Proposal Title –

Phase III Double-blind, Placebo-controlled Study of BXCL701 for the Treatment of Metastatic Castration-Resistant Prostate Cancer in Men

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1.0 Introduction and Background

Prostate cancer is a form of cancer that begins in the gland cells of the prostate, which is found only in the male population. Early stages of prostate cancer rely on testosterone to grow and sometimes, lowering testosterone can control growth. If prostate cancer spreads beyond the prostate, it is called "metastatic," and is found growing in other organs or tissues[3]. Metastatic castration-resistant prostate cancer is more commonly known as mCRPC. It can be difficult to treat. Advanced prostate cancer like this can be life threatening if it spreads to other parts of the body[7].

Castration-resistant prostate cancer (CRPC) is a form of advanced prostate cancer. With CRPC, the cancer no longer completely responds to treatments that lower testosterone. It shows signs of growth, like a rising PSA (prostate-specific antigen), even with low levels of testosterone[3]. With Metastatic CRPC (mCRPC), the cancer stops responding to hormone treatment, and it is found in other parts of the body. It can spread to nearby lymph nodes, bones, the bladder, rectum, liver, lungs, and maybe the brain[3].

BXCL701 is an intervention that is orally administered innate immune activator that is designed to initiate inflammation within the tumor microenvironment. The disease area is cancer, and the population is men with metastatic castration-resistant prostate cancer, or mCRPC[1]. The current

standard of care intervention, pembrolizumab does not address cancers that appear "cold" or uninflamed[6]. BXCL701 is created to render those "cold" tumors as "hot" to make them detectable by adaptive immune systems. Thus, this trial must be conducted because it is targeting an unaddressed research area.

This double-blinded, two parallel group superiority design study evaluates the impact of BXCL701 on mCRPC in male adults[2]. Specifically, the ***clinical question*** for this trial is: Among males aged 18 years and older, is there a statistically significant improvement in composite response rates for mCRPC patients who are orally administered BXCL701 in combination with Pembrolizumab daily compared to patients who are administrated only Pembrolizumab daily?

The target population is male adults who are 18 years old or older who are affected by mCRPC. The intervention group will be utilizing both the new intervention, BXCL701, and the current standard of care intervention, pembrolizumab. The patients will receive a fixed-dose of pembrolizumab (200 mg IV q21-days) once orally, along with BXCL701 orally on days 1-14 at recommended Phase 2 dose (RP2D) schedule, which is BXCL701 0.3 mg BID. The time of follow up for each individual patient will be 21 days[1]. The control group will be utilizing only the current standard of care, pembrolizumab. The patients will receive a fixed-dose of pembrolizumab (200 mg IV q21-days) once orally. The time of follow up for each individual patient will be 21 days[1].

2.0 Objectives

i) Primary

The primary objective is assessing the composite response rate for the combination of BXCL701 and Pembrolizumab. The time frame is up to 36 months[3].

Null hypothesis (H_0) with Alpha = 0.05:

- For patients who orally consume BXCL701 in combination with Pembrolizumab, the mean change of composite response rates from the baseline is the same as the patients who only take Pembrolizumab at the end of the 21-day treatment period.

Alternative hypotheses (H_a) with Alpha = 0.05:

- For patients who orally consume BXCL701 in combination with Pembrolizumab, the mean change of composite response rates from the baseline is *higher* than those patients who only take Pembrolizumab at the end of the 21-day treatment period.
- For patients who orally consume BXCL701 in combination with Pembrolizumab, the mean change of composite response rates from the baseline is *lower* than those patients who only take Pembrolizumab at the end of the 21-day treatment period.

Direction of clinical interest will be one-sided test to assess if the addition of BXCL701 is superior to the current standard of care by itself. The analysis of primary outcome will utilize a two-sided test to see whether there is any difference between the two group in terms of mean change in composite response rates. The two-sided test is appropriate because two groups are under comparison and we want to see if either group can alter the composite response rate, and how they are altered.

Patient important outcomes will be used in this trial because the composite response rate is the patient important outcome that directly reflects the patient's functionality. The baseline score for the composite response rate along with the composite rate at the end of the trial will be recorded for every single patient. The scores will demonstrate the effectiveness of the interventions on mCRPC.

ii) Secondary

1. Assessing the pharmacodynamic profile of BXCL701 and Pembrolizumab. The time frame is up to 36 months[3]. Assessed by measuring relevant effects on those cytokines previously shown to be modulated by BXCL701 in humans[5].

