PASS INFORMATION

Title	Identification of Risk Factors for Low Lymphocyte Count in Patients with Type 2 Diabetes: An Analysis of Data From the SAVOR Study
Version identifier of the final study report	DRAFT
Date of last version of the final study report	N/A
EU PAS register number	N/A
Active substance	Saxagliptin Dipeptidyl peptidase-4 (DPP4) inhibitor, ATC code: A10BH03 Saxagliptin/metformin HCl (XR or IR) Saxagliptin/metformin HCl IR, ATC code: A10BD Saxagliptin/metformin HCl XR, ATC code: A10BD
Medicinal product	ONGLYZA ®(saxagliptin) film-coated tablets 2.5 mg ONGLYZA (saxagliptin) film-coated tablets 5 mg KOMBOGLYZE (saxagliptin/metformin HCl immediate release) tablets 2.5 mg/850 mg KOMBOGLYZE (saxagliptin/metformin HCl immediate release) tablets 2.5 mg/1000 mg KOMBIGLYZE XR (saxagliptin/metformin HCl extended release) tablets 5 mg/500 mg KOMBIGLYZE XR (saxagliptin/metformin HCl extended release) tablets 5 mg/1000 mg KOMBIGLYZE XR (saxagliptin/metformin HCl extended release) tablets 2.5 mg/1000 mg
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Marketing authorisation holder(s)	AstraZeneca AB
Joint PASS	Yes

Research question and objectives	Research question: What are the risk factors associated with low lymphocyte count in patients with T2DM? Primary Objective: To identify risk factors for lymphopenia, defined as an absolute lymphocyte count <500 cells/µl or investigator reported lymphopenia, among patients with T2DM. Secondary Objective 1: To identify risk factors for low lymphocyte count, defined as an absolute lymphocyte count <750 cells/µl or investigator reported lymphopenia, among patients with T2DM. Secondary Objective 2: To identify risk factors for decreasing lymphocyte counts (>=30% decrease from baseline) in patients with T2DM. Secondary Objective 3: To evaluate whether risk factors for low lymphocyte counts or decreasing lymphocyte count differ between saxagliptin users and non-users. Exploratory Objective: To determine whether time-varying covariates are associated with changes in lymphocyte counts, and whether those associations differ by exposure.
Country(-ies) of study	Study is a secondary analysis of a previous study, SAVOR. SAVOR was conducted in Canada, United States, France, Germany, Italy, Netherlands, Spain, Sweden, United Kingdom, Czech Republic, Hungary, Israel, Poland, Russia, Argentina, Brazil, Chile, Mexico, Peru, People's Republic of China, Hong Kong, India, Taiwan, Thailand, Australia, and South Africa.
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TABLE OF CONTENTS

1 ABSTRACT

Title

Identification of Risk Factors for Low Lymphocyte Count in Patients with Type 2 Diabetes: An Analysis of Data from the SAVOR Study

Keywords

Saxagliptin, lymphopenia, lymphocyte count, diabetes mellitus

Rationale and background

A dose-related decrease in lymphocyte count was observed among saxagliptin users in the pre-authorization saxagliptin clinical program. This post-authorization safety study was conducted to assess risk factors associated with low lymphocyte count in patients with Type 2 Diabetes Mellitus (T2DM).

Research question and objectives

Research Question: What are the risk factors associated with low lymphocyte count in patients with T2DM?

Primary Objective: To identify risk factors for lymphopenia, defined as an absolute lymphocyte count <500 cells/µl or investigator reported lymphopenia, among patients with T2DM.

Secondary Objective 1: To identify risk factors for low lymphocyte count, defined as an absolute lymphocyte count <750 cells/µl or investigator reported lymphopenia, among patients with T2DM.

Secondary Objective 2: To identify risk factors for decreasing lymphocyte counts (≥30% decrease from baseline) in patients with T2DM.

Secondary Objective 3: To evaluate whether risk factors for low lymphocyte counts or decreasing lymphocyte count differ between saxagliptin users and non-users.

Exploratory Objective: To determine whether time-varying covariates are associated with changes in lymphocyte counts, and whether those associations differ by exposure.

Study design

This was a secondary analysis using data collected as part of the Saxagliptin Assessment of Vascular Outcomes Recorded in Patients with Diabetes Mellitus (SAVOR) study. SAVOR was a multicenter, randomized, double blind, placebo-controlled Phase IV study designed to evaluate whether treatment with saxagliptin can reduce the composite endpoint of CV death, non-fatal MI or non-fatal ischemic stroke in patients with T2DM, and to definitively exclude unacceptable CV toxicity. Patients were randomized to receive either saxagliptin (2.5 mg or 5 mg) or placebo, on top of whatever baseline treatment for diabetes the patient was already receiving (which could include other anti-diabetic medications, such as insulin, but which excluded incretin-based therapies.)

Setting

Approximately 16,500 patients with documented T2DM and either a history of a cardiovascular event or multiple risk factors for vascular disease were enrolled from sites throughout the world in the SAVOR study. Approximately 30% were from North America, 30% from Europe and the remaining 40% from rest of the world.

Subjects and study size, including dropouts

All randomized patients in SAVOR were eligible for this study. A total of 18,206 patients were enrolled in SAVOR of which 16,492 were randomized (8280 in the saxagliptin group and 8212 in the placebo group). Of the 1714 patients not randomized, 1413 (82.4%) failed inclusion or exclusion criteria and 301 (17.6%) were not randomized for other reasons. Approximately 97% of patients completed the study (8078 in the saxagliptin group and 7998 in the placebo group); the remaining 416 patients withdrew consent and 28 were lost to follow-up.

Variables and data sources

The primary outcome of this study (CV181-102) was lymphopenia, defined as a follow-up lymphocyte count <500 cells/µl or investigator-reported lymphopenia). Secondary outcomes of this study were low lymphocyte count, defined as a follow-up lymphocyte count <750 cells/µl or investigator-reported lymphopenia, and decreasing lymphocyte count, defined as relative decrease of ≥30% from the baseline lymphocyte count. An exploratory outcome was to evaluate changes in individuals' absolute lymphocyte count from baseline to each assessment time point.

The following data were used for all patients at baseline:

- age,
- sex,
- hemoglobin A1c,
- duration of type 2 diabetes,
- prescription drug use,
- cardiovascular and other medical comorbidities,
- physical measures (e.g., blood pressure, BMI), and
- laboratory data (including lymphocyte count).

For the exploratory objective, data collected at each follow-up were also assessed for association with concurrent lymphocyte count changes from baseline (e.g., HbA1c, blood pressure, newly reported co-morbidities, changes in concurrent medication use, etc.). All follow-up variables in SAVOR were routinely collected every 6 to 12 months.

Results

<u>Primary Objective</u>

Based on 71 (0.50%) patients with events, baseline variables found to be associated with lymphopenia (i.e. lymphocyte count <500 cell/µl or investigator reported lymphopenia) in the

multivariate Cox proportional hazards regression model included Region [Latin America (HR=4.17; 95% CI: 2.02, 8.60)], CABG involving >1 artery (HR=1.96; 95% CI: 1.10, 3.48), symptoms of claudication and amputation (HR=3.44; 95% CI: 1.01, 11.66); congestive heart failure (HR=2.14; 95% CI: 1.21, 3.81), atrial fibrillation/flutter (HR=1.96; 95% CI: 1.03, 3.73) and malignancy history (HR=2.16; 95% CI: 1.07, 4.35).

Secondary Objectives

Based on 236 (2%) patients with events, baseline variables associated with low lymphocyte count during follow-up (i.e. lymphocyte count <750 cell/µl or investigator reported lymphopenia) included baseline sulfonylurea (HR=1.34; 95% CI: 1.03, 1.75), increasing age (≥65 years (HR=1.63; 95% CI: 1.18, 2.23), region (Latin America HR=1.87; 95% CI: 1.97, 2.93) >2 coronary arteries with >50% stenosis (HR=1.45; 95% CI: 1.05, 2.00), atrial fibrillation/flutter (HR=2.02; 95% CI: 1.41, 2.90), and saxagliptin compared to placebo (HR=1.51; 1.16, 1.97).

Also based on 1930 (12%) patients with events in the multivariate Cox proportional hazards regression analysis, baseline variables found to be associated with a decreasing lymphocyte count of 30% or more during follow-up included residing in Latin America (HR=1.60; 95% CI: 1.27-2.02), CABG involving >1 artery (HR=1.14; 95% CI: 1.01, 1.28), atrial fibrillation/flutter (HR=1.35; 95% CI: 1.13, 1.60), being a current smoker (HR=1.15; 95% CI: 1.00, 1.31) and saxagliptin compared to placebo (HR=1.49; 95% CI: 1.36, 1.64).

There was no evidence of interaction to suggest that there were differences in any of the risk factors between saxagliptin users or placebo users.

Exploratory Objective

Analyses for the exploratory objective were not conducted because of the lack of definitive start and end dates of potential time-varying covariates.

Discussion

This study was conducted to identify risk factors for lymphopenia, a low lymphocyte count, and decreasing lymphocyte count in patients with T2DM. The study confirmed certain previously observed associations and identified additional potential risk factors that have not previously been reported including tobacco smoking and treatment with sulfonylurea.

The current study findings for saxagliptin are similar or consistent with findings from the pre-authorization saxagliptin clinical program and from the SAVOR study report, although events were defined slightly differently in each study.

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2 LIST OF ABBREVIATIONS

Term	Definition
ABI	Ankle Brachial Index
AEs	Adverse Events
AZ	AstraZeneca
CABG	Coronary Artery Bypass Graft Surgery
CVD	Cardiovascular disease
DPP4	Dipeptidyl peptidase IV
eGFR	Estimated glomerular filtration rate
FDA	Food and Drug Administration
FPG	Fasting plasma glucose
ITT	Intention-to-treat
HR	Hazard ratio
MDRD	Modification of Diet in Renal Disease
MRF	Multiple risk factors
NHANES	National Health and Nutrition Examination Survey
OAD	Oral Antidiabetic Drug
SAVOR	Saxagliptin Assessment of Vascular Outcomes Recorded in Patients with Diabetes Mellitus
SmPC	Summary of product characteristics
T2DM	Type 2 Diabetes Mellitus
TBI	Toe Brachial Index
UK	United Kingdom
US	United States

3 INVESTIGATORS

All sites that participated in the SAVOR clinical trial were included in CV181-102.

