TITLE PAGE

RETROSPECTIVE OBSERVATIONAL STUDY PROTOCOL

Protocol Title:	A retrospective observational study to evaluate the safety and effectiveness of Osilodrostat for the treatment of non-Cushing's disease Cushing's syndrome (LINC7 Study)
Protocol Number:	LCI699-RECAG-NI-0596
Protocol Version:	2.0
Protocol Date:	18 April 2023

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1 LIST OF ABBREVIATIONS

Abbreviation	Definition
ACTH	Adrenocorticotropic hormone
AE	Adverse Event
AESI	Adverse events of special interest
BL	Baseline
CD	Cushing's Disease
CRF	Case Report Form
CRH	Corticotropin-releasing hormone
CRO	Contract Research Organisation
CS	Cushing's Syndrome
CT	computed tomography
DCF	Data Clarification Form
DMP	Data Management Plan
DVS	Data Validation Specification
ECG	Electrocardiogram
eCRF	electronic case report form
EDC	Electronic Data Capture
EMA	European Medicines Agency
ENCePP	European Network of Centres for Pharmacoepidemiology and Pharmacovigilance
EU	European Union
FSH	Follicle Stimulating Hormone
GPP	Good Pharmacoepidemiology Practices
HbA1C	Glycated Haemoglobin
HCP	Health Care Providers
IEC	Independent Ethics Committee
IRB	Institutional Review Board
ITT	Intention to Treat
LH	Luteinizing Hormone
LTF	Loss to Follow-Up
MedDRA	Medical Dictionary for Regulatory Activities
NOL	No Objection Letter
PT	Preferred Term
QA	Quality Assurance
SAP	Statistical Analysis Plan
SD	Standard Deviation

SMP	Site Management Plan
sNDA	Supplemental New Drug Application
SOC	System Organ Class
TEAE	Treatment-emergent adverse events
TFL	Tables, Figures, Listings
UFC	Urinary free cortisol
ULN	Upper Limit Normal
WK	Week

2 RESPONSIBLE PARTIES

Sponsor	Recordati AG Rare Disease Branch Uferstrasse, 90 4057 Basel Ph. +41 61 205 61 00 Fax +41 61 205 61 39
Contract Research Organization (CRO)	PAREXEL International (IRL) Limited 70 Sir John Rogerson's Quay Dublin 2 Ireland
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3 PROTOCOL SIGNATURE PAGE (FOR SPONSOR)

PROTOCOL TITLE: A retrospective observational study to evaluate the safety and effectiveness of osilodrostat for the treatment of non-Cushing's disease Cushing's syndrome

PROTOCOL NUMBER: LCI699-RECAG-NI-0596

PROTOCOL VERSION: 2.0

Signature of Sponsor's authorized representative(s):

Alberto M. PEDRONCELLI, MD, PhD
Clinical Development and Medical Affairs Head

PPD

Name Signature Date

STATEMENT OF COMPLIANCE – INVESTIGATOR SIGNATURE PAGE

This study titled "*A retrospective observational study to evaluate the safety and effectiveness of Osilodrostat for the treatment of non-Cushing's disease Cushing's syndrome"* (Protocol Code: LCI699-RECAG-NI-0596)

will be conducted in accordance with International Conference on Harmonisation Good Clinical Practice (ICH GCP), the ethical principles that have their origin in the Declaration of Helsinki and the Code of Federal Regulations on the Protection of Human Subjects (45 CFR Part 46). The Principal Investigator will assure that no deviation from, or changes to the protocol will take place without prior agreement from the sponsor. All personnel involved in the conduct of this study have completed Human Subjects Protection and ICH GCP Training. Also, the French data authority has been notified about this study documents and no-objection letter.

I agree to ensure that all staff members involved in the conduct of this study are informed about their obligations in meeting the above commitments.

Principal Investigator: _____ _____

Signature Date

Name: _____

Address: _____

_____

_____

4 SYNOPSIS

Protocol Title:	A retrospective observational study to evaluate the safety and effectiveness of osilodrostat for the treatment of non-Cushing's disease Cushing's syndrome (LINC7 Study)
Protocol Number:	LCI699-RECAG-NI-0596
Protocol Version:	2.0
Protocol Date:	18 April 2023
Sponsor:	Recordati AG
Study Centre(s):	13 sites in France
Rationale and Background:	Osilodrostat-an orphan medicine-was approved by the European Medicnes Agency (EMA) on 9 January 2020 for the treatment of endogenous Cushing's Syndrome (CS) in adults, and by the United States (US) Food Drug Administration (FDA) on 6 March 2020 for the treatment of adult patients with Cushing's disease (CD) for whom pituitary surgery is not an option or has not been curative. Prior to approval and commercialization of osilodrostat in the european union (EU), patients with CS had already been initiated on osilodrostat under the French Autorisation Temporaire d'Utilisation (ATU) programme. Recordati proposes an FDA label expansion for osilodrostat to include all forms of CS. This study therefore intends to evaluate the safety and effectiveness of osilodrostat for the treatment of non-CD CS patients in support of an FDA supplemental New Drug Application (sNDA).
Research Question and Objective(s):	What is the effectiveness and safety of osilodrostat in non-CD CS patients treated in a real-world setting?
Study Design:	Retrospective, observational single-cohort study with up to 36 months of retrospective real-world data review period.

Population:	Patients treated with osilodrostat for CS under 1. The 2019 French Autorisation Temporaire d'Utilisation (ATU), or 2. routine clinical practice after commercialization of osilodrostat in France in June 2020
Data Sources:	Patients' medical records
Data Collection Method	Electronic Data Capture (EDC)
Study Size:	Approximately 100 patients
Data Analysis:	Descriptive

5 AMENDMENTS AND UPDATES

Amendment 1

Overall Rationale for the Amendment: This amendment was initiated in response to FDA Advice/Information Request letter dated 13 February 2023. In addition, other updates and clarifications have been made for consistency throughout the protocol and to ensure alignment with regulatory authority requirements.

Summary of changes are presented below. All underlined and bold texts denote new texts while strikethrough text is the deleted text in this Protocol Amendment 1.

Section # and Title	Description of Change	Brief Rationale
Section 4: Synopsis	**Study centre(s):** **Deleted text:** Approximately ~~20~~ sites in France **New text:** **13** sites in France **Study size:** **Deleted text:** ~~A minimum of 40-50~~ patients **New text:** Approximately **100** patients	Study centre(s) and study size are updated.
Section 7: Table-1 Design elements (New Secondary Endpoint)	**New text:** **Proportion of patients with mUFC ≤ ULN at weeks 24, 36 and every 12 weeks thereafter.**	To include "the proportion of patients with mUFC ≤ ULN at Weeks 24, 36, and every 12 weeks thereafter" as a secondary endpoint, to assess the long-term effects of osilodrostat in patients with non-Cushing's disease Cushing's syndrome.
Section 7: Table-1 Design elements (Secondary End point)	**Deleted text:** Change from baseline for biochemical variables to Week 4, 8, 12, ~~20, 28~~, 36, and to every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation **New text:** Change from baseline for biochemical variables to Week 4, 8, 12, **18, 24,** 36, and to every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation	To amend the protocol to replace study assessment visits.

Section # and Title	Description of Change	Brief Rationale
Section 7: Table-1 Design elements (Secondary Endpoint)	**Deleted text:** Actual and percent Change in BMI, ECG, blood pressure, metabolic, biochemical, and hormone levels from baseline for clinical outcomes to Week 4, 8, 12, ~~20, 28,~~ 36, and to every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation **New text:** Actual and percent Change in BMI, ECG, blood pressure, metabolic, biochemical, and hormone levels from baseline for clinical outcomes to Week 4, 8, 12, **18, 24,** 36, and to every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation	To amend the protocol to replace study assessment visits.
Section 7: Table-1 Design elements (Exploratory Endpoint)	**Deleted text:** Proportion of patients with normal late night salivary cortisol at Week 4, 8, 12, ~~20, 28~~, 36, and at every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation **New text:** Proportion of patients with normal late night salivary cortisol at Week 4, 8, 12, **18, 24**, 36, and at every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation	To amend the protocol to replace study assessment visits.
Section 7: Table 1: Design elements	**New text:** **Comprehensive metabolic panel (CMP), Complete blood count (CBC)**	Study variables made consistent across protocol

Section # and Title	Description of Change	Brief Rationale
Section 8.1: Figure 1: Study Schematic	**Updated with new figure**	Updated last patient treatment start date according to actual study start date, which is actual first study site initiation visit date and replaced study assessment visits.
Section 8.1: Study Design	**Deleted text:** Clinical practice medical records will be reviewed for all eligible patients who received osilodrostat between April 2019 and the study start date ~~(approximately Q4 2022)~~ at study sites in France. **New text:** Clinical practice medical records will be reviewed for all eligible patients who received osilodrostat between April 2019 and the study start date **(16 December 2022)** at study sites in France. **The study start date was the date the first site was initiated in the study, which was on 16 December 2022.** All patients treated with osilodrostat between April 2019 and the study start date **(16 December 2022)** will be enrolled in the study. A no-objection letter (NOL) will be requested from the patients at enrolment unless the patient has previously signed a general consent for research.	Updated last patient treatment start date according to actual study start date, which is actual first study site initiation visit date.

