Dose–exposure–IGF-I response of once-weekly somapacitan
in adults with GH deficiency

Supplementary Data
Supplementary methods

Bioanalysis
Somapacitan plasma concentrations were measured using a validated somapacitan specific luminescent oxygen channelling immunoassay developed by Novo Nordisk. The lower limit of quantification (LLOQ) was 0.500 ng/mL. The assay was validated according to the current guidelines for bioanalytical method validation and performed with a precision (% coefficient of variation [CV]) of 3.59 to 12.4% (21.5% at LLOQ). The accuracy (% relative error [RE]) was -7.46 to 2.84% across the full assay range. Insulin-like growth factor I (IGF-I) was analysed using commercially available assay kits (Immuno Diagnostic Systems immunoassay [ISYS] assay). The precision (%CV) was determined to be in the range of 2.7–3.3%.

Data sets and data cleaning
Patients with at least one pharmacokinetic (PK) measurement were included in the PK data set, and patients with at least one PK and IGF-I measurement were included in the PK/pharmacodynamic (PD) data set. A total of 333 patients were randomised to somapacitan in the phase 3 trials (Table 1), and 330 were included in the analyses (Table 2). Two patients were not included as they had no PK samples, and one patient was excluded during data cleaning due to a missing dosing diary.

A total of 4470 PK samples and 5137 IGF-I samples were obtained. During data cleaning, 106 (2.4%) PK and 259 (5.0%) IGF-I records were excluded, the majority of which were excluded due to incomplete or ambiguous dosing records or collection of samples later than 168 hours (7 days) after planned sampling time. No imputation was applied for missing PK and IGF-I data.
**Somapacitan population PK and PK/PD analysis methods**

The phase 3 PK/PD models were based on previously published models developed using phase 1 data (1). The PK of somapacitan was described by a one-compartment model with dual first- and zero-order absorption through a transit compartment and with saturable elimination, and the PK/PD model was an indirect response model with an additive maximum IGF-I production rate ($E_{max}$) effect relationship between somapacitan PK and IGF-I rate of production. Key PK parameters were related to the relative bioavailability (F), correlating with the total exposure per mg somapacitan. Key PD parameters were the baseline IGF-I production rate ($k_{in}$), correlating with the observed baseline IGF-I levels, and $E_{max}$, correlating with the maximum effect predicted with somapacitan. The PK model was fitted to estimate structural parameters, residual variability and inter-individual variability (IIV) equivalent to F. The PK/PD model was fitted to estimate structural parameters, residual variability and IIV on $k_{in}$ and $E_{max}$. Structural PK and PD parameters that were not identifiable with the sparse PK and IGF-I data sets obtained in phase 3 were fixed to those estimated with the phase 1 models based on full PK and IGF-I profiles (1).

**Covariate models for somapacitan PK and PK/PD**

Pre-defined covariates were tested in a stepwise manner for all parameters with IIV. Predefined covariates included body weight, sex (male, female without oral oestrogen, female with oral oestrogen), age group (>65, ≤65) and race (White/Other, Asian Japanese, Asian non-Japanese). Covariates significant at p<0.01 were retained in the final models (Supplementary Table 2). The reference profile was pre-specified as male, White, age <65 years and body weight of 85 kg. All pre-defined covariates significantly affected F and were included as covariates in the PK model. The following covariates significantly affected somapacitan PD parameters and were included in the PK/PD model: body weight, oral
oestrogen use and age on $k_{in}$, and body weight, sex, race (Asian non-Japanese) and age on
$E_{max}$.

In addition to pre-defined covariates, the following exploratory covariates were tested:
injection site (abdomen, thigh, unknown), growth hormone deficiency (GHD) onset
(education, adulthood), age group ($\leq 25$ years, $>25$ years), body mass index (BMI) group ($<25$
kg/m², 25–35 kg/m², 35–40 kg/m² and $>40$ kg/m²) and diabetes status (yes, no). No
exploratory covariates significantly affected PK or PD parameters of somapacitan.

All covariate values used in the models were baseline values. For the categorical covariate
variables, at least 20 patients were required in each category. Race groups with an insufficient
number of patients (Black or African American [n=4] and Hawaiian or Pacific Islander [n=1])
were pooled with the largest group (White) in the covariate analyses.

**Evaluation of somapacitan and somatropin models**

The final PK and PK/PD models were qualified by evaluation of the precision of fixed effect
parameters, the shrinkage of random effects and the goodness-of-fit by visual inspection of
model diagnostics plots, including prediction-corrected visual predictive checks (VPCs).

