CONFIDENTIAL

STATISTICAL ANALYSIS PLAN FOR PROTOCOL 206886

Dose Response of Three Experimental Dentifrices in Plaque Removal in a Single Brushing Model

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Glossary

AE	Adverse Event
ANCOVA	Analysis of Covariance
CI	Confidence Interval
ITT	Intention to Treat
κ	Kappa
MedDRA	Medical Dictionary for Regulatory Activities
OHT	Oral hard Tissue
OST	Oral Soft Tissue
PP	Per protocol
SOC	System of Organ
TPI	Turesky modification of Quigley Hein Plaque Index
w/w	Weight by weight

1 Introduction

This document describes the statistical methods and data presentations to be used in the summary and analysis of the final data from Protocol 206886.

2 Objectives

Primary	Endpoint
• To evaluate and compare the plaque removal efficacy, as measured by Turesky modification of Quigley Hein Plaque Index (TPI), of 67% w/w sodium bicarbonate, versus a 0% sodium bicarbonate toothpaste.	• Change from Pre-brushing to Post-brushing TPI
Secondary	**Endpoint**
• To evaluate and compare the plaque removal efficacy, as measured by TPI, of three toothpastes containing 20%, 35% and 50% w/w sodium bicarbonate, versus a 0% sodium bicarbonate toothpaste.	• Change from Pre-brushing to Post-brushing TPI
• To evaluate and compare the plaque removal efficacy, as measured by TPI, of three toothpastes containing 20%, 35% and 50% w/w sodium bicarbonate, versus a 67% sodium bicarbonate toothpaste.	• Change from Pre-brushing to Post-brushing TPI

3 Study Design

This will be a single centre, controlled, examiner blind, five treatment, five period, crossover design study in healthy volunteers. At the screening visit, following provision of written informed consent, all subjects will undergo an oral soft tissue (OST) examination and oral hard tissue (OHT) examination. Eligible subjects will be

provided with a standard wash-out toothpaste and toothbrush to use at home during the study; and for at least 7 days (maximum 28 days) prior to the first treatment visit (Visit 2).

For each treatment visit, subjects must abstain from oral hygiene for a period of 22 - 30 hours, immediately preceding the pre-brushing dental plaque evaluation.

At Visit 2, all the subjects will undergo an OST examination followed by disclosing and a pre-brushing dental plaque assessment (TPI). Subjects meeting the entry criteria will be to one of the five study treatments. Subjects will then perform a supervised brushing as per directions with the assigned test product. This will be followed by re-disclosing and a post-brushing plaque assessment. Subjects will brush with the washout paste following the post brushing plaque assessments to remove stain from the disclosing dye.

A 4 – 6 days washout period will follow each treatment period during which subjects will brush with the standard washout toothpaste. Subjects will complete five treatment visits and will brush once with each of the five test toothpastes throughout the course of the study.

At Visits 3, 4, 5 and 6, subjects will undergo the same assessments as performed at Visit 2.

At Visits 1, 2, 3, 4, 5 and 6, repeatability data will be generated for plaque assessment from replicate examinations on the same subject. If deemed necessary by the examiner, plaque may be re-disclosed if the dye has faded. Depending on subject visit scheduling, every effort will be made to complete repeatability examination for two subjects, that is, one in the morning and one in the afternoon on each assessment day. Repeatability examinations will be separated by a minimum of 10 minutes and, where possible, separated by another subject.

The five treatments of the study are as the following:

- **Test 1**: Experimental Dentifrice containing 20% w/w sodium bicarbonate;
- **Test 2**: Experimental Dentifrice containing 35% w/w sodium bicarbonate;
- **Test 3**: Experimental Dentifrice containing 50% w/w sodium bicarbonate;
- **Positive control**: Dentifrice containing 67% w/w sodium bicarbonate;
- **Negative control**: Dentifrice containing 0% w/w sodium bicarbonate.

