Long-term treatment with egg oral immunotherapy enhances sustained unresponsiveness that persists after cessation of therapy

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Background: We previously reported the results of a randomized placebo-controlled study of egg oral immunotherapy (eOIT) in which 27.5% of subjects achieved sustained unresponsiveness (SU) after 2 years. Here we report the results of treatment through 4 years and long-term follow-up.

Objective: We sought to evaluate the efficacy and safety of eOIT in participants treated up to 4 years.

Methods: Children with egg allergy (5-18 years old) received eOIT (n = 40) for up to 4 years or placebo (n = 15) for 1 year or less. The key outcome was the percentage of subjects achieving SU by year 4. Safety and immunologic assessments were performed, and long-term follow-up questionnaires (LFQs) were administered after study conclusion (LFQ-1) and 1 year later (LFQ-2).

Results: Of 40 eOIT-treated subjects, 20 (50.0%) of 40 demonstrated SU by year 4. For those subjects still dosing during years 3 and 4, mild symptoms were present in 12 (54.5%) of 22 subjects. At the time of the LFQ, more subjects receiving eOIT (LFQ-1, 23/34 [68%]; LFQ-2, 21/33 [64%]) were consuming unbaked and baked egg versus placebo (LFQ-1, 2/11 [18%], $P = .006$; LFQ-2, 3/12 [25%], $P = .04$). Of subjects achieving SU, 18 (90%) of 20 completed the LFQ, with 18 (100%) of 18 reporting consumption of all forms of egg. When compared with subjects not achieving SU, subjects achieving SU had higher IgG_4 values ($P = .001$) and lower egg skin prick test scores ($P = .0002$) over time and a lower median baseline ratio of egg-specific IgE to total IgE (1.1% vs 2.7%, $P = .04$).

Conclusions: SU after eOIT is enhanced with longer duration of therapy and increases the likelihood of tolerating unbaked egg in the diet. (J Allergy Clin Immunol 2016;137:1117-27.)

Key words: *Egg allergy, food allergy, oral immunotherapy, desensitization, sustained unresponsiveness, immune tolerance, IgE, follow-up*

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Abbreviations used
AUC: Area under the curve
eOIT: Egg oral immunotherapy
IQR: Interquartile range
kU_A: Kilounits of antibody
LFQ: Long-term follow-up questionnaire
OFC: Double-blind, placebo-controlled oral food challenge
OIT: Oral immunotherapy
OR: Odds ratio
SPT: Skin prick test
SU: Sustained unresponsiveness

Egg allergy is common in childhood, with a prevalence ranging from 0.5% to 2.5%.[1-4] Egg and egg-derived products are ubiquitous ingredients, and therefore avoiding accidental exposures leading to reactions is difficult.[5] Although the long-term prognosis of egg allergy is generally favorable, recent studies have suggested that resolution might occur more slowly than was previously appreciated, and a subset of patients with egg allergy have egg allergy that persists into adolescence.[6,7] Until and if tolerance develops spontaneously, patients are at risk for allergic reactions.

Several approaches to mitigating this risk have been examined in clinical trials. The best-studied approach is oral immunotherapy (OIT), a procedure that aims to decrease reactivity to allergen with gradual escalation of daily doses followed by a maintenance treatment period.[8] Our group previously reported that 10 months of treatment with egg oral immunotherapy (eOIT) was superior to placebo when comparing the successfully consumed dose during ongoing therapy (clinical desensitization) and that this benefit was enhanced with an additional year of therapy.[9] Sustained unresponsiveness (SU; defined as a lack of dose-limiting symptoms during a double-blind, placebo-controlled oral food challenge [OFC] to and subsequent open feeding of egg 4-6 weeks after stopping OIT) was achieved in 28% of the subjects receiving eOIT by month 24, with all reporting consumption of egg 1 year later. The results from this trial suggested that OIT might have a long-term disease-modifying effect, as noted for outcomes assessed during 2 years of therapy.

Evidence of such an outcome would be a major breakthrough in the development of a food allergy treatment. Currently, the stability of treatment effects after such trials are not well understood. Two recent studies have provided long-term follow-up data after milk and peanut OIT, and both demonstrated that regular oral intake of the allergenic food appears to be required to maintain the protective effect after OIT; however, continued intake was difficult for some patients to continue.[10,11] Another study comparing peanut OIT and sublingual immunotherapy demonstrated suppression of basophil effector cell function and dendritic cell–driven T_H2 cytokine responses after peanut OIT, with some reversibility of those responses noted when antigen was discontinued in those achieving SU.[12,13] These studies indicate that if allergen is avoided, clinical relapse can ensue, even among subjects previously considered to be treatment successes. This situation poses potential safety concerns if such subjects incorrectly believe themselves to be protected.[14] Additional long-term studies after OIT treatment are necessary to further examine the feasibility, safety, and durability of the treatment effect.

To investigate the effects of long-term OIT in patients with egg allergy, we continued the previously reported trial of eOIT for up to 4 years of treatment.[9] The proportion of subjects achieving SU in those 4 years was calculated. After the treatment phase of the study ended, an annual long-term follow-up questionnaire (LFQ) to assess egg consumption patterns was administered.

METHODS

Study design and end points assessed

The current study is an extension of a previously published multicenter, randomized, double-blind, placebo-controlled study of eOIT.[9] As previously reported, subjects were enrolled and treated with placebo or eOIT for 10 months. Placebo-treated subjects were discontinued from dosing after 10 months and were followed as treatment controls through year 2 and then surveyed for long-term follow-up. Subjects receiving eOIT continued dosing to year 4 and discontinued dosing after passing an OFC off therapy (ie, those achieving SU) after any yearly challenge point (years 2, 3, or 4). The key outcome of this study was the percentage of subjects with SU to egg after up to 4 years of eOIT. SU was defined as a lack of dose-limiting symptoms during a 10-g egg white powder (approximately 8 g of egg white protein) OFC and open feeding of a meal-sized portion of whole cooked egg 4 to 6 weeks after stopping eOIT while maintaining an egg-restricted diet. Secondary outcomes included safety during the additional years of treatment by using methods previously reported[9] and immunologic assessments. Egg consumption was evaluated after the last subject completed the treatment phase by having all available subjects complete an LFQ, which was repeated approximately 1 year later. Tolerance to baked egg consumption was not assessed during the study entry or at any point in the study.

Study population

Subjects were aged 5 to 18 years from 5 US sites, with inclusion/exclusion criteria previously reported.[9] The study was approved by each site's institutional review board, and written consent/assent was obtained. The study was conducted under a US Food and Drug Administration investigational new drug application and monitored by an independent data and safety monitoring board from the National Institute of Allergy and Infectious Diseases.

eOIT dosing and participant follow-up

Dried standard egg white powder (raw, uncooked egg) was purchased from a commercial manufacturer (Deb-El Food Products, Elizabeth, NJ) and manufactured for individual doses for eOIT dosing. The daily OIT dose was mixed in a vehicle, such as pudding or applesauce, for dosing. Limiting physical activity was recommended for all participants for the first 2 hours after OIT dosing. Subjects who attained SU at any challenge point were instructed to incorporate egg into their diets *ad libitum*; however, there were no specific recommendations made on the frequency, amount, or type of egg product.[9] Subjects who did not have SU at year 2 or year 3 were instructed to continue egg avoidance and to continue open-label dosing per protocol with 2000 mg/d eOIT for up to 4 years of treatment. Subjects who failed the SU OFC resumed eOIT maintenance dosing through dose escalations every 1 to 2 weeks beginning with 25% of their maintenance dose or their highest tolerated cumulative dose during the OFC, whichever was lowest. Subjects who withdrew from dosing for any reason other than achieving SU or were originally in the placebo treatment arm were instructed to continue dietary avoidance of egg. An exception included one site's institutional review board mandate to cross over placebo subjects to eOIT treatment after year 2 as part of a separate treatment protocol. Subjects who did not achieve SU after the 4-year study period were discontinued from dosing and instructed to continue dietary avoidance. For LFQ analysis, subjects were grouped into 4 categories based on their treatment and last known clinical outcome status: (1) eOIT-SU, (2) eOIT-desensitized, (3) eOIT-not desensitized, and (4) placebo.

Oral food challenge assessment

At years 2, 3, and 4, all subjects treated with eOIT underwent a 10-g (cumulative dose) oral food challenge (OFC) to egg white powder to assess desensitization (ie, the ability to consume egg while undergoing OIT). Those who passed the desensitization OFC discontinued OIT dosing for 4 to 6 weeks and had a second (10-g) OFC, followed by a 10-g open egg feeding, to assess for SU, as previously reported.[9] The food challenge dose was mixed in a vehicle, typically pudding or applesauce, for dosing.

