Document Coversheet

Study Title:

A Phase II Study of Type-1 Polarized Dendritic Cell (αDC1) based Treatment in Combination with Tumor-Selective Chemokine Modulation (CKM: Interferon alpha 2b, Rintatolimod and Celecoxib) in Melanoma Patients with Primary PD-1/PD-L1 Resistance

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Confidentiality Statement

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1 OBJECTIVES

1.1 Primary Objective

- To evaluate the objective response rate to treatment with an autologous αDC1/TBVA cell-based treatment (alpha-type-1-polarized dendritic cells loaded with tumor blood vessel-targeting antigenic peptides) plus cytokine modulating (CKM) regimen (rintatolimod, IFN-α2b and, celecoxib) in HLA-A2+ subjects with primary PD-1 resistant IO-refractory melanoma (who will continue the original PD-1/PD-L1-regimen for 12 weeks).

1.2 Secondary Objectives*

- To evaluate the immune-related objective response rate (ORR; per iRECIST;) in the above patient population treated with autologous αDC1/TBVA cell-based treatment plus cytokine CKM regimen followed by continued treatment with PD-1/PD-L1 blockade (± CTLA4 blockade or LAG3 blockade)
- Evaluate rate of durable responses (> 6 months) on the combination treatment in the above patient population treated with autologous αDC1/TBVA cell-based treatment plus cytokine CKM regimen followed by continued treatment with PD-1/PD-L1 blockade (± CTLA4 blockade or LAG3 blockade)

* Note: The secondary objectives are in reference to patients that continue on with either PD-1/PD-L1 and/or CTLA4 blockade after meeting the 12-week primary objective.

1.3 Exploratory Objective

- Examine whether the combination of peptide-loaded autologous αDC1 cell-based treatment and tumor-selective chemokine modulation (CKM: IFN-a2b, rintatolimod, and celecoxib) improves the OS and iPFS in HLA-A2+ subjects PD-1/PD-L1-refractory melanoma compared to the historical control of the best supportive care.
- Identify the intratumoral and systemic immune correlates of the response to treatment

2 BACKGROUND

The incidence of melanoma continues to rise, with the American Cancer Society estimating that over 91,000 patients will be diagnosed with this form of cancer in 2018, with an estimated death toll of over 9,320 (1). Despite recent therapeutic successes for immune checkpoint blockade in the setting of advanced-stage disease, only a minority of treated melanoma patients manifest durable benefit (2-5), reinforcing the need to develop 2nd-line therapies that are effective against checkpoint-refractory disease. Results from our recently completed phase II clinical trial UPCI 12-048 ("A Randomized Phase II Pilot Study of Type I-Polarized Autologous Dendritic Cell Vaccines Incorporating Tumor Blood Vessel Antigen (TBVA)-Derived Peptides in Combination with Dasatinib in Patients with Metastatic Melanoma"(6), suggests a link between patient objective clinical response and their ability to develop: i) a pro-inflammatory CK-dominated tumor microenvironment (i.e. increased expression of pro-inflammatory CKs CCL5/CCL19/CCL21/CXCL10 and reduced expression of regulatory CKs CCL22/CXCL12); ii) peripheral Type-1 CD8+ T cell responses against vaccine-incorporated antigens (as well as TBVA and melanoma antigens not in the vaccine formulation; i.e. as an index of "epitope spreading"), iii) increased TCR convergence (focus on specific MHC/peptide complexes) and the presence of

unique TIL clonotypes not detectable in peripheral blood (TCRseq) and, iv) a biomarker signature for tertiary lymphoid structures (TLS) within their tumors (including the canonical TLS-associated CKs CCL19, CCL21) 5 weeks after the initiation of therapy, and in advance of spreading in the CD8+ T cell repertoire or CT-documented objective clinical response (OCR; RECIST). Overall, **objective clinical benefit was observed in 6 of 13 (46%) evaluable patients, including 4/7 (57%) patients exhibiting prior primary resistance to PD1 blockade.** Importantly for the primary endpoint of this study, **all of the objective clinical responses were observed already within 9 weeks of αDC1/TBVA treatment** of PD1-non-responsive patients. New preliminary data developed in mouse melanoma models supports the superior anti-tumor efficacy of αDC1/TBVA vaccines in HLA-A2 Tg mice when combined with the CKM regimen vs. dasatinib, prompting the performance of our proposed innovative clinical trial.

2.1 Pre-Clinical Studies

Intratumoral density of effector T cells (CD8+ CTLs) in melanoma tissue predicts patients' responsiveness to PD-1/PD-L1 blockade, and overall clinical outcomes (7-17). Our preclinical data show that melanoma antigen-loaded alpha-type-1-polarized dendritic cells (αDC1s) induce highly effective melanoma-specific CD8+ CTLs that express chemokine receptors CCR5 and CXCR3. We also observed that the treatment of melanoma or other cancer tissues with interferon-alpha (IFNα) combined with a TLR3 ligand synergistically enhances local production of the relevant chemokines able to attract CCR5- and CXCR3-expressing CTLs (CXCL10/IP10 and CCL5/RANTES) selectively in the tumor lesions, rather than in marginal healthy tissues, while simultaneously suppressing local production of chemokines that attract undesirable Treg cells (CCL22). Our completed phase I of clinical trial NCT01545141 in patients with advanced colorectal cancer demonstrated that the chemokine modulatory combination (CKM) involving one-to two five-day-long cycles of (IFNα) (Intron-A, up to 20MU/M2; i. v.) with rintatolimod (Ampligen, short half-life selective TLR3 ligand formulated for i. v. use; 200 mg) and celecoxib (Celebrex, selective COX2 blocker, 2 x 200 mg; p.o.) is safe and is capable of significantly enhancing the ratios between CTL-attractants versus Treg attractants in patients' tumor tissues. Our additional experience within phase I/II trial NCT02151448 (four cycles of CKM combined with αDC1 vaccine loaded with autologous tumor material) further demonstrated very good tolerability of this combination treatment (unpublished data from University of Pittsburgh collaborators).

We and others have also shown that dendritic cell (DC) vaccines targeting antigens that are selectively (over)expressed by tumor associated vascular endothelial cells (VEC) or pericytes (TBVA: DLK1, EphA2, HBB, NRP1, RGS5 and TEM) (12), are both safe and effective against established murine tumors,(10, 15, 16) including B16 and BP ($BRAF^{V600E}$PTEN-/-) melanomas. Importantly, we failed to detect, (i.) vaccine-induced T cell reactivity against normal tissue pericytes or VEC, (ii.) delay in the kinetics of skin closure after full thickness wounding or, (iii.) alterations in the pregnancy or litter sizes of female mice vaccinated against TBVA (16, 18).

2.2 Clinical Studies

Based on the results developed in HLA-A2 Tg (HHD) mice, the FDA approved the translation of this dendritic cell-based treatment approach in a first-in-human phase II clinical trial (UPCI 12-048; NCT01876212) of an autologous alpha-DC1/TBVA peptide-based cell therapy (coordinately targeting 6 TBVA: DLK1, EphA2, HBB, NRP1, RGS5 and TEM) (19) for the treatment of

advanced-stage melanoma; observing their safety and ability to induce radiologic responses in patients with advanced melanomas who previously progressed on checkpoint blockade. Importantly, ***in the subset of patients with primary PD-1-reistance (7/13 patients), the objective response rate (ORR) was 57% (4 of 7 patients)*** providing a strong rationale to evaluate the ability of αDC1/TBVA to reverse the primary PD1 resistance.

2.3 Rationale

Our preliminary translational and clinical (UPCI 12-048/NCT01876212) data support the ability of treatment consisting of αDC1/TBVA + agents to augment pro-inflammatory CKs (and reduce regulatory CKs) in the TME to improve therapeutic TIL recruitment/function and yield OCR in melanoma patients exhibiting prior primary checkpoint resistance. Although we observed a 57% response rate to αDC1/TBVA vaccination + dasatinib-based immunotherapy in UPCI 12-048, our translational modeling suggests that treatment with a combination vaccine + CKM regimen may yield greater therapeutic efficacy based on more robust changes in CK profiles in the TME, including augmented expression of CCL5, CCL19, CCL21, CXCL10 and CXCL13 (associated with naïve/effector T cell recruitment and TLS formation) and reduced expression of regulatory cell-recruiting CKs CCL22 and CXCL12 (**Figure** 1)

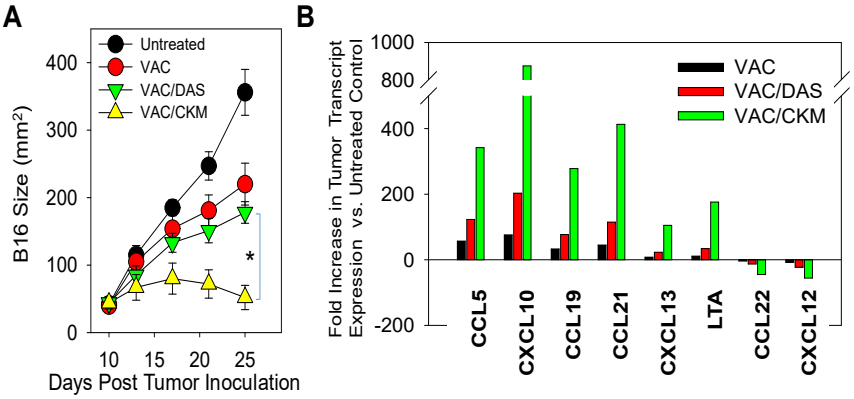


Figure 1. αDC1/TBVA vaccines exhibit greater anti-tumor efficacy when combined with CKM vs. dasatinib. HLA-A2 Tg mice bearing established day 10 subcutaneous (SC). B16 melanomas (right flank) were left untreated or they were treated by SC injection (left flank) with 5 x 10^5 syngeneic bone marrow-derived αDC1 loaded with TBVA peptides (VAC; identical to those used in UPCI 12-048/NCT01876212) on days 7, 14 and 21 ± i.p. dasatinib (DAS; 0.1 mg/mouse) or CKM [rintatolimod (50 μg), rmIFNα (10,000U), celecoxib (75 μg)]. Tumor cell growth was monitored every 3-4 days (A), with harvested tumor biopsies analyzed by qRT-PCR (B) for the indicated analytes on day 25 (n = 5 mice/group); *$p < 0.05$ for vaccine/CKM vs. vaccine/DAS on days > 13.

Our group has already demonstrated (NCT01545141, NCT02151448) that the CKM regimen (involving 1-2 five-day-long cycles of (IFNα) (Intron-A, up to 20 MU/M^2; i. v.) with rintatolimod (Ampligen, short half-life selective TLR3 ligand formulated for i. v. use; 200 mg) and celecoxib (Celebrex, selective COX2 blocker, 2 x 200 mg; p.o.) is both safe and capable of significantly enhancing the ratio between CTL-attractants (inflammatory CKs) versus Treg attractants (regulatory CKs) in patients' tumor tissues. In the case of NCT02151448, CKM treatment was combined with an αDC1-base vaccine (i.e. DC loaded with autologous tumor material) and found to exhibit very good tolerability.

In view of these preliminary data, we hypothesize that the treatment of PD-1/PD-L1-refractory melanoma patients with the combination of αDC1 vaccines loaded with DLK1, EphA2, HBB, NRP1, RGS5 and TEM peptides, and CKM (to enhance the entry of cytotoxic T-lymphocytes into melanoma lesions) will promote both systemic immunization and robust infiltration of vaccination-induced CD8+ CTLs to tumor lesions, and, subsequently, tumor regression with improved patient survival.

Rationale for using Bioferon as a component of CKM. While the original protocol involved INTRON-A (Merck-produced IFNα2b), as a component of the CKM regimen (in addition to rintatolimod and celecoxib), Merck's decision to discontinue the production of INTRON®-A, forced us to identify an alternative IFNa2b to perform the trial and revise the original BB-IND16,704. We have recently identified (Bioferon®, from Argentinian supplied by Biosidus S.A.), as a IFNα2b product used at the same 20 MU/M^2 dose as the INTRON®-A dose used in the trial conducted under BB-IND16,704. Bioferon® is not FDA-approved and is considered an investigational new drug in the United States. For safety reason, in analogy to the recently approved ND 163681 (also held by Roswell Park and FDA-approved on October 14, 2022), which covers the same CKM + PD-1/PD-L1 blockade regimen but without a DC vaccine component, we propose an additional lead-in phase I cohort of 3 patients to be treated with reduced-dose CKM including Bioferon at a dose of 10 MU/m^2. Following the testing of lower dose of IFNα2b in these 3 initial patients, and in the absence of dose-limiting adverse effects, we will continue the accrual of patients to be treated using a Bioferon dose of 20MU/m2. We will retain the original protocol design but consider safety as the formal primary endpoint for these additional 3 patients. The data from these patients will be shared between the lead-in and phase II component for overall treatment efficacy evaluations.

The use of Bioferon to replace INTRON A, was FDA approved on October 14, 2022 (IND 163681 - held by Roswell Park). The revision of BB-IND16,704 includes a letter of cross-reference to IND 163681.

3 INCLUSION AND EXCLUSION CRITERIA

This Phase II trial will be applicable to HLA-A2+ subjects with primary PD-1/PD-L1 resistant IO-refractory melanoma with normal organ and marrow function.

3.1 Inclusion Criteria

To be included in this study, participants must meet the following criteria:

1. Age ≥ 18 years of age.
2. Participant must be HLA-A2+. Retesting is not required for patients who have previous documented positivity.
3. Have IO-refractory melanoma with primary PD-1/PD-L1-resistance. *Note*: Any lines of prior therapies are allowed, but **the last line needs to include an anti PD-1 or anti PD-L1 agent.** The prior treatments may include any standard and/or experimental therapies.
4. Have ≥ 1 tumor site amenable to core needle biopsy that is not the site of disease used to measure antitumor response
5. Have measurable disease based on RECIST 1.1 criteria present,
6. Have an ECOG Performance Status of 0-2. Refer to Appendix A**.**

7. Have normal organ and marrow function as defined below:
    - Platelets ≥ 75,000/ microliter
    - Hemoglobin ≥ 9 g/ deciliter
    - Absolute Neutrophil Count (ANC) ≥ 1500/microliter
    - Creatinine < 1.5 X institutional upper limit of normal (ULN) OR creatinine clearance ≥ 50 mL/min by Cockroft-Gault formula for subjects with creatinine levels ≥ 1.5 x ULN (Appendix D).
    - Total bilirubin not greater than 1.5 X institutional ULN, except for patients with known Gilberts Syndrome, who are eligible to no more than 2 X institutional ULN.
    - AST(SGOT) and ALT(SGPT) no greater than 3 x institutional ULN OR, no greater than 5 x ULN for subjects with liver metastases
8. Participants of child-bearing potential must agree to use adequate contraceptive methods (e.g., hormonal or barrier method of birth control; abstinence) prior to study entry. Should a woman become pregnant or suspect she is pregnant while she or her partner is participating in this study, she should inform her treating physician immediately.
9. Participant must understand the investigational nature of this study and sign an Independent Ethics Committee/Institutional Review Board approved written informed consent form prior to receiving any study related procedure.
10. Candidate for continuation/resumption of anti-PD-1/PD-L1 blockade (in parallel to DC vaccine and CKM).

Refer to Appendix B for the Investigator Study Eligibility Verification Form: Inclusion Criteria.

