

Semaglutide s.c. Once-Weekly in Type 2 Diabetes: A Population Pharmacokinetic Analysis

Kristin Cecilie Carlsson Petri · Steen Hvass Ingwersen ·
Anne Flint · Jeppe Zacho · Rune Viig Overgaard

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ABSTRACT

Introduction: Semaglutide, a new treatment option approved for the treatment of patients with type 2 diabetes mellitus, is a glucagon-like peptide-1 receptor agonist to be injected subcutaneously once weekly. This analysis used a population pharmacokinetic model of semaglutide to identify clinically relevant covariates for exposure.

Methods: A total of 1612 patients with up to seven pharmacokinetic observations each were included in the analysis. All subjects had type 2 diabetes mellitus and were enrolled in one of five trials in the phase III development program for subcutaneous semaglutide once weekly (the SUSTAIN program). The treatment duration of the trials varied from 30 to 104 weeks.

Results: No clinically relevant effects on the exposure were seen for sex, age, race, ethnicity,

renal function, or injection site used, and semaglutide exposure was stable over time. Of the covariates chosen, only body weight had a relevant effect on the exposure of semaglutide. Few subjects developed semaglutide antibodies, and the antibodies had no effect on exposure. Dose proportionality was shown for the 0.5 mg and 1.0 mg maintenance doses of semaglutide.

Conclusion: The population pharmacokinetic study showed that semaglutide exposure is not affected by covariates other than body weight at either a maintenance dose of 0.5 or 1.0 mg semaglutide. Therefore, we conclude that no semaglutide dose adjustments are needed in different populations. This finding is to be further explored in an exposure-response analysis.
Trial Registration: The trials were registered at ClinicalTrials.gov (identifiers: NCT02054897, NCT01930188, NCT01885208, NCT01720446 and NCT02207374).

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K. C. Carlsson Petri (✉) · S. H. Ingwersen · A. Flint · J. Zacho · R. V. Overgaard
Novo Nordisk A/S, Vandtårnsvej 108, 2860 Søborg, Denmark
e-mail: KCC@NovoNordisk.com

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INTRODUCTION

Diabetes is of increasing global concern. Since 1980, the population of adults with diabetes has quadrupled worldwide. The age-standardized

prevalence of diabetes has increased, and no single country has managed to see a reduction in this prevalence [1]. Type 2 diabetes mellitus (T2DM) is the most common type of diabetes, making up approximately 90% of the total number of patients with diabetes in high-income countries, with an estimated 415 million adults with diabetes worldwide in 2015 [2].

T2DM is linked to a range of comorbidities, such as cardiovascular diseases (CVD), microvascular complications, and cancer [2, 3]. Patients with T2DM have twice the risk of CVD mortality compared with age-matched subjects without T2DM, approximately 10% increased risk of cancer, and poorer survival rates of cancer [4–6]. As the number of comorbidities a patient suffers from and the number of medications a patient is prescribed increase, his or her perceived quality of life decreases [7]. There are many available treatments, but there is still a need for more effective treatments for T2DM.

Semaglutide (marketed as Ozempic[®]) is a glucagon-like peptide-1 (GLP-1) receptor agonist approved for the treatment of T2DM. Native human GLP-1 functions in a glucose-dependent manner, increasing insulin secretion and decreasing glucagon secretion in the presence of elevated blood glucose [8, 9]. GLP-1 also decreases appetite and energy intake [10]. However, endogenous GLP-1 has a short half-life ($t_{1/2} = 2\text{--}3$ min) and is therefore not well suited for the treatment of T2DM [11]. Semaglutide retains 94% amino acid homology with native GLP-1, allowing it to retain GLP-1 signaling functionality but with a half-life of about 1 week. This allows for subcutaneous injection once weekly [12].

The Semaglutide Unabated Sustainability in Treatment of Type 2 Diabetes (SUSTAIN) phase III trials have shown that semaglutide has a strong glycated hemoglobin (HbA_{1c})-lowering effect in patients with T2DM when injected subcutaneously once weekly [13–19]. As with other GLP-1 receptor agonists (GLP-1RAs), treatment with semaglutide causes a reduction in body weight. Globally conducted clinical trials have shown that semaglutide can provide superior glycemic control and body weight loss compared with placebo, sitagliptin, exenatide extended-release or insulin glargine [13–19] and

compared with additional oral anti-diabetic drugs investigated in a dedicated Japanese trial [20]. In addition, semaglutide has been shown to reduce systolic blood pressure, and no cardiovascular risk was demonstrated [19].

In the present report, we document the pharmacokinetics of semaglutide in subjects with T2DM based on pharmacokinetic modeling of drug concentrations from the phase III data. The covariates included in the analysis were chosen to reflect important baseline characteristics in the broad spectrum of patients with T2DM as well as other possibly important factors related to semaglutide exposure. The population pharmacokinetic model tested the effect of sex, age, race, ethnicity, body weight, renal function, maintenance dose level used and injection site chosen on the individual average steady-state plasma concentrations of semaglutide. Additionally, the effects of time since first dose and presence of semaglutide antibodies on exposure were evaluated.

METHODS

Data Sources for the Population Pharmacokinetic Model of Semaglutide

The population pharmacokinetic model for semaglutide used data generated from five of the phase III SUSTAIN trials; these were SUSTAIN 1, 2, 3, 6, and SUSTAIN-Japan [13–15, 19, 20]. The design of these trials is summarized in Table 1.

The trials included in the model were all global trials, with the exception of the Japanese trial. Male and female subjects diagnosed with T2DM were included, with an age ≥ 18 years (Japanese patients ≥ 20 years). For all trials, subjects had a minimum HbA_{1c} of 7.0%, and there were no restrictions on body weight or body mass index (BMI). With the exception of SUSTAIN 6, inclusion and exclusion criteria were overall similar across all trials. Concomitant oral anti-diabetic drugs (OADs) were allowed for all trials (except in SUSTAIN 1, which was a monotherapy trial). Subjects enrolled in SUSTAIN 6 were also allowed basal or pre-mix insulin. SUSTAIN 6 was a pre-

