TT-06 STATISTICAL ANALYSIS PLAN COVER PAGE

Official trial title	Efficacy and safety of the SQ tree sublingual immunotherapy tablet in children and adolescents (5 through 17 years of age) with moderate to severe allergic rhinitis and/or conjunctivitis induced by pollen from birch and trees belonging to the birch homologous group
NCT number	NCT04878354
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Statistical Analysis Plan

Trial ID: TT-06

Efficacy and safety of the SQ tree sublingual immunotherapy tablet in children and adolescents (5 through 17 years of age) with moderate to severe allergic rhinitis and/or conjunctivitis induced by pollen from birch and trees belonging to the birch homologous group

Sponsor: ALK A/S
Bøge Alle 6-8
DK-2970 Hørsholm

Investigational medicinal product: SQ tree SLIT-tablet

Phase: III

EudraCT No.: 2020-004372-17

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ALK approval of statistical analysis plan

Trial statistician:

[redacted]

Signature _____ Date _____

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[redacted]

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Version History
This SAP for the trial TT-06 is based on the CTP version 3.0 dated 24-MAY-2022.

SAP Version	Date	Change	Rationale
1	**19-SEP-2023**	**Not applicable**	**Original version**

1 List of abbreviations

AE	adverse event
AHPS	alder-hazel pollen season
AR/C	allergic rhinoconjunctivitis
BPS	birch pollen season
eCRF	electronic case report form
CTP	clinical trial protocol
DMS	daily rhinoconjunctivitis medication score
DSS	daily rhinoconjunctivitis symptom score
DPS	data point sets
E	exclusion criteria
EAACI	European Academy of Allergy and Clinical Immunology
EOE	eosinophilic oesophagitis
EOT	end of treatment
ESI	event of special interest
EudraCT	European Union Drug Regulating Authorities Clinical Trials Database
FAS	full analysis set
[redacted]	[redacted]
[redacted]	[redacted]
FDA	Food and Drug Administration
FEV_1	forced expiratory volume in 1 second
FVC	forced vital capacity
GLMM	generalized linear mixed model
[redacted]	[redacted]
H	hypothesis

ICH	International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use
IE	intercurrent event
IgE	immunoglobulin E
IgE-BF	IgE-blocking factor
IgG_4	immunoglobulin G_4
IMP	investigational medicinal product
IRT	interactive response technology
LME	linear mixed effects
MAR	missing at random
MedDRA	Medical Dictionary for Regulatory Activities
MNAR	missing not at random
OPS	oak pollen season
PD	protocol deviation
PT	preferred term
RQLQ	rhinitis quality of life questionnaire
SABA	short-acting β_2-agonist
SAE	serious adverse event
SAF	safety analysis set
SAP	statistical analysis plan
SAS	statistical analysis system
SD	standard deviation
SOC	system organ class
SPT	skin prick test
SLIT	sublingual immunotherapy
SLIT-tablet	sublingual immunotherapy tablet
SUB	subgroup analysis set
TAS	total analysis set
TC	telephone call
TCS	total combined score
TEAE	treatment-emergent adverse event
TPS	tree pollen season
UV	unscheduled visit

V	visit
WHO	World Health Organisation

2 List of definitions

Cohort	A group of subjects receiving treatment during the pollen seasons of a particular calendar year (e.g., cohort 1 was treated during the pollen seasons of 2022)
Completed subject	A randomised subject is considered completed if he/she has not discontinued the trial before visit 6.
Concomitant medication	All medications continued by a subject on entry to the trial and all medications initiated during the trial including ALK rescue medication.
Date of last contact	Date of the last contact, either by telephone or in a visit.
End of trial	The overall end of the trial is defined as the date of last follow-up phone visit (TC4) for the last subject in the trial globally.
Estimand	A precise description of the treatment effect reflecting the clinical question of interest posed by the trial objective. It summarises at a population-level what the outcomes would be in the same patients under different treatment conditions being compared (ICH 2020).
ESI	ESIs are selected AEs that are considered critical for the evaluation of the product's safety profile and for which additional data will be collected.
IMP	A pharmaceutical form of an active substance or placebo being tested or used as a reference in a clinical trial, including products already with a marketing authorisation but used or assembled (formulated or packaged) in a way different from the authorised form, or when used for an unauthorised indication, or when used to gain further information about the authorised form.
IE	Events occurring after treatment initiation that affect either the interpretation or existence of the measurements associated with the clinical question of interest. It is necessary to address IEs when describing the clinical question of interest in order to precisely define the treatment effect that is to be estimated (ICH 2020).
IRT	Automated system that can be used for managing randomisation, subject enrolment, and trial supply management in a clinical trial.
Rescue medication	Medicinal products provided by ALK when the efficacy of the IMP is not sufficient or likely to cause an adverse event to the subject or to manage an emergency situation in relation to allergy symptoms in agreement with the EMA Definition of IMPs and use of AMPs

	consultation document (EMA 2016).
TEAEs	An adverse event with start date on or after the time of first IMP administration and no later than 7 days after last IMP administration.
Trial discontinuation date	Date of subject trial discontinuation. In case of subjects lost to follow-up, the trial discontinuation date is defined as the date the investigator/sponsor decides to discontinue the subject from the trial.

3 Introduction

The statistical analysis plan is a supplement to the statistical section in the CTP and provides additional details regarding estimands (where applicable), analysis sets, endpoints, and the statistical analyses.

Changes to the analyses and changes that might impact the analyses described in the CTP are documented in section 6.11.

Supporting documentation is provided in section 9. Section 9.1 ‘Definition of pollen seasons’ contains the definition of start and end dates for the different pollen seasons used in the trial. Section 9.2 ‘Derivations for the trial’ contains details about the derivations used in the trial.

In addition to above, the SAP includes several appendices (section 11). Appendix A includes a list of terms used to identify local administration site TEAEs, further details about the statistical analyses are provided in Appendix B, examples of SAS code are listed in Appendix C, and Appendix D includes the start and end dates of the pollen station.

This SAP has been written and approved before database lock and unblinding.

3.1 Objectives, endpoints, and estimands

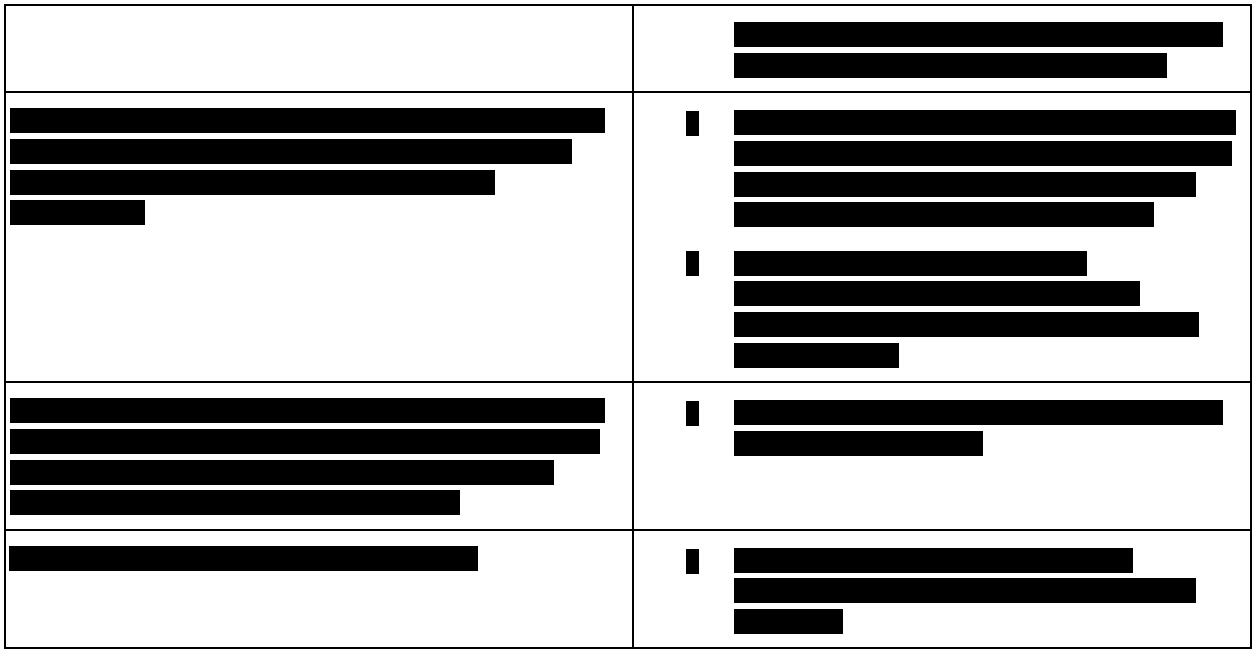
Table 1: Objective and endpoints

▪ **Objectives**	▪ **Endpoints**
▪ Primary	
To compare the efficacy of the SQ tree SLIT-tablet to placebo in the treatment of moderate to severe AR/C induced by pollen from birch and trees belonging to the birch homologous group in children and adolescents (5 through 17 years of age) based on the average TCS[1] during the BPS.	• Average TCS during the BPS
▪ Key secondary	
To compare the efficacy of the SQ tree SLIT-tablet to placebo based on average TCS during the TPS, and average rhinoconjunctivitis daily symptom score and average rhinoconjunctivitis daily medication score during the BPS and TPS.	• Average TCS during the TPS • Average DSS during the BPS • Average DSS during the TPS • Average DMS during the BPS • Average DMS during the TPS
▪ Secondary	

To compare the safety and tolerability of the SQ tree SLIT-tablet to placebo.	• TEAEs
	• ESIs
	  ○ Systemic allergic reactions including anaphylaxis
	  ○ Events treated with adrenaline/epinephrine
	  ○ Severe local swelling or oedema of the mouth and/or throat
	  ○ EoE
	  ○ Oral allergy syndrome
	  ○ Asthma exacerbations
	• Vital signs, physical examination, lung function tests and clinical laboratory values during treatment and at the end-of-treatment visit
	• Treatment-related local administration site TEAEs (defined according to a pre-specified list of MedDRA preferred terms)

[1] daily total combined score = daily symptom score + daily medication score

To compare the efficacy of the SQ tree SLIT-tablet to placebo using additional endpoints based on daily allergic rhinoconjunctivitis symptoms and rescue medication use.	• Average TCS, average DSS and average DMS during the AHPS and seasons OPS • Average TCS (EAACI scoring) during the BPS and TPS • Number of severe days during the BPS and TPS • Number of well days during the BPS and TPS • Number of symptom-free days during the BPS and TPS • Percentage of patients free of symptoms/signs and with no use of rescue medication during the BPS and TPS
To compare the efficacy of the SQ tree SLIT-tablet to placebo based on assessments of quality of life.	• Average weekly overall RQLQ score during the BPS and TPS (12-17 years only) • Overall PRQLQ score during the BPS (5-11 years only) • TSQM-9 evaluations
To compare the efficacy of the SQ tree SLIT-tablet to placebo based on patient treatment satisfaction.	• Patient-rated global evaluation of treatment efficacy
To compare the effect of the SQ tree SLIT-tablet to placebo on immunological parameters to birch, alder, hazel and oak pollen.	• Change from baseline in birch specific IgE, and IgG_4 • Change from baseline in alder, hazel and oak specific IgE and IgG_4 (for a subgroup of randomised subjects only) • Change from baseline in birch, alder and hazel specific IgE-BF (for a subgroup of randomised subjects only)
▪ Exploratory	
[redacted]	[redacted]



3.1.1 Estimands

A primary and secondary estimand is defined for each of the primary and key secondary endpoints.

3.1.1.1 Primary estimand

The primary estimand will be referred to as the 'trial product estimand'. The trial product estimand assesses the anticipated effect of the SQ tree SLIT-tablet if it is taken as instructed. This estimand is considered to be of most relevance to the patient, as it describes the potential benefit they could obtain from the SQ tree SLIT-tablet if they adhere to treatment for the planned duration. Adherence to treatment is considered important in allergy immunotherapy since the optimal effect of allergy immunotherapy is expected when taken consistently for a longer period.

Using the framework proposed in the ICH E9 (R1) addendum, the trial product estimand can be described by:

A. **Treatment:** SQ tree SLIT-tablet or placebo, with rescue medication taken as required
B. **Population**: Children and adolescents (5-17 years of age) with moderate to severe AR/C caused by pollen from birch
C. **Variable**: Average TCS during the BPS
D. **How to account for intercurrent events:** under the hypothetical situation where all subjects have completed treatment for the planned duration (hypothetical strategy)
E. **Population summary**: Absolute difference between treatments

A hypothetical strategy will be used to address the intercurrent event of discontinuation of trial treatment. As such, if a subject discontinues trial treatment, only diary data up until the time of

discontinuation will be included in the analysis. Any diary data recorded after discontinuation of trial treatment will be excluded from the analysis. Details of the handling of IEs and missing data are provided in section 6.3.3.1.