3.2 Exclusion Criteria

Participants will be excluded from this study for the following:

1. Is currently being treated with systemic immunosuppressive agents, including steroids: Subjects will be ineligible until 3 weeks after removal from immunosuppressive treatments, *except* when they are administered as replacement therapy for endocrine dysfunction (and receive no more than 10 mg prednisone or equivalent: inhaled steroids are allowed).
2. Has had prior anti-cancer therapy within 2 weeks prior to study day 1 or who has not recovered (i.e., no more than Grade 1 or at baseline) from adverse events due to a previously administered agent, except for neuropathy (no more than Grade 2) or alopecia or vitiligo (any grade).
3. Has a known additional malignancy that is progressing or requires active treatment.
4. Has known active central nervous system (CNS) metastases and/or carcinomatous meningitis. Treated brain metastases are allowed, if stable for more than 4 weeks (and receive no more than 10 mg prednisone or equivalent: inhaled steroids are allowed).
5. Has a history of cardiac event(s) (acute coronary syndrome, myocardial infarction, or ischemia (within 3 months of signing consent) or, subject has a New York Heart Association classification of III or IV (Appendix E).
6. Has an active infection requiring systemic therapy.
7. Has known active Hepatitis B or Hepatitis C infection.

8. Has known immunosuppressive disease (e.g. HIV, AIDS or other immune depressing disease). Testing is not mandatory.
9. Has known serious hypersensitivity reactions to peg-interferon alpha-2b or interferon alpha-2b.
10. Prior allergic reaction or hypersensitivity to sulfonamides, celecoxib, or NSAIDs.
11. Has received a blood transfusion in the two weeks prior to leukapheresis.
12. Women of childbearing potential who are pregnant or nursing.
13. Unwilling or unable to follow protocol requirements.
14. Any condition which in the Investigator's opinion deems the participant an unsuitable candidate or unacceptable risk to receive study drug regimen.
15. Patients who showed initial response to PD-1/PD-L1 blockade and developed secondary resistance.

Refer to Appendix C for the Investigator Study Eligibility Verification Form: Exclusion Criteria.

3.3 Special Populations

The following special populations will be excluded from this study:

- Cognitively impaired adults/adults with impaired decision-making capacity
- Individuals who are not yet adults (infants, children, teenagers)
- Pregnant women
- Prisoners

3.4 Inclusion of Women and Minorities

The integrated clinical trial is open to patients of any sex, race, or ethnic background. Based on the SEER Database, the incidence of melanoma is greater in males versus females and in Caucasians versus minorities. We expect to enroll minorities proportional to their representation in the Buffalo, NY regional population.

No patients will be excluded based on sex, race, or ethnic origin. An attempt to increase enrollment of women and minorities will be made by promoting the clinical trial through several unique Roswell Park programs designed to reach out to women and minorities, increase their awareness of clinical trial, and help support them should they choose to participate in clinical research protocols.

3.5 Inclusion of Children

Only subjects over 18 years of age will be included in the clinical protocol. Melanoma is rare in children under 18 years of age, and we do not expect to see a child under 18 who is qualified for inclusion. We would work in collaboration with our pediatric oncology colleagues should this occur.

4 LOCAL AND STUDY-WIDE NUMBER OF SUBJECTS

A maximum of 24 evaluable participants at Roswell Park will be enrolled (8 participants per year over a period of 3 years).

5 LOCAL AND STUDY-WIDE RECRUITMENT METHODS

Participants will be identified/recruited/screened from patients at the Melanoma Clinic at Roswell Park, referring physicians and, from multi-disciplinary conference discussion. Patients will also be recruited through The Roswell Park Care Network sites.

Roswell Park Care Network patients will be given the option to complete an internal HIPAA Authorization form. If obtained, this authorization will allow Roswell Park study teams to access patient medical records for review of potential eligibility for clinical trials. In addition, this authorization includes an agreement to be contacted by Roswell Park. Patients who have a signed authorization on file will have records reviewed for eligibility for this trial. Roswell Park staff will work with primary oncologist at Roswell Park Care Network to approach patients and patients will be presented with study information by approved study team members. The Roswell Park Care Network sites will not be engaged in research and all research activities will be carried out at Roswell Park Main Campus.

6 MULTI-SITE RESEARCH

Not applicable: This is a single-site study.

7 STUDY TIMELINES

Subjects will be on active experimental treatment for 12 weeks, with continued PD-1/PD-L1 blockade. In the absence of disease progression, patients will continue PD-1/PD-L1 blockade as per SoC and be followed for PFS and OS within routine care.

Patients with progressive disease at 12 weeks can at the physician's discretion, begin LAG-3 or CTLA-4 ± PD-1/PD-L1 inhibitor treatment and will be followed as per standard of care for this disease group.

Patients will continue to be monitored for PFS and OS within standard care visits (every 3 months for up to 2 years).

8 STUDY ENDPOINTS

8.1 Primary Endpoint

- Objective response rate (ORR) at 12 weeks (only in participants who have completed the first 3 treatment cycles) will be evaluated using RECIST 1.1criteria.

8.2 Secondary Endpoints

- Objective response rate (ORR) at 6 months (using iRECIST criteria)(20)
- Durable objective response rate (DORR ≥ 6 months): to be calculated from the time of first confirmed response

8.3 Exploratory/ Correlative Studies

- Immune-related progression-free survival (PFS; using iRECIST criteria)
- Overall Survival (OS)

- Changes in the density of CD8+ CTLs and molecular biomarkers (OmniSeq immune report card, IRC) in paired tumor tissues collected at pre- and post-treatment (changes in the molecular markers and magnitude and character of immune infiltrate may be evaluated in paired tumor tissues collected at pre- and post-treatment, using additional methods, depending on the availability of funding).

9 DESIGN

This is a single-arm Phase II study with a 3 patient safety lead-in cohort to evaluate the clinical efficacy and immunomodulatory action of a combination treatment regimen consisting of a type-1 polarized dendritic cell (αDC1)-based therapy and tumor-selective chemokine modulation (CKM: interferon alpha-2b, rintatolimod and celecoxib) in HLA-A2+ subjects with PD-1/PD-L1-refractory advanced-stage melanoma, who will continue the original SoC (PD-1/PDL-1 blockade). The study schema is depicted in **Figure 2**.

Figure 2 Study Schema

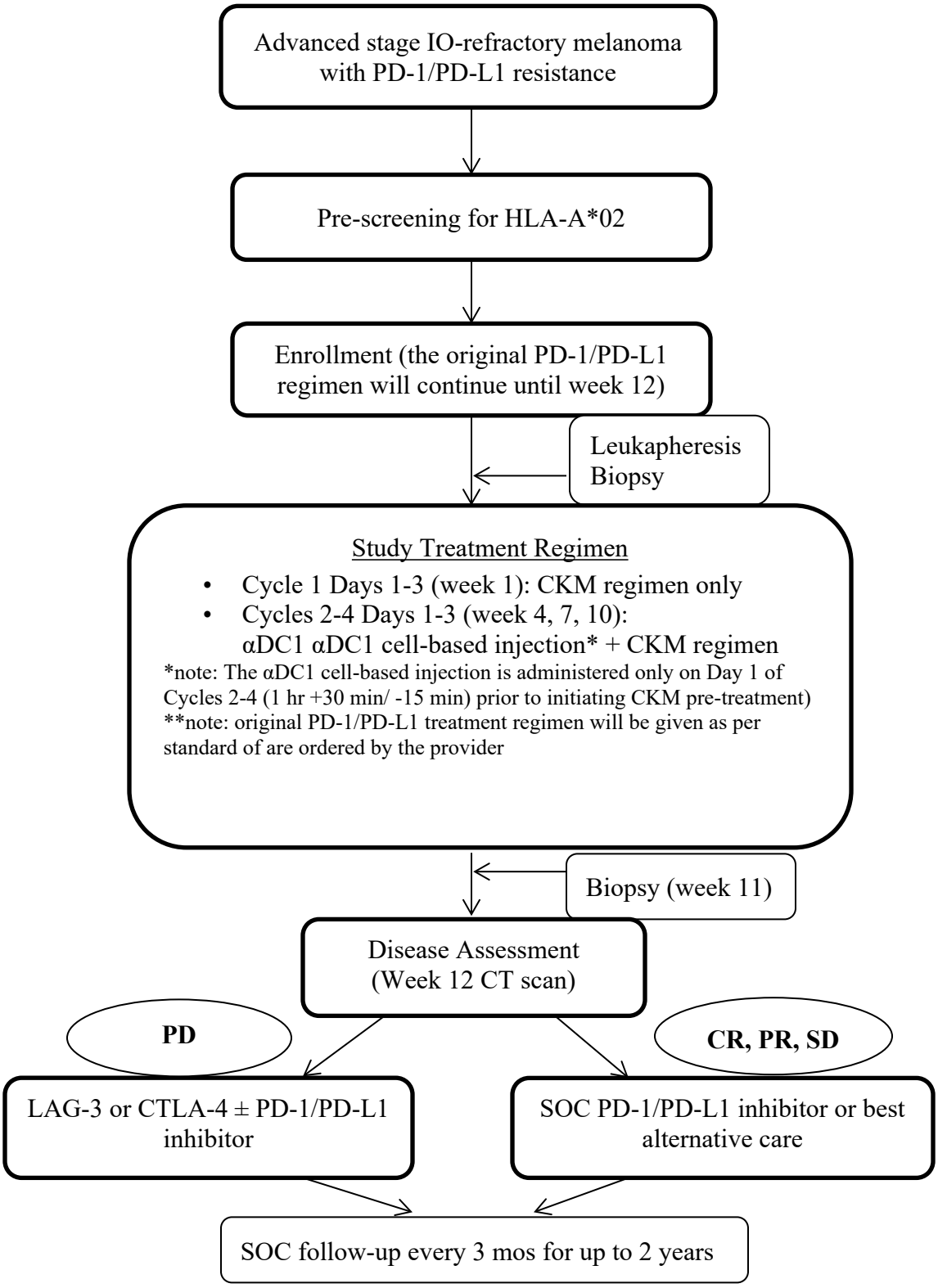
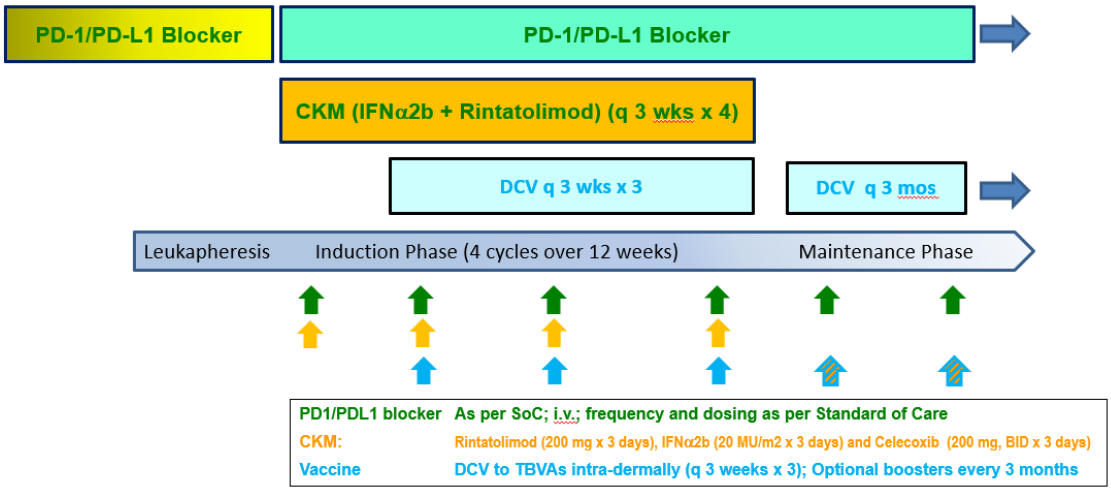


Figure 3

NCT04093323: ***αDC1 Vaccine Combined with CMK*** (Rintatolimod, IFNα2b & Celecoxib) in Patients with PD-1-Refractory HLA-A2+ Melanoma Receiving Continued PD1 Blockade



**Patients will continue the original PD-1/PD-L1 blockade as Standard of Care upon entering the study and will continue until the 12-week decision point shown in Figure 2, when a switch to a combination ICB (with CTLA4 or LAG3 blocker) can be made in case of PD.*

This is a single-arm Phase II study with a safety lead-in cohort, to evaluate the anti-tumor efficacy of combined αDC1/TBVA vaccination (Vx) + CKM in primary PD-1/PD-L1-refractory melanoma patients. DC cellular treatment will be produced by the Therapeutic Cell Production Shared Resource (TCP) at Roswell Park. OCR will be determined by PET/CT imaging after CKM cycle 4. Tumor biopsies (2 per patient), PBMC & serum (≥ 7 per patient; pre-/post-therapy) will be collected at for immunologic/molecular analysis. Follow-up treatment (post week 12) will be based on patient objective complete response (OCR) status.

At week 12, response will be evaluated by CT scan, performed as a part of standard care. Any patient with PD will be offered an opportunity to switch to PD-1/PD-L1 inhibitor combined with either CTLA4 blockade (Ipilimumab) or with LAG3 blockade, unless contra-indicated, and will be assessed as per standard of care guidelines. Any patient with a CR, PR or SD will be offered PD-1/PD-L1 blockade (according to the approved standard of care regimen) or best alternative care. Thereafter patients will be contacted by phone every 3 months for up to 2 years by medical record review/telephone call for survival status and other anti-cancer therapies.

10 TREATMENT

After CT/PET imaging confirmation of PD-1/PD-L1-resistance status, patients will be screened for HLA-A2+ status (using flow cytometry) and, for all relevant inclusion/exclusion criteria.

A patient apheresis will be performed to isolate peripheral blood monocytes to generate the autologous αDC1/TBVA cell-based treatment.

In each of 4 cycles of combination immunotherapy, enrolled subjects will receive a three-day-long course (days 1-3) of CKM (IFN alpha-2b [20 MU/m2 i. v. over 30 min], rintatolimod [200 mg i. v. over 2.5 h] and celecoxib [200 mg p.o. 30 min (can be up to 2 hours earlier) before IFN alpha-2b and 200 mg p.o. 12 hrs after IFN alpha-2b; 8-16 hours are acceptable)]). In addition, on ***day 1 only*** of cycles 2-4, subjects will receive the αDC1 cell-based treatment (10 million cells intradermal) prior to the chemokine modulating regimen.

On days of the αDC1 injection, patients will be monitored as outpatients (approximately every 20 minutes) for 1 hour after the injection Pre-treatment prior to the CKM regimen will begin 1 hr (± 10 min) following the αDC1 injection. Additionally, at approximately 1 hr (± 10 min) after the administration of the αDC1 cell-based treatment, vital signs will be obtained and assessment of the injection sites will be documented).

Treatment is intended for an outpatient setting.

Due to the need to replace INTRON-A (as originally proposed for use) with Bioferon, as we have recently done for FDA-approved IND 163681 which covers the same CKM + PD-1/PD-L1 blockade regimen but without a DC vaccine component, we propose inclusion of an additional lead-in phase I cohort of 3 patients to be treated with a reduced-dose CKM employing Bioferon at an initial dose of 10 MU/m^2. Following testing of this lower dose of IFNα2b in 3 patients, and in the absence of any dose-limiting adverse effects, we will continue the accrual patients to be treated at a Bioferon dose of 20MU/m^2. We will retain the original protocol design but will consider safety as the formal primary endpoint for these initial 3 patients to be included in the phase II efficacy evaluations.

10.1 Dosing and Administration

10.1.1 Chemokine Modulating (CKM) Regimen

The chemokine modulating regimen will be administered on an outpatient basis at Roswell Park over 3 consecutive days (days 1-3) for 4 treatment cycles (1 cycle = 21 days).