Table 1 Characteristics of trials included in the population PK analysis

	SUSTAIN 1	SUSTAIN 2	SUSTAIN 3	SUSTAIN 6 (CVOT)	SUSTAIN–Japan
Blinding	Double blind	Double blind	Open label	Double blind	Open label
Comparator	Placebo	Sitagliptin 100 mg	Exenatide ER 2.0 mg	Placebo	Additional OAD
Semaglutide maintenance dose	0.5, 1.0 mg	0.5, 1.0 mg	1.0 mg	0.5, 1.0 mg	0.5, 1.0 mg
Randomization	2:2:1:1 ^a	2:2:1:1 ^b	1:1	1:1:1:1 ^a	2:2:1 ^c
Planned number subjects randomized	390	1200	798	3260	595
Planned number subjects randomized to semaglutide	260	800	399	1630	480
Treatment duration	30 weeks	56 weeks	56 weeks	104 weeks	56 weeks
Background medication	None	1–2 OADs (either MET, PIO, ROSI, or a combination of either MET/PIO or MET/ROSI	1–2 OADs (MET and/ or TZD and SU)	0–2 OADs, basal or premixed insulins ± 0–2 OADs. Background medication was allowed to change during the trial	0–1 OAD (of SU, glinide, α-GI or TZD)
Clinicaltrials.gov identifier	NCT02054897	NCT01930188	NCT01885208	NCT01720446	NCT02207374

α-GI α-glucosidase inhibitor, CVOT cardiovascular outcomes trial, MET metformin, OAD oral antidiabetic drug, PIO pioglitazone, PK pharmacokinetic, ROSI rosiglitazone, SU sulfonylurea, SUSTAIN Semaglutide Unabated Sustainability in Treatment of Type 2 Diabetes, TZD thiazolidinediones

^a Relative proportions randomized to 0.5 mg semaglutide, 1.0 mg semaglutide, 0.5 mg placebo, and 1.0 mg placebo

^b Relative proportions randomized to 0.5 mg semaglutide: 1.0 mg semaglutide, sitagliptin with 0.5 mg semaglutide placebo, sitagliptin with 1.0 mg semaglutide placebo

^c Relative proportions randomized to 0.5 mg semaglutide, 1.0 mg semaglutide, additional OAD

^d The aim was to perform the assessments in approximately 60 subjects with severe renal impairment (GFR value 15–29 ml/min/1.73 m²) and 180 subjects without severe renal impairment (normal renal function as well as mild and moderate renal impairment)

approval cardiovascular and other long-term outcomes trial with an enriched CVD population (aged ≥ 50 years with clinical evidence of CVD or aged ≥ 60 years with cardiovascular risk

factors). Subjects with normal or mildly impaired renal function [defined as estimated glomerular filtration rate (eGFR) ≥ 90 ml/min/1.73 m² for normal function or eGFR of

60–89 ml/min/1.73 m² for mildly impaired function] were enrolled in all trials. SUSTAIN 1, SUSTAIN 6, and SUSTAIN-Japan also included subjects with moderate renal impairment (eGFR of 30–59 ml/min/1.73 m²). In SUSTAIN 6, the pharmacokinetic (PK) properties of semaglutide were assessed in subjects with severe impaired renal function (eGFR < 30 ml/min/1.73 m²) as well as subjects with less severe renal impairment or normal renal function. For more information on inclusion and exclusion criteria in the individual trials, see the trial publications [13–15, 19, 20].

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments and Good Clinical Practice [21, 22]. The trial protocols were approved by independent ethics committees and/or institutional review boards. All subjects provided written informed consent before initiation of any trial-related activities.

Subjects included in the population pharmacokinetic analysis were randomized to a maintenance dose of either 0.5 mg or 1.0 mg semaglutide once weekly. SUSTAIN 3 used only the 1.0 mg maintenance dose for the semaglutide treatment arm (Table 1). In all trials, subjects taking semaglutide followed a dose-escalation regimen with the aim of minimizing gastrointestinal adverse events common to the class of GLP-1RAs [23]. Subjects started with 4 weeks of treatment with 0.25 mg semaglutide once weekly before escalating to 0.5 mg semaglutide once weekly. Subjects randomized to the 1.0 mg semaglutide treatment arm would escalate to the 1.0 mg dose after 4 weeks of 0.5 mg semaglutide, remaining on this dose for the duration of the trial. The subcutaneous injections should be administered on the same day of each week, and subjects providing blood samples to be used in the population pharmacokinetic model were encouraged to inject in the same area of the body throughout the trial.

Blood samples for the PK measurement were drawn at approximately 4, 8, 16, and 30 weeks after the first dose in all trials, and all except SUSTAIN 1 included a sample during week 56.

SUSTAIN 6 had additional samples taken during week 2 and at end of treatment (minimum 104 weeks after first dose). There were no restrictions on the timing of the blood sample relative to dosing.

Subjects recorded the date, time, and injection site for the initial dose and the two doses prior to each blood sampling in a diary. This information was transferred, along with the date and time of blood sampling to an electronic case report form at each sampling visit.

Subjects exposed to at least one dose of semaglutide and with at least one valid PK measurement were included in the data set, which included dosing information for all recorded injections, semaglutide concentrations, and covariate values. Records with concentration values missing or below the lower limit of quantification, other dosing deviations, or missing information were excluded from the analysis. Dosing deviations were defined as missing injections or if the last two injections prior to blood sampling were administered less than 120 h apart. Missing information was defined as incomplete diary information for 2 weeks before blood sampling.

Semaglutide Assay

The semaglutide plasma concentrations were measured following protein precipitation using a validated liquid chromatography assay followed by a tandem mass spectrometry assay (Celerion Inc. Fehraltorf, Switzerland); see [24] for more details. The lower limit of quantification of the assay used for samples included in this population PK model was 0.729 nmol/l.

Population Pharmacokinetic Model

A pre-specified full model approach was used for the population PK analysis, including a base model without covariates and a full model with all covariates included [25, 26].

The base model was a one-compartment model with first-order absorption and elimination. This model has been shown to provide an adequate description of the PK of semaglutide (Novo Nordisk, data on file). The model was

parameterized for semaglutide in terms of k_a (absorption rate constant), CL/F (apparent clearance), and V/F (apparent volume of distribution). The semaglutide absorption rate constant (k_a) was set to 0.0286 h^{-1} based on data from clinical pharmacology trials with richly sampled PK profiles (Novo Nordisk, data on file). The model was estimated on un-transformed concentration values, and a proportional error model was used to describe the residual variability. Models were estimated using first-order conditional estimation with interaction (FOCE + I).

The full model was used for estimating the potential effects of individual covariates on semaglutide plasma exposure in terms of clearance. The average semaglutide concentration (C_{avg}) during the dosing interval was

$$C_{\text{avg}} = \frac{\text{AUC}_{\text{ss},0-168\text{h}}}{168 \text{ h}}$$

The area under the curve at steady state ($\text{AUC}_{\text{ss},0-168\text{h}}$) was calculated by

$$\text{AUC}_{\text{ss},0-168\text{h}} = \frac{\text{Dose}}{\text{CL}/F},$$

where ‘dose’ was the relevant maintenance dose for the subject, and CL/F was the individually estimated apparent clearance from the population pharmacokinetic model.

