaire data from 140 outpatients were analyzed. These patients were divided into two groups: natto allergy group (13 patients, M:F = 10:3, mean age 40.6 years) and non-natto allergy group (127 patients, M:F = 46:81, mean age 44.5 years). All patients with natto allergy had positive results in skin prick test and basophil activation test with PGA. Of these, 92.3% had a marine sport hobby, especially surfing (84.6%). PGA sensitization was independently associated with marine sports (odds ratio, 278.0, 95 percent confidence interval, 36.9–6315.9, $p < 0.001$) adjusted for male sex and sea bathing, but not with male sex or sea bathing. In addition, although there was no significant difference in the experience of marine sports between natto and non-natto allergy groups, the natto allergy group participated significantly more frequently in marine sports than the non-natto allergy group ($p < 0.001$). There was no significant difference between natto consumption amount and PGA sensitization.

Conclusions: Surfing is a risk factor for PGA sensitization in those with allergy to natto.

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Introduction

Natto is a Japanese traditional food made from soybeans fermented with natto bacteria, *Bacillus subtilis*. Fermented soybeans, such as natto and miso, are generally considered hypoallergenic foods for patients with soybean allergy because the fermentation process degrades allergen components.¹ However, in recent years, several cases of natto-induced late-onset anaphylaxis have been reported.^{2–6} The major natto allergen is a polypeptide, poly(γ -glutamic acid) (PGA), which is a constituent of natto mucilage produced by *B. subtilis* during fermentation.⁷ PGA is a water-soluble, biodegradable biopolymer consisting of D- and L-glutamic acid and has a high molecular weight ranging from 100,000 to over 1,000,000.⁷ We speculated that the time required to digest and absorb high molecular weight PGA in the gastrointestinal tract accounted for the late-onset anaphylaxis.²

Natural PGA has only been detected intracellularly in cnidarian nematocyst capsules, although it is present extracellularly in some prokaryotes, including natto bacteria.⁸ The cnidarians, such as jellyfish and sea anemone, produce PGA in nematocysts when stinging. In jellyfish, PGA attracts cations within the capsule and is critical for nematocyst discharge, which involves rapid changes in intracapsular osmotic pressure. Therefore, PGA in venoms might be in contact with skin via jellyfish stings while in the sea.

Recently, we reported a surfer who had allergy to both natto and jellyfish.⁹ In addition, we noticed while checking their food diaries that most of our patients had marine sport hobbies, especially surfing. Furthermore, most reported cases of natto-induced late-onset anaphylaxis lived in a limited area, especially Shonan and its neighboring areas, which is a famous surfing destination in Japan. Therefore, we hypothesized that surfing is a risk factor for sensitization to PGA in natto allergy because of the increased chance of jellyfish sting.⁹ To date, no other studies have reported an association between PGA sensitization and surfing.

To investigate a relationship between surfing and PGA sensitization, we performed a questionnaire survey of demographic and behavior factors, including experience of marine sports, in

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patients with food allergy, and statistically analyzed the replies by comparing natto allergy patients to non-natto allergy patients.

Methods

This study enrolled consecutive outpatients with food allergies who visited Yokohama City University Hospital for regular follow-up between November 1, 2015, and December 31, 2016. These patients were diagnosed with food allergies if they had a convincing history of allergic reactions after the ingestion of the causative food and had positive results on skin prick tests (SPTs) and/or challenge tests with the causative food. Open food challenges were performed as previously described, except in cases with a recent history (within 1 year) of anaphylaxis after ingestion of the causative food and SPT positive to it.^{10,11}

We evaluated each patient's clinical history to determine the involvement of cofactors. The time period accepted for considering a potential relationship with food-allergic reaction was 2 h before food ingestion, combined with the simultaneous intake of nonsteroidal anti-inflammatory drugs and the food.¹² For exercise, a 4-h period after food ingestion was established.

This study was designed to determine demographic and behavioral factors that might be associated with natto allergy with PGA sensitization. The data sources included questionnaires completed by patients and their medical records. Questionnaires regarding their clinical features and behavior were administered at the moment of enrollment (Supplementary Table 1). This study protocol (B151105001) was approved by the Institutional Review Board of Yokohama City University Hospital.