Section # and Title	Description of Change	Brief Rationale
Section 8.1: Study Design,	**New text:** **Patients who have previously signed a general consent for research, either alive or deceased, can be enrolled with immediate effect. If no general consent was signed, a signed no-objection letter (NOL) will be requested from patients who are still alive. Some patients might not send the NOL back to the investigator. In such instance, the investigator can document a tacit approval from the patient if there is no response after a month of the first shipment of the NOL and after 3 documented contact attempts. For deceased patients since their past treatment with osilodrostat, they can also be enrolled after the investigator has checked that the deceased patient did not leave any written document stating refusal to participate in any scientific research.**	To clarify NOL/tacit approval process.
Section 8.1: Study Design and	**<u>New text:</u>** **The only patients excluded from the primary efficacy analyses will be the ones who started treatment with osilodrostat less than 12 weeks prior to the end of the abstraction period, and are still treated with osilodrostat at the end of the abstraction period. These patients will be included only in the analyses of the secondary and exploratory endpoints including safety. The patients who discontinued treatment for any reason or were lost-to-follow up prior to 12 weeks will be considered non-responders for the primary endpoint.**	To clarify patients who discontinued treatment prior to 12 weeks for any reason or were lost-to-follow up will be considered non-responders for the primary endpoint.
Section 8.1: Table 2: Data collection schedule	• **Replaced Week 20 +/-4 weeks with Week 18+/- 2 weeks** • **Replaced Week 28 +/-4 weeks with Week 24 +/-4 weeks**	To amend the protocol to replace study assessment visits.

Section # and Title	Description of Change	Brief Rationale
Section 8.1: Table 2: Data collection schedule	**Deleted Assessment:** **Comorbidities**	Form to capture co-morbidities no longer present. The information whether a Medical history (MH)/Adverse Event (AE) is related to CS is enabled.
Section 8.1: Table 2: Data collection schedule	**New Assessment:** **Prior medications**	Updated prior medication collection period
Section 8.1: Table 2: Data collection schedule (footnote)	**Deleted footnote-b text:** ~~Any assessments occurred out of the tolerated time window will be collected as Unscheduled visits.~~ **New foot note-b text:** **The assessments which occurred closest to each defined visit should be recorded in the CRF, even if these assessments occurred on different dates.**	To clarify that there is no requirement to collect assessments out of tolerated time-window as unscheduled since there are no planned analysis with this data.
Section 8.1: Table 2: Data collection schedule (footnote)	**Deleted footnote-c text:** ~~A no-objection letter (NOL) will be requested from the patients at enrolment unless~~ the patient has previously signed a general consent for research. **New foot note-c text:** **Either** the patient has previously signed a general consent for research, **or a signed no-objection letter (NOL) will be requested from the patient if still alive. If patient does not send the NOL back to the investigator, the investigator can document a tacit approval from the patient if there is no response after a month of the first shipment of the NOL and after 3 documented contact attempts. For a deceased patient since his/her past treatment with osilodrostat, access after the investigator has checked that this deceased patient did not leave any written document stating his/her refusal to participate in any scientific research.**	To clarify conditions for getting access to patient's medical data before enrollment

Section # and Title	Description of Change	Brief Rationale
Section 8.1: Table 2: Data collection schedule (footnote)	**New foot note-e text:** **All prior medications taken within 6 months prior to the first dose of Osilodrostat must be collected.**	Updated prior medication collection period
Section 8.1: Table 2: Data collection schedule (footnote)	**New foot note-f text:** **Baseline mean urinary free cortisol (mUFC) data can be collected within 1 month prior to osilodrostat initiation.**	To clarify previous data collection period for mUFC
Section 8.1: Table 2: Data collection schedule (footnote)	**New foot note-g text:** **Footnote-a is not applicable to the following assessments: prior medications, concomitant medications, medical and surgical history, and mean urinary free cortisol.**	To clarify foot note-a is not applicable to a few assessments
Section 8.1: Table 2: Data collection schedule (footnote)	**New foot note-h text:** **All available medical history and surgical history related to CS must be collected. Medical and surgical history not related to CS that occurred within 6 months prior to the first dose of Osilodrostat must be collected.**	To clarify collection period for medical and surgical history related or not to CS
Section 8.1: Table 2: Data collection schedule (footnote)	**New foot note-$ text:** **Lipid variables include Total Cholesterol, HDL, LDL, and Triglycerides**	To clarify the lipid variables that will be captured.

Section # and Title	Description of Change	Brief Rationale
Section 8.2: Setting	**Deleted text:** The study population will consist of patients with non-CD CS. ~~They must be able to provide electronically or manually a signed NOL and~~ meet all the inclusion criteria and none of the exclusion criteria. **New text:** The study population will consist of patients with non-CD CS **who** meet all the inclusion criteria and none of the exclusion criteria. **The authorization for getting access to the medical data of the patients must also be ensured, through the existence of a signed general consent, or requesting a signed NOL, or fulfilling the conditions of a tacit approval or no written document stating they refused any participation to scientific research.**	To clarify conditions for getting access to patients medical data.
Section 8.2.1: Inclusion Criteria	**New text:** Patients treated with osilodrostat between April 2019 and the study start date **(16 December 2022)** as part of ATU programme or commercialisation.	Updated last patient treatment start date according to actual study start date, which is actual first study site initiation visit date.
Section 8.2.2: Exclusion Criteria 1	**New text:** Patients who participated in **an interventional** clinical trial anytime during the study period.	To clarify that patients who participated in an interventional clinical trial cannot be enrolled.

Section # and Title	Description of Change	Brief Rationale
Section 8.2.3: Patient Enrolment	**Deleted text:** Site policies and local regulations regarding ~~patient consent (NOL)~~ will be followed. **New text:** Site policies and local regulations regarding **the access to their medical records** will be followed All eligible patients identified at a site between April 2019 and study start date **(16 December 2022)** and consenting to be part of the study will be included.	Updated last patient treatment start date according to actual study start date, which is actual first study site initiation visit date. To clarify that the access to patient's medical records will be as per local regulations.
Section 8.3.2: secondary Variables, Section 8.7.7: Demographics and Baseline Disease Characteristics Table 2: Data collection schedule	**Deleted text:** Removed race and ethnicity details.	Race/ethnicity are not collected due to local regulations.
Section 8.3.2: Secondary Variables	**Deleted text:** ~~Co-morbidities (obesity, hypertension, osteopenia/osteoporosis, diabetes, impaired glucose tolerance, dyslipidaemia, poor wound healing, recurrent infections, oligomenorrhea or infertility, thromboembolic disease, and cardiovascular, neurologic, and psychiatric manifestations).~~	Form to capture co-morbidities no longer present. The information whether a MH/AE is related to CS is enabled.

Section # and Title	Description of Change	Brief Rationale
Section 8.3.2: Secondary Variables	**Deleted text:** ~~Late night salivary cortisol.~~ ~~Laboratory investigations (CMP, CBC, lipids, HBA1c).~~ **New text:** Morning serum cortisol **and mUFC.** Laboratory **abnormalities (sodium, potassium, calcium levels, CO_2, glucose, Glycated hemoglobin (HBA1c) and lipids.** **Comprehensive metabolic panel (CMP) and Complete blood count (CBC).**	Study variables made consistent across protocol. Removed late night salivary cortisol as it is not a secondary endpoint but exploratory endpoint.
Section 8.7.3: Analysis Population	**New text:** The ITT population consists of all enrolled participants who have met all inclusion/exclusion criteria and have received osilodrostat treatment for non-CD CS, **with a potential follow-up** of at least 12 weeks. **The only patients excluded from the primary efficacy analyses will be the ones who started treatment with osilodrostat less than 12 weeks prior to the end of the abstraction period, and are still treated with osilodrostat at the end of the abstraction period. These patients will be included only in the analyses of the secondary and exploratory endpoints including safety.**	To clarify all patients enrolled in the study are included in the Intention to Treat (ITT) population, instead of only patients who have received osilodrostat with at least 12 weeks follow up.

