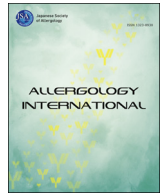




Contents lists available at ScienceDirect

Allergology International

journal homepage: <http://www.elsevier.com/locate/alit>

Original article

Long-term safety of subcutaneous immunotherapy with TO-204 in Japanese patients with house dust mite-induced allergic rhinitis and allergic bronchial asthma: Multicenter, open label clinical trial

Takao Fujisawa^{a,*}, Terufumi Shimoda^b, Keisuke Masuyama^c, Kimihiro Okubo^d, Kohei Honda^e, Mitsuhiro Okano^f, Toshio Katsunuma^g, Atsuo Urisu^h, Yasuto Kondo^h, Hiroshi Odajimaⁱ, Kazuyuki Kurihara^j, Makoto Nagata^k, Masami Taniguchi^l, Shoichiro Taniuchi^m, Satoru Doiⁿ, Tomoshige Matsumoto^o, Shoji Hashimoto^o, Akihiko Tanaka^p, Kensuke Natsui^q, Nahoko Abe^q, Hideki Ozaki^q

^a Allergy Center, Mie National Hospital, Mie, Japan^b Clinical Research Center, Fukuoka National Hospital, Fukuoka, Japan^c Department of Otorhinolaryngology, Head and Neck Surgery, University of Yamanashi, Yamanashi, Japan^d Department of Otorhinolaryngology, Nippon Medical School, Tokyo, Japan^e Department of Otorhinolaryngology, Head and Neck Surgery, Akita Graduate School of Medicine, Akita, Japan^f Department of Otolaryngology-Head and Neck Surgery, Okayama University Graduate School of Medicine, Dentistry and Pharmaceutical Sciences, Okayama, Japan^g Department of Pediatrics, Jikei University Daisan Hospital, Tokyo, Japan^h Department of Pediatrics, Fujita Health University, The Second Teaching Hospital, Nagoya, Japanⁱ Department of Pediatrics, Fukuoka National Hospital, Fukuoka, Japan^j Department of Allergy, Kanagawa Children's Medical Center, Yokohama, Japan^k Department of Respiratory Medicine, Saitama Medical University, Saitama, Japan^l Department of Allergy, Sagami National Hospital, Kanagawa, Japan^m Department of Pediatrics, Kansai Medical University Takii Hospital, Osaka, Japanⁿ Department of Pediatrics, Osaka Prefectural Hospital Organization Osaka Habikino Medical Center, Osaka, Japan^o Department of Allergy and Internal Medicine, Osaka Prefectural Hospital Organization Osaka Habikino Medical Center, Osaka, Japan^p Division of Respiratory Medicine and Allergology, Department of Medicine, Showa University, School of Medicine, Tokyo, Japan^q Department of Clinical Development, Torii Pharmaceutical Co., Ltd., Tokyo, Japan

ARTICLE INFO

Article history:

Received 19 June 2017

Received in revised form

21 October 2017

Accepted 24 October 2017

Available online xxx

Keywords:

Allergen immunotherapy

Allergic bronchial asthma

Allergic rhinitis

Clinical trial

House dust mite

Abbreviations:

AE, Adverse event; ADR, Adverse drug reaction; AIT, Allergen immunotherapy;

ABSTRACT

Background: To evaluate the long-term safety of subcutaneous immunotherapy with TO-204, a standardized house dust mite (HDM) allergen extracts, we conducted a multicenter, open label clinical trial.

Methods: Japanese patients aged 5–65 years were eligible for the study, if they had HDM-induced allergic rhinitis (AR), allergic bronchial asthma (BA), or both. TO-204 was administered in a dose titration scheme, and the maintenance dose was determined according to the predefined criteria. The treatment period was 52 weeks, and patients who were willing to continue the treatment received TO-204 beyond 52 weeks. This clinical trial is registered at the Japan Pharmaceutical Information Center (Japic CTI-121900).

Results: Between July 2012 and May 2015, 44 patients (28 with AR and 16 with allergic BA) were enrolled into the study. All patients were included in the analysis. The duration of treatment ranged from 23 to 142 weeks and the median maintenance dose was 200 Japanese allergy units (JAU). Adverse events occurred in 22 patients (50%). The most common adverse event was local reactions related to the injection sites. Four patients experienced anaphylactic reactions when they were treated with the dose of 500 JAU. Two patients experienced anaphylactic shock with the doses of 1000 JAU at onset. These 6 patients could continue the study with dose reduction.

* Corresponding author. Allergy Center, Mie National Hospital, 357 Osato-kubota, Tsu, Mie 514-0125, Japan.

E-mail address: fujisawa@mie-m.hosp.go.jp (T. Fujisawa).

Peer review under responsibility of Japanese Society of Allergology.

<https://doi.org/10.1016/j.alit.2017.11.004>

1323-8930/Copyright © 2017, Japanese Society of Allergology. Production and hosting by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

AR, Allergic rhinitis; BA, Bronchial asthma; *D. farinae*, *Dermatophagoides farinae*; *D. pteronyssinus*, *Dermatophagoides pteronyssinus*; IgE, Immunoglobulin E; IgG, Immunoglobulin G; IgG4, Immunoglobulin G4; IM, Intramuscular injection; IV, Intravenous infusion; HDM, House dust mite; JAU, Japanese allergy unit; JSA, Japanese Society of Allergology; SCIT, Subcutaneous immunotherapy; SD, Standard deviation; QOL, Quality of life; WAO, World Allergy Organization

Conclusions: Safety profile of TO-204 was acceptable in Japanese patients with HDM-induced AR or allergic BA. Higher doses should be administered carefully, because the risk of anaphylaxis increased at doses of 500 or 1000 JAU.

Copyright © 2017, Japanese Society of Allergology. Production and hosting by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

Introduction

Allergic disease is one of the most common chronic diseases. The basic treatment for this disease is to avoid the allergen exposure, and pharmacotherapy including antihistamines and topical corticosteroids are used if necessary. However, it is difficult to completely avoid exposure to allergens in daily life. Furthermore, the pharmacotherapy can alleviate various allergic symptoms, but they often relapse if it is discontinued.

Allergen immunotherapy (AIT) is a treatment that modify Th2-skewed immune responses in patients with allergic diseases through the administration of “culprit” allergens, and this therapy may overcome the problems mentioned above. The mechanisms of AIT that have been reported are the following: production of immunoglobulin G (IgG) antibodies that act as blocking antibodies to allergens; inhibition of the production of immunoglobulin E (IgE) antibodies; activation of T regulatory cells; or decreasing the number and inhibiting the activation of Th2 cells.^{1–3} It is reported that long-term AIT modifies the immunological conditions of patients and AIT is expected to be a curative treatment for allergic bronchial asthma (BA) and allergic rhinitis (AR).^{1–5} In addition, AIT inhibits the onset of allergic BA in patients with AR⁶ and new sensitization to other allergens.^{7–9} These effects may also lead to the favorable benefits.

House dust mite (HDM) is a globally important allergen to cause AR and allergic BA. While the use of standardized allergen extracts is recommended in subcutaneous immunotherapy (SCIT) for patients with HDM-induced allergy,¹ unstandardized house dust extracts have been used in Japan. Although the house dust extract preparations are effective in patients with HDM-induced allergy,^{10,11} the potency of mite contained in the preparations is not high enough compared to other standardized HDM extracts available in Western countries. Therefore, standardized HDM extracts have been required in clinical practice in Japan.

TO-204 is a HDM extract preparation standardized for SCIT by the United States Food and Drug Administration (FDA). The efficacy and safety of HDM extract preparations for SCIT have been reported in the previous studies conducted in Western countries.^{2,12–15} However, these preparations have never been used in Japan and their safety profiles remain uncertain. Because SCIT continues over the long term, it is especially important to evaluate the long-term safety of these preparations. Therefore, we conducted a clinical trial of TO-204 in Japanese patients with HDM-induced AR or allergic BA. The primary objective of this study was to evaluate the long-term safety of TO-204. Especially, we aimed to determine safety profile at maximal tolerable dose of TO-204 since efficacy and adverse events of allergen immunotherapy are dose-dependent^{16–18} and the maximal tolerable dose for Japanese patients is not known. Efficacy was analyzed in ad-hoc manner.

Methods

Study design and ethical considerations

This multicenter, open label clinical trial was conducted at 13 sites in Japan between July 2012 and May 2015. The study was conducted in accordance with the Declaration of Helsinki and Good Clinical Practice, and the protocol was approved by the institutional review board of each participating center. All patients or their guardians gave written informed consent. This clinical trial is registered at the Japan Pharmaceutical Information Center (Japic CTI-121900).

Patients

According to the international recommendation,¹⁹ patients aged 5–65 years who had AR, allergic BA, or both were eligible for the study if they met the following criteria: positive skin prick test results to HDM allergen; and serum specific HDM IgE levels ≥ 0.7 (kU/L, Immuno CAP). For patients with AR, the severity was determined according to the Practical Guideline for the Management of Allergic Rhinitis in Japan 2009²⁰ published by the Japanese Society of Allergology (JSA), and patients with mild to very severe AR were included. For patients aged 5–15 years who had allergic BA, the severity was determined according to the Japanese Pediatric Guideline for the Treatment and Management of Asthma 2012²¹ published by the Japanese Society of Pediatric Allergy and Clinical Immunology, and patients with intermittent, mild persistent, or moderate persistent allergic BA were included. The severity of allergic BA in patients aged 16 years or older was determined according to the Asthma Prevention and Management Guideline 2012²² published by the JSA, and patients with mild intermittent, mild persistent, or moderate persistent allergic BA were included. When patients had AR complicated by allergic BA, patients aged 5–15 years who had intermittent and mild persistent allergic BA and patients aged 16 years or older who had mild intermittent allergic BA were included since allergen immunotherapy has potential to prevent asthma progression²³ and to improve long term outcome.^{13,24}

Patients were excluded from the study if they had the following: uncontrolled asthma symptoms, asthma or rhinitis symptoms induced by allergen other than HDM throughout a year; a history of serious adverse reactions related to AIT; a history of anaphylactic shock or angioedema; concurrent drug allergy or a history of drug allergy; hepatic dysfunction; or renal impairment.