Statistical analysis

Fisher's exact test, chi-squared test and the Mann–Whitney U-test were used to compare differences in the distributions of patient demographic and behavioral factors between the natto allergy and non-natto allergy groups.

Associations between demographic and behavior factors, and PGA sensitization were assessed using a logistic regression model, resulting in the estimation of adjusted odds ratios (ORs) and 95 percent confidence intervals (% CIs). In a stepwise process, all factors with a trend toward an association with natto allergy on univariate analysis ($p < 0.05$) were included in the multivariate model. JMP version Pro 12 software (Cary, NC, USA) was used for all analyses.

Results

Clinical characteristics and marine sport experience of patients

One hundred and forty patients (56 males and 84 females) were enrolled in the study (Table 1). The mean age of the patients was 44.3 years (range, 1–84 years). Thirteen patients (9.3%) had natto allergy. All 13 patients in the natto allergy group had positive results for SPT and basophil activation test with PGA performed as previously described.⁹

Other causative foods included hen eggs ($n = 9$, 6.4%), cow milk ($n = 3$, 2.1%), wheat ($n = 48$, 34.3%), crustaceans ($n = 5$, 3.6%), fruits ($n = 37$, 26.4%), peanuts ($n = 3$, 2.1%), and tree nuts ($n = 6$, 4.3%). Based on the causative foods, participants in this study were divided into two groups: natto allergy group ($n = 13$, M:F = 10:3, mean age 40.6 years) and non-natto allergy group ($n = 127$, M:F = 46:81, mean age 44.5 years).

Analysis of questionnaire data

In the natto allergy group, 12 patients (92.3%) had a marine sports hobby: surfing ($n = 11$, 84.6%) and scuba diving ($n = 1$, 7.7%), and in the non-natto allergy group, 4 patients had experienced marine sports: scuba diving ($n = 2$, 1.6%), surfing ($n = 1$, 0.8%) and windsurfing ($n = 1$, 0.8%) (Fig. 1). In unadjusted comparisons, there was a significant association of experience of marine sports (OR, 369; 95% CI, 38.1–3571.9; $p < 0.005$) and surfing (OR, 693; 95% CI, 58.1–8261.1; $p < 0.005$) with PGA sensitization (Table 1). The unadjusted comparisons also showed a significant association between male sex (OR, 5.87; 95% CI, 1.70–27.2; $p < 0.005$), sea bathing (OR, 10.7, 95% CI, 2.68–42.1; $p < 0.005$), and occupation related to marine sports ($p < 0.01$), and PGA sensitization. However, there was no statistically significant association between the mean amount of natto consumed per month for 5 years prior to the onset of food allergy or age, and PGA sensitization ($p = 0.14$ and 0.29, respectively) (Table 1).

Next, the significant factors listed in Table 1 were entered into a regression analysis. PGA sensitization was independently associated with marine sports (OR, 278.0, 95% CI, 36.9–6315.9, $p < 0.001$) after controlling for potential confounders, such as male sex and sea bathing (Table 2). However, there was no association of PGA sensitization with male sex ($p = 0.14$) or sea bathing ($p = 0.56$) after adjustment for each potential confounder.

Furthermore, detailed data regarding the experience of marine sports were analyzed. In both groups, years of marine sport experience varied from less than 1 year to more than 20 years (Fig. 2). The mean number of weeks per year of marine sport experience ranged from more than 2 weeks to approximately 24 weeks in the natto allergy group, and from less than 1 week to 2 weeks in the non-natto allergy group (Fig. 3). Thus, although there was no significant difference in years of marine sport experience between the natto and non-natto allergy groups ($p = 0.08$), the mean number of weeks per year in marine sport experience prior to the onset of food allergy was significantly higher in the natto allergy group compared with the non-natto allergy group ($p < 0.05$) (Fig. 3). In terms of natto consumption, the mean amount of natto consumed for 5 years prior to the onset of food allergies was 378.2 g and 259.6 g in the natto allergy group and non-natto allergy group, respectively. There was no significant difference between natto consumption prior to the onset of natto allergy and PGA sensitization ($p = 0.14$).