On <u>each</u> CKM dosing day, the patient will be given the following medications in the following order:

- Pre-treatment: 500 mL Normal saline IV over 60 minutes
- Pre-meds: Acetaminophen (Tylenol) 650 mg by mouth x 1 dose; Prochlorperazine (Compazine) 10 mg by mouth x 1 dose – administered 30 minutes (±5 minutes) after starting pre-treatment hydration. The CKM treatment will begin 30 minutes (±5 minutes) after that, at completion of IV normal saline
- Celecoxib: 200 mg orally, administered along with pre-meds (2 hrs to 30 min prior to interferon alpha-2b)
- Interferon Alpha-2b: (20 million units/M^2, with the exception of the initial 3 patients who will receive a (reduced) dose of 10 MU/m^2) IV over 30 minutes
- Rintatolimod: 200 mg IV, initial administration should begin at a slow rate of infusion (approximately 20 cc/ hour) and increase to 40 cc/ hour after 30 minutes (15-45 mins range is acceptable). Tubing should be flushed with 30 to 50 mL of normal saline solution upon

completion. Administration will be followed by 1 hour of observation with vital signs at 30 and 60 minutes post infusion (± 5 minutes)

- Post-CKM Regimen:
    - Celecoxib: The patient will be instructed to take the second dose of celecoxib (200 mg orally) at home, approximately 12 hours following the initial dose of celecoxib.

Pre-treatment normal saline and pre-meds listed above are recommendations and may be modified based upon institutional SOC requirements and at the discretion of the investigator.

10.1.2 αDC1/TBVA Cellular Therapy

Dendritic cells (DC) are derived from autologous adherent mononuclear cells in the peripheral blood by 5-day or 6- day culture in the presence of interleukin 4 (IL-4) and granulocyte-macrophage colony stimulating factor (GM-CSF), followed by an additional period of antigen loading and maturation. To generate the autologous DC-based cellular treatment, patients will undergo a 90 minute leukapheresis to obtain an enriched population of monocytes as a precursor source for DC developed in culture.

Leukapheresis

Outpatient leukapheresis will be performed in the Therapeutic Apheresis unit. Antecubital veins will be used, or when necessary, a central catheter will be placed. A standard collection protocol will be used to obtain the peripheral white blood cells with settings modified according to the COBE Spectra Apheresis System recommendations to obtain the best mononuclear cell yield. In order to obtain sufficient monocytes cells, leukapheresis time may vary from 90 minutes to 240 minutes. Approximately 10 L to 15 L of blood will be circulated by leukapheresis in order to retrieve approximately 500 x 10^6 blood monocytes. The cell yield from a leukapheresis procedure is variable. Autologous leukapheresis product, counts, and recoveries will be monitored for each donation. The procedure will be performed according to leukapheresis SOPs at Roswell Park. The leukapheresis product will be transported to the cGMP Therapeutic Cell Production Facility by a designated staff member of the cGMP Therapeutic Cell Production Facility. Before moving the product from the collection area, the donor center staff and the designee will confirm the following information: Patient's Name, Medical Record Number, and Date/Time of Collection. If the labeling is incomplete it must be corrected before the product leaves the collection area. Monocytes will be cultured as described below. Other leukapheresis fractions will be used in the Immune Analysis Facility to assess immunologic parameters.

The cell yield from a leukapheresis procedure is variable. However, it is expected that 1 leukapheresis will yield enough cells for the 3 intradermal αDC1/TBVA injections. If a sufficient number of cells have not been obtained, an additional leukapheresis may be required in order to complete the treatment protocol. The second leukapheresis will be performed within 14 days of the first. If a sufficient number of cells for 3 intradermal αDC1/TBVA injections and proposed QC testing are still not obtained after the second leukapheresis, the patient will be withdrawn from the study.

Preparation of Dendritic Cells

The αDC1/TBVA cells will be generated in the cGMP Therapeutic Cell Production Shared Resource of Roswell Park under the Roswell Park-held FDA-approved BB-IND-16,704 with

qualified SOPs for generation, cryopreservation, and preparation for administration of the αDC1/TBVA cell-based treatment. The cGMP Facility staff will follow SOPs, perform Quality Control (QC) and Quality Assurance (QA) procedures and manage associated documentation pertaining to products manufactured for clinical trials in this facility. Qualified personnel who are familiar with the procedures, which minimize undue exposure to them and to the environment, will undertake the preparation, handling, and safe disposal of immunotherapeutic agents in a self-contained protective environment

Intradermal αDC1 Vaccination Administration

Treatment will be administered on an outpatient basis. When the patient is ready for the administration of the intradermal αDC1 cell treatment, the required dose (in a single tuberculin syringe with attached new 27 gauge safety-glide needle and suspended in 5% albumin) will be hand-carried by a designee to the patient area. The product will be transported on ice in sealed sterile plastic bag in a small "igloo"-type cooler. Prior to releasing the αDC1 cellular product from the cGMP Therapeutic Cell Production Shared Resource, the following information will be verified: Patient Identification Number (PIN) and Medical Record Number (MR#). Prior to injection, the same information will again be verified. Each 0.2 mL of the αDC1 cell suspension will contain approximately 10 x 10^6 cells and will be loaded onto a single tuberculin syringe. The αDC1 cell suspension will be administered intradermally (i. d.) by the investigator or qualified designee. The investigator/designee will inject approximately 0.1 mL of the syringe contents intradermally and then will remove the needle from the patient. Using the same needle, the investigator/designee will then inject the remaining 0.1 mL of the syringe contents intradermally on the opposite side of the body (the other thigh). The upper thighs are the preferred locations for the injections, however upper arms are also allowed. The injections will be administered within 6 hours of thawing and release (target: 2 hrs).

Patients will be monitored (every 20 min± 5 min) as outpatients for 1 hour after αDC1 cell injection. Monitoring will consist of vital sign measurements, and monitoring of any local/systemic reactions to the injection. This monitoring will include clinical assessment of pain, tenderness, warmth and swelling. Appropriate laboratory tests will be performed based on the judgment of the clinical investigators if clinical side effects are apparent.

10.2 Dose Modifications and Treatment Delays

The following dose modification rules will be used with respect to potential toxicity. Toxicity will be assessed according to the NCI Common Terminology Criteria for Adverse Events Version 5.0 (CTCAE v5.0). The initial 3 patients will receive a reduced dose of IFNα2b (10MU/m^2), to obtain initial safety confirmation for the new source of IFNα2b. In the absence of dose-limiting adverse effects, we will proceed to use the full dose IFNα2b (20MU/M^2) in the CKM regimen. In case of dose limiting toxicities, we will suspend accrual and consult the DSMC for the development of extended safety plan and protocol revision.

In case of observation of toxicities in the initial 3 patients, in additional patients we may suspend the PD-1/PD-L1 blockade for the 12 week duration of the experimental treatment and resume it (alone or combined with either CTLA4- or LAG3 blockade) at week 12.

10.2.1 Celecoxib Hematologic and Non-Hematologic Toxicities

Considered to be possibly, probably, or definitely related to Celecoxib (Interferon Alpha-2b and rintatolimod will be continued). Celecoxib will be discontinued for any grade ≥ 3 toxicity attributed to Celecoxib.

10.2.2 Interferon alpha-2b and/or rintatolimod Hematologic and Non-hematologic toxicities

Considered to be possibly, probably, or definitely related to Interferon Alpha-2b and rintatolimod:

Interferon alpha-2b

- **Hematologic Toxicities:**

  For grade 3 toxicity:

  - If < 7 days to resolution (transfusion are allowed): Treatment to continue if toxicity is resolved to ≤ grade 1 or baseline) at a dose reduction of 33%. The dose can be re-escalated if toxicity remains at ≤ grade 1 or baseline.
  - If > 7 days to resolution: Treatment to continue if toxicity is resolved to ≤ grade 1 or baseline at the next study visit at a dose reduction of 33%. The dose cannot be re-escalated.
  - 2nd incidence: Treatment to continue if toxicity is resolved to ≤ grade 1 or baseline at the next study visit at a dose reduction of 33%. (total 66% reduction if the dose was not re-escalated). The dose cannot be re-escalated.

  For Grade 4 toxicity:

  - If < 7 days to resolution (transfusions are allowed): Treatment to continue if toxicity is resolved to ≤ grade 1 or baseline at the next study visit at a dose reduction of 33%. The dose cannot be re-escalated.
  - If > 7 days to resolution: Discontinue

- **Non-Hematologic toxicities**

  For grade 3 toxicity:

  - 1st episode: Treatment to continue if toxicity is resolved to ≤ grade 1 or baseline) at a dose reduction of 33%. The dose can be re-escalated if toxicity remains at ≤ grade 1 or baseline.
  - 2nd episode: Treatment to continue if toxicity is resolved to ≤ grade 1 or baseline at the next study visit at a dose reduction of 33%. (a total of 66% reduction if the dose was not re-escalated). The dose cannot be re-escalated.

  For grade 4 toxicity:

  - 1st episode: Treatment to continue if toxicity is resolved to ≤ grade 1 or baseline at the next study visit at a dose reduction of 33%. The dose cannot be re-escalated.
  - 2nd episode: Off study, patient will be followed until the event resolves to ≤ grade 1 or baseline study (patient may receive pembrolizumab off of the protocol at treating physician's discretion)

Rintatolimod

- **Hematologic Toxicities:**

  For grade 3 toxicity:

  - If < 7 days to resolution (transfusion are allowed): Treatment to continue if toxicity is resolved to ≤ grade 1 or baseline) at a dose reduction of 33%. The dose can be re-escalated if toxicity remains at ≤ grade 1 or baseline.
  - If > 7 days to resolution: Treatment to continue if toxicity is resolved to ≤ grade 1 or baseline at the next study visit at a dose reduction of 33%. The dose cannot be re-escalated.
  - 2nd incidence: Treatment to continue if toxicity is resolved to ≤ grade 1 or baseline at the next study visit at a dose reduction of 33%. (a total of 66% reduction if the dose was not re-escalated). The dose cannot be re-escalated.

  For Grade 4 toxicity:

  - If < 7 days to resolution (transfusions are allowed): Treatment to continue if toxicity is resolved to ≤ grade 1 or baseline at the next study visit at a dose reduction of 33%. The dose cannot be re-escalated.
  - If > 7 days to resolution: Discontinue

- **Non-Hematologic toxicities:**

  For grade 3 toxicity:

  - 1st episode: Treatment to continue if toxicity is resolved to ≤ grade 1 or baseline) at a dose reduction of 20%. The dose can be re-escalated if toxicity remains at ≤ grade 1 or baseline.
  - 2nd episode: Treatment to continue if toxicity is resolved to ≤ grade 1 or baseline) at a dose reduction of 20% (total dose reduction 20% if dose was not re-escalated). The dose can be re-escalated if toxicity remains at ≤ grade 1 or baseline.

  For grade 4 toxicity:

  - 1st episode: Treatment to continue if toxicity is resolved to ≤ grade 1 or baseline) at a dose reduction of 20% (total dose reduction 20% if dose was not re-escalated). The dose can be re-escalated if toxicity remains at ≤ grade 1 or baseline.
  - 2nd episode: Off study (patient may receive pembrolizumab off of the protocol at treating physician's discretion)

10.2.3 Toxicity Management

The toxicity of high dose interferon (20 million units/ m^2/d) has been established by Kirkwood et al in a number of trials. Most notable was the E1684 trial (21) where HDI (20 million units/m^2/d) was administered daily for 5 days x 4weeks. In that trial (n=143), grade 3 toxicities were 67%, grade 4 toxicities were 9% (mainly constitutional and neurologic), and there were 2 treatment related mortalities (grade 5) due to hepatoxicity. The proportion of Grade 3 and 4 toxicities in that trial were 48.2% for constitutional toxicities (defined as 'worst grade of any constitutional toxicity, including fever, chills, flu-like symptoms, fatigue, malaise, and diaphoresis), and 66% for non-

constitutional toxicities (23.8% for myelosuppression, 13.9% for hepatotoxicity, 28% for neurological toxicity).

These are suggestions for specific toxicities and may be modified for institutional guidelines and treating physician preference.

Gastrointestinal Toxicity:

- Nausea and/or vomiting should be controlled with adequate antiemetic therapy. Prophylactic antiemetic therapy can be used at the discretion of the investigator/sub-investigator. Subjects are encouraged to take plenty of oral fluids.
- Diarrhea should be managed with appropriate antidiarrheal therapy. Subjects should be encouraged to take plenty of oral fluids. If symptoms do not decrease to grade 1 or less with adequate antidiarrheal therapy, all protocol drugs should be held until resolved to < grade 1.

Pain/Fever/Chills

- For chills and/or fever or mild local pain, acetaminophen will be utilized at the discretion of the investigator/sub-investigator or designee.

Hypersensitivity Reactions

Caution: Subjects who had a mild to moderate hypersensitivity reaction have been successfully re-challenged, but careful attention to prophylaxis and bedside monitoring of vital signs is recommended. Hypersensitivity reactions to Interferon Alpha-2b and/or rintatolimod will be managed as follows:

- Mild symptoms (e.g., mild flushing, rash, pruritus): Complete infusion. Supervise at bedside. No treatment required.
- Moderate symptoms (e.g., moderate rash, flushing, mild dyspnea, chest discomfort): Stop infusion. Give intravenous diphenhydramine 50 mg and intravenous famotidine 20 mg. Resume infusion after recovery of symptoms at a 66% slower rate, then, if no further symptoms, rate can be titrated up every 5-10 min to initial rate until infusion is complete. If symptoms recur, at lowest infusion rate stop the infusion and no further Interferon Alpha-2b administered on that day. If symptoms recur after the rate has been increased, stop the infusion, treat reaction as per institutional guidelines, consider dexamethasone 20 mg IV and restart at 66% slower rate (over 1 hour) without up-titration. Future doses pre-treatment modifications and rate modifications after discussion with the investigator. Record toxicity.
- Severe life-threatening symptoms (e.g., hypotension requiring pressor therapy, angioedema, respiratory distress requiring bronchodilation therapy, generalized urticaria): Stop infusion. Treat as per institutional guidelines. Subject should be removed from further protocol therapy. Report as serious adverse event.
- In case of observation of toxicities which will require removal of any of the initial 3 patients, in additional patients we will suspend the PD-1/PD-L1 blockade of the 12 week duration of the experimental treatment and resume it (alone or combined with either CTLA4- or LAG3 blockade) at week 12.

11 PROCEDURES INVOLVED

The study-specific assessments are outlined in Appendix I (Schedule of Procedures and Observations). Unless otherwise stated on the study calendar, Baseline and/or Screening assessments must be performed within **28** days prior to the first cycle of the treatment regimen. Any results falling outside of the reference ranges may be repeated at the discretion of the investigator.

Unless otherwise defined in the written protocol text, all procedures/assessments will be conducted in accordance with Roswell Park Clinical Trials Office Standard Operating Procedures.

Eligibility of each participant will be established prior to start of treatment.

Informed consent ***MUST*** be completed prior to receiving any study related procedures.

Pre-screening

- Buccal swab (or peripheral blood) for HLA-A2 testing.

HLA-A2 positivity is required for study eligibility. Samples for HLA-A2 typing for eligibility will performed by a CLIA certified laboratory, as selected by site.

Note: Participant must sign a separate informed consent prior to HLA testing.

Other than the HLA testing, the rest of the screening tests (as indicated in Appendix I) will be carried out after confirmation of HLA eligibility and informed consent and, are to be completed within 28 days prior to Cycle 1-Day 1 of the CKM regimen. Any results falling outside of the reference ranges may be repeated at the discretion of the investigator.

11.1 Post-Treatment Follow-Up Evaluations

To capture all possible delayed immune-related adverse events, follow-up safety evaluations will occur at least monthly for 90 days after last dose of study drug or until resolution of any drug-related toxicity (telephone contact is acceptable).

