A 2-arm, open label, prefatory study to explore changes in nasal mucociliary clearance between smokers and never smokers and to standardize nasal scraping procedure

STATISTICAL ANALYSIS PLAN

Version No. 1

24 February 2017

Prepared by:

[redacted]

Requested by:

INFLAMAX Research Inc

[redacted]

USA

TITLE PAGE

Study title:	A 2-arm, open label, prefatory study to explore changes in nasal mucociliary clearance between smokers and never smokers and to standardize nasal scraping procedure.
Study code:	15-LE-001-RD
Study type:	Exploratory study
Study drug:	Not applicable
Therapeutical area:	Not applicable
Sponsor:	INFLAMAX Research Inc [redacted] USA
Version no., date:	Version No. 1, 24 February 2017
Statistical report performed by:	[redacted]

SIGNATURE PAGE

STUDY TITLE: A 2-arm, open label, prefatory study to explore changes in nasal mucociliary clearance between smokers and never smokers and to standardize nasal scraping procedure.

PROTOCOL VERSION: Final 1.0

PROTOCOL DATE: 08-Dec-2016

SAP PREPARED BY:

[redacted]

[redacted] dd/mmm/yyyy

Senior Biostatistician

[redacted]

SAP REVIEWED BY:

[redacted]

[redacted] dd/mmm/yyyy

Senior Biostatistician

[redacted]

SAP APPROVED BY:

[redacted]

[redacted] dd/mmm/yyyy

Project Manager

Inflamax Research

TABLE OF CONTENTS

TABLE OF TABLES

TABLE OF FIGURES

TABLE OF LISTINGS

1 INTRODUCTION

This Statistical Analysis Plan (SAP) was written in accordance with Eurotrials' Standard Operating Procedures (SOP) ST.CO.01-01 for Biostatistics and intends to provide guidelines from which the analysis will proceed, create a common and clear understanding of the planned analysis by all involved, clarify issues which was not clarified in the protocol, expand statistical section of the protocol, provide basis for the statistical section of the statistical report and reduce the opportunity for bias by prospectively defining analysis. This SAP was prepared by the biostatistician who will be further responsible (if possible) for the statistical analysis of the study data.

This SAP, in particular table shells and examples of figures/graphs, was reviewed and approved by the Sponsor's Representative previously to database locking and performance of the statistical analysis.

Any changes to the planned statistical methodology/definitions described on this SAP during the statistical analysis of the study data will be documented in the Statistical Report.

This document was written in accordance with the information contained in the Study Protocol 15-LE-001-RD, Version Final 1.0 of 08-Dec-2016 and eCRF version Final V1.0 of 12Jan2017.

2 STUDY OBJECTIVES AND ENDPOINT

2.1 Primary objectives and endpoints

The primary objectives of this study are:

1. To evaluate the NMC over the course of 12 hours following single use of cigarette in smokers.
• STT value as assessed by STT test at each time point.

2. To compare NMC over the course of 12 hours in smokers following single use of cigarette relative to never smokers.
• STT value as assessed by STT test at each time point.

3. To examine the relationship between plasma nicotine levels and STT value in smokers and never smokers.
• STT value as assed by the STT test and plasma nicotine levels at each time point.

2.2 Secondary objectives and endpoints

The secondary objectives of this study are:

1. To standardize nasal scraping procedure using two methods.
• Collection of nasal epithelium for further histology.
• Evaluation of RNA quality and quantity.

2. To monitor the safety during the study.

• Vital signs.

• Adverse events (AE).

• Nasal and throat exam.

• Hematology, clinical chemistry and urine analysis.

• Brief physical examination.

• Concomitant medications.

3 STUDY DESIGN

This will be a single-center study in which 14 healthy adult male study participants, consisting of 7 cigarette smokers and 7 never smokers as a control group, will be enrolled. Study participants will not be replaced after being enrolled.

This study will have three visits on three separate days as described in Figure 1.