The SAVOR study was conducted at 790 study sites worldwide with a targeted distribution of patients as follows: at least 30% in North America (Canada [50 sites] and United States (US) [271 sites]), approximately 30% in Europe (France [10 sites], Germany [23 sites], Italy [22 sites], Netherlands [27 sites], Spain [15 sites], Sweden [17 sites], United Kingdom (UK) [18 sites],

Czech Republic [30 sites], Hungary [27 sites], Israel [27 sites], Poland [26 sites], and Russia [33 sites]) and the remainder in Mexico (23 sites), South America (Argentina [21 sites], Brazil [20 sites], Chile [13 sites], and Peru [18 sites]), Australia (12 sites), Asia (People's Republic of China [13 sites], Hong Kong [9 sites], India [22 sites], Taiwan [11 sites], and Thailand [11 sites]), and South Africa (21 sites).

There was no principal investigator. The international coordinating investigators for SAVOR were:

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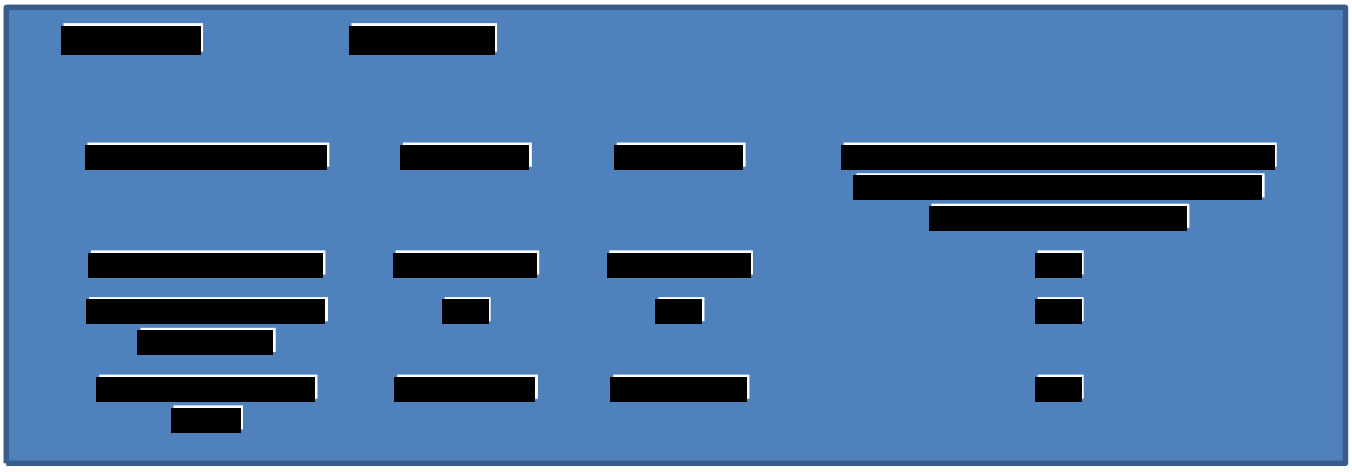
4 OTHER RESPONSIBLE PARTIES

The responsible party for this study includes the sponsor, AstraZeneca.

5 MILESTONES



6 RATIONALE AND BACKGROUND

Saxagliptin, a potent dipeptidyl peptidase-4 (DPP-4) inhibitor, is an oral anti-diabetic drug (OAD) to be used in combination with diet and exercise to control hyperglycemia in adults with type 2 diabetes mellitus (T2DM). It is prescribed as both monotherapy and combination therapy in the

United States (US), and as combination therapy in the European Union.[1,2] This OAD was approved for use by the US Food and Drug Administration (FDA) in July 2009 and European Medicines Agency in October 2009.

During the clinical program, a dose-related decrease in lymphocyte count was observed among saxagliptin users. The median change in lymphocyte count from baseline was a decrease of 110 cells/µL for the saxagliptin 5 mg group compared to 20 cells/µL for the placebo group. In the overall saxagliptin group, 9.4% of users experienced a 30% decrease in their lymphocyte count over a 12-month period, and 4% experienced "lymphopenia," which was defined as a lymphocyte count <750 cells/µl or investigator reported lymphopenia, during the short-term and long-term follow-up. There did not appear to be a higher frequency of infection-related adverse events or a different pattern in the type of infections reported among saxagliptin-treated patients compared to the active comparison group in the clinical trials.

No published studies were identified for known risk factors of lymphopenia or for the incidence or prevalence of low lymphocyte count among patients with T2DM. To improve understanding of the relationship between lymphocyte levels and T2DM, epidemiologists from BMS and AZ used both National Health and Nutrition Examination Survey (NHANES) 1999-2006 data and the Ingenix LabRx database to evaluate lymphocyte levels among patients with and without T2DM.

An age-related decline in median lymphocyte count was observed in NHANES 1999-2006 data. The age group of 18-35 year olds had a median lymphocyte count of 2100 cells/µl, compared to those 75 years and older, who had a median lymphocyte count of 1700 cells/µl. In addition, women tended to have a slightly higher median lymphocyte count than men across all age groups (50 cells/µl difference overall and the highest difference was 300 cells/µl in the 65-74 year age group between men and women).

Among adults with T2DM in NHANES 1999-2006 (defined as HbA1c>=6.5% or self-report of ever having been told by a health professional to have diabetes (other than gestational diabetes)), overall median lymphocyte count was 2100 cells/µl, and the median lymphocyte count was higher than in the general population in all age strata. The age-related decline in lymphocyte count, and the persistently higher count among women than men across age groups, was also observed among patients with diabetes in NHANES, as well as in the Ingenix database. In the Ingenix database, the average lymphocyte count across all studied patients was 1,966 cells/µl (SD = 955) for males and 2,145 cells/µl (SD = 954) for females, and there was an inverse relationship between age and lymphocyte count.

Approximately 0.05% of the adult general population of NHANES 1999-2006 had a lymphocyte count <500 cells/µl, while 1.7% had a lymphocyte count <1000 cells/µl, and 90.1% had a lymphocyte count within the "normal" range of 1000 to 3500 cells/µl, inclusive. The proportion with low lymphocyte counts increased with age, such that 0.5% of those in the 18-34 year old age group had a lymphocyte count <1000 cells/µl, while 5.7% of those age 75 or older did. Interestingly, although median lymphocyte counts were higher among patients with T2DM, higher percentages of patients reached very low levels of lymphocyte counts (0.12% had lymphocyte

counts <500 cells/µl and 3.3% had a lymphocyte count <1000 cells/µl), and a lower percentage had lymphocyte counts in the normal range (85.7%), than among all adults.

In the Ingenix database, analyses among a sample of patients with T2DM demonstrated that the prevalence of low lymphocyte count (defined in this study as a lymphocyte count <1000 cells/µl) was 2.6% in women and 4.1% in men. Prevalence of low lymphocyte count increased with age (1.2% among 18- to 34-year-olds and 6.2% among those age 65 or older). An important limitation of this study is that lymphocyte counts available in this administrative claims and laboratory database were not systematically captured for all patients. Those who were tested would be more likely than the average patient to have a clinical finding that would necessitate testing; therefore, the prevalence suggested in this study must be considered with caution due to potential surveillance bias.

This Post-Authorization Safety Study is being conducted as part of the BMS/AZ Saxagliptin Risk Management Plan to assess the risk factors associated with low lymphocyte count in patients with T2DM.

7 RESEARCH QUESTION AND OBJECTIVES

The research question was as follows: what are the risk factors associated with low lymphocyte count in patients with T2DM?

The primary objective was to identify risk factors for lymphopenia, defined as an absolute lymphocyte count <500 cells/µl or investigator reported lymphopenia, among patients with T2DM.

The secondary objectives were:

1. To identify risk factors for low lymphocyte count, defined as an absolute lymphocyte count <750 cells/µl or investigator reported lymphopenia, among patients with T2DM;
2. To identify risk factors for decreasing lymphocyte counts (≥30% decrease from baseline) in patients with T2DM; and
3. To evaluate whether risk factors for low lymphocyte counts or decreasing lymphocyte count differ between saxagliptin users and non-users.

The exploratory objective was to determine whether time-varying covariates are associated with changes in lymphocyte counts, and whether those associations differ by exposure.

8 AMENDMENTS AND UPDATES

None.

9 RESEARCH METHODS

9.1 Study Design

This study was a secondary analysis of data collected as part of the Saxagliptin Assessment of Vascular Outcomes Recorded in Patients with Diabetes Mellitus (SAVOR) study. The primary outcome of this present study was lymphopenia, defined as a follow-up lymphocyte count <500 cells/µl or investigator-reported lymphopenia. Secondary outcomes of this study were low lymphocyte count, defined as a follow-up lymphocyte count <750 cells/µl or investigator-reported lymphopenia and decreasing lymphocyte count, defined as relative decrease of ≥30% from the baseline lymphocyte count. An exploratory outcome was to evaluate changes in individuals' absolute lymphocyte count from baseline to each assessment time point. The study design was consistent with Section 5.1 of Protocol CV181-102.

A prospective study with systematic capture of laboratory data is ideal for studying lymphocyte counts. Lymphocyte counts are not routinely captured as part of normal clinical practice for patients with T2DM and are often done when the clinician has a suspicion for clinical abnormality within that patient, such as a suspect infection. A study that relied on routine clinical practice would be limited to evaluating lymphocyte counts only among those patients tested and testing would often be conducted only when a patient was most likely to have an abnormal lymphocyte count. Evaluation of risk factors in that setting might confuse risk factors for low lymphocyte count with risk factors for testing for lymphocyte counts. As lymphocyte counts were collected annually per SAVOR study protocol, annual testing would not be related to clinical suspicion and, therefore, provided a more appropriate setting for risk factor analysis. However, investigators were also allowed to report a patient as having lymphopenia in addition to lab results. The investigator reports were not supported by lab results and may be due to clinical suspicion.