4 Sample Size Determination

Sufficient healthy subjects will be screened by the study site so that a maximum of 56

subjects who fulfill all the entry criteria will be randomized, which should ensure that at least 50 evaluable subjects complete all study visits (thus allowing for at most a 10% drop-out).

With 50 subjects completing all study visits, the study has 90% power to detect a treatment difference of 0.15 in plaque index in a paired t-test of significance level 0.05. The standard deviation of difference (between treatments) is 0.32 as reviewed from the results of RH01455. As this is an exploratory study, multiplicity adjustment will not be applied.

5 Data Considerations

5.1 Analysis Populations

Safety population is defined as all subjects who are randomized and have received at least one dose of study products.

The intent to treat (ITT) population is defined as those subjects who are randomized, receive at least one dose of study product and have at least one post-baseline efficacy measurement.

The Per Protocol (PP) population will be a subset of the ITT population. Subjects with a protocol violation that is deemed to affect efficacy for all efficacy assessments will be excluded from the PP population. Subjects with a protocol violation that is deemed to affect efficacy for only some (but not all) of the efficacy assessments will be part of the PP population, but their data will be excluded from the assessment at which the protocol violation occurred. Efficacy analysis will be based on ITT population. A PP analysis will be performed only if 10% or more ITT subjects are excluded from PP population.

The repeatability population is defined as all subjects who have a repeat clinical assessment (TPI) at any visit.

Any of the following will be considered a protocol violation which will warrant exclusion from efficacy analysis:

- Violation of inclusion or exclusion criteria that are deemed to affect efficacy.
- Medical history which is deemed to affect efficacy.
- Use of prohibited treatment or medication before or during the study, which is felt to affect the assessment of efficacy.

- Not receiving randomized treatment.
- Noncompliance on treatment washout

Further data listings will be included in the review of protocol violations but will be reviewed on a case-by-case basis to determine whether the data should be excluded from a PP. Full data listings will be provided in review listing requirement document.

Protocol violations which warrant exclusion from efficacy analysis will be identified between the statistician and medical director or designee, ahead of database lock and breaking the study blind.

5.2 Subgroups/Stratification

There is no subgroup/Stratification in this study.

5.3 Time Windows

The study schedule should be followed as per protocol. Deviations from the study schedule with respect to visit timings will be reviewed on a case-by-case basis to determine whether the data should be excluded from PP analysis. Required time windows are presented below:

- Visit 2 — 7-28 days from Visit 1
- Visit 3 — 4-6 days from Visit 2
- Visit 4 — 4-6 days from Visit 3
- Visit 5 — 4-6 days from Visit 4
- Visit 6 — 4-6 days from Visit 5.

6 Demographics and Baseline Characteristics

6.1 Subject Disposition

The subject disposition summary will include the number of screened subjects and screen failures overall and the number of subjects randomised per treatment group and overall.

The number and percentage of subjects, in the Safety, ITT and PP populations will be presented per treatment group and overall. The percentages will be based upon the total number of subjects randomised.

The number and percentage of subjects completing the study and not completing the study, including a breakdown of the reasons for not completing the study, will be presented per treatment group and overall. The percentages are based upon the total number of subjects randomised.

A separate summary table of protocol violations leading to exclusion from PP analyses will be produced indicating the number and percentage of subjects with each violation per treatment group and overall. Percentages will be based on the ITT population.

6.2 Demographics

Descriptive statistics (number of subjects, mean, standard deviation, median, minimum and maximum for continuous variables, and frequency and percentage for categorical variables) will be provided for demographic data. These data include age, gender and ethnicity and will be presented for the Safety, ITT and PP populations.

6.3 Baseline Characteristics

Baseline efficacy measurements will be summarised in efficacy tables.

7 Treatment Compliance and Concomitant Medications

7.1 Treatment Compliance

Treatment compliance will be reviewed during blinded review and a listing will be produced for evaluation of protocol violations only. Non-compliance for extra/missed brushing will be assessed on a subject by subject basis. The data which are regarded as influenced by treatment non-compliance will be excluded from PP analysis. Any subject and/or time point excluded from PP analysis will be clearly documented in population definition document.