VPCs are shown in **Supplementary Fig. 1** for the somapacitan PK model (**Supplementary
Fig. 1A**), the somapacitan PK/PD model (**Supplementary Fig. 1B**) and the somatropin
PK/PD model (**Supplementary Fig. 1C**). The models reproduced the overall trends of the
phase 3 data, including the median and variability. Hence, the models provided accurate
prediction of the observed data.

Sensitivity analyses showed similar results when dividing age groups at 60 years ($>60$, $\leq 60$)
and 65 years ($>65$, $\leq 65$) in the PK and PK/PD models. Different age groups were used for
starting dose groups ($>60$, $\leq 60$) and for covariate age groups in the modelling analysis ($>65$,
$\leq 65$), due to a discrepancy between current clinical treatment guidelines (2) and the definition
of geriatric populations in regulatory guidelines (3).
Dose–exposure-IGF-I estimation based on final somapacitan PK and PK/PD models

Estimates for the individual dose–exposure–IGF-I-response pairs at maintenance dose levels after titration were constructed based on individual estimates for PK and PK/PD parameters from the final PK and PK/PD models. Fixed dose levels were defined as the median of doses registered for an individual after the last titration visit. Average exposure (C_{avg}) and average IGF-I response (IGF-I_{avg}) were estimated from the area under the curve (AUC) of individual predicted profiles during maintenance treatment for the fixed dose level. No imputation was performed for subjects not completing titration (n = 6).

The individual dose–exposure–IGF-I-response curves across the somapacitan dose range of 0.1–8 mg were estimated from the AUC of individual predicted profiles using the final PK and PK/PD models.

Somatropin PK/PD analysis methods

The somatropin PK/PD model was based on data from three phase 3 trials in patients with adult growth hormone deficiency (AGHD) (Tables 1 and 2) (4-6). The PK of somatropin was described by a one-compartment model with linear absorption and clearance, while somatropin PK/PD was described using an indirect response model with additive E_{max} effect relationship between PK and IGF-I, like the PK/PD model for somapacitan. Somatropin model development and dose–response analyses were performed similarly to the procedures described for somapacitan.

Software

R version 3.2.3 or above (R Foundation for Statistical Computing, Vienna, Austria) was used for data file processing, explorative data analysis, exposure-response analyses, for plotting and simulation. NONMEM version 7.3 (ICON Development Solutions, Ellicott City, MD,
USA) and PsN (version 4.6.0) was used for the population PK and PK/PD analysis and simulation.
| Country | Ethics committee                                                                                                                                                                                                 | Trial   |
|---------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|---------|
| AU      | 1. Bellberry human Research Ethics committee                                                                                                                                                                | REAL 1  |
|         | 2. Melbourne Health Human Research Ethics Committee                                                                                                                                                    | REAL 1  |
| DE      | 1. Landesamt für Gesundheit und Soziales Ethik-Kommission des Landes Berlin                                                                                                                                  | REAL 1, 2|
| DK      | 1. Regionshuset Viborg, De Videnskabsetiske Komiteer for Region Midtjylland                                                                                                                                   | REAL 2  |
| FR      | 1. CPP Ouest II, Maison de la Recherche Clinique                                                                                                                                                    | REAL 2  |
| IL      | 1. Souraski Helsinki Committee                                                                                                                                                                            | REAL 1  |
| IN      | 1. Institutional Ethics Committee, KPC Medical College & Hospital                                                                                                                                         | REAL 1  |
|         | 2. Institutional Ethics Committee of Topiwala National Medical College and BYL Nair Charitable Hospital G                                                                                               | REAL 1  |
|         | 3. Institutional Ethical Committee, Care Hospital, Care Convergence Centre, H.                                                                                                                            | REAL 1  |
|         | 4. Ethics Committee, Apollo Hospitals                                                                                                                                                                    | REAL 1  |
|         | 5. Institutional Ethics Committee, Amrita Institute of Medical and Research Centre                                                                                                                       | REAL 1  |
| JP      | 1. Hokkaido University Hospital IRB                                                                                                                                                                    | REAL 1  |
|         | 2. Tokyo Heart Center IRB                                                                                                                                                                                   | REAL 1  |
|         | 3. Chiba University Hospital IRB                                                                                                                                                                          | REAL 1, JP|
|         | 4. Toranomon Hospital IRB                                                                                                                  | REAL 1  |
|         | 5. Nippon Medical School Hospital IRB                                                                                                         | REAL 1, 2, JP|
|         | 6. Kitasato University Sagamihara IRB                                                                                                           | REAL 1, 2, JP|
|         | 7. Kyoto Medical Center IRB                                                                                                                      | REAL 1, 2|
|         | 8. Central IRB of Clinical Research Network Fukuoka                                                                                             | REAL 1  |
|         | 9. Yamagata University Hospital IRB                                                                                                              | REAL 1, JP|
|         | 10. Okayama University Hospital IRB                                                                                                             | REAL 1, JP|
|         | 11. Kagoshima University Hospital IRB                                                                                                           | REAL 1, JP|
|         | 12. Teikyo University Hospital IRB                                                                                                              | REAL 1, 2|
|         | 13. Yokohama Rosai Hospital IRB                                                                                                                 | REAL 1, JP|
|         | 14. IRB of Adachi Kyosai Hospital                                                                                                               | REAL 1, JP|
|         | 15. Kobe University Hospital IRB                                                                                                                 | REAL 2, JP|
16. IRB of Takahashi Clinic
17. Shimane University Hospital IRB
18. Institutional Review Board of National Hospital Organization Kyoto Medical Center
19. Fukuoka University Chikushi Hospital IRB
20. Osaka University Hospital IRB