9.2 Setting

This study was a secondary analysis of data from the SAVOR study, a multicenter, randomized, double-blind, placebo-controlled Phase IV study. Approximately 16,500 patients with documented T2DM and either a history of a cardiovascular event or multiple risk factors for vascular disease were enrolled from sites throughout the world in the SAVOR study. Approximately 30% were from North America, 30% from Europe and the remaining 40% from rest of the world (Protocol CV181-102, Section 5.2). The SAVOR study were initiated from May 2010 (first patient, first visit) and ended by May 2013 (last patient, last visit).

9.3 Subjects

All patients randomized in SAVOR were eligible to be included in this study, consistent with Protocol CV181-102 (Sections 5.2.1, 5.2.2).

Key eligibility criteria for SAVOR included:

- Diagnosis with T2DM,
- Baseline HbA1c ≥6.5%, age 40 or older, and

- High risk for a cardiovascular event (either had established CV disease or multiple risk factors (MRF)).

Key exclusion criteria for SAVOR included:

- Current or previous (within 6 months) incretin-based therapy,
- Acute vascular event within 2 months of randomization,
- Chronic dialysis and/or renal transplant and/or serum creatinine > 6.0 mg/dL, pregnancy,
- HIV/AIDS,
- Severe autoimmune disease,
- Current chronic steroid use, and
- A variety of other exclusions based on laboratory or physical measurements.[3]

A total of 18,206 patients were enrolled in SAVOR of which 16,492 were randomized (8280 in the saxagliptin group and 8212 in the placebo group. Of the 1714 patients not randomized, 1413 (82.4%) failed inclusion or exclusion criteria and 301 (17.6%) were not randomized for other reasons. Approximately 97% of patients completed the study (8078 in the saxagliptin group and 7998 in the placebo group); the remaining 4% withdrew consent or were loss to follow-up.

Of all randomized patients, 31.9% and 42.2% were enrolled at sites in North America and Europe, respectively, with the remaining patients enrolled at sites in Latin America and Asia/Pacific regions. Patients with multiple risk factors for vascular disease comprised 21.4% of the total population with the remaining 78.6% of patients had documented cardiovascular disease. Most patients had normal renal function or mild renal impairment at baseline (84.4%); 13.6% and 2% had moderate and severe renal impairment, respectively, at baseline.

9.4 Variables

Demographic and clinical data on patients included in this study were obtained from the SAVOR study database, consistent with Protocol CV181-102 (Section 6.2).

9.4.1 Independent Variables

Treatment is defined as exposure to saxagliptin or placebo on the date of randomization to the SAVOR study. Patients were randomized to receive either saxagliptin (2.5 mg for patients with moderate to severe renal function or 5 mg in patients with normal or mild renal function) or placebo, on top of whatever baseline treatment for diabetes the patient was already receiving (which included other anti-diabetic medications, including insulin but excluding incretin-based therapies). In primary analyses, an "as-treated" approach was used and saxagliptin or placebo exposure ended 30 days after the patient stopped taking the assigned treatment. In sensitivity analyses, exposure to saxagliptin or placebo was considered to have ended at the end of the study period or discontinuation from the SAVOR study, using an intention-to-treat approach.

Other variables collected at baseline included: age, sex, hemoglobin A1c, blood pressure, duration of type 2 diabetes, prescription drug use, cardiovascular and other medical co-morbidities, physical measures, and laboratory data. See Table 9.4.1-1 for operational definitions of each.

Table 9.4.1-1: Evaluated risk factors for lymphocyte endpoints

Variable	Categories
Age	<65, ≥65, ≥75
Gender	Female, Male
Race	White, Black or African American, Asian, American Indian or Alaska, Native, Native Hawaiian or Other, Pacific Islander, Multiracial, Unknown
BMI	< 30 kg/m2 and ≥30 kg/m2
Duration of T2DM	<5 years (1826 days) , ≥5 - <10 years,(1826-3653 days) , ≥10 - <15 years,(3653 – 5479 days), ≥15 - <20 years,(5479 – 7306 days) , ≥20 years (7306,days)
HbA1c	<6.5%, ≥6.5% - <7%, ≥7% - <8%,,≥8% - <9%, ≥9%
Baseline FPG	<126 mg/dL, ≥126 – < 150 mg/dL,,≥150 - <220 mg/dL, ≥ 220 - <300,mg/dL, ≥300 mg/dL
Sitting systolic blood pressure	≤130 mm/HgL, >130mm/HgL
Sitting diastolic blood pressure	≤80 mm/HgL, >80mm/HgL
eGFR[1]	>50 mL/min, ≤30-≤50 mL/min, <30 mL/min
Region	North America, Europe, Latin,America, Asia/Pacific
Baseline diabetic medication use	Insulin, Sulfonylurea, Thiazolidinediones, Metformin, Any other DM medications, no other DM medications
Baseline CV medication use	Aspirin, Statin, ACE inhibitor, Angiotensin receptor blocker, any other CV medications, no other CV, medications
Medical history baseline comorbidities	Myocardial Infarction Ischemic Stroke PCI of >1 artery History of stent CABG of >1 artery >/=2 Coronary arteries with >50% stenosis Peripheral Artery Disease Current symptoms of claudication ABI or TBI < 0.90 Prior Peripheral Revascularization Amputation due to arterial disease Angina Pectoris Congestive heart failure Atrial fibrillation / flutter Renal function - estimated GFR (MDRD) Creatinine

Table 9.4.1-1: Evaluated risk factors for lymphocyte endpoints

Variable	Categories
	Hypertension
	Dyslipidemia
	Current smoker
	HbA1c >6.5% in previous 6 months
	Date of DM Diagnosis (year)
	Retinopathy
	Nephropathy
	Amputation due to limb ischemia or diabetic complication
	History of malignancy
	History of Thrombocytopenia (Platelets <100,000/mm^3)
	Lymphocyte count (continuous)

[1] GFR estimated using Modification of Diet in Renal Disease (MDRD) equation.

9.4.2 Dependent Variables

The primary outcome of this study was lymphopenia, defined as a follow-up lymphocyte count <500 cells/µl or investigator-reported lymphopenia.

To investigate risk factors for less severe decreases in lymphocyte count, secondary outcomes of this study were low lymphocyte count, defined as a follow-up lymphocyte count <750 cells/µl or investigator-reported lymphopenia and decreasing lymphocyte count, defined as relative decrease of ≥30% from the baseline lymphocyte count.

An exploratory outcome was to evaluate changes in individuals' absolute lymphocyte count from baseline to each assessment. Across outcomes, follow-up occurred until the first low lymphocyte count (event) or patient discontinued from study or study drug.

9.5 Data Sources and Measurement

This study used data from the Saxagliptin Assessment of Vascular Outcomes Recorded in Patients with Diabetes Mellitus (SAVOR) study. SAVOR was a multicentre randomized double-blind placebo-controlled Phase IV study designed to determine whether treatment with saxagliptin compared with placebo will result in a reduction in the composite endpoint of cardiovascular death non-fatal myocardial infarction or non-fatal ischaemic stroke in patients with T2DM. Patients were randomized to receive either saxagliptin (2.5 mg or 5 mg) or placebo on top of whatever baseline treatment for diabetes the patient is already receiving (which can include other anti-diabetic medications including insulin but excluding incretin-based therapies).

Baseline data collected for all patients included patient demographics prescription drug use cardiovascular and other medical co-morbidities physical measures (e.g. blood pressure height weight) and laboratory data [including diabetes specific measures such as HbA1c and Fasting Plasma Glucose (FPG)] measured by a central laboratory. Lymphocyte counts were included in the baseline laboratory data collected. Patients were contacted every three months between clinic

visits via telephone and were asked to provide information about events concomitant medications and adherence to treatment. At months 6, 18, 30, 42, and 54 patients engaged in a clinic visit where the same information collected via telephone interview was recorded and a few additional non-lymphocyte related data elements were collected. Visits at months 12, 24, 36, 48, and 60 (study closeout) were the most comprehensive; the same data above were collected in addition to non-fasting blood measurements (including lymphocyte counts), limited urinalysis, physical exam and patient-reported outcomes. Lymphocyte counts were systematically measured on an annual basis per SAVOR study protocol. Lymphocyte counts obtained by local laboratories in addition to annual systematic assessments were also included in the SAVOR study as well as this current analysis. Additionally, patients or providers reports of adverse events were recorded, including lymphopenia or decreased lymphocyte count, during one of the telephone or clinic visits, or spontaneously at any time during the course of the study.

9.6 Bias

The SAVOR study is a randomized double-blind study with systematic capture of laboratory and other measures. Comparability across treatment groups on baseline measures were evaluated as part of the primary SAVOR study analyses and those results showed comparability across treatment groups. Despite the benefits of randomization for increasing comparability across treatment groups, randomization does not give comparability across other potential risk factors for lymphopenia. Thus, unmeasured confounding may still exist between other variables and lymphopenia. This study will also be limited in its ability to assess changes in risk factors over time (e.g. presence of an infection) which could also influence primary study results.

In addition, there is variability in the assessment of the laboratory data. Thus, for the change in lymphocyte analysis, only patients with both baseline and post-baseline lymphocyte measurements are included. Although lymphocyte counts were systematically measured on an annual basis per the SAVOR protocol, additional lymphocyte counts obtained by local laboratories (i.e. investigator ordered) were also included in the SAVOR study as well as this current analysis. This additional monitoring of lymphocyte counts during follow up could be differential with respect to potential risk factors, and result in bias.

Lastly, all investigator-reported adverse events of low lymphocyte count were considered an event even if there was no supporting lab data. Such reports could be subject to ascertainment bias if the treating clinician diagnosed a patient with lymphopenia without supporting lab data and this occurred disproportionately across a potential risk factor.

9.7 Study Size

The study size is consistent with Protocol CV181-102 Section 6.4. Sample size was based upon power calculation for the primary endpoint of the SAVOR study. Although the sample size was not based upon statistical considerations for the lymphocyte study, this sample size allowed for a group of patients with clinically meaningful reduction in lymphocytes to be investigated for risk factors for development of low lymphocyte count.