Treatment

TO-204 100,000 JAU/mL (ALK-Abellò, Round Rock, TX, USA), which is a mixture of allergen extracts from *Dermatophagoides farinae* (*D. farinae*) and *Dermatophagoides pteronyssinus* (*D.*

pteronysinus) in equal amounts, was used as the study drug. TO-204 contains major allergens Der f 1, Der f 2, Der p 1, and Der p 2 of HDM, and its potency is equivalent to 10,000 AU/mL determined by the FDA. Because the JSA originally standardized HDM allergen extract for AIT in Japan in 2014,²⁵ the potency of TO-204 is expressed in Japanese allergy units (JAU) in this paper.

This study consisted of a run-in period, up-dosing phase, and maintenance phase (Supplementary Fig. 1). In the run-in period, the threshold concentration in each patient was determined according to the intradermal reaction. TO-204 serially diluted by 10-fold (i.e., 100,000-, 10,000-, 1000-, and 100-fold) were administered intradermally in ascending order, and the lowest concentration positive (diameter of erythema is larger than 20 mm or the diameter of wheal is larger than 9 mm) to the intradermal test was determined to be the threshold concentration (Supplementary Table 1).

In the up-dosing phase, the initial dose was set as follows: 1/10 of the threshold concentration for patients with allergic BA, the threshold concentration for patients with AR, and 1/10 of the threshold concentration for patients with both AR and allergic BA. The initial dose (volume of 0.2–0.5 mL) was administered subcutaneously to upper arm or forearm. In addition, the investigators selected one of the following dose-titration schemes according to protocols that they employ in regular clinical practice: administering the study drug once or twice a week and increasing the dose by 50% or 100% for several months (conventional schedule for immunotherapy); rush protocol administering the drug several times a day and increasing the dose in an interval of 3–14 days, or cluster protocol administering the drug several times a day and increasing the dose in 1 or 2 days weekly (rush schedule for immunotherapy). The maximum tolerable dose in each patient was determined by the investigators on the basis of patient's sensitivity and local reactions against the preceding injection.

In the maintenance phase, the maximum tolerable dose was subcutaneously administered approximately once a month. After each injection, the patient's condition was monitored by the investigator for at least 30 min. When adverse events (AEs) occurred, the dose was reduced or interrupted. The treatment period of TO-204 was 52 weeks. For patients who were willing to continue the treatment, TO-204 was administered beyond 52 weeks. Changes in any medications (including those in dosage and administration) other than SCIT for AR and allergic BA were not allowed throughout the study.

Outcomes

The onset of AEs was regularly monitored during the study. For patients with AR, each nasal symptom (runny nose, blocked nose, sneezing, and itchy nose) was assessed on a 0–3 point scale from no symptoms to severe symptoms based on the European Medicines Agency guideline,²⁶ and the total nasal symptom score was calculated by adding up the scores for individual symptoms. For patients with allergic BA, the asthma control test²⁷ (or the childhood asthma control test²⁸ for patients aged 11 years or younger) was performed. Furthermore, quality of life (QOL) was assessed. In the assessment of QOL for patients with AR, Japanese Rhinconjunctivitis QOL questionnaire²⁰ was used. For patients aged 16 years or older with allergic BA, the asthma health questionnaire-Japan²⁹ was used. For patients aged 15 years or younger with allergic BA and their guardians, the QOL questionnaire for asthmatic children and their parents or caregivers ver. SF 2008 (Gifu)³⁰ was used. For patients with both AR and allergic BA, the investigator selected one of these as the primary disease and evaluated the efficacy of TO-204 in the primary disease. These efficacy outcomes were assessed at baseline and Week 12, 26, 38, and 52. If

patients continued the treatment, efficacy outcomes were also assessed at Week 78 and once after Week 78. In addition, the plasma concentrations of total IgE, specific IgE, and specific immunoglobulin G4 (IgG4) for *D. farinae* and *D. pteronyssinus* were measured as exploratory outcomes.

Statistical considerations

A sample size of 30 patients was determined on the basis of feasibility. AEs that occurred after the initiation of treatment were classified in accordance with the system organ class and preferred term as defined in the Medical Dictionary for Regulatory Activities Terminology/Japanese version 15.0, and the AEs possibly related to the study drug were categorized as adverse drug reactions (ADRs). The results of the efficacy outcomes were descriptively evaluated, and their values before treatment and at Week 12, 26, 38, 52, 78 and last observation visit were compared using the Wilcoxon signed-rank test as an exploratory analysis (Supplementary Table 2, 3). Additionally, the Dunnett test was performed as a post hoc analysis. All reported *P* values are two-sided with the significance level of 5%. All data were analyzed using SAS version 9.3 (SAS Institute, Cary, NC, USA).

Results

Trial profile

A flowchart of the study patients is shown in Figure 1. A total of 44 patients received the study drug and all patients were included in the analysis. Of these, 41 patients completed the 52-week treatment and 39 were willing to continue the treatment more than 52 weeks. In 34 patients, the last efficacy outcomes were obtained between Week 95 and 120, and 33 patients completed the extended treatment. Three patients discontinued the study before Week 52, including 1 who discontinued the study in the up-dosing phase. This patient was hospitalized because of AEs (intervertebral disc protrusion and osteoarthritis), which the investigator considered to be unrelated to the study drug. Five patients discontinued the study after Week 52. Of these, 1 patient discontinued the study because the investigator considered allergic BA was poorly controlled, which was possibly related to the study drug. Other reasons for discontinuation included consent withdraws and others. The duration of treatment ranged from 23 to 142 weeks.

The demographic and baseline characteristics of 44 patients who received the study drug are shown in Table 1. Patient's ages ranged from 5 to 53 years, including 18 patients (41%) aged 11 years or younger. The primary diseases were AR in 28 patients and allergic BA in 16 patients (note: 16 patients had both diseases). In the up-dosing phase, the conventional schedule was applied for 21 patients and the rush schedule was applied for 23 patients. Among the patients for whom the conventional schedule was applied, 6 patients (29%) were aged 11 years or younger and 15 patients (71%) were aged 12 years or older, including 2 patients aged 50 years or older (Supplementary Table 4). Among patients for whom the rush schedule was applied, 12 patients (52%) were aged 11 years or younger and 11 patients (48%) were aged 12 years or older; the patients aged 11 years or younger accounted for approximately half of these patients. The rush schedule was not used in any patients aged 30 years or older.

The number of patients by maintenance doses is shown in Table 2. The maintenance dose ranged from 0.2 to 2000 JAU. The median dose was 500 JAU in patients aged 11 years or younger, 200 JAU in patients aged 12 years or older, and 200 JAU in all patients. No maintenance dose accounted for a particularly large

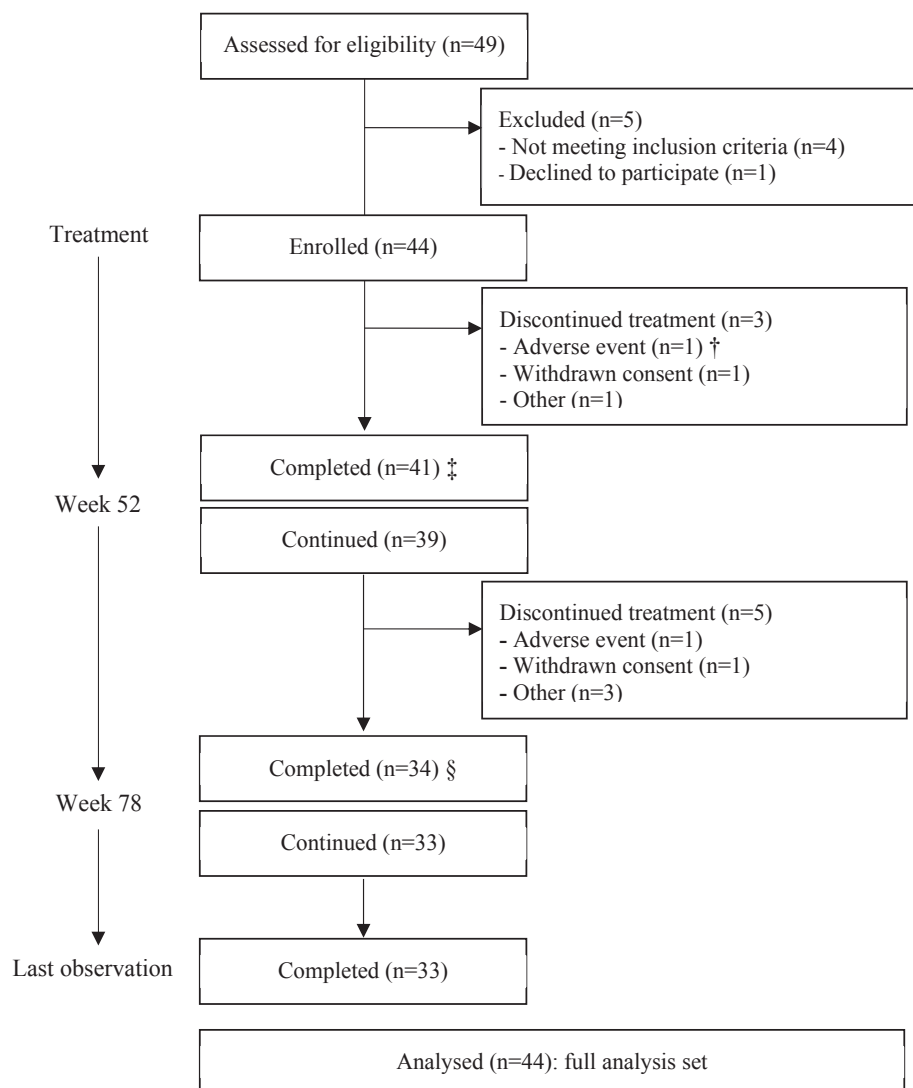


Fig. 1. Flowchart of the study patients. † One Patient discontinued the study in the up-dosing phase. ‡ Two patients were not willing to continue the treatment and completed at week 52. § One patient was not willing to continue the treatment and completed at week 104.

proportion. In addition, no correlations were found between the maintenance dose and the initial dose, age, or up-dosing method.

Safety

A total of 594 AEs occurred in 42 patients (95%) during the study period (Table 3). Serious AEs occurred in 7 patients (16%). Of these, 4 patients had AEs that were possibly related to the study drug. These AEs comprised urticaria, anaphylactic reaction, anaphylactic shock, and asthma. Serious AEs leading to treatment discontinuation were intervertebral disc protrusion and osteoarthritis that occurred in 1 patient.

The most common AE was nasopharyngitis, which occurred in 32 patients (73%). Other common AEs were influenza in 18 patients (41%) and headache in 16 patients (36%). The following number of patients had AEs in each treatment interval: 40 patients (91%) before Week 26, 31 (72%) between Week 27 and 52, 34 (87%) between Week 53 and 78, and 30 (88%) after 78 week. Most of the AEs occurred before Week 26. No AEs increased its incidence depending on the treatment duration.