Discussion

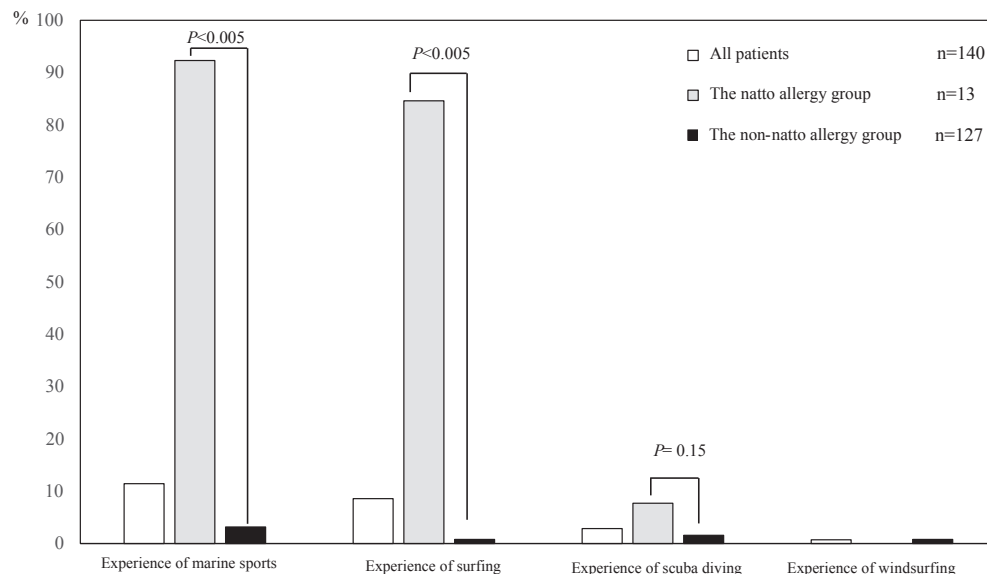
This questionnaire survey showed that marine sports, especially surfing, are a major risk factor for PGA sensitization in natto allergy. Marine sports were consistently and statistically associated with PGA sensitization after adjustment for potential confounders, such as male sex and sea bathing, which were not associated with PGA sensitization. In addition, marine sport enthusiasts in the natto allergy group, had a significantly greater frequency of experiencing marine sports than those in the non-natto allergy group. Therefore, more frequent surfing experiences may contribute to PGA sensitization. To our knowledge, this is the first study demonstrating the impact of marine sports on PGA sensitization, resulting in the onset of natto allergy.

The routes of PGA sensitization are unclear for natto-induced late-onset anaphylaxis. Considering the distribution of PGA in nature, there are two potential routes for its contact and sensitization of subjects: oral ingestion of PGA derived from natto, or epicutaneous stings from Cnidaria. Our study showed that PGA sensitization was significantly associated with an experience of marine

Table 1

Unadjusted Odds ratios and 95% confidential intervals measuring association between natto allergy and subject demographic, clinical and behavior factors.

	Reply	All patients (n = 140)	Natto allergy (n = 13)	Non-natto allergy (n = 127)	Unadjusted OR	95% CI	P value
Male sex	Yes	56 (40%)	10 (76.9%)	46 (36.2%)	5.87	1.70–27.2	0.004
	No [†]	84 (64%)	3 (23.1%)	81 (63.8%)	1.00		
Age group (years)	Average	44.3	40.6	44.5			0.29*
	0–19	20 (14.3%)	0 (0%)	20 (15.7%)			
	20–39	31 (22.1%)	6 (46.2%)	25 (19.7%)			
	40–59	56 (40.0%)	7 (53.8%)	49 (38.6%)			
	60–79	31 (22.1%)	0 (0%)	31 (24.4%)			
	80–	2 (1.4%)	0 (0%)	2 (1.6%)			
Experience of marine sports	Yes	16 (11.4%)	12 (92.3%)	4 (3.1%)	369	38.1–3571.9	<0.0001
	No [†]	124 (88.6%)	1 (7.7%)	123 (96.9%)	1.00		
Experience of surfing	Yes	13 (9.3%)	11 (84.6%)	2 (1.6%)	693	58.1–8261.1	<0.0001
	No [†]	127 (90.7%)	2 (15.4%)	125 (90.7%)	1.00		
Experience of sea bathing	Yes	14 (10.0%)	5 (38.5%)	7 (5.5%)	10.7	2.68–42.1	<0.0001
	No [†]	126 (90.0%)	8 (61.5%)	120 (94.5%)	1.00		
Occupation related to marine sports	Yes	2 (1.4%)	2 (15.4%)	0 (0%)	N.D.	N.D.	<0.01**
	No	138 (98.6%)	11 (84.6%)	127 (100%)			
Amount of natto consumed (gram per month) [‡]	Mean	286.8 (n = 45)	378.2 (n = 13)	259.6 (n = 32)			0.14*
	0	6 (13.3%)	0 (0%)	6 (18.8%)			
	1–200	27 (60%)	8 (61.5%)	19 (59.3%)			
	201–400	5 (11.1%)	1 (7.7%)	4 (12.5%)			
	401–	7 (15.6%)	4 (30.8%)	3 (9.4%)			