A total of 18206 patients were enrolled in the SAVOR study of which 16492 were randomized (8280 in the saxagliptin group and 8212 in the placebo group). Of the 1714 patients not

randomized, 1413 (82.4%) failed inclusion or exclusion criteria and 301 (17.6%) were not randomized for other reasons. Approximately 97% of patients completed the study; the remaining 4% withdrew consent or were loss to follow-up. There were a total of 83 cases of lymphopenia defined as <500 cells/µl or investigator reported, and 296 cases of low lymphocyte count, defined as lymphocyte count <750 cells/µl or investigator reported.

9.8 Data Transformation

The analytic datasets from SAVOR were obtained for analysis. Categorical and continuous data were handled as outlined in Statistical Methods (Section 9.9).

9.9 Statistical Methods

9.9.1 Main Summary Measures

Continuous variables were summarized by means and standard deviations (SDs) or medians and interquartile ranges (IQRs) where appropriate; and categorical variables were summarized by frequencies and proportions.

Evaluations of risk factors associated with the outcome are reported using hazard ratios (HR) with 95% confidence intervals (CIs) and p-values.

9.9.2 Main Statistical Methods

The Primary Objective identified risk factors for lymphopenia, across the study population, using data collected at baseline. Secondary Objective 1 identified risk factors for low lymphocyte count, across the study population, using data collected at baseline. Secondary Objective 2 identified risk factors for a decrease in lymphocyte count ≥ 30% from baseline across the study population. Any risk factors identified in the Primary Objective or Secondary Objectives 1 or 2 were further assessed in Secondary Objective 3, to determine whether or not there was effect modification with the treatment exposure group (saxagliptin or placebo). The Exploratory Objective assessed whether or not risk factors that changed over time were associated with changes in lymphocyte count.

The current study used an ‘as-treated’ approach, wherein follow-up was censored 30 days after the patient stopped taking the assigned treatment.

9.9.2.1 Primary Objective

Unadjusted associations between potential risk factors and lymphopenia were assessed using Cox proportional hazards regression. Variables with $p<0.20$ were included in the multivariate Cox regression analysis to determine whether or not variables associated with an increased risk of low lymphocyte count remained statistically significant following adjustment for other potential risk factors. Biologically important variables and known confounders (i.e., baseline lymphocyte count, treatment assignment, age, sex, renal impairment status and primary/secondary CV prevention) were kept in the model regardless of their significance level.

Only those patients with baseline lymphocyte count ≥500 cells/µl and no investigator reported lymphopenia at baseline were included in this analysis.

9.9.2.2 Secondary Objectives

Analyses for the secondary objective 1 were the same as the primary objective. The only exception is that analyses evaluated potential risk factors for low lymphocyte count.

Only those patients with baseline lymphocyte count ≥750 cells/µl and no investigator reported lymphopenia were included in this analysis.

For secondary objective 2, relative differences in lymphocyte count from baseline were calculated and categorized into a decrease of >=30%. A follow up decreasing lymphocyte count ≥30% was defined as an absolute decrease in lymphocyte count from the baseline until the first decrease ≥30% during the follow up. The post-baseline lymphocyte count could be based on annual assessments (per SAVOR protocol) or on lymphocyte counts obtained by local laboratories (investigator ordered). Cox proportional hazard models were conducted to assess associations between potential risk factors and decrease in lymphocyte count. Variables with $p<0.20$ were included in the multivariate model to determine risk factors associated with a decrease in lymphocyte count. Biologically important variables and known confounders (such as baseline lymphocyte count, treatment assignment, age, sex, renal impairment status, and primary/secondary CV prevention) were kept in the model regardless of their significance level. Potential risk factors included demographic, clinical, physical, and laboratory measures assessed at baseline, as in the primary objective. Only those patients with baseline and post-baseline lymphocyte count assessments were included in this analysis.

For secondary analysis 3, Cox proportional hazard models with interaction variables between the identified risk factor (from Primary Objective or Secondary Objectives 1 or 2) and treatment risk group (saxagliptin vs. placebo) were used to determine whether or not there was a statistically significant effect modification by exposure. That is, whether the risk factor has a different impact on reaching lymphocyte cut-offs for those exposed to saxagliptin compared to those exposed to placebo

9.9.2.3 Exploratory Objectives

Exploratory analysis using mixed models for repeated measures to assess whether changes in variables over time were associated with changes in lymphocyte count were not conducted for the following reasons:

1. Start dates and end dates were not collected or were missing for time varying covariates (i.e. cardiovascular medications, diabetes medications); and
2. Very few patients with lymphopenia also reported other AEs together with reported lymphopenia at the same time during the study period; two patients with lymphopenia also experienced other AEs at the same time

Due to the lack of definitive start and end dates, the analysis was unable to determine whether the change in medication occurred before or after the outcome. Moreover, the low number of patients who experienced both lymphopenia and a clinical event limited the ability to conduct meaningful analyses.

9.9.3 Missing Values

Missing values were not imputed. In SAVOR, laboratory values from the closest unscheduled visit were used when scheduled laboratory visits were missing.

9.9.4 Sensitivity Analyses (Intention-to-treat analyses)

All analyses described in Sections 9.9.2 above were duplicated; however, exposure to saxagliptin or placebo was considered to have ended at the end of the study period or discontinuation from the SAVOR study, using an intention-to-treat approach.

9.9.5 Amendments to the Statistical Analysis Plan

N/A

9.10 Quality Control

This was a secondary analysis of SAVOR data; all data management activities were performed as part of SAVOR. Data management for SAVOR was performed by AstraZeneca Data Management Centre staff.

The data collected through third-party sources were obtained and reconciled against study data.

AEs were classified according to the terminology of the latest version of the MedDRA. Medications were classified according to the AstraZeneca Drug Dictionary. All coding was performed by AstraZeneca Data Management Centre staff, with the exception of SAE coding, which was completed by BMS.

Data queries were raised for inconsistent, impossible or missing data. The data were validated as defined in the Data Management Plan for SAVOR. Quality control procedures were applied to each stage of data handling to ensure that all data are reliable and have been processed correctly.

10 RESULTS

10.1 Participants

For this analysis, the "as treated" population included 16,413 patients. Only patients without lymphopenia at baseline were included in the analyses for objectives 1 and 2; for objective 3, only patients with baseline and post-baseline lymphocyte count assessments were included. The number of patients included in the analysis for each of the objectives is listed below:

- The primary objective included 15717 patients with ≥500 cell/µl and no history of lymphopenia at baseline.
- The analysis for secondary objective 1 included 15621 patients with ≥750 cell/µl and no history of lymphopenia at baseline.
- The analysis for secondary objective 2 included 14898 patients with both baseline and post-baseline lymphocyte count assessments.

10.2 Descriptive Data

Table 10.2-1 describes the prevalence of lymphopenia, low lymphocyte count, and decreasing lymphocyte count by patient subgroup. Note that this table provides frequencies of the total "as treated" population (n=16413), prior to the removal of patients with baseline lymphopenia.

Table 10.2-1: Description of the prevalence of outcome definitions by patient subgroup, As Treated population (N=16,413)

		Outcome					
		Lymphopenia		Low Lymphocyte Count		Decreasing Lymphocyte Count	
	N	n	%	N	%	n	%
Treatment							
Saxagliptin	8240	47	0.57	182	2.21	1158	14.05
Placebo	8173	36	0.44	114	1.39	772	9.45
CVD Risk Group							
CVD Risk	12892	62	0.48	233	1.81	1471	11.41
MRF Risk	3521	21	0.60	63	1.79	459	13.04
Age							
<65 years	7898	30	0.38	72	0.91	890	11.27
≥65 years	2313	18	0.78	90	3.89	290	12.54
Sex							
Female	10984	57	0.52	216	1.97	1278	11.64
Male	5429	26	0.48	80	1.47	652	12.01
Race							
White	12335	62	0.50	242	1.96	1422	11.53
Non-White	4078	21	0.51	54	1.32	508	12.46
BMI							
BMI <=30	7609	36	0.47	139	1.83	941	12.37
BMI >30	8777	47	0.54	157	1.79	987	11.25
BMI Unknown	27	0	0	0	0	2	7.41
Duration of Diabetes:							
<5 years	3900	13	0.33	49	1.26	449	11.51
>=5 to <10 years	3906	21	0.54	65	1.66	448	11.47
>=10 to <15 years	3476	24	0.69	67	1.93	425	12.23
>=15 to <20 years	2107	8	0.38	38	1.80	258	12.24
>= 20 years	3012	17	0.56	77	2.56	349	11.59
Unknown	12	0	0	0	0	1	8.33

Table 10.2-1: Description of the prevalence of outcome definitions by patient subgroup, As Treated population (N=16,413)

		Outcome					
		Lymphopenia		Low Lymphocyte Count		Decreasing Lymphocyte Count	
	N	n	%	N	%	n	%
HbA1c Category:							
<6.5	1261	8	0.63	27	2.14	164	13.01
>=6.5 to <7	2846	15	0.53	66	2.32	349	12.26
>=7 to <8	5385	30	0.56	105	1.95	652	12.11
>=8 to <9	3124	3	0.10	43	1.38	358	11.46
>=9	3516	25	0.71	48	1.37	404	11.49
Not Reported	281	2	0.71	7	2.49	3	1.07
Region							
North America	5239	21	0.40	122	2.33	583	11.13
Latin America	2707	20	0.74	51	1.88	413	15.26
Asia/Pacific	1546	2	0.13	15	0.97	167	10.80
Europe	6921	40	0.58	108	1.56	767	11.08

Source: Appendix 1

10.3 Outcome Data

The primary outcome of this study was lymphopenia, defined as a follow-up lymphocyte count <500 cells/µl or investigator-reported lymphopenia. After accounting for the inclusion and exclusion criteria for these analyses, 71 patients were included in the primary analysis.

The secondary outcomes of this study were low lymphocyte count, defined as a follow-up lymphocyte count <750 cells/µl or investigator-reported lymphopenia and decreasing lymphocyte count, defined as relative decrease of ≥30% from the baseline lymphocyte count. After accounting for the inclusion and exclusion criteria for these analyses, 236 and 1930 patients were included in the secondary objective 1 and 2 analyses, respectively.

Across outcomes, follow-up occurred until the first event or patient discontinuation from study or study drug.