7.2 Concomitant Medications

Concomitant medication data will not be presented in the study report. A listing of concomitant medications will be produced for evaluation of protocol violators only.

8 Efficacy Analysis

Turesky modification of Quigley Hein Plaque Index (TPI) is the efficacy measure of the study. Treatment comparisons are under the hypotheses: H_0: 'treatment difference

is null' vs. H_a: 'treatment difference is not null'. All statistical tests will be conducted at the two-sided 5% significance level. All analysis will be conducted in SAS 9.2. The primary population for analysis will be ITT population.

8.1 Primary Efficacy Analysis

Primary efficacy endpoint will be the TPI score change from pre-brushing after a single brushing treatment. TPI score will be calculated as the average index over all tooth sites. Mixed effect ANCOVA model will be applied with treatment, study period as fixed effects, subject as a random effect and two baseline terms as covariates; (i) the subject-level baseline score calculated as the mean pre-brushing score across all periods within a subject, and (ii) the period level baseline minus the subject-level baseline. P-values for treatment comparisons, adjusted means of all treatments and treatment differences and their 95%CIs will be provided.

The primary analysis will be the comparison between the positive control and the negative control.

The assumption of residual normality and variance homogeneity in ANCOVA analysis will be investigated. If violated, data transformation or a non-parametric method (e.g., the Wilcoxon Signed-Rank test) will be applied. If violation is caused by several extreme values, a sensitivity analysis may be conducted by removing the extreme values.

8.2 Secondary Efficacy Analysis

Only if the primary objective is met (comparison of 67% w/w sodium bicarbonate, versus a 0% sodium bicarbonate is significant at two-sided 5% level), will the remaining secondary analyses be fully conducted. Otherwise no P-values will be provided for secondary comparisons. Only the estimates of treatment differences and confidence intervals will be provided.

Secondary analyses include the following treatment comparisons:

1. 50% sodium bicarbonate dentifrice versus a 0% sodium bicarbonate dentifrice;
2. 35% sodium bicarbonate dentifrice versus a 0% sodium bicarbonate dentifrice;
3. 20% sodium bicarbonate dentifrice versus a 0% sodium bicarbonate dentifrice;
4. 50% sodium bicarbonate dentifrice versus a 67% sodium bicarbonate dentifrice;
5. 35% sodium bicarbonate dentifrice versus a 67% sodium bicarbonate dentifrice;
6. 20% sodium bicarbonate dentifrice versus a 67% sodium bicarbonate dentifrice.

The analyses will be carried out by the same ANCOVA model in primary analysis.

In the ANCOVA analysis of the effects of the five treatments with different levels of sodium bicarbonate (0%, 20%, 35%, 50%, 67%), the linear and quadratic contracts will be tested for dose-response trend if the difference between the highest dose (67%) and lowest dose (0%) is significant. When a trend is significant (significance level 5%), a mixed effect regression (linear, quadratic etc) will be run to provide dose-response regression curve. For example the quadratic regression will take dose and dose^2 as explanatory variables and subject as random effect. The regression curve will be plotted together with the dose-response data. If neither linear model nor quadratic model appears to fit the data well (through residual check), other nonlinear models may also be considered.

8.3 Other Efficacy Analysis

All subjects who have repeat plaque (TPI) assessments (conducted by the examiner) form the repeatability population which will be used for repeatability analysis. The repeat assessments will be compared to the original assessments. The repeat assessments will not be used in any efficacy analysis. The first and second assessments on each tooth at a given visit will be cross-tabulated and a weighted Kappa coefficient (κ) will be calculated, along with the 95% confidence interval, to assess the intra-examiner repeatability. Repeatability will be deemed:

- Excellent, if $\kappa > 0.75$
- Fair to good, if $0.4 \leq \kappa \geq 0.75$
- Poor if $\kappa < 0.4$

All subjects who have repeatability data will be included in this analysis.