p values were calculated by the chi-square test, unless otherwise noted. OR denotes odds ratio, and CI confidence interval.**p* values were calculated by Mann Whitney U test.***p* values were calculated by Fisher's exact test.[†] This group served as the reference category.[‡] Total samples were different from other variables because not all participants answered all questions.**Fig. 1.** Frequency in experience of marine sports in all participants and each group. The natto group had a significantly greater experience of marine sports compared with the non natto-group ($p < 0.005$). Among marine sports, surfing, but not scuba diving and windsurfing, had a significant association with natto allergy. Some of the replies by a responding patient from a single group overlapped. Statistical analysis was performed by Fisher's exact test.**Table 2**

Adjusted ORs and 95% CIs measuring association between PGA sensitization and demographic and behavioral factors.

	Adjusted OR	95% CI	<i>p</i> value
Male sex	4.7	0.59–51.1	0.14
Experience of marine sports	278.0	36.9–6315.9	<0.0001
Experience of sea bathing	2.0	0.19–26.2	0.56

PGA, poly(γ -glutamic acid); OR, odds ratio; and CI, confidence interval.In stepwise process, all factors with a trend toward an association with PGA sensitization allergy on univariate analysis ($p < 0.05$) were included in multivariate model. *p* Value was calculated by the chi-square test, unless otherwise noted.

sports but not with the amount of natto consumed prior to the onset of natto allergy. Therefore, epicutaneous PGA exposure by cnidarian stings might significantly affect PGA sensitization rather than oral exposure by ingesting of natto.

Jellyfish envenomation causes local and systemic injuries in humans and may also provoke immunological responses.^{13,14} Thousand jellyfish tubules penetrate human skin and release toxin combined with other constituents while stings. Although the effect of each venom component on the human immune system is unclear, there are potential stimuli in nematocyst envenomation: the venom components, such as porins and secretion substances

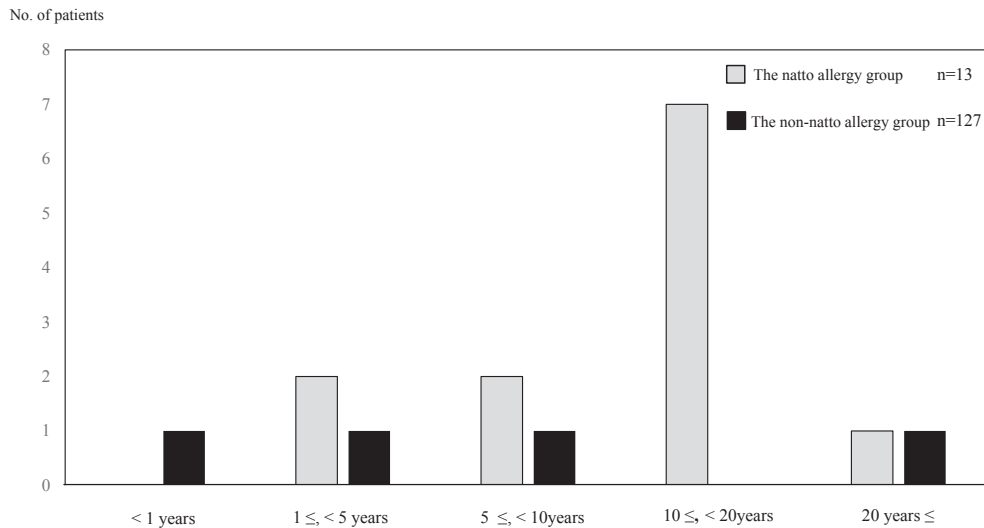


Fig. 2. Years of marine-sport experience in each group. There was no significant difference in years of marine-sport experience between the natto and the non-natto groups ($p = 0.08$). The p value was calculated by Mann–Whitney U -test.