10.4 Main Results

All results are presented by outcome, which included variables with $p<0.20$ in the univariate analyses.

Results from the sensitivity analysis (Section 9.9.4 Sensitivity Analyses) are included in Appendix 3 and are described below only in instances where the interpretation of the findings were different from the primary analysis.

10.4.1 Primary Objective

10.4.1.1 Primary Objective - Univariate Analysis

Univariate Cox proportional hazards regression was used to identify baseline characteristics that were associated with a diagnosis of lymphopenia, defined as a lymphocyte count during follow-up of <500 cell/µl or investigator reported lymphopenia. Variables with a statistically significant (p-value <0.20) association with lymphopenia are included in Table 10.4.1.1-1. All other univariate results or those variables with a p-value >0.20 are in Appendix 2.1.1.

Three kidney disease markers (GFR, nephropathy, and microalbuminuria) and three cardiovascular disease markers (CABG involving > 1 artery, congestive heart failure and atrial fibrillation/flutter) as well as region where the patient lives, current symptoms of claudication and amputation due to arterial disease, and a history of malignancy were associated with lymphopenia in univariate analyses. (Table 10.4.1.1-1)

Table 10.4.1.1-1: Univariate Cox Regression of lymphopenia <500 cell/µl or Investigator Reported Events and No History of lymphopenia

Variable	N	Hazard Ratio	95% CI	p-Value
GFR Category:				0.02
>50	59	*		
>=30 to <=50	7	0.83	0.38, 1.81	0.63
<30	5	3.67	1.47, 9.15	0.01
Region:				0.05
North America	18	*		
Latin America	19	2.00	1.05, 3.83	0.04
Asia/Pacific	1	0.24	0.03, 1.80	0.17
Europe	33	1.18	0.66, 2.10	0.57
CABG >1 Artery				
Yes	22	1.80	1.80, 2.98	0.02
No	49	*		
Current Symptoms of Claudication and Amputation due to Arterial Disease				
Yes	3	3.66	1.15, 11.64	0.03
No	68	*		
Congestive Heart Failure:				
Yes	19	2.95	1.74, 5.00	<0.0001
No	52	*		
Atrial Fibrillation/flutter:				
Yes	14	3.43	1.91, 6.16	<0.0001

Table 10.4.1.1-1: Univariate Cox Regression of lymphopenia <500 cell/µl or Investigator Reported Events and No History of lymphopenia

Variable	N	Hazard Ratio	95% CI	p-Value
No	57	*		
Nephropathy:				
Yes	22	2.14	1.29-3.54	0.003
No	49	*		
Microalbuminuria:				
Yes	16	1.82	1.04-3.17	0.0352
No	55	*		
Malignancy History				
Yes	10	2.68	1.37, 5.24	0.004
No	61	*		

Source: Appendix 2.1.1
^Events occurring more than 30 days after last dose are censored. Table only includes variables with p-value < 0.20.
*Reference group

10.4.1.2 Primary Objective - Multivariate Analysis

Based on 71 patients with events , variables associated with lymphocyte count <500 cells/µl or investigator reported lymphopenia included: residing in Latin America (HR=4.17; 95% CI=2.02, 8.60), current symptoms of claudication and amputation due to arterial disease (HR=3.44; 95% CI: 1.01, 11.66), CABG >1 Artery (HR=1.96; 95% CI: 1.10, 3.48), congestive heart failure (HR=2.14; 95% CI: 1.21, 3.81), atrial fibrillation/flutter (HR=1.96, 95% CI: 1.03, 3.73) and malignancy history (HR=2.16; 95% CI: 1.07, 4.35).

Table 10.4.1.2-1: Multivariate Cox Regression of Lymphocytopenia <500 cell/µl or Investigator Reported Events and No History of Lymphocytopenia^

Variable	N	Odd Ratio	95% CI	p-Value
GFR Category:				0.12
>50	59	*		
>=30 to <=50	7	0.44	0.19, 1.00	
<30	5	1.17	0.41, 3.31	
Region:				0.001
North America	18	*		
Latin America	19	4.17	2.02, 8.60	
Asia/Pacific	1	0.43	0.06, 3.23	
Europe	33	1.79	0.97, 3.28	
CABG >1 Artery				

Table 10.4.1.2-1: Multivariate Cox Regression of Lymphocytopenia <500 cell/µl or Investigator Reported Events and No History of Lymphocytopenia^

Variable	N	Odd Ratio	95% CI	p-Value
Yes	22	1.96	1.10, 3.48	0.02
No	49	*		
Current Symptoms of Claudication and Amputation due to Arterial Disease				
Yes	3	3.44	1.01, 11.66	0.047
No	68	*		
Congestive Heart Failure:				
Yes	19	2.14	1.21, 3.81	0.01
No	52	*		
Atrial Fibrillation/flutter:				
Yes	14	1.96	1.03, 3.73	0.04
No	57	*		
Nephropathy:				
Yes	22	1.72	0.64, 4.58	0.28
No	49	*		
Microalbuminuria:				
Yes	16	0.77	0.28, 2.11	0.61
No	55	*		
Malignancy History				
Yes	10	2.16	1.07, 4.35	0.03
No	61	*		

Source: Appendix 2.2.3
^Events occurring more than 30 days after last dose are censored. Based on variables with p-value <0.20 in the univariate regression models.
*Reference group

10.4.2 Secondary Objective 1

10.4.2.1 Secondary Objective 1 Univariate Analysis

Univariate Cox proportional hazards regression was used to identify baseline characteristics that were associated with a low lymphocyte count during follow-up of <750 cell/µl or investigator reported lymphopenia. Variables with a statistically significant (p-value<0.20) association with lymphopenia are included in Table 10.4.2.1-1. All other univariate results or those variables with a p-value>0.20 are in Appendix 2.2.1.

In univariate analyses, saxagliptin compared to placebo was associated (HR=1.57; 95% CI: 1.20, 2.03) with a low lymphocyte count. Demographic variables associated with a low lymphocyte

count in this analysis include increasing age, race, and female sex. Similar to the univariate models for lymphopenia (defined as <500 cell/µl), kidney disease markers and cardiovascular disease markers were associated with a low lymphocyte count as well as a history of malignancy. Duration of diabetes and diabetic complications, specifically amputations, were also associated with a low lymphocyte count (Table 10.4.2.1-1).

Table 10.4.2.1-1: Univariate Cox Regression of Low Lymphocyte Count <750 cell/µl or Investigator Reported Events

Variable	N	Hazard Ratio	95% CI	p-Value
Treatment:				
Saxagliptin	145	1.57	1.20, 2.03	0.0008
Placebo	91	*		
Age:				
<65 years	58	*		
≥65 years	178	2.91	2.16, 3.91	<0.0001
Sex:				
Female	175	*		
Male	61	0.67	0.50-0.90	0.007
Race:				
White	190	*		
Non-white	46	0.81	0.58, 1.11	0.19
Duration of Diabetes:				0.01
<5 years	41	*		
>=5 to <10 years	51	1.26	0.83, 1.90	0.30
>=10 to <15 years	54	1.52	1.01, 2.28	0.04
>=15 to <20 years	32	1.48	0.93, 2.35	0.10
>= 20 years	58	1.98	1.33, 2.96	0.0008
HbA1c Category:				0.14
<6.5	23	*		
>=6.5 to <7	57	1.09	0.67, 1.78	0.72
>=7 to <8	80	0.85	0.53, 1.35	0.48
>=8 to <9	36	0.69	0.41, 1.17	0.17
>=9	40	0.71	0.43, 1.19	0.20
Sitting Diastolic Blood Pressure				
≤ 80	158	*		
> 80	78	0.57	0.43, 0.75	<0.0001
GFR Category:				<0.0016

Table 10.4.2.1-1: Univariate Cox Regression of Low Lymphocyte Count <750 cell/µl or Investigator Reported Events

Variable	N	Hazard Ratio	95% CI	p-Value
>50	183	*		
≥30 - ≤50	45	1.72	1.24, 2.38	0.001
<30	8	1.94	0.95, 3.93	0.07
Region:				0.001
North America	95	*		
Latin America	43	0.83	0.58, 1.20	0.32
Asia/Pacific	10	0.41	0.21, 0.79	0.01
Europe	88	0.60	0.45, 0.80	0.001
Baseline Sulfonylurea				
Yes	106	1.21	0.94, 1.57	0.14
No	130	*		
Baseline Metformin				
Yes	139	0.58	0.45, 0.75	<0.0001
No	97	*		
Any Other Baseline CVD Medications				
Yes	185	1.34	0.98, 1.83	0.06
No	51	*		
Myocardial Infarction				
Yes	95	1.31	1.01, 1.71	0.04
No	141	*		
PCI > 1 Artery				
Yes	62	1.31	0.98, 1.75	0.07
No	174	*		
Subject received stent				
Yes	55	1.34	0.99, 1.81	0.06
No	181	*		
At least 1 drug-eluting stent				
Yes	35	1.42	0.99, 2.03	0.06
No	201	*		
CABG >1 Artery:				
Yes	82	2.05	1.56, 2.68	<0.0001
No	154	*		

Table 10.4.2.1-1: Univariate Cox Regression of Low Lymphocyte Count <750 cell/µl or Investigator Reported Events

Variable	N	Hazard Ratio	95% CI	p-Value
≥2 Coronary Arteries with >50% stenosis:				
Yes	73	1.90	1.44, 2.51	<0.0001
No	163	*		
Peripheral Artery Disease				
Yes	35	1.52	1.06, 2.18	0.02
No	201	*		
Current Symptoms of Claudication				
Yes	34	1.53	1.06, 2.20	0.02
No	202	*		
Angina Pectoris				
Yes	62	1.37	1.02, 1.83	0.03
No	174	*		
Congestive Heart Failure:				
Yes	45	1.88	1.36, 2.61	0.0001
No	191	*		
Arterial Fibrillation/ Flutter:				
Yes	42	3.11	2.23, 4.34	<0.0001
No	194	*		
Dyslipidemia				
Yes	175	1.22	0.91, 1.64	0.18
No	61	*		
Current Smoker:				
Yes	19	0.54	0.34, 0.87	0.01
No	217	*		
Retinopathy				
Yes	39	1.45	1.03, 2.05	0.03
No	197	*		
Laser Treatment:				
Yes	23	1.94	1.26, 2.98	0.003
No	213	*		
Nephropathy:				