Null hypothesis (H_0) with Alpha = 0.05:

- For patients who orally consume BXCL701 in combination with Pembrolizumab, the pharmacodynamic profile is the same as patients who only take Pembrolizumab at the end of the 21-day treatment period.

Alternative hypothesis (H_a) with Alpha = 0.05:

- For patients who orally consume BXCL701 in combination with Pembrolizumab, the pharmacodynamic profile is *not* the same as patients who only take Pembrolizumab at the end of the 21-day treatment period.

2. Determining the risk profile for the use of BXCL701 in combination with Pembrolizumab. The time frame is up to 36 months[3]. Determined by the frequency and severity of known and unknown adverse events with the use of BXCL701 in combination with Pembrolizumab[5].

Null hypothesis (H_0) with Alpha = 0.05:

- For patients who orally consume BXCL701 in combination with Pembrolizumab, the risk profile is the same as patients who only take Pembrolizumab at the end of the 21-day treatment period.

Alternative hypothesis (H_a) with Alpha = 0.05:

- For patients who orally consume BXCL701 in combination with Pembrolizumab, the risk profile is *not* the same as patients who only take Pembrolizumab at the end of the 21-day treatment period.

3. Estimating the median overall survival for the combination of BXCL701 and Pembrolizumab in groups A and B. The time frame is up to 36 months. Estimated with the median time frame with overall survival with the use of BXCL701 in combination with Pembrolizumab[5].

Null hypothesis (H_0) with Alpha = 0.05:

- For patients who orally consume BXCL701 in combination with Pembrolizumab, the estimated median overall survival is the same as patients who only take Pembrolizumab at the end of the 21-day treatment period.

Alternative hypotheses (H_a) with Alpha = 0.05:

- For patients who orally consume BXCL701 in combination with Pembrolizumab, the estimated median overall survival is *higher* than the patients who only take Pembrolizumab at the end of the 21-day treatment period.
- For patients who orally consume BXCL701 in combination with Pembrolizumab, the estimated median overall survival is *lower* than the patients who only take Pembrolizumab at the end of the 21-day treatment period.

iii) Safety

First:

- Adverse reactions toward BXCL701, such as hypertension, fatigue, or rashes. This will be measured using the coordinators' judgment daily, and the response will be recorded as categorical answers, specifically YES or NO.

Second:

- Allergic reactions toward BXCL701. This will be measured using a skin prick test to check for immediate allergic reactions, and the response will be recorded as categorical answers, specifically YES or NO. Data will be collected and assessed by coordinators daily.

Third:

- Significant increase in stress level. This will be measured using the Perceived Stress Scale, and it will be recorded as continuous variables, specifically a scale from 0 to 10. It will be assessed by coordinators daily.

3.0 Trial Design

i) RCT Features

This trial is a double-blinded, two parallel group superiority design study. It incorporates an achieves the following seven features of an idea RCT as seen below:

Prospective: Longitudinal cohort study conducted over 26 months to see how the two patient groups will react to the new treatment intervention.

Intervention: The intervention is an active treatment, BXCL701, that is administrated by the workers.

Control Group: This trial consists of two groups, with one being the treatment group (active intervention and active standard of care) and one control group (active standard of care).

Randomization: The assignment of patients into the two groups is by chance.

Double blinding: There will be double blinding to ensure that the doctors, their staff, and the patients themselves do not know the interventions to which the patients are assigned to.

Intent-To-Treat Primary Analysis: The patients will be assessed according to the groups they were originally assigned to.

Complete Follow-up: The follow-up period for each patient is 21 days and must be completed after assessment.

ii) Blinding

This study will be a double-dummy scheme. Group A will be randomized to BXCL701 and pembrolizumab. Patients will receive two sets of identical medication bottles. One set contains active BXCL701, and one set contains active pembrolizumab. Group B will be randomized to just pembrolizumab. Patients will receive two sets of identical medication bottles. One set contains placebo BXCL701, and one set contains active pembrolizumab. The double-dummy scheme is a medication masking system that ensures blinding, specifically blinding the doctors, their staff, and the patients themselves so that they do not know the interventions to which the patients are assigned to.

iii) Randomization

This is a randomized controlled trial, the best method to prove causality in spite of various limitations. Random allocation is a technique where individuals are assigned to groups entirely by chance with no regard to the will of researchers or patients' condition and preference. This allows researchers to control all known and unknown factors that could possibly affect results in treatment groups and control groups[7].