Section # and Title	Description of Change	Brief Rationale
Section 8.7.3: Analysis Population	**Deleted text:** The ITT population consists of all participants who ~~have signed a general consent or the No-objection letter~~, have met all inclusion/exclusion criteria and have received osilodrostat treatment for non-CD CS, with at least 12 week ~~follow up~~. **New text:** The ITT population consists of all **enrolled** participants who have met all inclusion/exclusion criteria and have received osilodrostat treatment for non-CD CS, with a potential follow-up of at least 12 weeks.	To clarify about patients included in the ITT population.
Section 8.7.4: Control of Bias and Confounding	**New text:** Investigators will include all patients treated with osilodrostat between April 2019 and the study start date **(16 December 2022)**; to avoid selection bias in participant recruitment, all eligible patients will be included in the study.	Updated last patient treatment start date according to actual study start date, which is actual first study site initiation visit date.
Section 8.7.8: Analysis of Primary Endpoint	**New text:** **The patients who discontinued treatment for any reason or were lost-to-follow up prior to 12 weeks will be considered non-responders for the primary endpoint**. **The patients who started treatment with osilodrostat less than 12 weeks prior to the end of the abstraction period, and are still treated with osilodrostat at the end of the abstraction period will be excluded from the analysis.**	To clarify patients who discontinued treatment for any reason or were lost-to-follow up prior to 12 weeks will be considered non-responders for the primary endpoint and those treated for less than 12 weeks but were still under treatment at the time of the end of the abstraction period should be excluded from the primary analysis.

Section # and Title	Description of Change	Brief Rationale
Section 8.7.9: Analysis of Secondary Endpoints,	**Deleted text:** Changes from baseline for biochemical variables and clinical outcomes to Week 4, 8, 12, ~~20, 28,~~ 36, and to every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation will be summarised descriptively and provided as listing. **New text:** Changes from baseline for biochemical variables and clinical outcomes to Week 4, 8, 12, **18, 24**, 36, and to every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation will be summarised descriptively and provided as listing.	To amend the protocol to replace study assessment visits.

Section # and Title	Description of Change	Brief Rationale
Section 8.7.10: Analysis of Exploratory Endpoints	**Deleted text:** Late night salivary cortisol observed values at baseline and changes from baseline to Week 4, 8, 12, ~~20, 28~~, 36 and to every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation will be summarised descriptively and provided as listing. Where applicable, proportion of patients with normal and/abnormal late night salivary cortisol at Week 4, 8, 12, ~~20, 28~~, 30, 36 and at every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation. **New text:** Late night salivary cortisol observed values at baseline and changes from baseline to Week 4, 8, 12, **18, 24**, 36 and to every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation will be summarised descriptively and provided as listing. Where applicable, proportion of patients with normal and/abnormal late night salivary cortisol at Week 4, 8, 12, **18, 24,** 30, 36 and at every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation.	To amend the protocol to replace study assessment visits.

Section # and Title	Description of Change	Brief Rationale
Section 8.7.12: Stratified Analysis	**New text:** **The number and proportion of patients with mean urinary free cortisol (mUFC) ≤ULN at 12 weeks will be stratified if they received or not other therapies for Cushing's syndrome (regardless of the type of therapy) and will be summarised descriptively along with associated 95% CI. Differences in the proportions and 95% CI of the different groups will be evaluated. Further details will be described in the final SAP.**	To specify how to handle those patients who are on other therapies for Cushing's syndrome and who may confound the interpretation of the osilodrostat-related safety and efficacy results of the study.
Section 9.2: Informed Consent	**Deleted text:** ~~However, a NOL for study participation has to be signed from the potential patients~~ **New text:** **The authorization for getting access to the medical data of the patients must also be ensured, through the existence of a signed general consent, or requesting a signed NOL, or fulfilling the conditions of a tacit approval or no written document stating they refused any participation to scientific research.**	To clarify conditions for getting access to patients' medical data.
Section 9.4: Confidentiality	**Deleted text:** Documents not to be submitted to Parexel that identify the patient ~~(e.g., the signed NOL)~~ must be maintained in confidence by the investigator.	To clarify that documents that could identify the patient should not be submitted to Parexel.
Section 10: Management and reporting of adverse events/adverse drug reactions	**New text:** Being a retrospective chart review, it does not require active reporting of AEs/ADRs **to regulatory authorities**.	To clarify that reporting of AEs/ADRs refers to regulatory authorities, and not to investigator reporting responsibilities.

6 . BACKGROUND AND RATIONALE

Endogenous Cushing Syndrome (CS) is a rare endocrine disorder characterised by sustained elevated cortisol levels, posing risk of multiple comorbidities leading to cardiovascular diseases and mortality. Cushing's disease (CD) represents the most frequent cause of CS in 65-80% of cases. Ectopic Adrenocorticotropic hormone (ACTH)-secreting tumours (including neuroendocrine tumours) correspond to 10-15% of CS cases but may be underdiagnosed. Adrenal adenomas and carcinomas are the cause of ~20% of CS cases, and the <10% remaining cases are caused by rarer diseases such as bilateral macronodular adrenal hyperplasia, primary pigmented nodular adrenocortical disease, or ectopic Corticotropin-releasing hormone (CRH) production.

Endogenous CS is characterised by chronic hypercortisolism, resulting in metabolic dysregulation and comorbidities. Manifestations may be overt or subtle depending on severity of hypercortisolism. Comorbidities include menstrual irregularities and hirsutism in women, dermatological problems with striae, bruising, atrophy, hyperpigmentation, and fungal infections, obesity with moon facies, enlarged supraclavicular fat pads, and dorsal fat pad, glucose intolerance and diabetes, sleep apnoea, cardiovascular disease, venous thromboembolic events, bone loss, neuropsychiatric disease, and potentially severe infections. Whereas untreated Cushing's syndrome had a 50% five-year mortality rate, treatment to control cortisol production should substantially mitigate the risk of death due to hypercortisolism (Nieman et al. 2015).

Endogenous CS is a rare disease. Estimates of regional incidence have been limited and vary by aetiology and possibly methodology. CD is the most common cause of endogenous CS, with an estimated incidence in Europe of up to 2.4 per million and in the US of up to 7.6 per million depending on the series (Etxabe and Vazquez 1994; Broder et al. 2015; Tabarin et al. 2022). Adrenal tumours represent ~20% of CS cases (but may be higher in some countries such as Japan (Tanaka et al. 2020) and Italy (Invitti et al. 1999). Ectopic ACTH secretion from neuroendocrine tumours represent 10-15% of known CS cases. Different diseases causing CS manifest at different ages, i.e., smoking-related non-small lung cancer increases after the age of 50 years, neuroendocrine (carcinoid) tumours

occur earlier in adulthood, and adrenal tumours have bimodal age peaks (1st decade and 40-50 years old), whereas the peak incidence for CD is in women 25-45 years old (Bertagna and Orth 1981; Luton et al. 1990). CD occurs approximately 3 times than in men, women have a 4-5 times greater incidence of adrenal tumour causing CS, and neuroendocrine tumours causing CS are similar in incidence between the sexes (Carpenter 1988; Nieman et al. 2015). Heterogeneity in the cause and populations affected by non-CS CS, support the need of a dedicated efficacy and safety study focusing on non-CD CS population. Patients with CS have increased mortality, which applies both to patients with pituitary-dependent CS and adrenal-dependent CS, as well as patients with ectopic CS who have the worst prognosis (Ragnarsson et al. 2019).

The Endocrine Society guidelines (Nieman et al. 2015) recommend normalisation of cortisol in patients with overt Cushing's syndrome to mitigate signs, symptoms and comorbidities. Although first line of treatment for endogenous CS is often surgery or other disease-specific treatment (such as chemotherapy for a neuroendocrine tumour), patients often demonstrate the need for secondary treatments such pharmacotherapy because of persistence or recurrence, lack of surgical candidacy, and poor success rates of surgery other than bilateral adrenalectomy. Bilateral adrenalectomy ultimately may be required for control of unremittent disease in surgical candidates, resulting in permanent adrenal insufficiency.

Pasireotide is a second-generation somatostatin receptor ligand indicated for the medical treatment of Cushing's disease, suppresses corticotroph ACTH secretion that ultimately decreases cortisol production in the adrenals. Cabergoline has also historically been used without regulatory approval to treat Cushing's disease by inhibiting ACTH secretion at the pituitary level.

Mifepristone is a glucocorticoid receptor antagonist approved in the US for the treatment of hyperglycaemia secondary to hypercortisolism in adult patients with endogenous Cushing's syndrome who have type 2 diabetes mellitus or glucose intolerance.

Steroidogenesis inhibitors are often preferred, but use may be limited by availability, efficacy, compliance, or toxicity. Ketoconazole and metyrapone are not licenced in the United States: they tend to have a short onset of action and half-lives; metyrapone is

often a preferred class agent in Europe. Etomidate is also a steroidogenesis inhibitor with a rapid onset of action indicated for induction of general anaesthesia, requiring intensive monitoring by trained personnel. Mitotane is approved as an adrenolytic agent for adrenal cancer and has a relatively slow onset and long half-life.

Osilodrostat is a potent oral 11β-hydroxylase inhibitor, blocking the cortisol synthesis process. The LINC-3 (Pivonello et al. 2020) demonstrated its rapid efficacy and sustained control of mean 24-hour urinary free cortisol (UFC) alongside improvements in clinical signs of hypercortisolism. Also, it was well-tolerated by patients.