LT 1. Lithuanian Bioethics Committee

LV 1. Ethics Committee for Clinical Research at Pauls Stradins Clinical University

Hospital Development Society

MY 1. Medical Research & Ethics Committee, National Institute of Health

PL 1. Komisja Bioetyczna przy Okręgowej Izbie Lekarskiej w Krakowie

RO 1. National Bioethics Committee for Medicines and Medical Devices

RU 1. Ethics Committee at Ministry of Health of the Russian Federation

2. Local Ethics Committee of Federal State Budget Institution Endocrinology Research Centre

3. Ethics Committee of the State Budget Educational Institutional of Higher Professional Education “Siberian State Medical University” of the Ministry of Healthcare of Russian Federation

4. Ethics Committe of the State Budget Educational Institutional “Kazan State Medical Academy of the Ministry of Healthcare of the Russian Federation”. The Local Ethics Committee was changed on 20-Oct-2016 to: Local Ethics Committee of the Limited Liability Company of the Research Medical Complex “Your Health”

5. Ethics Committee within SBEI of HPE “Novosibirsk State Medical University of Ministry of Healthcare”

6. Ethics Committe of State Federal-Funded Educational Institution of Higher Professional Training I.M. Sechenov First Moscow State Medical University of the Ministry of Health of the Russian Federation

SE 1. Regionala Etikprövningsnämnden i Göteborg

TR 1. Istanbul University Cerrahpaşa Medical Faculty Ethics Committee

UA 1. The Ethic Committee State Institution “Institute of endocrinology and metabolism named after V.P. Komisarenko of the Academy of Medical Science of Ukraine”
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3. Martin Cooper, Research & Development Royal Devon and Exeter Hospital
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9. Dr Christopher Counsell, UHB Research Governance Office Education Centre

US
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2. Columbia University Medical Center IRB
3. Baylor College of Medicine IRB
4. Copernicus Group IRB
5. UCLA Office of the Human Research Protection Program
6. Cleveland Clinic IRB
7. University of Pennsylvania Office of Regulatory Affairs
8. Washington University Human Research Protection Office
9. VA Nebraska-Western Iowa Healthcare System Subcommittee of Human Studies of the Research and Development Committee
10. Western Institutional Review Board
11. Michigan State University Office of Regulatory Affairs Human Research Protection Program
12. USC Health Sciences Campus Institutional Review Board
13. OHSU Research Integrity
14. UNMC Institutional Review Board
15. Johns Hopkins Medicine Institutional Review Board

ZA
1. Wits Human Research Ethics Committee (WHREC)
Ethics committees are included for all trial sites that randomised and/or screened subjects in REAL 1, REAL 2 or REAL JP.

IRB, institutional review board.

**Supplementary Table 2: Covariates included in the final PK and PK/PD models of somapacitan**

| PK parameter | Covariates included in the PK model | p-value | PK/PD parameter | Covariates included in the PK/PD model | p-value |
|--------------|--------------------------------------|---------|-----------------|----------------------------------------|---------|
| F            | Body weight                          | <0.0001 | k<sub>in</sub>   | Body weight                            | <0.0001 |
|              | Sex                                  | <0.0001 |                | Oral oestrogen intake                   | <0.0001 |
|              | Oral oestrogen intake                 | <0.0001 |                | Age                                    | 0.0089  |
|              | Race (Asian Japanese)                 | <0.0001 | E<sub>max</sub>  | Body weight                            | 0.0001  |
|              | Race (Asian non-Japanese)             | <0.0001 |                | Sex                                    | 0.0002  |
|              | Age                                  | 0.0006  |                | Race (Asian non-Japanese)               | 0.0005  |
|              |                                       |         |                | Age                                    | 0.0013  |

Age groups (>65, ≤65). Race groups (White/Other, Asian Japanese, Asian non-Japanese). White/Other was set as reference. p-values were derived from the maximum likelihood difference between models with and without the covariate. Race effects on PK were tested as a covariate with two levels (Asian Japanese and Asian Non-Japanese versus White). No statistically significant difference was observed for White/Other versus Asian Japanese on E<sub>max</sub>.

