[redacted]

A phase 2, open-label, single-arm, multicenter study of SOT101 in combination with pembrolizumab to evaluate the efficacy and safety in patients with selected advanced/refractory solid tumors

Study interventions	Product code: SOT101 (previously SO-C101, RLI-15); International Nonproprietary Name: Nanrilkefusp alfa Pembrolizumab
Regulatory agency identifier numbers	EudraCT number: 2021-005774-25 IND number: 140011
Protocol number	SC104
Phase	2
Version	Protocol Amendment 1
Date	20Apr2023
Sponsor	SOTIO Biotech AG
Legal registered address	Lichtstrasse 35 - WSJ-210 4056 Basel Switzerland

[redacted]

SIGNATURES/PROTOCOL APPROVAL AND RELEASE: SOTIO

We, the undersigned, have read this Protocol and agree that it contains all the necessary information required for the conduct of this clinical trial.

For SOTIO Biotech AG:

[redacted]

Signature: ....……

Date: ....……

Signature: ....……

Date: ....……

SIGNATURES/PROTOCOL APPROVAL AND RELEASE: COORDINATING INVESTIGATOR

I, the undersigned, have read this Protocol and agree that it contains all the necessary information required for the conduct of this clinical trial.

Coordinating investigator:

Stéphane Champiat, M.D., Ph.D.
Assistant Professor
Institut Gustave Roussy
Villejuif, France

Signature: ……

Date: ……

INVESTIGATOR'S DECLARATION

I have read this Protocol and I agree that it contains all the necessary details for carrying out this clinical trial. I agree to personally conduct or supervise the clinical trial as described in accordance with the relevant current Protocol and within the time designated. I will only make changes after receiving the sponsor's approval, except when necessary to protect the safety, rights, or welfare of patients.

I verify that I am suitably qualified by education, scientific medical training, and experience to conduct the clinical trial. Documentation of my qualifications and professional affiliations are contained in my up-to-date curriculum vitae.

I will provide the Protocol and all information relating to pre-clinical and previous clinical experience (e.g., Investigator's Brochure) to all associates, colleagues, and staff assisting in the conduct of this clinical trial. I will discuss the material with them to ensure that they are fully conversant with the Protocol, the medical treatment, and the conduct of the clinical trial, and that they will handle the data and information generated in the clinical trial confidentially. I agree to ensure that they are informed about their obligations in meeting the investigator's commitments listed in this Investigator's Declaration, as delegated and applicable to them.

I agree to personally conduct or supervise the clinical trial in accordance with: i) the current version of the Declaration of Helsinki; ii) the current version of International Council for Harmonisation (ICH) guidelines on Good Clinical Practice (ICH E6); iii) the moral, ethical, and scientific principles that justify medical research; and iv) all relevant national and regional laws and regulations relating to clinical trials and the protection of patients of the country in which the clinical trial will be performed.

I will ensure that an institutional review board (IRB) or independent ethics committee (IEC; in the US)/ethics committee (EC; in the EU)/equivalent committee (outside the US and EU) that complies with the requirements of national and regional legislation and the Declaration of Helsinki, and that follows the recommendations in ICH E6, will be responsible for the initial and continuing review and approval of the clinical investigation. I also agree to promptly report to the IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) all changes in research activity and all unanticipated problems involving risks to patients or others. Additionally, I will not make any changes in the research without IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) approval, except where necessary to eliminate apparent immediate hazards to patients.

I agree to inform all patients and associates, colleagues, and staff assisting in the conduct of this clinical trial that the drugs are being used for investigational purposes, and I will ensure that the requirements relating to obtaining informed consent and IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) review and approval are met in accordance with national and regional legislation and the Declaration of Helsinki, and consistent with the recommendations in ICH E6.

All patients will be informed that they may withdraw from the clinical trial at their discretion at any time. I will use only the information sheet and consent form approved by the sponsor and the IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) which has reviewed this clinical trial.

I will provide the sponsor with any material written by myself (e.g., clinical trial summary) which is given to the IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the

US and EU) in support of the application. I agree to maintain adequate and accurate records and to make those records available for inspection in accordance with national and regional legislation and the Declaration of Helsinki, and consistent with the recommendations in ICH E6. I agree to the audit and monitoring procedures that involve verification of clinical trial records against the original records by direct access. In case the source documentation and clinical trial data are kept electronically, I agree to ensure that these comply with the requirements on computerized systems and their validation in line with national and regional legislation and recommendations in ICH E6.

I will retain the trial-related essential documents until the sponsor informs me that these documents are no longer needed but no sooner than the requirements in national and regional legislation.

I certify that any laboratory, excluding the central laboratory (laboratories) appointed for the clinical trial, in which laboratory parameters will be determined, is subject to regular external quality control.

I agree to report to the sponsor adverse events that occur in the course of the clinical trial in accordance with national and regional legislation and the Declaration of Helsinki, and consistent with the recommendations in ICH E6. I have read and understand the information in the Investigator's Brochure, including the potential risks and side effects of the study intervention.

I agree to the collection, processing, transfer, use, and storing of my personal data and details relating to my professional activities for the purposes of the clinical trial by the sponsor and/or by a delegated party (e.g., a contract research organization).

I understand that the (e)CRFs and other data pertinent to this clinical trial are the property of the sponsor and are confidential. I will supply the sponsor with the clinical trial data in such a way that the patient's personal information and identity are protected.

I agree to comply with all other requirements regarding obligations of clinical investigators and with all other pertinent requirements in accordance with national and regional legislation and the Declaration of Helsinki. I also agree to follow the recommendations in ICH E6.

Investigator's signature: _____

Date: _____

Printed name: _____

Street address: _____

Telephone number: _____

Other contact information

Full contact details for each investigational site, the sponsor, and key coordinating and operational personnel involved in this clinical trial will be maintained in the Trial Master File and in the Investigator Site File.

PROTOCOL AMENDMENT SUMMARY OF CHANGES

Change number	Description of change	Rationale for change	Section(s) in Protocol Amendment 1
1	Clarification that during futility analyses, recruitment may be paused	Clarification	9.4 Interim analysis
2	Benefit/risk assessment update	Update	2.3 Benefit/risk assessment
3	Reduction of pharmacokinetics sampling time points	To reduce the volume of blood drawn and number of samples to be withdrawn from patients	1.3.5 Nanrilkefusp alfa PK sampling
4	Addition of a time window for blood sampling	To allow the sites to collect blood within a defined time interval	1.3.3 Collection of blood samples, nanrilkefusp alfa combined with pembrolizumab
5	Guidance for prevention of fever before dosing	Some patients who were hospitalized to monitor whether there is a further increase which in some cases resulted in a serious adverse event and/or next dose was not given.	6.5.1 Dose modification and toxicity management for nanrilkefusp alfa
6	Clarification that tumor imaging is to be continued irrespective of clinical progression	Clarification	8.1.1.3 End of treatment and follow-up tumor imaging
7	Reduction of nanrilkefusp alfa anti-drug antibody sampling time points	To reduce the volume of blood drawn from patients	1.3.3 Collection of blood samples, nanrilkefusp alfa combined with pembrolizumab
8	Re-screening to be allowed	To allow more patients to be enrolled	5.4 Screen failures
9	Reduction of prostate cancer-specific biomarker sampling time points	To reduce the volume of blood drawn from patients	1.3.3 Collection of blood samples, nanrilkefusp alfa combined with pembrolizumab

Change number	Description of change	Rationale for change	Section(s) in Protocol Amendment 1
10	Clarification that legally speaking, all contractors of SOTIO Biotech AG are third parties	Clarification	10.1.12 Future research
11	Change of the sponsor's address	Sponsor moved	Title page
12	Clarification that not all benchmark treatments consist of pembrolizumab alone	Clarification/ Simplification	1.1 Synopsis, 9.4 Interim analysis
13	Correction of a typo in the definition of circulating tumor cell count conversion: from ">5" to "≥5"	Typo	1.1 Synopsis, 3 OBJECTIVES AND ENDPOINTS, 5.1 Inclusion criteria, 9.3.3.2 Efficacy endpoints
14	Replacement of the product code "SOT101" with the International Nonproprietary Name "nanrilkefusp alfa" in the body of the protocol for better clarity; the product code remains the same, i.e., "SOT101"	Clarification	Throughout the protocol
15	Clarification that day 6 blood sampling should be performed irrespective of any cytokine trends	RNA analysis is needed for all patients on day 6 to have relevant results	1.3 Schedule of activities
16	Clarification that for ovarian cancer, the last progression on platinum-based therapy should be considered in the calculation of the 6 months within when the study treatment should start	Clarification	1.1 Synopsis, 1.2 Schema, 4.2 Scientific rationale for study design, 5.1 Inclusion criteria

Change number	Description of change	Rationale for change	Section(s) in Protocol Amendment 1
17	Clarification that left ventricular ejection fraction should be determined by echocardiography or multigated acquisition scanning	Clarification	1.3 Schedule of activities, 8.2.4 Electrocardiography and left ventricular ejection fraction
18	Change of the progression-free survival and time to response censoring rule when no tumor assessments were performed from "eligibility" to "the first day of study treatment"	The censoring rules were not aligned with the study population. As only treated patients are considered, the first day of study treatment should be the censoring timepoint (in case of no tumor assessments) in order to avoid negative values.	9.3.3.2 Efficacy endpoints
19	Clarification that nanrilkefusp alfa and pembrolizumab concentrations over time are analyzed in serum not plasma	Clarification	1.1 Synopsis, 3 OBJECTIVES AND ENDPOINTS
20	Correction that serum clearance is not evaluated after oral administration	Typo	8.4 Pharmacokinetics
21	Reduction of the number of ECG assessments	No significant cardiac toxicity has been observed in a 13-week repeated dose toxicology study in cynomolgus monkeys and in clinical studies with nanrilkefusp alfa	1.3.1 Schedule of activities, nanrilkefusp alfa combined with pembrolizumab
22	Clarification that a fresh biopsy must be taken during screening unless the biopsy cannot be obtained due to safety reasons or non-accessibility of the tumor site. If it is not possible to obtain a fresh biopsy, every effort	To allow inclusion of patients without accessible tumor tissue	1.3.1 Schedule of activities, nanrilkefusp alfa combined with pembrolizumab, 5.1 Inclusion criteria

Change number	Description of change	Rationale for change	Section(s) in Protocol Amendment 1
	should be taken to retrieve an archival biopsy. Archived, fixed tumor tissue may only be collected if taken preferentially after completion of the most recent systemic tumor therapy and within 12 months prior to the first dose of study treatment. Investigators are highly encouraged to obtain an optional biopsy at cycle 2 day 13 and at disease progression or at any other clinically relevant event if it does not expose the patient to an increased risk.		
23	Change to exclusion criterion 3: from "before study entry (ICF signature)" to "before study treatment (cycle 1 day 1)"	Four weeks plus up to another 4 weeks of screening is too long for patients to be off systemic therapy	5.2 Exclusion criteria
24	Clarification of exclusion criterion 8.3 (added text underlined, deleted text crossed out): "and any clinically significant history of coronary heart disease and clinically significant ~~peripheral and/or carotid~~ artery disease within the past 5 years"	Clarification	5.2 Exclusion criteria
25	Standardization of adverse event (AE) terminology according to NCI CTCAE version 5.0	To standardize AE reporting	8.3.8.1 Adverse events, 8.3.8.2 Documenting on eCRFs
26	Deletion of the row "Whole blood biomarker	Redundant	1.3.3 Collection of blood samples,

Change number	Description of change	Rationale for change	Section(s) in Protocol Amendment 1
	analysis" in the table "Collection of blood samples, nanrilkefusp alfa combined with pembrolizumab"		nanrilkefusp alfa combined with pembrolizumab
27	Clarification that a maximum of 3 lines of previous treatment are allowed for patients with advanced hepatocellular carcinoma and a maximum of 4 lines are allowed for patients with metastatic castration-resistant prostate cancer and ovarian cancer	To clarify the number of previous lines of therapy	1.2 Schema, 4.2 Scientific rationale for study design, 5.1 Inclusion criteria,
28	Update of the information on the combination recommended phase 2 dose	To update the information on the combination recommended phase 2 dose	2.1 Study rationale, 4.3 Justification for dose
29	Clarification of the end of the study	Clarification	1.1 Synopsis, 1.3 Schedule of activities, 4.1.3 Follow-up, 4.4 End of study definition
30	Addition that live/attenuated vaccines are prohibited 90 days after the last dose of nanrilkefusp alfa and/or pembrolizumab, whichever is later	Revised according to the Czech State Institute for Drug Control (SUKL)'s recommendation (SUKL RFI 25Mar2022): *We request to add to the protocol that live/attenuated vaccines are prohibited even 90 days after the last dose of study treatment. This is standard procedure in immunotherapy studies.*	6.8.2 Prohibited medications
31	Addition that serology testing is mandatory	Revised according to SUKL's recommendation (SUKL RFI 25Mar2022): *We request to state in the protocol that HIV, HBV and*	5.1 Inclusion criteria, 5.2 Exclusion criteria, 8.2.5.5 Serology testing for infections

Change number	Description of change	Rationale for change	Section(s) in Protocol Amendment 1
		HCV tests are a mandatory part of screening in the Czech Republic. These infections are in exclusion criteria and therefore pose a risk to clinical trial subjects. This is a standard requirement for immunotherapy studies.	
32	Addition of cytokine release syndrome management recommendations	Revised according to SUKL's recommendation (SUKL RFI 25Mar2022): *We require that the exact cytokine release syndrome (CRS) management procedure be added to the protocol. It is not enough to just refer to standard workplace procedures, the protocol should include the management of all major toxicities, especially those for the tested product.*	6.5.1 Dose modification and toxicity management for nanrilkefusp alfa, 6.5.3 Clinical management of cytokine release syndrome
33	Addition of more detailed information about nanrilkefusp alfa application	Revised according to SUKL's recommendation (SUKL RFI 25Mar2022): *The protocol is missing more detailed information about the SOT101 application. Where will it be administered subcutaneously (abdomen, buttocks,...), will application site change, what exactly will be the procedure? We require proper adjustment of the protocol.*	6.5.1 Dose modification and toxicity management for nanrilkefusp alfa
34	Addition of information about SUSAR reporting	Revised according to SUKL's recommendation (SUKL RFI 25Mar2022): *The protocol does not mention SUSAR reporting. We request that the protocol be*	8.3.5 Regulatory reporting requirements for SAEs

Change number	Description of change	Rationale for change	Section(s) in Protocol Amendment 1
		supplemented in accordance with KLH-21.	
35	Removal of the possibility of nanrilkefusp alfa monotherapy treatment after pembrolizumab discontinuation (applicable in the Czech Republic only)	Revised according to SUKL's recommendation (SUKL RFI 14Apr2022): *We acknowledge that SOT101 monotherapy is only considered when pembrolizumab is discontinued due to unacceptable toxicity. However, we do not agree with the sponsor's opinion, monotherapy with SOT101 is unacceptable. Following discontinuation of pembrolizumab, there are other proven lines of treatment to which patients are entitled. To keep the patients on the experimental medicinal product with uncertainty of its effect would be unethical. Especially in patients who will be on the first / second line of treatment. Therefore, we request to remove information from the protocol about the possibility of continuing SOT101 monotherapy in case of discontinuation of pembrolizumab.*	1.3.1 Schedule of activities, nanrilkefusp alfa combined with pembrolizumab, 7.1 Discontinuation of study intervention
36	Addition of cytokine release syndrome management recommendations	Revised in response to the Spanish Agency of Medicines and Health Products (AEMPS)'s request for information (AEMPS RFI 21Apr2022): *In protocol section 6.5.1 on toxicity management and SOT101 dose modification, it is specified that the procedures and changes will be discussed on a case-by-case basis with*	6.5.1 Dose modification and toxicity management for nanrilkefusp alfa, 6.5.3 Clinical management of cytokine release syndrome

Change number	Description of change	Rationale for change	Section(s) in Protocol Amendment 1
		the sponsor's medical monitor primarily to avoid symptoms related to cytokine release syndrome. Justify or clarify the reason for not standardizing the procedures for managing these symptoms per protocol.	
37	Addition of specific risk minimization measures for nanrilkefusp alfa-related hepatic toxicities	Revised in response to the Belgian Federal Agency for Medicines and Health Products (AFMPS)'s requirement dated 16May2022: *The sponsor is required to add specific RMM for Liver Events in the study protocol...*	6.5.1 Dose modification and toxicity management for nanrilkefusp alfa
38	Addition of instructions for reporting of certain liver adverse events	Revised in response to AFMPS's requirement dated 16May2022: *In addition the sponsor is required to add in the protocol instructions for Reporting of Liver Adverse Event (AE) including, for immediate and expedited reporting, requirements from Investigator to Sponsor and from Sponsor to Health Authority, Investigators, and EC/IRB of 'Suspected Unexpected Serious Adverse Reaction'...*	8.3.1.3 Adverse events of special interest
39	Clarification that the study investigator will make every effort to obtain consent from the pregnant patient to follow the outcome of the pregnancy and report the condition of the fetus or newborn to the sponsor or designee	Revised in response to the Belgian central ethics committee's request dated 27Apr2022 concerning the Informed Consent Form for pregnant patients / pregnant partners	8.3.6 Pregnancy
40	Clarification that egg and sperm donation is	Revised in response to the Belgian central ethics	8.3.6 Pregnancy

Change number	Description of change	Rationale for change	Section(s) in Protocol Amendment 1
	not allowed during the treatment period and for at least 120 days after the last dose of pembrolizumab or at least 30 days after the last dose of nanrilkefusp alfa, whichever is later	committee's request dated 27Apr2022	
41	Addition that serology testing is mandatory	Revised in response to the Italian Medicines Agency (AIFA)'s request (AIFA RFI 17Jun2022): *Section 5.1-Inclusion Criteria-Hepatitis: Baseline HCV and HBV testing should be required in order to assure appropriate patient management and monitoring, according to medical judgement.*	5.1 Inclusion criteria, 5.2 Exclusion criteria, 8.2.5.5 Serology testing for infections
42	Addition of toxicity management recommendations for nanrilkefusp alfa	Revised in response to AIFA's request (AIFA RFI 17Jun2022): *Section 6.5.1 Dose modification and toxicity management for SOT101: for the sake of safety and in order to assure homogeneity in toxicity management, the dose modification should not be only discussed on a case-by-case basis. General recommendation should be reported in the study protocol.*	6.5.1 Dose modification and toxicity management for nanrilkefusp alfa, 6.5.3 Clinical management of cytokine release syndrome
43	Specification of study population in response to the French National Agency for the Safety of Medicines and Health Products (ANSM)'s request	Revised in response to ANSM's request (ANSM 22Apr2022): *To amend inclusion criterion 4 for disease characteristics as following :* *- NSCLC: add to the description of the advanced*	1.1 Synopsis, 1.2 Schema, 4.2 Scientific rationale for study design, 5.1 Inclusion criteria

Change number	Description of change	Rationale for change	Section(s) in Protocol Amendment 1
		setting "not amenable to curative treatment" and change "and/or" by "and" for immune checkpoint inhibitor and platinum containing regimen *- cSCC : mention that inclusion of patients is possible only in cSCC when association with RT is not feasible* *In addition the sponsor is requested to further substantiate the rationale for a direct rechallenge with immunotherapy instead of using cytotoxic chemotherapy* *- mCRPC : make clear that patients should have received abiraterone and enzalutamide*	
44	Addition of management recommendations for cytokine release syndrome, shortening of the QT interval, and injection site reaction	Revised in response to ANSM's request (ANSM 22Apr2022): *To amend study protocol section 6.5 so as to include description of toxicity management for identified and potential risks of SOT101, especially CRS, shortening of QT and injection site reaction. Indeed, the sole reference of "discussion on a case-by-case basis between the site and the sponsor's medical monitor" is not considered sufficient. Protocol should include or refers to guidelines for the management of the identified and potential risks for the experimental treatment. Management of increased ALT and AST is adequately described in table 6.2. Considering the risk of CRS, the protocol should include*	6.5.1 Dose modification and toxicity management for nanrilkefusp alfa, 6.5.3 Clinical management of cytokine release syndrome

Change number	Description of change	Rationale for change	Section(s) in Protocol Amendment 1
		stopping rules for the experimental product in addition to the reference to guidelines for the management of CRS.	
45	Addition of a biochemistry test on day 8 of cycle 3	Revised in response to ANSM's request (ANSM 22Apr2022): *To amend protocol section "1.3.3 Collection of blood samples, SOT101 combined with pembrolizumab" so as to add a Biochemistry test on day 8 of cycle 3.*	1.3 Schedule of activities
46	Addition of pregnancy testing until the end of mandatory contraception defined in the protocol	Revised in response to ANSM's request (ANSM 22Apr2022): *To amend schedule of activities so as to extend pregnancy testing, for WOCBP until the end of mandatory contraception defined in the protocol, per CTFG guideline.*	1.3.1 Schedule of activities, nanrilkefusp alfa combined with pembrolizumab
47	Specification of study population in response to ANSM's request	Revised in response to ANSM's request (ANSM 19Aug2022): *Regarding NSCLC, our request for amending "and/or" by "and" for immune checkpoint inhibitor and platinum containing regimen is maintained…*	1.1 Synopsis, 1.2 Schema, 4.2 Scientific rationale for study design, 5.1 Inclusion criteria, 9.4 Interim analysis, 9.5 Sample size determination
48	Specification of the study population in response to the request of the Hungarian central ethics committee (CEC)	Revised in response to the Hungarian CEC's request dated 09Sep2022: *The inclusion criteria should be supplemented; mCRPC: for the second-line or later-line treatment of patients with mCRPC after recurrence or failure of docetaxel therapy. In the above indication, the*	1.1 Synopsis, 1.2 Schema, 4.2 Scientific rationale for study design, 5.1 Inclusion criteria

Change number	Description of change	Rationale for change	Section(s) in Protocol Amendment 1
		criterion recommended by the Committee for the efficacy of the androgen receptor inhibitor therapy is as follows: *"For the second-line or later-line treatment of patients with mCRPC after recurrence or failure of docetaxel and androgen receptor target therapy"*	
49	Update of dose modification and toxicity management guidelines for hypothyroidism and myocarditis associated with pembrolizumab	To provide investigators with up-to-date information on the management of irAEs associated with pembrolizumab	6.5.2.1.3 Restarting study interventions
50	Clarification that any licensed COVID-19 vaccine (including for emergency use) in a particular country is allowed in the study as long as they are mRNA vaccines, adenoviral vaccines, or inactivated vaccines	Clarification	6.8.2 Prohibited medications
51	Correction of typographical errors, minor editing	To improve clarity	Throughout the document

DOCUMENT HISTORY	
Version	**Date**
Protocol	18Nov2021
Protocol Amendment CZ-1	04Apr2022
Protocol Amendment CZ-2	28Apr2022
Protocol Amendment ES-1	29Apr2022
Protocol Amendment BE-1	30Jun2022
Protocol Amendment FR-1	12Jul2022
Protocol Amendment IT-1	12Jul2022
Protocol Amendment FR-2	23Aug2022
Protocol Amendment HU-1	16Sep2022
Protocol Amendment UK-1	11Oct2022
Protocol Amendment 1	20Apr2023