9 Safety Analysis

The safety profile of the study treatments will be assessed with respect to adverse events (AEs). Oral soft tissue (OST) abnormalities are included as AEs if they appear or worsen after the initial assessment. All safety data will be reported for the Safety population as per treatment received (using variable ATRT). All AEs will be reviewed by the Clinical Research Director or Scientist prior to database lock and unblinding and will be coded using the Medical Dictionary for Regulatory Activities

(MedDRA). During this review stage, AEs will be further categorized as oral or non-oral. AEs will be regarded as treatment emergent if they occur on or after the start time of the first treatment application (as determined by EXSTDT and EXSTTM from the EXPOSURE panel if this date is missing a suitable alternative will be used eg date and time of randomisation). All other AEs prior to this will be considered non-treatment emergent. The following summary tables and listings will be presented by treatment group.

- Table of treatment emergent AEs by Oral/Non-Oral and Preferred Term
- Table of treatment emergent AEs by SOC and Preferred Term
- Treatment emergent treatment related AEs by Oral/Non-Oral and Preferred Term
- Listing of all AEs (including Non-treatment emergent).
- Listing of serious AEs. (if there are none, a null listing will be produced)

No inferential analyses will be performed to compare treatments with respect to safety.

10 Interim Analysis

There is no interim analysis planned for this study.

11 Topline Summary

11.1 Variables for topline

Efficacy

TPI score change from pre-brushing after a single brushing treatment. Both primary comparison and secondary comparisons will be provided.

Safety

Adverse events

11.2 Outputs for topline

Datasets/Tables	Description
Datasets	PONNFL, POPNEXCL, RANDOM ADSL, ADAE, ADTPI, STAT1, STAT2
Tables – Efficacy	No efficacy tables required if information can be obtained directly from stats datasets otherwise define here
Tables Safety	9.4.1 – Listing Of Adverse Events □ Safety Population If there are <10 AEs the listing will be enough else:- 9.4.2 – Summary Of Treatment Emergent Adverse Events □ Safety Population
Figures	Generated from data from stats datasets
Non priority outputs	Table 9.1.1 – Subject Disposition

12 Changes to Planned Analysis

There is no change to the planned analysis.

13 References

GSKCH clinical study RH01445 □ Plaque Removal Efficacy of Four Dentifrices in a Single Brushing Model.

Appendix 1 Study Schedule

Activity	Visit 1 Screening		Visit 2 Treatment Period 1		Visit 3 Treatment Period 2		Visit 4 Treatment Period 3		Visit 5 Treatment Period 4		Visit 6 Treatment Period 5
Informed Consent	X	7 to 28 days wash-in		4 to 6 days washout		4 to 6 days washout		4 to 6 days washout		4 to 6 days washout	
Demographics & Medical History	X										
Current/concomitant medication	X		X		X		X		X		X
Oral soft tissue examination	X		X		X		X		X		X
Oral hard tissue examination	X										
Plaque disclosure	X										
Plaque assessment	X										
Repeatability of plaque assessment in selection of subjects	X		X		X		X		X		X
Inclusion/Exclusion criteria	X[1]		X[1]								
Dispense wash-out toothpaste, toothbrush, countdown timer and diary card with verbal instructions	X										
Return wash-out toothpaste, toothbrush and diary card			X		X		X		X		X
Re-dispense wash-out toothpaste, toothbrush and diary card			X		X		X		X		
Pre-brushing plaque disclosure			X		X		X		X		X
Pre-brushing plaque assessment			X		X		X		X		X
Randomisation			X								
Supervised brushing with assigned toothpaste			X		X		X		X		X
Post brushing plaque disclosure			X		X		X		X		X
Post-brushing plaque assessment[2]			X		X		X		X		X
Brushing with washout toothpaste to remove stain from disclosing dye	X		X		X		X		X		X
Compliance check			X		X		X		X		X
Subject Adherence/Eligibility check			X		X		X		X		X
Adverse events	X		X		X		X		X		X
Study Conclusion											X

[1] Plaque (inclusion criteria 4 C) will be assessed at Visit 1 and Visit 2; use of antibiotics and Chlorhexidine mouthwashes (exclusion criteria 7 A and 8 K) at Visit 2 to determine eligibility to continue.