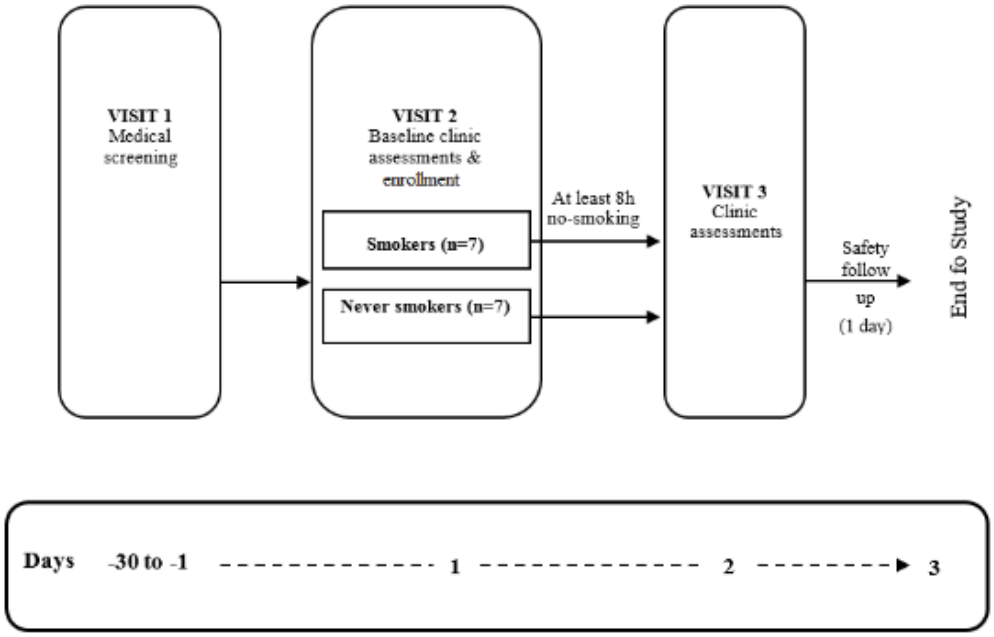

Figure 1 – Study flow chart

4 STUDY POPULATION

4.1 Inclusion criteria

1. Informed of the nature of the study and have agreed to and are able to read, review, and sign the informed consent form (ICF) prior to Screening. The subject must be willing to comply with the study procedures described in the informed consent. The informed consent document will be written in English, therefore the volunteer must have the ability to read and communicate in English.
2. Male subject aged ≥25 to ≤40 years old.
3. BMI between 18.0 kg/m^2 to 32.0 kg/m^2, inclusive.
4. Judged by the Principal Investigator or designee to be in good health as documented by the medical history, physical examination, vital sign assessments, clinical laboratory assessments, and by general observations.
5. Belong to one of the following two groups:
    a. Non-menthol cigarette smoker (meets all of the following criteria at Visit 1 and at Visit 2):
        i. A positive urine cotinine test (≥200 ng/mL).
        ii. Smoked at least 20 cigarettes per day for at least the past 5 years.
        iii. eCO levels >10 parts per million (ppm).
        iv. No plans to quit smoking in the next 3 months.
    b. Never smoker (meets all of the following criteria at Visit 1 and at Visit 2):
        i. Subject who has smoked less than 100 cigarettes throughout their lifetime and no cigarettes in the past 3 years.
        ii. A negative urine cotinine test (<200 ng/mL).
        iii. eCO levels ≤ 5 ppm.
6. Completed the Screening process within 30 days prior to Visit 2.
7. Availability for the entire study period and willingness to comply with study procedures, including smoking interruptions, as evidenced by a signed ICF and at Visit 2.

4.2 Exclusion criteria

1. As per the Principal Investigator or designee's judgment, the subject cannot participate in the study for any reason (e.g., medical, psychiatric, and/or social reason).
2. Subject is legally incompetent, or physically or mentally incapable of giving consent (e.g., emergency situation, under guardianship, prisoners, or subjects who are involuntarily incarcerated).
3. Presence of confounding allergies including allergic rhinitis and non-allergic rhinitis during the course of the study based on medical history and SPT.
4. Clinical significant abnormality on their nasal and throat exam, at the discretion of the Principal Investigator or designee at Visit 1 and/or at Visit 2.
5. Cigarette smoker who smoke/use any tobacco or nicotine products (other than CC), such as cigars, pipe, menthol cigarettes or electronic cigarettes in the previous 3 months, as self-reported at Visit 1 or Visit 2.
6. Never smoker who smoke/use any tobacco or nicotine products, such as cigars, pipe, menthol cigarettes or electronic cigarettes in the previous 3 years, as self-reported at Visit 1 or Visit 2.

7. Cigarette smokers who state they will be unable to abstain from smoking for up to 24 hours.

8. Inability to taste sweet within 60 minutes in the STT test.

9. Subjects who routinely use or who have used in the previous 4 weeks nasal sprays, inhalers or other nasal products, such as nasal irrigation (for example, Neti Pot) prior to Visit 1 and/or Visit 2.

10. Subjects who have taken any of the following medication without the indicated minimum washout period:

Prohibited Medication	Restriction period (with Principal Investigator or designee discretion)
Short-acting antihistamines including intranasal antihistamines	3 days before Visit 1 until Visit 2
Long-acting antihistamines (i.e. Loratadine, Desloratadine)	7 days before Visit 1 until Visit 2
Over-the-counter cough and cold preparations or sleep aids containing antihistamines	3 days before Visit 1 until Visit 2
Leukotriene inhibitors	14 days before Visit 1 until Visit 2
Oral or intra-articular steroid	30 days before Visit 1 until Visit 2
Intranasal and inhaled corticosteroids	14 days before Visit 1 until Visit 2
Use of monoamine oxidase inhibitors	14 days before Visit 1 until Visit 2
Decongestants	48 hours before Visit 1until Visit 2
Cromolyn products	14 days before Visit 1 until Visit 2
Beta-adrenergic blockers (i.e. Acebutolol, Atenolol, etc.)	14 days before Visit 1 until Visit 2
Anticholinergics	7 days before Visit 1 until Visit 2
Herbal or natural product remedies for allergy symptoms	On the day of Visit 1 until Visit 2
Short-Acting Beta Agonists	6 hours prior to spirometry (except as per protocol before spirometry)
Long-Acting Beta Agonists	3 days before Visit 1 until Visit 2
Phosphodiesterase 5 inhibitors (i.e. Sildenafil, Vardenafil, Tadalafil)	7 days before Visit 1 until Visit 2
Amiloride	3 days before Visit 1 until Visit 2
Macrolide antibiotics	7 days before Visit 1 until Visit 2
Guaifenesin	3 days before Visit 1 until Visit 2
Mucolytic agents	14 days before Visit 1 until Visit 2
Topical menthol products	14 days before Visit 1 until Visit 2
Topical nasal medication	4 weeks before Visit 1 until Visit 2
Topical ocular medication	4 weeks before Visit 1 until Visit 2
Any other medication at the Principal Investigator or designee's discretion that might interfere with the endpoints or procedures.	As per Principal Investigator or designee.

Table 1 – Prohibited medication.

11. Subjects with evidence of COPD, Global Initiative for Chronic Obstructive Lung Disease (GOLD) stage I or greater, and a forced expiratory volume 1 / the forced vital capacity ratio (FEV1/FVC ratio) <0.7.
12. Any condition the Principal Investigator or designee has cause to believe would interfere with the procedures for upper or lower airway function. This could include, but is not limited to, nasal/septum deviations, or nasal polyps or nasal allergies which will be identified by the Principal Investigator or designee.
13. Upper or lower respiratory diseases in the 4 weeks prior to Visit 2.
14. History of nasal or sinus surgery in the 5 years prior to Visit 2.
15. As per the Principal Investigator or designee's judgment, the subject has medical conditions which require or will require in the course of the study, a medical intervention (e.g., start of treatment, surgery, hospitalization) which may interfere with the study participation and/or study results.
16. The subject has a positive alcohol test and/or a history of alcohol abuse that could interfere with the subject's participation in study at Visit 1 or Visit 2.
17. Positive urine drug screen at Visit 1 or Visit 2.
18. The subject has positive serology test for human immunodeficiency virus (HIV)1/2, Hepatitis B or Hepatitis C.
19. Subject has donated or been in receipt of whole blood or blood products within 3 months prior to Visit 1.
20. Subject is a current or former employee of the tobacco industry or of their first-degree relatives (spouse, legal patner, parent, sibling, and child).
21. Subject is an employee of the investigational site or any other parties involved in the study or of their first-degree relatives (spouse, legal patner, parent, sibling, and child).
22. Subject has been in receipt of last dose from another clinical study within 3 months prior to Visit 1.
23. Subject has been previously screened in this study.