The covariates were included to investigate exposures in relevant sub-populations [2, 27, 28] and the dosing characteristics of semaglutide. Covariates were categorical, with the exception of body weight. Age was categorized into three groups, < 64 , $65-74$, or ≥ 75 years old at baseline. Renal impairment groups were categorized [by estimated glomerular filtration rate (eGFR)] as normal function ($\text{eGFR} \geq 90 \text{ ml/min/1.73 m}^2$), mild ($\text{eGFR} = 60-89 \text{ ml/min/1.73 m}^2$), moderate ($\text{eGFR} = 30-59 \text{ ml/min/1.73 m}^2$), or severe ($\text{eGFR} < 30 \text{ ml/min/1.73 m}^2$) renal impairment. Baseline body weight was included as a continuous covariate. For semaglutide, there are two maintenance doses (0.5 and 1.0 mg), which were included as covariates to assess the dose dependency of semaglutide exposure. The injection site (abdomen, thigh or upper arm)

was also included as a covariate, whereby the most frequently used injection site for an individual patient was used as the covariate value. Additionally, race and ethnicity were included as covariates.

The reference subject profile was defined as a non-Hispanic or non-Latino, white female < 65 years old, with a body weight of 85 kg (pre-specified, representing the expected approximate median body weight of the study population) with normal renal function and dosed in the abdomen with semaglutide 1.0 mg once weekly.

The model was parameterized as:

$$\begin{aligned} CL_i/F &= CL_{\text{typ}} \cdot E_{\text{dose}} \cdot E_{\text{weight}} \cdot E_{\text{sex}} \cdot E_{\text{age}} \cdot E_{\text{GFR}} \cdot \\ &E_{\text{race}} \cdot E_{\text{ethnicity}} \cdot E_{\text{inj.site}} \cdot \exp(\eta_i) \\ E_{\text{dose}} &= (\theta_{\text{dose}0.5\text{mg}})^{\text{dose}0.5\text{mg}} \\ E_{\text{weight}} &= \left(\frac{\text{weight}}{85 \text{ kg}} \right)^{\theta_{\text{wt}}} \\ E_{\text{sex}} &= (\theta_{\text{male}})^{\text{male}} \\ E_{\text{age}} &= (\theta_{\text{age}65-74\text{y}})^{\text{age}65-74\text{y}} \cdot (\theta_{\text{age} \geq 75\text{y}})^{\text{age} \geq 75\text{y}} \\ E_{\text{GFR}} &= (\theta_{\text{GFRmild}})^{\text{GFRmild}} \cdot (\theta_{\text{GFRmoderate}})^{\text{GFRmoderate}} \cdot \\ &(\theta_{\text{GFRsevere}})^{\text{GFRsevere}} \\ E_{\text{race}} &= (\theta_{\text{BlackAfrAm}})^{\text{BlackAfrAm}} \cdot (\theta_{\text{Asian}})^{\text{Asian}} \cdot (\theta_{\text{Other}})^{\text{Other}} \\ E_{\text{ethnicity}} &= (\theta_{\text{Hispanic}})^{\text{Hispanic}} \\ E_{\text{inj.site}} &= (\theta_{\text{Thigh}})^{\text{Thigh}} \cdot (\theta_{\text{Upperarm}})^{\text{Upperarm}} \end{aligned}$$

where CL_{typ} was the typical semaglutide clearance (CL/F) for the reference subject, and θ was used for covariate effect parameters. Exponents used for categorical covariate relations are indicator variables assigned the value 1 for the actual category and else 0, e.g., 1 for males and 0 for females. Groups that contained < 20 subjects were merged with the largest covariate group. Between-subject variability was assumed to be log-normally distributed and was included as η .

Between-subject variability was estimated for CL/F and V/F in both the base and full PK models to account for the degree of variability that could be explained by inclusion of the covariates. If the dose level was missing in a subject’s dosing diary, it was assumed to be the planned dose.

Data Analysis Software

The software program R (version 2.14.2, R Foundation; Revolution Analytics, Mountain View, CA, USA, version 6) was used for data file processing, explorative data analysis, and plotting. NONMEM (ICON Development Solutions, Ellicott City, MD, USA), version 7.1.2, was used for the population pharmacokinetic analysis. Both of these programs were run as validated server installations. PsN [29, 30] was used for the visual predictive check, and data processing was done with R.

RESULTS

Demographics

A total of 1683 subjects treated with semaglutide were scheduled for inclusion in the population pharmacokinetic assessment. After data cleaning, the final data set included 1612 subjects with a total of 6781 PK measurements, a mean 4.2 semaglutide concentrations per subject. Subjects were excluded because of missing PK data (19 subjects), semaglutide concentrations below the lower level of quantification (33 subjects), or incomplete or missing dosing history (19 subjects), resulting in an exclusion of 8.3% of the PK samples.

The subjects included in the population pharmacokinetic analysis covered a broad range of baseline characteristics: age ranged from 20 to 86 years, body weight ranged from 39.7 to 198.3 kg, duration of diabetes ranged from 0 to 48.9 years, and HbA_{1c} ranged from 5.9% to 13.1%. Both sexes were well represented, and subjects covered several races and different ethnicities and were recruited from many countries. Subjects with normal to severely impaired renal function were also included. A summary of the baseline characteristics is presented in Table 2 [additional characteristics are provided in Table S1 in the electronic supplementary material (ESM)].

Model Qualification

A one-compartment model with first-order absorption and elimination successfully described the pharmacokinetics of semaglutide. The parameter estimates for the base model can be seen in Table S2 in the ESM. Based on the full population pharmacokinetic model, values for CL/F and V/F in the reference subject profile were estimated to be 0.0478 l/h and 12.2 l, respectively (Table S3 in the ESM).

The full model was robust toward changes in k_a , evaluated using sensitivity analyses with two alternative models with modified k_a values ($\pm 25\%$ of the fixed value). Neither of these models had any relevant differences in terms of exposure compared with the presented model; moreover, the model was robust toward exclusion of data with high residuals, i.e., weighted residuals above 4 and below -4 (Table S4 in the ESM).