A total of 151 ADRs occurred in 22 patients (50%). The incidence of ADRs per injection was 7.6% (151 events out of 1988 injections)

and most ADRs (143 events) were mild. The incidence was 50% in both patient groups aged 5–11 years and ≥ 12 years. The number of patients with ADRs by treatment phase was 21 patients (48%) during the up-dosing phase and 11 (26%) during the maintenance phase with more patients experiencing ADRs during the up-dosing phase (Supplementary Table 5). The most common ADRs were those related to the injection site (97 events) (Supplementary Table 6). Local reactions occurred in 14 patients (32%) and systemic reactions occurred in 17 patients (39%). The systemic reactions occurred most frequently in the skin (8 patients) and respiratory organs (8 patients). The number of patients with systemic reaction at the median maintenance dose was 8 patients at <200 JAU, and 13 patients at ≥ 200 JAU. The number of patients with ADRs by treatment duration was the following: 21 patients (48%) before Week 26; 5 (12%) between Week 27 and 52; 7 (18%) between Week 53 and 78; and 3 (9%) after Week 78.

The numbers of ADRs by treatment duration and dose are shown in Table 4. For both local and systemic reactions, ADRs occurred frequently at doses of less than 100 JAU before Week 26. A total of 5 systemic reactions occurred in 4 patients after Week 26 and no systemic reactions occurred after Week 78. Of these, 3 events were either anaphylactic reaction or anaphylactic shock,

Table 1
Demographic and baseline characteristics of the study patients.

	HDM AR (n = 28)	HDM BA (n = 16)	Total (n = 44)
Sex, n (%)			
Male	17 (61)	8 (50)	25 (57)
Female	11 (39)	8 (50)	19 (43)
Age (years), n (%)			
5 ≤ to < 12	12 (43)	6 (38)	18 (41)
12 ≤ to < 16	4 (14)	2 (13)	6 (14)
16 ≤	12 (43)	8 (50)	20 (45)
Age (years), Descriptive statistics			
Mean ± SD	20.0 ± 14.7	22.8 ± 15.4	21.0 ± 14.8
Median (minimum–maximum)	13.5 (5–53)	17.5 (7–53)	13.5 (5–53)
Complicating disease, n (%)			
HDM BA	4 (14)	–	–
HDM AR	–	12 (75)	–
Severity of allergic rhinitis, n (%)			
Mild	6 (21)	–	–
Moderate	3 (11)	–	–
Severe	13 (46)	–	–
Very severe	6 (21)	–	–
Severity of bronchial asthma (16 years ≤), n (%)		n = 8	
Mild intermittent	–	3 (38)	–
Mild persistent	–	1 (13)	–
Moderate persistent	–	4 (50)	–
Severity of bronchial asthma (5 ≤ to < 16 years), n (%)		n = 8	
Intermittent	–	5 (63)	–
Mild persistent	–	3 (38)	–
Moderate persistent	–	0 (0)	–
Schedule for immunotherapy, n (%)			
Conventional	13 (46)	8 (50)	21 (48)
Rush	15 (54)	8 (50)	23 (52)

HDM AR, house dust mite induced allergic rhinitis; HDM BA, house dust mite induced allergic bronchial asthma; SD, standard deviation.

Table 2
Number of the patients by maintenance dose of TO-204.

Maintenance dose	Age		Total (n = 44)
	5–11 years (n = 18)	12 years ≤ (n = 26)	
	n (%)	n (%)	n (%)
<100	2 (11)	7 (27) [†]	9 (20)
100 ≤ to < 200 JAU	2 (11)	5 (19) [†]	7 (16)
200 ≤ to < 300 JAU	3 (17)	4 (15)	7 (16)
300 ≤ to < 500 JAU	0 (0)	1 (4)	1 (2)
500 ≤ to < 1000 JAU	4 (22)	5 (19)	9 (20)
1000 JAU ≤	7 (39)	4 (15)	11 (25)
Median (minimum–maximum)	500 (2–2000)	200 (0.2–1000)	200 (0.2–2000)

[†] Including the final dose of 2 patients who discontinued the study in the up-dosing phase.

which occurred at doses of 500 JAU or higher. The frequencies of local reactions after Week 26 were less. The incidence of ADRs did not associate with the treatment duration or dose level.

ADRs which occurred within 30 min after injection accounted for 58.3% (88/151 events) of the total ADRs, 72% (70/97 events) of the local reactions, and 33% (18/54 events) of the systemic reactions (Supplementary Table 7). ADRs which occurred at 2 h or more after injection were mainly systemic reactions, and the most common reactions by system organ class was skin disorders (6 events), followed by gastrointestinal disorders (4 events).

Anaphylactic reaction possibly related to the study drug occurred in 4 patients (9%) and anaphylactic shock occurred in 2 patients (5%). The incidences of anaphylactic reaction, anaphylactic shock, and anaphylaxis as a percentage of the total number of

injections were 0.2% (4 events out of 1988 injections), 0.1% (2 events), and 0.3% (6 events), respectively. No near fatal anaphylaxis-related event occurred. Anaphylactic reaction and anaphylactic shock are listed in Table 5. The doses at onset were 500 JAU in 4 patients with anaphylactic reaction and 1000 JAU in 2 patients with anaphylactic shock. These events occurred in 3 patients within 15 min after injection, in 1 patient between >30 and 60 min, and in 2 patients between >60 and 120 min. The most immediate onset was observed at 3 min after injection.

The symptoms and signs of anaphylactic reaction and anaphylactic shock were classified by organ system based on the guidelines³¹ of the World Allergy Organization (WAO). As a result, skin, subcutaneous tissue, and mucosa occurred in 6 patients, respiratory in 5 patients, gastrointestinal in 4 patients, cardiovascular system in 1 patient, central nervous system in 2 patients. Initial treatments for anaphylaxis were promptly administered in accordance with the guidelines, and medications included epinephrine (adrenaline) intramuscular injections, intravenous or oral H1-antihistamines, glucocorticoids, and beta-2 adrenergic agonists given by nebulizer. All patients recovered within 1 day after the treatment, and no patients experienced a recurrence of anaphylaxis after injection. Study treatment was continued at a reduced dose that was subsequently increased again, and no patients discontinued the study because of anaphylaxis.

Efficacy

In patients with AR, the total nasal symptom score decreased by Week 26, and the score was maintained thereafter. The mean score at the last observation was lower by approximately 2.0 points than the baseline value ($P = 0.016$) (Fig. 2A). The QOL scores at the last observation decreased from the baseline in all domains of daily life, outdoor activities, social life, sleep, the body, and spiritual life (Supplementary Table 3, 8). Nasal symptoms were also relieved (Supplementary Table 2).

In patients with allergic BA, the scores for the asthma control tests were maintained in 20 points or higher (well-controlled) throughout the treatment period in most patients aged ≥ 12 years and 5–11 years (Fig. 2B, C). The QOL scores remained unchanged during the treatment period in all domains (data not shown); in patients aged ≥ 12 years, the domains included asthmatic symptoms, emotion, daily activity, factors which worsened symptoms, social activity, and economic; and in patients aged 5–11 years old, the domains included mental burden, cause of attacks, instability of asthmatic symptoms, acceptance of asthma, and exercise tolerance. In the pulmonary function test, the mean value for percentage predicted forced expiratory volume in 1 s unchanged after the treatment (Supplementary Table 9).

The plasma concentrations of specific IgE and IgG4 during the study are shown in Figure 3A, B, respectively. No significant changes were found in the IgE levels, whereas the IgG4 levels kept increasing from baseline throughout the treatment period with the highest level obtained at the last observation ($P < 0.001$).

Discussion

In this study, most patients completed the 52-week study treatment, and TO-204 showed acceptable safety. The median maintenance dose was 200 JAU, which was similar to the typical maintenance dose seen with the conventional schedule for immunotherapy (administration protocol with dose increase by 100%) described in the guidance published by the JSA.³² In our study, each investigator aimed to achieve the highest dose but, to our surprise, resulting maintenance dose was far lower than the dose recommended in US at 500–2000 AU,⁴ or 5000–20,000 JAU.²⁵

Table 3
Summary of safety.

	Total (n = 44)		Week 1–26 (n = 44)		Week 27–52 (n = 43)		Week 53–78 (n = 39)		Week 79 ≤ (n = 34)	
	AEs	Patients (%)	AEs	Patients (%)	AEs	Patients (%)	AEs	Patients (%)	AEs	Patients (%)
AE	594	42 (95)	261	40 (91)	79	31 (72)	118	34 (87)	136	30 (88)
Serious AE	9	7 (16)	2	2 (5)	3	3 (7)	3	2 (5)	1	1 (3)
ADR	151	22 (50)	104	21 (48)	14	5 (12)	20	7 (18)	13	3 (9)
Serious ADR	5	4 (9)	1	1 (2)	1	1 (2)	3	2 (5)	0	0 (0)
Significant AEs										
Anaphylactic reaction	4	4 (9)	3	3 (7)	1	1 (2)	0	0 (0)	0	0 (0)
Anaphylactic shock	2	2 (5)	0	0 (0)	0	0 (0)	2	2 (5)	0	0 (0)
AEs leading to discontinuation	2	1 (2)	1	1 (2)	1	1 (2)	0	0 (0)	0	0 (0)

AE, adverse event; ADR, adverse drug reaction; AEs, Number of adverse events; Patients, Number of patients with adverse events or adverse drug reactions; %, Percentage of patients with adverse events or adverse drug reactions.

Table 4
Number of adverse drug reactions by treatment duration and dose at onset.

		Number of patients		Week 1–26 (n = 44)		Week 27–52 (n = 43)		Week 53–78 (n = 39)		Week 79 < (n = 34)		Total (n = 44)	
		Number of injections		1061		313		248		366		1988	
				ADRs	Patients	ADRs	Patients	ADRs	Patients	ADRs	Patients	ADRs	Patients
Total				104	21	14	5	20	7	13	3	151	22
Local reaction	Total			55	13	13	4	16	5	13	3	97	14
	<100 JAU			26	11	4	3	0	0	1	1	31	11
	100 ≤ to < 500 JAU			10	4	1	1	10	3	2	2	23	8
	500 ≤ to < 1000 JAU			6	3	6	1	5	1	10	1	27	3
Systemic reaction	1000 ≤			13	3	2	1	1	1	0	0	16	5
	Total			49	15	1	1	4	3	0	0	54	17
	<100 JAU			25	6	0	0	0	0	0	0	25	6
	100 ≤ to < 500 JAU			2	1	0	0	2	2	0	0	4	3
	500 ≤ to < 1000 JAU			6	4 [†]	1	1 [‡]	0	0	0	0	7	5
	1000 ≤			16	5	0	0	2	2 [§]	0	0	18	7

ADR, adverse drug reaction; ADRs, Number of adverse drug reactions; Patients, Number of patients with adverse drug reactions.