- Concomitant medications: List any ongoing medications with dose changes, as applicable.
- Adverse events
- Survival status

11.2 Long-Term Follow-Up Evaluations

After 90-day post treatment follow-up visit, participants will be followed within the scope of standard of care to assess survival status (e.g., every 3 months or according to institutional guidelines if patient starts on an alternative therapy). Participants who are unavailable for follow-up evaluations should be classified as lost to follow-up for 1 of the following reasons:

- Lost to follow-up: For a participant to be considered lost to follow-up, the investigator must make two separate attempts to re-establish contact with the participant. The attempts to re-establish participant contact must be documented (e.g., certified letter).
- Death: Date and cause of death will be recorded for those participants who die within 30 days after last dose of study drug (telephone verification is acceptable).

11.3 Correlative Studies

The correlative studies will involve testing to be performed by the Kokolus lab (within their scope), depending on the available funding.

11.3.1 Fresh Biopsy Specimens

A fresh biopsy is required at the following time points:

- Baseline (prior to Cycle 1)
- Week 11

At each time point, we will obtain 4 core biopsies of the tumor and 1 core biopsy of the surrounding non-tumor tissue. Samples are to be labeled with the study-specific subject ID number, the clinical study number, the protocol time-point and protocol day. One (1) core of tumor tissue for research will be placed in formalin and sent to Clinical Research Laboratory Services for FFPE processing. Dr. Kokolus's lab will be notified when a block is available for pick-up. Blocks will be stored in Dr. Kokolus's lab for future analysis. The remaining 3 cores of the tumor tissues and the core of non-tumor tissue will be placed in cold Dulbecco's Phosphate-Buffered saline and kept on wet ice for delivery to the Kokolus lab within 2 hours of collection (a maximum of within 4 hours of collection is allowed). Samples will then be cryopreserved and stored in Dr. Kokolus's lab until analysis.

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Note: All investigator or analyzing research laboratories housing research samples need to maintain current **Temperature Logs** and study-specific **Sample Tracking and Shipping Logs**. The Principal Investigator/Laboratory Manager **must** ensure that the stated lab(s) have a process in place to document the receipt/processing/storage/shipping of study-related samples/specimens. This is required for both observational and interventional clinical studies collecting clinical samples.

11.3.2 Blood Samples for Correlative Analysis

Blood samples will be collected via venipuncture for immune biomarker analysis. Samples will be collected using 6, 10 mL green-top heparinized tubes.

Samples for correlative analysis will be obtained on:

- Day 1 and Day 3 of Cycles 1, 2, 3 and 4
    - Pre-infusion and,
    - 1 hr (± 10 minutes) post-rintatolimod infusion
- End of Treatment (± 3 days)
- Week 12 (± 1 week)

Tubes will be labeled with the participant's MR number, participant's initials, participant's study number, clinical study number, time of collection and protocol day. Samples will be sent at room temperature to the attention of Clinical Research Laboratory Services (pneumatic Station 19) where they will get accessioned for tracking. Once specimen receipt has been documented, the specimens will be sent at ambient temperature to pneumatic station 641 (located in CCC LOB-4th floor). The Kokolus Laboratory will be notified via telephone *and* e-mail (all contacts to be copied with each e-mail: see contact information below) prior to sample shipment and, the samples will be held in Clinical Research Laboratory Services until a response is received acknowledging that personnel are available to procure the samples from the pneumatic tube station. Samples will be processed and stored in the Kokolus Laboratory for future analysis.

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12 WITHDRAWAL OF SUBJECTS

Upon treatment discontinuation all end of treatment evaluations and tests will be conducted. All participants who discontinue due to an AE must be followed until the event resolves or stabilizes. Appropriate medical care should be provided until signs and symptoms have abated, stabilized, or until abnormal laboratory findings have returned to acceptable or pre-study limits. The final status of the AE will be reported in the participant's medical records and the appropriate eCRF.

Reasons for treatment discontinuation should be classified as follows:

- Death
- Progressive disease
- Toxicity; treatment related or unrelated
- Investigator judgment
    - The Investigator may discontinue a participant if, in his/her judgment, it is in the best interest of the participant to do so.
- Noncompliance
- Participant voluntary withdrawal
    - A participant may withdraw from the study at any time, for any reason. If a participant discontinues treatment, an attempt should be made to obtain information regarding the reason for withdrawal.

12.1 Replacement

- Participants will be withdrawn from the study and replaced if 1) they do not meet the remaining eligibility requirements after having been tested as HLA-A2 positive, 2) if after a second leukapheresis (if required), a sufficient number of cells for 3 intradermal αDC1 cell-based injections and proposed QC testing are not obtained and if 3), the patient withdraws prior to Cycle 1-Day 1, there is evidence of disease progression from time of enrollment until prior to Cycle 1-Day 1 or, physician's decision to withdraw the patient prior to the start of the treatment regimen.
- A participant will be deemed **unevaluable** for the primary analysis (i.e., response evaluation at Week 12) if they have not completed 3 treatment cycles (Cycle 1: CKM only and Cycles 2-3: αDC1 cell-based treatment +CKM). One missed cycle is allowed.

13 RISKS TO SUBJECTS

Potential risks associated with patient treatment include:

- *DC-based therapy* (development of autoimmune diseases such as vitiligo, development of allergic reaction to the cellular therapy)
- *Leukapheresis* (anemia, numbness, chills, abdominal cramps, nausea, vomiting, local pain at site of venipuncture, and allergic reaction to topical anesthetic)
- *IFN alpha-2b* (fever, chills, flu-like symptoms, loss of appetite, nausea, vomiting, diarrhea, abdominal pain, fatigue, lowered white blood count, lowered platelets with increased bruising/bleeding, hair loss, drowsiness; behavioral alterations, numbness and/or tingling in the hands and/or feet, skin rashes, inflammation of the pancreas)
- *Celecoxib* (dyspepsia, headaches, elevated liver function tests)
- *Rintatolimod* (mild flu-like symptoms, transient headache, fever, myalgia, arthralgia, injection site reaction, pruritus, vasodilation, diarrhea, and fatigue/malaise)
- *Venipuncture* (mild pain, discomfort, bruising at the needle insertion site, inflammation of the vein, possible infection and bleeding)

- *Tumor Biopsy* (pain, discomfort, and/or soreness at the site where biopsy was done. The soreness is generally diminished within 48 hours of the procedure)

14 POTENTIAL BENEFITS TO SUBJECTS

This Phase II trial is designed to determine potential efficacy of the αDC1/TBVA cell-based treatment combined with the CKM (rintatolimod, IFN alpha-2b and celecoxib) regimen. The CKM regimen has proven to be safe and well-tolerated by colorectal cancer patients in ongoing trials.

The CKM regimen is anticipated to facilitate the direction of anti-TBVA/anti-tumor T cells into melanoma tissue, potentially leading to the induction of tumor regression and prolonged patient survival. The full potential benefit to the subjects however is not known.

This study will allow us to gain a greater understanding of the anti-tumor immune responses in the TME and may demonstrate advantage of potential treatments with αDC1/TBVA cell-based treatment and CKM in patients receiving concurrent anti-PD1 or anti-PD-L1. Although, based on our previous preclinical and clinical experience, we anticipate that αDC1/TBVA cell-based treatment plus CKM may be partially effective by themselves, the inclusion of continued immune checkpoint blockade in combination with αDC/CKM -based treatment protocol is expected to result in optimal improve anti-tumor efficacy.

15 DATA AND SPECIMEN BANKING

All samples for correlative analysis will be sent to Dr. Kokolus's Laboratory for processing (CCC-410-416). Samples will be used for planned study assays as well as for future analysis for other yet to be identified biomarkers that may be related to the clinical outcome of the study population. Any clinical data that is associated with the samples, will be stored on a secure server in the Department of Medicine, will be accessible only by the PI, Co-Investigators and PI designated data manager and, will be password protected. All computer entry and networking programs will be done using PIDs only. Any clinical data and/or specimens for future research will require verification of an IRB-approved protocol and will be de-identified before being released.

Note: All investigator or analyzing research laboratories housing research samples need to maintain current Temperature Logs and study-specific Sample Tracking and Shipping Logs. The Principal Investigator/Laboratory Manager must ensure that the stated lab(s) have a process in place to document the receipt/processing/storage/shipping of study-related samples/specimens. This is required for both observational and interventional clinical studies collecting clinical samples.

16 MEASUREMENT OF EFFECT

Objective response rate and progression will be evaluated at Week 12 (primary objective) using the revised Response Evaluation Criteria in Solid Tumors (RECIST) guideline (version 1.1) (22). Please refer to Appendix F for a summary of tumor response assessment according to RECIST 1.1 criteria.

Response and progression following Week 12 will be evaluated in this study using the immune-related response criteria (iRECIST): Please refer to Appendix G.

For the purposes of this study, patients should be re-evaluated for recurrence or progression of disease using CT scan as the primary modality. Other imaging modalities are allowed based on clinical circumstances (e.g. CT contrast allergy).

In addition to the scans during the treatment phase of the protocol, CT scans should also be obtained during the follow up period in accordance with standard of care, but no later than every 8-12 weeks following initial documentation of objective response.

Note: The following section on response and stable disease duration is part of the original submission (supplementary appendix) by Seymour et al, 2017(20) and has been peer reviewed.

16.1 Response and Stable Disease Duration (RECIST 1.1 and iRECIST)

Response duration will be measured from the time measurement criteria for CR/PR or iCR/iPR (whichever is first recorded) are first met until the first date that recurrent or progressive disease is objectively documented, taking as reference the smallest measurements recorded on study (including baseline).

Stable disease duration will be measured from the time of start of treatment until the criteria for progression are met, taking as reference the smallest sum on study (including baseline).

16.2 Methods of Measurement

The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up. Assessments should be identified on a calendar schedule and should not be affected by delays in therapy. While on study, all lesions recorded at baseline should have their actual measurements recorded at each subsequent evaluation, even when very small (e.g. 2 mm). If it is the opinion of the radiologist that the lesion has likely disappeared, the measurement should be recorded as 0 mm. If the lesion is believed to be present and is faintly seen but too small to measure, a default value of 5 mm should be assigned. For lesions which fragment/split add together the longest diameters of the fragmented portions; for lesions which coalesce, measure the maximal longest diameter for the "merged lesion".

16.2.1 Clinical Lesions

Clinical lesions will only be considered measurable when they are superficial and ≥10 mm as assessed using calipers (e.g. skin nodules). For the case of skin lesions, documentation by color photography including a ruler to estimate the size of the lesion is recommended. If feasible, imaging is preferred.

16.2.2 Chest X-ray

Chest CT is preferred over chest X-ray, particularly when progression is an important endpoint, since CT is more sensitive than X-ray, particularly in identifying new lesions. However, lesions ≥20 mm on chest X-ray may be considered measurable if they are clearly defined and surrounded by aerated lung.

16.2.3 CT, MRI

CT is the best currently available and reproducible method to measure lesions selected for response assessment. This guideline has defined measurability of lesions on CT scan based on the assumption that CT slice thickness is 5 mm or less. When CT scans have slice thickness greater

than 5 mm, the minimum size for a measurable lesion should be twice the slice thickness. MRI is also acceptable in certain situations (e.g. for body scans). Other specialized imaging or other techniques may also be appropriate for individual case.4 For example, while PET scans are not considered adequate to measure lesions, PET-CT scans may be used providing that the measures are obtained from the CT scan and the CT scan is of identical diagnostic quality to a diagnostic CT (with IV and oral contrast).

16.2.4 Ultrasound

Ultrasound is not useful in assessment of lesion size and should not be used as a method of measurement. If new lesions are identified by ultrasound in the course of the study, confirmation by CT is advised.

16.2.5 Endoscopy, Laparoscopy

The utilization of these techniques for objective tumour evaluation is not advised. However, they can be useful to confirm complete pathological response when biopsies are obtained or to determine relapse in trials where recurrence following complete response or surgical resection is an endpoint.

16.2.6 Tumor Markers

Tumour markers alone cannot be used to assess objective tumour response. If markers are initially above the upper normal limit, however, they must normalize for a patient to be considered in complete response.

16.2.7 Cytology, Histology

These techniques can be used to differentiate between PR and CR in rare cases if required by protocol (for example, residual lesions in tumour types such as germ cell tumors, where known residual benign tumors can remain). When effusions are known to be a potential adverse effect of treatment (e.g., with certain taxane compounds or angiogenesis inhibitors), the cytological confirmation of the neoplastic origin of any effusion that appears or worsens during treatment when the measurable tumour has met criteria for response or stable disease is advised to differentiate between response or stable disease and progressive disease.

17 SAFETY EVALUATION

17.1 Adverse Events

An adverse event or adverse experience (AE) is any untoward medical occurrence associated with the use of a drug in humans, whether or not considered drug related. Therefore, an AE can be ANY unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not considered related to the medicinal (investigational) product (attribution of ‘unrelated’, ‘unlikely’, ‘possible’, ‘probable’, or ‘definite’).

An AE is considered “unexpected” if it is not listed in the investigator brochure or is not listed at the specificity or severity that has been observed; or if an investigator brochure is not required or available, is not consistent with the risk information described in the general investigational plan in other study-related documents.

17.2 Diagnosis Versus Signs and Symptoms

If known, a diagnosis should be recorded on the CRF rather than individual signs and symptoms (e.g., record only liver failure or hepatitis rather than jaundice, asterixis, and elevated transaminases). However, if a constellation of signs and/or symptoms cannot be clinically characterized as a single diagnosis or syndrome at the time of reporting, each individual event should be recorded as an AE or SAE on the CRF. If a diagnosis is subsequently established, it should be reported as follow-up information.

17.3 Adverse Events Occurring Secondary to Other Events

In general, AEs occurring secondary to other events (e.g., cascade events or clinical sequelae) should be identified by their primary cause. For example, if severe diarrhea is known to have resulted in dehydration, it is sufficient to record only diarrhea as an AE or SAE on the CRF.

However, clinically significant AEs occurring secondary to an initiating event that are separated in time should be recorded as independent events on the CRF. For example, if a severe gastrointestinal hemorrhage leads to renal failure, both events should be recorded separately on the CRF.

17.4 Abnormal Laboratory Values

Only clinically significant laboratory abnormalities that require active management will be recorded as AEs or SAEs on the CRF (e.g., abnormalities that require study drug dose modification, discontinuation of study treatment, more frequent follow-up assessments, further diagnostic investigation, etc.).

If the clinically significant laboratory abnormality is a sign of a disease or syndrome (e.g., alkaline phosphatase and bilirubin 5 x the upper limit of normal associated with cholecystitis), only the diagnosis (e.g., cholecystitis) needs to be recorded on the Adverse Event CRF.

If the clinically significant laboratory abnormality is not a sign of a disease or syndrome, the abnormality itself should be recorded as an AE or SAE on the CRF. If the laboratory abnormality can be characterized by a precise clinical term, the clinical term should be recorded as the AE or SAE. For example, an elevated blood potassium level of 7 mEq/L should be recorded as “hyperkalemia”.

Observations of the same clinically significant laboratory abnormality from visit to visit should not be repeatedly recorded as AEs or SAEs on the CRF, unless their severity, seriousness, or etiology changes.

17.5 Preexisting Medical Conditions (Baseline Conditions)

A preexisting medical condition should be recorded as an AE or SAE only if the frequency, severity, or character of the condition worsens during the study. When recording such events on an Adverse Event CRF, it is important to convey the concept that the preexisting condition has changed by including applicable descriptors (e.g., “more frequent headaches”).

17.6 Grading and Reporting Adverse Events

Grading and Relationship to Drug

The descriptions and grading scales found in the CTEP Version 5 of the NCI Common Terminology Criteria for Adverse Events (CTCAE) will be utilized for AE reporting. CTEP Version 5.0 of the CTCAE is identified and located at:

http:// ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm#ctc_50.