3.1.1.2 Secondary Estimand

The secondary estimand will be referred to as the 'treatment policy estimand'. The treatment policy estimand assesses the treatment effect regardless of adherence to treatment and provides a broad perspective of the treatment effect in clinical practice in the selected population of patients. This estimand is in line with to the 'intention to treat' principle and provides a robust assessment of the efficacy of the SQ tree SLIT-tablet.

The treatment policy estimand can similarly be described by:

A. **Treatment:** SQ tree SLIT-tablet or placebo, with rescue medication taken as required
B. **Population**: children and adolescents (5-17 years of age) with moderate to severe AR/C caused by pollen from birch
C. **Variable**: Average TCS during the BPS
D. **How to account for intercurrent events:** regardless of whether subjects complete treatment for the planned duration (treatment policy strategy)
E. **Population summary**: Absolute difference between treatments

A treatment policy strategy will be used to address the intercurrent event of discontinuation of trial treatment. As such, if a subject discontinues trial treatment, all available diary data will be included in the analysis, regardless of adherence to treatment. Details of the handling of IEs and missing data are provided in section 6.3.3.2.

3.1.1.3 Intercurrent events

The IE that has been identified as relevant is discontinuation of trial treatment. No additional IEs were identified to have an impact on the overall efficacy assessment.

Due to the long duration of allergy immunotherapy clinical trials, rescue medication is dispensed to every subject for ethical reasons. The use of rescue medication is, therefore, part of the treatment policy and as such, it is not classified as an IE for this trial.

The number of IEs will be summarised by reason for discontinuation of trial treatment and treatment.

3.1.1.4 Estimands for key secondary objectives

The trial product estimand and treatment policy estimand, specified for the primary objective in sections 3.1.1.1 and 3.1.1.2 respectively, are defined similarly for each of the key secondary objectives, where the relevant key secondary endpoint of interest is specified in attribute C.

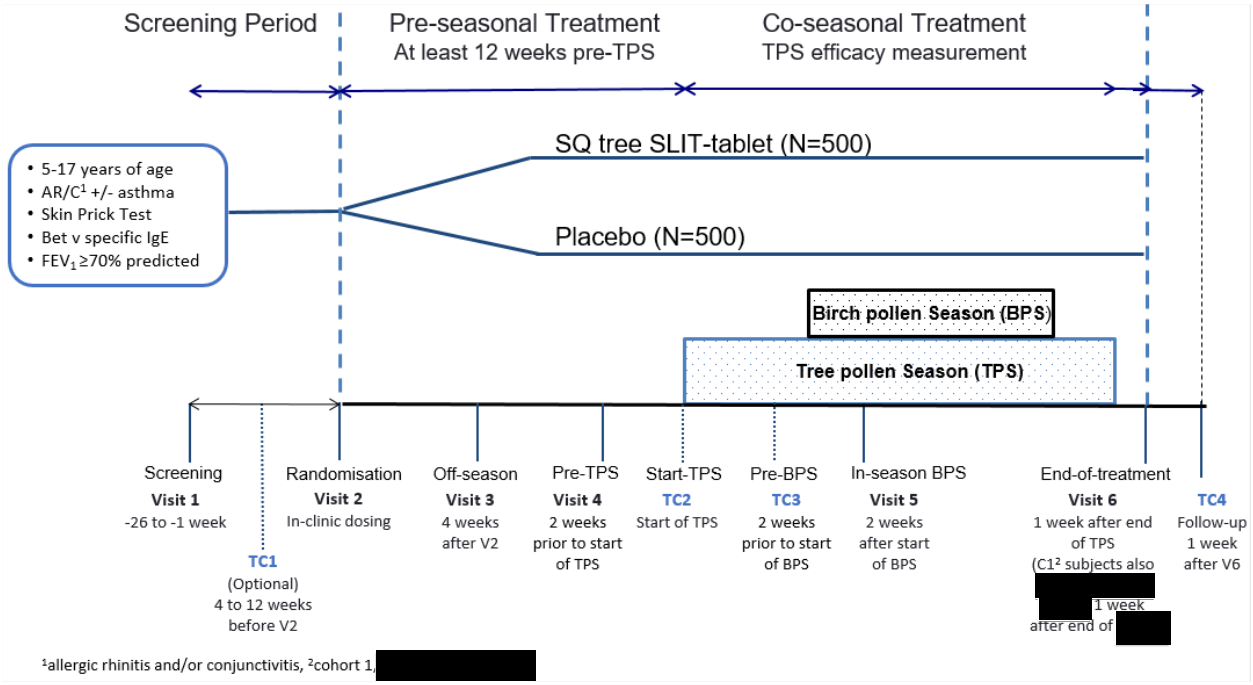
3.2 Trial design

This is a phase III, randomised, parallel-group, double-blind, placebo-controlled, multi-regional trial in children (5-11 years) and adolescents (12-17 years) with AR/C, with or without asthma, induced by pollen from birch. Based on previous trials, it is anticipated that most subjects allergic to birch will also have AR/C induced by pollen from the birch homologous group.

Approximately 1000 subjects were to be randomised (1:1) and receive treatment with the SQ tree SLIT-tablet or placebo. The randomisation was stratified by geographical location (country) and by age group (5-11 years and 12-17 years).

The trial consisted of 3 periods: a screening period, a treatment period, which included pre-seasonal and co-seasonal treatment, and a follow-up period (Figure 1). The trial consisted of 2 cohorts; subjects in cohort 1 were randomised in 2021 and received treatment during the pollen seasons of 2022; subjects in cohort 2 were randomised in 2022 and received treatment during the pollen seasons of 2023.

Figure 1: Overall trial design



The screening visit took place up to approximately 6 months prior to randomisation. Once randomised, subjects were treated for at least 12 weeks prior to the start of the TPS. Treatment continued until 1 week after the end of the TPS, corresponding to up to approximately 12 months of treatment.

[redacted]Open-label rescue medication for allergic rhinoconjunctivitis was provided during the TPS [redacted]. Open-label asthma medication was provided to subjects with asthma during the TPS [redacted]. A follow-up phone visit was planned 1 week after the final treatment.

3.2.1 Flow chart

The flow chart of the trial assessments by visits are shown below in Table 2.

Table 2: Flow chart

Visit ID:	**V1**	**TC1**[2]	**V2**	**V3**	**V4**	**TC2**	**TC3**	**V5**	**TC** [redacted]	**V6**	**TC4**	**UV**[3]
Name of visit	Screen-ing	Retention	Randomi-sation	Off season	Pre-TPS	Start of TPS	Pre-BPS	In season BPS	Only for cohort 1 subjects also [redacted]	End-of-treatment	Follow-up	Unsched-uled
Scheduled week	1-26 weeks prior to V2	4-12 weeks prior to V2	At least 12 weeks prior to expected start of TPS[4]	4 weeks after V2	2 weeks prior to expected start of TPS		2 weeks prior to expected start of BPS	2 weeks after expected start of BPS[5]	1 week after expected end of TPS	1 week after expected end of TPS Cohort 1 subjects also [redacted] 1 week after expected end of [redacted][6]	1 week after V6	
Visit window				+7 days	-14 to +7 days	-4 days	+7 days	+7 days	+7 days	+7 days	+7 days	
Administrative procedures												
Informed consent[7]	X											

[2] Only required if V1 (screening) is performed more than 12 weeks prior to randomisation.

[3] Unscheduled visits should be conducted as necessary. The evaluations/examinations listed with brackets should only be performed if deemed necessary by the investigator.

[4] Randomisation (V2) must be performed at least 12 weeks prior to expected TPS but no later than 31 October.

[5] Visit 5 can be scheduled based on the actual BPS start date.

[6] In cohort 1 subjects also [redacted] V6 is to be performed 1 week after the expected end of [redacted]. End of [redacted] can be no later than 31 August 2022.

[7] Obtain written informed consent for the trial, storage of serum samples in the ALK Research Biobank, and for pharmacogenetics testing before any other trial procedures are performed.

Visit ID:	**V1**	**TC1**[2]	**V2**	**V3**	**V4**	**TC2**	**TC3**	**V5**	**TC** [redacted]	**V6**	**TC4**	**UV**[3]
Name of visit	Screen-ing	Retention	Randomi-sation	Off season	Pre-TPS	Start of TPS	Pre-BPS	In season BPS	Only for cohort 1 subjects also [redacted]	End-of-treatment	Follow-up	Unsched-uled
Inclusion/exclusion criteria	X		X									
Randomisation			X									
Demography	X											
Smoking habit	X											
Medical history	X											
Record prior and concomitant medication	X	X	X	X	X	X	X	X	X	X	X	X
TC to ensure that subject is still interested in the trial		X										
Dispense IMP			X		X			X				(X)
Instruct the subject in use of IMP			X									(X)
Collect IMP and perform drug accountability					X			X		X		
Perform IMP compliance check				X	X			X		X		
Dispense rhinoconjunctivitis and asthma rescue medication					X			X				(X)
Collect rescue medication and perform drug accountability								X		X		
Handout eDiary					X							

Visit ID:	**V1**	**TC1**[2]	**V2**	**V3**	**V4**	**TC2**	**TC3**	**V5**	**TC** [redacted]	**V6**	**TC4**	**UV**[3]
Name of visit	Screen-ing	Retention	Randomi-sation	Off season	Pre-TPS	Start of TPS	Pre-BPS	In season BPS	Only for cohort 1 subjects also [redacted]	End-of-treatment	Follow-up	Unsched-uled
Instruct in the use of the eDiary incl. training in symptom scoring					X	X	X	X	X			
Show and/or discuss trial video					X	X	X	X				
Inform that TPS is starting						X						
Clinical procedures/assessments												
Physical examination	X		X							X		(X)
Height			X							X		(X)
Weight	X		X							X		(X)
Vital signs	X									X		(X)
Lung function test[8]			X							X		(X)
SPT[9]	X		(X)									
Assess AEs	X	X	X	X	X	X	X	X	X	X	X	X
Assess symptoms of eosinophilic oesophagitis	X		X	X	X	X	X	X	X	X	X	X
Intake of IMP at clinic			X									

[8] Measure FVC and FEV_1 while subject is on their usual asthma medication (if applicable) following at least a 6 hour washout of SABA.
[9] To be taken after a urine pregnancy test (if applicable). A positive SPT is defined in the SPT guideline. Briefly, a positive SPT is defined as a wheal size of ≥3 mm. If medication that could interfere with the SPT, according to prohibited and restricted medication table, has not been washed out, the SPT must be performed at V2 after the interfering medication has been washed out.

Visit ID:	**V1**	**TC1**[2]	**V2**	**V3**	**V4**	**TC2**	**TC3**	**V5**	**TC** [redacted]	**V6**	**TC4**	**UV**[3]
Name of visit	Screen-ing	Retention	Randomi-sation	Off season	Pre-TPS	Start of TPS	Pre-BPS	In season BPS	Only for cohort 1 subjects also [redacted]	End-of-treatment	Follow-up	Unsched-uled
All subjects (except for cohort 1 subjects with [redacted]) **Daily eDiary recording:** •AR/C symptoms •AR/C medication • [redacted] **Weekly eDiary recording:** •RQLQ(S)[10]						X ----------X						
Cohort 1 subjects with [redacted]						X ----------X						

[10] RQLQ(S) should be performed only for subjects who were 12-17 years at the randomisation visit.

Visit ID:	**V1**	**TC1**[2]	**V2**	**V3**	**V4**	**TC2**	**TC3**	**V5**	**TC** [redacted]	**V6**	**TC4**	**UV**[3]
Name of visit	Screen-ing	Retention	Randomi-sation	Off season	Pre-TPS	Start of TPS	Pre-BPS	In season BPS	Only for cohort 1 subjects also [redacted]	End-of-treatment	Follow-up	Unsched-uled
Daily eDiary recording: •AR/C symptoms AR/C medication												
Cohort 1 subjects with [redacted] **Daily eDiary recording:** •[redacted] •[redacted] [redacted] •[redacted] •[redacted] •[redacted] **Weekly eDiary recording:** RQLQ(S)[10]						X ----------X						
Review eDiary recordings and compliance						X----------X						
Collect eDiary										X		

Visit ID:	**V1**	**TC1**[2]	**V2**	**V3**	**V4**	**TC2**	**TC3**	**V5**	**TC** [redacted]	**V6**	**TC4**	**UV**[3]
Name of visit	Screen-ing	Retention	Randomi-sation	Off season	Pre-TPS	Start of TPS	Pre-BPS	In season BPS	Only for cohort 1 subjects also [redacted]	End-of-treatment	Follow-up	Unsched-uled
PRQLQ[11]								X				
All subjects (except for cohort 1 subjects with [redacted]): •Global evaluation of treatment efficacy •TSQM-9										X		
Cohort 1 subjects with [redacted]**:** •Global evaluation of treatment efficacy TSQM-9									X			
Laboratory procedures/assessments												
Blood sample for specific IgE against *Bet v*	X											(X)
Blood and urine samples for safety laboratory assessments	X									X		(X)
Urine pregnancy test[12]	X		X	X	X			X		X		(X)

[11] Perform PRQLQ interview, only for subjects who were 5-11 years at the randomisation visit.
[12] For female subjects of childbearing potential only, additional urine pregnancy tests should be performed during the trial, if a menstrual period is missed.