Table 10.4.2.1-1: Univariate Cox Regression of Low Lymphocyte Count <750 cell/µl or Investigator Reported Events

Variable	N	Hazard Ratio	95% CI	p-Value
Yes	64	1.77	1.33, 2.35	0.0001
No	172	*		
Microalbuminuria:				
Yes	53	1.81	1.33, 2.45	0.0002
No	183	*		
Amputation due to limb ischemia or diabetic complication:				
Yes	11	2.12	1.16, 3.89	0.01
No	225	*		
Malignancy History:				
Yes	27	2.16	1.45, 3.23	0.0002
No	209	*		

Source: Appendix 2.2.1
^Events occurring more than 30 days after last dose are censored. Table only includes variables with p-value < 0.20.
*Reference group

10.4.2.2 Secondary Objective 1 Multivariate Analysis

Based on 236 patients with events defined as a follow up lymphocyte count <750 cells/µl or investigator reported lymphopenia included in multivariate Cox regression analysis, baseline variables including residing in Latin America (HR=1.87; 95% CI: 1.19, 2.93), age 65 years and older (HR=1.63; 95% CI: 1.18, 2.23), >2 coronary arteries >50% stenosis (HR=1.45; 95% CI: 1.05, 2.00), atrial fibrillation/flutter (HR=2.02; 95% CI: 1.41, 2.90], baseline sulfonylurea (HR=1.34; 95% CI: 1.03, 1.75), and saxagliptin compared to placebo (HR=1.51; 95% CI: 1.16, 1.97) were found to be associated with increased rate of low lymphocyte count, defined as a lymphocyte count < 750 cells/µl or investigator-reported lymphopenia during follow up (Table 10.4.2.2-1).

Table 10.4.2.2-1: Multivariate Cox Regression of Low Lymphocyte Count <750 cell/µl or Investigator Reported Events^

Variable	N	Hazard Ratio	95% CI	p-Value
Treatment:				
Saxagliptin	145	1.51	1.16, 1.97	0.002
Placebo	91			
Age:				
<65 years	58	*		

Table 10.4.2.2-1: Multivariate Cox Regression of Low Lymphocyte Count <750 cell/µl or Investigator Reported Events^

Variable	N	Hazard Ratio	95% CI	p-Value
≥65 years	178	1.63	1.18, 2.23	0.003
Sex:				
Female	175	*		
Male	61	1.00	0.74, 1.37	0.98
Race:				
White	190	*		
Non-white	46	1.22	0.81, 1.84	0.34
Duration of Diabetes:				0.82
<5 years	41	*		
≥5 to <10 years	51	1.12	0.74, 1.71	0.58
≥10 to <15 years	54	1.19	0.78, 1.82	0.41
≥15 to <20 years	32	0.99	0.61, 1.60	0.98
≥ 20 years	58	0.97	0.63, 1.50	0.90
HbA1c Category:				0.38
<6.5	23	*		
≥6.5 to <7	57	1.42	0.87, 2.32	0.16
≥7 to <8	80	1.08	0.67, 1.75	0.74
≥8 to <9	36	1.02	0.59, 1.75	0.96
≥9	40	1.30	0.76, 2.21	0.34
Sitting Diastolic Blood Pressure				
≤80	158	*		
>80	78	0.78	0.58, 1.05	0.98
GFR Category:				0.56
>50	183	*		
≥30 - ≤50	45	0.82	0.57, 1.18	0.83
<30	8	0.84	0.40, 1.80	0.66
Region:				0.003
North America	95	*		
Latin America	43	1.87	1.19, 2.93	0.006
Asia/Pacific	10	0.59	0.28, 1.22	0.15
Europe	88	1.01	0.74, 1.38	0.95
Baseline Sulfonylurea				
Yes	106	1.34	1.03, 1.75	0.03

Table 10.4.2.2-1: Multivariate Cox Regression of Low Lymphocyte Count <750 cell/µl or Investigator Reported Events^

Variable	N	Hazard Ratio	95% CI	p-Value
No	130	*		
Baseline Metformin				
Yes	139	0.74	0.56, 0.99	0.04
No	97	*		
Any Other Baseline CVD Medications				
Yes	185	0.95	0.68, 1.33	0.75
No	51	*		
Myocardial Infarction				
Yes	95	1.09	0.81, 1.47	0.57
No	141	*		
PCI > 1 Artery				
Yes	62	1.11	0.51, 2.40	0.79
No	174	*		
Subject received stent				
Yes	55	1.01	0.42, 2.41	0.99
No	181	*		
At least 1 drug-eluting stent				
Yes	35	1.00	0.57, 1.76	0.999
No	201	*		
CABG >1 Artery:				
Yes	82	1.32	0.96, 1.84	0.09
No	154	*		
>2 Coronary Arteries with >50% stenosis:				
Yes	73	1.45	1.05, 2.00	0.03
No	163	*		
Peripheral Artery Disease				
Yes	35	0.97	0.14, 6.94	0.98
No	201	*		
Current Symptoms of Claudication				
Yes	34	1.58	0.22, 11.62	0.65
No	202	*		

Table 10.4.2.2-1: Multivariate Cox Regression of Low Lymphocyte Count <750 cell/µl or Investigator Reported Events^

Variable	N	Hazard Ratio	95% CI	p-Value
Angina Pectoris				
Yes	62	1.18	0.86, 1.62	0.31
No	174	*		
Congestive Heart Failure:				
Yes	45	1.10	0.76, 1.59	0.61
No	191	*		
Arterial Fibrillation/ Flutter:				
Yes	42	2.02	1.41, 2.90	0.0001
No	194	*		
Dyslipidemia				
Yes	175	1.09	0.80, 1.48	0.59
No	61	*		
Current Smoker:				
Yes	19	1.08	0.67, 1.75	0.74
No	217	*		
Retinopathy				
Yes	39	0.97	0.57, 1.64	0.91
No	197	*		
Laser Treatment:				
Yes	23	1.82	0.94, 3.51	0.07
No	213	*		
Nephropathy:				
Yes	64	0.97	0.50, 1.88	0.93
No	172	*		
Microalbuminuria:				
Yes	53	1.29	0.66, 2.52	0.45
No	183	*		
Amputation due to limb ischemia or diabetic complication:				
Yes	11	1.03	0.52, 2.05	0.94
No	225	*		
Malignancy History:				

Table 10.4.2.2-1: Multivariate Cox Regression of Low Lymphocyte Count <750 cell/µl or Investigator Reported Events^

Variable	N	Hazard Ratio	95% CI	p-Value
Yes	27	1.29	0.85, 1.96	0.23
No	209	*		

Source: Appendix 2.2.3
^Events occurring more than 30 days after last dose are censored. Based on variables with p-value <0.20 in the univariate regression models.
*Reference group

10.4.3 Secondary Objective 2

10.4.3.1 Secondary Objective 2 Univariate Analysis

Univariate Cox proportional hazards regression was used to identify baseline characteristics that were associated with a decreasing lymphocyte count of 30% or more during follow-up. Variables with a statistically significant (p-value <0.20) association with decreasing lymphocyte count are included in Table 10.4.3.1-1. All other univariate results or those variables with a p-value >0.20 are in Appendix 2.3.1.

The demographic variables associated with a decreasing lymphocyte count in this analysis included increasing age (≥65 years HR=1.10; 95% CI: 1.01-1.21), race (HR=1.21; 95% CI: 1.10, 1.34), and ethnicity (HR=1.28; 95% CI: 1.15, 1.41). Region was also associated with a reduction in lymphocyte count [Latin America (HR=1.39; 95% CI: 1.21-1.59), Asia (HR=1.16; 95% CI: 0.97, 1.38) and Europe (HR=0.84; 95% CI: 0.75, 0.93)]. Cardiovascular disease markers continue to be associated with a decreasing lymphocyte count as well as a history of diabetic complications, specifically amputations (HR=1.56; 95% CI: 1.09-2.23 & HR=1.46; 95% CI: 1.11-1.93). Saxagliptin compared to placebo was also associated (HR=1.51; 95% CI: 1.37-1.65) with a decreasing lymphocyte count (Table 10.4.3.1-1).

Table 10.4.3.1-1: Univariate Cox Regression for Patients with a Lymphocyte Count Decrease of 30% or more from Baseline for Variables Included in the Multivariate Cox Regression Model^

Variable	N	Hazard Ratio	95% CI	p-Value
Treatment:				
Saxagliptin	1158	1.51	1.37, 1.65	<0.0001
Placebo	772	*		
Age:				
<65 years	890	*		
≥65 years	1040	1.10	1.01, 1.21	0.03
Race				
White	1422	*		
Non-white	508	1.21	1.10, 1.34	0.0002

Table 10.4.3.1-1: Univariate Cox Regression for Patients with a Lymphocyte Count Decrease of 30% or more from Baseline for Variables Included in the Multivariate Cox Regression Model^

Variable	N	Hazard Ratio	95% CI	p-Value
Ethnicity				
Non-Hispanic/Latino	1450	*		
Hispanic or Latino	480	1.28	1.15, 1.41	<0.0001
BMI Category				
≤ 30	941	*		
> 30	987	0.85	0.78, 0.93	0.0006
Fasting Plasma Glucose FPG (mg/dl):				<0.0001
<126 mg/dl	725	*		
≥126 to <150 mg/dl	416	0.79	0.70, 0.89	0.0001
≥150 to <220 mg/dl	546	0.68	0.61, 0.76	<0.0001
≥220 to <300 mg/dl	147	0.69	0.58, 0.82	<0.0001
≥300 mg/dl	26	0.51	0.35, 0.76	0.0009
Sitting Systolic Blood Pressure				
≤ 130	705	*		
> 130	1225	0.88	0.80, 0.97	0.007
Sitting Diastolic Blood Pressure				
≤ 80	1105	*		
> 80	825	0.86	0.78, 0.94	0.0008
GFR Category:				0.009
>50	1611	*		
≥30 - ≤50	268	1.17	1.03, 1.33	0.02
<30	51	1.36	1.03, 1.80	0.03
Region:				<0.0001
North America	583	*		
Latin America	413	1.32	1.16, 1.50	<0.0001
Asia/Pacific	167	1.16	0.97, 1.38	0.0978
Europe	767	0.84	0.75, 0.93	0.0012
Baseline Metformin				
Yes	1331	0.90	0.82, 0.99	0.04
No	599	*		
Baseline Thiazolidinediones:				
Yes	95	0.76	0.62, 0.94	0.01