The model was qualified in accordance with regulatory guidelines [31, 32]. The model fits for both the base and full model were acceptable, and there were no critical trends in the conditional weighted residuals vs. either semaglutide concentration or time. The individual clearance and volume of distribution estimates appeared to approximate log-normal distributions. Model evaluation was performed through visual predictive checks using PsN and R. One thousand simulated data sets were generated, with a stratification by dose level. The 2.5th, 50th, and 97.5th percentiles of the experimental data were calculated. Then, the 95% confidence intervals of the 2.5th, 50th, and 97.5th percentiles were computed and displayed graphically together with the observed percentiles. The visual predictive check showed that the full model was able to reproduce the median concentrations of the population and that the simulated 95% confidence intervals were in line with the observed data (see Fig. S2, S3 and S4 in the ESM).

Population Pharmacokinetic Analysis

The mean semaglutide plasma concentration (C_{avg}) for the reference subject profile at steady

Table 2 Baseline characteristics of subjects included in the population PK analysis

Category	Group	Total
All	N	1612 (100%)
Sex	Male	927 (57.5%)
	Female	685 (42.5%)
Age group	18–64 years	1203 (74.6%)
	65–74 years	353 (21.9%)
	≥ 75 years	56 (3.5%)
Race	White	838 (52.0%)
	Asian	658 (40.8%)
	Black or African American	73 (4.5%)
	American Indian or Alaska Native ^a	2 (0.1%)
	Unknown ^a	41 (2.5%)
Ethnicity	Not Hispanic or Latino	1371 (85.0%)
	Hispanic or Latino	241 (15.0%)
Renal function ^b	Normal	997 (61.8%)
	Mild impairment	533 (33.1%)
	Moderate impairment	49 (3%)
	Severe impairment	33 (2.0%)
Maintenance dose	Semaglutide 0.5 mg	634 (39.3%)
	Semaglutide 1.0 mg	978 (60.7%)
Body weight, kg	Mean (SD)	86.2 (22.5)
	Range	[39.7–198.3]
BMI, kg/m ²	Mean (SD)	31.1 (7.1)
	Range	[16.3–72.8]
Duration of diabetes, years	Mean (SD)	8.1 (6.6)
	Range	[0–48.9]

Table 2 continued

Category	Group	Total
HbA _{1c} , %	Mean (SD)	8.2 (1)
	Range	[5.9–13.1]

Categories are ordered with categorical variables first followed by continuous variables. For parameters for each trial, see the electronic supplementary material

BMI body mass index, *HbA_{1c}* glycated hemoglobin A1c, *SD* standard deviation

^a The two groups ‘American Indian or Alaska Native’ and ‘unknown’ were merged with the group ‘white’ for the covariate analysis. Subjects without information on race were from France (*n* = 20), Mexico (*n* = 13), Canada (*n* = 2), USA (*n* = 2), Australia (*n* = 1), Norway (*n* = 1), South Africa (*n* = 1) and the UK (*n* = 1)

^b Renal function was based on eGRF defined as normal function: ≥ 90 ml/min/1.73 m², mild impairment: 60–89 ml/min/1.73 m², moderate impairment: 30–59 ml/min/1.73 m², and severe: < 30 ml/min/1.73 m²

state was estimated to be 15.8 nmol/l [95% confidence interval (CI) 15.6–16.1] with 0.5 mg semaglutide s.c. once weekly at steady state and about twice that at 29.8 nmol/l (95% CI 29.4–30.2) with 1.0 mg semaglutide at steady state (Table 3).

There was a large overlap in the exposure at the two doses, and exposure was generally similar at the same dose level between trials (Fig. 1). Subjects in the Japanese trial appeared to have slightly higher exposures compared with those in the global trials. As explained below, this was mainly due to lower body weights in Japanese individuals and not the influence of Japanese descent.

The population pharmacokinetic model estimated the effects of the chosen covariates on semaglutide exposure. Covariate effects were considered not to be important for exposure if the 90% CI of the relative exposure was within the 0.8–1.25 standard equivalence range. The estimated effect of each analyzed covariate is presented in Fig. 2. There were no important changes in exposure dependent on sex, age, race, ethnicity, renal function, or injection site used. Of the analyzed covariates, only body weight was considered important for the exposure of semaglutide. Dose-normalized exposure

Table 3 Summary of model-derived semaglutide exposures from the PK population

	Maintenance dose	
	0.5 mg	1.0 mg
Number of subjects		
<i>N</i> (%)	634 (39.3%)	978 (60.7%)
<i>C</i> _{avg} (nmol/l)		
Geometric mean (95% CI)	15.8 (15.6–16.1)	29.8 (29.4–30.2)
Range	[8.3–30.2]	[14.8–61.3]
Median	15.8	30.0
95% CI	[10.1–24.6]	[18.8–46.9]
AUC (h·nmol/L)		
Geometric mean (95% CI)	2660 (2614–2707)	5006 (4934–5079)
Range	[1388–5080]	[2485–10,299]
Median	2663	5034
95% CI	[1689–4134]	[3166–7875]

AUC area under the concentration-time curve, *C*_{avg} average semaglutide concentrations at steady state, *CI* confidence interval

was similar in the two dose groups, indicating dose proportionality (Fig. 2).

The between-subject variability of CL/F in the base model, in terms of the coefficient of variation (CV) %, was 26.6% and was reduced to 12.9% by inclusion of all covariates in the full model. This corresponds to 75.8% of the variability being explained by covariates.

Semaglutide exposure was inversely related to body weight. Compared with the reference subject of 85 kg, a subject weighing 55 kg had on average a 40% increased semaglutide exposure and a 127 kg subject had a 27% lower semaglutide exposure. These weights represent the 5% and 95% percentiles of the subjects in the trials.

The simulated mean semaglutide profile during three dosing intervals after s.c. dosing at steady state semaglutide 0.5 mg and 1.0 mg in subjects with T2DM is shown in Fig. 3. The baseline body weights of 70 kg and 100 kg represent the 25% and 75% weight quartiles in the studied population. The profiles were relatively flat, supporting low variability over time as well

as low variability between subjects. There also appeared to be an overlap in predicted exposure between the two weight groups at the same dose.

The observed inverse relationship between exposure and body weight over the entire body weight range is shown in Fig. 4 and was similar between males and females (Fig. 4a). Body weight differences may also explain the apparent difference in exposure values in the Japanese trials mentioned above; at the same body weight, the same exposure was seen in Asian (Japanese or non-Japanese) and non-Asian subjects (Fig. 4b).

Exposure of semaglutide was constant over the 2 years data were collected and appeared to be time-independent (Fig. S1 in the ESM).