5 STATISTICAL METHODS

5.1 Sample size

This is an exploratory study. This study will include 14 study participants: 7 cigarette smokers and 7 never smokers. No formal powering has occurred. However, with an expected standard deviation of 2.5 minutes, a difference in STT of 3.75 minutes can be detected.

5.2 Statistical software

The statistical analyses will be conducted through the software SAS® (version 9.4, SAS Institute Inc, Cary).

5.3 Analysis datasets

5.3.1 Full Analysis Set (FAS)

All study participants who have at least one evaluable STT test at Visit 3 will be included in FAS population.

5.3.2 Per Protocol (PP) Population

All study participants in Full Analysis Set who have no major protocol deviation will be included in the PP population.

5.3.3 Safety Population

All enrolled subjects will be included in safety population.

5.4 Study hypothesis

There are no statistical hypotheses to be tested for the primary analysis.

5.5 Statistical analysis

All quantitative variables will be summarized through descriptive statistics namely mean, median, standard deviation, quartiles (Q1 and Q3) and minimum and maximum values. Qualitative variables will be summarized through number (n) and frequency distribution (%).

All descriptive analyses will be performed by smoker and never smoker group.

Statistical tests will be two-tailed considering a significance level of 5%.

5.5.1 Subjects' disposition

The number of subjects included in this study will be described in this section. The number (n) and frequency distribution (%) of subjects who completed the study, subjects who discontinued the study and distribution for each study dataset will be also summarized by total, smoker and never smoker groups.
Subjects' disposition will be presented as described in **Table A 1** and listed in **Listing 1**. Protocol deviations will be listed by subject in **Listing 2**.

5.5.2 Demographic and other clinical assessments

Demographics at visit 1 will be summarized by total number of observations (n), namely mean, median, standard deviation, quartiles (Q1 and Q3) and minimum and maximum values for continuous variables, and total number of observations, number (n) and frequency distribution (%) for categorical variables.

Demographics and other assessments will be presented for FAS and PP dataset as described in **Table A 2** and **Table A 3**, respectively and listed in **Listing 3**.

Smoking history, medical history, prior and concomitant medication, exhaled carbon monoxide, drug/alcohol test, skin prick test and tobacco consumption will be listed by subject in **Listing 4** to **Listing *10***, respectively.

5.5.3 Primary analysis

STT for each time point will be summarized by smoker and never smoker groups using the descriptive statistics: mean, median, standard deviation, quartiles (Q1 and Q3), minimum and maximum values and 95% confidence interval (CI).
Changes in STT from T0 will be computed at T4, T8 and T12 and summarized by smoker and never smoker group using the descriptive statistics: mean, minimum, maximum, median, quartiles (Q1 and Q3), standard deviation (SD) and 95% confidence interval (CI).
The plasma nicotine levels for each time point will be summarized by smoker and never smoker group using the descriptive statistics mean, minimum, maximum, median, quartiles, standard deviation (SD) and 95% confidence interval (CI). The relationship between plasma nicotine levels and STT at each time point will be evaluated by spearman correlation as well as graphical presentation of these endpoints by time point and smoker and never smoker group.

Results regarding STT, plasma nicotine and STT versus plasma nicotine will be presented for FAS and PP dataset as described in **Table A 4** to **Table A 8** and in **Figure A 1** to **Figure A 4**.
Data regarding STT test will be listed by subject in **Listing 11**.

5.5.4 Secondary analysis

5.5.4.1 Nasal scraping procedures

Descriptive statistics of RNA results will be summarized by smoker and never smoker groups using total number of observations (n), mean, median, standard deviation, quartiles (Q1 and Q3) and minimum and maximum values.
Nasal scraping procedures will be presented for FAS dataset as described in **Table A 9**.

5.5.4.2 Safety analysis

Vital signs (systolic blood pressure, diastolic blood pressure, pulse rate and respiratory rate) will be summarized by total number of observations (n), mean, median, standard deviation, quartiles (Q1 and Q3) and minimum and maximum values.

An overall summary of AEs will be presented by smoker and never smoker groups showing the number of events and percent of study participants who experienced AEs, SAEs, severe AEs and AEs leading to study discontinuation.
A summary of AEs by System Organ Class (SOC) and Preferred Term (PT) according to Medical Dictionary for Regulatory Activities (MedDRA) by smoker and never smoker groups will be presented.

Each parameter of hematology, clinical chemistry and urine will be summarized over time by smoker and never smoker group thought total number of observations (n), mean, median, standard deviation, quartiles (Q1 and Q3) and minimum and maximum values. Number (n) and frequency distribution (%) of normal and abnormal values will also be present by each parameter.

All safety analyses will be performed for the safety dataset only.

Vital signs measures will be presented for safety dataset in **Table A 10** and listed in **Listing 12**

Incidence and frequency distribution of AEs, SAEs, AEs with reasonable possibility of relationship with study drug will be presented in **Table A 11** and **Table A 12**. All adverse events and serious adverse events will also be listed by subject in **Listing 13** and **Listing 14**.

Regarding laboratory parameters, results of hematology, chemistry and urinalysis will be presented for safety dataset in **Table A 13**, **Table A 14** and **Table A 15** and listed by subject in **Listing 15**.
Results on serology at visit 1 will be presented in **Table A 16**.

5.6 Missing values

Missing STT values for a time point will be treated as missing. When STT detection takes longer than 1 hour, the STT will be considered as 1 hour. When the subject is not able to test saccharine on direct application, the STT values should be treated as missing, and these subjects should not be included in the STT analysis.