Visit ID:	**V1**	**TC1**[2]	**V2**	**V3**	**V4**	**TC2**	**TC3**	**V5**	**TC** [redacted]	**V6**	**TC4**	**UV**[3]
Name of visit	Screen-ing	Retention	Randomi-sation	Off season	Pre-TPS	Start of TPS	Pre-BPS	In season BPS	Only for cohort 1 subjects also [redacted]	End-of-treatment	Follow-up	Unsched-uled
Blood sample for Immunological assessments	X				X					X		(X)
Blood sample for biobank[13]	X				X					X		(X)
Blood sample for pharmacogenetics[14]										X		(X)

[13] Only for subjects where informed consent for biobank sampling has been obtained.
[14] Only for subjects where informed consent for pharmacogenetics sampling has been obtained.

3.2.2 Actual trial timelines

For cohort 2 the randomisation window was extended based on regional pollen data instead of dates for national pollen data used for cohort 1.

The schedule of cohorts is presented in Table 3.

Table 3:Schedule of cohorts

	Cohort 1	Cohort 2
First subject first visit	08-Apr-2021	10-Mar-2022
Last subject randomised	01-Nov-2021	21-Dec-2022
Last subject last visit	24-Oct-2022	31-July-2023

3.2.3 Randomisation

The randomisation list has been generated by the IRT provider and will not be accessible to trial personnel involved in the conduct of the trial, until the database has been locked. The randomisation was administered centrally via IRT and was stratified based on country and age group.

The randomisation codes will only be made available for data analysis when the clinical database has been locked and all PDs have been identified, evaluated, categorised, and closed.

4 Statistical hypotheses

4.1.1 Multiplicity adjustment

For the trial product estimand based on the primary efficacy endpoint and the key secondary efficacy endpoints, the analyses for the main estimators will be controlled for multiplicity to ensure a maximum overall type I error rate of 5% in the hypothesis testing of these endpoints.

The control for multiplicity will be performed through hierarchical testing, using a pre-specified order of hypothesis to be tested. For each endpoint, the null hypothesis to be tested is the hypothesis of no difference between placebo and active SQ tree SLIT-tablet. Let μ_1 denote the mean in the SQ tree SLIT-tablet group, and μ_2 denote the mean in the placebo group. Then the null hypothesis (H_0) and the alternative hypothesis (H_A) are given as follows:

H_0: $\mu_1 = \mu_2$ and H_A: $\mu_1 \neq \mu_2$

The order of the hypotheses to be tested is:

1. The primary efficacy analysis of average TCS during BPS
2. The key secondary efficacy analysis of average TCS during TPS
3. The key secondary efficacy analysis of average DSS during BPS
4. The key secondary efficacy analysis of average DSS during TPS
5. The key secondary efficacy analysis of average DMS during BPS
6. The key secondary efficacy analysis of average DMS during TPS

The first hypothesis in the hierarchy will be tested at a two-sided 5% significance level. Testing will proceed to the second hypothesis only if the first test is statistically significant ($p<0.05$). Similarly, testing will proceed to the next level of the hierarchy, only if the previous comparison is statistically significant at the 5% level. If a test is not statistically significant ($p >0.05$) then no further testing within the hierarchy will be conducted. Thus, the 2nd, 3rd, 4th, 5th, and 6th hypothesis tests in the hierarchy will only be conducted if all previously tested hypotheses are statistically significant at the 5% level.

Furthermore, analysis of the quality of life endpoints will be included as the final steps in the testing hierarchy, where the order of the hypotheses to be tested is:

7. The secondary efficacy analysis of average weekly overall RQLQ score during the BPS
8. The secondary efficacy analysis of average weekly overall RQLQ score during the TPS
9. The secondary efficacy analysis of overall PRQLQ score during the BPS

Additional endpoints and analyses are supportive in nature and are not controlled for multiplicity.

5 Analysis sets

For the purposes of analysis, the analysis sets presented in

Table 4 are defined.

Table 4: Analysis set definitions

Participant analysis set	Description	Application
TAS	All subjects who signed informed consent and thus includes screening failures.	Outputs related to disposition.
FAS	All randomised subjects. Subjects will be analysed as randomised i.e., according to their randomised assignment of treatment.	Outputs related to baseline and efficacy outputs.
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
SAF	All randomised subjects who received at least one dose of IMP. Subjects will be analysed as treated i.e., according to treatment they actually received.	Outputs related to safety reporting.
SUB	Subjects in a random subset sample stratified by treatment group and age group for cohort 1.	Endpoints related to alder, hazel and oak specific IgE and IgG_4, and birch, alder, and hazel specific IgE-BF.

[redacted]

[redacted]

The following DPS are defined:

DPS1: For subjects who complete the trial treatment, all data is included.

For subjects who discontinue trial treatment, all data collected after the time of trial treatment discontinuation will not be included for the trial product estimand.

DPS2: All data is included.

FAS and DPS1 are used to estimate the trial product estimand, FAS and DPS2 are used to estimate the treatment policy estimand and observed case.

6 Statistical analyses

Statistical analyses will be carried out by ALK, Biometrics, Hørsholm, Denmark. All computation will be performed using SAS, SAS® version 9.4 or later.

The analyses described in this section along with the supporting information supplied in section 9 and Appendix B specify the statistical analyses.

6.1 General considerations

Continuous data will be summarised by treatment group using mean, standard deviation, median, minimum, and maximum. Categorical data will be summarised by treatment using frequency tables displaying numbers and percentages. For the tabulations of data by visit an observed case approach is used.

All the statistical tests described in this document use a significance level of 5% and all tests and confidence intervals are two-sided. The null hypothesis is the hypothesis of no difference between treatment groups and the alternative to the null hypothesis is the hypothesis of difference. For each analysis the specific test is described in more detail.

The number of imputations will be increased from 1000 to 10000 if the actual significance level of the performed test is estimated to be between 4%-6%. This will be done to ensure the precision of estimated standard error, and thus the right hypothesis testing conclusion will be made.

Baseline values are defined as the latest available observation at or prior to the date of the first administration of IMP, unless otherwise defined.

Collected age at randomisation will be used to derive the medication score throughout the pollen season.

6.2 Overall strategy of efficacy analyses

An overall overview of the strategies for efficacy analyses is listed in Table 5 . A detailed description of each endpoint will be described in following sections.

Table 5: Overall overview of strategies for efficacy analyses

	Hypothetical strategy	Treatment policy strategy	observed case
Primary endpoint	- Main estimator - Sensitivity estimator - Sensitivity estimator (tipping point)	- Main estimator - Sensitivity estimator	√
Key secondary 2-6[1]	- Main estimator - Sensitivity estimator	- Main estimator - Sensitivity estimator	√
Secondary 7-9[1]	- Main estimator - Sensitivity estimator		√
Secondary endpoints[2]	- Main estimator		
Immunology parameters			√
[redacted]			

[1] Key secondary and secondary endpoints in the testing hierarchy
[2] Binomial endpoints will be analysed under the hypothetical strategy using the same imputation rules as for the primary endpoints, however the estimator is different.

6.3 Primary endpoint analysis

6.3.1 Definition of endpoint

The primary endpoint is the average TCS during the BPS. The average TCS during the BPS is the average of the daily sum of the DSS and DMS during the BPS.

6.3.2 Main estimator, sensitivity estimator(s) and statistical analysis for the trial product and treatment policy estimands

The main estimator for both estimands will be the absolute treatment difference.

The main estimator for both estimands will be based on an identical method of statistical analysis (specified below), although the actual data used in each analysis may vary according to the different strategies taken for the handling of intercurrent events and missing data. Further details of the different strategies for each estimand are provided in sections 6.3.3.1, 6.3.3.2, and 6.4.1.

Analyses will be based on all randomised subjects. Subjects for whom the efficacy endpoint is missing will be included in the analysis through multiple imputation, where missing data will be

imputed from the same age group according to the approaches specified in sections 6.3.3.1, 6.3.3.2, and 6.4.1. Imputation will use the method of unrestricted random sampling with replacement (seed=7127) and 1000 multiply imputed datasets will be created. For each dataset, the efficacy endpoint will be square root transformed in order for the residuals to fulfil the underlying assumption of following the so-called statistical normal distribution and analysed using a linear mixed effects model assuming unequal variances in the 2 treatment groups. The model will include treatment, cohort (1, 2), and age group (5-11 years, 12-17 years) as fixed effects and pollen station within cohort as a random effect, with different residual errors specified for each treatment group. The model will be estimated using REML, and the denominator degrees of freedom will be calculated using the Kenward and Roger's approximation (Kenward and Roger 1997).

Rubin's rule is used to combine the 1000 analysis results across the multiple imputed datasets. The p-value for the absolute difference is reported as the test result.

The results are back-transformed as follows; after Rubin's rule is used to combine results across imputations, estimated least square means on the square root transformed scale are output along with the associated covariance matrix. The absolute difference is calculated by squaring the adjusted means, and then calculating the difference. For the absolute difference the SE is approximated by using the first order Delta method (first order Taylor approximation), and from this the 95% CI is calculated. For the relative difference, Fieller's theorem (Fieller 1954) is used to calculate the 95% CI. More details can be found in Appendix B.

6.3.3 Handling of intercurrent events and missing data

Subjects for whom the endpoint is missing because of missing diary data or missing pollen exposure (see Section 9.1), multiple imputation will be from own treatment group in case of no intercurrent event has occurred, otherwise below imputation rules will be applied. Data is assumed to be MAR.

Subjects with imputed endpoint, will be included in the statistical model with their original treatment group, age group, cohort, and pollen station.

6.3.3.1 Handling of intercurrent events and missing data for the trial product estimand

For the trial product estimand, diary data occurring after discontinuation of trial treatment will be excluded from the derivation of the primary endpoint (see section 9.2.2.3). Subjects for whom the primary endpoint is missing (because of either missing diary data or the exclusion of diary data due to discontinuation of trial treatment) will be included in the analysis through multiple imputation under the hypothetical situation where subjects continued to take trial treatment as planned.

Multiple imputation for subjects missing the primary endpoint will be conducted as follows:

- For subjects that discontinue trial treatment due to lack of efficacy, multiple imputation of the missing endpoint will be from the placebo group. This assumes that had the subject continued to take trial treatment, they would have experienced similar efficacy to subjects in the placebo arm. Data is assumed to be MNAR.
- For subjects that discontinue trial treatment due to treatment-related AEs, multiple imputation of the missing endpoint will be from their own treatment group. This assumes that the level of efficacy that would have been experienced by the subject if they had

continued to take trial treatment is unrelated to the occurrence of the AE and, if they had continued to take trial treatment, they would have experienced similar efficacy to subjects in their own treatment group. Data is assumed to be MAR.

- For subjects that discontinue trial treatment due to any other non-treatment related reason, multiple imputation of the missing endpoint will be from their own treatment group. This assumes that the level of efficacy that would have been experienced by the subject had they continued to take trial treatment is not related to their discontinuation of trial treatment. Data is assumed to be MAR.

6.3.3.2 Handling of intercurrent events and missing data for the treatment policy estimand

For subjects discontinuing trial treatment, all diary data (including data collected after discontinuation of trial treatment) will be included in the derivation of the primary endpoint (see section 9.2.1.4). Subjects for whom the primary endpoint is missing will be included in the analysis through multiple imputation as follows:

- For subjects discontinuing trial treatment due to treatment-related AEs or lack of efficacy, multiple imputation of the missing endpoint will be from the placebo group. This assumes that missing data following treatment-related discontinuation of trial treatment is MNAR and that subjects no longer taking active treatment for these reasons would have shown similar efficacy to subjects receiving placebo.
- For subjects discontinuing trial treatment due to non-treatment related reasons, multiple imputation of the missing endpoint will be their own treatment group. This assumes that the missing data are MAR.

6.3.3.3 Sensitivity analyses

The sensitivity estimator for both estimands will be the absolute treatment difference.

The sensitivity estimator for both estimands will be based on an identical method of statistical analysis although the actual data used in each analysis may vary according to the different strategies taken for the handling of intercurrent events and missing data. Further details of the different strategies for each estimand are provided in sections 6.3.3.4 and 6.3.3.5.

6.3.3.4 Sensitivity analysis for the product trial estimand

The sensitivity estimator for the trial product estimand will use the multiple imputation policy described for the main estimator (see section 6.3.3.1) except that:

- For subjects that discontinue trial treatment due to treatment-related AEs, multiple imputation of the missing endpoint will be from the placebo group. This assumes subjects reporting treatment-related AEs would have been experienced a level of efficacy similar to subjects in the placebo group if they had continued to take trial treatment. Data is assumed to be MNAR.