Few subjects (*N* = 29) developed anti-semaglutide antibodies, and the presence of antibodies did not appear to be related to a difference in exposure, as exposures in subjects with antibodies were similar to those observed in subjects without antibodies (data on file).

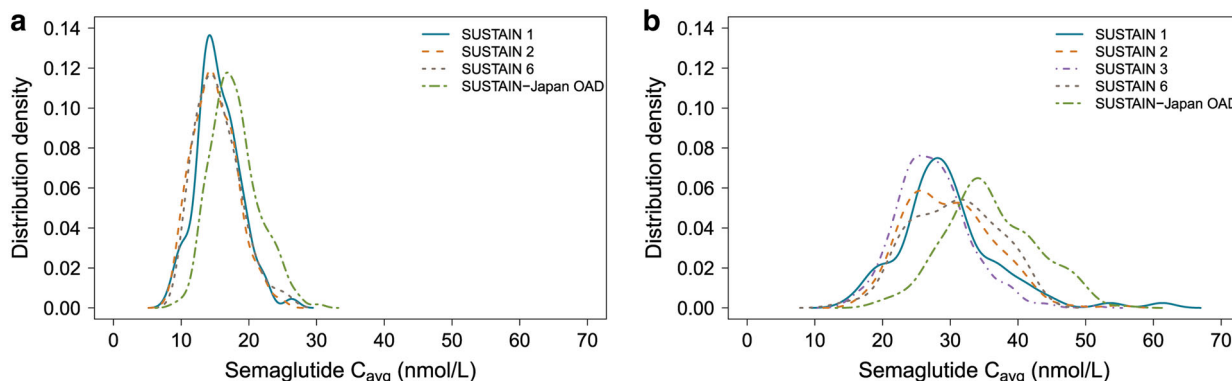


Fig. 1 Distribution of exposure values from trials included in the population PK analysis. **a** Exposure for subjects treated with 0.5 mg semaglutide; **b** exposure for subjects

treated with 1.0 mg semaglutide. *PK* pharmacokinetic, *SUSTAIN* Semaglutide Unabated Sustainability in Treatment of Type 2 Diabetes

Covariate	Test category	Reference category	Relative Exposure (Cavg)	Ratio [90% CI]
Sex	Male (N:927)	Female (N:685)	■	0.96 [0.95;0.98]
Age group	65–74 years (N:353)	18–64 years (N:1203)	■	1.01 [0.99;1.03]
	>74 years (N:56)		■	1.04 [1.00;1.08]
Race	Black or African American (N:73)	White (N:838)	■	1.03 [0.99;1.07]
	Asian (N:658)		■	1.01 [0.99;1.03]
Ethnicity	Hispanic or Latino (N:241)	Non-Hispanic or Latino (N:1371)	■	0.94 [0.92;0.96]
Body weight	55 kg	85 kg	■	1.40 [1.38;1.42]
	127 kg		■	0.73 [0.72;0.74]
Renal function	Mild impairment (N:533)	Normal (N:997)	■	1.06 [1.04;1.07]
	Moderate impairment (N:49)		■	1.05 [1.00;1.09]
	Severe impairment (N:33)		■	1.09 [1.03;1.15]
Maintenance dose	0.5 mg (N:634)	1.0 mg (N:978)	■	1.00 [0.98;1.01]
Injection site	Thigh (N:86)	Abdomen (N:1456)	■	0.97 [0.93;1.00]
	Upper arm (N:70)		■	0.93 [0.90;0.96]

Fig. 2 Forest plot of covariate analysis for semaglutide exposure expressed as steady-state dose-normalized average semaglutide concentrations relative to a reference subject. The reference subject profile was non-Hispanic or Latino, white, female, below 65 years, with a body weight of 85 kg, with normal renal function, and who was dosed in the abdomen with semaglutide 1 mg. The column to the right shows means and 90% CI for the relative exposures. Two additional race groups (American Indian or Alaska native, $n = 3$ subjects, and unknown, $n = 41$) were included in the analysis without a separate race covariate, i.e., modeled

as the reference race group (white) in the covariate analysis. Subjects without information on race were from France ($n = 20$), Mexico ($n = 13$), Canada and the USA ($n = 2$ each), Australia, Norway, South Africa, and the UK ($n = 1$ each). Body weight test categories (55 and 127 kg) represent the 5% and 95% percentiles, respectively, in the data set. Vertical dotted lines indicate the acceptance interval for bioequivalence (0.80; 1.25). C_{avg} average semaglutide concentrations at steady state; *CI* confidence interval

The dosing recommendations given to subjects during the semaglutide phase III program for delayed or missed doses were tested using

this model. Subjects were recommended to take a missed semaglutide dose as soon as possible within 5 days of the planned dose; a dose

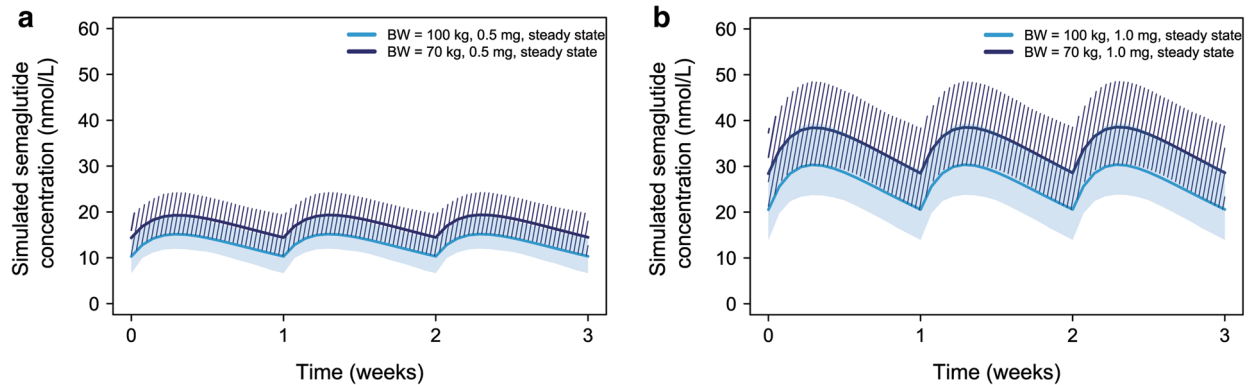


Fig. 3 Simulated concentration profiles for semaglutide 0.5 mg (a) or 1.0 mg (b) at steady state over 3 weeks, with variability. The shaded area illustrates the simulated 95%

concentration range predicted from the between-subject variability in the full population PK model ($N = 1000$ replications in each group). *BW* body weight