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1 PROTOCOL SUMMARY

1.1 Synopsis

<table>
<tr><td>Study title</td><td>A phase 2, open-label, single-arm, multicenter study of SOT101 in combination with pembrolizumab to evaluate the efficacy and safety in patients with selected advanced/refractory solid tumors</td></tr>
<tr><td>Rationale</td><td>Nanrilkefusp alfa is a fusion protein which consists of the N-terminal domain of human interleukin 15 (IL-15) receptor α covalently coupled to human IL-15. Pembrolizumab is a potent humanized IgG4 monoclonal antibody with high specificity of binding to the programmed cell death protein 1 receptor, thus inhibiting its interaction with programmed cell death ligand 1 and programmed cell death ligand 2. The combination of nanrilkefusp alfa and pembrolizumab is expected to lead to synergistic outcomes in the induction and/or maintenance of antitumor responses.</td></tr>
<tr><td>Overall design</td><td>This is a phase 2, open-label, single-arm, multicenter study of nanrilkefusp alfa in combination with pembrolizumab to evaluate the efficacy and safety in patients with selected advanced/refractory solid tumors.</td></tr>
<tr><td>Objectives and endpoints</td><td>
<table>
<tr><th>Objective</th><th>Endpoint(s)</th></tr>
<tr><td colspan="2">Primary</td></tr>
<tr><td>• To estimate the antitumor efficacy of nanrilkefusp alfa in combination with pembrolizumab</td><td>• Objective response rate (ORR) according to Response Evaluation Criteria In Solid Tumors version 1.1 (RECIST 1.1) in patients with measurable disease</td></tr>
<tr><td colspan="2">Secondary</td></tr>
<tr><td>• To assess the safety and tolerability of nanrilkefusp alfa in combination with pembrolizumab according to Common Terminology Criteria for Adverse Events (CTCAE) version 5.0</td><td>• Type, frequency, and severity of treatment-emergent adverse events (TEAEs); adverse events of special interest (AESIs); safety laboratory findings; vital signs; electrocardiography (ECG) findings</td></tr>
</table>
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	• To further evaluate the antitumor efficacy of nanrilkefusp alfa in combination with pembrolizumab	• ORR according to RECIST for immune-based therapeutics (iRECIST) (iORR) in patients with measurable disease • Best overall response according to RECIST 1.1 (BOR) and iRECIST (iBOR) in patients with measurable disease • Duration of response according to RECIST 1.1 (DoR), iRECIST (iDoR), and Prostate Cancer Clinical Trials Working Group 3 (PCWG3)-modified RECIST 1.1 (metastatic castration-resistant prostate cancer [mCRPC] only) • Clinical benefit rate according to RECIST 1.1 (CBR), iRECIST (iCBR), and PCWG3-modified RECIST 1.1 (mCRPC only) • Progression-free survival according to RECIST 1.1 (PFS), iRECIST (iPFS), and PCWG3-modified RECIST 1.1 (mCRPC only) • Time to response according to RECIST 1.1 (TtR) and iRECIST (iTtR) in patients with measurable disease • mCRPC only: o Circulating tumor cell (CTC) count conversion from ≥5 to <5 cells per 7.5 mL of blood o Confirmed prostate-specific antigen (PSA) decline of ≥50% o Time to confirmed PSA progression

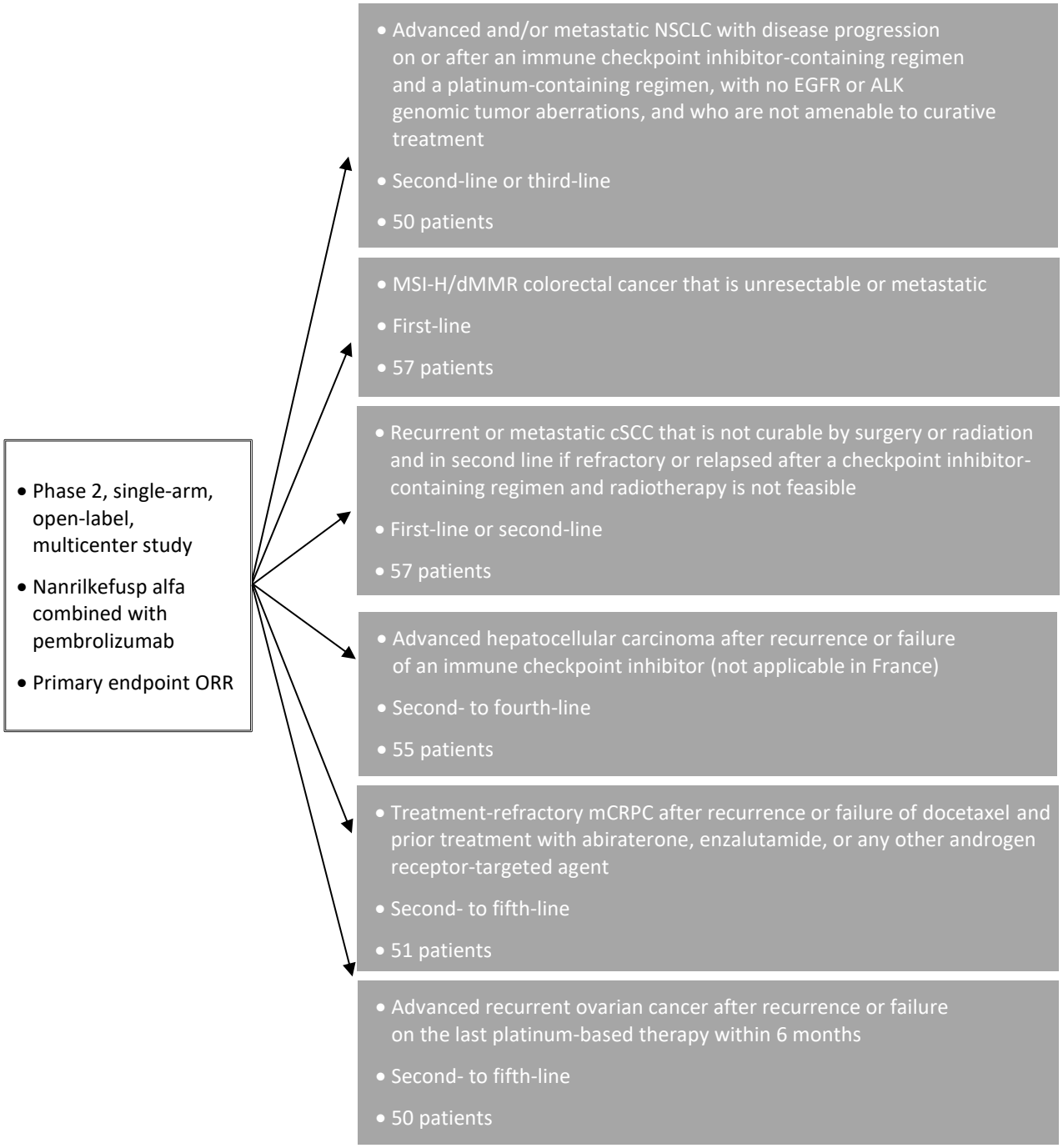
	• (Population) pharmacokinetics (PK) of nanrilkefusp alfa in combination with pembrolizumab	• Concentrations of nanrilkefusp alfa over time
	• To determine the immunogenicity of nanrilkefusp alfa in combination with pembrolizumab	• Incidence, titer, and time course of anti-drug antibodies (ADAs) against nanrilkefusp alfa
	Exploratory	
	• To identify immune and molecular (including genomic, metabolic, and/or proteomic) biomarker(s) in archival and/or fresh tumor tissue and blood that may be indicative of clinical response/resistance, safety, pharmacodynamic (PD) activity, and/or the mechanism of action of nanrilkefusp alfa and pembrolizumab	• Changes in the expression of immune biomarkers as compared to baseline in tumor tissue • Circulating tumor DNA fraction (mCRPC only) • Status of immune, molecular, disease-related, and other exploratory biomarkers in blood and archival and/or freshly obtained tumor tissue
	• To determine the immunogenicity of pembrolizumab in combination with nanrilkefusp alfa	• Incidence, titer, and time course of ADAs against pembrolizumab
	• (Population) PK of pembrolizumab in combination with nanrilkefusp alfa	• Concentrations of pembrolizumab over time
	• To further evaluate the antitumor efficacy of nanrilkefusp alfa in combination with pembrolizumab	• Overall survival (OS)

Study interventions	Patients will be treated with nanrilkefusp alfa 12 µg/kg subcutaneously on day 1 (±1 day for the cycle start), day 2, day 8, and day 9 in combination with pembrolizumab 200 mg intravenously (IV) on day 1 in 3-week cycles after all procedures and assessments have been completed. Pembrolizumab will be administered as an IV infusion via peripheral or central venous line within 30 minutes after the first dose (day 1) of nanrilkefusp alfa in each 3-week cycle.
Study population	Based on encouraging data on the combination therapy with efficacy signals as well as on pembrolizumab single-agent treatment, the following indications have been selected for this study: • Advanced and/or metastatic non-small cell lung cancer (NSCLC) with disease progression on or after an immune checkpoint inhibitor-containing regimen and a platinum-containing regimen, with no epidermal growth factor receptor (EGFR) or anaplastic lymphoma kinase (ALK) genomic tumor aberrations, and who are not amenable to curative treatment • Microsatellite instability-high (MSI-H) or mismatch repair deficient (dMMR) colorectal cancer that is unresectable or metastatic • Recurrent or metastatic cutaneous squamous cell carcinoma (cSCC) that is not curable by surgery or radiation and in second line if refractory or relapsed after a checkpoint inhibitor-containing regimen and radiotherapy is not feasible • Advanced hepatocellular carcinoma after recurrence or failure of an immune checkpoint inhibitor (not applicable in France) • Treatment-refractory mCRPC after recurrence or failure of docetaxel and prior treatment with abiraterone, enzalutamide, or any other androgen receptor-targeted agent • Advanced recurrent ovarian cancer after recurrence or failure on the last platinum-based therapy within 6 months
Study duration	The study will end one year after the last patient's last dose of study interventions or 3 years after the last patient's first dose of study interventions (whichever occurs earlier).
Estimated sample size	• NSCLC: 50 patients • MSI-H/dMMR colorectal cancer: 57 patients • cSCC: 57 patients • Advanced hepatocellular carcinoma: 55 patients (not applicable in France) • mCRPC: 51 patients • Recurrent ovarian cancer: 50 patients

Statistical considerations	**Interim analysis, stopping/discontinuation criteria**
	An analysis for futility will be performed when the sample size is considered enough for such analysis and for each indication, as described below in this section. Efficacy data and outputs will also be a part of the Independent Data Monitoring Committee (IDMC) review.
	Assuming a desired ORR, the futility analysis will be based on a comparison against a minimal ORR considered as both statistically and clinically relevant improvement as compared to the benchmark ORR. An 80% confidence interval (CI) using the exact method for the ORR (alpha = 0.2, alpha = 0.1 one-sided) will be used.
	If, for a certain indication, the ORR is:
	• lower than the minimal ORR, and
	• the 80% CI for the ORR does not include the minimal ORR,
	it will be concluded that the combination treatment is futile as compared to benchmark treatment and thus the indication will be discontinued. However, patients still on treatment in the indication can continue combination therapy if recommended by the IDMC.
	The criteria for conclusion of futility, for each indication, are defined as follows:
	• Indication 1 (NSCLC): Benchmark ORR = 15%; minimal ORR = 23%; desired ORR = 30.6%
	  ○ N = 21 patients if the number of responses is less than 2 (r < 2)
	• Indication 2 (MSI-H/dMMR colorectal cancer): Benchmark ORR = 44%; minimal ORR = 54.1%; desired ORR = 62.5%
	  ○ N = 12 patients if the number of responses is less than 4 (r < 4)
	• Indication 3 (cSCC): Benchmark ORR = 44%; minimal ORR = 54.1%; desired ORR = 62.5%
	  ○ N = 12 patients if the number of responses is less than 4 (r < 4)
	• Indication 4 (advanced hepatocellular carcinoma): Benchmark ORR = 17%; minimal ORR = 25.3%; desired ORR = 33% (not applicable in France)
	  ○ N = 19 patients if the number of responses is less than 2 (r < 2)
	• Indication 5 (mCRPC): Benchmark ORR = 5%; minimal ORR = 10.4%; desired ORR = 16.4%

	○ N = 35 patients if the number of responses is less than 1 (r < 1) • Indication 6 (recurrent ovarian cancer): Benchmark ORR = 15%; minimal ORR = 23%; desired ORR = 30.6% ○ N = 21 patients if the number of responses is less than 2 (r < 2) Additionally, the ORR will be evaluated on an ongoing basis (without stopping of recruitment). Other efficacy endpoints will be used as supportive information. **Statistical analyses** Analyses will be descriptive. The ORR will be presented with 95% CIs. Kaplan-Meier estimations of time-to-event data will be performed, together with an estimation of the median (if reached). If appropriate regarding the number of patients, descriptive statistics for other endpoints will be presented with 95% CIs.
Independent Data Monitoring Committee	An IDMC will be established for this study to safeguard the interest and safety of the patients participating in the study and provide independent review and assessment of the efficacy and safety data in a systematic manner. In addition, the IDMC will review safety data from the first 10-12 patients across all indications.

1.2 Schema



ALK, anaplastic lymphoma kinase; cSCC, cutaneous squamous cell carcinoma; dMMR, mismatch repair deficient; EGFR, epidermal growth factor receptor; mCRPC, metastatic castration-resistant prostate cancer; MSI-H, microsatellite instability-high; NSCLC, non-small cell lung cancer; ORR, objective response rate

1.3 Schedule of activities

1.3.1 Schedule of activities, nanrilkefusp alfa combined with pembrolizumab

Cycle	Screening Up to 28 days before day 1 of cycle 1	Cycle 1 and cycle 2						From cycle 3 onwards				End of treatment[1] Within 7 (+7) days after the last dose of nanrilkefusp alfa and/or pembrolizumab (whichever occurs later)	Follow-up
Visit		**Day 1**	**Day 2**	**Day 6**	**Day 8**	**Day 9**	**Day 13**	**Day 1**	**Day 2**	**Day 8**	**Day 9**		
Informed consent[2]	X												
Demography[3]	X												
Cancer[4] and medical history	X												
Height	X												
Pregnancy test	X (blood)	X (urine or blood)[5]						X (urine or blood)[5]					X (urine or blood)[6]
Physical examination	X	X[7]	X[7]	X	X[7]	X[7]	X	X[7]	X[7]	X[7]	X[7]	X	X[8]
Body weight	X	X[5]						X[5]				X	
Vital signs	X	X[9]	X[9]	X	X[9]	X[9]	X	X[9]	X[9]	X[9]	X[9]	X	X[8]
Urinalysis[10]	X	X[5]			X[5]			X[5]		X[5]		X	
Electrocardiography	X	X[5]						X[5]				X	
Left ventricular ejection fraction[11]	X	End of each second cycle (i.e., cycle 2, 4, 6…; allowed interval from day 9 of the current cycle to day 2 of the next cycle)										X	
Eastern Cooperative Oncology Group performance score	X	X[5]						X[5]				X	X[8]
Tumor assessment (CT/MRI/bone scan)	X[12]	Every 6 weeks (±2 weeks) starting from day 1 of cycle 1 until disease progression identified by the investigator, start of a new anti-cancer treatment, pregnancy of the patient, withdrawal of consent by the patient, or end of the study											
Nanrilkefusp alfa administration[13]		X	X		X	X		X	X	X	X		
Pembrolizumab administration[14]		Every 3 weeks (day 1) according to the prescribing information											
Tumor biopsy[15]	X	X											
Adverse events	X	X											X[8]
Concomitant medication/ non-drug therapies	X	X											X[8]
Survival information													X[16]

1. Criteria for discontinuation of study interventions are listed in section 7.1. In case nanrilkefusp alfa needs to be discontinued for reasons other than disease progression, pembrolizumab treatment can continue for up to a total of 35 administrations (approximately 2 years) in patients without disease progression or unacceptable toxicity. A schedule of activities for patients who continue pembrolizumab after discontinuation of nanrilkefusp alfa is shown in section 1.3.2. In case pembrolizumab needs to be discontinued, nanrilkefusp alfa treatment can continue until disease progression or unacceptable toxicity and the schedule of activities for patients receiving nanrilkefusp alfa combined with pembrolizumab will apply (not applicable in the Czech Republic). In the Czech Republic: In case pembrolizumab needs to be discontinued, nanrilkefusp alfa treatment will be discontinued as well.
2. No study-specific procedures are to be performed before ICF signature.
3. Age at screening and gender
4. Primary tumor location, histology/cytology, initial diagnosis date, lines of previous treatment, start and stop dates of the treatments before this study, any prior mutations/genetic analysis (e.g., EGFR, ALK mutations), and the date of the latest disease progression if not coinciding with the stop date
5. Before nanrilkefusp alfa administration
6. Every 30 (±2) days until 120 (±2) days after the last dose of pembrolizumab or 30 (±2) days after the last dose of nanrilkefusp alfa, whichever is later
7. Before nanrilkefusp alfa administration and afterwards as clinically required
8. Follow-up visits 30 (±2) days and 90 (+2) days after the last dose of nanrilkefusp alfa and/or pembrolizumab (whichever occurs later)
9. Before and after nanrilkefusp alfa administration as described in section 8.2.3
10. In case of proteinuria ≥100 mg/dL at screening, a 24-hour urine analysis will have to be performed (before the start of nanrilkefusp alfa treatment) to document 24-hour proteinuria levels and a urine test will continue during the treatment period. In case of increase of proteinuria with ≥300 mg/dL (at any time), a 24-hour urine analysis will be performed.
11. Using either echocardiography or MUGA; the method chosen needs to stay the same throughout the trial
12. Tumor scans performed as part of routine clinical management are acceptable for screening if they are of acceptable diagnostic quality and performed within 28 days of day 1 of cycle 1.
13. Time to be recorded
14. Time to be recorded; pembrolizumab will be administered within 30 minutes following nanrilkefusp alfa administration
15. A fresh biopsy must be taken during screening unless the biopsy cannot be obtained due to safety reasons or non-accessibility of the tumor site. If it is not possible to obtain a fresh biopsy, every effort should be taken to retrieve an archival biopsy. Archived, fixed tumor tissue may only be collected if taken preferentially after completion of the most recent systemic tumor therapy and within 12 months prior to the first dose of study treatment. Investigators are highly encouraged to obtain an optional biopsy at cycle 2 day 13 and at disease progression or at any other clinically relevant event if it does not expose the patient to an increased risk.
16. Contacts every 3 months (±2 weeks) during year 1 and then every 6 months (±2 weeks) until the end of the study

ALK, anaplastic lymphoma kinase; CT, computed tomography; EGFR, epidermal growth factor receptor; ICF, Informed Consent Form; MRI, magnetic resonance imaging; MUGA, multigated acquisition scanning

1.3.2 Schedule of activities, for patients who continue pembrolizumab after discontinuation of nanrilkefusp alfa

Visit	**Day 1** of each cycle	**End of treatment** Within 7 (+7) days after the last dose of pembrolizumab	**Follow-up**
Pregnancy test	X (urine or blood)[1]		X (urine or blood)[2]
Physical examination	X[1]	X	X[3]
Body weight	X[1]	X	
Vital signs	X[1]	X	X[3]
Urinalysis[4]	X[1]	X	
Electrocardiography	X[1]	X	
Left ventricular ejection fraction[5]		X	
Eastern Cooperative Oncology Group performance score	X[1]	X	X[3]
Tumor assessment (CT/MRI/bone scan)	Every 6 weeks (±2 weeks) starting from day 1 of cycle 1 until disease progression identified by the investigator, start of a new anti-cancer treatment, pregnancy of the patient, withdrawal of consent by the patient, or end of the study		
Pembrolizumab administration	Every 3 weeks (day 1) according to the prescribing information		
Adverse events	X		X[3]
Concomitant medication/non-drug therapies	X		X[3]
Survival information			X[6]

1. Before pembrolizumab administration
2. Every 30 (±2) days until 120 (±2) days after the last dose of pembrolizumab
3. Follow-up visits 30 (±2) days and 90 (+2) days after the last dose of pembrolizumab
4. In case of proteinuria ≥100 mg/dL at screening, a 24-hour urine analysis will have to be performed (before the start of nanrilkefusp alfa treatment) to document 24-hour proteinuria levels and a urine test will continue during the treatment period. In case of increase of proteinuria with ≥300 mg/dL (at any time), a 24-hour urine analysis will be performed.
5. Using either echocardiography or MUGA; the method chosen needs to stay the same throughout the trial
6. Contacts every 3 months (±2 weeks) during year 1 and then every 6 months (±2 weeks) until the end of the study

CT, computed tomography; MRI, magnetic resonance imaging; MUGA, multigated acquisition scanning

1.3.3 Collection of blood samples, nanrilkefusp alfa combined with pembrolizumab

All attempts should be made to collect the samples on time.

Cycle	Screening	Cycle 1						Cycle 2					Cycle 3				From cycle 4				End of treatment
Day		**1**	**2**	**6**	**8**	**9**	**13**	**1**	**2**	**6**	**8**	**9**	**1**	**2**	**8**	**9**	**1**	**2**	**8**	**9**	
Nanrilkefusp alfa administration[1]		X	X		X	X		X	X		X	X	X	X	X	X	X	X	X	X	
Pembrolizumab administration[2]		X						X					X				X				
Serum for nanrilkefusp alfa PK	See section 1.3.5																				
Serum for cytokines[3]		X	X		X	X		X	X		X	X									
Serum for nanrilkefusp alfa immunogenicity[4]		X						X									Cycle 4 only				X
Serum for pembrolizumab PK[5]		X						X									Cycles 4 and 8 only				X
Serum for pembrolizumab immunogenicity[6]		X						X									Cycle 4 only				X
Whole blood for RNA and DNA analysis[7]		X		X				X		X											
HIV, HBV, and HCV	X																				
Hematology (full complete blood count including absolute lymphocyte count)	X	X^{4}	X^{8}	X	X^{4}	X^{8}	X	X^{4}		X	X^{4}		X^{4}		X^{4}		X^{4}		X^{4}		X
Biochemistry (including C-reactive protein)	X	X^{4}	X^{8}	X		X^{8}	X	X^{4}		X		X^{8}	X^{4}		X^{4}		X^{4}		X^{4}		X
Creatinine clearance	X	X^{4}		X		X^{8}		X^{4}		X		X^{8}	X^{4}		X^{4}		X^{4}		X^{4}		
C-reactive protein					X^{4}				X^{8}		X^{4}										
Coagulation	X	X^{4}			X^{4}			X^{4}			X^{4}		X^{4}		X^{4}		X^{4}		X^{4}		X
TSH, T3 or free T3, free T4	X	X^{4}											X^{4}				$X^{4,9}$				X
Patients with mCRPC: PSA, alkaline phosphatase, lactate dehydrogenase, CTCs, circulating tumor DNA fraction	X	X^{4}											X^{4}				$X^{4,9}$				
Serum pregnancy	X																				

1. Time to be recorded
2. Time to be recorded; pembrolizumab will be administered within 30 minutes following nanrilkefusp alfa administration.
3. Before nanrilkefusp alfa administration and 4 hours (±30 min) after nanrilkefusp alfa administration
4. Within 1 day before nanrilkefusp alfa administration
5. Up to 24 hours before pembrolizumab infusion at cycles 1, 2, 4, and 8; within 30 minutes of the end of pembrolizumab infusion at cycles 1, 2, and 8; and at the End of treatment visit; sampling times to be recorded
6. Up to 24 hours before pembrolizumab infusion at cycles 1, 2, and 4; and at the End of treatment visit; sampling times to be recorded
7. Samples for RNA analysis: day 1 (before nanrilkefusp alfa administration) and day 6 of cycles 1 and 2; samples for DNA analysis: day 1 (before nanrilkefusp alfa administration) of cycle 1 only
8. On the day of dosing before nanrilkefusp alfa administration
9. Every other cycle

CTC, circulating tumor cell; HBV, hepatitis B; HCV, hepatitis C; mCRPC, metastatic castration-resistant prostate cancer; PK, pharmacokinetics; PSA, prostate-specific antigen; T3, triiodothyronine; T4, thyroxine; TSH, thyroid stimulating hormone

1.3.4 Collection of blood samples, for patients who continue pembrolizumab after discontinuation of nanrilkefusp alfa

All attempts should be made to collect the samples on time.