An additional sensitivity estimator will be performed to investigate the MAR assumption for the imputation in the sensitivity estimator above. The analysis will be performed as follows: a penalty (a number) is added to all imputed values in the active treatment group, and the analysis is repeated on the FAS population for the trial product estimand. The penalty is gradually increased until the point, the tipping point, where the null hypothesis is no longer

rejected. If the tipping point is considered a clinical plausible difference, the tipping-point analysis does not support the sensitivity estimator.

6.3.3.5 Sensitivity analysis for the treatment policy estimand

The sensitivity estimator for the treatment policy estimand will use multiple imputation from the placebo group for all subjects that discontinue trial treatment regardless of the reason for discontinuation. Data is assumed to be MNAR.

6.3.4 Supportive analyses

For the primary and key secondary efficacy endpoints, an observed case analysis based on all subjects with at least one diary entry in the relevant pollen season will be performed as a supportive analysis. This analysis will allow comparison of results both to previous trials where the primary efficacy analyses were based on observed cases and also to acknowledged criteria for clinical success, which were established based on observed case analyses.

6.3.5 Supplementary analyses

A supplementary analysis for the primary endpoint will be performed using the main estimator of the trial product estimand (see section 6.3.2). Subjects with potential data issues listed in (VV-CLIN-005043) will be excluded from the analysis.

6.4 Key secondary endpoint analyses

6.4.1 Estimators for estimands defined for key secondary efficacy objectives

The key secondary endpoints, average TCS during the TPS, average DSS during the BPS and TPS, and average DMS during the BPS and TPS will be analysed identically to the primary endpoint using the definitions of the trial product estimand and treatment policy estimand (see sections 3.1.1.1 and 3.1.1.2 respectively) and the main, sensitivity, and supportive estimators described in section 6.3.3, except for the tipping point analysis.

6.5 Summary of primary and key secondary endpoints analyses and figures

An overview of analyses for the primary and key secondary endpoints are presented in Table 6.

Table 6: Overview of analyses for the primary and key secondary endpoints

Endpoint	Description	Population	Model	Analysis name	Multiple imputation		
					Reason for IMP discontinuation' and not 'IE handling	Imputation group	Assumption
Average TCS during BPS	Trial product estimand	FAS	LME with MI	**Main estimator**	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Own treatment group • Own treatment group	• MNAR • MAR • MAR
				Sensitivity estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Placebo • Own treatment group	• MNAR • MNAR • MAR
				Sensitivity (tipping point)	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Placebo • Own treatment group (penalty on active group)	• MNAR • MNAR • MAR
				Supplementary	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Own treatment group • Own treatment group	• MNAR • MAR • MAR
	Treatment policy estimand		LME with MI	Main estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Placebo • Own treatment group	• MNAR • MNAR • MAR
				Sensitivity estimator	• All reasons	• Placebo	• MNAR

	Observed case		LME	Supportive	-	-	-
Average TCS during TPS	Trial product estimand	FAS	LME with MI	Main estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Own treatment group • Own treatment group	• MNAR • MAR • MAR
				Sensitivity estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Placebo • Own treatment group	• MNAR • MNAR • MAR
	Treatment policy estimand		LME with MI	Main estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Placebo • Own treatment group	• MNAR • MNAR • MAR
				Sensitivity estimator	• All reasons	• Placebo	• MNAR
	Observed case		LME	Supportive	-	-	-
Average DSS during BPS	Trial product estimand	FAS	LME with MI	Main estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reason	• Placebo • Own treatment group • Own treatment group	• MNAR • MAR • MAR
				Sensitivity estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Placebo • Own treatment group	• MNAR • MNAR • MAR
	Treatment policy estimand		LME with MI	Main estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Placebo • Own treatment group	• MNAR • MNAR • MAR
				Sensitivity estimator	• All reasons	• Placebo	• MNAR

	Observed case		LME	Supportive	-	-	-
Average DSS during TPS	Trial product estimand	FAS	LME with MI	Main estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Own treatment group • Own treatment group	• MNAR • MAR • MAR
				Sensitivity estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Placebo • Own treatment group	• MNAR • MNAR • MAR
	Treatment policy estimand		LME with MI	Primary	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Placebo • Own treatment group	• MNAR • MNAR • MAR
				Sensitivity estimator	• All reasons	• Placebo	• MNAR
	Observed case		LME	Supportive	-	-	-
Average DMS during BPS	Trial product estimand	FAS	LME with MI	Main Estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Own treatment group • Own treatment group	• MNAR • MAR • MAR
				Sensitivity estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Placebo • Own treatment group	• MNAR • MNAR • MAR
	Treatment policy estimand		LME with MI	Main estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Placebo • Own treatment group	• MNAR • MNAR • MAR
				Sensitivity estimator	• All reasons	• Placebo	• MNAR

	Observed case		LME	Supportive	-	-	-
Average DMS during TPS	Trial product estimand	FAS	LME with MI	Main estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Own treatment group • Own treatment group	• MNAR • MAR • MAR
				Sensitivity estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Placebo • Own treatment group	• MNAR • MNAR • MAR
	Treatment policy estimand		LME with MI	Main estimator	• Lack of efficacy • Treatment-related AEs • Other non treatment-related reasons	• Placebo • Placebo • Own treatment group	• MNAR • MNAR • MAR
				Sensitivity estimator	• All reasons	• Placebo	• MNAR
	Observed case		LME	Supportive	-	-	-

6.5.1 Figures for primary and key secondary endpoints

A plot with the results from the statistical analyses in the testing hierarchy will be presented. In addition, plots with 2 y-axes will be presented (the daily pollen count and average TCS, DSS and DMS) relative to day from TPS start for each cohort.

6.6 Additional endpoints analyses

6.7 Secondary endpoints

The following secondary endpoints will be analysed identically to the primary endpoint using the main estimator for the trial product estimand:

- Average TCS during the AHPS and OPS
- Average DSS during the AHPS and OPS
- Average DMS during the AHPS and OPS
- Average TCS (EAACI scoring) during BPS and TPS

Following endpoints will be analysed under the hypothetical strategy using the same intercurrent events and imputation approach defined for the main estimator.

- Proportion of severe days during BPS and TPS
- Proportion of well days during BPS and TPS
- Proportion of symptom-free days during BPS and TPS

The proportion of severe days during BPS will be analysed using a GLMM with a logit link function. For the proportion of severe days during BPS the model will includes “number of severe days/total number of days in BPS” as the response variable and treatment, cohort and age group as fixed effects and pollen station within cohort as random effects. From the GLMM model, the odds ratio of having a severe day of active treatment relative to placebo will be presented together with the corresponding p-values and 95% confidence limits. The endpoints proportion of severe days during TPS, proportion of well days during BPS and TPS, and symptom-free days during BPS and TPS will be analysed using the same model as for severe days during BPS. The imputation of missing data of aforementioned endpoints, will be done by using unrestricted random sampling method, which utilise selection with equal probability and with replacement. For each subject with missing data, the number of days in pollen season as well as the number of days with the endpoint of interest will be replaced from the pool of completed subjects with non-missing data in the same age group and the treatment group according to hypothetical strategy.

A plot with the results from the statistical analyses of the endpoints proportion of severe days, proportion of well days and proportion of symptom-free days during BPS will be presented. The plot will include estimated proportions of having a severe day, well day and symptom free days, the relative difference including 95% CI, and a p-values.

The endpoint percentage of patients free of symptoms and with no use of rescue medication during the BPS and TPS will be summarised. The endpoint will be categorised as a binary variable (0/1). A subject will be considered as responder if he/she has no symptoms and has not used rescue medication during an entire season (BPS or TPS).

6.7.1 Questionnaires

The endpoint average weekly overall RQLQ(S) score during the BPS and TPS respectively will be analysed using the same model as for the main estimator and sensitivity estimator for the trial product estimand and supportive analysis, except for not square rooting the endpoint and including age group as a fixed effect, i.e., the model will include treatment, and cohort as fixed effects and pollen station within cohort as a random effect, with different residual errors specified for each treatment group. The overall PRQLQ will be analysed using the same approach and model as for RQLQ.

In addition to above the average weekly overall RQLQ(S) score during the BPS and TPS will be summarised.

The endpoint patient-rated global evaluation of treatment efficacy at EOT categorised as a binary variable (improvement versus no improvement) will be analysed by using a GLIMM with a logit link function, under the hypothetical strategy using the same intercurrent event and imputation approach defined for the main estimator. The assessment is made at EOT and each subjects had to respond to the question using the 5-point scale: much better, better, the same, worse, much worse. A subject is considered as responder if she/he feels better or much better (improvement=1), otherwise it's considered as a non-responder (improvement=0). The model includes the binary endpoint (improvement/no improvement) as response variable and treatment, cohort and age group as fixed effects and pollen station within cohort as random effects. From the GLMM model, the odds ratio of having an improvement at EOT of active treatment relative to placebo will be presented together with the corresponding p-values and 95% CI.

TSQM-9 using the convenience domain for subjects 5 to less than 12 years at EOT will be analysed similar to average weekly overall RQLQ(S) score and overall PRQLQ using the main estimator for trial product estimand.

Summary of TSQM-9 by domains (effectiveness, convenience, global satisfaction) and treatment will be presented.

6.7.2 Immunology

The following immunology endpoints will be summarised by visit and treatment including change from baseline:

- Birch specific IgE, $\log_{10}$(IgE), $\log_{10}$(IgG_4).
- Alder, hazel and oak specific log10(IgE) and log10(IgG_4) (SUB)
- Birch, alder, and hazel specific IgE-BF (SUB)

Change from baseline in birch specific log10(IgE) and log10(IgG4) at visit 4 and 6 will be analysed using repeated measures linear mixed model. The model includes change from baseline of the log-transformed immunological parameter as response variable and the log-transformed immunological parameter at baseline, treatment, visit and treatment*visit interaction as fixed effects, and subject as random effect. The active group will be compared to placebo at each visit using a t-test in the linear mixed model. The corresponding difference in adjusted means (placebo vs active) will be calculated together with the associated p-value and 95% CI. In addition, the back-transformed adjusted mean and relative difference in back-transformed adjusted means including 95% CI will be listed. The analysis will be based on observed case and FAS.

The birch specific IgE-BF will be analysed using the same model as for IgE and IgG_4, without log transforming the endpoint and the fixed effect IgE-BF parameter at baseline. The analysis will be based on observed case and SUB.

Plots of change from baseline in birch specific $log_{10}(IgE)$, $log_{10}(IgG_4)$, and IgE-BF in adjusted means will be displayed by treatment with 95% CI.

When taking the logarithm, values below the LLOQ of IgE and IgG_4 will be given the minimum value measured above 0 for IgE and IgG_4 and. Otherwise, data is used as measured.

[redacted]

6.9 Safety analyses

All safety summary tables, figures, and listings will be based on the SAF unless otherwise specified.

6.9.1 Extent of exposure

6.9.1.1 Exposure

Exposure is a priori defined as the difference between the number of tablets dispensed and the number of tablets returned or lost. However, for a substantial number of subjects the information

about tablets lost is missing and is therefore assumed to be 0. To account for this, exposure will be calculated as the minimum of the difference between the number of tablets dispensed and the number of tablets returned or lost, and the treatment duration.

The number of tablets taken will be summarised by treatment group.

6.9.1.2 Treatment duration

The duration of treatment for each subject will be calculated from the date of first dose up until (and including) the date of last IMP administration.

Treatment duration (days) will be summarised by treatment group.

6.9.1.3 IMP compliance

IMP compliance (%) will be calculated for each subject as number of tablets taken divided by the treatment duration (days) multiplied by 100.

IMP compliance (%) will be summarised by treatment group.

Percentage IMP compliance will be summarised by treatment group.

6.9.1.4 eDiary compliance

eDiary compliance (%) for the TPS will be calculated as the number of non-missing eDiary entries in the TPS divided by total number of days in the TPS.

eDiary compliance (%) will be summarised by treatment group.

6.9.2 Adverse events

MedDRA version 23.1 will be used for reporting of AEs.

6.9.2.1 Events of special interest

Events of special interest for this trial are:

- Systemic allergic reactions including anaphylaxis
- Events treated with adrenaline/epinephrine
- Severe local swelling or oedema of the mouth and/or throat
- Oral allergy syndrome
- Eosinophilic oesophagitis
- Asthma exacerbations

6.9.2.2 Reporting of adverse events

AE is considered a TEAE if the time of onset is after the time of first dose of IMP and at most 7 days after last dose of IMP.