Table 10.4.3.1-1: Univariate Cox Regression for Patients with a Lymphocyte Count Decrease of 30% or more from Baseline for Variables Included in the Multivariate Cox Regression Model^

Variable	N	Hazard Ratio	95% CI	p-Value
No	1835	*		
Baseline Aspirin:				
Yes	1468	1.18	1.06, 1.31	0.002
No	462	*		
Baseline Angiotensin Receptor Blocker:				
Yes	567	1.07	0.97, 1.18	0.19
No	1363	*		
Myocardial Infarction				
Yes	721	1.18	1.07, 1.29	0.0006
No	1209	*		
CABG >1 Artery:				
Yes	466	1.23	1.10, 1.36	0.0001
No	1464	*		
≥2 Coronary Arteries with >50% Stenosis:				
Yes	427	1.21	1.09, 1.35	0.0005
No	1503	*		
Peripheral Artery Disease:				
Yes	256	1.38	1.21, 1.57	<0.0001
No	1674	*		
Current Symptoms of Claudication:				
Yes	248	1.38	1.21, 1.58	<0.0001
No	1682	*		
Congestive Heart Failure				
Yes	253	1.22	1.07, 1.39	0.003
No	1677	*		
Atrial Fibrillation/Flutter:				
Yes	166	1.35	1.15, 1.58	0.0002
No	1764	*		
Low level of high-density lipoprotein:				
Yes	778	0.94	0.86, 1.03	0.18

Table 10.4.3.1-1: Univariate Cox Regression for Patients with a Lymphocyte Count Decrease of 30% or more from Baseline for Variables Included in the Multivariate Cox Regression Model^

Variable	N	Hazard Ratio	95% CI	p-Value
No	1152	*		
Current Smoker:				
Yes	288	1.11	0.98, 1.26	0.11
No	1642	*		
HbA1c ≥6.5% in previous 6 months:				
Yes	1926	1.95	0.73, 5.20	0.18
No	4	*		
Nephropathy:				
Yes	368	1.12	1.00, 1.25	0.06
No	1562	*		
Microalbuminuria:				
Yes	289	1.10	0.97, 1.24	0.14
No	1641	*		
Amputation due to Limb Ischemia or Diabetic Complication:				
Yes	62	1.47	1.14, 1.89	0.03
No	1868	*		

Source: Appendix 2.3.1
^Includes subjects who had both baseline and at least one post baseline lymphocyte measurement. Events occurring more than 30 days after last dose were censored. Based on variables with p-value <0.20.
*Reference group

10.4.3.2 Secondary Objective 2 Multivariate Analysis

Based on 1930 patients with events defined as a follow up decreasing lymphocyte count of >30% or more included in multivariate Cox regression analysis, baseline variables including residing in Latin America (HR=1.60; 95% CI=1.27, 2.02), CABG >1 artery (HR=1.14; 95% CI:1.01, 1.28), atrial fibrillation/flutter (HR=1.35; 95% CI: 1.13, 1.60), current smoker (HR=1.15; 95% CI: 1.00, 1.31), and saxagliptin compared to placebo [on top of whatever baseline treatment for diabetes the patient was already receiving (which could include other anti-diabetic medications, such as insulin, but which excluded incretin-based therapies)](HR=1.49; 95% CI: 1.36, 1.64) were found to be associated with events defined as decreasing lymphocyte count of >30% or more during follow up (Table 10.4.3.2-1).

Table 10.4.3.2-1: Multivariate Cox Regression for Patients with a Lymphocyte Count Decrease of 30% or more from Baseline^

Variable	N	Odd Ratio	95% CI	p-Value
Treatment:				
Saxagliptin	1158	1.49	1.36, 1.64	<0.0001
Placebo	772	*		
Age:				
<65 years	890	*		
≥65 years	1040	1.09	0.99, 1.21	0.07
Race				
White	1422	*		
Non-white	508	0.93	0.81, 1.07	0.33
Ethnicity				
Non-Hispanic/Latino	1450	*		
Hispanic or Latino	480	0.87	0.70, 1.07	0.19
BMI Category				
≤ 30	941	*		
> 30	987	0.88	0.80, 0.97	0.01
Fasting Plasma Glucose FPG (mg/dl):				<0.0001
<126 mg/dl	725	*		
≥126 to <150 mg/dl	416	0.81	0.72, 0.91	0.0007
≥150 to <220 mg/dl	546	0.71	0.64, 0.80	<0.0001
≥220 to <300 mg/dl	147	0.70	0.56, 0.84	0.0001
≥300 mg/dl	26	0.52	0.35, 0.77	0.001
Sitting Systolic Blood Pressure				
≤ 130	705	*		
> 130	1225	0.97	0.87, 1.08	0.60
Sitting Diastolic Blood Pressure				
≤ 80	1105	*		
> 80	825	1.00	0.90, 1.11	0.97
GFR Category:				0.41
>50	1611	*		
≥30 - ≤50	268	1.03	0.89, 1.19	0.67
<30	51	1.22	0.91, 1.65	0.19
Region:				

Table 10.4.3.2-1: Multivariate Cox Regression for Patients with a Lymphocyte Count Decrease of 30% or more from Baseline^

Variable	N	Odd Ratio	95% CI	p-Value
North America	583	*		
Latin America	413	1.60	1.27, 2.02	<0.0001
Asia/Pacific	167	1.13	0.92, 1.40	0.25
Europe	767	0.85	0.76, 0.97	0.01
Baseline Metformin				
Yes	1331	0.98	0.89, 1.09	0.76
No	599	*		
Baseline Thiazolidinediones:				
Yes	95	0.79	0.63, 0.97	0.03
No	1835	*		
Baseline Aspirin:				
Yes	1468	1.07	0.95, 1.19	0.28
No	462	*		
Baseline Angiotensin Receptor Blocker:				
Yes	567	1.09	0.98, 1.20	0.10
No	1363	*		
Myocardial Infarction				
Yes	721	1.08	0.97, 1.21	0.16
No	1209	*		
CABG >1 Artery:				
Yes	466	1.14	1.01, 1.28	0.03
No	1464	*		
≥2 Coronary Arteries with >50% Stenosis:				
Yes	427	1.08	0.96, 1.22	0.22
No	1503	*		
Peripheral Artery Disease:				
Yes	256	1.07	0.52, 2.23	0.85
No	1674	*		
Current Symptoms of Claudication:				
Yes	248	1.11	0.53, 2.34	0.77
No	1682	*		

Table 10.4.3.2-1: Multivariate Cox Regression for Patients with a Lymphocyte Count Decrease of 30% or more from Baseline^

Variable	N	Odd Ratio	95% CI	p-Value
Congestive Heart Failure				
Yes	253	1.09	0.95, 1.27	0.22
No	1677	*		
Atrial Fibrillation/Flutter:				
Yes	166	1.35	1.13, 1.60	0.0006
No	1764	*		
Low level of high-density lipoprotein:				
Yes	778	0.94	0.85, 1.03	0.20
No	1152	*		
Current Smoker:				
Yes	288	1.15	1.00, 1.31	0.04
No	1642	*		
HbA1c ≥6.5% in previous 6 months:				
Yes	1926	1.98	0.74, 5.28	0.17
No	4	*		
Nephropathy:				
Yes	368	1.08	0.85, 1.38	0.53
No	1562	*		
Microalbuminuria:				
Yes	289	1.01	0.78, 1.31	0.95
No	1641	*		
Amputation due to Limb Ischemia or Diabetic Complication:				
Yes	62	1.14	0.86, 1.51	0.37
No	1868	*		

Source: Appendix 2.3.3
^Includes subjects who had both baseline and at least one post baseline lymphocyte measurement. Events occurring more than 30 days after last dose were censored. Based on variables with p-value <0.20 in the univariate regression models
*Reference group

10.4.4 Secondary Objective 3

Interaction variables between treatment and the identified risk factors from Sections 10.4.1, 10.4.2 and 10.4.3 were added to each respective multivariate model. For primary objective 1 and

secondary objective 1, interaction variables could not be added due to the high number of covariates relative to the low number of cases; the model did not converge when the interaction variable was added. For secondary objective 2, interaction variables were added to the multivariate model. Results indicated no interaction between the identified risk factors and saxagliptin or placebo (Appendix 2.3.2).

10.4.5 Exploratory Objective

The exploratory analysis to determine whether changes in variables over time were associated with changes in lymphocyte count over time were not conducted for the following reasons:

1. Start dates and end dates were not collected for cardiovascular medications
2. Start dates were missing for the majority of diabetes medications and end dates were not collected
3. Very few patients with lymphopenia also experienced any AEs together with reported lymphopenia diagnosis at the same time during the study period (n=1 saxagliptin patient, n=1 placebo patient)

The low number of patients who experienced both lymphopenia and a clinical event limited the ability to conduct meaningful analyses.

10.5 Sensitivity Analyses

All analyses were duplicated in which exposure to saxagliptin or placebo was considered to have ended at the end of the study period or discontinuation from the SAVOR study, using an intention-to-treat approach. These analyses are included in Appendix 3. All findings in these analyses are consistent with the interpretation of the results described above.

10.6 Adverse Events/Adverse Reactions

Adverse event reporting is outlined in section 6.4.4 of the SAVOR study protocol (Study Code D1680C00003). No additional AE reporting was required for this study.