F, relative bioavailability; k<sub>in</sub>, baseline IGF-I production rate; E<sub>max</sub>, maximum IGF-I production rate; PD, pharmacodynamic; PK, pharmacokinetic.
### Supplementary Table 3: Demographics and characteristics for starting dose groups

| Category          | Group       | Patients ≤60 years | Patients >60 years | Females on oral oestrogen |
|-------------------|-------------|--------------------|--------------------|--------------------------|
| All               | N           | 188                | 70                 | 72                       |
| Gender (%)        | Female      | 37                 | 36                 | 100                      |
|                   | Male        | 63                 | 64                 | 0                        |
| Race (%)          | White/Other | 63                 | 53                 | 72                       |
|                   | Asian Japanese | 27              | 47                 | 14                       |
|                   | Asian Non-Japanese | 10            | 0                  | 14                       |
| Body weight (kg)  | Mean        | 77                 | 78                 | 67                       |
| Age (years)       | Mean        | 43                 | 67                 | 36                       |

### Supplementary Table 4: Overview of dose adjustments in the titration period of the phase 3 trials

| Titration group | Number of subjects (%) | Number of subjects needing 0 to 5 dose adjustments |
|-----------------|------------------------|---------------------------------------------------|
|                 |                        | 0      | 1     | 2     | 3     | 4\(^a\) | 5\(^b\) |
| Titrating up    | 186 (56%)              | 0      | 56 (17%) | 53 (16%) | 51 (15%) | 26 (8%) | 0         |
| Titrating down  | 39 (12%)               | 0      | 29 (9%)  | 7 (2%)   | 3 (1%)   | 0      | 0         |
| Maintaining     | 105 (32%)              | 0      | 103 (31%) | 2 (1%)\(^c\) | 0      | 0      | 0         |
| starting dose   | Total                   | 330 (100%) | 103 (31%) | 85 (26%) | 62 (19%) | 54 (16%) | 26 (8%) |

Number of dose adjustments are included for the first titration period on somapacitan for each patient.

\(^a\) Maximum number of dose adjustments allowed in REAL 1 and REAL 2.

\(^b\) Maximum number of dose adjustments allowed in REAL JP.

\(^c\) Two subjects titrated up or down but returned to starting dose level.
**Supplementary Table 5: Predicted somapacitan and somatropin doses for similar mean IGF-I\textsubscript{avg}**

| Mean IGF-I\textsubscript{avg} SDS | Somapacitan dose (mg/week) | Somatropin dose (mg/day) | Somapacitan (mg/week) vs somatropin (mg/day) ratio |
|----------------------------------|-----------------------------|--------------------------|--------------------------------------------------|
| 0.0                              | 2.0                         | 0.25                     | 8.1                                              |
| 0.6                              | 3.0                         | 0.37                     | 8.2                                              |
| 1.1                              | 4.0                         | 0.49                     | 8.2                                              |
| 1.5                              | 5.0                         | 0.61                     | 8.2                                              |
| 1.9                              | 6.0                         | 0.74                     | 8.1                                              |

Predicted somapacitan and somatropin doses for similar mean IGF-I\textsubscript{avg} SDS in the phase 3 somapacitan and somatropin populations, respectively.

Avg, average; IGF-I, insulin-like growth factor I; SDS standard deviation score.
Supplementary Fig. 1: Prediction-corrected visual predictive checks

A  Somapacitan concentrations after first dose

B  IGF-I concentrations after first somapacitan dose

C  IGF-I concentrations after first somatropin dose
Data are prediction-corrected medians with 5\textsuperscript{th} and 95\textsuperscript{th} percentiles (red and blue lines) of somapacitan concentrations after first dose using the somapacitan PK model (a), IGF-I concentrations after first somapacitan dose using the somapacitan PK/PD model (b) or IGF-I concentrations after first somatropin dose using the somatropin PK/PD model (c). Shaded areas are 95\% confidence intervals around the prediction corrected medians and 5\textsuperscript{th} and 95\textsuperscript{th} percentiles based on 1000 simulations with the somapacitan and somatropin models. IGF-I, insulin-like growth factor I; PD, pharmacodynamic; PK, pharmacokinetic.
References

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