All TEAEs will be summarised by treatment group according to causality, severity (mild, moderate, severe), seriousness, action taken, outcome, and whether the event led to IMP discontinuation. The number of subjects in each treatment group, the number and frequency of

subjects having the event, as well as the number of events and proportion of events will be displayed. A similar summary table will be produced for all IMP-related TEAEs.

All TEAEs and all IMP-related TEAEs will be summarised (in separate tables) by treatment group, SOC, and PT, displaying number of subjects in treatment group, and number and frequency of subjects having the event. The tables will be sorted according to most frequent SOC and PT in the active treatment group, and the most common TEAEs (preferred term from MedDRA) defined as those that are present in at least 5% of subjects in the active treatment group will be marked.

Severe TEAEs will be listed only, if there are fewer than 10 events in total. Otherwise, severe TEAEs will also be summarised similar to how TEAEs are presented. The same applies for SAEs, IMP-related SAEs, TEAEs leading to IMP interruption, and TEAEs leading to IMP discontinuation.

For most frequent IMP-related TEAEs (present in at least 5% of subject in the active treatment group) onset and duration will be presented as follows: Average time to onset and average duration both in days, will be presented by PT and treatment. In addition, for day 1 of treatment average time to onset in day and minutes will be presented by SOC, PT, and treatment.

For the most frequent IMP-related TEAEs which are recurrent, the average daily duration and the average duration in days from first to last occurrence will be presented by SOC, PT and treatment.

Most frequent non-serious TEAEs in any arm (≥1 and ≥5%) will be summarised by system organ class and preferred term.

A table with the overall summary of the safety profile will be produced showing subjects with TEAEs, IMP-related TEAEs, IMP-related severe TEAEs, IMP-related treatment-emergent SAEs, IMP-related TEAEs leading to IMP discontinuation, and each type of IMP-related treatment-emergent ESI by treatment group.

Serious TEAEs, severe TEAEs, TEAEs leading to discontinuation, individual ESIs, and all non-treatment emergent adverse events will be listed.

The 6 different types of ESIs will be summarised by SOC, PT, and treatment group.

Medication errors, including overdose, abuse and misuse of the IMP will be listed.

In addition to above a plot of the most frequent (≥5%, in active arm) IMP-related TEAS by SOC and PT will be produced. Furthermore, plots of the median duration in days and time to onset in days of most frequently (≥5%) reported IMP-related AEs of subjects in active treatment group.

6.9.2.3 Local administration site TEAEs

The AEs identified as local administration site TEAEs, by comparison against a pre-defined list of MedDRA PTs, will be summarised separately. The pre-defined list of PTs is given in section Appendix A.

IMP-related local administration site reactions TEAEs will be summarised similar to all TEAEs by treatment group according to causality, severity (mild, moderate, severe), seriousness, action taken, outcome, and whether the event led to IMP discontinuation.

6.9.3 Additional safety assessments

Shift tables will be produced for physical examination, and laboratory assessments (chemistry, haematology, and urinalysis) for baseline to EOT.

In addition to above laboratory assessments (chemistry and haematology), vital signs and the pulmonary function test parameters (FVC, FVC percent predicted, FEV_1, and FEV_1 percent predicted) will be summarised by visit and treatment group.

Results from the pregnancy test for female subjects of child-bearing potential will be listed.

6.10 Interim analysis

No interim analysis was planned.

6.11 Changes and/or deviations to protocol-planned analyses

In addition to the analyses specified in the CTP a supplementary analysis for the primary endpoint has been added where subjects with potential data issues will be excluded from the main analysis.

The secondary endpoints (average TCS during the AHPS and OPS, average DSS during the AHPS and OPS, average DMS during the AHPS and OPS, and average TCS (EAACI scoring) during BPS and TPS) will be analysed by using the hypothetical estimand and main estimator only.

Local administration site reactions are specified as 'treatment-related local administration site TEAEs.

The reasons for IMP discontinuation will be visualised by a band plot, instead of a Kaplan-Meier plot as the band plot also shows the different reasons for discontinuation.

Physical examination will not be summarized by visit as stated in the CTP, instead a shift table will be made from baseline to end of treatment. 5-, 25-, 75-, and 95 percentiles will not be presented for tabulation of data by visit as stated in the CTP.

The exploratory assessment of the levels of serum antibodies/serum components/serum markers to relevant allergens will not be performed nor reported in a separate document as stated in CTP.

Due to the amount of missing pollen data, it was deemed necessary to perform a visual inspection of all species by pollen station instead of the pollen plots (pollen grains/m3 versus time) as the approach chosen is more precise.

7 Sample size determination

Sample size calculations are based on the primary endpoint, the average TCS during the BPS, which will be square root transformed prior to analysis in order to better approximate the normal distribution. Based on previous ALK SLIT-tablet studies in both adult and paediatric populations, it is assumed that the mean of the square root transformed average TCS during the BPS in the placebo group is 2.5 with corresponding common standard deviation of 1.4 in both placebo and active groups.

Analyses for the primary and secondary estimands will include all randomised subjects and use different multiple imputation strategies to account for missing data. For the purposes of sample size estimation, it is assumed that 5% of observations in the active arm are imputed from the placebo arm. A true relative treatment effect of 25% is assumed, which corresponds to an effective relative treatment effect of 23.75% after placebo imputation.

For a range of sample sizes and a 1:1 randomisation ratio between active and placebo groups, 1000 trial datasets were simulated under the above assumptions and each trial was analysed using a linear model with a fixed term for treatment. For each choice of sample size, the probability of the trial fulfilling the regulatory requirements of both the EMA and FDA for the primary endpoint was assessed over all the simulated trials.

The following regulatory requirements were considered:

1. Statistical significance of the absolute treatment effect (EMA)
2. Point estimate of the absolute treatment effect greater than 1 (EMA)
3. Point estimate of the relative treatment effect greater than 15% (FDA)
4. Lower limit of the 95% CI for the relative treatment effect greater than 10% (FDA)

Based on the results of the simulation trial, 500 randomised subjects per arm (1000 subjects in total) was chosen to provide at least 85% power to satisfy regulatory requirements 1, 2 and 3 for the analyses based on all randomised subjects (assuming 5% imputation of placebo in the active arm for missing data). Assuming 10% of the 1000 randomised subjects do not have any diary data during the BPS, the power for an observed case (OC) analysis to meet the requirements is also presented.

Table 7: Power to meet regulatory criteria for FAS and Observed Case Analyses

	Statistical significance of absolute treatment effect	Point estimate of absolute treatment effect >1	Point estimate of relative treatment effect >15%	Lower limit of 95% CI for relative treatment effect >10%
FAS analysis[1,3] n=500[2] per arm	95.0%	88.3%	92.8%	60.2%
OC analysis[1,4] n=450[2] per arm[3]	94.7%	89.9%	93.9%	62.2%

[1] FAS analysis includes all randomised subjects, OC analysis includes all randomised subjects with diary data in the BPS
[2] n=number of subjects included in the analysis
[3] Assumptions: placebo mean=2.5, SD=1.4 (active & placebo), true relative treatment effect=25%, 5% of active arm imputed from placebo
[4] Assumptions: placebo mean=2.5, SD=1.4 (active and placebo), true relative treatment effect=25%, no MI, 10% of 1000 randomised subjects excluded due to missing data

8 Demographics and baseline characteristics

8.1 Screening failures

A summary of reasons for screening failure will be presented for the total analysis set.

8.2 Protocol deviations

Overall important PDs will be summarised by trial-, country-, site- and subject-level. Furthermore, the important PDs will be summarized by the subject-level deviations and treatment.

A listing with all important PDs will be presented. In addition, a listing with subjects who had TC4 before V6 will be presented.

8.3 Subject disposition

Subject disposition will be summarised by number and percentage of subjects screened, randomised, included in the analysis sets, completed trial, discontinued trial, reason of discontinuation of trial, completed IMP, discontinued IMP, and reason for IMP discontinuation by treatment group.

IMP discontinuations over time by different reasons will be visualised by a band plot showing proportions on treatment, having discontinued treatment due to AE, withdrawal by subject, and all other reasons (pooled).

Screening failures will be summarised by reason.

8.4 Baseline characteristics

Demographic variables (age, age group, sex, race, ethnic origin, and country) will be summarised by treatment group.

Baseline characteristics (mono/poly sensitised, duration of AR/C induced by birch pollen, skin prick test results, skin prick test wheal sizes (birch), specific IgE to birch, and asthma status) will be summarised by treatment group.

8.5 Medical history

Medical history is coded using the MedDRA version 23.1 and will be summarised by treatment group.

Separate tables will be created for allergy or asthma history and other medical history. In addition, the other medical history will also be summarised by PT and SOC.

8.6 Prior and concomitant therapy

Prior and concomitant medication will be coded according to the WHO drug dictionary and summarised separately by treatment group.

9 Supporting documentation

9.1 Definition of pollen seasons

Each site was allocated to a pollen region prior to unblinding.

The different pollen seasons varied across the selected pollen regions both in intensity and duration.

BPS

For each pollen region and year the BPS were defined as follows:

Start date:	The start date of the BPS is defined as the first day of 3 consecutive days with pollen count larger than or equal to 30 grains/m^3
Stop date:	The stop date of the BPS is defined as the last day in the last occurrence of 3 consecutive days with pollen count larger than or equal to 30 grains/m^3

Alder, Hazel and Oak

The alder, hazel, oak pollen seasons were defined as the days included in any of the alder, hazel, and oak pollen seasons:

Start date:	The start date of the pollen season is defined as the first day of 3 consecutive days with pollen count larger than or equal to 10 grains/m^3
Stop date:	The stop date of the pollen season is defined as the last day in the last occurrence of 3 consecutive days with pollen count larger than or equal to 10 grains/m^3

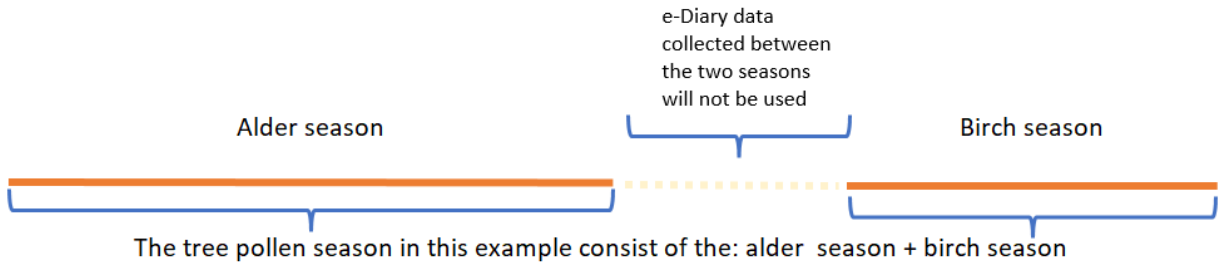
Based on experience from trial TT-04, it is anticipated that there will be a high degree of overlap between the alder and hazel seasons. As such, a combined alder-hazel season is defined for each pollen station as follows:

Start date:	The start date of the alder-hazel pollen season is the earliest of the alder season start date and hazel season start date
Stop date:	The stop date of the alder-hazel pollen season is the latest of the alder season stop date and the hazel season stop date

TPS

The TPS is defined as all days included in any of the alder, birch, hazel, oak pollen seasons. The TPS is not necessarily a continuous period, an example is illustrated in Figure 3 .

Figure 3: Example of tree pollen season with non-continuous period



[redacted]

In case of missing pollen counts for specific pollen stations one of the 3 approaches was applied prior to unblinding:

- For pollen stations with missing data between observed start and stop date of the season LOCF was applied.
- For pollen stations with either very sparse data or completely missing data, each station was assigned a neighboring station as close as possible to the original station where data was available. Pollen counts from the backup station was used and in addition a sanity check was made by looking into the historical pollen data for the original pollen station.
- For pollen stations where data was missing for a short period of time and it was deemed realistic that seasons had started or not yet ended LOCF was applied.

Due to the Ukraine crisis data from the pollen station RUMOSC (Moscow, Russia) was not retrievable. Start and stop dates for all pollen seasons in Russia in the season 2022 have been estimated by Uwe Berger[15] based on the past 5 years of pollen data.

In addition, a visual inspection of the pollen count for all species is made to validate the pollen counts in the season based on the criteria described below. This procedure can identify long periods with low pollen counts within the season.

The final pollen seasons are listed in Appendix D.

9.2 Derivations for the trial

9.2.1 Derivations of endpoints

9.2.1.1 Definition of DSS and DMS

During the diary completion period, symptoms and medication use are recorded daily by the subject in the e-diary. In addition, missed school days for patient, missed workdays for caregiver, and school days where performance is affected by AR/C are collected in the e-diary.

9.2.1.2 DSS

The 6 symptoms of AR/C DSS consist of 4 rhinitis symptoms (runny nose, blocked nose, sneezing, itchy nose) and 2 conjunctivitis symptoms (gritty feeling/red/itchy eyes, watery eyes). The subject rates each symptom as none, mild, moderate, or severe. Each rating is then assigned to a score from 0 to 3 based on the subject's daily rate:

0 = no symptoms
1 = mild symptoms
2 = moderate symptoms
3 = severe symptoms

The DSS is the sum of the 6 individual rhinoconjunctivitis symptom scores each day and ranges from 0 to 18.