LFQ

A scripted 25-item LFQ was administered twice (LFQ-1 after the last subject completed dosing and study OFCs and LFQ-2 approximately 1 year later). This survey (see this article's Online Repository at www.jacionline.org) captured patterns of unbaked and baked egg consumption in egg eOIT participants and those previously treated with placebo OIT. In the LFQ unbaked egg products referred to scrambled eggs, fried eggs, raw eggs, undercooked eggs, French toast, or custard; baked egg products referred to cakes, muffins, and waffles. Subjects were asked to rate the frequency of symptoms to various forms of egg ingested to assess consumption patterns and adverse symptoms.

Assessment of immunologic markers/mechanisms

Immunologic mechanisms were assessed during the study, as previously described.[9] Skin prick test (SPT) responses, serum egg-specific IgE and IgG_4 antibody levels, and basophil activation study results were performed at 6-month intervals through the end of the study (year 4). End point titration SPTs were performed at year 4 by measuring SPT wheal size with 5 serial weight/volume egg extract dilutions using commercially available egg white extract from Greer Laboratories (Lenoir, NC; 1:20, 1:200, 1:2,000, 1:20,000, 1:200,000).[15]

Statistical methods

The study design was previously described[9] and, in brief, was adequately powered to compare the placebo arm with the eOIT arm for the primary end point (ie, at 2 years) but was not specifically designed to ensure power for long-term comparisons of interest. The key outcome of interest for the extension phase was the cumulative proportion of eOIT-treated subjects achieving SU through 4 years. The proportion of eOIT-treated subjects who did not achieve SU at 2 years but did achieve SU when treated after 2 years was also estimated. Comparisons of interest for immunologic assessments were between eOIT-treated subjects who achieved SU and those who did not. Differences between groups in continuous variables were assessed by using exact Wilcoxon rank sum tests where computationally possible and the asymptotic Wilcoxon rank sum P value in all other cases. Egg end point titration area under the curve (AUC) was calculated as the sum of the wheal scores at each of the 5 serial weight/volume egg extract dilutions and assessed by using the Wilcoxon rank sum test. Basophil activation, immunoglobulin levels, and SPT wheal scores were each evaluated in repeated-measurement models, with the baseline value, study visit, and SU status at year 4 as covariates and heterogeneous compound symmetry within-person covariance. Hypothesis testing was performed by using $\log_{10}$ transformation for IgE and IgG_4, whereas summary statistics are reported on the observed scale. For the LFQ, analysis was primarily descriptive, although differences between original treatment groups (placebo vs eOIT) were evaluated in proportions of categorical variables by using the Fisher exact test and in continuous variables by using the Wilcoxon rank sum test. Logistic regression was used to identify baseline and week 44 clinical and mechanistic factors (log-transformed, where noted) that might identify SU. All analyses were performed with SAS software, version 9.2 (SAS Institute, Cary, NC).

RESULTS

Study population

Fifty-five subjects enrolled initially (Fig 1), 6 (15%) of 40 eOIT-treated subjects and 2 (13.3%) of 15 placebo-treated subjects withdrew from dosing before 24 months, 1 eOIT-treated subject withdrew after 24 months but before resuming dosing, and 8 additional eOIT-treated subjects withdrew from dosing after 24 months but before the end of the study at year 4 (see Table E1 in this article's Online Repository at www.jacionline.org). One of these subjects was withdrawn at month 30 for noncompliance because of regular whole egg consumption before passing any study OFC.

SU is enhanced with duration of eOIT dosing

As previously reported, 0 (0%) of 15 placebo-treated and 22 (55%) of 40 eOIT-treated subjects were desensitized to 5 g of egg white powder after 10 months of therapy, and 30 (75%) of 40 eOIT-treated subjects were desensitized to 10 g of egg white powder after 22 months of therapy.[9] The number of eOIT-treated subjects attaining desensitization reached 31 (77.5%) of 40 at years 3 and 4 (Table I). SU at year 2 occurred in 11 (27.5%) of 40 eOIT-treated subjects[9] and increased to 18 (45.0%) of 40 in year 3 and 20 (50.0%) of 40 in year 4. Among the 22 eOIT-treated subjects receiving treatment after year 2, 41% achieved SU by year 4. Logistic regression identified no baseline clinical features (eg, age, atopic dermatitis, or asthma) as significantly predictive of SU at year 4.

Long-term OIT dosing induces mild symptoms in a majority of subjects

Of 22 subjects receiving eOIT in years 3 to 4, 12 (54.5%) experienced symptoms with dosing, all categorized as mild (see Table E2 in this article's Online Repository at www.jacionline.org). Three (13.6%) subjects reported oral/pharyngeal symptoms only; 9 (40.9%) subjects reported skin, respiratory, or gastrointestinal symptoms; and 6 (27.3%) subjects were treated. Of 8925 doses administered, 95% (8482 doses) were symptom free (Table II). Only 1.9% of doses resulted in symptoms lasting more than 30 minutes reported during home dosing; 1.6% of doses required treatment with antihistamines during either clinic (2 doses) or home (144 doses) dosing. None required treatment with epinephrine for dosing-related symptoms during years 3 to 4. Three subjects received 1 dose of epinephrine each during OFCs assessing SU. Dosing symptoms before and after year 2 were summarized among the 22 subjects with eOIT dosing after year 2. Before year 2, the median percentage of doses per subject with any symptoms was 8.0% for these 22 subjects. After year 2, the median percentage of doses per subject was 0.2% for these same 22 subjects, representing a reduction in dosing symptoms after year 2 of the study.

Pre–week 44 dosing reactions predictive of SU

We examined pre–week 44 dosing reactions to determine whether early dosing reactions were associated with long-term clinical outcomes (see Table E3 in this article's Online Repository at www.jacionline.org). Reactions were compared between eOIT-treated subjects achieving SU and all other eOIT-treated subjects. The only statistically significantly difference was in the percentage of doses with moderate symptoms per subject ($P = .03$, Wilcoxon test). Subjects with no moderate symptoms before week 44 were significantly more likely to achieve SU by year 4 compared with subjects who had any moderate symptoms

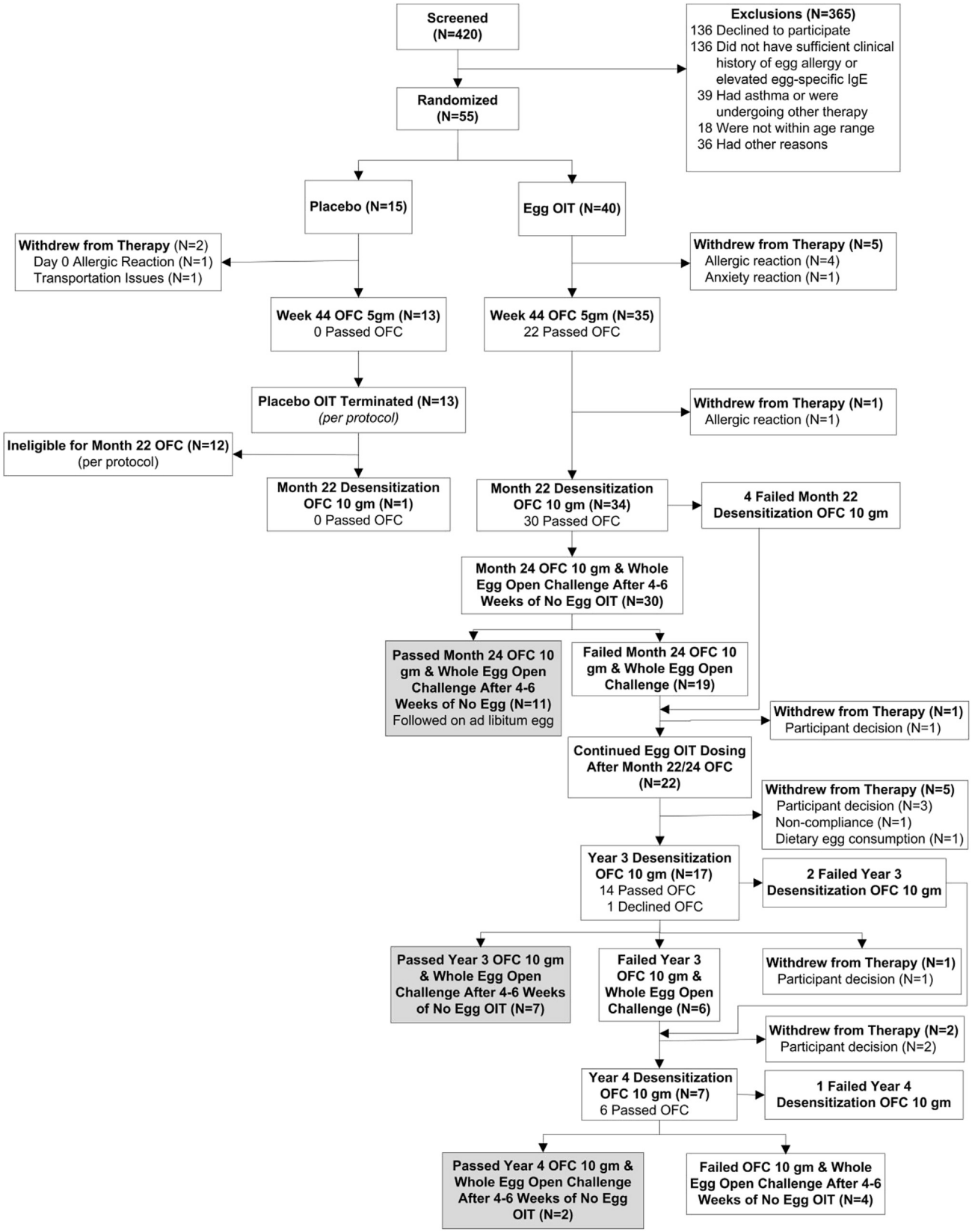

FIG 1. Study enrollment, randomization, and treatment outcomes through year 4. *Shaded boxes* represent participants achieving SU.