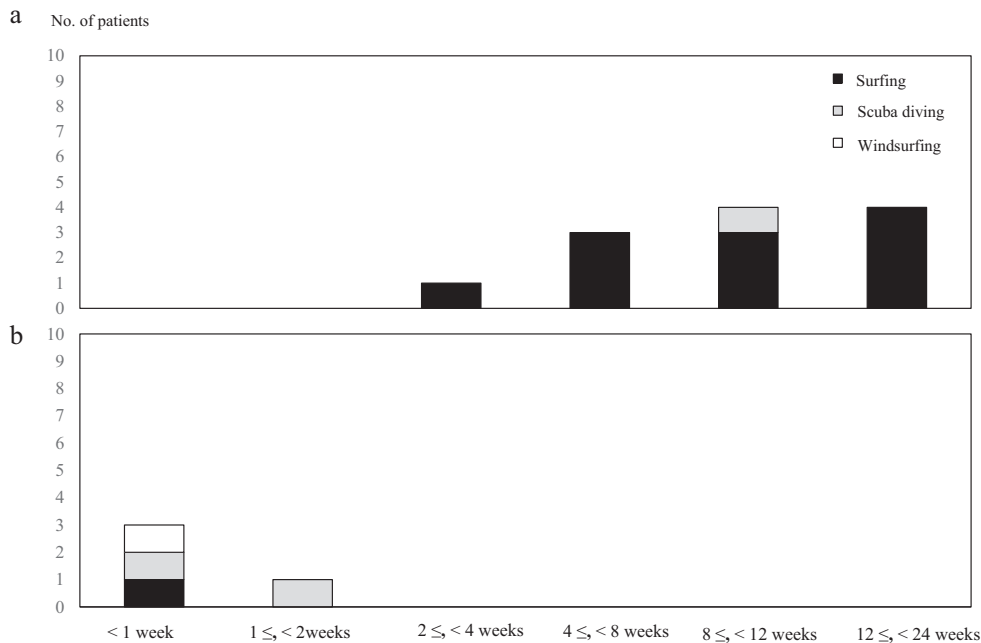


Fig. 3. The mean number of weeks per year of marine-sport experience between the natto group (a) and the non-natto group (b). There was a significant difference in the mean number of weeks per year of marine-sport experience between the natto and the non-natto groups ($p < 0.005$). The p value was calculated by the Mann–Whitney U -test.

(bioactive lipids and amines), the components of the nematocyst tubule, and physical changes (hypo-osmolality, acidification, and reactive oxygen species) at the sting site.^{15,16} Especially, porin is the main constitute of venom and activates keratinocytes and dermal mast cells.¹³ Repeated stings may induce IgE-mediated allergic reactions to toxin or tubule components.¹³ Indeed, the stings of pelagic jellyfish, which inhabit all oceans, cause wheals and pruritus.¹⁷ The case of anaphylaxis after a jellyfish sting was reported. In that case, the patient's basophils immediately released histamine in response to exposure to a crude extract of venom derived from tentacles of another jellyfish *Chrysaora quinquecirrha*, suggesting the presence of triggers, which induce degranulation from mast cells and basophils, in the venom, or the development of IgE-mediated allergic response to a venom-derived antigen.¹⁸ Thus,

jellyfish envenomation may have an adjuvant effect in sensitization to venom constituents, containing PGA.

Surfing is more significantly associated with PGA sensitization than other marine sports. However, the reason for this strong association between surfing and PGA sensitization is unclear. Surfing is a surface watersport and jellyfish are surface dwellers. Therefore, surfers might have a greater chance to be stung by jellyfish compared with other marine sports. Indeed, all surfers in the natto allergy group often experienced jellyfish stings, especially in the summer. Surfers wait for an ideal wave for several hours per visit and often only wear sea bathing suits rather than wetsuits; therefore, most of the body is exposed in the sea for long time, increasing the risk of jellyfish stings. In contrast, scuba divers regularly wear wetsuits and windsurfers only float in the sea for short time.