LINC 3 was a 4-period prospective, multicentre, open-label, phase III study with a double-blind randomised withdrawal period, which studied osilodrostat in 137 adults with confirmed persistent or de novo CD. Participants were initiated and treated to target mean UFC (mUFC) with open-label osilodrostat during Weeks 1 to 12, continued on a therapeutic dose of osilodrostat during weeks 13-24; eligible patients were randomised to either continue osilodrostat or switch to matching-placebo at Week 26 until Week 34, and then given open-label osilodrostat from Week 34 until study end at Week 48 whereas non—eligible patients continued open-label osilodrostat until the end of the core phase. At Week 34, maintenance of complete response was found in more patients in the osilodrostat group compared with the placebo group (31 [86%] vs 10 [29%]; odds ratio 13·7 [95% CI 3·7–53·4]; p<0·0001). At Week 24, The proportion of patients with normal mUFC at week 24 is 93/137 (67.9) (CI (59.37, 75.60). Nausea (57/137 [42%]), headache (46/137 [34%]), fatigue (39/137 [28%]), and adrenal insufficiency (38/137 [28%]) were the most common adverse events (AEs) (i.e., occurred in >25% of participants). Hypocortisolism-related AE were observed in (51%; n=70) and adrenal hormone precursors-related AE in (42%; n=58) of study patients. Also, other adverse events included anemia (8%), hypokalaemia (18 [13%]) and hypertension (17 [12%]). Non-serious cardiac adverse events included QT interval prolongation in five (4%) patients and pituitary adverse events included pituitary tumour enlargement, pituitary tumour volume increase, increased adenoma size, or pituitary tumour growth in four (3%) patients. This trial demonstrated adequate safety and efficacy of osilodrostat in patients with CD (Pivonello et al. 2020).

LINC-4 was a multinational phase III study of osilodrostat which assessed 73 adults with CD, with an upfront 12-week randomised, double-blind, placebo-controlled period followed by 36 weeks of open-label treatment. The LINC-4 study demonstrated that about 9 times more patients treated with osilodrostat (77%) compared with placebo (8%) achieved mUFC ≤ upper limit of normal (ULN) at Week 12 (OR 43.4; 95% CI 7.1-343.2; P<0.0001). The study showed that 80.8% of patients – including both patients initially randomised to osilodrostat or to placebo and then switched to active osilodrostat at Week 12 - had normal mUFC at week 36 with a maintained benefit at the end of treatment. During the overall study period, the most common AEs reported were consistent with the ones reported in LINC3 with the exception of adrenal insufficiency that was less frequently observed in LINC4 than in LINC3. The most common AEs were arthralgia, decreased appetite, nausea, and fatigue (Gadelha et al. 2022).

Additionally, a phase II, single-arm, open-label, dose-titration, multicentre study (Tanaka et al. 2020) evaluating osilodrostat in Japanese patients with non-CD CS, demonstrated a reduction in mUFC in all patients with endogenous CS other than CD, regardless of disease type with a safety profile that was consistent with that reported previously in CD patients. Of the nine patients enrolled in the study, seven completed the 12-week core treatment period and two discontinued at or prior to Week 12 due to AEs. Of the seven patients who completed 12 weeks of study treatment, two completed 48 weeks of study treatment. Median osilodrostat exposure was 12 weeks. Median (range) average dose including dose interruption (0 mg/day) was 2.143 (1.16–7.54) mg/day. Median (range, population) percentage change in mUFC was –94.47% (–99.0% to –52.6%, n=7) at Week 12. At Week 12, 6/9 patients were complete responders (mUFC ≤ULN) and 1/9 was a partial responder (mUFC >ULN but decreased by ≥50% from baseline). Most frequent AEs were adrenal insufficiency (n=7), gamma-glutamyl transferase increase, malaise, and nasopharyngitis (n=3 each). Serious AEs were seen in four patients. No deaths occurred in this study.

Proven clinical evidence from such previous studies (Pivonello et al. 2020; Gadelha et al. 2022) have made osilodrostat an effective orphan medicine with regulatory approval of its use in Europe and Japan for the treatment of endogenous CS and in the US for CD.

Prior to approval and commercialization of osilodrostat in the EU, patients with CS had already been initiated on osilodrostat within the French Autorisation Temporaire d'Utilisation (ATU) scheme as of April 2019.

Recordati proposes an FDA label expansion for osilodrostat to include all forms of endogenous CS. This study therefore intends to evaluate the safety and effectiveness of osilodrostat for the treatment of non-CD CS patients in support of an FDA supplemental New Drug Application (sNDA).

7 RESEARCH QUESTION AND OBJECTIVES, ENDPOINTS, AND VARIABLES

The proposed study will address the following questions:

The overall objectives of this study are to retrospectively evaluate the safety and effectiveness of osilodrostat in the treatment of non-CD CS in France and to characterise clinical parameters of CS patients treated with osilodrostat.

The proposed study will address the following research questions:

- What are the safety and effectiveness of osilodrostat in the management of non-CD CS patients in a real-world setting?
- What is the cohort demography, and how do clinical characteristics change with osilodrostat treatment?
- How is osilodrostat dosed and titrated in this patient cohort?
- What are the outcomes in these patients, including long term?
- How are these patients monitored (i.e., biochemically, and follow up characteristics)?
- What is the incidence of overall treatment-emergent adverse events (TEAEs) and adverse events of special interest (AESI)?

Table 1 Design Elements

	Objective	**Endpoint**	**Variable**
Primary	To estimate the effectiveness of osilodrostat in the treatment of non-CD Cushing's Syndrome (CS)	Number and Proportion of patients with mean urinary free cortisol (mUFC) ≤upper limit of normal (ULN) at 12 weeks	Mean Urinary free cortisol (mUFC)

	Objective	Endpoint	Variable
Secondary	To assess the long-term effects of osilodrostat in the treatment of non-CD Cushing's Syndrome (CS)	Proportion of patients with mUFC ≤ ULN at weeks 24, 36 and every 12 weeks thereafter.	mUFC
Secondary	To assess the change in biochemical variables associated with osilodrostat treatment in non-CD CS patients	Change from baseline for biochemical variables to Week 4, 8, 12, 18, 24, 36, and to every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation	mUFC; morning serum cortisol (normal and abnormal values as per standard definition)
Secondary	To assess actual and change in clinical outcomes associated with osilodrostat treatment in non-CD CS patients	Actual and percent Change in BMI, ECG, blood pressure, metabolic, biochemical, and hormone levels from baseline for clinical outcomes to Week 4, 8, 12, 18, 24, 36, and to every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation	Body Mass Index (BMI), electrocardiogram (ECG), Co-morbidities, Blood pressure, CS-specific physical exam findings Laboratory abnormalities (sodium, potassium, calcium levels, CO_2, glucose, Glycated haemoglobin (HBA1c), lipids), and hormones including: ▪ Plasma ACTH ▪ Serum 11-Deoxycortisol ▪ Plasma 11-Deoxycorticosterone ▪ Plasma Aldosterone, ▪ Plasma Renin ▪ Total Serum Testosterone or oestradiol per patient sex ▪ Serum LH ▪ Serum FSH

	Objective	Endpoint	Variable
	To characterise initiating doses, up- and down--titration regimens for non-CD CS patients treated with osilodrostat	Change from initial dose	Dose
	To estimate the incidence of overall treatment-emergent adverse events (TEAEs) and adverse events of special interest (AESI) for non-CD CS patients treated with osilodrostat	Proportion of patients who experienced TEAE and AESI	TEAE overall; TEAE specific to CS; AESI, Comprehensive metabolic panel (CMP), Complete blood count (CBC)
Exploratory	To assess change in late night salivary cortisol level from baseline to various timepoints for non-CD CS patients treated with osilodrostat	Proportion of patients with normal late night salivary cortisol at Week 4, 8, 12, 18, 24, 36, and at every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation	Late night salivary cortisol

8 RESEARCH METHODS

8.1 Study Design

This is a multi-centre, observational, non-comparative, retrospective cohort study designed to evaluate the long-term safety and effectiveness of osilodrostat in non-CD CS patients. Patients treated with oral osilodrostat regardless of the duration of their treatment will be followed retrospectively for up to 36 months after initiating osilodrostat. Follow-up for each patient will be variable, rather than fixed. Using a variable follow-up, rather than a fixed follow-up period for each patient, maximises the

person-time of patient follow-up in the study. This methodological approach allows for increased precision of findings compared to a fixed follow-up period because the unit of observation in analyses is person-time accrued aggregately across all eligible patients, rather than a count denominator of patients exposed to osilodrostat. Robust statistical properties are also associated with this study design feature.