Cycle	From cycle 1	End of treatment
Day	**1**	
Pembrolizumab administration[1]	X	
Hematology	X[2]	X
Biochemistry	X[2]	X
Coagulation	X[2]	X
TSH, T3 or free T3, free T4	X[2,3]	X

1. Time to be recorded
2. Before pembrolizumab administration
3. Every other cycle

T3, triiodothyronine; T4, thyroxine; TSH, thyroid stimulating hormone

1.3.5 Nanrilkefusp alfa pharmacokinetic sampling

All attempts should be made to collect the samples on time.

Cycle, day	Time point (pre-dose or time after nanrilkefusp alfa administration)	12 patients per indication	Rest of the patients
Cycle 1 day 1	Pre-dose to nanrilkefusp alfa	X	
	30 min (±5 min)	X	
	1 h (±5 min)	X	
	2 h (±15 min)	X	
	4 h (±30 min)	X	X
	12 h (±2 h) (optional)	X	
Cycle 1 day 2	Pre-dose to nanrilkefusp alfa (-2 h [-15 min] to 0 h)	X	X
Cycle 1 day 8	Pre-dose to nanrilkefusp alfa (-2 h [-15 min] to 0 h)	X	
Cycle 1 day 9	Pre-dose to nanrilkefusp alfa (-2 h [-15 min] to 0 h)	X	
	2 h (±15 min)	X	
	4 h (±30 min)	X	X
Cycle 2 day 1	Pre-dose to nanrilkefusp alfa (-2 h [-15 min] to 0 h)	X	
	2 h (±15 min)	X	
	4 h (±30 min)	X	X
Cycle 2 day 9	Pre-dose to nanrilkefusp alfa (-2 h [-15 min] to 0 h)	X	
	2 h (±15 min)	X	
	4 h (±30 min)	X	X
Cycle 3 day 1	Pre-dose to nanrilkefusp alfa (-2 h [-15 min] to 0 h)	X	
	2 h (±15 min)	X	X

2 INTRODUCTION

2.1 Study rationale

The involvement of the immune system in the control of tumor growth is well accepted and immunotherapy is taking a major place in the treatment of cancers.[1] Current cancer immunotherapies that have been investigated for their efficacy include, among others, cytokine therapy and immune checkpoint inhibition. Emerging clinical data showed that cytokine therapy is safe and feasible in the outpatient setting and can be safely combined with immune checkpoint inhibitors.[2,3] We hypothesize that this combination leads to synergistic outcomes in the induction and/or maintenance of the antitumor responses by activation of both innate and adaptive cellular immune responses against tumor cells.

This basket study is planned based on results from pre-clinical studies of nanrilkefusp alfa (SOT101) and preliminary results from an ongoing clinical study of nanrilkefusp alfa (study SC103 [AURELIO-03, NCT04234113], a multicenter, open-label, phase 1/1b study to evaluate the safety and preliminary efficacy of SO-C101 [nanrilkefusp alfa] as monotherapy and in combination with pembrolizumab in patients with selected advanced/metastatic solid tumors). Study SC103 (AURELIO-03) shows that the combination of nanrilkefusp alfa and pembrolizumab can be safely administered. The safe recommended dose of nanrilkefusp alfa in combination with pembrolizumab is 12 µg/kg body weight on days 1, 2, 8, and 9 every 3 weeks, the same as for nanrilkefusp alfa monotherapy. Preliminary efficacy signals were seen both in monotherapy and even stronger in combination therapy.

2.2 Background

Therapy with recombinant interleukin (IL)-2, a T-cell and natural killer (NK)-cell growth factor, was one of the earliest successes in immunotherapy. IL-2 (International Nonproprietary Name: aldesleukin) was approved in some countries in the EU to treat metastatic renal cell carcinoma and by the US Food and Drug Administration (FDA) to treat metastatic renal cell carcinoma and malignant melanoma.[4,5]

IL-15 is a cytokine that has many overlapping functions like IL-2, including the ability to promote antitumor responses, but with distinct advantages over IL-2. While both IL-2 and IL-15 signal through the common IL-2 receptor (IL-2R) chains β and γ (IL-2Rβγ or IL-15 receptor [IL-15R] βγ) complex, which is responsible for intracellular signaling through the JAK/STAT, MAPK, and PI3K pathways in T cells and NK cells, only IL-2 engages the high-affinity IL-2R chain α (IL-2Rα). As IL-2Rα is expressed on regulatory T cells (Tregs), IL-15 does not induce the expansion of these immunosuppressive cells.[6-8] Because IL-15 can engage IL-2Rβγ without engaging IL-2Rα on Tregs, it is thought to have an improved therapeutic index compared with IL-2.[9-11]

Despite the promising preclinical responses of recombinant IL-15,[12] success in the clinical setting was limited by its short *in vivo* half-life. To date, 3 main modifications of IL-15 have been made to generate more potent soluble IL-15 agonists: 1) combining IL-15 with a soluble IL-15R chain α (IL-15Rα) or the sushi domain of IL-15Rα; 2) fusing IL-15Rα to the Fc portion of human IgG1; 3) mutating IL-15 to increase its affinity to IL-2Rβγ. All formulations of IL-15 agonists have demonstrated increased strength and duration of IL-15R signaling and subsequent enhanced antitumor immunity in nonclinical studies.

In order to make use of the enhanced biological activity of the IL-15/IL-15Rα complex for the treatment of cancer and to overcome the need for endogenous IL-15Rα or co-administration of IL-15 with IL-15Rα, we engineered nanrilkefusp alfa, a fusion protein which consists of the N-terminal sushi domain of human IL-15Rα covalently coupled via a linker of 20 amino acids to human IL-15. The biological activity of nanrilkefusp alfa stems from the function of the IL-15 component, which acts as a pleiotropic cytokine and plays a critical role in stimulating both innate and adaptive immune responses. Experimental data have indicated that the biological activity of IL-15 was enhanced when co-administered with recombinant soluble IL-15Rα. Thus, nanrilkefusp alfa, which fuses the IL-15 component to the IL-15Rα component, may be more potent than IL-15 alone and may benefit from an extended half-life.

Nanrilkefusp alfa promoted mobilization, expansion, and activation of human NK and CD8+ T cells in humanized mice and murine NK and CD8+ T cells in syngeneic mice. This NK- and CD8+ T-cell activation correlated with the potent anti-cancer therapeutic effects of nanrilkefusp alfa.[13,14]

For detailed background information on nanrilkefusp alfa refer to the Investigator's Brochure (IB).

Pembrolizumab is a potent humanized IgG4 monoclonal antibody (mAb) with high specificity of binding to the programmed cell death protein 1 (PD-1) receptor, thus inhibiting its interaction with programmed cell death ligand 1 (PD-L1) and programmed cell death ligand 2 (PD-L2). Based on preclinical *in vitro* data, pembrolizumab has high affinity and potent receptor blocking activity for PD-1. Pembrolizumab has an acceptable preclinical safety profile and is in clinical development as an intravenous (IV) immunotherapy for advanced malignancies. Keytruda® (pembrolizumab) is indicated for the treatment of patients across a number of indications. For more details on specific indications refer to the IB.

For detailed background information on pembrolizumab refer to the IB.

The importance of intact immune surveillance function in controlling outgrowth of neoplastic transformations has been known for decades.[15] Accumulating evidence shows a correlation between tumor-infiltrating lymphocytes in cancer tissue and favorable prognosis in various malignancies. In particular, the presence of CD8+ T cells and the ratio of CD8+ effector T cells/FoxP3+ Tregs correlates with improved prognosis and long-term survival in solid malignancies, such as ovarian, colorectal, and pancreatic cancer; hepatocellular carcinoma; malignant melanoma; and renal cell carcinoma. Tumor-infiltrating lymphocytes can be expanded *ex vivo* and reinfused, inducing durable objective tumor responses in cancers such as melanoma.[16,17]

The PD-1 receptor-ligand interaction is a major pathway hijacked by tumors to suppress immune control. The normal function of PD-1, expressed on the cell surface of activated T cells under healthy conditions, is to down-modulate unwanted or excessive immune responses, including autoimmune reactions. PD-1 (encoded by the gene *Pdcd1*) is an Ig superfamily member related to CD28 and cytotoxic T lymphocyte-associated protein 4 (CTLA-4) that has been shown to negatively regulate antigen receptor signaling upon engagement of its ligands (PD-L1 and/or PD-L2).[18,19]

The structure of murine PD-1 has been resolved.[20] PD-1 and its family members are type I transmembrane glycoproteins containing an Ig-variable type domain responsible for ligand binding and a cytoplasmic tail responsible for the binding of signaling molecules. The cytoplasmic tail of PD-1 contains 2 tyrosine-based signaling motifs, an immunoreceptor

tyrosine-based inhibition motif, and an immunoreceptor tyrosine-based switch motif. Following T-cell stimulation, PD-1 recruits the tyrosine phosphatases, SHP-1 and SHP-2, to the immunoreceptor tyrosine-based switch motif within its cytoplasmic tail, leading to the dephosphorylation of effector molecules such as CD3 zeta, protein kinase C-theta, and zeta-chain-associated protein kinase ZAP70, which are involved in the CD3 T-cell signaling cascade.[19,21-23] The mechanism by which PD-1 down-modulates T-cell responses is similar to, but distinct from, that of CTLA-4, because both molecules regulate an overlapping set of signaling proteins.[24,25] As a consequence, the PD-1/PD-L1 pathway is an attractive target for therapeutic intervention in various cancers.

Therapeutic studies in mouse models have shown that administration of antibodies blocking PD-1/PD-L1 interaction enhances infiltration of tumor-specific CD8+ T cells and ultimately leads to tumor rejection, either as a monotherapy or in combination with other treatment modalities.[26-32] Anti-mouse PD-1 or anti-mouse PD-L1 antibodies have demonstrated antitumor responses in models of squamous cell carcinoma, pancreatic carcinoma, melanoma, acute myeloid leukemia, and colorectal carcinoma.[20,29,31-33] In such studies, tumor infiltration by CD8+ T cells and increased interferon gamma, granzyme B, and perforin expression were observed, indicating that the mechanism underlying the antitumor activity of PD-1 checkpoint inhibition involved local infiltration and activation of effector T-cell function *in vivo*.[31] Experiments have confirmed the *in vivo* efficacy of anti-mouse PD-1 antibody as a monotherapy, as well as in combination with chemotherapy, in syngeneic mouse tumor models (see the IB).

2.3 Benefit/risk assessment

The risk assessment of nanrilkefusp alfa is based on nonclinical studies in addition to clinical experience from completed and ongoing trials with nanrilkefusp alfa as monotherapy and in combination with pembrolizumab. Clinical safety data from study SC103 (AURELIO-03) in patients with advanced/metastatic solid tumors for nanrilkefusp alfa in combination with pembrolizumab showed a similar profile as for nanrilkefusp alfa monotherapy; no relevant overlapping toxicities were reported (cut-off date 11Apr2022). The majority of treatment-emergent adverse events (TEAEs) were of grade 1 or 2. The most common TEAEs for nanrilkefusp alfa monotherapy / nanrilkefusp alfa in combination with pembrolizumab were pyrexia (70.0/81.0%), decreased lymphocyte count (66.7/47.6%), anemia (60.0/52.4%), injection site reaction (56.7/57.1%), and chills (50.0/61.9%). The most common TEAEs of grade >2 were decreased lymphocyte, anemia, and pyrexia. These TEAEs could be well managed and were self-limiting upon discontinuation of nanrilkefusp alfa.

Preliminary efficacy results in study SC103 (AURELIO-03) in advanced/metastatic solid tumors were encouraging, showing clinical benefit for most patients in the combination part of study, including patients pretreated with immune checkpoint inhibitors. Maximum activation of NK cells was observed already at low dose levels of nanrilkefusp alfa, and maximum activation of CD8+ T cells was reached at 9 to 12 µg/kg nanrilkefusp alfa.

More recent information about the known and expected benefits and risks and adverse events (AEs) that could be associated with the administration of nanrilkefusp alfa and pembrolizumab may be found in the nanrilkefusp alfa and pembrolizumab IBs.

In conclusion, the current nonclinical and clinical data on nanrilkefusp alfa in patients with advanced/metastatic solid tumors suggest a favorable benefit/risk ratio and justify further clinical development of nanrilkefusp alfa.

3 OBJECTIVES AND ENDPOINTS

Objective	Endpoint(s)
Primary	
• To estimate the antitumor efficacy of nanrilkefusp alfa in combination with pembrolizumab	• Objective response rate (ORR) according to Response Evaluation Criteria In Solid Tumors version 1.1 (RECIST 1.1)[34] in patients with measurable disease
Secondary	
• To assess the safety and tolerability of nanrilkefusp alfa in combination with pembrolizumab according to Common Terminology Criteria for Adverse Events (CTCAE) version 5.0[35]	• Type, frequency, and severity of TEAEs; AEs of special interest (AESIs); safety laboratory findings; vital signs; electrocardiography (ECG) findings
• To further evaluate the antitumor efficacy of nanrilkefusp alfa in combination with pembrolizumab	• ORR according to RECIST for immune-based therapeutics (iRECIST)[36] (iORR) in patients with measurable disease • Best overall response according to RECIST 1.1 (BOR) and iRECIST (iBOR) in patients with measurable disease • Duration of response according to RECIST 1.1 (DoR), iRECIST (iDoR), and Prostate Cancer Clinical Trials Working Group 3 (PCWG3)-modified RECIST 1.1 (metastatic castration-resistant prostate cancer [mCRPC] only)[37] • Clinical benefit rate according to RECIST 1.1 (CBR), iRECIST (iCBR), and PCWG3-modified RECIST 1.1 (mCRPC only) • Progression-free survival according to RECIST 1.1 (PFS), iRECIST (iPFS), and PCWG3-modified RECIST 1.1 (mCRPC only)

Objective	Endpoint(s)

	• Time to response according to RECIST 1.1 (TtR) and iRECIST (iTtR) in patients with measurable disease
	• mCRPC only:
	  ○ Circulating tumor cell (CTC) count conversion from ≥5 to <5 cells per 7.5 mL of blood
	  ○ Confirmed prostate-specific antigen (PSA) decline of ≥50%
	  ○ Time to confirmed PSA progression

• (Population) pharmacokinetics (PK) of nanrilkefusp alfa in combination with pembrolizumab	• Concentrations of nanrilkefusp alfa over time
• To determine the immunogenicity of nanrilkefusp alfa in combination with pembrolizumab	• Incidence, titer, and time course of anti-drug antibodies (ADAs) against nanrilkefusp alfa
Exploratory	
• To identify immune and molecular (including genomic, metabolic, and/or proteomic) biomarker(s) in archival and/or fresh tumor tissue and blood that may be indicative of clinical response/resistance, safety, pharmacodynamic (PD) activity, and/or the mechanism of action of nanrilkefusp alfa and pembrolizumab	• Changes in the expression of immune biomarkers as compared to baseline in tumor tissue • Circulating tumor DNA fraction (mCRPC only)[38] • Status of immune, molecular, disease-related, and other exploratory biomarkers in blood and archival and/or freshly obtained tumor tissue
• To determine the immunogenicity of pembrolizumab in combination with nanrilkefusp alfa	• Incidence, titer, and time course of ADAs against pembrolizumab
• (Population) PK of pembrolizumab in combination with nanrilkefusp alfa	• Concentrations of pembrolizumab over time
• To further evaluate the antitumor efficacy of nanrilkefusp alfa in combination with pembrolizumab	• Overall survival (OS)

4 STUDY DESIGN

4.1 Overall design

This is a phase 2, open-label, single-arm, multicenter study of nanrilkefusp alfa in combination with pembrolizumab to evaluate the efficacy and safety in patients with selected advanced/refractory solid tumors.

Participation of each patient will consist of the following study periods:

4.1.1 Screening

Patients will be screened within a period of not more than 28 days, which will start when the Informed Consent Form (ICF) has been signed and end before day 1 of cycle 1. A medical monitor will verify and confirm patients' eligibility.

4.1.2 Treatment

During the treatment period, the trial assessments will be performed as outlined in the Schedule of activities. Study interventions will be administered as described in section 6.

Patients will be treated with nanrilkefusp alfa together with pembrolizumab until any of the criteria for treatment discontinuation is met.

After termination of study interventions, patients will be evaluated at an End of treatment visit. This visit will be scheduled within 7 days (+7 days) after the patients' last dose of nanrilkefusp alfa and/or pembrolizumab (whichever occurs later).

Reasons for study intervention discontinuation are listed in section 7.

4.1.3 Follow-up

All patients will come to the clinic 30 (±2) days and 90 (+2) days after their last dose of nanrilkefusp alfa and/or pembrolizumab (whichever occurs later).

Patients will be followed up for survival every 3 months (±2 weeks) during year 1 and then every 6 months (±2 weeks) until the end of the study.

4.2 Scientific rationale for study design

Based on previous encouraging data on the combination therapy of nanrilkefusp alfa with pembrolizumab from study SC103 (AURELIO-03) with efficacy signals as well as on pembrolizumab single-agent treatment, the following indications have been selected for this study SC104 entitled "A phase 2, open-label, single-arm, multicenter study of SOT101 in combination with pembrolizumab to evaluate the efficacy and safety in patients with selected advanced/refractory solid tumors":

- Advanced and/or metastatic non-small cell lung cancer (NSCLC) with disease progression on or after an immune checkpoint inhibitor-containing regimen and a platinum-containing regimen, with no epidermal growth factor receptor (EGFR) or anaplastic lymphoma kinase (ALK) genomic tumor aberrations, and who are not amenable to curative treatment
    - Pembrolizumab was investigated in the following clinical trials:
        - KEYNOTE-010 (NCT01905657), a randomized, multicenter, open-label, active-controlled trial conducted in 1033 patients with metastatic NSCLC who had

progressed following platinum-containing chemotherapy, and if appropriate, targeted therapy for EGFR or ALK genomic tumor aberrations. In this trial, pembrolizumab showed superiority over second-line docetaxel.[39]

- KEYNOTE-024 (NCT02142738), a randomized, multicenter, open-label, active-controlled trial conducted in 305 patients with previously untreated NSCLC with a PD-L1 tumor proportion score of at least 50% and no sensitizing EGFR or ALK alterations. In this trial, pembrolizumab provided a durable, clinically meaningful long-term OS benefit versus chemotherapy as first-line therapy for metastatic NSCLC with PD-L1 tumor proportion score of at least 50%.[40]
- KEYNOTE-189 (NCT02578680), a randomized, multicenter, double-blind, active-controlled trial conducted in 616 patients with metastatic non-squamous NSCLC, regardless of PD-L1 tumor expression status, who had not previously received systemic therapy for metastatic disease and in whom there were no EGFR or ALK genomic tumor aberrations. In this trial, the addition of pembrolizumab to standard chemotherapy of pemetrexed and a platinum-based drug showed significantly longer OS and PFS than chemotherapy alone.[41]
- PROLUNG (NCT02574598), a randomized, open-label, crossover trial conducted in 78 patients with advanced NSCLC. In this trial, the combination of pembrolizumab plus docetaxel was well tolerated and substantially improved ORR and PFS in patients with advanced NSCLC who had previous progression after platinum-based chemotherapy, including NSCLC with EGFR variations.[42]

- Atezolizumab was investigated in IMpower110 (NCT02409342), a randomized, multicenter, open-label trial conducted in 572 patients with metastatic non-squamous or squamous NSCLC who had not previously received chemotherapy and who had PD-L1 expression on at least 1% of tumor cells or at least 1% of tumor-infiltrating immune cells. In this trial, atezolizumab showed superiority over first-line chemotherapy.[43]
- This indication is therefore chosen to test the combination of nanrilkefusp alfa and pembrolizumab as second-line or third-line treatment after an immune checkpoint inhibitor-containing regimen and a platinum-containing regimen.

- Microsatellite instability-high (MSI-H) or mismatch repair deficient (dMMR) colorectal cancer that is unresectable or metastatic
  - Pembrolizumab was investigated in KEYNOTE-177 (NCT02563002), a randomized, multicenter, open-label trial conducted in 307 patients with advanced MSI-H/dMMR colorectal carcinoma. In this trial, first-line pembrolizumab showed superiority over first-line chemotherapy.[44]
  - This indication is therefore chosen to test the ability of the combination of nanrilkefusp alfa and first-line pembrolizumab to improve pembrolizumab efficacy while maintaining safety (based on data from KEYNOTE-177).
- Recurrent or metastatic cutaneous squamous cell carcinoma (cSCC) that is not curable by surgery or radiation and in second line if refractory or relapsed after a checkpoint inhibitor-containing regimen and radiotherapy is not feasible
  - Pembrolizumab was investigated in KEYNOTE-629 (NCT03284424), a single-arm trial conducted in 105 patients with recurrent or metastatic cSCC. In this trial,

pembrolizumab demonstrated effective antitumor activity; clinically meaningful, durable responses; and acceptable safety in primarily elderly patients with recurrent or metastatic cSCC.[45]

- Cemiplimab was investigated in a single-arm, multicenter trial (NCT02760498) in 78 patients with locally advanced cSCC for whom there was no widely accepted standard of care. In this trial, cemiplimab showed antitumor activity and an acceptable safety profile.[46]
- Patients treated with nanrilkefusp alfa showed responses in study SC103 (AURELIO-03) after resistance to or failure on immune checkpoint inhibitors.
- This indication is therefore chosen to test the ability of the combination of nanrilkefusp alfa and pembrolizumab to improve pembrolizumab efficacy while maintaining safety as first-line treatment (based on data from KEYNOTE-629) and as second-line treatment if refractory or relapsed after an immune checkpoint inhibitor-containing regimen (based on data from study SC103 [AURELIO-03]).