In this trial, enrollment will have screened individuals as n = a, and screened failures as n = b, where the excluded number of patients is b. Then, they will be randomized to where n = a – b. After this enrollment step is allocation, where the patients, now randomized, will be allocated to either the combination of BXCL701 and Pembrolizumab, or just Pembrolizumab. This can be seen as n = (a – b) / 2. Following this allocation is follow-up, where the patients will receive the interventions. This can be seen as n = (a – b) / 2 – c. During this phase, the patients who are either discontinued or lost to follow-up will be noted as n = c for each arm intervention. The last part of this is the analysis, where the patients will be analyzed such that n = (a – b) / 2 – c, for each arm intervention.

There will be randomly permuted blocks to ensure that the size of the following block is completely randomized. The treatment assignment will be generated by using published tables of random numbers and assigning subjects based on the following number that is listed on the table. No stratification will be used since there is already an overall randomization assignment for the participants as well as all other factors. Block sizes of different values with pre-specified proportions will be inputted.

iv) Inclusion and Exclusion Criteria

Main inclusion criteria[5]:

1. Patient is 18 years or older.
2. Patient has signed informed consent.
3. Patient can adhere to study visit schedule along with other protocol requirements.
4. Patient has progressive, metastatic castration-resistant disease, as defined by the PCWG3 criteria.
5. Progression during or following completion of at least 1 prior line of systemic therapy for locally advanced or metastatic prostate cancer.
6. Patient has serum testosterone <50 ng/dL during Screening
7. Patient has Eastern Cooperative Oncology Group performance status of 0 to 2.
8. Patient's acute toxic effects of previous anticancer therapy have resolved to ≤Grade 1
9. Patient has adequate baseline organ and hematologic function.
10. Male patients and their female partners must agree and commit to use a barrier contraception throughout the duration of the study until at least 6 months following the last dose of study drug, in addition to their female partners using either an intrauterine device or hormonal contraception and continuing until at least 6 months following the last dose of study drug.

Main exclusion criteria[5]:

1. Patient has received treatment with >2 cytotoxic chemotherapy regimens for castration-resistant prostate cancer (CRPC).
2. Patient has received external-beam radiation or another systemic anticancer therapy within 14 days or 5 half-lives, whichever is shorter, prior to study treatment.

3. Patient has received treatment with an investigational systemic anticancer agent within 14 days prior to study drug administration.
4. Patient has clinically significant cardiovascular disease.
5. QT interval corrected for heart rate using Bazett's formula (QTcB) >480 msec at Screening.
6. Patient has uncontrolled pulmonary disease, symptomatic brain metastases, active autoimmune disease, immunodeficiency, uncontrolled intercurrent illness, human immunodeficiency virus, hepatitis B/C, or any medical condition which, in the opinion of investigator, puts the patient at an unacceptably high risk for toxicity.
7. Patient has known positive status for human immunodeficiency virus, active or chronic Hepatitis B, or Hepatitis C. Screening is not required.

v) Enrolling Centers

Enrolling centers will include research hospitals, medical colleges, research centers, and through telehealth.

vi) Data Coordination and Trial Management

There will be Data Coordinating Center and Clinical Trial Management resources at a level appropriate for this trial since it is a multicenter trial. The role of the DCC is to ensure quality standard of data management and analysis, provide necessary manuscripts, document, and information for data collection and other aspects of the trial, coordinate and monitor the study. It is responsible for the collection, verification, and storage of all data collected from all the sites that are involved in this multicenter trial. The role of the CTM is to ensure the clinical trials will

be completed within time frame, budgets, and desired quality, and to lead the trial activities where necessary. It serves as a single-centralized, web-based enterprise resource to support the clinical research studies conducted across all sites of the trial.

4.0 Data Collection and Patient Follow-up

i) Outcome Details

Primary outcome:

Change from baseline in composite response rate for the combination of BXCL701 and Pembrolizumab in 36 months[1]. This is a continuous variable and will be assessed using the response evaluation criteria for solid tumors includes circulating tumor cell conversion from >5/7.5 mL to <5/7.5 mL per Veridex assay, along with 50% or greater prostate-specific antigen decline from baseline[3]. It will be tracked and assessed weekly by the coordinators using the evaluation criteria. This instrument accurately reflects the outcome of interest because the coordinators are professionally trained, and the patients will be visited by the coordinators daily at the hospitals they are staying at. The composite response rate is a continuous variable. The data coordinators from the Data Coordinating Center will record the participants composite response rates for assessment.

Secondary outcome:

First:

- Change from baseline in the pharmacodynamic profile as assessed by measuring relevant effects on those cytokines previously shown to be modulated by BXCL701 in humans.