TABLE I. Food challenge–defined clinical outcomes with long-term eOIT

Time from eOIT initiation	Desensitization	SU
Year 2*	30/40 (75%)	11/40 (27.5%)
Year 3	31/40 (77.5%)	18/40 (45.0%)
Year 4	31/40 (77.5%)	20/40 (50.0%)†

*Previously reported in Burks et al.[9]
†Among the 22 eOIT-treated subjects treated after 2 years, 41% (95% CI, 21% to 64%) achieved SU. One eOIT-treated subject in whom the 2-year tolerance OFC failed did not resume eOIT dosing.

in that time period (logistic regression: $P = .047$; odds ratio [OR], 4.64; 95% CI, 1.02-21.00; see Table E4 in this article's Online Repository at www.jacionline.org).

Thirty-five food-induced reactions not related to study product occurred in eOIT-treated subjects in years 3 to 4, with only 3 being egg related. Two occurred in eOIT-desensitized subjects with symptoms of mouth/throat itching and 1 treated with antihistamines only. One reaction included abdominal pain and vomiting requiring epinephrine after the subject consumed lightly cooked egg without any precipitating triggers (eg, fever, viral infection, or exercise). This subject had achieved SU 19.4 months before and subsequently reported unrestricted asymptomatic egg consumption of baked and unbaked egg several times per week on their LFQs. A total of 141 adverse events (events that were not dosing symptom/OFC related) were reported in 21 of 22 subjects during years 3 and 4 of eOIT dosing, mostly Medical Dictionary for Regulatory Activities categorized as System Organ Class Infections/Infestations (40.4%). Adverse events were predominantly mild (95.7%), with 6 (4.3%) being moderate.

LFQ suggests persistence of SU after treatment cessation

Forty-five (82%) of 55 subjects completed the LFQ-1 and LFQ-2 (see Table E5 in this article's Online Repository at www.jacionline.org). LFQ-1 was completed for eOIT- and placebo-treated subjects, respectively, a median of 61.7 months (interquartile range [IQR], 57.0-64.7 months) and 58.5 months (IQR, 56.6-65.6 months) after study enrollment ($P = .74$), 15.6 months (IQR, 9.9-26.1 months), and 37.1 months (IQR, 34.4-39.4 months) after the last clinic visit ($P < .001$) and at a median age of 12.3 years (IQR, 11.2-13.7 years) and 13.0 years (IQR, 10.7-14.0 years; $P = .93$).

SU after OIT is associated with higher rates of successful egg consumption when compared with other treatment groups. During both LFQ periods, a significantly higher proportion of eOIT-treated subjects reported consumption of unbaked and baked egg compared with placebo-treated subjects, potentially driven by those achieving SU (Tables III and IV). However, when considering dietary consumption of any form of egg (unbaked or baked), only during the LFQ-1 were eOIT-treated subjects significantly different than placebo-treated subjects ($P = .007$, Table III).

Of eOIT-treated subjects achieving SU, 18 (100%) of 18 surveyed reported dietary consumption of all forms of egg in LFQ-1 and LFQ-2, whereas only 4 (44%) of 9 in LFQ-1 and 2 (25%) of 8 in LFQ-2 of eOIT-desensitized subjects and 1 (14%) of 7 in both LFQ-1 and LFQ-2 of eOIT-not desensitized subjects consumed unbaked egg (Table IV). Overall, eOIT-treated subjects achieving SU reported increased frequency and amount of egg consumption in all forms when compared with other outcome groups (Fig 2 and Table V and see Table E6 in this article's Online Repository at www.jacionline.org). Of subjects achieving SU, 3 (17%) of 18 had increased frequency of symptoms with egg consumption between LFQ-1 and LFQ-2, and 1 (6%) of 18 had symptom reduction. Three subjects at LFQ-1 and 4 subjects at LFQ-2 with SU reported symptoms after ingesting unbaked egg; 3 had increased symptoms to unbaked egg at LFQ-2 compared with LFQ-1, and 1 had decreased symptoms to unbaked egg. One subject with SU reported asymptomatic baked egg consumption at LFQ-1 but increased symptoms with baked egg consumption during LFQ-2. The longest period of time (median) that subjects achieving SU reported going without consuming unbaked egg was 14 days (IQR, 5-21 days) in LFQ-1 and 14 days (IQR, 5-14 days) in LFQ-2, and the longest period of time that subjects achieving SU reported going without consuming baked egg was 5.5 days (IQR, 2-14 days) in LFQ-1 and 3.5 days (IQR, 2-7 days) in LFQ-2. For eOIT-treated subjects not achieving SU and those who were placebo treated, dietary restriction of egg was variable, and successful dietary egg consumption in any form was reported at a lower frequency and lesser amounts were consumed than among subjects with SU (Fig 2 and Tables IV and V).

Immunologic data over time

Mechanistic data were compared between subjects achieving SU and those who did not both at year 4 specifically and over time. At year 4, only egg SPT score (median, 3.0 vs 7.0 mm; $P = .04$) and end point titration SPT AUC (median, 3.5 vs 11.0 mm; $P = .01$) were significantly changed in subjects achieving SU at year 4 (Fig 3 and see Tables E7-E9 and Fig E1 in this article's Online Repository at www.jacionline.org). A repeated-measures analysis was performed to examine whether there was a difference in immunologic values over time, after adjusting for baseline value and study visit, between subjects who reached SU and those who did not. Over time, egg IgG_4 levels were significantly greater in those achieving SU ($P = .001$). Similarly, egg SPT scores were significantly lower in those achieving SU ($P = .0002$). For egg IgE levels and basophil activation, there was a decrease over time in those achieving SU; however, the differences were not significant and therefore did not discriminate between those who achieved SU and those who did not.

Immunologic predictors of SU

The baseline median egg-specific IgE level was lower but statistically insignificant in subjects achieving SU compared with those who did not (9.4 kilounits of antibody [kU_A/L] vs 18.5 kU_A/L, $P = .07$). Median percentages of egg IgE at baseline (ie, the proportion of egg-specific IgE to total IgE multiplied by 100) were significantly lower in subjects with SU (1.1% vs 2.7%, $P = .04$); however, logistic regression analysis was not statistically significant (OR, 0.89; 95% CI, 0.78-1.01; $P = .072$). Logistic regression of mechanistic outcomes identified baseline $\log_{10}$ egg IgE level (OR, 0.21; 95% CI, 0.04-0.99; $P = .049$), week 44 $\log_{10}$ egg IgG_4 level (OR, 5.48; 95% CI, 1.23-24.38; $P = .026$), and week 44 $\log_{10}$ egg IgG_4/IgE ratio (OR, 4.62; 95% CI, 1.34-15.84; $P = .015$) as statistically significantly predictive of SU at year 4; however, the AUC for

TABLE II. OIT doses (percentages)* associated with symptoms during years 3 and 4 of the study

Visit type	No. of doses	Any symptom	Symptom type					Persist >30 min	Treated	Symptom severity		
			Oral/pharyngeal	Skin	Respiratory	Gastrointestinal	Other			Mild	Moderate	Severe
Clinic	194	14.9	11.3	0.5	1.5	1.0	1.5	0.0	1.0	4.1	0.0	0.0
Home	8731	4.7	2.2	1.1	2.1	0.2	0.2	2.0	1.6	3.0	0.0	0.0
All	8925	5.0	2.4	1.1	2.1	0.3	0.2	1.9	1.6	3.0	0.0	0.0

*With the exception of number of doses, values are percentages of doses.

TABLE III. LFQ: Egg consumption by treatment group

	eOIT, no. (%)	Placebo,* no. (%)	*P* value
Eating any egg in diet			
LFQ-1	28/34 (82)	4/11 (36)	.007
LFQ-2	28/33 (85)	8/12 (67)	.22
Eating unbaked and baked egg in diet			
LFQ-1	23/34 (68)	2/11 (18)	.006
LFQ-2	21/33 (64)	3/12 (25)	.04

*By the time of the LFQ-2, 2 placebo-treated subjects were receiving eOIT in other open-label study protocols. One had crossed over to active OIT at year 2 per the site's institutional review board requirements and was eating unbaked and baked egg for LFQ-1 and LFQ-2. This subject is included in both the group eating any egg in diet and the group eating unbaked and baked egg in diet for LFQ-1 and LFQ-2. The other subject entered a new study protocol after completion of LFQ-1 and was fully restricting egg except for study product eOIT dosing. For the LFQ-2, this subject is included in the group eating any egg in diet.

each model was less than 0.75 (log_{10} baseline egg IgE level: AUC, 0.67 [95% CI. 0.50-0.84]; week 44 log_{10} IgG_4 level: AUC, 0.71 [95% CI, 0.53-0.88], and week 44 log_{10} IgG_4/IgE ratio: AUC, 0.74 [95% CI, 0.59-0.90]), indicating limited utility as a predictor of treatment response.