9.2.1.3 DMS

The AR/C DMS is derived using the scoring system in Table 8 and takes values between 0 and 20 (maximum allowable daily rescue medication used). Note that the recommended dose of Desloratadine is scored 6, and that doses below are scored proportionally, i.e. if a 12-year-old takes 5 ml once daily, this is scored as 3. Similarly lower doses than recommended use of Loratadine tablets, Olopatadine eye drops, and Mometasone furoate nasal spray is also scored proportionally based on the recommended daily dose. Subjects taking Loratadine tablets, although they should have taken Desloratadine oral solution are scored the same as if they should have taken tablets, i.e. a score of 6 for 1 tablet, and a score of 3 for half a tablet. Even if a subject takes more than the allowed dosage, then it cannot score more than the max daily score for that type of rescue medication. The following rules will be applied:

- A half tablet will be scored to 0.5 x "tablet score".

[15] Uwe E. Berger, Head of Aerobiology and Pollen, Information Research Unit, AZ Pollen Research, GmbH, Friedrich Schoeffel-G. 6, 2000 Stockerau, Austria

- A maximum daily score of 20 will be ensured by capping individual items scores.

Table 8: Scoring for allergic rhinoconjunctivitis rescue medication usage

Rescue medication	Dosage	Score/ Dose unit	Max daily score
Loratadine, 10 mg Or Desloratadine, oral solution 0.5 mg/mL	6-12 years old and >30 kg: 1 tablet once daily as needed	6 per tablet	6
	>12 years old: 1 tablet once daily as needed		
	5 years old: 2.5 mL solution once daily as needed	6 per 2.5mL	
	6-11 years old: 5.0 mL solution once daily as needed	6 per 5 mL	
	≥12 years old: 10 mL solution once daily as needed	6 per 10 mL	
Olopatadine eye drops, 1 mg/mL	1 drop in the affected eye(s) twice daily, morning and evening as needed	1.5 pr. drop	6
Mometasone nasal spray, 50 µg/dose	5-11 years old: 1 puff in each nostril once daily as needed (max 2 puffs)	4 per puff	8
	≥12 years old: 2 puffs in each nostril once daily as needed (max 4 puffs)	2 per puff	
Maximum allergic rhinoconjunctivitis daily medication score (DMS)			20

9.2.1.4 TCS

The AR/C daily TCS is the sum of the daily DSS and DMS (that is, daily TCS=DSS+DMS) and takes values between 0 and 38.

[redacted]

9.2.1.6 Daily EAACI medication score

The daily EAACI medication score (DMSEAACI) is defined according to the use of symptom relieving medication as follows:

Table 9: EAACI scoring for allergic rhinoconjunctivitis rescue medication usage

Rescue medication	Dosage	EAACI component score if medication is used (0 otherwise)
Loratadine, 10 mg Or Desloratadine, oral solution 0.5 mg/mL	6-12 years old and >30 kg: 1 tablet once daily as needed	1
	>12 years old: 1 tablet once daily as needed	
	5 years old: 2.5 mL solution once daily as needed	
	6-11 years old: 5.0 mL solution once daily as needed	
	≥12 years old: 10 mL solution once daily as needed	
Olopatadine eye drops, 1 mg/mL	1 drop in the affected eye(s) twice daily, morning and evening as needed	1
Mometasone nasal spray, 50 µg/dose	5-11 years old: 1 puff in each nostril once daily as needed (max 2 puffs)	2
	≥12 years old: 2 puffs in each nostril once daily as needed (max 4 puffs)	
Maximum allergic rhinoconjunctivitis daily medication score (DMSEAACI)		2
DMSEAACI is defined as the greatest of the EAACI components for each rescue medication type i.e., DMSEAACI can takes values of 0, 1, or 2. Note that in DMSEAACI can take a value of 3 if a subject uses “Oral corticosteroids with/without INS, with/without H1A” (Pfaar et al. 2014). This is not relevant for this trial as oral corticosteroids are not supplied as rescue medication.		

9.2.1.7 TCS with EAACI medication scoring (TCSEAACI)

The TCS with EAACI medication scoring (TCSEAACI) is the sum of DSS/6 and DMSEAACI and ranges from 0 to 5.

9.2.2 Derivation of average symptom and medication scores

9.2.2.1 Average Daily Rhinoconjuctivitis Symptom Score (average DSS)

The average DSS during a given pollen season is the mean of all non-missing DSS over all days in the selected pollen season. Average DSS will be derived for the BPS, TPS, AHPS, OPS [redacted]

9.2.2.2 Average Daily Rhinoconjuctivitis Medication Score (average DMS)

The average DMS during a given pollen season is the mean of all non-missing DMS over all days in the selected pollen season. Average DMS will be derived for the BPS, TPS, AHPS, OPS [redacted]

9.2.2.3 Average Daily Rhinoconjuctivitis Total Combined score (average TCS)

The average TCS during a given pollen season is the mean of all non-missing daily TCS over all days in the selected pollen season. Average TCS will be derived for the BPS, TPS, AHPS, OPS [redacted] The definitions of the pollen seasons are provided in section 9.1.

[redacted]

9.2.2.5 Average Daily Total Combined Score with EAACI medication scoring (average TCSEAACI)

The average TCS (EAACI scoring) is the mean of the daily non-missing values of the AR/C daily TCS with EAACI scoring.

The average TCS (EAACI scoring) will be derived in the BPS and TPS.

9.2.3 Derivation of daily binary variables

9.2.3.1 Proportion of well days

A well day is defined as a day with no use of AR/C rescue medication (i.e., DMS=0) and DSS less than or equal to 2. A day that meets the definition of well day will take the value 1, and 0 otherwise.

The proportion of well days during BPS and TPS is the number of well days/total number of days in a season (BPS or TPS).

9.2.3.2 Proportion of severe days

A severe day is a day with a DSS greater than or equal to 6 and at least 2 moderate symptoms or 1 severe symptom. A day that meets the definition of severe day will take the value 1, and 0 otherwise.

The proportion of severe days during BPS and TPS is the number of severe days/total number of days in a season (BPS or TPS).

9.2.3.3 Proportion of symptom-free days

A symptom-free day is defined as a day with no symptoms and with no use of rescue medication (i.e., daily TCS=0). A day that meets the definition of symptom-free day will take the value 1, and 0 otherwise.

The proportion of symptom-free days during BPS and TPS is the number of symptom-free days/total number of days in a season (BPS or TPS).

[redacted]

9.2.4 Other derivations of endpoints

9.2.4.1 RQLQ

The RQLQ(S) consists of 28 items covering 7 domains (activities, sleep, no-nose/eye symptoms, practical problems, nasal symptoms, eye symptoms, emotional). Each item is scored on a 7-point (0-6) scale, where lower scores indicate better quality of life and all items within each domain are weighted equally. The domain scores are derived as the mean of all item scores within each domain. The overall RQLQ score is the mean of the 28 item scores.

The RQLQ(S) is completed weekly in the eDiary during TPS by subjects aged 12-17 years at randomisation.

9.2.4.2 PRQLQ

The PRQLQ consists of 23 items covering 5 domains (nasal symptoms, eye symptoms, practical problems, activity limitation and other symptoms). Each item is scored on a 7-point (0-6) scale, where lower scores indicate better quality of life and all items within each domain are weighted equally. The domain scores are derived as the mean of all item scores within each domain. The overall PRQLQ score is the mean of the 23 item scores.

The PRQLQ is completed once during the BPS by subjects aged 5-11 years at randomisation.

9.2.4.3 TSQM-9

The TSQM-9 consists of 9 items covering 3 domains (Effectiveness, Convenience, Global satisfaction). Items are scored on either a 5-point (1-5) scale or 7-point (1-7) scale, where higher scores indicate greater treatment satisfaction. For each domain, item scores are summed and normalised to a 0-100 scale, where 0 is the worst possible level of satisfaction and 100 is the best possible level of satisfaction. The calculations specific to each domain are presented in detail in the Treatment Satisfaction Questionnaire for Medication (TSQM) User Manual, version 1.8, 08Jun2020.

9.2.4.4 Patient-rated global evaluation of treatment efficacy

Subjects will be asked the following question regarding their perception of the efficacy of the treatment they received during the birch/tree pollen season: “Compared to your rhinitis and/or conjunctivitis symptoms in the previous birch/tree pollen season, how have you felt overall in this birch/tree pollen season?”

Subjects will respond to the question using the 5-point scale: much better, better, the same, worse, much worse.

‘Better’ or “much better” will be categorised as “improved”, and “much worse”, “worse” or “the same” will be categorised as “not-improved”.

9.2.4.5 Lung function

Measurements of FVC, FVC percent predicted, FEV1 and FEV1 percent predicted will be recorded by the trial sites. The formulae used for calculating predicted FVC and predicted FEV1 can vary slightly depending on the spirometer used at each site. Therefore, for reporting purposes, FVC percent predicted and FEV1 percent predicted will be recalculated using values of predicted FVC and predicted FEV1 calculated from the Quanjer equations (Quanjer et al. 1995). This is to ensure a standard approach to the derivation of the endpoints. The Quanjer formulae for predicted FVC and FEV1 are a function of age, height, and race.

9.2.5 Imputation of dates

9.2.5.1 Partial dates in adverse event reporting

Partial dates and time for **start** of AEs are handled in the following way:

- Missing day is imputed as maximum of first day of month and day of first IMP
- Missing month is imputed as maximum of first day in January and date of first IMP

Partial dates and time for **end** of AEs are handled in the following way:

- Missing day is imputed as minimum of last day in month and day of last contact
- Missing month is imputed as minimum last day of December and date of last contact

With these imputation rules, AEs are considered treatment emergent when dates are partial, and the existing information does not exclude the possibility of the event being treatment emergent.

9.2.5.2 Incomplete date for last IMP

Any partial date of last dose is imputed to as late as possible but no later than the date of last contact. Missing dates of last dose are imputed as the date of last contact.

10 References

EMA & PDCO. EMA/PDCO Standard Paediatric Investigation Plan for Allergen Products for Specific Immunotherapy. EMA/PDCO/737605/2009, Revision 4, 1-13. 2015

Fieller, EC. (1954). "Some problems in interval estimation". Journal of the Royal Statistical Society, Series B. 16 (2): 175–185

ICH E9 Statistical Principles for clinical Trials, Feb 1998

ICH E9 (R1) Addendum on estimands and sensitivity analysis in clinical trials to the guideline on statistical Principles for Clinical Trials, November 2019

Kenward, M. G., and Roger, J. H. (1997). “Small Sample Inference for Fixed Effects from Restricted Maximum Likelihood.” Biometrics 53:983–997.

Pfaar,O., Demoly,P., Gerth van,W.R., Bonini,S., Bousquet,J., Canonica,G.W., Durham,S.R., Jacobsen,L., Malling,H.J., Mosges,R., Papadopoulos,N.G., Rak,S., Rodriguez Del,R.P., Valovirta,E., Wahn,U. & Calderon,M.A. 2014. Recommendations for the standardization of clinical outcomes used in allergen immunotherapy trials for allergic rhinoconjunctivitis: an EAACI Position Paper. Allergy.

11 Appendices

Appendix A Local administration site TEAEs:

High-Level Group Terms (HLGTs)

- Oral soft tissue conditions
- Salivary gland conditions
- Tongue conditions

High-Level Terms (HLTs)

- Gingival disorders, signs, and symptoms NEC
- Gingival haemorrhages
- Dental and oral soft tissue infections

Appendix B Further details to statistical analyses

Table 10: LME analyses

LME output	Adjusted means for each treatment group, the absolute treatment difference (Placebo – Active) with 95% confidence interval and p-value, and the relative treatment difference (Placebo – Active/ Placebo) with 95% confidence interval will be presented.
Without multiple imputation	
Endpoint	The endpoint is analysed as the response variable in a LME model which includes relevant fixed and random effects, and with different residual errors specified for each treatment group. The model will be estimated using REML, and the Kenward and Roger's approximation (Kenward and Roger 1997) is used to calculate the denominator degrees of freedom. The p-value for the absolute difference is reported as the test result. The 95% CI for the relative difference will be calculated using Fieller's theorem (Fieller 1954).
Square root transformed endpoint	The square root transformed endpoint is analysed as the response variable in a LME model which includes relevant fixed and random effects, and with different residual errors specified for each treatment group. The model will be estimated using REML, and the Kenward and Roger's approximation (Kenward and Roger 1997) is used to calculate the denominator degrees of freedom. The results are back-transformed as follows: estimated least square means of the square root transformed endpoint are output along with the associated covariance matrix. The absolute difference is calculated by squaring the adjusted means, and then calculating the difference. For the absolute difference the SE is approximated by using the first order Delta method (first order Taylor approximation), and from this the 95% CI is calculated. For the relative difference, Fieller's theorem (Fieller 1954) is used to calculate the 95% CI for X/Y, and then the 95% confidence bounds for $(Y^2-X^2)/Y^2 = 1 - (X/Y)^2$ are found by applying the monotone transformation $f(r)= 1 - r^2$ to the confidence interval.
With multiple imputation	
Imputation will be done using the method of unrestricted random sampling with replacement (seed=7127) and 1000 multiple imputed datasets will be created.	
Endpoint	Each of the 1000 multiple imputed datasets is analysed using the model specified for the endpoint. The endpoint is analysed as the response variable in a LME model which includes relevant fixed and random effects. The model will be estimated using REML, and the Kenward and Roger's approximation (Kenward and Roger 1997) is used to calculate the denominator degrees of freedom. Rubin's rule is used to combine the 1000 analysis results across the multiple imputed datasets. The p-value for the absolute difference is reported as the test result. The 95% confidence interval for the relative difference is calculated using Fieller's theorem (Fieller 1954).