11 DISCUSSION

This study was conducted as part of the post approval commitments included in the saxagliptin risk management plan. The MAH would like to start with acknowledging the overlap in this analysis and the one conducted as part of the SAVOR study itself. The primary rationale to conduct this risk factor analysis using SAVOR data is non-differential screening of lymphocyte count planned per SAVOR study protocol; the planned lymphocyte assessment is non differential on an annual basis between saxagliptin and placebo, and not measure for a health reason. In a real world setting lymphocyte count is often assessed due to health concerns. In addition to the annual measurement per SAVOR study protocol, lymphocyte counts obtained by local laboratories as part of the SAVOR study were also included in this current analysis as were patient or investigator reports of lymphopenia or decreased lymphocyte count recorded during one of the telephone or clinic visits or spontaneously at any time during the course of the study. Thus, it is important to note the additional lymphocyte counts obtained by local laboratories during the follow up could be considered as due to health reasons.

In this current study, events of interest were defined as follows 1) lymphopenia defined as a follow-up lymphocyte count <500 cells/µl or investigator reported lymphopenia; 2) low lymphocyte count defined as a follow up lymphocyte count <750 cells/µl or investigator reported lymphopenia; and 3) decreasing lymphocyte count, defined as a follow-up decreasing lymphocyte count of 30% or more. In the SAVOR study, a decrease in lymphocyte count was defined differently. The numbers of patients meeting the definitions used in the present study described above were 71, 236, and 1930, respectively.

There is limited information regarding risk factors for lymphopenia and a low lymphocyte count in the general and T2DM population. Several previous studies have looked at risk factors of lymphopenia in special populations often with a specific disease or treatment.[4-14] Overall in these studies the identified risk factors includes history of infections, lymphoma, other malignancies, cardiac failure, autoimmune disorders, sarcoidosis, chemotherapy, immunosuppressive agents, and steroids.

11.1 Key Results

11.2 Primary Objective

Based on 71 patients with events in the primary analysis, baseline variables associated with lymphopenia, defined as lymphocyte count < 500 cells/µl or investigator-reported lymphopenia during follow-up, include: current symptoms of claudication and amputation due to arterial disease, history of cardiovascular conditions (CABG involving >1 Artery, congestive heart failure, atrial fibrillation/flutter), malignancy history, and region (Latin America).

The finding of an association between a history of congestive heart failure and malignancy respectively with lymphopenia is consistent with previous evidence.[5] An association between systemic disease such as autoimmune disorders, lymphoma, other malignancies, sarcoidosis, renal or cardiac failure, and history of infections with lymphopenia has also been suggested.[4-7,9,10,12,14]

The association found between lymphopenia and symptoms of claudication, a history of atrial fibrillation/flutter, CABG involving >1 Artery, or amputation due to arterial disease, have to our knowledge not been reported previously in the scientific literature. These findings must be interpreted with caution in view of many associations being analyzed without a clear hypothesis based on previous observations. It cannot be excluded that one or more of these associations could be due to chance or to unmeasured confounding factors. The observed associations to a history of atrial fibrillation/flutter, amputation due to arterial disease, symptoms of claudication and CABG involving >1 artery are consistent with an association to cardiovascular disease in general and these specific characteristics may not have been analyzed in previous studies.

Another finding is the association between residence in Latin America and lymphopenia. This finding has not been reported previously and the underlying reasons are not clear. In view of multiple comparisons in this study it could be due to chance. Notably, race and/or ethnicity were not found to be associated with lymphopenia in this study. It can be speculated that this finding could be related to a potential disparity in access to health care in Latin America compared to other regions of the world in general as well as among participants in a clinical trial.

11.3 Secondary Objectives

Lymphocyte count < 750 cells/µl or investigator-reported lymphopenia

In the secondary analysis, based on 236 patients with events included in multivariate Cox regression analysis, variables associated with low lymphocyte count (< 750 cells/µl or investigator-reported lymphopenia during follow-up) included the following: increasing age (>65 years), history of cardiovascular conditions (>2 Arteries with >50% stenosis, atrial fibrillation/flutter), baseline sulfonylurea, saxagliptin compared to placebo, and region (Latin America).

The finding of an association between older age and a low lymphocyte count is consistent with previous findings in the general population.[5] Previous evidence indicated that elderly patients have a tendency to develop lymphopenia and it has been suggested that if lymphocyte count is > 500 cell/µl in the absence of other concerning symptoms then no further investigation is required.[5]

We were unable to identify any publications specifically reporting on an association between >2 coronary arteries with >50% stenosis or AF and lymphopenia in patients with type 2 diabetes. The observed association to >2 coronary arteries with >50% stenosis or AF is consistent with an association to cardiovascular disease in general and these specific characteristics may not have been analyzed in previous studies.

In this analysis, baseline use of a sulfonylurea was also associated with a 34% increased risk of developing a low lymphocyte count. A review of the product labels of these drugs (i.e. glyburide, gliclazide, glimepiride and glipizide) did not identify any known lymphopenia adverse reactions. In addition, the association between sulfonylurea and a low lymphocyte count has to our knowledge not been reported previously. This observation must be interpreted with caution due to possible impact of confounding by indication..

Decreasing lymphocyte count of 30% or more

In the secondary analysis, based on 1930 patients with events, history of cardiovascular conditions (CABG involving >1 Artery, atrial fibrillation/flutter), current smoker, saxagliptin compared to placebo, and region (Latin America) were found to be associated with events decreasing lymphocyte count, defined as decreasing lymphocyte count of 30% or more during follow-up.

The finding of Saxagliptin compared to placebo associated with a low lymphocyte count is consistent with previous finding in the pre-authorization saxagliptin clinical program (Saxagliptin [BMS-477118] MODULE 2.7.4 Summary of Clinical Safety) as well as in the SAVOR study (SACOR CSR). In the saxagliptin pre-authorization clinical program, a dose-related decrease in lymphocyte count was observed among saxagliptin users when compared to placebo, and the median change in lymphocyte count from baseline was a decrease of 110 cells/µl for the saxagliptin 5 mg group compared to 20 cells/µl for the placebo group; in the overall saxagliptin group, 9.4% of users experienced a 30% decrease in their lymphocyte count over a 12-month period while 5% of the placebo group also experienced a 30% decrease. In addition, 1.3% of overall saxagliptin groups and 0.4% of the placebo group experienced an absolute lymphocyte count <750 cells/µl, during the short-term and long-term follow-up [Saxagliptin (BMS-477118) MODULE 2.7.4 Summary of Clinical Safety]. In the SAVOR study report, numerically more

subjects in the saxagliptin group had on-treatment lymphocyte counts <500 cells/µl compared with the placebo group [SAVOR CSR: 24/8240 (0.3%) vs 15/8173 (0.2%)]. The results of the present study are consistent with these previously reported findings, though low lymphocyte counts were defined differently in each study.

We were unable to identify any publications specifically reporting on an association between CABG >1 artery or AF and deceasing lymphocyte count of 30% or more in patients with type 2 diabetes. The observed associations to CABG involving >1 artery or AF is consistent with an association to cardiovascular disease in general and these specific characteristics may not have been analyzed in previous studies.

Previous studies suggest that smoking is associated with elevated blood lymphocyte levels.[15] In contrast, our study found smoking to be associated with a ≥30% reduction in lymphocyte count during the follow-up. Tobacco smoking is a well-known risk factor for peripheral vascular disease and certain types of malignancies. Our findings may be confounded by one of these or another unmeasured variable.

For all analyses, there was no evidence of interaction to suggest that there were differences in risk factors between saxagliptin users or placebo users.

11.4 Exploratory Analysis

Analyses for the exploratory objective were not conducted because of the lack of definitive start and end dates of potential time-varying covariates.

11.5 Limitations

As a secondary objective, this study assessed effects of saxagliptin on decrease in lymphocyte counts ≥30%. While SAVOR is a well-balanced study designed to study cardiovascular events as the primary outcomes, it was not designed to study changes in lymphocyte count as the primary outcome.

This study is limited in its ability to adjust for all potential confounders. Due to the low number of lymphopenia cases in SAVOR, we were limited in the number of covariates that could be added to the Cox regression models, and could not include interaction variables to assess whether risk factors varied by saxagliptin user and placebo user.

It is important to note that other known risk factors and potential confounders are not evaluated in this study. Variables during follow up such as anti-diabetic medication use (e.g., DPP4, sitagliptin, linagliptin, SGLT2, GLP1, sulfonylurea, insulin), recent surgeries, immunosuppressive agents, chemotherapy, autoimmune disorders, lymphoma, other malignancies, sarcoidosis, renal or cardiac failure, and history of infections were not included for evaluation in this current study.

There is also potential differential monitoring on ordered tests of lymphocyte count during follow up. Some of the SAVOR subjects had additional tests of lymphocyte count ordered outside the pre-planned annual data collection. Lymphocyte counts from both local and central laboratories were included in the analyses. The lymphocyte counts from the local laboratory in addition to the annual data collection could be biased by the underlying health conditions potentially associated with low lymphocyte count; such information was not included for evaluation in the current study.

Lastly, while the clinically "normal" range for lymphocyte count in adults is between 1000 - 3500 cells/µL, patients with an abnormally low lymphocyte count (<1000 cells/µL) at baseline are included in these analysis.

11.6 Strengths of Research Methods

This secondary analysis used data the SAVOR study, which was a randomized double-blind study with systematic capture of laboratory and other measures.

11.7 Interpretation

This study was conducted to identify risk factors for lymphopenia, and a low lymphocyte count in patients with T2DM. The study confirmed certain previously observed associations and identified additional potential risk factors that have not previously been reported including tobacco smoking and treatment with sulfonylurea. The current study findings for saxagliptin are similar or consistent with findings from the pre-authorization saxagliptin clinical program and from the SAVOR study report, although events were defined slightly differently in each study. .

11.8 Generalizability

The SAVOR patient population represented a subset of the general T2DM patient population. SAVOR included T2DM patients who were at least 40 years and older (more than 50% of SAVOR patients were age of 65 years and older), and had either established CV disease or multiple risk factors for CV events (but without established CV disease). Among approximately 16500 randomized T2DM patients in SAVOR, at least 800 patients had moderate to severe renal impairment and 300 of them had severe renal impairment.

Thus, results from this study are generalizable to T2DM patients meeting the SAVOR inclusion and exclusion criteria and having similar characteristics to the SAVOR study population. Results will be less generalizable to T2DM patients in the real world setting who may have different patient demographics, medications, and comorbid conditions.

12 OTHER INFORMATION

Not applicable.

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