Participating institutions will be where the past treatment of patients took place using osilodrostat under ATU and/or as a part of routine clinical practice. Data will be abstracted from patients' existing electronic, or paper institutional medical records collected during the French ATU and/or routine clinical care. Importantly, the historical decisions of health care providers (HCPs) to treat patients with osilodrostat were made purely at the discretion of the treating physicians as discussed with their patients. Thus, there is no relationship between the decision to treat patients with osilodrostat and inclusion in this study.

The study schematic is presented in Figure 1 below and illustrates the dates corresponding to the study's retrospective follow-up period, the French ATU, the latest date an osilodrostat-treated patient is eligible for inclusion in the cohort study, and general timelines.

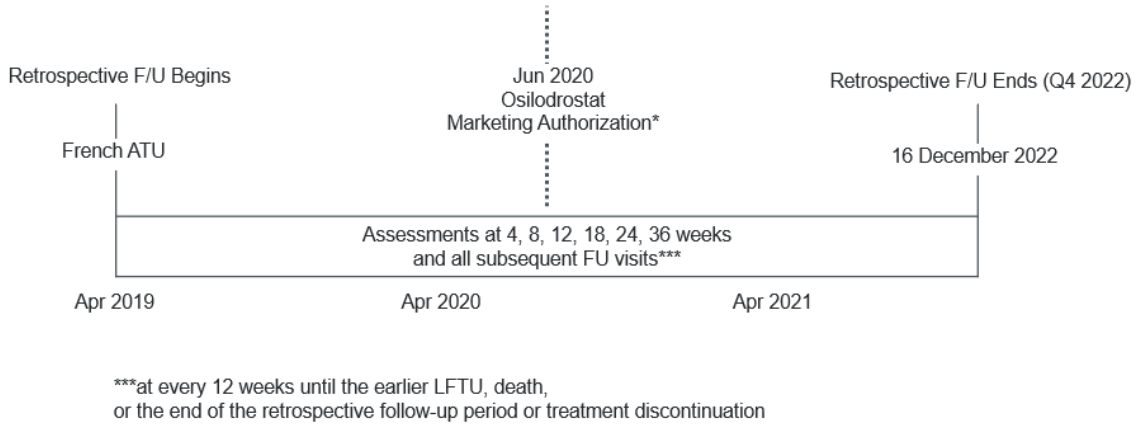

Figure 1. Study Schematic

Abbreviations : ATU : Autorisation Temporaire d'Utilisation. ; F/U : Follow Up ; LTFU : Lost to Follow Up.

Clinical practice medical records will be reviewed for all eligible patients who received osilodrostat between April 2019 and the study start date at study sites in France. The study start date was the date the first site was initiated in the study, which was on 16 December 2022. The study will screen all patients at each site who were treated with osilodrostat in France within the frame of the ATU programme prior to commercial availability along with the patients who were treated with commercially available osilodrostat. All patients treated with osilodrostat between April 2019 and the study start date (16 December 2022) will be enrolled in the study.

Patients who have previously signed a general consent for research, either alive or deceased, can be enrolled with immediate effect. If no general consent was signed, a signed no-objection letter (NOL) will be requested from patients who are still alive. Some patients might not send the NOL back to the investigator. In such instance, the investigator can document a tacit approval from the patient if there is no response after a month of the first shipment of the NOL and after 3 documented contact attempts. For deceased patients since their past treatment with osilodrostat, they can also be enrolled after the investigator has checked that the deceased patient did not leave any written document stating refusal to participate in any scientific research.

Investigators will manually review and abstract clinical information from selected patients' medical records and will enter information into the electronic case report form (eCRF) from the date of initiating Osilodrostat through the follow-up period. All data are safeguarded using industry-best security practices to prevent unlawful disclosure and practices are compliant with Title 21 Part 11 policies of the Code of Federal Regulations. The HIMS software used for EDC purposes include password protection and encryption for all transmitted data. The platform will be approved by Recordati prior to study data abstraction or during site selection procedure.

The follow-up period will be variable, from the start of their osilodrostat onset period until the earlier of loss to follow-up, death, the end of the retrospective follow-up period or treatment discontinuation occurs. The patients with a potential follow-up of at least 12 weeks will be part of the primary analysis. The only patients excluded from the primary efficacy analyses will be the ones who started treatment with osilodrostat less than 12 weeks prior to the end of the abstraction period, and are still treated with osilodrostat at the end of the abstraction period. These patients will be included only in the analyses of the secondary and exploratory endpoints including safety. The patients who discontinued treatment for any reason or were lost-to-follow up prior to 12 weeks will be considered non-responders for the primary endpoint. The end of study is defined as the last available assessment after treatment initiation for the last patient.

The timing of data collection is summarised in the Figure 1/Table 2

Table 2 Data Collection Schedule

	Data Collection Points						
Assessment	**BL[a]**	**Wk 4[b]**	**Wk 8[b]**	**Wk 12[b]**	**Wk 18[b]**	**Wk 24[b]**	**Wk 36 and every 12 wks until death, loss to F/U or end of patient data collection***
Window period		**± 2 wks**	**± 2 wks**	**± 4 wks**	**± 2 wks**	**± 4 wks**	**± 4 wks**
Authorisation of access to patient's data for enrollment[c]	X						
Inclusion-Exclusion Criteria	X						
Demographics (age at diagnosis of CS, gender)	X						
BMI	X	X	X	X	X	X	X
Clinical (disease) characteristics including date of diagnosis	X						
Medical and surgical history[h]	X[g]						
CS-Specific Physical Exam Findings (after BMI per every visit)[@]	X	X	X	X	X	X	X

	Data Collection Points						
Assessment	**BL[a]**	**Wk 4[b]**	**Wk 8[b]**	**Wk 12[b]**	**Wk 18[b]**	**Wk 24[b]**	**Wk 36 and every 12 wks until death, loss to F/U or end of patient data collection***
Window period		**± 2 wks**	**± 2 wks**	**± 4 wks**	**± 2 wks**	**± 4 wks**	**± 4 wks**
Vital signs (blood pressure)	X	X	X	X	X	X	X
Electrocardiogram	X	X	X	X	X	X	X
Prior CS treatment(s)	X						
Prior medications[e]	X[g]						
Concomitant medications	X[g]	X	X	X	X	X	X
Osilodrostat initiating dose (date)[d]	X	X	X	X	X	X	X
Up /or down-titration regimen for Osilodrostat (date)[d]		X	X	X	X	X	X
Comprehensive Metabolic Panel (CMP[&]), Complete Blood Count (CBC[#]), lipids[$]	X	X	X	X	X	X	X
HBA1c	X	X	X	X	X	X	X

	Data Collection Points						
Assessment	**BL[a]**	**Wk 4[b]**	**Wk 8[b]**	**Wk 12[b]**	**Wk 18[b]**	**Wk 24[b]**	**Wk 36 and every 12 wks until death, loss to F/U or end of patient data collection***
Window period		**± 2 wks**	**± 2 wks**	**± 4 wks**	**± 2 wks**	**± 4 wks**	**± 4 wks**
Mean urinary free cortisol[f]	X[g]	X	X	X	X	X	X
Morning serum cortisol	X	X	X	X	X	X	X
Plasma Adrenocorticotropic hormone (ACTH), Serum 11-Deoxycortisol, Plasma 11-Deoxycorticosterone ,Plasma Aldosterone, Plasma Renin, Total Serum Testosterone or oestradiol (per patient sex), Serum LH, Serum FSH	X	X	X	X	X	X	X
Late night salivary cortisol	X	X	X	X	X	X	X

	Data Collection Points						
Assessment	**BL[a]**	**Wk 4[b]**	**Wk 8[b]**	**Wk 12[b]**	**Wk 18[b]**	**Wk 24[b]**	**Wk 36 and every 12 wks until death, loss to F/U or end of patient data collection***
Window period		**± 2 wks**	**± 2 wks**	**± 4 wks**	**± 2 wks**	**± 4 wks**	**± 4 wks**
Adverse event	X	X	X	X	X	X	X
Osilodrostat discontinuation with reason for discontinuation		X	X	X	X	X	X

BL=Baseline

a. At the time of osilodrostat initiation or the closest value within 2 weeks prior to osilodrostat initiation.

b. Assessments at 4, 8, 12, 18, 24, 36 weeks will be collected, as well as all subsequent follow-up visits (at every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation) under routine clinical practice, if available. The assessments which occurred closest to each defined visit should be recorded in the CRF, even if these assessments occurred on different dates.

c. Either the patient has previously signed a general consent for research, or a signed no-objection letter (NOL) will be requested from the patient if still alive. If patient does not send the NOL back to the investigator, the investigator can document a tacit approval from the patient if there is no response after a month of the first shipment of the NOL and after 3 documented contact attempts. For a deceased patient since his/her past treatment with osilodrostat, access after the investigator has checked that this deceased patient did not leave any written document stating his/her refusal to participate in any scientific research.

d. Osilodrostat initiation dose and up- and/or down-titration regimens (including dates) and reasons for titration will be recorded in the database during the study

e. All prior medications taken within 6 months prior to the first dose of Osilodrostat must be collected.

f. Baseline mean urinary free cortisol data can be collected within 1 month prior to osilodrostat initiation.

g. Footnote-a is not applicable to the following assessments: prior medications, concomitant medications, medical and surgical history, and mean urinary free cortisol.

h. All available medical history and surgical history related to CS must be collected. Medical and surgical history not related to CS that occurred within 6 months prior to the first dose of Osilodrostat must be collected.