- Advanced hepatocellular carcinoma after recurrence or failure of an immune checkpoint inhibitor (not applicable in France)
  - Pembrolizumab was investigated in KEYNOTE-224 (NCT02702414), a single-arm, multicenter trial in 156 patients with hepatocellular carcinoma who had disease progression on or after sorafenib or who had not received prior systemic therapy. In this trial, pembrolizumab was effective and tolerable in patients who had previously been treated with sorafenib. First-line pembrolizumab monotherapy provided durable antitumor activity and promising OS.[47,48]
  - The combination of atezolizumab and bevacizumab was investigated in IMbrave150 (NCT03434379), a randomized, multicenter, open-label trial in 558 patients with unresectable hepatocellular carcinoma who had not previously received systemic treatment. Atezolizumab plus bevacizumab resulted in better outcomes than sorafenib.[49]
  - This indication is therefore chosen to investigate the combination of nanrilkefusp alfa and pembrolizumab as second- to fourth-line treatment in patients who have received immune checkpoint inhibitor therapy.
- Treatment-refractory mCRPC after recurrence or failure of docetaxel and prior treatment with abiraterone, enzalutamide, or any other androgen receptor-targeted agent
  - Pembrolizumab was investigated in KEYNOTE-199 (NCT02787005), a multicohort, open-label trial in 370 patients with mCRPC. In this trial, pembrolizumab monotherapy showed antitumor activity in a subset of patients with RECIST-measurable and bone-predominant mCRPC previously treated with docetaxel and targeted endocrine therapy. Observed responses seemed to be durable, and OS estimates were encouraging.[50]
  - This indication is therefore chosen to test the ability of the combination of nanrilkefusp alfa and pembrolizumab as second- to fifth-line treatment to improve pembrolizumab efficacy (based on data from KEYNOTE-199).

- Advanced recurrent ovarian cancer after recurrence or failure on the last platinum-based therapy within 6 months
  - Pembrolizumab was investigated in KEYNOTE-100 (NCT02674061), a single-arm, multicenter study conducted in 376 patients with advanced recurrent ovarian cancer. In this trial, single-agent pembrolizumab showed modest activity. Higher PD-L1 expression was correlated with higher response.[51]
  - This indication is therefore chosen to investigate the combination of nanrilkefusp alfa and pembrolizumab as second- to fifth-line treatment for advanced recurrent ovarian cancer (based on data from KEYNOTE-100).

4.3 Justification for dose

4.3.1 Nanrilkefusp alfa

The planned dose of nanrilkefusp alfa for this study is 12 µg/kg body weight on days 1, 2, 8, and 9 every 3 weeks, which is the recommended phase 2 dose defined based on the totality of data generated in study SC103 (AURELIO-03) entitled "A multicenter open-label phase 1/1b study to evaluate the safety and preliminary efficacy of SO-C101 as monotherapy and in combination with pembrolizumab in patients with selected advanced/metastatic solid tumors".

4.3.2 Pembrolizumab

The planned dose of pembrolizumab for this study is 200 mg every 3 weeks (Q3W). Based on the totality of data generated in the Keytruda development program, 200 mg Q3W is an appropriate dose of pembrolizumab for adults across all indications. As outlined below, this dose is justified by:

- Clinical data from 8 randomized studies in melanoma and NSCLC indications demonstrating flat dose- and exposure-efficacy relationships from 2 mg/kg Q3W to 10 mg/kg every 2 weeks (Q2W) representing an approximate 5- to 7.5-fold exposure range (refer to the IB)
- Population PK analysis showing that both fixed dosing and weight-based dosing provides similar control of PK variability with considerable overlap in the distributions of exposures, supporting suitability of 200 mg Q3W
- Clinical data showing meaningful improvement in benefit-risk including OS at 200 mg Q3W across multiple indications
- Pharmacology data showing full target saturation in both systemic circulation (inferred from PK data) and tumor (inferred from physiologically-based PK analysis) at 200 mg Q3W

Among the 8 randomized dose-comparison studies, a total of 2262 patients were enrolled with melanoma and NSCLC, covering different disease settings (treatment naïve, previously treated, PD-L1 enriched, and all-comers) and different treatment settings (monotherapy and in combination with chemotherapy). Five studies compared 2 mg/kg Q3W versus 10 mg/kg Q2W (KN001 cohort B2, KN001 cohort D, KN002, KN010, and KN021), and 3 studies compared 10 mg/kg Q3W versus 10 mg/kg Q2W (KN001 cohort B3, KN001 cohort F2 and KN006). All of these studies demonstrated flat dose- and exposure-response relationships across the doses studied representing an approximate 5- to 7.5-fold difference in exposure. The 2 mg/kg (or

200 mg fixed-dose) Q3W provided similar responses to the highest doses studied. Subsequently, flat dose-exposure-response relationships were also observed in other tumor types including head and neck cancer, bladder cancer, gastric cancer, and classical Hodgkin lymphoma, confirming 200 mg Q3W as the appropriate dose independent of the tumor type. These findings are consistent with the mechanism of action of pembrolizumab, which acts by interaction with immune cells, and not via direct binding to cancer cells.

Additionally, pharmacology data clearly show target saturation at 200 mg Q3W. First, PK data in KN001 evaluating target-mediated drug disposition conclusively demonstrated saturation of PD-1 in systemic circulation at doses much lower than 200 mg Q3W. Second, a physiologically-based PK analysis was conducted to predict tumor PD-1 saturation over a wide range of tumor penetration and PD-1 expression. This evaluation concluded that pembrolizumab at 200 mg Q3W achieves full PD-1 saturation in both blood and tumor.

Finally, population PK analysis of pembrolizumab, which characterized the influence of body weight and other patient covariates on exposure, has shown that the fixed-dosing provides similar control of PK variability as weight-based dosing, with considerable overlap in the distribution of exposures from the 200 mg Q3W fixed dose and 2 mg/kg Q3W dose. Supported by these PK characteristics and given that fixed-dose has advantages of reduced dosing complexity and reduced potential of dosing errors, the 200 mg Q3W fixed-dose was selected for evaluation across all pembrolizumab protocols.

4.4 End of study definition

A patient is considered to have completed the study if s/he has completed all phases of the study including the last follow-up contact.

The study will end one year after the last patient's last dose of study interventions or 3 years after the last patient's first dose of study interventions (whichever occurs earlier).

The sponsor reserves the right to discontinue the study for medical reasons or any other reason at any time. If the study is prematurely terminated or suspended, the sponsor will promptly inform the investigators/institutions, regulatory agencies, and institutional review boards (IRBs)/independent ethics committees (IECs; in the US), ethics committees (ECs; in the EU), or equivalent committees (outside the US and EU) of the termination or suspension and the reason(s) for the termination or suspension.

The investigator reserves the right to discontinue the study should his/her judgment so dictate. If the investigator terminates or suspends the study without prior agreement of the sponsor, the investigator should inform the institution where applicable, and the investigator/institution should promptly inform the sponsor and the IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) and provide the sponsor and the IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) with a detailed written explanation of the termination or suspension.

5 STUDY POPULATION

5.1 Inclusion criteria

Patients will be eligible to be included in the study only if all of the following criteria apply:

Type of patients

1. Be ≥18 years of age on the day of signing informed consent
2. Ability to understand and sign written informed consent to participate in the study
3. Provides written informed consent for the study

Disease characteristics

4. Patients with the following histologically or cytologically confirmed solid tumor indications and line of treatment:
    - NSCLC
        - For the second-line or third-line treatment of patients with advanced and/or metastatic NSCLC with disease progression on or after an immune checkpoint inhibitor-containing regimen and a platinum-containing regimen, with no EGFR or ALK genomic tumor aberrations, and who are not amenable to curative treatment
    - Colorectal cancer
        - For the first-line treatment of patients with unresectable or metastatic MSI-H/dMMR colorectal cancer
    - cSCC
        - For the first-line treatment of patients with recurrent or metastatic cSCC or second line if refractory or relapsed after an immune checkpoint inhibitor-containing regimen, and in whom radiotherapy is not feasible (based on data from study SC103 [AURELIO-03])
    - Advanced hepatocellular carcinoma (not applicable in France)
        - For the second- to fourth-line treatment of patients with advanced hepatocellular carcinoma having progressed on or after a checkpoint inhibitor-containing regimen
    - mCRPC
        - For the second- to fifth-line treatment of patients with mCRPC after recurrence or failure of docetaxel and prior treatment with abiraterone, enzalutamide, or any other androgen receptor-targeted agent
    - Ovarian cancer
        - For the second- to fifth-line treatment of patients with advanced ovarian cancer after recurrence or failure on the last platinum-based therapy within 6 months
5. Have measurable disease per RECIST 1.1 as assessed by the local site investigator/radiology; lesions situated in a previously irradiated area are considered measurable if progression has been demonstrated in such lesions

mCRPC: Patients with both measurable and non-measurable disease will be enrolled. At least 35 patients with measurable disease will be enrolled. Patients with no measurable disease and only widespread bone disease must have a CTC count of ≥5 cells per 7.5 mL of blood.

6. Availability of tumor tissue from a fresh biopsy at screening unless the biopsy cannot be obtained due to safety reasons or non-accessibility of the tumor site. If it is not possible to obtain a fresh biopsy, every effort should be taken to retrieve an archival biopsy. Archived, fixed tumor tissue may only be collected if taken preferentially after completion of the most recent systemic tumor therapy and within 12 months prior to the first dose of study treatment.
7. Performance status: Eastern Cooperative Oncology Group (ECOG) performance score 0-1
8. Must have recovered from all AEs (except alopecia) due to previous therapies to grade ≤1 toxicity (excluding alopecia) or have stable grade 2 neuropathy

Organ function

Have adequate organ function as defined below. Specimens must be collected within 10 days prior to the start of study interventions.

9. Hematology:
   - 9.1. Absolute neutrophil count ≥1500/µL
   - 9.2. Platelets ≥100 000/µL
   - 9.3. Hemoglobin ≥9.0 g/dL (criteria must be met without packed red blood cell transfusion within the prior 2 weeks; patients can be on a stable dose of erythropoietin [≥ 3 months])
10. Renal function: Creatinine clearance as measured by glomerular filtration rate ≥30 mL/min using Cockcroft-Gault equation
11. Hepatic function: ALT/AST ≤2.5× upper limit of normal (ULN) and total bilirubin ≤1.5×ULN or direct bilirubin ≤ ULN in patients without liver metastasis (benign hereditary hyperbilirubinemias, e.g., Gilbert's syndrome, are permitted if total bilirubin is ≤3 mg/dL). In patients with liver metastasis, ALT/AST ≤5×ULN is allowed but total bilirubin must be ≤2×ULN.
12. Prothrombin time and activated partial thromboplastin time ≤1.5×ULN

Hepatitis

13. A locally performed hepatitis B (HBV) test is required during screening. Patients who are HBV surface antigen positive are eligible if they have received HBV anti-viral therapy for at least 4 weeks and have undetectable HBV viral load before study entry (ICF signature). Patients should remain on anti-viral therapy throughout study treatment and follow local guidelines for HBV anti-viral therapy post completion of study interventions.
14. A locally performed hepatitis C (HCV) test is required during screening. Patients with history of HCV infection are eligible if HCV viral load is undetectable at screening. Patients must have completed anti-viral therapy at least 4 weeks before study entry (ICF signature).

Special requirements for female patients and contraception

15. A female patient is eligible to participate if she is not pregnant, not breastfeeding, and one of the following conditions applies:
    15.1. Not a woman of childbearing potential (WOCBP). A WOCBP is defined as fertile, following menarche and until becoming post-menopausal unless permanently sterile. Permanent sterilization methods include hysterectomy, bilateral salpingectomy, and bilateral oophorectomy. A postmenopausal state is defined as no menses for 12 months without an alternative medical cause. A high follicle stimulating hormone level in the postmenopausal range may be used to confirm a post-menopausal state in women not using hormonal contraception or hormonal replacement therapy. However, in the absence of 12 months of amenorrhea, a single follicle stimulating hormone measurement is insufficient.
    15.2. A WOCBP who agrees to use a highly effective contraceptive method during the treatment period and for at least 120 days after the last dose of pembrolizumab or at least 30 days after the last dose of nanrilkefusp alfa, whichever is later
    - WOCBP can only be included after a negative serum pregnancy test at screening.
    - Highly effective contraception includes:
        - Combined (estrogen- and progestogen-containing) hormonal contraception associated with inhibition of ovulation:
            - Oral
            - Intravaginal
            - Transdermal
        - Progestogen-only hormonal contraception associated with inhibition of ovulation:
            - Oral
            - Injectable
            - Implantable
        - Intrauterine device
        - Intrauterine hormone-releasing system
        - Bilateral tubal occlusion
        - Vasectomized partner provided the partner is the sole sexual partner of the WOCBP trial participant and that the vasectomized partner has received medical assessment of the surgical success
        - Sexual abstinence defined as refraining from heterosexual intercourse during the entire treatment period and for at least 120 days after the last dose of pembrolizumab or at least 30 days after the last dose of nanrilkefusp alfa, whichever is later. The reliability of sexual abstinence needs to be evaluated in relation to the duration of the clinical trial and the preferred and usual lifestyle of the patient.
16. Male patients must agree to use a condom during the treatment period and for at least 120 days after the last dose of pembrolizumab or at least 30 days after the last dose of nanrilkefusp alfa, whichever is later.

5.2 Exclusion criteria

Patients will be excluded from the study if any of the following criteria apply:

Prior/concomitant therapy

1. Has received prior therapy with an anti-PD-1, anti-PD-L1, or anti-PD-L2 agent or with an agent directed to another stimulatory or co-inhibitory T-cell receptor (e.g., cytotoxic T-lymphocyte antigen 4, CD134 [OX40], CD137), and was discontinued from that treatment due to a grade ≥3 AE
2. Prior exposure to drugs that are agonists of IL-2 or IL-15
3. Prior systemic anti-cancer therapies, including investigational agents, before the first dose of study medication (day 1 of cycle 1):
   - 3.1. Less than 4 weeks for systemic chemotherapy and immuno-oncology therapies; and for tyrosine kinase inhibitors 4 weeks or 5 half-lives (whichever is shorter)
   - 3.2. Less than 4 weeks from major surgeries and not recovered adequately from the procedure and/or any complications from the surgery
4. Has received prior radiotherapy within 2 weeks of the start of study interventions or have had a history of radiation pneumonitis.

   Note: Patients must have recovered from all radiation-related toxicities and not require corticosteroids. A 1-week washout is permitted for palliative radiation (≤2 weeks of radiotherapy) to non-central nervous system disease.
5. NSCLC indication only: Has received radiation therapy to the lung that is >30 Gy within 6 months of the first dose of study interventions
6. Has received a live or live-attenuated vaccine within 30 days prior to the first dose of study interventions

Prior/concurrent clinical study experience

7. Is currently participating in or has participated in a study of an investigational agent or has used an investigational device within 4 weeks or 5 half-lives (whichever shorter) before study entry (ICF signature). Patients who have entered the follow-up phase of an investigational study may participate as long as it has been 4 weeks or 5 half-lives (whichever shorter) after the last dose of the previous investigational agent.

Medical conditions

8. Clinically significant cardiac abnormalities including prior history of any of the following:
   - 8.1. Cardiomyopathy, with left ventricular ejection fraction ≤50% at screening
   - 8.2. Congestive heart failure of New York Heart Association grade ≥2
   - 8.3. History of clinically significant (i.e., active) atherosclerotic cardiovascular disease, specifically myocardial infarction, unstable angina, cerebrovascular accident within 6 months prior to the first dose of study interventions, and any clinically significant history of coronary heart disease and clinically significant artery disease within the past 5 years
   - 8.4. Prolongation of QTcF >450 msec

8.5. Clinically significant cardiac arrythmia that cannot be controlled with adequate medication

9. Uncontrolled hypertension defined as systolic blood pressure >160 mmHg, diastolic blood pressure >110 mmHg. Patients with uncontrolled hypertension should be medically managed on a stable regimen to control hypertension prior to study entry (ICF signature).
10. Has undergone prior allogeneic hematopoietic stem cell transplantation within the last 5 years. Patients who have had a transplant more than 5 years ago are eligible as long as there are no symptoms of graft versus host disease.
11. Has had an allogeneic tissue/solid organ transplant
12. Has a diagnosis of immunodeficiency or is receiving chronic systemic steroid therapy (in dosing exceeding 10 mg daily of prednisone equivalent) or any other form of immunosuppressive therapy within 7 days prior to the first dose of study interventions
13. History of or serology positive for HIV. A locally performed HIV test is required during screening.
14. Has a known additional malignancy that is progressing or has required active treatment within the past 5 years. Patients with basal cell carcinoma of the skin or carcinoma *in situ* excluding carcinoma *in situ* of bladder that have undergone potentially curative therapy are not excluded.
15. Has known active central nervous system metastases and/or carcinomatous meningitis. Patients with previously treated brain metastases may participate provided they are radiologically stable, i.e., without evidence of progression for at least 4 weeks by repeat imaging (note that the repeat imaging should be performed during study screening), clinically stable and without requirement of steroid treatment for at least 14 days prior to the first dose of study interventions.
16. Has severe hypersensitivity (grade ≥3) to pembrolizumab and/or any of its excipients
17. Has an active autoimmune disease that has required systemic treatment in the past 2 years (i.e., with use of disease-modifying agents, corticosteroids, or immunosuppressive drugs). Replacement therapy (e.g., thyroxine, insulin, or physiologic corticosteroid replacement therapy for adrenal or pituitary insufficiency) is not considered a form of systemic treatment and is allowed.
18. Has a history of (non-infectious) pneumonitis/interstitial lung disease that required steroids or has current pneumonitis/interstitial lung disease
19. Has an active infection requiring systemic therapy
20. Has a history or current evidence of any condition, therapy, or laboratory abnormality, or other circumstance that might confound the results of the study or interfere with the patient's participation for the full duration of the study, such that it is not in the best interest of the patient to participate, in the opinion of the treating investigator
21. Has a known psychiatric or substance abuse disorder that would interfere with the patient's ability to cooperate with the requirements of the study

5.3 Lifestyle considerations

5.3.1 Meals and dietary restrictions

Patients should maintain a normal diet unless modifications are required to manage an AE such as diarrhea, nausea, or vomiting.

5.3.2 Contraception

Nanrilkefusp alfa and pembrolizumab may have adverse effects on a fetus *in utero*. Furthermore, it is not known if nanrilkefusp alfa and pembrolizumab have transient adverse effects on the composition of sperm.

Patients should be informed that taking the study medication may involve unknown risks to the fetus (unborn baby) if pregnancy were to occur during the study. If there is any question that a patient of childbearing potential will not reliably comply with the requirements for contraception, that patient should not be entered into the study.

Please also see section 5.1.

5.3.3 Use in nursing women

It is unknown whether nanrilkefusp alfa and pembrolizumab are excreted in human milk. Since many drugs are excreted in human milk, and because of the potential for serious adverse reactions in the nursing infant, patients who are breastfeeding are not eligible for enrollment.

Please also see section 5.1.

5.4 Screen failures

Screen failures are defined as patients who consent to participate in the clinical study but are not subsequently entered into the study. A minimal set of screen failure information is required to ensure transparent reporting of screen failure patients to meet the Consolidated Standards of Reporting Trials publishing requirements and to respond to queries from regulatory authorities. Minimal information includes demography, reason for screen failure, and AE/serious AE (SAE) as listed in section 8.3.2.

Patients may be rescreened once.

5.5 Criteria for temporarily delaying enrollment/administration of study intervention

See section 6.5.

6 STUDY INTERVENTIONS AND CONCOMITANT THERAPY

Study intervention is defined as any investigational intervention(s), marketed product(s), placebo, or medical device(s) intended to be administered to a study patient according to the study Protocol.

6.1 Study interventions administered

The interventions to be administered in this study are listed in Table 6.1.

Patients will be treated with nanrilkefusp alfa 12 µg/kg subcutaneously on day 1 (±1 day for the cycle start), day 2, day 8, and day 9 in combination with pembrolizumab 200 mg IV on day 1 in 3-week cycles after all procedures and assessments have been completed.

Pembrolizumab will be administered as an IV infusion via peripheral or central venous line within 30 minutes after the first dose (day 1) of nanrilkefusp alfa in each 3-week cycle.

Sites should make every effort to target pembrolizumab infusion timing to be as close to 30 minutes as possible. However, given the variability of infusion pumps from site to site, a window between -5 minutes and +10 minutes is permitted (i.e., infusion time is 30 minutes [-5 min/+10 min]).

The Pharmacy Manual contains specific instructions for the preparation of the nanrilkefusp alfa dose and pembrolizumab infusion and administration of nanrilkefusp alfa and pembrolizumab.

Table 6.1: Study interventions

Intervention name	Dosage formulation	Unit dose strength(s)	Dosage level(s)	Route of administration	Regimen/ treatment period	Sourcing
Product code: SOT101 International Nonproprietary Name: Nanrilkefusp alfa	Solution for injection	1.3 mg/vial	12 µg/kg	Subcutaneous	Administration on day 1 (±1 day for the cycle start), day 2, day 8, and day 9 of each 3-week cycle	Provided centrally by the sponsor
Pembrolizumab	Solution for infusion	100 mg/vial	200 mg	IV infusion via peripheral or central venous line	Administration within 30 minutes after the first dose of nanrilkefusp alfa (day 1) of each 3-week cycle	Provided centrally by the sponsor

6.2 Preparation, handling, storage, accountability

The investigator or designee must confirm appropriate temperature conditions have been maintained during transit for all study interventions received and any discrepancies are reported and resolved before use of the study interventions.

Only patients enrolled in the study may receive study interventions and only authorized site staff may supply or administer study interventions. All study interventions must be stored in a secure, environmentally controlled, and monitored (manual or automated) area in accordance with the labeled storage conditions with access limited to the investigator and authorized site staff.

The investigator, institution, or the head of the medical institution (where applicable) is responsible for study intervention accountability, reconciliation, and record maintenance (i.e., receipt, reconciliation, and final disposition records).

Further guidance and information for the final disposition of unused study interventions are provided in the Pharmacy Manual.

6.3 Measures to minimize bias: randomization and blinding

This single-arm clinical trial will not be blinded.

6.4 Study intervention compliance

Treatment for each patient will be recorded during the study.

6.5 Dose modification and toxicity management

6.5.1 Dose modification and toxicity management for nanrilkefusp alfa

Dose modification and toxicity management for nanrilkefusp alfa for safety reasons will be discussed on a case-by-case basis between the site and the sponsor's medical monitor.

Transient pyrexia is a frequent AE after nanrilkefusp alfa injection. The use of, e.g., ibuprofen, and other non-steroid premedication can be considered according to local standard practice for prevention and treatment of fever.

Recommendations for clinical management of cytokine release syndrome are provided in section 6.5.3.

To minimize the risk for injection site reaction, subcutaneous injection sites should be rotated to different areas of the body (upper and lower extremities, each of the 4 quadrants of the abdomen).

To minimize the risks associated with shortening of the QT interval, vital signs and ECG must be closely monitored as per Protocol and institutional guidelines.

Liver function tests for nanrilkefusp alfa toxicity evaluation in this study will include AST, ALT, total bilirubin, and alkaline phosphatase.

Nanrilkefusp alfa-related hepatic toxicities should be managed as shown in Table 6.2.

Table 6.2: Management of nanrilkefusp alfa-related hepatic toxicities

Nanrilkefusp alfa-related hepatic toxicity	Severity	Nanrilkefusp alfa dose modification
AST increased or ALT increased, or blood bilirubin increased	Grade 2 with AST or ALT >3 to ≤5×ULN or total bilirubin >1.5 to ≤3×ULN	Withhold nanrilkefusp alfa Resume nanrilkefusp alfa if increased values return to grade ≤1
	Grade ≥3 with AST or ALT >5×ULN or total bilirubin >3×ULN	Withhold nanrilkefusp alfa Resume nanrilkefusp alfa at 9 µg/kg if increased values return to grade ≤1 Permanently discontinue nanrilkefusp alfa on second occurrence of grade ≥3 hepatic event

Please also refer to the IB, section 6.6 (Management guidelines for identified and potential risks).