DISCUSSION

Numerous single-center and multicenter studies have shown a beneficial effect of OIT on allergic responses in children with food allergy, including short-term protection for allergen desensitization and longer-term benefits, such as SU.[9-11,16-25] However, studies have suggested that long-lasting effects of therapy are unlikely without ongoing antigen exposure and that the effects of therapy might even be transient, highlighting the potential risk for relapse if allergen exposure is not maintained.[10-14,26]

We previously reported successful desensitization in children with egg allergy that improved in subjects from year 1 (55%) to year 2 (75%) on eOIT.[9] A smaller subset of subjects (27.5%) had SU after discontinuing daily OIT treatment.[9] In this treatment extension and long-term follow-up study, we show improvements in achieving SU with continued eOIT, with 45% in year 3 and 50% in year 4. This outcome might be comparable or slightly higher to rates of natural egg allergy resolution, which have been reported to be 12% to 37% in a similar age group.[6] Findings are similar to those published for an open-label peanut OIT study in which 50% of subjects attained SU after 5 years of therapy[11] and slightly higher than reported in a milk OIT trial in which 40% of subjects achieved SU after 15 months of OIT.[10] In a more recent study of shorter duration comparing peanut OIT and sublingual immunotherapy, 3 (27%) of 11 OIT-treated subjects attained SU after 18 months of therapy.[13]

Wide-scale clinical application of OIT has been hindered by reports of undesirable allergic side effects in randomized trials. In this study we report long-term safety in years 3 and 4 of eOIT

TABLE IV. LFQ: Egg consumption by treatment outcome status

	Unbaked egg*		Baked egg only		No Egg in diet		Total	
	LFQ-1	LFQ-2	LFQ- 1	LFQ-2	LFQ-1	LFQ- 2	LFQ-1	LFQ-2
eOIT-SU								
Total	18	18	0	0	0	0	18	18
No symptoms	15	14	0	0	0	0	15	14
Symptoms	3	4	0	0	0	0	3	4
eOIT, desensitized								
Total	4	2	4	5	1	1	9	8
No symptoms	4	2	3	4	0	0	7	6
Symptoms	0	0	1	1	0	0	1	1
Not in diet	0	0	0	0	1	1	1	1
eOIT, not desensitized								
Total	1	1	1	2	5	4	7	7
No symptoms	1	0	0	1	0	0	1	1
Symptoms	0	1	1	1	0	0	1	2
Not in diet	0	0	0	0	5	4	5	4
Placebo								
Total	2	4	2	4	7	4	11	12
No symptoms	1	3	1	1	0	0	2	4
Symptoms	1	1	1	3	0	0	2	4
Not in diet	0	0	0	0	7	4	7	4
Total	25	25	7	11	13	9	45	45

*The unbaked egg group includes subjects who had unbaked egg in their diet even if they did not have baked egg in their diet (1 placebo-treated subject enrolled in an eOIT study after completing LFQ-1 and consumes egg white powder daily but does not consume baked egg). For subjects in the unbaked egg group, the symptoms shown are to unbaked egg only and not to unbaked or baked egg. For subjects with baked egg only in their diet (baked egg only group), the symptoms shown are to baked egg only and not to unbaked egg.

dosing. Mild symptoms were noted in 54.5% of subjects during dosing, with none reporting moderate or severe symptoms. During follow-up, 1 eOIT-treated subject reported anaphylaxis after consuming unbaked egg 19.4 months after achieving SU. Subsequently, the subject was able to resume *ad libitum* egg consumption of both unbaked and baked egg without limitations and without symptoms, as reported on LFQs. Interestingly, during years 1 and 2 of eOIT dosing, 15% of eOIT-treated subjects withdrew mostly because of dose-related symptoms (predominantly gastrointestinal),[9] a finding that was not present during years 3 and 4. Participants required unscheduled visits and staff telephone calls for advice during illness, further highlighting the importance of close monitoring and staff availability during OIT.

This is the first multicenter OIT study to comprehensively survey allergen consumption and symptoms for up to 4 years of treatment and after study completion through an annual questionnaire. Overall, 100% of subjects achieving SU were able to consume unbaked egg in their diet. As noted in the Results section, only 1 case of anaphylaxis (caused by unbaked egg) was noted in a subject with SU; however, that subject continued to