The present study revealed that the mean number of weeks per year in marine-sport experience was significantly greater in the natto allergy group than in the non-natto allergy group. All surfers constantly participated in surfing all year around. We speculated that the reason why surfers had the greatest PGA sensitization among marine sports might be because of the long floating time in the sea without a wetsuit. However, further investigations are needed to elucidate the association between surfing and PGA sensitization.

Sea bathing is more common than surfing and other marine sports. In the present study, there was no significant association of sea bathing with PGA sensitization after adjustment for marine sports and male sex. Although years of sea bathing experience in the natto allergy group was comparable to the non-natto allergy group, the mean number of weeks per year of sea bathing experience in the natto group was significantly longer than in the non-natto group. Therefore, occasional sea bathing has no risk for PGA sensitization.

One female patient had experienced neither marine sports nor sea bathing in the present study. In this case, the sensitization route of PGA is unknown. In a previous study, two pediatric cases, a 12-year-old boy and a 7-year-old girl, had also no experience of marine sports, including surfing.^{5,6} Currently, a commercial particle of PGA is widely used in cosmetics, drug vehicles, soft drinks, dressings, and seasonings to thicken their texture and enhance their taste.⁷ Indeed, one of our patients diagnosed with PGA allergy developed an anaphylactic reaction after the ingestion of commercial noodles with soup containing the commercial PGA.¹⁹ Therefore, exposure to the commercial PGA might induce allergic sensitization and induction through various routes: skin exposure of cosmetics and intravascular exposure of drugs. However, our female patient did not use cosmetics or other products containing PGA.

Meat can cause delayed-onset anaphylaxis in a similar manner to natto. The major allergen in meat-induced allergy is a protein containing oligosaccharide composed of galactose- α -1,3-galactose (α -gal) molecules.²⁰ Many meat allergy subjects live in areas with an epidemic of tick bites. Commins *et al.* suggested that bites from ectoparasitic ticks are the sensitizing event that leads to the development of IgE specific to the oligosaccharide α -gal, which results in a loss of tolerance to non-primate mammalian meat and related food products in some individuals.²⁰ Recently, there has been increasing evidence that the skin is an important route for IgE sensitization to proteins such as peanut and wheat.^{21,22} In 2008, Lack proposed a new “dual-allergen exposure hypothesis” that suggested the ingestion of food promoted immune tolerance to food antigens, whereas exposure to food antigens through inflamed skin facilitated allergic sensitization.²³ Interestingly, natto and red meat allergies share several clinical characteristics. First, sensitization to allergens might be induced by animal stings and bites in both allergies. In the case of red meat allergy, bites of larval lone star ticks, similar to adult ticks, can be intensely pruritic. However, bites from the deer tick *Ixodes scapularis*, which transmits Lyme disease, do not generally induce a pruritic skin response. In fact, itching after tick bites was associated with an increase in the risk of developing allergic sensitization to α -gal.²⁴ Some substances in tick saliva may induce Th2 responses, which promote IgE sensitization to α -gal.²⁰ Likewise, in PGA allergy, Cnidaria venom may have a similar adjuvant effect for PGA sensitization. Second, there is cross-reactivity between creatures belonging to unrelated categories by biological classification: insects and non-primate animals in α -gal allergy, and bacteria and cnidarians in PGA allergy. Third, many patients live in limited areas where there is an epidemic of the causative animal sting or bite. Furthermore, the causative allergens are simple structures such as a sugar chain consisting of galactose

in red meat allergy and a polymer consisting of glutamic acids in PGA allergy. Finally, the time frame from food ingestion to the onset of symptoms is longer in both allergies than in typical IgE-mediated food allergies. Therefore, considering natto and red meat allergies together may lead to a new category of epicutaneous sensitization as a mechanism of food allergy. In this category, stings and bites of animals induce the epicutaneous sensitization of antigens in their saliva and venom, resulting in cross-reactivity to food allergens and the consequent onset of food allergy. However, the process through which these antigens enter the skin and the time frame from exposure to antibody response is currently unknown.