AEs not covered by specific terminology listed should be reported with common medical terminology, and documented according to the grading scales provided in the CTCAE Version 5. The relationship of event to study drug will be documented by the Investigator as follows:

Unrelated: The event is clearly related to other factors such as the participant's clinical state, other therapeutic interventions or concomitant drugs administered to the participant.

Unlikely: The event is doubtfully related to investigational agent(s). The event was most likely related to other factors such as the participant's clinical state, other therapeutic interventions, or concomitant drugs.

Possible: The event follows a reasonable temporal sequence from the time of drug administration but could have been produced by other factors such as the participant's clinical state, other therapeutic interventions or concomitant drugs.

Probable: The event follows a reasonable temporal sequence from the time of drug administration and follows a known response pattern to the study drug. The event cannot be reasonably explained by other factors such as the participant's clinical state, therapeutic interventions or concomitant drugs.

Definite: The event follows a reasonable temporal sequence from the time of drug administration, follows a known response pattern to the study drug, cannot be reasonably explained by other factors such as the participant's condition, therapeutic interventions or concomitant drugs; AND occurs immediately following study drug administration, improves upon stopping the drug, or reappears on re-exposure.

Reporting Adverse Events

Routine AEs occurring between the starting date of the intervention until 30 days after the last intervention, or until the event has resolved, the study participant is lost to follow-up, the start of a new treatment, or until the study investigator assesses the event(s) as stable or irreversible, will be reported. New information will be reported after it is received.

Guidelines for Routine Adverse Event Reporting for Phase II Studies (Regardless of Expectedness)

Attribution	Grade 1	Grade 2	Grade 3	Grade 4
Unrelated			X	X
Unlikely			X	X
Possible	X	X	X	X
Probable	X	X	X	X
Definite	X	X	X	X

17.7 Serious Adverse Events

A serious adverse event (SAE) is any adverse event (experience) that in the opinion of either the investigator or sponsor results in **ANY** of the following:

- Death.
- A life-threatening adverse event (experience). Any AE that places a participant or participants, in the view of the Investigator or sponsor, at immediate risk of death from the reaction as it occurred. It does NOT include an AE that, had it occurred in a more severe form, might have caused death.
- Inpatient hospitalization or prolongation of existing hospitalization (for > 24 hours).
- A persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions.
- A congenital anomaly or birth defect.
- Important Medical Event (IME) that, based upon medical judgment, may jeopardize the participant and may require medical or surgical intervention to prevent one of the outcomes listed above.

Reporting Serious Adverse Events

All new SAEs occurring from the date the participant signs the study consent until 30 days after the last intervention or a new treatment is started, whichever comes first, will be reported. The Roswell Park SAE Source Form is to be completed with all available information, including a brief narrative describing the SAE and any other relevant information.

SAEs occurring after the 30 day follow-up period that the investigator determines to be possibly, probably or definitely related to the study intervention should be reported.

SAEs that are unexpected and possibly, probably or definitely related must be reported as an Unanticipated Problem. Please refer to **Section 17.10** for details on reporting Unanticipated Problems.

17.8 Follow-Up for Serious Adverse Events

All related SAEs should be followed to their resolution, until the study participant is lost to follow-up, the start of a new treatment, or until the study investigator assesses the event(s) as stable or irreversible. New information will be reported when it is received.

17.9 Unanticipated Problems

An Unanticipated Problem (UP) is any incident, experience, or outcome that meets all of the following criteria:

- Unexpected (in terms of nature, severity, or frequency) given:
  - The research procedures that are described in the study-related documents, including study deviations, as well as issues related to compromise of participant privacy or confidentiality of data.
  - The characteristics of the participant population being studied.
- Related or possibly related to participation in the research (possibly related means there is a reasonable possibility that the incident, experience, or outcome may have been caused by the procedures involved in the research).
- Suggests that the research places participants or others at a greater risk of harm (including physical, psychological, economic, or social harm) than was previously known or recognized and if in relation to an AE is also deemed Serious per Section 17.7.

Reporting Unanticipated Problems

The Reportable New Information (RNI) Form will be submitted to the CTO Quality Assurance (QA) Office within 1 business day of becoming aware of the Unanticipated Problem. After review, CTO QA Office will submit the RNI to the IRB.

When becoming aware of new information about an Unanticipated Problem, submit the updated information to CTO QA Office with an updated Reportable New Information Form. The site Investigator or designated research personnel will report all unanticipated problems to the IRB in accordance with their local institutional guidelines.

17.10 FDA Reporting

When Roswell Park is the IND holder the following describes the FDA reporting requirements by timeline for AEs and new safety findings that meet the criteria outlined below:

Within 7 Calendar Days

Any adverse event that meets **ALL** the following criteria:

- Related or possibly related to the use of the study drug;
- Unexpected; and
- Fatal or life-threatening.

Within 15 Calendar Days

Any adverse event that meets **ALL** the following criteria:

- Related or possibly related to the use of the study drug;
- Unexpected; and
- Serious but not fatal or life-threatening;

Or, meets **ANY** of the following criteria:

- A previous adverse event that is not initially deemed reportable but is later found to fit the criteria for reporting (report within 15 days from when event was deemed reportable).
- Any findings from other studies, including epidemiological studies, pooled analysis of multiple studies, or other clinical studies conducted with the study drug that suggest a significant risk in humans exposed to the drug.
- Any findings from animal or in vitro testing that suggest a significant risk for human participants including reports of mutagenicity, teratogenicity, or carcinogenicity or reports of significant organ toxicity at or near the expected human exposure.
- Any clinically important increase in the rate of occurrence of a serious, related or possibly related adverse event over that listed in the protocol or investigator brochure.

Sponsors are also required to identify in IND safety reports, all previous reports concerning similar adverse events and to analyze the significance of the current event in the light of the previous reports.

Reporting Process

The principal investigator or designee will complete and submit a FDA Form 3500A MedWatch for any event that meets the above criteria. Forms will be submitted to the CTO QA Office via email to CTO-QA@RoswellPark.org.

18 DATA MANAGEMENT AND CONFIDENTIALITY

18.1 Data Collection

Full build studies are managed by Roswell Park CTO Data Management for analysis by Roswell Park Biostatisticians. All electronic case report form (eCRF) data are captured for these studies.

Data management activities are performed using a CTMS system that enables the collection, cleaning and viewing of clinical trial data. CTO data management designs the study-specific database and facilitates development by the Information Technology team. Once the database design is approved by the Investigator, Statistician, and Clinical Research Coordinator, the database is put into production and data entry can begin. Data can be entered and changed only by those with the rights to do so into the eCRFs.

18.2 Maintenance of Study Document

Essential documents will be retained per Roswell Park's policy for 6 years from the study termination date. These documents could be retained for a longer period, however, if required by the applicable local regulatory requirements or by an agreement with Roswell Park.

18.3 Revisions to the Protocol

Roswell Park may make such changes to the protocol as it deems necessary for safety reasons or as may be required by the U.S. FDA or other regulatory agencies. Revisions will be submitted to the IRB/ERC for written approval before implementation.

18.4 Termination of the Study

Roswell Park may terminate the study at any time upon immediate notice if it believes termination is necessary for the safety of participants enrolled in the study.

19 STATISTICAL PLAN

19.1 Demographic and Baseline Characteristics

Descriptive statistics (as appropriate: n, percent, mean, median, min and max) will be used to summarize demographic and baseline characteristics.

19.2 Study Duration and Compliance

All study drug administration and compliance data will be summarized.

19.3 Prior and Concomitant Medication

All relevant prior medication and all concomitant medications will be summarized by frequencies and percentages.

19.4 Safety evaluations.

For the initial 3 patients, safety will constitute the formal primary endpoint. Overall safety profile will be characterized by type, frequency, severity (according to CTCAE version 5.0), timing, seriousness, and relationship to study treatment. Please see detailed description in points 19.10 and 19.11 below. The initial 3 patients will receive a reduced dose of IFNα2b (10MU/mM^2), to obtain initial safety confirmation for the new source of IFNα2b. In the absence of dose-limiting adverse effects, we will proceed to use the full dose of IFNα2b (20MU/M^2) in the CKM regimen. In case of dose limiting toxicities, we will suspend accrual and consult the IRBN and FDA for the development of extended safety plan and protocol revision.

19.5 Efficacy Analyses

This trial is designed as a single arm phase II trial of a type-1 polarized dendritic cell (αDC1)-based treatment in combination with tumor-selective chemokine modulation (CKM: Interferon-a2b, Rintatolimod and Celecoxib) in melanoma patients with primary PD-1/PD-L1 resistance. The primary endpoint is ORR at 12 weeks. The maximum sample size will be n=24.

The primary test of efficacy will be carried out by an exact binomial test of a proportion within a Simon two-stage design. The study design stops early for futility only. We will test that the null ORR = 0.20 versus the alternative ORR > 0.20, which leads to the following decision rule:

Stage 1: If 2 or less of the first n1 = 14 evaluable subjects have an OR, it will be concluded that the therapy and the study will end due to futility. Otherwise, the study will continue to the second stage.

Stage 2: We will accrue 10 additional subjects. If 8 or more of the total of n1 + n2 = 24 evaluable subjects have an OR, it will be concluded that the therapy is promising; otherwise, it will be concluded that the therapy is not worthy of further study.

19.6 Sample Size Determination

The nominal significance level of this design is alpha=0.10. The sample size calculation is based on testing the hypotheses concerning the proportion of the population with a response to the treatment. This two-stage design requires a maximum of n=24 in order to achieve approximately 1-beta=0.80 power to detect differences of 0.20 percentage points greater than the null hypothesized value of a 0.20 ORR at six months.

19.7 Secondary Analyses

We will evaluate the immune-related objective response rate (ORR; per iRECIST;) in patients that continue on with either PD-1/PD-L1 and/or CTLA4 blockade after meeting the 12-week primary objective treated with autologous αDC1/TBVA cellular treatment plus cytokine CKM regimen followed by re-treatment with PD-1/PD-L1 blockade (± CTLA4 blockade). In addition, we will evaluate rate of durable responses (> 6 months) on the combination treatment in the above patient population treated with autologous αDC1/TBVA cellular treatment plus cytokine CKM regimen followed by re-treatment with PD-1/PD-L1 blockade (± CTLA4 blockade). The analysis will be primarily descriptive and consist of sample proportions and the corresponding 95% confidence intervals.

19.8 Exploratory Analyses

Exploratory endpoints are to examine whether the combination of peptide-loaded autologous αDC1 cellular therapy and tumor-selective chemokine modulation with IFN-a2b, rintatolimod, and celecoxib improves the OS and irPFS in HLA-A2+ subjects PD-1/PD-L1-refractory melanoma compared to the historical control of the best supportive care and to identify the intratumoral and systemic immune correlates of the response to treatment. Time-to-event endpoints will be analyzed by a Cox regression model as a function of various biomarker combinations.

19.9 Randomization

This is a non-randomized study.

19.10 Safety and Toxicity Analyses

Adverse events monitoring and clinical findings including physical examinations, vital signs, laboratory test results, concomitant medications, and withdrawals/terminations will be used to assess safety.

19.11 Adverse Events and Serious Adverse Events

AEs will be categorized by SOC and Preferred Terms as per CTCAE version 5.0. The incidence of AEs as well as the severity and relationship to study drug will be presented and the incidence of AEs leading to withdrawal from the study and serious AEs (SAEs) will be summarized by frequency and percentages.

As per NCI CTCAE Version 5.0, the term toxicity is defined as adverse events that are classified as either possibly, probably, or definitely related to study treatment. The maximum grade for each type of toxicity will be recorded for each patient and will be used for reporting. Frequency tables will be reviewed to determine toxicity patterns. In addition, all adverse event data graded as 3, 4,

or 5 and classified as either “unrelated or unlikely to be related” to study treatment in the event of an actual relationship developing will be reviewed.

Serious adverse events will be similarly summarized. Clinical findings will include evaluation of physical examinations, vital signs and laboratory test results, concomitant medications, and withdrawals/terminations.

19.12 Procedure for Accounting for Missing, Unused and Spurious Data

Missing data will be indicated in the listings, but excluded from all descriptive analyses. All data will be listed, including otherwise unused data. Spurious data will be identified as such, wherever possible.

20 PROVISIONS TO MONITOR THE DATA TO ENSURE THE SAFETY OF SUBJECTS

Patients will be monitored at intervals as stipulated in the Study Calendar and/or dictated by standard care. Appropriate care will be provided to any patients experiencing difficulty while being treated, and patients are encouraged in the consent forms to report adverse experiences to the investigator/sub-investigator. This information is collected per the protocol, and treatment is administered as needed. Patients will be advised regarding the importance of reporting any side effects to their investigator/sub-investigator or the study coordinator so that the appropriate medical treatment can be administered to prevent life threatening or fatal adverse events. Study treatment will be immediately discontinued for any grade IV toxicity that becomes apparent. Treatment will be held for any grade III toxicity pending reversal of such toxicity. In the event of any adverse effect, appropriate medical treatment will be instituted and study treatment will be discontinued if the above toxicity remains. The results of all study tests will be discussed with the patient and this information will be protected under University, Hospital, and federal HIPAA policies. Medical records will be considered confidential and are stored in a locked cabinet in the administrative office of Dr. Igor Puzanov. The only persons with access to these records will be the clinical principal investigator and his medical team, representatives of the NCI, the U.S. Food and Drug Administration (FDA), the National Institutes of Health (NIH), and the Roswell Park Cancer Institute Protocol Review and Monitoring Committee and Institutional Review Board (IRB). Patients will not be identified in any report or publication of this study or of its results without prior written permission. A comprehensive Data Safety Monitoring Plan (DSMP) is available for all patients involved in clinical research studies at Roswell Park. If patients are injured due to their participation on trial, immediate medical care will be provided by the clinical PI and resources of Roswell Park as outlined in the informed consent document.

The Roswell Park Data Safety Monitoring Committee will assess the progress of the study, the safety data, and critical efficacy endpoints (Phase I studies will be reviewed quarterly Phase II, III and pilot investigator-initiated studies will be reviewed semi-annually). The DSMC will review the study and will make recommendations that include but not limited to; (a) continuation of the study, (b) modifications to the design, (c) suspension of, or (d) or termination of the study.

21 VULNERABLE POPULATIONS

Not Applicable.

22 COMMUNITY-BASED PARTICIPATORY RESEARCH

Not Applicable.

23 SHARING OF RESULTS WITH SUBJECTS

Individual response data is shared with the participant as a part of their clinical care.

24 SETTING

Participants will be identified/recruited/screened from patients at the Melanoma Clinic at Roswell Park, referring physicians and, from multi-disciplinary conference discussion.

Potential subjects will be identified by the examining physician and referred for evaluation by the principal investigator or co-investigators. Informed consent will be obtained on all subjects by the principal investigator or co-investigators or other designee prior to all screening procedures. No patient will be entered into the clinical trial without having a signed written consent form. The Clinical Research Coordinator will screen the subjects further, as per study PI orders, to determine if they would meet the inclusion/exclusion criteria.

25 PROVISIONS TO PROTECT THE PRIVACY INTERESTS OF SUBJECTS

Any data, specimens, forms, reports, video recordings, and other records that leave the site will be identified only by a participant identification number (Participant ID, PID) to maintain confidentiality. All records will be kept in a limited access environment. All computer entry and networking programs will be done using PIDs only. Information will not be released without written authorization of the participant.

26 RESOURCES AVAILABLE

Production of αDC1/TBVA cells for the trial will take place at the cGMP cell production facility (Therapeutic Cell Production Facility; TCPF) of the Center for Immunotherapy (CFI) at Roswell Park.