Regarding plasma nicotine samples:

- Missing sampling time: if the time of collection for a sample is unknown, that individual data point will be treated as missing data for descriptive statistics.
- Missing concentration data: missing concentration data will be treated as missing and replaced with a "." in the concentration dataset. No values will be imputed.
- Concentration data below lower limit of quantification (LLOQ=0.399 ng/mL): in the calculation of descriptive statistics for concentration data at each sampling time point, all LLOQ values will be treated as zero for time zero and as 1/2 LLOQ for the remaining time

6 CLARIFICATIONS TO THE STUDY PROTOCOL

In the study protocol, the populations that should be considered in each statistical analysis is not clear. In this SAP the following assumptions were considered:

- Statistical analysis for demographics will be performed for the FAS and PP population;
- Primary analysis will be performed for the FAS and PP population;
- Safety analysis will be performed for the safety population.

7 APPENDIX 1: DERIVED VARIABLES

The following variables will be derived:

Group of subjects – A dichotomous variable (smoker/never smoke) will be calculated.

Smoker – subjects who meet all of the following criteria at Visit 1 and at Visit 2:

- A positive urine cotinine test (=200 ng/mL).
- Smoked at least 20 cigarettes per day for at least the past 5 years.
- eCO levels >10 parts per million (ppm).
- No plans to quit smoking in the next 3 months.

Never smoker – subjects who meet all of the following criteria at Visit 1 and at Visit 2:

- Subject who has smoked less than 100 cigarettes throughout their lifetime and no cigarettes in the past 3 years.
- A negative urine cotinine test (<50 ng/mL).
- eCO levels = 5 ppm.

STT – At each time point, STT value will be obtained as the difference between end time and start time, in minutes, of STT test. When STT detection takes longer than 1 hour, the STT will be considered as 1 hour (60 minutes). When the subject is not able to test saccharine on direct application, the STT values should be treated as missing, and these subjects should not be included in the STT analysis.

8 APPENDIX 2: TABLES AND FIGURES

Table A 1 – Subjects' disposition

	Total	Smoker	Never smoker
No. of subjects included in the study, n (%)			
Subjects who completed the study, n (%)[a]			
Subjects who discontinued, n (%)[a]			
Adverse event			
Best interest for study participant			
Non-compliance with study procedures			
Study terminated by investigator			
Study terminated by sponsor			
Withdrawal by subject			
Other			
Analysis populations, n (%)[a]			
FAS			
PP			
Safety			

FAS: Full analysis set. PP: Per protocol.
a) Percentages calculated within included subjects.

Table A 2 – Demographic and anthropometric characteristics at visit 1 – FAS population

	Smoker (n=xx)	Never smoker (n=xx)
Age (years)		
N		
Mean		
Median		
Standard Deviation		
Q1		
Q3		
Minimum		
Maximum		
Gender, n (%)		
Male		
Female		
Total		
Ethnicity, n (%)		
Hispanic or Latino		
Not Hispanic or Latino		
Total		
Race, n (%)		
Asian		
American Indian or Alaska native		
Black or African American		
Native Hawaiian / other pacific islander		
White		
Total		
Height (cm)		
N		
Mean		
Median		
Standard Deviation		
Q1		
Q3		
Minimum		
Maximum		
Weight (kg)		
N		
Mean		
Median		
Standard Deviation		
Q1		
Q3		
Minimum		
Maximum		
BMI (kg/m^2)		
N		
Mean		
Median		
Standard Deviation		
Q1		
Q3		
Minimum		
Maximum		

BMI: body mass index.

Table A 3 – Demographic and anthropometric characteristics at visit 1 – PP population

	Smoker (n=xx)	Never smoker (n=xx)
Age (years)		
N		
Mean		
Median		
Standard Deviation		
Q1		
Q3		
Minimum		
Maximum		
Gender, n (%)		
Male		
Female		
Total		
Ethnicity, n (%)		
Hispanic or Latino		
Not Hispanic or Latino		
Total		
Race, n (%)		
Asian		
American Indian or Alaska native		
Black or African American		
Native Hawaiian / other pacific islander		
White		
Total		
Height (cm)		
N		
Mean		
Median		
Standard Deviation		
Q1		
Q3		
Minimum		
Maximum		
Weight (kg)		
N		
Mean		
Median		
Standard Deviation		
Q1		
Q3		
Minimum		
Maximum		
BMI (kg/m^2)		
N		
Mean		
Median		
Standard Deviation		
Q1		
Q3		
Minimum		
Maximum		

BMI: body mass index.

Table A 4 – Saccharin transit time – FAS population

	Visit 1		Visit 2		Visit 3 –T0		Visit 3 – T4		Visit 3 – T8		Visit 3 – T12	
	Smoker (n=xx)	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**
STT result (min)												
N												
Mean												
Median												
Standard Deviation												
Q1												
Q3												
Minimum												
Maximum												
95% CI												
Change from T0												
N	-	-	-	-	-	-						
Mean	-	-	-	-	-	-						
Median	-	-	-	-	-	-						
Standard Deviation	-	-	-	-	-	-						
Q1	-	-	-	-	-	-						
Q3	-	-	-	-	-	-						
Minimum	-	-	-	-	-	-						
Maximum	-	-	-	-	-	-						
95% CI												

STT value will be obtained as the difference between end time and start time, in minutes (see appendix 1).

Table A 5 – Saccharin transit time – PP population

	Visit 1		Visit 2		Visit 3 –T0		Visit 3 – T4		Visit 3 – T8		Visit 3 – T12	
	Smoker (n=xx)	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**
STT result (min)												
N												
Mean												
Median												
Standard Deviation												
Q1												
Q3												
Minimum												
Maximum												
95% CI												
Change from T0												
N	-	-	-	-	-	-						
Mean	-	-	-	-	-	-						
Median	-	-	-	-	-	-						
Standard Deviation	-	-	-	-	-	-						
Q1	-	-	-	-	-	-						
Q3	-	-	-	-	-	-						
Minimum	-	-	-	-	-	-						
Maximum	-	-	-	-	-	-						
95% CI												

STT value will be obtained as the difference between end time and start time, in minutes (see appendix 1).