[†] Anaphylactic reaction (n = 3).

[‡] Anaphylactic reaction (n = 1).

[§] Anaphylactic shock (n = 2).

Table 5
Summary of anaphylactic reaction and anaphylactic shock.

Age [†] /Sex	Adverse drug reaction	Method of up-dosing	Onset			Medication	Outcome
			Day (Phase)	Dose at onset	Time from last injection		
13/Male	Anaphylactic reaction	Rush	16 (up-dosing)	500 JAU (10,000 JAU/mL, 0.05 mL)	75 min	Epinephrine (Nebulizer), Glucocorticoid (Nebulizer, Oral), Others	Recovered
14/Male	Anaphylactic reaction	Rush	16 (up-dosing)	500 JAU (10,000 JAU/mL, 0.05 mL)	75 min	Epinephrine (Nebulizer), Glucocorticoid (Nebulizer, IV, Oral), H1-antihistamine (Oral), Others	Recovered
25/Male	Anaphylactic reaction	Cluster	61 (up-dosing)	500 JAU (10,000 JAU/mL, 0.05 mL)	60 min	Epinephrine (IM), Glucocorticoid (IV), Beta-2 adrenergic agonist (Nebulizer) Others	Recovered
28/Female	Anaphylactic reaction	Rush	241 (maintenance)	500 JAU (10,000 JAU/mL, 0.05 mL)	14 min	Epinephrine (IM, IV), Glucocorticoid (IV), H1-antihistamine (IV), Others	Recovered
46/Male	Anaphylactic shock	50% Conventional	440 (maintenance)	1000 JAU (10,000 JAU/mL, 0.1 mL)	4 min	Epinephrine (IM), Glucocorticoid (IV, Oral), H1-antihistamine (Oral), Beta-2 adrenergic agonist (Nebulizer)	Recovered
31/Female	Anaphylactic shock	50% Conventional	400 (up-dosing)	1000 JAU (10,000 JAU/mL, 0.1 mL)	3 min	Epinephrine (IM), Glucocorticoid (IV, Oral), H1-antihistamine (Oral)	Recovered

Others include Agents affecting digestive organs (Oral, IV), neophyllin (IV), leukotriene receptor antagonist (Oral), hydroxyzine hydrochloride (IV), analgesics, anti-itchings, astringents and anti-inflammatory agents (ointment, lotion).

IM, intramuscular injection; IV, intravenous infusion.

[†] Patient's age (years) when she/he gave written informed consent.

It should be emphasized that tolerable allergen dose in subcutaneous immunotherapy may be different among various nations and that this is the first study to identify the safe dose for Japanese patients.

At the dose of TO-204 in this study, ADRs occurred in similar proportion of patients irrespective of their ages with 50% of both

patients aged ≤11 years and ≥12 years having ADRs. Frequently reported ADRs were injection site-related events, events affecting the skin and respiratory organs, and anaphylaxis (anaphylactic reaction and anaphylactic shock). WAO has recommended a classification and grading system for SCIT-induced systemic reactions.³³ In this system, a systemic reaction is defined as an adverse reaction

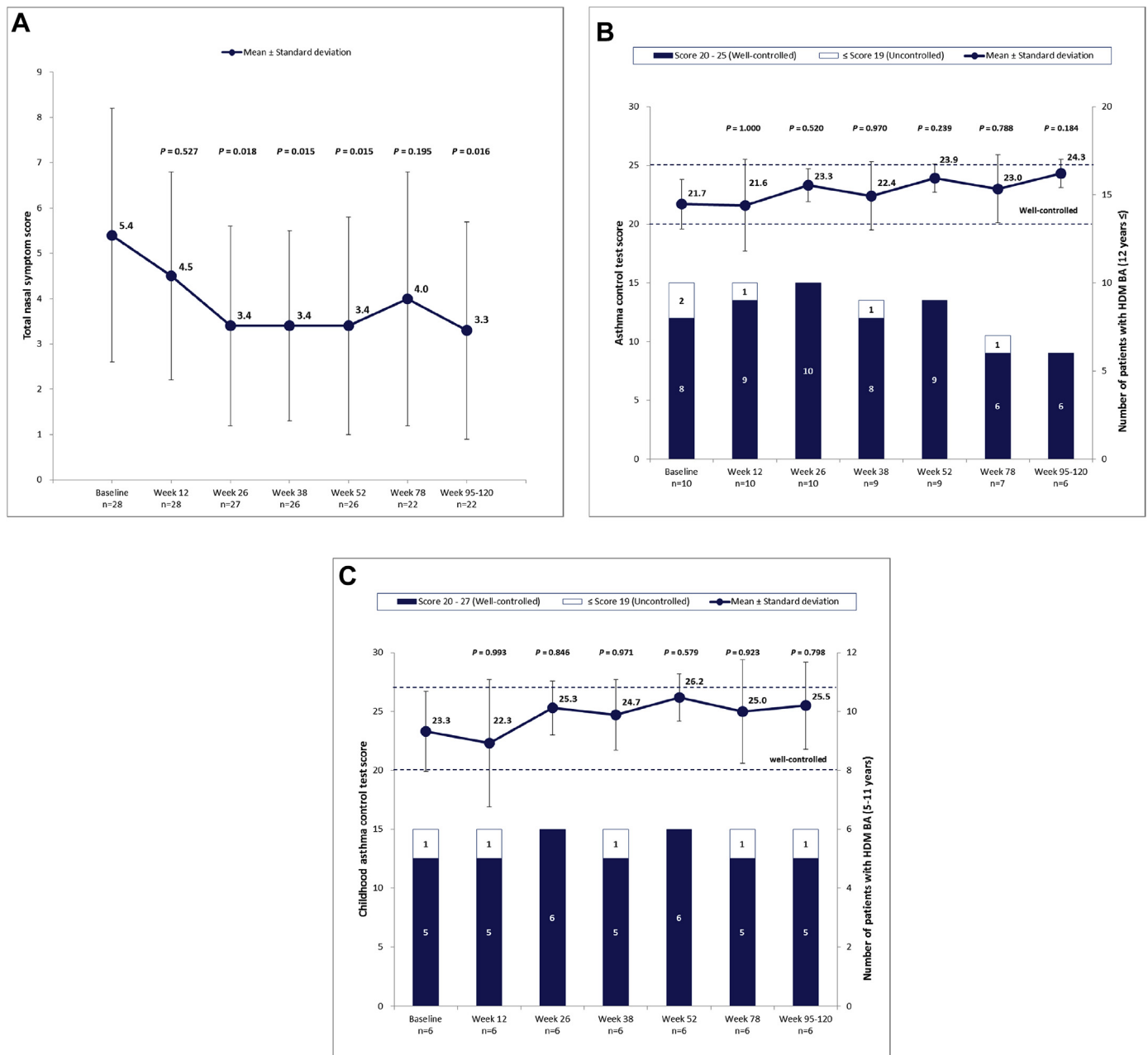


Fig. 2. Total nasal symptom score and Asthma control test. **(A)** Total nasal symptom score (Sum of watery discharge, blocked nose, sneezing, and itchy nose scores) in patients with HDM AR over the study. Data points (closed circles) are mean and vertical bars are standard deviation in total nasal symptom score. *P* values mean the results of Dunnett test (vs baseline). **(B)** Asthma control test score in patients with HDM BA (≥ 12 years) over the study. Data points (closed circles) are mean and vertical bars are standard deviation in asthma control test score. All points have no statistical difference (Dunnett test vs baseline). **(C)** Childhood asthma control test score in patients with HDM BA (5–11 years) over the study. Data points (closed circles) are mean and vertical bars are standard deviation in childhood asthma control test score. All points have no statistical difference (Dunnett test vs baseline). HDM AR, house dust mite induced allergic rhinitis; HDM BA, house dust mite induced allergic bronchial asthma.

that occurs in an organ system distant from the injection site. The organ systems are classified into the skin, subcutaneous tissue and mucosa, respiratory, gastrointestinal, cardiovascular system, central nervous system, and others. In accordance with this system, we classified ADRs occurring at the injection sites as local reactions and the other ADRs as systemic reactions. As a result, we found that both local and systemic reactions occurred frequently during the up-dosing phase in the early stage of treatment and the incidence subsequently decreased irrespective of the dose levels. This result was consistent with the previous results.^{33–36}

Both local and systemic reactions occurred frequently within 30 min after injection with TO-204. ADRs occurring after more than 30 min were mainly systemic reactions. Several studies have reported that SCIT-induced ADRs mostly occur within 30 min after injection,^{34,35} and our results are consistent with these reports. Furthermore, the AEs occurring within 30 min have been reported to be generally more severe, and those occurring within a few minutes may be a sign of severe anaphylaxis.^{4,33,36} In contrast, other studies have reported that delayed systemic reactions and anaphylaxis occurred beyond 30 min after injection,^{36–38} and 3