This study had some limitations including the small number of subjects enrolled. To date, only a few cases of natto allergy have been reported because the late onset of natto allergy might make its diagnosis difficult. In addition, commercial diagnostic procedures for PGA allergy are unavailable at present. Another limitation is the small number of patients with other food allergies because of the investigation was set in a single hospital setting. Therefore, further investigation of a large number of subjects from various areas of Japan is required to clarify whether PGA sensitization might be associated with marine sports and be promoted by Cnidaria stings.

In conclusion, this study identified surfing as a major risk factor for PGA sensitization by statistically comparing a natto allergy group with a non-natto allergy group. PGA sensitization may occur via inflamed skin following cnidarian stings while floating in the sea. If marine sport enthusiasts, especially surfers, visit clinics because of suspected food-induced anaphylaxis, natto allergy with PGA sensitization should be considered for allergic examinations.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at <https://doi.org/10.1016/j.alit.2017.11.001>.

Conflict of interest

The authors have no conflict of interest to declare.

Authors' contributions

NI and MA designed research; NI and MM summarized and analyzed clinical data; NI wrote the paper. All authors read and approved the final manuscript.

References

1. Frias J, Song YS, Martinez-Villaluenga C, Gonzalez de Mejia E, VidalValverde C. Immunoreactivity and amino acid content of fermented soybean products. *J Agric Food Chem* 2008;**56**:99–105.
2. Inomata N, Osuna H, Ikezawa Z. Late-onset anaphylaxis to *Bacillus natto*—fermented soybeans. *J Allergy Clin Immunol* 2004;**113**:998–1000.
3. Inomata N, Osuna H, Yanagimachi M, Ikezawa Z. Late-onset anaphylaxis to fermented soybeans: the first confirmation of food-induced, late-onset anaphylaxis by provocation test. *Ann Allergy Asthma Immunol* 2005;**94**:402–6.
4. Honda T, Michigami M, Miyachi Y, Kabashima K. A case of late-onset anaphylaxis to fermented soybeans (natto). *J Dermatol* 2014;**41**:940–1.
5. Mori S, Tsumagari S, Kurihara K. A case of a 7-year-old girl with late-onset anaphylaxis to fermented soybeans. *Pediatr Allergy Immunol* 2017;**28**:501–2.
6. Matsubayashi R, Matsubayashi T, Yokota T, Ohro Y, Fujita N, Nakashima Y, et al. Pediatric late-onset anaphylaxis caused by natto (fermented soybeans). *Pediatr Int* 2010;**52**:657–8.
7. Inomata N, Nomura Y, Ikezawa Z. Involvement of poly (γ - glutamic acid) as an allergen in late onset anaphylaxis due to fermented soybeans (natto). *J Dermatol* 2012;**39**:409–12.
8. Weber J. Poly(c-glutamic acid)s are the major constituents of nematocysts in Hydra (Hydrozoa, Cnidaria). *J Biol Chem* 1990;**265**:9664–9.
9. Inomata N, Chin K, Aihara M. Anaphylaxis caused by ingesting jellyfish in a subject with fermented soybean allergy: possibility of epicutaneous