*End of data collection may be due to earlier of loss-to-follow-up, death, end of retrospective follow-up period or treatment discontinuation

#CBC includes WBC, RBC, Haemoglobin, Haematocrit, MCV, MCH, MCHC, RDW, Platelet Count, MPV and Differential (Absolute and Percent - Neutrophils, Lymphocytes, Monocytes, Eosinophils, and Basophils)

&CMP includes Albumin, Alkaline Phosphatase, ALT, AST, Calcium, Carbon Dioxide, Chloride, Creatinine with GFR Estimated, Glucose, Potassium, Sodium, Total Bilirubin, Total Protein, (Blood) Urea Nitrogen

$Lipid variables include Total Cholesterol, HDL, LDL, and Triglycerides

@For list of CS-specific examination findings, please refer to 8.3.2.

8.1.1 Rationale for the Study Design

The observational aspect of this study provides an approach to document the clinical course, treatment, and outcome in a real-world setting. Given that this information is readily available in the clinical setting, a retrospective review of patient data will enable efficient use of accessible information.

8.2 Setting

The study population will consist of patients with non-CD CS who meet all the inclusion criteria and none of the exclusion criteria. The authorisation for getting access to the medical data of the patients must also be ensured, through the existence of a signed general consent, or requesting a signed NOL, or fulfilling the conditions of a tacit approval or no written document stating refusal for any participation to scientific research.

8.2.1 Inclusion Criteria

Patients will be entered into this study only if they meet all of the following criteria:

1. Male and female patients ≥18 years old with diagnosis of CS, except for CD (i.e., an aetiology of adrenal adenoma, adrenocortical carcinoma, adrenal hyperplasia,

or ectopic adrenocorticotropic hormone secretion). Patients should have a contemporaneously documented diagnosis of CS as per effective guidelines.

2. Patients treated with osilodrostat between April 2019 and study start date (16 December 2022) as part of ATU programme or commercialisation.

8.2.2 Exclusion Criteria

Patients' data will be considered for inclusion in this study only if they meet none of the following criteria:

1. Patients who participated in an interventional clinical trial anytime during the study period.

2. Patients with Pseudo-Cushing's syndrome, cyclic CS, or iatrogenic CS.

8.2.3 Patient Enrolment

Patients meeting eligibility criteria will be retrospectively identified and included in the study by site investigators based on a review of medical records at site. Site policies and local regulations regarding the access to their medical records will be followed. All eligible patients identified at a site between April 2019 and study start date (16 December 2022) and consenting to be part of the study will be included.

8.2.4 Patient Withdrawal and Replacement

Not applicable

8.2.5 Patient Identification Numbers

Each patient included in the study will be identified by a number assigned by the electronic database. The number will include a site identifier and a unique patient number that will be based solely on the order in which patients are entered into the database by the investigative site according to Recordati's nomenclature practices. For example, Patient 1010001 will be the first patient entered into the database at site 101. Investigative sites will retain a record of the patient numbers and corresponding identifying patient information.

The participating investigator will agree to complete a participant/patient identification and enrolment log to permit easy identification of each patient during and after the study. This document will be reviewed by the sponsor or the designated CRO and participating site contact for completeness. The patient identification and enrolment log will be treated as confidential and will be filed by the participating investigator in the study file. To ensure patient confidentiality, no copy will be made. All reports and communications relating to the study will identify patients by patient identification and age (if allowed) at initial informed consent. In cases where the patient is not enrolled for data collection in the study, the date seen and age at initial informed consent will be used. Where applicable, the participating investigator should also complete a patient screening log, which documents all patients who were seen to determine eligibility for data collection in the study.

8.3 Variables

8.3.1 Primary Variable

- Mean urinary free cortisol (mUFC).

8.3.2 Secondary Variables

- Demographics (age, gender).
- Clinical (disease) characteristics (adrenal adenoma, adrenocortical carcinoma, ectopic adrenocorticotropic hormone secretion, adrenal hyperplasia) including date of diagnosis.
- BMI.
- Concomitant medications.
- Medical and surgical history.
- CS-specific physical exam findings, such as:
    - Plethora (facial rubor)
    - Round face (moon faces)
    - Centripetal (abdominal) obesity
    - Female hirsutism

  - Ecchymoses (bruising)
  - Dorsal/supraclavicular fat pad
  - Purplish striae
  - Proximal muscle weakness
  - Acne
  - Oedema
  - Female balding
- Vital signs (blood pressure) and ECG.
- Morning serum cortisol and mUFC.
- Laboratory abnormalities (sodium, potassium, calcium levels, CO_2, glucose, Glycated haemoglobin (HBA1c), and lipids) .
- Hormone levels
  - Plasma ACTH
  - Serum 11-Deoxycortisol
  - Serum or plasma 11-Deoxycorticosterone
  - Plasma Aldosterone
  - Plasma Renin
  - Total Serum Testosterone or oestradiol per patient sex
  - Serum LH
  - Serum FSH.
- AEs, including treatment-emergent adverse events (TEAEs) and adverse events of special interest (AESI).
- Comprehensive metabolic panel (CMP) and Complete blood count (CBC)
- The exposure of interest (defined as osilodrostat treatment and is the basis of the start of follow-up for the retrospective cohort study to assess for the defined outcomes of interest). This will include initiating osilodrostat doses along with any dose titrations.

8.4 Data Sources

The primary data source for this study will be the existing electronic medical records stored in the hospital information management systems (HIMS) and laboratory information management systems (LIMS) at the investigative sites. Also, where applicable, data will be captured from paper medical records.

8.5 Study Size

There is no statistical hypothesis testing for this study; therefore, power was not formally calculated, and no formal sample size calculations were performed. The sample size is based on practical considerations.

Historical data will be collected from approximately 100 patients with a diagnosis of non-CD CS with an aetiology of adrenal adenoma, adrenocortical carcinoma, adrenal hyperplasia, or ectopic adrenocorticotropic hormone secretion treated with osilodrostat as follows:

a) Under the April 2019 French ATU (5th Apr 2019) or

b) Under conditions of routine clinical practice after commercialization of osilodrostat in France since June 2020.

8.6 Data Management

Standardised and validated procedures and systems will be utilized to collect, process and file the clinical data of this study. Any system used will be compliant with FDA 21 CFR Part 11 requirements.

A data management plan (DMP) will be prepared to describe the processes and dataflow within the clinical study timelines, versions for the computer systems and the coding will be defined in the DMP, and if applicable, sponsor specific requests will also be documented within.

A Data Validation Specification (DVS) will be created to outline the validation checks to be performed during the study. The DVS must be finalised before data validation. The eCRFs will include programmable edits to obtain immediate feedback if critical data are missing, out-of-range, illogical or potentially erroneous. High data quality standards will be maintained, and processes and procedures utilised to repeatedly ensure that the data are as clean and accurate as possible when presented for analysis. Data quality will be enhanced through a series of programmed data quality checks that automatically detect out-of-range or anomalous data.

The study centre will allow the study monitor and sponsor representative direct access to all study documents, medical files, and source documents to enable verification of the study data, while maintaining the anonymity of the subject and confidentiality of the data. The responsible study monitor will check data during the monitoring visits either on site or remotely. The investigator will ensure that the data collected are accurate and complete. Data will be monitored within the EDC by the study monitor and any changes made during monitoring will be documented with a full audit trail within the EDC.

The raw data intended for further processing will be checked by standard routines or according to the DVS and queries will be generated and sent to the investigator for review and resolution. Corrections resulting from these queries will be confirmed on the Data Clarification Forms (DCFs). This process will be repeated until no further discrepancies are found. The data will then be declared as clean. Applicable documentation will be stored in the study files.

Only trained study staff will have access to the clinical database and every change in data will have a full audit trail.

8.6.1 Data Collection

Data collected from the medical records will be incorporated into an eCRF, that will have restricted password-protected access and an audit trail for all data access and entry. All data entered within the EDC can be viewed by appropriate study team members. The system is 21 CFR Part 11 compliant.

All data collection will be performed by appropriately trained site personnel in accordance with site privacy policies.

8.6.2 Data Processing

All data will be entered by site personnel.

The data-review and data-handling document, to be developed during the initiation phase of the study, will include specifications for consistency and plausibility checks on data

and will also include data-handling rules for obvious data errors. Query/correction sheets for unresolved queries will be sent to the study monitors for resolution with the investigator. The database will be updated based on signed corrections.

A historical file of these procedures shall be maintained, including all revisions and the dates of such revisions. Any changes in data entries shall be documented.