The patients' pharmacodynamic profile change status will be recorded as a categorical variable, YES or NO, weekly for up to 36 months. The patient will be visited in person at the hospital for pharmacodynamic profile assessments. This instrument accurately reflects the outcome of interest because it is a precise measure of cytokine effects, and the patients will be visited by the coordinators weekly at the hospitals they are staying at. The score will be recorded by the data coordinator from the Data Coordinating Center.

Second:

- Change from baseline in risk profile as assessed using the frequency and severity of known and unknown adverse events with the use of BXCL701 in combination with Pembrolizumab. The patients' risk profile change status will be recorded as a categorical variable, YES or NO, weekly for up to 36 months. The patient will be visited in person at the hospital for their risk profile assessments. This instrument accurately reflects the outcome of interest because it is a precise measure regarding the frequency and severity of adverse events, and the patients will be visited by the coordinators weekly at the hospitals they are staying at. The adverse reaction status will be recorded by the data coordinator from the Data Coordinating Center.

Third:

- Change from baseline in median overall survival as assessed by calculating the median time frame with overall survival with the use of BXCL701 in combination with Pembrolizumab. Participants will take an assessment and receive a score on a continuous scale from 0 to 10 weekly for up to 36 months. The patient will be visited in person at the hospital for their scores and assessments. This instrument accurately reflects the outcome of interest because it is a precise measure of overall survival, and the patients will be

visited by the coordinators weekly at the hospitals they are staying at. The score will be recorded by the data coordinator from the Data Coordinating Center.

ii) Data Collection Mechanism

The mechanism for data collection will be a web-based data management system with Electronic Case Report Forms.

iii) Schedule of Visits

Table 1. Schedule of Visits

	Screening Period		**Treatment Period**						
Milestones			**Baseline**					**Last Treatment Dispenses**	**EOT EOS**
Visit #	1	2	3	4	5	6	7	8	9
Date #	1	8	15	22	29	36	43	36	43
Week #	1	2	3	4	5	6	7	8	9
Screening/Baseline:									
Informed consent									
Inclusion/exclusion									
Medical history									
Randomization									

IVRS/IWRS Contact									
Treatment:									
Trial Training									
Administer BXCL701									
Study BXCL701 dispensation									
Data collection assessment									
Safety Assessments:									
Perceived stress scale score									
Adverse reaction towards BXCL701									
Allergic reaction towards BXCL701									
Primary Outcome Assessment:									
Δ from baseline in composite response rate									

Secondary Outcomes Assessment:									
Δ from baseline in pharmacodynamic profile									
Δ from baseline in risk profile									
Δ from baseline in median overall survival									

iv) Trial Timeline

Design and startup before randomization –8 months

Randomization – 1 month

Day 1 – First patient visit

All patients follow-up period – Up to 36 months

Last Day – Last patient visit

Data cleaning – 5 months

Final database lock

Data analysis – 3 months

CSR

5.0 Statistical Considerations

i) Type of Outcome

The primary outcome, Change from baseline in composite response rate, is a continuous variable. Therefore, the two-sample t-test, assuming equal variance, is utilized.

Let μ_I = The true mean of change from baseline in composite response rate in the BXCL701 in combination with Pembrolizumab group.

μ_c = The true mean of change from baseline in BAI score in the Pembrolizumab group.

Null hypothesis (H_a): $\mu_c - \mu_I = 0$

Alternative hypothesis (H_0): $\mu_c - \mu_I \neq 0$

Statistical design: Superiority

Type I error: 0.05

Power: 0.9

Test: Two-sided

ii) Power Calculation:

The unadjusted effect size is calculated to be 0.90 using PASS when using a sample size of 135 and a standard deviation of 2 for each intervention arm. These values were chosen from the estimated enrollment values of the trial[5]. As for the adjusted effect size, we must assume that for both treatment and control group the percent crossover is 5%, non-compliers is 20%, and full - compliers is 75%. The adjusted effect size is calculated to be 0.63 (Appendix 1).

The effect size indicates the difference in outcomes between the intervention groups. It should be

noted that a small effect size depicts a small difference between the groups. Therefore, the least clinically meaningful effect size is 0.63. This will be used for the sample size calculation in place of the unadjusted effect size in order to adjust for non-compliances and crossovers throughout the trial.

iii) Sample Size

For an adjusted effect size of 0.63 unit decline in functional outcome, a total sample size of 430, or 215 in each intervention group, is needed to achieve 90% power.