Appendix 2 List of Tables, Figures & Listings

Table No.	Table Title (including population)	Standard	Template
9.1.1.1	Subject Disposition By Treatment Group – All Screened Subjects	X	
9.1.1.2	Subject Disposition By Treatment Group and Study Period – All Screened Subjects	X	
9.1.2	Protocol Violations Leading To Exclusion From Per Protocol Analysis – ITT Population	X	
9.2.1.1	Demographic Characteristics – Safety Population	X	
9.2.1.2	Demographic Characteristics – ITT Population	X	
9.3.1.1	Analysis Of Turesky Plaque Score Change from Pre-brushing – ITT Population		App 3
9.3.1.2*	Analysis Of Turesky Plaque Score Change from Pre-brushing – PP Population		9.3.1.1
9.3.2	Repeatability Analysis Of Turesky Plaque Index – Repeatability Population		App 3
9.4.1.1**	Listing Of Adverse Events – Safety Population	X	
9.4.1.2**	Listing Of Serious Adverse Events – Safety Population	X	
9.4.2	Treatment Emergent Adverse Events By Oral/Non Oral And PT – Safety Population	X	
9.4.3	Treatment Emergent Treatment Related Adverse Events By Oral/Non Oral And PT – Safety Population	X	
9.4.4	Treatment Emergent Adverse Events By SOC And PT – Safety Population	X	
9.4.5***	Treatment Emergent Non-Serious Adverse Events By SOC And PT – Safety Population	X	

*provided only if PP analysis is done.

** If there are non AEs a NULL listing will be provided. For 9.4.1.2, if there are more than 5 serious AEs a table will be done instead of listing.

*** Provided only if there are more than 5 serious AEs.

FIGURES

Figure No.	Figure Title (including population)	Standard	Template
9.1	Mean Turesky Plaque Score By Treatment – ITT Population		App 3
9.2	Scatter Plot With Regression Curve Over Dose Range [0,67] (% sodium bicarbonate) – ITT Population		App 3

Appendix 3 Templates for Tables, Figures & Listing

Treatment headers will be: Test 1; Test 2; Test 3; Positive Control; Negative Control.

For all tables add footnotes
Test 1: Experimental dentifrice containing 20% w/w sodium bicarbonate [redacted]
Test 2: Experimental dentifrice containing 35% w/w sodium bicarbonate
Test 3: Experimental dentifrice containing 50% w/w sodium bicarbonate
Positive Control: Dentifrice containing 67% w/w sodium bicarbonate (German marketed Parodontax Classic (non-fluoride) Toothpaste)
Negative Control: Dentifrice containing 0% w/w sodium bicarbonate (UK marketed Macleans Fresh Mint Toothpaste – 1450 ppm Fluoride)

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Table 9.3.1.1
Analysis Of Turesky Plaque Score Change from Pre-brushing
ITT Population

Study Population: ITT (N=xxx)

	Test 1 (N=xx)		Test 2 (N=xx)		...	Positive Control (N=xxx)		Negative Control (N=xx)	
	Raw	Change	Raw	Change	...	Raw	Change	Raw	Change
Pre-brushing									
N*	XX		XX		...	XX		XX	
MEAN	X.XX		X.XX		...	X.XX		X.XX	
SD	X.XXX		X.XXX		...	X.XXX		X.XXX	
SE	X.XXX		X.XXX		...	X.XXX		X.XXX	
MEDIAN	X.XX		X.XX		...	X.XX		X.XX	
MINIMUM	X.X		X.X		...	X.X		X.X	
MAXIMUM	X.X		X.X		...	X.X		X.X	
Post-brushing									
N*	XX	XX	XX	XX	...	XX	XX	XX	XX
MEAN	X.XX	X.XX	X.XX	X.XX	...	X.XX	X.XX	X.XX	X.XX
SD	X.XXX	X.XXX	X.XXX	X.XXX	...	X.XXX	X.XXX	X.XXX	X.XXX
SE	X.XXX	X.XXX	X.XXX	X.XXX	...	X.XXX	X.XXX	X.XXX	X.XXX
MEDIAN	X.XX	X.XX	X.XX	X.XX	...	X.XX	X.XX	X.XX	X.XX
MINIMUM	X.X	X.X	X.X	X.X	...	X.X	X.X	X.X	X.X
MAXIMUM	X.X	X.X	X.X	X.X	...	X.X	X.X	X.X	X.X
ADJUSTED MEAN [1]		X.XX	X.XX	X.XXX	...	X.XX	X.XX	X.XX	X.XX
SE [1]		X.XXX		X.XXX	...	X.XXX	X.XXX	X.XXX	X.XXX