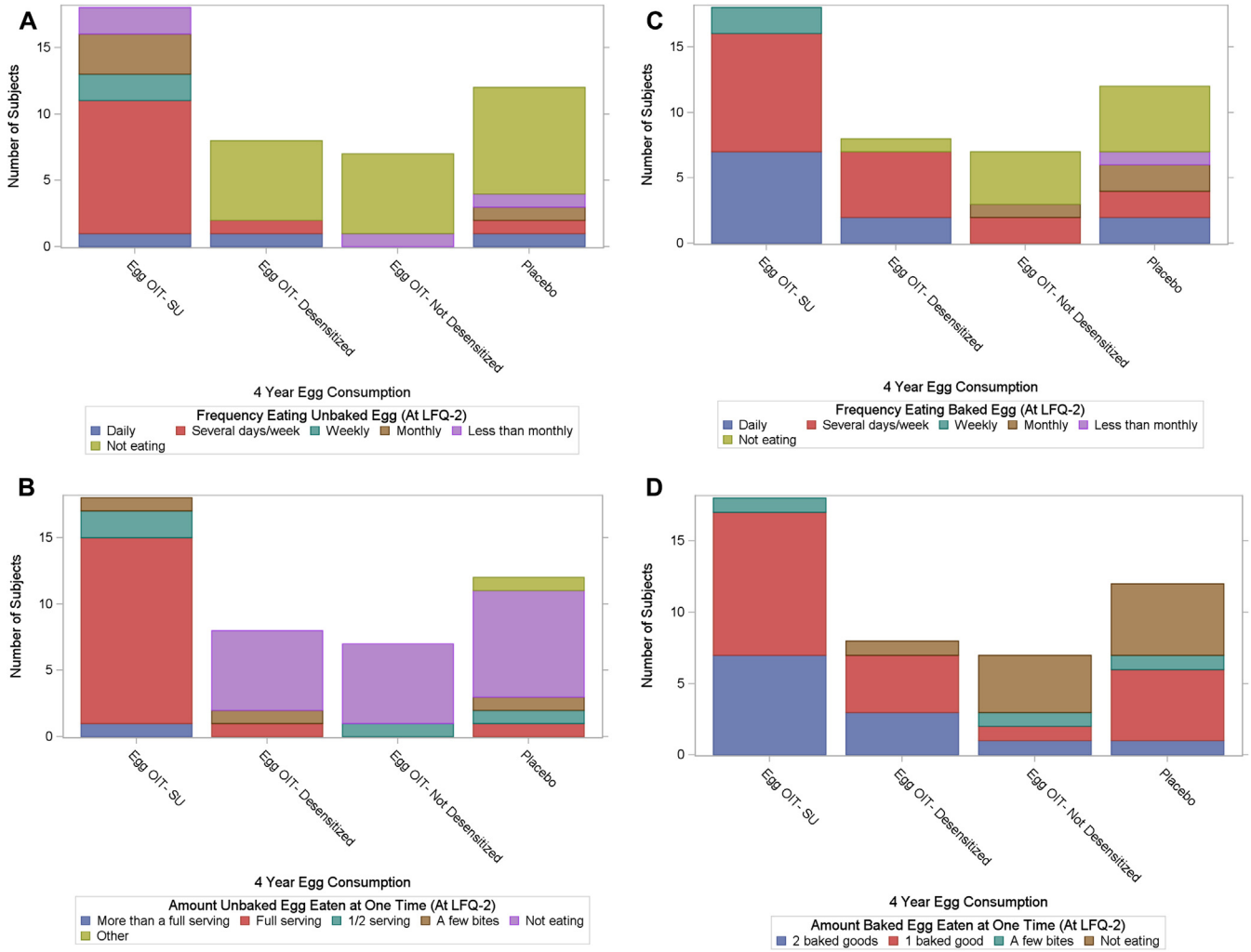

FIG 2. LFQ-2: Frequency and amount of egg consumption by treatment outcome status. **A,** Frequency of eating unbaked egg. The percentage of subjects who were eating unbaked egg several days per week or more was as follows: eOIT-SU group, 61%; eOIT-desensitized (eOIT-D) group, 25%; eOIT-not desensitized (eOIT-Not-D) group, 0%; and placebo group, 17%.* The percentage of subjects who were not eating unbaked egg was as follows: eOIT-SU group, 0%; eOIT-D group, 75%; eOIT-Not-D group, 86%; and placebo group, 67%. **B,** Amount of unbaked egg eaten at one time. The percentage of subjects who were eating a full serving or more of unbaked egg was as follows: eOIT-SU group, 83%; eOIT-D group, 13%; eOIT-Not-D group, 0%; and placebo group, 8%. **C,** Frequency of eating baked egg. The percentage of subjects who were eating baked egg several days per week or more was as follows: eOIT-SU group, 89%; eOIT-D group, 88%; eOIT-Not-D group, 29%; and placebo group, 33%.** The percentage of subjects who were not eating baked egg was as follows: eOIT-SU group, 0%; eOIT-D group, 13%; eOIT-Not-D group, 57%; and placebo group, 42%.* **D,** Amount of baked egg eaten at one time. The percentage of subjects who were eating 1 or more of baked egg goods was as follows: eOIT-SU group, 94%; eOIT-D group, 88%; eOIT-Not-D group, 29%; and placebo group, 50%.† *One placebo-treated subject enrolled in an eOIT study after completing LFQ-1 and consumes egg white powder daily but does not consume baked egg. †One placebo-treated subject crossed over to eOIT treatment per their site's institutional review board requirement after 2 years on study and consumes half a serving of concentrated egg less than monthly and 1 baked egg product daily.

consume unbaked egg in the diet without issues afterward. This finding in the SU treatment group was in direct contrast to that seen in other eOIT and placebo treatment groups, which demonstrated an overall inability to consistently consume unbaked egg. When comparing baked egg consumption among treatment groups, all eOIT and placebo groups were able to incorporate some amount of baked egg into their diets by the second questionnaire; however, the group achieving SU was able to consume baked egg without limitations more frequently and in higher amounts.

Despite instructions to continue an egg-free diet, placebo- or eOIT-treated subjects who did not attain SU reported unbaked and baked egg consumption, with many also reporting symptoms with consumption. Unfortunately, this difference in recommended diets and the lack of systemic evaluation of placebo-treated subjects is a clear source of bias because it is possible that, if systematically challenged, more placebo-treated subjects would be able to tolerate baked or unbaked egg than we found here. Full resolution of egg allergy in 2 (17%) of 12 placebo-treated subjects and partial improvement in dietary egg consumption in 7 (58%) of

TABLE V. LFQ-2: Frequency and amount of egg consumption by treatment outcome status

	eOIT, SU		eOIT, desensitized		eOIT, not desensitized		Placebo*	
	No.	Percent	No.	Percent	No.	Percent	No.	Percent
Total subjects	18	100.0	8	100.0	7	100.0	12	100.0
Frequency eating unbaked egg								
Daily	1	5.6	1	12.5	0	0	1	8.3
Several days/week	10	55.6	1	12.5	0	0	1	8.3
Weekly	2	11.1	0	0	0	0	0	0
Monthly	3	16.7	0	0	0	0	1	8.3
Less than monthly	2	11.1	0	0	1	14.3	1	8.3
Not eating	0	0	6	75.0	6	85.7	8	66.7
Amount of unbaked egg eaten at one time								
More than a full serving	1	5.6	0	0	0	0	0	0
Full serving	14	77.8	1	12.5	0	0	1	8.3
Half serving	2	11.1	0	0	1	14.3	1	8.3
A few bites	1	5.6	1	12.5	0	0	1	8.3
Other	0	0	0	0	0	0	1	8.3
Not eating	0	0	6	75.0	6	85.7	8	66.7
Frequency eating baked egg								
Daily	7	38.9	2	25.0	0	0	2	16.7
Several days/week	9	50.0	5	62.5	2	28.6	2	16.7
Weekly	2	11.1	0	0	0	0	0	0
Monthly	0	0	0	0	1	14.3	2	16.7
Less than monthly	0	0	0	0	0	0	1	8.3
Not eating	0	0	1	12.5	4	57.1	5	41.7
Amount of baked egg eaten at one time								
Two baked goods	7	38.9	3	37.5	1	14.3	1	8.3
One baked good	10	55.6	4	50.0	1	14.3	5	41.7
A few bites	1	5.6	0	0	1	14.3	1	8.3
Not eating	0	0	1	12.5	4	57.1	5	41.7

*One placebo-treated subject enrolled in an eOIT study after completing the LFQ-1 and consumes egg white powder daily but does not consume baked egg.

12 placebo-treated subjects was noted by the second follow-up questionnaire (although 1 of these was receiving open-label eOIT in another study protocol), suggesting the potential for underestimation of the effect of natural history.[6] In fact, at the time of the second questionnaire, there was no longer a significant difference in the inclusion of any form of egg in the diet between the active and placebo-treated participants; however, significant differences were noted when considering dietary consumption of all forms of egg.

Our results support the hypothesis that administering OIT for 3 to 4 years results in enhanced rates of SU when compared with administration for only 2 years. We speculate that this effect is due to ongoing immunomodulation in specific T- and B-regulatory processes over time, but we cannot rule out the role of natural history in this uncontrolled study. Furthermore, ongoing intermittent dietary allergen exposure might be required to maintain SU. Importantly, 17% of those achieving SU reported increased symptoms with variable egg consumption from daily to less than monthly, but all reported continued egg consumption, indicating that variable consumption might have led to symptoms in these subjects. In contrast, another study demonstrated that 100% of peanut OIT–treated subjects treated over 5 years maintained symptom-free *ad libitum* peanut ingestion while consuming an average of 555 mg/d (range, 0-4000 mg/d) at a frequency of 3 times/week (range, 0-7 times/week).[11] These results indicate that continued follow-up of patients with SU is critical to monitor for relapse.

Although multiple studies have indicated that significant immunomodulation is associated with a positive response to OIT,[9,11,19,24,27,28] none of the immune parameters tested have been consistently predictive of treatment outcomes. In this study only egg SPT and end point titration SPT responses were different at year 4 for subjects with SU, and only egg SPT responses and $\log_{10}$ egg IgG_4 levels changed significantly over time. Although baseline $\log_{10}$ egg IgE and week 44 $\log_{10}$ egg IgG_4 levels were predictive of SU, the *P* values were marginal and unadjusted for multiple comparisons, and the AUCs were low, we would not consider these immune parameters to be strong predictors. Egg-specific immunoglobulin components might provide additional predictors for success.[29] In the 5-year peanut OIT study, the baseline peanut-specific IgE/total IgE ratio and the baseline and end-of-study peanut-specific IgE levels were predictive of treatment success.[11] In a recent peanut OIT study, basophil activation and dendritic cell–driven T_H2 cytokine responses to peanut were reduced in subjects achieving SU after only 18 months of treatment; however, these parameters reversed in the majority of subjects after OIT was discontinued and were not defined predictors of treatment success.[12] Our findings and interpretations are limited by the small size of the study. Further work is needed to identify biomarkers reliably predictive of a successful treatment response to OIT.

This study has several limitations. First, the initial placebo treatment in this study lasted only 10 months without subsequent food challenges, and therefore the long-term efficacy and safety parameters reported are not controlled.

Second, as noted in our first report, the study population was enrolled based on egg allergy criteria that would predict persistence of allergy past 2 years of treatment, even though the