$\mu_I = 1.63,\ \mu_c = 1$

Standard Deviation = 2

Significance Level: 0.05, two-sided

iv) Sensitivity Analysis

Total Sample Size (power = 0.9)			
	$\delta = 0.5$	$\delta = 0.63$	$\delta = 0.75$
$\sigma = 1$	172	108	76
$\sigma = 2$	682	430	304
$\sigma = 2.5$	1064	670	474

As seen in the table above, the sample size varies as the standard deviation and effect sizes change. Standard deviation of 2 and effect size of 0.63 are chosen because they are the values used for the adjusted sample size calculation. The other two values were chosen to depict how the sample size varies accordingly. As standard deviation increases, the sample size also

increases. As the effect size increases, the sample size decreases. For this trial, the adjusted sample size of 430 is within the optimal range.

v) Interim Analysis Plan

For the group sequential design, three number of looks will be used. The maximum time will be 1, and information will be 5, 10, and 15. The O'Brien-Fleming method will be utilized when calculating the upper and lower stopping boundaries. Lower levels of significance will be ensured to the type I error. There is no placebo control intervention in this trial. The boundaries and their corresponding alpha values can be seen below. It is important to note that the lower boundaries should all be negative values.

Upper boundaries (positive):

Look	Time	Info	Lower Bndry	Upper Bndry	Nominal Alpha	Inc Alpha	Total Alpha	Inc Power	Total Power
1	0.33333	5	-3.71030	3.71030	0.00021	0.00021	0.00021	0.03402	0.03402
2	0.66667	10	-2.51141	2.51141	0.01202	0.01189	0.01210	0.52797	0.56200
3	1.00000	15	-1.99302	1.99302	0.04626	0.03790	0.05000	0.33892	0.90091

Lower boundaries (negative):

Look	Time	Info	Lower Bndry	Upper Bndry	Nominal Alpha	Inc Alpha	Total Alpha	Inc Power	Total Power
1	0.33333	5		3.20010	0.00069	0.00069	0.00069	0.06758	0.06758
2	0.66667	10		2.14080	0.01615	0.01569	0.01637	0.54048	0.60806
3	1.00000	15		1.69478	0.04506	0.03363	0.05000	0.29213	0.90020

6.0 Safety Considerations

i) How Safety Outcomes are Measured

1) Significant increase in stress levels will be measured through the perceived stress scale as continuous variables, from 0 to 40. Scores ranging from 0-13 would be considered low stress, 14-26 would be considered moderate stress, and 27-40 would be considered high perceived stress. A coordinator will visit the hospital weekly to obtain and assess the scores.
2) Adverse reactions towards BXCL701 will be measured as categorical variables, specifically YES and NO. A coordinator will visit the hospital weekly to assess whether any adverse reactions have occurred. It is important to note that there is 24/7 emergency service, and that the patients should call for help immediately if they encounter any adverse reactions prior to assessments.
3) Allergic reactions towards BXCL701 will be measured as categorical answer of YES or No. A coordinator will visit the hospital weekly to conduct skin prick tests in order to check for allergic reactions.

ii) Specific Safety Outcome Importance

1) Significant increase in stress levels is important to monitor in regards to mCRPC because stress will not only worsen patient condition mentally, but physically as well. This will negatively impact mCPRC and make it more difficult to recover.
2) Adverse reaction towards BXCL701 is important to monitor due to its severely negative impact on patient health and puts them at risk of a variety of medical emergencies.
3) Allergic reaction towards BXCL701 is important to monitor for the same reasons, as it severely negatively impacts patient health and puts them at risk of a variety of medical emergencies.

iii) General Safety Outcomes

This trial will also monitor urination. mCPRC symptoms often include trouble urinating in addition to bloody urination. A coordinator will visit the hospital weekly to assess urination status, urination frequency, and urination color.

7.0 Limitations and Late-breaking Problems

The perceived stress scale scores may be skewed due to self-reporting. Since the test only encompasses 10 questions with a scale of 0-4 for each question, it is likely that patient interpretations will vary greatly thus lowering its comparability and validity.

8.0 References

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9.0 Appendix

Appendix 1:

Unadjusted effect size $\delta = 0.90$

Then, $\mu_I - \mu_c = 0.90$

Let $\mu_I = 1.90,\ \mu_c = 1$

For intervention group, adjusted mean outcome is:

$$1 * 0.05 + 0.2 * \frac{2.90}{2} + 0.75 * 1.90$$

This is equal to 1.76.

For intervention group, adjusted mean outcome is:

$$1.90 * 0.05 + 0.2 * \frac{2.90}{2} + 0.75 * 1$$

This is equal to 1.13.

Therefore, adjusted effect size is equal to 1.76 minus 1.13, which equals 0.63.