TREATMENT COMPARISONS [1]	DIFFERENCE(CI)[1][2]	P-VALUE[1]
Pos.Control vs. Neg.Control	X.XX (X.XX, X.XX)	0.XXXX
Test 1 vs. Neg.Control	X.XX (X.XX, X.XX)	0.XXXX**
Test 2 vs. Neg.Control	X.XX (X.XX, X.XX)	0.XXXX**
Test.3 vs. Neg.Control	X.XX (X.XX, X.XX)	0.XXXX**
Test 1 vs. Pos.Control	X.XX (X.XX, X.XX)	0.XXXX**
Test.2 vs. Pos.Control	X.XX (X.XX, X.XX)	0.XXXX**
Test 3 vs. Pos.Control	X.XX (X.XX, X.XX)	0.XXXX**
Linear Contrast		0.XXXX**
Quadratic Contrast		0.XXXX**

*Number of subjects with non-missing values
**P-value for test products against two controls will be provided only if Pos.Control vs neg.Control is significant.
[1] From ANCOVA analysis for change from pre-brushing with treatment and period as fixed effect, subject as random effect, subject-level baseline and period-level minus subject-level baseline as covariates.
[2] Difference is first named treatment minus second named treatment such that a negative difference favors the first named treatment

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Table 9.3.2
Repeatability Analysis of Turesky Plaque Index
Repeatability Population

Study Population: Repeatability (N=xx)

	Second Assessment					
First Assessment [1]	Missing	0	1	2	3	4
MISSING	xx	xx	xx	xx	xx	xx
0	xx	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)
1	xx	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)
2	xx	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)
3	xx	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)
4	xx	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)	xx (xx.x%)

WEIGHTED KAPPA = 0.xxx

95% C.I. = 0.xxx, 0.xxx

Note: Percentages are based on total of all non-missing combinations
[1] The first assessment is the one used in the efficacy analysis.
0: No plaque
1: Slight flecks of plaque at the cervical margin of the tooth
2: A thin continuous band of plaque (1 mm or smaller) at the cervical margin of the tooth
3: A band of plaque wider than 1 mm but covering less than 1/3 of the crown of the tooth
4: Plaque covering at least 1/3 but less than 2/3 of the crown of the tooth
5: Plaque covering 2/3 or more of the crown of the tooth

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Figure 9.1
Mean TPI change from pre-brushing (± SE) by treatment
ITT Population

Study Population: ITT (N=xxx)