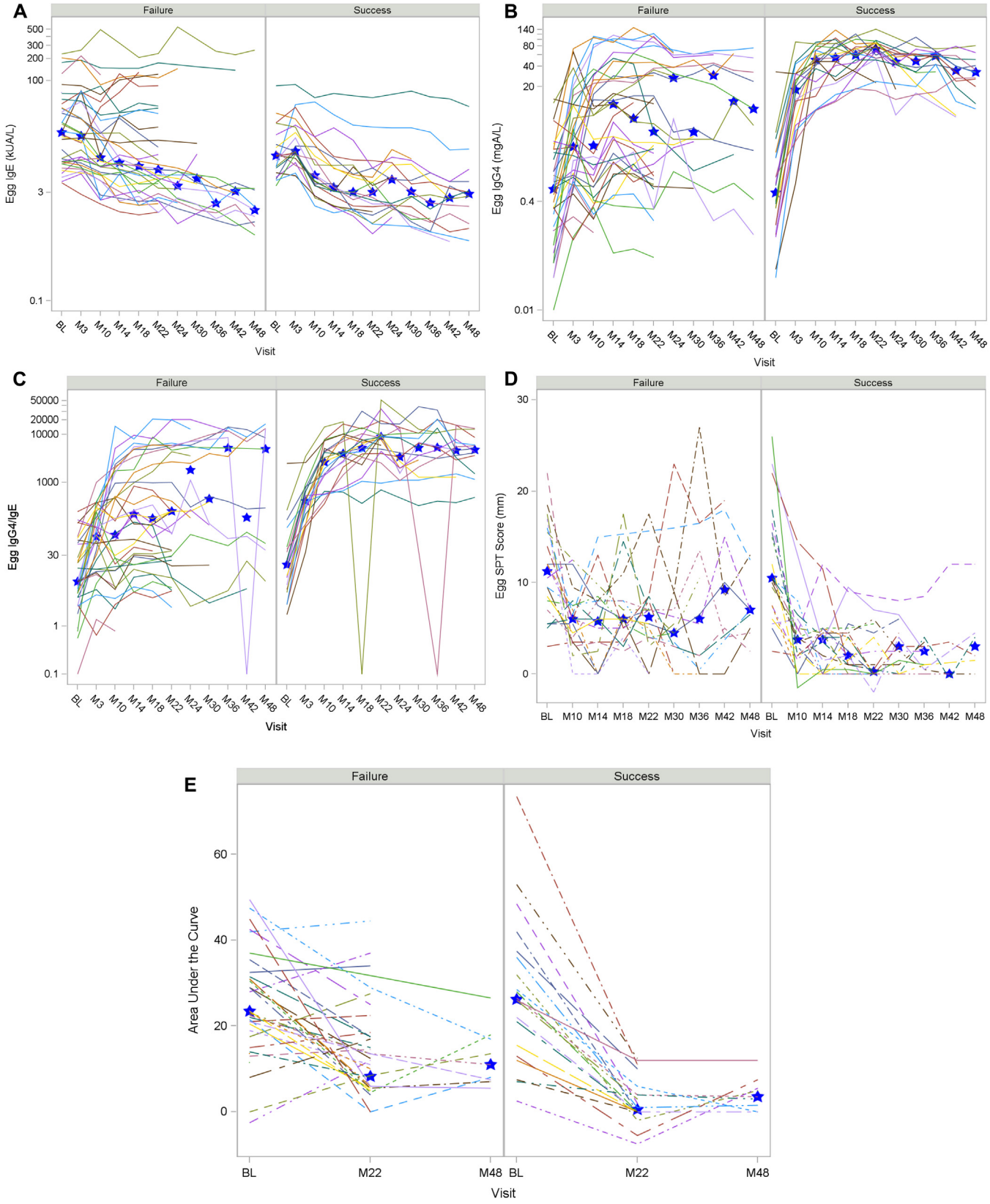

FIG 3. Immune mechanistic assessments by year 4 treatment outcome status for eOIT-treated subjects. **A,** Change in egg-specific IgE levels over time. **B,** Change in egg-specific IgG_4 levels over time. **C,** Change in egg IgG_4/IgE ratio over time. **D,** Change in egg skin prick test response over time. **E,** Change in egg end point titration AUC over time. The egg IgG_4/IgE ratio was calculated by converting IgG_4 levels from milligrams of antibody per liter to nanograms per milliliter and converting IgE levels from kilounits of antibody per liter to nanograms per milliliter with the formula ($IgG_4 \times 1000$/IgE $\times$ 2.4). Egg end point titration AUC was calculated by adding together the scores from all 5 dilutions. Graphs designate *Success* as those subjects treated with eOIT who achieved SU versus *Failure* as those subjects treated with eOIT who did not achieve SU. *Stars* represent median values. *BL*, Baseline; *M*, month.

ability to predict is limited because the subjects did not have a baseline OFC. Thus natural history might play a role in this long-term analysis. This study was not adequately controlled to assess spontaneous resolution of egg allergy; however, we believe that the majority of a group of children included with increased egg-specific IgE levels and history of egg allergy at enrollment are unlikely to have tolerance naturally. Participants were also not assessed for reactivity to baked egg at baseline, and therefore some might have been baked egg tolerant at study entry.

Third, because of the expanding numbers of OIT-treated subjects achieving SU and subject dropout over the 4-year assessment period, the number of participants left to fully evaluate immunologic parameters was small. Thus the end-of-study analysis is likely underpowered to detect differences that might exist.

Finally, the long-term questionnaire data are based on recall reporting rather than prospective longitudinal data collection.

In summary, we demonstrate that the likelihood of SU to eOIT is enhanced with therapy exceeding 2 years, with those achieving SU demonstrating an increased likelihood of tolerating unbaked egg in their diet compared with those who did not achieve SU. Half of the eOIT-treated subjects achieved SU by year 4, a treatment response that might require ongoing intermittent dietary egg exposure to be maintained. For the majority of subjects, eOIT was administered with mild symptoms reported throughout the study. Extensive longitudinal immune evaluation did not identify biomarkers consistently predictive of treatment success. Future trials should focus on expansion of study population size and diversity of subjects with attention to immunologic assays that can help personalize OIT to enhance predictable success.

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Key messages

- **Longer duration of treatment with eOIT is associated with enhanced clinical response and increased likelihood of tolerating unbaked egg.**
- **Mild symptoms are commonly encountered throughout OIT treatment.**
- **Immune parameters studied do not consistently predict treatment response.**

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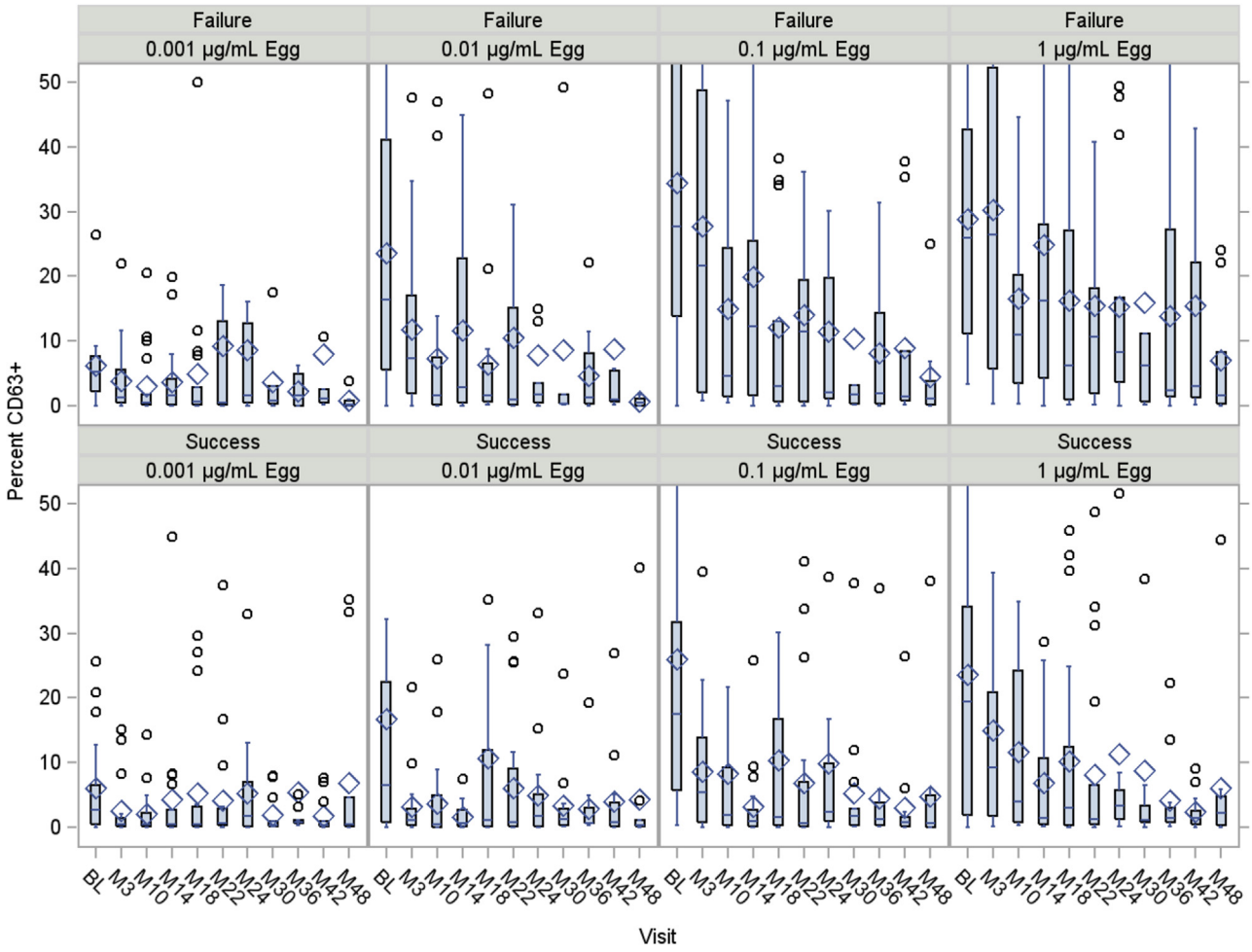

FIG E1. Effect of eOIT on basophil activation (percentage of $CD63^+$ basophils) by year 4 treatment outcome status for eOIT-treated subjects. *Top row*, eOIT treatment failure. *Bottom row*, eOIT treatment success. Graphs designate *Success* because those subjects treated with eOIT who achieved SU versus *Failure* as those subjects treated with eOIT who did not achieve SU. Cells were stimulated with 0.001 μg/mL egg white extract *(column 1)*, 0.01 μg mL egg white extract *(column 2)*, 0.1 μg/mL egg white extract *(column 3)*, or 1 μg/mL egg white extract *(column 4)*. *Diamonds* represent mean values. *BL*, Baseline; *M*, month.