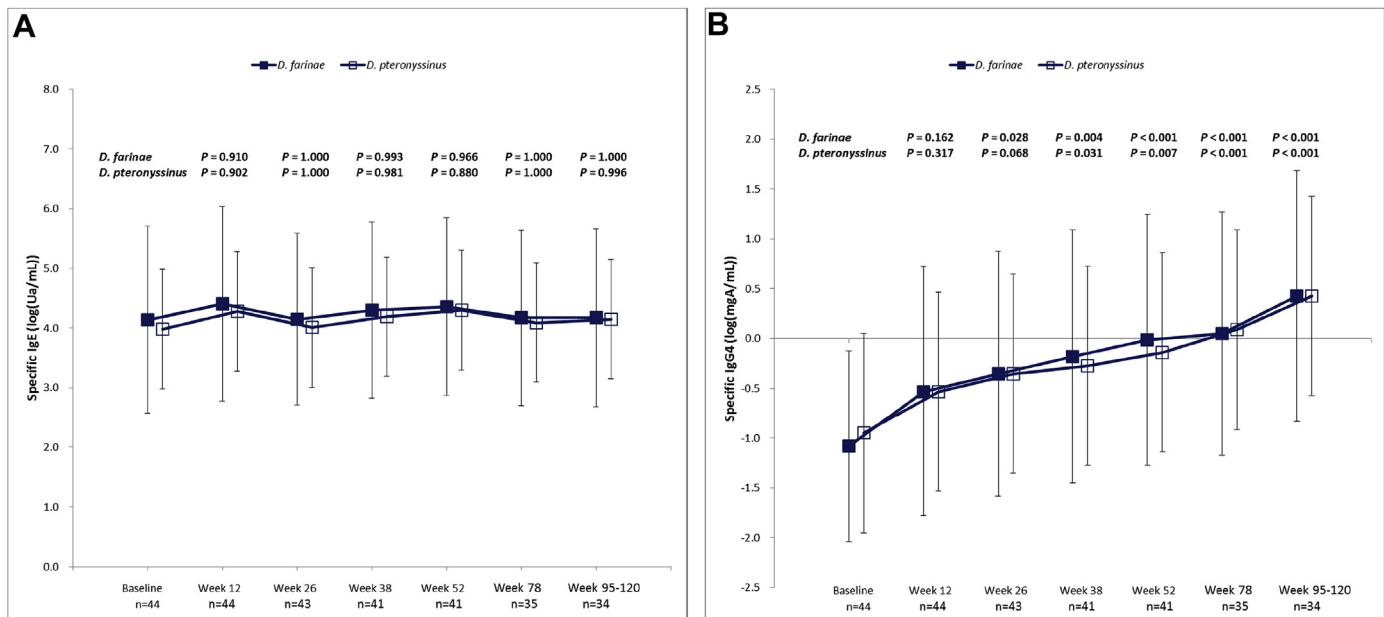


Fig. 3. Plasma concentrations of house dust mite specific IgE and IgG4 during the study. (A) HDM-Specific IgE concentrations against *D. farinae* and *D. pteronyssinus*. Data were log-transformed. Data points (opened squares and closed squares) are mean and vertical bars are standard deviation HDM-Specific IgE. All points have no statistical difference (Dunnett test vs baseline). (B) HDM-Specific IgG4 concentrations against *D. farinae* and *D. pteronyssinus*. Data were log-transformed. Data points (opened squares and closed squares) are mean and vertical bars are standard deviation in HDM-Specific IgG4. P values mean the results of Dunnett test (vs baseline). HDM, house dust mite; IgE, Immunoglobulin E; IgG4, Immunoglobulin G4.

patients had anaphylaxis after more than 30 min in our study. Considering these, while we have to observe any signs of anaphylaxis in the first 30 min after injection, careful monitoring should be continued thereafter. Action plan for patients to manage late anaphylaxis should be implemented.^{4,39}

Anaphylaxis occurred in 6 patients and the incidence per injection was 0.3% (6 events out of 1988 injections). WAO summarized clinical studies for SCIT and reported that the incidence of systemic reactions per injection in conventional schedules was approximately 0.2% (range: 0.026%–0.37% in the USA, and 0.01%–0.3% in Europe).³³ Our results were comparable with this report, although the grading system we employed was different. Of these 6 patients, the dose of 4 patients who had anaphylactic reaction was 500 JAU and the dose of 2 patients with anaphylactic shock was 1000 JAU. Although the safety review for SCIT has concluded that it is difficult to compare the results between the different studies,⁴⁰ we consider that TO-204 can be administered safely at doses of less than 500 JAU by comparing the incidence of anaphylaxis in our study with the reports on other serious AEs. However, anaphylaxis occurred in 3 patients before Week 26, in 1 patient between Week 26 and 52, and in 2 patients after Week 52, which indicates no relationship between the incidence of anaphylaxis and the treatment duration. Therefore, it is necessary to pay attention to the onset of anaphylaxis while treating patients with TO-204.

This study was conducted at the selected institutions where expert physicians with enough knowledge about SCIT-induced ADRs could treat patients immediately after the onset of anaphylaxis. Furthermore, the investigators carefully observed the condition of patients before injection and monitored their clinical courses for at least 30 min after injection. In case anaphylaxis occurs after the monitoring period of 30 min, an action plan^{4,39} for unexpected anaphylaxis for the patients had been implemented and 3 patients who developed late anaphylaxis were able to seek appropriate treatment. Because the patients who experienced anaphylaxis recovered after the treatment recommended by WAO, they were not withdrawn from the study. Therefore, we consider

that SCIT with TO-204 is a safe and sustainable therapy for a long term if patients are treated by the physicians who can manage for anaphylaxis. It is important to treat patients with allergic diseases at the early disease stage, and the current guidelines for AIT^{1,41–43} including that published by the JSA³² recommend SCIT for children aged 5 years or older. Because this study included patients aged 5 years or older, we consider that TO-204 can be administered to children in this age group as long as they are monitored by the qualified physicians.

In the efficacy analysis, the total nasal symptom score decreased and the individual symptoms were well controlled in patients with AR. In patients with allergic BA, the scores of the asthma control test and the childhood asthma control test were maintained and the symptoms of most patients were well controlled throughout the treatment period. A systematic review of the efficacy of HDM SCIT and a meta-analysis of randomized controlled trials has shown the improvements in symptom scores for AR and allergic BA, medication scores, and QOL.^{12,44,45} Although this was a single-arm study without control group, the change in AR symptoms was similar to those reported in previous studies and BA symptoms were well controlled.

It has been reported that while the efficacy of SCIT increases in a dose-dependent manner, the risk of anaphylaxis also increases according to the dose.^{45–47} Therefore, when using TO-204, we should consider its benefits and risks to each patient, and individually determine the dose level according to the indicated potency. Even these days, when sublingual immunotherapy drugs have been launched for the treatment of allergic disease,^{48,49} standardized allergen extracts for SCIT play an important role in the treatment of AR and allergic BA. The potency of TO-204 is approximately 100-fold stronger than that of the unstandardized “house dust extract preparation (its potency is equivalent to 1000 JAU/mL)” that has been used in Japan. Therefore, when the house dust extract preparation is to be replaced by TO-204, the dose of TO-204 should be determined on the basis of this potency rate and should be adjusted according to the presence/absence of allergic reaction.

Our study has some limitations. First, because the study subjects included children, we could not administer placebo to these patients over a long period. As a result, we adopted a single-arm, open label design, which led to the limitation in efficacy evaluation. Second, although AIT is recommended to continue for 3–5 years,^{1,4} the treatment period was approximately 3 years at the longest. However, there was no relationship between the treatment duration and the incidence of ADRs in the 3-year follow-up. Third, because there is no established up-dosing protocol for SCIT, the dose of TO-204 was determined by the investigators without unified up-dosing criteria.

Despite these limitations, we consider that the safety profile of TO-204 was well confirmed because the study was conducted for approximately 3 years in practical clinical setting. Unlike pharmacotherapy, AIT is expected to have disease-modifying effects on allergic diseases, and to change the natural course of allergic diseases. Furthermore, the clinical effect may be maintained for several years after the treatment discontinuation.^{1,41,43} Further studies are necessary to clarify the risks and benefits of TO-204 over a longer period. It is also needed to explore the patient groups in whom the efficacy of TO-204 is maximized and to establish the risk management program in different age groups.

In conclusion, safety profile of TO-204 was acceptable in Japanese patients aged 5 years or older who had HDM-induced AR or allergic BA at the main dose of 200 JAU for a long period, which was lower than recommended dose in the US. However, higher dose should be administered carefully because the risk of anaphylaxis increased at doses of 500 or 1000 JAU.

Acknowledgments

We thank all the patients who participated in this study and their family members as well as all investigators and site staff. This trial was sponsored by Torii Pharmaceutical Co., Ltd, whose representatives were involved in data collection (along with the investigators) and in the interpretation and analysis of the data and writing this report to the study protocol. Representatives of company were involved in the design of the study, with scientific advice obtained from external experts on allergic disease. Editing assistance was provided by Kenichi Hayashi (Alamedic Co., Ltd., Tokyo, Japan). This assistance was funded by Torii Pharmaceutical Co., Ltd.

Appendix A. Supplementary data

Supplementary data related to this article can be found at <https://doi.org/10.1016/j.alit.2017.11.004>.

Conflict of interest

TF received lecture fee from GlaxoSmithKline, Maruho, MSD, and writing fee from AstraZeneca. KM received lecture and writing fee from Torii. KO received lecture fee from Hisamitsu, Kyorin, Meiji Seika Pharma, Mitsubishi Tanabe, Taiho, Torii and writing fee from Torii. MO received lecture fee from Sanofi, Shionogi. TK received lecture fee from Kyorin. MN received lecture fee from AstraZeneca, Kyorin, MSD, Torii. MT received lecture fee from AstraZeneca, Kyorin. AT received lecture fee from Torii. KN, NA and HOZ are employees of Torii. The rest of authors have no conflict of interest.

Authors' contributions

TF, TS, KM, KO, KH, MO, TK, AU, YK, HOD, KK, MN, MT, ST, SD, TM, SH, principal investigator, contributed to the conduct of the study and data collection and interpretation.

TF wrote the manuscript. AT, who is a study safety adviser, gave advice with study design and interpretation of the data. HOZ and NA designed and operated the study. KN was responsible for editorial, and journal submission assistance for this manuscript. All the listed authors were involved in the review and approved the final content.