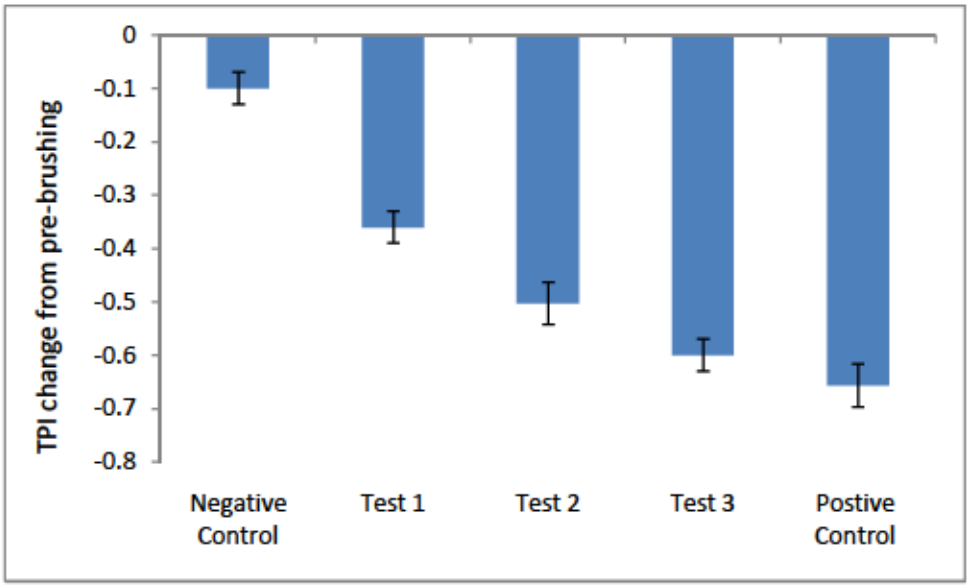

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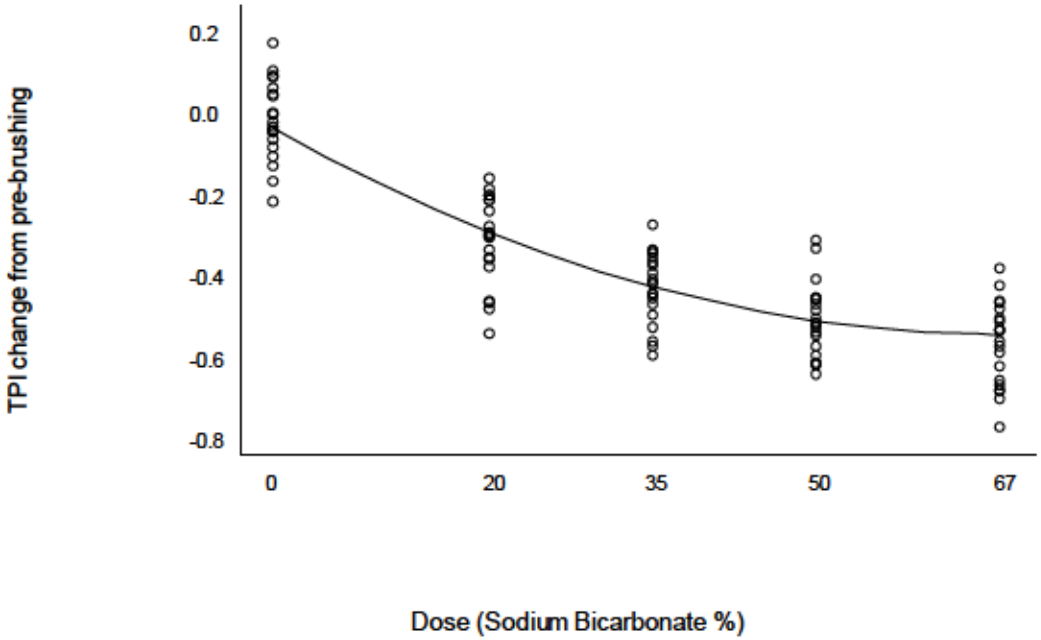
Figure 9.2
Scatter plot with regression curve over dose range [0,67] (% sodium bicarbonate)
ITT Population

Study Population: ITT (N=xxx)



Regression curve: TPI change = a + b*dose + c*dose^2 from mixed effect model with dose, dose^2 as regressors and subject as random effect.

SIGNATURE PAGE

206886 SAP

Date	Signed By
03-Jan-2017 10:59:49	PPD
Justification	Biostatistics Approval

Date	Signed By
04-Jan-2017 09:07:47	PPD
Justification	Approved

Date	Signed By
Justification	

Date	Signed By
Justification	

Date	Signed By
Justification	

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Justification	