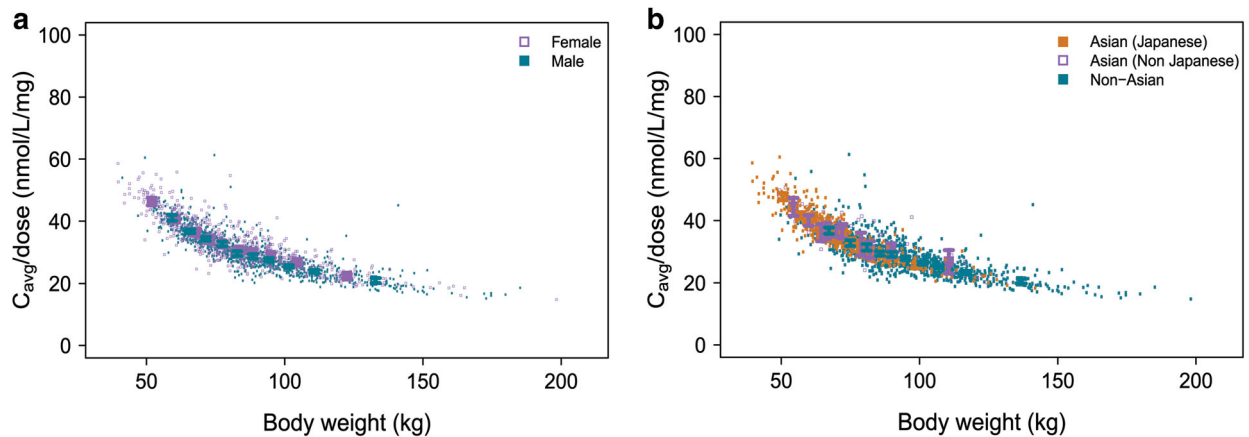


Fig. 4 Semaglutide exposure versus body weight. Data are dose-normalized individual average semaglutide concentrations (C_{avg}) versus baseline body weight (small rectangles)

and mean exposure estimates versus body weight presented in 10 quantiles by sex (a) or by ethnicity (b)

delayed more than 5 days should be skipped before resuming the planned dosing schedule. The full population pharmacokinetic model was used to generate semaglutide concentration profiles in a reference subject to predict exposure following missed or delayed dosing. If a dose was missed, the simulated profile showed that a 48% decrease in minimum concentration was expected before the next planned dose (Fig. 5a). A 5-day delayed dose should cause minimum semaglutide concentrations to be 37% lower and maximum concentrations to be 14% higher compared with the same subject at normal weekly steady state (Fig. 5b). In both cases, semaglutide concentrations will be close

to regular steady-state concentrations after 3 weeks.

DISCUSSION

The population pharmacokinetic model reported here successfully describes the pharmacokinetics of semaglutide, reproducing the values for CL/F and V/F of approximately 0.05 l/h and 12.5 l, respectively, obtained from clinical pharmacology trials with richly sampled PK profiles in subjects with T2DM (Novo Nordisk, data on file). The population pharmacokinetic analysis showed that the semaglutide plasma

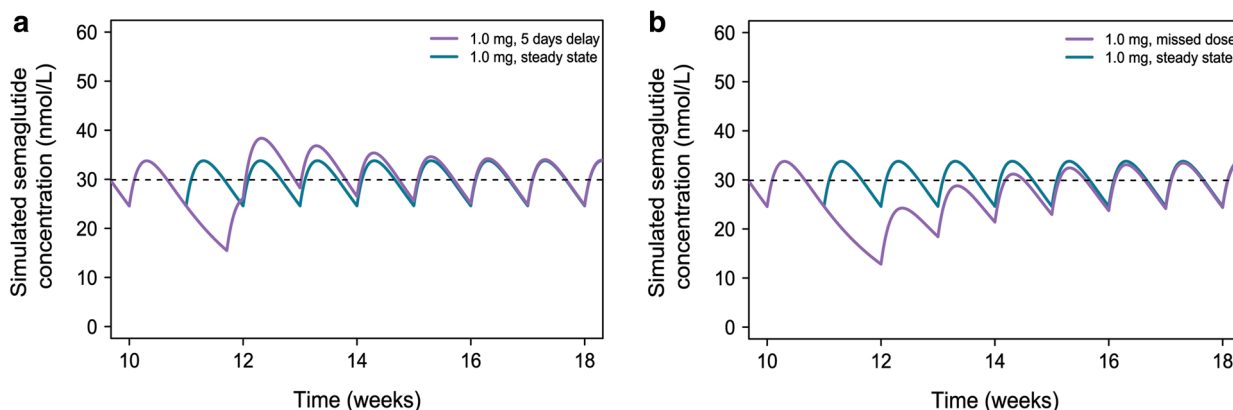


Fig. 5 Simulated semaglutide concentration profiles following missed or delayed doses. Data are simulated concentrations during once-weekly dosing at steady-state concentrations with one missed dose at week 11 (a) and for a dose with a delay of 5 days at week 11 (b) compared

with a steady-state profile for semaglutide dosed at weekly intervals. Simulations are for a reference subject profile (non-Hispanic or Latino, white female < 65 years, with a body weight of 85 kg, with normal renal function, and dosed in the abdomen with semaglutide 1.0 mg)

concentration is mainly dependent on a single baseline characteristic of a patient. The covariates of sex, age, race, ethnicity, and renal function did not have relevant effects on the predicted exposure; body weight was the only covariate of importance for exposure. Different injection sites were not of importance for exposure, suggesting that patients can use them interchangeably if desired. Semaglutide was similar to other GLP-1RAs regarding covariate-dependent exposure [33–36]. In a dedicated clinical pharmacology study, renal impairment did not affect semaglutide exposure [37]. Within the dosing regimen (starting at a low dose and increasing the dose based on efficacy and tolerability), no dose adjustments are necessary.

There were also no changes in exposure over time (up to 104 weeks) or relevant exposure differences in subjects with antibody development.

The population pharmacokinetic model was a one-compartment model with first-order absorption and elimination. Investigations using data from clinical pharmacology trials with richly sampled PK profiles indicate that the one-compartment model adequately describes the PK of semaglutide (data not shown).