References

- Bousquet J, Lockey R, Malling HJ. Allergen immunotherapy: therapeutic vaccines for allergic diseases. A WHO position paper. *J Allergy Clin Immunol* 1998;**102**:558–62.
- Burks AW, Calderon MA, Casale T, Cox L, Demoly P, Jutel M, et al. Update on allergy immunotherapy: American Academy of Allergy, Asthma & Immunology/European Academy of Allergy and Clinical Immunology/PRACTALL consensus report. *J Allergy Clin Immunol* 2013;**131**:1288–96. e3.
- Jutel M, Agache I, Bonini S, Burks AW, Calderon M, Canonica W, et al. International consensus on allergen immunotherapy II: mechanisms, standardization, and pharmacoeconomics. *J Allergy Clin Immunol* 2016;**137**:358–68.
- Cox L, Nelson H, Lockey R, Calabria C, Chacko T, Finegold I, et al. Allergen immunotherapy: a practice parameter third update. *J Allergy Clin Immunol* 2011;**127**:S1–55.
- Jutel M, Agache I, Bonini S, Burks AW, Calderon M, Canonica W, et al. International consensus on allergen immunotherapy. *J Allergy Clin Immunol* 2015;**136**:556–68.
- Des Roches A, Paradis L, Menardo JL, Bouges S, Daures JP, Bousquet J. Immunotherapy with a standardized Dermatophagoides pteronyssinus extract. VI. Specific immunotherapy prevents the onset of new sensitizations in children. *J Allergy Clin Immunol* 1997;**99**:450–3.
- Jacobsen L, Niggemann B, Dreborg S, Ferdousi HA, Halken S, Host A, et al. Specific immunotherapy has long-term preventive effect of seasonal and perennial asthma: 10-year follow-up on the PAT study. *Allergy* 2007;**62**:943–8.
- Marogna M, Spadolini I, Massolo A, Canonica GW, Passalacqua G. Long-lasting effects of sublingual immunotherapy according to its duration: a 15-year prospective study. *J Allergy Clin Immunol* 2010;**126**:969–75.
- Niggemann B, Jacobsen L, Dreborg S, Ferdousi HA, Halken S, Host A, et al. Five-year follow-up on the PAT study: specific immunotherapy and long-term prevention of asthma in children. *Allergy* 2006;**61**:855–9.
- Nagata M, Tanaka K. [An evaluation of the long-term effect of clustered immunotherapy in house-dust-mite-sensitive adult bronchial asthma]. *Arerugi* 2001;**50**:435–9 (in Japanese).
- Tanaka Y, Okafuji I, Narabayashi S, Tsuruta S. [Safety and efficacy of one-year rush subcutaneous immunotherapy in Japanese children, using house dust extract]. *Arerugi* 2015;**64**:1160–8 (in Japanese).
- Abramson MJ, Puy RM, Weiner JM. Injection allergen immunotherapy for asthma. *Cochrane Database Syst Rev* 2010;**8**: CD001186.
- Blumberg G, Groes L, Dahl R. SQ-standardized house dust mite immunotherapy as an immunomodulatory treatment in patients with asthma. *Allergy* 2011;**66**:178–85.
- Pajno GB, Barberio G, De Luca F, Morabito L, Parmiani S. Prevention of new sensitizations in asthmatic children monosensitized to house dust mite by specific immunotherapy. A six-year follow-up study. *Clin Exp Allergy* 2001;**31**:1392–7.
- Varney VA, Tabbah K, Mavroleon G, Frew AJ. Usefulness of specific immunotherapy in patients with severe perennial allergic rhinitis induced by house dust mite: a double-blind, randomized, placebo-controlled trial. *Clin Exp Allergy* 2003;**33**:1076–82.
- Dhami S, Kakourou A, Asamoah F, Agache I, Lau S, Jutel M, et al. Allergen immunotherapy for allergic asthma: a systematic review and meta-analysis. *Allergy* 2017;**72**:1825–48.
- Dhami S, Nurmatov U, Arasi S, Khan T, Asaria M, Zaman H, et al. Allergen immunotherapy for allergic rhinoconjunctivitis: a systematic review and meta-analysis. *Allergy* 2017;**72**:1597–631.
- Erekosima N, Suarez-Cuervo C, Ramanathan M, Kim JM, Chelladurai Y, Segal JB, et al. Effectiveness of subcutaneous immunotherapy for allergic rhinoconjunctivitis and asthma: a systematic review. *Laryngoscope* 2014;**124**:616–27.
- Zuberbier T, Bachert C, Bousquet PJ, Passalacqua G, Walter Canonica G, Merk H, et al. GA(2) LEN/EAACI pocket guide for allergen-specific immunotherapy for allergic rhinitis and asthma. *Allergy* 2010;**65**:1525–30.
- Okubo K, Kurono Y, Fujieda S, Ogino S, Uchio E, Odajima H, et al. Japanese guideline for allergic rhinitis. *Allergol Int* 2011;**60**:171–89.
- Hamasaki Y, Kohno Y, Ebisawa M, Kondo N, Nishima S, Nishimura T, et al. Japanese guideline for childhood asthma 2014. *Allergol Int* 2014;**63**:335–6.
- Ohta K, Ichinose M, Nagase H, Yamaguchi M, Sugiura H, Tohda Y, et al. Japanese guideline for adult asthma 2014. *Allergol Int* 2014;**63**:293–333.
- Stelmach I, Sobocinska A, Majak P, Smejda K, Jerzynska J, Stelmach W. Comparison of the long-term efficacy of 3- and 5-year house dust mite allergen immunotherapy. *Ann Allergy Asthma Immunol* 2012;**109**:274–8.
- Marogna M, Tomassetti D, Bernasconi A, Colombo F, Massolo A, Businco AD, et al. Preventive effects of sublingual immunotherapy in childhood: an open randomized controlled study. *Ann Allergy Asthma Immunol* 2008;**101**:206–11.
- Takai T, Okamoto Y, Okubo K, Nagata M, Sakaguchi M, Fukutomi Y, et al. Japanese Society of Allergy task force report on standardization of house dust mite allergen vaccines – secondary publication. *Allergol Int* 2015;**64**:181–6.
- European Medicines Agency. Guideline on the Clinical Development of Products for Specific Immunotherapy for the Treatment of Allergic Diseases 2008. Available at: http://www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2009/09/WC500003605.pdf

27. Nathan RA, Sorkness CA, Kosinski M, Schatz M, Li JT, Marcus P, et al. Development of the asthma control test: a survey for assessing asthma control. *J Allergy Clin Immunol* 2004;**113**:59–65.
28. Liu AH, Zeiger R, Sorkness C, Mahr T, Ostrom N, Burgess S, et al. Development and cross-sectional validation of the childhood asthma control test. *J Allergy Clin Immunol* 2007;**119**:817–25.
29. Arioka H. [Bronchial asthma and QOL]. *Nihon Naika Gakkai Zasshi [Jpn Soc Int Med]* 2006;**95**:1507–13 (in Japanese).
30. Kondo N, Hirayama K, Matsui E, Teramoto T, Kaneko H, Fukao T, et al. [QOL questionnaire for pediatric patients with bronchial asthma and their parents or caregivers. Preparation and evaluation of the short form version 2008 (Gifu)]. *Arerugi* 2008;**57**:1022–33 (in Japanese).
31. Simons FE, Arduoso LR, Bilo MB, El-Gamal YM, Ledford DK, Ring J, et al. World allergy organization guidelines for the assessment and management of anaphylaxis. *World Allergy Organ J* 2011;**4**:13–37.
32. [Guidance of Allergen Immunotherapy for Mite Allergy]. Tokyo: Japanese Society of Allergology; 2015 (in Japanese). Available at: http://www.jsaweb.jp/modules/journal/index.php?content_id=4. [Accessed 20 September 2017].
33. Cox L, Larenas-Linnemann D, Lockey RF, Passalacqua G. Speaking the same language: the World Allergy Organization Subcutaneous Immunotherapy Systemic Reaction Grading System. *J Allergy Clin Immunol* 2010;**125**:569–74. e7.
34. DaVeiga SP, Liu X, Caruso K, Golubski S, Xu M, Lang DM. Systemic reactions associated with subcutaneous allergen immunotherapy: timing and risk assessment. *Ann Allergy Asthma Immunol* 2011;**106**:533–7. e2.
35. Epstein TG, Liss GM, Murphy-Berendts K, Bernstein DI. AAAAI/ACAAI surveillance study of subcutaneous immunotherapy, years 2008–2012: an update on fatal and nonfatal systemic allergic reactions. *J Allergy Clin Immunol Pract* 2014;**2**:161–7.
36. Epstein TG, Liss GM, Murphy-Berendts K, Bernstein DI. Immediate and delayed-onset systemic reactions after subcutaneous immunotherapy injections: AAAAI/AAAAI surveillance study of subcutaneous immunotherapy: year 2. *Ann Allergy Asthma Immunol* 2011;**107**:426–31. e1.
37. Pfaar O, Nell MJ, Boot JD, Versteeg SA, van Ree R, Roger A, et al. A randomized, 5-arm dose finding study with a mite allergoid SCIT in allergic rhinoconjunctivitis patients. *Allergy* 2016;**71**:967–76.
38. Rieker-Schwienbacher J, Nell MJ, Diamant Z, van Ree R, Distler A, Boot JD, et al. Open-label parallel dose tolerability study of three subcutaneous immunotherapy regimens in house dust mite allergic patients. *Clin Transl Allergy* 2013;**3**:16.
39. Lieberman P. The risk and management of anaphylaxis in the setting of immunotherapy. *Am J Rhinol Allergy* 2012;**26**:469–74.
40. Caminati M, Dama AR, Djuric I, Montagni M, Schiappoli M, Ridolo E, et al. Incidence and risk factors for subcutaneous immunotherapy anaphylaxis: the optimization of safety. *Expert Rev Clin Immunol* 2015;**11**:233–45.
41. Brozek JL, Bousquet J, Baena-Cagnani CE, Bonini S, Canonica GW, Casale TB, et al. Allergic rhinitis and its impact on asthma (ARIA) guidelines: 2010 revision. *J Allergy Clin Immunol* 2010;**126**:466–76.
42. Okamoto Y, Ohta N, Okano M, Kamijo A, Gotoh M, Suzuki M, et al. Guiding principles of subcutaneous immunotherapy for allergic rhinitis in Japan. *Auris Nasus Larynx* 2014;**41**:1–5.
43. U.S. Department of Health and Human Services. National asthma education and prevention program. Expert Panel Report 3 (EPR-3): guidelines for the diagnosis and management of asthma-Summary report 2007. *J Allergy Clin Immunol* 2007;**120**:S94–138.
44. Calderon MA, Alves B, Jacobson M, Hurwitz B, Sheikh A, Durham S. Allergen injection immunotherapy for seasonal allergic rhinitis. *Cochrane Database Syst Rev* 2007;**1**: CD001936.
45. Calderon MA, Casale TB, Togias A, Bousquet J, Durham SR, Demoly P. Allergen-specific immunotherapy for respiratory allergies: from meta-analysis to registration and beyond. *J Allergy Clin Immunol* 2011;**127**:30–8.
46. Frew AJ, Powell RJ, Corrigan CJ, Durham SR. Efficacy and safety of specific immunotherapy with SQ allergen extract in treatment-resistant seasonal allergic rhinoconjunctivitis. *J Allergy Clin Immunol* 2006;**117**:319–25.
47. Calderon MA, Casale TB, Nelson HS, Demoly P. An evidence-based analysis of house dust mite allergen immunotherapy: a call for more rigorous clinical studies. *J Allergy Clin Immunol* 2013;**132**:1322–36.
48. Okamoto Y, Okubo K, Yonekura S, Hashiguchi K, Goto M, Otsuka T, et al. Efficacy and safety of sublingual immunotherapy for two seasons in patients with Japanese cedar pollinosis. *Int Arch Allergy Immunol* 2015;**166**:177–88.
49. Okubo K, Masuyama K, Imai T, Okamiya K, Stage BS, Seitzberg D, et al. Efficacy and safety of the SQ house dust mite sublingual immunotherapy tablet in Japanese adults and adolescents with house dust mite-induced allergic rhinitis. *J Allergy Clin Immunol* 2017;**139**:1840–8. e10.