Table A 6 – Plasma nicotine sample at Visit 3 – FAS population

	Visit 3 –T0		Visit 3 – T4		Visit 3 – T8		Visit 3 – T12	
	Smoker (n=xx)	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**
Result (%)								
N								
Mean								
Median								
Standard Deviation								
Q1								
Q3								
Minimum								
Maximum								
95% CI								

Table A 7 – Plasma nicotine sample at Visit 3 – PP population

	Visit 3 –T0		Visit 3 – T4		Visit 3 – T8		Visit 3 – T12	
	Smoker (n=xx)	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**
Result (%)								
N								
Mean								
Median								
Standard Deviation								
Q1								
Q3								
Minimum								
Maximum								
95% CI								

Table A 8 – Plasma nicotine sample versus STT

	FAS population		PP population	
	Smoker (n=xx)	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**
Plasma nicotine vs STT at Visit 3 - T0				
p-value				
Spearman correlation coefficient				
Plasma nicotine vs STT at Visit 3 - T4				
p-value				
Spearman correlation coefficient				
Plasma nicotine vs STT at Visit 3 - T8				
p-value				
Spearman correlation coefficient				
Plasma nicotine vs STT at Visit 3 - T12				
p-value				
Spearman correlation coefficient				

STT: Saccharin transit time.

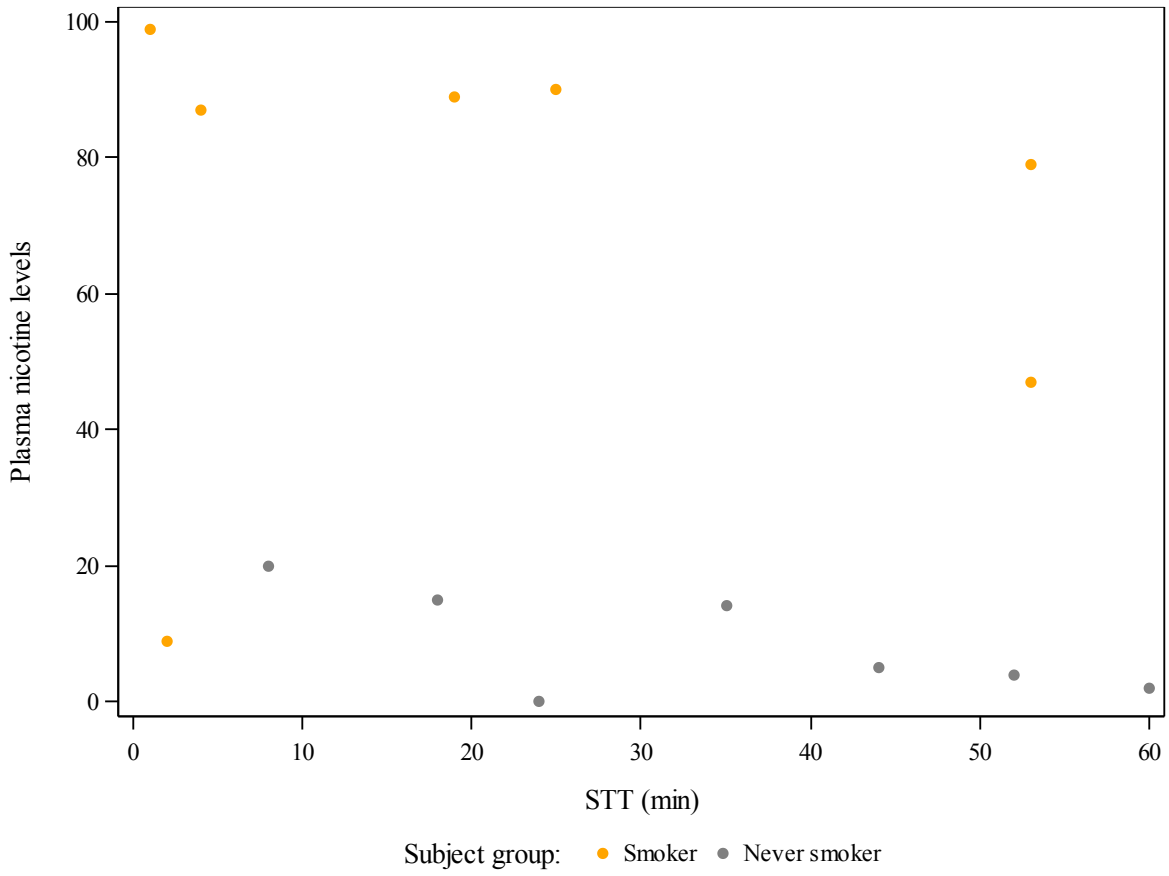

Figure A 1 – Plasma nicotine levels versus STT by smokers and never smokers, at T0 – FAS population

(data is illustrative only)