Covariate effects were limited to effects on clearance. Due to the sparse sampling, the data

did not support accurate estimates of individual volumes of distribution, which resulted in relatively high shrinkage for the volume. Covariate effects were not included for V/F, and k_a was fixed for the entire population. Because of the limited fluctuation in semaglutide concentration at steady state, this was not considered a serious limitation. All PK samples were assumed to be at steady state (with the exception of the observation at week 2 in SUSTAIN 6), and previous doses were assumed to be according to the study plan. Since the subject diary only included information for the previous two injections before sampling, any dosing deviations 2 weeks prior to sampling were not accounted for. Semaglutide has a $t_{1/2}$ of approximately 1 week and 75% of steady state is reached after 2 weekly doses of semaglutide. Consequently, any dose deviations prior to 2 weeks before sampling should have a limited effect on exposure, and this was therefore evaluated to have limited effect on the model.

Baseline body weight was included as a covariate, with the assumption that changes in body weight during the course of the trials would have a negligible effect on the exposure. Since semaglutide has a body weight-reducing effect [12], exposure to semaglutide may increase over the course of treatment. On average, subjects on semaglutide experienced an

approximately 5% weight loss [13–15, 19]. Based on the model estimates, this would lead to approximately 4% change in exposure, and a body weight loss of 10% would result in 7.8% increased exposure. These changes are below the day-to-day variability of ~ 10% and therefore not considered to be of importance.

In the model, data were assumed to be missing at random. The risk of bias caused by treatment discontinuation was evaluated to be minimal because few subjects were excluded because of missing PK samples.

SUSTAIN 6 was the only trial in the population pharmacokinetic model to include subjects with severe renal impairment ($n = 33$), and the covariate effect for severe renal impairment might therefore be confounded by a trial effect. The SUSTAIN 6 trial also included approximately equal numbers of subjects from each renal impairment group (including subjects with normal renal function), and a possible trial effect should therefore not exclusively affect the severe renal impairment covariate. In addition, a dedicated clinical pharmacology trial observed no clinically relevant effect of renal impairment on exposure after adjusting for covariates (age, sex, and body weight) [38]. This limitation was therefore considered to have a limited effect on the validity of the results for subjects with severe renal impairment.

CONCLUSIONS

The population pharmacokinetics of semaglutide have been characterized and were demonstrated to be predictable with limited variability following s.c. administration. From a PK point of view, no dose adjustment of semaglutide was shown to be required for different populations based on sex, age, race, ethnicity, or renal function. Choice of injection site had no significant effect on exposure and can therefore be chosen interchangeably. The two doses of semaglutide indicated dose proportionality. The only covariate of importance for the exposure of semaglutide in the population studied was body weight, which was inversely correlated with semaglutide exposure. Therefore, based on PK considerations, no dose adjustments of

semaglutide are necessary in different populations. This finding is to be further explored in an exposure-response analysis. The option of adjusting the dose from 0.5 mg to 1.0 mg remains.

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Compliance with Ethics Guidelines. All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or

national research committee and with the 1964 Helsinki Declaration and its later amendments and Good Clinical Practice [21, 22]. The trial protocols were approved by independent ethics committees and/or institutional review boards. All subjects provided written informed consent before initiation of any trial-related activities.

Data Availability. The data sets generated during and/or analyzed during the current study are not publicly available but NONMEM control streams may be made available from the corresponding author on reasonable request.

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REFERENCES

1. Collaboration NCDRF. Worldwide trends in diabetes since 1980: a pooled analysis of 751 population-based studies with 4.4 million participants. *Lancet*. 2016;387:1513–30.
2. International Diabetes Federation. IDF diabetes atlas. 7th ed. Brussels: International Diabetes Federation; 2015.
3. Giovannucci E, Harlan DM, Archer MC, Bergenstal RM, Gapstur SM, Habel LA, et al. Diabetes and cancer: a consensus report. *Diabetes Care*. 2010;33:1674–85.
4. Laakso M. Cardiovascular disease in type 2 diabetes from population to man to mechanisms: the Kelly West Award Lecture 2008. *Diabetes Care*. 2010;33:442–9.
5. Noto H, Tsujimoto T, Sasazuki T, Noda M. Significantly increased risk of cancer in patients with diabetes mellitus: a systematic review and meta-analysis. *Endocr Pract*. 2011;17:616–28.
6. Barone BB, Yeh HC, Snyder CF, Peairs KS, Stein KB, Derr RL, et al. Long-term all-cause mortality in cancer patients with preexisting diabetes mellitus: a systematic review and meta-analysis. *JAMA*. 2008;300:2754–64.
7. Wexler D, Grant R, Wittenberg E, Bosch J, Cagliero E, Delahanty L, et al. Correlates of health-related quality of life in type 2 diabetes. *Diabetologia*. 2006;49:1489–97.
8. Holst JJ, Vilsbøll T, Deacon CF. The incretin system and its role in type 2 diabetes mellitus. *Mol Cell Endocrinol*. 2009;297:127–36.
9. Kieffer TJ, Habener JF. The glucagon-like peptides. *Endocr Rev*. 1999;20:876–913.
10. Flint A, Raben A, Astrup A, Holst JJ. Glucagon-like peptide 1 promotes satiety and suppresses energy intake in humans. *J Clin Invest*. 1998;101:515–20.
11. Meier JJ. GLP-1 receptor agonists for individualized treatment of type 2 diabetes mellitus. *Nat Rev Endocrinol*. 2012;8:728–42.
12. Lau J, Bloch P, Schäffer L, Pettersson I, Spetzler J, Kofoed J, et al. Discovery of the once-weekly glucagon-like peptide-1 (GLP-1) analogue semaglutide. *J Med Chem*. 2015;58:7370–80.
13. Sorli CHS, Harashima SI, Tsoukas GM, Unger J, Karsbol JD, Hansen T, Bain SC. Efficacy and safety of once-weekly semaglutide monotherapy versus placebo in patients with type 2 diabetes (SUSTAIN 1): a double-blind, randomised, placebo-controlled, parallel-group, multinational, multicentre phase 3a trial. *Lancet Diabetes Endocrinol*. 2017;5:251–60.
14. Åhrén B, Comas LM, Kumar H, Sargin M, Derving Karsbøl J, Jacobsen SH, et al. Efficacy and safety of once-weekly semaglutide versus sitagliptin as an add-on to metformin, thiazolidinediones, or both, in patients with type 2 diabetes (SUSTAIN 2): a 56-week, double-blind, phase 3a, randomised trial. *Lancet Diabetes Endocrinol*. 2017;5:341–54.
15. Ahmann AJ, Capehorn M, Charpentier G, Dotta F, Henkel E, Lingvay I, et al. Efficacy and safety of once-weekly semaglutide versus exenatide ER in subjects with type 2 diabetes (SUSTAIN 3): A 56-week, open-label, randomized clinical trial. *Diabetes Care*. 2018;41:258–66.
16. Aroda VR, Bain SC, Cariou B, Piletic M, Rose L, Axelsen M, et al. Efficacy and safety of once-weekly semaglutide versus once-daily insulin glargine as add-on to metformin (with or without sulfonylureas) in insulin-naïve patients with type 2 diabetes (SUSTAIN 4): a randomised, open-label, parallel-group, multicentre, multinational, phase 3a trial. *Lancet Diabetes Endocrinol*. 2017;5:355–66.