Supplementary Tables

Supplementary Table 1

Summary of intradermal test and treatments administered.

Intradermal test†	Prior to the initiation of treatment, an intradermal test was performed with TO-204 to select the initial dose. The study drug was diluted immediately before use with 10-fold serial dilution and the intradermal test was performed in the order of low to higher concentrations. The dose volume was 0.02 mL and administered intradermally. The test results were determined as being positive (erythema [red flare], diameter ≥ 20 mm or wheal diameter ≥ 9 mm) or not between 15 and 30 minutes after administration, and the minimum concentration inducing a positive response was set as the threshold concentration.
Up-dosing phase‡	[Dose] The initial dose was subcutaneously administered at between 0.02 and 0.05 mL. In the case of the conventional schedule for immunotherapy, the next and subsequent doses were increased by 50% or 100%. When the dose volume reached 0.5 mL, the concentration of next dose was 10-fold higher as a rough indication. The dose was increased up to the maximum tolerable dose while paying careful attention to safety. In either of the administration protocols, taking the degree of sensitization and any local reaction at the previous injection into consideration, the maximum tolerable dose was determined by the investigator. [Number of doses] In the conventional schedule for immunotherapy, the drug was administered twice a week to once every 2 weeks as a rough indication. In the rush protocol or cluster protocol, the drug was administered once to 5 times a day as a rough indication.
Maintenance phase‡	[Dose] The dose volume was between 0.1 and 0.5 mL as a rough indication. The dose during the maintenance phase (maintenance dose) was the maximum tolerable dose. [Number of doses] The drug was administered once a month as a rough indication.
Cautions at the time of treatment	• If the patient was suffering from a fever or the common cold on the date of treatment, treatment was postponed. • During the up-dosing phase and maintenance phase, the investigator or sub-investigator kept the patient under observation for at least 30 minutes after injection. • During the up-dosing phase and maintenance phase, when the drug could not be routinely administered because of the onset of AEs, the condition of the patient, possible allergic reactions that occurred in connection with previous injections or a prolonged the time interval from the last injection, dose reduction or treatment interruption was considered. • When the lot of study drug was changed, the dose was reduced to between 50% and 75% of the previous dose and then increased again.

†TO-204 was diluted with a diluent ("Control solution TORII for diagnostic allergen intradermal extract", Torii Pharmaceutical Co., Ltd., Tokyo, Japan) for the intradermal test.

‡ TO-204 was diluted with a diluent ("Diluent TORII for immunotherapeutic allergen extract ", Torii Pharmaceutical Co., Ltd., Tokyo, Japan) for SCIT.

Supplementary Table 2

Nasal findings in HDM AR during the course of the study.

		Baseline (n = 28)		Week 12 (n = 28)		Week 26 (n = 27)		Week 38 (n = 26)		Week 52 (n = 26)		Week 78 (n = 22)		Week 95-120 (n = 22)	
		Patients	(%)	Patients	(%)	Patients	(%)	Patients	(%)	Patients	(%)	Patients	(%)	Patients	(%)
Swelling of the inferior nasal concha mucosa	—	1	(4)	3	(11)	0	(0)	3	(12)	4	(15)	5	(23)	4	(18)
	1+	9	(32)	13	(46)	17	(63)	16	(62)	16	(62)	10	(45)	13	(59)
	2+	15	(54)	9	(32)	9	(33)	6	(23)	6	(23)	7	(32)	5	(23)
	3+	3	(11)	3	(11)	1	(4)	1	(4)	0	(0)	0	(0)	0	(0)
	<i>P</i> value	—		0.186		0.112		0.009		0.002		0.021		0.004	
Color of the inferior nasal concha mucosa	—	1	(4)	3	(11)	2	(7)	5	(19)	3	(12)	4	(18)	5	(23)
	1+	13	(46)	10	(36)	12	(44)	15	(58)	18	(69)	12	(55)	12	(55)
	2+	5	(18)	8	(29)	9	(33)	4	(15)	3	(12)	4	(18)	3	(14)
	3+	9	(32)	7	(25)	4	(15)	2	(8)	2	(8)	2	(9)	2	(9)
	<i>P</i> value	—		0.575		0.225		0.003		0.006		0.007		0.004	
Amount of watery discharge	—	3	(11)	7	(25)	11	(41)	9	(35)	10	(38)	10	(45)	8	(36)
	1+	14	(50)	9	(32)	12	(44)	14	(54)	13	(50)	10	(45)	12	(55)
	2+	10	(36)	10	(36)	4	(15)	3	(12)	3	(12)	2	(9)	2	(9)
	3+	1	(4)	2	(7)	0	(0)	0	(0)	0	(0)	0	(0)	0	(0)
	<i>P</i> value	—		0.593		0.002		0.001		< 0.001		0.003		0.004	
Property of nasal discharge	—	4	(14)	9	(32)	12	(44)	11	(42)	10	(38)	14	(64)	13	(59)
	1+	1	(4)	1	(4)	2	(7)	2	(8)	3	(12)	0	0	1	(5)
	2+	7	(25)	9	(32)	4	(15)	5	(19)	6	(23)	5	(23)	1	(5)
	3+	16	(57)	9	(32)	9	(33)	8	(31)	7	(27)	3	(14)	7	(32)
	<i>P</i> value	—		0.059		0.014		0.004		< 0.001		< 0.001		< 0.001	

Abbreviation: HDM AR = house dust mite induced allergic rhinitis

Patients = Number of patients, % = Percentage of patients. *P* values mean the results of Wilcoxon signed-rank test.

Swelling of the inferior nasal concha mucosa: —; None, 1+; Center of middle concha is visible, 2+; Between 3+ and 1+, 3+; Middle concha is invisible

Color of the inferior nasal concha mucosa: —; None, 1+; Pink, 2+; Redness, 3+; Pallor

Amount of watery discharge: —; None, 1+; Just attached on surface, 2+; Between 3+ and 1+, 3+; Full

Property of nasal discharge: —; None, 1+; Purulent, 2+; Viscous, 3+; Watery

Supplementary Table 3

JRQLQ general state in HDM AR during the course of the study.

		Baseline (n = 28)		Week 26 (n = 27)		Week 52 (n = 26)		Week 78 (n = 22)		Week 95-120 (n = 22)	
		Patients	(%)	Patients	(%)	Patients	(%)	Patients	(%)	Patients	(%)
Evaluation index	0: Fine	3	(11)	6	(22)	6	(23)	9	(41)	6	(27)
	1	4	(14)	13	(48)	10	(38)	9	(41)	13	(59)
	2	14	(50)	5	(19)	8	(31)	2	(9)	3	(14)
	3	6	(21)	3	(11)	1	(4)	2	(9)	0	(0)
	4: Crying	1	(4)	0	(0)	1	(4)	0	(0)	0	(0)
<i>P</i> value		—		< 0.001		0.020		< 0.001		< 0.001	

Abbreviations: HDM AR = house dust mite induced allergic rhinitis, JRQLQ = Japanese Rhinoconjunctivitis Quality of Life Questionnaire
 Patients = Number of patients, % = Percentage of patients. *P* values mean the results of Wilcoxon signed-rank test.

Supplementary Table 4A

Individual threshold concentration, initial dose and maintenance dose data in HDM AR.

Primary disease†	Schedule for immunotherapy	Age	Threshold concentration (JAU/mL)	Initial dose (JAU)	Up-dosing phase		Maintenance phase	Whole treatment duration (Weeks)
					Duration (Weeks)	Number of injection	Maintenance dose (JAU)	
HDM AR	Conventional schedule for immunotherapy							
	Administration protocol with increase by 50%	9	1	0.002	30	27	2	133
		10	1	0.002	34	32	2	136
		32	0.1	0.0002	23	19	0.2‡	23
		47	0.001	0.000005	64	59	100	120
	Administration protocol with increase by 100 to 200%	5	—§	0.05	19	17	200	125
		8	0.000001	0.00000005	24	20	200	130
		11	1	0.05	21	17	200	139
		22	10	0.5	15	11	1000	130
		40	1	0.05	19	17	200	139
		41	1	0.05	20	14	100	124
		44	1	0.05	9	8	10	52
		44	10	0.5	10	5	10	34
		53	0.1	0.005	18	17	300	129
	Rush schedule for immunotherapy							
	Rush protocol	6	0.000001	0.0000001	8	27	1000	75
		6	0.1	0.001	5	17	500	142
		9	0.000001	0.00000001	8	28	1000	75
		10	0.01	0.00005	36	29	500	125
		11	1	0.05	23	20	2000	120
		11	0.1	0.0005	4	30	100	124
		13	0.0001	0.0000001	2	24	1000	77
		13	1	0.05	6	15	200	120
		14	1	0.05	7	15	200	118
		14	0.1	0.0005	4	31	40	121
		17	0.001	0.00002	7	22	700	127
		17	0.001	0.000005	28	33	500	124
		22	0.001	0.00005	13	21	200	125
	Cluster protocol	6	0.01	0.001	25	22	1000	132
		25	0.001	0.000005	27	36	500	124

Abbreviation: HDM AR = house dust mite induced allergic rhinitis

† For patients with both AR and allergic BA, the investigator selected one of these as the primary disease.

‡ Maintenance dose data of patients who discontinued during the up-dosing phase complemented the final dose.

§ No available.

Supplementary Table 4B

Individual threshold concentration, initial dose and maintenance dose data in HDM BA.