8.6.3 Archiving Study Records

According to International Society for Pharmacoepidemiology Guidelines on Good Pharmacoepidemiology Practices (GPP), the study archive should be maintained for at least 5 years after final report or first publication of study results, whichever comes later. The archive should include all source data and, where feasible, any biologic specimens.

8.7 Data Analysis

Statistical analysis of this study will be the responsibility of the sponsor's designee.

Any change to the data analysis methods described in the protocol will require an amendment only if it changes a principal feature of the protocol. Any other change to the data analysis methods described in the protocol, and justification for making the change, will be described in the final statistical analysis plan (SAP). The SAP will be developed as a separate document and approved prior to database lock, containing a description of the planned statistical analysis in detail with Tables, Figures and Listings (TFLs) templates.

Any deviations from the planned analyses will be described and justified in the final study report.

The overview of statistical analysis is detailed in the below section.

8.7.1 Statistical Hypothesis

The study plans to summarize only descriptive outputs and there is no inferential endpoint associated with the study. Hence no formal statistical hypothesis will be tested for this study.

8.7.2 General Considerations

An SAP describing the planned statistical analysis in detail with TLF templates will be developed as a separate document. All statistical analyses will be performed using Statistical Analysis System® (version 9.4 or higher) unless otherwise stated.

No formal statistical hypothesis testing will be performed, and all the analyses will be primarily descriptive in nature. When appropriate, and unless otherwise specified, 95% confidence intervals (CIs) will be displayed.

Descriptive statistics will include the number of available data, number of missing data and the following:

- Mean and associated standard error of mean (SE) standard deviation (SD), minimum, median, maximum, first and third quartiles and 95% CIs for means/medians, minimum (Min), maximum (Max).
- Counts and percentages of each category for categorical variables with 95% CIs.

Percentages will be based on the number of patients in safety or ITT population.

8.7.3 Analysis Population(s)

The ITT population consists of all enrolled participants who , have met all inclusion/exclusion criteria and have received osilodrostat treatment for non-CD CS, with a potential follow-up of at least 12 weeks. The only patients excluded from the primary efficacy analyses will be the ones who started treatment with osilodrostat less than 12 weeks prior to the end of the abstraction period, and are still treated with osilodrostat at the end of the abstraction period. These patients will be included only in the analyses of the secondary and exploratory endpoints including safety.

The Safety population consists of all participants who receive at least one dose of osilodrostat.

All effectiveness analysis will be performed on ITT population. Safety analysis will be performed on the safety population.

8.7.4 Control of Bias and Confounding

The study will be conducted in France through the ATU prior to commercial availability along with the patients who were treated with commercially available osilodrostat in France. Investigators will include all patients treated with osilodrostat between April 2019 and the study start date (16 December 2022); to avoid selection bias in participant recruitment, all eligible patients will be included in the study. Patients with CD should be strictly avoided by sites as a potential confounder and excluded from analysis if enrolled. Also, this will be the same for patients with Pseudo-Cushing's syndrome, cyclic CS, or iatrogenic CS, who have been treated with osilodrostat: they should be strictly avoided and if enrolled, they will be excluded from analysis.

Investigators will manually review and abstract clinical information from selected patients' medical records and will enter information into the eCRF developed for the study.

8.7.5 Patient Disposition

The numbers and percentages of participants in each population will be tabulated. The reasons for participant exclusions from data analysis sets for each of the populations will also be tabulated. In addition, the number of participants who were treated, who died will be tabulated. Primary reasons for osilodrostat treatment discontinuation will be tabulated.

8.7.6 Protocol Deviation

In this context of a retrospective observational study with no additional ancillary activities or protocol-specific procedures, protocol deviations are defined as those functional changes from the protocol that are likely to have an impact on the patient's

rights and/or the validity of the data for analysis. All protocol deviations will be reported in the clinical study report (CSR).

The number of patients with protocol deviations will be summarised.

Protocol deviations will be listed with date and study day of occurrence, description and analysis populations from which patient is excluded.

8.7.7 Demographics and Baseline Disease Characteristics

The demographics and baseline clinical characteristic will be summarised descriptively and using descriptive statistics such as n, mean, 95% CI of the mean, SD, median, minimum, maximum or frequency of counts will be provided.

- Demographics will include age, gender.
- Clinical (disease) characteristics will include putative causation (i.e., adrenal adenoma, adrenocortical carcinoma, ectopic adrenocorticotropic hormone secretion, adrenal hyperplasia) and date of Cushing's syndrome diagnosis.

8.7.8 Analysis of Primary Endpoint

The number and proportion of patients with mean urinary free cortisol (mUFC) ≤ULN at 12 weeks will be summarised descriptively along with associated 95% CI. It is a fixed value, which is upper limit of normal range. The patients who discontinued treatment prior to 12 weeks for any reason or were lost-to-follow up will be considered non-responders for the primary endpoint. The patients who started treatment with osilodrostat less than 12 weeks prior to the end of the abstraction period, and are still treated with osilodrostat at the end of the abstraction period will be excluded from the analysis.

8.7.9 Analysis of Secondary Endpoints

Changes from baseline for biochemical variables and clinical outcomes to Week 4, 8, 12, 18, 24, 36, and to every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation will be summarised descriptively and provided as listing.

Baseline is defined as the available value at the time of osilodrostat initiation or the closest value within 2 weeks prior to osilodrostat initiation. Change from baseline will be calculated as the visit value of interest minus the baseline value.

For secondary variables, refer to Table 1.

Change from initial dose

The extent of exposure will be provided by summarising the description of initiating dose (and date) of osilodrostat, and up- and/or down-titration regimen (dose increases/decreases, date).

Proportion of patients who experienced TEAEs and AESIs

An overall summary table of all treatment-emergent adverse events (TEAEs) will be presented with the number and proportion of participants and the number of events. Summary incidence tables of all TEAEs (through Week 12 and follow-up at every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period, or treatment discontinuation), SAEs and TEAEs by severity grade and relationship to study drug will be provided, classified by primary System Organ Class (SOC) and Preferred term (PT).

All AEs will be coded according to the latest version of the Medical Dictionary for Regulatory Activities and will be classified by MedDRA PT and SOC.

All safety data will be included in the participant data listings, such as listings of all TEAEs, SAEs, TEAEs leading to study drug withdrawal and listings of deaths.

Summary incidence tables of select AEs specific to CS (AEs of fatigue, nausea, vomiting, headache, backache, oedema, clinical features including acne, hirsutism, laboratory abnormalities including hypokalaemia or hyperkalaemia, and hypertension) by severity grade and relationship to study drug will be provided, classified by primary SOC and PT.

Adverse Events of Special Interest (AESI) will be defined as following AEs:

- Adrenal hormone precursor accumulation-related AEs
- Hypocortisolism-related AEs
- Pituitary tumour enlargement-related AEs
- Arrhythmogenic potential and QT prolongation AEs

The frequency and percentage of patients experiencing treatment emergent AESI will be summarised by severity grade and relationship to study drug and will be provided, classified by primary SOC and PT.

8.7.10 Analysis of Exploratory Endpoint

Late night salivary cortisol observed values at baseline and changes from baseline to Week 4, 8, 12, 18, 24, 36 and to every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation will be summarised descriptively and provided as listing. Where applicable, proportion of patients with normal and/abnormal late night salivary cortisol at Week 4, 8, 12, 18, 24, 36 and at every 12 weeks until the earlier of loss to follow-up, death, or the end of the retrospective follow-up period or treatment discontinuation.

8.7.11 Interim Analyses

Not applicable.

8.7.12 Stratified Analysis

The number and proportion of patients with mean urinary free cortisol (mUFC) ≤ULN at 12 weeks will be stratified if they received or not other therapies for Cushing's syndrome (regardless of the type of therapy) and will be summarised descriptively along with associated 95% CI. Differences in the proportions and 95% CI of the different groups will be evaluated. Further details will be described in the final SAP.

8.8 Quality Control

8.8.1 Data Quality Assurance

Recordati or delegate will conduct a site visit to verify the qualifications of each investigator, inspect the site facilities, and inform the investigator of responsibilities and the procedures for ensuring adequate and correct documentation.

The investigator is required to prepare and maintain adequate and accurate case histories designed to record all observations and other data pertinent to the study for each study participant. All information recorded on the eCRFs for this study must be consistent with the patients' source documentation (i.e., medical records).

8.8.2 Electronic Data Capture/Electronic Case Report Forms and Source Documentation

All data obtained during this study should be entered in the eCRFs promptly using CISIV EDC tool. All source documents from which eCRF entries are derived should be placed in the patient's medical records (electronic or paper). Measurements for which source documents are usually available include laboratory assessments, electrocardiogram (ECG) recordings, computed tomography (CT) scans, and X-rays.

