**December 18, 2023**

## Official Title of the study:

**Efficacy of Semaglutide in glycemic control, weight loss, and improving lipid profiles- the role of baseline vitamin D**

**Study number: 0034-23-LEU**

## Introduction

Semaglutide, a glucagon-like peptide-1 (GLP-1) receptor agonist, was initially approved for the treatment of type 2 diabetes mellitus (T2DM) (1) due to its superior glucose-lowering efficacy compared to oral antidiabetic agents, with a favorable safety profile regarding hypoglycemia risk (2-5). In June 2021, the U.S. Food and Drug Administration (FDA) approved higher doses of semaglutide for long-term weight management, marking the first pharmacologic advancement in obesity treatment since 2014 (FDA, 2021).

Semaglutide exerts its effects through multiple mechanisms, including delayed gastric emptying, appetite suppression, enhanced glucose-stimulated insulin secretion from pancreatic β-cells, and reduced glucagon secretion. In individuals with overweight and obese, semaglutide induces substantial and sustained weight reduction, with an average decrease of approximately 14.9% of baseline body weight (6).

The pathophysiology of T2DM varies across racial and ethnic groups. For example, T2DM in Caucasian populations is primarily characterized by obesity and insulin resistance, whereas in East Asians, including the Japanese, early-onset β-cell dysfunction and insulin deficiency predominate (7,8). Consequently, the glycemic response to semaglutide is not uniform across populations. Notably, semaglutide predominantly enhances first-phase insulin secretion, an effect that may be particularly beneficial in individuals with insulin deficiency (9,10). Several blinded, controlled trials have demonstrated superior glycemic control with semaglutide in Asian populations, reinforcing the significance of insulin resistance versus insulin deficiency in determining its net metabolic effects (7,8,11,12,13).

While genetic and ethnic determinants of semaglutide response are largely non-modifiable, certain adjunctive interventions may optimize its therapeutic effects. Among these, vitamin D has emerged as a potential modulator of molecular pathways involved in metabolic regulation. In regard to diabetes control, vitamin D is a key immunomodulatory factor, and preclinical studies have demonstrated its protective effects against β-cell dysfunction in diabetes. In murine models, early vitamin D administration mitigated the inflammatory insults responsible for β-cell destruction, thereby preventing autoimmune diabetes onset (14). These findings are supported by human studies in prediabetic and newly diagnosed T2DM individuals, in whom long-term vitamin D supplementation (six months) significantly improved peripheral insulin sensitivity and β-cell function, potentially delaying diabetes progression (15).

Obesity has long been linked to vitamin D deficiency. While the role of vitamin D in weight loss remains debated, a recent meta-analysis highlighted its potential clinical

efficacy in optimizing weight reduction (16). Specifically, three months of vitamin D supplementation in obese women significantly augmented weight loss and improved metabolic parameters (17).

Given the high prevalence of vitamin D deficiency, particularly among individuals with obesity and T2DM, supplementation may offer metabolic benefits.

To date, the relationship between vitamin D status and the metabolic effects of semaglutide has not been well characterized. In this retrospective cross-sectional study, the investigators aimed to investigate the association between baseline vitamin D levels and semaglutide-induced glycemic control and weight loss in an Israeli T2DM population.

## Materials and Methods

### Study Population

The investigators conducted a population-based study among adult members of Leumit Health Services (LHS), a large Israeli nationwide health maintenance organization (HMO), which provides health services to nearly 730,000 members. LHS has a comprehensive computerized database, continuously updated regarding the demographics, medical diagnoses and clinic visits, hospitalizations, and laboratory tests of insured members.

The socio-economic status (SES) was defined according to the home address. The Israeli Central Bureau of Statistics categorizes all cities and settlements into 20 SES levels. Classification at levels 1–9 is considered low–medium SES, while levels 10–20 represent the medium–high SES. Ethnicity was also defined according to the home address of the HMO members, and categorized into three groups: general population, ultra-orthodox Jews, and Arabs.

All LHS members have identical health insurance coverage and access to healthcare services. Relevant diagnoses are entered or updated according to the International Classification of Diseases 9th revision (ICD-9). The validity of chronic diagnoses in the registry has been previously established (Hamood et al., 2016; Rennert and Peterburg, 2001). The study population included all LHS members aged 18 or older, with T2DM who were prescribed semaglutide since 2019 and fulfilled the following criteria:

- Consumed semaglutide regularly for at leastsix months.
- Have at least one plasma 25(OH)D level prior to semaglutide initiation.

Baseline medical conditions including obesity, T2DM, hypertension, asthma, chronic obstructive pulmonary disease, ischemic heart disease, the presence of malignancy, and chronic kidney disease, were recorded. Obesity was defined as BMI > 30 kg/m2. According to LHS guidelines, vitamin D tests were collected after overnight fasting and transported on ice to the central laboratory for processing within 4 h of collection using the DiaSorin Chemiluminescence assay (18-21). For categorization of vitamin D levels, the investigators calculated the median value and divided the patients into three categories: 0-15 ng/ml, 15-25 mg/dl, and > 25 ng/ml. The study protocol was approved by the LHS Institutional Review Board (0034-23-LEU).

**3 .Statistical Analysis**

Descriptive statistics, including mean, standard deviation, median, and percentiles, were reported for all study parameters. Group differences were assessed using the t-test for continuous variables and Fisher's exact test for categorical variables. To evaluate the impact of vitamin D level categories on changes in HbA1c, BMI, and LDL cholesterol following GLP-1 treatment, linear regression models were fitted with vitamin D category as the main explanatory variable. Models were adjusted for age, gender, and the last recorded values of HbA1c, BMI, and LDL cholesterol before treatment initiation.

Variables included in the multivariate analysis were selected based on their statistical significance in univariate analyses. A p-value < 0.05 was considered statistically significant. All statistical analyses were conducted using R version 4.4.0.

Only inpatients aged ≥18 years with a definite diagnosis of T2DM (recorded in the diabetes mellitus registry) were included in the study. T2DM was defined according to the American Diabetes Association classification (26). The exclusion criteria were as follows: (1) with missing serum 25-hydroxyvitamin D (25[OH]D) data; (2) pregnant or lactating females

**Data Collection**

General demographic information, including age, sex, smoking status, duration of diabetes, and family history of diabetes were collected. Body mass index (BMI) was calculated as weight divided by height squared.

The laboratory measurements collected in this study included: serum 25(OH)D, albumin, triglycerides, total cholesterol, high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), fasting plasma glucose, glycated hemoglobin (HbA1c), and serum creatinine. Notably, serum 25(OH)D concentration was determined by chemiluminescence assay (Siemens ADVIA Centaur XP, Germany) and the detection limit was <10.5 nmol/L. Estimated glomerular filtration rate (eGFR) was

calculated using the abbreviated Modification of Diet in Renal Disease (MDRD) equation: 186× (serum creatinine) −1.154×(age)−0.203× (0.742 if female).

In addition, diabetic complications (i.e., DR, DKD, DFU, diabetic peripheral neuropathy [DPN]) and related comorbidities (i.e., hypertension, dyslipidemia, coronary heart disease, cerebrovascular disease) were also evaluated.

Participants were divided into three groups by vitamin D level (i.e., 0.1-14.9 ng/ml; 15–24.9 ng/ml; And 25-100 ng/ml). Participants were also categorized into four groups based on the levels of BMI: underweight (<18.5 kg/m2), normal weight (18.5–24.9 kg/m2), overweight (25.0–29.9 kg/m2), and obese (≥30.0 kg/m2). Glycemic control was classified based on HbA1c levels.