TABLE E1. eOIT-treated subjects who discontinued dosing/withdrew from the study in years 3 to 4

Subject	Last OFC	Study day	Drug discontinuation/study withdrawal comments	LFQ	Status at last study contact
1	2 y, 10 g D-OFC	846	Study dosing discontinuation was initiated by the PI and confirmed by the subject's mother because of prolonged cessation of dosing caused by illness in combination with a repetitive history of study drug noncompliance.	Yes	Not fully restricting egg; eats baked egg only
2	2 y, 10 g D-OFC	907	Subject was eating egg, as well as taking their daily dose of egg powder, and was withdrawn by the PI.	Yes	Not restricting egg; eats concentrated and baked egg
3	2 y, 10 g D-OFC	951	Subject stated they could not take the doses any longer.	No	Does not eat whole egg; eats baked egg in diet without reactions
4*	2 y, 10 g SU-OFC	760	After failing the 2-year SU OFC, subject decided not to participate in the extension phase of the study and did not receive any eOIT dosing after 2 years.	Yes	Not restricting egg; eats concentrated and baked egg
5	2 y, 10 g SU-OFC	876	Subject refused to take any more doses because he or she was tired of it.	Yes	Fully restricting egg; does not eat concentrated or baked egg
6	2 y, 10 g SU-OFC	1148	Mom verbalized that she was going to stop dosing subject, and study contact could not be maintained.	No	Egg avoidance
7	3 y, 10 g D-OFC	1138	Subject passed their 3-year 10-g desensitization OFC but did not want to stop eOIT therapy to test SU.	Yes	Not fully restricting egg; eats baked egg only
8	3 y, 10 g D-OFC	1340	Subject stopped daily egg dose because of family circumstances. Subject was unable to complete end-of-study food challenges because he or she stopped doses without informing study staff. Eating egg in baked goods in their diet per mother's report.	No	Does not eat whole egg in diet; eats baked egg without reactions
9	3 y, 10 g SU-OFC	1222	Mother elected to discontinue dosing and pursue treatment of subject's egg allergy outside of the study.	Yes	Not restricting egg; eats concentrated and baked egg

D-OFC, Desensitization OFC; *PI*, principal investigator.
*Subject did not resume study dosing after 2-year OFC.

TABLE E2. Consortium of Food Allergy Research allergic reaction toxicity grading

Grade 1: Mild	Grade 2: Moderate	Grade 3: Severe	Grade 4: Life-threatening	Grade 5: Death
Transient or mild discomforts (<48 h), no or minimal medical intervention/therapy required. These symptoms might include pruritus, swelling or rash, abdominal discomfort, or other transient symptoms.	Symptoms that produce mild-to-moderate limitation in activity some assistance might be needed; no or minimal intervention/therapy is required. Hospitalization is possible. These symptoms can include persistent hives, wheezing without dyspnea, abdominal discomfort/increased vomiting, or other symptoms.	Marked limitation in activity, some assistance usually required; medical intervention/ therapy required, hospitalization is possible. Symptoms might include bronchospasm with dyspnea, severe abdominal pain, throat tightness with hoarseness, and transient hypotension among others. Parenteral medication or medications are usually indicated.	Extreme limitation in activity, significant assistance required; significant medical/therapy. Intervention is required; hospitalization is probable. Symptoms can include persistent hypotension and/or hypoxia with resultant decreased level of consciousness associated with collapse and/or incontinence or other life-threatening symptoms.	Death

TABLE E3. Percentage of pre–week 44 dosing symptoms per subject by 4-year treatment outcome status

	No.	Mean	SD	Median	Lower quartile	Upper quartile	Minimum	Maximum
Total no. of doses administered								
eOIT-SU	20	328.1	17.0	331.5	318.5	342.0	292.0	353.0
eOIT-D	11	329.5	19.9	334.0	315.0	348.0	297.0	356.0
eOIT-Not-D	9	191.9	142.9	163.0	70.0	334.0	15.0	339.0
Placebo	15	268.1	100.5	302.0	284.0	318.0	10.0	334.0
Any symptoms								
eOIT-SU	20	27.2	36.0	8.2	3.9	55.1	0.0	99.4
eOIT-D	11	20.0	25.4	10.6	7.1	15.7	2.4	87.8
eOIT-Not-D	9	26.1	16.7	23.6	14.8	26.3	8.0	53.9
Placebo	15	4.7	6.7	1.0	0.0	10.5	0.0	20.0
Any symptoms, excluding oral/pharyngeal								
eOIT-SU	20	13.1	19.5	5.2	2.7	13.2	0.0	67.1
eOIT-D	11	14.8	23.8	7.9	4.6	12.6	1.7	85.2
eOIT-Not-D	9	22.1	16.9	14.8	11.4	21.8	8.0	53.3
Placebo	15	4.6	6.8	0.9	0.0	10.5	0.0	20.0
Duration >30 min								
eOIT-SU	20	2.8	5.9	1.1	0.4	2.2	0.0	26.3
eOIT-D	11	8.7	21.9	2.0	0.6	4.0	0.0	74.4
eOIT-Not-D	9	4.0	3.9	3.6	1.4	4.5	0.0	13.3
Placebo	15	1.8	3.5	0.0	0.0	1.5	0.0	10.4
Treatment used								
eOIT-SU	20	1.7	2.2	1.2	0.5	1.7	0.0	10.3
eOIT-D	11	7.1	18.2	0.6	0.6	3.4	0.0	61.6
eOIT-Not-D	9	5.9	8.0	3.7	2.7	3.9	1.2	26.7
Placebo	15	1.1	2.6	0.0	0.0	0.6	0.0	10.0
Oral/pharyngeal symptoms								
eOIT-SU	20	20.5	32.4	3.0	0.8	30.3	0.0	96.0
eOIT-D	11	9.2	13.0	5.7	0.6	8.9	0.0	36.4
eOIT-Not-D	9	6.0	7.8	1.8	0.0	7.8	0.0	20.4
Placebo	15	0.2	0.5	0.0	0.0	0.0	0.0	1.6
Skin symptoms								
eOIT-SU	20	2.8	5.8	0.9	0.3	2.8	0.0	26.1
eOIT-D	11	7.7	18.5	1.1	0.6	3.0	0.0	63.1
eOIT-Not-D	9	2.7	2.4	1.8	0.6	4.9	0.0	6.7
Placebo	15	0.7	1.1	0.0	0.0	1.1	0.0	3.1
Respiratory symptoms								
eOIT-SU	20	6.5	15.0	0.9	0.5	3.0	0.0	61.8
eOIT-D	11	7.5	16.7	1.9	0.3	7.8	0.0	56.8
eOIT-Not-D	9	9.0	13.9	3.7	1.4	9.8	0.0	43.7
Placebo	15	2.8	4.5	0.3	0.0	6.8	0.0	13.4
Gastrointestinal symptoms								
eOIT-SU	20	4.3	6.2	1.8	0.3	6.3	0.0	24.4
eOIT-D	11	5.6	8.8	3.7	0.3	6.9	0.0	30.7
eOIT-Not-D	9	13.5	16.4	10.2	2.7	15.6	0.0	53.3
Placebo	15	1.6	5.1	0.0	0.0	0.9	0.0	20.0
Other symptoms								
eOIT-SU	20	2.0	5.0	0.5	0.3	1.0	0.0	21.8
eOIT-D	11	2.6	5.9	0.6	0.3	2.2	0.0	20.2
eOIT-Not-D	9	1.3	1.4	0.6	0.3	2.1	0.0	3.7
Placebo	15	0.7	1.1	0.0	0.0	1.4	0.0	3.5
Mild symptoms								
eOIT-SU	20	12.6	19.3	5.0	2.7	13.0	0.0	66.0
eOIT-D	11	13.6	23.6	5.6	4.6	11.0	1.3	83.5
eOIT-Not-D	9	20.5	15.6	14.8	10.0	20.0	6.1	47.6
Placebo	15	3.9	5.5	0.9	0.0	10.0	0.0	15.8
Moderate symptoms								
eOIT-SU	20	0.4	0.6	0.3	0.0	0.7	0.0	1.9
eOIT-D	11	1.2	1.6	0.6	0.3	1.7	0.0	5.6
eOIT-Not-D	9	1.6	2.0	0.9	0.6	1.8	0.0	6.7
Placebo	15	0.7	2.6	0.0	0.0	0.0	0.0	10.0

eOIT-D, eOIT-desensitized; *eOIT-Not-D*, eOIT-not desensitized.