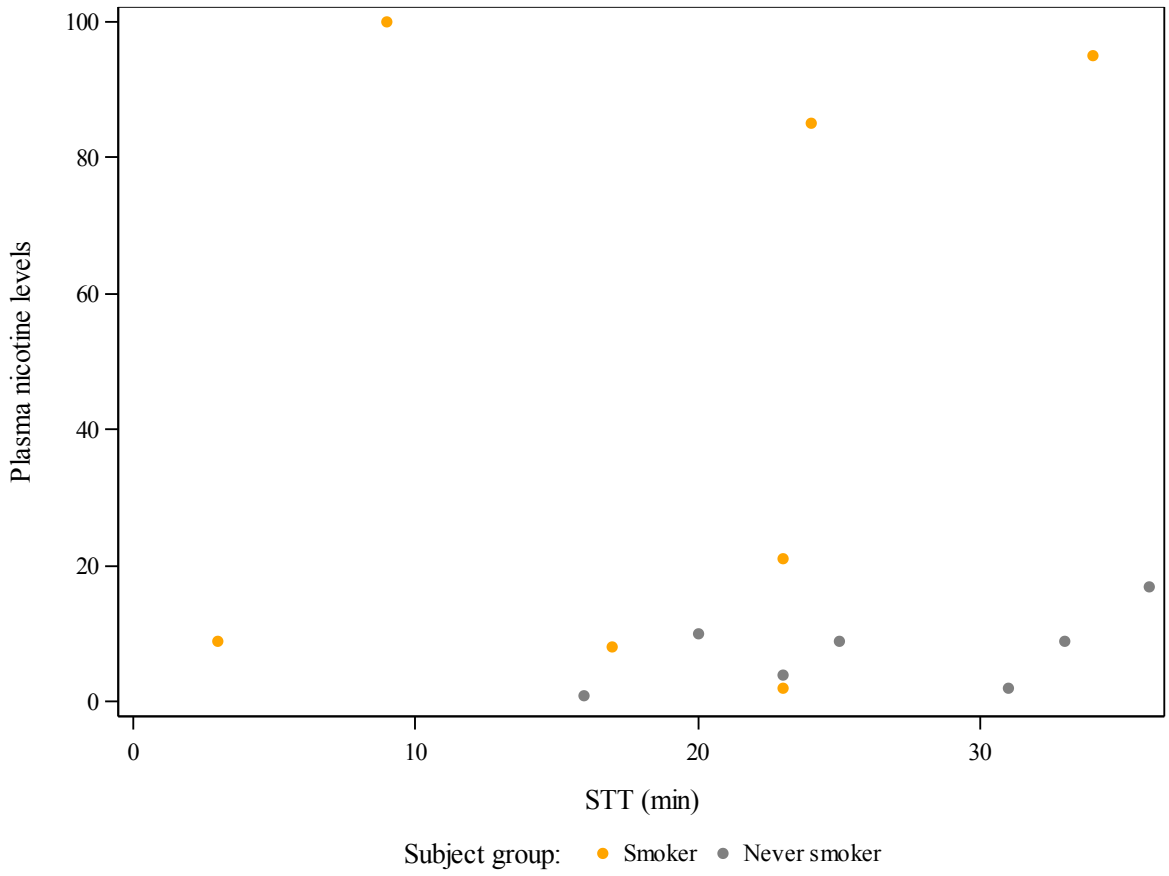

Figure A 2 – Plasma nicotine levels versus STT by smokers and never smokers, at T4 – FAS population

(data is illustrative only)

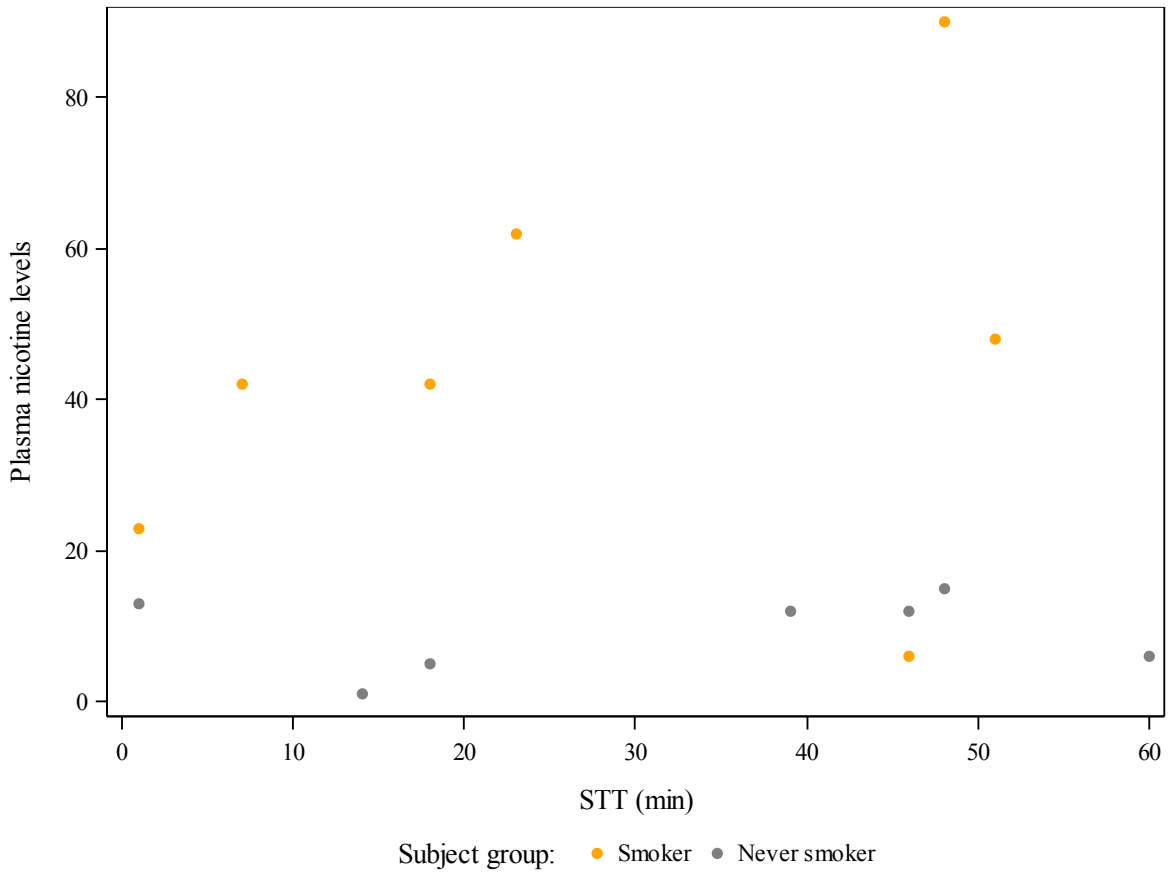

Figure A 3 – Plasma nicotine levels versus STT by smokers and never smokers, at T8 – FAS population

(data is illustrative only)