Square root transformed endpoint	Each of the 1000 multiple imputed datasets is analysed using the model specified for square root transformed endpoint. The square root transformed endpoint is analysed as the response variable in a LME model which includes relevant fixed and random effects, and with different residual errors specified for each treatment group. The model will be estimated using REML, and the Kenward and Roger's approximation (Kenward and Roger 1997) is used to calculate the denominator degrees of freedom. Rubin's rule is used to combine the 1000 analysis results across the multiple imputed datasets. The p-value for the absolute difference is reported as the test result. The results are back-transformed as follows; after Rubin's rule is used to combine results across imputations, estimated least square means on the square root transformed scale are output along with the associated covariance matrix. The absolute difference is calculated by squaring the adjusted means, and then calculating the difference. For the absolute difference the SE is approximated by using the first order Delta method (first order Taylor approximation), and from this the 95% CI is calculated. For the relative difference, Fieller's theorem (Fieller 1954) is used to calculate the 95% CI for X/Y, and then the 95% confidence bounds for $(Y^2-X^2)/Ys2 = 1 - (X/Y)^2$ are found by applying the monotone transformation $f(r)= 1 - r^2$ to the confidence interval.

Convergence issues:

If the model does not converge due, then the random effect pollen station will be pooled into larger groups. If the model still does not converge, the random effect of pollen within cohort will be removed from the model.

Model checking:

The assumption of normally distributed residuals underlying the LME model will be evaluated by visual inspection of quantile-quantile plots for the primary and key secondary endpoints

Square root transformation:

The LME relies on the assumption of normally distributed residuals. Therefore, the square root transformation is generally applied, as this usually results in a good approximation to the so-called normal distribution. In addition, the square root transformation is used as the DMS scores are skewed with a lot if 0's, which is the reason for not using a log transformation.

Mean and SE

Let $\mu_{placebo}$, μ_{Active}, ${\sigma^2}_{Placebo}$, and ${\sigma^2}_{Active}$ be the resulting means and variances for placebo and active treatment and cov be the covariance. Then the back-transformed means are given by:

- $\hat{\mu}_{Placebo} = (\mu_{Placebo})^2$
- $\hat{\mu}_{Active} = (\mu_{Active})^2$
- $\hat{\mu}_{Difference} = (\mu_{Placebo})^2 - (\mu_{Active})^2$

and the back-transformed standard errors are given by

- $SE_{Active} = \sqrt{(2\mu_{Active})^2\sigma_{Active}}$
- $SE_{Placebo} = \sqrt{(2\mu_{Placebo})^2\sigma_{Placebo}}$
- $SE_{Difference} = \sqrt{(2\mu_{Placebo})^2\sigma_{Placebo} + (-2\mu_{Active})^2\sigma_{Active} + 2(2\mu_{Placebo})(-2\mu_{Active})cov}$

Appendix C Examples of SAS code

Table 11: SAS code for primary efficacy analysis

SAS code

proc mixed data= method=reml plots=studentpanel(marginal conditional);
class treatment cohort age_group pollen_station;
model TCS_{qrt}= treatment cohort age_group / ddfm = KenwardRogers residual;
random pollen_station (cohort);
repeated/group=treatment;
lsmeans treatment / cl cov diff;
run;

TCS_{sqrt} is the square root of the average TCS.

Table 12: Example for calculating proportion endpoints

SAS code

proc glimmix data =;
class treatment cohort age_group pollen_station;
model aval / datadays = treatment cohort agegroup / dist=binomial link=logit;
random pollen_station(cohort);
lsmeans treatment / diff ilink oddsratio cl;
run;

Appendix D Final pollen season

pollen_ station	Season	Cohort	Start_date	End_date	Average grains/m3	Neighbouring	comments
ATWIEN	ALDER	1	05-02-2022	30-03-2022	95,6		
CZTRIN	ALDER	1	13-02-2022	30-03-2022	110,1		
DEAACH	ALDER	1	09-02-2022	08-03-2022	47		
DEBOCH	ALDER	1	07-02-2022	13-03-2022	54,7		
DEBORS	ALDER	1	27-01-2022	25-03-2022	109,8		
DEDRES	ALDER	1	12-02-2022	24-03-2022	55		
DELEVE	ALDER	1	07-02-2022	13-03-2022	27,2		
DELOEW	ALDER	1	04-02-2022	22-03-2022	52,8		
DEMARB	ALDER	1	08-02-2022	23-03-2022	64,7		
DEMOEN	ALDER	1	07-02-2022	13-03-2022	34,7		
DKCOPE	ALDER	1	12-02-2022	15-03-2022	20		
FRSTRA	ALDER	1	02-02-2022	28-03-2022	324,8		
HUGYOE	ALDER	1	03-02-2022	26-03-2022	84,7		
HUMISK	ALDER	1	05-02-2022	01-05-2022	36,3		
HUPEST	ALDER	1	04-02-2022	30-03-2022	118,2		
HUSALG	ALDER	1	05-02-2022	03-04-2022	107,4		
HUSZE2	ALDER	1	15-02-2022	29-03-2022	65,2		
LTSIAU	ALDER	1	24-02-2022	28-03-2022	82,3		
LTVILN	ALDER	1	25-02-2022	05-04-2022	63,5		
PLBYDG	ALDER	1	13-02-2022	06-04-2022	171,7		
PLCRAC	ALDER	1	13-02-2022	28-03-2022	156,3		
PLKATO	ALDER	1	08-02-2022	04-04-2022	126,5		
PLLODZ	ALDER	1	13-02-2022	02-04-2022	180,8		
PLLUBL	ALDER	1	15-02-2022	29-03-2022	120,6		
PLPTRY	ALDER	1	08-02-2022	05-04-2022	175,1		
PLSZCZ	ALDER	1	05-02-2022	14-03-2022	51,5		
RUMOSC	ALDER	1	14-03-2022	12-04-2022			
SKBRAT	ALDER	1	09-02-2022	09-04-2022	113,6		
ATWIEN	ALDER	2	20-02-2023	11-03-2023	19,3		
BEBRUS	ALDER	2	08-02-2023	23-03-2023	133,4	NLLEID	completely missing
BEMARC	ALDER	2	07-02-2023	24-03-2023	153,8	DEMOEN	completely missing
CZTRIN	ALDER	2	07-03-2023	10-03-2023	27,5		
DEBOCH	ALDER	2	13-02-2023	22-03-2023	141,2		
DEDRES	ALDER	2	16-02-2023	03-03-2023	71,8		
DELEVE	ALDER	2	07-02-2023	24-03-2023	153,8	DEMOEN	partially missing
DELIPP	ALDER	2	16-02-2023	21-03-2023	83,1		

DELOEW	ALDER	2	15-02-2023	06-03-2023	76,5		
DEMARB	ALDER	2	22-02-2023	18-03-2023	63,4		
DEMOEN	ALDER	2	07-02-2023	24-03-2023	153,8		
DEMUNC	ALDER	2	16-02-2023	10-03-2023	38		
DKCOPE	ALDER	2	11-02-2023	03-04-2023	41,8		
DKVIBO	ALDER	2	01-03-2023	03-04-2023	12,8		
FRSTRA	ALDER	2	02-02-2023	14-03-2023	127,7		
HUGYOE	ALDER	2	17-02-2023	10-03-2023	45,3		
HUMISK	ALDER	2	21-02-2023	05-03-2023	18,9		
HUPEST	ALDER	2	17-02-2023	10-03-2023	25,3		
HUSALG	ALDER	2	18-02-2023	17-03-2023	25,3		
HUSZE2	ALDER	2	17-02-2023	02-03-2023	35,6		
LTSIAU	ALDER	2	19-03-2023	11-04-2023	46		
LTVILN	ALDER	2	14-03-2023	25-03-2023	138,5		
MONTREAL	ALDER	2	09-04-2023	11-05-2023	35,8		
NLLEID	ALDER	2	08-02-2023	23-03-2023	133,4		
PLBIAL	ALDER	2	10-03-2023	24-03-2023	93,5		
PLBYDG	ALDER	2	17-02-2023	16-04-2023	24,3		
PLCRAC	ALDER	2	26-02-2023	14-03-2023	33,5		
PLKATO	ALDER	2	10-03-2023	14-03-2023	44,8		
PLLODZ	ALDER	2	03-03-2023	20-03-2023	54		
PLLUBL	ALDER	2	18-02-2023	31-03-2023	18,9	PLPTRY	completely missing
PLPTRY	ALDER	2	18-02-2023	31-03-2023	18,9		
PLSZCZ	ALDER	2	17-02-2023	16-04-2023	24,3	PLBYDG	completely missing
PLWROC	ALDER	2	01-03-2023	14-03-2023	46,5		
QUEBEC	ALDER	2	13-04-2023	09-05-2023	20,4		
SKBRAT	ALDER	2	17-02-2023	11-03-2023	43,2		
SKNITR	ALDER	2	17-02-2023	11-03-2023	43,2	SKBRAT	completely missing
ATWIEN	BIRCH	1	28-03-2022	03-05-2022	367,9		LOCF
CZTRIN	BIRCH	1	14-04-2022	15-05-2022	378,4		
DEAACH	BIRCH	1	23-03-2022	26-04-2022	260,5	DEMOEN	completely missing
DEBOCH	BIRCH	1	23-03-2022	29-04-2022	323,5		
DEBORS	BIRCH	1	16-04-2022	11-05-2022	324,8		
DEDRES	BIRCH	1	06-04-2022	11-05-2022	463,6		
DELEVE	BIRCH	1	22-03-2022	24-04-2022	221,4		
DELOEW	BIRCH	1	28-03-2022	09-05-2022	132,1		
DEMARB	BIRCH	1	09-04-2022	11-05-2022	556,8		
DEMOEN	BIRCH	1	23-03-2022	26-04-2022	260,5		
DKCOPE	BIRCH	1	27-04-2022	30-04-2022	41		

FRSTRA	BIRCH	1	24-03-2022	04-05-2022	228,2		
HUGYOE	BIRCH	1	28-03-2022	03-05-2022	185,2		
HUMISK	BIRCH	1	05-04-2022	14-05-2022	250,1		
HUPEST	BIRCH	1	26-03-2022	10-05-2022	175,5		
HUSALG	BIRCH	1	07-04-2022	12-05-2022	257,3		
HUSZE2	BIRCH	1	29-03-2022	14-05-2022	131,4		
LTSIAU	BIRCH	1	21-04-2022	24-05-2022	560,2		
LTVILN	BIRCH	1	26-04-2022	24-05-2022	389,2		
MONTREAL	BIRCH	1	08-05-2022	13-05-2022	150,9		
PLBYDG	BIRCH	1	13-04-2022	15-05-2022	1053,9		
PLCRAC	BIRCH	1	12-04-2022	13-05-2022	601		
PLKATO	BIRCH	1	08-04-2022	23-05-2022	1049,2		
PLLODZ	BIRCH	1	13-04-2022	14-05-2022	972,1		
PLLUBL	BIRCH	1	14-04-2022	13-05-2022	632,8		
PLPTRY	BIRCH	1	13-04-2022	14-05-2022	1002,3		
PLSZCZ	BIRCH	1	18-04-2022	03-05-2022	78,3		
QUEBEC	BIRCH	1	09-05-2022	14-05-2022	198,7		
RUMOSC	BIRCH	1	21-04-2022	13-05-2022			
SKBRAT	BIRCH	1	27-03-2022	12-05-2022	441,6		
ATWIEN	BIRCH	2	24-03-2023	24-04-2023	64,3		
BEBRUS	BIRCH	2	12-04-2023	01-05-2023	143,2	NLLEID	completely missing
BEMARC	BIRCH	2	02-04-2023	05-05-2023	329,1	DEMOEN	completely missing
CZTRIN	BIRCH	2	13-04-2023	06-05-2023	189,6		
DEBOCH	BIRCH	2	10-04-2023	05-05-2023	615,5		
DEDRES	BIRCH	2	17-04-2023	01-05-2023	68,2	DEMARB	partially missing
DELEVE	BIRCH	2	05-04-2023	14-04-2023	172,1		LOCF
DELIPP	BIRCH	2	17-04-2023	02-05-2023	212,9		
DELOEW	BIRCH	2	17-04-2023	05-05-2023	81,4		
DEMARB	BIRCH	2	17-04-2023	01-05-2023	68,2		
DEMOEN	BIRCH	2	02-04-2023	05-05-2023	329,1		
DEMUNC	BIRCH	2	21-04-2023	05-05-2023	158,5		
DKCOPE	BIRCH	2	19-04-2023	05-05-2023	248,4		
DKVIBO	BIRCH	2	20-04-2023	14-05-2023	250,4		
FRSTRA	BIRCH	2	22-03-2023	05-05-2023	80,4		
HUGYOE	BIRCH	2	23-03-2023	24-04-2023	80,8		
HUMISK	BIRCH	2	16-04-2023	26-04-2023	338,8		LOCF
HUPEST	BIRCH	2	24-03-2023	26-04-2023	151,4		
HUSALG	BIRCH	2	12-04-2023	24-04-2023	613,4		
HUSZE2	BIRCH	2	24-03-2023	24-04-2023	141,3		