This study will be performed under the IND held by Roswell Park, modified active BB-IND 16,704, covering the combination of αDC1/TBVA cell-based treatment (αDC1s loaded with tumor blood vessel-related antigens) and systemic CKM regimen (i. v. rintatolimod and IFN alpha-2b, combined with oral celecoxib). This IND was transferred to Roswell Park in mid-December 2017.

The transfer of the IND and the development of the clinical MTA and non-commercial use license for the use of Dr. Storkus' (University of Pittsburgh) peptides at Roswell Park (finalized in January 2018), allows us to extend the scope of the BB-IND 16,704 to include DC production for melanoma patients at Roswell Park.

Since Dr. Storkus holds a related active BB-IND 15,224, which covers the same αDC1/TBVA cell-based treatment, but combined with dasatinib (melanoma trial NCT01876212/UPCI 12-048), we also needed a clinical data-sharing agreement relevant to both trials, to assure appropriate reporting of safety and efficacy of all studies involving αDC1/TBVA cell-based treatment, at both Institutions

Pawel Kalinski, MD, PhD. is Professor, Department of Obstetrics, Gynecology & Reproductive Sciences University of Pittsburgh School of Medicine and Vice Chair of Translational Research

Development in Women's Health Magee-Women's Research Institute. Dr. Kalinski will receive de-identified data related to patients enrolled for the purpose of writing papers for publication. Dr. Kalinski will be receiving analyzed data for genomic profiling (via OmniSeq) and expression of RNA message (via RT-qPCR using TaqMan technology). Dr. Kalinski may work with Dr. Kokolus and Roswell lab team in review/interpretation of data. At this time, the trial is closed to accrual. The role of the external collaborator is to review aggregate data summary reports that have been deidentified and they will never have access to identifiable data. The Roswell Park legal department will ensure proper sharing agreements are in place with the University of Pittsburgh School of Medicine. Local IRB approval will be obtained from the University of Pittsburgh School of Medicine.

27 PRIOR APPROVALS

Pending: It is anticipated that the trial will be activated within 6 months of receiving NIH funding

28 COMPENSATION FOR RESEARCH-RELATED INJURY

If the subject believes they have been injured as a direct result of their participation in this research study, they will be advised to notify the Roswell Park Patient Advocate at (716) 845-1365 or the Study Doctor at (716) 845-7505.

Medical diagnosis and treatment for the injury will be offered, and a determination will be made regarding appropriate billing for the diagnosis and treatment of the injury. A financial counselor (716-845-4782) will be able to provide an explanation of coverage and to answer questions the subject may have regarding study related billing.

The subject is not prevented from seeking to collect compensation for injury related to malpractice, fault, or blame on the part of those involved in the research.

29 ECONOMIC BURDEN TO SUBJECTS

The participants will not be subject to any economic burden.

30 CONSENT PROCESS

The Roswell Park SOP: Informed Consent Process for Research (HRP-090) will be followed:

This study will not be initiated until the protocol and informed consent document(s) have been reviewed and approved by a properly constituted Institutional Review Board (IRB) or Independent Ethics Committee (IEC). Each participant (or legal guardian) shall read, understand, and sign an instrument of informed consent prior to performance of any study-specific procedure. It is the responsibility of the investigator to ensure that the participant is made aware of the investigational nature of the treatment and that informed consent is given.

The Investigator is responsible for the retention of the participant log and participant records; although personal information may be reviewed by authorized persons, that information will be treated as strictly confidential and will not be made publicly available. The investigator is also responsible for obtaining participant authorization to access medical records and other applicable study specific information according to Health Insurance Portability and Accountability Act regulations (where applicable).

This study will be conducted in compliance with all applicable laws and regulations of the state and/or country and institution where the participant is treated. The clinical trial should be conducted in accordance with the ethical principles embodied in the Belmont Report: Ethical Principles and Guidelines for the Protection of Human Subjects of Research, consistent with good clinical practice and the applicable regulatory requirements and according to the guidelines in this protocol, including attached appendices.

31 PROCESS TO DOCUMENT CONSENT IN WRITING

The Roswell Park "SOP: Written Documentation of Consent (HRP-091)" will be followed:

The Investigator (or IRB specified designee) is responsible for obtaining written consent from each participant in accordance with GCP guidelines using the approved informed consent form, before any study specific procedures (including screening procedures) are performed. The informed consent form acknowledges all information that must be given to the participant according to applicable GCP guidelines, including the purpose and nature of the study, the expected efficacy and possible side effects of the treatment(s), and specifying that refusal to participate will not influence further options for therapy. Any additional information that is applicable to the study must also be included. Additional national or institutionally mandated requirements for informed consent must also be adhered to. The participant should also be made aware that by signing the consent form, processing of sensitive clinical trial data and transfer to other countries for further processing is allowed.

The Investigator or designee shall provide a copy of the signed consent form to the participant and the signed original shall be maintained in the Investigator File. A copy of the signed consent form must be filed in the participant file. At any stage, the participant may withdraw from the study and such a decision will not affect any further treatment options.

32 DRUGS OR DEVICES

Roswell Park will hold the IND for this study.

IND NUMBER: BB-IND 16,704

32.1 Celecoxib

A sulfa non-steroidal anti-inflammatory drug (NSAID) used in the treatment of osteoarthritis, rheumatoid arthritis, acute pain, painful menstruation and menstrual symptoms, and to reduce numbers of colon and rectum polyps in patients with familial adenomatous polyposis.

32.1.1 Other Names

Celebrex, Celebra or Onsenal (commercially available)

32.1.2 Formulation and packaging

Celecoxib as capsules in the following dosages: 100 mg and 200 mg.

32.1.3 Drug Shipment

Celecoxib will be provided by Roswell Park (to be purchased with grant or startup funding).

32.1.4 Drug administration

Celecoxib 200mg will be administered orally twice a day approximately 12 hours apart on Days 1-3 of Cycles 1-4). The morning dose will be given to the participant at the same time as the other pre-medications for the CKM regimen. The evening dose of Celecoxib will be self-administered by the participant at home and documented on the patient diary Appendix H).

32.1.5 Drug Storage and accountability

The Investigator or designate will be responsible for ensuring that the investigational product is securely maintained in a locked, limited access facility in accordance with the applicable regulatory requirements.

Celecoxib is to be stored at room temperature (20-25°C), away from light and moisture. Brief storage between 59-86 °F (15-30 °C) is permitted.

Drug storage temperature will be maintained and recorded, as applicable.

32.2 Interferon Alpha-2b (Bioferon, replacing the discontinued INTRON-A)

32.2.1 Active Substance and Source

Interferon α-2b (BIOFERON®) is supplied in a single-dose vial containing sterile lyophilized powder. Each vial contains 10 million lnternational Units (MIU) of recombinant human interferon alfa 2b, glycine, dibasic sodium phosphate dodecahydrated, monobasic sodium phosphate anhydrous and human albumin. For its administration, lyophilized powder should be reconstituted by adding 1mL of sterile water for injection. Bioferon® is not FDA-approved and is considered an investigational new drug in the United States.

32.2.2 Drug Shipment

Interferon α-2b (Bioferon®) will be provided by Biosidus S.A. and shipped to the Roswell Park IDS.

The date of receipt and the amount of drug received will be documented. Drug shipment records will be retained by the investigational pharmacist or designee.

How supplied: Vial containing 10 million Units r-Hu-IFN α-2b freeze dried powder. Each package contains 1 solvent ampule with 1 mL sterile WFI.

32.2.3 Preparation

The lyophilized product is reconstituted as directed by the manufacturer. Investigational Drug Service Pharmacy (IDS) will prepare and dispense.

10 million units/mL, lyophilized powder, which must be reconstituted prior to administration.

- Vial size: 10 million units/vial
- Diluent: Interferon α-2b (Bioferon®) should be reconstituted with 1 mL sterile water to reach a final concentration of 10:1. IV dose should be diluted in sodium chloride 0.9%/100 mL given over 30 minutes. The final concentration of interferon α-2b should not be less than 10 million units/100 mL.

Once reconstituted, mix the suspension with gentle rotation movements, do not shake vigorously.

For IV injection, it is recommended that Interferon α-2b be administered as a solution of no less than 10 million units/100 mL to minimize adsorption of the drug to glass and plastic containers.

32.2.4 Storage and Stability

The Investigator or designate will be responsible for ensuring that the investigational product is securely maintained in a locked, limited-access facility, as specified and in accordance with the applicable regulatory requirements.

Interferon α-2b (BIOFERON®) should be kept refrigerated (2 - 8 °C). Do not freeze. Do not freeze the ampoule containing water for injection since it can leak. Once the Interferon α-2b is reconstituted, it should be used within the following 24 hours, storing the solution refrigerated (2 - 8ºC) and following strict aseptic conditions during powder reconstitution.

Vials containing reconstituted solution should be used within two hours if they are kept at room temperature, or within 24 hours if they are stored at 2 - 8ºC.

This preparation does not contain preservative agents. Therefore, it is advisable not to extract more than one dose from the vial to prevent contamination. Do not use this medication after the expiry date printed in its container.

Drug storage temperature will be maintained and recorded, as applicable.

32.2.5 Drug Administration

Interferon α-2b (10 million units/M^2 or 20 million units/M^2) will be administered intravenously over 30 minutes during all CKM cycles.

32.2.6 Concomitant Medications

Interactions between Interferon α-2b and other drugs have not been fully evaluated. Caution should be exercised when administering Interferon α-2b therapy in combination with other potentially myelosuppressive agents such as zidovudine. Concomitant use of Interferon α-2b and theophylline decreases theophylline clearance, resulting in a 100% increase in theophylline levels. Concomitant Interferon α-2b and REBETOL® (Ribavirin) use is contraindicated.

32.3 Rintatolimod (poly IC analog)

A substituted double stranded polyribonucleic acid (polyI: polyC12U), rintatolimod preserves activity of polyIC with a much improved systemic toxicity profile. The product has been studied extensively for use as a cellular treatment adjuvant and for its direct antiviral activity, as well in several cancer studies as a monotherapy, but most extensively in chronic fatigue syndrome (CFS).

32.3.1 Other Names

polyIC_{12}U, Ampligen®, poly I: polyC_{12}U; Polyinosinic: polycytidylic-polyuridylic acid; polyriboinosinic/polyribocytidylic (uridylic) acid

32.3.2 Formulation and Packaging

Rintatolimod is supplied as a liquid solution in glass bottles containing 200 mg (100 mg in case of toxicity) per 80 mL. Rintatolimod is a colorless solution containing 2.5 mg/mL in physiological salts (0.15 M NaCl, 0.01 M phosphate, 0.001 M Mg++). The product does not contain preservatives or antioxidants.

32.3.3 Drug Shipment

Rintatolimod will be provided by Hemispherix and shipped to the participating site.

The date of receipt and the amount of drug received will be documented. Drug shipment records will be retained by the investigational pharmacist or designee.

32.3.4 Preparing ad Dispensing

A vial of rintatolimod is suitable for direct IV infusion. IDS will prepare and dispense. Each vial should be taken from the refrigerator and allowed to equilibrate to room temperature.

32.3.6 Drug Administration

Rintatolimod 200 mg will be administered by intravenous infusion after Interferon Alpha-2b on Days 1-3 of Cycles 1-4 of the CKM regimen. Additional details on the procedures for receiving, storing and using rintatolimod (Ampligen®) can be found in a separate document entitled "Procedures for Receiving, Storing, and Using Ampligen® (Poly I:Poly $C_{12}U$) Liquid Solution".

The initial administration should begin at a slow rate of infusion (approximately 20 cc/ hour) and increase to 40cc/hour after 30 minutes. Tubing should be flushed with 30 to 50 mL of normal saline solution upon completion.

32.3.7 Handling and Disposal

The Investigator or designee will be responsible for dispensing and accounting for all investigational drug provided by Hemispherix exercising accepted medical and pharmaceutical practices. Study drugs must be handled as cytotoxic agents and appropriate precautions taken per the institution's environmentally safe handling procedures. All investigational drugs will be dispensed in accordance with the Investigator's prescription or written order.

All products dispensed will be recorded on a product accountability record. Records of product lot numbers and dates received will be entered on a product accountability form. This record will be reviewed by the Sponsor's staff or representative during periodic monitoring visits. It is the Investigator's responsibility to ensure that an accurate record of investigational drug issued and returned is maintained.

Used vials (excess drug) will be destroyed according to standard practices after properly accounting for the dispensing. Partially used vials of study drug will not be re-used for other participants.

Under no circumstances will the Investigator supply investigational drug to a third party or allow the investigational drug to be used in a manner other than as directed by this protocol.

In regard to drug receipt, accountability and storage, SOP IDS-601 will be followed.

32.4 Alpha-DC1/ TBVA Cellular Product

The αDC1 cellular product will be produced accordingly to the currently active IND sponsored by Roswell Park. Please refer to Section 2 (Background) for the description of the cellular treatment production and the preclinical and early clinical experience with αDC1/TBVA cell-based treatments and other αDC1-based cellular treatments.

32.4.1 Active Substance and Source

Dendritic cells (DC) are derived from autologous (the patient's own) adherent mononuclear cells (monocytes) in the peripheral blood by 5-day or 6- day culture in the presence of interleukin 4 (IL-

4) and granulocyte-macrophage colony stimulating factor (GM-CSF), followed by 1 day of maturation.

32.4.2 Formulation

The αDC1 cell-based treatment will be manufactured at the Roswell Park cGMP Therapeutic Cell Production Shared resource, in a process analogous to the Roswell Park-held IND (autologous monocyte-derived DCs loaded with UV/gamma radiation-killed autologous tumor material). The αDC1 cells are prepared under cGMP conditions and the final product is vialed and cryopreserved. Labels placed on all tubes contain a unique cellular product lot. The αDC1 cellular product release will follow the quality and safety testing of the cellular product lot.

32.4.3 Packaging and Labeling

All synthesis, production, formulation and packaging of the αDC1 will be in accordance with applicable current Good Manufacturing Practices and meet applicable criteria for use in humans.

32.4.4 Storage and Preparation

The αDC1cellular product will be stored at the cGMP Therapeutic Cell Production Shared Resource and laboratory staff will be responsible for ensuring that study cellular product is securely maintained in accordance with the applicable regulatory requirements. The αDC1 cellular product storage temperature will be maintained, recorded, dated and initialed daily as applicable.

Vials containing the final product, αDC1, will be viably frozen using a controlled rate freezer and stored in the vapor phase of LN2 freezer located on the 4th floor of the CCC Building, in the cGMP suite, Therapeutic Cell Production Shared Resource at Roswell Park.

αDC1s used in the cell-based treatment will be suspended in 5% human serum albumin (HSA) and delivered to the clinic for administration. For preparation of the αDC1 cell-based treatment, the labeled vials of cryopreserved αDC1 are removed from storage in liquid nitrogen and quickly thawed in a 37^{o}C water bath. After two washes, thawed αDC1 will be suspended in 5% HSA and placed in sterile syringes for intradermal or intraperitoneal administration to the patient. Each syringe will be labeled with a custom-designed label, identifying the patient, the cellular product, and the route of administration. The 5% HSA is clinical grade.

32.4.5 Stability

The αDC1 cellular product retains viability for at least 24 hours when stored in 5% HSA at 4^{o}C following thawing, washing and loading onto syringes. However, we observed gradual decrease in IL-12p70 producing function upon prolonged storage. For this reason and to assure uniform quality, DC cell-based injections will be administered within 6 hours after their release.