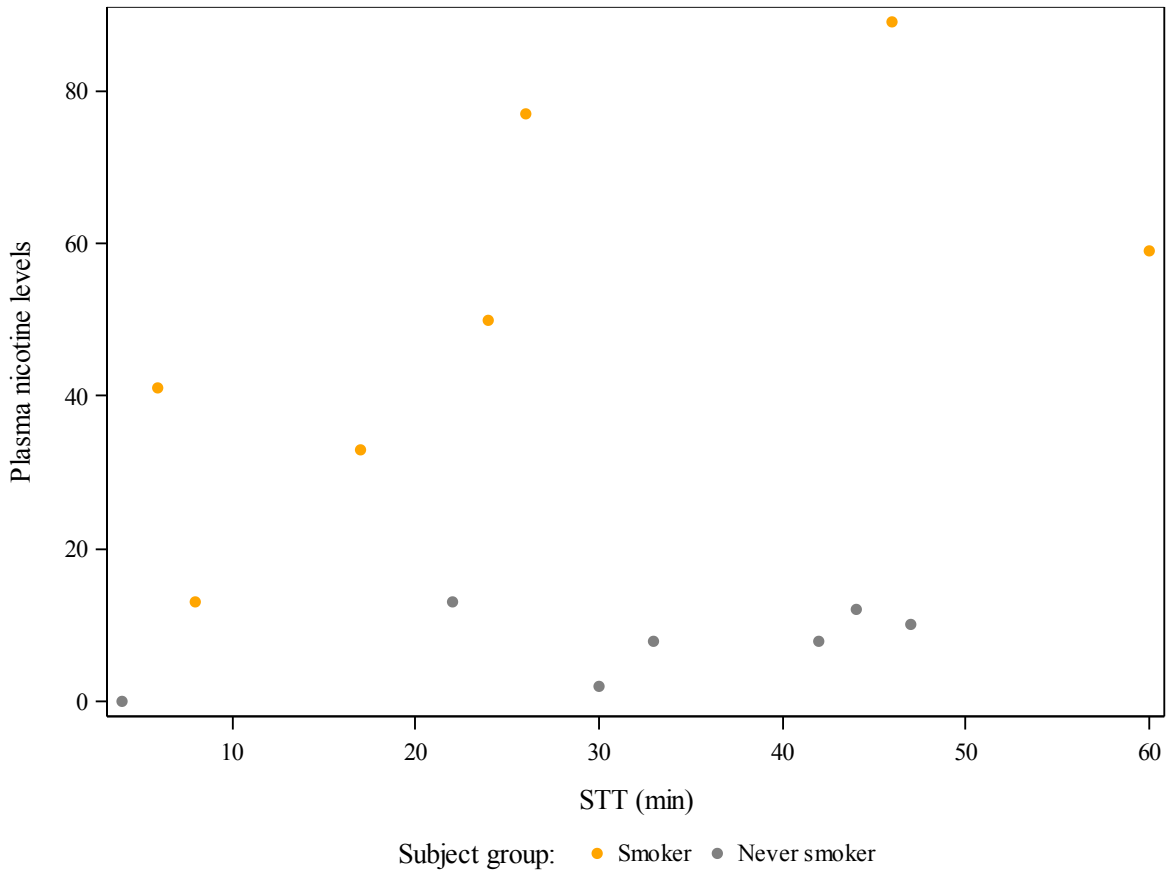

Figure A 4 – Plasma nicotine levels versus STT by smokers and never smokers, at T12 – FAS population

(data is illustrative only)

Table A 9 – Nasal scraping at visit 3 – FAS population

	Total (n=xx)
Method, n (%)	
Left nostril (method 1)	
Right nostril (method 2)	
Total	
Only method 1 (5 subjects)	
RNA	
N	
Mean	
Median	
Standard Deviation	
Q1	
Q3	
Minimum	
Maximum	
Only method 2 (5 subjects)	
RNA	
N	
Mean	
Median	
Standard Deviation	
Q1	
Q3	
Minimum	
Maximum	
Method 1 or 2 (10 subjects)	
RNA	
N	
Mean	
Median	
Standard Deviation	
Q1	
Q3	
Minimum	
Maximum	
Method 1 and 2 (4 subjects)	
RNA	
N	
Mean	
Median	
Standard Deviation	
Q1	
Q3	
Minimum	
Maximum	

Table A 10 – Vital signs – Safety population

	Visit 1		Visit 2		Visit 3	
	Smoker (n=xx)	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**
Systolic blood pressure (mmHg)						
N						
Mean						
Median						
Standard Deviation						
Q1						
Q3						
Minimum						
Maximum						
Diastolic blood pressure (mmHg)						
N						
Mean						
Median						
Standard Deviation						
Q1						
Q3						
Minimum						
Maximum						
Pulse rate (beats/min)						
N						
Mean						
Median						
Standard Deviation						
Q1						
Q3						
Minimum						
Maximum						
Respiratory rate (breaths/min)						
N						
Mean						
Median						
Standard Deviation						
Q1						
Q3						
Minimum						
Maximum						

Table A 11 – Overall summary of adverse events – Safety population

	Smoker (n=xx)		Never smoker (n=xx)	
	n (%)	**nAEs**	**n (%)**	**nAEs**
Any adverse event, n (%)				
Any serious adverse event, n (%)				
Any severe adverse event, n (%)				
Any adverse event leading to study discontinuation, n (%)				

n (%) = Number (percent) of subjects.
nAE = Number of adverse events.

Table A 12 – Adverse events – Safety population

SOC PT	Smoker (n=xx)		Never smoker (n=xx)	
	n (%)	nAEs	n (%)	nAEs
SOC 1, n (%)				
xxxxx				
xxxxx				
xxxxx				
SOC 2, n (%)				
xxxxx				
xxxxx				
xxxxx				
Etc…				

n (%) = Number (percent) of subjects.
nAE = Number of adverse events.

Table A 13 – Hematology – Safety population

	Visit 1		Visit 3	
	Smoker (n=xx)	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**
Hematocrit (unit)				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				
Total				
Hemoglobin (unit)				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				
Total				
MCH (unit)				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				
Total				
MCHC (unit)				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				
Total				
MCV (unit)				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				

Total

Platelet count (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

RBC (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

WBC (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Neutrophils (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Basophils (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Eosinophils (unit)
N
Mean
Median

Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Lymphocytes (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Monocytes (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Table A 14 – Clinical chemistry – Safety population

	Visit 1		Visit 3	
	Smoker (n=xx)	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**
Albumin (unit)				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				
Total				
Total protein (unit)				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				
Total				
AP (unit)				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				
Total				
ALT (unit)				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				
Total				
AST (unit)				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				

Total

BUN (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Creatinine (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Fasting glucose (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Fibrinogen (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

GGT (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

LDH (unit)
N
Mean
Median

Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Potassium (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Sodium (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Total bilirubin (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Direct bilirubin (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Total cholesterol (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Triglycerides (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

AP: Alkaline phosphatase. ALT: alanine aminotransferase. AST: aspartate aminotransferase. BUN: blood urea nitrogen. GGT: Gamma-glutamyl transferase. LDH: lactate dehydrogenase.