LTSIAU	BIRCH	2	19-04-2023	17-05-2023	162,7		
LTVILN	BIRCH	2	18-04-2023	15-05-2023	880,5		
MONTREAL	BIRCH	2	28-04-2023	24-05-2023	195,5		
NLLEID	BIRCH	2	12-04-2023	01-05-2023	143,2		
PLBIAL	BIRCH	2	17-04-2023	07-05-2023	277,5		
PLBYDG	BIRCH	2	17-04-2023	06-05-2023	283,7		
PLCRAC	BIRCH	2	13-04-2023	04-05-2023	260,2		
PLLODZ	BIRCH	2	16-04-2023	06-05-2023	464,1		
PLLUBL	BIRCH	2	16-04-2023	07-05-2023	327,4	PLPTRY	completely missing
PLPTRY	BIRCH	2	16-04-2023	07-05-2023	327,4		
PLSZCZ	BIRCH	2	17-04-2023	06-05-2023	283,7	PLBYDG	completely missing
PLWROC	BIRCH	2	15-04-2023	02-05-2023	409,7		
QUEBEC	BIRCH	2	05-05-2023	29-05-2023	243		LOCF
SKBRAT	BIRCH	2	25-03-2023	24-04-2023	138		
SKNITR	BIRCH	2	25-03-2023	24-04-2023	138	SKBRAT	completely missing

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ATWIEN	HAZEL	1	30-01-2022	17-03-2022	30		
CZTRIN	HAZEL	1	10-02-2022	23-03-2022	100,4		
DEAACH	HAZEL	1	09-02-2022	21-02-2022	81,1		
DEBOCH	HAZEL	1	28-01-2022	15-02-2022	55,3		
DEBORS	HAZEL	1	27-01-2022	24-02-2022	25,1		
DEDRES	HAZEL	1	28-01-2022	19-02-2022	31,9		
DELEVE	HAZEL	1	12-02-2022	18-02-2022	26,3		
DELOEW	HAZEL	1	28-01-2022	14-03-2022	23,2		
DEMARB	HAZEL	1	27-01-2022	13-03-2022	20,4		
DEMOEN	HAZEL	1	31-12-2021	15-02-2022	14,4		
FRSTRA	HAZEL	1	02-01-2022	13-03-2022	71,4		
HUGYOE	HAZEL	1	02-02-2022	24-03-2022	21,8		
HUMISK	HAZEL	1	05-02-2022	28-03-2022	53,9		
HUPEST	HAZEL	1	29-01-2022	31-03-2022	41,8		
HUSALG	HAZEL	1	05-02-2022	03-04-2022	69,3		

HUSZE2	HAZEL	1	03-02-2022	31-03-2022	47,1		
LTSIAU	HAZEL	1	12-03-2022	28-03-2022	64,2		
LTVILN	HAZEL	1	15-03-2022	26-03-2022	65,7		
PLBYDG	HAZEL	1	07-02-2022	24-03-2022	20,7		
PLCRAC	HAZEL	1	09-02-2022	21-03-2022	49		
PLKATO	HAZEL	1	09-02-2022	21-03-2022	46,2		
PLLODZ	HAZEL	1	12-02-2022	22-03-2022	22,3		LOCF
PLLUBL	HAZEL	1	13-02-2022	24-03-2022	42,1		
PLPTRY	HAZEL	1	09-02-2022	24-03-2022	31,4		
PLSZCZ	HAZEL	1	28-01-2022	30-01-2022	20,7		
RUMOSC	HAZEL	1	18-03-2022	12-04-2022			
SKBRAT	HAZEL	1	02-02-2022	01-05-2022	21,5		
ATWIEN	HAZEL	2	16-02-2023	10-03-2023	19,4		
BEBRUS	HAZEL	2	06-01-2023	18-02-2023	6	NLLEID	completely missing
BEMARC	HAZEL	2	31-12-2022	12-04-2023	28,9	DEMOEN	completely missing
CZTRIN	HAZEL	2	11-01-2023	26-02-2023	29,5		
DEBOCH	HAZEL	2	05-01-2023	23-02-2023	58,1		
DEDRES	HAZEL	2	03-02-2023	02-03-2023	37,3		
DELEVE	HAZEL	2	05-01-2023	07-01-2023	29,3		
DELIPP	HAZEL	2	13-02-2023	27-02-2023	39,9		
DELOEW	HAZEL	2	15-02-2023	06-03-2023	42,7		
DEMARB	HAZEL	2	05-01-2023	12-03-2023	10,3		
DEMOEN	HAZEL	2	31-12-2022	12-04-2023	28,9		
DEMUNC	HAZEL	2	04-01-2023	10-03-2023	31,7		
DKCOPE	HAZEL	2	11-02-2023	21-02-2023	25,1		
FRSTRA	HAZEL	2	02-01-2023	10-03-2023	89,6		
HUGYOE	HAZEL	2	13-01-2023	21-02-2023	5,8		
HUMISK	HAZEL	2	17-02-2023	13-03-2023	29,3		LOCF
HUPEST	HAZEL	2	01-01-2023	16-03-2023	17,4		
HUSALG	HAZEL	2	27-01-2023	16-03-2023	22,1		
HUSZE2	HAZEL	2	07-01-2023	10-03-2023	14,5		
LTSIAU	HAZEL	2	19-03-2023	22-03-2023	43,7		
LTVILN	HAZEL	2	18-03-2023	24-03-2023	51,4		
NLLEID	HAZEL	2	06-01-2023	18-02-2023	6		
PLBYDG	HAZEL	2	10-01-2023	19-03-2023	14		
PLCRAC	HAZEL	2	10-01-2023	14-03-2023	11,3		
PLKATO	HAZEL	2	21-02-2023	19-03-2023	18,7		
PLLODZ	HAZEL	2	17-03-2023	19-03-2023	14,7		
PLLUBL	HAZEL	2	06-02-2023	15-03-2023	15,6	PLPTRY	completely missing

PLPTRY	HAZEL	2	06-02-2023	15-03-2023	15,6		
PLSZCZ	HAZEL	2	10-01-2023	19-03-2023	14	PLBYDG	completely missing
SKBRAT	HAZEL	2	17-02-2023	11-03-2023	25,6		
SKNITR	HAZEL	2	17-02-2023	11-03-2023	25,6	SKBRAT	completely missing
ATWIEN	OAK	1	24-04-2022	20-05-2022	134,2		
CZTRIN	OAK	1	02-05-2022	22-05-2022	84,9		
DEAACH	OAK	1	14-04-2022	17-05-2022	253,3	DEMOEN	completely missing
DEBOCH	OAK	1	18-04-2022	14-05-2022	353,4		
DEBORS	OAK	1	01-05-2022	22-05-2022	786,7		
DEDRES	OAK	1	28-04-2022	11-05-2022	582,1		
DELEVE	OAK	1	14-04-2022	11-05-2022	193		
DELOEW	OAK	1	14-04-2022	13-05-2022	89,7		
DEMARB	OAK	1	03-05-2022	20-05-2022	430,1		
DEMOEN	OAK	1	14-04-2022	17-05-2022	253,3		
DKCOPE	OAK	1	11-05-2022	22-05-2022	60,5		
FRSTRA	OAK	1	13-04-2022	20-05-2022	136		
HUGYOE	OAK	1	30-04-2022	12-05-2022	32,2		
HUMISK	OAK	1	29-04-2022	22-05-2022	65,3		
HUPEST	OAK	1	24-04-2022	22-05-2022	91,3		
HUSALG	OAK	1	24-04-2022	23-05-2022	196,2		
HUSZE2	OAK	1	14-04-2022	19-05-2022	42,9		
LTSIAU	OAK	1	17-05-2022	28-05-2022	25,1		
LTVILN	OAK	1	19-05-2022	27-05-2022	40,4		
MONTREAL	OAK	1	13-05-2022	19-05-2022	32,6		LOCF
PLBYDG	OAK	1	28-04-2022	24-05-2022	74,9		
PLCRAC	OAK	1	28-04-2022	16-05-2022	101,2		
PLKATO	OAK	1	15-04-2022	21-05-2022	57,9		
PLLODZ	OAK	1	05-05-2022	21-05-2022	124,5		
PLLUBL	OAK	1	05-05-2022	21-05-2022	76,1		
PLPTRY	OAK	1	30-04-2022	23-05-2022	97,3		
PLSZCZ	OAK	1	07-05-2022	13-05-2022	24,3		
QUEBEC	OAK	1	18-05-2022	26-05-2022	94,2		
RUMOSC	OAK	1	07-05-2022	22-05-2022			
SKBRAT	OAK	1	25-03-2022	22-05-2022	115,4		
ATWIEN	OAK	2	21-04-2023	11-05-2023	39,5		
BEBRUS	OAK	2	03-05-2023	21-05-2023	46,1	NLLEID	completely missing
BEMARC	OAK	2	10-04-2023	21-05-2023	63,5	DEMOEN	completely missing
CZTRIN	OAK	2	11-05-2023	14-05-2023	29,8		
DEBOCH	OAK	2	09-04-2023	16-05-2023	89,2		

DEDRES	OAK	2	17-05-2023	25-05-2023	30,7	DEMARB	partially missing
DELEVE	OAK	2	25-04-2023	21-05-2023	83,7	DEMOEN	partially missing
DELIPP	OAK	2	28-04-2023	22-05-2023	112,1		LOCF
DELOEW	OAK	2	22-04-2023	10-05-2023	28,1		
DEMARB	OAK	2	17-05-2023	25-05-2023	30,7		
DEMOEN	OAK	2	10-04-2023	21-05-2023	63,5		
DEMUNC	OAK	2	03-05-2023	23-05-2023	33,1		
DKCOPE	OAK	2	10-05-2023	23-05-2023	27,5		
DKVIBO	OAK	2	12-05-2023	26-05-2023	91,8		
FRSTRA	OAK	2	23-03-2023	21-05-2023	31,9		
HUGYOE	OAK	2	22-04-2023	10-05-2023	12,4		
HUPEST	OAK	2	16-04-2023	09-05-2023	36,4		
HUSALG	OAK	2	28-04-2023	22-05-2023	76,7		
HUSZE2	OAK	2	11-04-2023	04-05-2023	55,7		
LTSIAU	OAK	2	11-05-2023	23-05-2023	39,7		
LTVILN	OAK	2	10-05-2023	16-05-2023	39,1		
MONTREAL	OAK	2	11-05-2023	22-05-2023	121,5		
NLLEID	OAK	2	03-05-2023	21-05-2023	46,1		
PLBYDG	OAK	2	23-04-2023	23-05-2023	18		
PLCRAC	OAK	2	23-04-2023	14-05-2023	49,1		
PLKATO	OAK	2	05-05-2023	18-05-2023	28,6		
PLLODZ	OAK	2	02-05-2023	18-05-2023	89,6		
PLLUBL	OAK	2	27-04-2023	23-05-2023	44,4	PLPTRY	completely missing
PLPTRY	OAK	2	27-04-2023	23-05-2023	44,4		
PLSZCZ	OAK	2	23-04-2023	23-05-2023	18	PLBYDG	completely missing
PLWROC	OAK	2	21-04-2023	18-05-2023	49,5		
QUEBEC	OAK	2	19-05-2023	29-05-2023	253,3		LOCF
SKBRAT	OAK	2	22-03-2023	11-05-2023	23,2		
SKNITR	OAK	2	22-03-2023	11-05-2023	23,2	SKBRAT	completely missing

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