Once the eCRF clinical data have been submitted to the central server at the independent data centre, corrections to the data fields will be captured in an audit trail. The reason for change, the name of the person who performed the change, together with the time and date will be logged to provide an audit trail.

If additional corrections are needed, the responsible monitor or data manager will raise a query in the EDC application. The appropriate staff at the study site will answer queries sent to the investigator. The name of the staff member responding to the query, and time and date stamp will be captured to provide an audit trail. Once all source data verification is complete and all queries are closed, the monitor will freeze the eCRF page.

The specific procedures to be used for data entry and query resolution using the EDC system/eCRF will be provided to study sites in a training manual. In addition, site personnel will receive training on the EDC system/eCRF.

8.8.3 Access to Source Data

During the study, a site monitor will perform remote monitoring and may also make site visits when required to review protocol compliance, compare eCRFs and individual patient's medical records to ensure that the study is being conducted according to pertinent regulatory requirements. Electronic CRF entries will be verified against source documentation. The review of medical records will be performed in a manner to ensure that patient confidentiality is maintained.

Checking of the eCRFs for completeness and clarity, and cross-checking with source documents, may be required to monitor the progress of the study. Moreover, regulatory authorities of certain countries, IRBs, IECs, and/or Recordati' s Clinical Quality Assurance Group may wish to carry out such source data checks and/or on-site audit inspections. Direct access to source data will be required for these inspections and audits; they will be carried out giving due consideration to data protection and medical confidentiality. The investigator assures Parexel and Recordati teams of the necessary support at all times. It is to be noted that Recordati' s visits or site audits covers their oversight responsibilities as a sponsor.

8.9 Limitations of the Research Methods

Due to the observational nature of this retrospective cohort study, conducted under conditions of routine clinical practice, bias is more prone to occur. Consequently, study findings may be subject to limitations in generalizability to the target population in availability of data, and variability in local treatment practices and guidelines.

This study is an observational study and relies on patients' existing medical records available at the time of notification of the protocol to the French Data Hub. Therefore, although all patients will be followed longitudinally using historical medical records, some data points may be missing. However, it is expected that any missing data will be

minimal and would not affect the study outcome, or that statistical methods of imputation and/or missingness will mitigate its effect.

Many factors can lead to biased study results. The possible sources of bias are selection bias, inconsistent data collection, measurement error with continuous variables, misclassification with categorical variables, confounding factors, methodical errors, and other errors. A good study plan, a good risk management plan, and the good quality control procedures are the key to reducing, avoiding, correcting predictable problems.

8.10 Other Aspects

Not applicable.

9 PROTECTION OF HUMAN SUBJECTS

9.1 Ethical Considerations

The procedures set out in this study protocol are designed to ensure that Recordati and investigator abide by the principles of the International Society for Pharmacoepidemiology GPP guidelines *(Ispe 2008)*. The study will also be carried out in compliance with local legal requirements MR-004.

9.2 Informed Consent

Informed consent is not required for this study, because this study is a secondary data collection study using anonymised patient data. The authorisation for getting access to the medical data of the patients must also be ensured, through the existence of a signed general consent, or requesting a signed NOL, or fulfilling the conditions of a tacit approval or no written document stating they refused any participation to scientific research.

9.3 Protocol Approval and Amendment

Before the start of the study, the French health data hub will be notified with the study protocol and/or other relevant documents, in accordance with local legal requirements.

Recordati must ensure that all ethical and legal requirements have been met before the first patient is enrolled in the study.

To alter the protocol, amendments must be sent to Competent Authority as notification prior to implementation.

Administrative changes (not affecting patient risk) may be made without the need for a formal amendment. All amendments will be distributed to all protocol recipients, with appropriate instructions.

9.4 Confidentiality

All study findings and documents will be regarded as confidential. The investigator and members of his/her research team must not disclose such information without prior written approval from Recordati.

The anonymity of participating patients must be maintained. Investigators shall ensure that personal identifiers will be removed from any study files that are accessible to non-study personnel in accordance with applicable laws and regulations. Patients will be identified on eCRFs, and other documents submitted to Parexel by their patient number, initials and/or birth date (if allowed), not by name. Documents not to be submitted to Parexel that identify the patient must be maintained in confidence by the investigator.

9.5 Duration of the Study

The study duration will be approximately up to 3 years. It will be retrospective follow-up period at every 12 weeks until the earlier of loss to follow-up (LTF), death, or the end of the retrospective follow-up period or treatment discontinuation.

10 MANAGEMENT AND REPORTING OF ADVERSE EVENTS/ADVERSE DRUG REACTIONS

Being retrospective chart review, it does not require active reporting of AEs/ADRs to regulatory authorities.

10.1 Definitions

Adverse events (AEs)

An AE is any untoward medical occurrence in a patient or clinical study participant administered a pharmaceutical product, whether or not considered related to the product. An AE can therefore be any unfavourable and unintended sign (including an abnormal laboratory finding, for example), symptom, or disease (new or exacerbated) temporally associated with the use of a product.

Treatment-emergent adverse events (TEAEs)

Any AE with an onset date on or after the study drug start date and no later than 30 days after last dose of study drug, or any worsening of any AE on or after the study drug start date.

Serious adverse events (SAEs)

An SAE is any AE that:

- Results in death.
- Is life-threatening. (Note: The term 'life-threatening' in the definition of 'serious' refers to an event in which the subject was at risk of death at the time of the event; it does not refer to an event that hypothetically might have caused death if it were more severe).
- Requires inpatient hospitalization or prolongation of existing hospitalization.
- Results in persistent or significant disability/incapacity.
- Is a congenital anomaly/birth defect.

- Is an important medical event. Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered an SAE when, based upon appropriate medical judgment, they may jeopardize the patient and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

Adverse drug reaction (ADR)

An ADR is defined as a response to a medicinal product which is noxious and unintended. Response in this context means that a causal relationship between a medicinal product and an AE is at least a reasonable possibility, i.e., the relationship cannot be ruled out.

10.1.1 Categorisation of Adverse events

10.1.1.1 Severity Assessment

All AEs will be classified using the current version of the Common Terminology Criteria for Adverse Events (CTCAE) version 5.0, developed, and maintained by the Cancer Therapy Evaluation Program at the National Cancer Institute.

Grade 1: Mild: asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated; The AE is easily tolerated and does not interfere with daily activities.

Grade 2: Moderate: minimal, local, or non-invasive intervention indicated; The AE interferes with daily activities, but the subject is still able to function.

Grade 3. Severe: not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; The AE is incapacitating and requires medical intervention. Every effort will be made to obtain an adequate evaluation of the severity.

Grade 4. Life threatening: consequences; urgent or emergent intervention needed.

Grade 5. Death related to or due to AE.

10.1.1.2 Causality Assessment

The following definitions of relationship to the study drug should be used to characterise the suspected causality of each AE, based on the investigator's consideration of all available information about the AE, including temporal relationship to drug administration, recognised association with drug product/class, pharmacological plausibility, and alternative aetiology (e.g., underlying illness, concurrent conditions, concomitant treatments):

• **Related** - An AE that follows a reasonable possibility of a causal relationship to study drug i.e., reasonable temporal sequence from administration of a drug (including the course after withdrawal of the drug), or for which possible involvement of the drug cannot be ruled out, although factors other than the drug, such as underlying diseases, complications, concomitant drugs, and concurrent treatments, may also be responsible

• **Not Related** - An AE that does not follow a reasonable possibility of a causal relationship to the study drug i.e., no reasonable temporal sequence from administration of a drug and/or that can reasonably be explained by other factors, such as underlying diseases, complications, concomitant drugs, and concurrent treatments.

10.2 Collection and Reporting of Adverse Events/Adverse Reactions

This study uses data from electronic and paper healthcare records. Consistent with regulatory requirements for a non-interventional study based on secondary use of data (EU Good Pharmacovigilance Practices VI.C.1.2.1), no reporting of AEs is required. It is assumed that any AEs identified in existing healthcare records have already been reported at the time it occurred in the past, therefore, no additional AE/SAE reporting will be made to regulatory authorities to avoid duplicate reports.

All adverse events/reactions collected for the study will be recorded and summarised in the final study report.

11 PLANS FOR DISSEMINATING AND COMMUNICATING STUDY RESULTS

The results of the study will be communicated after the end of the study to the investigator and to Competent Authorities/Ethics Committee, as appropriate. They can be submitted to Regulatory Authorities and can be either submitted for publications and/or posted in a publicly available database. The Sponsor reserves the right to review all manuscripts and abstracts before their submission for publication or presentation. This is not intended to restrict or hinder publication or presentation but to allow the sponsor to protect the confidentiality of information and to provide comments that may not yet be available to the investigator. Publications will comply with internal Recordati standards and the International Committee of Medical Journal Editors (ICMJE).

Ownership of all data will remain with the sponsor.

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13 APPENDICES

13.1 Appendix 1: List of Supplementary Documents

Number	***Title***
1	*No Objection Letter*
2	*Sample Case Report Form*