6.5.2 Dose modification and toxicity management for pembrolizumab

6.5.2.1 Dose modification and toxicity management for immune-related AEs associated with pembrolizumab

AEs associated with pembrolizumab combination exposure, including coadministration with additional compounds, may represent an immune-related response. These immune-related AEs (irAEs) may occur shortly after the first dose or several months after the last dose of pembrolizumab combination treatment and may affect more than one body system simultaneously. Therefore, early recognition and initiation of treatment is critical to reduce complications. Based on existing clinical study data, most irAEs were reversible and could be managed with interruptions of pembrolizumab combination treatment, administration of corticosteroids, and/or other supportive care. For suspected irAEs, ensure adequate evaluation to confirm etiology or exclude other causes. Additional procedures or tests such as bronchoscopy, endoscopy, skin biopsy may be included as part of the evaluation. Dose modification and toxicity management guidelines for irAEs associated with pembrolizumab are provided in Table 6.3.

6.5.2.1.1 Attribution of toxicity

When study interventions are administered in combination, attribution of an AE to a single component is likely to be difficult. Therefore, while the investigator may attribute a toxicity event to the combination, to nanrilkefusp alfa alone, or to pembrolizumab alone, for AEs listed in Table 6.3, both interventions must be held according to the criteria in Table 6.3 and these cases should be discussed with the sponsor's medical monitor.

6.5.2.1.2 Holding study interventions

When study interventions are administered in combination, if the AE is considered immune-related, both interventions should be held according to recommended dose modifications.

6.5.2.1.3 Restarting study interventions

Patients may not have any dose modifications (no change in dose or schedule) of pembrolizumab in this study, as described in Table 6.3.

If the toxicity does not resolve or the criteria for resuming treatment are not met, the patient must be discontinued from all study interventions.

If the toxicities do resolve and conditions are aligned with what is defined in Table 6.3, the combination of nanrilkefusp alfa and pembrolizumab may be restarted at the discretion of the investigator. In these cases where the toxicity is attributed to the combination or to nanrilkefusp alfa alone, re-initiation of pembrolizumab as a monotherapy may be considered after communication with and agreement by the sponsor.

Table 6.3: Dose modification and toxicity management guidelines for irAEs associated with pembrolizumab

General instructions:

1. Severe and life-threatening irAEs should be treated with IV corticosteroids followed by oral steroids. Other immunosuppressive treatment should begin if the irAEs are not controlled by corticosteroids.
2. Pembrolizumab must be permanently discontinued if the irAE does not resolve or the corticosteroid dose is not ≤10 mg/day within 12 weeks of the last pembrolizumab treatment.
3. Corticosteroid taper should begin when the irAE is grade ≤1 and continue at least 4 weeks.
4. If pembrolizumab has been withheld, pembrolizumab may resume after the irAE decreased to grade ≤1 after corticosteroid taper.

irAEs	**Toxicity grade (CTCAE version 5.0)**	**Action with pembrolizumab**	**Corticosteroid and/or other therapies**	**Monitoring and follow-up**
Pneumonitis	Grade 2	Withhold	Administer corticosteroids (initial dose of 1 to 2 mg/kg prednisone or equivalent) followed by taper Add prophylactic antibiotics for opportunistic infections	Monitor patients for signs and symptoms of pneumonitis Evaluate patients with suspected pneumonitis with radiographic imaging and initiate corticosteroid treatment
	Recurrent grade 2, grade 3, or grade 4	Permanently discontinue		

irAEs	Toxicity grade (CTCAE version 5.0)	Action with pembrolizumab	Corticosteroid and/or other therapies	Monitoring and follow-up
Diarrhea/colitis	Grade 2 or grade 3	Withhold	Administer corticosteroids (initial dose of 1 to 2 mg/kg prednisone or equivalent) followed by taper	Monitor patients for signs and symptoms of enterocolitis (i.e., diarrhea, abdominal pain, blood, or mucus in stool with or without fever) and of bowel perforation (i.e., peritoneal signs and ileus) Patients with grade ≥2 diarrhea suspecting colitis should consider gastrointestinal consultation and performing endoscopy to rule out colitis Patients with diarrhea/colitis should be advised to drink liberal quantities of clear fluids. If sufficient oral fluid intake is not feasible, fluid and electrolytes should be substituted via IV infusion
	Recurrent grade 3 or grade 4	Permanently discontinue		
AST or ALT elevation or increased bilirubin	Grade 2 [a]	Withhold	Administer corticosteroids (initial dose of 0.5 to 1 mg/kg prednisone or equivalent) followed by taper	Monitor with liver function tests (consider weekly or more frequently until liver enzyme value returned to baseline or is stable)
	Grade 3 [b] or grade 4 [c]	Permanently discontinue	Administer corticosteroids (initial dose of 1 to 2 mg/kg prednisone or equivalent) followed by taper	

irAEs	Toxicity grade (CTCAE version 5.0)	Action with pembrolizumab	Corticosteroid and/or other therapies	Monitoring and follow-up
T1DM or hyperglycemia	New onset T1DM or grade 3 or 4 hyperglycemia associated with evidence of β-cell failure	Withhold [d]	Initiate insulin replacement therapy for patients with T1DM Administer antihyperglycemics in patients with hyperglycemia	Monitor patients for hyperglycemia or other signs and symptoms of diabetes
Hypophysitis	Grade 2	Withhold	Administer corticosteroids and initiate hormonal replacements as clinically indicated	Monitor for signs and symptoms of hypophysitis (including hypopituitarism and adrenal insufficiency)
	Grade 3 or grade 4	Withhold or permanently discontinue [d]		
Hyperthyroidism	Grade 2	Continue	Treat with nonselective beta-blockers (e.g., propranolol) or thionamides as appropriate	Monitor for signs and symptoms of thyroid disorders
	Grade 3 or grade 4	Withhold or permanently discontinue [d]		
Hypothyroidism	Grade 2	Continue	Initiate thyroid replacement hormones (e.g., levothyroxine or liothyronine) per standard of care	Monitor for signs and symptoms of thyroid disorders
	Grade 3 or grade 4	Withhold or permanently discontinue		
Nephritis: grading according to increased creatinine or acute kidney injury	Grade 2	Withhold	Administer corticosteroids (prednisone 1 to 2 mg/kg or equivalent) followed by taper	Monitor changes of renal function
	Grade 3 or grade 4	Permanently discontinue		
Neurological toxicities	Grade 2	Withhold	Based on severity of AE administer corticosteroids	Ensure adequate evaluation to confirm etiology and/or exclude other causes
	Grade 3 or grade 4	Permanently discontinue		

irAEs	Toxicity grade (CTCAE version 5.0)	Action with pembrolizumab	Corticosteroid and/or other therapies	Monitoring and follow-up
Myocarditis	Asymptomatic cardiac enzyme elevation with clinical suspicion of myocarditis (previously CTCAE version 4.0 grade 1)	Withhold	Based on severity of AE administer corticosteroids	Ensure adequate evaluation to confirm etiology and/or exclude other causes
	Grade 2, grade 3, or grade 4	Permanently discontinue		
Exfoliative dermatologic conditions	Suspected SJS, TEN, or DRESS	Withhold	Based on severity of AE administer corticosteroids	Ensure adequate evaluation to confirm etiology or exclude other causes
	Confirmed SJS, TEN, or DRESS	Permanently discontinue		
All other irAEs	Persistent grade 2	Withhold	Based on severity of AE administer corticosteroids	Ensure adequate evaluation to confirm etiology or exclude other causes
	Grade 3	Withhold or discontinue based on the event [e]		
	Recurrent grade 3 or grade 4	Permanently discontinue		

irAEs	Toxicity grade (CTCAE version 5.0)	Action with pembrolizumab	Corticosteroid and/or other therapies	Monitoring and follow-up
AE(s), adverse event(s); ALT, alanine transaminase; AST, aspartate transaminase; CTCAE, Common Terminology Criteria for Adverse Events; DRESS, drug rash with eosinophilia and systemic symptom; irAE(s), immune-related AE(s); IV, intravenous; SJS, Stevens-Johnson syndrome; T1DM, type 1 diabetes mellitus; TEN, toxic epidermal necrolysis; ULN, upper limit of normal Note: Non-irAE will be managed as appropriate, following clinical practice recommendations. [a] AST/ALT: >3.0 to 5.0×ULN if baseline normal; >3.0 to 5.0×baseline, if baseline abnormal; bilirubin:>1.5 to 3.0×ULN if baseline normal; >1.5 to 3.0×baseline if baseline abnormal [b] AST/ALT: >5.0 to 20.0×ULN, if baseline normal; >5.0 to 20.0×baseline, if baseline abnormal; bilirubin:>3.0 to 10.0×ULN if baseline normal; >3.0 to 10.0×baseline if baseline abnormal [c] AST/ALT: >20.0×ULN, if baseline normal; >20.0×baseline, if baseline abnormal; bilirubin: >10.0×ULN if baseline normal; >10.0×baseline if baseline abnormal [d] The decision to withhold or permanently discontinue pembrolizumab is at the discretion of the investigator or treating physician. If control achieved or grade ≤2, pembrolizumab may be resumed. [e] Events that require discontinuation include but are not limited to: encephalitis and other clinically important irAEs (e.g., vasculitis and sclerosing cholangitis)				

6.5.2.2 Dose modification and toxicity management of infusion-reactions related to pembrolizumab

Pembrolizumab may cause severe or life-threatening infusion reactions including severe hypersensitivity or anaphylaxis. Signs and symptoms usually develop during or shortly after drug infusion and generally resolve completely within 24 hours of completion of infusion. Dose modification and toxicity management guidelines on pembrolizumab associated infusion reaction are provided in Table 6.4.

Table 6.4: Pembrolizumab infusion reaction dose modification and treatment guidelines

CTCAE grade	Treatment	Premedication at subsequent dosing
Grade 1 Mild reaction; infusion interruption not indicated; intervention not indicated	• Increase monitoring of vital signs as medically indicated until the patient is deemed medically stable in the opinion of the investigator.	None
Grade 2 Requires therapy or infusion interruption but responds promptly to	• **Stop infusion** • Additional appropriate medical therapy may	Patient may be premedicated 1.5 hour (±30 minutes) prior to infusion of study

CTCAE grade	Treatment	Premedication at subsequent dosing
symptomatic treatment (e.g., antihistamines, NSAIDs, narcotics, IV fluids); prophylactic medications indicated for ≤24 hours	include but is not limited to: ○ IV fluids ○ Antihistamines ○ NSAIDs ○ Acetaminophen ○ Narcotics • Increase monitoring of vital signs as medically indicated until the patient is deemed medically stable in the opinion of the investigator. • If symptoms resolve within 1 hour of stopping drug infusion, the infusion may be restarted at 50% of the original infusion rate (e.g., from 100 mL/hour to 50 mL/hour). Otherwise dosing will be held until symptoms resolve and the patient should be premedicated for the next scheduled dose. **Patients who develop grade 2 toxicity despite adequate premedication should be permanently discontinued from further study drug treatment.**	intervention with: • Diphenhydramine 50 mg po (or equivalent dose of antihistamine) • Acetaminophen 500-1000 mg po (or equivalent dose of analgesic)
Grade 3 or 4 **Grade 3:** Prolonged (i.e., not rapidly responsive to symptomatic medication and/or brief interruption of infusion); recurrence	• **Stop infusion** • Additional appropriate medical therapy may include but is not limited to: ○ Epinephrine** ○ IV fluids	No subsequent dosing

CTCAE grade	Treatment	Premedication at subsequent dosing
of symptoms following initial improvement; hospitalization indicated for other clinical sequelae (e.g., renal impairment, pulmonary infiltrates) **Grade 4:** Life-threatening; pressor or ventilator support indicated	○ Antihistamines ○ NSAIDs ○ Acetaminophen ○ Narcotics ○ Oxygen ○ Pressors ○ Corticosteroids • Increase monitoring of vital signs as medically indicated until the patient is deemed medically stable in the opinion of the investigator. • Hospitalization may be indicated. **In cases of anaphylaxis, epinephrine should be used immediately. **Patient is permanently discontinued from further study drug treatment.**	
Appropriate resuscitation equipment should be available at the bedside and a physician readily available during the period of drug administration. For further information, please refer to CTCAE version 5.0 at http://ctep.cancer.gov.		

6.5.2.3 Other allowed dose interruption for pembrolizumab

Pembrolizumab may be interrupted for situations other than treatment-related AEs such as medical or surgical events and/or unforeseen circumstances not related to study interventions. However, study interventions are to be restarted within 21 days of the originally scheduled dose and within 42 days of the previously administered dose, unless otherwise discussed with the sponsor. The reason for interruption is to be documented in the patient's study record.

6.5.3 Clinical management of cytokine release syndrome

The cytokine release syndrome management recommendations described in Table 6.5 can be modified by investigational sites as medically necessary or as appropriate without requiring a Protocol Amendment or being considered a Protocol deviation.

Table 6.5: Clinical management of cytokine release syndrome[52]

Grading assessment	Treatment and measures
Grade 1 • Fever, constitutional symptoms	• Vigilant supportive care • Assess for infections (Treat fever and neutropenia if present, monitor fluid balance, antipyretics, analgesics as needed)
Grade 2 • Hypotension: responds to fluids or one low dose pressor • Hypoxia: responds to <40% O_2 • Organ toxicity: grade 2	***Extensive comorbidities or older age?*** → No: Vigilant supportive care (Monitor cardiac and other organ function closely) → Yes: Vigilant supportive care Tocilizumab* ± corticosteroids
Grade 3 • Hypotension: requires multiple pressors or high dose pressors • Hypoxia: requires ≥40% O_2 • Organ toxicity: grade 3, grade 4 transaminitis	• Vigilant supportive care • Tocilizumab* ± corticosteroids
Grade 4 • Mechanical ventilation • Organ toxicity: grade 4, excluding transaminitis	• Vigilant supportive care • Tocilizumab* ± corticosteroids

*Should be used if no other treatment option, only

6.6 Continued access to study intervention after the end of the study

Patients who are still on study interventions at the time of study completion/termination may continue to receive study interventions if they are experiencing clinical benefit. The continued access to study interventions will end when a criterion for discontinuation is met or 35 doses of pembrolizumab have been administered (also see section 7.1).

6.7 Treatment of overdose

For this study, an overdose of nanrilkefusp alfa will be defined as any dose above 13 µg/kg.

For this study, an overdose of pembrolizumab will be defined as any dose of 1000 mg or greater.

No specific information is available on the treatment of overdose of nanrilkefusp alfa and/or pembrolizumab. In the event of overdose, the patient should be observed closely for signs of toxicity. Appropriate supportive treatment should be provided if clinically indicated.

6.8 Concomitant therapy

All treatments that the investigator considers necessary for a patient's welfare may be administered at the discretion of the investigator in keeping with the community standards of medical care.

All concomitant medications will be recorded on the electronic Case Report Form (eCRF) including all prescription, over-the-counter products, herbal supplements, and IV medications and fluids. If changes occur during the study period, documentation of drug dosage, frequency, route, and date should also be included on the eCRF.

All concomitant medications received within 28 days before the first dose of study interventions and up to 90 days after the last dose of study interventions should be recorded. All concomitant medications administered during SAEs or AESIs are to be recorded. SAEs and AESIs are defined in section 8.3.

6.8.1 Rescue medication and supportive care

Patients should receive appropriate supportive care measures as deemed necessary by the treating investigator. Suggested supportive care measures for the management of AEs with potential immunologic etiology are outlined along with the dose modification guidelines in section 6.5.

Note: If after the evaluation of the event it is determined not to be related to pembrolizumab, the investigator does not need to follow the treatment guidance. Refer to section 6.5 for guidelines regarding dose modification and supportive care.

It may be necessary to perform conditional procedures such as bronchoscopy, endoscopy, or skin photography as part of evaluation of the event.

6.8.2 Prohibited medications

Medications or vaccinations specifically prohibited in the exclusion criteria are not allowed during the ongoing trial. If there is a clinical indication for any medication or vaccination specifically prohibited during the trial, discontinuation from study interventions or vaccination may be required (also see section 7.1). The investigator is to discuss prohibited medication/vaccination with the sponsor's medical monitor. The final decision on any supportive therapy or vaccination rests with the investigator and/or the patient's primary physician. However, the decision to continue the patient on study interventions requires the mutual agreement of the investigator, the sponsor, and the patient.

The following medications and vaccinations are prohibited during the study:

- Concomitant use of drugs known to prolong the QT/QTc interval is prohibited during the study. Some of the medications known to prolong the QT interval are[53]: amiodarone, azithromycin, ciprofloxacin, chlorpromazine, citalopram, domperidone, donepezil, escitalopram, fluconazole, haloperidol, levofloxacin, levomepromazine, methadone, ondansetron, and sulpiride. A complete list of medications known to prolong the QT interval can be found at www.crediblemeds.org.

- Antineoplastic systemic chemotherapy or biological therapy
- Immunotherapy not specified in this Protocol
- Chemotherapy not specified in this Protocol
- Investigational agents other than nanrilkefusp alfa and pembrolizumab
- Radiation therapy

  Note: Radiation therapy to a symptomatic solitary lesion or to the brain may be allowed at the investigator's discretion. Palliative radiotherapy of, e.g., painful bone metastases not defined as indicator lesions is allowed.
- Live or live attenuated vaccines within 30 days prior to the first dose of study interventions, during treatment with nanrilkefusp alfa and/or pembrolizumab, and within 90 days after the last dose of nanrilkefusp alfa and/or pembrolizumab, whichever is later. Note: Killed vaccines are allowed.

  Note: Any licensed COVID-19 vaccine (including for emergency use) in a particular country is allowed in the study as long as they are mRNA vaccines, adenoviral vaccines, or inactivated vaccines. These vaccines will be treated just as any other concomitant therapy.

  Investigational vaccines (i.e., those not licensed or approved for emergency use) are not allowed.
- Systemic glucocorticoids except when used for the following purposes:
  - To modulate symptoms of an AE that is suspected to have an immunologic etiology
  - For the prevention of emesis
  - To premedicate for IV contrast allergies
  - To treat chronic obstructive pulmonary disease exacerbations (only short-term oral or IV use in doses >10 mg/day prednisone equivalent)
  - For chronic systemic replacement not to exceed 10 mg/day prednisone equivalent
  - Other glucocorticoid use except when used for the following purposes:
    - For topical use or ocular use
    - Intraarticular joint use
    - For inhalation in the management of asthma or chronic obstructive pulmonary disease

  Note: Inhaled steroids are allowed for management of asthma.

If the investigator determines that a patient requires any of the aforementioned treatments for any reason, study interventions must be discontinued.

7 DISCONTINUATION OF STUDY INTERVENTION AND PATIENT DISCONTINUATION/WITHDRAWAL

7.1 Discontinuation of study intervention

Discontinuation of study interventions does not represent withdrawal from the study.

A patient must be discontinued from study interventions but continue to be monitored in the study (if possible) for any of the following reasons:

- Confirmed radiographic disease progression (please refer to section 10.2)
- Clinical disease progression (investigator's assessment)
- AE (intercurrent illness or study intervention-related toxicity that would, in the judgment of the investigator, affect assessments of the patient's clinical status to a significant degree or require discontinuation of study interventions)
- Patient may withdraw from the study at any time for any reason; the investigator must make every effort to determine the reason for this decision and record it in source documentation of the patient
- Discontinuation of treatment may be considered for patients who have attained a confirmed complete response (CR) and have been treated for at least 8 cycles (at least 24 weeks), receiving 2 cycles of the combination including 2 doses of pembrolizumab and at least 80% of the planned doses of nanrilkefusp alfa beyond the date when the initial CR was declared
- Any study intervention-related toxicity specified as a reason for permanent discontinuation as defined in the guidelines for dose modification due to AEs in section 6.5
- Death of the patient
- Pregnancy of the patient
- Concomitant treatment with a prohibited medication, including further lines of systemic anti-cancer therapy (also see section 6.8.2)
- Patient's non-compliance
- Lost to follow-up
- Study terminated by the sponsor

Pembrolizumab will be discontinued after the completion of 35 administrations (approximately 2 years; also see section 6.6).

In case nanrilkefusp alfa needs to be discontinued for reasons other than disease progression, pembrolizumab treatment can continue for up to a total of 35 administrations (approximately 2 years) in patients without disease progression or unacceptable toxicity. In case pembrolizumab needs to be discontinued, nanrilkefusp alfa treatment can continue until disease progression or unacceptable toxicity (not applicable in the Czech Republic). In the Czech Republic: In case pembrolizumab needs to be discontinued, nanrilkefusp alfa treatment will be discontinued as well.

See the Schedule of activities for data to be collected at the time of intervention discontinuation and follow-up and for any further evaluations that need to be completed.

7.2 Patient discontinuation/withdrawal from the study

A patient may withdraw from the study at any time at his/her own request, or may be withdrawn at any time at the discretion of the investigator for safety, behavioral, compliance, or administrative reasons, or when the study is discontinued by the sponsor. This is expected to be uncommon.

The patient will be permanently discontinued both from the study interventions and from the study at that time.

If the patient withdraws consent for disclosure of future information, the sponsor may retain and continue to use any data collected before such a withdrawal of consent.

If a patient withdraws from the study, s/he may request destruction of any samples taken and not tested, and the investigator must document this in the site study records.

7.3 Lost to follow-up

A patient will be considered lost to follow-up if s/he repeatedly fails to return for scheduled visits and is unable to be contacted by the study site.

The following actions must be taken if a patient fails to return to the clinic for a required study visit:

- The site must attempt to contact the patient and reschedule the missed visit as soon as possible and counsel the patient on the importance of maintaining the assigned visit schedule and ascertain whether or not the patient wishes to and/or should continue in the study.
- Before a patient is deemed lost to follow-up, the investigator or designee must make every effort to regain contact with the patient (where possible, 3 telephone calls and, if necessary, a certified letter to the patient's last known mailing address or local equivalent methods). These contact attempts should be documented in the patient's medical record.
- Should the patient continue to be unreachable, s/he will be considered to have withdrawn from the study.

8 STUDY ASSESSMENTS AND PROCEDURES

Study procedures and their timing are summarized in the Schedule of activities. Protocol waivers or exemptions are not allowed.

Immediate safety concerns should be discussed with the sponsor immediately upon occurrence or awareness to determine if the patient should continue or discontinue study interventions.

Adherence to the study design requirements, including those specified in the Schedule of activities, is essential and required for study conduct.

All screening evaluations must be completed and reviewed to confirm that potential patients meet all eligibility criteria. The investigator will maintain a screening log to record details of all patients screened and to confirm eligibility or record reasons for screening failure, as applicable.

Procedures conducted as part of the patient's routine clinical management (e.g., blood count) and obtained before signing of the ICF may be utilized for screening or baseline purposes provided the procedures met the Protocol-specified criteria and were performed within the time frame defined in the Schedule of activities.

8.1 Efficacy assessments

8.1.1 Tumor imaging and assessment of disease

Throughout this section, the term "scan" refers to any medical imaging data used to assess tumor burden and may include cross-sectional imaging (such as computed tomography [CT] or magnetic resonance imaging [MRI]), medical photography, or other methods as specified in this Protocol.