17. Rodbard HW, Lingvay I, Reed J, de la Rosa R, Rose L, Sugimoto D, et al. Semaglutide added to basal insulin in type 2 diabetes (SUSTAIN 5): a randomized, controlled trial. *J Clin Endocrinol Metab.* 2018;103:2291–301.
18. Seino Y, Terauchi Y, Osonoi T, Yabe D, Abe N, Nishida T, et al. Safety and efficacy of semaglutide once weekly versus sitagliptin once daily, both as monotherapy in Japanese subjects with type 2 diabetes. *Diabetes Obes Metab.* 2018;20:378–88.
19. Marso SP, Bain SC, Consoli A, Eliaschewitz FG, Jódar E, Leiter LA, et al. Semaglutide and Cardiovascular Outcomes in Patients with Type 2 Diabetes. *N Engl J Med.* 2016;375:1834–44.
20. Kaku K, Yamada Y, Watada H, Abido A, Nishida T, Zacho J, Kiyosue A. Safety and efficacy of once-weekly semaglutide vs additional oral antidiabetic drugs in Japanese people with inadequately controlled type 2 diabetes: a randomized trial. *Diabetes Obes Metab.* 2018;20:1202–12.
21. World Medical Association. WMA Declaration of Helsinki—Ethical Principles for Medical Research Involving Human Subjects. Last amended by the 64th WMA General Assembly, Fortaleza, Brazil. 2013. <https://www.wma.net/policies-post/wma-declaration-of-helsinki-ethical-principles-for-medical-research-involving-human-subjects/>. Accessed 08 Mar 2018.
22. International Conference of Harmonisation. ICH Harmonised Tripartite Guideline E6(R1): Guideline for Good Clinical Practice. 1996. https://www.ich.org/fileadmin/Public_Web_Site/ICH_Products/Guidelines/Efficacy/E6/E6_R1_Guideline.pdf. Accessed 08 Mar 2018.
23. Nauck MA, Petrie JR, Sesti G, Mannucci E, Courrèges JP, Lindegaard ML, et al. A phase 2, randomized, dose-finding study of the novel once-weekly human GLP-1 analog, semaglutide, compared with placebo and open-label liraglutide in patients with type 2 diabetes. *Diabetes Care.* 2016;39:231–41.
24. Kapitzka C, Nosek L, Jensen L, Hartvig H, Jensen CB, Flint A. Semaglutide, a once-weekly human GLP-1 analog, does not reduce the bioavailability of the combined oral contraceptive, ethinylestradiol/levonorgestrel. *J Clin Pharmacol.* 2015;55:497–504.
25. Hu C, Zhang J, Zhou H. Confirmatory analysis for phase III population pharmacokinetics. *Pharm Stat.* 2011;10:14–26.
26. Marshall SF, Marshall S, Burghaus R, Cosson V, Cheung SY, Chenel M, et al. Good practices in model-informed drug discovery and development: practice, application, and documentation. *CPT Pharmacometrics Syst Pharmacol.* 2016;5:93–122.
27. Nguyen NT, Nguyen XM, Lane J, Wang P. Relationship between obesity and diabetes in a US adult population: findings from the National Health and Nutrition Examination Survey, 1999–2006. *Obes Surg.* 2011;21:351–5.
28. Gatineau M, Hancock C, Holman N, Outhwaite H, Oldridge L, Christie A, et al. Adult obesity and type 2 diabetes. In: England PH, editor. London: Public Health England; 2014.
29. Lindbom L, Pihlgren P, Jonsson EN. PsN-Toolkit—a collection of computer intensive statistical methods for non-linear mixed effect modeling using NONMEM. *Comput Methods Programs Biomed.* 2005;79:241–57.
30. Lindbom L, Ribbing J, Jonsson EN. Perl-speaks-NONMEM (PsN)—a Perl module for NONMEM related programming. *Comput Methods Progr Biomed.* 2004;75:85–94.
31. U.S. Food and Drug Administration, Center for Drug Evaluation and Research (CDER). Guidance for Industry. Population Pharmacokinetics. 1999. <https://www.fda.gov/downloads/drugs/guidances/UCM072137.pdf>. Accessed 08 Mar 2018.
32. European Medicines Agency, Committee for Medicinal Products for Human Use (CHMP). Guideline on reporting the results of population pharmacokinetic analyses (CHMP/EWP/185990/06). 21 June 2007. http://www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2009/09/WC500003067.pdf. Accessed 08 Mar 2018.
33. Overgaard RV, Petri KC, Jacobsen LV, Jensen CB. Liraglutide 3.0 mg for weight management: a population pharmacokinetic analysis. *Clin Pharmacokinet.* 2016;55:1413–22.
34. Jacobsen LV, Flint A, Olsen AK, Ingwersen SH. Liraglutide in type 2 diabetes mellitus: clinical pharmacokinetics and pharmacodynamics. *Clin Pharmacokinet.* 2016;55:657–72.
35. Geiser JS, Heathman MA, Cui X, Martin J, Loghin C, Chien JY, et al. Clinical pharmacokinetics of dulaglutide in patients with type 2 diabetes: analyses of data from clinical trials. *Clin Pharmacokinet.* 2015;55:625–34.
36. Cirincione B, Mager DE. Population pharmacokinetics of exenatide. *Pharmacokinetics.* 2016;83:517–26.
37. Jensen L, Kupčová V, Arold G, Pettersson J, Hjerpested JB. Pharmacokinetics and tolerability of semaglutide in people with hepatic impairment. *Diabetes Obes Metab.* 2018;20:998–1005.

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38. Marbury TC, Flint A, Jacobsen JB, Karsbøl JD, Lassetter K. Pharmacokinetics and tolerability of a single dose of semaglutide, a human glucagon-like peptide-1 analog, in subjects with and without renal impairment. *Clin Pharmacokinet.* 2017;56:1381–90.