TABLE E4. Logistic regression for eOIT-treated subjects where achieving SU counted as success with dosing symptoms before week 44 as predictors

Variable	*P* value, Wald χ^2	OR	OR lower CI	OR upper CI
Subjects with no moderate symptoms vs any moderate symptoms	.047	4.64	1.02	21.00
Subjects with no moderate symptoms vs ≥2 doses with moderate symptoms	.344	1.83	0.52	6.43
% Per subject with symptoms*	.632	1.01	0.98	1.03
% Per subject with symptoms, excluding oral/pharyngeal	.433	0.99	0.95	1.02
% Per subject, duration >30 min	.402	0.96	0.88	1.05
% Per subject, treatment used	.245	0.84	0.62	1.13
% Per subject, oral/pharyngeal symptoms	.133	1.03	0.99	1.06
% Per subject, skin symptoms	.456	0.97	0.90	1.05
% Per subject, respiratory symptoms	.719	0.99	0.95	1.04
% Per subject, gastrointestinal symptoms	.178	0.94	0.87	1.03
% Per subject, other symptoms	.977	1.00	0.88	1.15
% Per subject, mild symptoms	.511	0.99	0.96	1.02
% Per subject, moderate symptoms	.051	0.33	0.11	1.00

*Model had a significant lack of fit.

TABLE E5. Clinical outcome categories at time of LFQ

	LFQ-1, no. (%)	LFQ-2, no. (%)
eOIT	34/40 (85)	33/40 (83)
OIT-SU	18/20 (90)	18/20 (90)
OIT-D	9/11 (82)*	8/11 (73)
OIT-Not-D	7/9 (78)	7/9 (78)
Placebo	11/15 (73)	12/15 (80)†
Total	45/55 (82)	45/55 (82)

*One OIT-desensitized subject completed LFQ-1 only.
†One placebo-treated subject completed LFQ-2 only.

TABLE E6. LFQ egg exposure and reactions by 4-year egg consumption

	All eOIT				eOIT-SU				eOIT-D				eOIT-Not-D				Placebo*			
	LFQ-1		LFQ-2		LFQ-1		LFQ-2		LFQ-1		LFQ-2		LFQ-1		LFQ-2		LFQ-1		LFQ-2	
	No.	Percent	No.	Percent	No.	Percent	No.	Percent	No.	Percent	No.	Percent	No.	Percent	No.	Percent	No.	Percent	No.	Percent
Total subjects	34	100.0	33	100.0	18	100.0	18	100.0	9	100.0	8	100.0	7	100.0	7	100.0	11	100.0	12	100.0
No exposure to egg	4	11.8	4	12.1	0	0	0	0	1	11.1	1	12.5	3	42.9	3	42.9	6	54.5	4	33.3
Exposure to egg, no symptoms to egg	22	64.7	19	57.6	15	83.3	14	77.8	6	66.7	4	50.0	1	14.3	1	14.3	3	27.3	4	33.3
Exposure to egg, symptoms to egg	8	23.5	10	30.3	3	16.7	4	22.2	2	22.2	3	37.5	3	42.9	3	42.9	2	18.2	4	33.3

Placebo crossover subject 1: This subject enrolled in an eOIT open-label study protocol 51 days after completing the LFQ-1. He or she completed the LFQ-2 351 days after enrolling in the other study. On the LFQ-2, the subject reported that he or she eats concentrated egg in diet daily and indicated the amount of egg powder (which in Fig 2, *B*, is coded as "Other"). The subject did not consume baked egg in the diet and, other than the study treatment, is fully restricting egg. This subject was not counted as having improvement in egg consumption from LFQ-1 to LFQ-2. In the analysis the subject was counted as eating any egg in the diet for the LFQ-2 but was not counted as eating unbaked and baked egg in the diet (because he or she does not eat baked egg).

Placebo crossover subject 2: This subject crossed over to eOIT after year 2 per the site's institutional review board. The subject is eating concentrated egg and baked egg in their diet for the LFQ-1 and LFQ-2. The subject reported eating half a serving of concentrated egg weekly on LFQ-1 and half a serving less than monthly on the LFQ-2; he or she reported symptoms to concentrated egg rarely on the LFQ-1 and no symptoms on the LFQ-2. He or she reported eating 1 baked egg good (cookie, muffin, cake slice, pancake, or waffle) daily both on the LFQ-1 and LFQ-2. The subject reported symptoms to baked egg rarely on LFQ-1 and no symptoms on the LFQ-2. In the analysis the subject was counted as eating any egg in the diet and also counted as eating unbaked and baked egg in the diet for the LFQ-1 and LFQ-2.

eOIT-D, eOIT-desensitized; *eOIT-Not-D*, eOIT-not desensitized.

*Two placebo-treated subjects were crossed over to active treatment and received eOIT under another study protocol during the 4-year study period.

TABLE E7. Total IgE level, egg IgE level, egg IgG_4 level, and egg IgG_4/IgE ratio by 4-year egg consumption for eOIT-treated subjects

	Success				Failure			
	No.	**Median**	**Lower quartile**	**Upper quartile**	**No.**	**Median**	**Lower quartile**	**Upper quartile**
Egg IgE level (kU_A/L)								
BL	20	9.43	6.01	15.81	20	18.48	7.81	42.30
M10	20	5.10	3.73	12.91	20	7.36	4.74	18.87
M22	20	3.04	1.78	5.61	19	4.38	2.55	7.18
M48	12	2.86	1.53	5.18	9	1.71	1.19	3.23
Egg IgE: change from baseline								
BL	20	0.00	0.00	0.00	20	0.00	0.00	0.00
M10	20	−2.86	−7.65	−1.73	20	−6.43	−18.15	−2.05
M22	20	−6.36	−11.85	−3.96	19	−5.23	−22.74	−2.52
M48	12	−8.02	−11.37	−3.53	9	−5.39	−26.25	−4.44
Egg IgG_4 level (mg_A/L)								
BL	20	0.55	0.16	1.78	20	0.54	0.06	1.40
M10	20	49.71	21.45	69.20	20	17.30	5.43	33.62
M22	20	72.33	52.17	89.73	19	32.47	4.34	81.56
M48	12	32.80	23.05	46.30	9	9.43	2.29	32.80
Egg IgG_4: Change from baseline								
BL	20	0.00	0.00	0.00	20	0.00	0.00	0.00
M10	20	46.56	16.75	68.24	20	17.02	1.16	32.75
M22	20	71.08	51.46	88.60	19	32.42	0.29	80.95
M48	12	31.56	22.90	43.73	9	9.38	0.34	31.68
Egg IgG_4/IgE ratio								
BL	20	19.06	6.55	42.75	20	5.69	3.28	23.88
M10	20	2612.69	1028.27	4778.93	20	492.62	130.80	2069.84
M22	20	8981.02	4679.76	10339.76	19	2685.12	83.86	5909.44
M48	12	4720.20	2556.21	7284.80	9	4973.63	53.17	13011.70
Egg IgG_4/IgE ratio: Change from baseline								
BL	20	0.00	0.00	0.00	20	0.00	0.00	0.00
M10	20	2594.69	961.30	4637.21	20	476.73	94.59	2023.03
M22	20	8967.68	4190.39	10333.61	19	2680.15	81.27	5900.51
M48	12	4701.14	2542.88	7280.45	9	4972.86	48.36	13002.76

BL, Baseline; *M*, month.

TABLE E8. SPT responses by 4-year egg consumption for eOIT-treated subjects

	Success				Failure			
	No.	Median	Lower quartile	Upper quartile	No.	Median	Lower quartile	Upper quartile
Egg SPT response (mm)								
BL	20	10.50	7.50	16.00	20	11.25	7.50	15.75
M10	20	3.75	1.75	5.50	20	6.00	3.75	7.75
M22	20	0.25	0.00	4.25	18	6.25	4.00	7.00
M48	6	3.00	1.50	4.50	9	7.00	5.00	7.00
Change from baseline in Egg SPT response (mm)								
BL	20	0.00	0.00	0.00	20	0.00	0.00	0.00
M10	20	−6.50	−11.50	−4.25	20	−3.50	−8.50	0.00
M22	20	−9.50	−11.50	−6.25	18	−5.25	−8.50	0.00
M48	6	−4.50	−10.50	−1.00	9	−4.50	−5.00	−2.50

BL, Baseline; *M*, month.

TABLE E9. Egg end point titration AUC by 4-year egg consumption for eOIT-treated subjects

	Success				Failure			
	No.	**Median**	**Lower quartile**	**Upper quartile**	**No.**	**Median**	**Lower quartile**	**Upper quartile**
AUC								
Baseline	20	26.25	14.27	36.75	20	23.50	21.25	34.00
Month 22	20	0.50	0.00	5.00	18	8.25	5.00	13.50
Month 48	9	3.50	1.50	5.50	9	11.00	7.50	17.00
Change from baseline in AUC								
Baseline	20	0.00	0.00	0.00	20	0.00	0.00	0.00
Month 22	20	−22.50	−34.50	−14.77	18	−18.00	−25.00	−7.50
Month 48	9	−22.00	−27.00	−5.50	9	−13.50	−24.00	−11.50