Scan evaluation by a local radiologist or by the investigator will be used for the main analyses. However, all scheduled scans for patients will be collected. In case of a deemed need for health authority interactions, a sensitivity analysis will be considered based on evaluation of imaging documentation by an independent blinded radiologist. A scan that is obtained at an unscheduled time point, for any reason (including suspicion of progression or other clinical reason), will also be collected if it shows disease progression, or if it is used to support a response assessment. In addition, historical scans that were obtained at disease progression on previous systemic anti-cancer treatment will be collected.

The process for scan collection can be found in the Site Imaging Manual. CT scans are preferred over other tumor imaging methods. For the abdomen and pelvis, contrast-enhanced MRI may be used when CT with iodinated contrast is contraindicated, or when mandated by local practice. The same type of scan should be used in a patient throughout the study to optimize the reproducibility of the assessment of existing and new tumor burden and improve the accuracy of the assessment of response or progression based on imaging. Note: for the purposes of assessing tumor scans, the term "investigator" refers to the local investigator at the site and/or the radiological reviewer located at the site or at an offsite facility.

If brain scans are performed, magnetic resonance scans are preferred; however, CT scans are acceptable if MRI is medically contraindicated.

Bone scans may be performed to evaluate bone metastases. Any supplemental scans done to support a positive or negative bone scan, such as plain X-rays acquired for correlation, should also be collected.

At screening, patient eligibility will require radiographic documentation of at least one lesion that meets the requirements for selection as a target lesion (as defined by RECIST 1.1) before the start of study interventions.

8.1.1.1 Initial tumor scans

Initial tumor scans at screening must be performed within 28 days prior to day 1 of cycle 1. The site study team must review screening scans to confirm the patient has measurable disease per RECIST 1.1.

Tumor scans performed as part of routine clinical management are acceptable for screening if they are of acceptable diagnostic quality and performed within 28 days of day 1 of cycle 1.

If brain scans are performed to document the stability of existing metastases, the brain MRI should be acquired during screening. If MRI is medically contraindicated, CT with contrast is an acceptable alternative.

8.1.1.2 Tumor scans during the study

The first on-study scan assessment should be performed at 6 weeks (±2 weeks; starting from cycle 1 day 1). Subsequent tumor scans should be performed every 6 weeks (±2 weeks) or more frequently if clinically indicated. Scan timing should follow calendar days and should not be adjusted for delays in cycle starts.

All supplemental imaging must be collected.

Objective response should be confirmed by repeat scan performed at least 4 weeks after the first indication of a response is observed. Patients will then return to the regular scan scheduled, starting with the next scheduled time point. Patients who receive additional scans for confirmation do not need to undergo the next scheduled scan if it is fewer than 4 weeks later; scans may resume at the subsequent scheduled time point.

When radiological disease progression is identified by the investigator in clinically stable patients, disease progression is to be confirmed by another set of scans performed 4 to 8 weeks later, per iRECIST guidelines in section 8.1.1.5.

If disease progression is not confirmed, clinically stable patients are to continue study interventions until progression is confirmed. Patients are to return to their regular scan schedule. If the next scheduled scan will occur in less than 4 weeks, this scheduled scan may be skipped.

If disease progression is confirmed, study interventions will be discontinued. Exceptions are detailed in section 8.1.1.5.

8.1.1.3 End of treatment and follow-up tumor imaging

If patients discontinue study interventions, tumor scans should be performed at the time of discontinuation (±4-week window) unless previous scans were obtained within 4 weeks of discontinuation. If patients discontinue study interventions due to documented disease progression, this is the final required tumor scan if the investigator elects not to implement iRECIST.

If patients discontinue study interventions without documented disease progression, every effort should be made to monitor disease status by acquiring tumor scans using the same schedule used while on treatment (every 6 weeks [±2 weeks]).

Scans are to be continued until one of the following conditions are met:

- Disease progression identified by the investigator (radiological progression per [i]RECIST [1.1] or mCRPC-specific progression)
- Start of a new anti-cancer treatment
- Pregnancy of the patient
- Withdrawal of consent by the patient
- End of the study

For clinically unstable patients, a scan obtained within 2 weeks after clinical progression is recommended. Scans are to be continued as per regular schedule (section 1.3).

8.1.1.4 RECIST 1.1 assessment of disease

RECIST 1.1 will be used as the primary measure for assessment of tumor response, date of disease progression, and as a basis for all Protocol guidelines related to disease status (e.g., discontinuation of study interventions).

8.1.1.5 iRECIST assessment of disease

iRECIST is based on RECIST 1.1 but adapted to account for the unique tumor response seen with immunotherapeutic drugs. iRECIST will be used by the investigator to assess tumor response and progression and make treatment decisions. When clinically stable, patients may continue study interventions beyond RECIST 1.1 progression with continued assessment of response according to the rules outlined in section 10.2. iRECIST reflects that some patients can have a transient tumor flare after the start of immunotherapy, then experience subsequent disease response. This data will be captured in the clinical database.

- If the patient is clinically stable, continue study interventions per Protocol
  - Continue scans per Protocol schedule (the next scheduled scan should be ≥4 weeks from most recent scan acquired)
  - Continue investigator assessment per iRECIST
  - Collect scans
- If the patient is not clinically stable, best medical practice is to be applied

For the purpose of this decision process, lack of clinical stability is defined as:

- Unacceptable toxicity
- Clinical signs or symptoms indicating clinically significant disease progression
- Decline in performance status
- Rapid disease progression or threat to vital organs or critical anatomical sites (e.g., central nervous system metastasis, respiratory failure due to tumor compression, spinal cord compression) requiring urgent alternative medical intervention

8.1.1.6 PD-1 treatment progression

PD-1 treatment progression is defined by meeting all of the following criteria:

1. Has received at least 2 doses of an approved anti-PD-1/L1 mAb
2. Has demonstrated disease progression after PD-1/L1
3. Progressive disease has been documented within 12 weeks from the last dose of anti-PD-1/L1 mAb
   - 3.1. Progressive disease is determined according to iRECIST
   - 3.2. This determination is made by the investigator. Once progressive disease is confirmed, the initial date of disease progression documentation will be considered the date of disease progression.

8.2 Safety assessments

If patients feel unwell at any point in time during the study after being discharged from the hospital, they should contact their study investigator as indicated in the ICF.

8.2.1 Physical examinations, ECOG performance status, body height, and body weight

General physical examination with organ/system-specific physical examination will be carried out by a licensed physician (or the physician's assistant or a nurse practitioner).

For screening, a complete physical examination will be performed including head (eyes, ear, nose, and throat), neck, cardiovascular, chest/lungs, abdomen (including liver and spleen size), extremities, neurological, skin, and lymph nodes. For subsequent visits, a physical examination is to be done with the focus on abdomen (including liver and spleen size), lymph nodes, and any other system that may contribute to clinical disease assessments.

An ECOG performance status will be assigned as indicated in the Schedule of activities.

Body height and body weight will be measured. Body height will be measured only at screening. Body weight will be measured before the start of each treatment cycle and as indicated in the Schedule of activities.

8.2.2 Vital signs

Vital signs will include blood pressure (systolic and diastolic, after ≥5 minutes of rest), body temperature, heart rate, and respiratory rate.

8.2.3 Special assessments on treatment days

8.2.3.1 Cycle 1 day 1

Patients will be observed in the hospital up to 12 hours following nanrilkefusp alfa administration. Vital signs (systolic and diastolic blood pressure after ≥5 minutes of rest, body temperature, heart rate, and respiratory rate) will be documented at the following frequency at minimum unless clinically required otherwise:

- Before nanrilkefusp alfa administration
- 15 minutes (±5 minutes) after nanrilkefusp alfa administration

- 30 minutes (±5 minutes) after nanrilkefusp alfa administration
- 2 hours (±15 minutes) after nanrilkefusp alfa administration
- 4 hours (±30 minutes) after nanrilkefusp alfa administration
- 12 hours (±2 hours) after nanrilkefusp alfa administration

8.2.3.2 Cycle 1 day 2, day 8, and day 9; and treatment days of cycle 2

Patients will be observed in the hospital up to 4 hours following nanrilkefusp alfa administration. Vital signs (systolic and diastolic blood pressure after ≥5 minutes of rest, body temperature, heart rate, and respiratory rate) will be documented at the following frequency until discharge:

- Before nanrilkefusp alfa administration
- 15 minutes (±5 minutes) after nanrilkefusp alfa administration
- 30 minutes (±5 minutes) after nanrilkefusp alfa administration
- 2 hours (±15 minutes) after nanrilkefusp alfa administration
- 4 hours (±30 minutes) after nanrilkefusp alfa administration

8.2.3.3 Treatment days of cycle 3 and onwards

Patients will be observed in the hospital up to 2 hours following nanrilkefusp alfa administration. Vital signs (systolic and diastolic blood pressure after ≥5 minutes of rest, body temperature, heart rate, and respiratory rate) will be documented at the following frequency until discharge:

- Before nanrilkefusp alfa administration
- 15 minutes (±5 minutes) after nanrilkefusp alfa administration
- 30 minutes (±5 minutes) after nanrilkefusp alfa administration
- 2 hours (±15 minutes) after nanrilkefusp alfa administration

8.2.4 Electrocardiography and left ventricular ejection fraction

During the study, patients with signs or symptoms of chest pain, murmurs, gallops, irregular rhythm, or palpitations must be further assessed if clinically indicated, including the need for hospitalization. Standard 12-lead ECG will be done locally at the site for assessment of any change in QTcF interval and other parameters.

Left ventricular ejection fraction will be assessed using either echocardiography or multigated acquisition scanning (MUGA). However, the method chosen needs to stay the same throughout the trial.

8.2.5 Clinical safety laboratory assessments

Instructions on sample collection, handling, storage, and shipment of samples are detailed in the study-specific Laboratory Manual provided to the study site.

8.2.5.1 Coagulation

Coagulation tests will include prothrombin time, activated partial thromboplastin time, international normalized ratio, D-dimer, and fibrinogen.

8.2.5.2 Hematology

Hematology tests will include hemoglobin, glycated hemoglobin at screening, hematocrit, red blood cell count, reticulocytes, white blood cell count (with full differentiation), absolute lymphocyte count, and platelet count.

8.2.5.3 Biochemistry

Blood biochemistry tests will include Na, K, Cl, phosphate, Mg, Ca, albumin, total protein, ALT, AST, bilirubin (direct, total), alkaline phosphatase, lactate dehydrogenase, creatinine clearance calculated by the Cockcroft-Gault formula, creatinine, glucose (preferably fasting), urea or blood urea nitrogen, cholesterol, triglyceride, C-reactive protein, uric acid, amylase, and lipase.

8.2.5.4 Urinalysis

The following parameters are to be analyzed: pH, glucose, protein, bilirubin, urobilinogen. Microscopic examination (mandated only if clinically indicated): red blood cell count, white blood cell count, epithelial cells, bacteria.

In case of proteinuria ≥100 mg/dL at screening, a 24-hour urine analysis will have to be performed (before the start of nanrilkefusp alfa treatment) to document 24-hour proteinuria levels and a urine test will continue during the treatment period. In case of increase of proteinuria with ≥300 mg/dL (at any time), a 24-hour urine analysis will be performed.

8.2.5.5 Serology testing for infections

Mandatory serology testing for infections will include testing for HIV, HBV, and HCV. Should the patient refuse testing, his/her participation will be terminated.

8.2.6 Monitoring of thyroid function

Thyroid function will be monitored by means of thyroid stimulating hormone (TSH), triiodothyronine (T3) or free T3, and free thyroxine (T4) testing.

8.2.7 Pregnancy testing

WOCBP will be tested for pregnancy.

8.3 Adverse events, serious adverse events, and other safety reporting

8.3.1 Definitions

8.3.1.1 Adverse events

International Council for Harmonisation (ICH) guideline E2A defines an AE as any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product and which does not necessarily have to have a causal relationship with this treatment.

An AE can therefore be any unfavorable and unintended sign (e.g., tachycardia, enlarged liver), symptom (e.g., nausea, chest pain), abnormal result of an investigation (e.g., laboratory finding), or disease temporarily associated with the use of a medicinal product, whether or not considered related to the medicinal product.

According to the FDA (21CFR312.32), an AE means any untoward medical occurrence associated with the use of a drug in humans, whether or not considered drug related.

8.3.1.2 Serious adverse events

A SAE is any untoward medical occurrence that at any dose fulfills one or more of the following criteria:

- Results in death
- Is immediately life-threatening

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

  A hospitalization is defined as an inpatient overnight stay, but this can be shorter than 24 hours.
- Is another medically significant event defined as an event that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient or may require intervention to prevent any of the above listed outcomes

8.3.1.3 Adverse events of special interest

The following AEs are defined for this Protocol as AESIs and must be reported to the sponsor or designee within 24 hours of awareness:

1. For patients without liver metastases or liver cancer or patients who do not receive hepatotoxic concomitant medications, the following AESIs must be reported:
   a. On-treatment (i.e., from the first dose of study treatment to 90 days after the last dose of study treatment) ALT or AST increase that is greater than 3×ULN in combination with clinical jaundice
   b. On-treatment ALT increase that is greater than or equal to 3×ULN and international normalized ratio increase that is greater than 1.5
   c. Recurrence of CTCAE grade 3 ALT and/or AST increase in combination with total bilirubin increase greater than 2×ULN or greater than 2×baseline level
2. For patients with liver metastases or liver cancer or patients who receive hepatotoxic concomitant medications, the following AESIs must be reported:
   a. On-treatment ALT or AST increase that is greater than 5×ULN in combination with total bilirubin increase greater than 2×ULN (of which more than or equal to 35% is direct bilirubin)

b. On-treatment ALT or AST increase that is greater than 5×ULN in combination with clinical jaundice

3. An overdose of pembrolizumab, as defined in section 6.7

4. An elevated AST or ALT laboratory value that is greater than or equal to 3×ULN and an elevated total bilirubin laboratory value that is greater than or equal to 2×ULN and, at the same time, an alkaline phosphatase laboratory value that is less than 2×ULN, as determined by way of Protocol-specified laboratory testing or unscheduled laboratory testing*

   *Note: These criteria are based upon available regulatory guidance documents. The purpose of the criteria is to specify a threshold of abnormal hepatic tests that may require an additional evaluation for an underlying etiology. The trial site guidance for assessment and follow-up of these criteria can be made available. It may also be appropriate to conduct additional evaluation for an underlying etiology in the setting of abnormalities of liver blood tests including AST, ALT, bilirubin, and alkaline phosphatase that do not meet the criteria noted above. In these cases, the decision to proceed with additional evaluation will be made through consultation between the study investigators and the sponsor's medical monitor for the trial.

   However, abnormalities of liver blood tests that do not meet the criteria noted above are not AESIs for this trial.

8.3.1.4 Excluded events

Hospitalization for the following reasons will not be regarded as serious (not immediately reportable):

- Routine treatment or monitoring of the disease under study, including hospitalization due to study-related procedures, not associated with any deterioration of the patient's status
- Elective or pre-planned treatment (before signing the ICF) for a pre-existing condition that is unrelated to the disease under study and has not worsened since signing the ICF
- Social reasons, respite care, and in the absence of a medical condition (e.g., for observational purposes without any intervention)

8.3.1.5 Severity/intensity vs. seriousness

ICH E2A: The term "severe" is often used to describe the intensity (severity) of a specific event (as mild, moderate, or severe myocardial infarction); the event itself, however, may be of a relatively minor medical significance (such as a severe headache). This is not the same as "serious", which is based on patient/event outcome or action criteria usually associated with events that pose a threat to a patient's life or functioning. Seriousness (not severity) serves as a guide for defining regulatory reporting obligations.

8.3.2 Time period and frequency for collecting AE, SAE, and other reportable safety event information

Every effort should be taken to collect all AEs, AESIs, and SAEs from the date of the patient's signing the ICF until 90 days after the final administration of nanrilkefusp alfa and/or pembrolizumab (whichever is later) or until the start of a new anti-cancer therapy, whichever is earlier.

Additionally, any SAE brought to the attention of an investigator at any time outside of the time period specified above must be reported immediately to the sponsor or designee if the event is considered as having a suspected causal relationship to nanrilkefusp alfa and/or pembrolizumab per the investigator's judgment.

Pregnancies of the patient or patient's female partner must be reported from the date of the patient's signing the ICF up to 120 days following the last dose of pembrolizumab or 30 days after the last dose of nanrilkefusp alfa, whichever is later.

8.3.3 Method of detecting AEs and SAEs

Care will be taken not to introduce bias when detecting AEs and/or SAEs. Open-ended and non-leading verbal questioning of the patient is the preferred method to inquire about AE occurrences. A standardized question such as "Have you had any health problems since your last visit or since you were last questioned?" will be given by the investigator or the investigational site personnel at each contact with the patient.

The investigator assesses at each visit (or more frequently, if necessary) if there are any changes in AE diagnosis, severity, suspected causal relationship to clinical trial medication/procedure, interventions required to treat the event, and AE outcome.

8.3.4 Follow-up of AEs and SAEs

After the initial (S)AE report, the investigator is required to proactively follow each patient at subsequent visits/contacts (or more frequently, if necessary).

AEs are monitored (followed up) until resolution or until 90 days after the last dose of nanrilkefusp alfa and/or pembrolizumab, whichever is later.

All SAEs and AESIs will be followed until resolution, stabilization (becoming a permanent condition), or the patient is lost to follow-up as defined in section 7.3.

8.3.5 Regulatory reporting requirements for SAEs

Prompt notification by the investigator to the sponsor or designee of an SAE is essential so that legal obligations and ethical responsibilities towards the safety of patients and the safety of a study intervention under clinical investigation are met.

The sponsor has a legal responsibility to notify both the local regulatory authority and other regulatory agencies about the safety of an investigational product under clinical investigation. The sponsor or designee will comply with country-specific regulatory requirements relating to safety reporting to the regulatory authority, IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU), and investigators.

An investigator who receives a periodic or expedited safety report (e.g., suspected unexpected serious adverse reaction report, summary or line listing of suspected unexpected serious adverse reactions) or other specific safety information from the sponsor or designee will review and then file it along with the IB and will notify the IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU), if appropriate according to local requirements.

8.3.6 Pregnancy

Pregnancy is not considered an AE unless it meets any criteria for becoming serious (see the definitions in section 8.3.1.2). Patients must inform the investigator of any newly identified pregnancy or pregnancy of their partners without delay.

If a patient inadvertently becomes pregnant while on treatment with nanrilkefusp alfa and/or pembrolizumab, the patient will be immediately discontinued from study interventions (see section 7.1). The site will contact the patient at least monthly and document the patient's or patient's female partner status until the pregnancy has been completed or terminated. The outcome of the pregnancy will be reported to the sponsor or designee within 24 hours of awareness. In addition, if the outcome is a SAE in line with section 8.3.1.2, e.g., death, abortion, congenital anomaly, or other disabling or life-threatening complication to the mother or newborn, the SAE report form needs to be completed. The study investigator will make every effort to obtain permission (consent) from the pregnant patient to follow the outcome of the pregnancy and report the condition of the fetus or newborn to the sponsor or designee. Every infant must be followed up for 2 months after delivery.

Consent to report information on the outcome of the pregnancy of male patients' partners needs to be obtained from the pregnant partners.

Egg and sperm donation is not allowed during the treatment period and for at least 120 days after the last dose of pembrolizumab or at least 30 days after the last dose of nanrilkefusp alfa, whichever is later.

8.3.7 Assessing AEs

Information about adverse reactions (causally related events) already known for the investigational medicinal products can be found in the nanrilkefusp alfa and pembrolizumab IBs and Keytruda's current package insert/US prescribing information (for the US) and current Summary of Product Characteristics (for the EU)[54,55] or will be communicated between IB updates in the form of "Dear Investigator Letter".

8.3.7.1 Causality

The investigator needs to assess the causal relationship of any AE to:

- Nanrilkefusp alfa and/or pembrolizumab
- Other suspected cause(s) of the event (e.g., concurrent disease, concomitant medication)

This assessment is based on the investigator's clinical judgment, taking into account all relevant information available at the time of AE reporting including (but not limited to):

- Temporal association of the event onset with administration of the medication/procedure
- Known type of reaction for any of the administered drugs
- Disappearance or abating of symptoms when the drug is discontinued or the dose is reduced
- Reappearance of symptoms when the drug is re-administered
- Event may or may not be caused by the patient's health condition
- Presence of risks or factors not related to nanrilkefusp alfa and/or pembrolizumab or procedure that are known to be associated with the occurrence of the event

Causal relationship of all AEs will be classified as follows:

- **Not suspected**: It is not plausible that the AE is caused by medication/procedure and a likely alternative explanation exists.

  No reasonable possibility of a causal or temporal relationship.

- **Suspected**: It is plausible that the AE is caused by medication/procedure.

  Reasonable possibility of a causal relationship.

*For the purposes of safety reporting, **"reasonable possibility"** means there is evidence to suggest a causal relationship between one of the drugs/procedure and the AE.*

8.3.7.2 Severity/intensity

Severity or intensity of an AE has to be assessed according to the CTCAE, version 5.0. A grading scale is provided for AE terms displaying grades 1 through 5 with unique clinical descriptions of severity for each AE.

Grading refers to the severity of the AE.

8.3.8 Reporting by the investigational site

8.3.8.1 Adverse events

Any AE, whether or not considered to be causally related to the trial medication and regardless of its seriousness, must be reported (described and recorded) in the AE section of the patient's eCRF on an ongoing basis. Each AE will be evaluated by the investigator to determine its term and grade according to the CTCAE, version 5.0. In case a suitable CTCAE term is not available, an appropriate medical verbatim term may be used instead.

Only clinically relevant abnormal laboratory values, vital signs, or examination abnormalities need to be documented as AEs. Whenever possible, a diagnosis rather than symptoms should be provided on the AE eCRF page (e.g., anemia instead of low hemoglobin).

Clinically relevant here means: induce clinical signs or symptoms, require therapy (e.g., hematologic abnormality requiring treatment), require a change in trial medication(s), or require a change in the clinical trial schedule or per investigator's discretion.

Physical examination findings will be compared with the baseline status and any significant change, as assessed by the investigator, must be documented as an AE.

A surgical procedure is not an AE but a therapeutic measure for a condition that necessitates surgery. Therefore, the condition for which the surgery is required has to be reported as an AE.

Any pre-planned surgery (i.e., planned before signature of the ICF) or other intervention permitted by this study Protocol and the condition leading to that measure are not AEs. In such cases, the underlying condition needs to be documented in the patient's medical history on the eCRF.

Death itself is not an AE term but the outcome of an event, which needs to be described using medical terminology. Information about death will be captured on the respective eCRF page along with relevant details (date of death, immediate and underlying causes of death).

8.3.8.2 Documenting on eCRFs

The reported term should be a medical diagnosis or sign/symptom of the event according to the CTCAE, version 5.0 (whenever possible), not a procedure. Each symptom in a constellation of symptoms should be listed separately if the investigator has not set a diagnosis.

Fluctuations or re-occurrences of a condition, which are considered normal for the patient and are recorded on the patient's eCRF medical history, do not need to be reported as an AE. However, if the condition deteriorates during the trial, it needs to be captured as an AE.

If the same AE occurs repeatedly, it must be assessed and documented separately each time.