Primary disease†	Schedule for immunotherapy	Age	Threshold concentration (JAU/mL)	Initial dose (JAU)	Up-dosing phase		Maintenance phase	Whole treatment duration (Weeks)
					Duration (Weeks)	Number of injection	Maintenance dose (JAU)	
HDM BA	Conventional schedule for immunotherapy							
	Administration protocol with increase by 50%	31	0.1	0.0005	69	67	150‡	69
		46	0.01	0.00005	49	45	100	78
	Administration protocol with increase by 100 to 200%	9	1	0.005	17	14	100	120
		22	1	0.001	28	26	1000	135
		29	0.00001	0.00000005	81	64	100	104
		32	10	0.05	12	7	10	52
		45	10	0.05	5	4	0.2	33
		53	0.001	0.00001	23	29	500	127
		Rush schedule for immunotherapy						
	Rush protocol	7	100	1	5	14	2000	122
		7	0.0001	0.000002	7	21	500	121
		10	0.00001	0.0000002	6	24	1000	138
		10	0.1	0.001	7	17	500	141
		13	0.01	0.0001	7	20	500	141
		28	0.01	0.00005	17	31	50	125
	Cluster protocol	10	0.01	0.0001	25	24	1000	132
		12	0.01	0.0001	25	24	1000	132

Abbreviation: HDM BA = house dust mite induced allergic bronchial asthma

† For patients with both AR and allergic BA, the investigator selected one of these as the primary disease.

‡ Maintenance dose data of patients who discontinued during the up-dosing phase complemented the final dose.

Supplementary Table 5

Rates of adverse drug reaction in different subgroups divided according to age, treatment period, and up-dosing method.

			Rates of adverse drug reactions		
			ADRs	Patients	(%)
Total		(n = 44)	151	22	(50)
Age	5-11 years	(n = 18)	37	9	(50)
	12 years ≤	(n = 26)	114	13	(50)
Treatment period	Up-dosing phase	(n = 44)	105	21	(48)
	Maintenance phase	(n = 42)	46	11	(26)
Schedule for immunotherapy	Conventional	(n = 21)	42	9	(43)
	Rush	(n = 23)	109	13	(57)

Abbreviation: ADR = adverse drug reaction

ADRs = Number of adverse drug reactions, Patients = Number of patients with adverse drug reactions, % = Percentage of patients with adverse drug reactions

Supplementary Table 6
Incidence of adverse drug reactions.

System organ class	Preferred term	Rates of adverse drug reactions			
		No. of ADRs	(%) [†]	No. of patients	(%) [‡]
Total		151	(7.6)	22	(50)
Local reaction	Total	97	(4.9)	14	(32)
	Injection site erythema	14	(0.7)	5	(11)
	Injection site induration	28	(1.4)	2	(5)
	Injection site pain	11	(0.6)	7	(16)
	Injection site pruritus	10	(0.5)	6	(14)
	Injection site reaction	6	(0.3)	5	(11)
	Injection site warmth	5	(0.3)	3	(7)
	Injection site swelling	22	(1.1)	7	(16)
	Injection site discomfort	1	(0.1)	1	(2)
Systemic reaction	Total	54	(2.7)	17	(39)
Blood and lymphatic system disorders	Total	2	(0.1)	1	(2)
	Lymphadenopathy	2	(0.1)	1	(2)
Immune system disorders	Total	6	(0.3)	6	(14)
	Anaphylactic reaction	4	(0.2)	4	(9)
	Anaphylactic shock	2	(0.1)	2	(5)
Nervous system disorders	Total	2	(0.1)	2	(5)
	Headache	1	(0.1)	1	(2)
	Sensory disturbance	1	(0.1)	1	(2)
Eye disorders	Total	1	(0.1)	1	(2)
	Ocular hyperaemia	1	(0.1)	1	(2)
Ear and labyrinth disorders	Total	1	(0.1)	1	(2)
	Ear pruritus	1	(0.1)	1	(2)
Vascular disorders	Total	2	(0.1)	2	(5)
	Flushing	2	(0.1)	2	(5)
Respiratory, thoracic and mediastinal disorders	Total	12	(0.6)	8	(18)
	Asthma	2	(0.1)	2	(5)
	Cough	5	(0.3)	5	(11)
	Dyspnoea	1	(0.1)	1	(2)
	Sneezing	1	(0.1)	1	(2)
	Wheezing	1	(0.1)	1	(2)
	Oropharyngeal discomfort	2	(0.1)	2	(5)
Gastrointestinal disorders	Total	4	(0.2)	3	(7)
	Nausea	2	(0.1)	1	(2)
	Vomiting	2	(0.1)	2	(5)
Skin and subcutaneous tissue disorders	Total	21	(1.1)	8	(18)
	Erythema	3	(0.2)	2	(5)
	Pruritus	4	(0.2)	2	(5)
	Urticaria	13	(0.7)	6	(14)
	Plantar erythema	1	(0.1)	1	(2)
General disorders and administration site conditions §	Total	3	(0.2)	3	(7)
	Malaise	1	(0.1)	1	(2)
	Oedema peripheral	1	(0.1)	1	(2)
	Pyrexia	1	(0.1)	1	(2)

Abbreviation: ADR = adverse drug reaction

[†] Percentage of adverse drug reactions for a total of 1988 injections

[‡] Percentage of patients with adverse drug reactions for a total of 44 patients

§ Excluded administration site conditions (administration site condition was included in the local reaction)

Supplementary Table 7

Number of adverse drug reactions by time to onset.

	Time to onset												
	≤15 minutes		16-30 minutes		31-45 minutes		46-60 minutes		61-120 minutes		121 minutes <		Whole time
	(n = 14)		(n = 9)		(n = 6)		(n = 2)		(n = 9)		(n = 13)		(n = 22)
	ADRs	(%)	ADRs	(%)	ADRs	(%)	ADRs	(%)	ADRs	(%)	ADRs	(%)	Total
Total	39	(25.8)	49	(32.5)	13	(8.6)	3	(2.0)	21	(13.9)	26	(17.2)	151
Local reaction	29	(30)	41	(42)	7	(7)	2	(2)	10	(10)	8	(8)	97
Systemic reaction	10	(19)	8	(15)	6	(11)	1	(2)	11	(20)	18	(33)	54

Abbreviation: ADR = adverse drug reaction

ADRs = Number of adverse drug reactions, % = Percentage of adverse drug reactions at each interval

Supplementary Table 8

JRQLQ regional scores in HDM AR during the course of the study.

	Baseline	Week 26		Week 52		Week 78		Week 95-120	
	(n = 28)	(n = 27)	<i>P</i> value	(n = 26)	<i>P</i> value	(n = 22)	<i>P</i> value	(n = 22)	<i>P</i> value
Daily life	1.00 ± 1.08 0.60 (0.0-4.0)	0.37 ± 0.50 0.00 (0.0-1.8)	0.003	0.45 ± 0.62 0.00 (0.0-2.0)	0.014	0.39 ± 0.54 0.00 (0.0-1.6)	0.008	0.24 ± 0.49‡ 0.00 (0.0-1.8)	< 0.001
Outdoor activities	0.64 ± 1.06 0.00 (0.0-4.0)	0.35 ± 0.59 0.00 (0.0-2.0)	0.363	0.42 ± 0.92 0.00 (0.0-4.0)	0.641	0.16 ± 0.36 0.00 (0.0-1.0)	0.051	0.05 ± 0.15‡ 0.00 (0.0-0.5)	0.011
Social life	0.41 ± 0.76 † 0.00 (0.0-2.7)	0.11 ± 0.29 0.00 (0.0-1.3)	0.118	0.27 ± 0.83 0.00 (0.0-4.0)	0.774	0.06 ± 0.20 0.00 (0.0-0.7)	0.067	0.00 ± 0.00‡ 0.00 (0.0-0.0)	0.024
Sleep	1.07 ± 1.36 1.00 (0.0-4.0)	0.44 ± 0.85 0.00 (0.0-3.0)	0.031	0.35 ± 0.69 0.00 (0.0-2.0)	0.010	0.14 ± 0.47 0.00 (0.0-2.0)	< 0.001	0.00 ± 0.00§ 0.00 (0.0-0.0)	< 0.001
The body	1.02 ± 1.30 0.25 (0.0-4.0)	0.46 ± 0.69 0.00 (0.0-2.0)	0.080	0.52 ± 0.94 0.00 (0.0-4.0)	0.144	0.34 ± 0.61 0.00 (0.0-2.0)	0.031	0.25 ± 0.72§ 0.00 (0.0-3.0)	0.014
Spiritual life	0.71 ± 1.18 0.25 (0.0-4.0)	0.31 ± 0.70 0.00 (0.0-3.0)	0.101	0.19 ± 0.37 0.00 (0.0-1.3)	0.017	0.23 ± 0.39 0.00 (0.0-1.3)	0.041	0.05 ± 0.17§ 0.00 (0.0-0.8)	0.003

Abbreviations: HDM AR = house dust mite induced allergic rhinitis, JRQLQ = Japan allergic rhinitis standard QOL questionnaire

Data are presented as means ± standard deviation and medians (minimum-maximum), *P* value; Dunnett test, Full analysis set

† Excluded 1 patient who ticked "definitely do not relate to the symptoms in nose and eye" in the baseline of Social life.

‡ Excluded 1 patient who ticked "definitely do not relate to the symptoms in nose and eye" in the Week 95-120 of daily life, outdoor activities and social life.

§ Excluded 1 patient who ticked "definitely do not relate to the symptoms in nose and eye" in the Week 95-120 of sleep, the body and spiritual life.

Supplementary Table 9

Percentage predicted forced expiratory volume in one second in HDM BA during the course of the study.

	Baseline (n = 16)	Week 12 (n = 16)	Week 26 (n = 16)	Week 38 (n = 15)	Week 52 (n = 15)	Week 78 (n = 13)	Week 95-120 (n = 12)
Mean	94.29	96.73	97.61	92.43	94.69	97.08	95.83
Standard deviation	12.44	12.02	15.32	12.39	13.52	15.50	14.64
Median	95.95	95.70	91.95	90.80	94.90	93.90	96.50
(Minimum- Maximum)	(72.8-113.9)	(72.2-115.0)	(71.1-124.3)	(70.6-112.2)	(73.7-114.3)	(72.5-116.0)	(72.5-116.4)

Abbreviation: HDM BA = house dust mite induced allergic bronchial asthma

Percentage predicted forced expiratory volume in one second (%)

Supplementary Fig. 1.

[illegible]

† Patients who met the eligibility criteria received an intradermal test with TO-204 to determine the threshold concentration.

‡ Patients with HDM AR were assessed nasal symptom score, patients with HDM BA were assessed asthma control test (12 years ≤) and childhood asthma control (5-11 years)

§ Last observation of efficacy was assessed once after Week 78.

¶ Baseline of efficacy outcomes.