32.4.6. Dosing and Administration

Each 0.2 mL of the αDC1 cell suspension will contain approximately 10 x 10^6 cells and will be loaded onto a single tuberculin syringe. The αDC1 cell suspension will be administered intradermally (i. d.) by the investigator or qualified designee. The investigator/designee will inject approximately 0.1 mL of the syringe contents intradermally and then will remove the needle from the patient. Using the same needle, the investigator/designee will then inject the remaining 0.1 mL of the syringe contents intradermally on the opposite side of the body (the other thigh). The upper

thighs are the preferred locations for the injections, however upper arms are also allowed. The injections will be administered within 6 hours of thawing and release (target: 2 hrs).

32.4.7 Handling and Disposal

The Investigator will be responsible for dispensing and accounting for all investigational drug provided by the cGMP Therapeutic Cell Production Facility at Roswell Park. All investigational drugs will be dispensed in accordance with the Investigator's prescription or written order.

All products dispensed will be recorded on a product accountability record. Records of product receipt, date, time, and temperature will be entered on a product accountability form. Accurate records must be kept regarding dispensing and return of study cellular treatment, for each individual patient in the study. Reasons for departure from the expected dispensing or return of study cellular treatment must be recorded. The *cellular treatment* transportation container with temperature recorder must be returned to the cGMP Therapeutic Cell Production Facility after administration. The cellular treatment will be transported directly from the cGMP Facility without need to go to the Investigational Drug Service (IDS) in the pharmacy department.

Study drug accountability records will be available for verification by the Clinical Trials Office (CTO) monitor appointed from the CTO office at each monitoring visit. At the completion of the study, there will be a final reconciliation of all study drugs.

An investigational agent dispensing record will be kept current and will contain the following information:

- Patient's identification information (i.e., patient identification number and medical record number).
- Date and quantity of drug dispensed.
- Date and quantity of drug returned to the Investigator/pharmacy (if any).
- Date and quantity of accidental loss of study drug (if any).

These inventories must be made available for inspection by the CTO monitor. It is the Investigator's responsibility to ensure that an accurate record of investigational drug issued and returned is maintained.

At the end of the study, the CTO monitor will also collect the original investigational drug dispensing records. Remaining unused αDC1 cellular product will be stored at the cGMP Therapeutic Cell Production Facility for a period that is consistent with applicable regulations. Unused αDC1 cellular product will be used for laboratory research or disposed. Any disposal of investigational agent shall be noted on the investigational drug disposition log and signed-off by a second person. At the end of the study, the CTO monitor will collect the original drug disposition record.

Under no circumstances will the Investigator supply investigational drug to a third party or allow the investigational drug to be used in a manner other than as directed by this protocol.

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34 APPENDICES

Appendix A ECOG Performance Status Scores

Description	Status
Fully active, able to carry on all pre-disease performance without restriction.	0
Restricted in physically strenuous activity but ambulatory and able to carry out work of a light or sedentary nature, e.g., light housework, office work.	1
Ambulatory and capable of all self-care but unable to carry out any work activities.	2
Capable of only limited self-care, confined to bed or chair more than 50% of waking hours.	3
Completely disabled. Cannot carry on any self-care. Totally confined to bed or chair.	4
Dead	5

Appendix B INVESTIGATOR STUDY ELIGIBILITY VERIFICATION FORM INCLUSION CRITERIA

Participant Name: _____

Medical Record No.: _____

Title: A Phase II Study of Type-1 Polarized Dendritic Cell (αDC1)-based Treatment in Combination with Tumor-Selective Chemokine Modulation (CKM: Interferon alpha-2b, Rintatolimod and Celecoxib) in Melanoma Patients with Primary PD-1/PD-L1 Resistance

INCLUSION CRITERIA				
Yes	**No**	**N/A**	**All answers must be "Yes" or "N/A" for participant enrollment.**	**Date**
☐	☐	☐	1. Age ≥ 18 years of age.	
☐	☐	☐	2. Participant must be HLA-A2+, Retesting is not required for patients who have previous documented positivity	
☐	☐	☐	3. Have IO-refractory melanoma with primary PD-1/PD-L1-resistance. *Note*: Any lines of prior therapies are allowed, but the last line needs to include an anti PD-1 or anti PD-L1 agent. The prior treatments may include any standard and/or experimental therapies.	
☐	☐	☐	4. Have ≥ 1 tumor site amenable to core needle biopsy that is not the site of disease used to measure antitumor response	
☐	☐	☐	5. Have measurable disease based on RECIST 1.1 criteria present.	
☐	☐	☐	6. Have an ECOG Performance Status of 0-2. Refer to **Appendix A**.	
☐	☐	☐	7. Have normal organ and marrow function as defined below: • Platelets ≥ 75,000/ microliter • Hemoglobin ≥ 9 g/ deciliter • Absolute Neutrophil Count (ANC) ≥ 1000/microliter • Creatinine < 1.5 X institutional upper limit of normal (ULN) OR creatinine clearance ≥ 50 mL/min by Cockroft-Gault formula for subjects with creatinine levels ≥ 1.5 x ULN (**Appendix D**). • Total bilirubin not greater than 1.5 X institutional ULN, except for patients with known Gilberts Syndrome, who are eligible to no more than 2 X institutional ULN. • AST(SGOT) and ALT(SGPT) no greater than 3 x institutional ULN OR, no greater than 5 x ULN for subjects with liver metastases	
☐	☐	☐	8. Participants of child-bearing potential must agree to use adequate contraceptive methods (e.g., hormonal or barrier method of birth control; abstinence) prior to study entry. Should a woman become pregnant or suspect she is pregnant while she or her partner is participating in this study, she should inform her treating physician immediately.	

INCLUSION CRITERIA				
Yes	**No**	**N/A**	**All answers must be "Yes" or "N/A" for participant enrollment.**	**Date**
☐	☐	☐	9. Participant must understand the investigational nature of this study and sign an Independent Ethics Committee/Institutional Review Board approved written informed consent form prior to receiving any study related procedure.	
☐	☐	☐	10. Candidate for continuation/resumption of anti-PD-1/PD-L1 blockade (in parallel to DC vaccine and CKM).	

Investigator Signature: _____ **Date:** _____

Printed Name of Investigator: _____

Appendix C INVESTIGATOR STUDY ELIGIBILITY VERIFICATION FORM EXCLUSION CRITERIA

Participant Name: _____

Medical Record No.:_____

Title: A Phase II Study of Type-1 Polarized Dendritic Cell (αDC1)-based Treatment in Combination with Tumor-Selective Chemokine Modulation (CKM: Interferon alpha-2b, Rintatolimod and Celecoxib) in Melanoma Patients with Primary PD-1/PD-L1 Resistance

EXCLUSION CRITERIA				
Yes	**No**	**N/A**	**All answers must be "Yes" or "N/A" for participant enrollment.**	**Date**
☐	☐	☐	1. Is currently being treated with systemic immunosuppressive agents, including steroids: Subjects will be ineligible until 3 weeks after removal from immunosuppressive treatments, *except* when they are administered as replacement therapy for endocrine dysfunction (and receive no more than 10 mg prednisone or equivalent: inhaled steroids are allowed.	
☐	☐	☐	2. Has had prior anti-cancer therapy within 2 weeks prior to study day 1 or who has not recovered (i.e., no more than Grade 1 or at baseline) from adverse events due to a previously administered agent, except for neuropathy (no more than Grade 2) or alopecia or vitiligo (any grade)..	
☐	☐	☐	3. Has a known additional malignancy that is progressing or requires active treatment.	
☐	☐	☐	4. Has known active central nervous system (CNS) metastases and/or carcinomatous meningitis. Treated brain metastases are allowed, if stable for more than 4 weeks (and receive no more than 10 mg prednisone or equivalent: inhaled steroids are allowed).	
☐	☐	☐	5. Has a history of cardiac event(s) (acute coronary syndrome, myocardial infarction, or ischemia (within 3 months of signing consent) or, subject has a New York Heart Association classification of III or IV.	
☐	☐	☐	6. Has an active infection requiring systemic therapy.	
☐	☐	☐	7. Has known active Hepatitis B or Hepatitis C infection.	
☐	☐	☐	8. Has known immunosuppressive disease (e.g. HIV, AIDS or other immune depressing disease). Testing is not mandatory.	
☐	☐	☐	9. Has known serious hypersensitivity reactions to peg-interferon alpha-2b or interferon alpha-2b.	
☐	☐	☐	10. Prior allergic reaction or hypersensitivity to sulfonamides, celecoxib, or NSAIDs.	
☐	☐	☐	11. Has received a blood transfusion in the two weeks prior to leukapheresis.	
☐	☐	☐	12. Women of child bearing potential who are pregnant or nursing.	
☐	☐	☐	13. Unwilling or unable to follow protocol requirements.	
☐	☐	☐	14. Any condition which in the Investigator's opinion deems the participant an unsuitable candidate to receive study drug.	

EXCLUSION CRITERIA				
Yes	**No**	**N/A**	**All answers must be "Yes" or "N/A" for participant enrollment.**	**Date**
☐	☐	☐	15. Patients who showed initial response to PD-1/PD-L1 blockade and developed secondary resistance.	

Participant meets all entry criteria: ☐ **Yes** ☐ **No**

If "NO", do not enroll participant in study.

Investigator Signature: _____ Date: _____

Printed Name of Investigator: _____

Appendix D Cockcroft-Gault Equation

Cockcroft-Gault Equation*

Men: CrCl = [(140-YR) x IBW] / (PCr x 72)

Women: CrCl = 0.85 x [(140-YR) x IBW] / (PCr x 72)

Where:

CrCl is creatine clearance (mL/min)

IBW is ideal body weight (kg)

PCr is plasma creatinine (mg/dL)

YR is age (years)

Appendix E NYHA CLASSIFICATION

NEW YORK HEART ASSOCIATION CLASSIFICATION OF CARDIAC DISEASE

Class	Functional Capacity	Objective Assessment
I	Patients with cardiac disease but without resulting limitations of physical activity. Ordinary physical activity does not cause undue fatigue, palpitation, dyspnea, or anginal pain.	No objective evidence of cardiovascular disease.
II	Patients with cardiac disease resulting in slight limitation of physical activity. They are comfortable at rest. Ordinary physical activity results in fatigue, palpitation, dyspnea, or anginal pain.	Objective evidence of minimal cardiovascular disease.
III	Patients with cardiac disease resulting in marked limitation of physical activity. They are comfortable at rest. Less than ordinary activity causes fatigue, palpitation, dyspnea, or anginal pain.	Objective evidence of moderately severe cardiovascular disease.
IV	Patients with cardiac disease resulting in inability to carry on any physical activity without discomfort. Symptoms of heart failure or the anginal syndrome may be present even at rest. If any physical activity is undertaken, discomfort is increased.	Objective evidence of severe cardiovascular disease.

Appendix F RECIST 1.1 Criteria

Objective Tumor Response

All protocol-defined imaging studies must be performed at the investigative site or sponsor-approved facility using protocol-defined parameters. The same method of assessment and the same technique should be used to characterize each identified and reported lesion at baseline and during follow-up. RECIST 1.1 will be used to assess objective tumor response.

Target Lesions

All measurable lesions up to a maximum of 2 lesions per organ and 5 lesions in total, representative of all involved organs, will be identified as target lesions and recorded and measured at baseline. Target lesions will be selected on the basis of their size. Lesions with the longest diameter (short axis for lymph nodes) and are ≥ 10 mm (CT and MRI), ≥ 15 mm lymph nodes, > 20 mm CXR and are for accurate repetitive measurements (either by imaging techniques or clinically) will be chosen. A sum of the longest diameter (short axis for lymph nodes) of all target lesions will be calculated and reported as the baseline sum diameters. This will be used as reference to further characterize the objective tumor response of the measurable dimension of the disease.

- **Complete Response (CR):** Disappearance of all target lesions. Any lymph nodes must have a reduction in short axis to < 10 mm. Changes in tumor measurements must be confirmed by repeat studies performed no less than 6 weeks after the criteria for response are first met.
- **Partial Response (PR):** At least a 30% decrease in the sum of diameters of target lesions, taking as reference the baseline sum diameters. Changes in tumor measurements must be confirmed by repeat studies performed no less than 6 weeks after the criteria for response are first met.
- **Progressive Disease (PD):** At least a 20% increase in the sum of diameters of target lesions, taking as references the smallest sum on study (this includes the baseline sum if that is the smallest on study). In addition to the relative increase of 20%, the sum must also demonstrate an absolute increase of at least 5 mm. The appearance of one or more new lesions is also considered progression.
- **Stable Disease (SD):** Neither a sufficient shrinkage to qualify for PR nor sufficient increase to qualify for PD, taking as reference the smallest sum diameter while on study. Participants having a documented response with no confirmation of the response will be listed with stable disease.

Non-Target Lesions

All other small lesions (longest diameter < 10 mm or lymph nodes ≥ 10 mm to < 15 mm short axis) and non-measurable lesions (i.e., leptomeningeal disease, ascites, pleural or pericardial effusion, inflammatory breast disease, lymphangitic involvement of skin or lung, blastic bone lesions, or abdominal masses / abdominal organomegaly identified by physical exam that is not measurable by imaging) should be identified as non-target lesions and indicated as present in the source documents at baseline. The general location will also be documented on the images drawing a regularly-shaped Region of Interest. Measurements of the non-target lesions will not be

performed, but the presence or absence of each should be noted throughout follow-up and evaluation.

Complete Response: Disappearance of all non-target lesions and normalization of tumor marker level, if applicable. All lymph nodes must be non-pathological in size (< 10 mm short axis).

Non-Complete Response/Non-Progressive Disease: Persistence of 1 or more non-target lesion(s) and/or maintenance of tumor marker level above the upper limits of normal.

Progressive Disease: Appearance of 1 or more new lesions or the unequivocal progression of existing non-target lesions. Although a clear progression of non-target lesions is exceptional, in such circumstances, the opinion of the treating physician should prevail and the progression status should be confirmed at a later time.

Evaluation of Response

Time point response assessments will be performed every 12 weeks [starting with Week 12 ± 1 week)]. To determine time point response, refer to the tables below:

Time Point Response Criteria: target (± non-target disease)

Target Lesions	Non-Target Lesions	New Lesions	Overall Response
CR	CR	No	CR
CR	Non-CR/Non-PD	No	PR
CR	Not evaluated	No	PR
PR	Non-PD or not all evaluated	No	PR
SD	Non-PD or not all evaluated	No	SD
Not all evaluated	Non-PD	No	NE
PD	Any	Yes or No	PD
Any	PD	Yes or No	PD
Any	Any	Yes	PD

Time Point Response Criteria: non-target disease only

Non-Target Lesions	New Lesions	Overall Response
CR	No	CR
Non-CR/non-PD	No	Non-CR/non-PD[1]
Not all evaluated	No	NE
Unequivocal PD	Yes or No	PD
Any	Yes	PD

1 Non-CR/non-PD is preferred over SD for non-target disease since SD is used as endpoint for assessment of efficacy in trials so to assign this category when no lesions can be measured is not advised.

The best overall response is the best response recorded from the start of study treatment until disease progression, taking into account any requirement for confirmation. In general, the participant's best response assignment will depend on the achievement of both measurement and confirmation criteria and will be determined by combining the participant's status of target lesions, non-target lesions, and new lesions.

- Symptomatic Deterioration: Participants with global deterioration of health status requiring discontinuation of treatment without objective evidence of disease progression at that time, and not related to study treatment or other medical conditions should be reported as progressive disease due to "symptomatic deterioration." Every effort should be made to document objective progression even after discontinuation of treatment due to symptomatic deterioration. Symptomatic deterioration that may lead to discontinuation of treatment include, but is not limited to, symptoms such as:
- Weight loss > 10% of body weight.
- Worsening of disease-related symptoms (e.g., worsening dyspnea, increasing pain/increasing requirement for narcotic analgesics).
- Decline in performance status of > 1 level on ECOG scale.