If possible, each AE should be evaluated to determine:

- Event term (according to the CTCAE, version 5.0, not as reported by the patient) or a description of the AE in medical terms
- Severity grade or intensity of the event as assessed by the investigator (1-5 according to the CTCAE, version 5.0)
- Its causal relationship to nanrilkefusp alfa and/or pembrolizumab, as assessed by the investigator (suspected; not suspected)
- Other suspected cause(s) of the event (e.g., concurrent disease, concomitant medication)
- Event duration, including onset date and end date
- Action taken with nanrilkefusp alfa and/or pembrolizumab due to the reported event
- Other action taken
- Event seriousness (non-serious or serious AE)
- Event outcome

8.3.8.3 Immediately reportable events

The investigator or any investigational site staff must immediately (**within 24 hours of awareness at the latest**) notify/report to the sponsor or designee any initial or medically relevant follow-up information about these events:

- SAE
- AESI
- Pregnancy of the patient or patient's female partner

The initial notification can be made by phone on +420 725 385 443. This notification must be followed within an additional 24 hours by a written report (i.e., a completed SAE Report Form or Pregnancy Data Collection Form), providing all available information and a detailed narrative description. A formless notification (without a report form) is not required if the initial/follow-up information is reported on the appropriate form within 24 hours of knowledge.

The investigator must not wait to receive additional information to fully document the event before notifying the sponsor or designee **primarily via the eCRF system**; or via:

Email: safety@sotio.com

Fax: +420 224 175 498

Phone: +420 725 385 443

Follow-up information must be sent within the same timelines using the same contact details as outlined above.

Additionally, refer to the safety reporting instructions for sites for information on how to report these events.

Minimum notification/reporting requirements

The following information must be provided for a valid notification/report:

1. Identification of the notifying/reporting person (e.g., name of the reporter)
2. Identification of the patient (e.g., patient number)
3. Concerned study treatment(s) (nanrilkefusp alfa and/or pembrolizumab) or clinical trial (e.g., SC104)
4. Reason for notification/reporting (i.e., SAE, AESI, pregnancy of the patient or patient's female partner)
5. Event term

*In addition, providing the **assessment of the causal relationship** is necessary for comprehensive evaluation by the sponsor and potential regulatory submission.*

8.3.8.4 Report forms

The SAE Report Form, **primarily completed within the eCRF system** for the study, will be used for reporting of SAEs and AESIs and submitted to the sponsor or designee. In case the eCRF system is not available/accessible, a paper SAE Report Form is filled out and sent to the sponsor or designee (see details above).

For reporting of pregnancies, the paper Pregnancy Data Collection Form is to be used. Completion guidelines provide information on format and details of the information required.

The originals of the paper report forms must be kept in the site study file.

The report forms need to be completed in English.

All immediately reportable events from the investigational site to the sponsor or designee (i.e., SAEs, AESIs, and patients' or partners' pregnancies) must also be recorded in the site's source documentation and on the eCRF as appropriate.

8.4 Pharmacokinetics

The following PK parameters will be analyzed (nanrilkefusp alfa and pembrolizumab; pembrolizumab samples will be collected to be analyzed later during the course of the study if needed):

- Area under the curve
- Measured maximal concentration
- Time corresponding to occurrence of maximal concentration
- Apparent terminal elimination half-life
- Apparent total serum clearance after administration

- Apparent volume of distribution during terminal phase
- Accumulation ratio

Further details will be specified in the PK Analysis Plan.

The actual date and time (24-hour clock time) of each sample will be recorded.

Instructions on sample collection, handling, storage, and shipment of samples are detailed in the study-specific Laboratory Manual provided to the study site.

8.5 Genetics

Participation in genetic research is optional. Patients who do not wish to participate in genetic research may still participate in the study.

Genetic research will include the following: DNA analysis such as targeted sequencing (tumor mutational burden [TMB] in blood and tumor), MSI analysis, and circulating tumor DNA analysis.

Instructions on sample collection, handling, storage, and shipment of samples are detailed in the study-specific Laboratory Manual provided to the study site.

8.6 Biomarkers

Collection of samples for biomarker research is also part of this study. The following samples will be collected from patients in this study:

- Archival and/or freshly obtained tumor tissue: Newly obtained tissue is preferred to an archival sample. Formalin-fixed, paraffin-embedded block specimens are preferred to slides. Fine needle aspirate samples are not acceptable.
- Blood

DNA and RNA analyses are optional. Patients who do not wish to participate in this research may still participate in the study.

8.6.1 Tumor tissue

Tumor PD-L1 expression (tumor proportion score ≥1% or combined positive score ≥1%) will be determined by an FDA-approved immunohistochemistry assay performed by a central laboratory using previously obtained archival tumor tissue or tissue obtained from a biopsy at screening. For patients whose initial tumor tissue sample is PD-L1 negative, a biopsy can be performed at screening to submit fresh tissue for testing PD-L1 status.

PD-(L)1 expression status is not for eligibility.

Other immune-related features to be measured may include but are not limited to the presence of tumor-infiltrating lymphocytes (T cells, NK cells, FoxP3+ cells) and myeloid cells, immune-related messenger RNA expression signatures, and mutational burden and MSI present in the collected biospecimens. Appropriate methods will be used including but not limited to immunohistochemistry, targeted sequencing, gene expression profiling using RNA sequencing analysis or NanoString analysis, PCR, metabolomics, and/or measurement of other analytes.

8.6.2 Blood sample for RNA and DNA analyses and biomarker studies

Immune-related features measured in blood samples may include immune-related messenger RNA expression signatures, blood TMB, and/or measurement of other analytes.

Additional translational research might be conducted as appropriate including but not limited to the assessment of T-cell clonality, neoantigen expression, metabolomics, proteomics, and presence and changes in the circulating tumor markers such as circulating tumor DNA at screening and following nanrilkefusp alfa and pembrolizumab treatment.

The correlation/relationship between biomarkers in tumor tissue and blood and measures of clinical efficacy, safety, PD activity, and/or mechanism of action will be evaluated.

For patients with mCRPC, CTC count and circulating tumor DNA will be analyzed.

Instructions on sample collection, handling, storage, and shipment of samples are detailed in the study-specific Laboratory Manual provided to the study site.

Samples may be stored for a maximum of 20 years (or according to local regulations) after the end of the study at a facility selected by the sponsor to enable further analysis of biomarker responses to nanrilkefusp alfa in combination with pembrolizumab.

8.7 Immunogenicity assessments

Samples for ADAs will be collected to assess nanrilkefusp alfa tolerability, correlation with PK, and potential AEs associated with ADAs against nanrilkefusp alfa and for the prediction of nanrilkefusp alfa ADA production.

Samples for pembrolizumab ADAs will be collected. These samples will be stored and a decision about their analysis will be taken at a later date.

Instructions on sample collection, handling, storage, and shipment of samples is detailed in the study-specific Laboratory Manual provided to the study site.

Samples may be stored for a maximum of 20 years (or according to local regulations) after the end of the study at a facility selected by the sponsor to enable further analysis of immune responses to nanrilkefusp alfa in combination with pembrolizumab.

8.8 Health economics

Health economics parameters will not be evaluated in this study.

8.9 Pharmacodynamics

PD markers will be assessed in paired tumor tissue collected before and after treatment with nanrilkefusp alfa in combination with pembrolizumab. Activation/expansion of various immune cell populations (leukocytes including NK cells, CD8+ and CD4+ T cells, myeloid cells) will be detected based on the expression of specific markers by immunohistochemistry analysis or genetic analysis (optional).

9 STATISTICAL CONSIDERATIONS

9.1 Statistical hypotheses

No formal testing of statistical hypotheses is planned in this open-label, single-arm trial.

The primary objective is to estimate the antitumor efficacy of nanrilkefusp alfa in combination with pembrolizumab according to RECIST 1.1 by means of ORR, for each indication or disease cohort separately. Other efficacy endpoints (e.g., iORR, [i]PFS, [i]BOR) will also be explored to further evaluate the antitumor efficacy of the combination therapy.

9.2 Analysis sets

9.2.1 All-subjects-as-treated population

The all-subjects-as-treated (ASaT) population will consist of all patients exposed to nanrilkefusp alfa or pembrolizumab.

All safety analyses will be performed on the ASaT population.

9.2.2 PK population

The PK population will consist of all patients who are PK-evaluable. Please also see sections 1.3.5 and 8.4.

9.2.3 Efficacy population

The efficacy population will consist of all patients exposed to the combination therapy for at least one treatment cycle.

This will be the main population for the analyses of the primary endpoint and efficacy, secondary, and exploratory endpoints.

9.2.4 Per Protocol population

The per Protocol (PP) population is defined as all patients who had at least one full treatment cycle of nanrilkefusp alfa and pembrolizumab, did not violate any eligibility criteria, and did not have any major Protocol deviations.

9.3 Statistical analyses

9.3.1 General considerations

Analyses will be descriptive (i.e., without a comparison). Efficacy analyses will be performed for each indication separately. Safety will be analyzed as described in section 9.3.3.

Summary statistics will include:

- Counts and percentages (categorical data)
- Number of observations, mean, standard deviation, median, minimum, and maximum (continuous data)

The ORR, iORR, BOR, iBOR, CBR, iCBR, CTC response, and PSA response will use the exact method based on the binomial distribution[56] to derive 95% confidence intervals (CIs).

Kaplan-Meier estimations and estimations of median (if reached) with log-log 95% CI, Q1 (25th percentile), and Q3 (75th percentile) will be used for time to confirmed PSA progression (mCRPC only), DoR, iDoR, PFS, iPFS, TtR, iTtR, and OS. In case of a confirmed response or disease progression, the date of the first tumor assessment or PSA evaluated as response or progression will be used for the time-to-event variables.

Depending on the amount of missing data, a sensitivity analysis will be performed for time-to-event endpoints. If such an analysis is planned, it will be described in the final Statistical Analysis Plan (SAP).

The last value of PSA, CTC count, CT scan, bone scan, or MRI on or up to 28 days before the date of the first study treatment will be used as the baseline value for each assessment. Only tumor assessments prior to or at the date of initiation of further-line therapy and using CT scan, bone scan, or MRI will be used for the evaluation of tumor response.

9.3.2 Primary endpoint analysis

The ORR is defined as the proportion of patients with CR or partial response (PR). ORR will be analyzed and presented as a point estimate and a 95% CI for each indication separately. Patients with missing data will be considered as non-responders.

Sensitivity analyses will include the use of:

- ASaT and PP populations
- No imputation of missing data

9.3.3 Secondary endpoints analysis

9.3.3.1 Safety endpoints

- TEAEs

A TEAE is defined as an AE that started or worsened at or after the start of study treatment.

TEAEs will be summarized with counts and percentages. These will be provided for the incidence of, but not limited to: any TEAE, any serious TEAE, any grade 3-5 TEAE, any drug-related TEAE, any serious and drug-related TEAE, any grade 3-5 and drug-related TEAE, temporarily discontinued due to a TEAE, permanently discontinued due to a TEAE, death. TEAE incidence frequencies by system organ class (SOC) and preferred term (PT) will also be provided.

- AESIs

AESIs will be summarized with counts and percentages by SOC and PT.

- Safety laboratory findings, vital signs, and ECG

Safety laboratory findings, vital signs measurements, and ECG data will be summarized accordingly (if categorical or continuous data). Laboratory results will be summarized using SI units as appropriate.

Further details on analyses to be performed by indication and overall will be specified in the final SAP.

9.3.3.2 Efficacy endpoints

- iORR

The iORR is defined as the proportion of patients with complete response (iCR) or partial response (iPR) and will be analyzed and presented as a point estimate and a 95% CI for each indication separately. Patients with missing data will be considered as non-responders.

- BOR and iBOR

The BOR and iBOR are defined as the best response from the start of study treatment until progression of disease or death and will be summarized with counts and percentages together with a 95% CI for each indication separately. Stable disease (SD, iSD) needs to last at least 6 weeks from the baseline scan; if not, at least one follow-up scan is required to declare stable disease.

- DoR and iDoR

The DoR and iDoR are defined as the time until progression of disease for patients with partial response (PR, iPR) or complete response (CR, iCR) and will be summarized using Kaplan-Meier estimates for each indication separately. Responders will be considered to have an ongoing response if they: i) have not progressed, and ii) have not started a new anti-cancer therapy, and iii) have not been lost to follow-up, and iv) are alive.

Patients with missing data will be censored/considered as having an event as specified below:

Situation	Date of progression or censoring	Outcome
No progression, no death. No start of new anti-cancer therapy.	Date of the last evaluable tumor assessment.	Censored
No progression, no death. Start of new anti-cancer therapy.	Date of the last tumor assessment with non-progression before the start of new anti-cancer therapy.	Censored
Progression or death after one missed adequate tumor assessment.	Date of progression or death, whichever is earliest (if both occur).	Progressed
Progression or death after more than one missed adequate tumor assessment.	Date of the last evaluable tumor assessment.	Censored

- CBR and iCBR

The CBR and iCBR are defined as the number of partial responses (PR, iPR), complete responses (CR, iCR), and stable diseases (SD, iSD) and will be analyzed and presented as a point estimate and a 95% CI for each indication separately. Stable disease (SD, iSD) needs to last at least 6 weeks from the baseline scan; if not, at least one follow-up scan is required to declare stable disease. Patients with missing data will be considered as non-responders.

- PFS and iPFS

PFS and iPFS are defined as the time from the first day of study treatment until the first date of radiological disease progression or death and will be summarized using Kaplan-Meier estimates for each indication separately.

Patients with missing data will be censored/considered as having an event as specified below:

Situation	Date of progression or censoring	Outcome
Incomplete or no baseline tumor assessment	Date of the first day of study treatment	Censored
Start of new anti-cancer therapy	Date of the last tumor assessment with non-progression before the start of new anti-cancer therapy	Censored
Death before the first disease progression assessment	Date of death	Progressed
Death between adequate tumor assessment visits	Date of death	Progressed
Progression or death after one missed adequate tumor assessment	Date of progression or death, whichever is earliest (if both occur)	Progressed
Progression or death after more than one missed adequate tumor assessment	Date of the last evaluable tumor assessment	Censored
No progression, no death	Date of the last evaluable tumor assessment	Censored

- TtR and iTtR

TtR and iTtR are defined as the time from the first day of study treatment until the first date of partial response (PR, iPR) or complete response (CR, iCR) and will be summarized using Kaplan-Meier estimates for each indication separately. Patients with missing data will be censored at the last assessment date, date of death, or date of the first day of study treatment (if incomplete or no baseline tumor assessments), whichever occurs latest.

- CTC count conversion from ≥5 to <5 cells per 7.5 mL of blood

The CTC response is defined as the proportion of patients with CTC count conversion from ≥5 to <5 cells per 7.5 mL of blood and will be analyzed and presented as a point estimate and a 95% CI for the mCRPC indication. Patients with missing data will be considered as non-responders.

- Confirmed PSA decline of ≥50%

The PSA response is defined as the proportion of patients with a PSA decline of ≥50% and will be analyzed and presented as a point estimate and a 95% CI for the mCRPC indication.

Responses will need confirmation by a second consecutive value obtained 4 or more weeks after the first value indicated a response. Response will be evaluated 6 months post trial entry. Evaluable patients with no confirmed response as defined above will be classified as non-responders.

- Time to confirmed PSA progression

Time to confirmed PSA progression is defined as the time from the first day of study treatment to the date of PSA progression and will be summarized using Kaplan-Meier estimates for the mCRPC indication.

PSA progression is defined as the date when an increase of 25% or more and an absolute increase of 2 ng/mL or more from the nadir are documented. For patients who have a decline in PSA during treatment, PSA progression must be confirmed by a second value 3 or more weeks later increased with respect to the nadir PSA.

9.3.4 Exploratory endpoint analysis

- OS

OS is defined as the time from the first day of study treatment until the date of death and will be summarized using Kaplan-Meier estimates for each indication separately. Patients with missing data will be censored at the last time known to be alive.

9.3.5 Other analyses

Other sensitivity, secondary, and exploratory analyses such as PK, immunogenicity, biomarker exploratory analyses, and circulating tumor DNA fraction (mCRPC only) will be described in the final SAP. The population PK analysis will be presented separately from the main Clinical Study Report.

9.4 Interim analysis

An analysis for futility will be performed when the sample size is considered enough for such analysis and for each indication, as described below in this section. Efficacy data and outputs will also be a part of the Independent Data Monitoring Committee (IDMC) review.

During this period, patient recruitment may be paused if the sponsor deems it appropriate to wait for the results of the futility analysis before enrolling additional patients.

Assuming a desired ORR, the futility analysis will be based on a comparison against a minimal ORR considered as both statistically and clinically relevant improvement as compared to the benchmark ORR. An 80% CI using the exact method[56] for the ORR (alpha = 0.2, alpha = 0.1 one-sided) will be used.

The analysis will be based on the efficacy population.

If, for a certain indication, the ORR is:

1. lower than the minimal ORR, and
2. the 80% CI for the ORR does not include the minimal ORR,

it will be concluded that the combination treatment is futile as compared to benchmark treatment and thus the indication will be discontinued. However, patients still on treatment in the indication can continue combination therapy if recommended by the IDMC.

The criteria for conclusion of futility, for each indication, are defined as follows:

- Indication 1 (NSCLC): Benchmark ORR = 15%; minimal ORR = 23%; desired ORR = 30.6%
  - N = 21 patients if the number of responses is less than 2 (r < 2)
- Indication 2 (MSI-H/dMMR colorectal cancer): Benchmark ORR = 44%; minimal ORR = 54.1%; desired ORR = 62.5%
  - N = 12 patients if the number of responses is less than 4 (r < 4)
- Indication 3 (cSCC): Benchmark ORR = 44%; minimal ORR = 54.1%; desired ORR = 62.5%
  - N = 12 patients if the number of responses is less than 4 (r < 4)
- Indication 4 (advanced hepatocellular carcinoma): Benchmark ORR = 17%; minimal ORR = 25.3%; desired ORR = 33% (not applicable in France)
  - N = 19 patients if the number of responses is less than 2 (r < 2)
- Indication 5 (mCRPC): Benchmark ORR = 5%; minimal ORR = 10.4%; desired ORR = 16.4%
  - N = 35 patients if the number of responses is less than 1 (r < 1)
- Indication 6 (recurrent ovarian cancer): Benchmark ORR = 15%; minimal ORR = 23%; desired ORR = 30.6%
  - N = 21 patients if the number of responses is less than 2 (r < 2)

Additionally, the ORR will be evaluated on an ongoing basis (without stopping of recruitment). Other efficacy endpoints will be used as supportive information.

9.5 Sample size determination

The number of patients per indication to be treated for at least one cycle is based on a Bayesian calculation: for each indication separately, Markov chain Monte Carlo has been used for the sample size determination. Assuming a desired ORR for the number of responses, and a uniform prior distribution between 0 and 1, there will be at least 80% posterior probability to achieve an effect above the minimal ORR:

- Indication 1 (NSCLC): 50 patients
- Indication 2 (MSI-H/dMMR colorectal cancer): 57 patients
- Indication 3 (cSCC): 57 patients

- Indication 4 (advanced hepatocellular carcinoma): 55 patients (not applicable in France)
- Indication 5 (mCRPC): 51 patients
- Indication 6 (recurrent ovarian cancer): 50 patients

This results in a total of 320 patients.

10 SUPPORTING DOCUMENTATION AND OPERATIONAL CONSIDERATIONS

10.1 Appendix 1: Regulatory, ethical, and study oversight considerations

10.1.1 Regulatory and ethical considerations

The Protocol, ICF, and appropriate related documents must be reviewed and approved by an IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) constituted and functioning in accordance with ICH Guideline E6 Good Clinical Practice (GCP)[57] and any local regulations, and by the competent authority (CA). Any Protocol Amendment or revision to the ICF will be resubmitted to the IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) and to the CA for review and approval, except for changes involving only logistical or administrative aspects of the study (e.g., change of clinical research associates [CRAs], change of telephone numbers). Documentation of IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) compliance with ICH E6 and any local regulations regarding constitution and review conduct will be provided to the sponsor.

A signed letter of study approval from the IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) chairman must be sent to the principal investigator or, if regionally required, the head of the medical institution with a copy to the sponsor before study start and the release of study interventions to the site by the sponsor or its designee. If the IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) decides to suspend or terminate the study, the investigator (or if regionally required, the head of the medical institution) will immediately send the notice of study suspension or termination by the IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) to the sponsor.

Study progress is to be reported to IRBs/IECs (in the US)/ECs (in the EU)/equivalent committees (outside the US and EU) and to CAs annually (or as required) by the investigator or sponsor, depending on local regulatory obligations. If the investigator is required to report to the IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU), s/he will forward a copy to the sponsor at the time of each periodic report.

The investigator(s) or the sponsor will submit, depending on local regulations, periodic reports and inform the IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) (or if regionally required, the investigator and the relevant IRB/IEC [in the US]/EC [in the EU]/equivalent committee [outside the US and EU] via the head of the medical institution) and the CA of any reportable AEs per ICH guidelines and local IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) standards of practice. Upon completion of the study, the investigator or sponsor will provide the IRBs/IECs (in the US)/ECs (in the EU)/equivalent committees (outside the US and EU) and the CAs with a brief report of the outcome of the study, if required.

At the end of the study, the sponsor should notify the IRBs/IECs (in the US)/ECs (in the EU)/equivalent committees (outside the US and EU) and CAs as regionally required.

This study will be conducted in accordance with standard operating procedures of the sponsor (or designee), which are designed to ensure adherence to applicable regulatory requirements, and in accordance with the current version of the Declaration of Helsinki and ICH guidelines on GCP.

10.1.2 Financial disclosure

Information on potential financial interests will be provided by all participating investigators. This information will be collected by the sponsor (or a delegated party) before the initiation of the clinical trial, during the course of the clinical trial whenever the financial interests may change, and for a period of one year after the completion of the clinical trial, or termination of a particular individual's participation in this trial.

10.1.3 Informed consent process

10.1.3.1 Clinical trial participation

Written informed consent will be given by each patient before any procedure of this clinical trial is performed. The process of obtaining the informed consent must comply with applicable ICH GCP E6 guidelines as implemented in EU guidelines and national regulatory requirements.

It is the responsibility of the investigator that patients are clearly and fully informed about the purpose of the clinical trial, its potential risks and benefits, and other critical issues regarding the clinical trial, in which the patient volunteers to participate, before undergoing any clinical trial-specific procedure.

The IRB/IEC- (in the US)/EC- (in the EU)/equivalent committee- (outside the US and EU) approved written ICF, which complies with the above-mentioned regulations, will be provided to each patient. The patient should be given ample time to read and to understand the ICF, and to get the answers to any enquiry related to the clinical trial s/he may have. The ICF should be signed personally by the patient and subsequently by the delegated investigator who obtains the consent. The patient will be provided with a fully signed ICF printout, and with any other written information, before participation in the clinical trial.

The process of obtaining informed consent should be documented in the patient's source documents.

The sponsor will provide the investigator with a master ICF that complies with ICH GCP guidelines and regulatory requirements and is considered appropriate for this clinical trial. Any changes to this master ICF suggested by the investigator must be agreed to by the sponsor before being submitted to the IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU). A copy of the approved version, together with all accompanying approvals, must be provided to the sponsor.

The ICF and any other information provided to patients are subject to changes and revisions whenever important new information relevant to patients' willingness to continue participation in the clinical trial becomes available. Once the IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) approval/favorable opinion on this new information is obtained, the delegated investigator should inform each patient about this newly emerging information as soon as possible. All procedures and regulations to be followed and mentioned above apply also to this scenario, and this process should also be documented in the patient's source documents.