Table A 15 – Urine – Safety population

	Visit 1		Visit 3	
	Smoker (n=xx)	**Never smoker (n=xx)**	**Smoker (n=xx)**	**Never smoker (n=xx)**
pH				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				
Total				
Bilirubin (unit)				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				
Total				
Glucose (unit)				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				
Total				
Nitrite (unit)				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				
Total				
Red blood cells trace (unit)				
N				
Mean				
Median				
Standard Deviation				
Q1				
Q3				
Minimum				
Maximum				
Normal/abnormal, n (%)				
Normal				
Abnormal				

Total

Protein (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Specific gravity (unit)
N
Mean
Median
Standard Deviation
Q1
Q3
Minimum
Maximum

Normal/abnormal, n (%)
Normal
Abnormal
Total

Table A 16 – Serology at visit 1 – Safety population

	Smoker (n=xx)	Never smoker (n=xx)
Hepatitis B surface antigen, n (%)		
Positive		
Negative		
Total		
Hepatitis C virus, n (%)		
Positive		
Negative		
Total		
Anti-HIV1/2, n (%)		
Positive		
Negative		
Total		

HIV: human immunodeficiency virus.

9 APPENDIX 2: LISTINGS

Listing 1 – Subjects' disposition

Subject	Subject complete the study	Reason for discontinuation	Group	FAS	PP	Safety

FAS: Full analysis set. PP: Per protocol.

Listing 2 – Protocol deviations

Subject	Protocol deviation	Group	FAS	PP	Safety

FAS: Full analysis set. PP: Per protocol.

Listing 3 – Demographic and anthropometric characteristics at visit 1

Subject	Age	Sex	Ethnicity	Race	Height (cm)	Weight (kg)	BMI (kg/m^2)	Group	FAS	PP	Safety

FAS: Full analysis set. PP: Per protocol.

Listing 4 – Smoking history

Subject	Subject smoked 100 cigarettes or more in their life	Subject planning to quit smoking in the next 3 months	Subject smoked at least 20 cigarettes per day for at least the past 5 consecutive years	No. of years that subject smoked cigarettes in their entire life	No. of cigarettes smoked per day	Subject mostly exhales smoke through the nose	Use of E-cigarette over the last 3 months	Use of Cigar over the last 3 months	Use of Pipe or hookah over the last 3 months

Use of Snuff or Chewing Tobacco over the last 3 months	Use of Nicotine product to help quit smoking over the last 3 months	Use of other over the last 3 months	Use of other tobacco/nicotine containing products over the last 3 months	Subject smoked any cigarettes or more in the past 3 years	Use of E-cigarette over the last 3 years	Use of Cigar over the last 3 years	Use of Pipe or hookah over the last 3 years

Use of Snuff or Chewing Tobacco over the last 3 years	Use of Nicotine product to help quit smoking over the last 3 years	Use of other over the last 3 years	Use of other tobacco/nicotine containing products over the last 3 years	Brand of cigarettes	Group	FAS	PP	Safety

FAS: Full analysis set. PP: Per protocol.

Listing 5 – Medical history

Subject	MH subcategory	MH term	Start date	Ongoing	End date	Group	FAS	PP	Safety

FAS: Full analysis set. PP: Per protocol.

Listing 6 – Prior and concomitant medication

Subject	Medication	Dose	Dose units	Frequency	Route	Start date	ongoing	End date	Indication	Used to treat AE or MH?	AE or MH	Group	FAS	PP	Safety

AE: Adverse event. MH: Medical history. FAS: Full analysis set. PP: Per protocol.

Listing 7 – Exhaled Carbon Monoxide

Subject	Visit	Collection date	Exhaled Carbon Monoxide level (ppm)	Group	FAS	PP	Safety

FAS: Full analysis set. PP: Per protocol.

Listing 8 – Drug/alcohol test

Subject	Collection time	Panel name	Test name	Result	Group	FAS	PP	Safety

FAS: Full analysis set. PP: Per protocol.

Listing 9 – Skin prick test

Subject	Skin prick test date	Allergen	Mean wheal diameter (mm)	Group	FAS	PP	Safety

FAS: Full analysis set. PP: Per protocol.

Listing 10 – Tobacco consumption

Subject	Visit	Current use of tobacco	Tobacco consumption date	Tobacco consumption time	Group	FAS	PP	Safety

FAS: Full analysis set. PP: Per protocol.

Listing 11 – Saccharin transit time test

Subject	Visit	Date	STT test performed	STT test date	Start time	End time	Was the subject able to perceive sweet taste within 60 minutes time?	Did the study participant taste the saccharin after direct application?	Group	FAS	PP	Safety

STT: Saccharin transit time. FAS: Full analysis set. PP: Per protocol.

Listing 12 – Vital signs

Subject	Visit	Systolic blood pressure (mmHg)	Diastolic blood pressure (mmHg)	Pulse rate (beats/min)	Respiratory rate (breaths/min)	Group	FAS	PP	Safety

FAS: Full analysis set. PP: Per protocol.

Listing 13 – Adverse events

Subject	AE	Start date	ongoing	End date	Serious	Severity	Relationship to study procedures	Action taken	Outcome	Study discontinuation	Group	FAS	PP	Safety

AE: Adverse event. FAS: Full analysis set. PP: Per protocol.

Listing 14 – Serious adverse events

Subject	AE	Congenital anomaly or birth defect	Significant disability	Death	Hospitalization	Life threatening	Other medical important event	Group	FAS	PP	Safety

AE: Adverse event. FAS: Full analysis set. PP: Per protocol.

Listing 15 – Labs (hematology, chemistry and urine analysis)

Subject	Collection date	Panel name	Test name	Result	Normal range	Normal range indicator	Clinically significant	Is abnormal test result related with an adverse event or concomitant medication?	Group	FAS	PP	Safety

FAS: Full analysis set. PP: Per protocol.