Confirmation Measurement

If Week 12 CT scan demonstrates stable disease/response, patients will continue in follow-up with CT imaging every 12 weeks until progression, clinical deterioration or withdrawal from study.

Methods of Measurement

Refer to protocol **Section 16.2**

Appendix G Immune-Related Response Criteria (iRECIST)

Tumor response will be assessed using the Immune-Related response Criteria (iRESIST) as described by Seymour et al, 2017(20)*.

iRECIST Response Assessment

Overall response will also be assessed using iRECIST. Immunotherapeutics may result in infiltration of immune cells leading to transient increase in the size in malignant lesions, or undetectable lesions becoming detectable. The criteria are identical to those of RECIST 1.1 in many respects but have been adapted to account for instances where an increase in tumour burden, or the appearance of new lesions, does not reflect true tumour progression.

Key differences are described below. All responses defined using iRECIST criteria are designated with a prefix. iRECIST time-point and best overall responses will be recorded separately.

Confirming Progression

Unlike RECIST 1.1, iRECIST requires the confirmation of progression and uses the terms iUPD (unconfirmed progression) and iCPD (confirmed progression). Confirmatory scans should be performed at least 4 weeks, but no longer than 8 weeks after iUPD.

iCPD is confirmed if further increase in tumour burden, compared to the last assessment, is seen as evidenced by one or more of the following:

- Continued increase in tumour burden (from iUPD) where RECIST 1.1 definitions of progression had been met (from nadir) in target, non-target disease or new lesions
    - Progression in target disease worsens with an increase of at least 5 mm in the absolute value of the sum
    - Continued unequivocal progression in non-target disease with an increase in tumour burden
    - Increase in size of previously identified new lesion (s) (an increase of at least 5 mm in the absolute value of the sum of those considered to be target new lesions) or additional new lesions.
- RECIST 1.1 criteria are met in lesions types (target or non-target or new lesions) where progression was not previously identified, including the appearance of additional new lesions.

If iUPD is not confirmed at the next assessment, then the appropriate response will be assigned (iUPD if the criteria are still met, but no worsening, or iSD, iPR or iCR if those criteria are met compared to baseline). As can be seen in table 2, the prior documentation of iUPD does not preclude assigning iCR, iPR, or iSD in subsequent time-point assessments or as best overall response (BOR) providing that iCPD is not documented at the next assessment after iUPD.

New lesions

New lesions should be assessed and measured as they appear using RECIST 1.1 criteria (maximum of 5 lesions, no more than 2 per site, at least 10 mm in long axis (or 15 mm in short axis for nodal lesions), and recorded as New Lesions-Target (NLT) and New Lesion-Non-Target (NLNT) to allow clear differentiation from baseline target and non-target lesions.

New lesions may either meet the criteria of NLT or NLNT to drive iUPD (or iCPD). However, the measurements of target lesions should NOT be included in the sum of measures of original target lesions identified at baseline. Rather, these measurements will be collected on a separate table in the case record form.

PD is confirmed in the New Lesion category if the next imaging assessment, conducted at least 4 weeks (but not more than 8 weeks) after iUPD confirms further progression from iUPD with either an increase of at least 5 mm in the absolute value of the sum of NLT OR an increase (but not necessarily unequivocal increase) in the size of NLNT lesions OR the appearance of additional new lesions.

The following tables describe the time-point response (Table A) and best overall response (Table B) assessment when using iRECIST.

Table A: Time-point (TP) iResponse

Target Lesions*	Non-Target Lesions*	New Lesions*	Time Point Response	
			No prior iUPD*	Prior iUPD**; ***
iCR	iCR	No	iCR	iCR
iCR	Non-iCR/Non-	No	iPR	iPR
iPR	Non-iCR/Non-	No	iPR	iPR
iSD	Non-iCR/Non-	No	iSD	iSD
iUPD with no change OR decrease from las	iUPD with no change O R decrease from last TP	Yes	NA	NLs confirms iCPD if NLs were previously identified and increase in size (≥5 mm in SOM for NLT or any increase for NLNT) or number. If no change in NLs (size or number) from last TP, remains iUPD
iSD	iUPD	No	iUPD	Remains iUPD unless iCPD confirmed based in further increase in size of NT disease (need not meet RECIST 1.1 criteria for unequivocal PD)
iUPD	Non-iCR/Non-iUPD	No	iUPD	Remains iUPD unless iCPD confirmed based on: o further increase in SOM of at least 5 mm, otherwise remains iUPD
iUPD	iUPD	No	iUPD	Remains iUPD unless iCPD confirmed based on further increase in: o previously identified T lesion iUPD SOM ≥5 mm and / or o NT lesion iUPD (prior assessment - need not be unequivocal PD)
iUPD	iUPD	Yes	iUPD	Remains iUPD unless iCPD confirmed based on further increase in: o previously identified T lesion iUPD ≥5 mm and / or o previously identified NT lesion iUPD (need not be unequivocal) and /or o size or number of new lesions previously identified
Non-iUPD/PD	Non-iUPD/PD	Yes	iUPD	Remains iUPD unless iCPD confirmed based on o increase in size or number of new lesions previously identified
* Using RECIST 1.1 principles. If no PSPD occurs, RECIST 1.1 and iRECIST categories for CR, PR and SD would be the same. ** in any lesion category. *** previously identified in assessment immediately prior to this TP.				

All patients will have their iBOR from the start of study treatment until the end of treatment classified as outlined below.

Table B: iRECIST Best Overall Response (iBOR)

TPR1	TPR2	TPR3	TPR4	TPR5	iBOR
iCR	iCR, iP	iCR, iPR, iUPD, NE	iUPD	iCPD	iCR
iUPD	iPR, iSD, NE	iCR	iCR, iPR, iSD, iUPD, NE	iCR, iPR, iSD, iUPD, iCPD, NE	iCR
iUPD	iPR	iPR, iSD, iUPD, NE	iPR, iSD, iUPD, NE,	iPR, iSD, iUPD, NE, iCPD	iPR
iUPD	iSD, NE	PR	iPR, iSD, iUPD, NE	iPR, iSD, iUPD, iCPD, NE	iPR
iUPD	iSD	iSD, iUPD, NE	iSD, iUPD, iCPD, NE	iSD, iUPD, ICPD, NE	iSD
iUPD	iCPD	Anything	Anything	Anything	iCPD
iUPD	iUPD	iCPD	Anything	Anything	iCPD
iUPD	NE	NE	NE	NE	iUPD

- Table assumes a randomized study where confirmation of CR or PR is not required.
- NE = not evaluable that cycle.
- Designation "I" for BOR can be used to indicate prior iUPD to aid in data interpretation.
- For patients with non-target disease only at baseline, only CR or non-CR/non-PD can be assigned at each TPR but is not shown in the table for ease of presentation.

Methods of Measurement

Refer to protocol **Section 16.2**.

* For additional information, refer to the following article and online supplemental information:

Seymour L, Bogaerts J, Perrone A, Ford R, Schwartz LH, Mandrekar S, Lin NU, Litiere S, Dancey J, Chen A, Hodi FS, Therasse P, Hoekstra OS, Shankar LK, Wolchok JD, Ballinger M, Caramella C, de Vries EG, group Rw. iRECIST: guidelines for response criteria for use in trials testing immunotherapeutics. Lancet Oncol. 2017;18(3):e143-e52. doi: 10.1016/S1470-2045(17)30074-8. PubMed PMID: 28271869.

Appendix H Celecoxib Patient Diary

Protocol # _____ Patient Name: _____
Drug Name: Celecoxib

Med. Record #: ___ ___ ___ ___ ___ ___

Study Medication Calendar

You are participating in a study that requires Celecoxib 200 mg to be given twice daily, approximately 12 hours apart on certain days. Your morning dose of Celecoxib will be given to you along with your pre-medications while you are in clinic. Please complete this calendar to document your evening doses of Celecoxib. Your nurse will let you know what time you receive your morning dose of Celecoxib so you can take your evening dose approximately 12 hours after.

Cycle # #_____:

Date			
Time that morning dose was taken (to be completed by the nurse in clinic)			
Evening Dose			
Time			
Patient's Initials			

Please remember to bring this calendar and your pill bottle (including any unused pills) with you to your next study appointment.

Coordinator use only
Date of return: _____
# of pills
(dispensed: _____ -- returned: _____)
_____ x 100 = % adherence: _____
# of pills scheduled _____

Patient signature: _____ Date: _____

Investigator signature: _____ Date: _____

Appendix I Schedule of Procedures and Observations

Evaluation	Screening/ Baseline[1]	Within 2 weeks (± 3 days) prior to Cycle 1	Cycle 1 Days 1-3 (Week 1)	Cycles 2-4 Days 1-3 (Weeks 4, 7, 10)	Treatment Discontinuation (± 3 days)	Week 12[2] (±1 week)	Post-Treatment Follow-Up		
							SafetyFollow-Up[3]	Follow-Up at 3 and 6 months	Long Term Follow-Up
Medical History	X[11]								
Physical Examination (including height and weight)[4]	X		X	X	X	X			
Vital signs[5]	X		X	X	X	X			
ECOG Performance Status	X		X (Day 1 only)	X (Day 1only of each cycle)	X	X			
Concomitant Medications	X	X	X	X	X	X	X	X	
Adverse Events		X	X	X	X	X	X	X	
Survival Status							X	X	X[3a]

Evaluation	Screening/ Baseline[1]	Within 2 weeks (± 3 days) prior to Cycle 1	Cycle 1 Days 1-3 (Week 1)	Cycles 2-4 Days 1-3 (Weeks 4, 7, 10)	Treatment Discontinuation (± 3 days)	Week 12[2] (±1 week)	Post-Treatment Follow-Up		
							SafetyFollow-Up[3]	Follow-Up at 3 and 6 months	Long Term Follow-Up
Laboratory Procedures									
Infectious Disease Panel (HBV and HCV)	X								
Hematology[6]	X		X[6a]	X[6a]	X	X			
Chemistry[7]	X		X[7a]	X[7a]	X	X			
TSH, T4[14]	X		X[14a]	X[14b]					
HLA-A2 typing (confirmation of HLA eligibility)[15]	X								
Urine βHCG pregnancy test[8]	X								

Evaluation	Screening/ Baseline[1]	Within 2 weeks (± 3 days) prior to Cycle 1	Cycle 1 Days 1-3 (Week 1)	Cycles 2-4 Days 1-3 (Weeks 4, 7, 10)	Treatment Discontinuation (± 3 days)	Week 12[2] (±1 week)	Post-Treatment Follow-Up		
							SafetyFollow-Up[3]	Follow-Up at 3 and 6 months	Long Term Follow-Up
Troponin I level[9]	X								
Leukapheresis[10]		X							
Blood Draw for Correlative Analysis[12]			X[12a]	X[12a]	X	X			
Imaging / Other Procedures									
12-lead EKG[9]	X								
Tumor/Disease Assessment (CT scan of the chest abdomen and pelvis)[11]	X					X[11a]		X[11a]	X[11a]
Biopsy[13]		X				X			
Treatment Regimen Administration									

Evaluation	Screening/ Baseline[1]	Within 2 weeks (± 3 days) prior to Cycle 1	Cycle 1 Days 1-3 (Week 1)	Cycles 2-4 Days 1-3 (Weeks 4, 7, 10)	Treatment Discontinuation (± 3 days)	Week 12[2] (±1 week)	Post-Treatment Follow-Up		
							SafetyFollow-Up[3]	Follow-Up at 3 and 6 months	Long Term Follow-Up
Chemokine modulation (CKM)			X	X					
αDC1/TBVA cell-based injection[16]				X					

Footnotes

1. Baseline and/or Screening assessments must be performed within **28** days prior to the first cycle of the CKM treatment regimen.
2. Week 12: Response rate will be evaluated by CT scan, performed as a part of standard care at week 12 (± 1 wk). Any patient with PD will be offered an opportunity to switch to the best alternative treatment, per physician discretion [(e.g., LAG-3 (OPDUALAG™) or CTLA4 blockade (Ipilimumab) ± PD-1/PD-L1 inhibitor (unless contra-indicated)] and will be assessed as per standard of care guidelines. Any patient with a CR, PR or SD will be offered PD-1/PD-L1 blockade (according to the approved standard of care regimen) or best alternative care (per physician discretion). Thereafter, patients will be contacted by phone every 3 months for up to 2 years by medical record review/telephone call for survival status and other anti-cancer therapies.
3. To capture all possible delayed immune-related adverse events, follow-up safety evaluations will occur at least monthly for 90 days after last dose of study drug or until resolution of any drug-related toxicity (medical record review or telephone contact is acceptable).
   3a: Patients will be contacted by phone every 3 months up to 2 years by medical record review/telephone call for survival status and other anti-cancer therapies.

4. Physical exam to include assessment of Head/Ears/Eyes/Nose/Throat (HEENT), Skin, Chest/Lungs, Heart, Neck/Lymph nodes, Abdomen, Extremities and weight. Height required at baseline only.
5. Vital Signs include blood pressure, pulse, temperature and respiratory rate. On days that rintatolimod is given, vital signs will be obtained pre-treatment, 30 minutes (± 5 minutes) and 60 minutes (± 5 minutes) post rintatolimod. Following the αDC1 vaccination patients will be monitored as outpatients (every 20 min ± 5 min) for 1 hour. Monitoring will consist of vital sign measurements, and monitoring of any local/systemic reactions to the injection. This monitoring will include clinical assessment of pain, tenderness, warmth and swelling.
6. CBC with automated differential
   6a: Labs are ONLY taken once per cycle: Day 1 of each cycle unless otherwise clinically indicated.
7. CMP
   7a: Only Day 1 of each cycle unless otherwise clinically indicated.
8. Within 7 days of Day 0 in women of child-bearing potential (and as clinically indicated once on treatment).
9. A single 12-lead ECG and Troponin I level to be performed at screening and then only as clinically indicated thereafter.
10. Patient MUST meet inclusion/exclusion criteria prior to leukapheresis. Leukapheresis to occur approximately 2 weeks prior to Cycle 1-Day 1 (first CKM treatment regimen). It is expected that 1 leukapheresis will yield enough cells for at least 3 i. d. αDC1 cell-based injections. If a sufficient number of cells have not been obtained, an additional leukapheresis may be required in order to complete the treatment protocol. If a sufficient number of cells for 3 i. d. αDC1 cell-based injections and proposed QC testing are still not obtained after the second leukapheresis, the patient will be withdrawn from the study and replaced.
11. Baseline disease assessment may be within 28 days prior to Day 0.
    11a: If Week 12 CT scan demonstrates stable disease/response, patients will continue in follow-up with CT imaging every 12 weeks until progression, clinical deterioration or withdrawal from study. Any patient with PD will be offered an opportunity to switch to CTLA4 blockade (Ipilimumab) ± PD-1/PD-L1 inhibitor and will be assessed as per standard of care guidelines.
12. ONLY Day 1 and Day 3: Refer to Section 0.
    12a: Day 1 and Day 3 of Cycles 1, 2, 3 and 4: Pre-infusion and at 1 hr (± 10 minutes) post-rintatolimod infusion
13. Refer to Section 0: Fresh Biopsy Specimens.
14. Thyroid: TSH and T4 (only twice per protocol):
    14a: Cycle 1 Day 1 only
    14b: Cycle 3 Day 1 only

15. Participant must sign a separate informed consent prior to HLA testing. HLA-A2 positivity is required for study eligibility. Samples for HLA-A2 typing for eligibility will performed by a CLIA certified laboratory, as selected by site.
16. The αDC1 cell-based injection is administered only on Day 1 of Cycles 2-4 (1 hr +30 min/-15 min) prior to initiating CKM pre-treatment.