10.1.3.2 Pharmacogenomic research

Patients will be asked to participate in pharmacogenomic research (see sections 8.5, 8.6, and 8.9). Depending on local laws and regulations, the patients might be required to provide their

written consent to this research by signing a separate ICF. In such a case, the consent to participate in the pharmacogenomic research will be optional and not a prerequisite for entry into the clinical trial.

10.1.4 Data protection

In order to ensure that personal information of each patient is kept confidential and protected, names and any other information that allow direct identification of a patient will not be on the eCRFs or included in any records or samples provided to the sponsor or sponsor's authorized representatives; such information will be pseudonymized, i.e., all such information will be replaced by a specific code (patient number) assigned by a study doctor and all patients will be identified on eCRFs or any other records or samples by a patient number only.

The personal information collected for the purposes of this study will be held by the study sites, the sponsor, and sponsor's authorized representatives, which together are responsible for processing of personal information in accordance with the applicable data protection laws, including the Swiss Data Protection Act, the Swiss revised Data Protection Act, the General Data Protection Regulation (EU) 2016/679, and any corresponding local legislation.

The sponsor and its authorized representatives will analyze and use the personal information they receive for the purposes of this study only. These include:

- checking patients' suitability to take part in the study,
- monitoring patients' health during treatment with nanrilkefusp alfa in combination with pembrolizumab,
- comparing and pooling study results,
- establishing whether nanrilkefusp alfa in combination with pembrolizumab meets the appropriate standards of safety set by the authorities,
- establishing whether nanrilkefusp alfa in combination with pembrolizumab is effective,
- supporting the clinical development of nanrilkefusp alfa in combination with pembrolizumab,
- supporting the licensing application for regulatory approval of nanrilkefusp alfa in combination with pembrolizumab anywhere in the world,
- supporting the marketing, distribution, sale, and use of nanrilkefusp alfa in combination with pembrolizumab anywhere in the world,
- complying with specific regulations governing clinical trials.

Participation of patients in this study is voluntary and they may withdraw from the study at any time by informing the investigator. Their participation in the study will then end and the study personnel will stop collecting personal information from the patients, but the sponsor will need to retain and use the pseudonymized personal information and associated research results that have already been collected from the patient. The sponsor must do this to comply with its legal and regulatory obligations, to maintain the scientific integrity of the study, and to complete the marketing authorization process for nanrilkefusp alfa in combination with pembrolizumab. It may be necessary to retain certain aspects of pseudonymized (coded) personal information for at least 25 years following the end of the study to comply with applicable laws and regulatory requirements and to ensure the scientific integrity of the study.

If necessary for the study purposes mentioned above, the sponsor may communicate such pseudonymized personal information to third parties (such as service providers, contractors, and research institutions that support the study) and regulatory or other governmental agencies that need to check the results of the study.

These third parties may be located in countries of the European Economic Area (EEA), in Switzerland, in the United States, and in other countries that are outside of the EEA. Some non-EEA countries may not offer the same level of privacy protection. However, the sponsor will keep personal information it receives as confidential as possible within the limits of the law. The sponsor will implement appropriate contractual measures, including the standard data protection contractual clauses, to ensure that the relevant recipients outside the EEA provide an adequate level of protection to personal information as set out in this form and as required by applicable law.

The sponsor, either alone or together with other researchers, may publish or present the results of the study; however, personal information will not be disclosed in any publication or presentation.

All persons have certain rights to gain access to and correct any inaccuracies in the personal information held about them. In certain circumstances, they can also request restriction of processing of their personal information, object to certain types of processing of their personal information, request their personal information be erased and have their personal information provided to them or a third party in a digital format. The sponsor shall comply with the above requests to the fullest extent consistent with other legal and regulatory obligations and where required by law.

Personal data cannot be erased, even after patients finish or terminate their participation in the study, in order to guarantee the validity of the clinical research and to comply with statutory duties and drug authorization requirements.

Representatives from government agencies, the local EC and sponsor or its authorized representatives may also need access to medical records and study records for the purpose of checking data collected for the study.

The sponsor shall process all personal information of the patients in the study in accordance with the applicable data protection laws, including the Swiss Data Protection Act, the Swiss revised Data Protection Act, the General Data Protection Regulation (EU) 2016/679, any applicable local legislation, and the internal data protection policies reflecting organizational and technical arrangements to avoid unauthorized access, disclosure, dissemination, alteration, or loss of information and personal data processed. The organizational and technical measures introduced by the sponsor in relation to a protection of personal information of the patients involve the above mentioned pseudonymization of personal information, appropriate controls to restrict its employees access to the personal information, a physical access control to any premises where the personal information is stored, an electronic access and system control logging for any systems containing personal information, data entry and data transfer control, availability control (back-up and recovery concept), network protection including firewalls and penetration testing procedures for regular testing, industry-standard security policies and procedures including assessment and evaluation of processes and regular training procedures. In the event of any security breach, the incident management procedures would be implemented, and the sponsor would notify such breach as applicable.

10.1.5 Organizational structure of the study

The sponsor of the clinical trial, SOTIO, may delegate certain tasks to designees, e.g., a contract research organization (CRO) or other third-party vendors. A list of such designated collaborators, including their contact details, will be documented in the investigator's manuals/Investigator Site File.

10.1.6 Dissemination of clinical study data

By signing the Protocol (INVESTIGATOR'S DECLARATION), every participating investigator agrees to keep all information and results concerning the clinical trial and the investigational product strictly confidential. The confidentiality obligation also applies to all personnel involved at the investigational site.

10.1.7 Data quality assurance

An eCRF is required and must be completed for each patient by qualified and authorized personnel. All data on the eCRF must reflect the corresponding source document. Any correction to entries made on the eCRF must be documented in a valid audit trail where the correction is dated, the individual making the correction is identified, the reason for the change is stated, and the original data are not obscured. Only data required by the Protocol for the purposes of the study should be collected. The investigator must sign the eCRFs.

10.1.8 Trial monitoring, access to source documentation, and data retention

Before trial initiation, at the investigators' meeting or during the Site initiation visit, a representative of the sponsor or a CRO will review the Protocol and eCRFs with the site staff. During the clinical trial, the CRA will oversee the progress of the clinical trial and will visit the site regularly to verify the completeness of patient records, reliability and accuracy of entries on eCRFs, the site's adherence to the Protocol and GCP, the progress of enrollment, and that study interventions are being stored, dispensed, and accounted for according to specifications. The CRA will also ensure that the safety and rights of the patients are not compromised. Key clinical trial personnel must be available to assist the CRA during these visits.

For each patient recruited into the clinical trial, the investigator must maintain source documents, which should consist of case and visit notes (hospital or outpatient clinic medical records) containing demographic and medical information, laboratory data, and results of any other tests or assessments. All information on eCRFs must be traceable to these source documents in the patient's file. The investigator must also keep the original of the ICF signed by the patient (the second signed original must be given to the patient).

The investigator must allow the CRAs or other delegated representatives to visit all site locations, and to allow direct access to all clinical trial-related documentation and data, as well as to the patient's source documents and other charts and records.

Monitoring standards of the sponsor require full verification for the presence of a fully signed and dated ICF, adherence to the inclusion/exclusion criteria, documentation of SAEs, and recording of data that will be used for all primary and safety variables. Additional verification of the consistency of the source data with the eCRFs will be performed according to a clinical trial-specific Monitoring Plan.

Monitoring details describing strategy (e.g., risk-based initiatives in operations and quality such as risk management and mitigation strategies and analytical risk-based monitoring), methods, responsibilities and requirements, including handling of noncompliance issues and monitoring techniques (central, remote, or on-site monitoring) are provided in the Monitoring Plan.

The sponsor assumes accountability for actions delegated to other individuals (e.g., CROs).

The trial may also be evaluated by any other designee delegated by the sponsor, IRB/IEC (in the US)/EC (in the EU)/equivalent committee (outside the US and EU) representatives, or by any independent institutional or government inspectors, who must be given direct access to the same as mentioned above. Such a possibility must also be clearly mentioned in the patient's ICF.

The investigator must promptly notify the sponsor or its delegated representatives any time the request for inspection is raised by any regulatory agency, and to provide copies of all documentation received from such an agency.

The investigator or institution must retain all trial-related records, materials provided by the sponsor or its delegated collaborators, copies of eCRFs, and source documents of patients for a specified period of time. This period of time is derived from the locally applicable regulation guidelines or requirements, institution procedures, or requirements stipulated by the sponsor or its representatives, whichever is longer.

It is requested that at the completion of the required retention period, or should the investigator retire or relocate, the investigator contact the sponsor, allowing the sponsor the option of permanently retaining the study records.

10.1.9 Study and site start and closure

The first act of recruitment is the signature of the ICF by the first patient enrolled and will be the study start date.

The end of the study is defined in section 4.4.

10.1.10 Publication policy

The results of this clinical trial will be published and/or presented at scientific meetings in their totality in a timely manner. No single institution presentation/publication of data is foreseen if not agreed by the sponsor and all relevant investigators of the study. Any formal publication of clinical trial results will be a collaborative effort between the sponsor and the investigator(s). All manuscripts or abstracts will be reviewed and approved in written by the sponsor before submission. The sponsor may request a delay in publication if there are important intellectual property concerns but does not have the right to suppress the publication of the clinical trial results indefinitely.

Authorship will be determined by mutual agreement, with the coordinating investigator of this clinical trial being given priority for first authorship. Publications will include all clinical trial investigators in the order of their relative patient contribution, taking into account also contributions during Protocol development and data analyses.

10.1.11 Independent Data Monitoring Committee

An IDMC will be established for this study to safeguard the interest and safety of the patients participating in the study and provide independent review and assessment of the efficacy and safety data in a systematic manner. In addition, the IDMC will review safety data from the first 10-12 patients across all indications.

The efficacy results will be presented via descriptive statistics during systematic reviews, no testing is planned.

The IDMC will be tasked with making a recommendation to the sponsor to continue, modify, or stop recruitment or the trial based on their assessment of efficacy and safety information. The membership, key responsibilities of the IDMC, and corresponding procedures will be defined in the IDMC charter.

10.1.12 Future research

Biological samples collected during this study may be stored for future research by the sponsor. Storage conditions will be in compliance with the standards for repositories of biological samples, according to the "Recommendation CM/Rec (2016)6 of the Committee of Ministers to member States on research on biological materials of human origin".[58]

The repository will have independent monitoring that will guarantee protection of the data and the patients' interests (also see section 10.1.4). Except for contractors of the sponsor, the samples will not be transferred to third parties, and they will not be sold to third parties. The exploratory studies that will be conducted with the samples will undergo a rigorous independent review evaluating both ethical and scientific aspects.

10.2 Appendix 2: Description of the iRECIST process for assessment of disease progression

iRECIST is based on RECIST 1.1 but adapted to account for the unique tumor response seen with immunotherapeutic drugs. iRECIST will be used by the investigator to assess tumor response and progression, and to guide decisions about changes in management.

10.2.1 Assessment at screening and prior to RECIST 1.1 progression

Until radiographic disease progression based on RECIST 1.1, there is no distinct iRECIST assessment.

10.2.2 Assessment and decision at RECIST 1.1 progression

Patients who show radiological disease progression by RECIST 1.1 will continue treatment until repeat scans are obtained, as described in section 8.1.1.

Tumor flare may manifest as any factor causing radiographic progression per RECIST 1.1, including:

- Increase in the sum of diameters of target lesion(s) identified at baseline to ≥20% and ≥5 mm from nadir

  Note: The iRECIST publication uses the terminology "sum of measurements", but "sum of diameters" will be used in this Protocol, consistent with the original RECIST 1.1 terminology.

- Unequivocal progression of non-target lesion(s) identified at baseline
- Development of new lesion(s)

iRECIST defines new response categories, including unconfirmed progressive disease (iUPD) and confirmed progressive disease (iCPD). For purposes of iRECIST assessment, the first visit showing progression according to RECIST 1.1 will be assigned a visit (overall) response of iUPD, regardless of which factors caused the progression.

At this visit, target and non-target lesions identified at baseline by RECIST 1.1 will be assessed as usual.

New lesions will be classified as measurable or non-measurable, using the same size thresholds and rules as for baseline lesion assessment in RECIST 1.1. From measurable new lesions, up to 5 lesions total (up to 2 per organ), may be selected as new lesions – target. The sum of diameters of these lesions will be calculated and kept distinct from the sum of diameters for target lesions at baseline. All other new lesions will be followed qualitatively as new lesions – non-target.

10.2.3 Assessment at the confirmatory scans

On the confirmatory scans, the patient will be classified as progression confirmed (with an overall response of iCPD), or as showing persistent unconfirmed progression (with an overall response of iUPD), or as showing disease stability or response (iSD/iPR/iCR according to iRECIST).

10.2.4 Confirmation of progression

Progression is considered confirmed, and the overall response will be iCPD, if ANY of the following occurs:

- Any of the factors that were the basis for the initial iUPD to show worsening
  - For target lesions, worsening is a further increase in the sum of diameters of ≥5 mm, compared to any prior iUPD time point.
  - For non-target lesions, worsening is any significant growth in lesions overall, compared to a prior iUPD time point; this does not have to meet the "unequivocal" standard of RECIST 1.1.
  - For new lesions, worsening is any of these:
    - An increase in the new lesion sum of diameters by ≥5 mm from a prior iUPD time point
    - Visible growth of new non-target lesions
    - The appearance of additional new lesions
- Any new factor appears that would have triggered disease progression by RECIST 1.1

10.2.5 Persistent iUPD

Progression is considered not confirmed, and the overall response remains iUPD, if:

- none of the progression-confirming factors identified above occurs, AND

- the target lesion sum of diameters (initial target lesions) remains above the initial disease progression threshold (by RECIST 1.1).

Additional scans for confirmation are to be scheduled 4 to 8 weeks from the scans on which iUPD is seen. This may correspond to the next visit in the original visit schedule. The assessment of the subsequent confirmation scan proceeds in an identical manner, with possible outcomes of iCPD, iUPD, and iSD/iPR/iCR.

10.2.6 Resolution of iUPD

Progression is considered not confirmed, and the overall response becomes iSD/iPR/iCR, if:

- none of the progression-confirming factors identified above occurs, AND
- the target lesion sum of diameters (initial target lesions) is not above the initial disease progression threshold.

The response is classified as iSD or iPR (depending on the sum of diameters of the target lesions), or iCR if all lesions resolve.

In this case, the initial iUPD is considered to be pseudoprogression, and the level of suspicion for progression is "reset". This means that the next visit that shows radiographic progression, whenever it occurs, is again classified as iUPD by iRECIST, and the confirmation process is repeated before a response of iCPD can be assigned.

10.2.7 Management following the confirmatory scan

If repeat scans do not confirm disease progression per iRECIST, as assessed by the investigator, and the patient continues to be clinically stable, study interventions are to continue and the regular scan schedule is to be followed. If disease progression is confirmed, patients may be discontinued from study interventions.

Note: If a patient has iCPD and clinically meaningful, study interventions may be continued after consultation with the sponsor. In this case, if study interventions are continued, tumor imaging should continue to be performed following the intervals as outlined in section 8.1.1.

10.2.8 Detection of progression at visits after pseudoprogression resolves

After resolution of pseudoprogression (i.e., after iSD/iPR/iCR), another instance of progression (another iUPD) is indicated by any of the following events:

- Target lesions
  - Sum of diameters reaches the disease progression threshold (≥20% and ≥5 mm increase from nadir) either for the first time or after resolution of previous pseudoprogression. The nadir is always the smallest sum of diameters seen during the entire trial, either before or after an instance of pseudoprogression.
- Non-target lesions
  - If non-target lesions have never shown unequivocal progression, their doing so for the first time results in iUPD.
  - If non-target lesions have shown previous unequivocal progression, and this progression has not resolved, iUPD results from any significant further growth of non-target lesions, taken as a whole.

- New lesions
  - New lesions appear for the first time
  - Additional new lesions appear
  - Previously identified new target lesions show an increase of ≥5 mm in the new lesion sum of diameters, from the nadir value of that sum
  - Previously identified non-target lesions show any significant growth

If any of the events above occur, the overall response for that visit is iUPD, and the iUPD evaluation process (see section 10.2.3) is repeated. Progression must be confirmed before iCPD can occur.

The decision process on the subsequent iUPD is identical to the iUPD confirmation process for the initial disease progression, with one exception, which can occur if new lesions had occurred at a prior instance of iUPD, had not resolved, then worsened (increase in size or number) leading to the second iUPD. If new lesion worsening has not resolved at the confirmatory scan, then iUPD cannot resolve to iSD or iPR. It will remain iUPD until either a decrease in the new lesion burden allows resolution to iSD or iPR, or until new or worsening causes of progression indicates iCPD.

Additional details about iRECIST are provided in the iRECIST publication.[36]

10.3 Appendix 3: Abbreviations

Abbreviation	Term
ADA	Anti-drug antibody
AE	Adverse event
AESI	Adverse event of special interest
ALK	Anaplastic lymphoma kinase
ALT	Alanine transaminase
ASaT	All-subjects-as-treated
AST	Aspartate transaminase
BOR	Best overall response (according to RECIST 1.1)
CA	Competent authority
CBR	Clinical benefit rate (according to RECIST 1.1)
CD	Cluster of differentiation
CI	Confidence interval
CR	Complete response (according to RECIST 1.1)
CRA	Clinical research associate
CRO	Contract research organization
cSCC	Cutaneous squamous cell carcinoma
CT	Computed tomography
CTC	Circulating tumor cell
CTCAE	Common Terminology Criteria for Adverse Events
CTLA-4	Cytotoxic T lymphocyte-associated protein 4
dMMR	Mismatch repair deficient
DoR	Duration of response (according to RECIST 1.1)
DRESS	Drug rash with eosinophilia and systemic symptom
EC	Ethics committee
ECG	Electrocardiography
ECOG	Eastern Cooperative Oncology Group
eCRF	Electronic Case Report Form
EEA	European Economic Area
EGFR	Epidermal growth factor receptor
Fc	Fragment crystallizable
FDA	US Food and Drug Administration

Abbreviation	Term
FoxP3	Forkhead box P3
GCP	Good Clinical Practice
HBV	Hepatitis B
HCV	Hepatitis C
IB	Investigator's Brochure
iBOR	Best overall response (according to iRECIST)
iCBR	Clinical benefit rate (according to iRECIST)
ICF	Informed Consent Form
ICH	International Council for Harmonisation
iCPD	Confirmed progressive disease (according to iRECIST)
iCR	Complete response (according to iRECIST)
IDMC	Independent Data Monitoring Committee
iDoR	Duration of response (according to iRECIST)
IEC	Independent ethics committee
Ig	Immunoglobulin
IL	Interleukin
IL-2R	IL-2 receptor
IL-15R	IL-15 receptor
iORR	Objective response rate (according to iRECIST)
iPFS	Progression-free survival (according to iRECIST)
iPR	Partial response (according to iRECIST)
irAE	Immune-related adverse event
IRB	Institutional review board
iRECIST	Response Evaluation Criteria In Solid Tumors for immune-based therapeutics
iSD	Stable disease (according to iRECIST)
iTtR	Time to response (according to iRECIST)
iUPD	Unconfirmed progressive disease (according to iRECIST)
IV	Intravenous
JAK	Janus kinase
mAb	Monoclonal antibody
MAPK	Mitogen-activated protein kinase

Abbreviation	Term
mCRPC	Metastatic castration-resistant prostate cancer
MRI	Magnetic resonance imaging
MSI(-H)	Microsatellite instability(-high)
MUGA	Multigated acquisition scanning
NK	Natural killer
NSAID	Non-steroidal anti-inflammatory drug
NSCLC	Non-small cell lung cancer
ORR	Objective response rate (according to RECIST 1.1)
OS	Overall survival
PCWG3	Prostate Cancer Clinical Trials Working Group 3
PD	Pharmacodynamic(s)
PD-1	Programmed cell death protein 1
PD-L1	Programmed cell death ligand 1
PD-L2	Programmed cell death ligand 2
PFS	Progression-free survival (according to RECIST 1.1)
PI3K	Phosphatidylinositol-4,5-bisphosphate 3-kinase
PK	Pharmacokinetic(s)
PP	Per Protocol
PR	Partial response (according to RECIST 1.1)
PSA	Prostate-specific antigen
PT	Preferred term
Q1	25th percentile
Q2W	Every 2 weeks
Q3	75th percentile
Q3W	Every 3 weeks
RECIST 1.1	Response Evaluation Criteria In Solid Tumors version 1.1
SAE	Serious adverse event
SAP	Statistical Analysis Plan
SD	Stable disease (according to RECIST 1.1)
SHP-1	Tyrosine phosphatase
SHP-2	Tyrosine phosphatase
SJS	Stevens-Johnson syndrome

Abbreviation	Term
SOC	System organ class
STAT	Signal transducer and activator of transcription
T1DM	Type 1 diabetes mellitus
T3	Triiodothyronine
T4	Thyroxine
TEAE	Treatment-emergent adverse event
TEN	Toxic epidermal necrolysis
TMB	Tumor mutational burden
Treg	Regulatory T cell
TSH	Thyroid stimulating hormone
TtR	Time to response (according to RECIST 1.1)
ULN	Upper limit of normal
WOCBP	Woman of childbearing potential
ZAP70	Zeta-chain-associated protein kinase

Abbreviations of commonly used weight, height, and volume measures are not listed above.

10.4 Appendix 4: Protocol Amendment history

The PROTOCOL AMENDMENT SUMMARY OF CHANGES for the current Protocol Amendment is located directly before the TABLE OF CONTENTS.

10.5 Appendix 5: Country-specific requirements/differences

Country	Section(s) in Protocol Amendment 1	Country-specific requirements/differences
Czech Republic (per country-specific amendment CZ-2 dated 28Apr2022)	1.3.1 Schedule of activities, nanrilkefusp alfa combined with pembrolizumab	"In case pembrolizumab needs to be discontinued, nanrilkefusp alfa treatment can continue until disease progression or unacceptable toxicity and the schedule of activities for patients receiving nanrilkefusp alfa combined with pembrolizumab will apply" was replaced with "In case pembrolizumab needs to be discontinued, nanrilkefusp alfa treatment will be discontinued as well."
	7.1 Discontinuation of study intervention	"In case pembrolizumab needs to be discontinued, nanrilkefusp alfa treatment can continue until disease progression or unacceptable toxicity" was replaced with "In case pembrolizumab needs to be discontinued, nanrilkefusp alfa treatment will be discontinued as well."
France (per country-specific amendment FR-2 dated 23Aug2022)	1.1 Synopsis, Study population; 1.2 Schema; 4.2 Scientific rationale for study design	"Advanced hepatocellular carcinoma after recurrence or failure of an immune checkpoint inhibitor" is not applicable
	1.1 Synopsis, Estimated sample size	"Advanced hepatocellular carcinoma: 55 patients" is not applicable
	1.1 Synopsis, Statistical considerations; 9.4 Interim analysis	"Indication 4 (advanced hepatocellular carcinoma): Benchmark ORR = 17%; minimal ORR = 25.3%; desired ORR = 33%" is not applicable
	5.1 Inclusion criteria, criterion 4	"Advanced hepatocellular carcinoma" is not applicable
	9.5 Sample size determination	"Indication 4 (advanced hepatocellular carcinoma): 55 patients" is not applicable

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