- sensitization to poly- gamma- glutamic acid by jellyfish stings. *J Dermatol* 2014;**41**:752–3.
10. Nakamura K, Inomata N, Ikezawa Z. Dramatic augmentation of wheat allergy by aspirin in a dose-dependent manner. *Ann Allergy Asthma Immunol* 2006;**97**: 712–3.
 11. Bolhaar ST, van de Weg WE, van Ree R, Gonzalez-Mancebo E, Zuidmeer L, Bruijnzeel-Koomen CA, et al. In vivo assessment with prick-to-prick testing and double-blind, placebo-controlled food challenge of allergenicity of apple cultivars. *J Allergy Clin Immunol* 2005;**116**:1080–6.
 12. Pascal M, Muñoz-Cano R, Reina Z, Palacín A, Vilella R, Picado C, et al. Lipid transfer protein syndrome: clinical pattern, cofactor effect and profile of molecular sensitization to plant-foods and pollens. *Clin Exp Allergy* 2012;**42**: 1529–39.
 13. Tibballs J, Yanagihara AA, Turner HC, Winkel K. Immunological and toxino- logical responses to jellyfish stings. *Inflamm Allergy Drug Targets* 2011;**10**: 438–46.
 14. Metz M, Maurer M. Innate immunity and allergy in the skin. *Curr Opin Immunol* 2009;**21**:687–93.
 15. Chung JJ, Ratnapala LA, Cooke IM, Yanagihara AA. Partial purification and characterization of a hemolysin (CAH1) from Hawaiian box jellyfish (*Carybdea alata*) venom. *Toxicon* 2001;**39**:981–90.
 16. Prasad P, Yanagihara AA, Small-Howard AL, Turner H, Stokes AJ. Secretogranin III directs secretory vesicle biogenesis in mast cells in a manner dependent upon interaction with chromogranin A. *J Immunol* 2008;**181**:5024–34.
 17. Tibballs J. Australian venomous jellyfish, envenomation syndromes, toxins and therapy. *Toxicon* 2006;**48**:830–59.
 18. Togias AG, Burnett JW, Kagey-Sobotka A, Lichtenstein LM. Anaphylaxis after contact with a jellyfish. *J Allergy Clin Immunol* 1985;**75**:672–5.
 19. Inomata N, Chin K, Nagashima M, Ikezawa Z. Late-onset anaphylaxis due to poly (γ -glutamic acid) in the soup of commercial cold Chinese noodles in a patient with allergy to fermented soybeans (natto). *Allergol Int* 2011;**60**:393–6.
 20. Commins SP, Satinover SM, Hosen J, Mozena J, Borish L, Lewis BD, et al. Delayed anaphylaxis, angioedema, or urticarial after consumption of red meat in patients with IgE antibodies specific for galactose- α -1,3-galactose. *J Allergy Clin Immunol* 2009;**123**:426–33.
 21. Brough HA, Simpson A, Makinson K, Hankinson J, Brown S, Douiri A, et al. Peanut allergy: effect of environmental peanut exposure in children with filaggrin loss-of-function mutations. *J Allergy Clin Immunol* 2014;**134**: 867–75. e1.
 22. Yokooji T, Kurihara S, Murakami T, Chinuki Y, Takahashi H, Morita E, et al. Characterization of causative allergens for wheat-dependent exercise-induced anaphylaxis sensitized with hydrolyzed wheat proteins in facial soap. *Allergol Int* 2013;**62**:435–45.
 23. Lack G, Fox D, Northstone K, Golding J. Avon Longitudinal Study of Parents and Children Study Team. Factors associated with the development of peanut allergy in childhood. *N Engl J Med* 2003;**348**:977–85.
 24. Burke G, Wikel SK, Spielman A, Telford SR, McKay K, Krause PJ, et al. Hyper- sensitivity to ticks and Lyme disease risk. *Emerg Infect Dis* 2005;**11**:36–41.

Supplementary Table 1 Questionnaire

1. What kinds food cause your food allergy?

- 1.1 Hen egg
- 1.2 Cow milk
- 1.3 Wheat
- 1.4 Peanuts
- 1.5 Tree nuts
- 1.6 Crustaceans
- 1.7 Fruits/vegetables
- 1.8 Natto
- 1.9 Others

2. Had you taken part in any marine sports prior to the onset of food allergy ?

- 2.1 Yes
- 2.2 No

3. What type of marine sports do you often take part in?

- 3.1 Surfing
- 3.2 Scuba diving
- 3.3 Others ()

4. How long have you been taken part in marine sports?

- 4.1 <1 years
- 4.2 1years ≤, <5 years
- 4.3 5 years ≤, < 10years
- 4.4 10 years ≤, < 20 years
- 4.5 20 years ≤

5. How many weeks per years do you take part in marine sports?

- 5.1 <1 week
- 5.2 1 week ≤, <2weeks
- 5.3 2 weeks ≤, <4 weeks
- 5.4 4 weeks ≤, <8 weeks
- 5.5 8 weeks ≤, <12 weeks
- 5.6 12 weeks ≤, < 24 weeks
- 5.6 24 weeks≤

6. Dose your occupation involve marine sports?

- 6.1 Yes
- 6.2 No

7. How often had you consumed natto in the 5 years prior to the onset of food allergy?

8. How much natto do you consume in one serving?
